

Mobilization of bacteriophages from soil matrix to soil water in the Attert River basin: A case study

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vEGU general assembly 2021



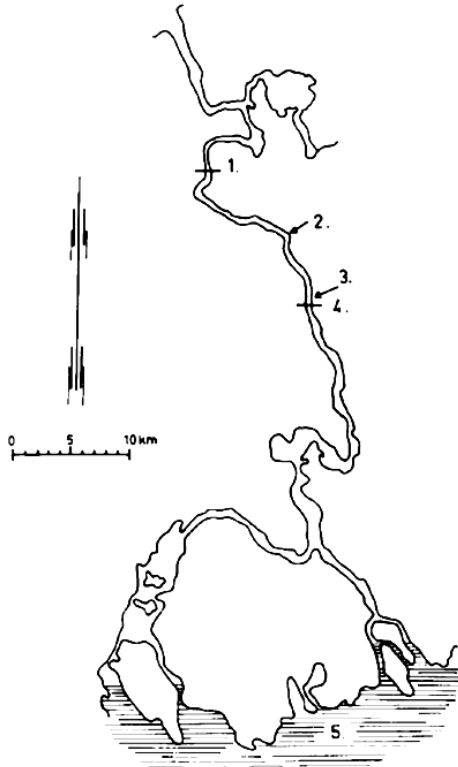
30th April, 2021



Premises of viruses in hydrology

In 1968, Kinnunen and Niemelä ¹

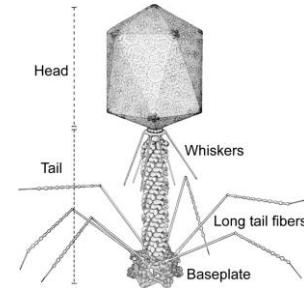
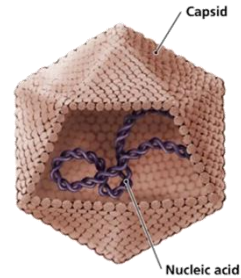
- First tracing experiment using particular viruses



River Kymijoki, Finland

Bacteriophages

Viruses which specifically infect bacteria



Bacteriophages in tracing experiments

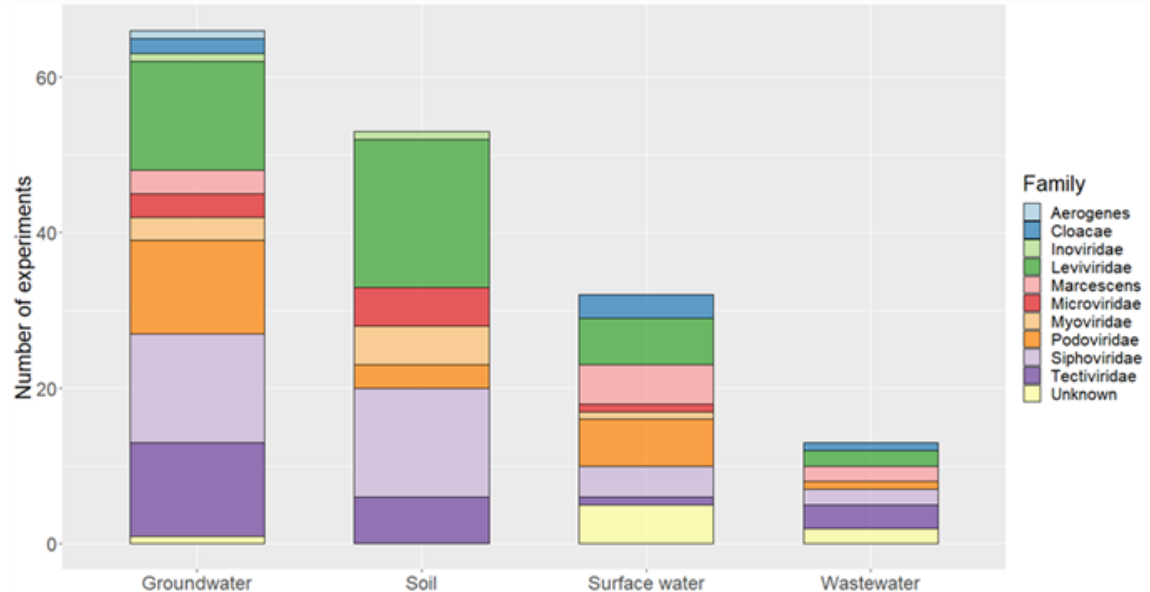
ADVANTAGES

- 1°) bacterial specificity
- 2°) low detection limit
- 3°) eco-friendly property

In total,

- > 100 tracing experiments
- Injection method
- 30 different phages

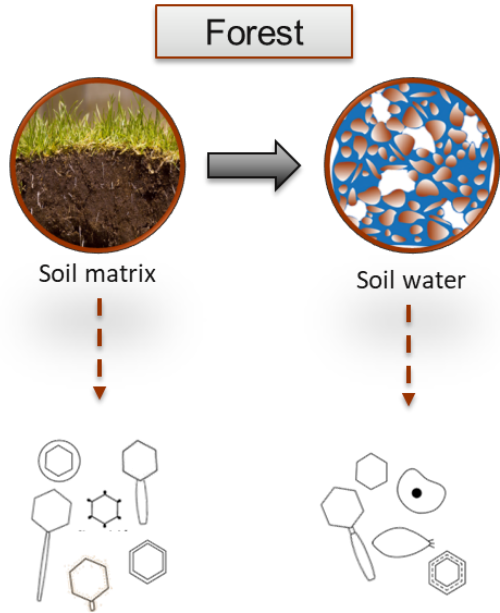
Most studied compartment: groundwater



Prev.

Next

What do we expect?

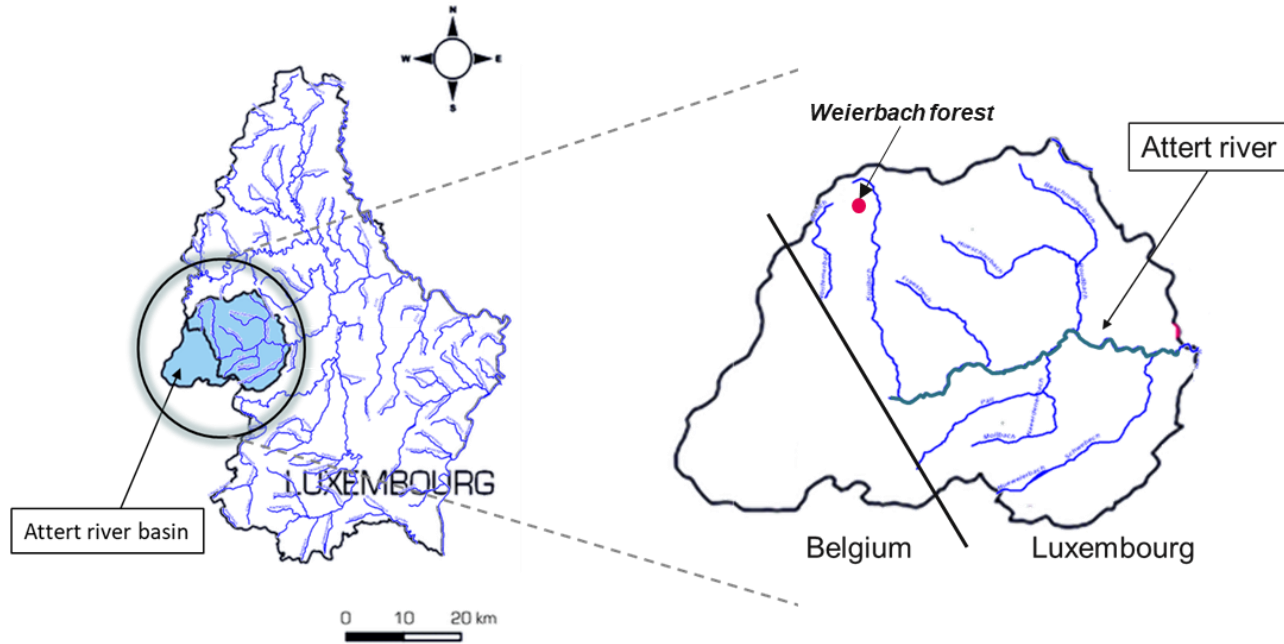


Similarities/Dissimilarities?

Hypothesis:

Some phages of the existing viral community of soil matrices are preferentially transferred towards the soil water during a rainfall events, depending on the phage and soil characteristics

The studied site: the Attert River basin



Weierbach forest

Prev.

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Methodology: shotgun metagenomics



DNA extraction

- *To extract viral DNA*
- *PowerSoil kit*



Library preparation

- *To prepare library sequencing for small genomes*
- *Nextera XT kit*



Sequencing

- *To determine nucleic acid sequences*
- *Illumina NovaSeq 6000*



Bio-informatics

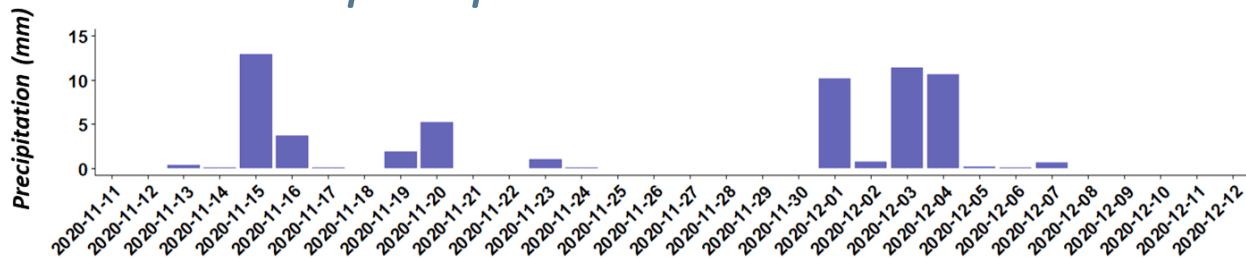
- *Sort sequences*
- *Identification of phage populations*



Prev.

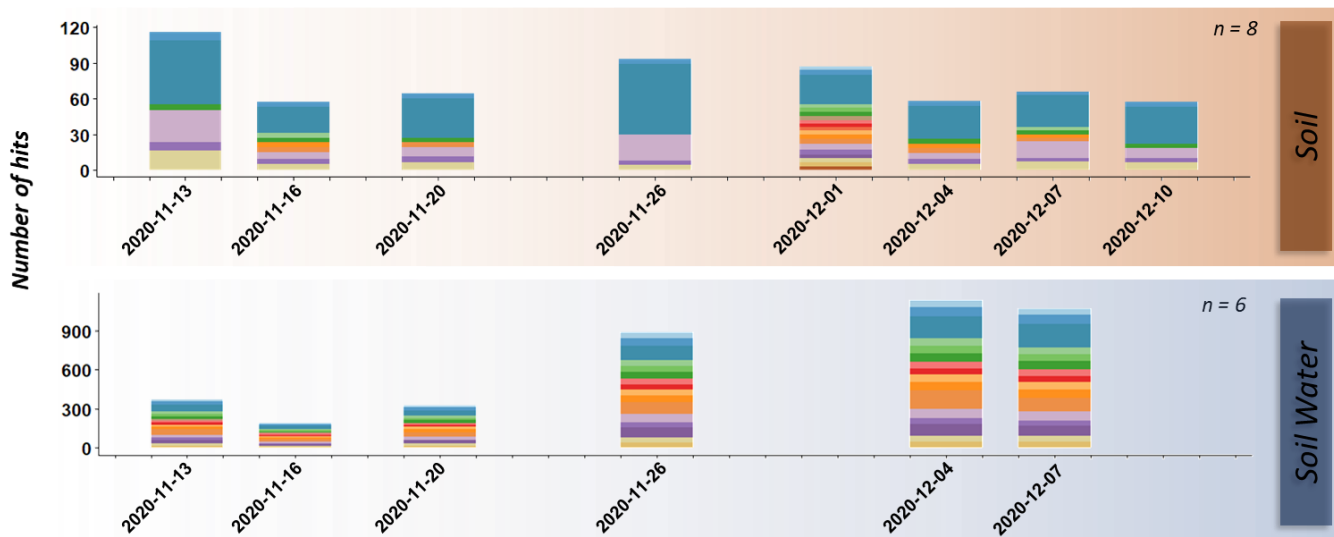
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Results #1: Phage species in both soil and soil water regarding the precipitation events



Variation in the species populations occur:

- In soil, it decreases while a precipitation happens and increases during a drought period
- In soil water, the population increases after the first precipitation event



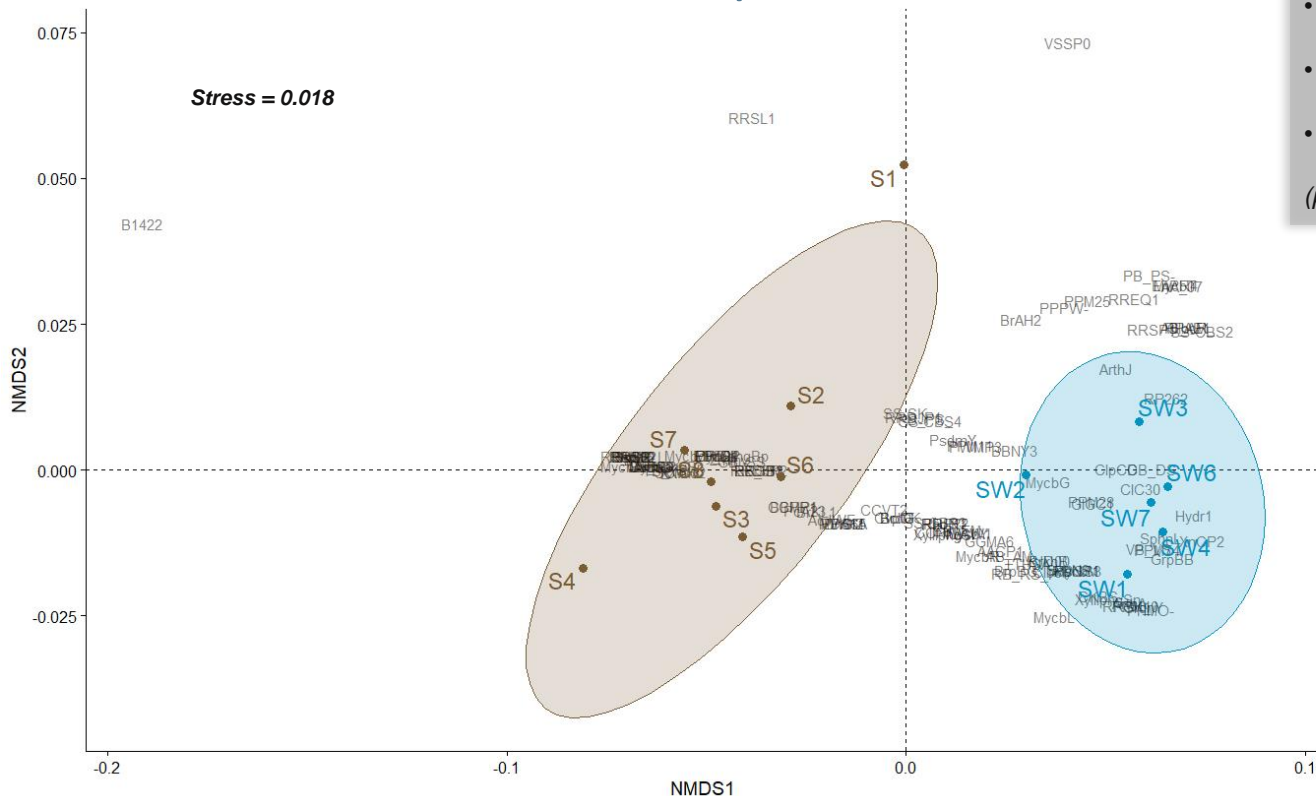
Species

- Arthrobacter phage Adat
- Arthrobacter phage Jasmine
- Bdellovibrio phage phi1422
- Caulobacter phage CcrColossus
- Caulobacter phage Cr30
- Dinoroseobacter phage vB_DshS-R5C
- Gluconobacter phage GC1
- Gordonia phage BritBrat
- Hydrogenobaculum phage 1
- Mycobacterium phage Gaia
- Pseudomonas phage PaMx28
- Pseudomonas phage PaMx74
- Ralstonia phage phiRSL1
- Rhodoferrax phage P26218
- Sphingobium phage Lacusarx
- Vibrio phage SSP002
- Vibrio phage vB_VpaS_MAR10
- Xanthomonas citri phage CP2

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Results #2: Representation of phage species according to the studied compartments



- Soil samples are all similar from one another
- Soil water samples have some dissimilarities
- Soil and soil water are significantly different from each other ($p.value < 2e-16, \alpha = 0.05$)

Samples	Kruskal - DunnTest
S1	a
S2	a
S3	a
S4	a
S5	a
S6	a
S7	a
S8	a
SW1	bc
SW2	b
SW3	c
SW4	d
SW6	d
SW7	d

$\alpha = 0.05$

Same letter = similar; Different letter = different

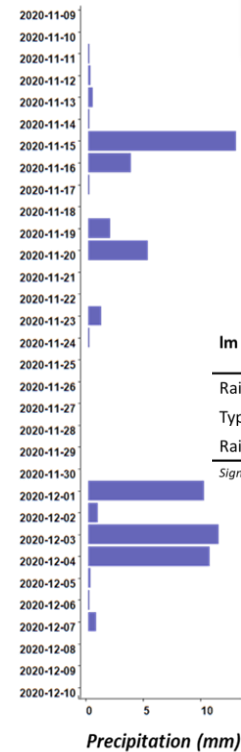
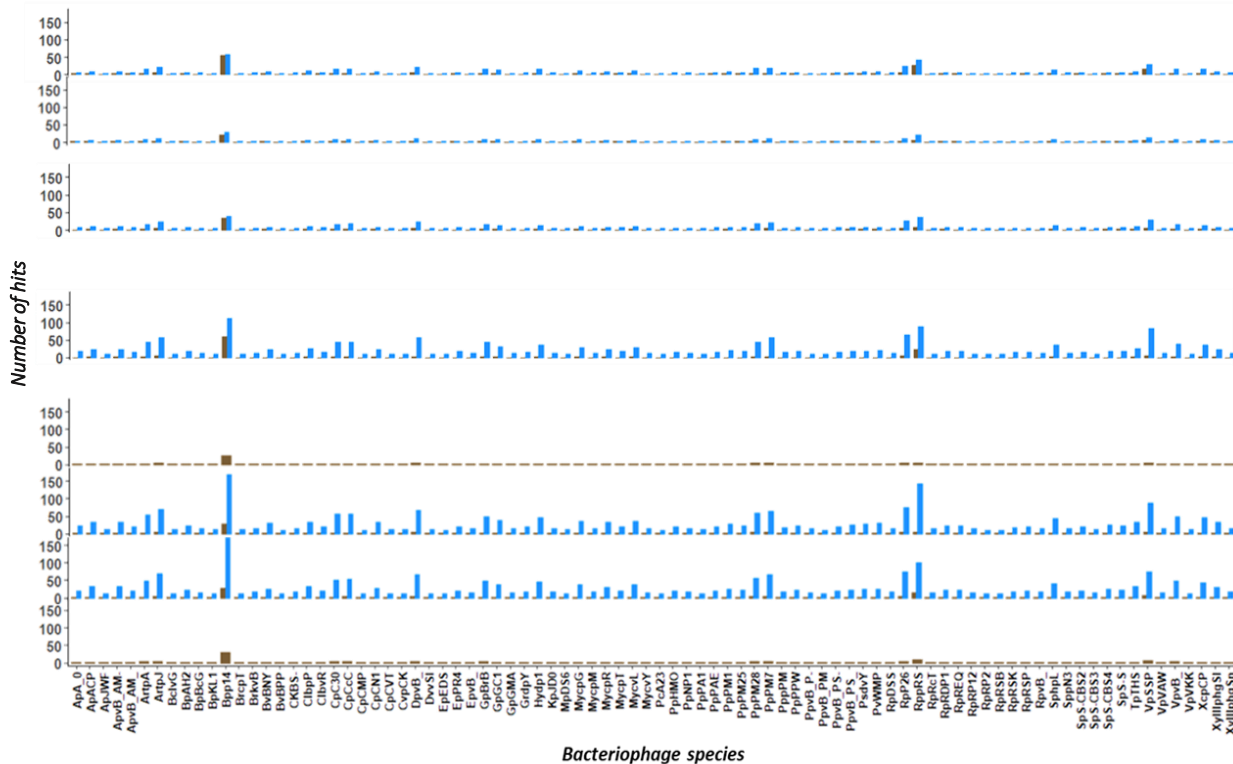
Fig. Non-metric multidimensional scaling (NMDS) of species (in grey) and compartments (brown for soil and blue for soil water)



Results #3: Number of phage species for all samples

- Precipitation alone has not effect on the species distribution
- The species distribution is impacted by both rain event and type of compartment

Compartments: ■ Soil (N = 8) ■ Soil water (N = 6)



ANOVA	df	P-value
Rain	1	0.34820
Type	1	< 2e-16 ***
Rain:Type	1	0.00307 **
Residuals	1592	

Sign: **** 0.001 *** 0.01 ** 0.05 * 0.1 ' ' 1

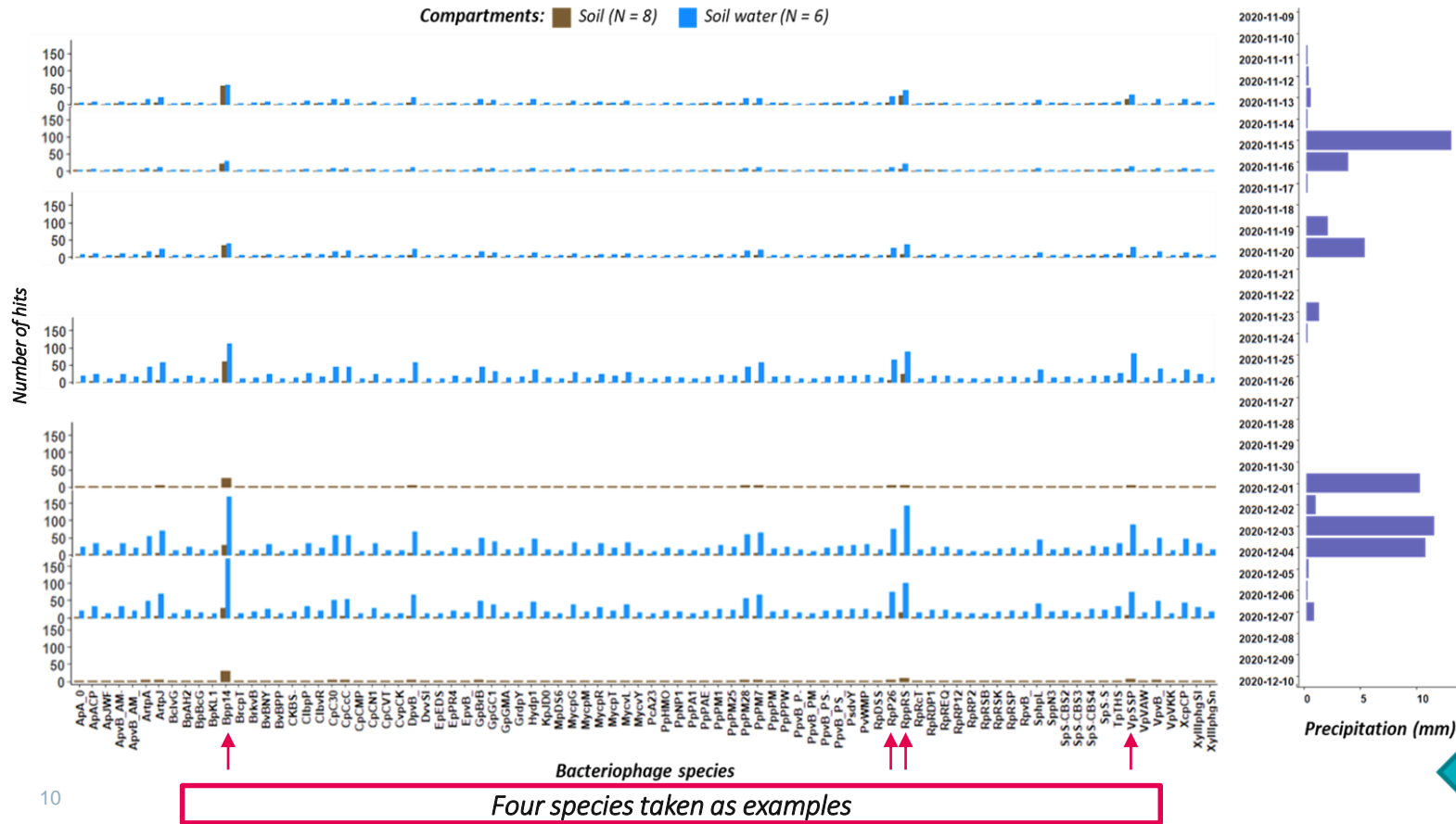
lm	Regression coefficients	Std. error	P-value
Rain	0.2064	0.0777	0.00798 **
Type	7.7174	0.5815	< 2e-16 ***
Rain:Type	0.3258	0.1099	0.00307 **

Sign: **** 0.001 *** 0.01 ** 0.05 * 0.1 ' ' 1

*Hit = correspondence between a query sequence and the database

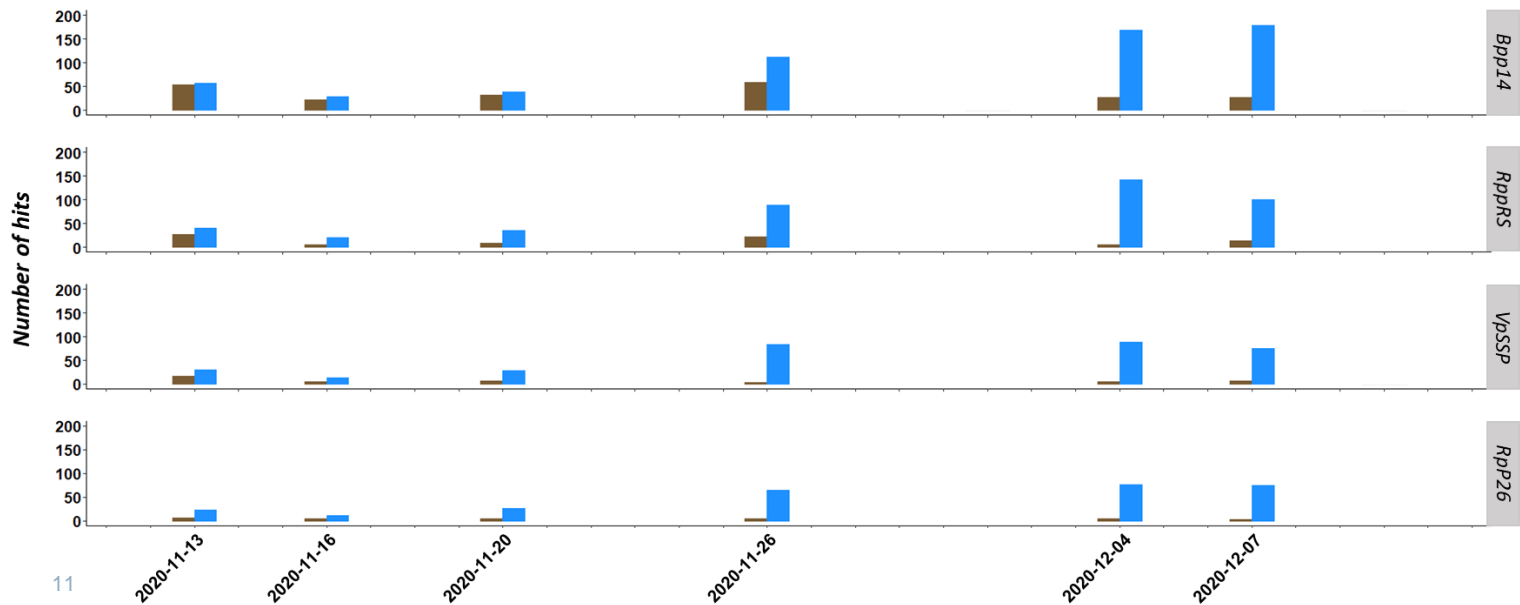
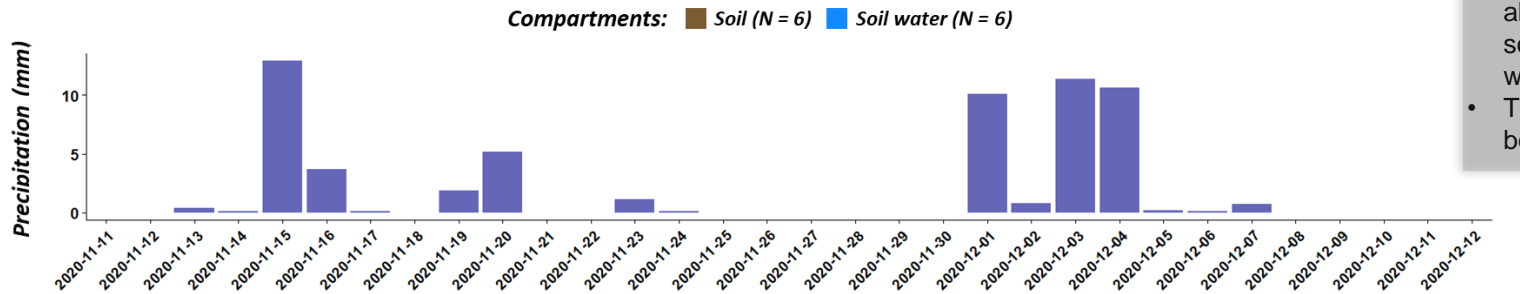


Results #3: Number of phage species for all samples



Results #3: Number of phage species for all samples

- For a same specie, their abundance decreases in soil while it increases in soil water
- This forms a gap between both kind of populations



Conclusion

- The bacteriophage populations in soil and soil water are well different from one another
- More precisely, some species from soil samples are less abundant than in their respective soil water samples

➔ This gives an insight about the mobilization of bacteriophage from soil to soil water

However:

- Is the precipitation responsible for this change ?
- If so, how does the change in population respond to the rain event ?