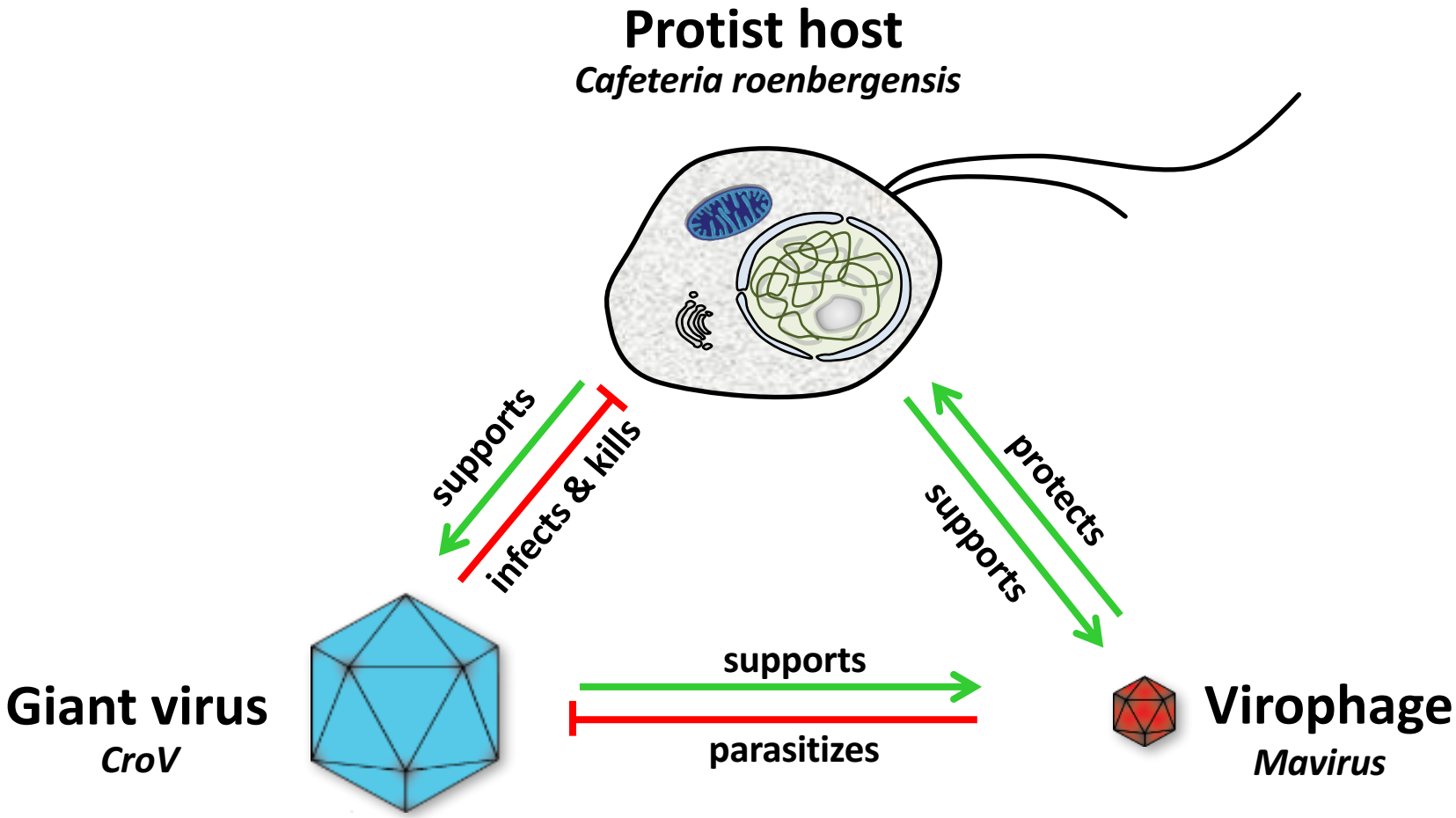
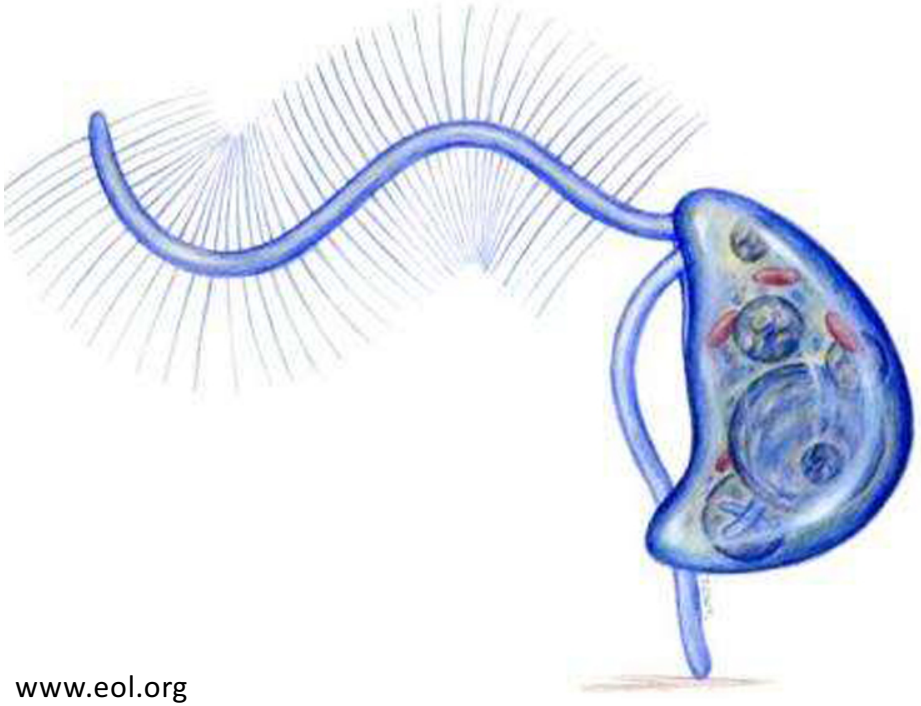
Heterotrophic protists as a genomic hub for virophages and other mobile genetic elements



Matthias Fischer
MPI for Medical Research
Heidelberg, Germany

MAX-PLANCK-GESELLSCHAFT
MPI for Medical Research

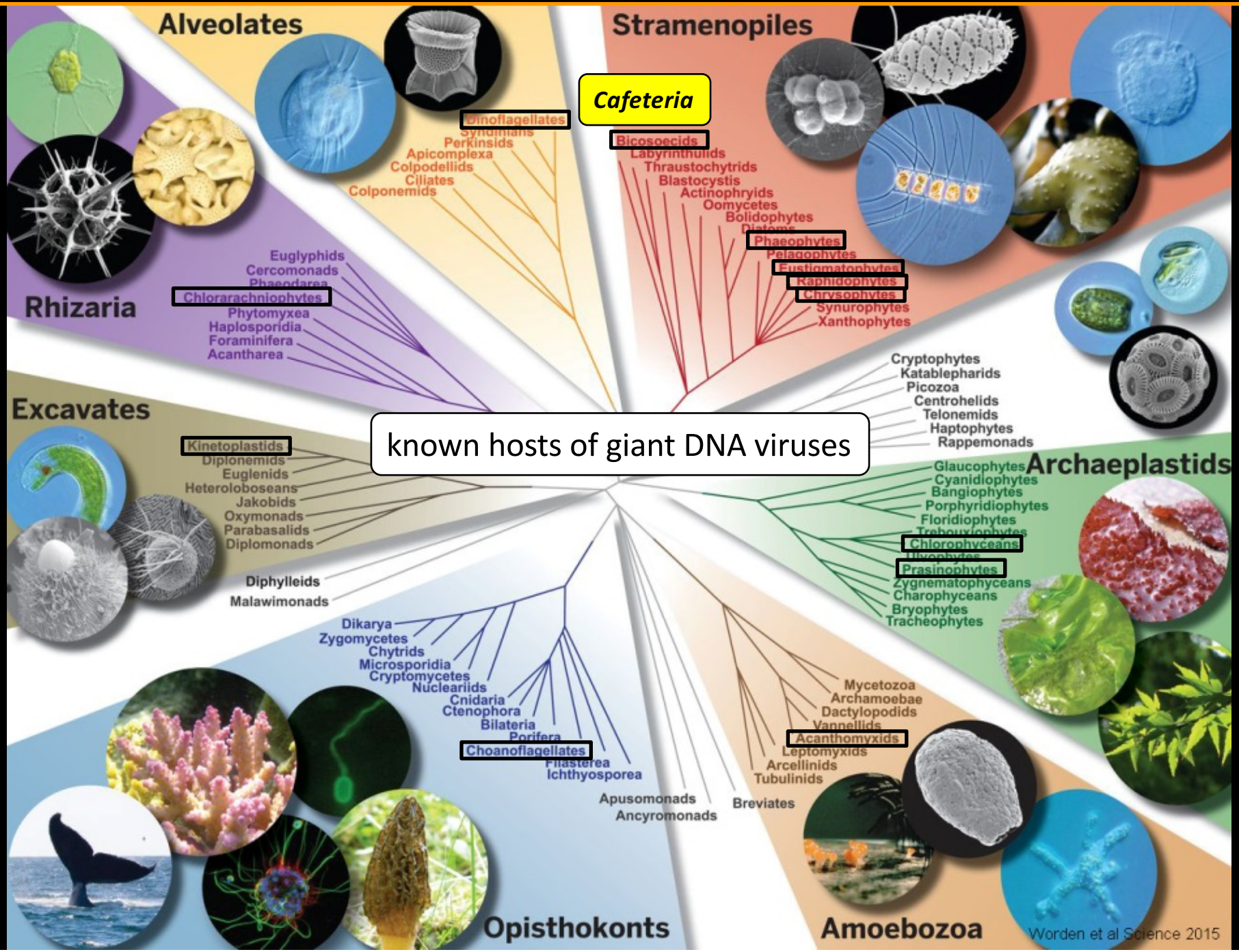
Host: *Cafeteria roenbergensis*



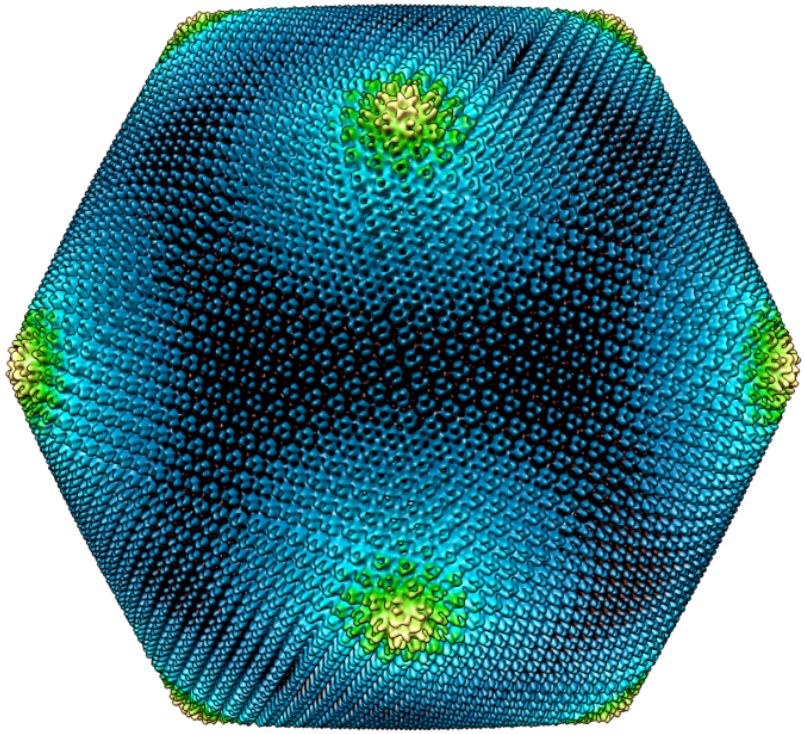
www.eol.org

- marine heterotrophic nanoflagellate
- ecologically important bacterivore
- Ø 5-10 µm
- 30 Mpb diploid genome, asexual (?)

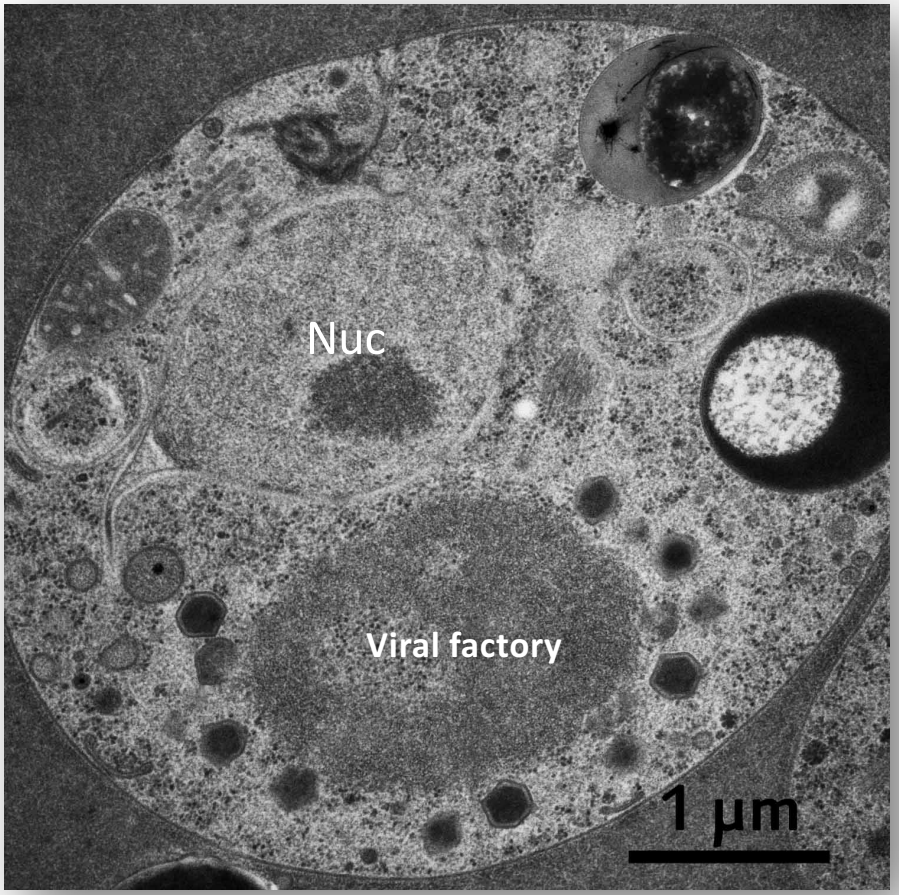
Cafeteria is a deep-branching stramenopile



Parasite: Cafeteria roenbergensis virus (CroV)



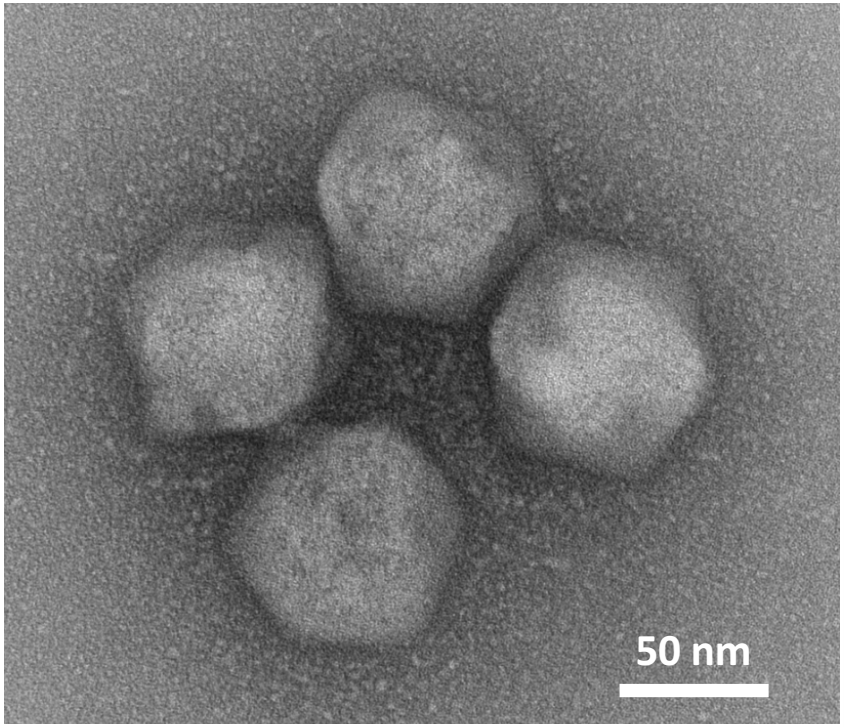
- capsid Ø = 300 nm
- T number = 499
- 691,790 bp dsDNA
- > 600 proteins
- 57 tRNA genes
- member of *Mimiviridae*

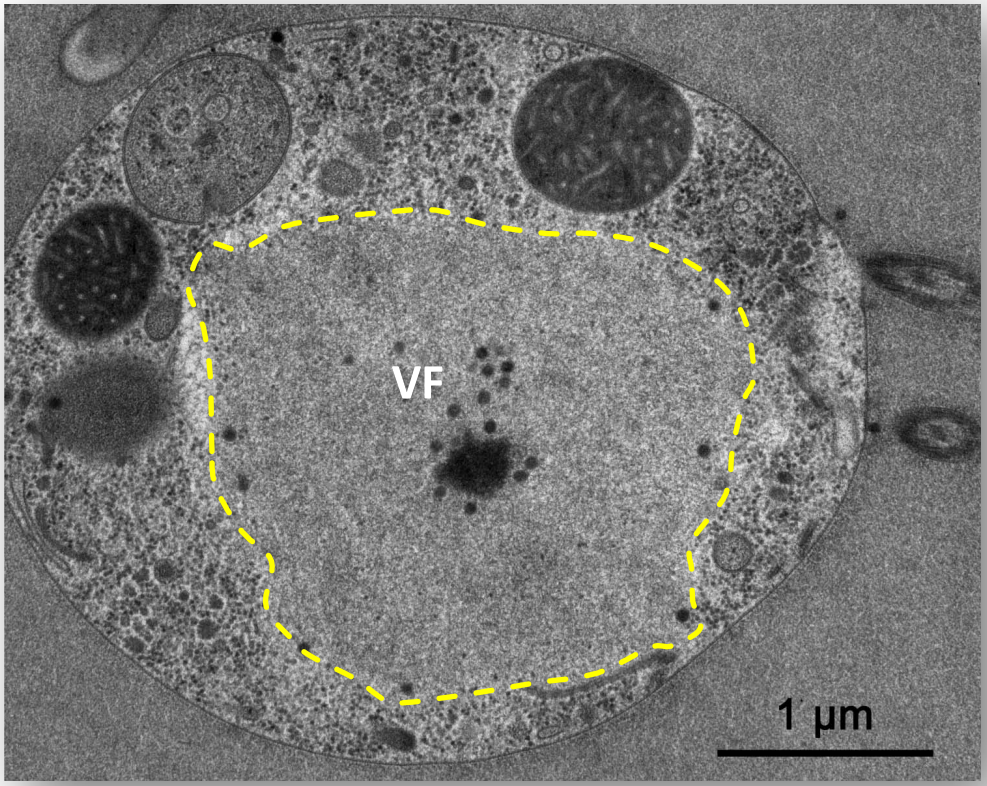


Electron microscopy by Ulli Mersdorf, MPImF

Fischer et al (2010) **PNAS** 107:19508
Xiao, Fischer et al (2017) **Sci Rep** 7:5484

Hyperparasite: the virophage mavirus





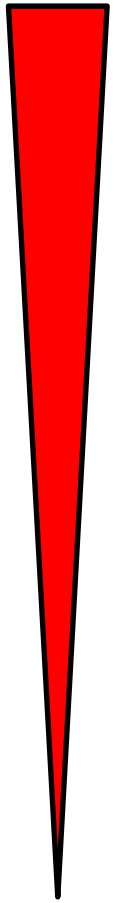
capsid Ø = 75 nm
19 kb dsDNA
20 ORFs

→ Mavirus is highly virulent for CroV

Fischer & Suttle (2011) **Science** 332:231

Virulence towards giant viruses varies among virophages

strong inhibition

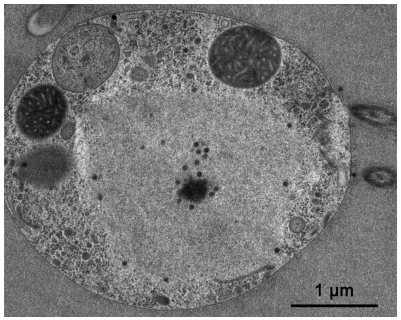


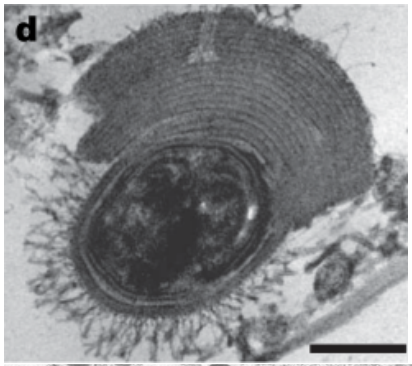
no inhibition

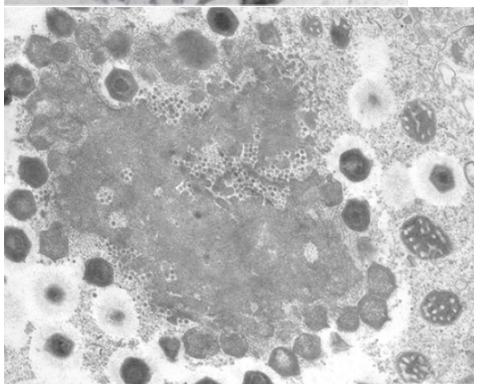
Mavirus ⊣ **CroV**

Sputnik ⊣ **Mimivirus**

Zamilon ⊣ **Mimivirus**





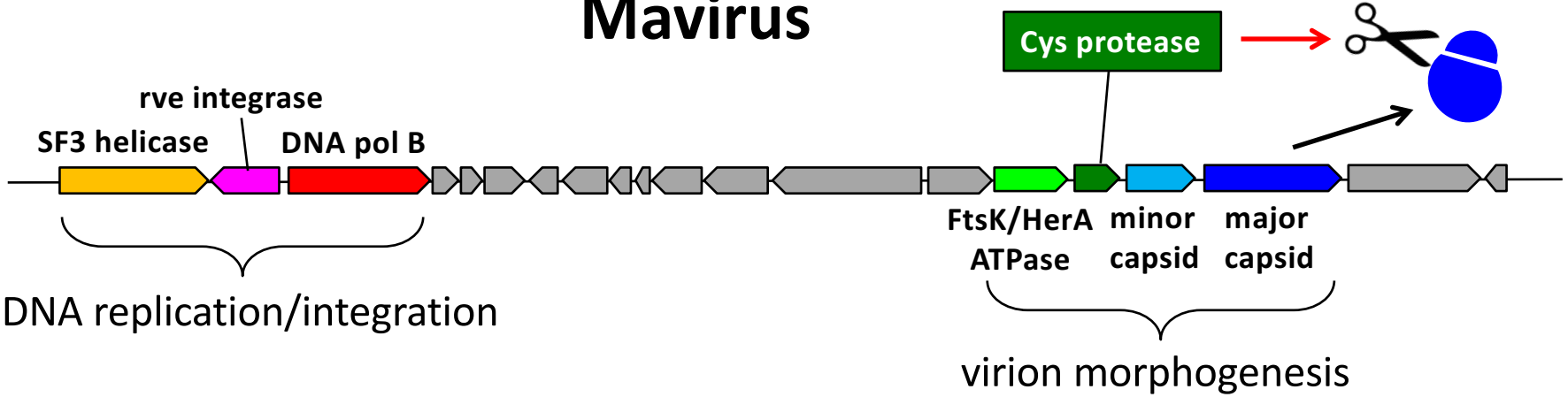


Fischer & Suttle (2011) **Science** 332:231
La Scola, Desnues et al (2008) **Nature** 455:100
Gaia et al (2014) **PLoS One** 9:e94923

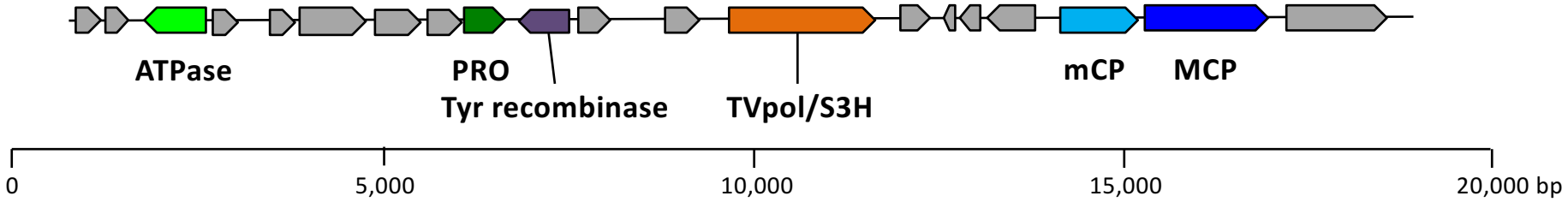
Virophages genomes are modular

Born et al (2018) **PNAS** Jun 25

Mavirus

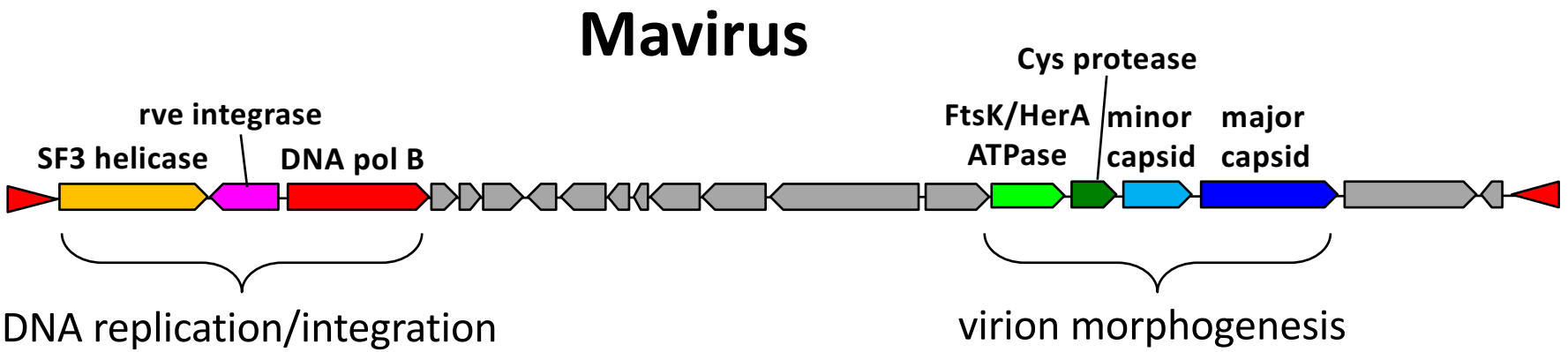


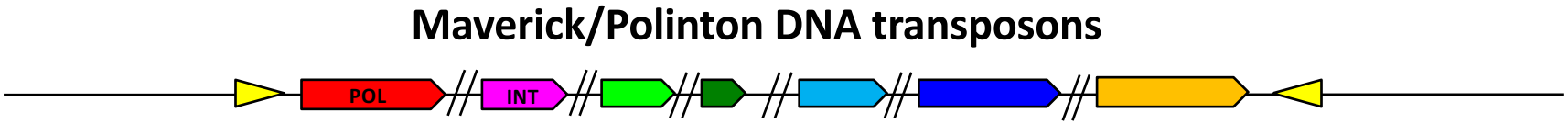
Sputnik



Conserved morphogenesis genes:

Mavirus is related to endogenous eukaryotic elements





- 15-20 kb long
- flanked by inverted repeats
- found in protists, nematodes, insects, vertebrates, fungi

Trichomonas vaginalis:
30% of genome are polintons

Feschotte & Pritham (2005) **Trends Genet** 21
Kapitonov & Jurka (2006) **PNAS** 103

Virophage genome integration

Sputnik integrated in mimivirus:

Provirophages and transpovirons as the diverse mobilome of giant viruses (2012) **PNAS** 109

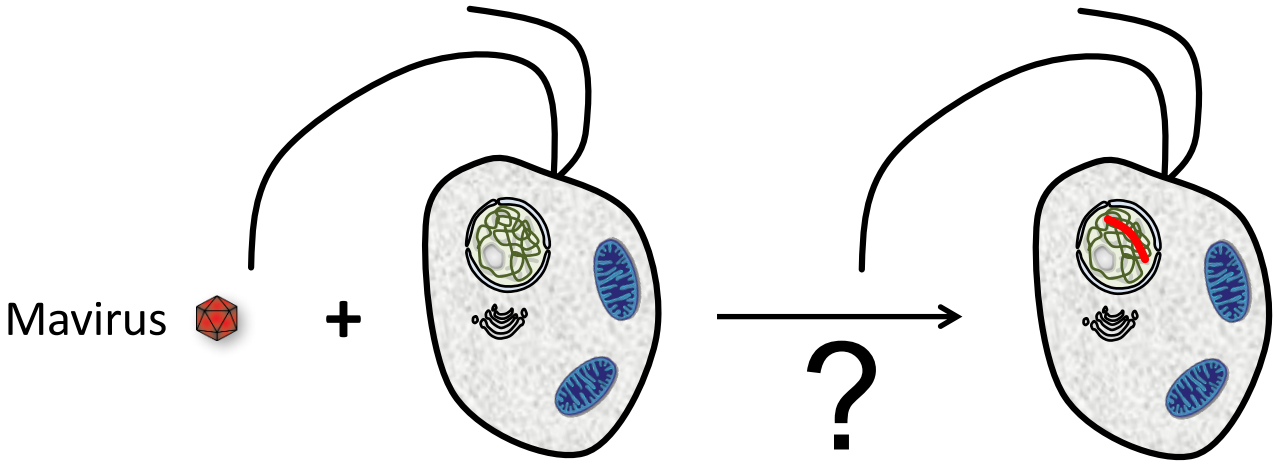
Christelle Desnues[a,1], Bernard La Scola[a,1], Natalya Yutin[b], Ghislain Fournous[a], Catherine Robert[a], Saïd Azza[a], Priscilla Jardot[a], Sonia Monteil[a], Angélique Campocasso[a], Eugene V. Koonin[b], and Didier Raoult[a,2]

Endogenous virophages in an algal genome:

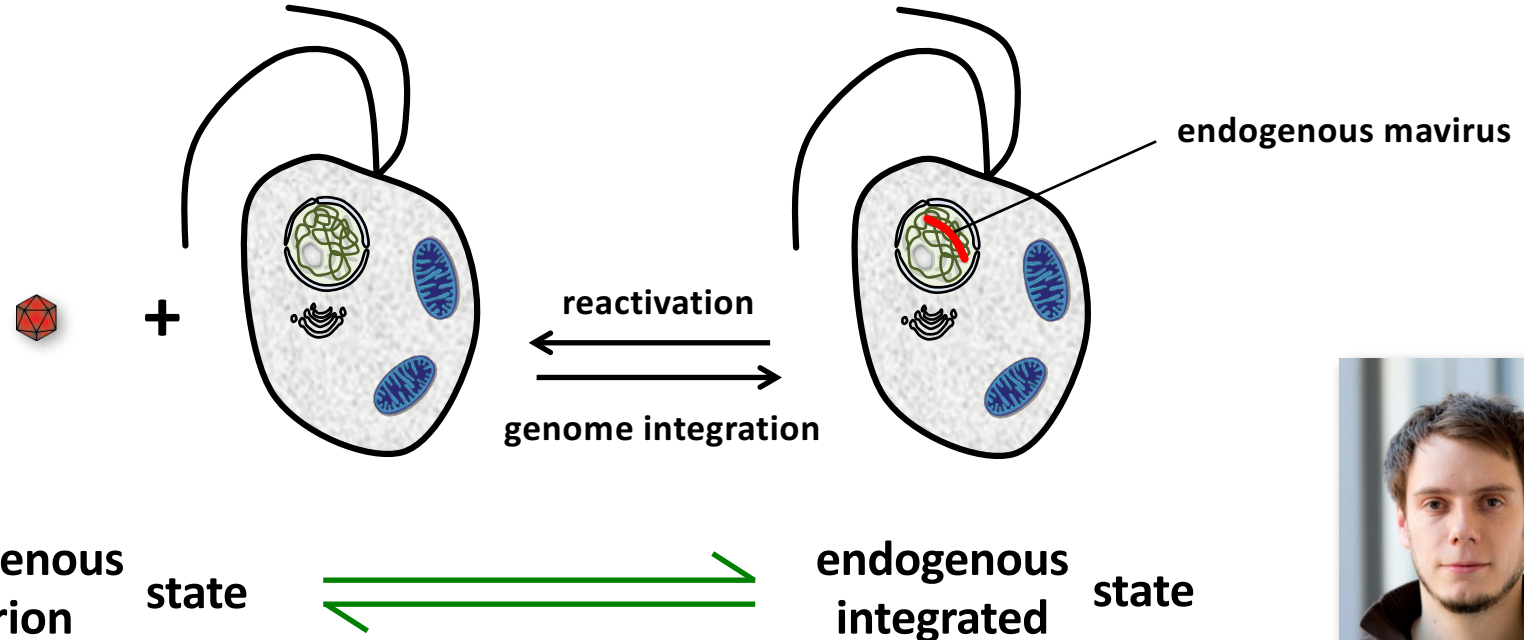
Provirophages in the Bigelowiella genome bear testimony to past encounters with giant viruses

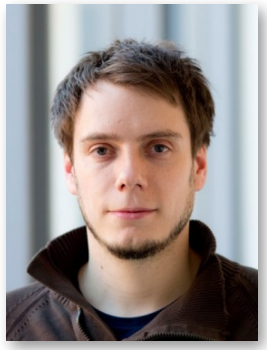
Guillaume Blanc[a,1,2], Lucie Gallot-Lavallée[a], and Florian Maumus[b,1,2] (2015) **PNAS** 112

Endogenous mavirus in *Cafeteria* genome?

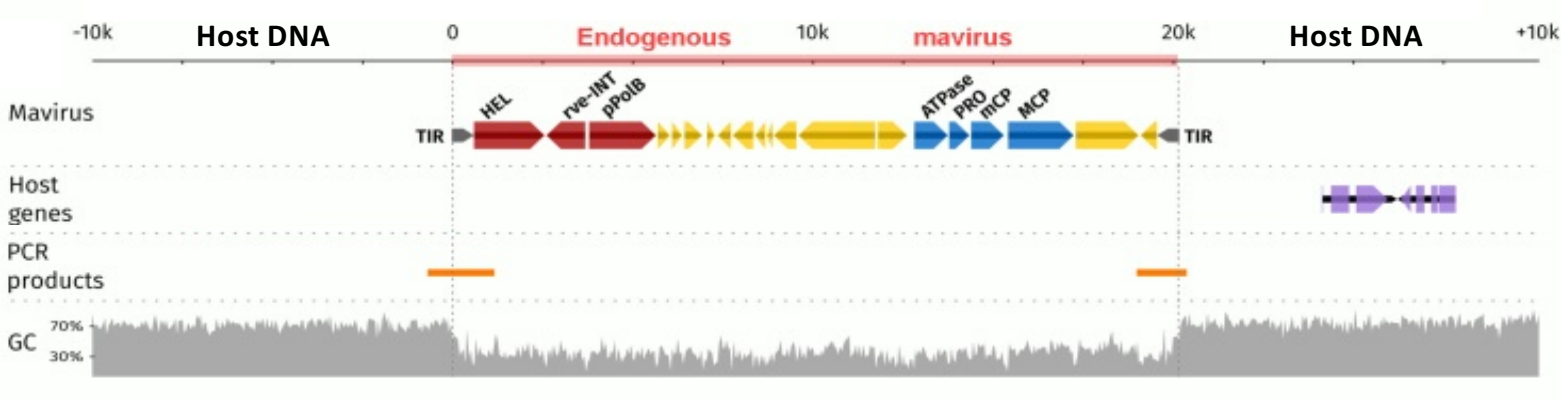


Mavirus integrates into the host genome



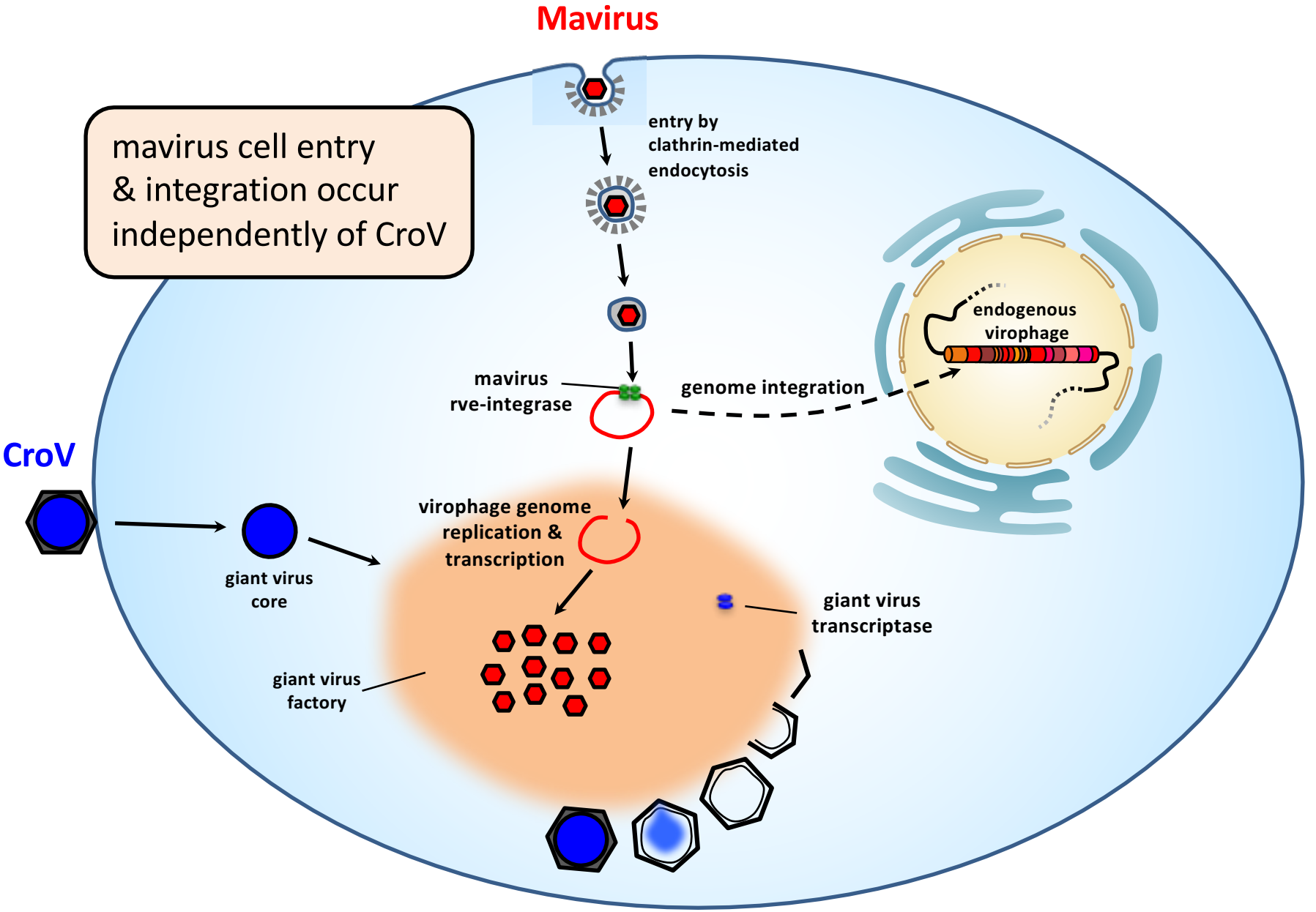


Thomas Hackl



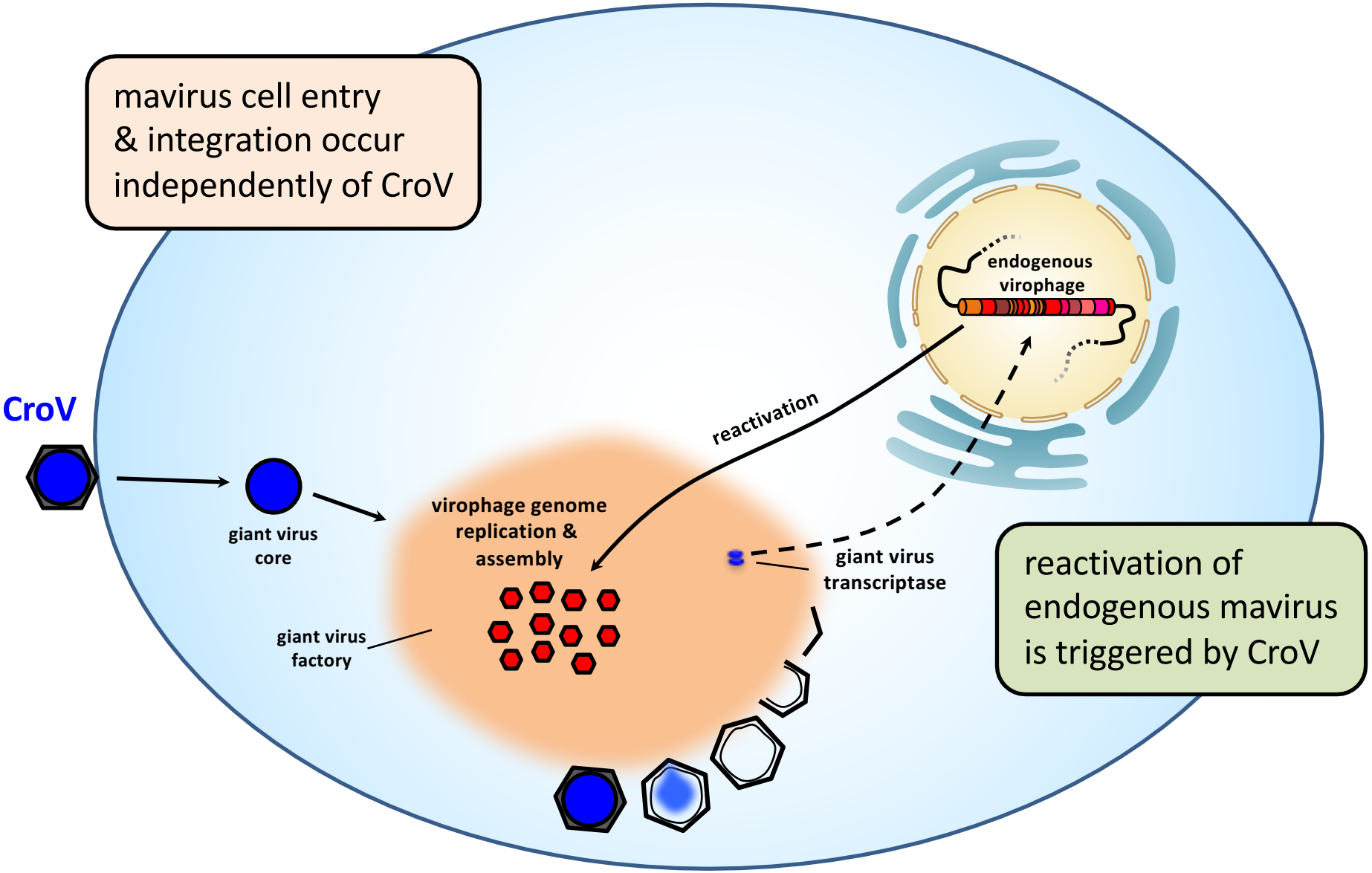
Fischer & Hackl (2016) **Nature** 540:288

The mavirus infection cycle

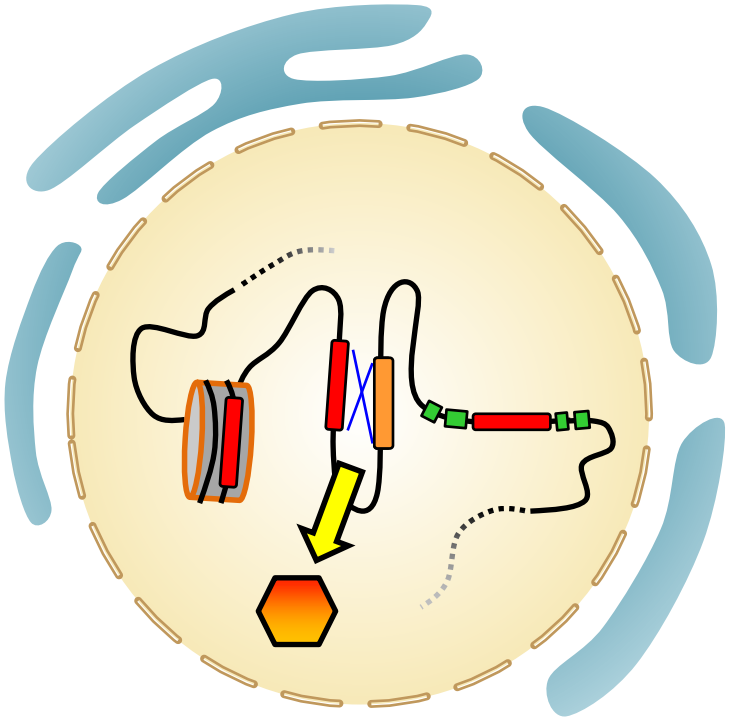


Born et al (2018) **PNAS** Jun 25

The mavirus infection cycle

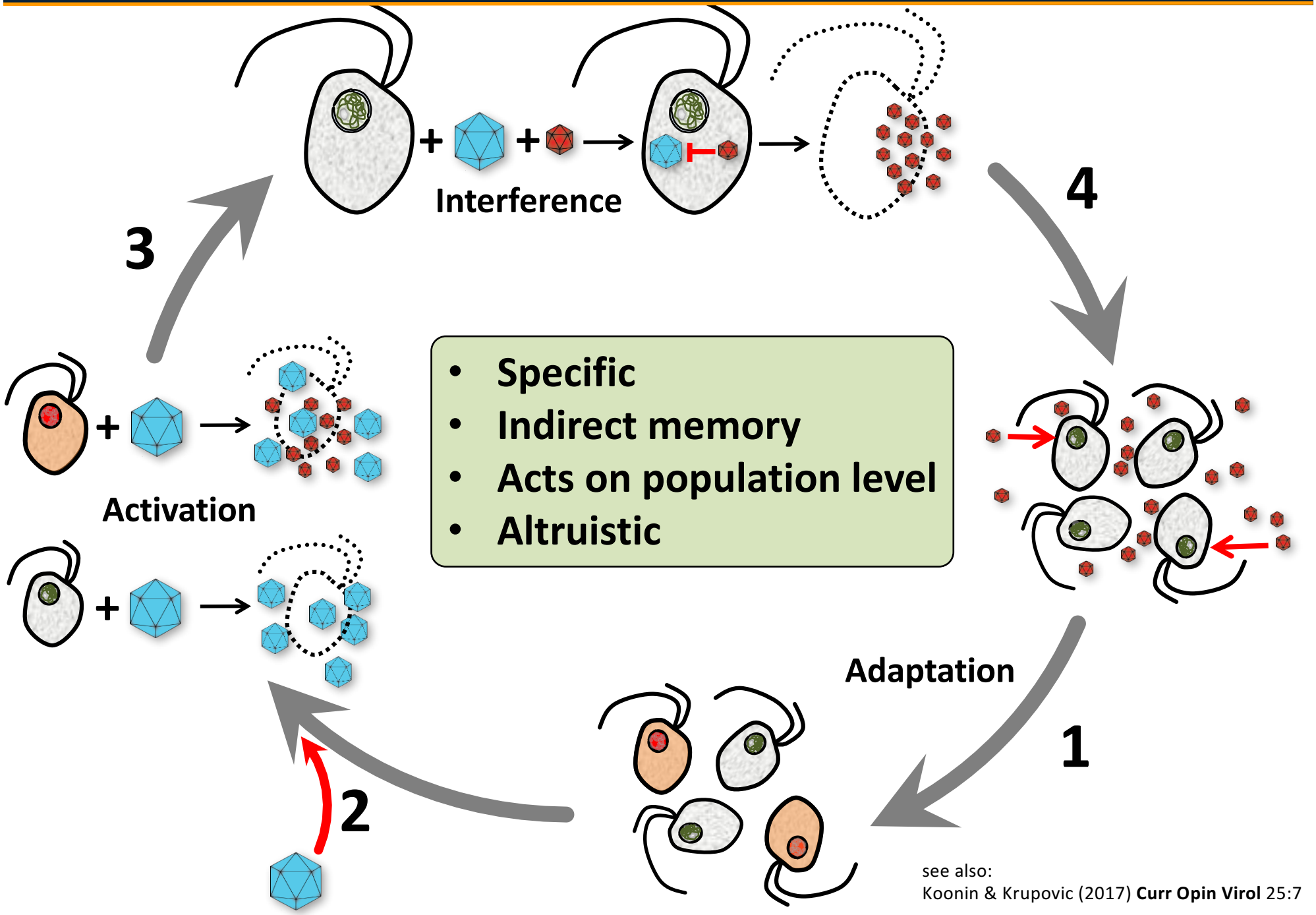


Cost imposed by endogenous virophages?



- cost of replicating an endogenous virophage genome (20 kb in 30 Mb → 0.07%)
- positional effects?
  - insertion into heterochromatin → not reactivatable
  - insertion into host genes → less fit / lethal phenotype
- multiple insertions → genomic rearrangements → instability or novelty?
- recombination with other endogenous viruses/transposons → new properties

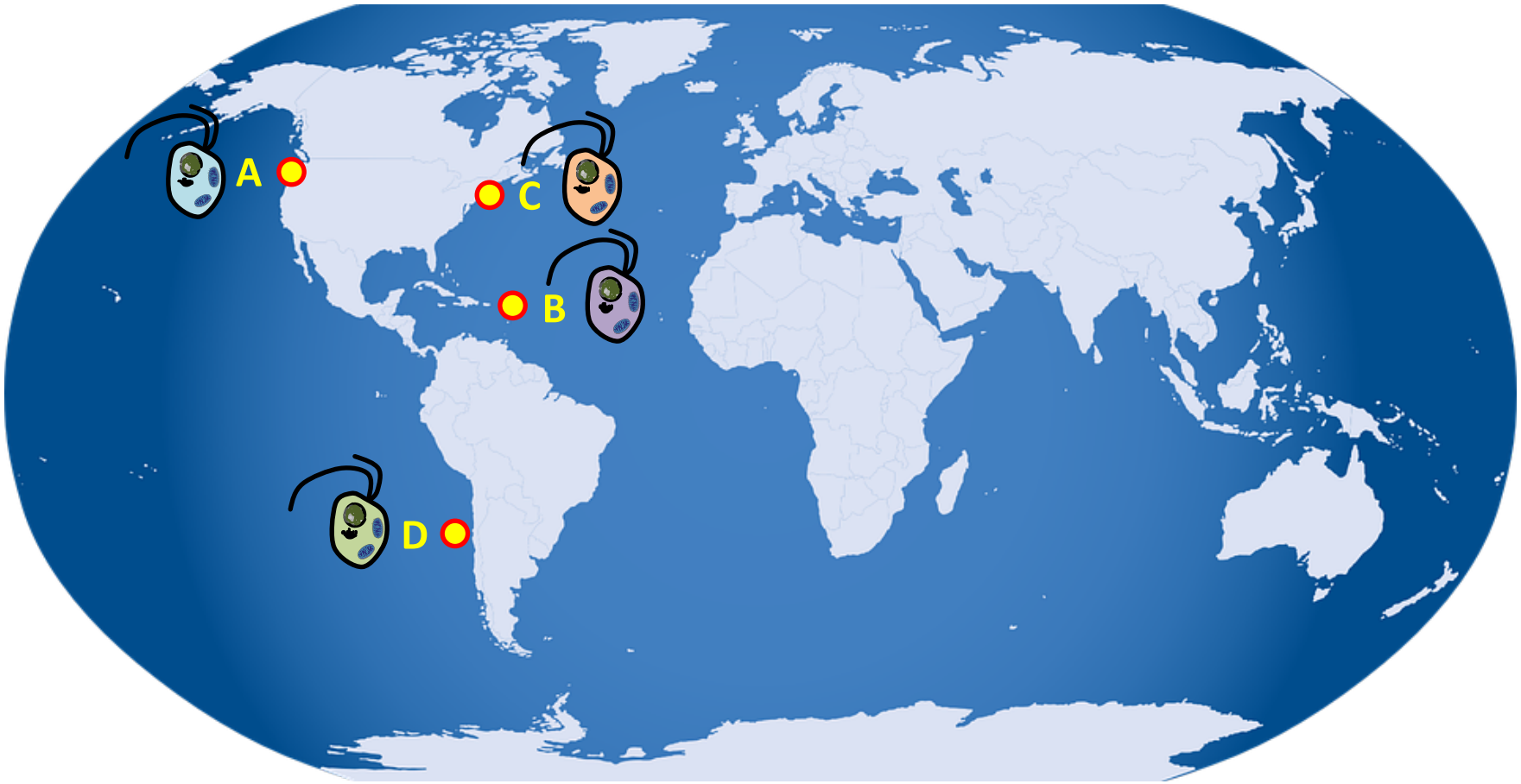
Virophages may provide adaptive population immunity



see also:
Koonin & Krupovic (2017) **Curr Opin Virol** 25:7

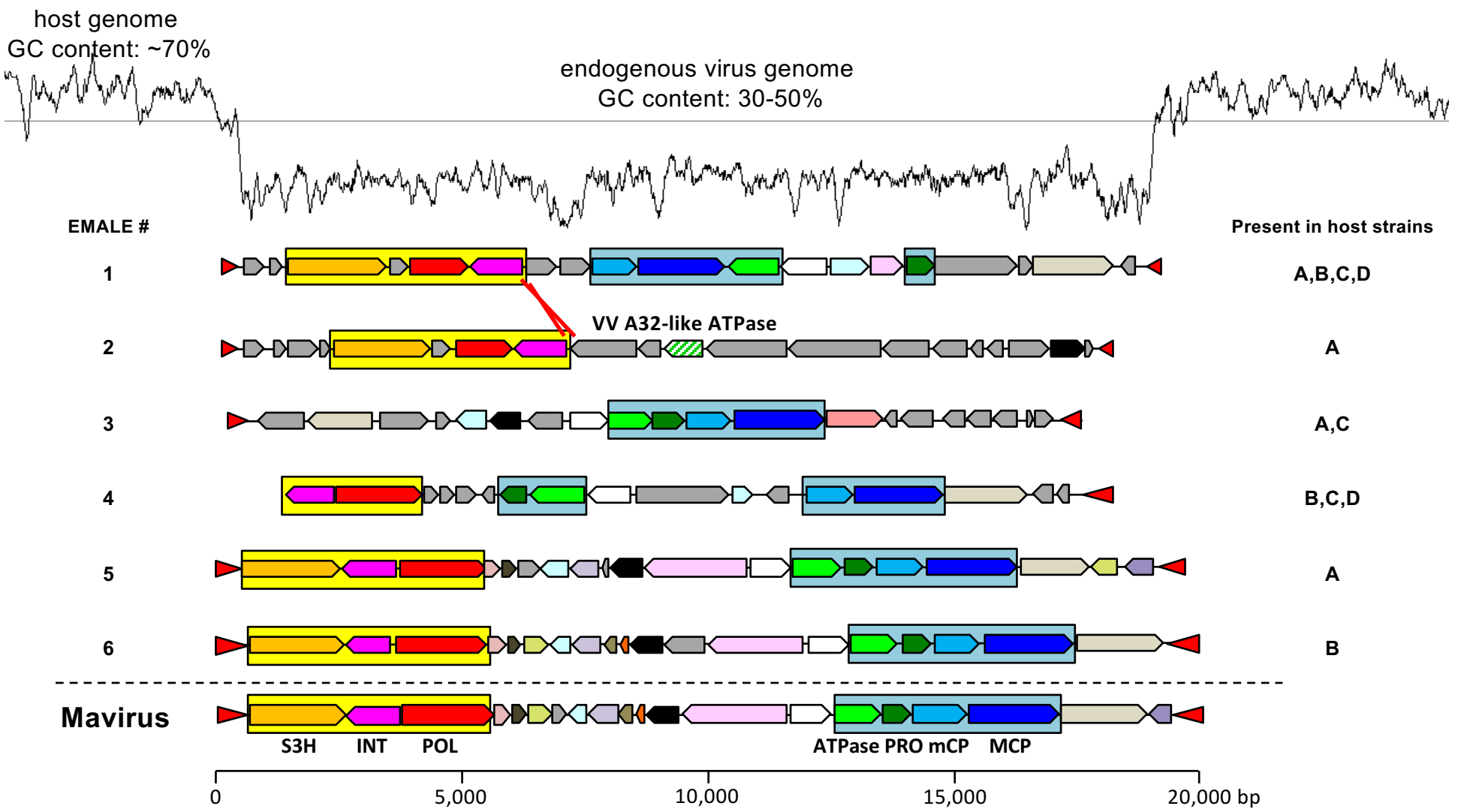
How widespread are endogenous virophages?

Four *Cafeteria roenbergensis* strains
Genome sequencing (MiSeq & PacBio)
Assembly (Proovread)



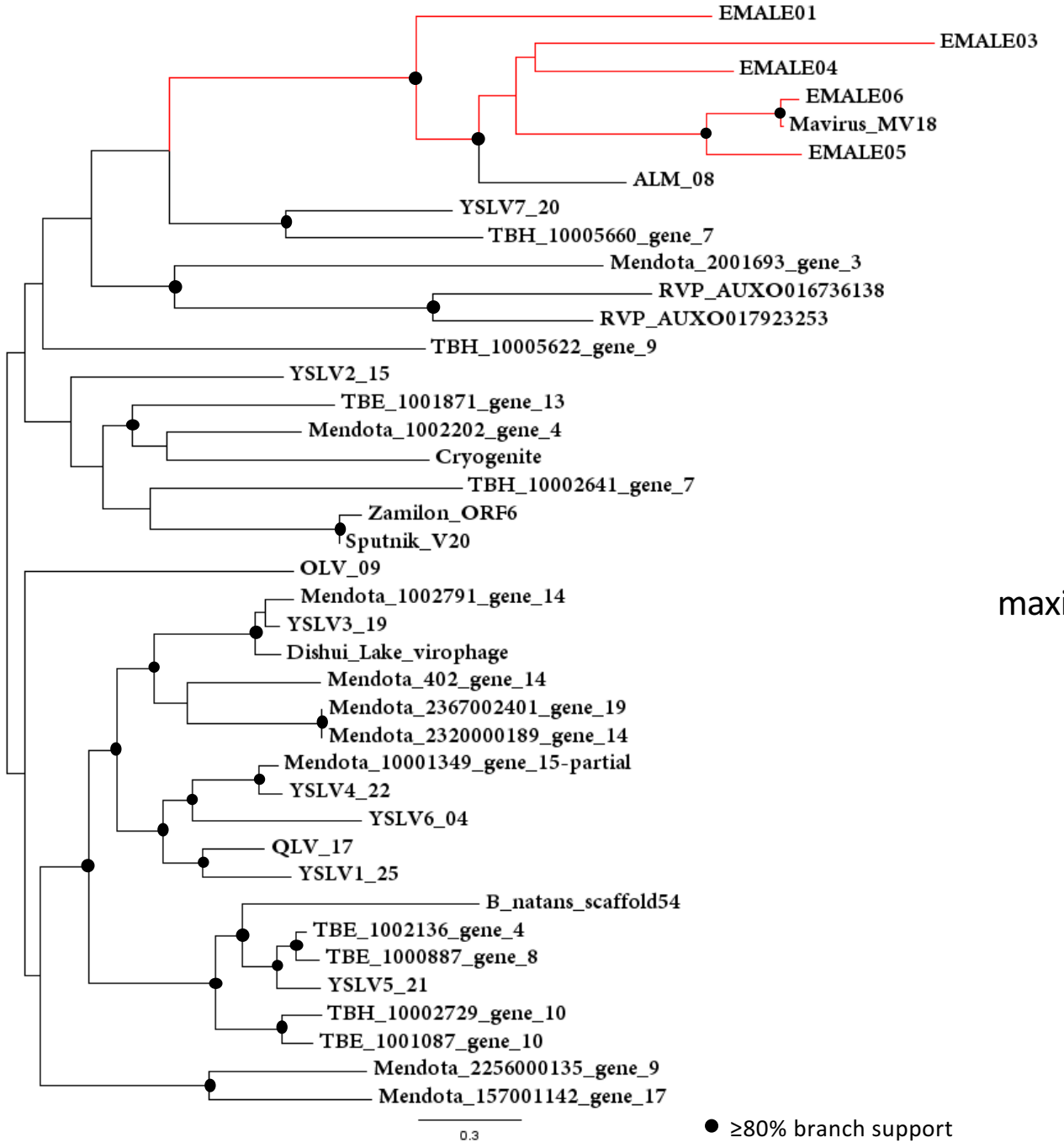
Endogenous virophages in natural *Cafeteria* strains

Six distinct versions of Endogenous Mavirus-Like Elements (EMALEs)



Hackl, Pfitzer & Fischer, in preparation

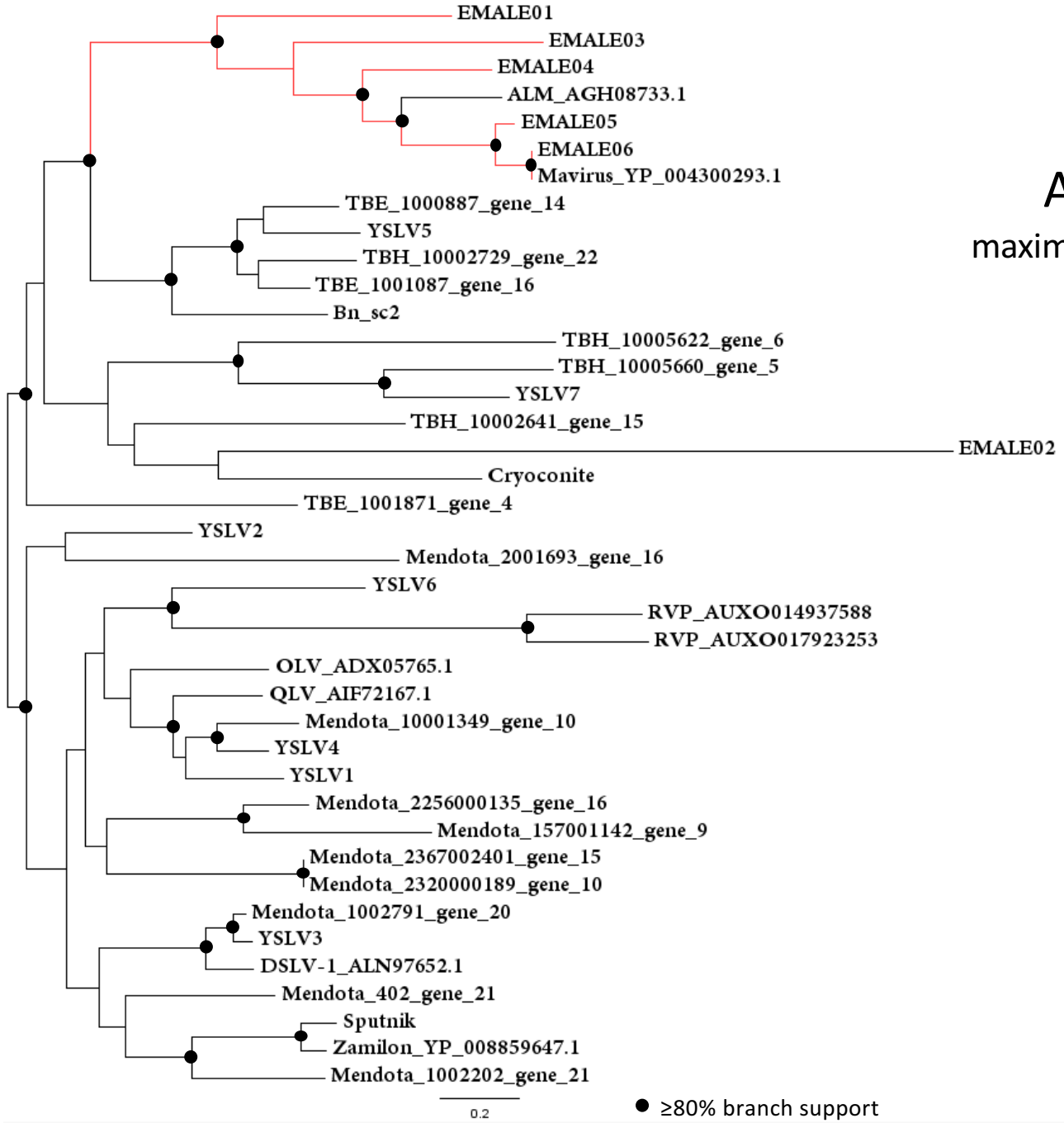
Major capsid protein phylogeny of virophages



MCP

maximum likelihood

FtsK-HerA-type ATPase phylogeny of virophages

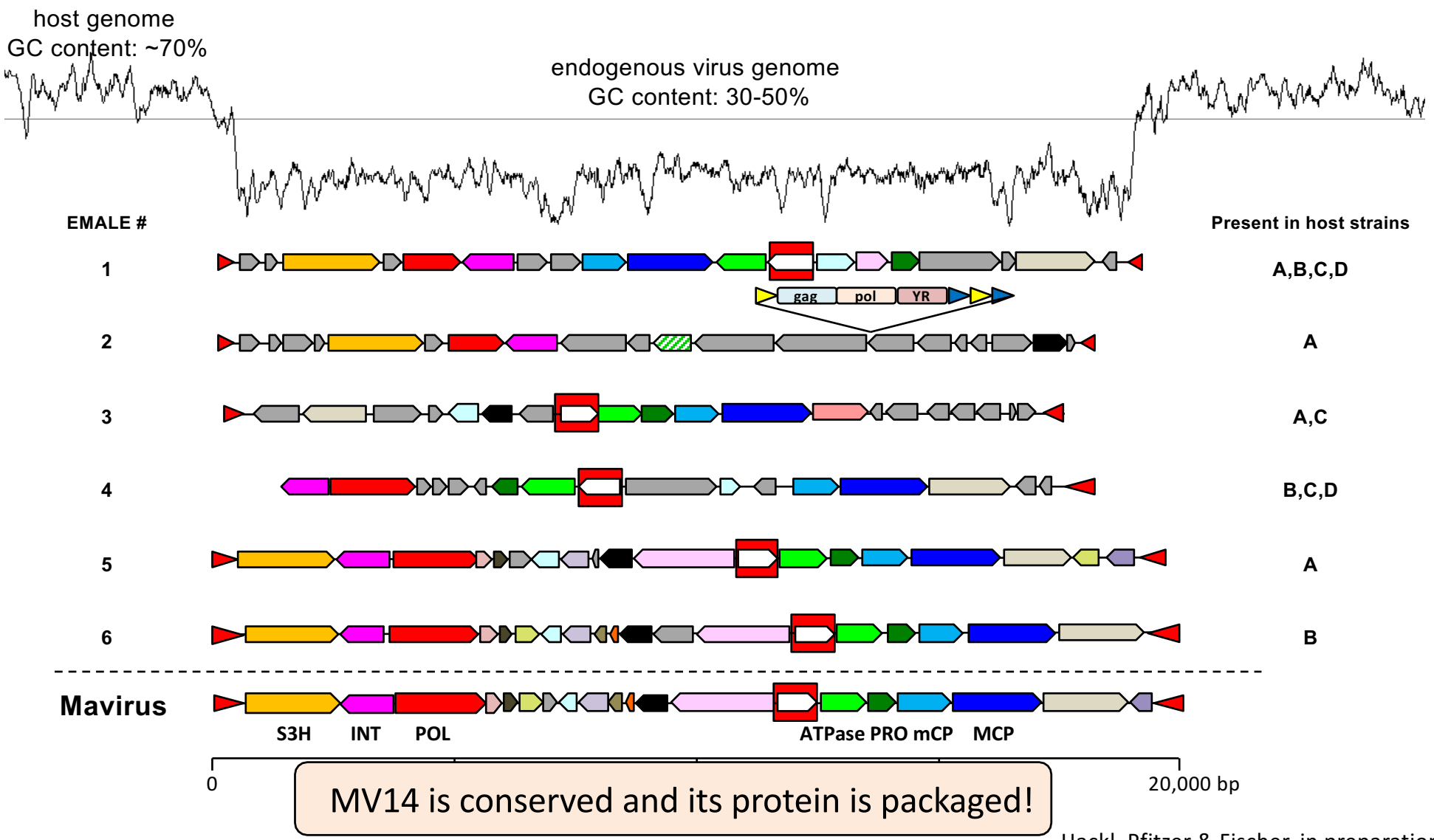


ATPase

maximum likelihood

Endogenous virophages in natural *Cafeteria* strains

Six distinct versions of Endogenous Mavirus-Like Elements (EMALEs)

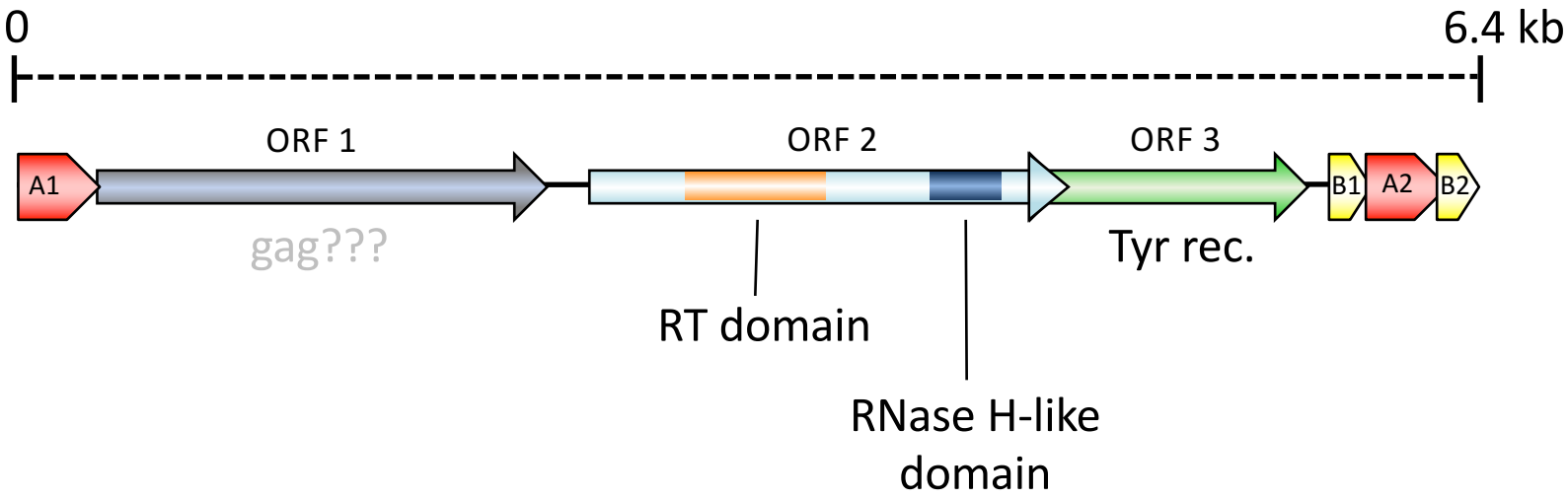


MV14 is conserved and its protein is packaged!

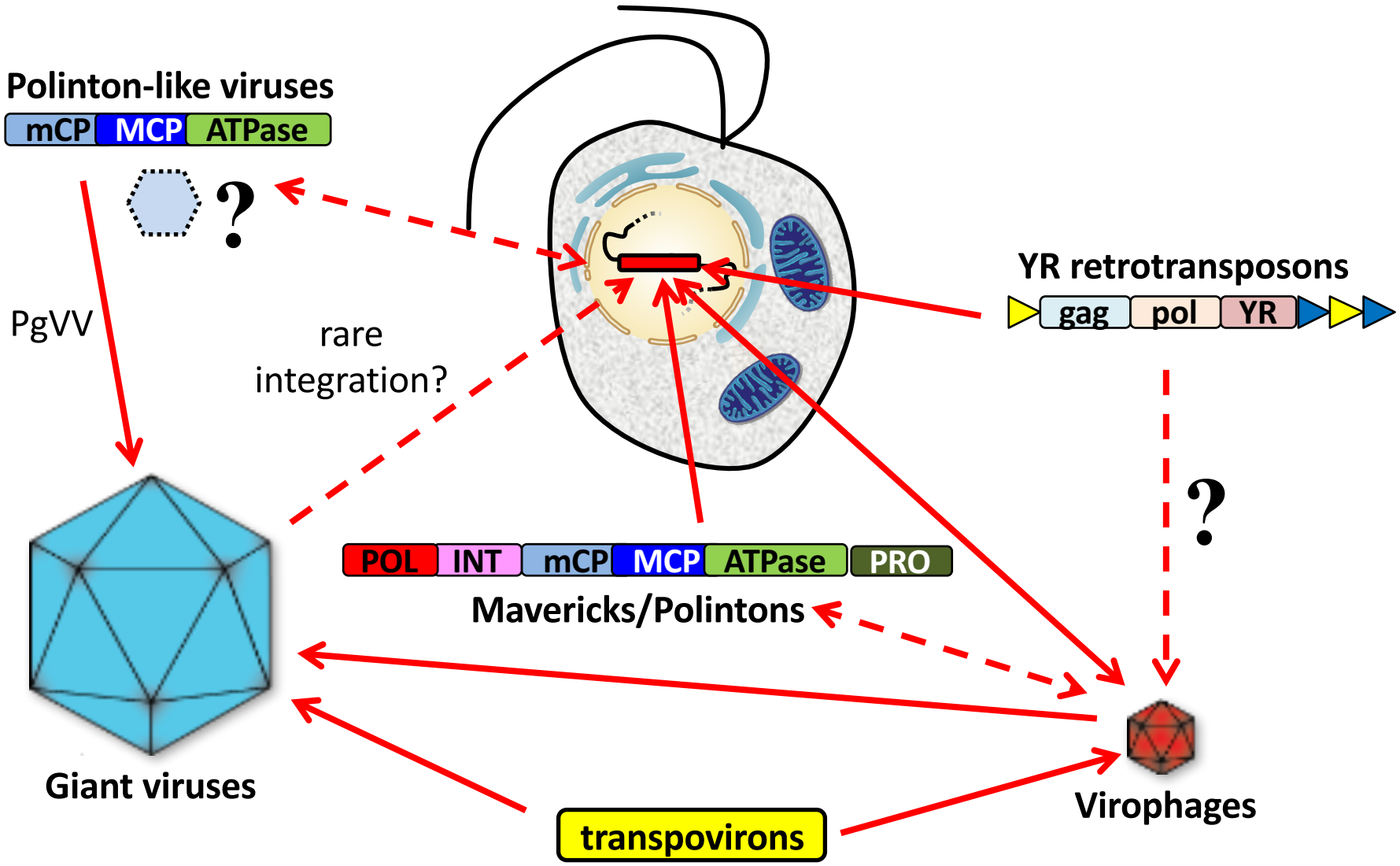
Hackl, Pfitzer & Fischer, in preparation

Some EMALEs carry Tyr recombinase retrotransposons

Ngaro retrotransposons (DIRS-like)
Non-LTR retrotransposons with split direct repeats

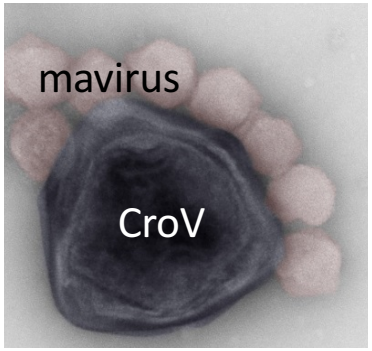


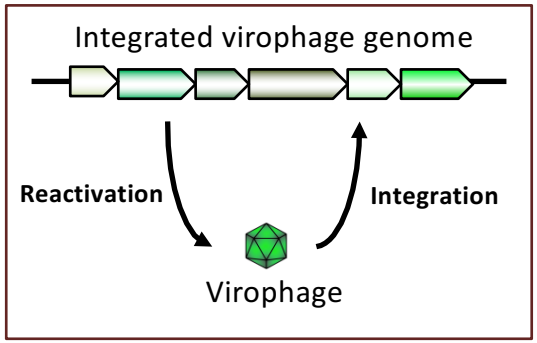
Heterotrophic protists and their mobile genetic elements

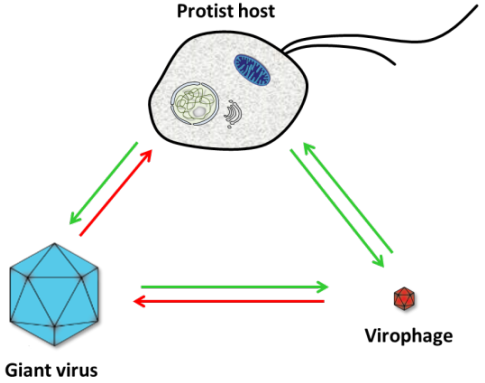


PLVs: Yutin et al (2015) **BMC Biol** 13:95
PgVV: Santini et al (2013) **PNAS** 110:10800
YR-RT: e.g. Piednoël et al (2011) **BMC Genom** 12:621
Transpovirons: Desnues et al (2012) **PNAS** 109:18078
Endog. giant viruses: e.g. Gallot-Lavallée et al (2017) **Viruses** 9:17

Take home points







1. Virophages are parasites of giant viruses and MGE in heterotrophic flagellates

2. Mavirus leads a dual lifestyle and displays high mobility

3. Virophages may act as an adaptive immune system in *Cafeteria*-like flagellates

Open questions:
- ecological significance?
- diversity/specificity/stability/interactions?
- virophages as HGT agents?
- …

Acknowledgements

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U. Amsterdam:
Corina Brussaard

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EMBO

Chica and Heinz Schaller Foundation