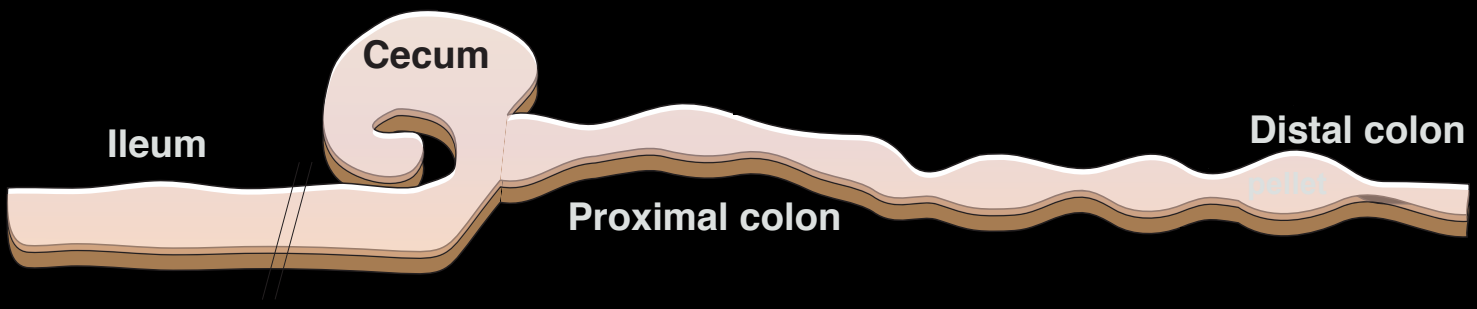
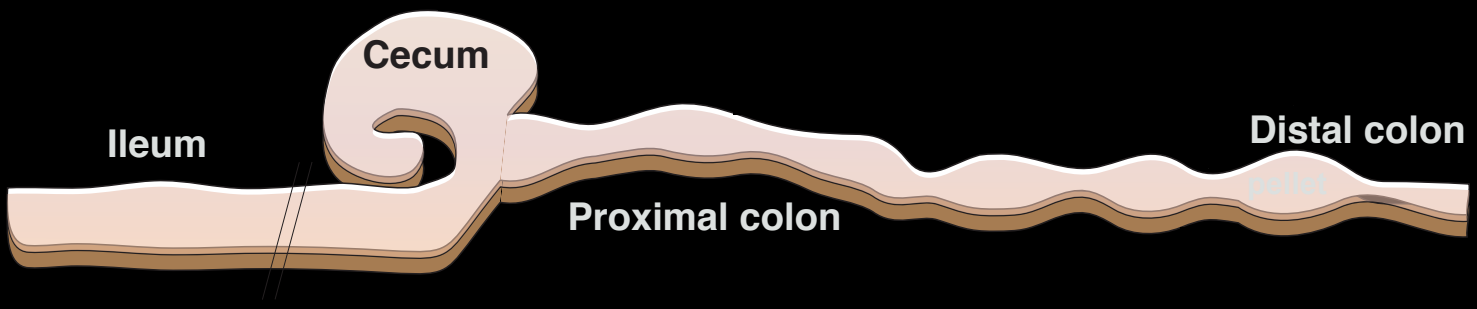
The physical environment constrains species survival

The gut as a host-controlled tube



The gut is composed of divergent environments



bacterial load

antimicrobials

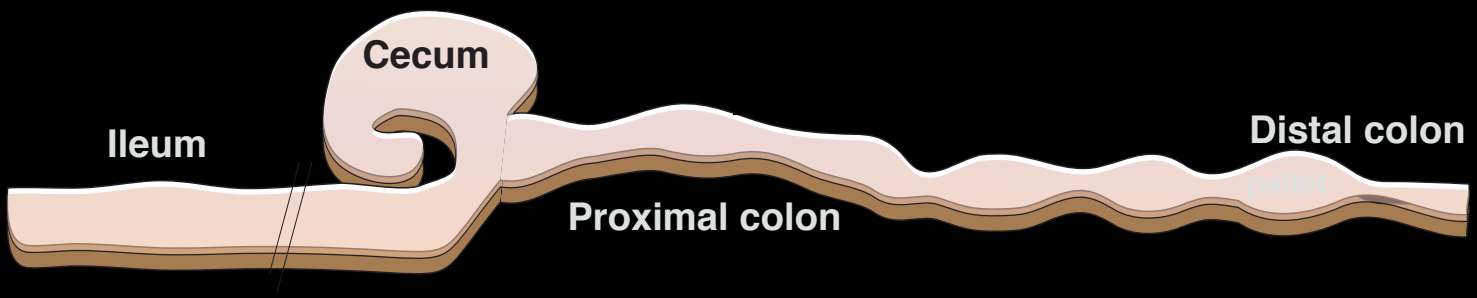
O_2

pH

'easy' nutrients

Fiber

The gut is composed of divergent environments



Bacteroidaceae, Prevotellaceae,

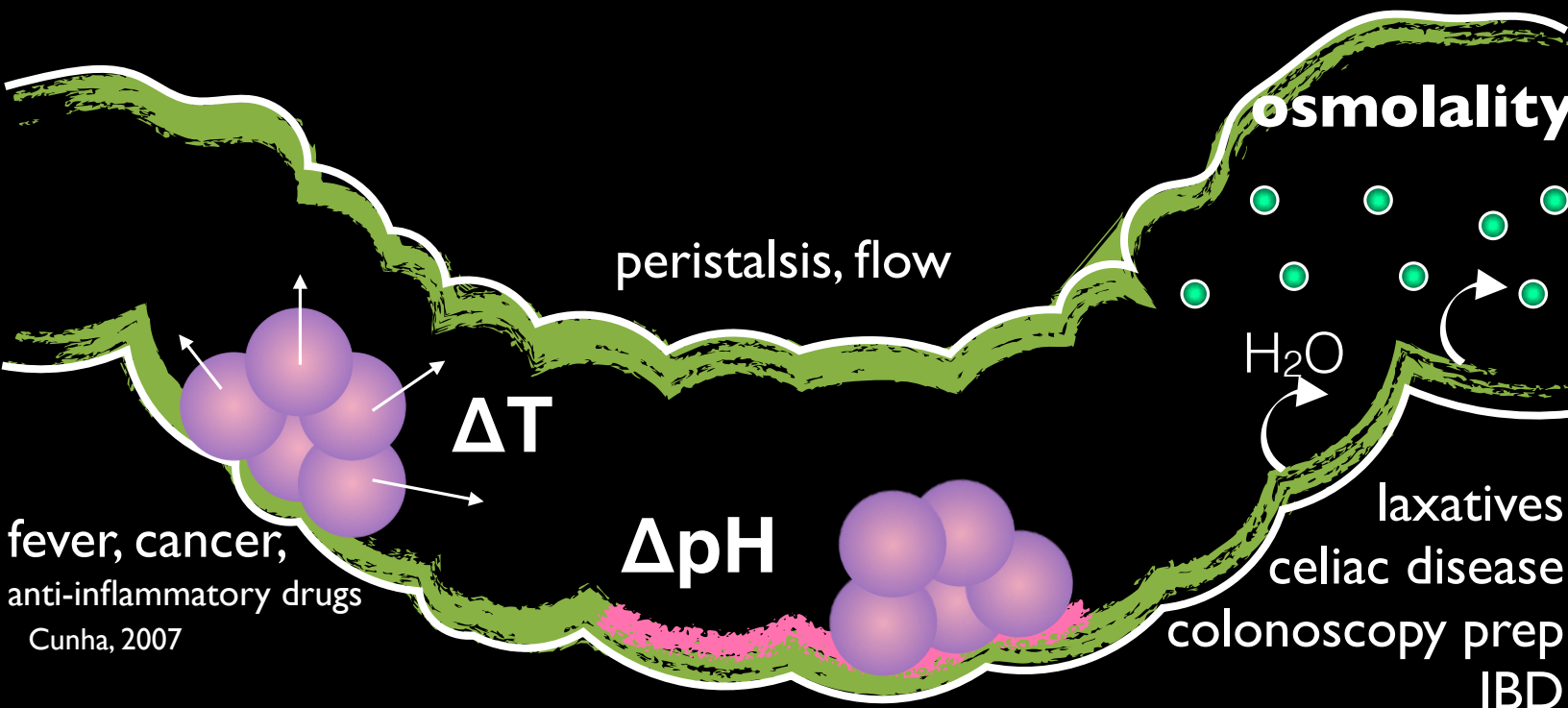
Lactobacillaceae,
Enterobacteriaceae

Rikenellaceae, Lachnospiraceae,
Ruminococcaceae

Tropini, et al. Cell Host and Microbe 2017

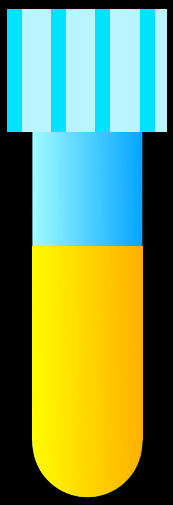
Donaldson et al. Nat Rev Microbiol 2016

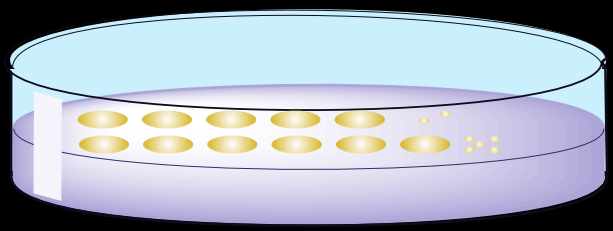
The physical environment in the gut impacts our microbiota



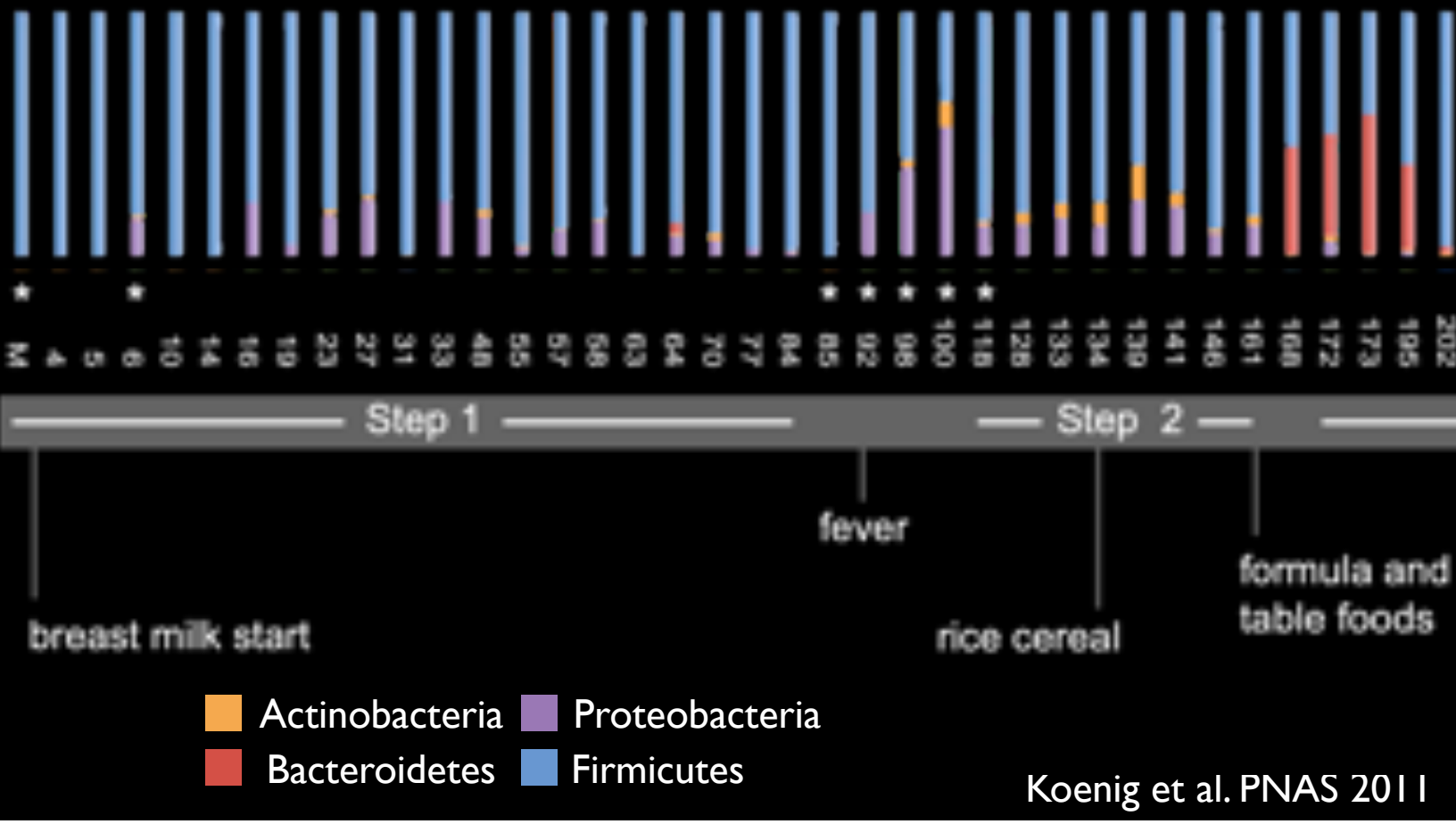
We have purposely fixed the physical environment we grow cells in

- pH
- Osmolality
- Temperature
- Material stiffness
- Oxygen tension
- […]



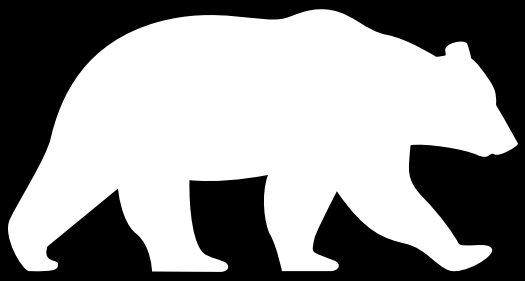


The human microbiome is disrupted by fever



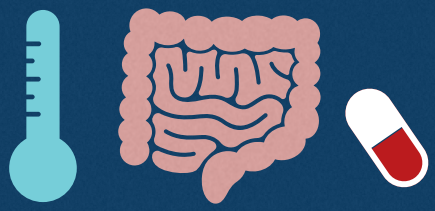
Koenig et al. PNAS 2011

Abiotic perturbations affect organisms at all scales

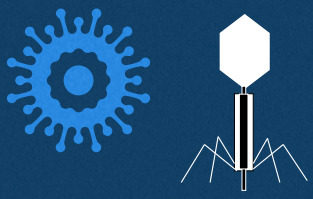


Global warming
1.5-2°C difference

10^6 species face extinction



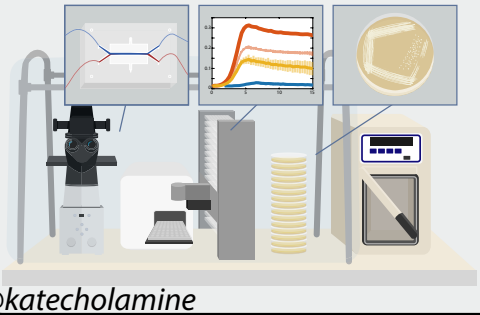
Abiotic Perturbations
due to Disease and Drugs



Phages driving
microbiota dynamics

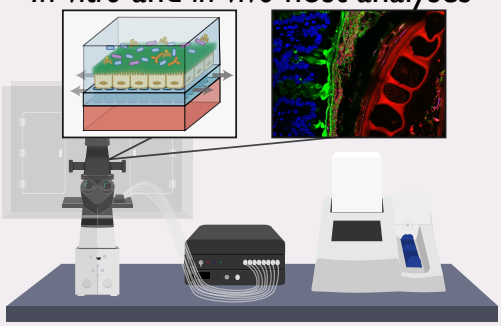
Microbiota
Engineering

In vitro bacterial analysis

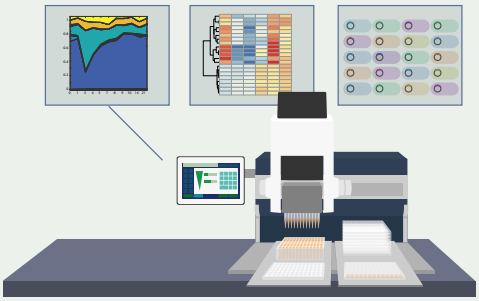


@katecholamine

In vitro and *in vivo* host analyses



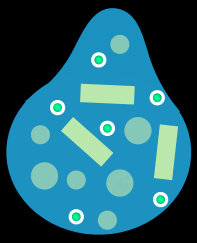
Genomic analysis & engineering



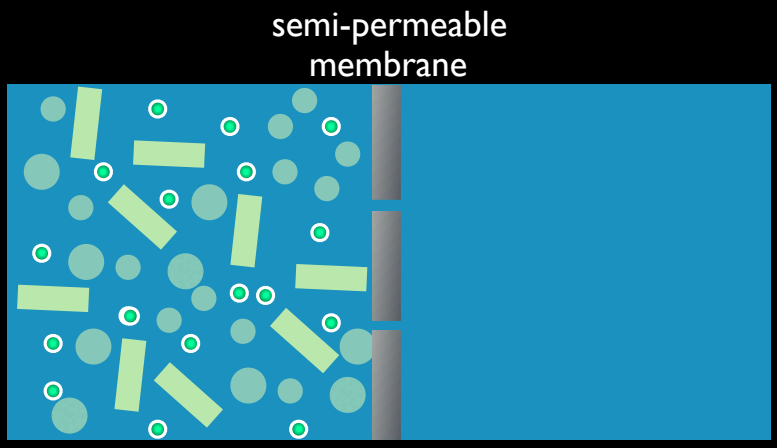
Open questions for the group

- What basic numbers are we missing?
- How to connect different measurements?
- How simple should our models be?
- How many model organisms for the gut microbiota?
- How important is spatial understanding? How much are we losing by sequencing stool?

Osmolality

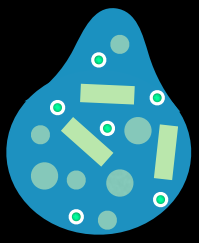


concentration of a solution:
number of particles/ Kg

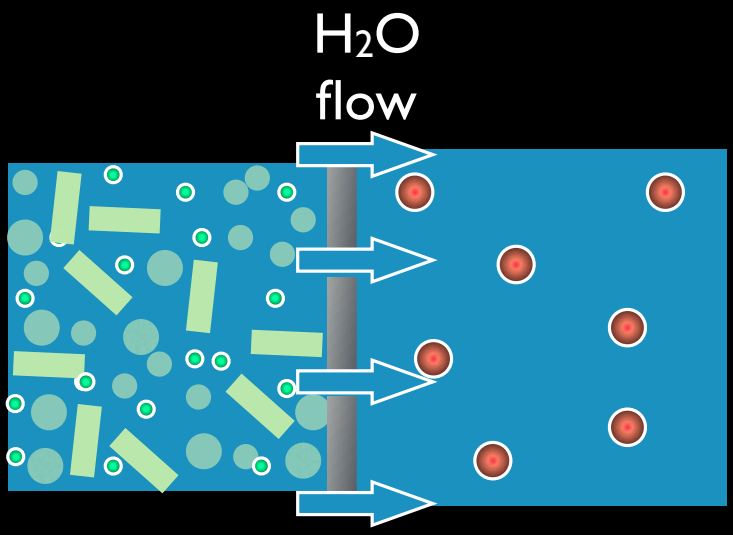


Cell interior
High *P*

Osmolality



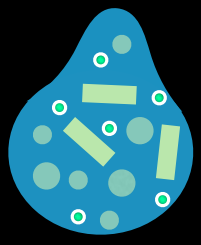
concentration of a solution:
number of particles/ Kg



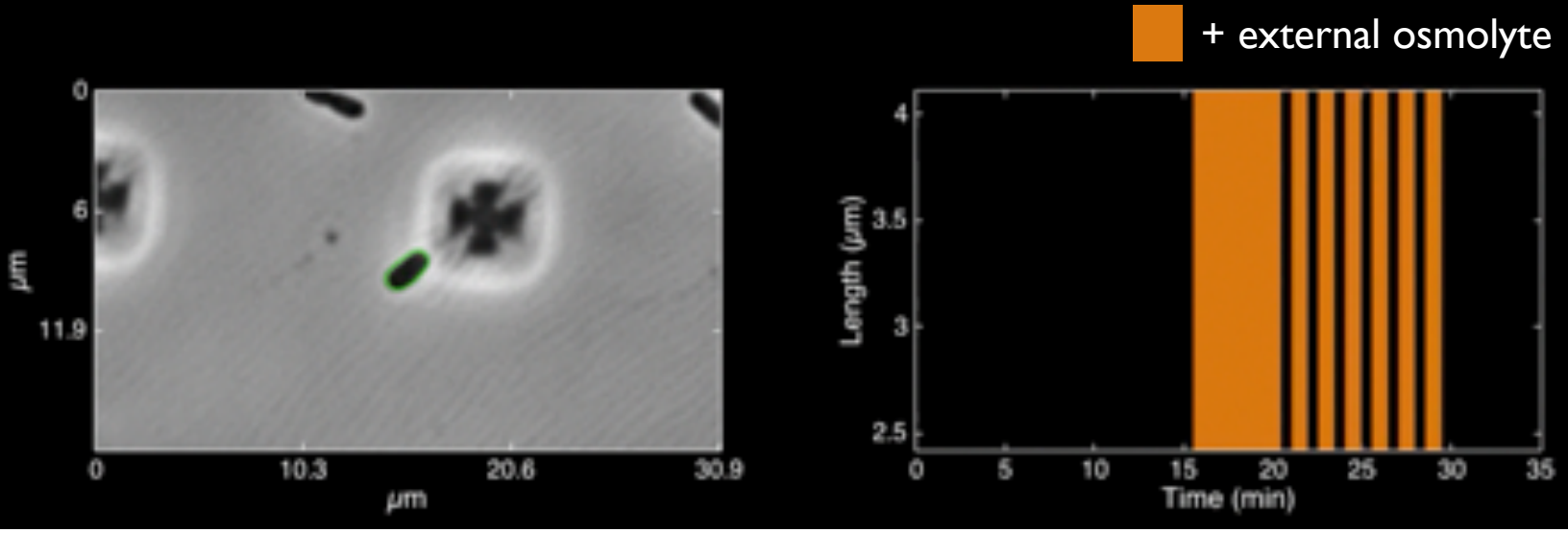
Cell interior
Lower *P*

External osmolyte

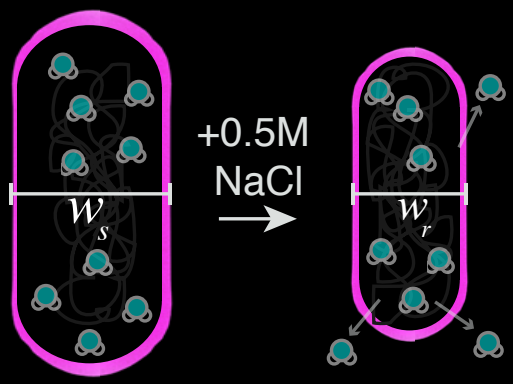
Osmolality

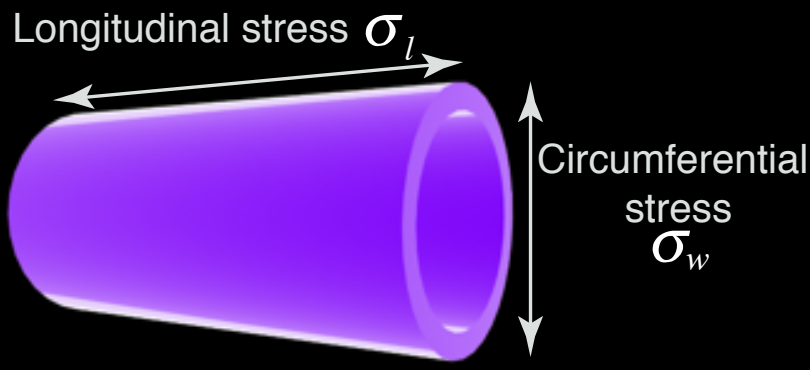


concentration of a solution:
number of particles/ Kg

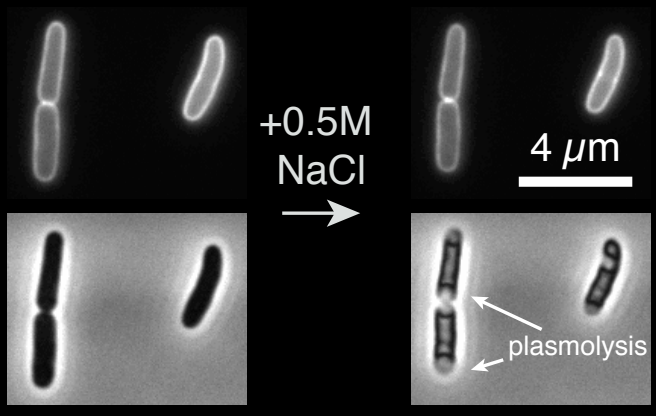


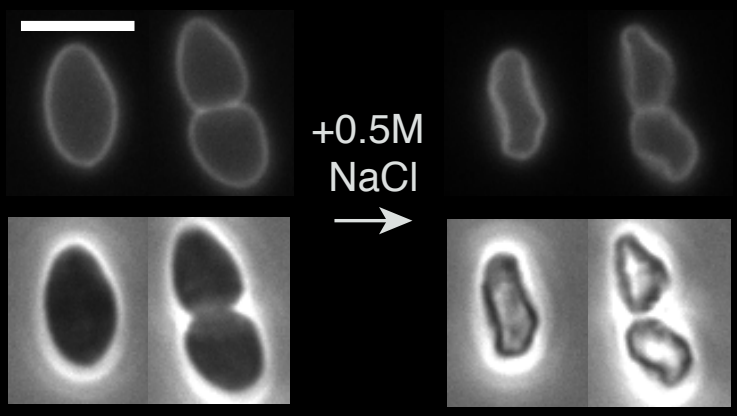
Shocking bacterial response





$$\varepsilon_w = \frac{w_s - w_r}{w_r} = \frac{\sigma_w}{Y}$$





Tropini *et al.* Cell Reports 2014

Osmotic perturbations are prevalent

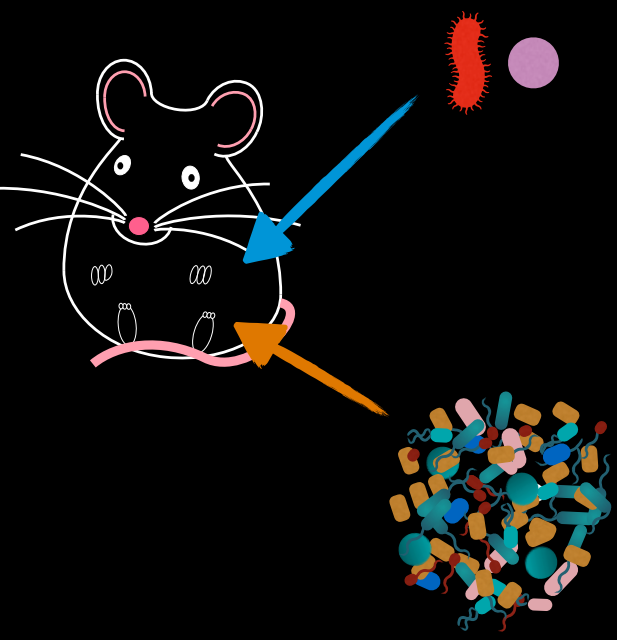
MiraLAX #2 top digestive remedy in US*

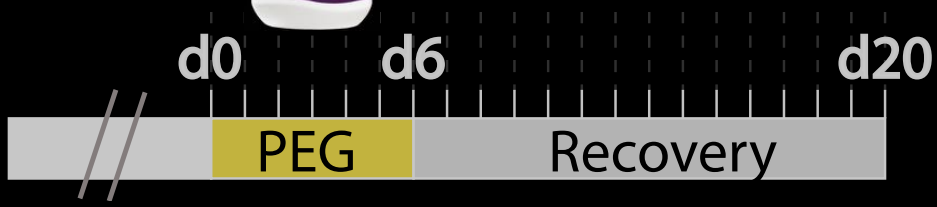
$186.1 m sales*
~5.3 million kg/year

*statista - drug store news

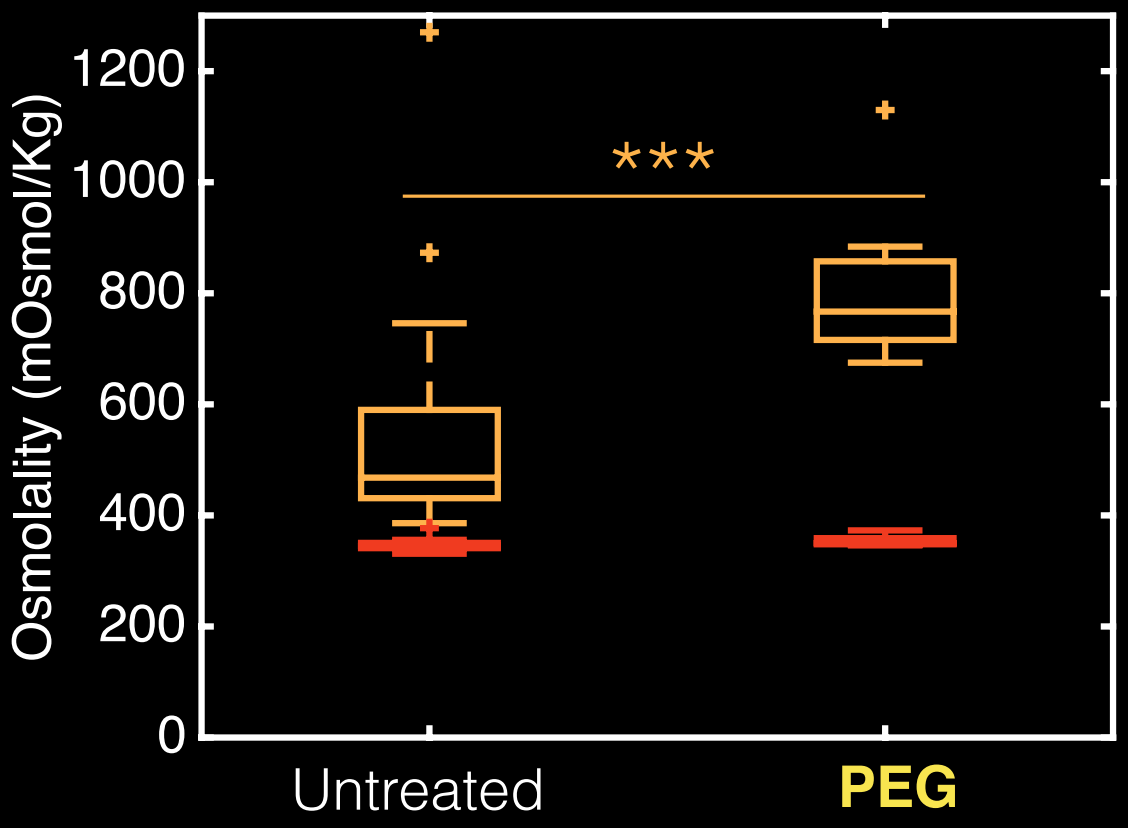
A model for osmotic perturbation

MiraLAX= PEG
polyethylene glycol



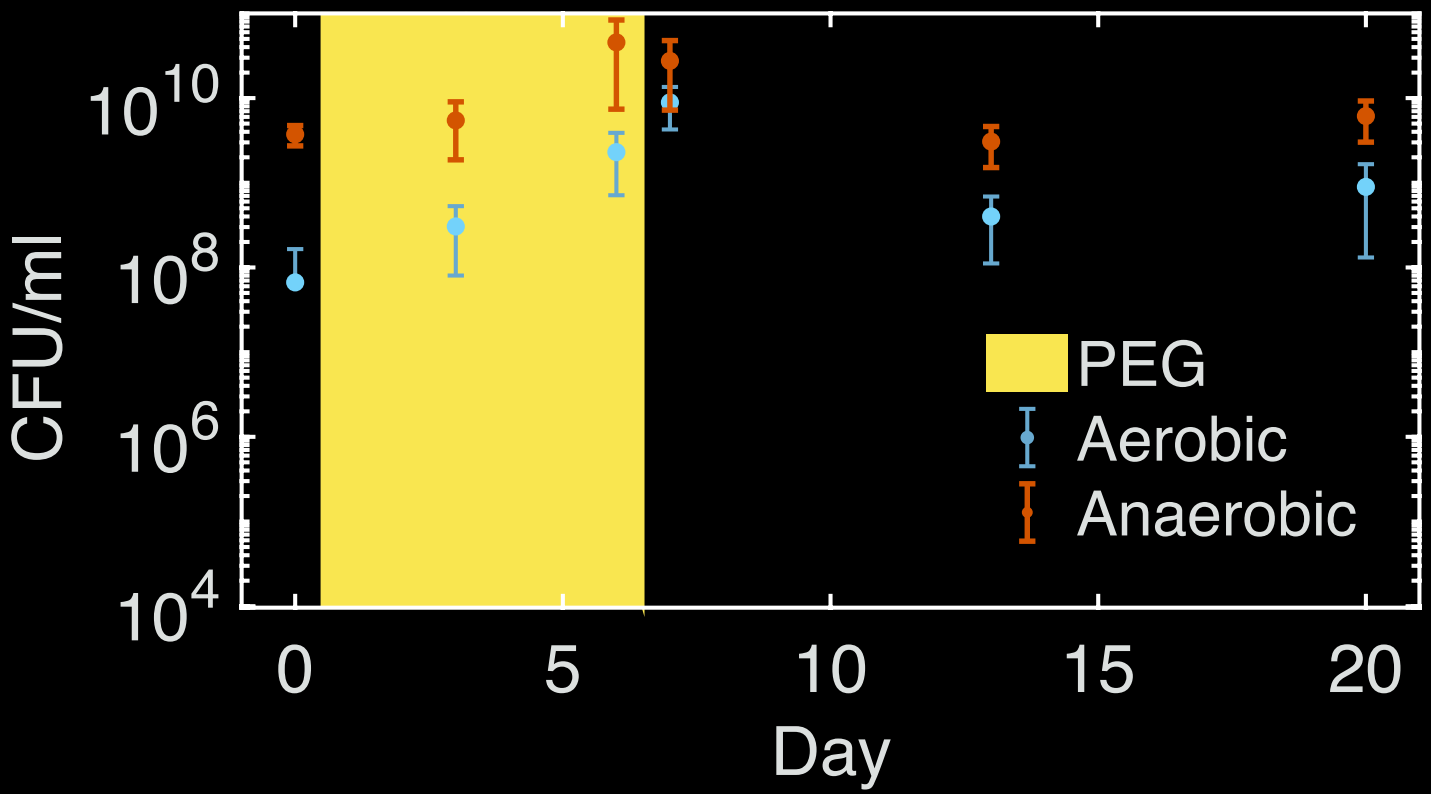


Cecum osmolality increases during PEG treatment



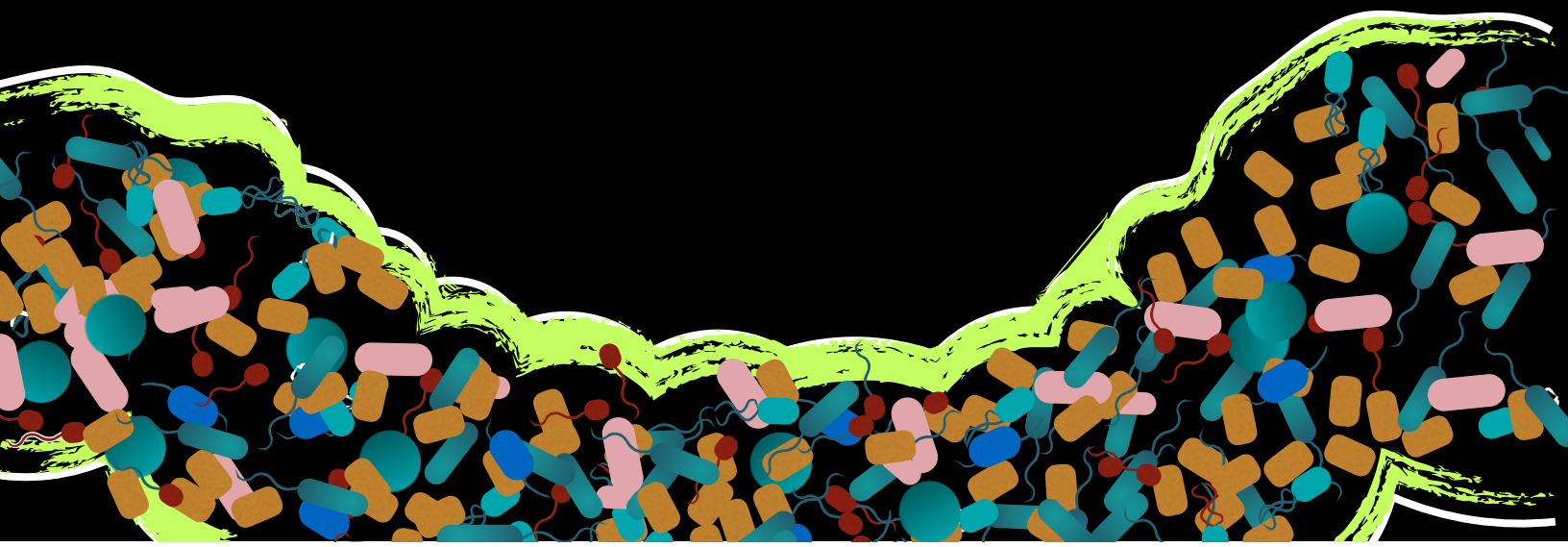
Tropini *et al.* June 2018 *Cell*

Culturable bacterial load is not decreased by diarrhea



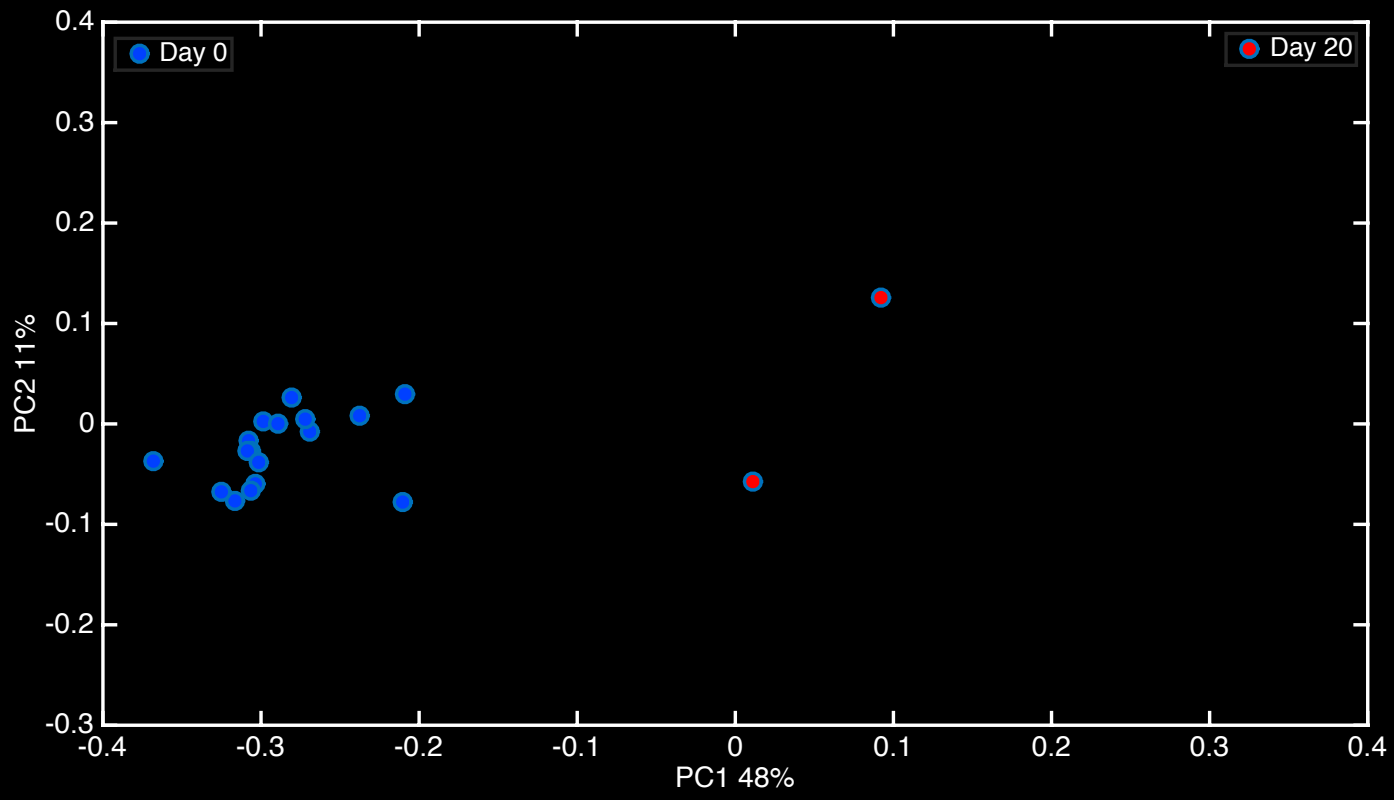
Overall entire content over GI tract of culturables is constant

Does osmotic perturbation affect the gut microbiota?



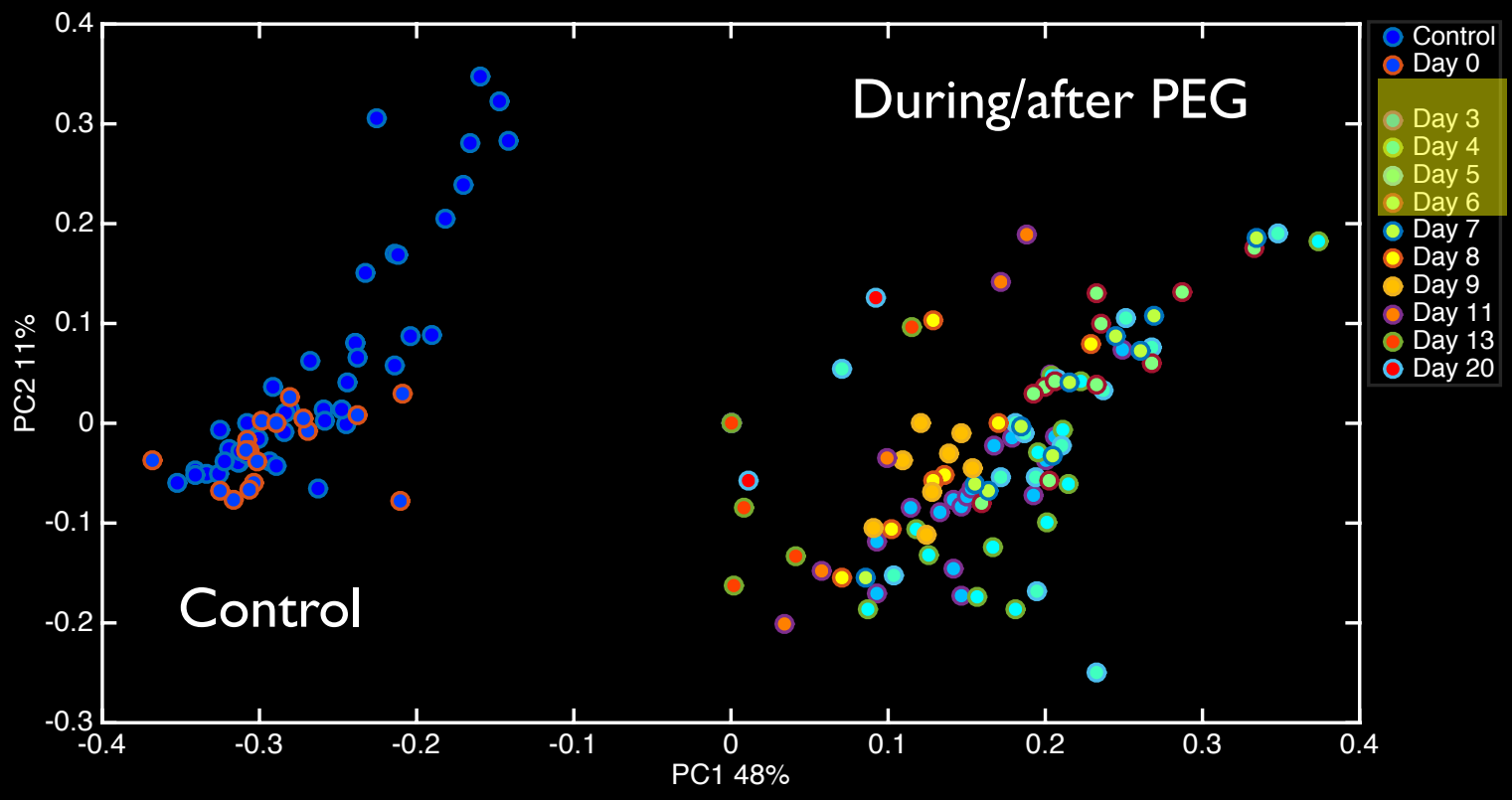
PCoA of bacterial beta-diversity

PEG



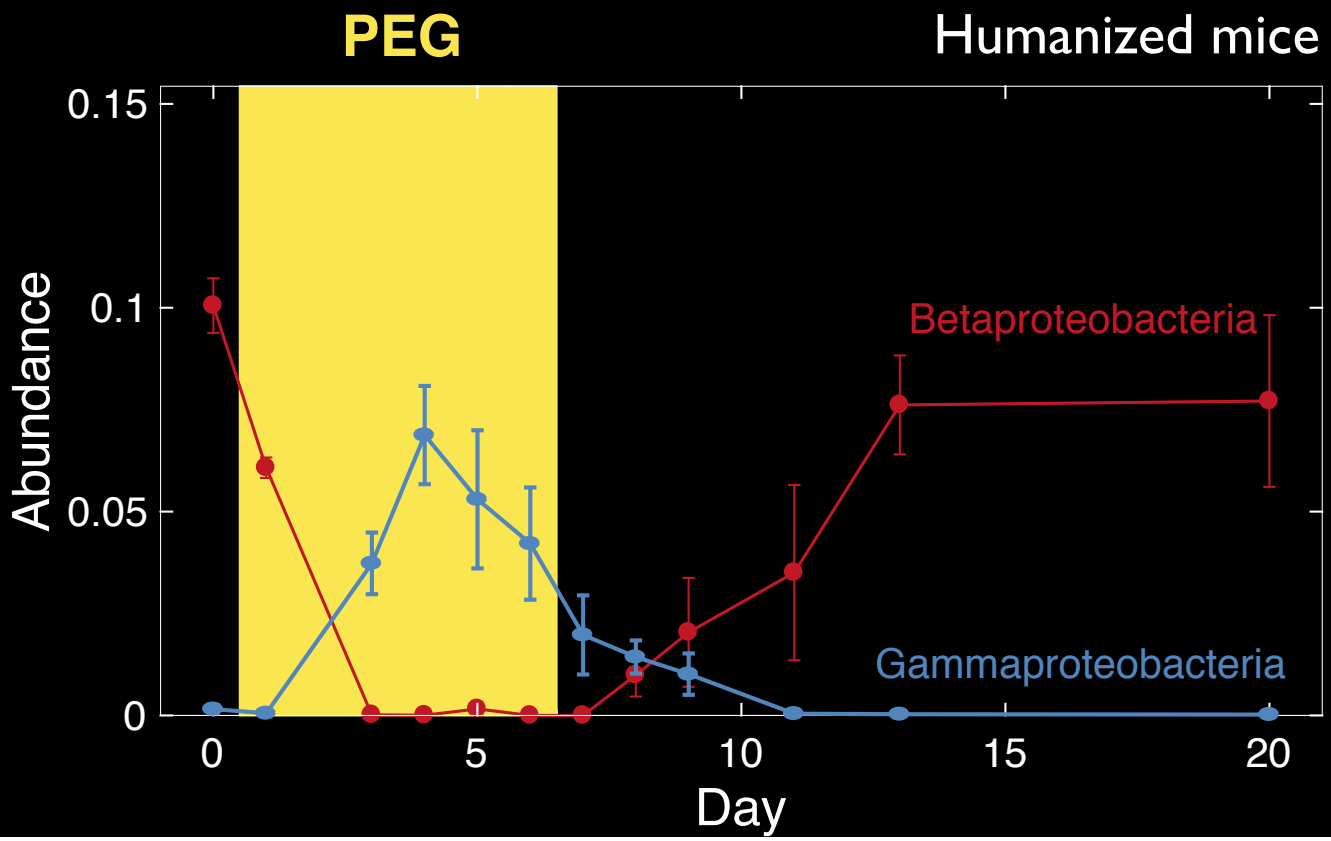
Tropini *et al.* June 2018 *Cell*

Osmotic perturbation reshapes microbiota composition

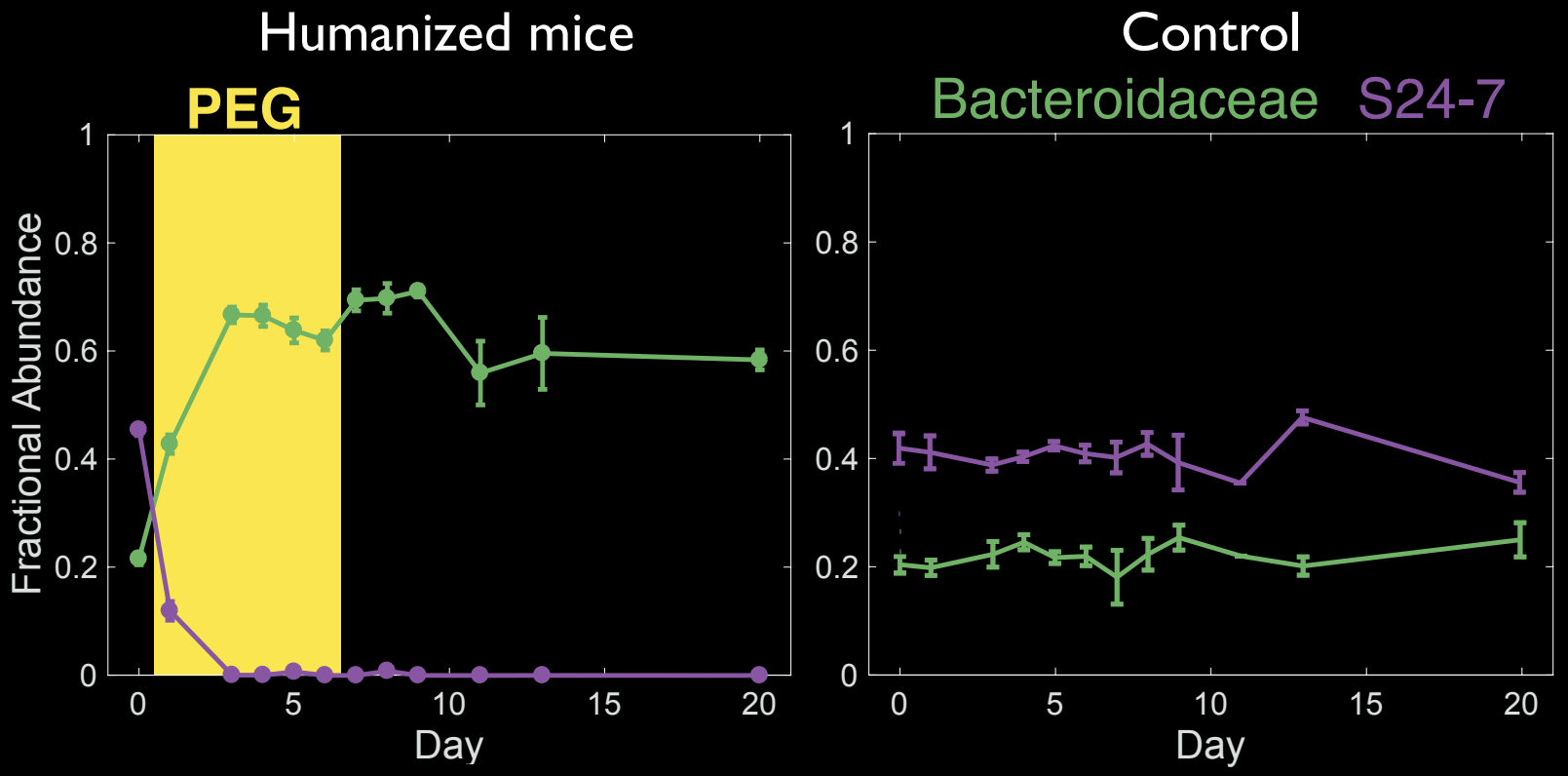


This tells us the microbiota has changed. Why? How?

Osmotic perturbation causes transient changes

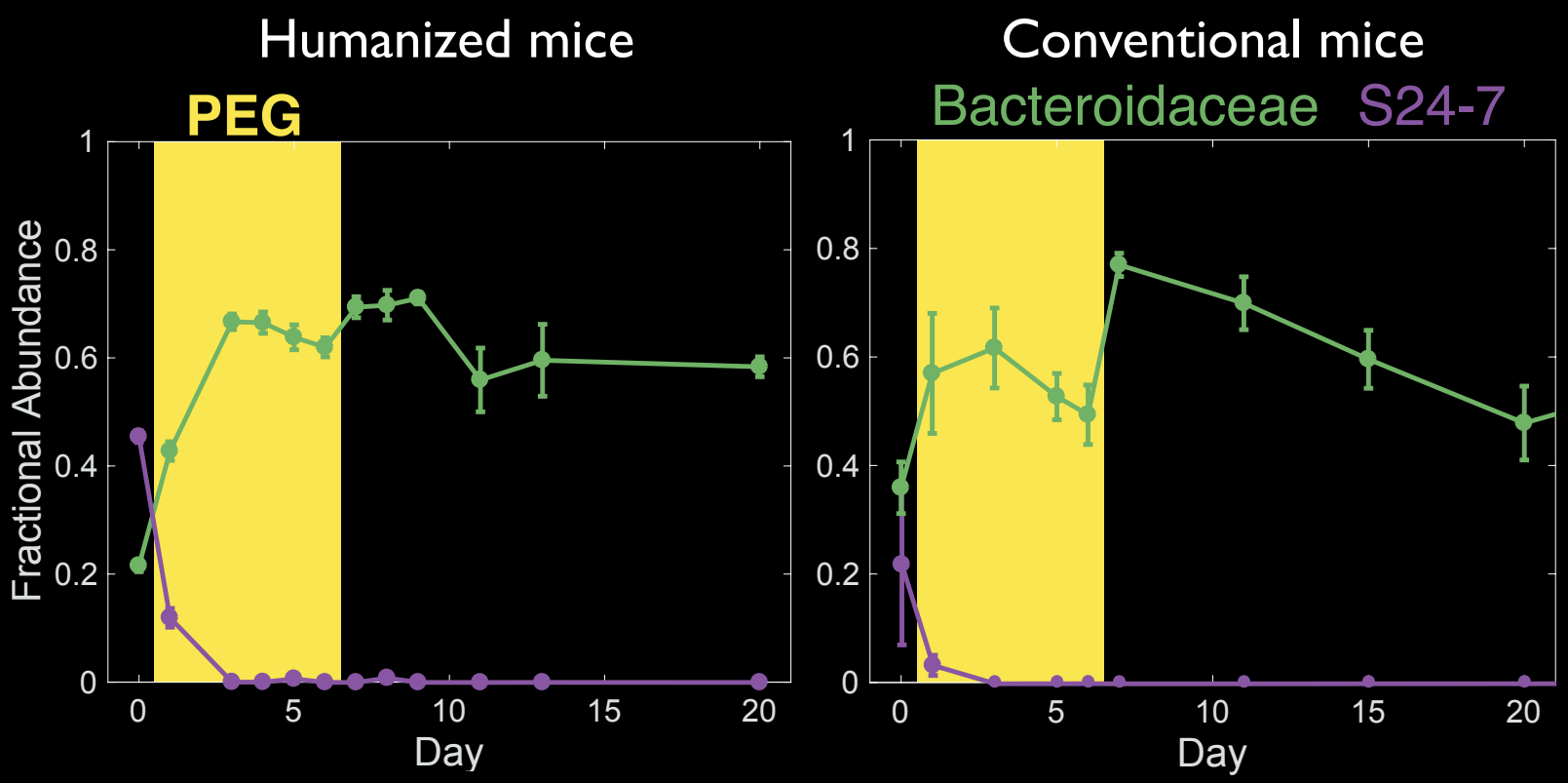


~50% of microbiota disappears



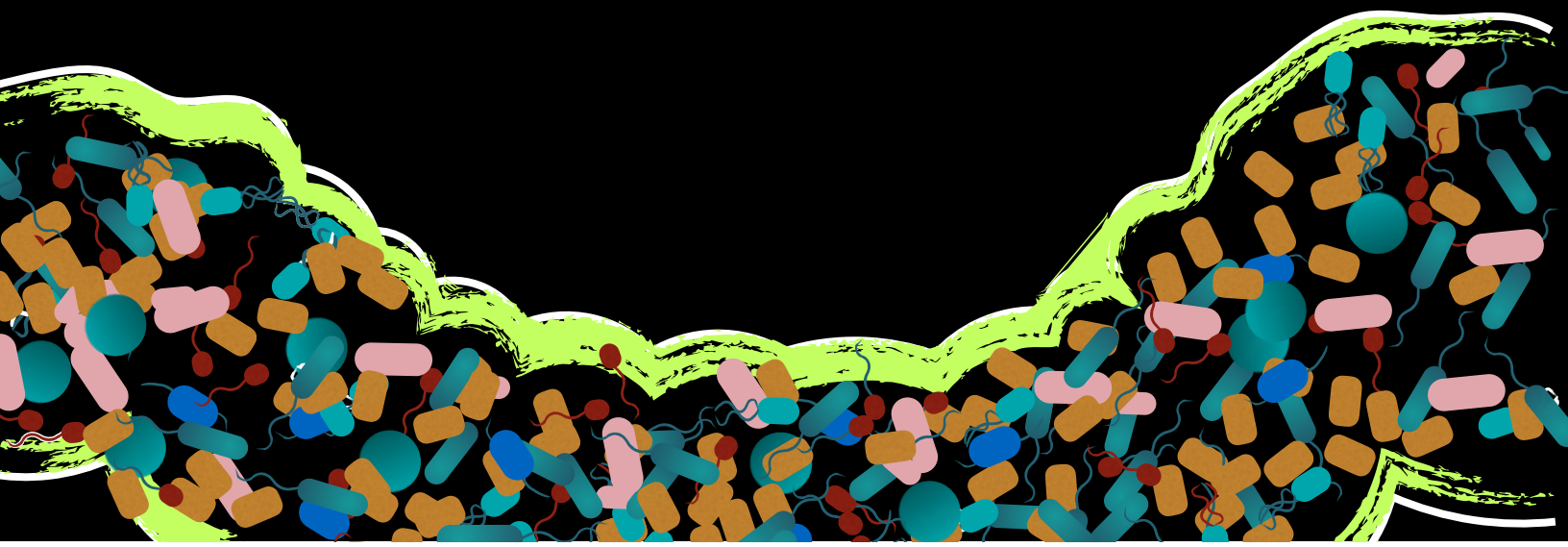
Tropini *et al.* June 2018 *Cell*

Dynamics are independent of microbiota composition

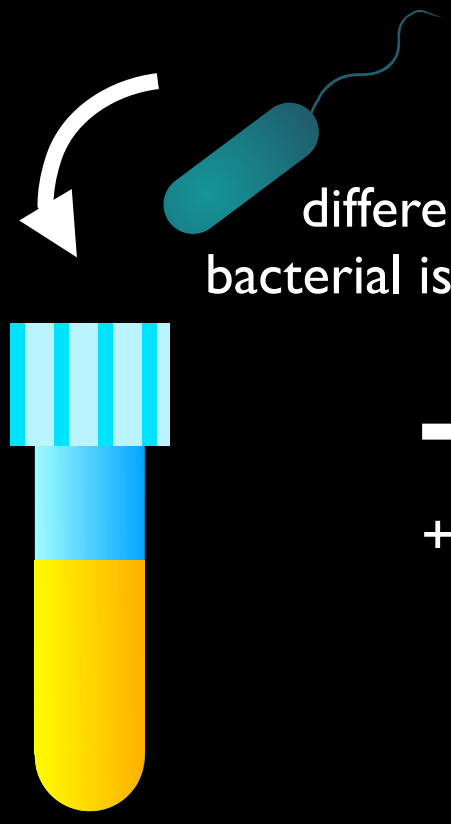


Tropini *et al.* June 2018 *Cell*

Why is S24-7 driven to disappearance during PEG treatment?

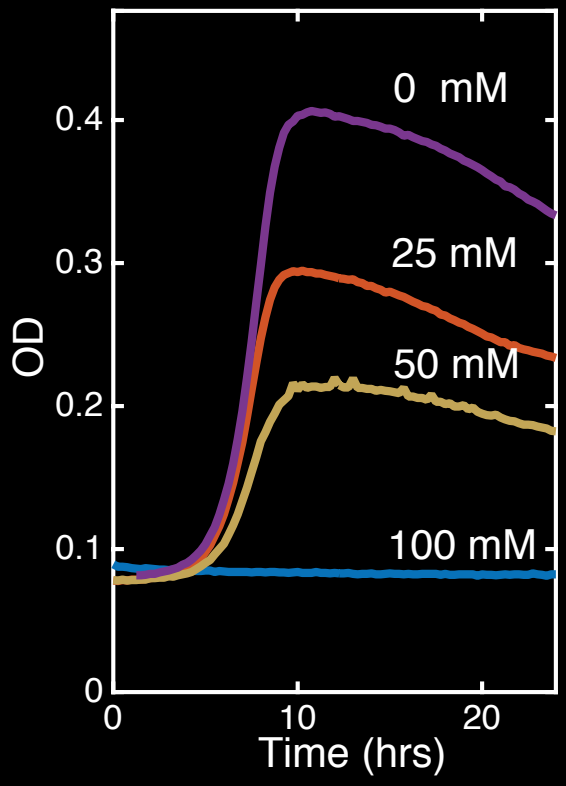


How does osmolality affect bacteria in absence of the host?

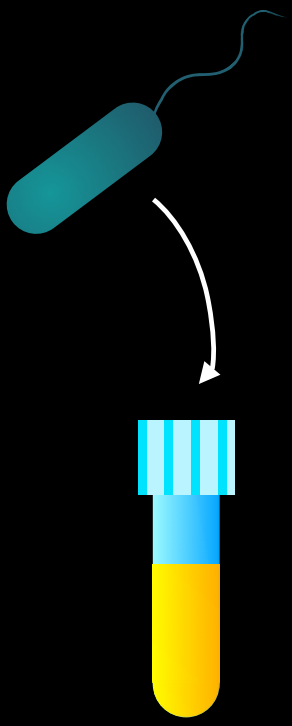


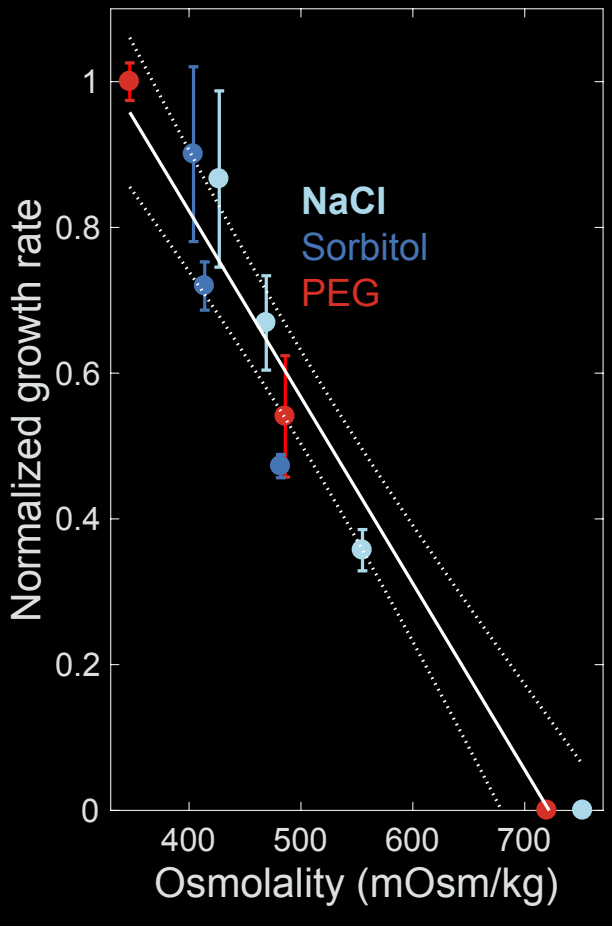
different
bacterial isolates

+/- osmolyte



S24-7 is impaired by increased osmolality *in vitro*

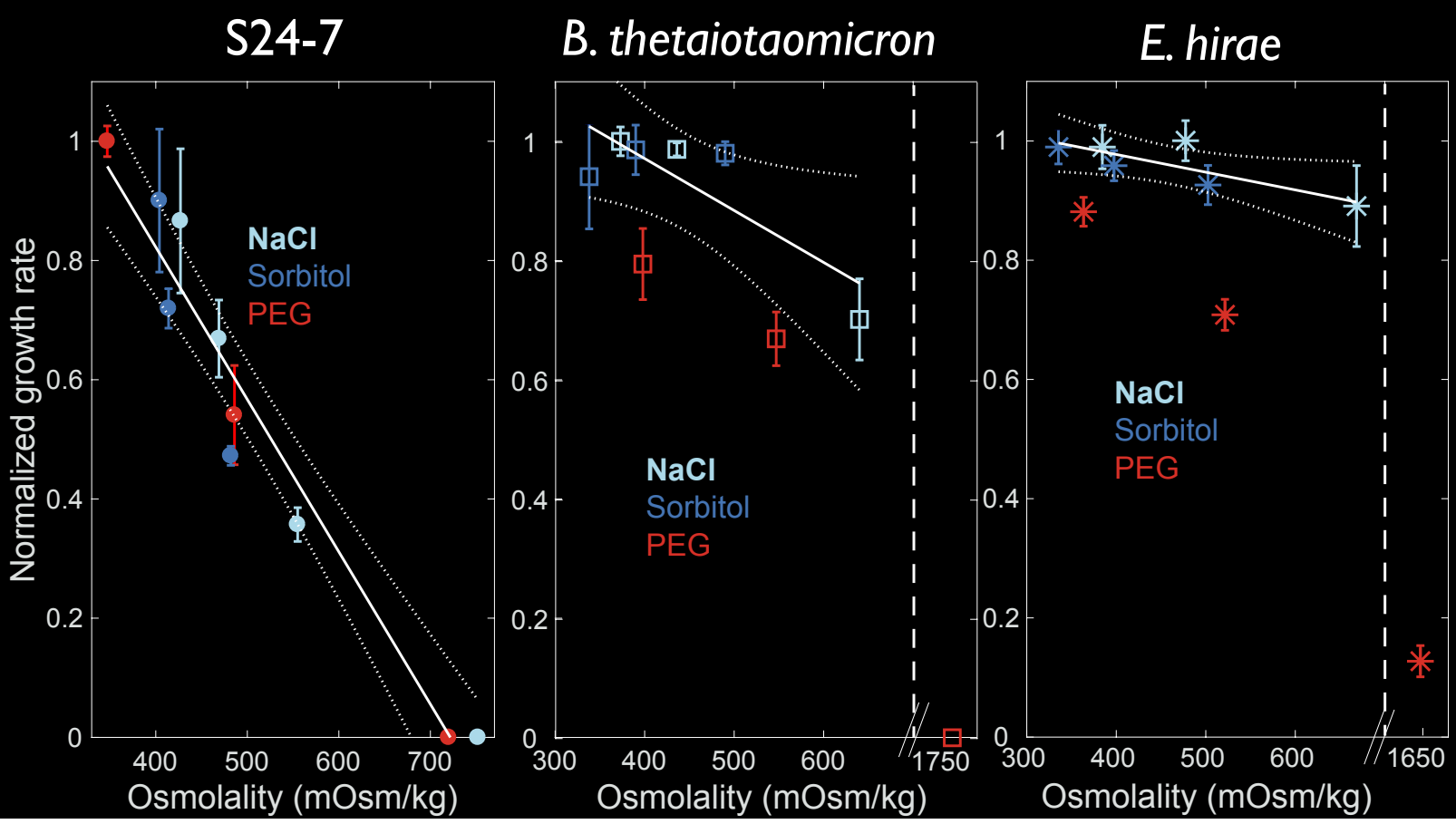




in vivo
PEG: 810

Tropini *et al.* June 2018 *Cell*

Other taxa are not as affected by osmolality *in vitro*

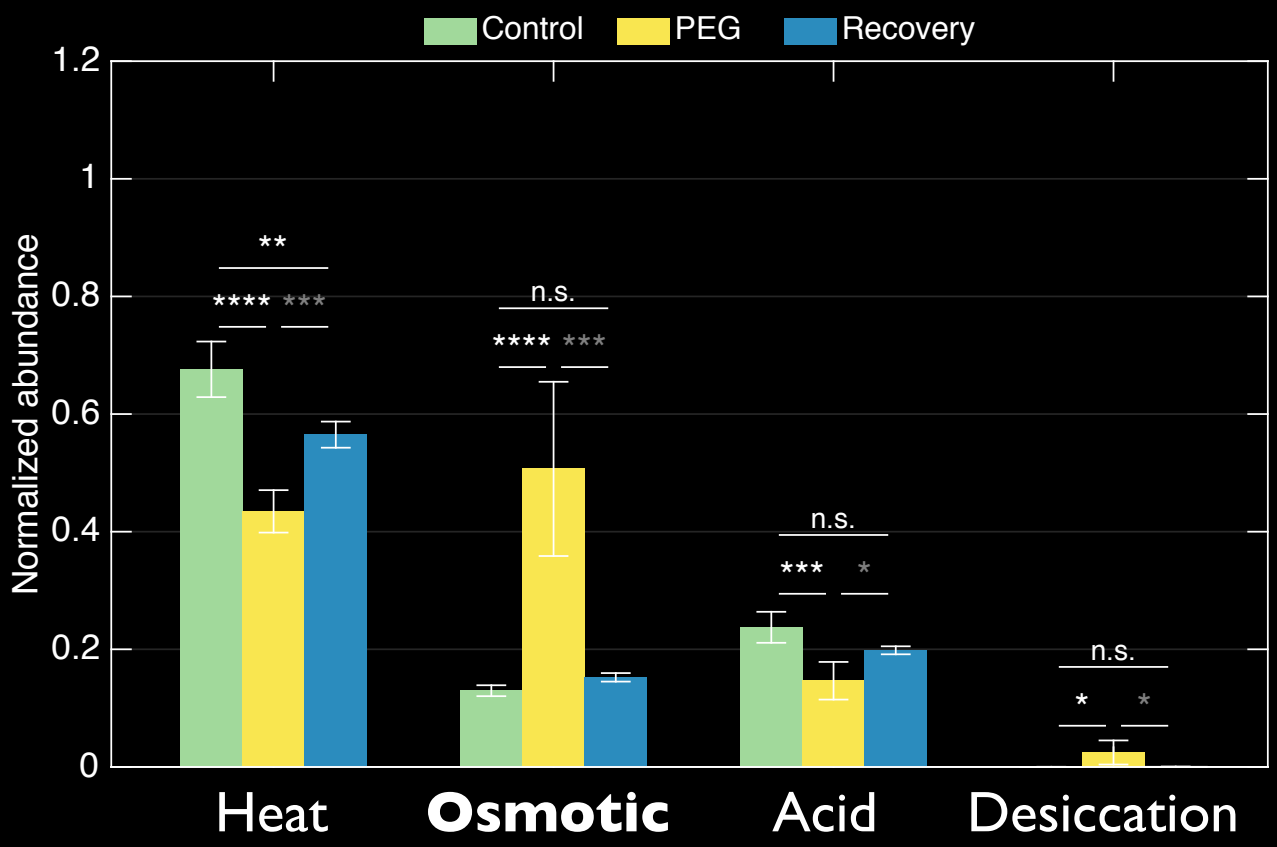


The mechanism for S24-7 reduced tolerance to osmolality

Deanna Pepin, M.Sc.

Sarah Popple

Osmotic perturbation increases abundance of osmotic stress genes



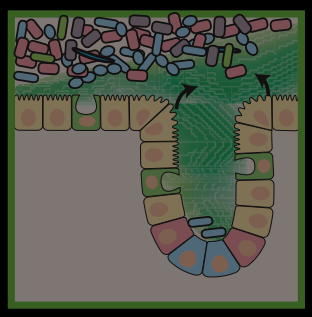
Tropini *et al.* June 2018 *Cell*

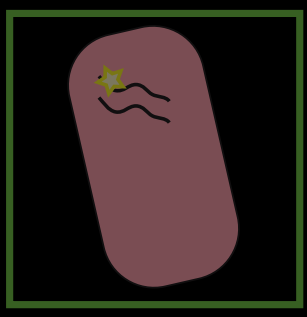
The gut microbiota is **not** resilient to osmotic perturbation

Increased osmolality impairs bacterial growth *in vitro*

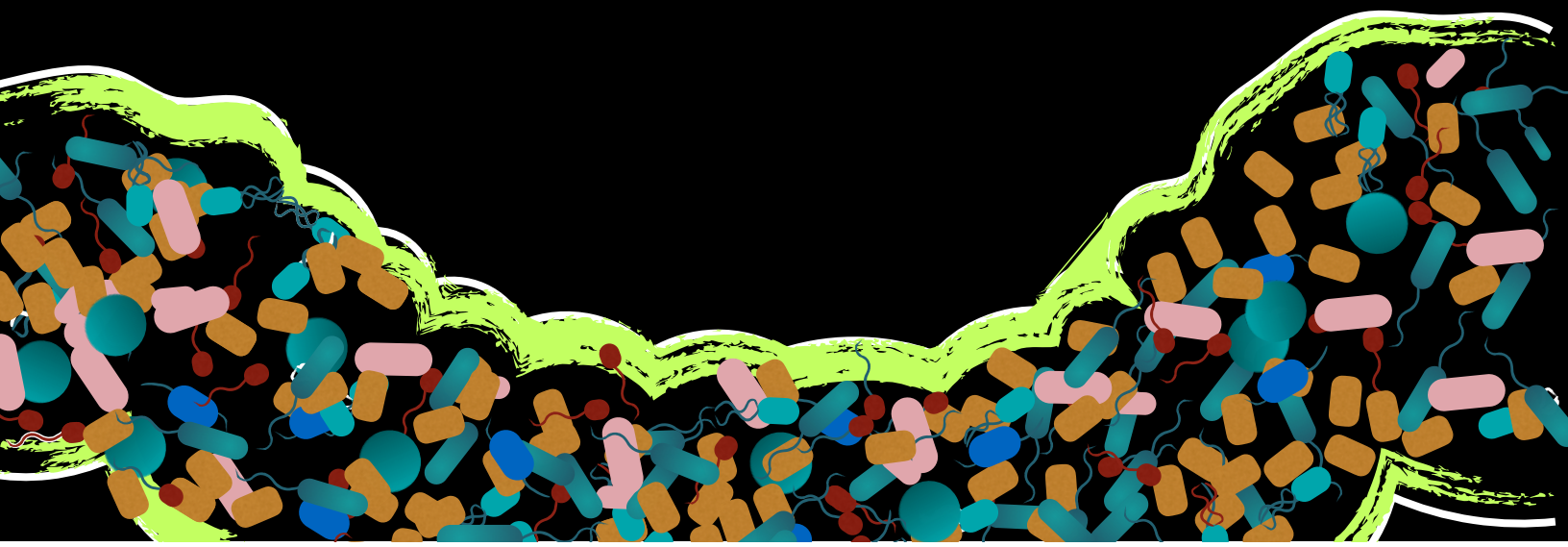
Osmotic stress genes over-represented during osmotic perturbation



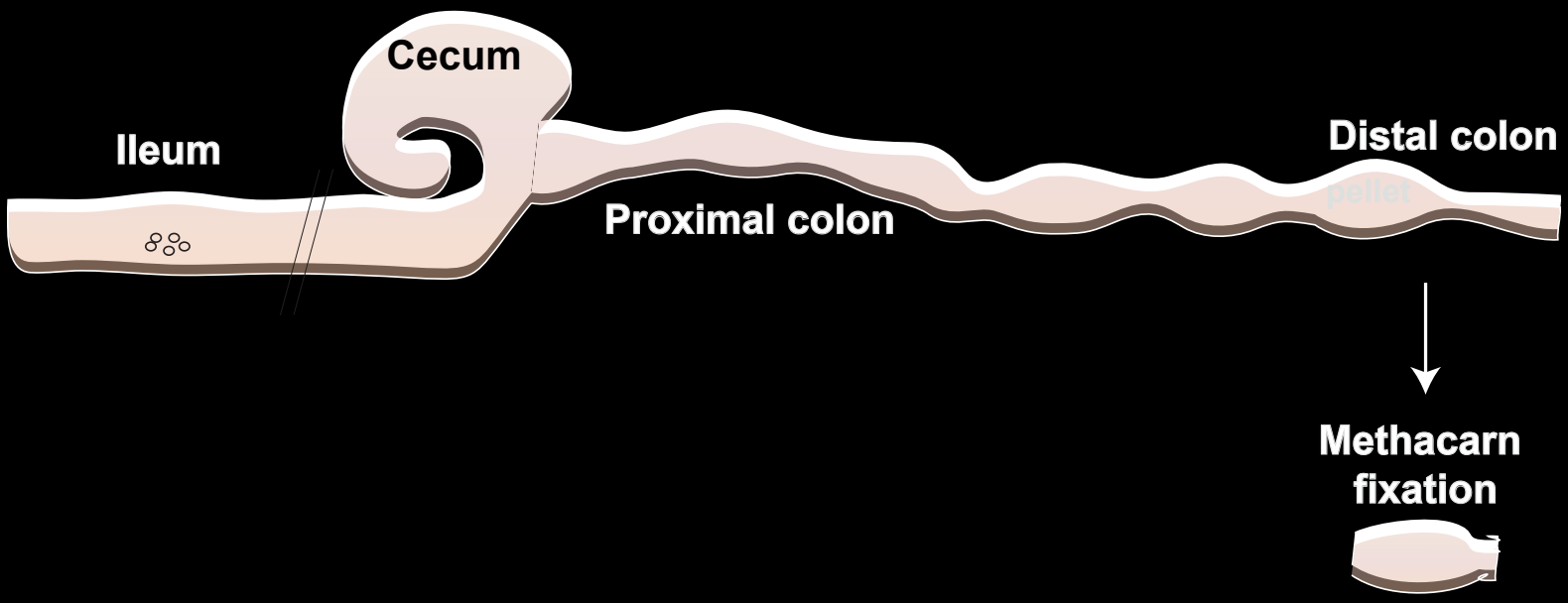




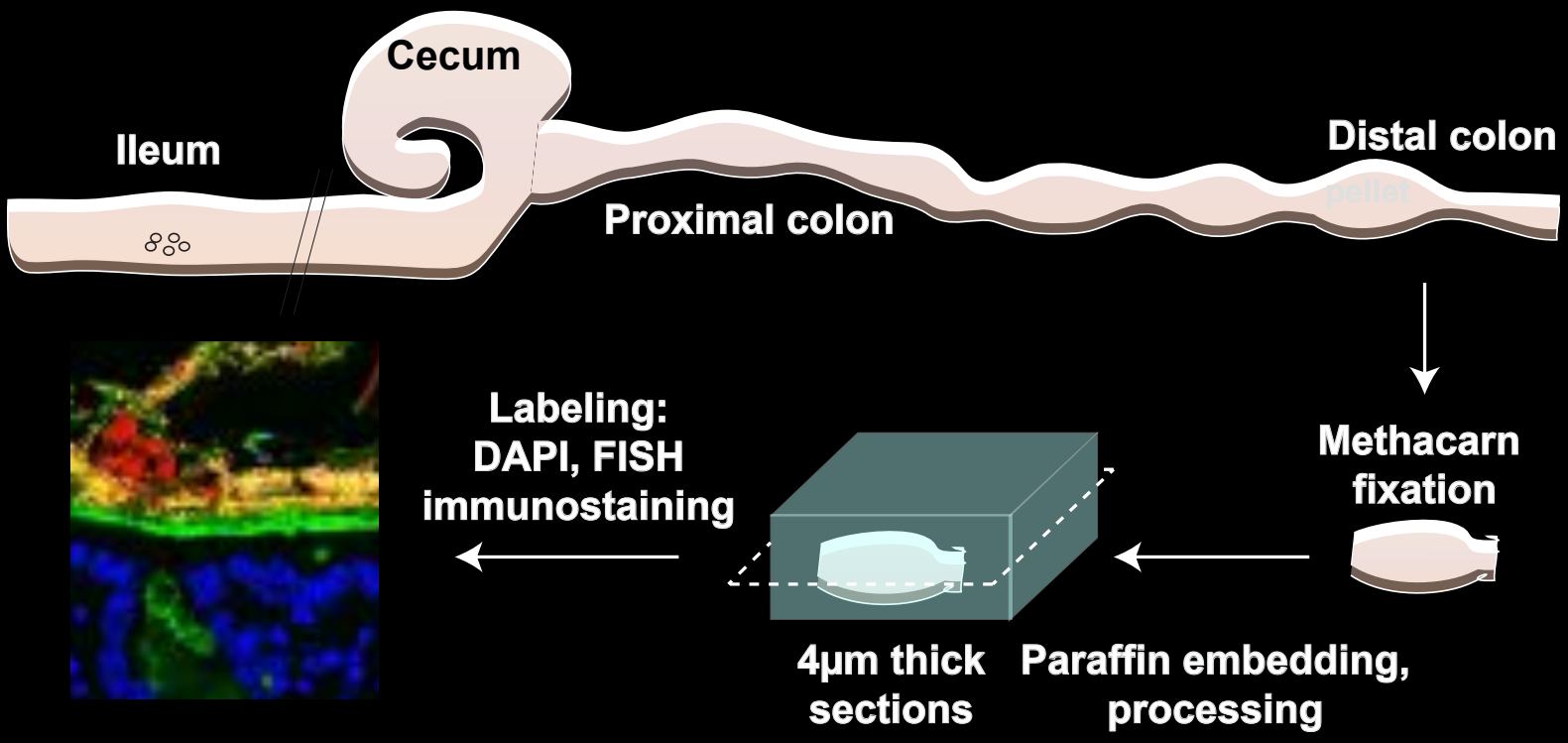
What about non-osmotic effects?

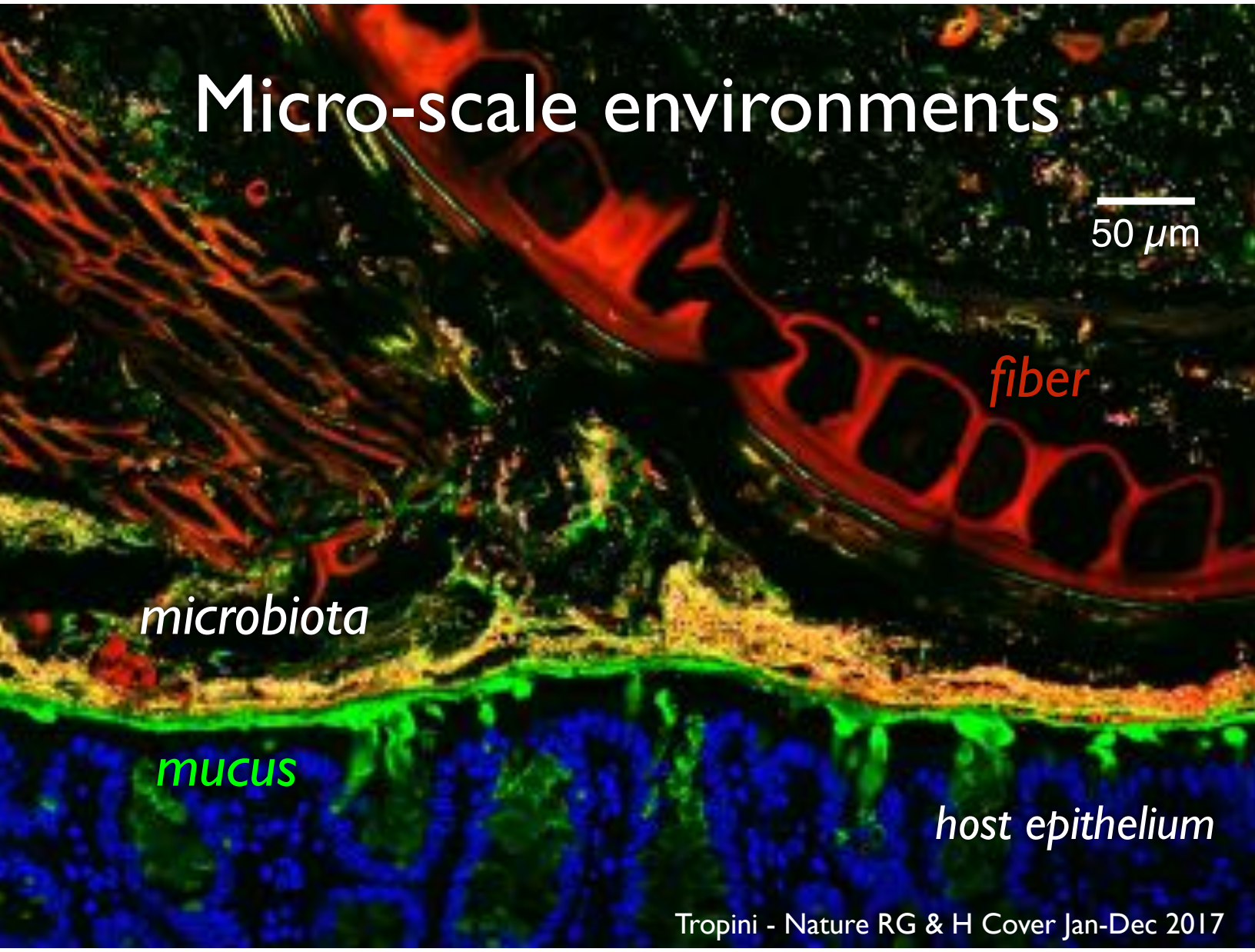


Imaging the host-microbiota interface



Imaging the host-microbiota interface

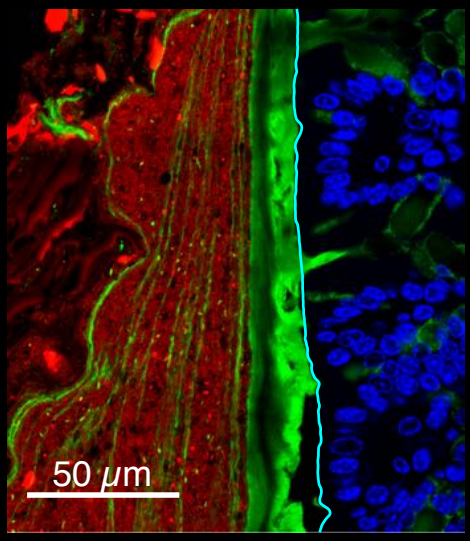



Micro-scale environments
50 μm
fiber
microbiota
mucus
host epithelium
Tropini - Nature RG & H Cover Jan-Dec 2017

Imaging the host-microbiota interface

Distal colon

Bacteria **Mucus** **Nuclei**

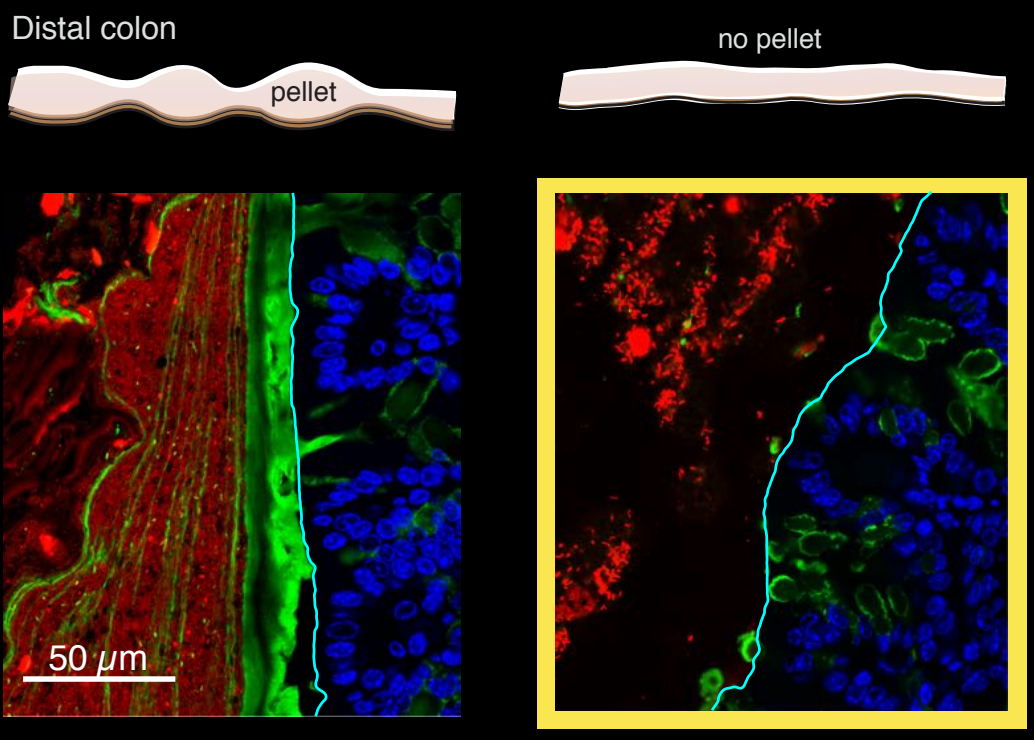


Untreated

Tropini *et al.* June 2018 *Cell*

Osmotic perturbation alters gut landscape and depletes mucus layer

Bacteria Mucus Nuclei

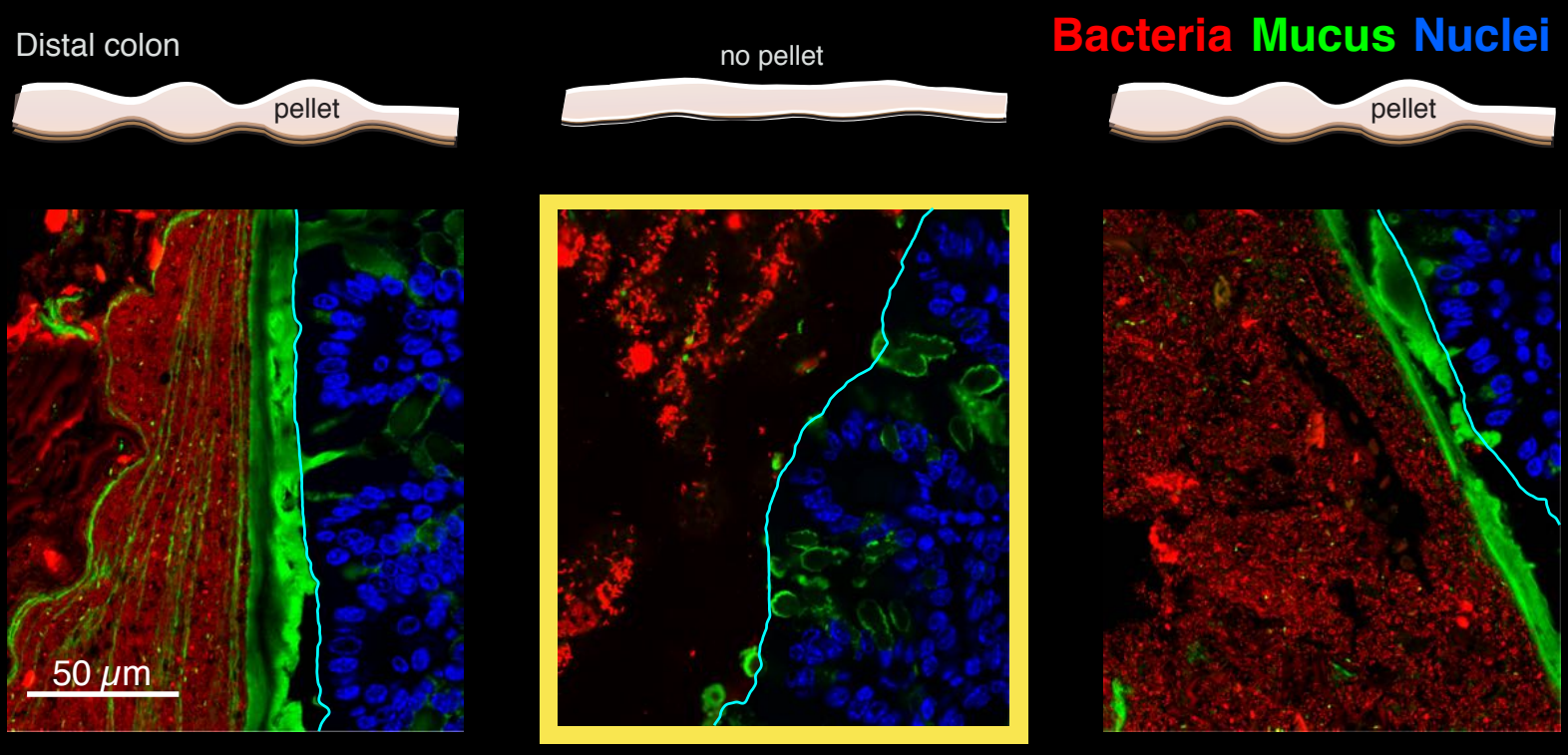


Untreated

PEG (6d)

Tropini *et al.* June 2018 *Cell*

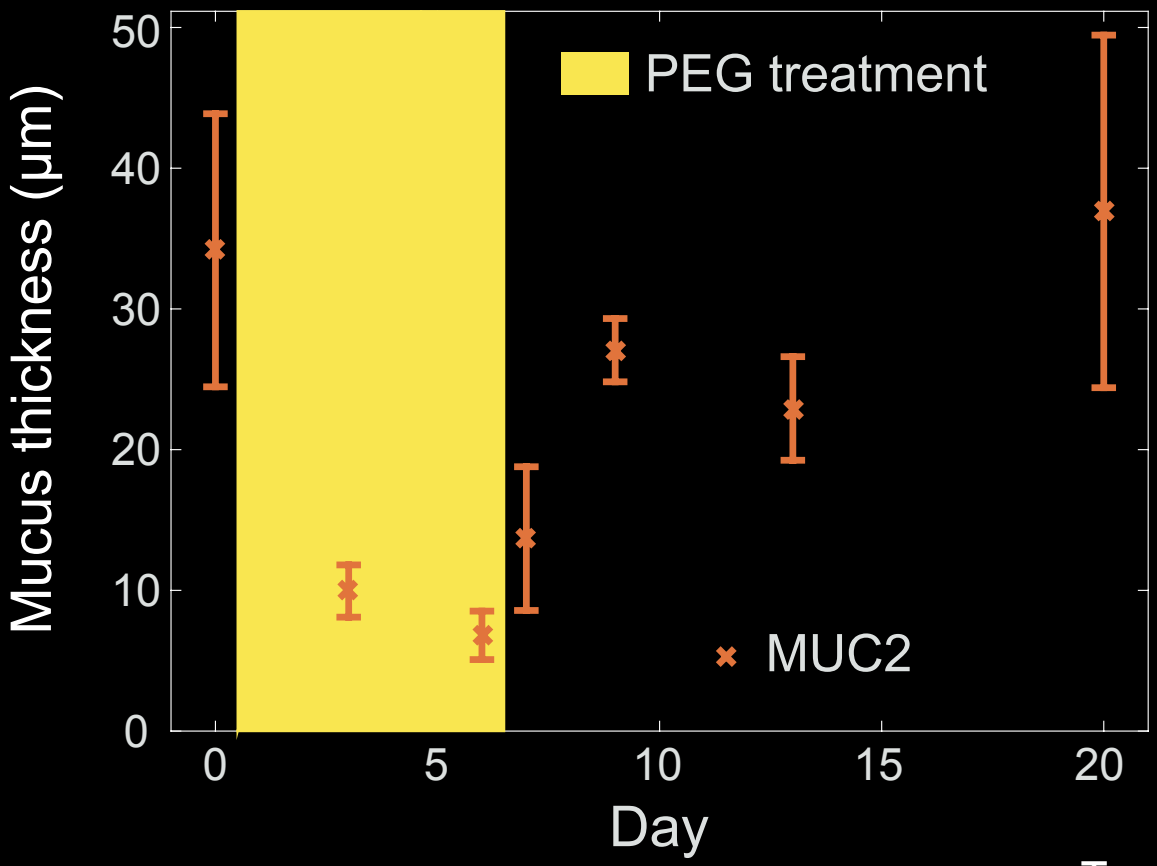
Osmotic perturbation alters gut landscape and depletes mucus layer

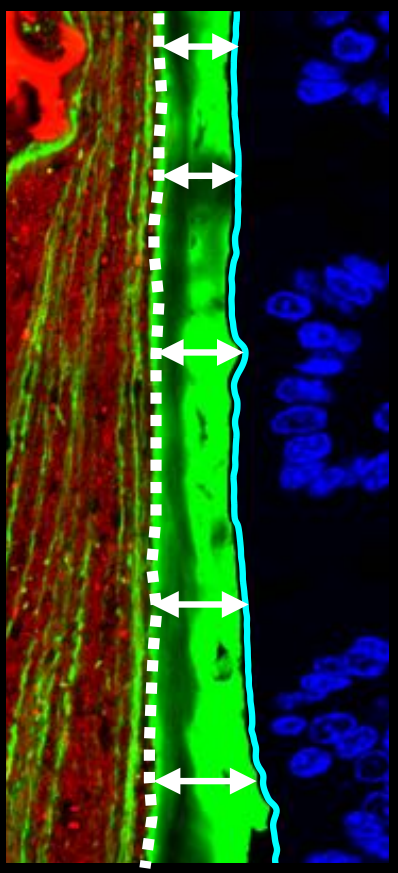


Untreated PEG (6d) 14 d post PEG

Tropini *et al.* June 2018 *Cell*

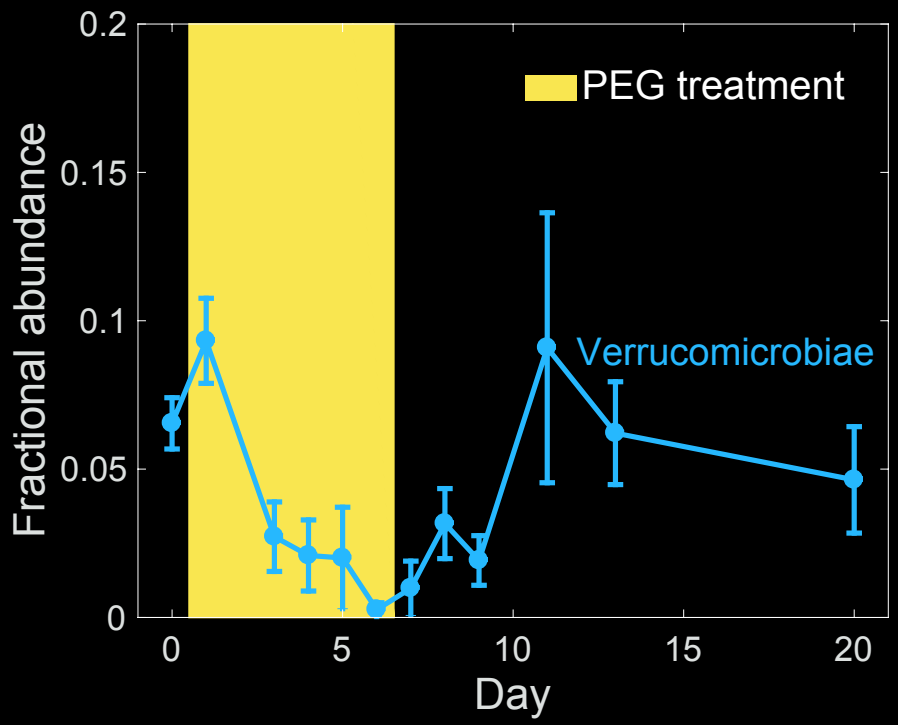
Osmotic perturbation alters gut landscape and depletes mucus layer





Tropini *et al.* June 2018 *Cell*

Mucin-utilizing bacteria reduced in abundance when mucus is low

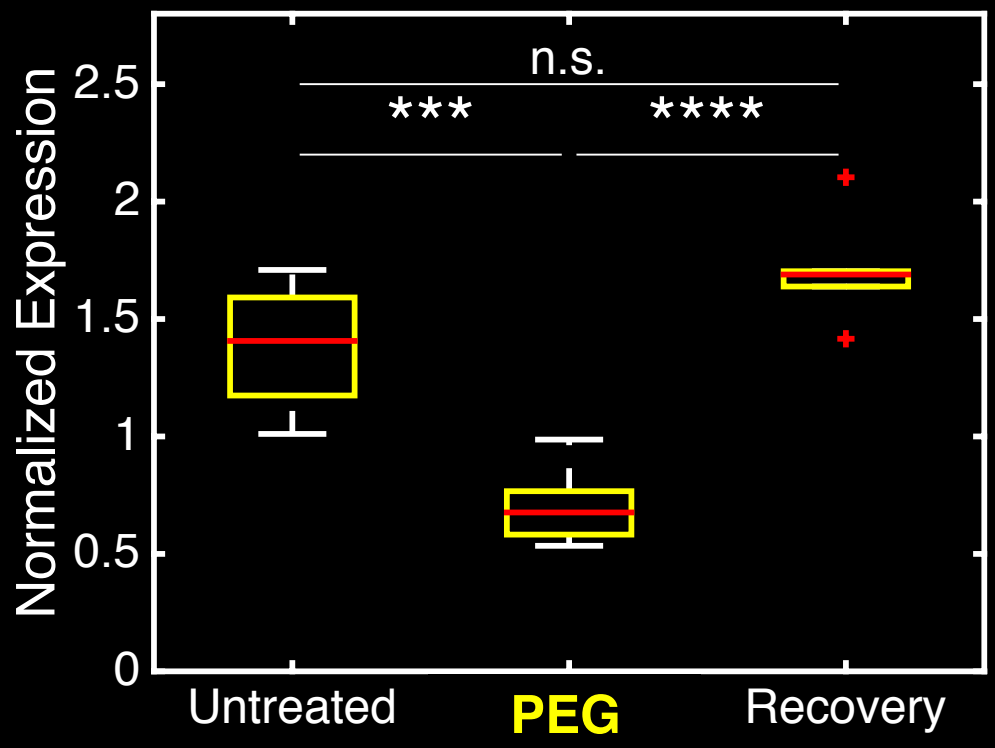


Does the changed environment cause gene expression changes?

Tropini *et al.* June 2018 *Cell*

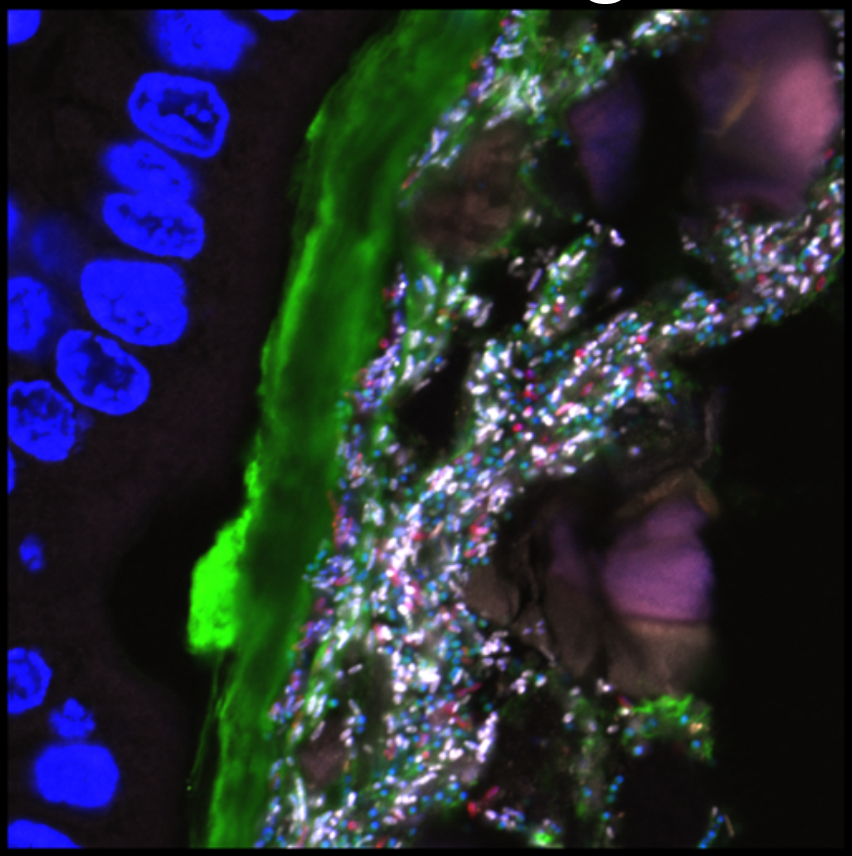
Mucin utilization genes down regulated when mucus is low

(defined community *B. thetaiotaomicron, E. hirae, Lachnospiraceae*)

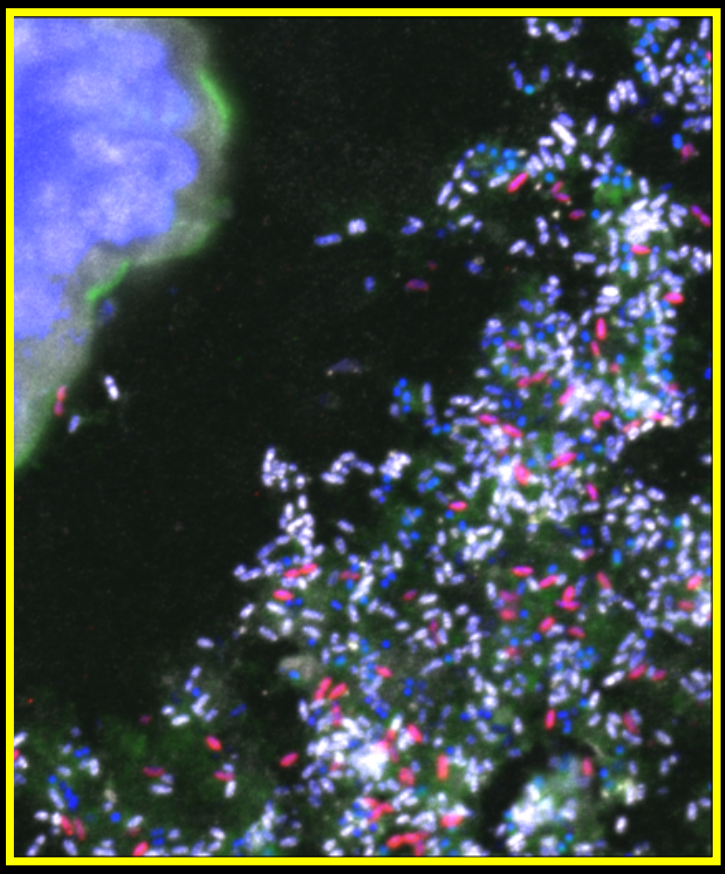


Tropini *et al.* June 2018 *Cell*

Bacterial organization changes during PEG treatment



Normal

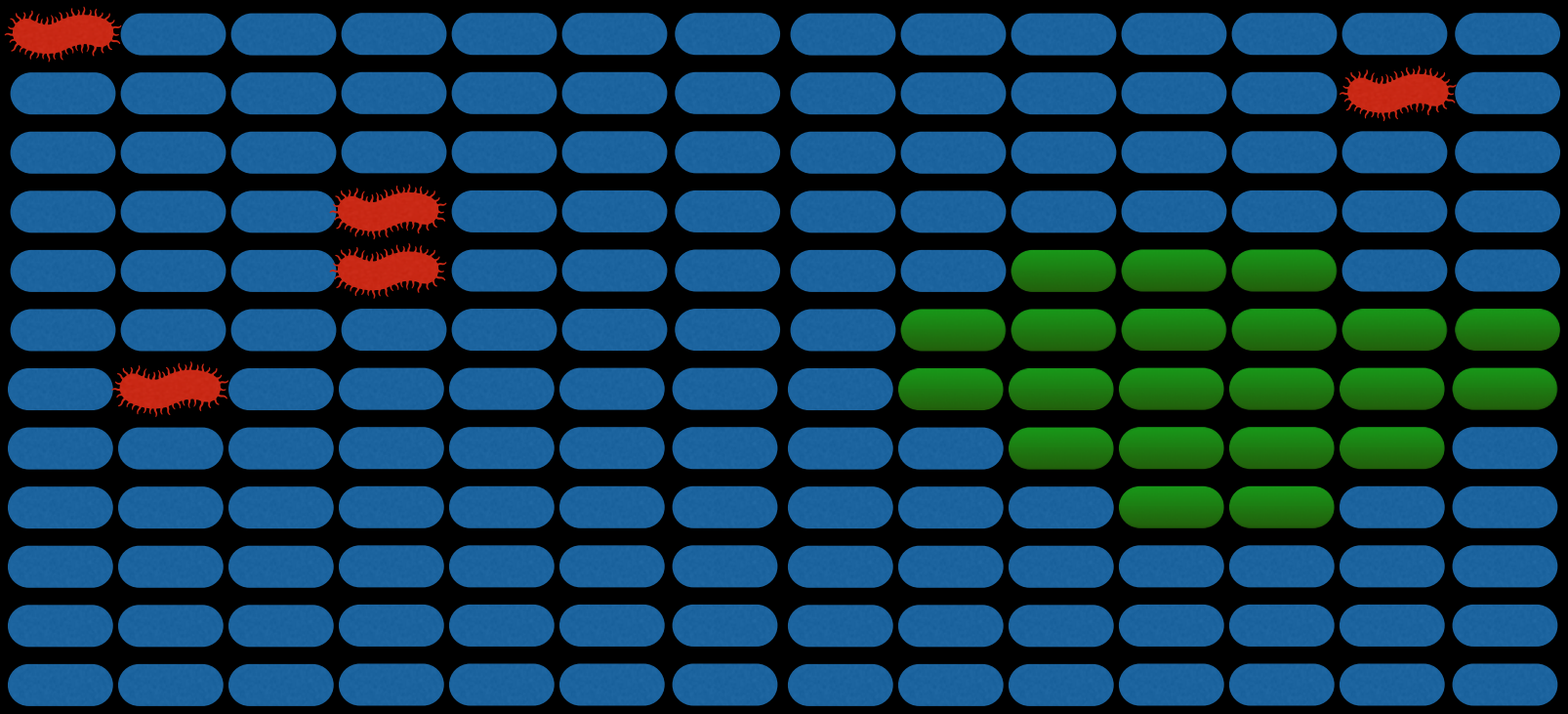


PEG

Enterococcus **Bacteroides** **Lachnospiraceae** **Host nuclei** **Mucus**

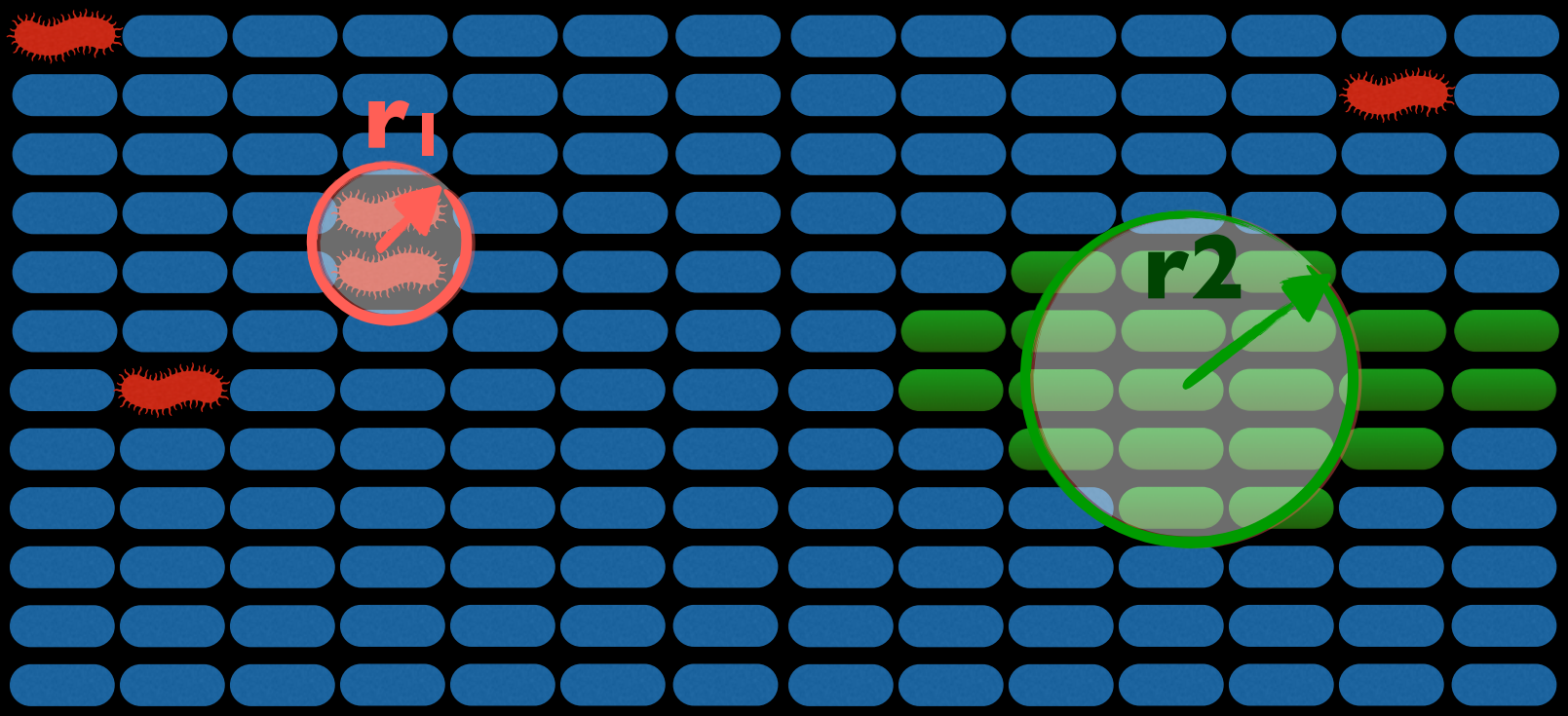
Quantifying bacterial organization

Distance between  and  ,  and  ?



Quantifying bacterial organization

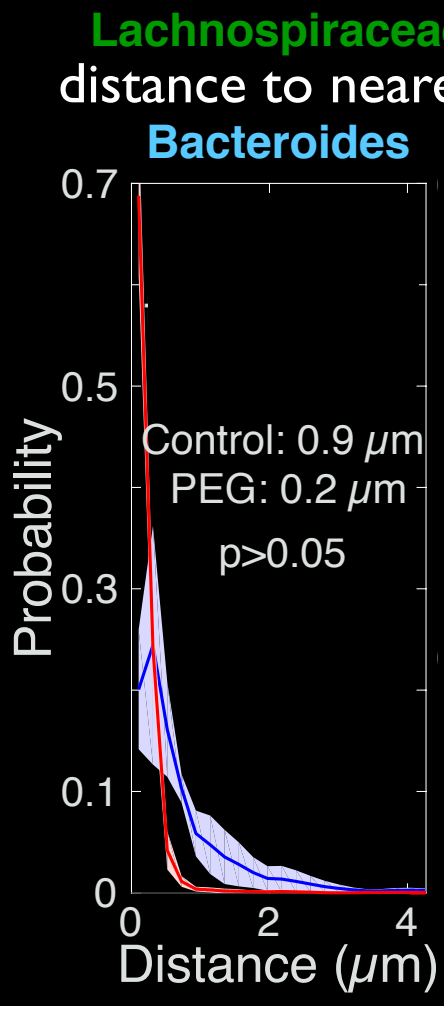
Distance between and , and ?

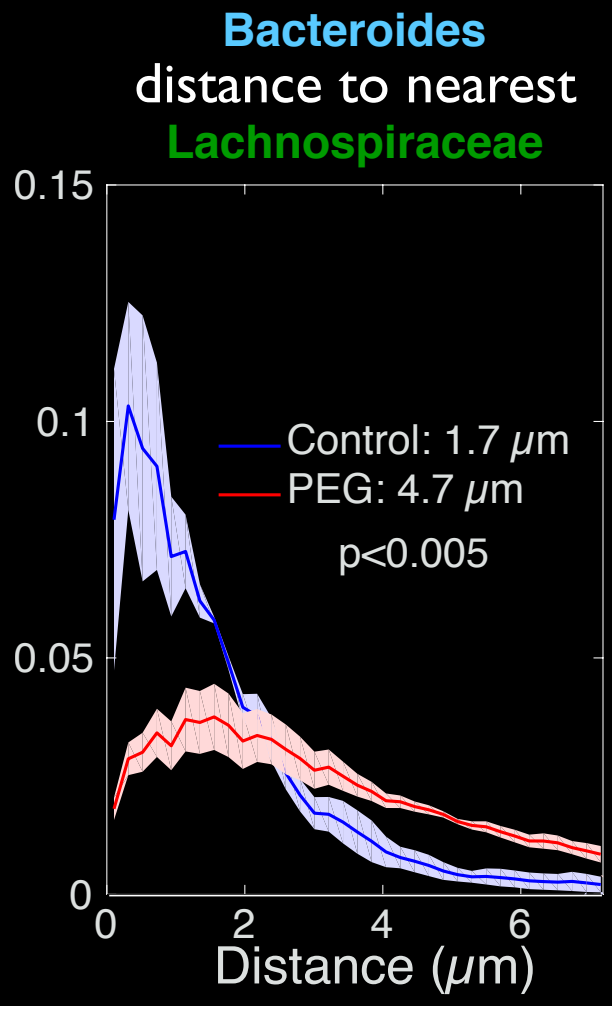


Quantifying bacterial organization

Distance between and , and ?

Bacterial organization changes during PEG





Mild osmotic diarrhea

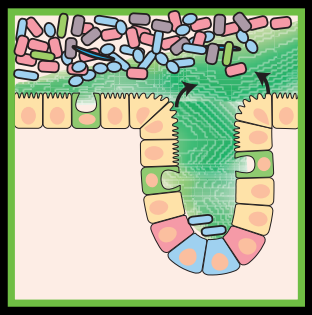
- Mucus,
- Mucus-consuming microbes,
- Mucus utilization genes

Reduced/down-regulated

Bacterial organization is affected

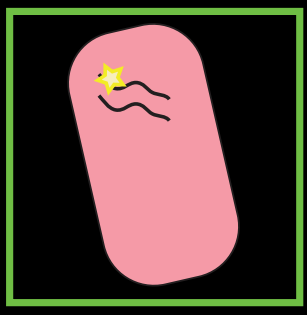
Host

Organ



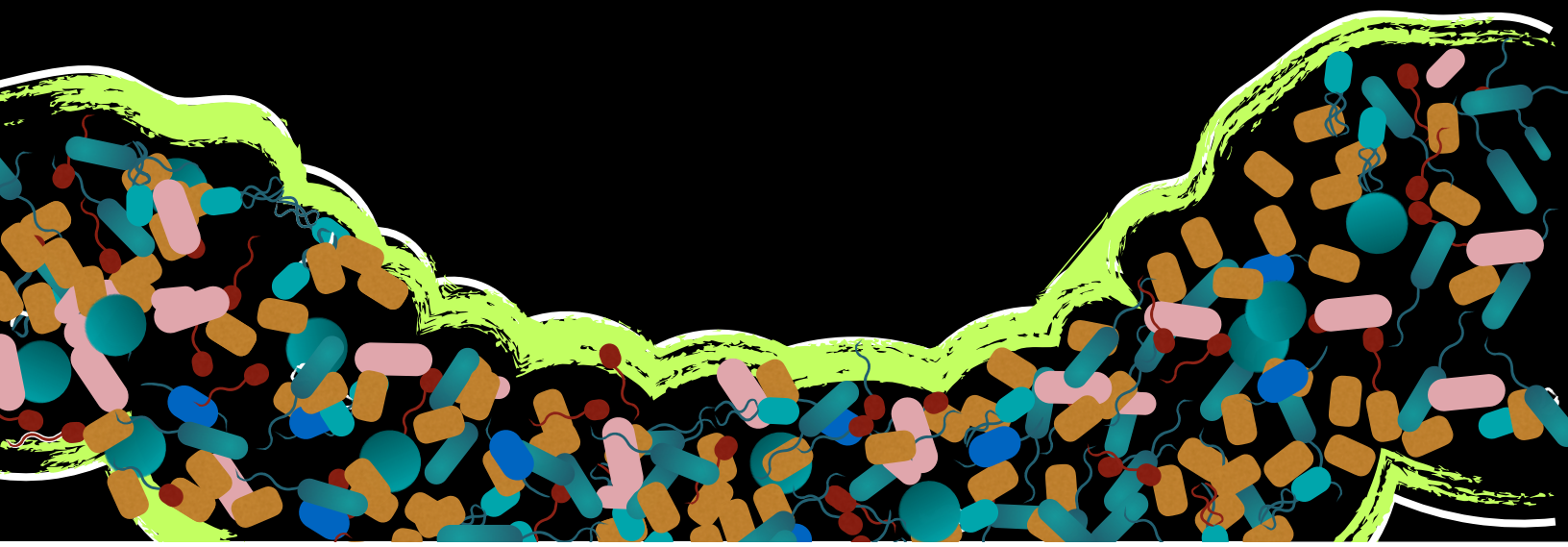
Interface

Bacterial communities

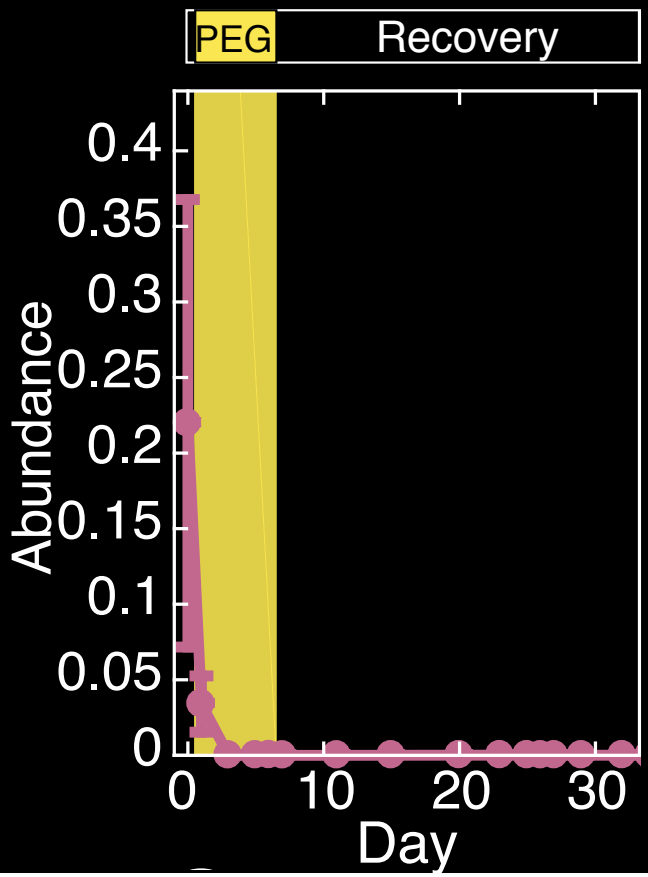


(Sub)cellular processes

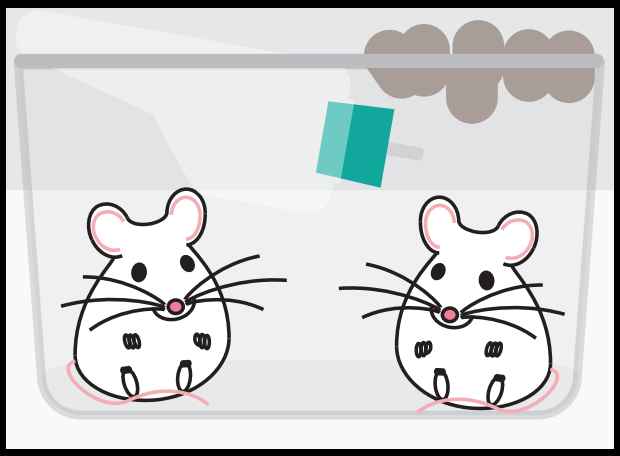
Can species disappearance be avoided/compensated?



Can reseeding occur after microbiota stabilizes?

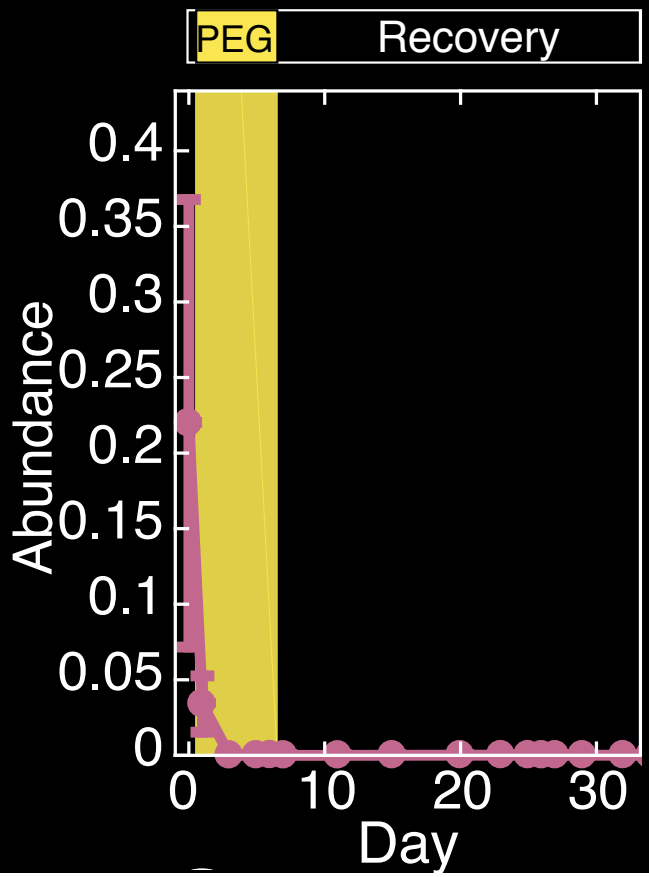


Add healthy microbiota to cage



Tropini *et al.* June 2018 *Cell*

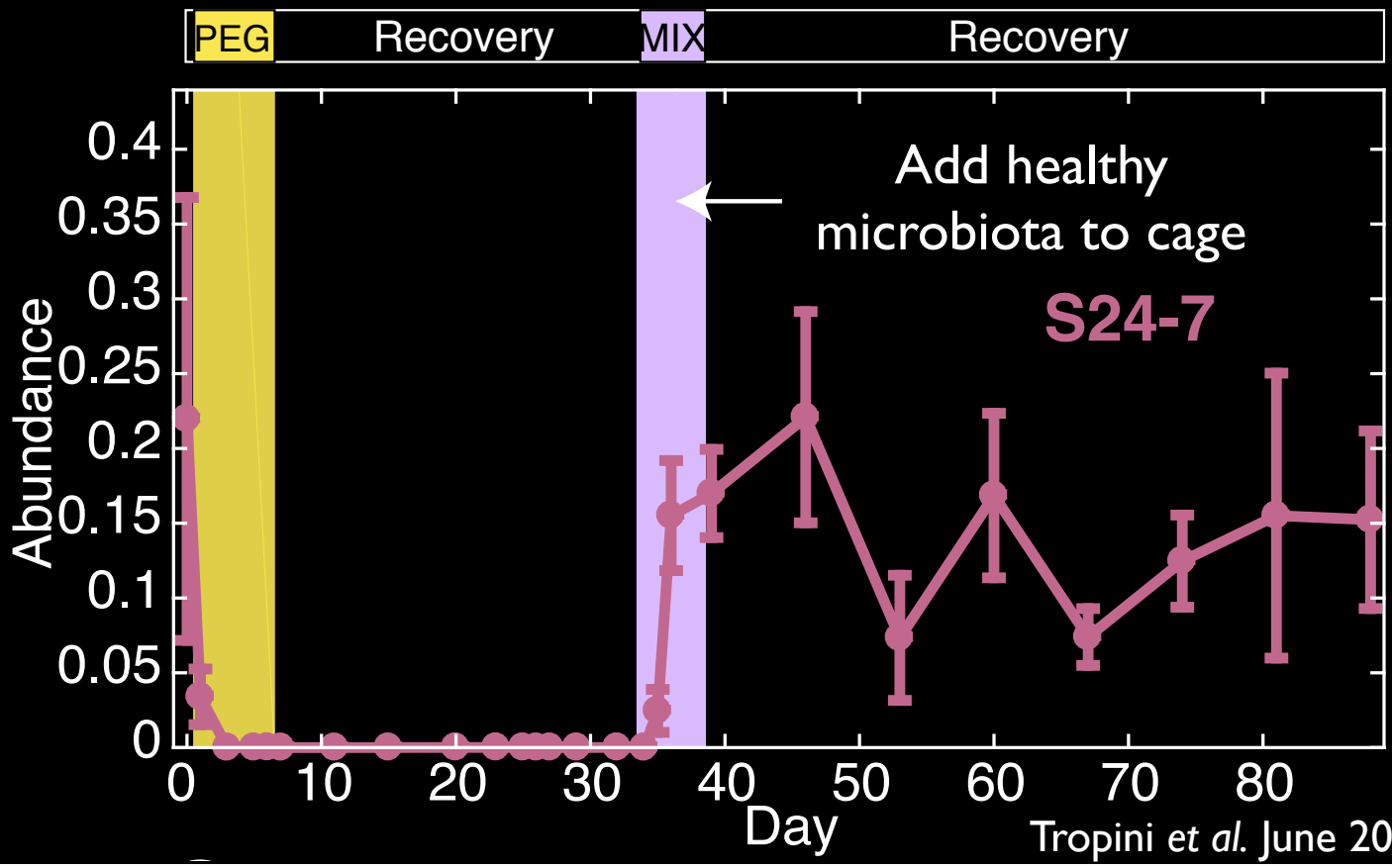
Can reseeding occur after microbiota stabilizes?



Add healthy microbiota to cage

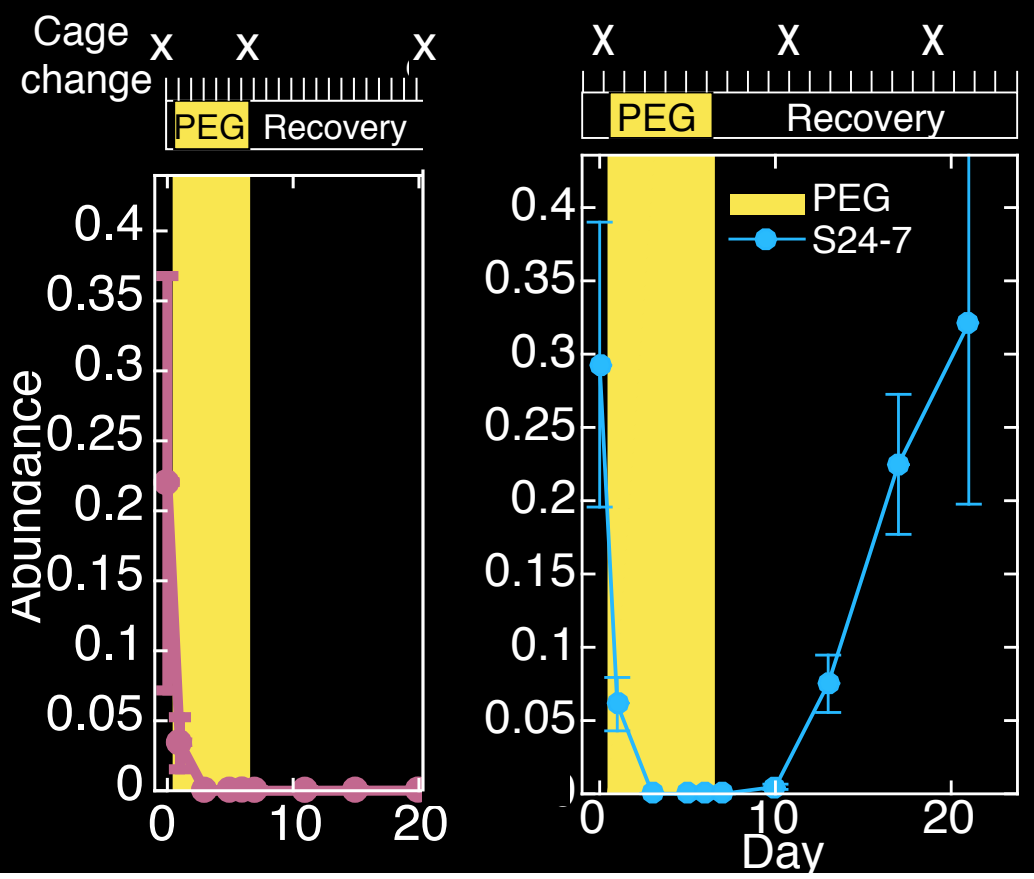
Tropini *et al.* June 2018 *Cell*

Reseeding can occur after microbiota has been at steady-state for weeks

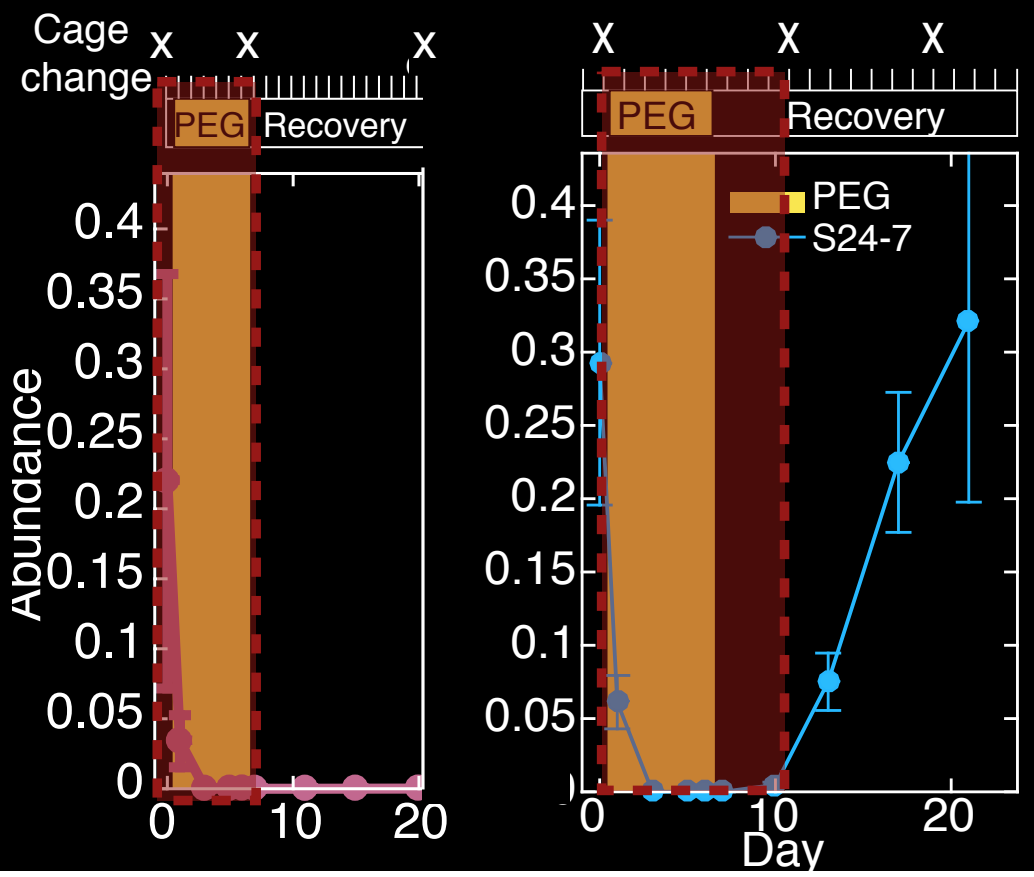


Tropini *et al.* June 2018 *Cell*

Microbiota reprogramming and the physical environment

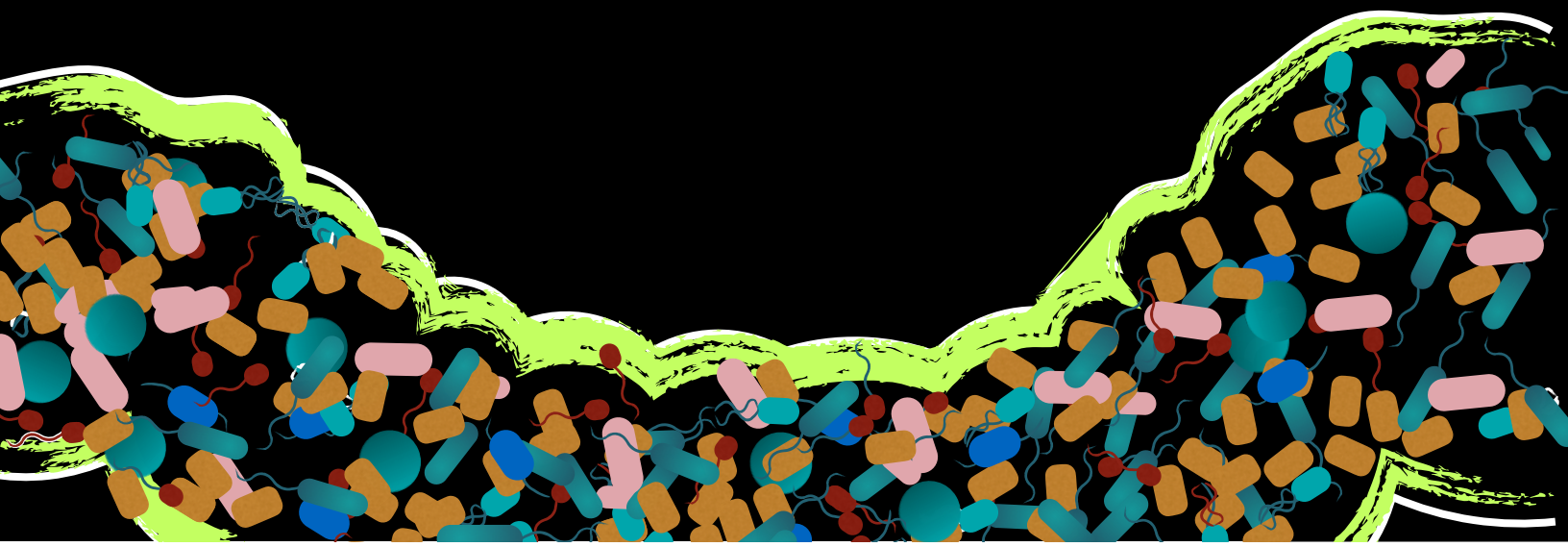


Microbiota reprogramming and the physical environment

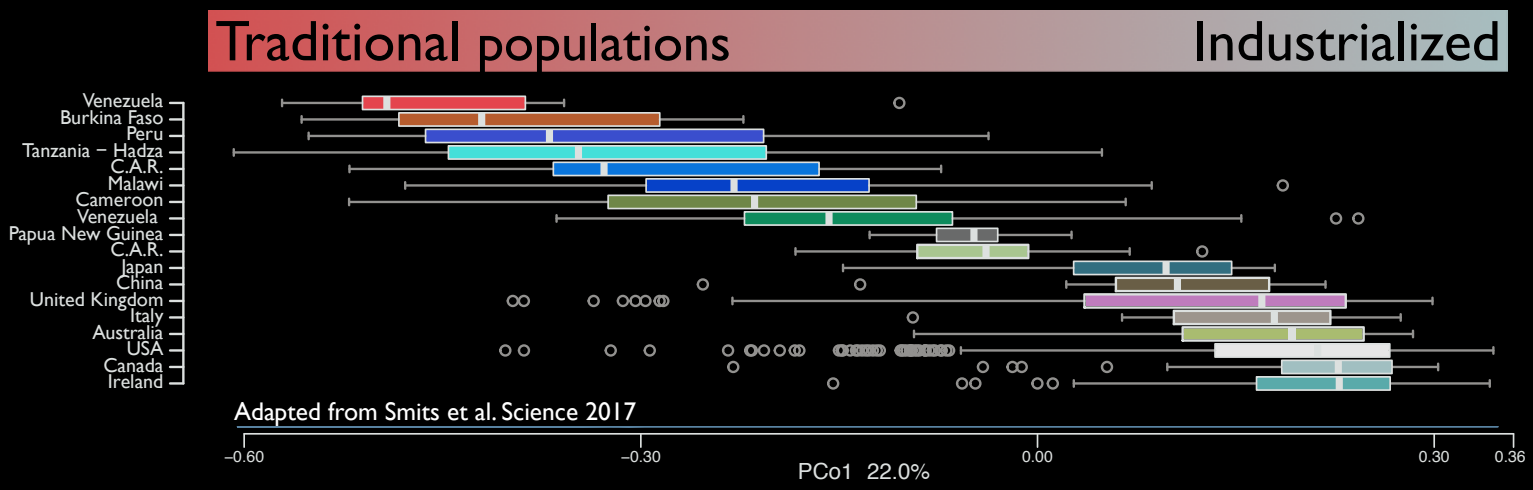


- physical context necessary insight
- resilience aided by reduced sanitation
- timing matters!

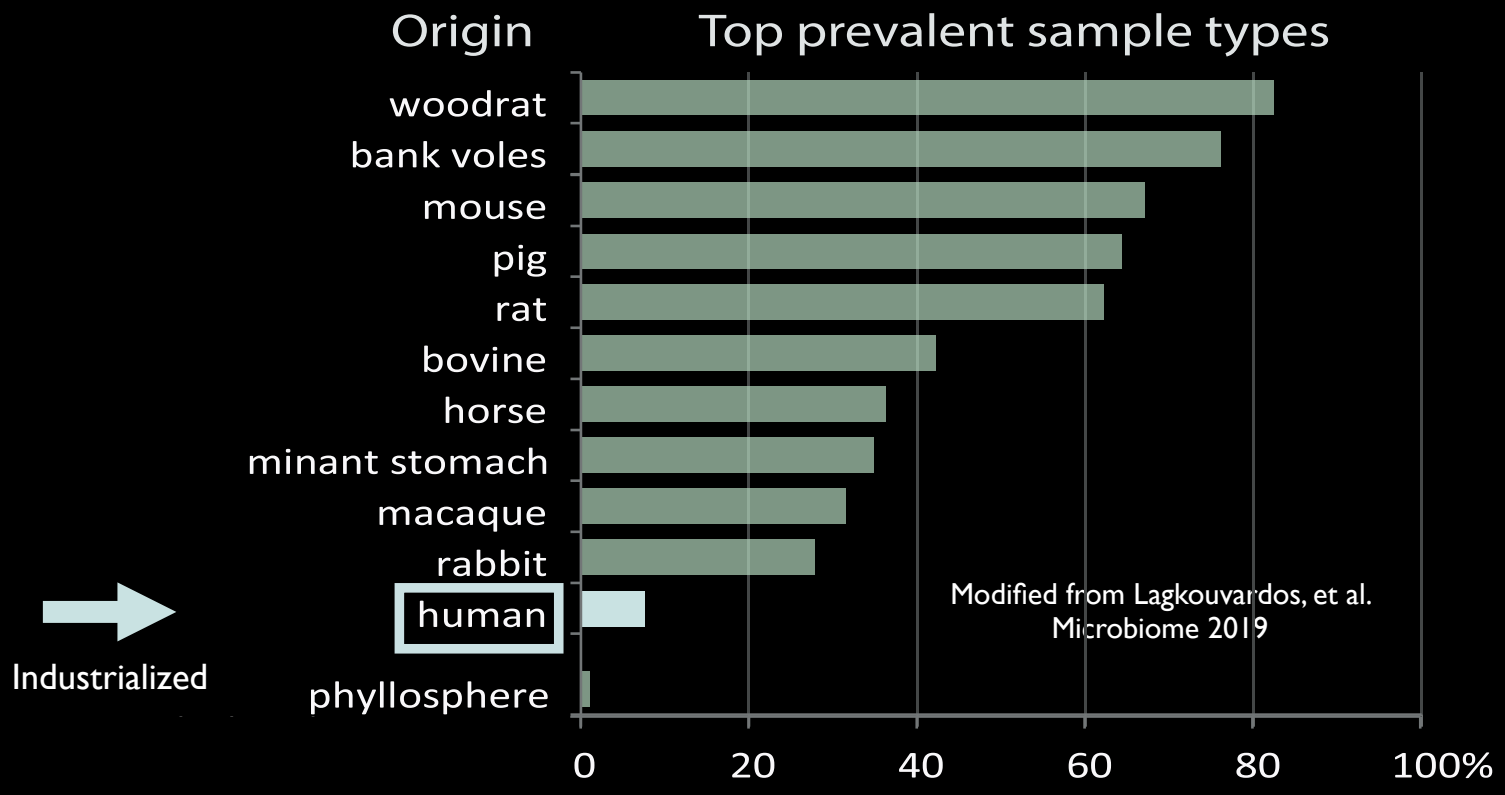
What have we learned about microbiota ecology?



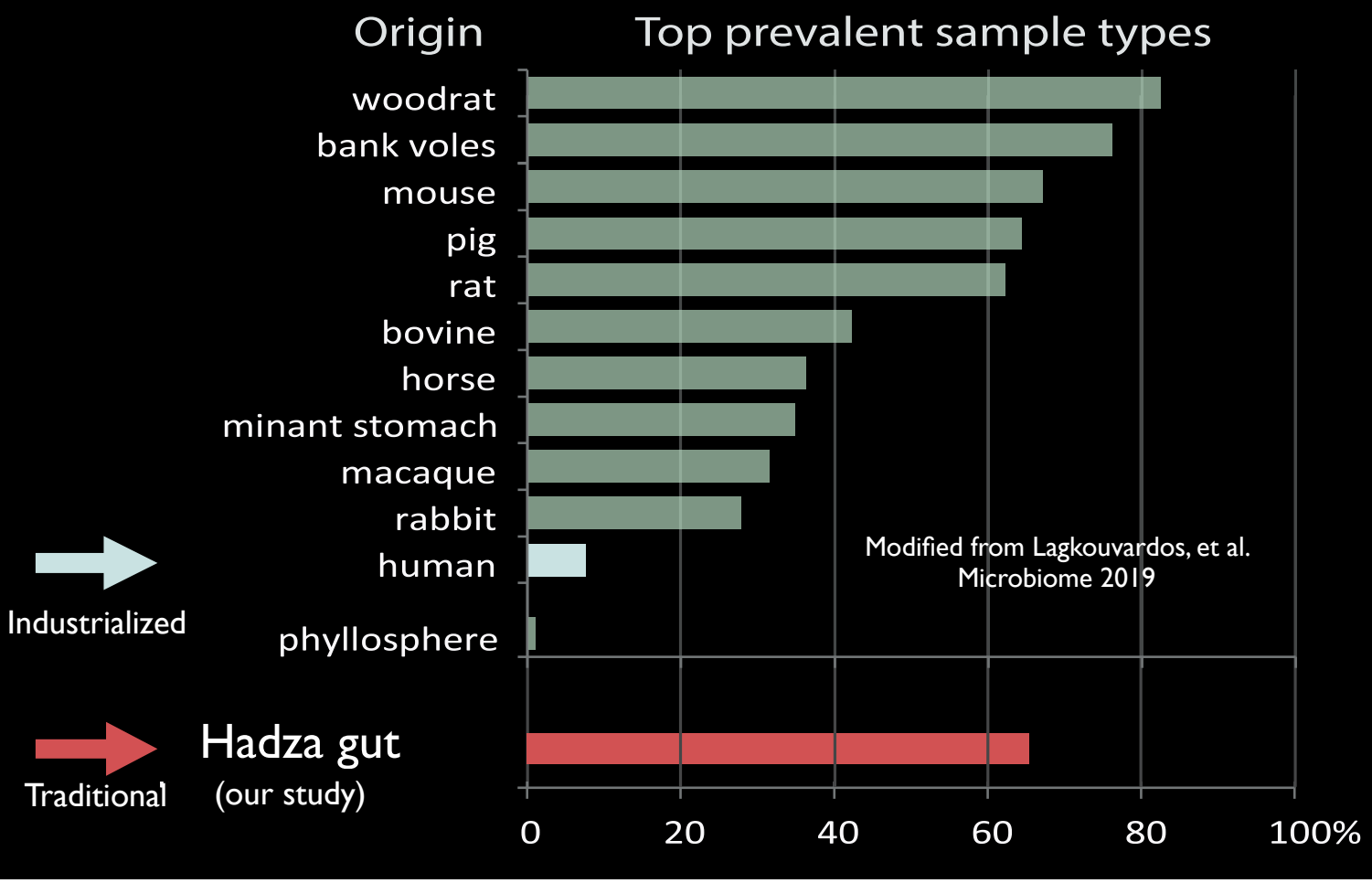
Large scale community engineering



S24-7 is prevalent in animals but not western humans

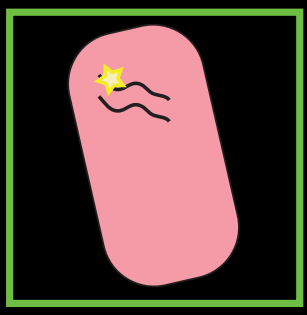


Vanishing taxa - S24-7



The gut ecosystem exhibits limited **resilience** to osmotic diarrhea

Reduced mucin utilization genes expression



Cell-shape changes over short time-scale

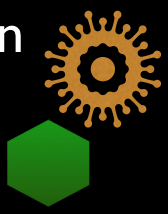
(Sub)cellular processes

Changes in capsular genes

Δ osmolality

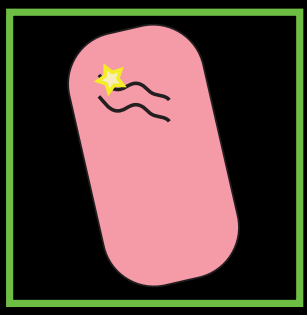
The gut ecosystem exhibits limited **resilience** to osmotic diarrhea

Large shifts in bacterial/phage community composition



Variation in tolerance to changed osmolality

Bacterial communities



(Sub)cellular processes

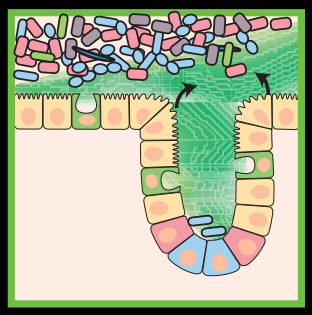
Δ osmolality

The gut ecosystem exhibits limited **resilience** to osmotic diarrhea

Mucus layer depleted

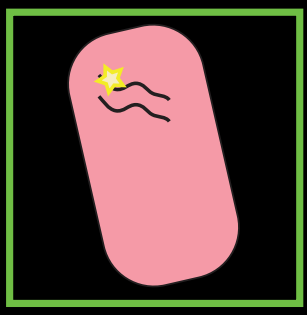
Immune response to commensals

No host inflammation



Interface

Bacterial communities

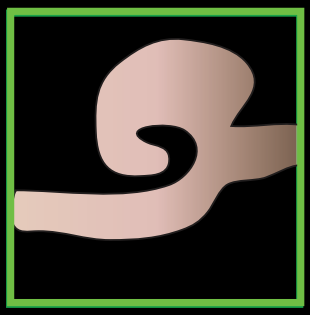


(Sub)cellular processes

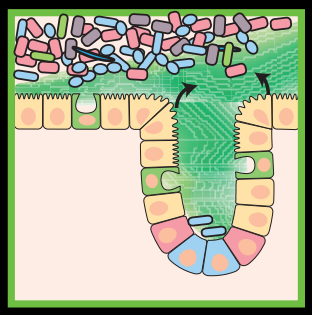
Δ osmolality

The gut ecosystem exhibits limited **resilience** to osmotic diarrhea

Increased stool motility, no significant epithelial damage

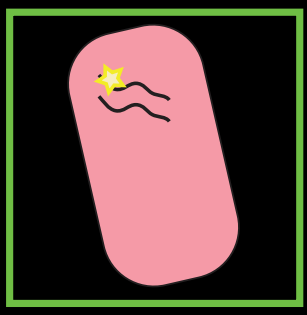


Organ



Interface

Bacterial communities

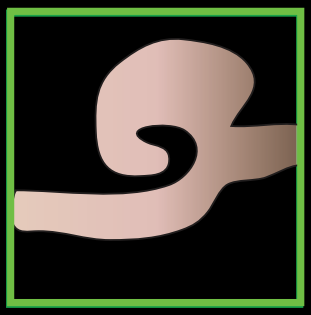


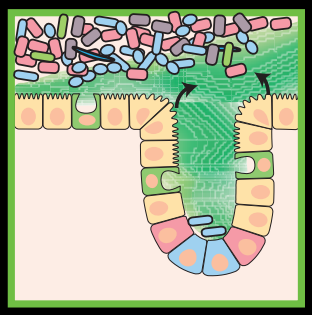
(Sub)cellular processes

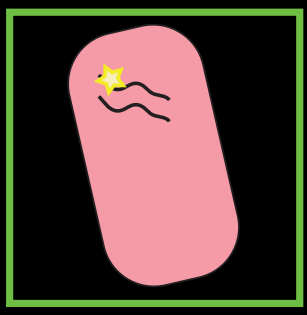
Δ osmolality

The gut ecosystem exhibits limited **resilience** to osmotic diarrhea

Weight loss,
dehydration







Host

Organ

Interface

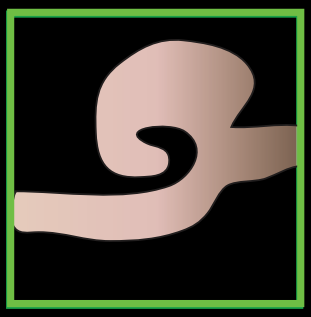
Bacterial communities

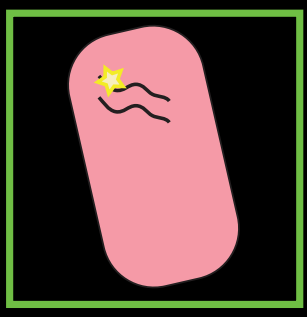
(Sub)cellular processes

Δ osmolality

The gut ecosystem exhibits **limited resilience** to osmotic diarrhea

- Multi-scale understanding is essential
- Host recovers quickly (days) after perturbation
- Microbiota may be perturbed long-term, S24-7 disappearance
- Implications for IBD

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