would reduce the size of dystrophic neurites and improve neuronal function — why would reducing a major pathway of protein degradation be beneficial in a disorder of toxic-protein accumulation? Yuan *et al.* provided tantalizing evidence that dystrophic neurites are major sites through which extracellular amyloid-β is brought into neurons for recycling or degradation. Perhaps PLD3 ablation prevents the endolysosomal pathway from being overwhelmed by a never-ending task of degrading extracellular amyloid-β at dystrophic neurites.

This mechanism complements the idea that early inefficiencies in endolysosome–autophagy pathways in Alzheimer's disease can prevent proper breakdown of amyloid-β, promoting a cascade of events that leads to plaque formation[8]. If a faulty endolysosome–autophagy system triggers this cascade, Yuan and colleagues' work suggests that precise ablation of some of the pathway's components might increase axonal health, by preventing amyloid-β from being brought into neurons in the vicinity of amyloid plaques.

Yuan *et al.* stop short of investigating the cellular mechanisms that underlie the beneficial effects of PLD3 ablation in their animal model. Furthermore, it remains unclear exactly how PLD3 variants increase the risk of Alzheimer's disease and promote accumulation of organelles in dystrophic neurites. Until this knowledge gap is filled, it is premature to propose PLD3-based interventions as potential therapies. Nevertheless, the study provides strong evidence that dystrophic neurites and PLD3 play a crucial part in nerve-conduction deficits in Alzheimer's disease.

It's been more than 30 years since synaptic abnormalities and loss were identified as the earliest, best predictors of cognitive decline in Alzheimer's disease[9]. Synaptic dysfunction (including synapse loss and defects in the plasticity of synaptic connections) is triggered by low concentrations of soluble amyloid-β, and is independent of insoluble amyloid plaques[10,11]. These and other findings cemented the idea that Alzheimer's disease is fundamentally a disorder of synaptic failure and brain-network dysfunction[12,13] — an idea strengthened by the finding that amyloid-based therapies effectively clear plaques, but produce little cognitive benefit[14]. Now, Yuan *et al.* provide support for the relevance of the plaque microenvironment, and potentially point to an inability of amyloid-based therapies to resolve axonal impairments after plaques are cleared[15]. Going forward, approaches to analyse the function of neuronal circuits and networks in the human brain will be needed to truly understand the mechanisms that underlie this disease, if we are to develop successful therapeutics to treat it.

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Microbiology

# A viral cocktail calms gut inflammation

**Alice Bertocchi & Fiona Powrie**

Abnormalities in gut bacteria can contribute to hard-to-treat illnesses, such as inflammatory bowel diseases. Efforts to harness bacterium-targeting viruses reveal a promising way to tackle these conditions.

Inflammatory bowel diseases (IBDs) are chronic conditions of the gut in which genetic and environmental maladaptations drive a breakdown in communication between the cells of the host and the diverse gut bacteria, termed the microbiota[1,2]. Writing in *Cell*, Federici *et al.*[3] present their highly ambitious and systematic approach to targeting bacteria associated with the development of IBD.

The composition of the microbiota varies substantially from person to person, but dysbiosis — lower-than-normal richness and diversity of gut-bacterial species — is a common feature of IBD[2]. This reduction in diversity is associated with an impaired immune response and problems with the cellular barrier that usually blocks bacterial entry from the gut lumen into gut tissue. These malfunctions can cause problems with antibacterial defence mechanisms, leading to the emergence of potentially disease-causing bacteria that thrive in an inflamed gut. Such observations have fuelled attempts to target dysbiosis in IBD. However, therapeutic approaches ranging from drugs such as antibiotics to the use of faecal transfers to populate the microbiota have yielded mixed and inconsistent results[2,4].

The authors harnessed bacteriophages (also known as phages) — viruses that can infect and kill bacteria. Phages can specifically target particular bacterial strains, and therefore offer a therapeutic strategy for IBD in which one or more specific disease-causing bacteria are selected for destruction[5]. Federici and colleagues show that an orally delivered cocktail of phages that target an IBD-associated strain of the bacterium *Klebsiella pneumoniae* attenuated intestinal inflammation in mice (Fig. 1), providing a proof of concept for the use of phage therapy for this condition.

*Klebsiella pneumoniae* is frequently present in oral tissue, but can colonize the gut during dysbiosis, sometimes leading to inflammation[6]. Federici and colleagues collected clinical data from people with IBD and from healthy individuals in four locations around the world. The authors observed that 39% of people with IBD had an increased proportion of *K. pneumoniae* in their stool samples compared with healthy individuals. This also indicates that *K. pneumoniae* colonization of the gut occurs across many diets and lifestyles. Previous work identified a strain of *K. pneumoniae*, called Kp-2H7, in the saliva of people with IBD, that triggers a pro-inflammatory response in the mouse gut[6].

Federici *et al.* investigated *K. pneumoniae* strains in their human samples using a DNA-sequencing approach followed by bioinformatic analyses, and identified one strain (which they named Kp KSB1_4E) as being significantly more abundant in people with IBD than in healthy individuals. Interestingly, this strain belongs to the same genetic branch (clade) as Kp-2H7, which the authors term collectively as an IBD-associated Kp2 clade.

To test whether such Kp2 strains have a causal role in driving gut inflammation, the authors isolated Kp2 and non-Kp2 strains from stool samples of people with IBD, and tested the functional features of these strains using *in vivo* tests in mice. When introduced into

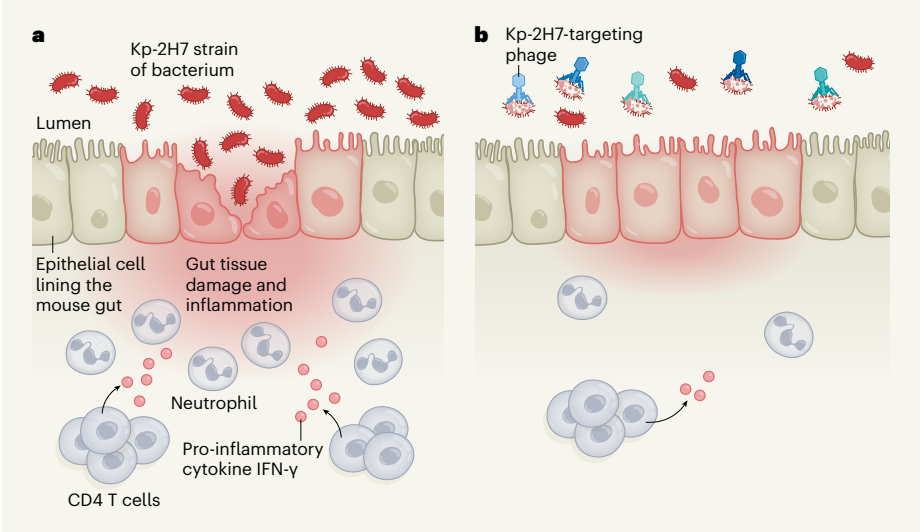


**Figure 1 | A viral treatment that limits gut inflammation.** **a**, Using a mouse model of a gut-inflammation condition called colitis, Federici *et al.*[3] showed that a strain of the bacterium *Klebsiella pneumoniae*, termed Kp-2H7, in the gut lumen caused inflammation. Mice whose guts were colonized only with Kp-2H7 had typical features of gut inflammation (pink indicates inflamed gut tissue), including high numbers of immune cells, called CD4 T cells, that produce the protein IFN-γ and abundant immune cells called neutrophils. **b**, Treatment with a cocktail of five viruses (called phages; different shades of blue indicate different phages) that specifically target the Kp-2H7 strain for destruction lessened the inflammation and tissue damage induced by Kp-2H7 infection.

mice lacking their own microbiota (germ-free mice), both types of strain drove comparable production of the pro-inflammatory protein IFN-γ (a type of immune-signalling molecule known as a cytokine) by pro-inflammatory T cells of the immune system. However, Kp2 strains induced a response with higher levels of IFN-γ relative to the levels of the anti-inflammatory cytokine IL-10, presumably reflecting a more pronounced inflammatory environment after Kp2-strain colonization compared with non-Kp2-strain colonization.

Consistent with these results and those of a previous study[6], germ-free mice that lacked the gene encoding IL-10, and that were solely colonized (mono-colonized) with Kp2 strains, developed severe gut inflammation (colitis). The animals showed marked gut infiltration of immune cells compared uninfected mice that lacked the gene encoding IL-10, confirming the potential of Kp2 strains to drive gut inflammation. Further studies will be needed to determine whether the ability to induce colitis is unique to Kp2 strains or is a common feature of other *K. pneumoniae* strains. It would also be worth testing whether Kp2 strains drive colitis in model systems that have a more diverse and physiological microbiota than that of mono-colonized germ-free mice.

The authors then determined whether targeting Kp2 strains through phage therapy would reduce intestinal inflammation. They undertook the mammoth task of screening environmentally sourced phages to identify those that target Kp2 strains. A cocktail of five phages was identified that consistently reduced the *K. pneumoniae* levels in antibiotic-pretreated Kp2-infected mice (such antibiotic use allows *K. pneumoniae* to efficiently colonize the gut).

Federici *et al.* next used an acute model of colitis that disrupts the gut barrier, allowing abnormal bacterial movement from the gut lumen into the gut tissue and promoting inflammation. Importantly, this model allowed them to show that the phage cocktail inhibited colitis in Kp-2H7-mono-colonized mice compared with untreated control mice. Phage administration reduced *K. pneumoniae*, diminished immune-cell infiltration driven by the bacterium in the gut and lowered the production of inflammatory cytokines, such as IFN-γ. This and previous work[7] demonstrate that phage therapy is effective when given as a preventive treatment. However, whether it can successfully combat established inflammation, a scenario that is more relevant in a clinical setting, is unknown.

Federici *et al.* carried out a pilot interventional study in healthy volunteers. The authors showed that Kp2-targeting phages could be detected in stool samples six days after people received phage treatment, indicating that these phages could survive in the gut. The treatment was well tolerated and did not induce bacterial dysbiosis. Although preliminary, these results are encouraging and worthy of further validation. Because *K. pneumoniae* abundance is low in people who do not have IBD, it was not possible to assess the specificity of the phage targeting of Kp2 strains or its off-target effects on other bacteria in a complex microbiota.

The study by Federici and colleagues is impressive for its scope, spanning clinical observations, mechanistic analysis in animal models and human studies. Moreover, the research moves the field a step nearer to the development of personalized medicine for the treatment of IBD by attempting to target specific disease-causing bacteria present in individual patients.

However, questions remain. For example, the phage cocktail was administered to mice that were colonized only with *K. pneumoniae*, so it is unknown whether this treatment would be as efficient in a gut with a complex bacterial community. Studies[1,2,4] indicate that changes in the bacterial community occur in IBD, raising the question of whether targeting one bacterial strain would suffice, or whether it would be necessary to use combinations of phages to target different species simultaneously.

Phages are normally abundant in the gut environment[5]. Over the past decade, research has identified IBD-specific changes in gut-dwelling phages that might shape the composition of the microbiota[8–10]. When looking at this through the lens of a predator–prey relationship, people with IBD have an increased amount of bacteriophage (predator) and reduced bacterial (prey) diversity compared with the case in healthy individuals[8–10]. This raises the question of how the addition of phages would affect an already overpopulated phage ecosystem in IBD; a rise in inflammation might occur as a result of further phage-mediated bacterial killing.

Federici and colleagues' study paves the way for further analysis of the use of phages as a treatment for IBD. Testing such an approach in people with the condition will be a key next step and one worth pursuing, because success might also usher in an era of personalized medicines. Lessons learnt from such approaches for IBD might also inform similar strategies for other conditions associated with maladapted host–microbiota responses, such as colorectal cancer and obesity[11,12].

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