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VIROLOGY

Phages make a group decision

It emerges that phage viruses, which infect bacteria, use small peptides to communicate with each other. This observation of intercellular communication also reveals how viruses make a key developmental decision.

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When a major decision looms, people often seek advice from friends or relatives. A study online in *Nature* by Erez *et al.*[1] reveals that bacterium-targeting viruses known as phages also show a collaborative aspect to a developmental decision-making process. In this case, intercellular communication occurs through the use of a viral peptide. Intercellular molecular communication between viruses hadn't been observed before, and this study illuminates a previously unknown mechanism of viral action.

After injecting the phage genome into a bacterial host cell, most phages have two life-cycle options. The phage can enter the lytic developmental pathway and eventually destroy its bacterial host in a process that will release dozens of new phage particles into the environment. Alternatively, it can follow the pathway of lysogeny, sparing the host cell, protecting it from subsequent phage infection, and integrating the phage genome into the host genome[2]. Depending on the prevailing conditions, either lytic or lysogenic phage development might be more advantageous for the survival of a phage population.

The importance of the decision between lytic and lysogenic development for the evolutionary success of phages is emphasized by the complexity of the regulatory mechanisms involved. Hundreds of research papers have been generated from studies of the decision between lysis or lysogeny for the phage lambda, which infects the bacterium *Escherichia coli*, and this model system has been described in Mark Ptashne's widely read book *A Genetic Switch*[3]. However, even this highly studied system is not fully understood[4]. These previous studies focused on the many intracellular signalling pathways and genetic circuits that contribute to the process of deciding between lysis and lysogeny. The remarkable finding by Erez *et al.* is that the choice in some phages is also profoundly influenced by communication, by means of small molecules, between phages infecting other cells in a culture.

The initial goal of Erez and colleagues' study was to test whether bacteria that have been infected by a phage might produce and secrete molecules to alert other bacterial cells to phage infection. To test this hypothesis, the authors infected *Bacillus subtilis* bacteria with four different phages and screened the culture medium three hours later for the presence of molecules that could inhibit phage infection. Surprisingly, rather than identifying a bacterially produced molecule, the authors found a molecule that was synthesized by one of the phages, phi3T. This viral molecule protected *B. subtilis* cells from infection by phi3T, but not from infection by the other phages tested. Additional experiments showed that this molecule was a small peptide that protected bacterial cells from phage-induced lysis by promoting phage lysogeny.

An idea for how this viral communication system might work came from a comparison with bacterial communication systems. Quorum sensing is a mechanism used by some bacteria to sense the population level of their nearby close relatives[5]. In the Gram-positive group of bacteria, this process often involves the secretion of a small peptide (frequently less than 10 amino-acid residues in length) from one bacterium that is then taken up by another bacterium, stimulating changes in gene expression[6]. Erez *et al.* sequenced the phi3T genome and identified a gene encoding a protein 43 amino-acid residues long that they named AimP. This protein shows strikingly similar features to those of proteins involved in

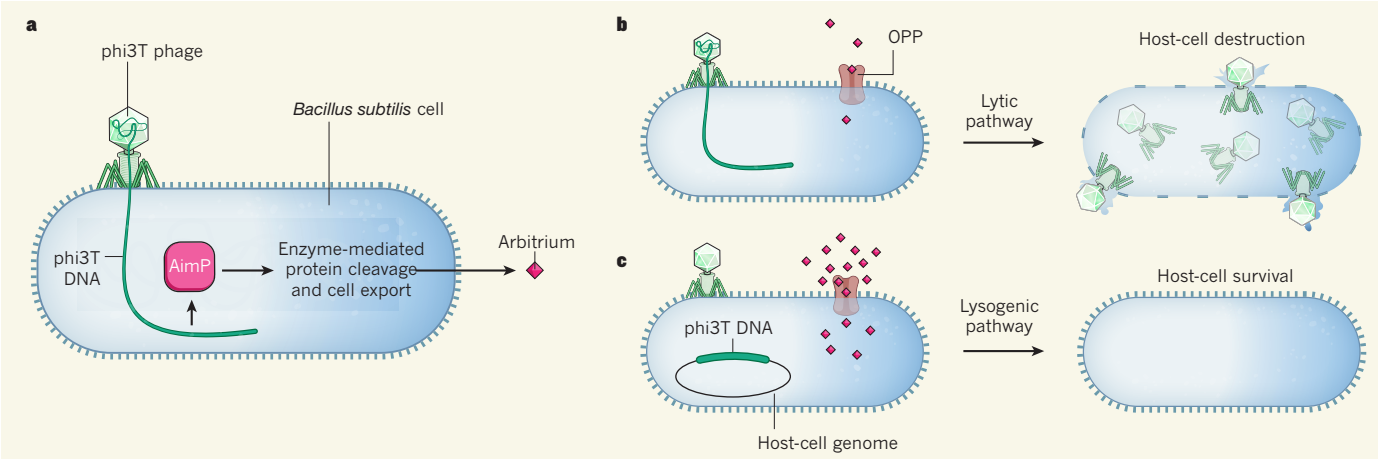

Figure 1 | Phages use a small peptide to communicate with each other. **a**, Erez *et al.*[1] investigated *Bacillus subtilis* bacterial cells infected by a phage virus called phi3T. They identified a phage protein, AimP, that undergoes enzyme-mediated cleavage to form a peptide fragment that is exported from the bacterial cell. This fragment, termed arbitrium, enables intercellular viral communication, a phenomenon that had not been observed previously. **b**, Arbitrium is taken up by neighbouring bacterial cells through the bacterial OPP transporter protein. If low levels of arbitrium are present in a bacterial cell, phage infection has a higher probability of following the lytic developmental pathway that eventually destroys the host cell and releases further virus particles into the environment. **c**, High levels of arbitrium in a bacterial cell increase the probability that phage infection will result in the lysogenic developmental pathway, which results in the phage genome being integrated into the host-cell genome and in host-cell survival.

quorum sensing and other peptide-mediated signalling pathways in *B. subtilis*.

Erez *et al*. found that, during bacterial infection by phage phi3T, enzyme-mediated cleavage of AimP produced a six-residue peptide, called arbitrium by the authors, which was secreted from the bacteria. Uptake of arbitrium by other bacterial cells through the bacterial oligopeptide permease (OPP) transporter protein increased the probability that phi3T infection of the bacteria would result in lysogeny. At the high concentrations of arbitrium reached during the late stages of a phage-infection process, bacterial-cell lysis was strongly suppressed because of the high frequency of phage lysogeny (Fig. 1).

The authors found that arbitrium acts by binding to an intracellular phage protein, AimR, and inhibiting its activity. AimR can bind to a specific site on the phage genome and activate transcription of the *aimX* gene, which promotes the lytic pathway through an unknown mechanism. Erez and colleagues observed that arbitrium in bacterial cells reduces the expression of *aimX*, and thus increases the probability of lysogeny.

Notwithstanding the complexity of the arbitrium system, the logic is simple. During the initial period of phage infection, when the concentration of phage particles is low and that of bacteria high, the production of phage particles through the lytic pathway could provide a successful bacterial-infection strategy. However, as the number of phage particles increases, the concentration of host cells might decrease to a level at which phage particles could no longer find a host cell to infect. In this context, an approach to preserve host cells and phage genomes by promoting the phage's lysogenic life cycle would be preferable. Interestingly, the probability of phage lambda lysogeny increases when multiple phages simultaneously infect the same cell[7]. However, the phi3T system provides the first example that I know of in which phages infecting different cells have been shown to communicate with one another through a small molecule, and this communication aspect is what makes the work by Erez and colleagues so exciting.

The authors found that communication systems similar to that involving arbitrium exist in more than 100 phages, indicating their general utility for phage survival. Furthermore, Erez *et al.* demonstrated that the arbitrium system of another phage operates in a similar manner, although it uses a different peptide sequence. These arbitrium peptides affect only the phage that produces them. The authors observed a wide diversity of peptide sequences among phages that had arbitrium-like systems, implying an evolutionary drive for phage specificity. These phages are 'speaking' different molecular languages and so convey messages only to their own kind.

Small-molecule communication between phages enables the viruses to strongly influence the decisions of their descendants. Such communication provides a stunning example of the complexity and nuance of function that can be achieved by 'simple' entities. Despite extensive investigation of phage genomes, most have many genes of as yet unknown function. Erez and colleagues' discoveries provide a hint that some uncharacterized phage genes might be involved in mechanisms that increase the viruses' evolutionary fitness. Perhaps we will find that phages can communicate with each other on many other topics, too. ■

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