

Induction Mechanisms and Strategies Underlying Inter-Prophage Competition During Polylysogeny

Justin E. Silpe^{1,2,3}, Olivia P. Duddy^{1,3}, and Bonnie L. Bassler^{1,2,*}

1 Department of Molecular Biology, Princeton University, Princeton, NJ, USA

2 Howard Hughes Medical Institute, Chevy Chase, MD, USA

3 These authors contributed equally

*Correspondence: bbassler@princeton.edu

INTRODUCTION

Phages play central roles in shaping bacterial community biology. For example, lytic phages, by eliminating particular subpopulations of bacteria, control the composition of bacterial biofilm communities (Figure 1) [1–3]. Temperate phages can infect and persist in bacteria, a state called lysogeny [4]. As inhabitants, lysogenic phages drive bacterial genome evolution via the introduction of viral genes that endow the hosts with new capabilities or that regulate host biochemical or signaling pathways. For instance, some cyanophages encode photosystem components that enhance host light harvesting ability [5,6]. In *Shigella flexneri*, phage-encoded enzymes modify O-antigen sugars enabling serotype conversion [7]. Similarly, phage enzymes hydrolyze the polysaccharide in the *Acinetobacter baumannii* capsule, altering biofilm formation [8]. Underscoring their importance to human health, temperate phages supply their bacterial hosts with the toxin-encoding genes responsible for diseases including cholera (caused by phage CTX of *Vibrio cholerae*), dysentery (caused by STX phages of *Escherichia coli*), diphtheria (caused by phage Beta of *Corynebacterium diphtheriae*), and botulism (caused by neurotoxin-encoding phages of *Clostridium botulinum*) [9–12]. These and other examples highlight fascinating connections between phage biology and bacterial biology. In some cases, phage infection confers benefits to the host bacterium, for example, enhancing colonization or dissemination from eukaryotic hosts. Phages are also frequently bacterial parasites and, consequently, bacteria are under the pervasive threat of infection by phages that can exploit resources for continued propagation and, moreover, that can kill host bacteria in response to particular conditions.

Following host infection, temperate phages, undertake one of two lifestyle programs (Figure 1) [13,14]. They can enter the lytic cycle in which the phage uses host resources to replicate and

package its genome into viral particles. The viral particles are subsequently released, killing the host cell and promoting phage dissemination to new host cells. Alternatively, temperate phages can enter into lysogeny and exist as prophages [13,14]. Commonly, the phage integrates its DNA at a discrete site in the host bacterial genome. Less commonly, the phage remains as an extra-chromosomal element in a plasmid-like state [15,16]. In both cases, prophage replication during host cell division ensures transmission to progeny. Phages that enact lysogeny typically convert to the lytic route when the host bacterium experiences stress. The canonical trigger for the lysogeny-to-lysis transition is activation of the bacterial SOS response following host DNA damage [13,14,17]. The notion is that by tuning into host SOS, the phage connects its lifestyle decision-making to host vitality. Phages thereby “abandon” their current hosts when host long-term survivability becomes uncertain.

Advances in high-throughput culturing, metagenomic sequencing, and genome assembly techniques are revealing new intricacies concerning the nature of phage-host and phage-phage interactions in real-world settings [18–23]. For example, most bacteria are predicted to harbor more than one prophage, a condition called polylysogeny [24–26]. Moreover, a growing number of phages isolated from different niches cannot be induced in the laboratory using standard induction conditions [27,28]. The majority of phage genes have no known functions [29], suggesting that, perhaps, some of these genes encode novel induction pathway components that enable multiple prophages to co-exist and compete with one another. In this Pearl, we highlight newly discovered mechanisms underlying phage-phage competition with a focus on prophage induction.

THE CANONICAL PROPHAGE INDUCTION CUE: DNA DAMAGE

Extensive knowledge exists concerning molecular mechanisms underlying temperate lambdoid phages. Typically, entry into the lytic or lysogenic cycle is controlled by the master repressor of lysis, called *cl* [13]. *cl*-type proteins bind particular operator sequences in phage genomes where they function to repress transcription of lytic genes, driving lysogeny. Following host SOS induction, *cl*-type proteins are inactivated either by proteolysis, mediated by the host RecA protein, or by antirepression conferred by another phage protein called an antirepressor [14,30,31]. Consequently, lytic genes are derepressed, launch of the lytic cascade occurs, and the bacterial host cell is killed.

Activation of the host SOS response is regarded as a “universal” inducer of prophages. Agents commonly used in the laboratory to drive prophage induction include DNA-damaging antibiotics and modulators of reactive oxygen species [32]. The discovery that most bacteria are polylysogens, coupled with the dogma that prophages are induced by host SOS, suggests that once the host cell is stressed, competition among phages would occur for appropriation of the host cell resources required for propagation [33,34]. Consequently, differences in replication rates, packaging rates, burst size, and other intrinsic phage properties that aid in the monopolization of host cell resources should dictate each phage’s success in producing progeny virions (Figure 2). Indeed, experiments show that only one temperate phage is predominantly recovered following SOS induction of a polylysogen, presumably the winner of the phage-phage

competition [33,34]. Likewise in *E. coli*, in cases in which two lambdoid phages could be detected following induction, overall virion production was lower in the polylysogen than in a monolysogen [34]. Specifically, one phage exhibited its baseline productivity level while virion production by the other phage declined, or there was a loss in productivity of both phages [34]. These findings suggest that the host resources required for viral reproduction are limiting and/or interactions between prophage lytic programs cause mutual interference in the harnessing of host resources.

NON-CANONICAL PROPHAGE INDUCTION CUES

A limitation of the above phage competition studies is that commonly used DNA-damaging agents fail to induce all prophages in the laboratory. Thus, DNA-damaging agents may not reflect the authentic cues that drive prophage induction in natural environments. Moreover, bacteria encounter environmental stressors that do not invoke the SOS response, suggesting that if prophage induction occurs under such conditions, induction must be triggered by SOS-independent cues. In support of this notion, accumulating evidence indicates that prophages have evolved to selectively tune into SOS-independent facets of host physiology, however, we note that such cues are generally orders of magnitude less potent at inducing the lysogeny-to-lysis transition than are SOS-dependent inducers. For example, salt stress and the intracellular ionic environment can bias phage λ^{imm434} toward lytic development, presumably because interactions between the λ^{imm434} *ci* repressor and its target operator DNA are sensitive to salt [35]. Administration of EDTA increases induction of lambdoid STX prophages [36]. The hypothesis is that divalent cation chelation disrupts the integrity of the bacterial outer membrane, which, by an unknown mechanism, functions as an inducing signal. Finally, overproduction of the RcsA or the DsrA small RNAs that control biofilm formation increases lambdoid prophage induction in *E. coli*

[37–39]. Low temperature is an environmental stressor known to induce RcsA and DsrA, so their artificial overproduction likely mimics this condition [38]. The molecular mechanisms connecting the above prophage inducers to alterations in phage gene expression are mostly unknown. Also unknown is whether the stresses/mechanisms are highly specific for a single prophage or a single species of bacteria or, like DNA damage, are general.

Recent explorations into exotic prophage induction cues reveal that prophages surveil diverse aspects of bacterial physiology beyond host stress [40–45]. As one illustration, prophages detect bacterial-produced extracellular quorum-sensing (QS) signal molecules called autoinducers (AI) [41–43]. QS is a chemical communication process bacteria use to synchronize collective behaviors [46]. Prophages harbor QS AI receptors, and detection of accumulated AIs launches the lytic cycle and release of viral particles. Thus, host killing occurs exclusively at high bacterial cell density when the probability of phage infection of another cell is maximized. For example, prophage ARM81Id of *Aeromonas* sp. ARM81 encodes a homolog of the bacterial LuxR-type QS receptor called LuxR_{ARM81Id} [41,47]. LuxR_{ARM81Id} detects its host-*Aeromonas*-produced C4-homoserine lactone (HSL) AI, and in response, activates expression of a gene encoding an antirepressor [48]. The antirepressor inactivates the ARM81Id *ci* repressor, thus launching the phage lytic cascade. ARM81Id is also SOS-inducible via proteolysis of its *ci* protein [48].

Aeromonas sp. ARM81 has recently been developed as a model polylysogenic bacterium as it harbors two prophages that employ distinct strategies for persistence. As noted, prophage ARM81Id is both SOS- and QS-inducible. The second *Aeromonas* sp. ARM81 prophage is called ARM81mr, and it is SOS- but not QS-inducible [48]. This arrangement makes it so that particular conditions could favor induction of one prophage over the other (Figure 2). Indeed, the ARM81mr

prophage exhibits higher levels of spontaneous induction than does the ARM81ld prophage, and following DNA-damage and SOS activation, higher levels of phage ARM81mr viral particles are released than are particles of phage ARM81ld [47,48]. By contrast, high levels of the C4-HSL QS AI induce the ARM81ld prophage but not the ARM81mr prophage, so almost exclusively phage ARM81ld viral particles are released [48]. Thus, whereas phage ARM81ld productivity is likely limited by competition with phage ARM81mr during SOS induction, selectively tuning into QS cues enables exclusive acquisition of host resources for phage ARM81ld propagation. In a fascinating twist, non-cognate HSL AIs produced by non-*Aeromonas* bacteria repress $\text{LuxR}_{\text{ARM81ld}}$ activity, thereby dampening ARM81ld prophage induction [43]. This feature could guard against premature launch of the prophage ARM81ld lytic cascade when the probability of encountering a suitable host for infection is low. Thus, particular environmental conditions apparently drive the outcomes of inter-prophage competition in polylysogens.

CONCLUDING REMARKS

Here, we considered inter-prophage competitive interactions during prophage induction in polylysogenic bacteria. Beyond the well-characterized SOS induction pathway, new studies are uncovering varied cues and physiological conditions that trigger prophage lytic programs and viral dissemination. Presumably, competition among prophages, together with relevant environmental cues, drive the evolution of their induction strategies. As results in this developing field of study continue to unfold, defining the molecular underpinnings of inter-prophage interactions, either competitive or cooperative, could be key to understanding how multiple prophages alter host biology in polylysogenic bacteria [49–51]. While not covered in this Pearl, cooperativity among prophages and prophage-like elements occurs, and indeed, can underpin host pathogenicity [31,52–54]. For instance, in *Staphylococcus aureus*, prophage-like mobile pathogenicity islands

are repressed by a *ci*-type protein [55]. Their induction, and consequently, toxin production, occurs in response to co-residing helper prophages that supply the needed antirepressor. Curiously, polylysogeny is widespread in bacterial pathogens suggesting that the pathogenic lifestyle could select for acquisition and/or retention of multiple prophages. Given that virulence determinants are frequently encoded within prophages and can be produced during prophage induction, cross-regulatory prophage interactions could become promising targets for therapeutic intervention in clinically-important bacterial pathogens.

REFERENCES

1. Pires DP, Melo LDR, Azeredo J. Understanding the Complex Phage-Host Interactions in Biofilm Communities. *Annual Review of Virology*. 2021;8: 73–94. doi:10.1146/annurev-virology-091919-074222
2. Bond MC, Vidakovic L, Singh PK, Drescher K, Nadell CD. Matrix-trapped viruses can prevent invasion of bacterial biofilms by colonizing cells. Shou W, Storz G, Shou W, editors. *eLife*. 2021;10: e65355. doi:10.7554/eLife.65355
3. Brüssow H, Hendrix RW. Phage Genomics: Small Is Beautiful. *Cell*. 2002;108: 13–16. doi:10.1016/S0092-8674(01)00637-7
4. Lwoff A. Lysogeny. *Bacteriol Rev*. 1953;17: 269–337.
5. Mann NH, Cook A, Millard A, Bailey S, Clokie M. Bacterial photosynthesis genes in a virus. *Nature*. 2003;424: 741–741. doi:10.1038/424741a
6. Frank JA, Lorimer D, Youle M, Witte P, Craig T, Abendroth J, et al. Structure and function of a cyanophage-encoded peptide deformylase. *ISME J*. 2013;7: 1150–1160. doi:10.1038/ismej.2013.4
7. Allison GE, Verma NK. Serotype-converting bacteriophages and O-antigen modification in *Shigella flexneri*. *Trends in Microbiology*. 2000;8: 17–23. doi:10.1016/S0966-842X(99)01646-7
8. Shahed-Al-Mahmud Md, Roy R, Sugiokto FG, Islam MdN, Lin M-D, Lin L-C, et al. Phage ϕ AB6-Borne Depolymerase Combats *Acinetobacter baumannii* Biofilm Formation and Infection. *Antibiotics (Basel)*. 2021;10: 279. doi:10.3390/antibiotics10030279
9. Waldor MK, Mekalanos JJ. Lysogenic Conversion by a Filamentous Phage Encoding Cholera Toxin. *Science*. 1996;272: 1910–1914. doi:10.1126/science.272.5270.1910
10. O'Brien AD, Newland JW, Miller SF, Holmes RK, Smith HW, Formal SB. Shiga-Like Toxin-Converting Phages from *Escherichia coli* Strains That Cause Hemorrhagic Colitis or Infantile Diarrhea. *Science*. 1984;226: 694–696. doi:10.1126/science.6387911
11. Groman NB. Conversion by corynebacteriophages and its role in the natural history of diphtheria. *J Hyg (Lond)*. 1984;93: 405–417.
12. Hauser D, Eklund MW, Kurazono H, Binz T, Niemann H, Gill DM, et al. Nucleotide sequence of *Clostridium botulinum* C1 neurotoxin. *Nucleic Acids Res*. 1990;18: 4924.

13. Ptashne M. A Genetic Switch, Third Edition: Phage Lambda Revisited. 3rd edition. Cold Spring Harbor, N.Y: Cold Spring Harbor Laboratory Press; 2004.
14. Ofir G, Sorek R. Contemporary Phage Biology: From Classic Models to New Insights. *Cell*. 2018;172: 1260–1270. doi:10.1016/j.cell.2017.10.045
15. Ravin NV. N15: The linear phage-plasmid. *Plasmid*. 2011;65: 102–109. doi:10.1016/j.plasmid.2010.12.004
16. Łobocka MB, Rose DJ, Plunkett G, Rusin M, Samojedny A, Lehnher H, et al. Genome of Bacteriophage P1. *J Bacteriol*. 2004;186: 7032–7068. doi:10.1128/JB.186.21.7032-7068.2004
17. Little JW. Autodigestion of *lexA* and phage lambda repressors. *PNAS*. 1984;81: 1375–1379. doi:10.1073/pnas.81.5.1375
18. Clooney AG, Sutton TDS, Shkoporov AN, Holohan RK, Daly KM, O'Regan O, et al. Whole-Virome Analysis Sheds Light on Viral Dark Matter in Inflammatory Bowel Disease. *Cell Host & Microbe*. 2019;26: 764-778.e5. doi:10.1016/j.chom.2019.10.009
19. Hendrix RW. Bacteriophages: Evolution of the Majority. *Theoretical Population Biology*. 2002;61: 471–480. doi:10.1006/tpbi.2002.1590
20. Marsh P, Wellington EMH. Phage-host interactions in soil. *FEMS Microbiology Ecology*. 1994;15: 99–107. doi:10.1111/j.1574-6941.1994.tb00234.x
21. de Jonge PA, Wortelboer K, Scheithauer TPM, van den Born B-JH, Zwinderman AH, Nobrega FL, et al. Gut virome profiling identifies a widespread bacteriophage family associated with metabolic syndrome. *Nat Commun*. 2022;13: 3594. doi:10.1038/s41467-022-31390-5
22. Fremin BJ, Bhatt AS, Kyrpides NC, Sengupta A, Sczyrba A, Maria da Silva A, et al. Thousands of small, novel genes predicted in global phage genomes. *Cell Reports*. 2022;39: 110984. doi:10.1016/j.celrep.2022.110984
23. Nayfach S, Pérez-Espino D, Call L, Low SJ, Sberro H, Ivanova NN, et al. Metagenomic compendium of 189,680 DNA viruses from the human gut microbiome. *Nat Microbiol*. 2021;6: 960–970. doi:10.1038/s41564-021-00928-6
24. Hendrix RW, Smith MCM, Burns RN, Ford ME, Hatfull GF. Evolutionary relationships among diverse bacteriophages and prophages: All the world's a phage. *PNAS*. 1999;96: 2192–2197. doi:10.1073/pnas.96.5.2192
25. Howard-Varona C, Hargreaves KR, Abedon ST, Sullivan MB. Lysogeny in nature: mechanisms, impact and ecology of temperate phages. *ISME J*. 2017;11: 1511–1520. doi:10.1038/ismej.2017.16
26. Chen F, Wang K, Stewart J, Belas R. Induction of Multiple Prophages from a Marine Bacterium: a Genomic Approach. *Appl Environ Microbiol*. 2006;72: 4995–5001. doi:10.1128/AEM.00056-06
27. Campbell DE, Ly LK, Ridlon JM, Hsiao A, Whitaker RJ, Degnan PH. Infection with Bacteroides Phage BV01 Alters the Host Transcriptome and Bile Acid Metabolism in a Common Human Gut Microbe. *Cell Reports*. 2020;32: 108142. doi:10.1016/j.celrep.2020.108142
28. Cornuault JK, Petit M-A, Mariadassou M, Benevides L, Moncaut E, Langella P, et al. Phages infecting *Faecalibacterium prausnitzii* belong to novel viral genera that help to decipher intestinal viromes. *Microbiome*. 2018;6. doi:10.1186/s40168-018-0452-1
29. Hatfull GF, Hendrix RW. Bacteriophages and their Genomes. *Curr Opin Virol*. 2011;1: 298–303. doi:10.1016/j.coviro.2011.06.009
30. Kim M, Kim HJ, Son SH, Yoon HJ, Lim Y, Lee JW, et al. Noncanonical DNA-binding mode of repressor and its disassembly by antirepressor. *PNAS*. 2016;113: E2480–E2488. doi:10.1073/pnas.1602618113

31. Lemire S, Figueroa-Bossi N, Bossi L. Bacteriophage Crosstalk: Coordination of Prophage Induction by Trans-Acting Antirepressors. *PLOS Genetics*. 2011;7: e1002149. doi:10.1371/journal.pgen.1002149
32. DeMarini DM, Lawrence BK. Prophage induction by DNA topoisomerase II poisons and reactive-oxygen species: Role of DNA breaks. *Mutation Research/Fundamental and Molecular Mechanisms of Mutagenesis*. 1992;267: 1–17. doi:10.1016/0027-5107(92)90106-C
33. Guo Q, Chen B, Tu Y, Du S, Chen X. Prophage LambdaSo uses replication interference to suppress reproduction of coexisting temperate phage MuSo2 in *Shewanella oneidensis* MR-1. *Environ Microbiol*. 2019;21: 2079–2094. doi:10.1111/1462-2920.14592
34. Refardt D. Within-host competition determines reproductive success of temperate bacteriophages. *ISME J*. 2011;5: 1451–1460. doi:10.1038/ismej.2011.30
35. Shkilnyj P, Koudelka GB. Effect of Salt Shock on Stability of λ imm434 Lysogens. *J Bacteriol*. 2007;189: 3115–3123. doi:10.1128/JB.01857-06
36. Imamovic L, Muniesa M. Characterizing RecA-independent induction of Shiga toxin2-encoding phages by EDTA treatment. *PLoS One*. 2012;7: e32393. doi:10.1371/journal.pone.0032393
37. Rozanov DV, D'Ari R, Sineoky SP. RecA-Independent Pathways of Lambdoid Prophage Induction in *Escherichia coli*. *Journal of Bacteriology*. 1998;180: 6306–6315. doi:10.1128/JB.180.23.6306-6315.1998
38. Sledjeski DD, Gupta A, Gottesman S. The small RNA, DsrA, is essential for the low temperature expression of RpoS during exponential growth in *Escherichia coli*. *The EMBO Journal*. 1996;15: 3993–4000. doi:10.1002/j.1460-2075.1996.tb00773.x
39. Repoila F, Gottesman S. Signal Transduction Cascade for Regulation of RpoS: Temperature Regulation of DsrA. *J Bacteriol*. 2001;183: 4012–4023. doi:10.1128/JB.183.13.4012-4023.2001
40. Erez Z, Steinberger-Levy I, Shamir M, Doron S, Stokar-Avihail A, Peleg Y, et al. Communication between viruses guides lysis–lysogeny decisions. *Nature*. 2017;541: 488–493. doi:10.1038/nature21049
41. Silpe JE, Bassler BL. Phage-Encoded LuxR-Type Receptors Responsive to Host-Produced Bacterial Quorum-Sensing Autoinducers. *mBio*. 2019;10. doi:10.1128/mBio.00638-19
42. Silpe JE, Bassler BL. A Host-Produced Quorum-Sensing Autoinducer Controls a Phage Lysis-Lysogeny Decision. *Cell*. 2019;176: 268–280.e13. doi:10.1016/j.cell.2018.10.059
43. Silpe JE, Duddy OP, Bassler BL. Natural and synthetic inhibitors of a phage-encoded quorum-sensing receptor affect phage–host dynamics in mixed bacterial communities. *Proceedings of the National Academy of Sciences*. 2022;119: e2217813119. doi:10.1073/pnas.2217813119
44. Laganenka L, Sander T, Lagonenko A, Chen Y, Link H, Sourjik V. Quorum Sensing and Metabolic State of the Host Control Lysogeny-Lysis Switch of Bacteriophage T1. *mBio*. 2019;10. doi:10.1128/mBio.01884-19
45. Jancheva M, Böttcher T. A Metabolite of *Pseudomonas* Triggers Prophage-Selective Lysogenic to Lytic Conversion in *Staphylococcus aureus*. *J Am Chem Soc*. 2021;143: 8344–8351. doi:10.1021/jacs.1c01275
46. Papenfort K, Bassler BL. Quorum sensing signal–response systems in Gram-negative bacteria. *Nature Reviews Microbiology*. 2016;14: 576–588. doi:10.1038/nrmicro.2016.89
47. Dziewit L, Radlinska M. Two novel temperate bacteriophages co-existing in *Aeromonas* sp. ARM81 – characterization of their genomes, proteomes and DNA methyltransferases. *Journal of General Virology*. 2016;97: 2008–2022. doi:10.1099/jgv.0.000504
48. Silpe JE, Duddy OP, Hussain FA, Forsberg KJ, Bassler BL. Small protein modules dictate prophage fates during polylysogeny. *bioRxiv*; 2022. p. 2022.09.16.508337. doi:10.1101/2022.09.16.508337

49. Fillol-Salom A, Rostøl JT, Ojiogu AD, Chen J, Douce G, Humphrey S, et al. Bacteriophages benefit from mobilizing pathogenicity islands encoding immune systems against competitors. *Cell*. 2022;185: 3248-3262.e20. doi:10.1016/j.cell.2022.07.014
50. Argov T, Sapir SR, Pasechnek A, Azulay G, Stadnyuk O, Rabinovich L, et al. Coordination of cohabiting phage elements supports bacteria–phage cooperation. *Nat Commun*. 2019;10: 5288. doi:10.1038/s41467-019-13296-x
51. Azulay G, Pasechnek A, Stadnyuk O, Ran-Sapir S, Fleisacher AM, Borovok I, et al. A dual-function phage regulator controls the response of cohabiting phage elements via regulation of the bacterial SOS response. *Cell Reports*. 2022;39: 110723. doi:10.1016/j.celrep.2022.110723
52. Tormo-Más MÁ, Mir I, Shrestha A, Tallent SM, Campoy S, Lasa Í, et al. Moonlighting bacteriophage proteins derepress staphylococcal pathogenicity islands. *Nature*. 2010;465: 779–782. doi:10.1038/nature09065
53. Miguel-Romero L, Alqasmi M, Bacarizo J, Tan JA, Cogdell RJ, Chen J, et al. Non-canonical *Staphylococcus aureus* pathogenicity island repression. *Nucleic Acids Research*. 2022;50: 11109–11127. doi:10.1093/nar/gkac855
54. Alqurainy N, Miguel-Romero L, Moura de Sousa J, Chen J, Rocha EPC, Fillol-Salom A, et al. A widespread family of phage-inducible chromosomal islands only steals bacteriophage tails to spread in nature. *Cell Host & Microbe*. 2023;31: 69-82.e5. doi:10.1016/j.chom.2022.12.001
55. Haag AF, Podkowik M, Ibarra-Chávez R, Gallego del Sol F, Ram G, Chen J, et al. A regulatory cascade controls *Staphylococcus aureus* pathogenicity island activation. *Nat Microbiol*. 2021;6: 1300–1308. doi:10.1038/s41564-021-00956-2

FUNDING

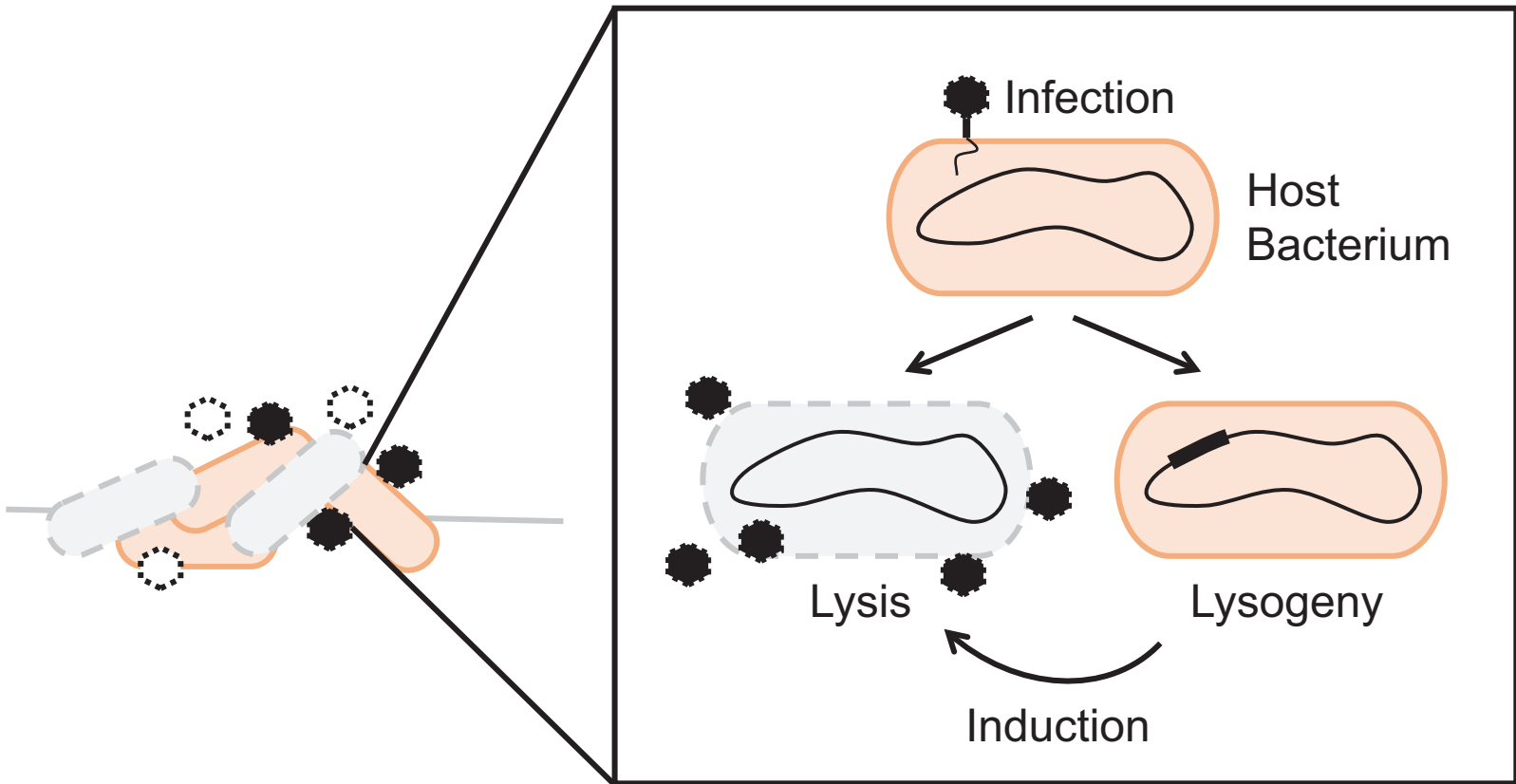
The authors acknowledge financial support from the Jane Coffin Childs Memorial Fund for Medical Research (J.E.S), Howard Hughes Medical Institute (J.E.S and B.L.B), National Science Foundation grant MCB-2043238 (B.L.B.), the National Institutes of Health grant R37GM065859 (B.L.B.), and NIGMS training grant T32GM007388 (O.P.D.). The funders had no role in study design, data collection and analysis, decision to publish, or preparation of the manuscript.

AUTHOR CONTRIBUTIONS

All authors contributed to the conceptualization, supervision, and writing (original draft preparation and review & editing) of this piece.

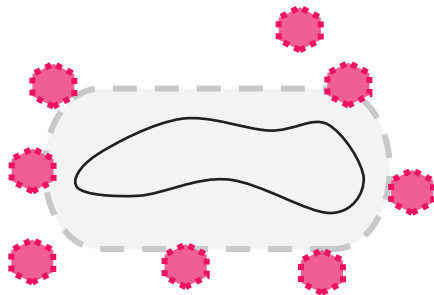
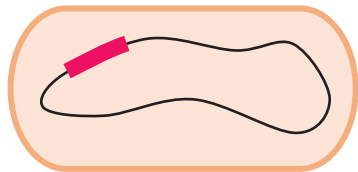
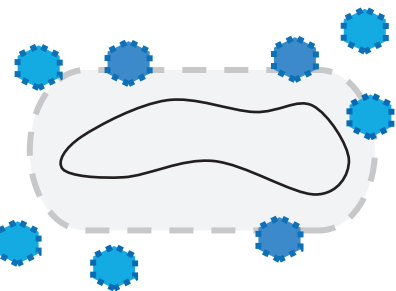
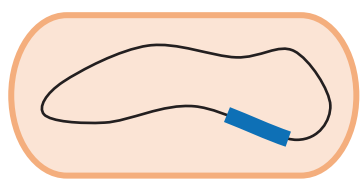
COMPETING INTERESTS

The authors have declared that no competing interests exist.

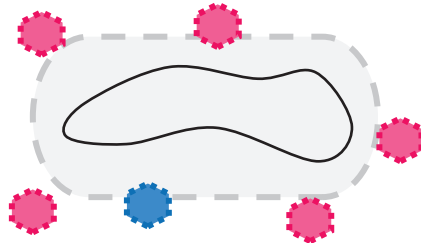
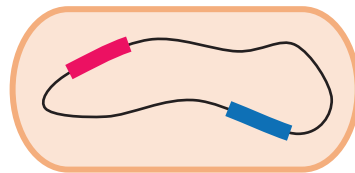


Canonical Prophage Induction Cue (SOS)

Monolysogens



Polylysogen



Non-canonical Induction Cue

Polylysogen

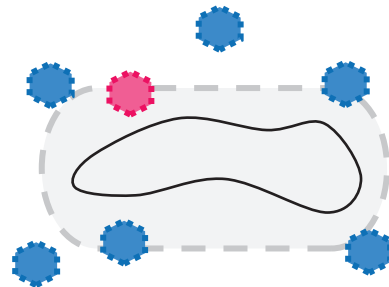
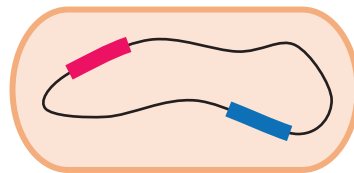


FIGURE LEGENDS

Figure 1: Prophage induction drives changes in bacterial communities.

The cartoon shows possible effects of phage activity on a bacterial community. Black and white phage particles represent different phages present in the community. Orange and gray cells represent live and dead bacteria, respectively, the latter shown with hash symbols to denote disrupted membranes. Inset: The lysis-lysogeny switch is a phage-encoded regulatory decision-making program. Lysis leads to killing of the host bacterium and release of new phage particles. Lysogeny allows the phage to persist in the host cell and be passed down to progeny. The thick black region in the genome of the lysogenized bacterium represents the integrated phage genome. We note that, while not a main topic of this piece, phage infection in bacterial biofilms could be particularly interesting. Biofilms are surface-bound bacterial communities held together by an extracellular matrix. Biofilms can be composed of one or more bacterial species. Studies of prophage induction in bacterial biofilms show that, depending on the host ranges of the resident phages and the species composition of the bacteria present, induction of one prophage may kill the entire community or may target and eliminate specific bacterial species within the biofilm.

Figure 2. Phage productivity depends on the presence or absence of co-inhabiting prophages and the mechanism of induction.

Left: Two different prophages (blue and pink segments of DNA in the host genomes) inhabiting different bacterial cells (designated monolysogens). Viral particles may be produced in equal quantities (blue and pink particles) following induction by the canonical SOS trigger. Middle: If the two prophages inhabit the same bacterium (designated polylysogen), SOS induction leads to inter-phage competition for host resources. Consequently, the phage that “wins” the competition (pink phage in the cartoon) will produce more virions than the other co-resident phage (blue phage in the cartoon). Particles of the most competitive (pink) phage will primarily be released to spread within the bacterial community at the expense of the less-competitive (blue) phage. Nonetheless, overall virion production is lowered in the polylysogen compared to the monolysogen. Right: In polylysogens, prophages that encode non-canonical induction pathways controlling their lysis-lysogeny programs (designated non-canonical induction cue) can outcompete co-residing prophages. This outcome occurs when the non-canonical induction cue is encountered. Only the phage that is responsive to the non-canonical induction cue (blue phage in the cartoon) replicates. In the examples in the figure, the pink phage “wins” the competition when the inducer is the canonical SOS cue and the blue phage “wins” the competition when the inducer is the non-canonical cue. Thus, the particular induction cue dictates the outcome of the phage-phage competition.