Faure, G., Shmakov, S.A., Yan, W.X., Cheng, D.R., Scott, D.A., Peters, J.E., Makarova, K.S., and Koonin, E.V. (2019). CRISPR-Cas in mobile genetic elements: counter-defence and beyond. Nat. Rev. Microbiol. *17*, 513–525.

Fujii, S., and Fuchs, R.P. (2020). A Comprehensive View of Translesion Synthesis in Escherichia coli. Microbiol. Mol. Biol. Rev. *84*, e00002–20.

Hossain, A.A., McGinn, J., Meeske, A.J., Modell, J.W., and Marraffini, L.A. (2021). Viral recombination systems limit CRISPR-Cas targeting through the generation of escape mutations. Cell Host Microbe *29*, 1482–1495.

Koonin, E.V., Makarova, K.S., and Zhang, F. (2017). Diversity, classification and evolution of CRISPR-Cas systems. Curr. Opin. Microbiol. *37*, 67–78.

Lopes, A., Amarir-Bouhram, J., Faure, G., Petit, M.A., and Guerois, R. (2010). Detection of novel recombinases in bacteriophage genomes unveils Rad52, Rad51 and Gp2.5 remote homologs. Nucleic Acids Res. *38*, 3952–3962.

Myers, R.S., and Stahl, F.W. (1994). Chi and the RecBC D enzyme of Escherichia coli. Annu. Rev. Genet. *28*, 49–70.

Pickar-Oliver, A., and Gersbach, C.A. (2019). The next generation of CRISPR-Cas technologies and applications. Nat. Rev. Mol. Cell Biol. *20*, 490–507.

Poteete, A.R. (2001). What makes the bacteriophage lambda Red system useful for genetic engineering: molecular mechanism and biological function. FEMS Microbiol. Lett. *201*, 9–14.

Thomason, L.C., Sawitzke, J.A., Li, X., Costantino, N., and Court, D.L. (2014). Recombineering: genetic engineering in bacteria using homologous recombination. Curr. Protoc. Mol. Biol. *106*, 1.16.1–39.

Stuck on you: Bacterial-auxin-mediated bacterial colonization of plant roots

Jing-Mei Qian[1,2,3,4] and Yang Bai[1,2,3,4,*]
[1]State Key Laboratory of Plant Genomics, Institute of Genetics and Developmental Biology, Innovation Academy for Seed Design, Chinese Academy of Sciences, 100101 Beijing, China
[2]CAS Center for Excellence in Biotic Interactions, College of Advanced Agricultural Sciences, University of Chinese Academy of Sciences, 100049 Beijing, China
[3]CAS-JIC Centre of Excellence for Plant and Microbial Science, Institute of Genetics and Developmental Biology, Chinese Academy of Sciences, 100101 Beijing, China
[4]College of Advanced Agricultural Sciences, University of Chinese Academy of Sciences, 100049 Beijing, China
*Correspondence: ybai@genetics.ac.cn
https://doi.org/10.1016/j.chom.2021.09.014

Auxin secreted by root-associated bacteria promotes plant growth, yet benefits to bacteria themselves are ill-defined. In this issue of *Cell Host & Microbe*, Tzipilevich et al. (2021) demonstrate that auxin and plant EFR-triggered response are essential for root colonization of *B. velezensis*, indicating potential co-evolution of plants and root commensal bacteria.

Plant roots are colonized by diverse commensal bacteria, a process vital to plant growth and development. Many basic scientific questions are behind the interactions between these bacteria and host plants. For half a century, it has been evolutionarily puzzling that a significant proportion of root-associated bacteria synthesize plant hormones (e.g., auxin) that promote plant growth (Fuentes-Ramirez et al., 1993; Kaper and Veldstra, 1958; Spaepen et al., 2007). According to the theory of adaptive evolution, bacteria should accumulate genes that benefit themselves—not their host plants. The benefits of bacterial auxin to the root-dwelling bacteria that produce this hormone are unknown.

In this issue of *Cell Host and Microbe*, Tzipilevich et al. (2021) provide insights into the role and genetic mechanism of bacterial auxin in the colonization of commensal bacteria on plant roots. The authors studied the interaction between *Arabidopsis thaliana* and *Bacillus velezensis* FZB42, a model gram-positive soil bacterium that synthesizes auxin and stimulates lateral root formation and plant biomass accumulation. Tzipilevich et al. (2021) discovered that bacterial auxin is necessary and sufficient for *B. velezensis* colonization on *Arabidopsis* roots by helping bacteria tolerate the toxic effects of plant reactive oxygen species (ROS) (Figure 1). Further, they found that plant EFR-triggered response and bacterial flagellum production are essential for the adhesion of the bacteria to plant roots and protect plants against infection of the fungal pathogen *Rhizoctonia solani*. Finally, the authors showed that auxin-induced colonization is a bacterium-specific phenomenon by examining four other auxin-secreting bacteria.

To explore the role of bacterial auxin in root colonization, Tzipilevich et al. (2021) inoculated *Arabidopsis* seedlings with wild-type *B. velezensis* and *ΔysnE*, a mutant strain deficient in auxin production. The *ΔysnE* strain failed to proliferate and adhere on *Arabidopsis* roots. Exogenous auxin restored the colonization defect of the Δ*ysnE* mutant, suggesting that bacterial auxin is necessary and sufficient for the colonization of *B. velezensis* on *Arabidopsis* roots. The authors hypothesized that bacterial auxin is needed for this bacterium to antagonize plant immune response. They found that plant immune response mediated by the receptor EFR restricted the colonization of the auxin-deficient bacterial mutant *ΔysnE*. Mutation of *EFR* restored the

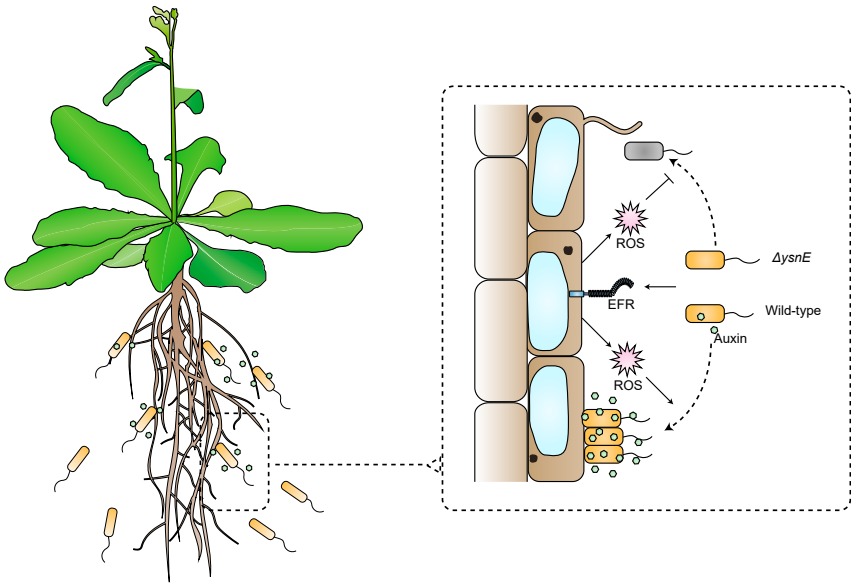

Figure 1. Diagram of interactions between auxin-producing bacteria and EFR-triggered plant immune response

Wild-type bacteria and *ΔysnE*, a mutant strain deficient in auxin (green hexagons) production, are recognized by plant immune receptor EFR. Then EFR stimulates plant ROS production. ROS trigger the accumulation of bioactive auxin in wild-type bacteria while restricting the proliferation of *ΔysnE* mutants without bacterial auxin. Bacterial auxin and the plant EFR-triggered response are essential for the colonization of *B. velezensis* on *Arabidopsis* roots.

proliferation defect of *ΔysnE*, suggesting that bacterial auxin plays an important role in overcoming the restriction effect of the plant immune system on bacterial root colonization.

To identify the components of the plant immune system that restrict the proliferation of *B. velezensis ΔysnE*, the authors examined colony counts of *ΔysnE* bacteria associated with roots in several *Arabidopsis* mutants with deficient immune responses. The proliferation defect of the *ΔysnE* mutant strain was restored on the roots of *rbohd rbohf* double-mutant plants, which are defective in immune-triggered ROS production. ROS are important compounds involved in plant defense responses against invading pathogens (Fones and Preston, 2012). Tzipilevich et al. (2021) treated wild-type and *ΔysnE B. velezensis* strains with superoxide (O_2^-) *in vitro* and found that O_2^- was less toxic to wild-type bacteria than to the *ΔysnE* mutant. Treatment with the exogenous auxin increased the survival of wild-type and mutant strains, suggesting that bacterial auxin is necessary and sufficient to help bacteria tolerate the toxic effects of plant ROS.

Tzipilevich et al. (2021) then compared gene expression changes in wild-type and *ΔysnE* strains after O_2^- treatment. Genes involved in the SOS response, DNA repair, and iron homeostasis were specifically upregulated in the wild-type bacteria compared to *ΔysnE*. The expression of these genes in *ΔysnE* bacteria significantly enhanced bacterial proliferation on roots, indicating that these genes function downstream of bacterial auxin. Notably, ROS induced the accumulation of bacterial auxin, as ROS treatment enhanced the fluorescence of YsnE-GFP in bacteria. In addition, a supernatant prepared from ROS-treated *B. velezensis* showed greater induction of fluorescence in DR5::GFP *Arabidopsis* plants than the untreated control, demonstrating that ROS trigger the efficient production of bacterial auxin with bioactivity.

Tzipilevich et al. (2021) also investigated the adhesion of bacteria to *Arabidopsis* roots in response to EFR-triggered immune response. Although the proliferation defect of *ΔysnE* bacteria was restored on *efr2* roots, these bacteria failed to efficiently adhere to *efr2* roots. Wild-type bacteria also showed less efficient adhesion to *efr2* roots than to wild-type Col-0 plants, suggesting that EFR-triggered response is essential for inducing bacterial adhesion. Interestingly, exogenous auxin treatment promoted the adhesion of wild-type bacteria to the roots of both Col-0 and *efr2* plants, suggesting that bacterial auxin induces both the proliferation and adhesion of bacteria in the plant root environment. Tzipilevich et al. (2021) then examined a series of colonization-related bacterial mutants impaired in motility, adhesion, and biofilm formation under exogenous auxin treatment. Auxin treatment failed to restore the root adhesion of the flagellum-production-deficient mutants *Δhag* and *ΔswrA*, but not the flagellum-motility-deficient mutant *ΔmotA*, suggesting that flagellar production, but not flagellar movement, is involved in auxin-induced bacterial adhesion to plant roots.

Next, Tzipilevich et al. (2021) investigated the benefits of auxin-induced colonization on bacteria and plants. They co-inoculated *B. velezensis* with *P. polymyxa* or *Arthrobacter* MF161 onto the roots of Col-0 and *efr2 Arabidopsis* plants. *B. velezensis* outcompeted the growth of *P. polymyxa* on Col-0 roots in the elongation and maturation zones, while *P. polymyxa* outcompeted the growth of *B. velezensis* on roots of the *efr2* mutant, suggesting that EFR-triggered response enhanced the colonization of *B. velezensis* competing with other root commensal bacteria in the same niche. Notably, the root colonization enhanced by EFR-triggered response increased the ability of *B. velezensis* to protect plants against pathogen infection. The authors reduced the bacterial root colonization using plant *efr2* and bacterial *Δhag* mutants and checked the effect of decreased colonization of *B. velezensis* on its protective function against infection of root fungal pathogen *Rhizoctonia solani*. They found that wild-type *B. velezensis* protected Col-0 plants better than the *efr2* mutant and that the bacterial *Δhag* mutant strain showed less protective function on Col-0 plants, suggesting that enhanced adhesion and colony formation help *B. velezensis* protect plants from fungal infection.

Tzipilevich et al. (2021) also examined the roles of bacterial auxin in root colonization using four other auxin-secreting bacteria: *P. polymyxa*, *Arthrobacter* MF161, *Pseudomonas* species 65, and WCS374. The root colonization of *P. polymyxa* and

CellPress

Arthrobacter MF161 was enhanced by exogenous auxin, whereas *Pseudomonas* species 65 and WCS374 showed no response to the treatment, suggesting that auxin-induced colonization is a bacterium-specific phenomenon.

This important study uncovers a mechanism for the interplay between auxin-secreting bacteria and the plant immune system. The authors demonstrated that bacterial auxin is essential for root colonization of *B. velezensis* and its survival under EFR-triggered response, revealing benefits of bacterial auxin to the bacteria themselves. The observation that auxin induced bacterial colonization on roots is consistent with previous findings showing that auxin-overproducing *S. meliloti* enhanced nodulation in *Medicago* species and that *P. syringae* strain DC3000 showed higher bacterial loads on *Arabidopsis* leaves than bacterial mutants deficient in auxin production (McClerklin et al., 2018; Pii et al., 2007). The regulation of auxin concentrations in roots by host plants and commensal bacteria is vital for plant growth and development (Dinneny and Benfey, 2008; Finkel et al., 2020). Enhanced colonization by EFR-triggered response helps *B. velezensis* compete with other bacteria in the same niche and protects plants from fungal infection, indicating the potential co-evolution of plants and root commensal bacteria.

ACKNOWLEDGMENTS

This work was supported by grants from the Strategic Priority Research Program of the Chinese Academy of Sciences (XDA24020104 to Y.B.; XDA28030200 to J.-M.Q.), the Key Research Program of Frontier Sciences of the Chinese Academy of Sciences (QYZDB-SSW-SMC021 to Y.B.), and the National Natural Science Foundation of China (31772400 and 31761143017 to Y.B.).

REFERENCES

Dinneny, J.R., and Benfey, P.N. (2008). Plant stem cell niches: standing the test of time. Cell *132*, 553–557.

Finkel, O.M., Salas-González, I., Castrillo, G., Conway, J.M., Law, T.F., Teixeira, P.J.P.L., Wilson, E.D., Fitzpatrick, C.R., Jones, C.D., and Dangl, J.L. (2020). A single bacterial genus maintains root growth in a complex microbiome. Nature *587*, 103–108. https://doi.org/10.1038/s41586-020-2778-7.

Fones, H., and Preston, G.M. (2012). Reactive oxygen and oxidative stress tolerance in plant pathogenic Pseudomonas. FEMS Microbiol. Lett. *327*, 1–8. https://doi.org/10.1111/j.1574-6968.2011.02449.x.

Fuentes-Ramirez, L.E., Jimenez-Salgado, T., Abarca-Ocampo, I., and Caballero-Mellado, J. (1993). Acetobacter diazotrophicus, an indoleacetic acid producing bacterium isolated from sugarcane cultivars of Mexico. Plant Soil *154*, 145–150.

Kaper, J.M., and Veldstra, H. (1958). On the metabolism of tryptophan by Agrobacterium tumefaciens. Biochim. Biophys. Acta *30*, 401–420.

McClerklin, S.A., Lee, S.G., Harper, C.P., Nwumeh, R., Jez, J.M., and Kunkel, B.N. (2018). Indole-3-acetaldehyde dehydrogenase-dependent auxin synthesis contributes to virulence of Pseudomonas syringae strain DC3000. PLoS Pathog. *14*, e1006811. https://doi.org/10.1371/journal.ppat.1006811.

Pii, Y., Crimi, M., Cremonese, G., Spena, A., and Pandolfini, T. (2007). Auxin and nitric oxide control indeterminate nodule formation. BMC Plant Biol. *7*, 21. https://doi.org/10.1186/1471-2229-7-21.

Spaepen, S., Vanderleyden, J., and Remans, R. (2007). Indole-3-acetic acid in microbial and microorganism-plant signaling. FEMS Microbiol. Rev. *31*, 425–448. https://doi.org/10.1111/j.1574-6976.2007.00072.x.

Tzipilevich, E., Russ, D., Dangl, J.L., and Benfey, P.N. (2021). Plant immune system activation is necessary for efficient root colonization by auxin secreting beneficial bacteria. Cell Host Microbe *29*, 1507–1520.

Shigella shuts down the pyrop-technic show

Cristina Giogha[1,2] and Jaclyn S. Pearson[1,2,3,*]
[1]Centre for Innate Immunity and Infectious Diseases, Hudson Institute of Medical Research, Clayton, Victoria, Australia
[2]Department of Molecular and Translational Science, Monash University, Clayton, Victoria, Australia
[3]Department of Microbiology, Monash University, Clayton, Victoria, Australia
*Correspondence: jaclyn.pearson@hudson.org.au
https://doi.org/10.1016/j.chom.2021.09.012

***Shigella* is a highly infectious human pathogen, yet mice are naturally resistant to infection. In this issue of *Cell Host & Microbe*, Luchetti et al. (2021) discuss this species specificity, demonstrating that *Shigella* directly targets the pore-forming protein Gasdermin D for degradation, thus preventing pyroptosis to enable infection of human cells.**

Shigella species are the causative agents of bacillary dysentery, characterized by severe diarrhea that can be accompanied by blood and mucous. They impose an immense burden of disease on the global human population, causing ~270 million cases of shigellosis and 210,000 deaths annually (Khalil et al., 2018). Although *Shigella* has long been considered a major diarrheal pathogen of low-income countries, it has recently re-emerged as a sexually transmitted infection causing epidemics across high-income countries (Baker et al., 2018; Ingle et al., 2019). Of critical public health concern is the recent emergence of multidrug-resistant *Shigella*, leading to a situation in which there are now circulating strains that are entirely resistant to oral antibiotics (Ingle et al., 2019). As there is currently no vaccine for *Shigella*, there is an urgent need to comprehensively understand its pathogenesis in order to develop alternative therapeutics to combat the global spread of drug-resistant *Shigella*. Here, Luchetti et al. (2021) make an important contribution to the understanding of host manipulation by *Shigella* and how we can use mice to model human disease outcomes.