As is the nature of discovery, many questions remain. How do the co-housed CRON mice enable AMER mice to respond to the CRON diet? The most straightforward explanation is that the co-housed CRON mice simply complement individual microbes lost by long-term AMER exposure. Other explanations need to be considered, however. Perhaps these co-housed mice provide exposure to a more diverse microbial ecosystem that allows for re-emergence of "lost" microbes that remained at low levels, but dormant or suppressed, in the AMER microbiota. Differentiating between these potential mechanisms could help elucidate key principles underlying maintenance of gut microbial ecology and would have important implications for strategies to overcome persistent, diet-induced changes in the microbiota.

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The CAT(2) Comes Back

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Genetic and biochemical evidence supporting CATALASE2 as a salicylic acid (SA) receptor has finally emerged. In this issue of *Cell Host & Microbe*, Yuan et al. (2017) show that SA binds to CATALASE2 to inhibit auxin and jasmonic acid biosynthetic enzymes as a means to strengthen plant immunity against biotrophic pathogens.

Plants use contrasting defense strategies against biotrophic and necrotrophic pathogens because the former live off live host cells, whereas the latter kill the host cells to obtain nutrients. Salicylic acid (SA) plays a major role in the plant defense response against biotrophic pathogens, such as *Pseudomonas syringae pv. tomato* DC3000 (*Pst*). Upon exposure to *Pst*, SA production increases and promotes the nuclear translocation of the master immune regulator, NON-EXPRESSOR OF PATHOGENESIS-RELATED GENES 1 (NPR1) to reprogram the plant's transcriptome. NPR1 induces genes encoding antimicrobial peptides and the endoplasmic reticulum-resident proteins involved in the secretion of these peptides into the apoplast, establishing systemic acquired resistance (SAR). A large number of genes are also repressed during SAR, including those involved in mediating responses to the growth hormone auxin and the stress hormone jasmonic acid (JA) used mainly against necrotrophic pathogens (Spoel and Dong, 2008). The inhibitory effects of SA on the auxin and JA signaling pathways have been shown to be a part of SA-mediated resistance, but the molecular mechanisms of this crosstalk remain elusive. To understand the multi-layered immune responses mediated by SA, it is critical to identify the corresponding cellular targets of SA. Recently, evidence has emerged that NPR1 and its paralogs, NPR3 and NPR4, are nuclear receptors for SA (Fu et al., 2012; Wu et al., 2012). SA binding allows NPR3 and NPR4 to regulate stability of the NPR1 protein through their Cullin 3 ubiquitin ligase adaptor activity. However, these recent findings do not explain all of the physiological functions of SA, including the NPR1-independent immune response to biotrophic pathogens. Prior to the discovery of NPRs, catalases were the first proposed SA receptors (Chen et al., 1993). SA binding to catalases was shown to inhibit their enzymatic activities, resulting in the accumulation of H_2O_2 and the subsequent induction of plant defense genes. However, questions were raised about the specificity of SA binding as it appeared to be not unique to catalase but rather a general property of iron-containing enzymes of plant origin (Rüffer et al., 1995). Furthermore, it was found that exogenous application of H_2O_2 failed to activate SAR, adding controversy to the SA-catalase ligand-receptor story (Bi et al., 1995).

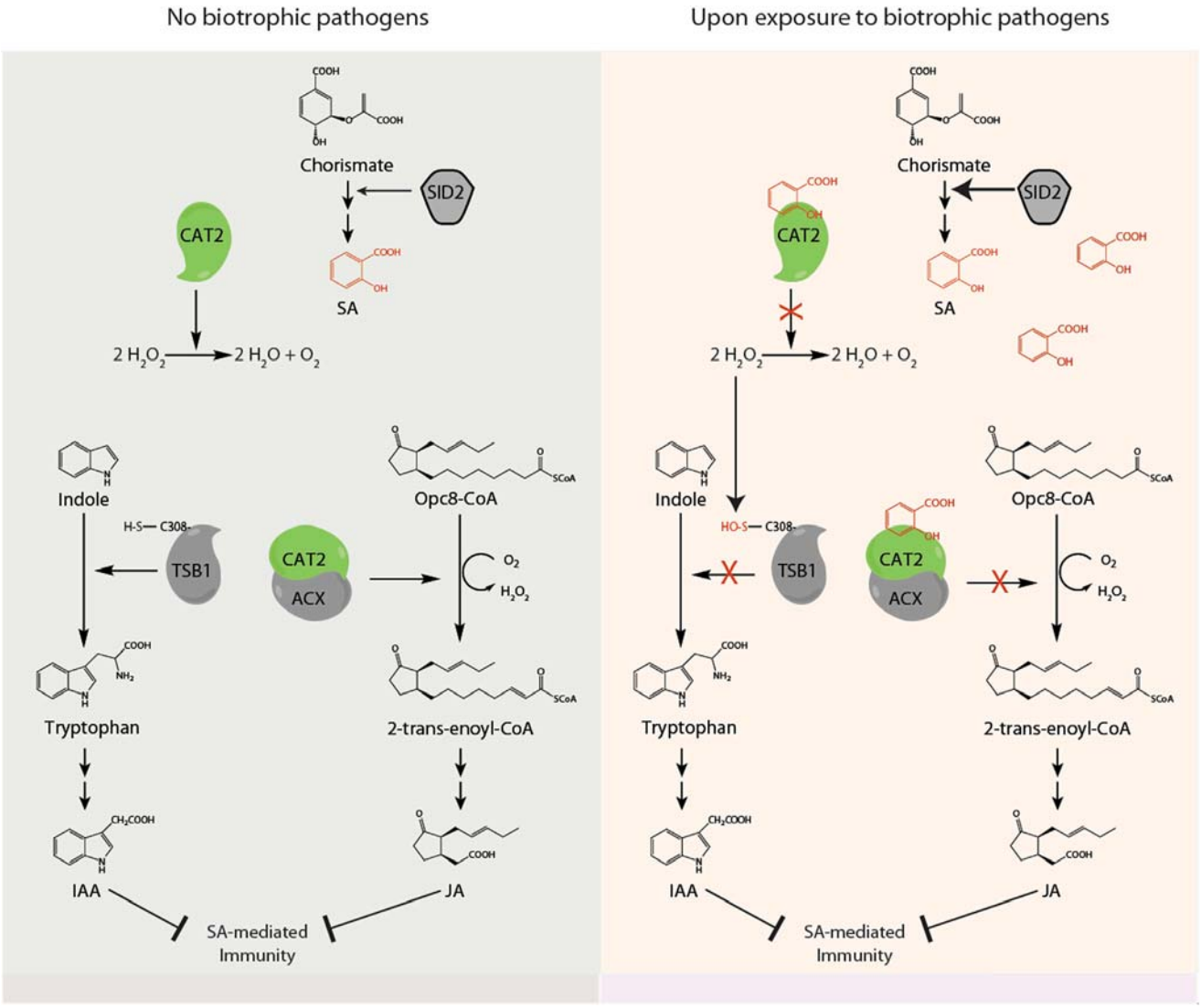

Figure 1. A Model Showing How SA Binding to CATALASE2 Represses Auxin and JA Biosynthesis to Strengthen Plant Immunity against Biotrophic Pathogens

In the absence of biotrophic pathogens, CAT2 efficiently converts H_2O_2 to H_2O. Upon exposure to biotrophic pathogens, SA accumulates and binds to CAT2 to inhibit its enzymatic activity. The accumulating H_2O_2 sulfenylates TSB1 to repress IAA production. SA binding to CAT2 also blocks CAT2's role in facilitating JA biosynthesis through ACX. The resulting reduction in IAA and JA levels strengthens plant immunity against biotrophic pathogens.

In this issue of *Cell Host & Microbe*, Yuan and coworkers present convincing data to demonstrate that CATALASE2 (CAT2) is a biologically functional SA receptor that contributes to SA-mediated resistance through inhibition of enzymes involved in auxin (indole acetic-acid, IAA) and JA biosynthesis (Yuan et al., 2017). The use of a genetic approach to validate CAT2 as a SA receptor, instead of relying on exogenous application of SA or H_2O_2, was key to the success of the study. The authors showed that in the SA biosynthetic gene mutant, *sid2*, pathogen-triggered inhibition of catalase activity was alleviated, preventing the accumulation of H_2O_2. A cross between *cat2* and *sid2* plants partially rescued the *sid2* SA-deficient phenotypes, including the enhanced disease susceptibility and compromised inhibition of auxin and JA biosynthesis, demonstrating that one of the functions of SA is to repress CAT2 activity.

To elucidate the functional consequences of SA-mediated inhibition of CAT2 activity, the authors focused on the effects on auxin and JA, the two hormones known to be inhibited during SAR. They examined protein sulfenylation as a possible signaling mechanism for H_2O_2 and found that the IAA-biosynthesis protein TRYPTOPHAN SYNTHETASE β SUBUNIT 1 (TSB1) is sulfenylated. Using both in vitro and in vivo enzymatic assays, as well as overexpression of TSB1 in the *cat2* mutant plant, the authors established a strong causal link between H_2O_2-mediated sulfenylation of cysteine 308 in TSB1, inhibition of the TSB1 enzymatic activity, and decreased IAA levels during SA-mediated resistance to biotrophic pathogens (Figure 1).

While SA mediates resistance against biotrophic pathogens, JA is produced during plant responses to necrotrophic

pathogens, such as *Botrytis cinerea*. The increased susceptibility to *Botrytis cinerea* observed in the *cat2* mutant plants encouraged the authors to study the physical interaction between CAT2 and the JA biosynthesis enzymes ACYL-CoA OXIDASES 2 and 3 (ACX2/3), which they detected in a yeast two-hybrid analysis. They found that the enzymatic activity of ACX2/3 is enhanced by its interaction with CAT2, and this enhancement can be suppressed by SA, suggesting that an enzymatically active CAT2 is required. Since ACX2/3 catalyzes the dehydrogenation reaction of acyl-CoAs resulting in the production of H_2O_2, we hypothesize that CAT2, in proximity, may increase the ACX2/3 activity by actively removing H_2O_2. Reduced JA levels in response to SA could compromise resistance to necrotrophic pathogens while enhancing defense against biotrophic pathogens (Figure 1).

It is intriguing that among the three *Arabidopsis* catalases, only CAT2 appeared to affect IAA and JA synthesis, even though SA has been shown to bind and inhibit all catalase isoforms of tobacco (Durner and Klessig, 1996). Though not tested in this study, this may be due to the time of the day when the catalase gene is expressed, as all three *Arabidopsis* catalase genes, as well as SA and JA synthesis, have been shown to be regulated by the circadian clock (Michael et al., 2008; Goodspeed et al., 2012). *CAT2* and *ACX* are morning-phased genes, whereas *CAT1* and *CAT3* are evening-phased genes.

Altogether, this study shows that CAT2 contributes to SA-mediated resistance by inhibiting IAA and JA biosynthesis, thereby providing functional evidence for its activity as a SA receptor. This work further supports the argument that there may be multiple SA receptors, each important for a subset of the many roles that SA plays in plant physiology.

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Strange New World: Bacteria Catalyze Ubiquitylation via ADP Ribosylation

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Three recent papers, including one by Kotewicz et al. (2016) in this issue of *Cell Host & Microbe*, show that *Legionella* deploys a novel form of ubiquitylation to generate its replicative vacuole. Without E1 and E2 enzymes, SidE effectors ubiquitylate serine residues in substrates via an ADP-ribosylated ubiquitin intermediate.

Legionella pneumophila is an opportunistic pathogen that typically spreads via inhaled aerosols from contaminated water sources such as air conditioners or water fountains. Upon ingestion by alveolar macrophages, bacteria replicate intracellularly and cause pneumonia during so-called Legionnaires' disease (Cunha et al., 2016). The natural hosts of *L. pneumophila* are free-living amoeba where, similar to human macrophages, bacterial replication requires manipulation of the original phagosome to avoid fusion with lysosomes and to enable the establishment of the *Legionella*-containing vacuole (LCV). LCVs are unique organelles that acquire several endoplasmic reticulum (ER) markers. To create its replicative niche, *L. pneumophila* deploys a type IVB secretion system (T4SS), known as Dot/Icm, that translocates effector proteins into the host cell. The extent of host cell manipulation by *L. pneumophila* can be appreciated from the sheer number of Dot/Icm effectors encoded in the bacterial genome: more than 300 genes encode