PLANT PATHOLOGY

Receptor networks underpin plant immunity

Plant-pathogen coevolution led to complex immune receptor networks

By **Chih-Hang Wu, Lida Derevnina, Sophien Kamoun**

Plants are attacked by a multitude of pathogens and pests, some of which cause epidemics that threaten food security. Yet a fundamental concept in plant pathology is that most plants are actively resistant to most pathogens and pests. Plants fend off their innumerable biotic foes primarily through innate immune receptors that detect the invading pathogens and trigger a robust immune response. The conceptual basis of such interactions was elegantly articulated by Harold H. Flor, who, in 1942, proposed the hypothesis that single genes in plants and pathogens define the outcome of their interactions; that is, a plant harboring a specific gene displays resistance against a pathogen that carries an interacting virulence gene (*1*). This gene-for-gene model was hugely insightful and influential—it has helped to guide applied and basic research on disease resistance. However, recent findings are taking the field beyond this simplified binary view of plant-pathogen interactions. Plants carry extremely diverse and dynamic repertoires of immune receptors that are interconnected in complex ways. Conversely, plant pathogens secrete a diversity of virulence proteins and metabolites called effectors, and pathogen genomics has revealed hundreds of effector genes in many species. These effectors have evidently evolved to favor pathogen infection and spread, but a subset of them inadvertently activate plant immune receptors. The emerging paradigm is that dynamic webs of genetic and biochemical networks underpin the early stages of plant-pathogen interactions.

Three layers of network complexity underpin plant response to pathogens (see the figure). First, sensor immune receptors can directly or indirectly detect molecules derived from pathogens. Second, many sensor receptors require coreceptors, helper receptors, or receptor-like regulatory scaffolds to translate pathogen recognition events into immune responses. Third, receptor complexes recruit distinct, but overlapping, downstream signaling components to generate a multifaceted innate immune response. Cross-talk at the receptor level is prevalent in plant immune networks and contributes to adjusting phenotypic outputs to biotic and abiotic environmental conditions, such as light and temperature (*2*, *3*).

Plant immune receptors are classed into cell surface receptors, including receptor-like kinases (RLKs) and receptor-like proteins (RLPs), or intracellular receptors of the nucleotide-binding domain and leucine-rich repeat–containing (NLR) family, both of which appear to form intricate receptor networks to mediate immune responses. The network feature of cell surface receptors is well established. RLKs and RLPs form various preformed and pathogen-activated complexes at the cell plasma membrane, which recruit receptor-like cytoplasmic kinases (RLCKs) to activate downstream immune signaling (*4*, *5*). RLKs in the somatic embryogenesis receptor kinase (SERK) family, such as SERK3 [also called BRASSINOSTEROID-ASSOCIATED KINASE 1 (BAK1)], participate in diverse cellular processes involved in development and immunity (*4*). Systemic analysis revealed that the extracellular leucine-rich repeat (LRR) domains of RLKs govern interactions between different receptors, forming an intricate network with multiple operational modules. Several RLKs, such as SERK3/BAK1 and ARTICULATION POINT EXECUTIVE (APEX), serve as interconnected cross-talk nodes or articulation points that help maintain network integrity. This reflects their essential roles in integrating multiple environmental stimuli and ensuring appropriate response modulation (*3*). Another RLK, SUPPRESSOR OF BIR1-1 (SOBIR1), is genetically and biochemically linked to RLPs in a network that also includes SERK3/BAK1 (*5*). An increased understanding of cell surface receptor networks and their connections to downstream components would enable manipulation of network nodes and fine-tuning of signaling outputs.

NLR proteins also engage in genetic and biochemical interactions that are more complex than anticipated (*6*, *7*). Many NLRs, which sense host-translocated pathogen effectors, require other NLR proteins to function, forming connections that vary from NLR pairs to networks. NLR pairs often operate through negative regulation, with the sensor NLR releasing its inhibition of helper NLR autoactivity upon pathogen perception. The concept of NLR networks is more recent, culminated through observations that some helper NLRs are functionally redundant and are required for the activities of multiple sensor NLRs. In asterid

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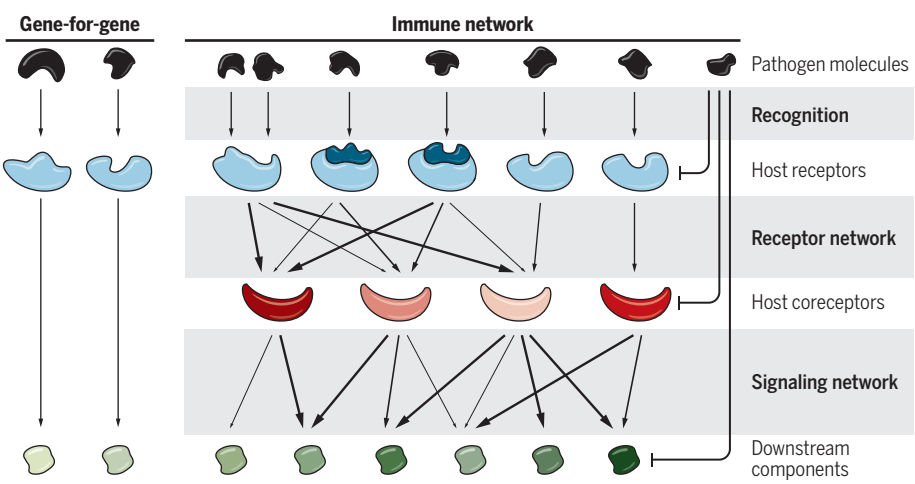
There are three mechanistic layers that contribute to immune complexity; each layer is represented by network nodes that may display functional redundancy, differential dependency (represented by arrow intensity), and specificity. These layers may also be targeted by pathogen effectors. This contrasts with the simplified binary view of the gene-for-gene hypothesis.



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plants, such as tomato and tobacco, NRCs (NLR-required for cell death proteins) and NRC-dependent NLRs form a complex genetic network that operates against various pathogens and pests (*7*). The NRC network has evolved from an ancestral NLR pair that expanded to half of all NLRs in some asterid species. Similarly, paralogs of the plant *Arabidopsis thaliana* ACTIVATED DISEASE RESISTANCE PROTEIN 1 (ADR1) helper NLR family are required for the function of several *A. thaliana* NLRs, although their evolutionary relationships are currently unknown (*8*). Whether additional phylogenetic clades of NLRs and cell surface receptors form evolutionarily defined networks remains to be determined. It will be interesting to further explore the network architecture of these highly diversified receptors in an evolutionary context, as this may unravel genetic and biochemical relationships among plant immune receptors.

Networks require finely-tuned regulation for optimal function. Receptor network homeostasis is achieved through different regulatory processes, including gene expression, receptor complex formation, and activation of signal transduction. Genes encoding paired NLRs overwhelmingly occur in a head-to-head genomic configuration, possibly because they share the same promoter elements for coregulation (*9*). Presumably, tight coexpression is important for optimal function and to avoid autoimmunity and fitness costs. How plants regulate genetically unlinked receptors that engage in complex networks remains poorly understood. Transcription factors, chromatin regulation, alternative splicing, and small RNAs are among the mechanisms that modulate NLR transcript dosage and maintain network homeostasis (*10*, *11*). Plants have also evolved regulatory mechanisms to control receptor complex formation. Cell surface receptors form ligand-dependent complexes with other receptor proteins, which may function as bona fide coreceptors or as regulatory scaffolds that establish signaling complexes (*3*). Homeostasis by negative biochemical regulation is also a common process. Cell surface receptor complexes are negatively regulated by RLCKs, pseudokinases, phosphatases, and E3 ubiquitin ligases, resulting in multiple paths of network feedback mechanisms (*5*).

Why have immune receptor systems evolved into complex network architectures? Networks enhance evolvability and maintain robustness, prominent features of immunity in many biological systems (*12*). A key feature of plant immune receptor networks is the uncoupling of plant pathogen perception from initiation of downstream signaling and immune response. Thus, receptors devoted to sensing pathogen molecules are free to expand and evolve rapidly with reduced constraints. In addition, functionally redundant core elements bypass the need for specific signaling mechanisms against each pathogen and increase resilience to environmental or internal disturbances. The SERK family of RLKs and the NRC family of NLRs are examples of core elements that display some degree of redundancy (*4*, *7*). Such elements ensure that the immune system remains operational whenever a receptor becomes nonfunctional owing to mutation, deletion, or immunosuppression by pathogens.

Plant pathogens counteract host immunity by deploying effectors that target the multiple layers of immune networks. Pathogen effectors are highly redundant and repeatedly suppress key host pathways, but their activities can also activate NLR immunity (*13*). Thus, effector redundancy can be viewed as a form of evolutionary bet hedging (whereby an organism suffers a fitness disadvantage in normal conditions for increased fitness under stress) that enables pathogen populations to keep up with plant immunity. Conversely, redundant nodes in immune receptor networks may help the plant evade suppression by pathogen effectors, further contributing to the robustness of the immune system. That pathogen effectors can act as both triggers and suppressors of receptor-mediated immunity further highlights the complex coevolutionary dynamics that drive these interactions.

"...understanding of receptor network topology directly affects strategies for retooling the immune system of crop plants..."

How does the network view of plant immune systems affect breeding for disease resistance? The effectiveness of the plant immune system rests on its capacity to detect invading pathogens. To date, most approaches to improve plant immune receptors have focused on expanding their pathogen detection spectrum (*14*). A detailed understanding of receptor network topology directly affects strategies for retooling the immune system of crop plants for enhanced disease resistance. These include boosting the expression of coreceptors to intensify the immune response mediated by multiple receptors. Network architecture can be manipulated for broader receptor-coreceptor connections to extend the capacity of the immune network. Transfer of individual receptor genes across distantly related crops often fails. Delivery of receptor-coreceptors as paired or subnetwork units may increase the success of broadening disease resistance in crops. Finally, given that pathogens have evolved effectors to target immune receptors, it should be possible to generate receptor variants that evade pathogen suppression. This could prove particularly useful when the pathogen-targeted receptor is a critical node in the immune network.

A new field, studying plant immune receptor networks, is emerging. We postulate that Flor's intuitive gene-for-gene model is superseded by the systems view that plant immune receptors form networks with complex topology (*3*, *4*, *7*). Although some plant immune receptors may function as singletons, possibly combining both recognition and signaling activities into a single molecule, most cell surface and intracellular immune receptors appear to engage other receptors to function. In the network context, node connectivity between pathogen effectors and sensor receptors, sensors, and helper or coreceptors, as well as helpers and signaling elements, is dynamic and obeys rules of specificity and signal strength. Receptor networks also govern immunity in animals, with Toll-like cell surface receptors forming homo- or heterocomplexes, and NLRs such as neuronal apoptosis inhibitory protein (NAIP) immune receptors forming inflammasome megacomplexes with their NLRC4 (NLR family CARD domain–containing protein 4) helper (*6*, *15*). Some plant receptors are known to form complexes, but the degree to which they form megacomplexes similar to inflammasomes is unclear. Decrypting the biochemical codes that rule receptor-network wiring will help define the molecular basis of host immunity. Ultimately, an improved knowledge of immune systems could enable optimal use and deployment of immune receptors in agriculture. ■

REFERENCES

1. H. H. Flor, *Phytopathology* **32**, 653 (1942).
2. Y. Belkhadir, Y. Jaillais, *New Phytol.* **206**, 522 (2015).
3. E. Smakowska-Luzan *et al.*, *Nature* **553**, 342 (2018).
4. X. Ma *et al.*, *Trends Plant Sci.* **21**, 1017 (2016).
5. D. Couto, C. Zipfel, *Nat. Rev. Immunol.* **16**, 537 (2016).
6. J. D. G. Jones *et al.*, *Science* **354**, 1174 (2016).
7. C.-H. Wu *et al.*, *Proc. Natl. Acad. Sci. U.S.A.* **114**, 8113 (2017).
8. V. Bonardi *et al.*, *Proc. Natl. Acad. Sci. U.S.A.* **108**, 16463 (2011).
9. J. C. Stein *et al.*, *Nat. Genet.* **50**, 285 (2018).
10. Y. Lai, T. Eulgem, *Mol. Plant. Pathol.* **19**, 1267 (2017).
11. Y. Zhang *et al.*, *Mol. Biol. Evol.* **33**, 2692 (2016).
12. H. Kitano, K. Oda, *Mol. Syst. Biol.* **2**, 2006.0022 (2006).
13. J. Win *et al.*, *Cold Spring Harb. Symp. Quant. Biol.* **77**, 235 (2012).
14. S. Cesari, *New Phytol.* **219**, 17 (2018).
15. N. Gay *et al.*, *Nat. Rev. Immunol.* **14**, 546 (2014).

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