植物学报 *Chinese Bulletin of Botany* 2021, **56** (2): 142–146, www.chinbullbotany.com
doi: 10.11983/CBB21042

·热点评述·

两类免疫受体强强联手筑牢植物免疫防线

王伟, 唐定中*

福建农林大学植物免疫中心, 福州 350002

摘要 植物先天免疫系统在抵御病原菌入侵过程中发挥至关重要的作用, 主要包括两个层次, 即病原菌相关分子模式和效应因子分别触发的PTI和ETI免疫反应。PTI和ETI分别由植物细胞膜表面模式识别受体(PRRs)和胞内免疫受体(NLRs)激活, 具有特异的激活机制, 但是两者激活的下游免疫事件相互重叠。PTI和ETI是否为泾渭分明的两道防线, 以及ETI与PTI下游事件为何如此相似, 一直是植物免疫领域最受关注的问题之一。最近, 中国科学院分子植物科学卓越创新中心辛秀芳团队与合作者利用拟南芥(*Arabidopsis thaliana*)与丁香假单胞杆菌(*Pseudomonas syringae*)互作系统对PTI和ETI在机制上的联系进行了研究。他们发现PRRs和共受体参与ETI, 而活性氧的产生是联系PRRs和NLRs所介导的免疫早期信号事件。他们还发现NLRs信号能够迅速增强PTI关键因子的转录和蛋白水平, PTI的增强在ETI免疫反应中不可或缺。该研究从机制上解析了植物免疫领域中长期悬而未决的PTI与ETI相似性之谜, 是该领域的一项突破性进展, 为未来作物分子设计育种提供了新的启示。

关键词 植物免疫, 病原菌相关分子模式触发的免疫反应(PTI), 效应因子触发的免疫反应(ETI), 活性氧, NLRs, 模式识别受体(PRRs)

王伟, 唐定中 (2021). 两类免疫受体强强联手筑牢植物免疫防线. 植物学报 **56**, 142–146.

当受到病原菌侵染时, 植物会利用位于细胞膜表面的模式识别受体(pattern-recognition receptors, PRRs)迅速识别病原菌保守的相关分子模式(pathogen-associated molecular patterns, PAMPs), 通过与共受体(co-receptor)结合并磷酸化激活胞质型类受体激酶(receptor-like cytoplasmic kinases, RLCKs), 如BIK1 (BOTRYTIS-INDUCED KINASE 1)和BSK1 (BR-SIGNALING KINASE 1) (Lu et al., 2010; Zhang et al., 2010; Shi et al., 2013), 进而激活丝裂原激活蛋白激酶(mitogen-activated protein kinases, MAPKs)级联信号通路、钙依赖蛋白激酶(calcium dependent protein kinases, CDPKs)通路和引起活性氧(reactive oxygen species, ROS)爆发等, 这些反应统称为PTI免疫反应(pattern-triggered immunity) (Couto and Zipfel, 2016; Tang et al., 2017; Wang et al., 2020; Zhou and Zhang, 2020)。

为了抑制植物PTI免疫反应, 一些病原菌向宿主细胞分泌效应因子(effectors)以干扰免疫进程, 使植物感病(Jones and Dangl, 2006)。面对病原菌的入侵, 植物进化出细胞内受体蛋白NLRs (nucleotide-binding site and leucine-rich repeat domain receptors), 通过识别效应因子激活更强烈的ETI免疫反应(effector-triggered immunity), 通常在病原菌入侵位点诱导超敏反应(hypersensitive response, HR)抑制病原菌的生长(Cui et al., 2015; Monteiro and Nishimura, 2018)。ETI免疫反应同样可以引起MAPKs信号通路激活和ROS爆发(Qi et al., 2011)。这些过程的调控与PTI相关组分是否有关尚不清楚, PTI和ETI之间的联系也有待研究。最近, 中国科学院分子植物科学卓越创新中心辛秀芳团队与合作者利用拟南芥(*Arabidopsis thaliana*)与丁香假单胞杆菌(*Pseudomonas syringae*)互作系统开展研究, 发现拟南芥ETI免疫反应的发生也依赖PTI通路中的多种元件, 进而揭示了PRRs与NLRs协同激活植物免疫的新机制, 增进了人们对植物先天免疫系统的认识, 是植物免疫研究领域的一项突破性进展(Yuan et al., 2021) (图1)。

收稿日期: 2021-03-02; 接受日期: 2021-03-05
基金项目: 国家自然科学基金(No.31830077)
* 通讯作者。E-mail: dztang@genetics.ac.cn

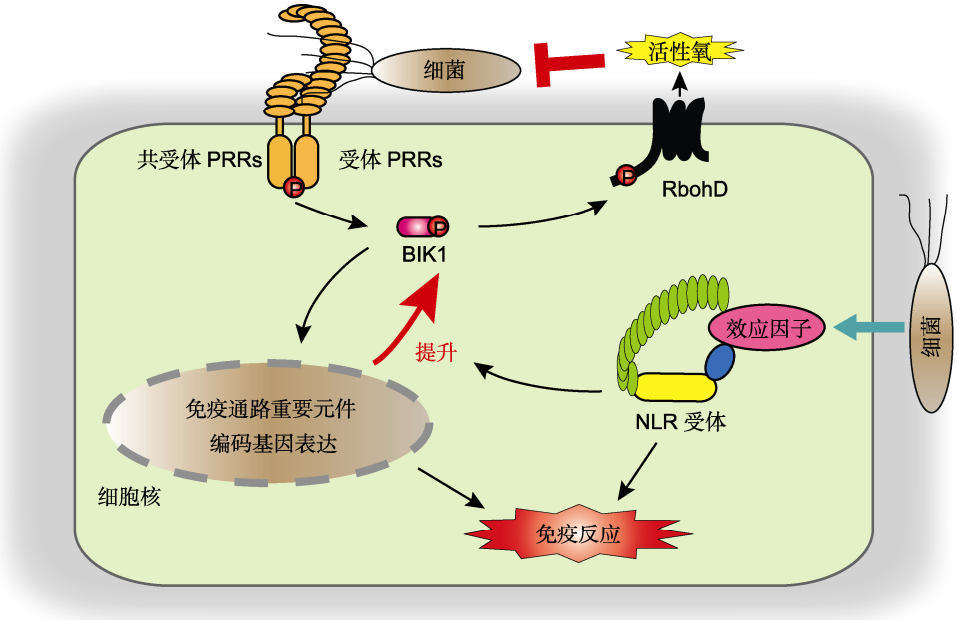

图 1 植物先天免疫系统示意图

植物通过细胞膜表面 PRRs 受体和共受体识别病原菌 PAMPs，并磷酸化胞质型类受体激酶 BIK1。激活的 BIK1 通过磷酸化 RbohD 正调控活性氧(ROS)爆发，抑制病原菌生长。成功入侵的病原菌通过向植物细胞内分泌效应因子抑制免疫反应。宿主细胞内的 NLRs 受体识别特定的效应因子后，不仅能够激活下游免疫反应，而且能提升 BIK1 和 RbohD 等免疫关键蛋白的丰度，增强活性氧爆发和免疫通路重要基因的表达，导致更加强烈的免疫反应。

Figure 1 Schematic representation of the plant immune system

Plants recognize the PAMPs of pathogens via the cell surface-localized PRRs, leads to phosphorylation of the receptor-like cytoplasmic kinase BIK1. The activated BIK1 positively regulates reactive oxygen species (ROS) burst by phosphorylation of RbohD, resulting in the inhibition of pathogen growth. Successful invaded pathogens deliver effectors into plant cell to inhibit immune responses. Upon perception of specific effectors, the host intracellular NLRs activate downstream immune responses, and boost the abundance of key immune-related proteins such as BIK1 and RbohD to enhance ROS burst and the transcripts of important genes in immune signaling, leading to robust immune responses.

野生型拟南芥通过NLRs蛋白识别特定效应因子，引起强烈的ETI免疫反应，抑制丁香假单胞杆菌*Pto* DC3000相应菌株的生长(Cesari, 2018; Monteiro and Nishimura, 2018)。但是，辛秀芳团队的研究发现，在缺失多种重要PRRs或共受体的拟南芥突变体*fls2*/*efr*/*cerk1* (*fec*)和*bak1*/*bkk1*/*cerk1* (*bbc*)中，由效应因子avrRpt2、avrPphB和avrRps4诱导的ETI免疫反应明显受到影响，说明ETI免疫反应也受PRRs受体的调控，暗示ETI依赖PTI (Yuan et al., 2021)。为了排除细菌内源效应因子等对免疫反应的影响，他们利用敲除了所有36个内源效应因子和冠菌素(coronatine)合成基因的*Pto* DC3000突变菌株D36E (Wei et al., 2015)进行研究。通过将*avrRpt2*转入D36E，并侵染拟南芥不同材料，发现D36E (*avrRpt2*)在野生型植物中的生长数量明显少于D36E菌株；而在缺失PRRs或共受体的突变体中，2种菌株的生长数量无显著差异(Yuan et al., 2021)，进一步说明avrRpt2诱导的ETI免疫反应依赖PRRs及其共受体。

细菌效应因子avrRpt2可以切割细胞内的RIN4 (RPM1-INTERACTING PROTEIN 4)，激活NLR蛋白RPS2 (RESISTANT TO P. SYRINGAE 2) (Day et al., 2005; Kim et al., 2005)。而辛秀芳团队研究发现，与野生型相比，PRRs或共受体多重突变体中D36E (*avrRpt2*)诱导的RIN4蛋白减少和*RPS2*转录本增加均未受影响，且MPK3/6的磷酸化程度也一致。为了深入探究PRRs或共受体多重突变体如何影响ETI免疫反应，他们重点研究了在PTI和ETI反应中都发挥重要作用的ROS产生(Qi et al., 2017)。非常有意思的是，通过对地塞米松(dexamethasone, DEX)诱导型*avrRpt2*转基因植株进行研究，他们发现flg22和DEX

共处理不仅可以快速诱导ROS的爆发(PTI-ROS), 而且在处理2–3小时后还出现了第2次持续性ROS爆发, 被称为ETI-ROS。ETI-ROS爆发同样依赖于flg22处理, 且在共受体多重突变体中ETI-ROS爆发几乎丧失, 说明PRRs信号通路的持续激活是ETI-ROS爆发的关键。同时, flg22处理可以加速拟南芥中avrRpt2诱导的HR反应, 说明ETI-ROS爆发可能促进ETI免疫反应(Yuan et al., 2021)。

PTI-ROS和ETI-ROS的产生是否依赖相同的亚细胞组分依然未知。早期有研究报道, 植物NADPH氧化酶和过氧化物酶(peroxidases)在病原菌诱导的PTI-ROS爆发过程中发挥关键作用(Torres et al., 2002; Daudi et al., 2012)。随后, 辛秀芳团队通过化学抑制剂筛选处理, 发现ETI-ROS的产生需要NADPH氧化酶RbohD (respiratory burst oxidase homolog D)。同时, 在D36E (*avrRpt2*)诱导的ETI免疫反应中, *RbohD*转录本水平出现上调(Yuan et al., 2021)。通过后续的遗传学和生物化学实验, 他们进一步明确ETI-ROS的产生依赖BIK1, 而BIK1通过磷酸化RbohD第343和347位丝氨酸(S343/347)调控ETI-ROS的爆发(Yuan et al., 2021)。前期已有研究表明, BIK1的S343/347磷酸化在细菌侵染时PTI反应中的ROS产生以及ETI介导的抗病反应中发挥重要作用(Kadota et al., 2019)。辛秀芳团队的研究结果进一步揭示了PRRs信号和NLRs信号通过协调RbohD活性和丰度来调控ETI-ROS爆发的重要性。

为了进一步探明是否存在其它PTI免疫通路成员参与调控ETI反应, 辛秀芳团队利用RNA-Seq技术分析了拟南芥野生型和共受体多重突变体分别接种D36E和D36E (*avrRpt2*)菌株3小时后的转录组差异。他们发现D36E (*avrRpt2*)处理后, ETI的激活可以在很大程度上恢复共受体多重突变体中PTI相关基因表达的缺陷。但是, 与野生型拟南芥相比, D36E (*avrRpt2*)处理后的共受体多重突变体中仍有272个基因表达存在差异, 其中包括许多PTI免疫通路标志基因, 如*WRKY22*/*29*和*FRK1*, 且这些基因的表达同样受到BIK1的调控, 说明BIK1是ETI免疫反应中ROS爆发和免疫基因表达调控的一个重要节点。此外, 无论是在野生型还是在共受体多重突变体中, D36E (*avrRpt2*)处理均引起大量PTI免疫通路关键元件编码基因的表达上调(Yuan et al., 2021)。最近, 英国塞恩斯伯里实验室Jonathan DG Jones课题组也发现PRRs和NLRs两类免疫受体互相协作, 单独激活PRRs或NLRs介导的抗性都不能有效抵御*Pto* DC3000入侵(Ngou et al., 2021)。相关研究成果以“背靠背”形式发表在*Nature*杂志上。在以往的研究中, PTI和ETI被认为是两条平行的免疫通路。但以上研究结果表明, PTI和ETI并非泾渭分明的两道防线, ETI的激活需要PRRs和共受体参与, ETI免疫信号可以调动PTI免疫通路, 进而放大PTI反应, 表明植物免疫反应的全面激活需要PRRs信号和NLRs信号的强强联手。这项研究工作从机制上阐明了植物免疫领域中长期悬而未决的PTI与ETI相似性之谜, 增进了人们对植物先天免疫系统的理解, 为未来作物的抗病育种提供了新的启示。

参考文献

Cesari S (2018). Multiple strategies for pathogen perception by plant immune receptors. *New Phytol* **219**, 17–24.

Couto D, Zipfel C (2016). Regulation of pattern recognition receptor signaling in plants. *Nat Rev Immunol* **16**, 537–552.

Cui HT, Tsuda K, Parker JE (2015). Effector-triggered immunity: from pathogen perception to robust defense. *Annu Rev Plant Biol* **66**, 487–511.

Daudi A, Cheng ZY, O'Brien JA, Mammarella N, Khan S, Ausubel FM, Bolwell GP (2012). The apoplastic oxidative burst peroxidase in *Arabidopsis* is a major component of pattern-triggered immunity. *Plant Cell* **24**, 275–287.

Day B, Dahlbeck D, Huang J, Chisholm ST, Li DH, Staskawicz BJ (2005). Molecular basis for the RIN4 negative regulation of RPS2 disease resistance. *Plant Cell* **17**, 1292–1305.

Jones JDG, Dangl JL (2006). The plant immune system. *Nature* **444**, 323–329.

Kadota Y, Liebrand TWH, Goto Y, Sklenar J, Derbyshire P, Menke FLH, Torres MA, Molina A, Zipfel C, Coaker G, Shirasu K (2019). Quantitative phosphoproteomic analysis reveals common regulatory mechanisms between effector- and PAMP-triggered immunity in plants. *New Phytol* **221**, 2160–2175.

Kim HS, Desveaux D, Singer AU, Patel P, Sondek J, Dangl JL (2005). The *Pseudomonas syringae* effector AvrRpt2 cleaves its C-terminally acylated target, RIN4, from *Arabidopsis* membranes to block RPM1 activation.

Proc Natl Acad Sci USA **102**, 6496–6501.

Lu DP, Wu SJ, Gao XQ, Zhang YL, Shan LB, He P (2010). A receptor-like cytoplasmic kinase, BIK1, associates with a flagellin receptor complex to initiate plant innate immunity. *Proc Natl Acad Sci USA* **107**, 496–501.

Monteiro F, Nishimura MT (2018). Structural, functional, and genomic diversity of plant NLR proteins: an evolved resource for rational engineering of plant immunity. *Annu Rev Phytopathol* **56**, 243–267.

Ngou BPM, Ahn HK, Ding PT, Jones JDG (2021). Mutual potentiation of plant immunity by cell-surface and intracellular receptors. *Nature* doi: 10.1038/s41586-021-03315-7.

Qi JS, Wang JL, Gong ZZ, Zhou JM (2017). Apoplastic ROS signaling in plant immunity. *Curr Opin Plant Biol* **38**, 92–100.

Qi YP, Tsuda K, Glazebrook J, Katagiri F (2011). Physical association of pattern-triggered immunity (PTI) and effector-triggered immunity (ETI) immune receptors in *Arabidopsis*. *Mol Plant Pathol* **12**, 702–708.

Shi H, Shen QJ, Qi YP, Yan HJ, Nie HZ, Chen YF, Zhao T, Katagiri F, Tang DZ (2013). BR-SIGNALING KINASE 1 physically associates with FLAGELLIN SENSING 2 and regulates plant innate immunity in *Arabidopsis*. *Plant Cell* **25**, 1143–1157.

Tang DZ, Wang GX, Zhou JM (2017). Receptor kinases in plant-pathogen interactions: more than pattern recognition. *Plant Cell* **29**, 618–637.

Torres MA, Dangl JL, Jones JDG (2002). *Arabidopsis* gp91^{phox} homologues *AtrbohD* and *AtrbohF* are required for accumulation of reactive oxygen intermediates in the plant defense response. *Proc Natl Acad Sci USA* **99**, 517–522.

Wang W, Feng BM, Zhou JM, Tang DZ (2020). Plant immune signaling: advancing on two frontiers. *J Integr Plant Biol* **62**, 2–24.

Wei HL, Chakravarthy S, Mathieu J, Helmann TC, Stodghill P, Swingle B, Martin GB, Collmer A (2015). *Pseudomonas syringae* pv. *tomato* DC3000 Type III secretion effector polymutants reveal an interplay between HopAD1 and AvrPtoB. *Cell Host Microbe* **17**, 752–762.

Yuan M, Jiang Z, Bi G, Nomura K, Liu M, Wang Y, Cai B, Zhou JM, He SY, Xin XF (2021). Pattern-recognition receptors are required for NLR-mediated plant immunity. *Nature* doi: 10.1038/s41586-021-03316-6.

Zhang J, Li W, Xiang TT, Liu ZX, Laluk K, Ding XJ, Zou Y, Gao MH, Zhang XJ, Chen S, Mengiste T, Zhang YL, Zhou JM (2010). Receptor-like cytoplasmic kinases integrate signaling from multiple plant immune receptors and are targeted by a *Pseudomonas syringae* effector. *Cell Host Microbe* **7**, 290–301.

Zhou JM, Zhang YL (2020). Plant immunity: danger perception and signaling. *Cell* **181**, 978–989.

Synergistic Cooperation Between Cell Surface and Intracellular Immune Receptors Potentiates to Activate Robust Plant Defense

Wei Wang, Dingzhong Tang*

Plant Immunity Center, Fujian Agriculture and Forestry University, Fuzhou 350002, China

Abstract Innate immune system plays a crucial role to defend against pathogens attack and is classified into two layers, which include pathogen-associated molecular pattern-triggered immunity (PTI) and effector-triggered immunity (ETI). The PTI and ETI are activated by cell-surface localized pattern-recognition receptors (PRRs) and mostly intracellularly-localized nucleotide-binding, leucine-rich repeat receptors (NLRs), respectively, with specific activation mechanisms, but largely overlapped downstream immune events and components. One of the top unanswered questions in the field of plant immunity is whether ETI and PTI are really distinct, considering the high similarity of the downstream of the recognition processes and components. Recently, a team led by Prof. Xiufang Xin, CAS Center for Excellence in Molecular Plant Sciences, Chinese Academy of Sciences, used the *Arabidopsis thaliana* and *Pseudomonas syringae* pathosystem to study the functional link between PTI and ETI, and demonstrated that PRRs and the co-receptor of PRRs contribute to ETI, and the production of reactive oxygen species (ROS) is the early signal event that connects PTI and ETI. They also showed that ETI enhances the transcript and protein levels of key components of PTI, and the increased PTI is crucial for full activation of ETI. This study provides mechanistic explanation to a long-lasting enigma in the field of plant immunity regarding the mechanistic connections of PTI and ETI, and the high similarity of these two layers of immunity. This work represents an important breakthrough in the field of plant immunity, and will have implications for the future molecular breeding in crops.

Key words plant immunity, pattern-triggered immunity (PTI), effector-triggered immunity (ETI), reactive oxygen species, nucleotide-binding site and leucine-rich repeat domain receptors (NLRs), pattern-recognition receptors (PRRs)

Wang W, Tang DZ (2021). Synergistic cooperation between cell surface and intracellular immune receptors potentiates to activate robust plant defense. *Chin Bull Bot* **56**, 142–146.

---

* Author for correspondence. E-mail: dztang@genetics.ac.cn

(责任编辑: 白羽红)