



Plant Immunity and Pathogen Resistance: Recent Discoveries

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Abstract

Plants and pathogens are natural enemies of each other, and understanding how plant immunity is developed against pathogen attacks is very important to ensure food security. In the last few decades, major progress has been made to determine how plants recognize pathogens and defend themselves at the molecular level, which has led to improve forms of plants. This article summarises some recent developments on plant immunity with emphasis on pathogen recognition, hormonal signaling and genetic engineering for enhanced resistance in crop plants. It is important to note that plants own an elaborate immune system, which includes innate as well as acquired immunity. The plants cells bear some receptors called pattern recognition receptors (PRRs) which detect the pathogen-associated molecular patterns (PAMPs) and leads to pattern-triggered immunity (PTI), whereas the effector-triggered immunity (ETI) involve receptors called resistance (R) genes that recognize pathogen effectors. Thus, not only have phytohormones like salicylic acid (SA), jasmonic acid (JA) and ethylene (ET) been identified to be involved in regulation of immune response and in the fine control of defense and growth trade-off. Owing to rapid progeny in genomics and biotechnology, numerous new resistance genes have been discovered, thus improving knowledge of the resistance of pathogens. As perhaps the best example, the CRISPR/Cas9 technology, along with other similar gene-editing processes, can proceed with research on the generation of crops that are resistant to most of the new strains of pathogens. Also, it is the micro and macro organisms that make up the plant microbiomes that can improve pathogen resistance through mutual symbiotic relationships with plants. The recent revelation of epigenetic regulation of immunity-gene further complicates plant



defense mechanisms, and it can be assumed that plants have memory of past pathogens. Although issues like pathogen evolution and breakdown of resistance persist, such knowledge continues to be gradually produced from research in these fields to deliver permanent solutions to disease resistance in crops. It also shows the need for cross-sectional studies in informing the next course of action towards eradicating the menace of plant diseases as well as stabilizing the agriculture sector.

Introduction

Plant defence and pathogen infection are critical consiliaries to the plant health and crop yield. With the increased demand of food in the world, there is need to guard the crops from ailments attributed to different pathogens hence the concern in this paper. In the last several decades the study of plant immunity has painted a picture of complex and constantly evolving system that enables plants to discern between self and non self microbial pathogens including bacteria, fungi and viruses. These immune mechanisms are not only fascinating from the plant biology perspective but they give directions to breed crops that is resistant to diseases. There are innate and adaptive immunity in plants that work as a society and each organism on its own. The first level of defense called pattern-triggered immunity (PTI) activated when PRRs on plant cell surface identify pathogen-associated molecular patterns (PAMPs). Subsequently, broad and more effective defense mechanisms– effector-triggered immunity (ETI) –are initiated by plant resistance (R) genes that recognize pathogen effectors that result into a stronger reaction. Novel advances have revealed multiple new factors that play important roles in these processes, that open a door to understand how plants prevent pathogens. Besides these genetic factors, phytohormones including SA, JA and ET are largely involved in regulation of immune responses. These hormones regulate how the body will defend itself against different stressors as well as growth and the immune response. In addition, the relation plant endophytes and pathogens also presented as crucial influence in defense mechanisms. It is useful to know that beneficial microbes can improve the immune system reactivity of plants that can then create a barrier against pathogens.

Plant Immunity Mechanisms

Many plants have developed complex defense mechanism that guard them from diverse pathogen agents such as bacteria, fungi, viruses and nematodes. Plant immunity mechanisms are broadly classified into two main types: Somatic immunity and hormonal immunity which involves important functions in protection against infections. Primary defense that is inherent comprises of plant intrinsic immune system which is rapid to be initiated once the plant recognizes the PAMPs through PRRs on the cell membrane. This recognition leads to Pattern-Triggered Immunity (PTI), a broad or non-specific immune response. PTI engages several defensive measures for instance enzyme activated ROS, actively obliterate pathogens and antimicrobial peptides to directly attack the pathogens. This response is usually followed by physical barriers such as the thickening of plant cell wall. The acquired immunity is more specific and effective type of immunity which occurs when the plant meets with pathogen's secreted effector proteins. There is effector-triggered immunity (ETI) an additional layer of defense that is activated in plant cells by the recognition of these effectors by plant resistance genes (R genes). This reciprocal recognition by pathogen effectors and R proteins results in a highly localized cell death called hypersensitive response (HR) to prevent the further invasion of the pathogen. Effectors are recognized by R proteins in very specific manners, this gives plants the advantage of response towards a specific pathogen. Apart from PTI and ETI, SAR is an enduring form of immunity, which offers profound protection against subsequent attack. SAR is triggered by localized infection through a signaling molecule, salicylic acid (SA) that has a relay of defense signals all over the plant. Combined these mechanisms create a layered system that enables plants to identify and combat pathogen threats effectively so that plants may live in hostile conditions.

Understanding of molecular breathe of pathogen resistance

Originally, resistance of a host plant to a pathogen lies in a combination of the genetics, biochemistry, and cell biology of the plant. Pathogens have been identified by plants through resistance (R) genes that are proteins that are involved in both the reception and defense against



pathogen invasions. Understanding pathogen resistance involves the gene-for-gene hypothesis whereby for every effector gene of the pathogen, there is an R gene in the plant. This identification brings about an immune response that either hampers or reduces the generation of pathogen. R proteins are crucial to plant defense and are categorized into many classes of which the most-researched are the NBS-LRR or Nucleotide Binding Site-Leucine-Rich Repeat proteins. Signal is initiated by a nucleotide-binding domain which is a part of these proteins while the pathogen recognition is due to the leucine-rich repeat segment in the proteins. When a pathogen injects effector molecules into plant cells, an R protein perceives specific pathogen-derived effectors by detecting molecular patterns of pathogen, thereby leading to effector-triggered immunity (ETI). This usually results in a rapid hypersensitive response (HR) in which localized cell death occurs to the extent of containing the pathogens. However, plants have their own receptors called PRRs that recognize conserved pathogen associated factors such as flagellin or chitin, apart from the R genes. PTI is a broader defense mechanism that triggers other downstream defense responses such as synthesis of ROS, thickening of cell wall and release of antibiotics. Also, plants use the hormone signaling to regulate or increase resistance. These include the hormones such as salicylic acid (SA) as a component of defense hormone, jasmonic acid (JA) and ethylene (ET). Activation of SA signal is important in immune response to biotrophic pathogens while JA and ET pathways are important in defense against necrotrophic pathogens. In summary, the molecular aspects of the plant defense against pathogens are an integrated genetic hormonal and biochemical system through which plants are able to detect, protect and counteract against a plethora of pathogen invasions.

Hormonal Signalling for Plant Protection

Plant hormones are central to the orchestration of defense mechanism that occur in plants in response to infection by different pathogens. To engage and modulate defense reactions, plants apply numerous kinds of hormones, including salicylic acid, jasmonic acid and ethylene. Bot01, Bot02, ABA and GA are such hormones acting to co-ordinate plant defense responses with growth and development, in order to efficiently utilize available resources during stress. It is

worth, therefore, to note that the regulation of defense against the biotrophic pathogens that feed on living plant tissues involve the Salicylic Acid (SA). When plants come across these pathogens, SA is stocked in the plants and it sets off a chain of immune responses. Perhaps the most publicized impact of SA is the elicitation of Systemic Acquired Resistance (SAR), which is a long duration defense mechanism against a broad spectrum of pathogens. SA also increases the extent of the synthesis of resistance (R) proteins involved in effector-triggered immunity (ETI). SAR responses are mainly dependent on the signaling of SA and are most important for combating viral and fungi. While, Jasmonic Acid (JA) is more engaged in react to necrotrophic pathogens, these are types of pathogens that kill, or cause plants tissues to die, and then feed on the dead plant cells. JA also plays imperative roles in defence against insect herbivores.” After pathogen recognition, JA accumulation leads activation of defense genes that synthesis the antimicrobial proteins such as proteinase inhibitors and defensins. JA signaling is widely connected with ISR which is a great defense to stresses that affect plant health all over the plant. Another hormone called Ethylene (ET) is also participate in plant immune response under the regulation of SA and JA suppressing mechanisms. ET plays a predictable role in modulating the responses to both biotic and abiotic stress. SA, JA and ET are well linked in the cross talk which defines the particular immune response to the pathogen or stress by a particular plant type.

New Insights on Resistance Gene Identification

Gene for resistance (R) genes have been pivotal in comprehending plant defence and breeding for disease resistance crops. Tom_pushButton_IS1 In the last decade important developments in genomics, bioinformatics, and functional genomics have allowed an enhanced pace in the discovery and description of R genes, creating new prospects for durable enhancements of crop resistance against various pathogens. Originally, R genes were detected in classical genetic analyses and plant breeding, when certain genes were localized to specific resistance factors. Still, new approach to identification of R genes became possible because of the recent advancements in high-throughput sequencing technologies referred to as next-generation sequencing (NGS) and whole-genome sequencing. These technologies have also said the



investigation of plant immune system at relatively deeper level revealing relationships between R genes, pathogen effectors and the plant immune signaling pathways.

Perhaps the single largest advance made in resistance gene discovery has been the elucidation of ETI processes and cloning of R genes required for the perception of specific pathogen effectors. Effector proteins are released by pathogens to down regulate plant defense mechanisms and R genes are able to recognize these effectors and activate defenses. Current work has made it possible to clone many R genes from crops such as wheat, rice and tomato and make selective breeding for disease-resistant varieties possible. Furthermore, with the advancement of CRISPR/Cas9 system and other gene editing tools, there are possibilities for the functional studies of R genes at detailed level and molecular manipulation of plant genome for germline transformation of disease resistance. In addition, with the advances in metagenomic analysis and the understanding of plant microbiome new observations were made that microbial members interact with plant immunity. Some microbial species can trigger a systemic acquired resistance (SAR) or may even compete with pathogens in the process. Studying the relationships of R genes with the microbiome is now a major research focus in plant defense mechanisms.

Epigenetics of plant-osphere interactions: Modulation of plant immunity.

Plant immunity is implicated in epigenetic regulation that determines how the plant will respond to pathogen attacks. Hypermethylation and hypomethylation are types of epigenetic regulation of gene expression on that do not entail any alteration of the element DNA sequence itself. These changes may be both heritable and adaptive and are crucial to monoand polysaccharide perception to fine-tune the plant's immune responses to biotic and abiotic stresses. Studies accomplished in the last decade have established that epigenetic regulations, such as DNA methylation, histone modification, and non coding RNAs, can play crucial functions in regulating plant defense and improving their resistance to diseases. DNA methylation is the process by which methyl is attached to the DNA, for instance to specific cytosine's which results to gene silencing. In plants, DNA methylation is now known to control the defense response genes, the resistance (R) genes which are involved in the recognition of



pathogen effector and subsequent immune responses. Other work has shown that DNA methylation results in the repression of the R genes while DNA demethylation leads to activation of the genes in a way that can either strengthen or weaken the capacity of a plant to organize an immune response. This dynamic regulation is critical so that plants can effectively manage both defense mechanisms and growth, with immunity only being invoked when necessary.

The other epigenetic regulation includes Histone modification, in which functional groups are added to Histones (proteins that DNA is condensed around), and is also critical in controlling the expression of immune related genes. These include acetylation, methylation and phosphorylation; the above made can either enhance or inhibit expression of genes depending on the changes of chromatin. In the context of plant immunity, histone modifications can also regulate the expression of genes related with the perception and signaling of the pathogen invasions and thus function as a switch to trigger the plant defense systems at once. Further, Noncoding RNA, small interfering RNAs (siRNAs) and microRNAs (miRNAs) also play the role in epigenetic regulation by either repressing the genes associate with pathogen permissiveness or by boosting immunity response. These RNA molecules can act on one or several genes, or segments of genes, that regulate the plant immune response.

Plant and Microbial Partnerships and Their Connection to Disease Fighter

Plant microbiome comprises bacteria, fungi, viruses, and archaeal populations as well as occurring biofilms which has potential impact on plant health and diseases. In recent years, the importance of plant-microbiome symbiosis for plant nutrient acquisition, abiotic stress tolerance, and the improvement in plant-pathogen interactions has been well documented by various scholars. It has been established that the microbiome can prevent plant diseases in several ways. First, beneficial microbes may employ exclusion: beneficial microbes including plant growth promoting rhizobacteria (PGPR) would consume and occupy all the space and nutrients needed by the pathogens. This is referred to as competition exclusion whereby advantageous microbes limit the growth of pathogenic microbes for the same resources and metabolic die quickly while the advantageous use up all available nutrients. Moreover, specific microbiomes start the plant



immune response which leads to Systemic Acquired Resistance (SAR) or Induced Systemic Resistance (ISR). These processes use actions to induce defense response even in specific portions of the plant that are not affected by pathogens. Microbes, for instance, those from the rhizosphere or leaf surface spew out chemical compounds like jasmonic acid (JA), salicylic acid (SA) activate the plant's immune system. For instance, *Pseudomonas fluorescens*, *Bacillus subtilis* have been reported to elicit ISR which favors protection against diverse pathogens. Another defense vector through which microbiomes contribute to disease immunity involves production of antimicrobial substances like the antibiotics and hydrogen peroxide. Such compounds immobilise or destroy the pathogens point blank thus adding another layer of defence to plants against diseases. Additionally, the plant microbiome control the development of the plant as a response to pathogens by changing their own hormonal signaling pathways and controlling the expression of resistance (R) genes. Beneficial microbes can wake up the plant's defence, making it better placed to combat subsequent invasions by pathogens.

Strategies Involving application of biotechnology for improved pathogen resistance

Biotechnology provides numerous opportunities in breeding plants to be resistant to pathogens including a variety of tools and techniques to address the continuously increasing problem of plant diseases. Although, in some occasions classical breeding for resistance has proven to work, this approach has shortcomings due to the limited diversity that is inherent in crop species and because of the processes involved. Whereas, the Application of modern biotechnological methodologies, more precise, faster and sustainable in developing disease resistance plants. Probably the most hydrologized biotechnological strategy is genetic engineering whereby genes conferring resistance (R) are introduced into the crops to enable them defend against pathogens. For example, the transgenic expression of genes that encode PRRs or effectors is an example of using genes which were introduced to provide resistance against bacterial, fungal or viral pathogens. Because Bt cotton/Corn contain the insecticidal protein from *Bacillus thuringiensis*, Bt cotton/Corn are successful examples of Genetically Modified crops which offer inherent protection against selected insect pests. One of the other disruptive technologies is gene editing



with a more well-known tool, CRISPR/Cas9. This tool is helpful in making specific alterations to plant genomes either to trigger indigenous protective responses or to bring in new characteristics. For example, it has been applied to treat underlying mutations of the signaling pathways in which immune system components exist by improving the plants' capacity to discover those components in pathogenic organisms. Further, RNA interference (RNAi) has been used to knock down genes related to susceptibility to pathogens or to enhance defense gene expression as a precise approach for enhancing disease tolerance. Another research area within genetic engineering of plants is synthetic biology. Using targeted microbiome manipulation, or through genetic modification of plants, the scientist can develop new pathways of resistance that would not otherwise be available in the selected type of organism. This includes the capability of producing plants with new types of antimicrobial peptides or new small molecules to destroy pathogens.

These biotechnological approaches have in fact improved plant disease management by presenting new prospects for increasing resistance against the pathogen in crops. However, conventional methods of breeding, despite being useful, have been hampered by time and gene constraints. Unlike deleterious gene-splicing, BT, however, is a specific, accurate and effective means to create disease-resistant plants that are necessary to meet emerging pathogen issues, environmental changes, and sustainable agriculture. Molecular breeding, new generation genetic engineering tool CRISPR/Cas9 and RNA interference technology is already expanding the horizon of improving plant resistance mechanisms. These technologies allow specific genetic manipulation of the genes which are responsible for pathogen perception, signaling and defense pathways thereby increasing the plants intrinsic disease resistance. Consequently, the ability to promote or down regulate specific resistance genes has catapulted the engineering of crops that are more resistant to diseases ranging from fungal and bacterial infections, to viral ailments. However, the future looks very promising due to the synthetic biology and engineering of plant microbiomes as the potential for the development of new resistance genes or mechanisms as improved resistance compounds or plant-microbe communication. Not only do these approaches



help increase disease tolerance but also, in combination with marker-assisted breeding, it also helps in the enhancement of few chemistries needed crop, which in turn reduces the global effects of the agriculture industry. These biotechnological tools offer a lot of promise, however they should come with thoughts on their regulatory, ecological, and moral advantage. However, realization of these technologies will depend on the acceptance of people in the society social chains, and proper management of the technologies once adopted. The other epigenetic regulation includes Histone modification, in which functional groups are added to Histones (proteins that DNA is condensed around), and is also critical in controlling the expression of immune related genes. These include acetylation, methylation and phosphorylation; the above made can either enhance or inhibit expression of genes depending on the changes of chromatin. In the context of plant immunity, histone modifications can also regulate the expression of genes related with the perception and signaling of the pathogen invasions and thus function as a switch to trigger the plant defense systems at once. Further, Noncoding RNA, small interfering RNAs (siRNAs) and microRNAs (miRNAs) also play the role in epigenetic regulation by either repressing the genes associate with pathogen permissiveness or by boosting immunity response. These RNA molecules can act on one or several genes, or segments of genes, that regulate the plant immune response.

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Conclusion

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