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Role of resistant-proteins in plant innate immunity- A review

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ABSTRACT

Plants are affected by an extensive range of microbes, such as fungi, bacteria, viruses, viroids, and protozoa. Over the advancement, plants have evolved to defend against pathogens by developing numerous defense mechanisms in the plant cell that respond to chemical and physical barriers, These are present in plants locally or integrally. Plants have an unacquired immunity system to battle against pathogens, such as microbe-associated molecular patterns (MAMPs) are recognized through pattern recognition receptors in a primitive immune system which intermediates the defense response. Pattern recognition receptors (PRRs) they play a major role in plant innate immune system. They encode host sensors of PRRs, they can able to detect molecules of the pathogens such as macrophages, dendritic cells, neutrophils and epithelial cells containing proteins that are present in plant immune system. In this system, they identify two classes of molecules such as pathogen-associated molecular patterns (PAMPs), which are conjoined with microbe associated molecular patterns (MAMPS), and damage-associated molecular patterns (DAMPs). These are correlated with host cells released during cell damage or cell death, these are evolved before adaptive immunity. These extracellular receptors of Pattern Recognition Receptors were first identified in plants and many plant PRRs have been identified by using genomic analysis (rice and Arabidopsis). There are six classes of R-proteins, playing the major role of plant innate immunity which help to activate a defense mechanism from different microbes. Many of plant R genes encode proteins which are nucleotide-binding site and leucine-rich repeats (NB-LRR) region that contact with pathogen effectors to cause defense responses. Pathogen infection in plants is usually limited by an accomplished defense response activated by resistance genes.

Key words: DAMPS, MAMPS, NBS– LRRS, Pattern recognition receptors, RLKS, PAMPS, RLPS.

Plants have resistance (R) genes which conserve uniform observation over the infecting pathogens (Joshi and Nayak, 2011). Pathogen Recognition takes place at two different relevant groups of host receptors. Pathogen-associated molecular patterns (PAMPs) are recognized by pattern recognition receptors (PRRs) of the host. These PAMPS are present in pathogen itself but not present in the host. The first group called PRRs, these containing extracellular receptor-like proteins (RLPs) and receptor-like kinases (RLKs) are in contact with the cell membrane. They resemble toll-like receptors (TLRs) present in an animal in terms of their structure and function in plant innate immunity. PAMP-triggered immunity (PTI) is to provide resistance against pathogens. Uematsu *et al*., (2006), Altenbach *et al*., (2007), Iriti *et al*., (2007), (Zipfel, 2008). Intracellular receptors are another group which contain resistance proteins (R proteins). Most of the identified Resistance proteins belong to the family of Nucleotide Binding Site-Leucine Rich Repeat (NBS-LRR) (Lukasik and Takken, 2009). Pamp triggered immunity suppressed due to these pathogen effector is introduced into the host cell. NBS-LRR genes resistant against only obligate biotrophs or hemibiotrophic pathogens, but not necrotrophs (Glazebrook, 2005).The pathogen produced virulence effectors which suppress the PAMP triggered immunity (PTI), which results in effector-triggered susceptibility (ETS) because plants do not have appropriate receptors. If a host has susceptible receptors, pathogen having avirulence effectors that initiate the defense response (Fig 1) is referred to as effector-triggered immunity (ETI) Jones and Dangle, (2006); De Wit, (2007). ETI is mediated by intracellular immune receptors and directly or indirectly depends on resistance genes (R genes). R genes can be divided into at least five groups. The largest group is the nucleotide-binding site-leucine-rich repeat (NBS–LRR) genes (2002). NBS–LRR genes are spread widely in plants.

MAMPS: First the microbe recognition is performed by extracellular proteins named as Pattern Recognition Receptors (PRRs). These PRRs recognized the MAMPS in plants these are small molecule sequences with groups of microbes that are recognized by cells of the innate immune system and PRRs of the host. PAMPS: The first level of microbe recognition is performed by membrane proteins termed pattern recognition receptors (PRRs), which perceive molecular signatures characteristic of a whole class of microbes, termed pathogen-associated (or microbe-

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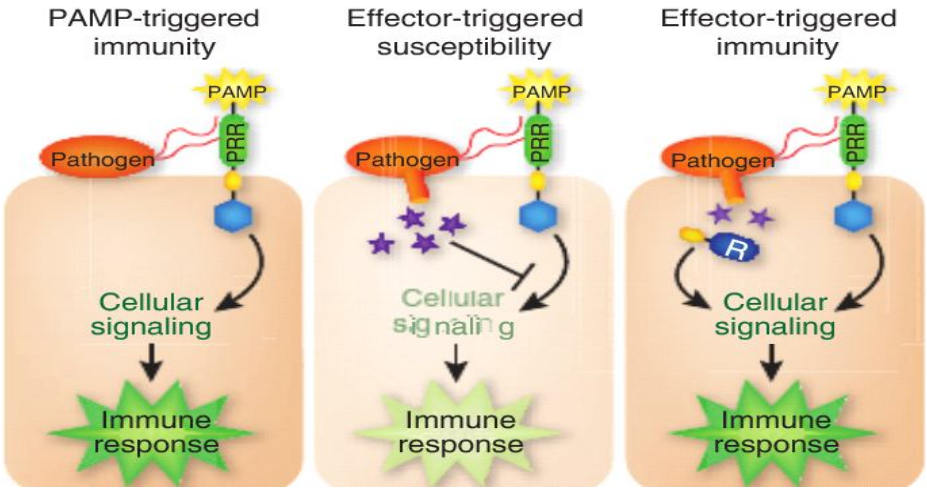


Fig 1: Upon pathogen attack, pathogen-associated molecular patterns (PAMPs) activate pattern-recognition receptors (PRRs) in the host, resulting in downstream signaling that leads to PAMP-triggered immunity (PTI). Virulent pathogens have acquired effectors (purple stars) that suppress PTI, resulting in effector-triggered susceptibility (ETS). In turn, plants have acquired resistance (R) proteins that recognize these attacker-specific effectors, resulting in a secondary immune response called effector-triggered immunity (ETI). (Pieterse *et al*.,2009).

associated) molecular patterns Perception of PAMPs by PRRs is common to all multicellular organisms and leads to an array of defense responses and redeployment of cellular energy in a fast, efficient, and multiresponse manner, which prevents further pathogen ingress. PAMP recognition leads to a chain of signaling events, broadly referred to as general defense responses in plants (Nicaise,2009). DAMPS: Many plant pathogens produce lytic enzymes to breach the structural barriers of plant tissues. The products generated by these enzymes may function as endogenous elicitors or DAMPs. Such DAMPs typically appear in the apoplast and, as in the case of MAMPs, can serve as danger signals to induce innate immunity Boller and Felix, (2009).

Plant PRRS: Plant PRRs that are structural and functionally with drosophila TOLL and mammalian TLRs. Xa21was the first identified PRR in plant or animal protein, resistance to the Gram-negative bacterial pathogen Xanthomonas oryzae pv oryzae (Bahar *et al*, 2014). Since that time two other plants PRRs, Arabidopsis FLS2 (flagellin) and EFR (elongation factor Tu receptor) have been isolated (Song *et al*, 1995). The corresponding PAMPs for FLS2 and EFR have been identified Boller and Felix, (2009). Upon ligand recognition, the plant PRRs transduce "PAMP-triggered immunity" (PTI). Plant immune systems also encode resistance proteins that resemble NOD-like receptors, that feature NBS and domains and can also carry other conserved interaction domains such as the TIR cytoplasmic domain found in Toll and Interleukin Receptors (Chisholm *et al*, 2006). The NBS-LRR proteins are required for effector-triggered immunity (ETI) (Hale *et al*, 2006).

Structure of PRRs: Receptors detecting microbial patterns can be divided into Surface receptors and Intracellular receptors. Surface receptors are known to detect both M/P/DAMPs these surface receptors are no.1.Receptor-like kinases (RLK) 2., Receptor-like proteins (RLP). In *Arabidopsis*, 610 RLK and56 RLP has been identified, but only a limited number have been functionally characterized and even fewer are reported to act as immune receptors (Song *et al*, 1995)Many *Arabidopsis* genes encoding RLK and RLP were found to be induced upon flg22 or EF-Tu, suggesting that they may function as immune receptors .

RLK: PRRs were first discovered in plants, Since that time many plant PRRs have been predicted by genomic analysis (370 in rice; 47 in Arabidopsis) (Song *et al.*, 1995). Plant PRRs are composed of an extracellular domain, trans membrane domain, and intracellular kinase domain as part of a single protein**.** Recent evidence indicates that some identified PRRs are members of the RLK family, e.g., the flagellin receptor (FLS2) and elongation factor Tu (EF-Tu)-receptor (EFR). Indeed, sequencing of the *Arabidopsis thaliana* genome has revealed the presence of >400 RLK sequences with various receptor configurations, of which those containing a leucine-rich repeat (LRR) in the extra-cellular domain constitute the largest group with 216 members Gomez-Gomez and Boller, (2002). The diversity and a large number of plant RLKs suggest that they may be involved in the perception of a wide range of stimuli. Other PRRs are also found amongst known-RLK proteins such as *Glycine max* β-glucan elicitor binding protein (GmGBP), *Lycopersicon esculentum* ethylene-inducing xylanase (LeEIX2) and chitin elicitor-binding protein (CeBIP) for the perception of β-glucans (soybean), xylanase (tomato) and chitin fragments (rice) respectively (Ingle *et al* 2006). Other RLKs found associated with the cell wall include lectin receptor kinases, a subset of which are found in plasma

membrane-cell wall adhesions and proline-rich extensin-like receptor kinases (PERKs) involved in sensing of cell wall damage due to wounding by pathogens.

RLP: Consists of an extracellular domain and a membrane-spanning domain. As they lack an intracellular activation kinase domain, and consequently, they require interaction with adaptor molecules for signal transduction Zipfel, (2008). RLPs are structurally reminiscent of the toll-like receptors (TLR) that mediate MAMP recognition in animals Kopp and Medzhitov (2003).

Toll-like receptors (TLR): Recognition of extracellular or endosomal pathogen-associated molecular patterns is mediated by transmembrane proteins known as toll-like receptors (TLRs). TLRs share a typical structural motif, the Leucine-rich repeats (LRR), which give them their specific appearance and are also responsible for TLR functionality (Beutler *et al*, 2006). Toll-like receptors were first discovered in Drosophila and trigger the synthesis and secretion of cytokines and activation of other host defense programs that are necessary for both innate or adaptive immune responses.

Characteristic features of PRRs: PRRS sense the nonself molecules only: Microbe-Associated Molecular Patterns (MAMPs), Pathogen Associated Molecular Patterns (PAMPs), a molecule that is generated within the plants upon microbial invasion or Damage Associated Molecular Patterns (DAMPs).PRRs associated with kinases**:** A class of protein called Kinases is key to PRRs ability to respond to pathogens. PRRS is often succumbed to "powerful" pathogen secretions (effectors): They are often called "effectors". The effectors are capable of "defeating" PRRs signal mediated defense product.

Cell-surface receptors: Cell-surface receptors, also known as transmembrane receptors, are cell surface, membrane-anchored, or integral proteins that bind to external ligand molecules. This type of receptor spans the plasma membrane and performs signal transduction, converting an extracellular signal into an intracellular signal. Ligands that interact with cell-surface receptors do not have to enter the cell that they affect. Cell-surface receptors are also called cell-specific proteins or markers because they are specific to individual cell types. Each cell-surface receptor has three main components: an external ligand-binding domain (extra-cellular domain), a hydrophobic membrane-spanning region, and an intracellular domain inside the cell. The size and extent of each of these domains vary widely, depending on the type of receptor. Cell-surface receptors are involved in most of the signaling in multicellular organisms (Fig 2).

Intracellular receptors in Plants: Plant NB-LRR proteins (nucleotide-binding site–leucine-rich repeats) have been studied in detail and some members are well characterized as immune receptors. They are traditionally referred to as disease resistance proteins or *R*-genes and form the bridge between molecular cell biology in plant immunity and plant breeding for agriculture. They form one of the largest gene families in plants. There are more than 140 predicted members in *Arabidopsis* and more than 400 in rice. Their gene products promote resistance to viral, bacterial, fungal, and oomycete pathogens.

NB-LRR proteins (Fig 3) recognize pathogen-specific signals, most often effector molecules responsible for virulence, either directly or indirectly. Although these proteins reside within the cytoplasm, they are also mobile and can translocate into the nucleus, chloroplast or mitochondria. For example, barley MLA, tobacco N, and *Arabidopsis* RPS4 translocate into the nucleus. Activation of defense responses by extracellular and intracellular PRRs have been defined as primary and secondary immune responses, respectively Shen and Schulze-Lefert *et al*, (2007). Recent studies demonstrated that individual effectors could be recognized by the same intracellular receptor,

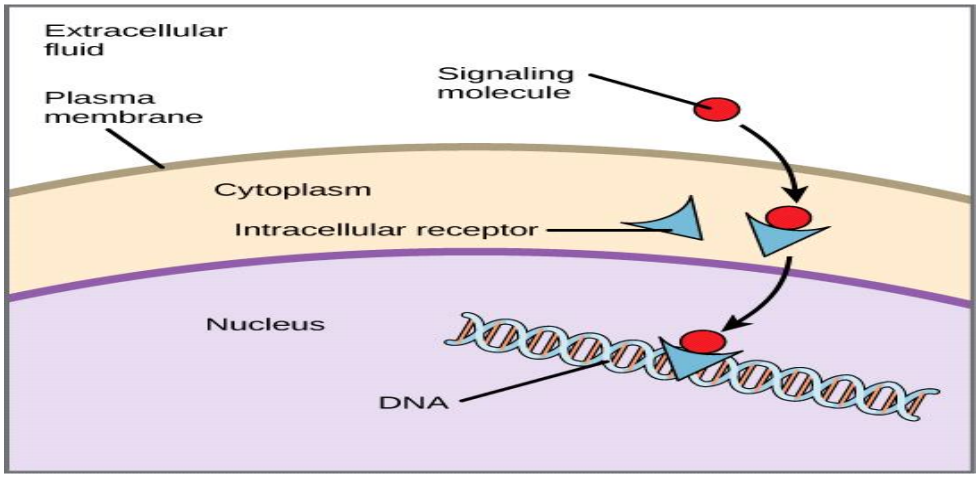
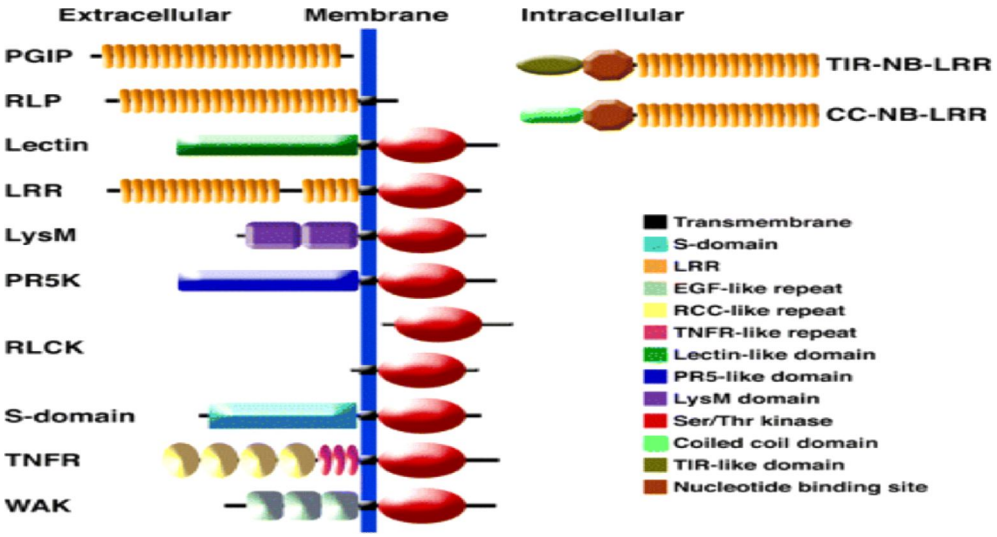


Fig 2: Hydrophobic signaling molecules typically diffuse across the plasma membrane and interact with intracellular receptors in the cytoplasm. Many intracellular receptors are transcription factors that interact with DNA in the nucleus and regulate gene expression (Sigalov, 2010).

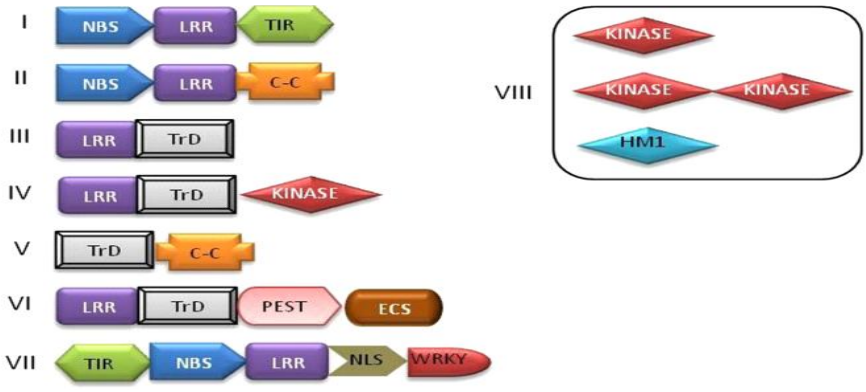
especially by those that recognize incoming effectors indirectly de Wit, (2007).

Major classes of R-proteins: Major classes of R proteins Plant resistance genes (Fig 4) can be broadly divided into eight groups based on their amino acid motif organization and their membrane-spanning domains. The LRRs (Leucine-rich repeats) represents the components having an important role for recognition specificity and these domains are present in the majority of R proteins. A first major class of R-genes includes the genes encoding for cytoplasm proteins with a nucleotide-binding site (NBS), a C-terminal leucine-rich repeat (LRR) and a putative coiled-



(Tor *et al*., 2009)

Fig 3: Domain organization of typical extracellular and intracellular receptors in plants. Receptor proteins act as surveillance mechanisms that plants utilize to detect signals including microbe-associated molecular pattern molecules (MAMPs), damage-associated molecular pattern molecules (DAMPs), virulence factors, secreted proteins, or processed peptides. ExtracellularPGIP, RLP, and RLK-type (classiûed according to their extracellular domains) receptors recognize these signals by direct or indirect interactions. Subsequentheterodimerization and phosphorylation events and possible conformational changes promote interactions with regulatory molecules such as RLCKs to distribute the message within the cell. Intracellular receptors, NB-LRR proteins, tend to recognize pathogen speciûc signals, most often effector molecules responsible for virulence, either directly or indirectly. Through conformational changes, they evoke the canonical signal-ling cascade and trigger resistance to the pathogen. AlthoughNB-LRR-type receptors constitutively reside within the cytoplasm they are also mobile and can translocate into the nucleus, chloroplast or mitochondria. RLCK-type proteins do not have extracellular domains, however, they are classiûed as RLKs. Abbreviations: PGIP, polygalacturonase inhibitor protein; RLP,receptor-like protein RLK, receptor-like kinase; LRR, leucine-rich repeats; LysM, lysine motif; PR5K, pathogenesis-related 5-likereceptor kinase; RLCK, receptor-like cytoplasmic kinase; S-domain, self-incompatibility domain; TNFR, tumour necrosis factor receptor; WAK, wall-associated kinase; NB, nucleotide binding; TIR, Toll and Interleukin 1 transmembrane receptor; CC, coiled-coil; EGF, epidermal growth factor; RCC, regulator of chromosome condensation; Ser/Thr, serine/threonine).



(Gururani *et al*., 2012)

Fig 4: (Major classes of plant resistance (R) genes based on the arrangement of the functional domains. LRR e Leucine-rich repeats; NBS e Nucleotide-binding site; TIRToll/Interleukin-1- receptors; C-C e Coiled-coil; TrD e Transmembrane domain; PEST e Protein degradation domain (proline-glycine-serine-threonine); ECS e Endocytosis cell signaling domain; NLS e Nuclear localization signal; WRKY e Amino acid domain; HM1 e*Helminthosporiumcarbonum* toxin reductase enzyme).

coil domain (CC) at the N- terminus. The examples of this class of resistance genes include the *P. syringae* RPS2 and RPM1 resistance genes of Arabidopsis and the tomato *Fusarium oxysporum* resistance gene I2. The second class of resistance genes consists of cytoplasmic proteins which possess LRR and NBS motifs and an N-terminal domain with homology to the mammalian toll-interleukin-1- receptor (TIR) domain. The tobacco N gene, flax L6 gene, and RPP5 gene are a few examples categorized under this class. A third major class of resistance genes family devoid of NBS motif consists of extra cytoplasmic leucine-rich repeats (eLRR), attached to a transmembrane domain (TrD). eLRRs are known to play an important role for certain defense proteins such as aspolygalacturonase-inhibiting proteins (PGIPs) even though they are not directly involved in pathogen recognition and activation of defense genes. The *C. fulvum* resistance genes (Cf-9, Cf-4, and Cf-2) having an extracellular LRR (eLRR), a membrane-spanning domain, and a short cytoplasmic C terminus are some examples of this class of resistance genes. The rice Xa21 resistance gene for Xanthomonas is an example of the fourth class of resistance genes which consists of an extracellular LRR domain, a transmembrane domain (TrD) and an intracellular serine-threonine kinase (KIN) domain Song *et al*, (1995). The fifth class of resistance genes contains the putative extracellular LRRs, along with a PEST (Pro-Glu-Ser-Thr) domain for protein degradation (found only in Ve2, and not Ve1), and short proteins motifs (ECS) that might target the protein for receptor-mediated endocytosis (e.g. tomato Ve1 and Ve2 genes) However, these Ve1 and Ve2 proteins have recently been proposed as PAMP receptors Thomma *et al*, (2011). The Arabidopsis RPW8 protein is an example of the sixth major class of resistance genes which contains a membrane protein domain (TrD), fused to a putative coiled-coil domain (CC) whereas, the seventh major class of resistance genes includes the Arabidopsis RRS1-R gene conferring resistance to the bacterial phytopathogen *Ralstonia solanacearum*, and it is a new member of the TIReNBSeLRR R protein class. RRS1-R has a C-terminal extension with a putative nuclear localization signal (NLS) and a WRKY domain Deslandes *et al*, (2003). The WRKY domain is a 60 amino acid region that is defined by the conserved amino acid sequence WRKYGQK at its N-terminal end, together with a novel zinc-finger-like motif. The eighth major class of resistance genes includes the enzymatic R-genes which contain neither LRR nor NBS groups. For example, the maize Hm1 gene which provides protection against southern corn leaf blight caused by the fungal pathogen *Cochliobolus carbonum* Johal and Briggs, (1992). Unlike other resistance genes, Hm1 codifies the enzyme HC toxin reductase, which detoxifies a specific cyclic tetrapeptide toxin produced by the fungus (HC toxin) that is essential for pathogenicity. Therefore, cereal resistance genes like Hm1 can be seen to codify a range of different proteins that in some cases have obviously very different functions. Another notable example, Pto protein in *P. syringae* contains a Ser-Thr kinase domain without LRRs. Whereas, the Rpg1 gene of barley which confers resistance to stem rust codified a receptor kinase-like protein with two tandem protein kinase (kinase) domains and does not contain a strong membrane-targeting motif and known receptor sequences Brueggeman *et al*, (2002). In Arabidopsis the SAR path way confers resistant to Pseudomonas syringae (P.v) maculicolaES 4326 and *Peranospora parasitica*.SAR has been reported in rice with *Pseudomonas syringae* pv.s*yringae* as the inducing pathogen(Kamal *et al*., 2015).

Evolution of R genes: When a plant was first attacked by a new pathogen strain, the plant probably had some genes. This pathogen was able to cause considerable damage to the plant and possibly killed many of the susceptible plants. During the evolutionary race for survival of the plant from the pathogen, a resistance (R1) gene evolved, e.g, by modification of one of the general resistance genes, and that gene allowed the plant to recognize one of the initial steps of infection by the new pathogen (race 1) (Fig 5) and to resist infection, such an individual plant and its progeny (variety 1) were selected for survival and so the plant and the R1 gene survived and multiplied. For example, by modification of one of the receptors involved in activating plant defenses against pathogens in general. Thus, the modified receptor1 product of the R1 gene recognizes

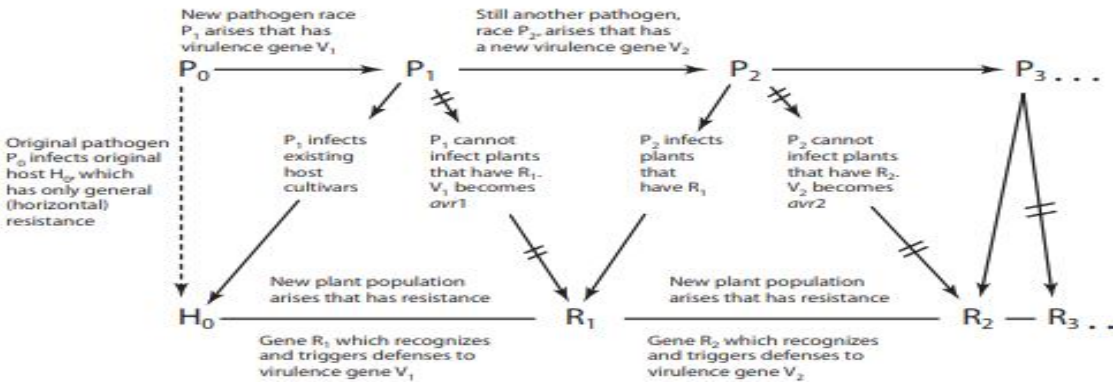


Fig 5: Steps in evolution of genes for virulence, resistance and avirulence **(**Agrios, 2005).

specifically particular compound (elicitor 1) produced by a pathogen gene, which gene, as a result, behaves like an avirulence (*avr1*) gene. Pathogens carrying this *avr1*gene (race 1) cannot survive on such R1 gene-carrying plants. If, however, in time, a mutation affects the *avr1*gene of race 1 of the pathogen, which gene until now was the cause of its virulence, the gene and the avirulence destroyed. As a result, the new offspring of the pathogen become virulent again, capable of attacking the so-far resistant variety 1 of the plant. This new virulent pathogen population could be called race 2. The host plant (variety 1) is now susceptible to race 2, which infects and may kill many plants. After the *avr2* gene conferring avirulence to the pathogen because it is recognized by the R2 gene of the plant. In this way, numerous, diverse R genes have evolved in a plant host to counteract corresponding virulence genes in the various races of one of its pathogens Agrios, (2005).

The classification of NBS-LRR proteins: The basis for the activity of the NBS-LRR family of plant R proteins is their ability to directly or indirectly recognize MAMPs and MIMPs typical for the particular pathogens prior to initiating the specific resistance response. This is only possible after recruiting a number of domains displaying various functions. Two such domains that are essential for classifying a protein as a member of the NBS-LRR family are the nucleotide-binding domain (NB) and the leucine-rich repeat domain (LRR). They are connected by a homologous region called ARC, which is present in plant R proteins, and in mammal APAF-1 protein and its homolog, CED4 from *Caenorhabditis elegans*. The NB domain combined within ARC motif is usually referred to as a nucleotide-binding site (NBS) Rairdan *et al*, (2007). Further classification of the NBS-LRR proteins depends on the presence of other domains. Two subfamilies of these proteins can be distinguished: TIR-NBSLRR(TNL) proteins containing the TIR domain, originally identified as an intracellular part of the Toll receptor in *Drosophila* and human interleukin 1receptor, and non-TIR-NBS-LRR proteins containing other domains, the largest of which is a group of CNL receptors with the coiled-coil (CC) domain.Such domains are present in various organisms, and they function in oligomerization processes like the TIR domain Oakley and Hollenbeck, (2001). So far, no TNL proteins have been detected in monocotyledonous plants. However, in papaya, a small number of genes encoding NBS-LRR proteins were detected. It has been suggested that the diversity of R proteins in papaya is achieved by alternative splicing among other factors; this would compensate for the relatively small genomic diversity of R proteins encoding genes (Porter *et al*, 2009). Monocot mannose binding lectins (MMBL) attract greater attention because of its antiviral and anti fungal activities which were confirmed by artificial bio assay experiment (Sathyapriya *et al*.,2012)

NBS-LRRS structure and function: The R proteins were identified in several plant species, such as *A. thaliana*, *Populus* or *Medicago trunculata*. The two main domains of plant R proteins, NBS and LRR, seem to be the most crucial in the pathogen recognition process and the activation of signal transduction in the response to pathogen attack.

LRR domain: The LRR domain was identified for the first time in the α 2-glycoprotein of human serum (Takanashi *et al*, 1985). In plants, the first NBS-LRR-encoding genes were cloned in1994 (Mc Hale *et al*, 2006). The essential structural element of the LRR domain is the tandem repeat of 20-30 amino acids containing a consensus sequence LxxLxLxxNxL, where L is a leucine residue or another aliphatic amino acid, N is asparagine, threonine, serine or cysteine, and x is any amino acid Kajava, (1998). A protein with an LRR domain has to contain at least two LRR repeats. The outer part of the domain, usually composed of α-helices, connected with β-strands by β-turns (Bella *et al*, 2008).The LRR domain is most frequently attributed the role of an intramolecular regulatory that inhibits the receptor activity when an appropriate elicitor is absent. Recent data showed that the dissociation of the LRR and NBS domains might not be required for the activation of the receptor (Rairdan and Moffett, 2006). Instead, repetitive rounds of dissociation and re-association could lead to the amplification of the signal originating from elicitor recognition.

NBS domain: The NBS domain was identified in NBS-LRR proteins based on its similarity to the homologous sequences in the animal APAF-1 and CED-4 proteins (Vanderbiezan and Jones, 1998). This domain is characterized by NTPase activity, and it is suggested to play a crucial role as a molecular switch activating signal transduction. In the signaling pathway, changes in the conformation of the NBS domain occur, caused by reversible nucleotide binding, which leads to the activation/deactivation of the whole receptor (Tameling *et al*, 2006). Research on the tomato mutants *I-2* and *Mi-2* proved that the NBS domain functions by switching R protein from its active to non-active state and showed that it has the capability to bind and hydrolyze ATP. A similar effect is shown by mutation of the MHD motif at the site of the aspartic acid residue of the flax L6 protein leading to spontaneous necroses. In this case, mutations in the nucleotide-binding regions (P loop) interacting with an elicitor (LRRdomain) neutralize the receptor activity Howles *et al*, (2005). Generally, mutations in the NB andARC2 subdomains are involved in the mechanism of auto-activation (Takken *et al*, 2006). Similar effects were reported for domain swapping between potato Rx proteins (which recognize the capsid protein of potato virus X) and GPA2 (which recognize the elicitors of pathogenic nematodes) Rairdan and Moffett, (2006). Egg plant or brinjal highly susceptible to various soil borne disease. The extensive use of chemical fungicides to combat these disease can be minimized by identification of resistant genes analogs

(RGAs) in wild species of cultivated plants. Nucleotide binding sites having conserved regions these were used to amplify RGAs from wild relatives of egg plant (Dev *et al*., 2018).

NBS-LRR Proteins evolution: NBS-LRR genes have been identified in most of the major plant groups. It is believed that TNL receptors developed as early as before the separation of the branch leading to bryophytes from the one leading to seed plants NBS-LRR genes were also found in gymnosperms. Genes from *Pinus monticola* were cloned and compared with the sequences of different R genes in *A. thaliana* and *Brassica* and were found to display phylogenic links with the TIR-NBS-LRR protein class (Liu and Ekramoddoullah, 2003). Genes cloned from *Pinus lambertiana*, determined as *P1_RGC-CC_1*, showed a significant similarity to the CC-NBS-LRR proteins of *A. thaliana*. A high degree of conservativeness of these genes was discovered, which makes them similar to *ADR1* (Activated Disease Resistance 1) of *A. thaliana*. One of the proposed explanations for this stability assumes that P1_RGC-CC_1 is an example of a guard protein (Jermstad *et al*, 2006). As mentioned before, no genes coding the TIR-NBS-LRR proteins were identified in monocotyledons, although extensive research was carried out involving database searches and sequence cloning. Since such genes were found in gymnosperms, and they are very common in dicotyledons, it is likely that they occurred in a common ancestor of angiosperms, and then the branch leading to monocotyledons lost the TIR-NBS-LRR genes (Bai *et al*, 2002). The probability of sucha scenario is also supported by a similar loss having also been observed in sugarbeet (*Beta vulgaris*), a dicotyledonTian *et al*, (2004). Research on twelve R genes derived from different plants (e.g. *Zea mays*, *A. thaliana*, *L. usitatissimum*) allowed the detection of traces of the differentiating selection in over 70% of the xxLxLxx motifs present in the LRR domain (Jiang *et al*, 2007).This phenomenon seems to be in contradiction to the pressure leading to the generation of new virulence factors in a pathogen population and the corresponding new R genes in a host population (Vanderhoon *et al*, 2002). It can be caused either by favorite influence of individual alleles of R genes on the host fitness in the presence of a pathogen or by negative influence when a pathogen is absent Meyers *et al*, (2005). A distinct selection of this type has been observed in the locus of the *A. thalianaPRP13* gene, which conditions resistance to *Peronospora parasitica* (Rose *et al*, 2004).

NBS-LRR role in plant immunity: Considerable evidence indicates the involvement of the NBS-LRR proteins in plant resistance to many pathogens, particularly the fact that many mapped regions of plant genomes containing different disease resistance genes codifiedproteins with NBS-LRR domains. By mapping these regions, it was possible to tolink CNL family genes with the resistance of some cultivars of ginger (*Zingiberofficinale*) to *Fusariumoxysporum (*Swethapriya and Subramanian, 2008). Also, the *A. thaliana*RRS1-R gene, preconditioning resistance to *Ralstonia solanacearum* or the RPP4 gene, responsible for resistance to different strains of *P. parasitica*, were found tobelong to the multigenic family of RPP5 receptors of the TNL type (Van der Biezen *et al*, 2002). Recentresults showed the roles of intracellular interactions between different domainsof R proteins and intercellular interactions between Avr and R and other hostproteins in the initiation of disease resistance (Rentel *et al*, 2008). The NBS-LRR and CC domains of Rx proteins, responsible for the resistance of *Solanaceae* plants to PVX (potato virus X), can interact in the *trans*conformation occurring during the co-expression of their coding sequences. It was shown that theintegration of the *Hyaloperonospora parasitica* effector gene *ATR13* (*A. thaliana* recognized 13) with the genome of Tu MV (Turnip Mosaic Virus) results in the resistance of *A. thaliana* plants to this virus. The interaction between *S. lycopersicum* and *P. syringae* allowed the description of the initiation of the resistance response by NBS-LRR receptors. It was proved that the resistance of *S. lycopersicum* to *P. syringae* results from the presence of Pto kinase and the NBS-LRR protein called Prf. Itseems that interactions between Avr Pto, Pto ,and Prf occur according to the decoyor guard model. In the first case, Pto is an operative target of the Avr Pto effectoror a decoy produced by the host to bind pathogen effectors, and Prf is anR protein. According to the guard model, Pto is a target for the AvrPto effector, guarded by Prf. Pto kinase seems to be an inhibitor of Prf activity, and Avr Ptoprobably interacts with Pto, inhibiting its kinase activity through interaction between two specific regions. Such a reaction brings about de-repression of there sistance response initiated by Prf (Vanderhoon and Kamoun, 2008).

CONCLUSION

R- protein-based resistance of plants, to various pathogens, is the main area of interest in plant innate immunity. Understanding the evolutionarily developed interactions between plants and pathogens is a necessary starting point to develop a strategy to protect economically important crops. Research on TLRS has also revealed that recognition of intracellular pathogen is mediated via a TLRS-Independent mechanism. Future efforts should focus on identifying new R genes and their corresponding pathogen effectors and modeling these proteins and their interactions.

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