Review

NPR1 and Redox Rhythm: Connections, between Circadian Clock and Plant Immunity

Jingjing Zhang [1,2]**, Ziyu Ren** [1,2]**, Yuqing Zhou** [1,2]**, Zheng Ma** [1,2,3]**, Yanqin Ma** [1,2,3]**, Dairu Hou** [1,2]**, Ziqin Xu** [1,2,3] **and Xuan Huang** [1,2,3,]*****

[1]   Provincial Key Laboratory of Biotechnology of Shaanxi, Xi'an 710069, China;
      twinklezhang@stumail.nwu.edu.cn (J.Z.); reneeziyu@stumail.nwu.edu.cn (Z.R.);
      desertree0302@163.com (Y.Z.); 0mazheng0@stumail.nwu.edu.cn (Z.M.);
      mayanqin@stumail.nwu.edu.cn (Y.M.); dairuhou@stumail.nwu.edu.cn (D.H.); ziqinxu@nwu.edu.cn (Z.X.)
[2]   College of Life Sciences, Northwest University, Xi'an 710069, China
[3]   Key Laboratory of Resource Biology and Biotechnology in Western China, Ministry of Education,
      Xi'an 710069, China
*   Correspondence: xuanhuang@nwu.edu.cn; Tel.: +86-29-8830-2411; Fax: +86-29-8830-3572

check for updates

Abstract: The circadian clock in plants synchronizes biological processes that display cyclic 24-h oscillation based on metabolic and physiological reactions. This clock is a precise timekeeping system, that helps anticipate diurnal changes; e.g., expression levels of clock-related genes move in synchrony with changes in pathogen infection and help prepare appropriate defense responses in advance. Salicylic acid (SA) is a plant hormone and immune signal involved in systemic acquired resistance (SAR)-mediated defense responses. SA signaling induces cellular redox changes, and degradation and rhythmic nuclear translocation of the non-expresser of *PR* genes 1 (NPR1) protein. Recent studies demonstrate the ability of the circadian clock to predict various potential attackers, and of redox signaling to determine appropriate defense against pathogen infection. Interaction of the circadian clock with redox rhythm promotes the balance between immunity and growth. We review here a variety of recent evidence for the intricate relationship between circadian clock and plant immune response, with a focus on the roles of redox rhythm and NPR1 in the circadian clock and plant immunity.

Keywords: plant immunity; circadian clock; SA-signaling network; NPR1; redox rhythm; NADH/nicotinamide adenine dinucleotide phosphate (NADPH); reactive oxygen species (ROS)

1. Introduction

Most eukaryotic and some prokaryotic organisms have some type of circadian clock that functions as an internal timing mechanism for the control of various normal physiological reactions under light/dark cycle, such as metabolism, hormone levels, body temperature, permitting vision, and brain activities processes [1–5]. The circadian clock is a rather robust system, especially in detecting the precise time of day processes in many metabolic and physiological reactions to help plants make the right reaction at the right time. Furthermore, it drives an endogenous oscillating rhythm strictly observe a period of approximately 24 h even if it is under continuous light or dark [6–9]. Diverse physiological pathways are connected with the circadian clock, including ion homeostasis, hormone signaling, sugar sensing, photoperiodic flowering and stress signaling. Increasing evidence has proved that plant immune response are also connected with the circadian clock [10–12]. Preserving circadian clock regulation during the day increases carbon dioxide fixation, growth, and fitness [13,14]. In addition, at least 30% of genes expression with robust oscillations in *Arabidopsis* are associated with physiological rhythms driven by the circadian clock [15].

Plants have evolved highly efficient defense mechanisms against a variety of major pathogens (*e.g.*, *Pseudomonas syringae*, *Golovinomyces cichoracearum*, *Botrytis cinerea*, *Alternaria brassicicola*) [16–18]. There are two "branches" of triggered immunity in plants. (i) PAMP/MAMP-triggered immunity (PTI/MTI) is triggered by conserved pathogen or microbe-associated molecular patterns (PAMPs, MAMPs) [19]. Specific pattern recognition receptors (PRRs), such as EF-Tu receptor (EFR) and FLAGELLIN-SENSING 2 (FLS2) [20], recognize PAMPs or MAMPs and trigger PTI/MTI for resistance to biotrophic cellular pathogens, viruses, and insects [21]. (ii) Effector-triggered immunity (ETI), which is triggered by specific recognition between pathogen effectors and immune receptors in a host cell, generally encompasses a programmed cell death at the site of pathogen recognition known as the hypersensitive response (HR), and the receptors encoded by resistance genes (*R* genes) [22–24]. Subsequently (hours or days later), systemic acquired resistance (SAR) occurred following HR, which evidently result from cooperative activation of numerous genes termed pathogenesis-related (*PR*) genes [25–28]. Salicylic acid (SA), a plant hormone involved in development and basal resistance against multiple pathogens, is a key regulator of pathogen-induced SAR [29–34].

Numerous studies indicate a relationship among circadian clock, plant immunity, and reduction-oxidation (redox) rhythm [35–37]. Accumulation of reactive oxygen species (ROS) induced by PTI may prevent pathogen invasion [38]. ETI induces programmed cell death associated with changes in nicotinamide adenine dinucleotide phosphate (NADPH) oxidase activity that has been evidenced as the source of ROS [39]. Non-expresser of *PR* genes 1 (*NPR1*, also known as *NIM1* and *SAI1*) [40] regulates expression of clock genes and is also involved in plant defense response through its role in SA signaling [41,42]. In coordination with circadian clock changes, SA regulates expression of downstream defense genes in response to pathogen behavior [43]. In this article, we review interactions between circadian clock and plant immune system, we focus on recent advances in rhythmic oscillation of both redox and clock, and on metabolic and physiological reactions that promote balance of plant immunity and growth.

2. Relationship between Circadian Clock and Immune Responses

The core element of the circadian systems is an oscillator based on a transcription-translation negative feedback loop [44,45]. The circadian clock regulates plant innate immunity to invasion by exogenous pathogens. The coordination between biotrophic pathogens and host provides a daily schedule for plants to defend against exogenous pathogens, that plant defense may related to circadian clock. [6,8,46]. However, the link between them has never been firmly established.

2.1. The Core Loop of Circadian Clock

A. thaliana has a core loop structure of interrelated morning and evening loops which encodes molecular components of the circadian oscillator. Two partially redundant morning-phased genes, *CCA1* (*CIRCADIAN CLOCK ASSOCIATED 1*) [47] and *LHY* (*LATE ELONGATED HYPOCOTYL*) [48] have transcript and protein levels that peak in the morning and activate expression of *PRR* family genes (*PRR5*, *PRR7*, and *PRR9*/NI; *PSEUDO-RESPONSE REGULATORs 5, 7, 9*/night inhibitor) through direct association with their promoters [49,50]. *PRR* family genes also suppress *CCA1* and *LHY* expression by binding to their promoters. *PRR1*, the eponymous member of the *PRR* family, is also referred to as evening-phased *TOC1* (*TIMING OF CAB 1*) [51,52]. In antiphase to *TOC1* peak in early evening, ~40% of TOC1 target genes have an early morning phase regulated by circadian clock [53].

Loss of core loop elemental genes notably speeds up the clock [54]. Feedback loops have been shown to form the circadian clock and to regulate rhythms of output genes whose expression is reflected in behavioral and physiological rhythms in response to environmental changes throughout the day [55,56]. LHY and CCA1 have both been shown to regulate *TOC1*, *LHY*, and *CCA1* gene expression negatively (Figure 1) [47,56,57]. TOC1 acts as a repressor that directly binds to *CCA1* and *LHY* promoters in early evening [58,59]. Mutations of *CCA1* (*cca1-1*), *LHY* (*lhy-20*) and *TOC1* (*toc1-2*) did not completely disrupt clock rhythm. Loss-of-function *Arabidopsis* mutants, in comparison with

wild-type, showed shorter appropriate duration of circadian rhythms and reduced fitness [13,52,60–62]. Semi-dominant *toc1-1* mutant revealed the strong effect of TOC1 on clock-controlled output processes (Table 1) [63]. CCA1 and LHY proteins evidently have analogous functions and, even if one of them is absent, the other is able to maintain circadian rhythm.

Table 1. Core loop genes are direct regulators of plant defense.

Gene Name	CCA1		LHY		TOC1
Expression phase	Morning		Morning		Evening
Clock phenotypes	Shorter periods		Shorter periods		Shorter periods
Relationship to core loop	Negatively regulates *TOC1*, *LHY* and *CCA1* gene expression		Negatively regulates *TOC1*, *LHY* and *CCA1* gene expression		Repressor directly binding to *CCA1* and *LHY* promoters in early night
Expression influencing factors	Affected by flg22, RPP4, and infection by *P. syringae*, *H. arabidopsidis*, and *B. cinerea*		Increased by SA		Increased by SA and suppressed by mutations disrupting *ICS1*, *NPR1*, or ROS signaling
Types	Mutants	Overexpression	Mutants	Overexpression	Mutants
Susceptibility	More susceptible to *P. syringae*, *H. arabidopsidis*, and *B. cinerea*	More susceptible to *P. syringae*, *T. ni* More resistant to *H. arabidopsidis*	More susceptible to *P. syringae*, *H. arabidopsidis*, and *B. cinerea*	More susceptible to *P. syringae*	Higher SA-induced resistance to *P. syringae*
References	[35,47,51,57,61,62,64–68]		[35,47,51,57,61,62,65,68]		[13,35,42,45,52,58,59, 65,69–72]

2.2. Regulation of Plant Immunity by Circadian Clock/Rhythm

Numerous studies have demonstrated direct regulation of plant immunity by circadian clock. PTI and ETI will be triggered through regulating evening-phased elements in plants involved in the circadian clock.

2.2.1. *CCA1* Reduces Plant Susceptibility to Pathogens

Clock gene *CCA1* suppresses plant susceptibility to pathogens. It enhances pathogen resistance at dawn, as evidenced by changes in loss-of-function mutants of susceptibility to *Hyaloperonospora arabidopsidis* (*Hpa*) Emwa1, the cause of downy mildew disease in *Arabidopsis* leaves (Figure 1) [73,74]. *CCA1* encodes the transcription factor that contain a single MYB domain. Resistance to downy mildew was enhanced in a *CCA1* overexpression mutant (*CCA1*-ox) (Table 1) [47]. Resistance to pathogens at dusk is reduced in *CCA1* deletion mutants (Table 1) [75]; these plants show greater resistance in the morning and greater susceptibility in the evening. Rhythmic susceptibility changes throughout the day in *Arabidopsis* interaction with virulent biotrophic pathogen *P. syringae pv. tomato* DC3000 (DC3000), which triggers PTI, were not observed in *CCA1*-ox and *EARLY FLOWERING 3-1* mutants (*elf3-1*), indicating the ability of circadian clock to regulate the host immune response [76]. Defense response of ETI is more intense than that of PTI. Some plants recognize effector molecules secreted by pathogens through *R* genes, and then activate ETI to strengthen the defense response [30].

(1) *CCA1*, *LHY*, and *TOC1* form the core loop of transcription-translation feedback loops that regulate daily timekeeping. *CCA1* and *LHY* have transcript and protein levels that peak in the morning, *TOC1* peaks in early evening and acts as a repressor that directly binds to *CCA1* and *LHY* promoters. ~40% of TOC1 target genes have an early morning phase regulated by the circadian clock. LHY and CCA1 have both been shown to regulate *TOC1* negatively. (2) *CCA1* and *LHY* activate expression of *PRR* family genes through direct association with their promoters. *PRR* family genes also suppress *CCA1* and *LHY* expression by binding to their promoters. (3) *CCA1* and *LHY* regulate *GRP7*, a key constituent of a slave oscillator that modulates stomatal activity related to plant immune response, is involved in regulation of plant immunity. Stomatal opening at dawn and

closure at noon. (4) Morning-phased *LHY* and *CCA1* positively regulate resistance against oomycete pathogen *Hpa* and bacterial pathogen *P. syringae*, whereas evening-phased *TOC1* negatively regulates resistance against bacteria. Spore formation of oomycete pathogen occurs mainly at night, and spores, therefore, germinate at dawn. (5) Maximal JA accumulation occurs around midday, whereas SA peaks around midnight, cytosolic NPR1 suppressed JA signaling. (6) NPR1 plays essential roles in binding of SA. NPR1 oligomers become monomers under pathogen infection, and this process triggers SA accumulation. In the absence of SA or pathogen challenge, NPR1 is degraded by the proteasome and its inhibitory effect on effector-triggered cell death and anti-pathogen defense is eliminated. (7) Under anti-pathogen defense response or SA induction, NPR1 is converted into monomers, which can be combined with TGA2 transcription factor to promote *PR* gene expression, and are released (transferred) from cytoplasmic sites to the nucleus. GSNO facilitates Cys156 disulfide bonding and promotes NPR1 oligomer formation. Both TRX-3h and TRX-5h catalyze monomerization of NPR1 and prevent its repolymerization.
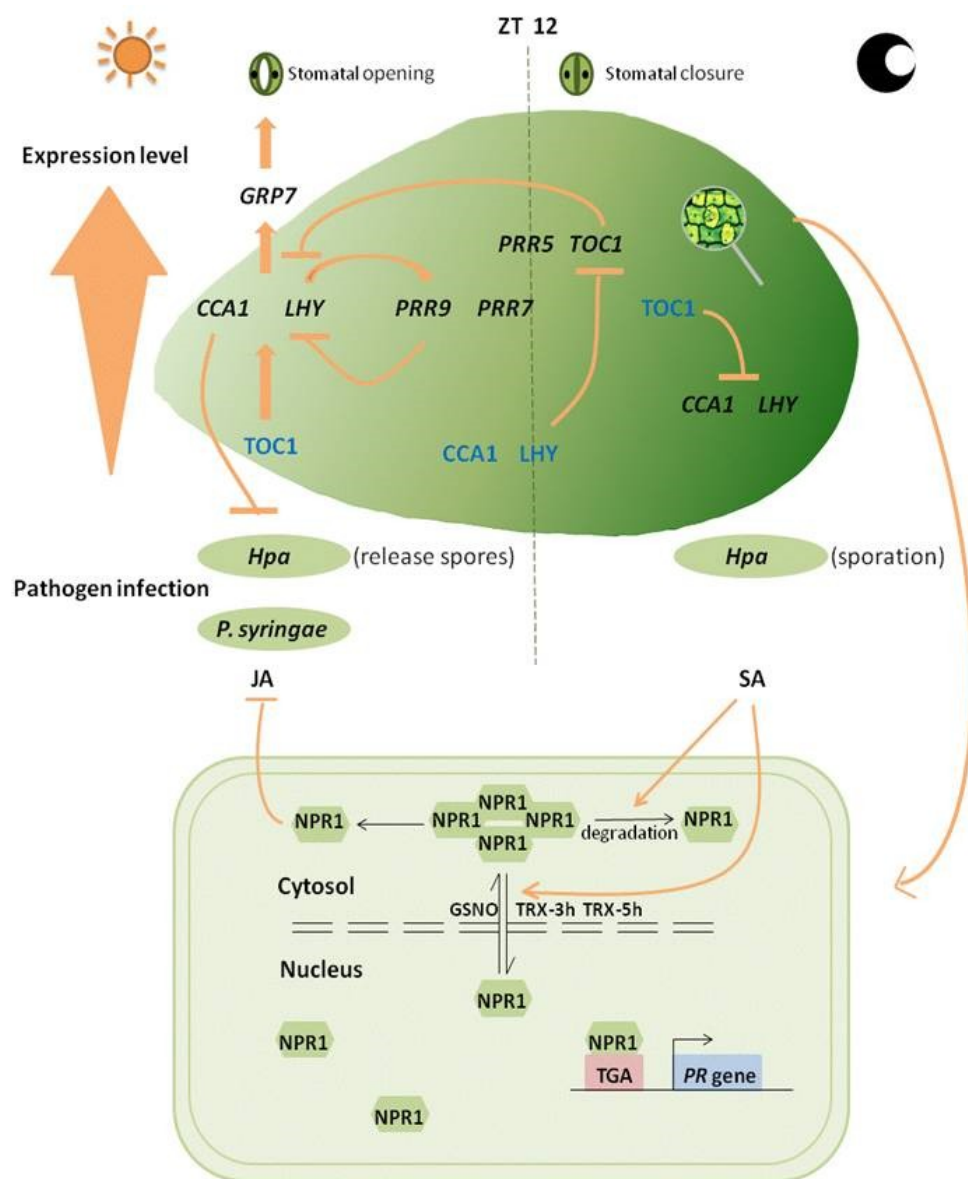

Figure 1. Circadian clock genes changes affect plant defense.

Even in the absence of pathogen invasion, plants are programmed to deal with pathogen infection on the basis of a circadian clock schedule. Appropriate clock function plays a key role in defense responses. Spore formation of oomycete pathogen occurs mainly at night, and spores therefore germinate at dawn [77]. Defense response-related genes have adapted to such timing by peaking around midnight and dawn (Figure 1); an example is *CCA1*, overexpression of which enhances resistance in *Arabidopsis* defense response [47]. *Hpa* EmwaI defense in *Arabidopsis* stimulated not only programmed cell death but also basal resistance through *RECOGNITION OF PERONOSPORA PARASITICA 4* (*RPP4*), whose promoters show close association with the circadian clock. Expression of *RPP4* and its target genes shows broad overlap with CCA1 rhythm, CCA1 can promote the expression of the target defense genes related to programmed cell death (PCD) by activating RPP4 directly [64]. Certain other RPP4-target defense genes independent of *RPP4* can also be activated by regular changing CCA1 level for basal defense. The dual activation mode is resulting in a more powerful and efficient defense response [12].

2.2.2. Does *CCA1* Regulate Non-Clock-Related Activities?

Does *CCA1* plays a role in regulation of non-clock-related activities for enhancement of anti-pathogen defense responses? As an output of the circadian clock, plant immune response is regulated by the clock under both constant light and circular lighting (12 hr light/12 hr dark). Under both these light conditions, susceptibility to avirulent *P. syringae* strain was greater for *CCA1*-ox plants than for wild-type, the opposite of this, *CCA1*-ox plants showed an enhanced resistance to *Hpa* [46,62]. Both *cca1-1/lhy-20* double mutant and *LHY*-ox plants showed enhanced susceptibility to avirulent *P. syringae*, and *cca1-1/lhy-20* double mutant also showed enhanced susceptibility to oomycete pathogen *Hpa* strains under circular lighting. Although *CCA1* and *LHY* mutants showed similarly shortened circadian period, *CCA1*-ox plants show enhanced immune response to *Hpa*, but LHY does not show a defense response against *Hpa* [64]. These findings suggest that the circadian clock regulates plant innate immunity as an output under both light conditions, however, whether the enhanced resistance to *Hpa* in *CCA1*-ox plants is caused by non-clock function resulting from *CCA1* overexpression is still unclear.

2.2.3. Interactions between Plant Immunity and Circadian Clock/Rhythm

Mutual regulation between the circadian clock and plant immune response was observed in the clock-related gene mutants mentioned above, and further investigation showed that defense response was under the control of the circadian clock. When circadian clock rhythm resulting from *CCA1* or *LHY* overexpression was disrupted, susceptibility to *P. syringae* strain was greatly enhanced. *CCA1* and *LHY* regulate their downstream target gene *GLYCINE-RICH RNA-BINDING PROTEIN 7* (*GRP7*), which is involved in regulation of plant immunity and is a key constituent of a slave oscillator [78–80] that modulates stomatal activity related to plant immune response [81]. Interestingly, the circadian clock regulates plant defense response, and defense response in turn regulates the circadian clock. For example, treatment of avirulent *P. syringae* strain infection or of flg22 (a resulting flagellin protein) is associated with modulation of clock activity [62].

SA can be transiently produced upon infection [82]. A previous finding has shown that the amplitude of clock rhythms will increase under SA treatment [42]. In contrast to this, a lately study observed both SA treatment and *Pst* DC3000 infection decrease the amplitude of clock rhythms [83]. The sharp different result may caused by different SA concentrations or plants age [84,85]. Notably, SA treatment leads to a negative phase shift, which instead a slight phase advance in *Pst* DC3000 infection plants, only *Pst* DC3000 infection plants lengthened the clock period, which has no evident change under SA treatment [83]. Altogether, these data suggests that the ultimate clock phenotypes show in infection plants is due to SA-dependent and -independent pathways integration.

The stress- and defense-related hormone JA undergoes a rhythm oscillator as well. The clock regulates JA levels with a peak in the midday [69]. In *Arabidopsis*, the clock protein CCA1 are able to

bind to the JA biosynthetic gene promoter *LIPOXYGENASE2* [86]. The circadian clock can regulate the expression of the JA receptor *CORONATINE INSENSITIVE1*, as well as the expression of *MYC2*, a positive transcription factor of JA signaling. TIME FOR COFFEE (TIC) acts as a key determinant in circadian clock, also acts as a negative transcription factor in JA-mediated defense responses. It was found to inhibit the transcription of *CORONATINE INSENSITIVE1* to repress MYC2 protein accumulation [87].

3. Plant Immune Responses Involving Salicylic Acid (SA) and Jasmonic Acid (JA)

The plant hormone SA is produced in response to biotrophic pathogens invasion and induces SAR and JA is essential for the defense against necrotrophic pathogens and herbivorous insects [31]. Oscillation of hormones level plays a key role in proper coordination of immune response and plant development. The SA-signaling network involves transcription co-activators NPR1, NPR3, and NPR4. Whether NPR1 is a receptor of SA remains unclear [88,89].

3.1. Role of SA and JA in Coordinating Plant Immune Response and Growth

Recent chromatin immunoprecipitation studies indicate that morning-phased *CCA1*, evening-phased *TOC1*, and *PPR* family genes can combine with hundreds of gene promoters [59,86,90–92]. Circadian clock-related elements are regulated directly by plant hormones such as SA and jasmonic acid (JA), not by chance [93]. Maximal JA accumulation occurs around midday, whereas SA peaks around midnight [69]. *ISOCHORISMATE SYNTHASE 1* (*ICS1*) is a direct clock target gene that encodes a key enzyme involved in SA biosynthesis. *ICS1* expression is controlled by *CCA1 HIKING EXPEDITION* (*CHE*), a transcription factor of evening-phased elements [65]. *ICS1* gene expression, like SA accumulation, peaks at midnight and is involved in pathogen infection in the morning.

Morning-phased *LHY* and *CCA1* positively regulate resistance against oomycete pathogen *Hpa* and bacterial pathogen *P. syringae* [62,87], whereas evening-phased *TOC1* negatively regulates resistance against bacteria. With SA treatment in the subjective morning or evening under 3 h constant light, defense-related genes showed increased expression mainly 3 h after morning SA application [42], whereas genes related to growth and development showed increased expression mainly after evening SA application. SA treatment has a consistent effect on redox rhythm, and morning application has a positive effect on plant defense [87,94], whereas evening application has a negative effect. Such a shift of plant defense during the course of a day may reflect adaptation to changing diurnal physical conditions; temperature and humidity in the morning are typically more conducive to pathogen challenge [64]. The clock system also regulates plant defenses via other clock-related genes such as phosphate transporter gene *PHOSPHATE TRANSPORTER 4;1* (*PHT4;1*), which *is* regulated by *CCA1*, a negative regulator of SA signaling [95]. SA functions in both local defense against biotrophs and as an essential signal in distal organization for broad-spectrum SAR [33].

A functional JA signaling pathway is required for oscillation in susceptibility to the *fungus Botrytis cinerea* [68]. In *Arabidopsis*, SA produced during pathogen infection strongly antagonizes both JA biosynthesis and JA-responsive gene expression, resulting in a large set of JA-responsive genes, including *LIPOXYGENASE2* (*LOX2*), *VEGETATIVE STORAGE PROTEIN (VSP)*, and *PLANT DEFENSIN 1.2* (*PDF1.2)* down regulated [93]. Plants may show negative responses to inappropriate stimuli; e.g., inappropriate induction of immunity at night may inhibit plant growth. Experimental SA treatment of plants under constant dark conditions helped maintain stronger circadian rhythm, but caused a major reduction of fresh weight. SA treatment also had an inhibitory effect on the expression of various defense-related aquaporin genes. An aqueous environment is generally essential for pathogen virulence. Thus, timing of SA-mediated immune responses in the morning helps avoid conflict between SA immune response and growth-related activities that require water transport at night [96].

3.2. Is NPR1 an SA Receptor?

Recently, some data clearly pointed out that NPR1 has high-affinity to bind with SA [97]. Increasing evidence strongly supports the idea that NPR1 plays a key role in SA signaling and as an SA receptor.

3.2.1. Molecular Structure of NPR1

The *NPR1* gene was first discovered through genetic screening of *Arabidopsis* mutants showing defects of SA-mediated pathogen resistance [24,25,91,98,99]. NPR1 was cloned as a novel protein containing a bipartite nuclear localization signal, two protein–protein interaction domains, N-terminal bric-a-brac, tramtrack, and BTB/POZ (broad-complex, tramtrack, and bric-à-brac/poxvirus, zinc finger) domains, and an ankyrin-repeat domain with diverse functions [100–102]. The BTB/POZ domain is found in proteins that function as substrate adapters of CUL3-based ubiquitin ligases for the degradation of specific substrates [103].

3.2.2. NPR1 Signal Transduction Pathway

Both increase of endogenous SA levels and exogenous SA application lead to gene transcription that is greatly altered under pathogen induction [104]. NPR1, a transcription co-activator known as a "master" regulator of plant immunity, triggers change of SA level. NPR1 function requires direct combination of the transition metal copper with SA, similar to the requirement of phytohormone ethylene for copper [105]. The SA/NPR1 complex is sensitive to chelation by Ethylene Diamine Tetraacetic Acid (EDTA), and the combination between SA and NPR1 could not be detected until testing of equilibrium dissociation constant (Kd) for incubation of SA with NPR1, coupled to the solid phase. The C-terminus of NPR1 contains two Cys residues (Cys521, Cys529) that play essential roles in binding SA to NPR1 [88].

The above findings clearly indicate that NPR1 is the SA receptor, and that they bind directly to each other. Partially reduced NPR1 oligomers become monomers under pathogen infection, and this process triggers SA accumulation (Figure 1) [33]. In the absence of SA or pathogen challenge, NPR1 is degraded by the proteasome and its inhibitory effect on effector-triggered cell death and anti-pathogen defense is eliminated [84]. For this, NPR1 requires Cullin-3 (CUL3)-based E3 ubiquitin ligase, a ligand with significant binding activity that can combine BTB domain-containing proteins [100,106–108]. NPR1 does not interact directly with CUL3, and it has, therefore, been proposed that association of CUL3 with NPR1 may be mediated by other adaptors that have the same domain as NPR1 [109]. NPR3 and NPR4, homologues of NPR1 that contain BTB domains, are good candidate adaptors [84,110]. *npr3-1/npr4-3* double mutant showed enhanced level of *PR* gene expression regardless of pathogen induction, and reduced susceptibility to both *P. syringae* pv. *maculicola* (*P.s.m.*) ES4326 and *Hyaloperonospora parasitica* Noco2. NPR1 protein stability thus makes the *npr3-1/npr4-3* double mutant insensitive to SAR induction. Binding affinity to SA is higher for NPR4 (K_d = 23.54 ± 2.742 nM) than for NPR3 (K_d = 176.7 ± 28.31 nM) [84,88,111]. Pathogen induction results in generation of an SA gradient whereby NPR1 accumulates in adjacent cells and promotes cell survival and SA-mediated resistance [112]. NPR1 may be degraded by NPR3 or NPR4 under stress, with suppression of programmed cell death during ETI at an infection site [23,89]. Surprisingly, the latest study shows that NPR1 functions separately with NPR3/NPR4. NPR3/NPR4 and may bind to SA at low levels to repress defense gene expression, the transcription co-activator NPR1 will promote defense gene expression on the accumulation of SA. Although both NPR1 and NPR3/4 are SA receptors, they function differently in transcriptional regulation of SA-induced defense gene expression, a NPR1 mutant loss the function in binding SA, it promotes SA-induced defense gene expression, whereas a NPR4 mutant represses SA-induced immune responses [97].

These findings, taken together, demonstrate the essential role of NPR1 as a receptor of SA, and also negative NPR3 and NPR4 controlling protein levels of NPR1 in plant immunity [84].

3.3. Plant Defense Network Is Controlled by Cytosolic NPR1

NPR1 as a key regulatory factor in the cross-talk between SA and JA signaling. Interaction between NPR1 and transcription factor TGA1 roles in the SA signaling, nuclear localization of NPR1 is not required for a plant defense network between SA and JA signaling, and therefore, the network is modulated by the NPR1 stays in cytosol [113]. Once the necrotrophic pathogens *Alternaria solani* and *Botrytis cinerea* are infected, the SA pathway will be manipulated through NPR1 and two JA-dependent defense genes expression including *PROTEINASE INHIBITORS I* and *II* will be suppressed for disease symptom development [68].

In brief, both JA and SA are of vital importance in plant immunity and thus coordinates the plant defense as well as growth regulation through various mechanisms.

4. Central Role of NPR1 in SA-Mediated Plant Defense Response

NPR1 is a key regulator of the SA-signaling network. SA induces degradation of NPR1, and NPR1 activates *PR1* transcription by combining with TGA factors following the transfer to the nucleus [33], as detailed in this section.

4.1. Role of NPR1 in Plant Immunity

Plant evolution involves continuous improvement and sophistication of immune systems. As has been talked above (2.2), *NPR1* shows defects in SA-mediated pathogen resistance on *Arabidopsis* mutants when it was first discovered; studies during the past two decades have clarified the role of NPR1 as a key transcriptional regulator affecting a wide variety of host genes involved in immune responses and biocompatibility [27,109,114]. This concept is supported by NPR1 overexpression experiments in *A. thaliana*. The conserved function of NPR1 has also been demonstrated in crop species such as rice, wheat, tomato, and apple [115]. Overexpression experiments with *Arabidopsis NPR1* (*AtNPR1*) homologues [116] in these species have revealed powerful resistance against fungal and bacterial pathogen attacks [117–119].

Two-hybrid screening studies of a yeast model have shown that NPR1 interacts with TGA transcription factors through its ankyrin repeat domain [120], and also acts as a transcription co-activator to regulate SAR gene expression. SA-mediated activation of *PR* genes is required for binding activity of NPR1 and TGA transcription factors to promoter elements [121,122]. TGA/OBF family members containing bZIP can also interact with NPR1 [120,121,123,124]. Following the binding of essential transcription factors to as-1 component, the *PR* gene promoter begins transcription and expression to initiate hypersensitive responses. NPR1 clearly functions as a key positive regulator in SA signaling, as well as a negative regulator in JA signaling [113].

4.2. Transfer of NPR1 Degradation Products to Nucleus

Under anti-pathogen defense response or SA induction, NPR1 is converted into monomers, which can be combined with TGA2 transcription factor to promote *PR* gene expression, and are released (transferred) from cytoplasmic sites to the nucleus (Figure 1) [24,25,70,99,125].

NPR1 negatively regulates programmed cell death. Occurrence of ETI in pathogen-infected cells leads to high SA accumulation, followed by NPR1 regulation of its target genes, CUL/ NPR3-mediated NPR1 turnover, and activation of cell death. CUL3/ NPR4-mediated degradation of NPR1 around infected cells is necessary to avoid unnecessary activation of resistance [70]. This event must occur prior to NPR1 regulation of its target genes, because NPR1 is a negative regulator of cell death by CUL3/ NPR4. Cys156 is the essential amino acid residue for NPR1 oligomerization. NPR1 can be assembled into a high-molecular weight oligomer in cytoplasm through S-nitrosylation among its Cys156 residues [126,127]. S-nitrosoglutathione (GSNO) facilitates Cys156 disulfide bonding and promotes NPR1 oligomer formation (Figure 1). Following SAR induction, oligomers in cytoplasm

release large amounts of NPR1 monomers which are transferred to the nucleus and subsequently turned over [128].

NPR1 monomer is preferentially degraded relative to NPR1 oligomer, and in *35S::NPR1-GFP* plants, NPR1-GFP oligomer's being converted to monomer is accelerated [70,102,128]. In the absence of new protein synthesis, the NPR1-GFP level is reduced or unchanged. In the presence of dexamethasone (DEX), it promotes nuclear translocation of a NPR1 fused to a DEX-responsive glucocorticoid receptor (NPR1-GR) and rapidly degraded [102,128]. In the absence of DEX, NPR1-GR stays localized mainly in cytoplasm. Constitutive degradation of the NPR1 monomer by proteasomes occurs only in the nucleus [128].

4.3. Differences between Systemic Acquired Resistance (SAR)-Induced and SAR-Uninduced Cells

NPR1 is a key regulator of the SA-signaling network during SAR development. Although SAR has important functions in plant immunity, preventing untimely SAR activation is essential for normal growth and development. CUL3-mediated degradation of NPR1 monomer prevents SAR activation in plants not under pathogen challenge [128]. In SA treatment experiments with *35S::NPR1-GFP* plants, NPR1 Ser11/15 phosphorylation level was greatly increased in SA-treated plants, whereas nuclear levels of phosphorylated endogenous NPR1 and NPR1-GFP were low in SA-nontreated plants. Turnover of phosphorylated NPR1 is essential for activation of SAR, and unphosphorylated NPR1 may be degraded in combination with target gene promoters. Gene transcription may be restricted by proteasomes, as a result of the destruction of transcription factors or proteolysis of co-activators, to protect assembly and installation of active transcription complexes [128]. Once the proteasome mediates NPR1 degradation, that SAR will respond stronger but more transiently [43].

4.4. Role of NPR1 in Circadian Clock Core Loop

NPR1 monomer level has recently been shown to peak at night, suggesting that rhythmic translocation of NPR1 to nucleus, controlled by rhythmic oscillation of endogenous SA, may regulate circadian clock genes. NPR1 antagonistics both SA and *P. syringae* infection in the regulation of circadian rhythms. The amplitude reduction and phase delay triggered by a transient SA treatment is prevented by NPR1, the amplitude reduction induced by *Pst* DC3000 infection was also enhanced in *npr1-1* mutant plants [83]. NPR1 may function as an intrinsic regulator of clock gene *TOC1* in response to rhythmic accumulation of endogenous SA [69]. NPR1 is most likely not the only regulator of *TOC1*. Treatment with 1 mM SA on 3-week-old soil-grown wild-type or on *NPR1-3* mutant plants does not significantly alter the rhythm cycle. TOC1 expression level is increased immediately by SA induction [42]. Reduction of TOC1 level shortens the rhythm cycle, whereas increase of the TOC1 level lengthens the cycle [71,129]. Other clock genes such as *LHY*, in contrast to *TOC1*, are sensitive to SA induction. The regulatory mechanism of clock genes in relation to plant immunity was elucidated by experiments using *LHYp:LUC* reporter (*LHYp (TBSm):LUC*). *LHY* expression peaked at dawn, while *TOC1* expression was delayed by SA treatment, because *LHY* is an antagonist of *TOC1* in the clock. NPR1 will strengthen the circadian clock when the redox rhythm is disrupted by challenges, through regulation of both morning-phased *LHY* and evening-phased *TOC1* [69].

5. Plant Immunity and Redox Rhythm as Related to the Circadian Clock

Redox rhythm is closely associated with plant immunity, and both are related to the circadian clock. Early studies of the fungus *Neurospora crassa* suggested regulation of cellular redox state by the circadian clock [130]. Daily timekeeping functions are driven by both transcription-translation feedback loops (circadian clock) and non-transcriptional redox oscillations in numerous organisms, including *Arabidopsis* [2–4]. Nicotinamide adenine dinucleotide oxidation state (NAD/NADH ratio) also presents a daily rhythm.

5.1. Reactive Oxygen Species (ROS) Oscillation Triggers Plant Immunity

The alteration of cellular redox state is one of the earliest occurring responses in a challenged cell [131]. ROS (e.g., H_2O_2) and reactive nitrogen species (RNS; e.g., nitric oxide [NO]) have closely related roles in plant immunity [132–134]. Plant immune responses are effectively triggered only if ROS and RNS signaling are activated simultaneously. The balance between ROS and RNS production is crucial in determining the fate of infected cells, and the success of plant immune responses [133,135].

Morning-phased *CCA1* controls ROS homeostasis by regulating expression of catalases (e.g., catalase mutant *CAT2*) involved in plant defense, Alvina et. al., pointed that *CCA1* is a master regulator of ROS homeostasis (Figure 2) [35]. In *Arabidopsis*, inhibition of *CAT2* activity by SA leads to increased H_2O_2 level (Figure 2) and subsequent sulfenylation (oxidation of thiol -SH into sulfenic acid -SOH) of tryptophan synthetase β-subunit 1 (TSB1) [136]. In contrast to historical and multigenerational circadian clocks based on differing transcription-translation feedback loops, the peroxiredoxin active site (responsible for redox rhythms) is conserved in a wide variety of organisms, indicating a possible origin of redox-based circadian clocks [2,96].
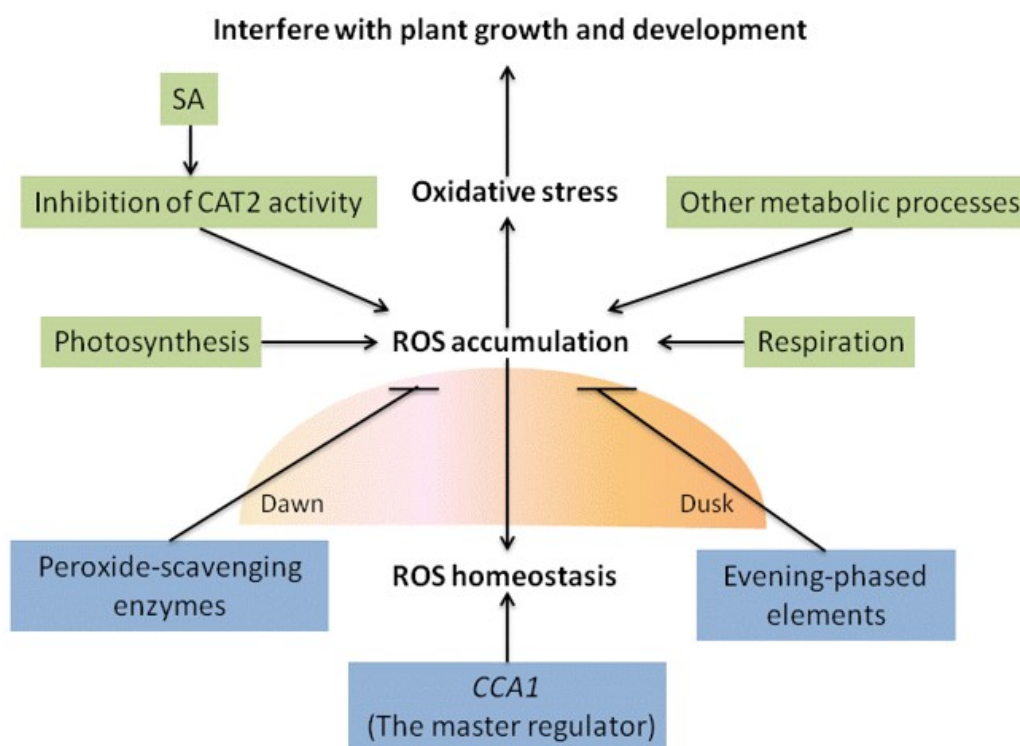
Figure 2. Reactive oxygen species (ROS) homeostasis. ROS, which are generated during photosynthesis, respiration, and other metabolic processes, peak around midday and are toxic to interfere with plant growth and development. *CCA1* is a master regulator of ROS homeostasis. In *Arabidopsis*, inhibition of *CAT2* activity by SA leads to increased H_2O_2 level. Oscillation of peroxide-scavenging enzyme levels is upregulated close to subjective dawn and inhibits ROS production. Evening-phased elements in promoters of antioxidant genes involved in transcriptional regulation were found to mitigate oxidative stress.

ROS, which are generated during photosynthesis, respiration, and other metabolic processes, and peak around midday, are toxic and can interfere with plant growth and development (Figure 2). In association with plant physiological responses, the level of redox species (which scavenge ROS) peaks at around the same time. Oscillation of peroxide-scavenging enzyme (antioxidant) levels is upregulated close to subjective dawn (Figure 2). ROS production can be inhibited by peroxide-scavenging enzymes (Figure 2). Circadian clock may, therefore, regulate ROS-related genes

to prevent changes in ROS homeostasis [137,138]. Experiments on an *Arabidopsis* model to test the hypothesis that the antioxidant network is regulated by the circadian clock at the transcriptional level revealed that the ROS homeostasis regulon transcription was altered in *CCA1* and *LHY* mutants. Evening-phased elements in promoters of antioxidant genes involved in transcriptional regulation were found to mitigate oxidative stress (Figure 2). These findings support the concept that central oscillators control oxidative stress through transcriptional regulation [35,139].

5.2. NADP/NADPH Oscillation Reflects Redox Rhythm

Circadian redox rhythm in anucleate red blood cells (erythrocytes) is reflected in oscillations of both oxidation state of ROS scavenger peroxiredoxins and NADP/NADPH levels, both of which lack transcription/translation activities [3,96]. Redox rhythm is reflected mainly in regular changes of redox coenzymes NADPH and NADP$^+$. Zhou et al. [42] found that, under constant light, NADPH peaked before subjective dawn while NADP$^+$ peaked before subjective dusk. Redox rhythm was sensitive to external perturbation; e.g., SA treatment disrupted NADPH/NADP$^+$ rhythms and altered their proportions to induce expression of defense-related genes.

5.3. Oscillation of NPR1 Regulation Pathway

Because the redox rhythm is regulated by NPR1, circadian redox rhythm in plants is coupled with a genetic clock. This coupling is mediated by cellular redox changes induced by endogenous SA level, which induce degradation and rhythmic nuclear translocation of NPR1 [69,70]. Endogenous SA levels oscillate in a circadian pattern [69]. Reduction by SA and glutathione of ethyl ester (GSHmee), a redox-altering reagent, enhanced expression of evening-phased *TOC1* in an NPR1-dependent manner [140]. NPR1 is a general redox sensor that modulates clock gene *TOC1*. NPR1 monomer level peaks at night and displays a circadian oscillation pattern. Basal expression of *TOC1* was reduced in both an *NPR1* mutant and in an SA mutant [100]. Supplemental SA increased *TOC1* expression in the SA mutant but not in the *NPR1* mutant, regardless of treatment duration [42].

Redox signaling in plants helps determine the most suitable defense against pathogen infection, and the circadian clock helps predict various potential/ imminent attackers [132,141,142]. Upon pathogen challenge in *Arabidopsis*, stomatal closure is induced by NADPH oxidase RESPIRATORY BURST OXIDASE HOMOLOGUE D (RBOHD) [143,144]. SA accumulation is induced by RBOHF, another NADPH oxidase [145]. Expression of SA-dependent defense genes is induced by ROS produced by apoplastic peroxidases PRX33 and PRX34 [146–149]. Both stomatal closure and defense gene induction required NO, produced by NO synthase-like activity [96,150–152]. NO functions as a redox regulator to promote rhythmic nuclear translocation of NPR1, and thus plays an important role in the modulation of plant immunity [153].

Reduced glutathione (GSH) and oxidized glutathione (GSSG) form an essential antioxidant buffer system. Reduction of the GSH/GSSG ratio during pathogen infection leads to increased oxidative stress. NPR1 reduction does not lead to an immediate increase of the GSH/GSSG ratio [70]. GSH is, therefore, not a direct reducing agent of NPR1, although it has some correlation with redox changes of NPR1 and GSH/GSSG ratio. Other redox-related molecules such as glutathione and thioredoxin may be involved in NPR1 reduction. Pathogen challenge creates an SA gradient which is associated with changes in cellular redox and reduces the activity of two cytosolic thioredoxins (TRX-3h, TRX-5h) that catalyze monomerization of NPR1 and prevent its repolymerization (Figure 1) [126].

6. Future Prospects

We have summarized here a variety of evidence for connections between circadian clock, plant immunity, and redox rhythm. Cellular redox state is regulated by circadian clock. SA treatment of plants disrupted oscillations of NADPH/ NADP$^+$ and altered their proportions, thereby triggering expression of defense-related genes. ROS have a toxic (biocidal) effect on invading bacteria, but can also injure host cells. NPR1 may be the core element that links together the circadian clock and

redox rhythm. Plant defense systems are associated with the circadian clock for regulation of daily physical activity via a complex network system. Defense responses create an environment conducive to plant survival, but often lead to growth inhibition, thus setting up a perpetual competition for resource allocation. Our understanding of plant immunity and the circadian clock has undergone major, sequential progress during the past decade, and research in both areas is well advanced. On the other hand, SA is the last of the major plant hormones whose receptors have an intricate control of SA responses. Future studies will further elucidate the mechanism of how SA receptors regulate plant defense responses and the core elements that coordinate the circadian clock with plant immunity.

Conflicts of Interest: The authors declare no conflict of interest. The funders had no role in the design of the study; in the collection, analyses, or interpretation of data; in the writing of the manuscript, or in the decision to publish the results.

References

1. Chi-Castañeda, D.; Ortega, A. Glial cells in the genesis and regulation of circadian rhythms. *Front. Physiol.* **2018**, *9*, 88–98.
2. Edgar, R.S.; Green, E.W.; Zhao, Y.; van Ooijen, G.; Olmedo, M.; Qin, X.; Xu, Y.; Pan, M.; Valekunja, U.K.; Feeney, K.A.; et al. Peroxiredoxins are conserved markers of circadian rhythms. *Nature* **2012**, *485*, 459–464. [CrossRef] [PubMed]
3. O'Neill, J.S.; Reddy, A.B. Circadian clocks in human red blood cells. *Nature* **2011**, *469*, 498–503. [CrossRef] [PubMed]
4. O'Neill, J.S.; Van Ooijen, G.; Dixon, L.E.; Troein, C.; Corellou, F.; Bouget, F.Y.; Reddy, A.B.; Millar, A.J. Circadian rhythms persist without transcription in a eukaryote. *Nature* **2011**, *469*, 554–558. [CrossRef] [PubMed]
5. Wright, K.P.; McHill, A.W.; Birks, B.R.; Griffin, B.; Rusterholz, R.T.; Chinoy, E.D. Entrainment of the human circadian clock to the natural light-dark cycle. *Curr. Biol.* **2013**, *23*, 1554–1558. [CrossRef] [PubMed]
6. Greenham, K.; Mcclung, C.R. Integrating circadian dynamics with physiological processes in plants. *Nat. Rev. Genet.* **2015**, *16*, 598–610. [CrossRef] [PubMed]
7. Hemmes, H.; Henriques, R.; Jang, I.C.; Kim, S.; Chua, N.H. Circadian clock regulates dynamic chromatin modifications associated with *Arabidopsis CCA1/LHY* and *TOC1* transcriptional rhythms. *Plant Cell Physiol.* **2012**, *53*, 2016–2029. [CrossRef] [PubMed]
8. Hsu, P.Y.; Harmer, S.L. Wheels within wheels: The plant circadian system. *Trends Plant Sci.* **2014**, *19*, 240–249. [CrossRef] [PubMed]
9. Mcclung, C.R. The genetics of plant clocks. *Adv Genet.* **2011**, *74*, 105–139. [PubMed]
10. Ingle, R.A.; Roden, L.C. Circadian regulation of plant immunity to pathogens. *Methods Mol. Biol.* **2014**, *1158*, 273–283. [PubMed]
11. Bolouri Moghaddam, M.R.; Van den, E.W. Sweet immunity in the plant circadian regulatory network. *J. Exp. Bot.* **2013**, *64*, 1439–1449. [CrossRef] [PubMed]
12. Korneli, C.; Danisman, S.; Staiger, D. Differential control of pre-invasive and post-invasive antibacterial defense by the *Arabidopsis* circadian clock. *Plant Cell Physiol.* **2014**, *55*, 1613–1622. [CrossRef] [PubMed]
13. Dodd, A.; Salathia, N.; Hall, A.; Kevei, E.; Toth, R.; Nagy, F.; Hibberd, J.M.; Millar, A.J.; Webb, A.A. Plant circadian clocks increase photosynthesis, growth, survival, and competitive advantage. *Science* **2005**, *309*, 630–633. [CrossRef] [PubMed]

14. Michael, T.P.; Salomé, P.A.; Yu, H.J.; Spencer, T.R.; Sharp, E.L.; McPeek, M.A.; Alonso, J.M.; Ecker, J.R.; McClung, C.R. Enhanced fitness conferred by naturally occurring variation in the circadian clock. *Science* **2003**, *302*, 1049–1053. [CrossRef] [PubMed]

15. Torres, M.; Becquet, D.; Franc, J.L.; François-Bellan, A.M. Circadian processes in the RNA life cycle. *Wiley Interdiscip. Rev. RNA* **2018**, *9*, e1467. [CrossRef] [PubMed]

16. Eggermont, K.; Mauch-Mani, B.; Vogelsang, R.; Broekaert, W.F. Separate jasmonate-dependent and salicylate-dependent defense-response pathways in *Arabidopsis* are essential for resistance to distinct microbial pathogens. *Proc. Natl. Acad. Sci. USA* **1998**, *95*, 15107–15111.

17. Fabro, G.; Di, R.J.; Voigt, C.A.; Savchenko, T.; Dehesh, T.K.; Somerville, S.; Alvarez, M.E. Genome-wide expression profiling *Arabidopsis* at the stage of *Golovinomyces cichoracearum* haustorium formation. *Plant Physiol.* **2008**, *146*, 1421–1439. [CrossRef] [PubMed]

18. Murray, S.L.; Ingle, R.A.; Petersen, L.N.; Denby, K.J. Basal resistance against *Pseudomonas syringae* in *Arabidopsis* involves WRKY53 and a protein with homology to a nematode resistance protein. *Mol. Plant Microbe Interact.* **2007**, *20*, 1431–1438. [CrossRef] [PubMed]

19. Li, L.; Yu, Y.; Zhou, Z.; Zhou, J.M. Plant pattern-recognition receptors controlling innate immunity. *Sci. China Life Sci.* **2016**, *59*, 878–888. [CrossRef] [PubMed]

20. Gheysen, G.; Mitchum, M.G. How nematodes manipulate plant development pathways for infection. *Curr. Opin. Plant Biol.* **2011**, *14*, 415–421. [CrossRef] [PubMed]

21. Erb, M.; Meldau, S.; Howe, G.A. Role of phytohormones in insect-specific plant reactions. *Trends Plant Sci.* **2012**, *17*, 250–259. [CrossRef] [PubMed]

22. Dodds, P.N.; Rathjen, J.P. Plant immunity: Towards an integrated view of plant-pathogen interactions. *Nat. Rev. Genet.* **2010**, *11*, 539–548. [CrossRef] [PubMed]

23. Rate, D.N.; Greenberg, J.T. The *Arabidopsis* aberrant growth and death 2 mutant shows resistance to *Pseudomonas syringae* and reveals a role for NPR1 in suppressing hypersensitive cell death. *Plant J.* **2001**, *27*, 203–211. [CrossRef] [PubMed]

24. Cao, H.; Bowling, S.A.; Gordon, A.S.; Dong, X. Characterization of an *Arabidopsis* mutant that is nonresponsive to inducers of systemic acquired resistance. *Plant Cell* **1994**, *6*, 1583–1592. [CrossRef] [PubMed]

25. Delaney, T.P.; Ryals, J.A. *Arabidopsis* signal transduction mutant defective in chemically and biologically induced disease resistance. *Proc. Natl. Acad. Sci. USA* **1995**, *92*, 6602–6606. [CrossRef] [PubMed]

26. Uknes, S.; Mauch-Mani, S.; Moyer, M.; Potter, S.; Williams, S.; Dincher, S.; Chandler, D.; Slusarenko, A.; Ward, E.; Ryals, J. Acquired resistance in *Arabidopsis*. *Plant Cell* **1992**, *4*, 645–656. [CrossRef] [PubMed]

27. Wang, D.; Weaver, N.D.; Kesarwani, M.; Dong, X. Induction of protein secretory pathway is required for systemic acquired resistance. *Science* **2005**, *308*, 1036–1040. [CrossRef] [PubMed]

28. Bari, R.; Jones, J.D.G. Role of plant hormones in plant defence responses. *Plant Mol. Biol.* **2009**, *69*, 473–488. [CrossRef] [PubMed]

29. Glazebrook, J. Contrasting mechanisms of defense against biotrophic and necrotrophic pathogens. *Annu. Rev. Phytopathol.* **2005**, *43*, 205–227. [CrossRef] [PubMed]

30. Jones, J.D.G.; Dangl, J.L. The plant immune system. *Nature* **2006**, *444*, 323–329. [CrossRef] [PubMed]

31. Pieterse, C.M.; Van der Does, D.; Zamioudis, C.; Leon-Reyes, A.; Van Wees, S.C. Hormonal modulation of plant immunity. *Annu. Rev. Cell Dev. Biol.* **2012**, *28*, 489–521. [CrossRef] [PubMed]

32. Robertseilaniantz, A.; Grant, M.; Jones, J.D.G. Hormone crosstalk in plant disease and defense: More than just jasmonate-salicylate antagonism. *Annu. Rev. Phytopathol.* **2011**, *49*, 317–343. [CrossRef] [PubMed]

33. Spoel, S.H.; Dong, X. How do plants achieve immunity? Defence without specialized immune cells. *Nat. Rev. Immunol.* **2012**, *12*, 89–100. [CrossRef] [PubMed]

34. Spoel, S.H.; Johnson, J.S.; Dong, X. Regulation of tradeoffs between plant defenses against pathogens with different lifestyles. *Proc. Natl. Acad. Sci. USA* **2007**, *104*, 18842–18847. [CrossRef] [PubMed]

35. Lai, A.G.; Doherty, C.J.; Muellerroeber, B.; Kay, S.A.; Schippers, J.H.; Dijkwel, P.P. *CIRCADIAN CLOCK-ASSOCIATED 1* regulates ROS homeostasis and oxidative stress responses. *Proc. Natl. Acad. Sci. USA* **2012**, *109*, 17129–17134. [CrossRef] [PubMed]

36. Peek, C.B.; Affinati, A.H.; Ramsey, K.M.; Kuo, H.Y.; Yu, W.; Sena, L.A.; IlKayeva, O.; Marcheva, B.; Kobayashi, Y.; Omura, C.; et al. Circadian clock NAD^+ cycle drives mitochondrial oxidative metabolism in mice. *Science* **2013**, *342*, 591–607. [CrossRef] [PubMed]

37. Ramsey, K.M.; Yoshino, J.; Brace, C.S.; Abrassart, D.; Kobayashi, Y.; Marcheva, B.; Hong, H.K.; Chong, J.L.; Buhr, E.D.; Lee, C.; et al. Circadian clock feedback cycle through nampt-mediated NAD$^+$ biosynthesis. *Science* **2009**, *324*, 651–654. [CrossRef] [PubMed]

38. Nürnberger, T.; Brunner, F.; Kemmerling, B.; Piater, L. Innate immunity in plants and animals: Striking similarities and obvious differences. *Immunol. Rev.* **2004**, *198*, 249–266. [CrossRef] [PubMed]

39. Elmayan, T.; Simon-Plas, F. Regulation of Plant NADPH Oxidase. *Plant Signal Behav.* **2007**, *2*, 505–507. [CrossRef] [PubMed]

40. Dong, X. NPR1, all things considered. *Curr. Opin. Plant Biol.* **2004**, *7*, 547–552. [CrossRef] [PubMed]

41. Kumar, D. Salicylic acid signaling in disease resistance. *Plant Sci.* **2014**, *228*, 127–134. [CrossRef] [PubMed]

42. Zhou, M.; Wei, W.; Kararpetyan, S.; Mwimba, M.; Marqués, J.; Buchler, N.E.; Dong, X. Redox rhythm reinforces the circadian clock to gate immune response. *Nature* **2015**, *523*, 472–476. [CrossRef] [PubMed]

43. Withers, J.; Dong, X. Posttranslational Modifications of NPR1: A single protein playing multiple roles in plant immunity and physiology. *PLoS Pathog.* **2016**, *12*, 1005707. [CrossRef] [PubMed]

44. Nagel, D.H.; Kay, S.A. Complexity in the wiring and regulation of plant circadian networks. *Curr. Biol.* **2012**, *22*, 648–657. [CrossRef] [PubMed]

45. Pokhilko, A.; Fernández, A.P.; Edwards, K.D.; Southern, M.M.; Halliday, K.J.; Millar, A.J. The clock gene circuit in *Arabidopsis* includes a repressilator with additional feedback loops. *Mol. Syst. Biol.* **2012**, *8*, 574–586. [CrossRef] [PubMed]

46. Roden, L.C.; Ingle, R.A. Lights, rhythms, infection: The role of light and the circadian clock in determining the outcome of plant-pathogen interactions. *Plant Cell* **2009**, *21*, 2546–2552. [CrossRef] [PubMed]

47. Wang, Z.Y.; Tobin, E.M. Constitutive expression of the *CIRCADIAN CLOCK ASSOCIATED 1*, (*CCA1*) gene disrupts circadian rhythms and suppresses its own expression. *Cell* **1998**, *93*, 1207–1217. [CrossRef]

48. Schaffer, R.; Ramsay, N.; Samach, A.; Corden, S.; Putterill, J.; Carrã, I.A.; Coupland, G. The *LATE ELONGATED HYPOCOTYL* mutation of *Arabidopsis* disrupts circadian rhythms and the photoperiodic control of flowering. *Cell* **1998**, *93*, 1219–1229. [CrossRef]

49. Farré, E.M.; Harmer, S.L.; Harmon, F.G.; Anovsky, M.J.; Kay, S.A. Overlapping and distinct roles of *PRR7* and *PRR9* in the *Arabidopsis* circadian clock. *Curr. Biol.* **2005**, *15*, 47–54. [CrossRef] [PubMed]

50. Nakamichi, N.; Kita, M.; Ito, S.; Sato, E.; Yamashino, T.; Mizuno, T. The *Arabidopsis* pseudo-response regulators, *PRR5* and *PRR7*, coordinately play essential roles for circadian clock function. *Plant Cell Physiol.* **2005**, *46*, 609–619. [CrossRef] [PubMed]

51. Matsushika, A.; Makino, S.; Kojima, M.; Yamashino, T.; Mizuno, T. The a *PRR1/TOC1* quintet implicated in circadian rhythms of *Arabidopsis thaliana*: II. characterization with *CCA1*-overexpressing plants. *Plant Cell Physiol.* **2002**, *43*, 118–122. [CrossRef] [PubMed]

52. Strayer, C.; Oyama, T.; Schultz, T.F.; Raman, R.; Somers, D.E.; Más, P.; Panda, S.; Kreps, J.A.; Kay, S.A. Cloning of the *Arabidopsis* clock gene *TOC1*, an autoregulatory response regulator homolog. *Science* **2000**, *289*, 768–771. [CrossRef] [PubMed]

53. Fujiwara, S.; Wang, L.; Han, L.; Suh, S.P.; Mcclung, C.; Somers, D. Post-translational regulation of the *Arabidopsis* circadian clock through selective proteolysis and phosphorylation of pseudo-response regulator proteins. *J. Biol. Chem.* **2008**, *283*, 23073–23083. [CrossRef] [PubMed]

54. Alabadí, D.; Oyama, T.; Yanovsky, M.J.; Harmon, F.G.; Más, P.; Kay, S.A. Reciprocal regulation between *TOC1* and *LHY/CCA1* within the *Arabidopsis* circadian clock. *Science* **2001**, *293*, 880–883. [CrossRef] [PubMed]

55. Imaizumi, T. *Arabidopsis* circadian clock and photoperiodism: Time to think about location. *Curr. Opin. Plant Biol.* **2010**, *13*, 83–89. [CrossRef] [PubMed]

56. Pruneda-Paz, J.L.; Kay, S.A. An expanding universe of circadian networks in higher plants. *Trends Plant Sci.* **2010**, *15*, 259–265. [CrossRef] [PubMed]

57. Fowler, S.; Lee, K.; Onouchi, H.; Samach, A.; Richardson, K.; Morris, B.; Coupland, G.; Putterill, J. Gigantea: A circadian clock-controlled gene that regulates photoperiodic flowering in *Arabidopsis* and encodes a protein with several possible membrane-spanning domains. *EMBO J.* **1999**, *18*, 4679–4688. [CrossRef] [PubMed]

58. Gendron, J.M.; Pruneda-Paz, J.L.; Doherty, C.J.; Gross, A.M.; Kang, S.E.; Kay, S.A. *Arabidopsis* circadian clock protein, TOC1, is a DNA-binding transcription factor. *Proc. Natl. Acad. Sci. USA* **2012**, *109*, 3167–3172. [CrossRef] [PubMed]

59. Huang, W.; Pérezgarcía, P.; Pokhilko, A.; Millar, A.J.; Antoshechkin, I.; Riechmann, J.L.; Mas, P. Mapping the core of the *Arabidopsis* circadian clock defines the network structure of the oscillator. *Science* **2012**, *336*, 75–79. [CrossRef] [PubMed]

60. Green, R.M.; Tingay, S.; Wang, Z.Y.; Tobin, E.M. Circadian rhythms confer a higher level of fitness to *Arabidopsis* plants. *Plant Physiol.* **2002**, *129*, 576–584. [CrossRef] [PubMed]

61. Green, R.M.; Tobin, E.M. Loss of the circadian clock-associated protein 1 in *Arabidopsis* results in altered clock-regulated gene expression. *Proc. Natl. Acad. Sci. USA* **1999**, *96*, 4176–4179. [CrossRef] [PubMed]

62. Zhang, C.; Xie, Q.; Anderson, R.G.; Ng, G.; Seitz, N.C.; Peterson, T.; McClung, C.R.; McDowell, J.M.; Kong, D.; Kwak, J.M.; et al. Crosstalk between the circadian clock and innate immunity in *Arabidopsis*. *PLoS Pathog.* **2013**, *9*, e1003370. [CrossRef] [PubMed]

63. Somers, D.E.; Webb, A.A.; Pearson, M.; Kay, S.A. The short-period mutant, *TOC1-1*, alters circadian clock regulation of multiple outputs throughout development in *Arabidopsis thaliana*. *Development* **1998**, *125*, 485–494. [PubMed]

64. Wang, W.; Barnaby, J.Y.; Tada, Y.; Li, H.; Tör, M.; Caldelari, D.; Lee, D.U.; Fu, X.D.; Dong, X. Timing of plant immune responses by a central circadian regulator. *Nature* **2011**, *470*, 110–114. [CrossRef] [PubMed]

65. Zheng, X.Y.; Zhou, M.; Yoo, H.; Pruneda-Paz, J.L.; Spivey, N.W.; Kay, S.A.; Dong, X. Inaugural article by a recently elected academy member: Spatial and temporal regulation of biosynthesis of the plant immune signal salicylic acid. *Proc. Natl. Acad. Sci. USA* **2015**, *112*, 9166–9173. [CrossRef] [PubMed]

66. Hevia, M.A.; Canessa, P.; Müller-Esparza, H.; Larrondo, L.F. A circadian oscillator in the fungus *Botrytis cinerea* regulates virulence when infecting *Arabidopsis thaliana*. *Pro Natl. Acad Sci. USA.* **2015**, *112*, 8744–8749. [CrossRef] [PubMed]

67. Seo, P.J.; Mas, P. Stressing the role of the plant circadian clock. *Trends Plant Sci.* **2015**, *20*, 230–237. [CrossRef] [PubMed]

68. Ingle, R.A.; Stoker, C.; Stone, W.; Adams, N.; Denby, K.J. Jasmonate signalling drives time-of-day differences in susceptibility of *Arabidopsis* to the fungal pathogen *Botrytis cinerea*. *Plant J.* **2015**, *84*, 937–948. [CrossRef] [PubMed]

69. Goodspeed, D.; Chehab, E.W.; Minvenditti, A.; Braam, J.; Covington, M.F. From the cover: *Arabidopsis* synchronizes jasmonate-mediated defense with insect circadian behavior. *Proc. Natl. Acad. Sci. USA* **2012**, *109*, 4674–4677. [CrossRef] [PubMed]

70. Mou, Z.; Fan, W.; Dong, X. Inducers of plant systemic acquired resistance regulate NPR1 function through redox changes. *Cell* **2003**, *113*, 935–944. [CrossRef]

71. Millar, A.J.; Carre, I.A.; Strayer, C.A.; Chua, N.H.; Kay, S.A. Circadian clock mutants in *Arabidopsis* identified by luciferase imaging. *Science* **1995**, *267*, 1161–1163. [CrossRef] [PubMed]

72. Más, P.; Kim, W.Y.; Somers, D.E.; Kay, S.A. Targeted degradation of TOC1 by ZTL modulates circadian function in *Arabidopsis thaliana*. *Nature* **2003**, *426*, 567–570. [CrossRef] [PubMed]

73. Donofrio, N.M.; Delaney, T.P. Abnormal callose response phenotype and hypersusceptibility to Peronospoara parasitica in defence-compromised Arabidopsis NIM1-1 and salicylate hydroxylase-expressing plants. *Mol. Plant Microbe Interact.* **2001**, *14*, 439–450.

74. Ea, V.D.B.; Freddie, C.T.; Kahn, K.; Parker, J.E.; Jones, J.D. *Arabidopsis RPP4* is a member the *RPP5* multigene family of TIR-NB-LRR genes and confers downy mildew resistance through multiple signaling components. *Plant J.* **2002**, *29*, 439–451.

75. Salomé, P.A.; Mcclung, C.R. *Pseudo-response regulator 7* and *9* are partially redundant genes essential for the temperature responsiveness of the arabidopsis circadian clock. *Plant Cell* **2005**, *3*, 791–803. [CrossRef] [PubMed]

76. Bhardwaj, V.; Meier, S.; Petersen, L.N.; Ingle, R.A.; Roden, L.C. Defence responses of *Arabidopsis thaliana* to infection by *Pseudomonas syringae* are regulated by the circadian clock. *PLoS ONE* **2011**, *6*, e26968. [CrossRef] [PubMed]

77. Hermanns, M.; Slusarenko, A.J.; Schlaich, N.L. Organ-specificity in a plant disease is determined independently of *R* gene signaling. *Mol. Plant Microbe Interact.* **2003**, *16*, 752–759. [CrossRef] [PubMed]

78. Fu, Z.Q.; Guo, M.; Jeong, B.R.; Tian, F.; Elthon, T.E.; Cerny, R.L.; Staiger, D.; Alfano, J.R. A type III effector ADP-ribosylates RNA-binding proteins and quells plant immunity. *Nature* **2007**, *447*, 284–288. [CrossRef] [PubMed]

79. Heintzen, C.; Nater, M.; Apel, K.; Staiger, D. AtGRP7, a nuclear RNA binding protein as a component of a circadian-regulated negative feedback loop in *Arabidopsis thaliana*. *Proc. Natl. Acad. Sci. USA* **1997**, *94*, 8515–8520. [CrossRef] [PubMed]

80. Staiger, D.; Zecca, L.; Wieczorek Kirk, D.A.; Apel, K.; Eckstein, L. The circadian clock regulated RNA-binding protein AtGRP7 autoregulates its expression by influencing alternative splicing of its own pre-mRNA. *Plant J.* **2003**, *33*, 361–371. [CrossRef] [PubMed]

81. Niwa, Y.; Ito, S.; Nakamichi, N.; Mizoguchi, T.; Niinuma, K.; Yamashino, T.; Mizuno, T. Genetic linkages of the circadian clock-associated genes, *TOC1*, *CCA1* and *LHY*, in the photoperiodic control of flowering time in *Arabidopsis thaliana*. *Plant Cell Physiol.* **2007**, *48*, 925–937. [CrossRef] [PubMed]

82. Ederli, L.; Madeo, L.; Calderini, O.; Gehring, C.; Moretti, C.; Buonaurio, R.; Paolocci, F.; Pasqualini, S. The *Arabidopsis thaliana* cysteine-rich receptor-like kinase CRK20 modulates host responses to *Pseudomonas syringae* pv. tomato DC3000 infection. *J. Plant Physiol.* **2011**, *15*, 1784–1794. [CrossRef] [PubMed]

83. Li, Z.; Bonaldi, K.; Uribe, F.; Pruneda-Paz, J.L. A localized *pseudomonas syringae*, infection triggers systemic clock responses in *Arabidopsis*. *Curr. Biol.* **2018**, *4*, 28–42. [CrossRef] [PubMed]

84. Fu, Z.Q.; Yan, S.; Saleh, A.; Wang, W.; Ruble, J.; Oka, N.; Mohan, R.; Spoel, S.H.; Tada, Y.; Zheng, N.; et al. NPR3 and NPR4 are receptors for the immune signal salicylic acid in plants. *Nature* **2012**, *486*, 228–232. [CrossRef] [PubMed]

85. Carella, P.; Wilson, D.C.; Cameron, R.K. Some things get better with age: Differences in salicylic acid accumulation and defense signaling in young and mature *Arabidopsis*. *Front Plant Sci.* **2015**, *5*, 775. [CrossRef] [PubMed]

86. Nagel, D.H.; Doherty, C.J.; Pruneda-Paz, J.L.; Schmitz, R.J.; Ecker, J.R.; Kay, S.A. Genome-wide identification of CCA1 targets uncovers an expanded clock network in *Arabidopsis*. *Proc. Natl. Acad. Sci. USA* **2015**, *112*, 4802–4810. [CrossRef] [PubMed]

87. Shin, J.; Heidrich, K.; Sanchez-Villarreal, A.; Parker, J.E.; Davis, S.J. Time for coffee represses accumulation of the MYC2 transcription factor to provide time-of-day regulation of jasmonate signaling in *Arabidopsis*. *Plant Cell* **2012**, *24*, 2470–2482. [CrossRef] [PubMed]

88. Wu, Y.; Zhang, D.; Chu, J.Y.; Boyle, P.; Wang, Y.; Brindle, I.; De Luca, V.; Després, C. The *Arabidopsis*, NPR1 protein is a receptor for the plant defense hormone salicylic acid. *Cell Rep.* **2012**, *1*, 639–647. [CrossRef] [PubMed]

89. Yan, S.; Dong, X. Perception of the plant immune signal salicylic acid. *Curr. Opin. Plant Biol.* **2014**, *20*, 64–68. [CrossRef] [PubMed]

90. Liu, T.; Carlsson, J.; Takeuchi, T.; Newton, L.; Farré, E.M. Direct regulation of abiotic responses by the *Arabidopsis* circadian clock component PRR7. *Plant J.* **2013**, *76*, 101–114. [CrossRef] [PubMed]

91. Liu, T.L.; Newton, L.; Liu, M.J.; Shiu, S.H.; Farré, E.M. A G-box-like motif is necessary for transcriptional regulation by circadian pseudo-response regulators in *Arabidopsis*. *Plant Physiol.* **2016**, *170*, 528–539. [CrossRef] [PubMed]

92. Nakamichi, N.; Kiba, T.; Kamioka, M.; Suzuki, T.; Yamashino, T.; Higashiyama, T.; Sakakibara, H.; Mizuno, T. Transcriptional repressor PRR5 directly regulates clock-output pathways. *Proc. Natl. Acad. Sci. USA* **2012**, *109*, 17123–17128. [CrossRef] [PubMed]

93. Atamian, H.S.; Harmer, S.L. Circadian regulation of hormone signaling and plant physiology. *Plant Mol. Biol.* **2016**, *91*, 691–702. [CrossRef] [PubMed]

94. Baldwin, I.T.; Meldau, S. Just in time: Circadian defense patterns and the optimal defense hypothesis. *Plant Signal Behav.* **2013**, *8*, e24410. [CrossRef] [PubMed]

95. Wang, G.; Zhang, C.; Battle, S.; Lu, H. The phosphate transporter *PHT4;1* is a salicylic acid regulator likely controlled by the circadian clock protein CCA1. *Front Plant Sci.* **2014**, *5*, 701–710. [CrossRef] [PubMed]

96. Karapetyan, S.; Dong, X. Redox and the circadian clock in plant immunity: A balancing act. *Free Radic. Biol. Med.* **2017**, *10*, e1044192. [CrossRef] [PubMed]

97. Ding, Y.; Sun, T.; Ao, K.; Peng, Y.; Zhang, Y.; Li, X.; Zhang, Y. Opposite roles of salicylic acid receptors NPR1 and NPR3/NPR4 in transcriptional regulation of plant immunity. *Cell* **2018**, *6*, 173–203. [CrossRef] [PubMed]

98. Glazebrook, J.; Rogers, E.E.; Ausubel, F.M. Isolation of *Arabidopsis* mutants with enhanced disease susceptibility by direct screening. *Genetics* **1996**, *143*, 973–982. [PubMed]

99. Shah, J.; Tsui, F.; Klessig, D.F. Characterization of a *salicylic acid-insensitive* mutant (*SAI1*) of *Arabidopsis thaliana*, identified in a selective screen utilizing the SA-inducible expression of the *TMS2* gene. *Mol. Plant Microbe Interact.* **1997**, *10*, 69–78. [CrossRef] [PubMed]

100. Cao, H.; Glazebrook, J.; Clarke, J.D.; Volko, S.; Dong, X. The *Arabidopsis NPR1* gene that controls systemic acquired resistance encodes a novel protein containing ankyrin repeats. *Cell* **1997**, *88*, 57–63. [CrossRef]

101. Aravind, L.; Koonin, E.V. Fold prediction and evolutionary analysis of the POZ domain: Structural and evolutionary relationship with the potassium channel tetramerization domain. *J. Mol. Biol.* **1999**, *285*, 1353–1361. [CrossRef] [PubMed]

102. Kinkema, M.; Fan, W.; Dong, X. Nuclear localization of NPR1 is required for activation of *PR* gene expression. *Plant Cell* **2000**, *12*, 2339–2350. [CrossRef] [PubMed]

103. Petroski, M.D.; Deshaies, R.J. Function and regulation of cullin-RING ubiquitin ligases. *Nat. Rev. Mol. Cell Biol.* **2005**, *6*, 9–20. [CrossRef] [PubMed]

104. Durrant, W.E.; Dong, X. Systemic acquired resistance. *Annu. Rev. Phytopathol.* **2004**, *42*, 185–209. [CrossRef] [PubMed]

105. Rodriguez, S.C.; López, B.; Chaves, A.R. Changes in polyamines and ethylene during the development and ripening of eggplant fruits (solanum melongena). *J. Agric. Food Chem.* **1999**, *47*, 1431–1434. [CrossRef] [PubMed]

106. Luke-Glaser, S.; Pintard, L.; Tyers, M.; Peter, M. The AAA-ATPase FIGL-1 controls mitotic progression, and its levels are regulated by the CUL-3^{MEL-26} E3 ligase in the *C. elegans* germ line. *J. Cell Sci.* **2007**, *120*, 3179–3187. [CrossRef] [PubMed]

107. Pintard, L.; Willems, A.; Peter, M. Cullin-based ubiquitin ligases: Cul3–BTB complexes join the family. *EMBO J.* **2004**, *23*, 1681–1687. [CrossRef] [PubMed]

108. Pintard, L.; Willis, J.H.; Willems, A.; Johnson, J.L.; Srayko, M.; Kurz, T.; Glaser, S.; Mains, P.E.; Tyers, M.; Bowerman, B.; et al. The BTB protein MEL-26 is a substrate-specific adaptor of the CUL-3 ubiquitin-ligase. *Nature* **2003**, *425*, 311–316. [CrossRef] [PubMed]

109. Mukhtar, M.S.; Nishimura, M.T.; Dangl, J. NPR1 in plant defense: it's not over 'til it's turned over. *Cell* **2009**, *137*, 804–806. [CrossRef] [PubMed]

110. Zhang, Y.; Yu, T.C.; Na, Q.; Zhao, Q.; Bi, D.; Xin, L. Negative regulation of defense responses in *Arabidopsis* by two NPR1 paralogs. *Plant J.* **2006**, *48*, 647–656. [CrossRef] [PubMed]

111. Manohar, M.; Tian, M.; Moreau, M.; Park, S.W.; Choi, H.W.; Fei, Z.; Friso, G.; Asif, M.; Manosalva, P.; Von Dahl, C.C.; et al. Identification of multiple salicylic acid-binding proteins using two high throughput screens. *Front Plant Sci.* **2015**, *5*, 777–791. [CrossRef] [PubMed]

112. Enyedi, A.J.; Yalpani, N.; Silverman, P.; Raskin, I. Localization, conjugation, and function of salicylic acid in tobacco during the hypersensitive reaction to tobacco mosaic virus. *Proc. Natl. Acad. Sci. USA* **1992**, *89*, 2480–2484. [CrossRef] [PubMed]

113. Spoel, S.H.; Koornneef, A.; Claessens, S.M.; Korzelius, J.P.; Pelt, J.A.; Mueller, M.J.; Buchala, A.J.; Métraux, J.; Brown, R.; Kazan, K.; et al. NPR1 modulates cross-talk between salicylate- and jasmonate-dependent defense pathways through a novel function in the cytosol. *Plant Cell* **2003**, *15*, 760–770. [CrossRef] [PubMed]

114. Heidel, A.J.; Dong, X. Fitness benefits of systemic acquired resistance during *Hyaloperonospora parasitica* infection in *Arabidopsis thaliana*. *Genetics* **2006**, *173*, 1621–1628. [CrossRef] [PubMed]

115. Fu, Z.Q.; Dong, X. Systemic acquired resistance: Turning local infection into global defense. *Annu. Rev. Plant Biol.* **2013**, *64*, 839–863. [CrossRef] [PubMed]

116. Cao, H.; Li, X.; Dong, X. Generation of broad-spectrum disease resistance by overexpression of an essential regulatory gene in systemic acquired resistance. *Proc. Natl. Acad. Sci. USA* **1998**, *95*, 6531–6536. [CrossRef] [PubMed]

117. Chern, M.S.; Fitzgerald, H.A.; Yadav, R.C.; Canlas, P.E.; Dong, X.; Ronald, P.C. Evidence for a disease-resistance pathway in rice similar to the NPR1-mediated signaling pathway in *Arabidopsis*. *Plant J.* **2001**, *27*, 101–113. [CrossRef] [PubMed]

118. Makandar, R.; Essig, J.S.; Schapaugh, M.A.; Trick, H.N.; Shah, J. Genetically engineered resistance to fusarium head blight in wheat by expression of *Arabidopsis* NPR1. *Mol. Plant Microbe Interact.* **2006**, *19*, 123–129. [CrossRef] [PubMed]

119. Malnoy, M.; Jin, Q.; Borejsza-Wysocka, E.E.; He, S.Y.; Aldwinckle, H.S. Overexpression of the apple *MpNPR1* gene confers increased disease resistance in Malus × domestica. *Mol. Plant Microbe Interact.* **2007**, *20*, 1568–1580. [CrossRef] [PubMed]

120. Zhou, J.M.; Trifa, Y.; Silva, H.; Pontier, D.; Lam, E.; Shah, J.; Klessing, D.F. NPR1 differentially interacts with members of the TGA/OBF family of transcription factors that bind an element of the *PR-1* gene required for induction by salicylic acid. *Mol. Plant Microbe Interact.* **2000**, *13*, 191–202. [CrossRef] [PubMed]

121. Després, C.; Delong, C.; Glaze, S.; Liu, E.W.; Fobert, P.R. The *Arabidopsis* NPR1/NIM1 protein enhances the DNA binding activity of a subgroup of the TGA family of bZIP transcription factors. *Plant Cell* **2000**, *12*, 279–290. [CrossRef] [PubMed]

122. Fan, W.; Dong, X. In vivo interaction between NPR1 and transcription factor TGA2 leads to salicylic acid–mediated gene activation in *Arabidopsis*. *Plant Cell* **2002**, *14*, 1377–1389. [CrossRef] [PubMed]

123. Zhang, Y.; Fan, W.; Kinkema, M.; Li, X.; Dong, X. Interaction of NPR1 with basic leucine zipper protein transcription factors that bind sequences required for salicylic acid induction of the *PR-1* gene. *Proc. Natl. Acad. Sci. USA* **1999**, *96*, 6523–6528. [CrossRef] [PubMed]

124. Subramaniam, R.; Desveaux, D.; Spickler, C.; Michnick, S.W.; Brisson, N. Direct visualization of protein interactions in plant cells. *Nat. Biotechnol.* **2001**, *19*, 769–772. [CrossRef] [PubMed]

125. Ryals, J.; Weymann, K.; Lawton, K.; Friedrich, L.; Ellis, D.; Steiner, H.Y.; Johnson, J.; Delaney, T.P.; Jesse, T.; Vos, P.; et al. The *Arabidopsis* NIM1 protein shows homology to the mammalian transcription factor inhibitor i kappa B. *Plant Cell* **1997**, *9*, 425–439. [CrossRef] [PubMed]

126. Tada, Y.; Spoel, S.H.; Pajerowska-Mukhtar, K.; Mou, Z.; Song, J.; Wang, C.; Zuo, J.; Dong, X. Plant immunity requires conformational changes of NPR1 via S-nitrosylation and thioredoxins. *Science* **2008**, *321*, 952–956. [CrossRef] [PubMed]

127. Spoel, S.H.; Tada, Y.; Loake, G.J. Post-translational protein modification as a tool for transcription reprogramming. *New Phytol.* **2010**, *186*, 333–339. [CrossRef] [PubMed]

128. Spoel, S.H.; Mou, Z.; Tada, Y.N.; Spivey, W.; Genschik, P.; Dong, X. Proteasome-mediated turnover of the transcription coactivator NPR1 plays dual roles in regulating plant immunity. *Cell* **2009**, *137*, 860–872. [CrossRef] [PubMed]

129. Más, P.; Alabadí, D.; Yanovsky, M.J.; Oyama, T.; Kay, S.A. Dual role of *TOC1* in the control of circadian and photomorphogenic responses in *Arabidopsis*. *Plant Cell* **2003**, *15*, 223–236. [CrossRef] [PubMed]

130. Brody, S.; Harris, S. Circadian rhythms in neurospora: Spatial differences in pyridine nucleotide levels. *Science* **1973**, *180*, 498–500. [CrossRef] [PubMed]

131. Dangl, J.L.; Jones, J.D. Plant pathogens and integrated defence responses to infection. *Nature* **2001**, *411*, 826–833. [CrossRef] [PubMed]

132. Frederickson Matika, D.E.; Loake, G.J. Redox regulation in plant immune function. *Antioxid. Redox Signal.* **2014**, *21*, 1373–1388. [CrossRef] [PubMed]

133. Delledonne, M.; Zeier, J.; Marocco, A.; Lamb, C. Signal interactions between nitric oxide and reactive oxygen intermediates in the plant hypersensitive disease resistance response. *Proc. Natl. Acad. Sci. USA* **2001**, *98*, 13454–13459. [CrossRef] [PubMed]

134. Levine, A.; Tenhaken, R.; Dixon, R.; Lamb, C. H_2O_2 from the oxidative burst orchestrates the plant hypersensitive disease resistance response. *Cell* **1994**, *79*, 583–593. [CrossRef]

135. Yun, B.W.; Feechan, A.; Yin, M.; Saidi, N.B.; Le, B.T.; Yu, M.; Moore, J.W.; Kang, J.G.; Kwon, E.; Spoel, S.H.; et al. S-nitrosylation of NADPH oxidase regulates cell death in plant immunity. *Nature* **2011**, *478*, 264–268. [CrossRef] [PubMed]

136. Yuan, H.M.; Liu, W.C.; Lu, Y.T. Catalase2 coordinates SA-mediated repression of both auxin accumulation and JA biosynthesis in plant defenses. *Cell Host Microbe* **2017**, *21*, 143–155. [CrossRef] [PubMed]

137. Covington, M.F.; Maloof, J.N.; Straume, M.; Kay, S.A.; Harmer, S.L. Global transcriptome analysis reveals circadian regulation of key pathways in plant growth and development. *Genome Biol.* **2008**, *9*, 2–18. [CrossRef] [PubMed]

138. Zhong, H.H.; McClung, C.R. The circadian clock gates expression of two *Arabidopsis* catalase genes to distinct and opposite circadian phases. *Mol. Gengenet.* **1996**, *251*, 196–203.

139. Bhattacharya, A.; Khanale, V.; Char, B. Plant circadian rhythm in stress signaling. *Indian J. Plant Physiol.* **2017**, *22*, 147–155. [CrossRef]

140. Jahan, M.S.; Ogawa, K.; Nakamura, Y.; Shimoishi, Y.; Mori, I.C.; Murata, Y. Deficient glutathione in guard cells facilitates abscisic acid-induced stomatal closure but does not affect light-induced stomatal opening. *J. Agric. Chem. Soc. Jpn.* **2008**, *72*, 2795–2798. [CrossRef] [PubMed]

141. Lu, H.; Mcclung, C.R.; Zhang, C. Tick tock: Circadian regulation of plant innate immunity. *Annu. Rev. Phytopathol.* **2017**, *55*, 287–311. [CrossRef] [PubMed]

142. Spoel, S.H.; Van, O.G. Circadian redox signaling in plant immunity and abiotic stress. *Antioxid. Redox Signal.* **2014**, *20*, 3024–3039. [CrossRef] [PubMed]

143. Kadota, Y.; Sklenar, J.; Derbyshire, P.; Stransfeld, L.; Asai, S.; Ntoukakis, V.; Jones, J.D.; Shirasu, K.; Menke, F.; Jones, A.; et al. Direct regulation of the NADPH oxidase RBOHD by the PRR-associated kinase BIK1 during plant immunity. *Mol. Cell* **2014**, *54*, 43–55. [CrossRef] [PubMed]

144. Li, L.; Li, M.; Yu, L.; Zhou, Z.; Liang, X.; Liu, Z.; Cai, G.; Gao, L.; Zhang, X.; Wang, Y.; et al. The FLS2-associated kinase BIK1 directly phosphorylates the NADPH oxidase rbohd to control plant immunity. *Cell Host Microbe* **2014**, *15*, 329–338. [CrossRef] [PubMed]

145. Chaouch, S.; Queval, G.; Noctor, G. *ATRbohF* is a crucial modulator of defence-associated metabolism and a key actor in the interplay between intracellular oxidative stress and pathogenesis responses in *Arabidopsis*. *Plant J.* **2012**, *69*, 613–627. [CrossRef] [PubMed]

146. Mammarella, N.D.; Cheng, Z.; Zheng, Q.F.; Daudi, A.; Bolwell, G.P.; Dong, X.; Ausubel, F.M. Apoplastic peroxidases are required for salicylic acid-mediated defense against *Pseudomonas syringae*. *Phytochemistry* **2015**, *112*, 110–121. [CrossRef] [PubMed]

147. Daudi, A.; Cheng, Z.; O'Brien, J.A.; Mammarella, N.; Khan, S.; Ausubel, F.M.; Bolwell, G.P. The apoplastic oxidative burst peroxidase in *Arabidopsis* is a major component of pattern-triggered immunity. *Plant Cell* **2012**, *24*, 275–287. [CrossRef] [PubMed]

148. O'Brien, J.A.; Daudi, A.; Finch, P.; Butt, V.S.; Whitelegge, J.P.; Souda, P.; Ausubel, F.M.; Bolwell, G.P. A peroxidase-dependent apoplastic oxidative burst in cultured *Arabidopsis* cells functions in MAMP-elicited defense. *Plant Physiol.* **2012**, *158*, 2013–2027. [CrossRef] [PubMed]

149. Bindschedler, L.V.; Dewdney, J.; Blee, K.A.; Stone, J.M.; Asai, T.; Plotnikov, J.; Denoux, C.; Hayes, T.; Gerrish, C.; Davies, D.R.; et al. Peroxidase-dependent apoplastic oxidative burst in *Arabidopsis* required for pathogen resistance. *Plant J.* **2006**, *47*, 851–863. [CrossRef] [PubMed]

150. Melotto, M.; Underwood, W.; Koczan, J.; Nomura, K.; He, S.Y. Plant stomata function in innate immunity against bacterial invasion. *Cell* **2006**, *126*, 969–980. [CrossRef] [PubMed]

151. Zeidler, D.; Zähringer, U.; Gerber, I.; Dubery, I.; Hartung, T.; Bors, W.; Hutzler, P.; Durner, J. Innate immunity in *Arabidopsis thaliana*: Lipopolysaccharides activate nitric oxide synthase (NOS) and induce defense genes. *Proc. Natl. Acad. Sci. USA* **2004**, *101*, 15811–15816. [CrossRef] [PubMed]

152. Durner, J.; Wendehenne, D.; Klessig, D.F. Defense gene induction in tobacco by nitric oxide, cyclic GMP, and cyclic ADP-Ribose. *Proc. Natl. Acad. Sci. USA* **1998**, *95*, 10328–10333. [CrossRef] [PubMed]

153. Sun, A.; Nie, S.; Xing, D. Nitric oxide-mediated maintenance of redox homeostasis contributes to NPR1-dependent plant innate immunity triggered by lipopolysaccharides. *Plant Physiol.* **2012**, *160*, 1081–1096. [CrossRef] [PubMed]