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# The exodomain of the impaired oomycete susceptibility 1 receptor mediates both endoplasmic reticulum stress responses and abscisic acid signalling during downy mildew infection of *Arabidopsis*

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## Abstract

The phytohormone abscisic acid (ABA) regulates cell growth and plant development, and contributes to defence responses to pathogens. We previously showed that the *Arabidopsis* malectin-like domain leucine-rich repeat receptor-like kinase (MLD-LRR-RLK) impaired oomycete susceptibility 1 (IOS1) attenuates ABA signalling during infection with the oomycete downy mildew pathogen *Hyaloperonospora arabidopsidis*. The exodomain of IOS1 with its MLD retains the receptor in the endoplasmic reticulum (ER), where it interacts with the ribophorin HAP6 to dampen a pathogen-induced ER stress response called the unfolded protein response (UPR). The down-regulation of both ABA and UPR signalling probably provides the pathogen with an advantage for infection. Here, we show that ABA-related phenotypes of the *ios1-1* mutant, such as up-regulated expression of ABA-responsive genes and hypersensitivity to exogenous ABA application, were reverted by expression of the IOS1 exodomain in the mutant background. Furthermore, knockdown mutants for ER-resident HAP6 showed similarly reduced UPR and ABA signalling, indicating that HAP6 positively regulates both pathways. Our data suggest that the IOS1 exodomain and HAP6 contribute in the ER to the IOS1-mediated interference with ABA and UPR signalling.

## KEYWORDS

abscisic acid, biotrophy, malectin-like domain leucine-rich repeat receptor-like kinase (MLD-LRR-RLK), unfolded protein response

## 1 | INTRODUCTION

The sesquiterpenoid phytohormone abscisic acid (ABA) is essential for plant development. It inhibits cell growth during stem and primary root elongation, and extends seed dormancy under adverse climatic

conditions. The hormone also contributes to plant defence responses to pathogens (Lee & Luan, 2012; Ton et al., 2009). Stomata constitute natural openings for the entry of microbes into plant tissues, and ABA-induced stomata closure is one of the most immediate plant immune responses against bacterial invasion (Melotto et al., 2006). Biotrophic

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oomycete pathogens, such as the downy mildew *Hyaloperonospora arabidopsidis* (Hpa), use stomatal openings for sporulation on leaf surfaces. ABA-induced stomatal closure thus provides an obstacle for the dissemination of these pathogens. The *Arabidopsis* malectin-like domain leucine-rich repeat receptor-like kinase (MLD-LRR-RLK) impaired oomycete susceptibility 1 (IOS1) has been shown to negatively regulate the expression of marker genes for ABA signalling. The *ios1-1* knockout mutant is hypersensitive to ABA when compared to the wild type, as evidenced by reduced seed germination and root elongation on growth media containing the hormone (Hok et al., 2014). IOS1 production on Hpa infection seems to attenuate ABA signalling and promote the development of the oomycete pathogen. By contrast, absence of IOS1 in *ios1-1* leads to ABA overactivation and decreased susceptibility to infection (Hok et al., 2014).

Excessive protein synthesis or the production of unfolded or misfolded proteins stimulate stress responses in the endoplasmic reticulum (ER) of eukaryotic cells. To restore homeostasis in the ER, cells engage processes such as the unfolded protein response (UPR), which leads to an arrest of protein synthesis and the repair or degradation of misfolded proteins. When restoration of homeostasis is not possible, the UPR engages in cell death programmes (Hetz, 2012; Hiramatsu et al., 2015). In plants, infection with microbial pathogens frequently results in the onset of the UPR, which can in turn contribute to immune responses (Moreno et al., 2012; Prasad & Sonnewald, 2013; Xu et al., 2019; Zhang et al., 2015) and programmed cell death, responses that aim at confining microbes and limiting the spreading of disease (Carvalho et al., 2014; Mishiba et al., 2013). To counteract such responses, oomycete pathogens from the genus *Phytophthora* attenuate the UPR by secreting effectors into host cells that interact with ER stress-regulatory proteins (Fan et al., 2018; Jing et al., 2016). The UPR signalling network involves two pathways in plants, which are similar to those found in animal cells. One is mediated by inositol-requiring protein 1 (IRE1) and the transcription factor (TF) bZIP60. The other involves the TFs bZIP28 and bZIP17, which are functional homologues of activating TF 6 (ATF6) in animals (Manghwar & Li, 2022). The third animal UPR signalling pathway, which is mediated by the protein kinase R-like endoplasmic reticulum kinase (PERK), has not yet been uncovered in plants (Howell, 2021). To avoid that unrestrained UPR leads to cell and tissue damage, eukaryotes possess regulatory mechanisms to tightly control the response (Angelos et al., 2017).

Functional links between ABA signalling and the UPR have been proposed (Afrin et al., 2020; Chen et al., 2020), but studies demonstrating these links are still rare. Recently, we reported that IOS1 interacts with the ER-residing ribophorin HAPLESS 6 (HAP6) from *Arabidopsis thaliana* and that the malectin-like domain (MLD) in the receptor's exodomain attenuates the pathogen-induced UPR on infection with Hpa (Giordano et al., 2022). The MLD of IOS1 consists of two subdomains, both of which share structural similarities with the ER-residing malectin protein from *Xenopus laevis* (Giordano et al., 2022). Malectins in animals are discrete ER membrane-associated proteins for which there are no equivalents in plants. They associate with ribophorins from the oligosaccharyltransferase (OST) complex, contribute

to N-glycosylation of proteins (Schallus et al., 2008), ensure quality control of these proteins (Galli et al., 2011), and regulate ER stress responses such as the UPR (Chen et al., 2011). Our observations thus suggest that the MLD of IOS1 has functional homologies with animal malectin, and that the pathogen uses the plant protein to attenuate a host response that is detrimental to infection.

Because IOS1 impairs both ABA signalling and the UPR during infection of *Arabidopsis* with Hpa, the present study aimed to determine whether these events are associated with each other during the plant-pathogen interaction. Among other parameters, we analysed the expression of marker genes for the UPR (Nagashima et al., 2011; Yang et al., 2014) and ABA responses (Yang et al., 2011) in *Arabidopsis*. Taken together, the results presented here indicate that the IOS1 exodomain and the ribophorin HAP6, which are responsible for modulating the pathogen-induced UPR, also contribute to the modulation of ABA signalling during infection.

## 2 | RESULTS

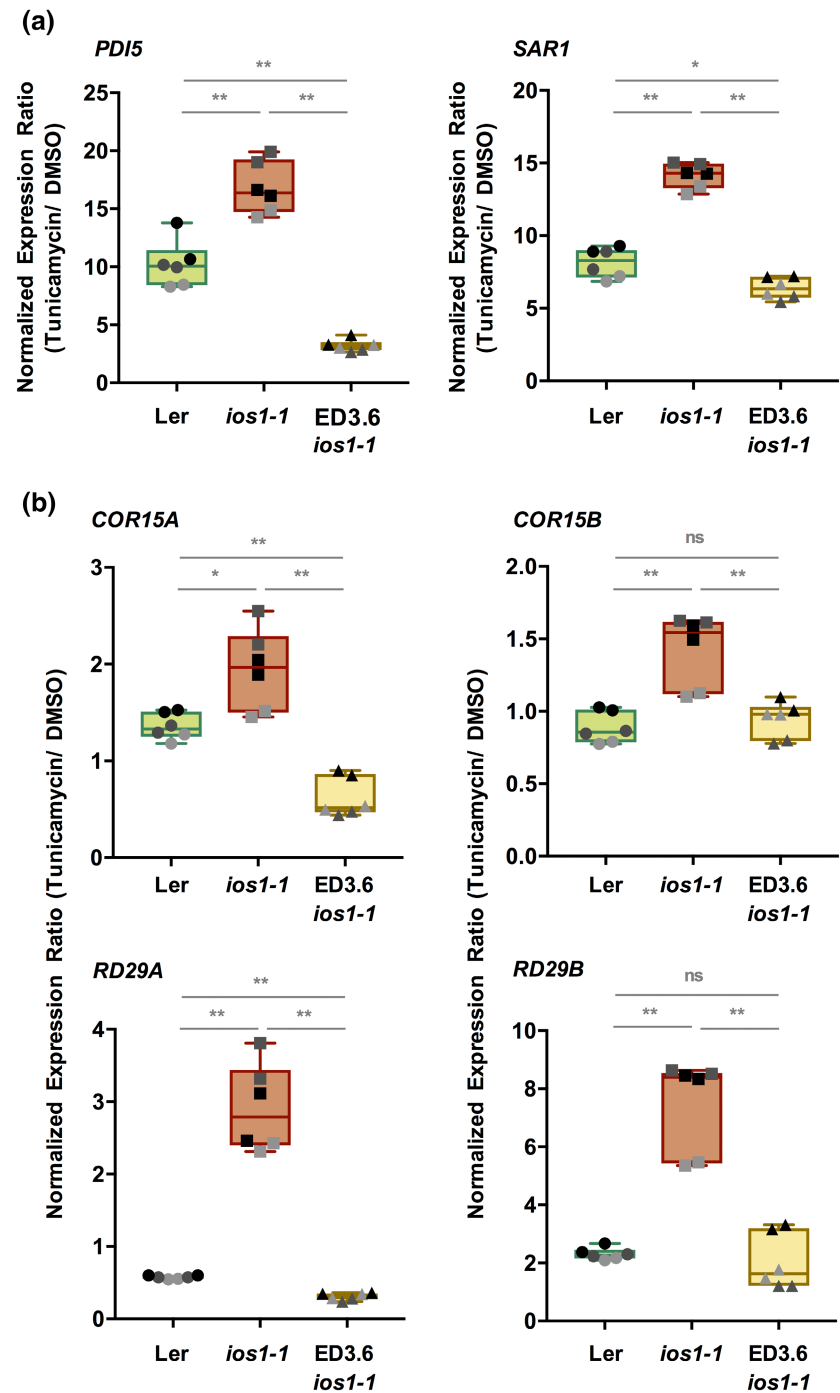
### 2.1 | ER stress-mediated ABA signalling is attenuated by the IOS1 exodomain

Tunicamycin inhibits N-glycosylation of proteins in the ER and triggers the UPR in *Arabidopsis* (Nagashima et al., 2011). To analyse whether or not a stimulation of the UPR may influence ABA signalling, we treated seedlings from the *Arabidopsis* Ler wild-type (Wt), the *ios1-1* mutant, and a line that expresses the IOS1 exodomain in the *ios1-1* mutant background (line ED3.6; Giordano et al., 2022) with this drug. In *ios1-1* mutant plants, the accumulation of transcripts from previously described UPR marker genes, such as *PDI5* and *SAR1* (Giordano et al., 2022), was significantly increased (Figure 1a). This increased mRNA accumulation in the absence of IOS1 was repressed in line ED3.6 (Figure 1a), confirming that the exodomain of IOS1 attenuates UPR signalling. We then analysed the tunicamycin-treated plants for the accumulation of transcripts from the marker genes for ABA signalling, *COR15A*, *COR15B*, *RD29A*, and *RD29B* (Hok et al., 2014) and found, similarly to what we observed for *PDI5* and *SAR1* transcripts, increased accumulation in the *ios1-1* mutant (Figure 1b). The increased expression of marker genes for ABA responses in *ios1-1* seedlings was reduced to wild-type levels (*COR15B*, *RD29B*) and below (*COR15A*, *RD29A*) when the IOS1 exodomain was expressed in the *ios1-1* background (line ED3.6; Figure 1b). Tunicamycin thus stimulates the UPR and the transcriptional activation of genes related to ABA signalling, and both events are attenuated by the IOS1 exodomain.

### 2.2 | ABA hypersensitivity of the *ios1-1* mutant is alleviated by the IOS1 exodomain

To determine if the exodomain is also responsible for the observed attenuated ABA response mediated by IOS1 during the interaction

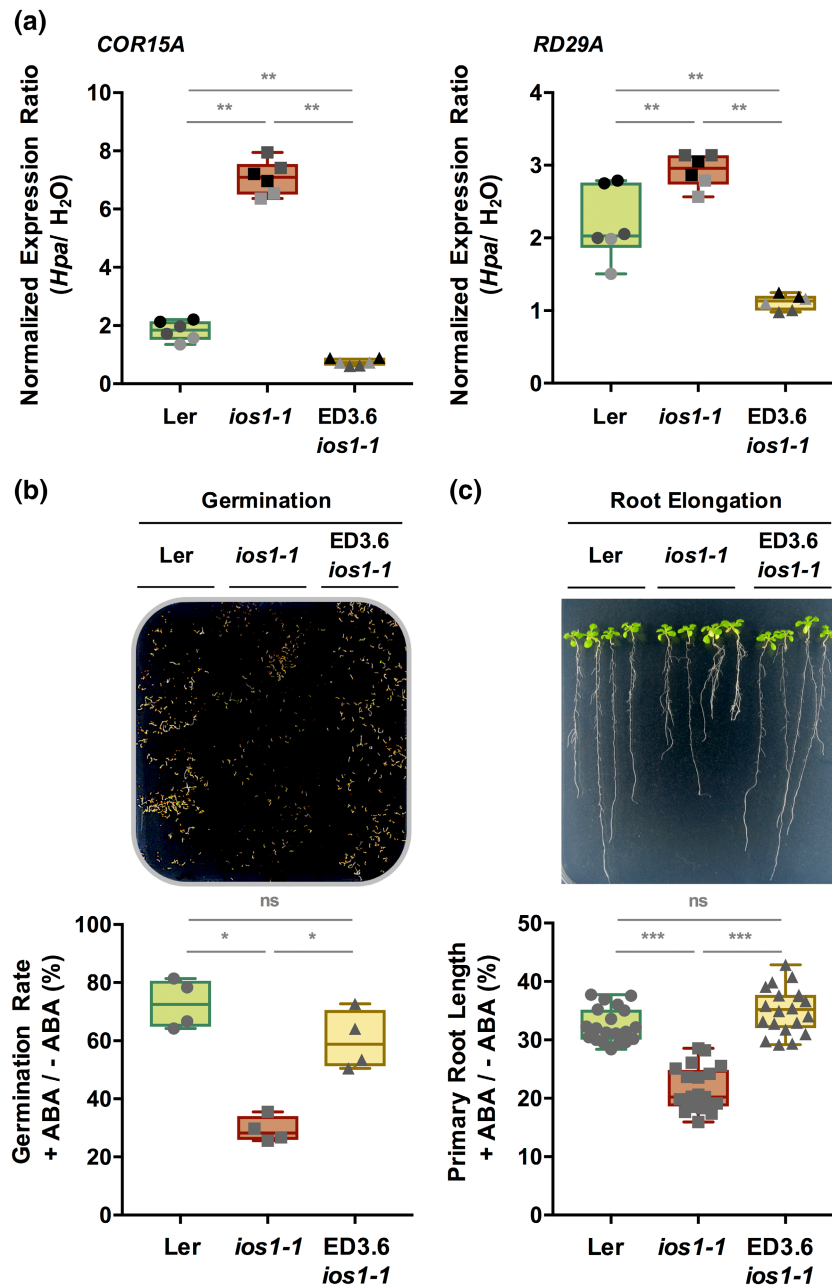
**FIGURE 1** Tunicamycin-induced unfolded protein response (UPR) and abscisic acid (ABA) responses. (a) The *ios1-1* mutant shows increased expression of marker genes for UPR signalling when compared to the wild type (Wt) (Ler). These increases are repressed after expression of the IOS1 exodomain in the mutant background (ED3.6 *ios1-1*), as revealed by analysis of transcript accumulation from the UPR marker genes *PDI5* and *SAR1*. (b) Tunicamycin treatment also leads to increases of ABA responses in *ios1-1* and their repression by the IOS1 exodomain, as revealed by analysis of transcript accumulation from the ABA marker genes *COR15A*, *COR15B*, *RD29A*, and *RD29B*. Reverse transcription-quantitative PCR data are expressed as the ratio between treated and control plants. Data are from three independent biological replicates each involving two technical replicates, and were analysed using *UBP22* and *UBQ10* as reference genes. Genes and primers are listed in Table S1. Asterisks in all graphs indicate statistically significant differences as determined by nonparametric Mann-Whitney test (\* $p < 0.05$ , \*\* $p < 0.01$ ). Biological replicates are represented by dots in different shades of grey and technical replicates are in the same shades of grey. ns, not significant



of *Arabidopsis* with Hpa, we analysed the expression of ABA-responsive genes in seedlings that were inoculated with the downy mildew pathogen. We observed stronger accumulation of *COR15A* and *RD29A* transcripts in the infected *ios1-1* mutant than in the Ler Wt (Figure 2a), reflecting the increased activation of ABA signalling on infection in the absence of IOS1 (Hok et al., 2014). This increased activation did not occur in line ED3.6, showing that the exodomain of IOS1 is sufficient to attenuate infection-responsive ABA signalling.

A reduction in seed germination and primary root elongation reflects the hypersensitivity of *ios1-1* to exogenous ABA application (Hok et al., 2014). The germination rate of Ler Wt seeds on medium

containing 10  $\mu\text{M}$  ABA was reduced by about 30% 5 days after sowing compared to germination on medium without ABA. For seeds of the *ios1-1* mutant, germination on medium containing ABA was reduced by about 70%, consistent with the previously observed phenotype. Expression of the extracellular IOS1 domain in the mutant background (line E3.6) was then sufficient to restore the ABA responsiveness of *ios1-1* seed germination to a level close to that of the Wt (Figure 2b). When seedlings growing on medium without ABA were transferred to fresh medium containing 10  $\mu\text{M}$  ABA, primary roots of Wt seedlings extended within 5 days to about 35% of the length that was gained in the absence of ABA. This ABA-induced growth



**FIGURE 2** Infection-responsive abscisic acid (ABA) signalling and ABA hypersensitivity. (a) Infection of *Arabidopsis* seedlings with the *Hyaloperonospora arabidopsidis* (Hpa) isolate Wela leads to increased ABA signalling in *ios1-1* when compared to the Ler wild type (Wt), as revealed by analysis of accumulation of transcripts from the ABA marker genes *COR15A* and *RD29A* 4 days postinoculation. The overactivation of marker genes for ABA responses is repressed on expression of the *IOS1* exodomain in the mutant background (ED3.6 *ios1-1*). Reverse transcription-quantitative PCR data were expressed as the ratio between inoculated and control plants. Data are from three biological replicates each involving two technical replicates, and were analysed using *ACT8* as reference gene. Biological replicates are represented by dots in different shades of grey and technical replicates are in the same shades of grey. (b) Hypersensitivity of *ios1-1* to exogenous ABA application leads to decreased seed germination. Seeds from the different lines were sown on medium containing or not containing 10  $\mu$ M ABA. Photographs of germinating seeds on ABA (top) were taken 7 days after sowing. For quantitative analyses, the number of seeds with and without emerging radicles was determined on four Petri dishes 5 days after sowing. (c) ABA hypersensitivity inhibits primary root elongation. Plants grown on medium without ABA were aligned on fresh medium containing or not containing 10  $\mu$ M ABA. The image (top) shows seedlings on ABA-containing medium 10 days after the transfer. Please note that the contrasts of the seed germination (b) and root elongation (c) images have been artificially enhanced to better identify radicles and roots. Graphs in (b) and (c) represent data expressed as the ratios of germination and root elongation between ABA-treated and untreated seeds and plantlets. Asterisks in all graphs indicate statistically significant differences as determined by nonparametric Mann-Whitney test (\* $p < 0.05$ , \*\* $p < 0.01$ , \*\*\* $p < 0.001$ ). ns, not significant

reduction was reinforced in the absence of IOS1 in the mutant, but restored to Wt levels on expression of the IOS1 exodomain in line ED3.6 (Figure 2c). Taken together, these results suggest that the exodomain is sufficient for the attenuation of ABA signalling defects caused in the *ios1-1* mutant.

### 2.3 | HAP6 stimulates ABA signalling on infection

IOS1 interacts with the ER-residing ribophorin HAP6, which positively regulates the UPR during infection with Hpa and thus antagonizes the activity of IOS1 in the ER (Giordano et al., 2022). We therefore addressed the question of whether HAP6, similar to IOS1, also affects ABA signalling. Because the *Arabidopsis hapless* full knockout mutant *hap6* has disrupted male functions and the corresponding mutation can only be maintained in plants that are heterozygous for the locus (Johnson et al., 2004), we inoculated the previously described HAP6 knockdown mutants *rpn2-1* and *rpn2-2* with Hpa. These mutants still produce HAP6 transcripts and exhibit relatively mild phenotypes with respect to the UPR and their interaction with pathogens, such as Hpa (Giordano et al., 2022) and the powdery mildew fungus (Weis et al., 2013). These phenotypes were more strongly pronounced in *rpn2-2* than in *rpn2-1* (Giordano et al., 2022; Weis et al., 2013). Accordingly, reductions in *COR15A* and *RD29A* transcript levels between inoculated Wt and *rpn2* mutants were weak, but they were significantly least in *rpn2-2* when compared to the Wt (Figure 3), thus indicating that HAP6 contributes to the stimulation of ABA signalling on infection. Taken together, our results show that the negative and positive effects on the UPR are reflected by the negative and positive effects of IOS1 and HAP6, respectively, on ABA signalling. These findings suggest that both proteins contribute to a crosstalk between ER stress and the hormonal pathways.

## 3 | DISCUSSION

Studies that have examined the relationship between the UPR and ABA signalling in plants are still scarce, but revealed regulatory elements that may coordinate both ER stress responses and ABA signalling (Chen et al., 2020). We show here that the exodomain of IOS1 and HAP6, which are responsible for modulating pathogen-induced ER stress (Giordano et al., 2022), also affect infection-associated ABA signalling during the interaction with the downy mildew pathogen. At present, we cannot reliably state if this occurs through independent mechanisms, if the effects on the UPR are a consequence of altered ABA signalling, or if the UPR is responsible for inhibiting or stimulating ABA responses (Figure 4). However, our observation that the UPR-inducing drug tunicamycin also activates transcription of marker genes for ABA signalling (Figure 1a) and the reported inefficiency of ABA treatments on bZIP60 mRNA splicing and stimulation of the UPR in *Arabidopsis* (Parra-Rojas et al., 2015) suggests that the regulation of ABA signalling is a consequence of ER stress rather than its trigger. This suggestion is supported by the finding that the TF bZIP17, a major player in one of the two UPR signalling pathways in plants (Manghwar & Li, 2022), is activated in maize during the UPR and interacts with the promoters of genes involved in both UPR and ABA signalling (Yang et al., 2013).

Another mechanistic link between UPR and ABA signalling might be provided by NAC (No apical meristem/*Arabidopsis thaliana* transcription activation factor/Cup-shaped cotyledon)-domain-containing TFs, which are highly diversified in plants and have 126 members in *Arabidopsis*. They integrate multiple stress signals in the positive or negative regulation of target gene expression. Members of the protein family coordinate responses to ABA (Fujita et al., 2004) and ER stress (Mendes et al., 2013). It is noteworthy that an effector protein from the oomycete pathogen *Phytophthora infestans* targets

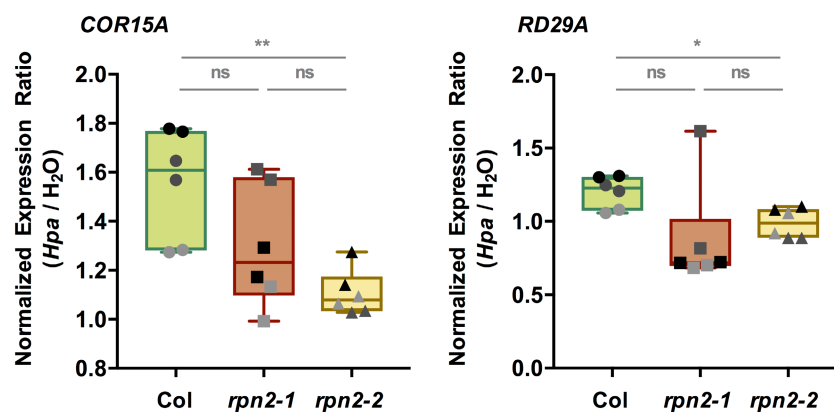
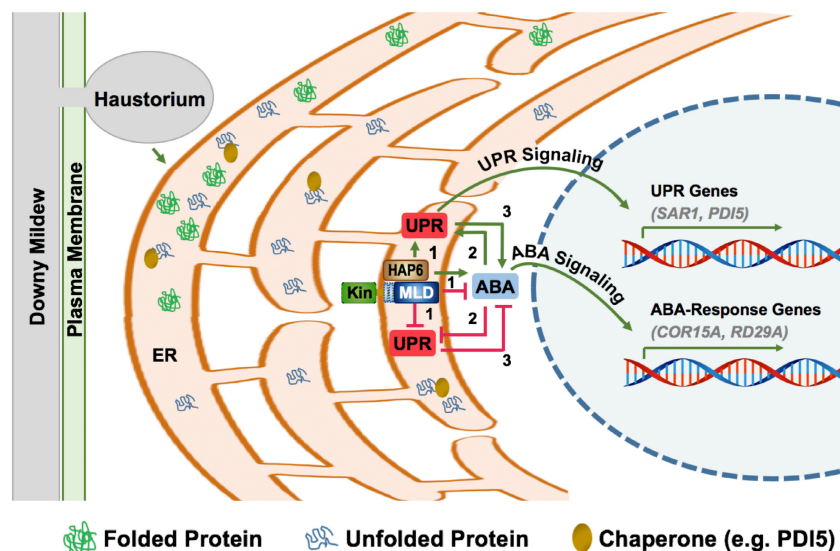


FIGURE 3 HAP6 stimulates abscisic acid (ABA) signalling on infection. Reduced HAP6 expression in *rpn2* knockdown mutants attenuates ABA signalling on infection. Seedlings from the wild type (Wt) (Col-0 background) and from the allelic mutant lines *rpn2-1* and *rpn2-2* (Giordano et al., 2022) were inoculated with *Hyaloperonospora arabidopsidis* isolate Noco2, and expression of the ABA marker genes *COR15A* and *RD29A* was analysed by reverse transcription-quantitative PCR 4 days after inoculation. Data are expressed as the ratio between inoculated and control plants. Data are from three biological replicates each involving two technical replicates, and were analysed using *UBP22* and *UBQ10* as reference genes. Biological replicates are represented by dots in different shades of grey and technical replicates are in the same shades of grey. Asterisks indicate statistically significant differences as determined by nonparametric Mann-Whitney test (\* $p < 0.05$ , \*\* $p < 0.01$ ). ns, not significant



**FIGURE 4** Proposed model for the regulation of the unfolded protein response (UPR) and abscisic acid (ABA) signalling during downy mildew infection. The biotrophic interaction provokes endoplasmic reticulum (ER) stress through excessive protein synthesis and the accumulation of unfolded proteins. Infection also induces IOS synthesis. IOS1 is partially retained in the ER by its exodomain, where it suppresses ER stress and interacts with the positive UPR regulator HAP6 (Giordano et al., 2022). The previously observed attenuation of ABA signalling (Hok et al., 2014) is also mediated by the exodomain of IOS1, whereas HAP6 tends to positively regulate ABA responses. Inhibition or stimulation of UPR and ABA may be achieved by independent mechanisms (1) or may be sequential events, with either the UPR being dependent on ABA (2) or the UPR being responsible for subsequent ABA responses (3). The IOS1 kinase domain (Kin) appears to be dispensable for the effects reported here. The chaperone PDI5, given here as an example, catalyses the formation of stabilizing disulphide bonds. MLD, malectin-like domain

two NAC-TFs in potato. The effector interacts with NAC proteins within plant cells and prevents their relocalization from the ER to the nucleus, which is required for target gene expression (McLellan et al., 2013). In *Arabidopsis*, NAC103 was identified as a direct target of spliced bZIP60, which transmits ER stress signals to downstream UPR genes (Sun et al., 2013). Moreover, NAC103 has been shown to regulate the expression of ABA-responsive genes, and its overexpression in transgenic *Arabidopsis* renders plants hypersensitive to the hormone (Sun et al., 2020). In our experiments, we observed that tunicamycin induced the expression of NAC103 and that this expression was down-regulated by the IOS1 exodomain (Figure S1). This observation makes the TF a potential candidate for integrating the effects of the IOS1 exodomain on ER stress responses and ABA signalling.

ABA signalling and its effects on tissue senescence (Asad et al., 2019) and stoma opening (Li et al., 2020) interfere with the biotrophic life cycle of downy mildew in *Arabidopsis*. In a similar way, ER stress responses like the UPR eventually influence cell survival and are detrimental to the development of filamentous biotrophs in plant tissues (Carvalho et al., 2014; Mishiba et al., 2013). Infection of *Arabidopsis* with Hpa triggers expression of the *IOS1* gene locally in cells harbouring haustoria (Hok et al., 2014). The *IOS1* exodomain suppresses UPR and ABA signals, and expression of this domain in a *ios1* knockout mutant makes plants susceptible to Hpa infection (Giordano et al., 2022). It is therefore tempting to speculate that downy mildew promotes *IOS1* synthesis to attenuate two host signalling pathways that negatively impact the biotrophic infection process.

In previous studies, we have shown that the transgenic expression of full-length *IOS1*, that a gene encoding the complete receptor with MLD, LRR, transmembrane, and kinase domains, can complement the ABA-related phenotypes of the *ios1-1* mutant (Hok et al., 2014). The data shown here and previous results (Giordano et al., 2022) suggest that the cytoplasmic *IOS1* kinase domain is not required for the described attenuation of infection-induced ABA and UPR responses in *Arabidopsis*. MLD-containing exodomains are part of more than 40 LRR-RLKs in *Arabidopsis* (Hok et al., 2011), but their function is not fully understood. The question of whether these domains from receptors other than *IOS1* can also affect UPR and ABA signalling, and interaction with microbes will certainly stimulate further research on the role of these exodomains in *Arabidopsis*. It should be noted that the sequence and predicted structure of the MLD of *IOS1* is similar to the MLD of the *Arabidopsis* receptor FERONIA (FER; Giordano et al., 2022). FER has multiple functions during the development, reproduction, and perception of environmental stimuli and was originally identified as an important regulator of female fertility in *Arabidopsis* (Huck et al., 2003). Similar to *IOS1*, FER promotes infection by a biotrophic filamentous pathogen (Kessler et al., 2010) and represses ABA signalling (Yu et al., 2012). Whether the exodomain of FER is responsible for these effects, similar to what we observed with *IOS1*, remains to be analysed. In conclusion, we believe that the present study will enrich the discussions on the relationship between ER stress responses and ABA signalling in plants, and on a possible role that exodomains of MLD-containing

receptors in plant cells might play in regulating the crosstalk between both events.

## 4 | EXPERIMENTAL PROCEDURES

### 4.1 | Biological material

*Arabidopsis* Wt ecotypes were Landsberg erecta (Ler) and Columbia-0 (Col). The *ios1-1* mutant (GT\_5\_22250) in the Ler background and the IOS1 exodomain-expressing *ios1-1* mutant line ED3.6 have been described previously (Giordano et al., 2022; Hok et al., 2011). Seeds from the homozygous *rpn2-1* (SALK\_019955) and *rpn2-2* (SALK\_017994) mutant lines are in the Col background and were obtained from the European Arabidopsis Stock Centre and have been characterized previously (Giordano et al., 2022; Weis et al., 2013). Seeds were sown on a soil/sand mixture, stratified for 2 days at 4°C, and then grown under a 12-h photoperiod in a growth chamber at 20°C. The downy mildew isolates Wela and Noco2 were used for inoculation of *Arabidopsis* lines with a Ler or Col genetic background, respectively. For infection, 10-day-old plants were spray-inoculated to saturation with a spore suspension of 40,000 spores/ml. Plants were kept in a growth cabinet at 16°C with a 12-h photoperiod.

### 4.2 | Plant treatments

For treatment with tunicamycin (SML1287; Sigma-Aldrich), about 200 sterilized *Arabidopsis* seeds were placed on a nylon net (0.4 mm pore size) on top of Gamborg agar plates (1% agar) with 1% sucrose. For germination over 10 days, the plates were placed at 21°C in an 8 h/16 h light/dark period. The mesh with the seedlings was then transferred onto plates with liquid Gamborg medium containing 7 µM tunicamycin and left overnight. Liquid Gamborg medium containing 0.1% dimethylsulphoxide c. was used as the control. For germination assays, seeds were sown on Gamborg's B5 medium including vitamins (Duchefa), which was complemented with 1% (wt/vol) sucrose and 10 µM ABA (A1049; Sigma-Aldrich). After stratification at 4°C for 2 days, the plates were placed at 21°C in an 8 h/16 h light/dark period. For root elongation assays, seeds were sown on medium without ABA before seedlings were aligned 7 days later on fresh medium containing or not containing 10 µM ABA. Root elongation was measured from this time point.

### 4.3 | Reverse transcription-quantitative PCR

Total RNA was prepared from plant tissues according to Hok et al. (2011, 2014) and 500 ng of RNA was reverse transcribed using SuperScript IV reverse transcriptase (ThermoFisher Scientific) according to the the supplier's instructions. The cDNA was diluted 10-fold, and 5 µl of this dilution was added to 10 µl of qRT-PCR

Brilliant III SYBR Master Mix (Agilent Technologies). Quantitative PCR (qPCR) was performed on an AriaMx Real-time PCR System (Agilent Technologies) with a programme consisting of 3 min of initial denaturation at 95°C, followed by 40 cycles consisting of 5 s at 95°C and 10 s at 60°C. Primers used for PCR experiments are listed in Table S1. All data were from three independent biological replicates each involving two technical replicates. Reference genes were AT4G05320, AT5G10790, and AT1G49240, as indicated in the figure legends.

### 4.4 | Quantification and statistical analysis

Data from AriaMx v1.0.8 were quantified with the qBase 1.3.5 Excel plugin (Biogazelle). Graph design and statistical analyses were performed using GraphPad Prism 7 software (GraphPad Prism Software Inc.). All graphs are represented as box and whisker plots. Statistically significant differences for RT-qPCR data and ABA response assays were determined by the nonparametric Mann-Whitney test. Significance groups are represented by stars above the boxplots in the respective graphs (\* $p < 0.05$ , \*\* $p < 0.01$ , \*\*\* $p < 0.001$ ).

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### CONFLICT OF INTEREST

The authors declare no conflict of interests.

### DATA AVAILABILITY STATEMENT

The data that support the findings are available from the corresponding author upon reasonable request.

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#### SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.

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