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REVIEW ARTICLE

Cold Stress Signaling Networks in Arabidopsis

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Abstract Cold is one of the critical environmental conditions that negatively affects plant growth and development and determines the geographic distribution of plants. Cold stress signaling is dynamic and interacts with many other signal transduction pathways to efficiently cope with adverse stress effects in plants. The cold signal is primarily perceived via Ca^{2+} channel proteins, membrane histidine kinases, or unknown sensors, which then activate the sophisticated cold-responsive signaling pathways in concert with phytohormone signaling, the circadian clock, and the developmental transition to flowering, as a part of the stress adaptation response. In this review, we focus on crosstalk between cold signaling and other signal transduction pathways in Arabidopsis.

Keywords: Circadian clock, Cold stress signaling, Crosstalk, Flowering, Phytohormone

Introduction

Plants are sessile organisms that are constantly exposed to various environmental stresses such as cold, drought, and high salinity. The environmental stresses limit plant growth and development and significantly reduce the yields of agriculturally important crops. Drought and high salinity affect > 10% of arable land and decrease average yields of most major crop plants by > 50%, and desertification and salinization are increasing on a global scale (Bray et al. 2000).

Cold adversely affects crop productivity and determines the geographic distribution of plants. Plants in tropical and subtropical regions are generally killed by a slight freeze, whereas plants in temperate regions exhibit varying degrees of freezing tolerance through the process of cold acclimation (Sakai and Larcher 1987). Cold acclimation is defined as the process in which plants adjust their metabolism to cold and acquire enhanced freezing tolerance after being exposed to nonfreezing cold temperatures (Thomashow 1999). A variety of numerous genes are up or down-regulated at the transcriptional level during cold acclimation, and their gene products play roles in the acquisition of stress tolerance (Shinozaki and Yamaguchi-Shinozaki 2000). Cold stress transcriptionally activates C-REPEAT/DEHYDRATION-RESPONSIVE ELEMENT BINDING FACTORS (CBFs/DREBs) that bind to the C-REPEAT/DEHYDRATION-RESPONSIVE ELEMENT (CRT/DRE), inducing expression of a number of their target genes. Forward and reverse genetic approaches have revealed several components that are involved in the CBF/DREB cold signaling pathway (Shinozaki et al. 2003; Chinnusamy et al. 2004, 2007; Kim 2007). Cold transcriptome analysis has shown that CBFs regulate only ~12% of the cold-responsive transcriptome (Fowler and Thomashow 2002), suggesting that other cold regulatory gene networks contribute to acquisition of freezing stress tolerance. Cold stress signaling integrates with various other signaling pathways to coordinate stress adaptation response with plant growth and development. Various phytohormones play roles in the cold acclimation process or freezing tolerance response. Cold can affect expression of genes involved in gibberellin (GA) signaling (Achard et al. 2008). Regulation of auxin homeostasis is involved in the stress adaptation response (Park et al. 2007). Several key components of cytokinin signaling play a role in cold signaling (Jeon et al. 2010; Jeon and Kim 2013). In addition to phytohormones, the circadian clock and developmental transition to flowering interact with cold signaling. This review describes cold stress signaling from early events of cold perception to gene regulation and signaling networks in Arabidopsis.

Early Events in Cold Signaling

A member of the transient receptor potential (TRP) cation channel family, TRP subfamily M member 8 (TRPM8),

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functions in cold temperature sensing in a subset of neurons (Peier et al. 2002; Karashima et al. 2009). TRPM8 is a nonselective cation channel stimulated by cold and a cooling agent, menthol (Peier et al. 2002). When temperature is lowered from 25°C to 15°C, an increase in intracellular calcium is observed in TRPM8-expressing mammalian cells. A stimulus at 10°C also evokes an influx of Ca^{2+}. This response is dependent on Ca^{2+} in the buffer, because removal of extracellular calcium suppresses this temperature response. TRPA1 has also been identified as a major sensor for noxious cold in mice (Karashima et al. 2009). Ca^{2+} channels may also be involved in early cold signaling events in higher plants. Cold can induce an immediate rise in cytosolic free calcium concentration in Arabidopsis and tobacco and activate calcium-permeable channels in Arabidopsis mesophyll cells (Knight et al. 1996; Plieth et al. 1999; Carpaneto et al. 2007). Alfalfa (*Medicago sativa*) cells treated with agents blocking Ca^{2+} influx are unable to cold-acclimate (Monroy et al. 1993). Conversely, chemical agents causing calcium influx induce *COLD ACCLIMATIZATION-SPECIFIC* (*CAS*) gene expression in alfalfa at 25°C. Arabidopsis treated with inhibitors of calcium flux also shows inhibited cold acclimation as well as expression of *COLD-INDUCED* (*KIN*) genes (Tähtiharju et al. 1997). A calcium sensor SYT1, a homolog of synaptotagmin that initiates exocytosis, functions in calcium-dependent freezing tolerance by resealing punctured plasma membrane sites (Yamazaki et al. 2008). A transcriptome analysis in response to increased calcium concentration has revealed various calcium-regulated promoter elements, implying plant signaling via increase in an intracellular calcium concentration in response to a variety of stimuli including cold (Whalley et al. 2011).

A correlation between a decrease in membrane fluidity and cold-induced gene expression has been reported in plants and microorganisms, indicating that the cold-induced decrease in membrane fluidity might be a sensing event during perception of cold signals. For example, membrane fluidizer benzyl alcohol inhibits induction of the *COLD ACCLIMATION-SPECIFIC* (*CAS30*) gene, calcium influx, and freezing tolerance at 4°C in alfalfa protoplast cells, whereas dimethyl sulfoxide acting as a membrane rigidifier induces *CAS30* gene expression, calcium influx, and freezing tolerance at 25°C (Õrvar et al. 2000). Plasma membrane protein histidine kinases have been proposed as the primary sensors of a decrease in temperature in the cyanobacterium *Synechocystis* sp. PCC 6803 (Murata and Los 1997; Suzuki et al. 2000). Cyanobacteria modulate the composition of membrane lipids in response to a temperature shift from 34°C to 22°C to increase fluidity of their membranes for adaptation to cold stress by enhancing expression of three fatty-acid *DESATURASE GENES* (*DES*): *DESA*, *DESB,* and *DESD* (Los et al. 1997). *HISTIDINE KINASE33* has been proposed to act as a sensor that detects a decrease in membrane fluidity caused by a temperature drop (Suzuki et al. 2001; Mikamiet et al. 2002). In Arabidopsis, ARABIDOPSIS HISTIDINE KINASE2 (AHK2) and AHK3 are involved in recognizing cold temperature at 1°C to induce expression of a subset of type-A *ARABIDOPSIS RESPONSE REGULATOR* (*ARR*) genes, *ARR5*, *ARR6*, *ARR7*, and *ARR15* (Jeon et al. 2010).

Cold activates proteolytic processing of a plasma-membrane-anchored NAC transcription factor NTL6 to elicit a pathogen-resistance response, in part, by inducing a small group of cold-inducible pathogenesis-related genes (Seo et al. 2010a; Seo et al. 2010b). NTL6 processing is stimulated by agents that reduce membrane fluidity, but is inhibited by treatment of linolenic acid, a 18:3 unsaturated fatty acid. A metalloprotease inhibitor blocks NTL6 processing. These results suggest that the reduction in membrane fluidity caused by cold stimulates the activity of a metalloprotease, cleaving the membrane-bound NTL6 transcription factor to migrate to the nucleus and induce a pathogen-resistance response.

Chromatins might be a site of sensing ambient low temperature (Kumar and Wigge 2010). *ACTIN-RELATED PROTEIN 6* (*ARP6*), which encodes a subunit of the SWITCH2/SUCROSE NONFERMENTABLE2 (SWI/SNF)-related adenosine triphosphatase complex, is necessary for inserting the alternative histone H2A.Z into nucleosomes in place of H2A (Deal et al. 2007). *arp6* mutants display a constitutive warm temperature developmental program under a low temperature such as 12°C, showing that nucleosomes containing the alternative histone H2A.Z are essential for perceiving ambient temperature. H2A.Z confers distinct DNA-unwrapping properties on nucleosomes, which might be a direct mechanism for perceiving ambient temperature through chromatin remodeling (Kumar and Wigge 2010).

CBF Cold Signaling Pathway

The CBF cold response pathway is the most well characterized cold signaling pathway at molecular levels in plants. *Arabidopsis thaliana* has three *CBF* genes (*CBF1/DREB1B*, *CBF2/DREB1C*, and *CBF3/DREB1A*) that encode closely related APETALA2/ETHYLENE RESPONSE FACTOR family transcription factors (Riechmann et al. 2000). These *CBF* genes are strongly and transiently induced by cold stress (Gilmour et al. 1998; Liu et al. 1998, Medina et al. 1999; Fowler and Thomashow 2002). *CBF3/DREB1A* or *CBF1/DREB1B*-overexpressing Arabidopsis plants exhibit strong tolerance to freezing, drought, and high salinity stresses (Jaglo-Ottosen et al. 1998; Liu et al. 1998; Kasuga et al. 1999). The CBFs/DREBs (hereafter CBF) bind the CRT/

DRE in the promoters of the CBF target genes, inducing expression of a few hundreds of genes including *COLD-REGULATED15A* (*COR15A*) referred to as the CBF regulon (Stockinger et al. 1997; Liu et al. 1998; Steponkus et al. 1998; Maruyama et al. 2004; Vogel et al. 2005). INDUCER OF CBF EXPRESSION 1 (ICE1), a MYC-like bHLH transcriptional activator, functions as a positive regulator of *CBF3* but has little effect on *CBF1* and *CBF2* expression (Chinnusamy et al. 2003). SIZ1 (yeast SCAFFOLD ATTACHMENT FACTOR A/B/ACINUS/PIAS DOMAIN and MSX-INTERACTING-ZINC FINGER1), SMALL-UBIQUITIN MODIFIER (SUMO) E3 ligase, mediates sumoylation of ICE1, leading to an increase in ICE1 activity or protein stability as an early response of cold signaling for *CBF3* expression (Miura et al. 2007). This activation response is attenuated by a RING-type ubiquitin E3 ligase, HIGH EXPRESSION OF OSMOTICALLY RESPONSIVE GENE (HOS1), by degrading ICE1 via ubiquitin-mediated proteasome pathway (Dong et al. 2006). Cytosolic HOS1 shuttles to the nucleus at a late time in response to cold and mediates degradation of ICE1 in the nucleus (Lee et al. 2001; Dong et al. 2006). This attenuation process might be a mechanism for transient expression of *CBF*s in response to cold allowing increased tolerance against freezing stress but without causing constitutive suppression of plant growth. The serine 403 of ICE1 is a key residue for attenuating the cold-stress response due to HOS1-mediated degradation of ICE1 (Miura et al. 2011). Consistent with this result, ICE1 (S403A)-overexpressing transgenic Arabidopsis plants show enhanced cold-responsive gene expression and freezing tolerance compared with those of ICE1-overexpressing transgenic plants. The abundance of ICE1 (S403A) proteins is not altered during cold signaling, whereas that of wild-type ICE proteins decreases. MYB15 encoding the R2R3-type MYB transcription factor binds to MYB recognition sequences in the *CBF* gene promoters as well (Agarwal et al. 2006).

Calcium signaling seems to be linked to the cold signaling pathway upstream of *CBF*s. The *CBF2* promoter has seven conserved DNA motifs (CM), called CM1 to CM7 (Doherty et al. 2009). The CM2 sequence contains the conserved CG-1 element consisting of a 5'-CGCG-3' sequence, which is a binding site for the calmodulin binding transcription activators (CAMTAs) in Arabidopsis (Finkler et al. 2007). CAMTA3 has been identified as a positive regulator of *CBF2* expression (Doherty et al. 2009). A *camta3* mutation impairs the cold induction of *CBF1*, *CBF2*, and *ZINC TRANSPORTER OF ARABIDOPSIS THALIANA12* (*ZAT12*) that have a CG-1 element in the promoter regions, suggesting that these genes might be direct targets of CAMTA3. *camta1 camta3* double mutants exhibit decreased freezing tolerance compared with that of the wild type. These results suggest that CAMTAs might be a direct link between calcium signals

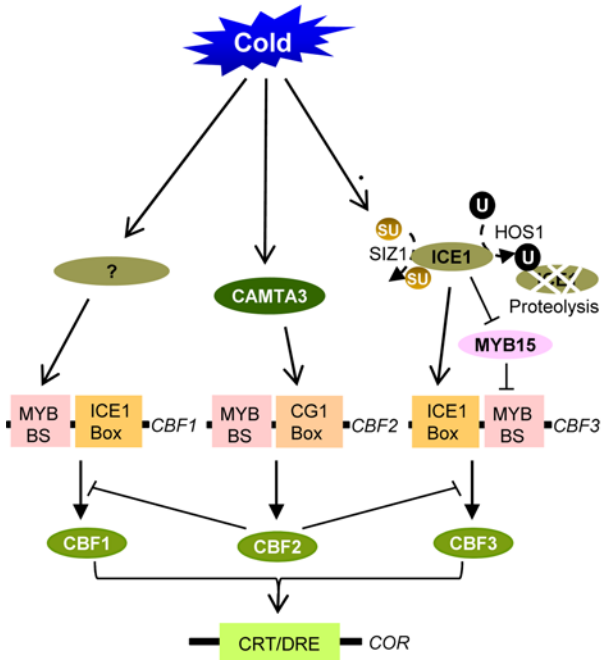

Fig. 1. The CBF cold signaling pathway. Broken arrows indicate post-translational regulation, and solid arrows indicate activation. Lines ending with a bar show negative regulation. SU, SUMO (small ubiquitin-related modifier); U, ubiquitin; CAMTA, calmodulin-binding transcription activator; CBF, C-repeat/dehydration-responsive element binding factor; COR, cold-regulated; CRT/DRE, C-repeat/dehydration-responsive element; MYBBS, MYB binding site; ICE1 box, ICE1 binding site; CBF/DREB, C-repeat binding factor/dehydration-responsive element binding factor; ICE1, inducer of *CBF* expression1; HOS1, high expression of osmotically responsive gene1; SIZ1, for yeast SAP and MIZ1.

and cold-regulated gene expression for cold acclimation. Fig. 1 depicts the regulatory networks of the CBF cold signaling pathway in Arabidopsis.

Microarray analysis has shown that CBFs regulate only a part of the cold-responsive transcriptome, suggesting that other multiple cold gene regulatory networks might contribute to the cold response and acquisition of freezing tolerance (Fowler and Thomashow 2002). For example, the *eskimo1* (*esk1*) mutant of Arabidopsis accumulates constitutively high levels of proline and is constitutively freezing tolerant (Xin and Browse 1998). Transcriptome profiling of the *esk1* mutant showed greater overlap with sets of genes regulated by salt, osmotic and abscisic acid treatments than genes regulated by cold acclimation or by CBF3 and ICE1. Thus, *esk1*-induced freezing tolerance might have a distinct molecular basis from that of CBF-dependent cold acclimation (Xin et al. 2007). *HOS9* encoding a putative homeodomain transcription factor also confers enhanced freezing tolerance to Arabidopsis by regulating genes that are independent of the CBF signaling pathway (Zhu et al. 2004). *AHK2* and *AHK3* mediate the cold signal for type-A *ARR* expression independently of the CBF pathway (Jeon et al. 2010).

However, AHK2 and AHK3 modulate some CBF3 target genes via negative regulation of MYB15, which acts as a negative regulator of *CBF3*, indicating that there is a link between the CBF3 response pathway and the AHK2/AHK3-responsive cold signaling pathway via MYB15 (Jeon and Kim 2013; Kim and Jeon 2013).

Crosstalk between Cold Signaling and Hormone Signaling

Hormones are involved in the plant adaptive responses to biotic and abiotic environmental stimuli. GAs play an important role in the abiotic stress response. Among the GA signaling components, DELLA proteins specifically arrest plant growth in adverse environmental conditions to promote survival (Achard et al. 2006, 2008). For example, salt slows growth through a DELLA-dependent mechanism that is associated with reduced accumulation of bioactive GAs (Achard et al. 2006). Quadruple-DELLA mutants show a reduced effect of salt slowing the leaf production rate, leaf expansion, and biomass accumulation. *CBF1*-overexpressing transgenic plants display retarded growth linked to enhanced freezing tolerance by accumulation of DELLA proteins resulting from reduced GA content caused by induction of GA2-OXIDASE inactivating GAs (Achard et al. 2008). The GA signal is perceived by GA-INSENSITIVE DWARF1 (GID1), promoting interaction between GID1 and the DELLA proteins, which then results in degradation of the DELLA proteins through the ubiquitin-proteasome pathway. Reduced GA levels induce the accumulation of DELLA proteins and restrain plant growth, causing a dwarf stature and delayed flowering. They further showed that DELLAs contribute significantly to the function of CBFs during cold acclimation and freezing tolerance by a distinct mechanism in which the CBF regulon such as *COR*s confers freezing tolerance.

Cold inhibits root basipetal auxin transport (Shibasaki et al. 2009). Cold causes dramatically reduced trafficking of the auxin efflux carrier PIN2 and inhibits the lateral relocalization of PIN3. These results suggest that cold stress negatively affects intracellular trafficking of auxin efflux carriers. Auxin mediates biotic and abiotic stress adaptation responses by modulating auxin homeostasis through a *GH3* gene encoding an auxin-conjugating enzyme (Park et al. 2007). *weso* (*wes*)1-*D*, an enhancer mutant of the IAA-conjugating GH3 enzyme, has been isolated, and shows retarded growth and induces expression of pathogenesis-related genes and *CBF*s. *wes1-D* mutants exhibit enhanced abiotic and biotic stress resistance, whereas a *wes1* T-DNA insertion mutant shows reduced stress resistance. These results suggest that regulation of auxin homeostasis by IAA-conjugating GH3 enzymes represents a way to modulate stress adaptation response through the fitness cost of induced resistance.

Several key components of the cytokinin signal transduction pathway play a role in cold signaling (Jeon et al. 2010; Jeon and Kim 2013). *AHK2* and *AHK3* encoding cytokinin receptors are involved in mediating the cold signal to express a subset of type-A *ARR*s, such as *ARR5*, *ARR6*, *ARR7*, and *ARR15* (Jeon et al. 2010). Overexpression of the cold-inducible *ARR7* in Arabidopsis results in a hypersensitivity response to freezing temperatures under cold acclimated conditions, whereas the type-A *arr* mutants, *arr5*, *arr6*, and *arr7*, show enhanced freezing tolerance, indicating that cold-inducible type-A *ARR*s play a negative regulatory role in cold stress signaling. ARR1, one of the type-B ARRs, plays a positive role in cold-induced expression of type-A *ARR*s (Jeon and Kim 2013). Results of cytokinin pretreatment experiments suggest that the cytokinin signal transduction pathways coact with the cold signaling during cold stress adaptation response (Jeon et al. 2010; Jeon and Kim, 2013). AHP2, AHP3, and AHP5 are redundantly involved in mediating the cold signal for type-A *ARR* expression. The cold transcriptome affected by *ahk2 ahk3* mutations has been identified by microarray analysis, revealing a new cold-responsive gene network regulated downstream of AHK2 and AHK3. Ten of the 57 genes identified as the cold transcriptome are ARR1-target genes and five of the 57 genes are CBF3 downstream genes. Many of the genes identified as the cold transcriptome are not regulated by CBF3 or ARR1, indicating the existence of unidentified transcription factors mediating the cold response downstream of AHK2 and AHK3. In addition to cold signaling, AHK2 and AHK3 are also involved in salt and dehydration stress tolerance responses (Nishiyama et al. 2011; Kang et al. 2012). Both cytokinin receptor-dependent and receptor-independent pathways operate during the dehydration response regulating *ARR* gene expression (Kang et al. 2013). Microarray analysis of *ahk2 ahk3* mutants has revealed significant networks among hormone signaling in Arabidopsis (Tran et al. 2007; Jeon and Kim 2012). AHK1 acts as a positive regulator of the drought and salt stress responses and ABA signaling (Tran et al. 2007). Microarray analysis of the *ahk1* mutant reveals down-regulation of many stress- and/or ABA-inducible genes, and DREB2A transcription factors and their downstream genes (Tran et al. 2007). These results suggest that there are extensive crosstalk and networks among plant hormone signaling pathways during the abiotic stress response including cold. The gene regulatory networks of the cold signaling and hormone signaling pathways are shown in Fig. 2.

Role of the Circadian Clock in Cold Signaling

Plants that have a correctly matched circadian clock to the

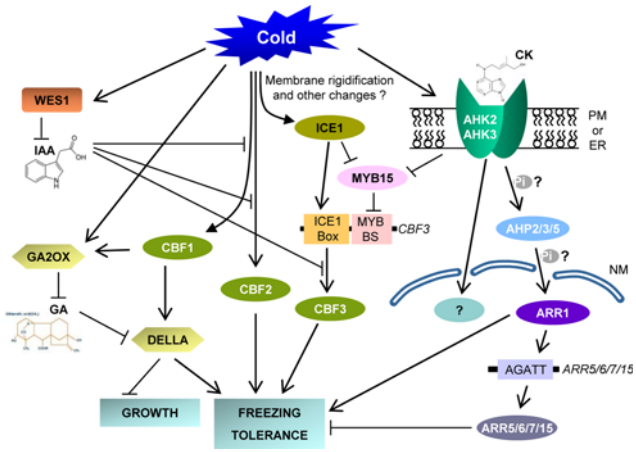

Fig. 2. Cold and hormone signaling networks in Arabidopsis. Arrows indicate activation, and lines ending with a bar show negative regulation. AHK, Arabidopsis histidine kinase; AHP, Arabidopsis histidine phosphotransfer protein; ARR, Arabidopsis response regulator; CK, cytokinin; ER, endoplasmic reticulum; IAA, indole acetic acid; GA, gibberellic acid; GA2OX, GA2 oxidase; NM, nuclear membrane; PM, plasma membrane; WES1, weso1 (IAA-conjugating GH3 enzymes). See Fig. 2 legend for other abbreviations.

environment contain more chlorophyll, fix more carbon, grow faster, and survive better than plants with circadian periods differing from their environment (Dodd et al. 2005). A variety of studies have revealed the critical importance of the circadian clock in the cold response. Increased chilling tolerance correlates with increased levels of polyunsaturated fatty acids in cotton during circadian clock (Rikin et al. 1993). *CBF1*, *CBF2*, and *CBF3* are subject to circadian regulation and their cold induction is gated by the circadian clock (Fowler et al. 2005). Similarly, the cold responsiveness of *RAV1* and *ZAT1*, the genes that are cold-induced in parallel with *CBF*s, is also subject to circadian regulation. The functional implication of the circadian rhythm in CBF-imparted cold tolerance has been demonstrated by an analysis of *circadian clock-associated 1* (*cca1*) and *late elongated hypocotyl* (*lhy*) double mutants exhibiting impaired freezing tolerance and diminished circadian regulation and cold induction of three CBF regulon genes (Dong et al. 2011). Overexpression of *CCA1*α confers freezing tolerance in Arabidopsis, whereas that of *CCA1*β results in increased sensitivity to freezing tolerance and moreover, cold temperatures reduces *CCA1*β production by suppressing *CCA1* alternative splicing, indicating that cold regulation of *CCA1* alternative splicing contributes to freezing tolerance (Seo et al. 2012). Arabidopsis *PSEUDO RESPONSE REGULATOR* (*PRR*) genes are components of the circadian clock. Arabidopsis *prr9 prr7 prr5* arrhythmic triple mutants exhibit enhanced freezing tolerance and highly express *CBF* transcripts compared to those of the wild-type (Nakamichi et al. 2009). Disruption of the Arabidopsis circadian clock causes extreme variation in the cold-responsive transcriptome (Bieniawska et al. 2008). Analysis of the *cis*-acting elements in the promoter regions of the AHK2/AHK3-responsive cold transcriptome shows over-representation of circadian responsive elements including EE and/or EE-like sequence elements and CCA1-binding sites (~65%), indicating that circadian rhythm might be an important aspect of cold responsive gene expression via AHK2 and AHK3 (Jeon and Kim 2013).

Cold Signaling and Flowering

Forward genetic and biochemical approaches suggest that FVE, a component of the autonomous pathway in Arabidopsis flowering time control, is a link between cold response and flowering time in Arabidopsis (Kim et al. 2004; Jeon and Kim 2011). They used a targeted genetic approach to isolate *acg1* mutant and identified ACG1 as a negative regulator of CBF pathway (Kim et al. 2004). The *acg1* mutant, a null allele of *fve*, shows increased expression of *CBF*s and their downstream genes such as *COR15A* and *COR47* with and without cold treatment compared with that in the wild-type. The *acg1* mutant exhibits late flowering with elevated expression of *FLOWERING LOCUS C* (*FLC*), a repressor of flowering. FVE encodes a homologue of the mammalian RETINOBLASTOMA-ASSOCIATED PROTEIN (RbAp), one component of a HISTONE DEACETYLASE (HDAC) complex involved in transcriptional repression, and has been shown to be involved in the deacetylation of *FLC* chromatin (Ausín et al. 2004). Chromatin immunoprecipitation experiments have revealed that FVE binds to the *FLC* and *COR15A* chromatin. Gel-filtration chromatography and the immunoprecipitation of putative FVE complexes showed that FVE forms a protein complex of approximately 1.0 MDa. These results indicate that FVE may exist as a multiprotein complex, similar to the mammalian HDAC complex harboring RbAp, to regulate flowering time and cold response by associating with *FLC* and *COR* chromatin.

SUPPRESSOR OF OVEREXPRESSION OF CONSTANS1 (*SOC1*), one of the key floral activators integrating multiple floral inductive pathways, negatively regulates expression of the cold response genes through direct repression of *CBF* transcription (Seo et al. 2009). Overexpression of cold-inducible *CBF*s causes late flowering through increased expression of *FLC*, an upstream negative regulator of *SOC1*. These results suggest that a feedback loop may operate between cold response and flowering-time regulation. This loop could prevent premature flowering under cold conditions in fall or early spring but suppress the cold response when floral induction occurs (Seo et al. 2009). *LONG VEGATATIVE PHASE 1* (*LOV1*), encoding a floral repressor that negatively regulates *CONSTANS* (*CO*) expression under long-day conditions, regulates the cold response by

upregulating *COR15A* and *KIN* expression (Yoo et al. 2007). This result suggests that LOV1 plays a role as a common regulator of two intersecting pathways that regulate flowering time and the cold response. Moreover, CO proteins are degraded by cold via an HOS1-dependent ubiquitination mechanism, resulting in suppression of the floral integrator *FLOWERING LOCUS T* (*FT*) and thus delaying flowering during cold stress (Jung et al. 2012).

Concluding Remarks

A variety of studies using genetic, reverse-genetic, and biochemical approaches have revealed many signaling components involved in the cold stress response in plants. Significant advances have been achieved in understanding the molecular mechanisms underlying cold stress signaling and integration and coordination with plant growth and development via interconnection with hormone signaling pathways, circadian clock, and flowering in Arabidopsis. However, much still remains to be elucidated regarding how the hormone signaling pathways and circadian clock are linked to the plant adaptation response to cold stress and how these signal transduction pathways coact with cold-responsive transcription factors during plant adaptation to cold stress. This knowledge will not only contribute to the molecular understanding of plant interaction with the environment but also provide efficient genetic tools to engineer stress tolerant crops without compromising yield associated with growth retardation.

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