Molecular genetic studies of the memory of winter

Sibum Sung and Richard M. Amasino*
Department of Biochemistry, 433 Babcock Drive, University of Wisconsin at Madison, Madison, WI 53706, USA

Received 19 May 2005; Accepted 30 June 2006

Abstract
Many plant species have evolved the ability to flower in the proper season by sensing environmental cues. The prolonged cold of winter is one such cue that certain plants use to acquire competence to flower the following spring. For example, biennials and winter annuals become established in one growing season and often flower quickly in the early spring of the following year to complete their life cycles. The process by which exposure to prolonged cold establishes competence to flower is known as vernalization. Many studies, starting with the classic work of Lang and Melchers, have shown that the vernalized state can be stable; i.e. after exposure to cold has ended, competence to flower, in certain species, can persist for many months and throughout many cell divisions in the shoot apical meristem. Thus, plants can exhibit a ‘memory of winter’ and vernalization can result in an epigenetic switch in the classic sense of the term: a change that is stable in the absence of the inducing signal. The nature of this epigenetic switch in Arabidopsis thaliana is discussed here.

Key words: Chromatin remodelling, epigenetics, histone code, memory of winter, vernalization.

Introduction
In the model plant Arabidopsis, FLOWERING LOCUS C (FLC) plays a major role in creating a requirement for vernalization (Michaels and Amasino, 1999; Sheldon et al., 1999). FLC is a strong repressor of flowering, and in the first growing season, a high level of FLC expression prevents plants from flowering unless vernalized (Michaels and Amasino, 2000; Sung and Amasino, 2005). FLC expression is repressed by prolonged exposure to cold, and FLC repression is maintained for the rest of plant life cycle even after cold exposure ends (Michaels and Amasino, 1999; Sheldon et al., 1999). Thus, in Arabidopsis, the ‘memory of winter’ is manifest as stable repression of FLC, and the vernalized state is an epigenetic switch of FLC expression (Sung and Amasino, 2005). Indeed, the vernalization-mediated repression of FLC provides an excellent system to study epigenetic regulation in an actual developmental context in plants.

Sensing winter
Cold exposure must trigger cellular signalling events for vernalization to take place, but little is known about the cold-sensing mechanism that initiates the necessary cellular signalling. In Arabidopsis, this signalling leads to the expression of a gene essential for the vernalization response, VERNALIZATION INSENSITIVE 3 (VIN3) (Sung and Amasino, 2004). VIN3 is expressed only during cold exposure, and the induction kinetics of VIN3 is correlated with the duration of cold exposure and the strength of the vernalization response. Clearly, regulatory components must exist to mediate cold-induced VIN3 expression. Dissecting the VIN3 regulatory system may lead to upstream components of the vernalization pathway that are involved in cold perception.

VIN3 encodes a Plant Homeo Domain (PHD) finger-containing protein (Sung and Amasino, 2004). PHD finger-containing proteins are often components of chromatin-remodelling complexes, and the PHD finger has been implicated in various biochemical functions including protein–protein interactions, nucleosome binding, and, as recently discovered, phospholipid binding (Fig. 1) (Gozani et al., 2003; Bienz, 2006). Interestingly, phospholipid binding is important for the proper localization of certain PHD finger-containing proteins (Gozani et al., 2003). The possibility that the binding of phospholipid by VIN3 could be part of the signalling pathway in the vernalization response is particularly intriguing, although the role, if any, of phospholipid binding remains to be investigated.

* To whom correspondence should be addressed. E-mail: amasino@biochem.wisc.edu

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Remembering winter
The epigenetic nature of the vernalized state and, in Arabidopsis, the mitotically stable repression of FLC indicates the involvement of a mechanism for conferring cellular memory. Of course, cellular memory is common in biology; many well-studied examples occur during the unfolding of animal development in which certain developmental genes are expressed or repressed at certain developmental stages in specific tissues (Ringrose and Paro, 2004). The pathway for cellular memory in animals is often developmentally pre-programmed, but the cellular memory of vernalization is triggered by the environmental cue of long-term cold temperature. Recent molecular studies have revealed that some components of cellular memory are conserved between plants and animals, but there are also some differences.

The involvement of Polycomb Repression Complex 2 in vernalization
Genetic studies have identified a number of genes that are involved in cellular memory in animals. For example, Polycomb Group genes (PcG) encode proteins required for the stable repression of certain developmental genes, whereas Trithorax Group genes (TrxG) are necessary for the maintenance of active expression of certain genes (Ringrose and Paro, 2004). Mutations in PcG and TrxG result in altered cell-identity phenotypes.

In plants, the first direct evidence of the involvement of PcG-mediated FLC repression in vernalization was the cloning of VERNALIZATION 2 (VRN2) (Gendall et al., 2001). VRN2 is required for the stable repression of FLC by vernalization. VRN2 encodes a homologue of Suppressor of Zeste 12 (Su(z)12), a PcG component first identified in Drosophila. Specifically, Su(z)12 is a part of Polycomb Repression Complex 2 (PRC2) (Cao et al., 2002; Kuzmichev et al., 2002; Muller et al., 2002). Many major components of PRC2 are evolutionally conserved between animals and plants (Hsieh et al., 2003; Schubert et al., 2005). In plants, PRC2 components have also been shown to be involved in the stable repression of certain developmentally regulated genes (Hsieh et al., 2003; Schubert et al., 2005). Thus, it is likely that some
form of PRC2 repression was established prior to divergence of plants and animals.

The winter code
In addition to the genetic information carried on the primary DNA sequence, chromatin structure has emerged as another source of relatively stable genetic information in eukaryotes (Jenuwein and Allis, 2001). For example, a gene is not typically expressed if it resides in highly condensed chromatin (i.e. heterochromatin), but if the chromatin is converted to a more open or relaxed state, the gene becomes available as a template for transcription (Richards and Elgin, 2002). An open, potentially active state of chromatin is often called euchromatin (Richards and Elgin, 2002). The ‘histone code hypothesis’ is that specific covalent modifications on chromatin serve as a ‘code’ to establish states of gene expression (Jenuwein and Allis, 2001). Numerous modifications have been identified specific proteins to form higher orders of chromatin structure that can be stable through cell generations (Jenuwein and Allis, 2001). Numerous modifications have been identified and the corresponding modifying enzymes are also being discovered (Table 1, and references therein).

Table 1. Histone modifications of eukaryotic chromatin

| Modifications          | Implications for gene expression in yeast and animals                                                                 | FLC chromatin vernalization change |
|------------------------|------------------------------------------------------------------------------------------------------------------------|-----------------------------------|
| H3K4 trimethylation    | Correlated with active gene at 5' end of transcripts (Bernstein et al., 2005; Liu et al., 2005; Margueron et al., 2005; Pokholok et al., 2005) | Decrease by vernalization (Sung et al., 2006) |
| H3K4 dimethylation     | Increase in the middle of ORFs (Bernstein et al., 2005; Liu et al., 2005)                                                 | Decrease by vernalization (Bastow et al., 2004) |
| H3K4 monomethylation   | Increase at 3' end of transcripts (Liu et al., 2005)                                                                    | Not examined.                     |
| H3K9/K14 acetylation   | Correlated with active genes at 5' end of transcripts (Liu et al., 2005; Pokholok et al., 2005; Wiren et al., 2005)          | Decrease by vernalization (Sung and Amasino, 2004) |
| H3S10 phosphorylation  | Correlated with active genes (Nowak and Corces, 2004)                                                                   | Not examined.                     |
| H3K36 dimethylation    | Correlated with active genes at 3' end of transcripts (Bannister et al., 2005; Rao et al., 2005)                           | Not examined.                     |
| H3K36 trimethylation   | Correlated with active genes at 3' end of transcripts (Bannister et al., 2005; Pokholok et al., 2005)                      | Not examined.                     |
| H3K79 dimethylation    | Correlated with silencing in telomere (Ng et al., 2002)                                                                  | Not examined.                     |
| H3K79 trimethylation   | No correlation with gene expression (Pokholok et al., 2005)                                                              | Not examined.                     |
| H4 hyperacetylation    | General correlation with active genes (He et al., 2003)                                                                  | Not examined.                     |
| H3K9 dimethylation     | Found in euchromatin (Martin and Zhang, 2005)                                                                          | Increase by vernalization (Sung and Amasino, 2004; Bastow et al., 2004) |
| H3K9 trimethylation    | Correlated with heterochromatin (Martin and Zhang, 2005)                                                                | Not examined.                     |
| H3K20 dimethylation    | Found in euchromatin (Martin and Zhang, 2005)                                                                          | Not examined.                     |
| H3K20 trimethylation   | Correlated with heterochromatin (Martin and Zhang, 2005)                                                                | Not examined.                     |
| H3K27 dimethylation    | Correlated with PcG-mediated repressed genes (Martin and Zhang, 2005)                                                   | Increase by vernalization (Sung and Amasino, 2004; Bastow et al., 2004) |
| H3K27 trimethylation   | Correlated with PcG-mediated repressed genes (Martin and Zhang, 2005)                                                   | Not examined.                     |
| H3S28 phosphorylation  | Chromosome condensation (Goto et al., 1999)                                                                            | Not examined.                     |
| H4R3 methylation       | Correlated with repressed genes (Huang et al., 2005)                                                                    | Not examined.                     |
| H2 variant replacement | Gene activation (Korber and Horz, 2004)                                                                                | Not examined.                     |
| H2AK119 ubiquitination | Correlated with PcG-mediated repressed genes (Wang et al., 2004)                                                         | Not examined.                     |

In animals, constitutive heterochromatin, such as in pericentric regions, is often enriched in methylated H3K9 (Maison and Almouzni, 2004), whereas methylated H3K27 is associated with PcG-mediated repressed chromatin which tend to be in euchromatic regions (Cao and Zhang, 2004).

The involvement of a homologue of a PRC2 component (Su(z)12/VRN2) and the PHD finger-containing VIN3 in vernalization-mediated FLC repression, as well as the mitotic stability of the vernalized state, prompted an examination of histone modifications at FLC chromatin during vernalization (Bastow et al., 2004; Sung and Amasino, 2004). The level of both H3K9 and H3K27 methylation at FLC chromatin increases during vernalization. Methylated H3K9 appears to be an important histone mark in the vernalization response because methylation of H3K27 alone is not sufficient to maintain stable FLC repression: in vrn1 mutants, which lack H3K9 methylation of FLC chromatin, FLC repression is not stably maintained, although FLC H3K27 methylation still increases during vernalization (Bastow et al., 2004; Sung and Amasino, 2004). Thus, compared with PcG repression in animals, which relies on H3K27 methylation, plants also appear to use H3K9 methylation to establish the vernalization-mediated formation of repressed chromatin.

In animals, genes repressed by PcG can also be activated by TrxG; in fact, competition between activating and repressing chromatin-modifying activities is thought to

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Trithorax) domain protein that is the observable single mutant phenotype (Chanvivattana 1997). Another E(z) homologue, SWN, does not exhibit any chromatin by vernalization have also not been identified among the three E(z) homologues with respect to vernalization genetically. Biochemical purification of VRN2-containing complexes would also serve as a TRE in Drosophila, in which PREs have been characterized, efforts to define the minimal PRE resulted in the identification of a 219 bp core PRE which contains binding sites for Zeste, Pleiohomeotic (PHO), and GAF (Dejardin and Cavalli, 2004; Dejardin et al., 2005). Binding sites for PHO and GAF in the PREs are necessary to recruit PRC2 (Dejardin et al., 2005). As noted above, the PRE also serves as a TRE in Drosophila and thus deletion of this element affects the maintenance of gene activation as well as repression.

The similarity between PRC2-mediated gene repression in Drosophila and vernalization suggested that FLC repression may involve a cis regulatory region similar to

**The winter encoders**

It has been difficult to pinpoint the specific genes encoding enzymes that catalyse vernalization-mediated histone modifications, perhaps due to redundancy. In animals, Enhancer of Zeste (E(z)) encodes the SET (Su(var)3–9, Enhancer-of-zeste, Trithorax) domain protein that is the actual H3K27 methyltransferase of the PRC2 complex (Cao and Zhang, 2004). In Arabidopsis, there are three homologues of E(z), MEDEA (MEA), CURLY LEAF (CLF), and SWINGER (SWN), and it is highly likely that one or more of these are in a complex with VRN2 that methylates target loci at H3K27. MEA is required for the repression of the MADS box gene PHERES (Kohler et al., 2003, 2005), whereas CLF is necessary for the repression of the MADS box gene AGAMOUS (Goodrich et al., 1997). Another E(z) homologue, SWN, does not exhibit any observable single mutant phenotype (Chanvivattana et al., 2004). Seed abortion of mea mutants makes it difficult to evaluate whether or not MEA has a role in vernalization, and both single mutants of clf and swn exhibit a normal vernalization response (MR Doyle and RM Amasino, unpublished data). It is suspected that there is redundancy among the three E(z) homologues with respect to vernalization similar to that observed for other developmental processes (Chanvivattana et al., 2004). Inducible expression systems to avoid lethality in double and/or triple mutants could be used to address the role of the E(z) homologues in vernalization genetically. Biochemical purification of VRN2-containing complexes would also reveal the identity of the components in this chromatin-modifying complex.

The enzymes responsible for H3K9 methylation of FLC chromatin by vernalization have also not been identified. First, it should be noted that a role for the plant E(z) homologues in H3K9 methylation has not been ruled out. Studies of histone modifications at target loci in E(z) mutants only indicates a role for E(z) in H3K27 methylation, although animal E(z) can methylate H3K9 in vitro (Kuzmichev et al., 2002). However, it is considered more likely that methylation of H3K9 is catalyzed by the same class of proteins that are responsible for this modification in yeast and animals, the Suppressor of Variegation 3–9 (Su(var)3–9) class of SET domain proteins (Rea et al., 2000). So far, none of the single mutants in these Su(var)3–9 homologues are affected in the vernalization response (Mylne et al., 2006), suggesting the possibility of redundancy among them. In addition, it has been shown that the maintenance of H3K9 methylation is distinct from the initial H3K9 methylation that occurs during cold exposure (Sung et al., 2006), raising the possibility that there are different chromatin-modifying complexes, perhaps with different H3K9 methyltransferases, involved in the initiation versus maintenance phase of FLC repression.

**DNA methylation does not appear to be involved in vernalization-mediated FLC repression**

DNA methylation is often associated with gene silencing, and there has been speculation about the involvement of DNA methylation in the vernalization response (Burn et al., 1993; Finnegan et al., 1998). Although recent studies indicate that some animal PRC2 complexes are associated with DNA methyltransferases (Vire et al., 2006), most examples of PRC2-mediated repression do not involve DNA methylation (Umlauf et al., 2004). Notably, there are no changes in DNA methylation of FLC brought about by vernalization (Finnegan et al., 2005).

**cis-acting components: regulatory DNA sequences in the vernalization response**

The target genes regulated by PcG and TrxG have cis regulatory elements to which PcG and TrxG proteins bind. Such cis elements are referred to as Polycomb response elements (PRE) or Trithorax response elements (TRE), and these bi-functional regions are often called ‘cellular memory modules’ (Ringrose and Paro, 2004).

In Drosophila, in which PREs have been characterized, efforts to define the minimal PRE resulted in the identification of a 219 bp core PRE which contains binding sites for Zeste, Pleiohomeotic (PHO), and GAF (Dejardin and Cavalli, 2004; Dejardin et al., 2005). Binding sites for PHO and GAF in the PREs are necessary to recruit PRC2 (Dejardin et al., 2005). As noted above, the PRE also serves as a TRE in Drosophila and thus deletion of this element affects the maintenance of gene activation as well as repression.
a PRE. An initial attempt to define such a vernalization element using a reporter transgene identified about 2 kb of the first intron of FLC as essential for maintenance of vernalization-mediated FLC repression (Sheldon et al., 2002). Recently, the region to 289 bp that was termed a vernalization response element (VRE) has been defined further, and it has been demonstrated that the VRE is required for vernalization-mediated H3K9 methylation (Sung et al., 2006).

In contrast to the bi-functional nature of PREs in Drosophila, deletion of the VRE, as well as deletions in the regions surrounding the VRE, did not compromise FLC activation (Sung et al., 2006). Thus the VRE may act differently from the Drosophila PRE in that the VRE only confers stable repression but is not necessary for activation. One possible regulatory region that could serve as an activating cis element like a TRE is in the 5′ untranslated region of FLC, a region that is enriched in H3K4 methylation when FLC is active (He et al., 2004).

Like Drosophila PREs, the VRE is likely to be necessary to recruit one or more chromatin remodelling complexes to FLC. It has been shown that the binding of DSP1, the Drosophila high mobility group gene B2 (HMGB2) homologue, to the PRE is a prerequisite for recruitment of PcG proteins including PHO (Dejardin et al., 2005). To date, VRN1 is the only DNA binding protein known to be involved in vernalization (Levy et al., 2002). Like HMGB1 and HMGB2, VRN1 exhibits low sequence-specific DNA binding activity in vitro (Levy et al., 2002). It is possible that VRN1 might be involved in the recognition of the VRE similar to the function of DSP1 in Drosophila. H3K9 methylation is not increased by vernalization in vrn1 mutants (Bastow et al., 2004; Sung and Amasino, 2004) and VRN1 protein is enriched at FLC chromatin by vernalization (Sung and Amasino, 2004). However, a vernalization-mediated increase in H3K27 still occurs in vrn1 mutants, suggesting that at least H3K27 methylation is independent of VRN1 recruitment to FLC chromatin.

**Deciphering the winter code**

A simplistic view of how the histone code can influence an epigenetic switch of gene expression is that once histones are marked with certain modifications, such modifications are recognized by modification-specific binding proteins, which, in turn, recruit repressive or activating complexes that maintain modifications through subsequent rounds of cell divisions. These feedback systems create stable states of higher order chromatin structure which maintain activation or repression of target genes. In Drosophila and mammals, Polycomb Repression Complex 1 (PRC1) contains Polycomb (Pc) protein which has a chromodomain. The Pc chromodomain binds preferentially to methylated H3K27, a histone modification characteristic of PcG-repressed chromatin (Fischle et al., 2003). The association of PRC1 mediates remodelling of the target chromatin into condensed chromatin and its maintenance in a condensed state (Levine et al., 2004).

Despite the conservation of PRC2 components in plants and animals, no PRC1 components, including Pc, have been found in plants (Hsieh et al., 2003; Schubert et al., 2005). Thus, plants must use a protein other than Pc to maintain vernalization-mediated histone modifications and stable repression. HETEROCHROMATIN PROTEIN 1 (HP1) is another chromodomain protein that binds methylated H3K9 (Fischle et al., 2003). This binding preference is consistent with the role of HP1 in the maintenance of constitutive heterochromatin in yeast and animals, because constitutive heterochromatin is enriched in methylated H3K9 (Maison and Almouzni, 2004). However, unlike the case for HP1 in yeast and animals, LIKE HETEROCHROMATIN PROTEIN 1 (LHP1), the only plant homologue of HP1, does not appear to have a role in the maintenance of constitutive heterochromatin (Malagnac et al., 2002; Libault et al., 2005; Nakahigashi et al., 2005). Rather, LHP1 is involved in the regulation of euchromatic genes, such as AGAMOUS and FLOWERING LOCUS T, and consistent with this role, LHP1 mainly localizes in euchromatic regions (Nakahigashi et al., 2005; Sung et al., 2006).

Recently, it has been shown that LHP1 is involved in the maintenance of stable FLC repression by vernalization (Mylne et al., 2006; Sung et al., 2006). Furthermore, vernalization results in an enrichment of LHP1 at FLC chromatin (Sung et al., 2006), suggesting that LHP1 may play a role in vernalization similar to the role of Pc in polycomb repression. Interestingly, LHP1 is required for maintaining increased H3K9 methylation at FLC, but not for the initiation of H3K9 methylation during cold exposure (Sung et al., 2006). As noted earlier, this suggests that there might be two different histone methylating components involved in the vernalization response.

Prior to vernalization, FLC chromatin is enriched in trimethylated Histone H3 Lys 4 (H3K4) (He et al., 2004) and vernalization results in a reduction of the level of trimethylated H3K4 (Sung et al., 2006). The mechanism by which trimethylated H3K4 levels are reduced is not known. The reduction could be mediated by histone demethylases; several histone demethylases have recently been discovered (Wysocka et al., 2005; Tsukada et al., 2006; Yamane et al., 2006), although trimethylated H3K4-specific demethylases are yet to be identified.

It is also interesting to note the recent discovery that the PHD finger can bind to trimethylated H3K4 (Li et al., 2006; Pena et al., 2006; Shi et al., 2006; Wysocka et al., 2006). This raises the possibility that recognition of trimethylated H3K4 by the PHD finger of VIN3 could contribute to the recruitment or activity of a silencing chromatin remodelling complex to FLC chromatin. Of course, VIN3 binding to the trimethylated H3K4 would not be the sole
mechanism to recruit repression complexes, since many other chromatin regions are enriched in trimethylated H3K4.

In addition to VIN3, there are a number of PHD finger-encoding genes found in plants. Although most of them have yet to be studied with respect to a possible involvement in chromatin remodelling, a VIN3 relative was identified, which was named VIL1, in a yeast two-hybrid screen for proteins that interact with VIN3 (S Sung and RM Amasino, unpublished results). VIL1 also contains a PHD finger in its N-terminal domain and its C-terminal domain is involved in the protein interaction between VIN3 and VIL1. Although VIL1 expression is not vernalization-specific, VIL1 is required for a proper vernalization response. Thus vernalization appears to require a chromatin remodelling complex that contains at least two PHD finger proteins, VIN3 and VIL1.

**Forgetting winter**

The vernalized state is stable throughout mitotic cell divisions in many species, but it is lost as repressed genes like FLC become reactivated in the next generation. It is tempting to speculate that this re-activation occurs during meiosis, but there is no direct evidence for this. Regarding a possible meiotic resetting mechanism, one interesting observation is that the VRN1 protein, which localizes to euchromatic regions in vegetative tissues, is below detection limits in pollen (Mylne et al., 2006). Also, VRN1 mRNA, as well as LHP1 mRNA, is reduced in pollen tissues, and it was suggested that the lower expression of such genes, which are essential for maintenance of the vernalized state, might contribute to the reactivation of FLC expression during meiosis (Mylne et al., 2006). However, it is important to consider that a large number of genes are poorly expressed in pollen (Schmid et al., 2005); indeed, the majority of vegetatively expressed genes are down-regulated in pollen (Schmid et al., 2005; Honys and Twell, 2003) including genes, such as **KRYPTONITE** and **CHROMODOMAIN DNA METHYLTRANSFERASE 3**, that are involved in the stable repression of target genes through meiosis (Jackson et al., 2002; Lindroth et al., 2004; Schmid et al., 2005). Moreover, any resetting model that involves the lack of key components as a mechanism to alleviate repression must also apply to the female gametophyte, but this has not been examined with respect to VRN1 and vernalization.

An alternative to a resetting model in which components are lacking, is that resetting involves the recruitment of components that can reactivate FLC chromatin. The dynamic nature of chromatin states gives an opportunity for resetting components to get access to repressed chromatin. For example, although HP1 and Pc are associated with a repressed chromatin structure, the binding of theses proteins to methylated histones is relatively weak (micromolar Kd; Fischle et al., 2003), and fluorescence bleaching assays indicate a relatively high rate of exchange of such repressive proteins on chromatin (Maison and Almouzni, 2004). Indeed, during cell division, heterochromatic regions need to be relaxed to permit DNA replication (Maison and Almouzni, 2004). During mitosis, cell cycle-specific kinases gain access to heterochromatin and phosphorylate, for example, Ser 10 of Histone H3 (H3S10) which in turn attenuates HP1 binding to methylated H3K9 (Fischle et al., 2005). After replication, removal of the phosphoryl group from H3S10 facilitates HP1 binding and the re-establishment of heterochromatin (Fischle et al., 2005). A similar mechanism may operate in the resetting of FLC repression. For example, during meiosis, increased phosphorylation of H3S10 could contribute to lowering the level of LHP1 binding to FLC chromatin which would, in turn, lower the levels of H3K9 methylation (Fig. 2) because the binding of LHP1 at FLC chromatin is required for the maintenance of methylated H3K9 (Sung et al., 2006). Recently, a JmjC-containing histone demethylase has been shown to demethylate H3K9 and this demethylation activity can attenuate HP1 binding to chromatin (Cloos et al., 2006; Klose et al., 2006; Tsukada et al., 2006; Yamane et al., 2006). Thus, it is possible that the H3K9-specific histone demethylase could have a role in the reversing H3K9 methyl marks at FLC (Fig. 2). An

![Hypothetical mechanism for resetting FLC chromatin during meiosis](https://academic.oup.com/jxb/article-abstract/57/13/3369/476321)

**Fig. 2.** Hypothetical mechanism for resetting FLC chromatin during meiosis. Vernalization causes LHP1 to associated with FLC chromatin and to ‘lock-in’ stable FLC repression as discussed in the text (top). Increasing phosphorylation at H3S10 could prevent LHP1 from binding (middle). Activating components gain access to FLC chromatin to reactivate FLC expression in the next generation (lower).
Arabidopsis JmjC homologue is involved in the regulation of FLC via chromatin remodelling (Noh et al., 2004), although its role in the ‘forgetting winter’ has not been determined. Much remains to be learned about the histone code in plants and, in particular, which parts of this code are used in the process of vernalization.

Acknowledgements

This paper is dedicated to Georges Bernier for his many basic contributions to our understanding of the flowering process, and to his tireless efforts to further interactions and understanding in the flowering field including his founding and stewardship of the Flowering Newsletter. We are grateful to Rachel Rodman and Robert Schmitz for their comments on the manuscript. Research in RMA’s lab was supported by the College of Agricultural and Life Sciences of the University of Wisconsin and by grants from the US Department of Agriculture National Research Initiative Competitive Grants Program and the National Science Foundation.

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