Development 139, 2267-2282 (2012) doi:10.1242/dev.070110

The Prdm family: expanding roles in stem cells and development

Tobias Hohenauer and Adrian W. Moore*

Summary

Members of the Prdm family are characterized by an N-terminal PR domain that is related to the SET methyltransferase domain, and multiple zinc fingers that mediate sequence-specific DNA binding and protein-protein interactions. Prdm factors either act as direct histone methyltransferases or recruit a suite of histone-modifying enzymes to target promoters. In this way, they function in many developmental contexts to drive and maintain cell state transitions and to modify the activity of developmental signalling pathways. Here, we provide an overview of the structure and function of Prdm family members and discuss the roles played by these proteins in stem cells and throughout development.

KEY WORDS: Cell fate, Chromatin, Differentiation, Prdm, SET, Transcription

Introduction

The Prdm family is defined on the basis of a conserved N-terminal domain. This domain was originally identified as shared between two proteins: PRDI-BF1 (positive regulatory domain I-binding factor 1), which is now commonly called Prdm1 (PR domain-containing 1); and RIZ1 (retinoblastoma protein-interacting zinc finger gene 1), now called Prdm2 (Buyse et al., 1995) and named the PR (PRDI-BF1-RIZ1 homologous) domain. PR domains are related to the catalytic SET domains (named after the *Drosophila* factors Suppressor of variegation 3-9, Enhancer of zeste and Trithorax) that define a large group of histone methyltransferases (HMTs, see Glossary, Box 1; see also Fig. 1) (Huang et al., 1998; Schneider et al., 2002; Sun et al., 2008; Wu et al., 2010).

Prmd1 was cloned as a human cDNA encoding a transcriptional repressor that bound the positive regulatory domain of the interferon β promoter (hence the original name PRDI-BF1) (Keller and Maniatis, 1991). Later, Blimp1 (B-lymphocyte-induced maturation protein 1) was independently identified in mouse as a primary regulator of plasma cell differentiation (Turner et al., 1994). PRDI-BF1 and Blimp1 are homologues (Huang, 1994) and to share a PR domain with Riz1, a retinoblastoma-binding protein with tumour suppressor activity in a broad array of tissues (Buyse et al., 1995; Huang et al., 1998; Kim et al., 2003; Steele-Perkins et al., 2001). The family grew further when the leukaemogenic oncogene Evi1 (ecotropic viral integration 1) was revealed to undergo an alternative splicing event, fusing it with what had previously been considered an independent upstream locus (myelodysplasia syndrome-associated protein 1; Mds1) (Fears et al., 1996). This fusion creates a PR domain-containing transcript called Mds1-Evi1, Mecom (Mds1 and Evi1 complex locus) or Prdm3. The same study noted that *Caenorhabditis elegans* factor EGL-43 (Garriga et al., 1993) also has a PR domain, demonstrating conservation of this family between invertebrates and vertebrates (Fig. 1B) (Fears et al., 1996).

As additional Prdm family members were identified, they were often implicated in neoplasia, including cervical and breast cancer, colorectal and gastric tumours, and leukaemia (see Box 2) (Fog et al., 2012; Morishita, 2007; Schneider et al., 2002). Additionally, a rapidly expanding set of studies demonstrated that Prdms act in stem cells and during cell differentiation and maturation in a wide range of developmental processes. In this review, we discuss recent studies that illustrate common themes and important differences in Prdm action during development, and we examine how these functions relate to the molecular mechanisms of Prdm action.

Prdm family molecular structure

Prdm family members control gene expression through modification of the chromatin state at target gene promoters. Members are characterized by the presence of an N-terminal PR domain found at various levels of conservation across species (Fig. 1A,B). In all but one family member (Prdm11 being the exception), the N-terminal PR domain is followed by repeated arrays of zinc fingers (Fumasoni et al., 2007; Kinameri et al., 2008; Sun et al., 2008). Included in the family of HMT SET domains, the PR domains are the most diverged from the canonical SET structure (Fig. 1C) (Fears et al., 1996; Sun et al., 2008; Wu et al., 2010). Furthermore, although most SET family members are enzymatically active, HMT activity has been found only in the PR domains of Prdm2, Prdm8 and Prdm9 (also called Meisetz; meiosis-induced factor containing PR/SET domain and zinc-finger motif) (Derunes et al., 2005; Eom et al., 2009; Hayashi et al., 2005). Activity has also been reported for Prdm6 (also called Prism – Prdm in smooth muscle); however, Prdm6 constructs that lack a PR domain still retain histone H4 methyltransferase activity and the nature of this activity requires further clarification (Wu et al., 2008).

Why some Prdm proteins exhibit intrinsic HMT activity and others do not is still unclear. In the larger SET family, mutations in the conserved H/RxxNHxC motif abolish catalytic activity (Rea et al., 2000). However, all PR domains lack this motif, and most significantly the essential histidine in position 2 of NHxC (Fig. 1C, orange box). In Prdm2, C106T mutations reduce methyltransferase activity (Fig. 1C, purple box). The cysteine in this position of Prdm2 corresponds to that in the NHxC motif in other SET proteins (Fig. 1C, orange and purple boxes). Furthermore, this cysteine is required for activity in other SET domains; mutations of this residue in the SET domain of SUV39H1, a human orthologue of suppressor of variegation 3-9, abolish HMT activity of the SUV39H1 protein (Derunes et al., 2005; Rea et al., 2000). Nevertheless, this important cysteine residue is not conserved among the PR domains of Prdm8 and Prdm9, which also exhibit HMT activity, nor in any other Prdm family members.

Disease Mechanism Research Core, RIKEN Brain Science Institute, Wako, Saitama, 351-0198, Japan.

*Author for correspondence (adrianm@brain.riken.jp)

Box 1. Glossary

Adipocytes (or lipocytes). Fat cells that make up adipose tissue and store chemical energy in the form of lipid droplets

Bipolar cells. A type of specialized sensory neuron that has two extensions. In the retina, these cells transmit signals from photoreceptors to ganglion cells.

Embryonic germ cells (EGCs). Cells that can be derived from primordial germ cells in vitro through yet uncharacterized mechanisms.

Embryonic stem cells (ESCs). Pluripotent stem cells derived from the inner cell mass of the blastocyst.

Endomesoderm. The common progenitor of endoderm and mesoderm that is found early in the development of many animals.

Enterocytes. Also called intestinal absorptive cells, these are epithelial cells of the small intestines and colon that carry out water and nutrient uptake.

Histone acetyltransferases (HATs). Enzymes that add acetyl groups to specific lysine residues on a histone protein.

Histone deacetylases (HDACs). Enzymes that remove acetyl groups from specific lysine residues on a histone protein.

Histone methyltransferases (HMTs). Enzymes that transfer one or more (up to three) methyl groups from co-factors onto lysine or arginine residues of histone proteins.

Induced pluripotent stem cells (iPSCs). Cells that are derived from a non-pluripotent cell via ectopic expression of four transcription factors (Oct3/4, Sox2, Myc and Klf4).

Interferon regulatory factors (IFRs). Transcriptional regulators involved in pathogen response, cytokine signalling, cell growth regulation and haematopoietic development.

Myocytes. Muscle cells that include specialized cardiac, skeletal and smooth muscle cells.

Notum mechanosensory organs. Organs found on the dorsal surface of the fly thorax consist of a bristle, which projects into the air-stream, and associated cells including an external sensory neuron that transfers sensory information to the central nervous system.

Olfactory receptor neurons (or olfactory sensory neurons). Neurons primarily found in the nose (vertebrates) or the antenna (flies) that directly detect odours from the environment and relay this information to the CNS.

Osteoclasts. Cells responsible for maintaining bone homeostasis.

Photoreceptor cells. Neurons in the retina that are specialized to absorb photons and transmit a resulting signal on to the central nervous system, indicating the presence of light in the visual field.

Primordial germ cells. Proliferative germ cell precursor that are yet to migrate to the gonads.

Box 2. Prdms in cancer and other diseases

Some Prdms act as tumour suppressors. Inactivation of Prdm1 can lead to diffuse large B cell lymphoma (DLBCL) (Calado et al., 2010; Mandelbaum et al., 2010) and natural killer cell lymphoma (Karube et al., 2011; Kucuk et al., 2011). Inactivation of Prdm2 is associated with a wide range of cancer types, including DLBCL and several solid, sometimes rare, tumours (Kim et al., 2003; Steele-Perkins et al., 2001). Prdm5 is also silenced in multiple tumour types (Shu et al., 2011), and Prdm12 is a putative tumour suppressor for chronic myeloid leukaemia (Reid and Nacheva, 2004).

Other Prdms are oncogenic. Expression of Prdm3 or Prdm16 isoforms that lack the PRDI-BF1-RIZ1 homologous domain during haematopoiesis can lead to acute myeloid leukaemia (Morishita, 2007). Prdm14 is overexpressed in breast cancer (Nishikawa et al., 2007) and causes lymphoid leukaemia (Dettman et al., 2011). Prdm3 is a susceptibility locus in nasopharyngeal carcinoma (Bei et al., 2010) and a variety of other solid tumours (Koos et al., 2011; Sugita et al., 2000). In addition, Prdm13 is a tumour antigen associated with medulloblastoma (Behrends et al., 2003).

Besides roles in tumourigenesis, Prdm1 is a susceptibility locus for the autoimmune diseases systemic lupus erythematosus (Gateva et al., 2009) and rheumatoid arthritis (Raychaudhuri et al., 2009). Furthermore, Prdm3 is a susceptibility locus for lung dysfunction (Soler Artigas et al., 2011) and is mutated in a mouse model of otitis media (Parkinson et al., 2006). Prdm5 is mutated in brittle cornea syndrome (Burkitt Wright et al., 2011) and Prdm16 is associated with migraine (Chasman et al., 2011).

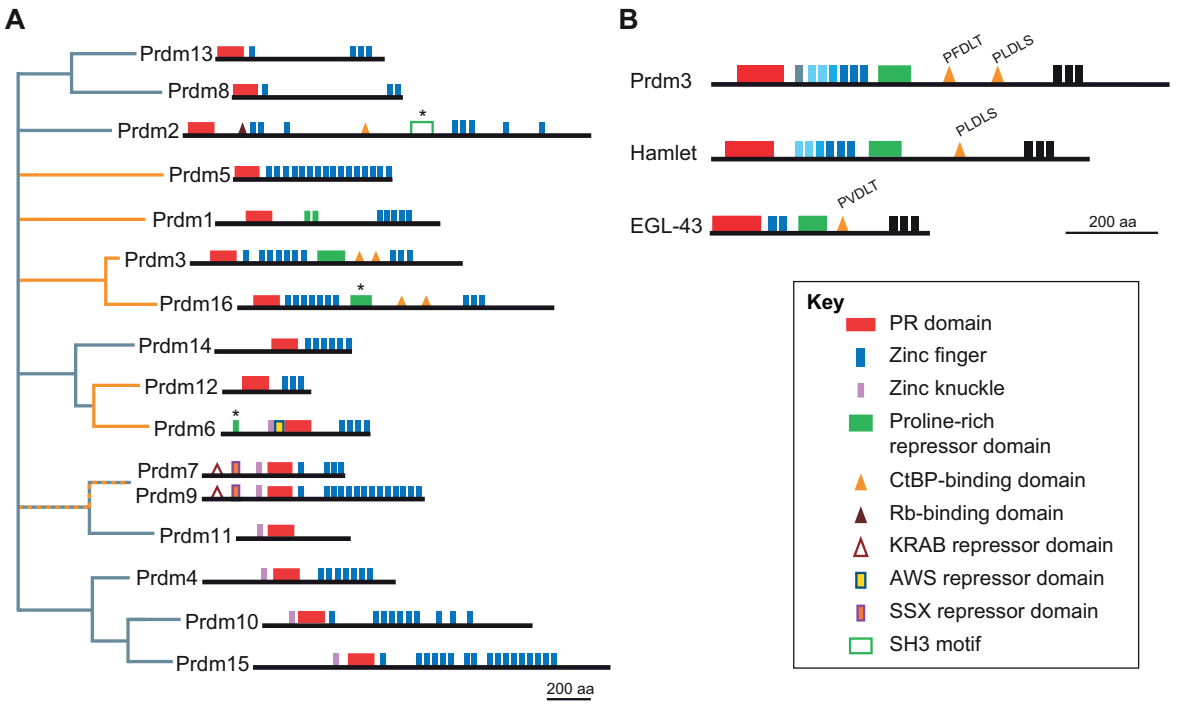
Investigation of the developmental roles of Prdm factors may help illuminate their pathological mechanisms. Nevertheless, the cell types affected in these diseases are often ill defined. As Prdm activity is cell type specific the correlations between developmental and pathological processes must be made with great care.

As most Prdm family members lack intrinsic enzymatic activity, they recruit histone-modifying enzymes to mediate their function. These enzymes include HMTs, which induce repressive chromatin states by methylation of histone lysines, in particular H3K9 (histone H3 lysine 9) by G9a and SuV39H, and H3K27 (histone H3 lysine 27) by the Polycomb repressive complex 2 (PRC2). They also include the enzyme Prmt5 (protein methyltransferase 5), which methylates arginines to create a repressive state, and Lsd1 (lysine-specific demethylase 1, Kdm1a), which primarily promotes repression through erasure of activating H3K4 (histone H3 lysine 4) methylation marks. In addition, Prdm factors can recruit histone deacetylases (HDACs, see Glossary, Box 1), which promote transcriptional repression, and the histone acetyltransferases (HATs; see Glossary, Box 1) p300/CBP (cAMP-responsive element-binding protein-binding protein) and P/CAF (p300/CBP-associated factor), which are associated with transcriptional activation. The activity of these factors is reviewed by Kouzarides (Kouzarides, 2007).

In some cases, histone-modifying enzyme recruitment occurs through the zinc fingers. In others, it is through additional domains, such as the proline-rich domains reported in Prdm1 (Huang et al., 1998) and Prdm3 (Bartholomew et al., 1997). Similarly, proline-rich sequence stretches can be found in Prdm6, in Prdm16 (also known as Mel1 – Mds1-like gene 1) and in the solitary invertebrate homologues of Prdm3 and Prdm16 – Hamlet in *Drosophila* and EGL-43 in *C. elegans* (Fig. 1). Prdm1 recruits the repressive chromatin-modifying factors Hdac2 (Yu et al., 2000) and Lsd1 (Su et al., 2009) through its central proline-rich domains. It also associates with G9a via both its zinc fingers (Gyory et al., 2004) and these proline-rich domains (Su et al., 2009). The PR domain of Prdm1 does not exert HMT activity, but it is also required for maximal repressive activity, suggesting that it could mediate as yet undefined protein-protein interactions (Gyory et al., 2003).

Prdm factors also recruit co-repressors that, in turn, associate with histone-modifying enzymes. Prdm1 recruits Groucho family members through its proline-rich domain (Ren et al., 1999), and Prdm2, Prdm3, Prdm16 and Hamlet all bind CtBP (C-terminal binding protein) through canonical PLDLS CtBP-binding sites (Fig. 1A,B) (Endo et al., 2012; Izutsu et al., 2001; Kajimura et al., 2008; Nishikata et al., 2011; Palmer et al., 2001; Quinlan et al., 2006; Van Campenhout et al., 2006). CtBP is used by Prdm16 to repress white fat cell differentiation (Kajimura et al., 2008), and by Hamlet to mediate repressive histone methylation events (H3K27) at target promoters during neuron diversification (Endo et al., 2012).

There are further motifs found in Prdm family members (Fig. 1A), the functions of which are not well characterized. These include an AWS (associated with SET) domain in Prdm6 that is



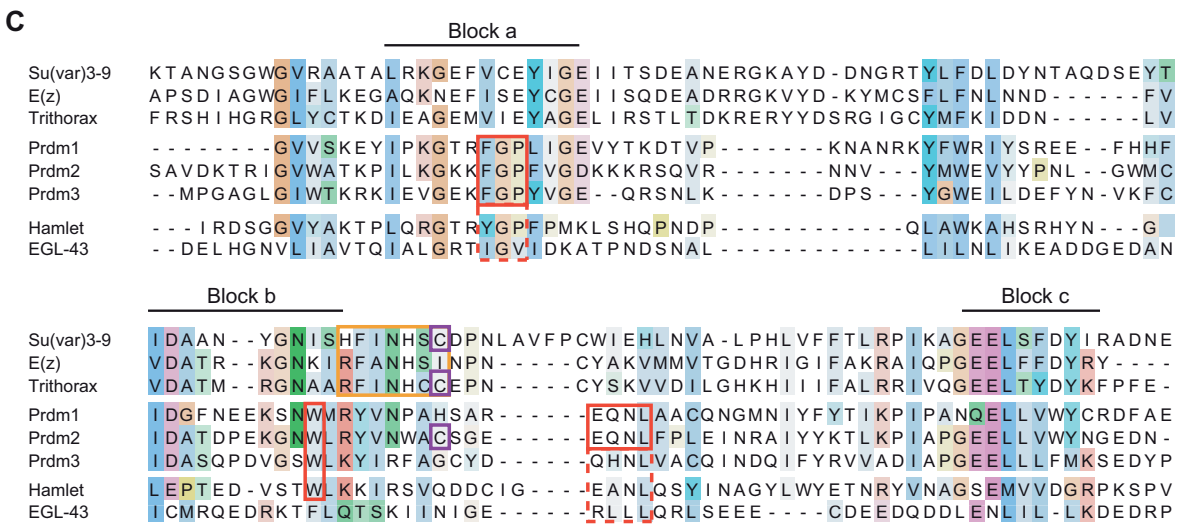


Fig. 1. Prdm family domain structure and relationships. (**A**) The domain structure for each human Prdm family member is illustrated, along with the relationships between their positive regulatory (PR) domains. Only the longest reported isoform is shown. Orange lines highlight the Prdms conserved in *C. elegans* and *Drosophila*; a putative Prdm9 without zinc fingers has been reported in *C. elegans*. Prdm11 alone does not contain zinc fingers; instead, it has a smaller protein-protein interaction motif known as a zinc knuckle that is also present in several other family members (Briknarova et al., 2008). Protein features marked with asterisks are derived from UniProt. (**B**) An example of cross-species conservation of Prdm structure. The orthologues of mouse Prdm3, *Drosophila* Hamlet and *C. elegans* EGL-43, are shown. Zinc fingers with high sequence similarity between the homologues are shown in the same colour, and the positions and sequences of shared CtBP-binding sites are also shown. AWS, associated with SET domain; CtBP, C-terminal binding protein; KRAB, Krüppel-associated box; Rb, retinoblastoma; SSX, synovial sarcoma X. (**C**) Sequence alignment of the SET domains of *Drosophila* Suppressor of variegation 3-9, Enhancer of zeste and Trithorax factors with the PR domains of the murine Prdm family founding factors Prdm1 and Prdm2, and with Prdm3 and its *Drosophila* and *C. elegans* homologues Hamlet and EGL-43 (made using JalView and ClustalW2 default parameters) (Waterhouse et al., 2009). Red boxes indicate motifs present in most PR domains but absent in most SET domains that can be used for discrimination. Blocks a to c indicate regions containing highly conserved amino acid sequences (Huang, 2002). The orange box denotes the H/RxxNHxC motif that is exclusive to SET domains and has been associated with intrinsic enzyme activity (Rea et al., 2000). Crucial cysteine residues within this motif are indicated with purple boxes. Dashed red boxes indicate conserved residues between the vertebrate Prdm factors and the Prdm3 homologues *Drosophila* Hamlet and *C. elegans* EGL-43.

necessary for PR-mediated repression (Davis et al., 2006), and SSX (synovial sarcoma X) and KRAB (Krüppel-associated box) repressor domains in Prdm7 and Prdm9 (Hochwagen and Marais, 2010). Notably, KRAB domain activity is involved in HAT and HMT recruitment in other proteins (Margolin et al., 1994; Pengue et al., 1994).

Table 1. Prdm enzymatic activity and binding partners

Factor	Intrinsic enzymatic activity	Binding to chromatin modifiers	Other co-repressors and binding partners
Prdm1		G9a (Gyory et al., 2004), Lsd1 (Su et al., 2009), Hdac2 (Yu et al., 2000), Prmt5 (Ancelin et al., 2006)	Groucho family (Tle1 and Tle2) (Ren et al., 1999) and Irf4 (Gupta et al., 2001)
Prdm2	H3K9 methlytransferase (Kim et al., 2003)	p300 (Carling et al., 2004)	Prdm2 homodimer (Huang et al., 1998), Rb1 (Buyse et al., 1995) and ER (Medici et al., 1999)
Prdm3		p300 (Chakraborty et al., 2001), P/CAF, HDACs (Alliston et al., 2005), SuV39H1 (Cattaneo and Nucifora, 2008), Dnmt3a/b (Senyuk et al., 2011), Mbd3 (Spensberger et al., 2008), Uxt (McGilvray et al., 2007) and PRC (Yoshimi et al., 2011)	CtBP (Izutsu et al., 2001; Palmer et al., 2001), Gata1 (Laricchia-Robbio et al., 2006), Jnk (Kurokawa et al., 2000; Spensberger et al., 2008), Runx1 (Senyuk et al., 2007), Smad1/2 (Alliston et al., 2005), Smad3 (Kurokawa et al., 1998), Pu.1 (Laricchia-Robbio et al., 2009), Pax2, Yy1 and Fos (Bard-Chapeau et al., 2012)
Prdm4		Hdac1-3 (Chittka et al., 2004)	p75 (Chittka et al., 2004)
Prdm5		G9a and Hdac1-3 (Duan et al., 2007)	Gfi1 (Duan et al., 2007)
Prdm6	H4K20 methyltransferase (Wu et al., 2008)	G9a, HDACs and p300 (Davis et al., 2006)	
Prdm8	H3K9 methyltransferase (Eom et al., 2009)		Bhlhb5 (Ross et al., 2012)
Prdm9	H3K4 methyltransferase (Hayashi et al., 2005)		
Prdm16		HDACs and p300 (Takahata et al., 2009)	Pparα/Pparγ (Seale et al., 2008), SKI (Takahata et al., 2009), Smad3 (Warner et al., 2007), CtBP (Kajimura et al., 2008), C/EBPβ (Kajimura et al., 2009) and Ppargc1α/Ppargc1β (Seale et al., 2007)

Bhlhb5, basic helix-loop-helix domain-containing class B5; C/EBPβ, (CCAAT/enhancer-binding protein β); CtBP, C-terminal binding protein; Dnmt, DNA methyltransferase; ER, oestrogen receptor; Gfi1, growth factor independent 1; HDAC, histone deacetylases; Irf4, interferon regulatory factor 4; Jnk, c-Jun N-terminal kinase; Lsd1, lysine-specific demethylase 1; Mbd3, methyl-CpG binding domain protein 3; Pu.1, SFFV proviral integration 1; P/CAF, p300/CBP-associated factor; Ppar, peroxisome-proliferator-activated receptor; Ppargc1, Pparγ co-activator 1α; Prc, Polycomb repressive complex; Prmt5, protein methyltransferase 5; Rb1, retinoblastoma 1; Runx1, runt-related transcription factor; Smad, MAD homologue; Tle, transducin-like enhancer of split; Uxt, ubiquitously transcribed tetratricopeptide repeat X chromosome; Yy1, yin-yang 1.

Prdm proteins can also bind DNA directly. Sequence-specific DNA binding via the zinc-finger domains has been demonstrated for Prdm1, Prdm3, Prdm5, Prdm9, Prdm14 and Prdm16 (Bard-Chapeau et al., 2012; Baudat et al., 2010; Chia et al., 2010; Delwel et al., 1993; Duan et al., 2007; Funabiki et al., 1994; Kuo and Calame, 2004; Ma et al., 2011; Seale et al., 2007). Zinc fingers one and two of the five zinc fingers of Prdm1 preferentially bind DNA sequences containing a GAAAG motif. This sequence can also be recognized by interferon regulatory factors (IFRs, see Glossary, Box 1), and Prdm1 and IRFs can antagonistically regulate promoter activity (Kuo and Calame, 2004). More recently, DNA binding of Prdm1 was shown to be abrogated by CpG methylation, adding complexity to transcriptional regulation by this protein, and suggesting a model in which Prdm1 could be actively excluded from a subset of its target sites, once silencing at the level of DNA methylation has been established (Doody et al., 2011).

Chromatin immunoprecipitation (ChIP) experiments followed by deep sequencing (ChIP-seq) data have been generated for Prdm1, and also for Prdm3 and Prdm14. These studies reveal binding at many sites across the genome, and a handful of targets has been validated (Bard-Chapeau et al., 2012; Chia et al., 2010; Doody et al., 2011; Ma et al., 2011; von Hofsten et al., 2008). It has also become clear that the same Prdm factor regulates divergent sets of targets in different cell types. Therefore, Prdm target selection is under context-dependent control. Recent ChIP-seq studies of Prdm3 found it to bind at many sites across the genome, and gave more information about how such Prdm context dependency might occur. Prdm3 has two separate domains of multiple zinc fingers with different DNA-binding specificities: the N-terminal zinc fingers predominantly bind a GATA-like motif (Delwel et al., 1993); and the C-terminal zinc fingers bind to an ETS-like sequence (Funabiki et al., 1994). Prdm3-bound ETS-like sites lie close to transcriptional start sites (TSS), whereas the Prdm3-bound GATA-like motifs tend to be localized further from TSS. Interestingly, Prdm3 binds to either of these motifs, but not to both motifs at the same promoter at the same time (Bard-Chapeau et al., 2012). Hence, binding site selection (and at least in the case of Prdm3 competition between different binding sites) represents one mechanism for context-dependent control of Prdm factor activity.

Prdm factors can complex with a wide variety of partners (summarized in Table 1), and partner selection might contribute to the context dependency of Prdm action. For example, the above-mentioned Prdm3 ChIP-seq studies, moreover, have revealed a strong enrichment for Ap1- (activator protein 1), Pax2- (paired box gene 2) or Yy1- (yin-yang 1) binding sites in close proximity to Prdm3 binding. As each of these factors is able to bind Prdm3 directly, binding partner selection may also control context-dependent activity. Notably, the Ap1 transcription factor commonly partners the proto-oncogene Fos, and knockdown of Prdm3 weakens Fos occupancy at Ap1 sites found in close proximity to the Prdm3-binding site, suggesting a regulatory interaction between these two factors (Bard-Chapeau et al., 2012).

In summary, Prdm factors act as direct HMTs or recruit a suite of additional histone-modifying enzymes to specific target promoters. They act simultaneously at multiple sites across the genome, and they exhibit context-dependent activity. These features create the capacity to drive and maintain cell state transitions, and to modify the transcriptional output of pleiotropic developmental signalling pathways.

Prdms direct a variety of cell differentiation events

A rapidly expanding body of work has now established Prdm factors as central players in development (summarized in Tables 2 and 3). Here, we discuss some of these recent studies, which

Table 2. An outline of reported Prdm factor embryonic expression

Factor	Embryonic gene expression patterns	References*
Prdm1	***Drosophila***: Gap gene pattern; sensory organ precursors; tracheal primordial; and trachea.	(Ng et al., 2006)
	Starfish and Sea urchin: Endomesoderm.	(Hinman et al., 2003; Livi and Davidson, 2006)
	Lamprey: Premigratory neural crest; branchial arches; eye; and somites.	(Nikitina et al., 2011)
	Xenopus: Anterior endomesoderm of the Spemann's organizer; Rohon-Beard sensory neurons; branchial arches; pharynx; eye; somites; and limb buds.	(de Souza et al., 1999; Rossi et al., 2008)
	Zebrafish: Prechordal plate; Rohon-Beard sensory neurons; branchial arches; pharynx; and limb buds (*Prdm1a*). Somites and retina (*Prdm1a* and *1b*).	(Sun et al., 2008; Roy and Ng, 2004)
	Mouse: Anterior endoderm; prechordal plate; head mesoderm; branchial arches; pharynx; limb buds; nascent photoreceptors; primordial germ cells; endocytes; epidermis including the sebaceous gland; and placenta.	(Chang et al., 2002; Harper et al., 2011; Horsley et al., 2006; Magnusdottir et al., 2007; Muncan et al., 2011; Robertson et al., 2007; Vincent et al., 2005)
Prdm3	***C. elegans* (*egl-43*)**: HSN and phasmid neurons; and cells of the developing uterus.	(Garriga et al., 1993; Hwang et al., 2007; Rimann and Hajnal, 2007)
	***Drosophila* (*hamlet*)**: External sensory organ and olfactory intermediate precursor cells and nascent neurons.	(Endo et al., 2012; Moore et al., 2002)
	Xenopus: Telencephalon; midbrain; hindbrain (initially rhombomere 4); branchial arches; limb buds; and pronephros.	(Van Campenhout et al., 2006)
	Chick: Branchial arches; limb buds; and pronephros.	(Van Campenhout et al., 2006)
	Zebrafish: Telencephalon; tegmentum; diencephalon; hindbrain; branchial arches; limb buds; and pronephros.	(Sun et al., 2008)
	Mouse: Primary head folds; rhombomere 4; branchial arches; limb buds; and mesonephros.	(Hoyt et al., 1997)
Prdm4	**Zebrafish**: Somites and muscle.	(Sun et al., 2008)
Prdm6	**Mouse**: Smooth muscle precursors in the aorta, outflow tract, lung, and bladder; and post-mitotic neurons in the spinal cord.	(Davis et al., 2006; Kinameri et al., 2008; Wu et al., 2008)
Prdm8	**Zebrafish**: Hindbrain and spinal cord (*Prdm8a*). Tegmentum, cerebellum and retina (*Prdm8b*).	(Sun et al., 2008)
	Mouse: Intermediate zone and cortical plate of the dorsal telencephalon; ventral spinal cord progenitors (p0, p1, p2 and pMN); retinal rod bipolar cells and a subpopulation amacrine cells.	(Kinameri et al., 2008; Komai et al., 2009; Ross et al., 2012)
Prdm9	**Mouse**: Germ cells entering meiotic prophase.	(Hayashi et al., 2005)
Prdm10	**Mouse**: Mesoderm-derived tissues.	(Park and Kim, 2010)
Prdm12	**Zebrafish**: Tegmentum; cerebellum; hindbrain; and olfactory placode.	(Sun et al., 2008)
	Mouse: Telencephalic ventricular zone; ventral spinal cord progenitors (p1); dorsal root ganglia; and cranial placodes.	(Kinameri et al., 2008)
Prdm13	**Zebrafish**: Tegmentum; hindbrain; spinal cord; retina; and olfactory placode.	(Sun et al., 2008)
	Mouse: Dorsal spinal cord progenitors.	(Kinameri et al., 2008)
Prdm14	**Mouse**: Inner cell mass and primordial germ cells.	(Yamaji et al., 2008)
Prdm15	**Zebrafish**: Cranial ganglia and muscle pioneer cells.	(Sun et al., 2008)
Prdm16	***Xenopus***: Head mesenchyme, forebrain; hindbrain; hyloid crest; retinal pigmented epithelium; and pronephros.	(Van Campenhout et al., 2006)
	Zebrafish: Forebrain; hindbrain; limb buds; and pronephros.	(Sun et al., 2008)
	Mouse: Craniofacial development including Meckel's cartilage, bones of the inner ear and nasal cartilage; telencephalic ventricular zone; hindbrain; retinal pigmented epithelium; cranial ganglia; limb buds becoming restricted to presumptive cartilage; and metanephros.	(Bjork et al., 2010; Chuikov et al., 2010; Kinameri et al., 2008)

Tissue-specific expressions have not been described for Prdm2, Prdm5 and Prdm11.
*For a complete description of the complex and dynamic expression patterns.

illustrate that Prdms often function to promote or repress developmental transitions in cell state by directing a series of chromatin modifications at target loci. We also discuss accumulating evidence that suggests Prdm factors drive cell fate by both promoting a cell type-specific program and by repressing alternative states that the cell can acquire.

Cross-species conservation of Prdm functions

The Prdm family first appeared in metazoans (Fumasoni et al., 2007). Three family members have been described in *Drosophila*, four in *C. elegans* and seven in sea urchin (*Strongylocentrotus purpuratus*) (Table 2). There is a base group of 15 Prdm family members in vertebrates (Fig. 1, Table 2). However, gene duplication and loss events have changed this number in different species. Prdm9 is lost from *Xenopus* and birds, but is duplicated in primates to create Prdm7 (Fumasoni et al., 2007; Oliver et al., 2009). Further paralogue groups (see Glossary, Box 1) of Prdm1a, Prdm1b and Prdm1c, and also Prdm8a and Prdm8b are found in zebrafish (Sun et al., 2008).

Prdm1 is the family member examined most extensively across species (Bikoff et al., 2009). It controls endomesoderm (see Glossary, Box 1) formation in sea urchin and starfish (Livi and Davidson, 2006), may perform a similar role in lamprey (Nikitina et al., 2011), and controls the formation of anterior endomesoderm in *Xenopus* and zebrafish (Baxendale et al., 2004; de Souza et al., 1999; Wilm and Solnica-Krezel, 2005). Further examples of putative evolutionarily shared function can be found in Tables 2 and 3, although the extent to which Prdm1-dependent

developmental mechanisms are conserved between analogous developmental events remains to be determined (Bikoff et al., 2009).

Prdm1 stabilizes photoreceptor identity

Evolutionary conservation of Prdm1 action is evident in the retina: disruption of *Prdm1* in both zebrafish and mouse leads to loss of photoreceptor cells (see Glossary, Box 1) (Brzezinski et al., 2010; Katoh et al., 2010; Wilm and Solnica-Krezel, 2005), and studies in the mouse have addressed the mechanism of Prdm1 action. Retinal bipolar cells (see Glossary, Box 1) and photoreceptor cells (see Glossary, Box 1) derive from a shared Chx10 (visual system homeobox 2, Vsx2)-positive proliferating precursor cell (Fig. 2). Prdm1 is transiently expressed in nascent photoreceptors. Ectopic expression of Prdm1 reduces the number of bipolar cells and increases the number of photoreceptors (Brzezinski et al., 2010; Katoh et al., 2010). Conversely, ectopic expression of Chx10 converts photoreceptors into bipolar cells (Livne-Bar et al., 2006). Prdm1 binds multiple canonical Prdm1-binding sites in the *Chx10* promoter in vivo, and represses expression of a *Chx10-luciferase* reporter construct in vitro (Brzezinski et al., 2010; Katoh et al., 2010). In *Prdm1* mutants, a nascent Chx10-negative photoreceptor forms, but as differentiation proceeds, Chx10 is re-expressed and the cell becomes re-specified to a bipolar or proliferating precursor state. These data suggest that the normal role of Prdm1 is not to induce photoreceptor formation, but rather to stabilize or maintain photoreceptor identity by repressing alternative differentiation programs (Fig. 2).

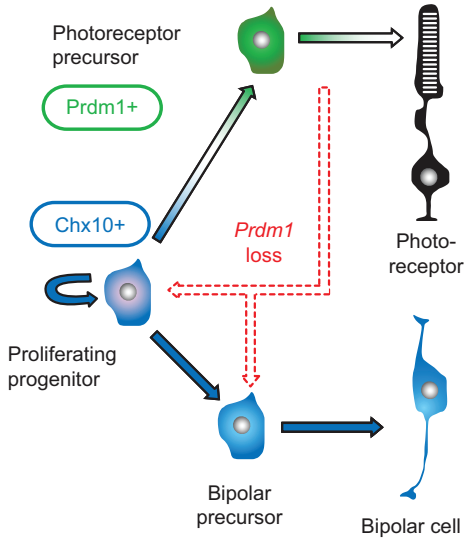


Fig. 2. Prdm1 regulates photoreceptor development. Chx10-positive progenitors give rise to both photoreceptors and bipolar cells. Daughters that maintain Chx10 become bipolar cells, whereas those that downregulate Chx10 become photoreceptors. In nascent photoreceptors, a pulse of Prdm1 expression keeps Chx10 repressed. Loss of *Prdm1* leads to re-expression of Chx10 in the nascent photoreceptors and they re-specify as Chx10-positive bipolar- or progenitor-like cells. Subsequently, many of these cells undergo apoptosis.

Prdm1 suppresses the adult enterocyte reprogramming event

Further recent reports emphasize the role of Prdm1 in maintaining one developmental state by repressing the transition to another. Neonatal mice feed solely on milk, and this requires enterocytes (see Glossary, Box 1) that are metabolically tuned for processing lactose-rich fluid. During weaning onto solid food, this requirement is changed. Thus, adult enterocytes exhibit reduced expression of factors required for milk processing and enhanced expression of factors required for breakdown of complex carbohydrates. Interestingly, Prdm1 is found only in neonatal and not in adult enterocytes, and, during weaning, neonatal Prdm1-positive enterocytes are systematically replaced by Prdm1-negative adult ones (Harper et al., 2011; Muncan et al., 2011). Lineage tracing shows that the adult Prdm1-negative enterocytes derive from a Prdm1-positive population, presumably a subset of neonatal enterocytes that have downregulated Prdm1 and undergone expansion (Harper et al., 2011). Importantly, *Prdm1*-deficient neonatal enterocytes show precocious activation of the adult gene expression program and simultaneous loss of the neonatal one (Harper et al., 2011; Muncan et al., 2011). Thus, Prdm1 represses adult and promotes neonatal enterocyte metabolism, and its loss is a trigger for the programmed developmental transition in enterocytes required for weaning.

Prdm1 action in the B- and T-cell lineages

Prdm1 action has been extensively examined in the B-cell lineage during the differentiation of antibody-secreting plasma cells (Turner et al., 1994). Prdm1 is not only needed to acquire plasmocytic properties, it is continually required for the maintenance of long-lived plasma cells, suggesting that, at some loci, it maintains a repressive state (Shapiro-Shelef et al., 2005). During plasma cell differentiation, Prdm1 directly represses the proliferation factor *Myc* through recruitment of HDACs (Yu et al., 2000); this leads to a strong reduction of histone 3 acetylation (H3Ac) at the *Myc* promoter. Prdm1 also represses *Ciita* (MHC class II trans-activator) through Lsd1 (Su et al., 2009). RNAi knockdown experiments showed that both Prdm1 and Lsd1 repress H3Ac and histone 3 lysine 4 dimethylation (H3K4me2) at this locus; Lsd1 also suppressed H3K4me3 (Su et al., 2009). Other targets of Prdm1 in plasma cell differentiation are *Pax5* (paired box 5) (which is required for B cell fate specification, must be downregulated during plasma cell differentiation and is also active in the germinal centre) and *Bcl6* (B-cell lymphoma 6), which promotes germinal centre B cell identity (reviewed by Belz and Kallies, 2010; Crotty et al., 2010; Martins and Calame, 2008). Notably, Bcl6 also acts as a transcriptional repressor and forms a negative-feedback loop with Prdm1 (reviewed by Crotty et al., 2010).

Prdm1 also acts as a repressor in subsets of effector $CD4^+$ and $CD8^+$ T cells (Cretney et al., 2011; Kallies et al., 2006; Kallies et al., 2009; Martins et al., 2006; Nutt et al., 2008; Rutishauser et al., 2009) and *Bcl6* is once more a target (Crotty et al., 2010). Nevertheless, Prdm1 appears to modulate predominantly different sets of targets between B- and T-cell lineages (Martins and Calame, 2008). One repressed target in T cells is the cytokine *Il2* (interleukin 2). Il2 regulates the initial expansion of naïve T cells then, acting through Stat5 (signal transducer and activator of transcription 5), it induces Prdm1 expression creating a feedback loop that downregulates its own expression during the later stages of T-cell differentiation (Martins et al., 2008). Prdm1 also functions in natural killer (NK) cells (Kallies et al., 2011; Smith et al., 2010), dendritic cells (Chan et al., 2009) and macrophages (Chang et al., 2000). (The extensive literature covering Prdm1 function in

immunology is beyond the scope of this review; thus, for comprehensive recent reviews, see Belz and Kallies, 2010; Crotty et al., 2010; Martins and Calame, 2008; Nutt et al., 2007.)

Interestingly, although Prdm1 is characterized as a transcriptional repressor, it can also promote transcriptional activation in combination with Irf4 (interferon regulatory factor 4) in regulatory T cells. At the *Il10* (interleukin 10) locus, Irf4 removes repressive H3K27 and Prdm1 promotes upregulation of activating H3K4 histone methylation (Cretney et al., 2011). Such bi-directional transcriptional control by Prdm1 may occur in other contexts. In particular, examination of putative Prdm1 targets (defined by the presence of canonical Prdm1 binding sites) in *Prdm1* mutant embryonic enterocytes noted a significant number of genes that showed downregulation, suggesting they may be positively regulated by this factor (Harper et al., 2011).

Prdm16 promotes brown fat and represses alternative states

How can a single Prdm act both as a repressor and activator in the same cell during a single developmental event? Recent studies of Prdm16 activity illustrate a mechanism through which this can occur. Prdm16 promotes brown adipose tissue (BAT) identity and simultaneously represses alternative fates (Kajimura et al., 2009; Kajimura et al., 2008; Seale et al., 2008; Seale et al., 2011; Seale et al., 2007). BAT is specialized to dissipate chemical energy by creating heat in defence against cold. It is common in human babies and small mammals. By contrast, the common form of human adult fat is white adipose tissue (WAT), which stores chemical energy. Weight gain is primarily caused by an energy imbalance; in other words, the excess of energy intake over energy consumption. Therefore, understanding the molecular principles underlying brown adipogenesis is a step towards BAT gene program manipulation, which could be used to combat human obesity (Fruhbeck et al., 2009). Lineage tracing shows that BAT derives from Myf5 (myogenic factor 5)-positive precursor cells that also give rise to skeletal muscle (Seale et al., 2008) (Fig. 3A). Removal of *Prdm16* from cultured brown adipocyte (see Glossary, Box 1) precursor cells by RNAi leads to loss of BAT-specific genes and upregulation of the myocyte (see Glossary, Box 1) program, leading to their re-specification as skeletal myocytes (see Glossary, Box 1). A similar, albeit less robust, phenotype is observed in *Prdm16* mutant mice (Seale et al., 2007; Seale et al., 2008) (Fig. 3A).

Prdm16 was originally identified as a leukaemogenic oncogene (Mochizuki et al., 2000; Morishita, 2007; Nishikata et al., 2003). Sequence-specific DNA binding through its two zinc-finger domains is crucial for its transcriptional activity in this pathogenic context (Nishikata et al., 2003). When Prdm16 promotes the BAT-specific genetic program, however, it does not target DNA itself. Rather Prdm16 binds to and is targeted by the sequence-specific DNA-binding protein C/EBPβ (CCAAT/enhancer-binding protein β) in order to initiate the BAT program (Fig. 3B) (Kajimura et al., 2009; Seale et al., 2009; Seale et al., 2007). Forced expression of Prdm16 and C/EBPβ can convert naïve fibroblasts into BAT, an exciting potential therapeutic tool (Kajimura et al., 2009; Seale et al., 2008). Among the factors induced by the Prdm16-C/EBPβ complex are Pparγ (peroxisome-proliferator-activated receptor γ) and Ppargc1α (Pparγ co-activator 1α). After induction of these factors, they now form new complexes with Prdm16 to again target its activity, this time to drive terminal BAT maturation (Fig. 3B).

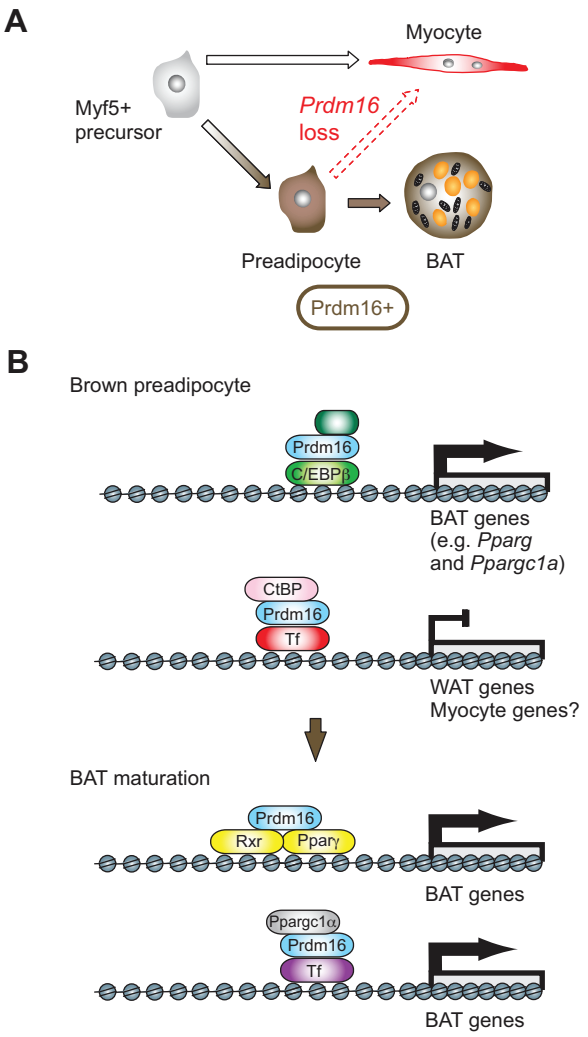


Fig. 3. Prdm16 promotes brown fat and represses white fat cell fate. (**A**) Myf5-positive precursors give rise to both skeletal muscle (myocytes) and brown adipose tissue (BAT). Prdm16 induces the formation of brown preadipocytes, and continued Prdm16 expression drives BAT differentiation. Loss of *Prdm16* from the preadipocytes causes loss of BAT gene expression with accompanying induction of myocyte gene expression and morphological differentiation. (**B**) Prdm16 (blue) associates with C/EBPβ (CCAAT/ enhancer-binding protein β; green) to promote the formation of a brown preadipocyte. Among the genes induced by Prdm16 are *Pparg* (peroxisome-proliferator-activated receptor γ; yellow) and *Ppargc1a* (PPARγ co-activator 1α; grey). These factors then associate with Prdm16 and other factors to promote full BAT maturation. Simultaneously, Prdm16 recruits CtBP (C-terminal-binding protein; pink) and acts as a repressor at a number of WAT (white adipose tissue) gene loci. Rxr, retinoid X receptor; Tf, additional unidentified transcription factors that may be involved in Prdm1b targeting.

Prdm16 also induces the formation of beige cells, BAT-like adipocytes that emerge in subcutaneous WAT as adaptive responses to cold exposure and to overfeeding (Seale et al., 2011). Beige cells may form via direct trans-differentiation of WAT; however, their definitive origin remains unclear (Barbatelli et al., 2011; Seale et al., 2011). In cultured adipocytes, Prdm16 is recruited to promoters of BAT-specific genes, activating their expression, and is simultaneously recruited to promoters of WAT-specific genes and represses their expression through recruitment of the co-repressor

CtBP (Fig. 3B) (Kajimura et al., 2008). These studies show how Prdm16 acts both as a transcriptional activator and repressor within the same cell by forming different molecular complexes in a promoter-context-dependent manner (Fig. 3B).

Sequence-specific targeting of Prdms through complex formation

An outstanding feature of Prdm16 action in BAT formation is that it is targeted to specific loci via other DNA-binding partners. Prdm3 may have a similar mode of action at the *Pbx1* (pre-B-cell leukaemia homeobox 1) locus (Shimabe et al., 2009), and Prdm8 at the *Cdh11* (cadherin 11) locus (Ross et al., 2012). Prdm8 is expressed in several regions of the developing nervous system (Table 2) (Kinameri et al., 2008; Komai et al., 2009) and is co-expressed in several places with the DNA sequence-specific bHLH (basic helix-loop-helix) transcription factor Bhlhb5 (Bhlhe22 – Mouse Genome Informatics) (Ross et al., 2012). Mice lacking *Prdm8* or *Bhlhb5* have very similar behavioural phenotypes and defects in axon outgrowth. Bhlhb5 targets several loci involved in neuronal development, including *Cdh11*, where it recruits Prdm8 that in turn represses transcription (Ross et al., 2012); whether this is through the intrinsic H3K9 methyltransferase activity of Prdm8 is unclear (Eom et al., 2009).

The above examples demonstrate how Prdms can promote cell fate choice and cell differentiation. Prdm factors can act by repressing the ability of a cell to transition to an alternative fate or to an alternative differentiation state. They can act in a cell context-dependent manner. For example, the targets of Prdm1 are different in B- and T-cell lineages. They can also act both as activators and repressors within the same cell in a promoter context-dependent manner. For Prdm16, this occurs because different binding partners control activator versus repressor activity. Furthermore, the DNA targeting activity of Prdm zinc fingers is well established; nevertheless, as demonstrated for Prdm16 and Prdm8, targeting can also occur through complex formation with sequence-specific DNA-binding partners. During BAT maturation, Prdm16 switches partners, thus enhancing context-dependent transcriptional flexibility.

Prdms promote and maintain stem and primordial germ cell identity

Pluripotent germ cells (see Glossary, Box 1), as well as pluripotent stem cells in culture- and tissue-specific stem cells, must all generate and maintain highly specialized cell states. Recent evidence demonstrates that several members of the Prdm family play important roles in creating these states.

Prdms in haematopoietic stem cells

In the haematopoietic system, defects in stem cell function can result in a wide range of malignant phenotypes, including leukaemia; both Prdm3 and Prdm16 were originally identified because their dysregulation causes myeloid malignancies (see Box 2) (reviewed by Morishita, 2007). The normal role of Prdm3 and Prdm16, however, is to maintain quiescent long-term and proliferating short-term haematopoietic stems cells (HSCs) (Aguilo et al., 2011; Chuikov et al., 2010; Goyama et al., 2008; Zhang et al., 2011). Leukaemic stem cells may share self-renewal machinery with normal HSCs, and elucidating of how Prdm3 and Prdm16 govern HSC self-renewal might provide biological insight into pathogenesis of these leukaemias (Goyama et al., 2008). Care is required in such interpretations; nevertheless, comparison of gene expression in wild-type and *Prdm3* mutant HSCs and further assessment against putative Prdm3 targets in acute myeloid leukaemia samples suggest some common pathways (Goyama et al., 2008). Prdm16 and Prdm3 have highly related molecular structures. They also both cause acute myeloid leukaemia and loss of either leads to similar phenotypes in HSCs. However, transcriptome examination of *Prdm3* mutant and *Prdm16* mutant HSCs suggests they are regulating somewhat different programs (Aguilo et al., 2011). For example, Prdm3 promotes *Pbx1* expression, whereas Prdm16 may repress it (Aguilo et al., 2011; Goyama et al., 2008; Shimabe et al., 2009). Interestingly, Prdm16, but not Prdm3, also maintains embryonic cortical stem cells, and this occurs in part through regulating oxidative stress (Chuikov et al., 2010; Kinameri et al., 2008).

Prdms in primordial germ cells

Prdm1 and Prdm14 play essential roles in the generation of pluripotent primordial germ cells (PGCs; see Glossary, Box 1), the precursors of oocytes in females and spermatozoa in males. In addition, after germ cell differentiation, Prdm9 positions recombination hotspots during meiosis (reviewed by Hochwagen and Marais, 2010) (see Box 3). Mouse PGCs are induced from proximal epiblast cells at the embryonic/extra-embryonic boundary in response to a bone morphogenic protein 4 (Bmp4) signal from the extra-embryonic ectoderm. Bmp4 induces both Prdm1 and Prdm14, and both *Prdm1* and *Prdm14* mutants cannot form PGCs (Ohinata et al., 2009; Ohinata et al., 2005; Vincent et al., 2005; Yamaji et al., 2008). However, in both mutants, a limited number of transient PGC-like cells do form and this has allowed the use of transcriptome profiling to determine the genetic programs controlled by these factors during PGC specification (Kurimoto et al., 2008; Yamaji et al., 2008). During PGC specification, the somatic epiblast gene expression program must be repressed, PGC-specific factors upregulated and a pluripotency program acquired. Prdm1 and Prdm14 coordinate these processes. In *Prdm1* mutant PGC-like cells, almost all somatic genes are not downregulated; instead, they remain expressed at the same level as in adjacent somatic cells (Kurimoto et al., 2008). By contrast, *Prdm14* mutant PGC-like cells repress the somatic program normally (Yamaji et al., 2008). However, both Prdm1 and Prdm14 are required for the proper initiation and coordination of the PGC-specific gene expression program, and initial evidence suggests they may control divergent sets of targets that remain to be fully characterized (Kurimoto et al., 2008; Yamaji et al., 2008). Finally, both Prdm1 and Prdm14 promote PGC pluripotency. The pluripotency regulator Sox2

Box 3. Prdm9 and meiotic recombination

Meiotic recombination in germ cells creates genetic diversity within a population, and thus provides the substrate for natural selection. The great majority of recombination events are concentrated at recombination hotspots. Prdm9 (also known as Meisetz) marks these hotspots and, in *Prdm9* mutant mice, progression through meiotic prophase is blocked. Prdm9 binds through its zinc fingers to a specific DNA sequence that defines hotspot identity; then, through its PRDI-BF1-RIZ1 domain, Prdm9 induces H3K4 trimethylation at the bound site. Prdm9 zinc fingers are evolving very fast and alterations in zinc-finger sequence have been linked to changes in hotspot DNA sequence. Prdm9 zinc-finger sequence changes create hybrid sterility between *Mus musculus musculus* and *Mus musculus domesticus*, and thus underpin the speciation event between these mice. Allelic differences in Prdm9 zinc fingers are also present in humans with different geographic origins. The DNA target sequence is changed and this correlates with altered hotspot distribution. [A recent review deals with Prdm9 function (Hochwagen and Marais, 2010).]

(SRY box-containing 2) is expressed in the epiblast before PGC induction; however, nascent PGCs and surrounding somatic epiblast cells both lose Sox2 expression. Sox2 then becomes re-expressed in the PGCs, and this requires both Prdm1 and Prdm14 activity (Kurimoto et al., 2008; Yamaji et al., 2008). Prdm1, but not Prdm14, is also required for the PGC expression of Nanog, a second pluripotency factor (Kurimoto et al., 2008). (For further reviews of this subject, see Bikoff and Robertson, 2008; Saitou, 2009.)

PGC specification also involves genome-wide epigenetic reprogramming (Saitou, 2009; Saitou et al., 2012). Early in PGC formation, Prdm1 associates with the arginine-specific HMT Prmt5, and this association results in high levels of dimethylated histone 2A and histone 4 arginine 3 (H2A/H4 R3me2) (Ancelin et al., 2006). As PGC differentiation continues, translocation of the Prdm1-Prmt5 complex from the nucleus into the cytoplasm correlates with loss of H2A/H4R3me2 levels (Ancelin et al., 2006). Prdm14 is required for the reduction of PGC H3K9me2 levels during the early steps of epigenetic reprogramming. This activity presumably involves Prdm14-mediated repression of Ehmt1 (euchromatic histone methyltransferase 1), a HMT that promotes H3K9me2 in early embryonic tissues. Prdm14 also acts via an undetermined mechanism to upregulate H3K27me3 levels 1 day later (Yamaji et al, 2008).

Prdms in embryonic and induced pluripotent stem cells

In culture, stem cells must actively maintain a pluripotent state, and they must simultaneously repress programs that will promote commitment and differentiation into specific cell types. These are roles carried out by Prdm1 and Prdm14 in the forming PGCs, so do these factors also function in cultured stem cells? PGCs can be changed by culturing into pluripotent stem cells known as embryonic germ cells (EGCs; see Glossary, Box 1). The formation of these EGCs requires Prdm14 activity and the loss of Prdm1 expression (Ancelin et al., 2006; Durcova-Hills et al., 2008; Yamaji et al., 2008). Somatic cells, such as adult fibroblasts, can also be artificially converted into a stem cell-like state in culture. These induced pluripotent stem cells (iPSCs; see Glossary, Box 1) cells are produced using a cocktail of Sox2, Oct4 (Pou5f1; POU domain, class 5, transcription factor 1), Myc and Klf4 (Kruppel-like factor 4). Notably, Prdm14 also has iPSC induction activity, and can enhance the activity of this cocktail, even if c-Myc or Klf4 are removed (Chia et al., 2010).

Conversely, neither Prdm1 nor Prdm14 are required for the derivation of mouse embryonic stem cells (ESCs; see Glossary, Box 1), although lineage tracing has shown that ESCs commonly arise from Prdm1-positive precursors (Chu et al., 2011). Nonetheless, once formed, both mouse and human ESCs continuously require Prdm14 to maintain the ESC state. *Prdm14* RNAi knockdown leads to downregulation of ESC genes and upregulation of the extra-embryonic endoderm program along with morphological signs of differentiation. The phenotype is slightly different between mouse and human ESCs, possibly reflecting the differing tissues from which these cells are derived (Chia et al., 2010; Ma et al., 2011; Tsuneyoshi et al., 2008). Genome-wide ChIP-seq studies followed by target validation found that Prdm14 binds and activates the *Oct4* promoter, and co-binds with Sox2, Nanog or Oct4 at many other sites (Chia et al., 2010; Ma et al., 2011).

In summary, Prdm factors promote and maintain germ and stem cell fate. The extent to which the same programs are controlled between different factors remains to be determined. Although very similar in structure, Prdm3 and Prdm16 control differing gene expression programs in HSCs. In PGCs Prdm1 and Prdm14 also control complementary programs rather than a shared one. To what degree Prdm14 function in PGCs relates to its activity in cultured stem cell types is also unclear.

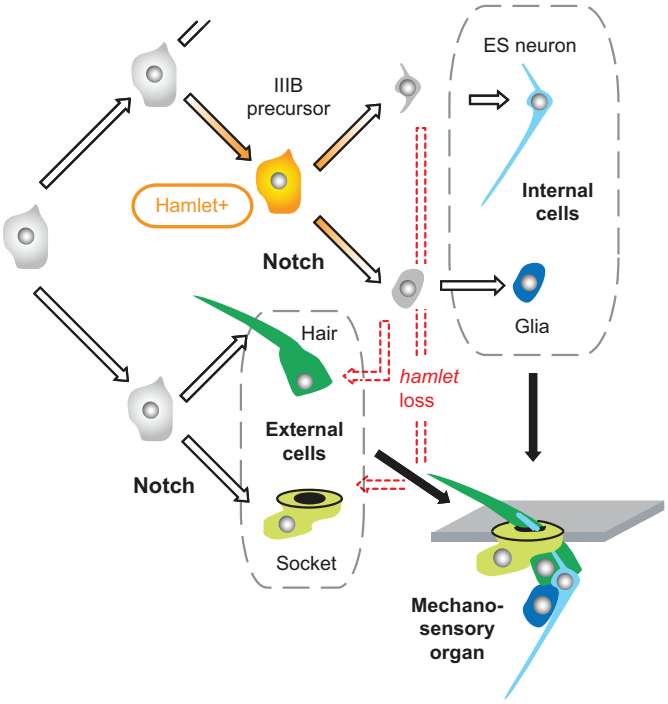


Fig. 4. Hamlet controls mechanosensory cell differentiation. The four cells of the *Drosophila* external sensory organ derive from a single precursor. The external cells – the hair and socket cells – are determined by a binary Notch (N) signalling event. The internal cells – the external sensory (ES) neuron and glial cells – are similarly determined. Hamlet expression initiates in the IIIB cell (orange), which is the precursor of ES neurons and glia. Its expression continues during IIIB division and is then downregulated in nascent internal cells. Loss of *hamlet* (red arrows) does not prevent initial expression of neural and glial cell markers. However, during differentiation, the cells lose these markers and re-specify as hair and socket cells. Remarkably, during these transformations, the internal cells re-specify to the opposite identity of that normally specified by N in the external cells.

Prdms modify developmental signalling pathways

Developmental processes produce a huge diversity of cell types. Surprisingly, in order to generate this diversity, nature uses only a handful of signalling systems, which function over and over again in different developmental contexts to create different outcomes (Pires-daSilva and Sommer, 2003). The output of signalling systems such as the transforming growth factor β (TGFβ) and Notch pathways is to regulate gene transcription through the action of dedicated sequence-specific transcription factors. Therefore, in order to ensure that these pleiotropic factors do not repeatedly activate the same targets in different developmental or cellular contexts, molecular mechanisms must exist to modulate the transcriptional activity in a 'context-dependent' manner. Recent studies, summarized below, suggest that Prdms can modify the transcriptional output of such developmental signalling pathways.

Modulation of TGFβ signalling

TGFβ signalling controls transcriptional regulation via sequence-specific Smad (MAD homologue) transcription factor activity (Moustakas and Heldin, 2009). A number of studies have shown

Table 3. An overview of Prdm factor loss- or reduction-of-function phenotypes

Factor	Loss- or reduction-of-function phenotypes	References
Prdm1	***C elegans***: Precocious onset of tail tip retraction[1,2].	(Nelson et al., 2011)
	Starfish, sea urchin: Failure to properly initiate the endoderm gene expression program[3,4].	(Hinman et al., 2003; Livi and Davidson, 2006; Livi and Davidson, 2007)
	Drosophila: Enlarged and irregular tracheal lumen. Precocious onset of prepupal to pupal transition[2,5].	(Agawa et al., 2007; Ng et al., 2006)
	Lamprey: Loss of anterior structures, including head[3].	(Nikitina et al., 2011)
	Xenopus: Loss of anterior structures, including head[4].	(de Souza et al., 1999)
	Zebrafish: Loss of anterior structures, including head[3]. Branchial arch defects possibly owing to loss of neural crest[3]. Failure of pectoral limb bud outgrowth[6]. Failure to specify precursors of Rohon-Beard neurons and neural crest[3,7]. Failure to induce slow twitch muscle program[6]. Loss of photoreceptors[3].	(Baxendale et al., 2004; van Eeden et al., 1996; Hernandez-Lagunas et al., 2005; Lee and Roy, 2006; Mercader et al., 2006; Wilm and Solnica-Krezel, 2005)
	Mouse: Failure to elaborate placental labyrinth layer[8,9]. Second and third branchial arches fail to form with apoptosis in the precursor mesenchyme[8,9]. Failed expansion of Prdm1-positive precursors that give rise to pharyngeal arches, ventricular septal defects and persistent truncus arteriosus[9-11]. Forelimb shows loss of posterior digits owing to failure to maintain zone of polarizing activity[9-11]. Loss of sensory vibrissae due to failure of precursor elaboration[9-11]. Nascent photoreceptors switch to a bipolar-like or precursor-like identity[10]. Failure of osteoclast differentiation, leading to loss of bone homeostasis[10]. Precocious expression of adult enterocyte metabolic and structural programs[9,10]. Maintenance of sebocyte stem cells and disrupted keratinocyte differentiation[10-12]. Failure to downregulate somatic and to upregulate PGC-specific gene programs[8,9,11,12].	(Harper et al., 2011; Horsley et al., 2006; Kallies et al., 2004; Magnusdottir et al., 2007; Miyauchi et al., 2010; Muncan et al., 2011; Nishikawa et al., 2010; Ohinata et al., 2005; Robertson et al., 2007; Shapiro-Shelef et al., 2003; Vincent et al., 2005)
Prdm2	**Mouse**: Oestrogen resistance in uterus, vagina and mammary gland[13].	(Carling et al., 2004; Steele-Perkins, 2001)
Prdm3	***C. elegans* (*egl-43*)**: Failed migration of HSN neurons and defective phasmid neuron structural differentiation. Failure of anchor cell invasion, failure of gene induction and modification of Notch signalling, leading to cell fate switches in the somatic gonad[14,15].	(Garriga et al., 1993; Hwang et al., 2007; Rimann and Hajnal, 2007)
	***Drosophila* (*hamlet*)**: Increased dendrite branching. Modification of Notch signalling and related olfactory receptor neuron class; external sensory organ cell fate switches[2,5,16].	(Endo et al., 2012; Moore et al., 2002; Moore et al., 2004)
	Xenopus: Disrupted pronephros patterning[4].	(Van Campenhout et al., 2006)
	Mouse: Loss of primitive haematopoiesis[17-21]. Failure to maintain embryonic and adult HSCs[17-21]. Forelimb patterning defects[22].	(Aguilo et al., 2011; Chuikov et al., 2010; Goyama et al., 2008; Hoyt et al., 1997; Parkinson et al., 2006; Sato et al., 2008; Zhang et al., 2011)
Prdm8	**Mouse**: Axon targeting defects and axon tract loss[23].	(Ross et al., 2012)
Prdm14	**Mouse**: Failure to upregulate multipotency and primordial germ cell-specific genes. Loss of primordial germ cells[24].	(Yamaji et al., 2008)
Prdm16	**Mouse**: Failure to maintain haematopoietic stems cell population and failure to maintain embryonic cortical stem cell population[25]. Failure of brown adipose tissue differentiation and ectopic expression of the myocyte gene program[25]. Cleft palate[25,26].	(Aguilo et al., 2011; Bjork et al., 2010; Chuikov et al., 2010; Kajimura et al., 2009; Seale et al., 2007; Seale et al., 2009)

[1]Prdm1(tm548), a gene deletion. [2]RNAi. [3]Morpholino. [4]Dominant negative. [5]Chromosomal deficiency spanning the gene locus. [6]Prdm1a^{tp39} (ubo), point mutant and putative hypomorph. [7]*Prdm1a*^{m805} (nrd), truncation and putative null. [8]*Prdm1*^{tm1Rob}, knockout. [9]*Prdm1*^{tm2Rob}, knockout. [10]*Prdm1*^{tm1Clme}, conditional knockout. [11]*Prdm1*^{tm1Nutt}, GFP knock-in and hypomorph. [12]*Prdm1*^{tm2Masu}, conditional knockout. [13]*Prdm2*^{tm1Shg}, knockout. [14]*egl43(n997* and *n1079)*, no molecular information. [15]*egl-43 (tm1802)*, gene deletion. [16]ham[1], truncation and null. [17]*Mecom*$^{tm1.1Miku}$, knockout. [18]*Mecom*^{tm1Miku}, conditional knockout. [19]*Mecom*$^{tm2.2Aspe}$, knockout. [20]*Mecom*^{tm1Aspe}, *lacZ* knock in and loss of function. [21]*Mecom*^{tm1Mmor}, a partial knockout that does not remove all *Prdm3* isoforms and leads to complex phenotypes with many aspects not seen in null knockouts (the mechanistic basis of these additional phenotypes is unclear and they are not reported here). [22]*Mecom*^{jbo}, a point mutant with a forelimb patterning phenotype not present in *Prdm3* knockouts. [23]*Prdm8*$^{-/-}$, no molecular information. [24]*Prdm14*^{tm1Sait}, knockout. [25]*Prdm16*$^{Gt(OST67423)Lex}$, *lacZ* genetrap and loss of function. [26]*Prdm16*^{csp1}, truncation and putative null.
Allele terminology from mouse genome informatics (http://www.informatics.jax.org/) and zebrafish model organism (www.zfin.org) databases.

that Prdm3 and Prdm16 bind Smads in order to repress Smad-mediated transcriptional activation (Alliston et al., 2005; Bjork et al., 2010; Sato et al., 2008; Takahata et al., 2009). This Prdm-mediated TGFβ repression requires recruitment of CtBP, which in turn recruits HDACs to deacetylate histones at Smad target promoters (Alliston et al., 2005; Izutsu et al., 2001). Smads usually recruit p300 to acetylate histones and thereby activate transcription. Surprisingly, however, binding of Prdm3 to Smads is also facilitated by p300. This means that the Prdm3-Smad complex, although primarily acting as a histone-deacetylating repressor complex, also contains putative histone-acetylating activity, providing a possible strategy for tuning Prdm3-Smad complex activity in different developmental contexts (Alliston et al., 2005).

Interplay with Notch signalling

In cells where Notch signalling is activated, the Notch intracellular domain (NICD) translocates to the nuclei where it binds to the DNA sequence-specific transcription factor CSL [C-promoter binding factor 1 (also called recombination signal binding protein J), Suppressor of Hairless and Lag-1)]. The resultant NICD/CSL complex activates Notch-target genes (Bray and Bernard, 2010).

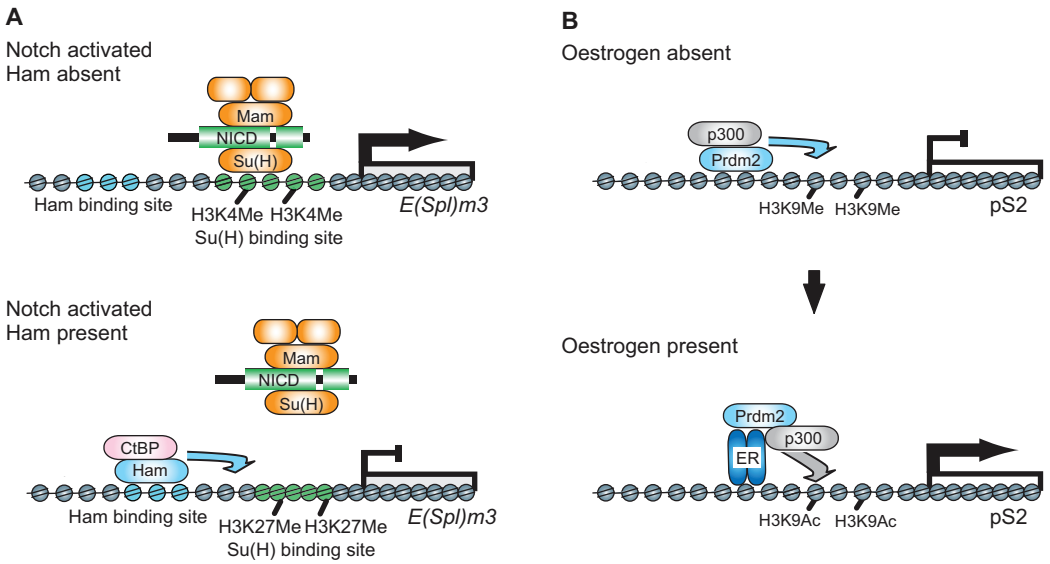


Fig. 5. Prdm interactions with signalling pathways. (**A**) Hamlet (Ham; light blue) modulates the Notch response at the *Enhancer of split m3* [*E(Spl)m3*] promoter. In Ham-positive cells, a Ham/CtBP (C-terminal-binding protein; pink) complex suppresses H3K4 and enhances H3K27 trimethylation at *E(Spl)m3*. This correlates with an increased histone density at the Suppressor of Hairless (SuH)-binding site and reduced accessibility of the transcription activator complex to the promoter, thus preventing *E(Spl)m3* induction. NICD, Notch intracellular domain (green); Mam, mastermind [NICD/Su(H)/Mam activator complex in green and orange]. (**B**) A Prdm2/p300 (Prdm2 in light blue; p300 in grey) complex modulates oestrogen receptor (ER; dark blue) signalling at the pS2 promoter. In the absence of oestrogen, the pS2 promoter is silenced by the endogenous H3K9 methyltransefase activity of Prdm2. Upon ER activation and translocation to the nucleus, the Prdm2/p300 complex associates with the ER, leading to a change whereby the histone acetylation activity of p300 now promotes transcriptional activation.

Recent studies in invertebrates have demonstrated a close relationship between Prdm function and NICD/CSL-mediated transcriptional regulation.

In *C. elegans*, EGL-43 (Fig. 1B,C) and Notch cooperate in two sequential signalling events during vulval development (Hwang et al., 2007; Rimann and Hajnal, 2007). Four precursor cells give rise to the vulva: one pair of cells directly adopts a ventral uterine precursor cell (VU) fate and the second pair undergoes a Notch-mediated binary cell fate choice. The cell with high Notch becomes another VU, whereas the cell with low Notch becomes the anchor cell (AC). EGL-43 is required to implement this Notch-mediated decision, as in *egl-43* mutants both cells adopt an AC fate. In the nascent VU cell, Notch is subsequently required to properly maintain EGL-43 expression. Finally, as the VU cells differentiate into π cells, Egl43 and Notch co-activate the *lin-11* homeobox gene (Hwang et al., 2007; Rimann and Hajnal, 2007). EGL-43 is additionally required in a Notch-independent manner for AC invasion of the vulval epithelium.

In *Drosophila*, the four cells of the notum mechanosensory organs (see Glossary, Box 1) derive from a single precursor via a stereotyped lineage (Lai and Orgogozo, 2004). The fates of all four cells are determined by the Notch signalling pathway. Thus, Notch-mediated binary cell fate decisions guide the formation of an external sensory (ES) neuron (low Notch) versus a glial cell (high Notch), and also the formation of a hair cell (low Notch) versus a socket cell (high Notch) (Fig. 4). Hamlet (Fig. 1B,C) is expressed in the IIIB intermediate precursor cell that gives rise to ES neurons and glia, and also in the nascent ES neurons and glia (Fig. 4) (Moore et al., 2002; Moore et al., 2004). When *hamlet* function is lost, the ES neuron and glia form, and they initially express neuronal and glial markers. However, as differentiation progresses, they re-specify into a second hair and socket cell (Moore et al., 2004). One peculiar aspect of this phenotype is that in *hamlet* mutants, the high-Notch glial cell transforms into a hair cell, but the hair cell is usually defined by a low-Notch state. Similarly, the low-Notch ES neuron converts into a socket, and this is a cell fate that is usually defined by high Notch activity (Fig. 4) (Moore et al., 2004).

Studies of *Drosophila* olfactory receptor neuron (ORN; see Glossary, Box 1) diversification have better illuminated the interaction between Hamlet and Notch (Endo et al., 2012). Hamlet acts as an epigenetic ‘reset switch’ that enables the Notch pathway to induce multiple distinct developmental outcomes for olfactory neurons. In *Drosophila*, there are three primary ORN identities, all of which are defined by Notch state (Endo et al., 2012). Two, Naa (high Notch) and Nab (low Notch), are sibling cells; the third, Nba (high Notch), is their cousin. Importantly, the Naa and Nab ORNs derive from a precursor cell, which was itself formed as the result of high Notch activity. Hamlet acts in the nascent Naa and Nab ORNs to erase the high-Notch transcriptional state of the parental cell. This then enables a successful new Notch signalling event to be used in the nascent ORNs. In *hamlet* mutants, the parental cell Notch state is not properly reduced, and the usually low Notch Nab ORN undergoes a fate switch to a high-Notch Nba identity (Endo et al., 2012). At the molecular level, Hamlet forms a complex with CtBP, and alters histone methylation status and organization at the promoter of the Notch target *Enhancer of split m3* [*E(spl)m3*]. At this locus, Hamlet upregulates H3K27me3, downregulates H3K4me3 and increases histone density at the CSL-binding site; correspondingly, the NICD/CSL activator complex is unable to bind properly to the *E(spl)m3* promoter and the locus becomes refractory to Notch-mediated induction (Fig. 5A) (Endo et al., 2012).

Transition from a precursor to an immature neuronal state also occurs during mouse cortical neurogenesis and is controlled by Notch signalling. During this transition, cells switch from Prdm16

to Prdm8 expression (Kinameri et al., 2008). However, whether these factors also influence Notch pathway activity is yet to be addressed.

Effects of Prdms on oestrogen signalling

A further study links the bi-directional transcriptional activity that can be associated with Prdm factor activity to the modification of signalling pathway activity. This study shows that Prdm2 acts at an oestrogen-activated locus, first as a repressor in advance of signalling and then, acting at the same locus, it switches to an activator during signalling (Fig. 5B). In this way, it accentuates the transcriptional response to oestrogen action. In line with this, *Prdm2* mutant mice have reduced reproductive capabilities and impaired responses to oestrogen signalling, such as markedly reduced mammopoiesis during pregnancy and in response to experimental hormone administration (Carling et al., 2004). At the molecular level, Prdm2 is associated with the oestrogen target *pS2* promoter in advance of oestrogen receipt. In this state, it mediates promoter repression via H3K9 methylation. Notably, Prdm2 is in a complex with p300, but p300 does not exert its activity when in this state. The activity of the Prdm2-p300 complex changes upon oestrogen activation. The estrogen receptor (ER) enters the nucleus, Prdm2 dissociates from the DNA, it binds directly to the ER, and together they create a Prdm2-p300-ER activation complex that drives histone acetylation, resulting in a transcriptionally active chromatin state (Carling et al., 2004) (Fig. 5B).

Taken together, these studies show that the molecular activity of Prdm factors makes them good candidates to modify the transcriptional output of signalling system activation. EGL-43 in *C. elegans* and Hamlet in *Drosophila* are linked to the Notch pathway (Endo et al., 2012; Hwang et al., 2007; Moore et al., 2004; Rimann and Hajnal, 2007), and Prdm3 and Prdm16 to TGFβ (Alliston et al., 2005; Bjork et al., 2010; Sato et al., 2008; Takahata et al., 2009). Bi-directional transcription regulation activity of Prdm2 at the *pS2* promoter provides one solution of how to amplify the outcome of signalling at a specific target promoter (Carling et al., 2004).

Conclusions

In summary, Prdm factors can act as direct HMTs or they can recruit a suite of histone-modifying enzymes to target promoters. Besides being associated with tumour formation, they function in many developmental contexts. They specify cell fate choice, and drive cell differentiation and maturation events. They can also maintain specialized cell fates, including that of HSCs and pluripotent ESCs (Aguilo et al., 2011; Chia et al., 2010; Chuikov et al., 2010; Goyama et al., 2008; Ma et al., 2011; Tsuneyoshi et al., 2008). Prdm9 has a divergent role in controlling meiotic crossing over (Hayashi et al., 2005; Hochwagen and Marais, 2010).

Importantly, Prdm factors show strong context dependency. Prdm-controlled genetic programs vary significantly for the same factor from cell type to cell type, e.g. Prdm1 in B and T cells (Martins and Calame, 2008). They also vary for closely related factors in the same cell type e.g. Prdm3 and Prdm16 in HSCs (Aguilo et al., 2011; Goyama et al., 2008). Even so, contained within these divergent programs may be key shared factors. For example, Prdm1 repression of *Bcl6* occurs in both B and T cell lineages (Crotty et al., 2010; Martins and Calame, 2008), and also in osteoclasts (see Glossary, Box 1) (Miyauchi et al., 2010; Nishikawa et al., 2010). Prdms can also have bi-directional transcriptional activity in the same process. For example, Prdm16 acts simultaneously at different loci as an activator or repressor during BAT specification (Kajimura et al., 2008) and Prdm2 transitions from repressor to activator at a single locus in response to oestrogen (Carling et al., 2004). What cellular contexts and promoter-specific mechanisms determine whether a Prdm factor will control gene transcription (i.e. how is cell type context dependency set) and whether it will act either as activator or repressor at a particular locus? Elucidating this will be central to our future understanding of how Prdm factors coordinate developmental processes.

Finally, recent evidence shows that Prdm factors can regulate the outcome of developmental signalling pathway activity. At the transcriptional level, this is best illustrated by EGL-43- and Hamlet-mediated regulation of Notch in invertebrates (Endo et al., 2012; Hwang et al., 2007; Rimann and Hajnal, 2007), and by Prdm3- and Prdm16-mediated regulation of Smad activity in vertebrates (Alliston et al., 2005; Bjork et al., 2010; Izutsu et al., 2001; Sato et al., 2008; Takahata et al., 2009). Because Prdm factors can create global changes in chromatin state, they have interesting potential to construct cell-type-specific chromatin contexts upon which developmental signalling pathways can operate.

Acknowledgements

We thank members of the Moore laboratory for many discussions, and we apologize to authors whose work was omitted from this article because of space limitations.

Funding

This work was funded by a Japan Society for Promotion of Sciences (JSPS) Grants-in-Aid Young Scientists (B) and a RIKEN Brain Science Institute (BSI) core grant to A.W.M., and by a RIKEN Foreign Postdoctoral Researcher award to T.H.

Competing interests statement

The authors declare no competing financial interests.

References

Agawa, Y., Sarhan, M., Kageyama, Y., Akagi, K., Takai, M., Hashiyama, K., Wada, T., Handa, H., Iwamatsu, A., Hirose, S. et al. (2007). Drosophila Blimp-1 is a transient transcriptional repressor that controls timing of the ecdysone-induced developmental pathway. *Mol. Cell. Biol.* **27**, 8739-8747.

Aguilo, F., Avagyan, S., Labar, A., Sevilla, A., Lee, D. F., Kumar, P., Lemischka, I. R., Zhou, B. Y. and Snoeck, H. W. (2011). Prdm16 is a physiologic regulator of hematopoietic stem cells. *Blood* **117**, 5057-5066.

Alliston, T., Ko, T. C., Cao, Y., Liang, Y. Y., Feng, X. H., Chang, C. and Derynck, R. (2005). Repression of bone morphogenetic protein and activin-inducible transcription by Evi-1. *J. Biol. Chem.* **280**, 24227-24238.

Ancelin, K., Lange, U. C., Hajkova, P., Schneider, R., Bannister, A. J., Kouzarides, T. and Surani, M. A. (2006). Blimp1 associates with Prmt5 and directs histone arginine methylation in mouse germ cells. *Nat. Cell Biol.* **8**, 623-630.

Barbatelli, G., Murano, I., Madsen, L., Hao, Q., Jimenez, M., Kristiansen, K., Giacobino, J. P., De Matteis, R. and Cinti, S. (2011). The emergence of cold-induced brown adipocytes in mouse white fat depots is determined predominantly by white to brown adipocyte transdifferentiation. *Am. J. Physiol. Endocrinol. Metab.* **298**, E1244-E1253.

Bard-Chapeau, E. A., Jeyakani, J., Kok, C. H., Muller, J., Chua, B. Q., Gunaratne, J., Batagov, A., Jenjaroenpun, P., Kuznetsov, V. A., Wei, C. L. et al. (2012). Ecotopic viral integration site 1 (EVI1) regulates multiple cellular processes important for cancer and is a synergistic partner for FOS protein in invasive tumors. *Proc. Natl. Acad. Sci. USA* **109**, 2168-2173.

Bartholomew, C., Kilbey, A., Clark, A. M. and Walker, M. (1997). The Evi-1 proto-oncogene encodes a transcriptional repressor activity associated with transformation. *Oncogene* **14**, 569-577.

Baudat, F., Buard, J., Grey, C., Fledel-Alon, A., Ober, C., Przeworski, M., Coop, G. and de Massy, B. (2010). PRDM9 is a major determinant of meiotic recombination hotspots in humans and mice. *Science* **327**, 836-840.

Baxendale, S., Davison, C., Muxworthy, C., Wolff, C., Ingham, P. W. and Roy, S. (2004). The B-cell maturation factor Blimp-1 specifies vertebrate slow-twitch muscle fiber identity in response to Hedgehog signaling. *Nat. Genet.* **36**, 88-93.

Behrends, U., Schneider, I., Rossler, S., Frauenknecht, H., Golbeck, A., Lechner, B., Eigenstetter, G., Zobywalski, C., Muller-Weihrich, S., Graubner, U. et al. (2003). Novel tumor antigens identified by autologous

antibody screening of childhood medulloblastoma cDNA libraries. *Int. J. Cancer* **106**, 244-251.
Bei, J. X., Li, Y., Jia, W. H., Feng, B. J., Zhou, G., Chen, L. Z., Feng, Q. S., Low, H. Q., Zhang, H., He, F. et al. (2010). A genome-wide association study of nasopharyngeal carcinoma identifies three new susceptibility loci. *Nat. Genet.* **42**, 599-603.
Belz, G. T. and Kallies, A. (2010). Effector and memory CD8+ T cell differentiation: toward a molecular understanding of fate determination. *Curr. Opin. Immunol.* **22**, 279-285.
Bikoff, E. K. and Robertson, E. J. (2008). One PRDM is not enough for germ cell development. *Nat. Genet.* **40**, 934-935.
Bikoff, E. K., Morgan, M. A. and Robertson, E. J. (2009). An expanding job description for Blimp-1/PRDM1. *Curr. Opin. Genet. Dev.* **19**, 379-385.
Bjork, B. C., Turbe-Doan, A., Prysak, M., Herron, B. J. and Beier, D. R. (2010). Prdm16 is required for normal palatogenesis in mice. *Hum. Mol. Genet.* **19**, 774-789.
Bray, S. and Bernard, F. (2010). Notch targets and their regulation. *Curr. Top. Dev. Biol.* **92**, 253-275.
Briknarova, K., Zhou, X., Satterthwait, A., Hoyt, D. W., Ely, K. R. and Huang, S. (2008). Structural studies of the SET domain from RIZ1 tumor suppressor. *Biochem. Biophys. Res. Commun.* **366**, 807-813.
Brzezinski, J. A., Lamba, D. A. and Reh, T. A. (2010). Blimp1 controls photoreceptor versus bipolar cell fate choice during retinal development. *Development* **137**, 619-629.
Burkitt Wright, E. M., Spencer, H. L., Daly, S. B., Manson, F. D., Zeef, L. A., Urquhart, J., Zoppi, N., Bonshek, R., Tosounidis, I., Mohan, M. et al. (2011). Mutations in PRDM5 in brittle cornea syndrome identify a pathway regulating extracellular matrix development and maintenance. *Am. J. Hum. Genet.* **88**, 767-777.
Buyse, I. M., Shao, G. and Huang, S. (1995). The retinoblastoma protein binds to RIZ, a zinc-finger protein that shares an epitope with the adenovirus E1A protein. *Proc. Natl. Acad. Sci. USA* **92**, 4467-4471.
Calado, D. P., Zhang, B., Srinivasan, L., Sasaki, Y., Seagal, J., Unitt, C., Rodig, S., Kutok, J., Tarakhovsky, A., Schmidt-Supprian, M. et al. (2010). Constitutive canonical NF-kappaB activation cooperates with disruption of BLIMP1 in the pathogenesis of activated B cell-like diffuse large cell lymphoma. *Cancer Cell* **18**, 580-589.
Carling, T., Kim, K. C., Yang, X. H., Gu, J., Zhang, X. K. and Huang, S. (2004). A histone methyltransferase is required for maximal response to female sex hormones. *Mol. Cell. Biol.* **24**, 7032-7042.
Cattaneo, F. and Nucifora, G. (2008). EVI1 recruits the histone methyltransferase SUV39H1 for transcription repression. *J. Cell Biochem.* **105**, 344-352.
Chakraborty, S., Senyuk, V., Sitailo, S., Chi, Y. and Nucifora, G. (2001). Interaction of EVI1 with cAMP-responsive element-binding protein-binding protein (CBP) and p300/CBP-associated factor (P/CAF) results in reversible acetylation of EVI1 and in co-localization in nuclear speckles. *J. Biol. Chem.* **276**, 44936-44943.
Chan, Y. H., Chiang, M. F., Tsai, Y. C., Su, S. T., Chen, M. H., Hou, M. S. and Lin, K. I. (2009). Absence of the transcriptional repressor Blimp-1 in hematopoietic lineages reveals its role in dendritic cell homeostatic development and function. *J. Immunol.* **183**, 7039-7046.
Chang, D. H., Angelin-Duclos, C. and Calame, K. (2000). BLIMP-1: trigger for differentiation of myeloid lineage. *Nat. Immunol.* **1**, 169-176.
Chang, D. H., Cattoretti, G. and Calame, K. L. (2002). The dynamic expression pattern of B lymphocyte induced maturation protein-1 (Blimp-1) during mouse embryonic development. *Mech. Dev.* **117**, 305-309.
Chasman, D. I., Schurks, M., Anttila, V., de Vries, B., Schminke, U., Launer, L. J., Terwindt, G. M., van den Maagdenberg, A. M., Fendrich, K., Volzke, H. et al. (2011). Genome-wide association study reveals three susceptibility loci for common migraine in the general population. *Nat. Genet.* **43**, 695-698.
Chia, N. Y., Chan, Y. S., Feng, B., Lu, X., Orlov, Y. L., Moreau, D., Kumar, P., Yang, L., Jiang, J., Lau, M. S. et al. (2010). A genome-wide RNAi screen reveals determinants of human embryonic stem cell identity. *Nature* **468**, 316-320.
Chittka, A., Arevalo, J. C., Rodriguez-Guzman, M., Perez, P., Chao, M. V. and Sendtner, M. (2004). The p75NTR-interacting protein SC1 inhibits cell cycle progression by transcriptional repression of cyclin E. *J. Cell Biol.* **164**, 985-996.
Chu, L. F., Surani, M. A., Jaenisch, R. and Zwaka, T. P. (2011). Blimp1 expression predicts embryonic stem cell development in vitro. *Curr. Biol.* **21**, 1759-1765.
Chuikov, S., Levi, B. P., Smith, M. L. and Morrison, S. J. (2010). Prdm16 promotes stem cell maintenance in multiple tissues, partly by regulating oxidative stress. *Nat. Cell Biol.* **12**, 999-1006.
Cretney, E., Xin, A., Shi, W., Minnich, M., Masson, F., Miasari, M., Belz, G. T., Smyth, G. K., Busslinger, M., Nutt, S. L. et al. (2011). The transcription factors Blimp-1 and IRF4 jointly control the differentiation and function of effector regulatory T cells. *Nat. Immunol.* **12**, 304-311.
Crotty, S., Johnston, R. J. and Schoenberger, S. P. (2010). Effectors and memories: Bcl-6 and Blimp-1 in T and B lymphocyte differentiation. *Nat. Immunol.* **11**, 114-120.
Davis, C. A., Haberland, M., Arnold, M. A., Sutherland, L. B., McDonald, O. G., Richardson, J. A., Childs, G., Harris, S., Owens, G. K. and Olson, E. N. (2006). PRISM/PRDM6, a transcriptional repressor that promotes the proliferative gene program in smooth muscle cells. *Mol. Cell. Biol.* **26**, 2626-2636.
de Souza, F. S., Gawantka, V., Gomez, A. P., Delius, H., Ang, S. L. and Niehrs, C. (1999). The zinc finger gene Xblimp1 controls anterior endomesodermal cell fate in Spemann's organizer. *EMBO J.* **18**, 6062-6072.
Delwel, R., Funabiki, T., Kreider, B. L., Morishita, K. and Ihle, J. N. (1993). Four of the seven zinc fingers of the Evi-1 myeloid-transforming gene are required for sequence-specific binding to GA(C/T)AAGA(T/C)AAGATAA. *Mol. Cell. Biol.* **13**, 4291-4300.
Derunes, C., Briknarova, K., Geng, L., Li, S., Gessner, C. R., Hewitt, K., Wu, S., Huang, S., Woods, V. I., Jr and Ely, K. R. (2005). Characterization of the PR domain of RIZ1 histone methyltransferase. *Biochem. Biophys. Res. Commun.* **333**, 925-934.
Dettman, E. J., Simko, S. J., Ayanga, B., Carofino, B. L., Margolin, J. F., Morse, H. C., 3rd and Justice, M. J. (2011). Prdm14 initiates lymphoblastic leukemia after expanding a population of cells resembling common lymphoid progenitors. *Oncogene* **30**, 2859-2873.
Doody, G. M., Care, M. A., Burgoyne, N. J., Bradford, J. R., Bota, M., Bonifer, C., Westhead, D. R. and Tooze, R. M. (2011). An extended set of PRDM1/BLIMP1 target genes links binding motif type to dynamic repression. *Nucleic Acids Res.* **38**, 5336-5350.
Duan, Z., Person, R. E., Lee, H. H., Huang, S., Donadieu, J., Badolato, R., Grimes, H. L., Papayannopoulou, T. and Horwitz, M. S. (2007). Epigenetic regulation of protein-coding and microRNA genes by the Gfi1-interacting tumor suppressor PRDM5. *Mol. Cell. Biol.* **27**, 6889-6902.
Durcova-Hills, G., Tang, F., Doody, G., Tooze, R. and Surani, M. A. (2008). Reprogramming primordial germ cells into pluripotent stem cells. *PLoS ONE* **3**, e3531.
Endo, K., Karim, M. R., Taniguchi, H., Krejci, A., Kinameri, E., Siebert, M., Ito, K., Bray, S. J. and Moore, A. W. (2012). Chromatin modification of Notch targets in olfactory receptor neuron diversification. *Nat. Neurosci.* **15**, 224-233.
Eom, G. H., Kim, K., Kim, S. M., Kee, H. J., Kim, J. Y., Jin, H. M., Kim, J. R., Kim, J. H., Choe, N., Kim, K. B. et al. (2009). Histone methyltransferase PRDM8 regulates mouse testis steroidogenesis. *Biochem. Biophys. Res. Commun.* **388**, 131-136.
Fears, S., Mathieu, C., Zeleznik-Le N., Huang, S., Rowley, J. D. and Nucifora, G. (1996). Intergenic splicing of MDS1 and EVI1 occurs in normal tissues as well as in myeloid leukemia and produces a new member of the PR domain family. *Proc. Natl. Acad. Sci. USA* **93**, 1642-1647.
Fog, C. K., Galli, G. G. and Lund, A. H. (2012). PRDM proteins: Important players in differentiation and disease. *BioEssays* **34**, 50-60.
Fruhbeck, G., Becerril, S., Sainz, N., Garrastachu, P. and Garcia-Velloso, M. J. (2009). BAT: a new target for human obesity? *Trends Pharmacol. Sci.* **30**, 387-396.
Fumasoni, I., Meani, N., Rambaldi, D., Scafetta, G., Alcalay, M. and Ciccarelli, F. D. (2007). Family expansion and gene rearrangements contributed to the functional specialization of PRDM genes in vertebrates. *BMC Evol. Biol.* **7**, 187.
Funabiki, T., Kreider, B. L. and Ihle, J. N. (1994). The carboxyl domain of zinc fingers of the Evi-1 myeloid transforming gene binds a consensus sequence of GAAGATGAG. *Oncogene* **9**, 1575-1581.
Garriga, G., Guenther, C. and Horvitz, H. R. (1993). Migrations of the Caenorhabditis elegans HSNs are regulated by egl-43, a gene encoding two zinc finger proteins. *Genes Dev.* **7**, 2097-2109.
Gateva, V., Sandling, J. K., Hom, G., Taylor, K. E., Chung, S. A., Sun, X., Ortmann, W., Kosoy, R., Ferreira, R. C., Nordmark, G. et al. (2009). A large-scale replication study identifies TNIP1, PRDM1, JAZF1, UHRF1BP1 and IL10 as risk loci for systemic lupus erythematosus. *Nat. Genet.* **41**, 1228-1233.
Goyama, S., Yamamoto, G., Shimabe, M., Sato, T., Ichikawa, M., Ogawa, S., Chiba, S. and Kurokawa, M. (2008). Evi-1 is a critical regulator for hematopoietic stem cells and transformed leukemic cells. *Cell Stem Cell* **3**, 207-220.
Gupta, S., Anthony, A. and Pernis, A. B. (2001). Stage-specific modulation of IFN-regulatory factor 4 function by Kruppel-type zinc finger proteins. *J. Immunol.* **166**, 6104-6111.
Gyory, I., Fejer, G., Ghosh, N., Seto, E. and Wright, K. L. (2003). Identification of a functionally impaired positive regulatory domain I binding factor 1 transcription repressor in myeloma cell lines. *J. Immunol.* **170**, 3125-3133.
Gyory, I., Wu, J., Fejer, G., Seto, E. and Wright, K. L. (2004). PRDI-BF1 recruits the histone H3 methyltransferase G9a in transcriptional silencing. *Nat. Immunol.* **5**, 299-308.
Harper, J., Mould, A., Andrews, R. M., Bikoff, E. K. and Robertson, E. J. (2011). The transcriptional repressor Blimp1/Prdm1 regulates postnatal reprogramming of intestinal enterocytes. *Proc. Natl. Acad. Sci. USA* **108**, 10585-10598.
Hayashi, K., Yoshida, K. and Matsui, Y. (2005). A histone H3 methyltransferase controls epigenetic events required for meiotic prophase. *Nature* **438**, 374-378.
Hernandez-Lagunas, L., Choi, I. F., Kaji, T., Simpson, P., Hershey, C., Zhou, Y., Zon, L., Mercola, M. and Artinger, K. B. (2005). Zebrafish narrowminded

disrupts the transcription factor prdm1 and is required for neural crest and sensory neuron specification. *Dev. Biol.* **278**, 347-357.
Hinman, V. F., Nguyen, A. T., Cameron, R. A. and Davidson, E. H. (2003). Developmental gene regulatory network architecture across 500 million years of echinoderm evolution. *Proc. Natl. Acad. Sci. USA* **100**, 13356-13361.
Hochwagen, A. and Marais, G. A. (2010). Meiosis: a PRDM9 guide to the hotspots of recombination. *Curr. Biol.* **20**, R271-R274.
Horsley, V., O'Carroll, D., Tooze, R., Ohinata, Y., Saitou, M., Obukhanych, T., Nussenzweig, M., Tarakhovsky, A. and Fuchs, E. (2006). Blimp1 defines a progenitor population that governs cellular input to the sebaceous gland. *Cell* **126**, 597-609.
Hoyt, P. R., Bartholomew, C., Davis, A. J., Yutzey, K., Gamer, L. W., Potter, S. S., Ihle, J. N. and Mucenski, M. L. (1997). The Evi1 proto-oncogene is required at midgestation for neural, heart, and paraxial mesenchyme development. *Mech. Dev.* **65**, 55-70.
Huang, S. (1994). Blimp-1 is the murine homolog of the human transcriptional repressor PRDI-BF1. *Cell* **78**, 9.
Huang, S. (2002). Histone methyltransferases, diet nutrients and tumour suppressors. *Nat. Rev. Cancer* **2**, 469-476.
Huang, S., Shao, G. and Liu, L. (1998). The PR domain of the Rb-binding zinc finger protein RIZ1 is a protein binding interface and is related to the SET domain functioning in chromatin-mediated gene expression. *J. Biol. Chem.* **273**, 15933-15939.
Hwang, B. J., Meruelo, A. D. and Sternberg, P. W. (2007). C. elegans EVI1 proto-oncogene, EGL-43, is necessary for Notch-mediated cell fate specification and regulates cell invasion. *Development* **134**, 669-679.
Izutsu, K., Kurokawa, M., Imai, Y., Maki, K., Mitani, K. and Hirai, H. (2001). The corepressor CtBP interacts with Evi-1 to repress transforming growth factor beta signaling. *Blood* **97**, 2815-2822.
Kajimura, S., Seale, P., Tomaru, T., Erdjument-Bromage, H., Cooper, M. P., Ruas, J. L., Chin, S., Tempst, P., Lazar, M. A. and Spiegelman, B. M. (2008). Regulation of the brown and white fat gene programs through a PRDM16/CtBP transcriptional complex. *Genes Dev.* **22**, 1397-1409.
Kajimura, S., Seale, P., Kubota, K., Lunsford, E., Frangioni, J. V., Gygi, S. P. and Spiegelman, B. M. (2009). Initiation of myoblast to brown fat switch by a PRDM16-C/EBP-beta transcriptional complex. *Nature* **460**, 1154-1158.
Kallies, A., Hasbold, J., Tarlinton, D. M., Dietrich, W., Corcoran, L. M., Hodgkin, P. D. and Nutt, S. L. (2004). Plasma cell ontogeny defined by quantitative changes in blimp-1 expression. *J. Exp. Med.* **200**, 967-977.
Kallies, A., Hawkins, E. D., Belz, G. T., Metcalf, D., Hommel, M., Corcoran, L. M., Hodgkin, P. D. and Nutt, S. L. (2006). Transcriptional repressor Blimp-1 is essential for T cell homeostasis and self-tolerance. *Nat. Immunol.* **7**, 466-474.
Kallies, A., Xin, A., Belz, G. T. and Nutt, S. L. (2009). Blimp-1 transcription factor is required for the differentiation of effector CD8(+) T cells and memory responses. *Immunity* **31**, 283-295.
Kallies, A., Carotta, S., Huntington, N. D., Bernard, N. J., Tarlinton, D. M., Smyth, M. J. and Nutt, S. L. (2011). A role for Blimp1 in the transcriptional network controlling natural killer cell maturation. *Blood* **117**, 1869-1879.
Karube, K., Nakagawa, M., Tsuzuki, S., Takeuchi, I., Honma, K., Nakashima, Y., Shimizu, N., Ko, Y. H., Morishima, Y., Ohshima, K. et al. (2011). Identification of FOXO3 and PRDM1 as tumor suppressor gene candidates in NK cell neoplasms by genomic and functional analyses. *Blood* **118**, 3195-3204.
Katoh, K., Omori, Y., Onishi, A., Sato, S., Kondo, M. and Furukawa, T. (2010). Blimp1 suppresses Chx10 expression in differentiating retinal photoreceptor precursors to ensure proper photoreceptor development. *J. Neurosci.* **30**, 6515-6526.
Keller, A. D. and Maniatis, T. (1991). Identification and characterization of a novel repressor of beta-interferon gene expression. *Genes Dev.* **5**, 868-879.
Kim, K. C., Geng, L. and Huang, S. (2003). Inactivation of a histone methyltransferase by mutations in human cancers. *Cancer Res.* **63**, 7619-7623.
Kinameri, E., Inoue, T., Aruga, J., Imayoshi, I., Kageyama, R., Shimogori, T. and Moore, A. W. (2008). Prdm proto-oncogene transcription factor family expression and interaction with the Notch-Hes pathway in mouse neurogenesis. *PLoS ONE* **3**, e3859.
Komai, T., Iwanari, H., Mochizuki, Y., Hamakubo, T. and Shinkai, Y. (2009). Expression of the mouse PR domain protein Prdm8 in the developing central nervous system. *Gene Expr. Patterns* **9**, 503-514.
Koos, B., Bender, S., Witt, H., Mertsch, S., Felsberg, J., Beschorner, R., Korshunov, A., Riesmeier, B., Pfister, S., Paulus, W. et al. (2011). The transcription factor evi-1 is overexpressed, promotes proliferation, and is prognostically unfavorable in infratentorial ependymomas. *Clin. Cancer Res.* **17**, 3631-3637.
Kouzarides, T. (2007). Chromatin modifications and their function. *Cell* **128**, 693-705.
Kucuk, C., Iqbal, J., Hu, X., Gaulard, P., De Leval, L., Srivastava, G., Au, W. Y., McKeithan, T. W. and Chan, W. C. (2011). PRDM1 is a tumor suppressor gene in natural killer cell malignancies. *Proc. Natl. Acad. Sci. USA* **108**, 20119-20124.
Kuo, T. C. and Calame, K. L. (2004). B lymphocyte-induced maturation protein (Blimp)-1, IFN regulatory factor (IRF)-1, and IRF-2 can bind to the same regulatory sites. *J. Immunol.* **173**, 5556-5563.
Kurimoto, K., Yabuta, Y., Ohinata, Y., Shigeta, M., Yamanaka, K. and Saitou, M. (2008). Complex genome-wide transcription dynamics orchestrated by Blimp1 for the specification of the germ cell lineage in mice. *Genes Dev.* **22**, 1617-1635.
Kurokawa, M., Mitani, K., Irie, K., Matsuyama, T., Takahashi, T., Chiba, S., Yazaki, Y., Matsumoto, K. and Hirai, H. (1998). The oncoprotein Evi-1 represses TGF-beta signalling by inhibiting Smad3. *Nature* **394**, 92-96.
Kurokawa, M., Mitani, K., Yamagata, T., Takahashi, T., Izutsu, K., Ogawa, S., Moriguchi, T., Nishida, E., Yazaki, Y. and Hirai, H. (2000). The evi-1 oncoprotein inhibits c-Jun N-terminal kinase and prevents stress-induced cell death. *EMBO J.* **19**, 2958-2968.
Lai, E. C. and Orgogozo, V. (2004). A hidden program in Drosophila peripheral neurogenesis revealed: fundamental principles underlying sensory organ diversity. *Dev. Biol.* **269**, 1-17.
Laricchia-Robbio, L., Fazzina, R., Li, D., Rinaldi, C. R., Sinha, K. K., Chakraborty, S. and Nucifora, G. (2006). Point mutations in two EVI1 Zn fingers abolish EVI1-GATA1 interaction and allow erythroid differentiation of murine bone marrow cells. *Mol. Cell. Biol.* **26**, 7658-7666.
Laricchia-Robbio, L., Premanand, K., Rinaldi, C. R. and Nucifora, G. (2009). EVI1 Impairs myelopoiesis by deregulation of PU.1 function. *Cancer Res.* **69**, 1633-1642.
Lee, B. C. and Roy, S. (2006). Blimp-1 is an essential component of the genetic program controlling development of the pectoral limb bud. *Dev. Biol.* **300**, 623-634.
Livi, C. B. and Davidson, E. H. (2006). Expression and function of blimp1/krox, an alternatively transcribed regulatory gene of the sea urchin endomesoderm network. *Dev. Biol.* **293**, 513-525.
Livi, C. B. and Davidson, E. H. (2007). Regulation of spblimp1/krox1a, an alternatively transcribed isoform expressed in midgut and hindgut of the sea urchin gastrula. *Gene Expr. Patterns* **7**, 1-7.
Livne-Bar, I., Pacal, M., Cheung, M. C., Hankin, M., Trogadis, J., Chen, D., Dorval, K. M. and Bremner, R. (2006). Chx10 is required to block photoreceptor differentiation but is dispensable for progenitor proliferation in the postnatal retina. *Proc. Natl. Acad. Sci. USA* **103**, 4988-4993.
Ma, Z., Swigut, T., Valouev, A., Rada-Iglesias, A. and Wysocka, J. (2011). Sequence-specific regulator Prdm14 safeguards mouse ESCs from entering extraembryonic endoderm fates. *Nat. Struct. Mol. Biol.* **18**, 120-127.
Magnusdottir, E., Kalachikov, S., Mizukoshi, K., Savitsky, D., Ishida-Yamamoto, A., Panteleyev, A. A. and Calame, K. (2007). Epidermal terminal differentiation depends on B lymphocyte-induced maturation protein-1. *Proc. Natl. Acad. Sci. USA* **104**, 14988-14998.
Mandelbaum, J., Bhagat, G., Tang, H., Mo, T., Brahmachary, M., Shen, Q., Chadburn, A., Rajewsky, K., Tarakhovsky, A., Pasqualucci, L. et al. (2010). BLIMP1 is a tumor suppressor gene frequently disrupted in activated B cell-like diffuse large B cell lymphoma. *Cancer Cell* **18**, 568-579.
Margolin, J. F., Friedman, J. R., Meyer, W. K., Vissing, H., Thiesen, H. J. and Rauscher, F. J., 3rd (1994). Kruppel-associated boxes are potent transcriptional repression domains. *Proc. Natl. Acad. Sci. USA* **91**, 4509-4513.
Martins, G. and Calame, K. (2008). Regulation and functions of Blimp-1 in T and B lymphocytes. *Annu. Rev. Immunol.* **26**, 133-169.
Martins, G. A., Cimmino, L., Shapiro-Shelef, M., Szabolcs, M., Herron, A., Magnusdottir, E. and Calame, K. (2006). Transcriptional repressor Blimp-1 regulates T cell homeostasis and function. *Nat. Immunol.* **7**, 457-465.
Martins, G. A., Cimmino, L., Liao, J., Magnusdottir, E. and Calame, K. (2008). Blimp-1 directly represses Il2 and the Il2 activator Fos, attenuating T cell proliferation and survival. *J. Exp. Med.* **205**, 1959-1965.
McGilvray, R., Walker, M. and Bartholomew, C. (2007). UXT interacts with the transcriptional repressor protein EVI1 and suppresses cell transformation. *FEBS J.* **274**, 3960-3971.
Medici, N., Abbondanza, C., Nigro, V., Rossi, V., Piluso, G., Belsito, A., Gallo, L., Roscigno, A., Bontempo, P., Puca, A. A. et al. (1999). Identification of a DNA binding protein cooperating with estrogen receptor as RIZ (retinoblastoma interacting zinc finger protein). *Biochem. Biophys. Res. Commun.* **264**, 983-989.
Mercader, N., Fischer, S. and Neumann, C. J. (2006). Prdm1 acts downstream of a sequential RA, Wnt and Fgf signaling cascade during zebrafish forelimb induction. *Development* **133**, 2805-2815.
Miyauchi, Y., Ninomiya, K., Miyamoto, H., Sakamoto, A., Iwasaki, R., Hoshi, H., Miyamoto, K., Hao, W., Yoshida, S., Morioka, H. et al. (2010). The Blimp1-Bcl6 axis is critical to regulate osteoclast differentiation and bone homeostasis. *J. Exp. Med.* **207**, 751-762.
Mochizuki, N., Shimizu, S., Nagasawa, T., Tanaka, H., Taniwaki, M., Yokota, J. and Morishita, K. (2000). A novel gene, MEL1, mapped to 1p36.3 is highly homologous to the MDS1/EVI1 gene and is transcriptionally activated in t(1;3)(p36;q21)-positive leukemia cells. *Blood* **96**, 3209-3214.
Moore, A. W., Jan, L. Y. and Jan, Y. N. (2002). hamlet, a binary genetic switch between single- and multiple- dendrite neuron morphology. *Science* **297**, 1355-1358.
Moore, A. W., Roegiers, F., Jan, L. Y. and Jan, Y. N. (2004). Conversion of neurons and glia to external-cell fates in the external sensory organs of

Drosophila hamlet mutants by a cousin-cousin cell-type respecification. *Genes Dev.* **18**, 623-628.
Morishita, K. (2007). Leukemogenesis of the EVI1/MEL1 gene family. *Int. J. Hematol.* **85**, 279-286.
Moustakas, A. and Heldin, C. H. (2009). The regulation of TGFbeta signal transduction. *Development* **136**, 3699-3714.
Muncan, V., Heijmans, J., Krasinski, S. D., Buller, N. V., Wildenberg, M. E., Meisner, S., Radonjic, M., Stapleton, K. A., Lamers, W. H., Biemond, I. et al. (2011). Blimp1 regulates the transition of neonatal to adult intestinal epithelium. *Nat. Commun.* **2**, 452.
Nelson, M. D., Zhou, E., Kiontke, K., Fradin, H., Maldonado, G., Martin, D., Shah, K. and Fitch, D. H. (2011). A bow-tie genetic architecture for morphogenesis suggested by a genome-wide RNAi screen in Caenorhabditis elegans. *PLoS Genet.* **7**, e1002010.
Ng, T., Yu, F. and Roy, S. (2006). A homologue of the vertebrate SET domain and zinc finger protein Blimp-1 regulates terminal differentiation of the tracheal system in the Drosophila embryo. *Dev. Genes Evol.* **216**, 243-252.
Nikitina, N., Tong, L. and Bronner, M. E. (2011). Ancestral network module regulating prdm1 expression in the lamprey neural plate border. *Dev. Dyn.* **240**, 2265-2271.
Nishikata, I., Sasaki, H., Iga, M., Tateno, Y., Imayoshi, S., Asou, N., Nakamura, T. and Morishita, K. (2003). A novel EVI1 gene family, MEL1, lacking a PR domain (MEL1S) is expressed mainly in t(1;3)(p36;q21)-positive AML and blocks G-CSF-induced myeloid differentiation. *Blood* **102**, 3323-3332.
Nishikata, I., Nakahata, S., Saito, Y., Kaneda, K., Ichihara, E., Yamakawa, N. and Morishita, K. (2011). Sumoylation of MEL1S at lysine 568 and its interaction with CtBP facilitates its repressor activity and the blockade of G-CSF-induced myeloid differentiation. *Oncogene* **30**, 4194-4207.
Nishikawa, N., Toyota, M., Suzuki, H., Honma, T., Fujikane, T., Ohmura, T., Nishidate, T., Ohe-Toyota, M., Maruyama, R., Sonoda, T. et al. (2007). Gene amplification and overexpression of PRDM14 in breast cancers. *Cancer Res.* **67**, 9649-9657.
Nishikawa, K., Nakashima, T., Hayashi, M., Fukunaga, T., Kato, S., Kodama, T., Takahashi, S., Calame, K. and Takayanagi, H. (2010). Blimp1-mediated repression of negative regulators is required for osteoclast differentiation. *Proc. Natl. Acad. Sci. USA* **107**, 3117-3122.
Nutt, S. L., Fairfax, K. A. and Kallies, A. (2007). BLIMP1 guides the fate of effector B and T cells. *Nat. Rev. Immunol.* **7**, 923-927.
Nutt, S. L., Kallies, A. and Belz, G. T. (2008). Blimp-1 connects the intrinsic and extrinsic regulation of T cell homeostasis. *J. Clin. Immunol.* **28**, 97-106.
Ohinata, Y., Payer, B., O'Carroll, D., Ancelin, K., Ono, Y., Sano, M., Barton, S. C., Obukhanych, T., Nussenzweig, M., Tarakhovsky, A. et al. (2005). Blimp1 is a critical determinant of the germ cell lineage in mice. *Nature* **436**, 207-213.
Ohinata, Y., Ohta, H., Shigeta, M., Yamanaka, K., Wakayama, T. and Saitou, M. (2009). A signaling principle for the specification of the germ cell lineage in mice. *Cell* **137**, 571-584.
Oliver, P. L., Goodstadt, L., Bayes, J. J., Birtle, Z., Roach, K. C., Phadnis, N., Beatson, S. A., Lunter, G., Malik, H. S. and Ponting, C. P. (2009). Accelerated evolution of the Prdm9 speciation gene across diverse metazoan taxa. *PLoS Genet.* **5**, e1000753.
Palmer, S., Brouillet, J. P., Kilbey, A., Fulton, R., Walker, M., Crossley, M. and Bartholomew, C. (2001). Evi-1 transforming and repressor activities are mediated by CtBP co-repressor proteins. *J. Biol. Chem.* **276**, 25834-25848.
Park, J. A. and Kim, K. C. (2010). Expression patterns of PRDM10 during mouse embryonic development. *BMB Rep.* **43**, 29-33.
Parkinson, N., Hardisty-Hughes, R. E., Tateossian, H., Tsai, H. T., Brooker, D., Morse, S., Lalane, Z., MacKenzie, F., Fray, M., Glenister, P. et al. (2006). Mutation at the Evi1 locus in Junbo mice causes susceptibility to otitis media. *PLoS Genet.* **2**, e149.
Pengue, G., Calabro, V., Bartoli, P. C., Pagliuca, A. and Lania, L. (1994). Repression of transcriptional activity at a distance by the evolutionarily conserved KRAB domain present in a subfamily of zinc finger proteins. *Nucleic Acids Res.* **22**, 2908-2914.
Pires-daSilva, A. and Sommer, R. J. (2003). The evolution of signalling pathways in animal development. *Nat. Rev. Genet.* **4**, 39-49.
Quinlan, K. G., Nardini, M., Verger, A., Francescato, P., Yaswen, P., Corda, D., Bolognesi, M. and Crossley, M. (2006). Specific recognition of ZNF217 and other zinc finger proteins at a surface groove of C-terminal binding proteins. *Mol. Cell. Biol.* **26**, 8159-8172.
Raychaudhuri, S., Thomson, B. P., Remmers, E. F., Eyre, S., Hinks, A., Guiducci, C., Catanese, J. J., Xie, G., Stahl, E. A., Chen, R. et al. (2009). Genetic variants at CD28, PRDM1 and CD2/CD58 are associated with rheumatoid arthritis risk. *Nat. Genet.* **41**, 1313-1318.
Rea, S., Eisenhaber, F., O'Carroll, D., Strahl, B. D., Sun, Z. W., Schmid, M., Opravil, S., Mechtler, K., Ponting, C. P., Allis, C. D. et al. (2000). Regulation of chromatin structure by site-specific histone H3 methyltransferases. *Nature* **406**, 593-599.
Reid, A. G. and Nacheva, E. P. (2004). A potential role for PRDM12 in the pathogenesis of chronic myeloid leukaemia with derivative chromosome 9 deletion. *Leukemia* **18**, 178-180.
Ren, B., Chee, K. J., Kim, T. H. and Maniatis, T. (1999). PRDI-BF1/Blimp-1 repression is mediated by corepressors of the Groucho family of proteins. *Genes Dev.* **13**, 125-137.
Rimann, I. and Hajnal, A. (2007). Regulation of anchor cell invasion and uterine cell fates by the egl-43 Evi-1 proto-oncogene in Caenorhabditis elegans. *Dev. Biol.* **308**, 187-195.
Robertson, E. J., Charatsi, I., Joyner, C. J., Koonce, C. H., Morgan, M., Islam, A., Paterson, C., Lejsek, E., Arnold, S. J., Kallies, A. et al. (2007). Blimp1 regulates development of the posterior forelimb, caudal pharyngeal arches, heart and sensory vibrissae in mice. *Development* **134**, 4335-4345.
Ross, S. E., McCord, A. E., Jung, C., Atan, D., Mok, S. I., Hemberg, M., Kim, T. K., Salogiannis, J., Hu, L., Cohen, S. et al. (2012). Bhlhb5 and prdm8 form a repressor complex involved in neuronal circuit assembly. *Neuron* **73**, 292-303.
Rossi, C. C., Hernandez-Lagunas, L., Zhang, C., Choi, I. F., Kwok, L., Klymkowsky, M. and Artinger, K. B. (2008). Rohon-Beard sensory neurons are induced by BMP4 expressing non-neural ectoderm in Xenopus laevis. *Dev. Biol.* **314**, 351-361.
Roy, S. and Ng, T. (2004). Blimp-1 specifies neural crest and sensory neuron progenitors in the zebrafish embryo. *Curr. Biol.* **14**, 1772-1777.
Rutishauser, R. L., Martins, G. A., Kalachikov, S., Chandele, A., Parish, I. A., Meffre, E., Jacob, J., Calame, K. and Kaech, S. M. (2009). Transcriptional repressor Blimp-1 promotes CD8(+) T cell terminal differentiation and represses the acquisition of central memory T cell properties. *Immunity* **31**, 296-308.
Saitou, M. (2009). Germ cell specification in mice. *Curr. Opin. Genet. Dev.* **19**, 386-395.
Saitou, M., Kagiwada, S. and Kurimoto, K. (2012). Epigenetic reprogramming in mouse pre-implantation development and primordial germ cells. *Development* **139**, 15-31.
Sato, T., Goyama, S., Nitta, E., Takeshita, M., Yoshimi, M., Nakagawa, M., Kawazu, M., Ichikawa, M. and Kurokawa, M. (2008). Evi-1 promotes para-aortic splanchnopleural hematopoiesis through up-regulation of GATA-2 and repression of TGF-b signaling. *Cancer Sci.* **99**, 1407-1413.
Schneider, R., Bannister, A. J. and Kouzarides, T. (2002). Unsafe SETs: histone lysine methyltransferases and cancer. *Trends Biochem. Sci.* **27**, 396-402.
Seale, P., Kajimura, S., Yang, W., Chin, S., Rohas, L. M., Uldry, M., Tavernier, G., Langin, D. and Spiegelman, B. M. (2007). Transcriptional control of brown fat determination by PRDM16. *Cell Metab.* **6**, 38-54.
Seale, P., Bjork, B., Yang, W., Kajimura, S., Chin, S., Kuang, S., Scime, A., Devarakonda, S., Conroe, H. M., Erdjument-Bromage, H. et al. (2008). PRDM16 controls a brown fat/skeletal muscle switch. *Nature* **454**, 961-967.
Seale, P., Kajimura, S. and Spiegelman, B. M. (2009). Transcriptional control of brown adipocyte development and physiological function-of mice and men. *Genes Dev.* **23**, 788-797.
Seale, P., Conroe, H. M., Estall, J., Kajimura, S., Frontini, A., Ishibashi, J., Cohen, P., Cinti, S. and Spiegelman, B. M. (2011). Prdm16 determines the thermogenic program of subcutaneous white adipose tissue in mice. *J. Clin. Invest* **121**, 96-105.
Senyuk, V., Sinha, K. K., Li, D., Rinaldi, C. R., Yanamandra, S. and Nucifora, G. (2007). Repression of RUNX1 activity by EVI1: a new role of EVI1 in leukemogenesis. *Cancer Res.* **67**, 5658-5666.
Senyuk, V., Premanand, K., Xu, P., Qian, Z. and Nucifora, G. (2011). The oncoprotein EVI1 and the DNA methyltransferase Dnmt3 co-operate in binding and de novo methylation of target DNA. *PLoS ONE* **6**, e20793.
Shapiro-Shelef, M., Lin, K. I., McHeyzer-Williams, L. J., Liao, J., McHeyzer-Williams, M. G. and Calame, K. (2003). Blimp-1 is required for the formation of immunoglobulin secreting plasma cells and pre-plasma memory B cells. *Immunity* **19**, 607-620.
Shapiro-Shelef, M., Lin, K. I., Savitsky, D., Liao, J. and Calame, K. (2005). Blimp-1 is required for maintenance of long-lived plasma cells in the bone marrow. *J. Exp. Med.* **202**, 1471-1476.
Shimabe, M., Goyama, S., Watanabe-Okochi, N., Yoshimi, A., Ichikawa, M., Imai, Y. and Kurokawa, M. (2009). Pbx1 is a downstream target of Evi-1 in hematopoietic stem/progenitors and leukemic cells. *Oncogene* **28**, 4364-4374.
Shu, X. S., Geng, H., Li, L., Ying, J., Ma, C., Wang, Y., Poon, F. F., Wang, X., Ying, Y., Yeo, W. et al. (2011). The epigenetic modifier PRDM5 functions as a tumor suppressor through modulating WNT/beta-catenin signaling and is frequently silenced in multiple tumors. *PLoS ONE* **6**, e27346.
Smith, M. A., Maurin, M., Cho, H. I., Becknell, B., Freud, A. G., Yu, J., Wei, S., Djeu, J., Celis, E., Caligiuri, M. A. et al. (2010). PRDM1/Blimp-1 controls effector cytokine production in human NK cells. *J. Immunol.* **185**, 6058-6067.
Soler Artigas, M., Loth, D. W., Wain, L. V., Gharib, S. A., Obeidat, M., Tang, W., Zhai, G., Zhao, J. H., Smith, A. V., Huffman, J. E. et al. (2011). Genome-wide association and large-scale follow up identifies 16 new loci influencing lung function. *Nat. Genet.* **43**, 1082-1090.
Spensberger, D., Vermeulen, M., Le Guezennec, X., Beekman, R., van Hoven, A., Bindels, E., Stunnenberg, H. and Delwel, R. (2008). Myeloid transforming protein Evi1 interacts with methyl-CpG binding domain protein 3 and inhibits in vitro histone deacetylation by Mbd3/Mi-2/NuRD. *Biochemistry* **47**, 6418-6426.

Steele-Perkins, G., Fang, W., Yang, X. H., Van Gele, M., Carling, T., Gu, J., Buyse, I. M., Fletcher, J. A., Liu, J., Bronson, R. et al. (2001). Tumor formation and inactivation of RIZ1, an Rb-binding member of a nuclear protein-methyltransferase superfamily. *Genes Dev.* **15**, 2250-2262.
Su, S. T., Ying, H. Y., Chiu, Y. K., Lin, F. R., Chen, M. Y. and Lin, K. I. (2009). Involvement of histone demethylase LSD1 in Blimp-1-mediated gene repression during plasma cell differentiation. *Mol. Cell. Biol.* **29**, 1421-1431.
Sugita, M., Tanaka, N., Davidson, S., Sekiya, S., Varella-Garcia, M., West, J., Drabkin, H. A. and Gemmill, R. M. (2000). Molecular definition of a small amplification domain within 3q26 in tumors of cervix, ovary, and lung. *Cancer Genet. Cytogenet.* **117**, 9-18.
Sun, X. J., Xu, P. F., Zhou, T., Hu, M., Fu, C. T., Zhang, Y., Jin, Y., Chen, Y., Chen, S. J., Huang, Q. H. et al. (2008). Genome-wide survey and developmental expression mapping of zebrafish SET domain-containing genes. *PLoS ONE* **3**, e1499.
Takahata, M., Inoue, Y., Tsuda, H., Imoto, I., Koinuma, D., Hayashi, M., Ichikura, T., Yamori, T., Nagasaki, K., Yoshida, M. et al. (2009). SKI and MEL1 cooperate to inhibit transforming growth factor-beta signal in gastric cancer cells. *J. Biol. Chem.* **284**, 3334-3344.
Tsuneyoshi, N., Sumi, T., Onda, H., Nojima, H., Nakatsuji, N. and Suemori, H. (2008). PRDM14 suppresses expression of differentiation marker genes in human embryonic stem cells. *Biochem. Biophys. Res. Commun.* **367**, 899-905.
Turner, C. A., Jr, Mack, D. H. and Davis, M. M. (1994). Blimp-1, a novel zinc finger-containing protein that can drive the maturation of B lymphocytes into immunoglobulin-secreting cells. *Cell* **77**, 297-306.
Van Campenhout, C., Nichane, M., Antoniou, A., Pendeville, H., Bronchain, O. J., Marine, J. C., Mazabraud, A., Voz, M. L. and Bellefroid, E. J. (2006). Evi1 is specifically expressed in the distal tubule and duct of the Xenopus pronephros and plays a role in its formation. *Dev. Biol.* **294**, 203-219.
van Eeden, F. J., Granato, M., Schach, U., Brand, M., Furutani-Seiki, M., Haffter, P., Hammerschmidt, M., Heisenberg, C. P., Jiang, Y. J., Kane, D. A. et al. (1996). Mutations affecting somite formation and patterning in the zebrafish, Danio rerio. *Development* **123**, 153-164.
Vincent, S. D., Dunn, N. R., Sciammas, R., Shapiro-Shalef, M., Davis, M. M., Calame, K., Bikoff, E. K. and Robertson, E. J. (2005). The zinc finger transcriptional repressor Blimp1/Prdm1 is dispensable for early axis formation but is required for specification of primordial germ cells in the mouse. *Development* **132**, 1315-1325.
von Hofsten, J., Elworthy, S., Gilchrist, M. J., Smith, J. C., Wardle, F. C. and Ingham, P. W. (2008). Prdm1- and Sox6-mediated transcriptional repression specifies muscle fibre type in the zebrafish embryo. *EMBO Rep.* **9**, 683-689.
Warner, D. R., Horn, K. H., Mudd, L., Webb, C. L., Greene, R. M. and Pisano, M. M. (2007). PRDM16/MEL1: a novel Smad binding protein expressed in murine embryonic orofacial tissue. *Biochim. Biophys. Acta* **1773**, 814-820.
Waterhouse, A. M., Procter, J. B., Martin, D. M., Clamp, M. and Barton, G. J. (2009). Jalview Version 2 – a multiple sequence alignment editor and analysis workbench. *Bioinformatics* **25**, 1189-1191.
Wilm, T. P. and Solnica-Krezel, L. (2005). Essential roles of a zebrafish prdm1/blimp1 homolog in embryo patterning and organogenesis. *Development* **132**, 393-404.
Wu, Y., Ferguson, J. E., 3rd, Wang, H., Kelley, R., Ren, R., McDonough, H., Meeker, J., Charles, P. C., Wang, H. and Patterson, C. (2008). PRDM6 is enriched in vascular precursors during development and inhibits endothelial cell proliferation, survival, and differentiation. *J. Mol. Cell. Cardiol.* **44**, 47-58.
Wu, H., Min, J., Lunin, V. V., Antoshenko, T., Dombrovski, L., Zeng, H., Allali-Hassani, A., Campagna-Slater, V., Vedadi, M., Arrowsmith, C. H. et al. (2010). Structural biology of human H3K9 methyltransferases. *PLoS ONE* **5**, e8570.
Yamaji, M., Seki, Y., Kurimoto, K., Yabuta, Y., Yuasa, M., Shigeta, M., Yamanaka, K., Ohinata, Y. and Saitou, M. (2008). Critical function of Prdm14 for the establishment of the germ cell lineage in mice. *Nat. Genet.* **40**, 1016-1022.
Yoshimi, A., Goyama, S., Watanabe-Okochi, N., Yoshiki, Y., Nannya, Y., Nitta, E., Arai, S., Sato, T., Shimabe, M., Nakagawa, M. et al. (2011). Evi1 represses PTEN expression and activates PI3K/AKT/mTOR via interactions with polycomb proteins. *Blood* **117**, 3617-3628.
Yu, J., Angelin-Duclos, C., Greenwood, J., Liao, J. and Calame, K. (2000). Transcriptional repression by blimp-1 (PRDI-BF1) involves recruitment of histone deacetylase. *Mol. Cell. Biol.* **20**, 2592-2603.
Zhang, Y., Stehling-Sun, S., Lezon-Geyda, K., Juneja, S. C., Coillard, L., Chatterjee, G., Wuertzer, C. A., Camargo, F. and Perkins, A. S. (2011). PR domain-containing Mds1-Evi1 is critical for long-term hematopoietic stem cells function. *Blood* **118**, 3853-3861.