


# Mononuclear phagocyte regulation by the transcription factor Blimp-1 in health and disease

Isabel Ulmert,<sup>1</sup>  
Luís Henriques-Oliveira,<sup>2</sup>  
Carlos-Filipe Pereira<sup>2,3,4</sup> and  
Katharina Lahl<sup>1,5</sup> 

<sup>1</sup>Division of Biopharma, Institute for Health Technology, Technical University of Denmark (DTU), Kongens Lyngby, Denmark, <sup>2</sup>Center for Neuroscience and Cell Biology, University of Coimbra, Coimbra, Portugal, <sup>3</sup>Cell Reprogramming in Hematopoiesis and Immunity Laboratory, Lund Stem Cell Center, Molecular Medicine and Gene Therapy, Lund University, Lund, Sweden, <sup>4</sup>Wallenberg Center for Molecular Medicine, Lund University, Lund, Sweden and <sup>5</sup>Immunology Section, Lund University, Lund, Sweden

doi:10.1111/imm.13249

Received 16 June 2020; revised 24 July 2020; accepted 24 July 2020.

Correspondence: Katharina Lahl, Möllevångsvägen 6F, 222 40 Lund, Sweden. Email: klah@dtu.dk  
Senior author: Katharina Lahl

## Summary

B lymphocyte-induced maturation protein-1 (Blimp-1), the transcription factor encoded by the gene *Prdm1*, plays a number of crucial roles in the adaptive immune system, which result in the maintenance of key effector functions of B- and T-cells. Emerging clinical data, as well as mechanistic evidence from mouse studies, have additionally identified critical functions of Blimp-1 in the maintenance of immune homeostasis by the mononuclear phagocyte (MNP) system. Blimp-1 regulation of gene expression affects various aspects of MNP biology, including developmental programmes such as fate decisions of monocytes entering peripheral tissue, and functional programmes such as activation, antigen presentation and secretion of soluble inflammatory mediators. The highly tissue-, subset- and state-specific regulation of Blimp-1 expression in MNPs suggests that Blimp-1 is a dynamic regulator of immune activation, integrating environmental cues to fine-tune the function of innate cells. In this review, we will discuss the current knowledge regarding Blimp-1 regulation and function in macrophages and dendritic cells.

**Keywords:** Blimp-1; dendritic cells; immune regulation; macrophages; transcriptional regulation.

## Introduction

B-lymphocyte-induced maturation protein-1 (Blimp-1) was first described in 1991 as a potent virus-induced interferon  $\beta$  (IFN $\beta$ ) repressor in humans. The 88-kD protein containing five zinc-finger motifs was designated PRDI-BF1 (positive regulatory domain 1-binding factor 1), due to its specific binding to the PRDI element in the IFN $\beta$  promoter.<sup>1</sup> Shortly thereafter, Mark Davis and colleagues identified a transcriptional repressor in the mouse

expressed in late-stage mature B-cells and plasma cells, and named it Blimp-1.<sup>2</sup> The mouse and human versions of Blimp-1, encoded by the gene *Prdm1* (positive regulatory domain containing 1, with zinc-finger domain), are highly homologous.<sup>3</sup> Blimp-1 serves as a transcriptional and epigenetic regulator of target genes across multiple cell types. It can directly bind DNA and recruit chromatin-modifying factors associated with inhibition of gene transcription, including histone deacetylases 1 and 2 (HDAC1/2), G9a histone methyltransferase and Groucho

Abbreviations: AhR, aryl hydrocarbon receptor; ATAC-seq, assay for transposase-accessible chromatin sequencing; Bcl-6, B-cell lymphoma 6; Blimp-1, B lymphocyte-induced maturation protein-1; BMDC, bone marrow-derived dendritic cell; BMP, bone morphogenic protein; cDC, conventional dendritic cell; ChIP, chromatin immunoprecipitation; CIITA, Class II major histocompatibility complex transactivator; CST3, cystatin C; CTSS, cathepsin S; ER $\alpha$ , estrogen receptor alpha; GM-CSF, granulocyte-macrophage colony-stimulating factor; GWAS, genome-wide association studies; Hobit, homologue of Blimp-1 in T-cells; IBD, inflammatory bowel disease; IRF4, interferon regulatory factor 4; IRF8, interferon regulatory factor 8; LN, lymph node; M-CSF, macrophage colony-stimulating factor; miRNA, microRNA; MNP, mononuclear phagocyte; Mo-DC, monocyte-derived dendritic cell; Mo-Mac, monocyte-derived macrophage; NK cells, natural killer cells; NLR, NOD-like receptor; NLRP12, NLR family pyrin domain containing 12; PRDI-BF1, positive regulatory domain 1-binding factor 1; *Prdm1*, positive regulatory domain containing 1, with zinc-finger domain; PRR, pattern recognition receptor; RANK, receptor activator of nuclear factor kappa-B; RANKL, receptor activator of nuclear factor kappa-B nonbinding ligand; SLE, systemic lupus erythematosus; SNP, single nucleotide polymorphism; SOCS1, suppressor of cytokine signalling 1; TLR, Toll-like receptor; Tregs, regulatory T-cells

family proteins.<sup>4–6</sup> In this review, we will discuss the role of Blimp-1 in regulating mononuclear phagocyte (MNP) development and function in health and disease. Unless otherwise stated, we focus this review on knowledge derived from murine experiments.

### **Blimp-1 is broadly expressed, and fulfills many different roles across various cell types**

Blimp-1 is expressed across many haematopoietic and non-haematopoietic cell types, and fulfills a broad array of functions. A growing body of literature covers the role of Blimp-1 as an important regulator during early developmental processes, across vertebrate species (reviewed in detail in Ref. [7]). Murine embryos deficient for Blimp-1 die at about embryonic day 10.5 due to placental insufficiency.<sup>8,9</sup> Dose-dependent bone morphogenetic protein (BMP)/Smad-induced Blimp-1 expression is essential for primordial germ cell specification,<sup>8,10</sup> where it acts in concert with the transcription factors AP2 $\gamma$  and PRDM14.<sup>11</sup> Blimp-1 is also broadly expressed in multipotent progenitor cells during tissue development, and guides morphogenesis of various tissues, including the posterior forelimb, the caudal pharyngeal arches, the cardiac outflow tract and the sensory vibrissae.<sup>12</sup>

Blimp-1 specifically plays important roles in epithelial cell differentiation and polarization. During the suckling phase, Blimp-1 is essential in maintaining the neonatal phenotype of intestinal epithelial cells. Epithelial cell-specific Blimp-1 deficiency leads to neonatal growth retardation and mortality owing to dysregulated expression of genes associated with metabolic functions.<sup>13,14</sup> Blimp-1 also represses expression of MHC Class I pathway genes, by directly competing with interferon regulatory factor (IRF)1 in the neonatal intestinal epithelium, thereby contributing to neonatal immune tolerance.<sup>15</sup> Outside of the intestine, Blimp-1 is also important for mammary gland formation by supporting proliferation and polarization of rare luminal progenitors.<sup>16</sup> Based on experiments using *in vitro* organoids, this is in part due to elevated IFN $\lambda$  expression in the epithelial cells.<sup>17</sup> In the cancerous mammary epithelium-derived cell line MCF7, high RelB/NF $\kappa$ B levels induce Blimp-1 expression, which in turn suppresses the estrogen receptor  $\alpha$  (ER $\alpha$ ), driving elevated migratory capacity due to reduced levels of E-cadherin and  $\gamma$ -catenin.<sup>18</sup> Transforming growth factor (TGF) $\beta$ -induced epithelial-to-mesenchymal transition in breast cancer cells is also orchestrated by Blimp-1: here, Blimp-1 represses BMP-5, leading to deregulation of Snail.<sup>19</sup> In the homeostatic skin, Blimp-1 has been shown to regulate sebaceous gland homeostasis by directly repressing c-Myc in sebocyte progenitors,<sup>20</sup> and it regulates the final steps of cornification, allowing for terminal epidermal differentiation.<sup>21</sup> Thus, Blimp-1 influences steady-state and pathogenic epithelial cell development and function at

multiple levels. This heterogeneous functionality in developmentally related cell types, as depicted here across epithelial cells, suggests a highly contextual action of Blimp-1.

Despite its broad expression and diverse functional impacts within the non-haematopoietic system, Blimp-1 is still best known for its crucial role as a key regulator of plasma cell development. During the differentiation of B-cells into plasma cells, IRF4 directly induces Blimp-1 expression,<sup>22</sup> and IRF4 and Blimp-1 are together required for the induction and maintenance of functional plasma cells.<sup>23,24</sup> Blimp-1 represses B-cell lymphoma 6 (Bcl-6) and c-Myc, key factors supporting germinal centre reactions, thereby allowing for the terminal differentiation of the plasma cell.<sup>25,26</sup> Importantly, Bcl-6 can also directly repress Blimp-1, placing these two transcription factors into the centre of mature B-cell trajectory decisions, together with the Blimp-1-inducing IRF4 and the Blimp-1-repressing IRF8 as upstream regulators.<sup>27,28</sup> A series of elegant studies showed that Blimp-1 directly regulates numerous pathways to affect plasma cell fate and function. One key effect is an increase in the plasma cell's capacity to produce and secrete vast amounts of antibody (reviewed in Ref. [29]). This is facilitated by Blimp-1-mediated upregulation of *Irf1*, which activates Xbp-1 through splicing, driving the required unfolded protein response pathway.<sup>30</sup> Other aspects of plasma cell biology regulated by Blimp-1 include chemokine receptors and adhesion molecules: Blimp-1 inhibits the expression of *Cxcr5*, *Ccr7*, *S1pr1*, *Sd22*, *Itgb7* and *Sell*, strongly suggesting that it affects the positioning of plasma cells after their maturation in secondary lymphoid organs.<sup>29</sup>

Parallel to the expression pattern in B-cells, Blimp-1 also marks terminal effector T-cells, although with the exception of T follicular helper cells, which require high expression of the mutually exclusive transcription factor Bcl-6 (reviewed in Refs [31,32]). A subset of regulatory T-cells (Tregs) primarily found in mucosal tissues depends on Blimp-1 for its high expression of interleukin (IL)-10. Indeed, deficiency of Blimp-1 in the T-cell compartment leads to spontaneous colitis onset at the age of 6 weeks.<sup>33,34</sup> Mirroring the regulatory network in plasma cells, Blimp-1 expression in Tregs requires induction by IRF4.<sup>34</sup> In intestinal ROR $\gamma$ t<sup>+</sup> Tregs, however, Blimp-1 has been shown to also directly inhibit IRF4 binding to the IL-17 locus, facilitating the maintenance of the regulatory state.<sup>35</sup> Likewise, Blimp-1 can stabilize the suppressive phenotype and correct localization of follicular Tregs, allowing them to inhibit germinal centre reactions.<sup>36</sup> However, the role of Blimp-1 in follicular Tregs may be context-dependent, as Blimp-1 expression induces an ST2<sup>+</sup> (Il1r1, IL-33R) allergy-promoting phenotype of Tregs in the house dust mite model of allergic asthma.<sup>37</sup> Interestingly, Blimp-1 shows particularly high expression levels in visceral adipose tissue Tregs in male mice, where

it is induced in response to, and is essential to counteract, the low-grade inflammatory signals sent by male visceral stroma. In this context, Blimp-1 directly induces expression of the regulatory cytokine IL-10, the chemokine receptor CCR2 (essential for positioning the cells within CCL2-abundant fat), and ST2 (important for the expansion of the visceral fat Treg population).<sup>38</sup>

In CD8 effector T-cells, Blimp-1 supports terminal differentiation along with high expression of effector molecules such as granzyme B.<sup>39,40</sup> Interestingly, IL-2-induced cytotoxicity in tumour-specific cytotoxic CD4 T-cells equally depends on Blimp-1 expression for optimal granzyme B expression, suggesting that Blimp-1 is generally required for the cytotoxic programme in T-cells.<sup>41</sup> Upon chronic viral infection, Blimp-1 drives CD8 T-cell exhaustion by directly repressing expression of the IL2 $\alpha$  chain and CD27.<sup>42,43</sup> Together with the transcription factor Hobit (homologue of Blimp-1 in T-cells), Blimp-1 was shown to support the formation of tissue-resident memory cells while suppressing circulating memory cells.<sup>44</sup>

Besides its profound role within the adaptive immune system, Blimp-1 is emerging as an important rheostat for innate immune cell subset identity, activation and function. In contrast to T- and B-cells, natural killer (NK) cells constitutively express Blimp-1. Similarly to its role in T-cells, Blimp-1 expression in NK cells is required for high granzyme B expression, but not for the secretion of cytokines or for their lytic capability. In sharp contrast to its regulation in adaptive immune cells, Blimp-1 expression in NK cells is independent of IRF4 and Bcl-6. Instead, steady-state expression of Blimp-1 in NK cells depends on T-bet expression, suggesting that Blimp-1 regulation is context-dependent across lymphocyte populations.<sup>45</sup> Blimp-1 was also shown to control the function of human NK cells, where it reportedly has broader effects: Blimp-1 inhibits secretion of pro-inflammatory cytokines such as tumour necrosis factor (TNF) $\alpha$  and IFN $\gamma$ , mirroring its function in CD4 T-cells.

Blimp-1-mediated polarization and regulation of terminal effector function appears to be a common modality across numerous cell lineages. In addition to the above-mentioned lineages, genome-wide association studies (GWAS), paired with mechanistic studies using animal models, paint an emerging picture of a role for Blimp-1 in the regulation of antigen-presenting cells with implications for immune homeostasis. In the remainder of this review, we will discuss the emerging role of Blimp-1 in MNPs. These include tissue-resident macrophages, monocyte-derived macrophages (Mo-Macs) and dendritic cells (Mo-DCs), and type 1 and type 2 subsets of conventional dendritic cells (cDC1 and cDC2; nomenclature defined in Ref. [46]). cDC1 depend on IRF8 and BATF3, are characterized by their expression of XCR1, and excel at cross-presenting antigen to CD8 T-cells, endowing them with a unique function in orchestrating the immune response

towards viruses and intracellular bacteria. cDC2 on the other hand express IRF4, are characterized by their expression of CD11b and Sirp- $\alpha$ , and present antigen to CD4 T-cells with high efficacy, leading to strong immunity particularly towards extracellular bacteria (summarized in Ref. [47]).

### Blimp-1 in mononuclear phagocytic development

Paralleling its widespread expression in lymphocyte subsets, Blimp-1 shows a broad expression pattern and functionality in MNPs (Fig. 1), which to date remains relatively unexplored in its complexity. An early study identified Blimp-1 as a myeloid lineage determinant *in vitro*. Blimp-1 expression is induced upon differentiation of pro-myelocytic cells into either macrophages or granulocytes.<sup>48</sup> Accordingly, *Prdm1* transcripts were also found to be expressed in human peripheral blood monocytes and granulocytes. Overexpression of Blimp-1 in pro-monocytic cells triggered the development of a partial macrophage morphology, including cell surface expression of CD11c and CD11b.<sup>48</sup>

Upon extravasation from the bloodstream, monocytes can differentiate into Mo-Macs or Mo-DCs.<sup>46</sup> Interestingly, Blimp-1 was recently discussed to act as an important positive regulator of Mo-DC differentiation.<sup>49</sup> The study showed that human monocytes express a Mo-Mac-biased transcriptomic signature, including MafB, CD163 and MerTK, and are by default directed towards a Mo-Mac fate. However, microenvironmental cues such as IL-4, TNF $\alpha$  and aryl hydrocarbon receptor (AhR) signalling supported a switch to an IRF4-dependent Mo-DC fate, a process suppressed by silencing of *Prdm1*.<sup>49</sup> Although the exact signalling network underlying Blimp-1-induced differentiation into Mo-DCs was not studied, the correlation between expression of IRF4 and Blimp-1 in Mo-DCs is unlikely to be coincidental, as IRF4 has been shown to act as a potent transcriptional activator of *Prdm1* in other settings.<sup>22,34,35,50</sup> Similarly to the pathway of B-cell maturation,<sup>51</sup> AhR signalling induced rapid *Prdm1* expression, and AhR signalling is required for the MHCII<sup>+</sup> CD226<sup>+</sup> subset of peritoneal MNP differentiation *in vivo*.<sup>49</sup> These cells are sensitive to antimicrobial treatment and therefore assumed to depend on the microbiome. To what extent IRF4, Blimp-1 and AhR signalling converge in supporting MNP fate decisions, and which transcriptional hierarchies define this network, remain to be investigated. In addition, given the assumption that the Blimp-1-sensitive subset requires microbial signalling for differentiation and is therefore likely to be influenced by environmental changes, specific analysis of Blimp-1-influenced monocyte fate differentiation and the specific role of Blimp-1 in MNP maturation in other tissues than the peritoneal cavity is warranted. Importantly, Blimp-1 has been described as a marker for a specific macrophage population

(defined as CD11c<sup>+</sup> CD206<sup>int</sup> CD121b<sup>+</sup>) located near the microbe-exposed surface in the colon,<sup>52</sup> suggesting that Blimp-1 expression predestines cells for certain fates, but does not lock MNPs into a DC phenotype per se. Instead, the accumulating evidence suggests that environmental factors, such as microbiota-derived AhR ligands, engage a transcriptional network involving Blimp-1 to allow for functional fine-tuning of plastic lineages.

Whether Blimp-1 plays a significant role in the development and subset differentiation of cDCs is unclear. A recent study found no effect on DC differentiation *in vitro* (GM-CSF + IL-4) upon silencing of *Prdm1*.<sup>53</sup> However, the deletion of Blimp-1 in the entire haematopoietic system resulted in the selective expansion of the cDC2 subset in spleen and peripheral lymph nodes (LNs), owing to an increased number of precursors. This may suggest that Blimp-1 negatively regulates cDC2, but secondary effects caused by the absence of Blimp-1 in other haematopoietic cells were not excluded.<sup>54</sup> Using reporter mice for Blimp-1, we detected high expression specifically in small intestinal cDC2, with no detectable reporter signal in splenic cDCs.<sup>55</sup> Importantly, and seemingly contradictory to what was suggested by Chan *et al.*<sup>54</sup>, *CD11c*.cre-driven deletion of Blimp-1 caused a specific loss of CD103<sup>+</sup> CD11b<sup>+</sup> cDC2 in the small intestinal lamina propria and the corresponding migratory population in the mesenteric LNs.<sup>55</sup> cDC2 are largely found in the marginal zone/bridging channel of the spleen and in the subcapsular sinus of peripheral LNs (reviewed in Ref. [56]), which are sites of relatively high antigen exposure, suggesting that steady-state Blimp-1 expression is a consequence of microenvironmental immune signalling. Antigen exposure in the small intestine is significantly higher, and our data suggest that high Blimp-1 expression in cDC2 stabilizes rather than regulates the cDC2 population. Further research is required to explore whether immune activation regulates Blimp-1 expression in cDC2 systemically, and whether this affects cDC2 abundance. Intriguingly, a recent study reported conserved expression of Blimp-1 between mouse and man in a subset of splenic cDC2 that also expressed RORγt, again suggesting an intricate network of transcriptional regulation in cDC2 likely affected by the environment and driving the suggested heterogeneity within cDC2.<sup>57</sup> While the mechanisms underlying the loss of intestinal cDC2 in the absence of Blimp-1 are entirely unexplored, Blimp-1 is exclusively expressed in IRF4-dependent cDC2, suggesting that the mutual antagonism of IRF4 and IRF8 described for B-cells and DCs alike may also result in overlapping regulatory circuits governing Blimp-1 expression in DC subsets. Of note, expression patterns of IRF4 and Blimp-1 are conserved across murine and human intestinal cDC2.<sup>55</sup>

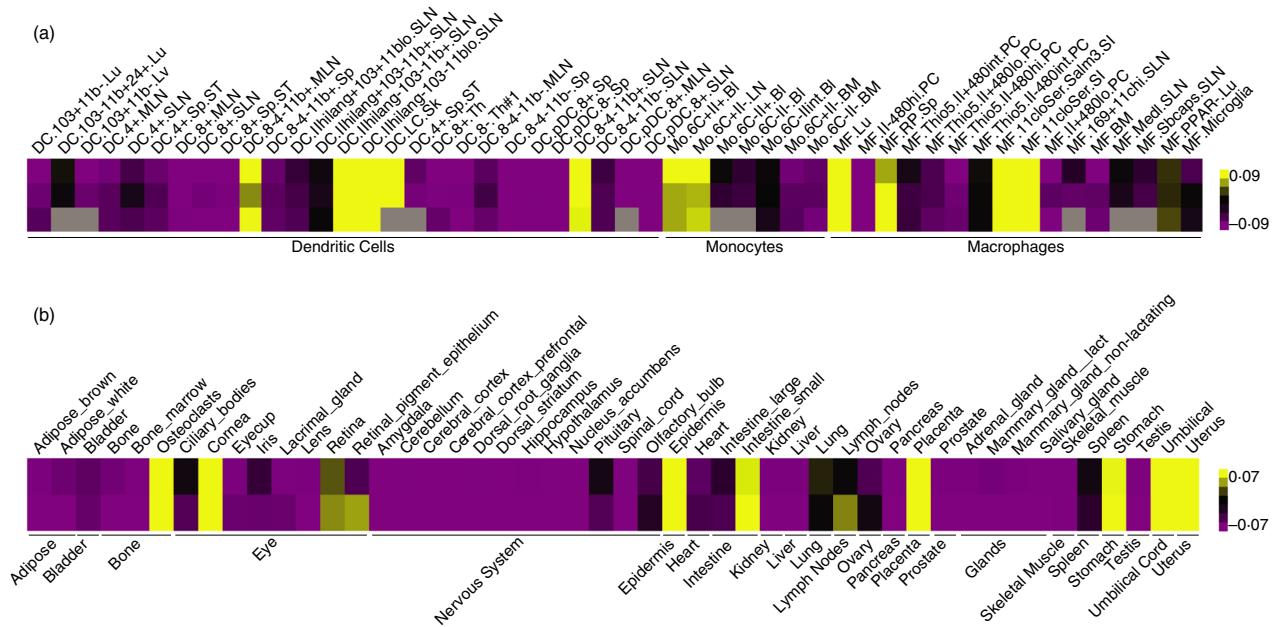
Blimp-1 also specifically supports the generation of osteoclasts, which are multi-nucleated cells derived from

the monocyte-macrophage lineage responsible for bone resorption. Osteoclasts develop from the fusion of haematopoietic myeloid precursors, and differentiate in response to receptor activator of nuclear factor kappa-B ligand (RANKL) and granulocyte-macrophage colony-stimulating factor (GM-CSF). Briefly, the interaction of RANKL with receptor activator of nuclear factor kappa-B (RANK) activates the initial expression of the master regulator NFATc1. This in turn induces expression of a gene signature essential for osteoclast differentiation and function (reviewed in Ref. [58]). The signature depends on RANK-induced Blimp-1 to inhibit the anti-osteoclastogenic genes *Bcl-6*, *IRF8* and *MafB*.<sup>59–61</sup> Blimp-1 deficiency in osteoclast progenitors consequently results in dysregulation of osteoclastogenesis, evident by aberrant bone formation *in vivo*. Intriguingly, IL-33 signalling through ST2 inhibits RANKL-induced osteoclast differentiation of macrophage colony-stimulating factor (M-CSF)- and RANKL-cultured bone marrow (BM)-cells by downregulating Blimp-1 mRNA while upregulating *IRF8* expression.<sup>62</sup> Although no direct signalling pathway has been proposed, this suggests Blimp-1 functions downstream of ST2, which is in sharp contrast to cells in the T-cell lineage mentioned above.<sup>37,38</sup>

### Regulation of Blimp-1 expression in mononuclear phagocytes

The molecular players driving Blimp-1 expression specifically in the MNP compartment have not been assessed in detail. However, some data suggest that Blimp-1 expression correlates with immune activation through pattern recognition receptors (PRRs): Toll-like receptors (TLRs) and NOD-like receptors (NLRs) engagement on MNPs induce Blimp-1 in various settings. GM-CSF-cultured bone marrow-derived dendritic cells (BMDCs) induced Blimp-1 expression upon LPS, CpG, poly(I:C) and TNFα stimulation. Pharmacological inhibition of p38, MAPK and NFκB abrogated Blimp-1 transcription in response to LPS.<sup>54</sup> Similarly, M-CSF-cultured BM-macrophages upregulated Blimp-1 transcripts rapidly upon exposure to LPS or pathogens such as *Listeria monocytogenes*, *Escherichia coli*, *Staphylococcus aureus* and Sendai virus.<sup>63,64</sup> Interestingly, Blimp-1 was often induced in two waves, one transient induction at 2 hr post-infection and a second peak induction after 24 hr, suggesting that Blimp-1 expression can result from both an immediate as well as a downstream PRR trigger.<sup>64</sup> TLR2-deficient BM-derived macrophages pretreated with an IL-1R antagonist failed to induce Blimp-1 transcription upon *L. monocytogenes* infection. Likewise, inhibition of the downstream signal transducers MyD88, MAPK and NFκB fully abrogated expression of Blimp-1. This implies that cell surface TLR2 and cytosolic PRRs regulating IL-1β production cooperate in the control of Blimp-1 transcription in BM-derived





**Figure 1.** B lymphocyte-induced maturation protein-1 (Blimp-1) expression across mononuclear phagocytes (MNPs) and murine tissues. (a) Heatmap showing Blimp-1 expression in MNP populations. Dendritic cell (DC), monocyte (Mo) and macrophage (MF) samples were extracted from the Immunological Genome Project microarray dataset (ImmGen, GSE15907). MLN, mesenteric lymph node; Lu, lung; Lv, liver; SLN, skin draining lymph node; Sp, spleen; LC, Langerhans cell; Sk, skin; Th, thymus; Bl, blood; BM, bone marrow; PC, peritoneal cavity; SI, small intestine; Medl, medullary; Sbcaps, subcapsular sinus; CNS, central nervous system; Ser, serosal; Salm, Salmonella-infected. (b) Heatmap showing Blimp-1 expression across multiple mouse tissues (GeneAtlas, MOE430), grouped according to their origin. Yellow indicates increased expression, and purple indicates decreased expression over the mean. Data from two–three replicates are shown. Unavailable data are depicted in grey. Gene expression data analysed in Gene Cluster 3-0 and displayed with Java Treeview.

macrophages upon *L. monocytogenes* infection.<sup>64</sup> Context-dependent expression of Blimp-1 in cDC2 *in vivo* in the small intestine<sup>55</sup> and inducible expression in lung cDC2 upon infection<sup>65</sup> further supports the relevance of immune activation in Blimp-1 induction.

It is intriguing to postulate that some other known activators of Blimp-1 in B- and T-cells also increase its expression in the MNP compartment. As described above, this has been shown for the AhR ligand FICZ rapidly increasing *Prdm1* expression in IRF4-dependent human monocyte cultures,<sup>49</sup> and in GM-CSF + IL-4 monocyte cultures, where IRF4 positively regulates Blimp-1 expression.<sup>50</sup> A recent study reports activation of *Prdm1* transcription by IL-10-induced STAT3-signalling in T-cells, resulting in a T<sub>H</sub>2 response upon nasal triggering in the lung, but not systemically.<sup>66</sup> Blimp-1 induction might reflect one mechanism by which STAT3 regulates MNP activation, given that both STAT3<sup>67</sup> and Blimp-1 (reviewed below, Fig. 2) negatively regulate CD11c<sup>+</sup> MNP function.

### Targets of Blimp-1 in mononuclear phagocytes

#### Blimp-1 in the regulation of inflammatory mediators

Conditional deletion of Blimp-1 in MNP subsets revealed a role for Blimp-1 in immune homeostasis and regulation

of inflammation. The identification of direct targets of Blimp-1 repression has just begun to explain this functional importance of Blimp-1 activity.

In M-CSF-cultured BM-macrophages, chromatin immunoprecipitation (ChIP) analysis identified *Ccl8* to be a direct target of Blimp-1, which was also evident *in vivo* as a steady-state increase of *Ccl8* transcripts and CCL8 protein in macrophages and in sera of *Prdm1<sup>fl/fl</sup>LysM.cre* mice.<sup>64</sup> Blimp-1 deficiency in the myeloid lineage rendered mice less susceptible to *L. monocytogenes* infection, as elevated CCL8 attracted more IL-17F-producing  $\gamma/\delta$  T-cells, which in turn increased neutrophil granulopoiesis and recruitment.<sup>64</sup>

In GM-CSF-cultured BMDs, a heterogeneous population of macrophages and DCs,<sup>68</sup> Blimp-1 directly represses *Il-6* and *Ccl2*.<sup>54</sup> Dysregulation of IL-6 expression by DCs *in vivo* was also described in the context of inflammatory bowel disease (IBD) and systemic lupus erythematosus (SLE) studies, where CD11c<sup>+</sup> cell-specific Blimp-1 deficiency led to enhanced immunopathology. In dextran sulphate sodium-induced colitis, severe disease state was specifically attributed to dysregulated IL-1 $\beta$  and IL-6 secretion by colonic CD103<sup>+</sup> DCs. Elevated pro-inflammatory cytokines resulted in the enhanced influx of neutrophils and activated macrophages into the colonic tissue. These macrophages expressed higher levels of

matrix metalloproteinases, as a direct consequence of increased IL-1 $\beta$  and IL-6 from Blimp-1-deficient colonic CD103<sup>+</sup> cDCs, leading to higher tissue destruction and exacerbated inflammation.<sup>69</sup> Blimp-1-deficient female mice spontaneously presented with an SLE-like phenotype, which could be rescued by additional knockout of IL-6, again suggesting the direct involvement of IL-6 from CD11c<sup>+</sup> cells.<sup>70</sup> In Flt3L-cultured BMDCs and splenic Blimp-1-deficient cDCs, IL-6 production was predominantly increased in female-derived cells upon LPS stimulation. This increased IL-6 production was responsible for driving the expansion of Tfh cells, resulting in the increased germinal centre formation, and ultimately in higher titres of IgG(2b) autoantibodies. The gender bias in the observed autoimmune phenotype could be explained by the role of ER $\alpha$  signalling in the positive regulation of IL-6 in BMDCs.<sup>71</sup> Blimp-1 also directly represses transcription of the microRNA (miRNA) *let-7c* in DCs.<sup>72</sup> One important *let-7c* target, suppressor of cytokine signalling 1 (SOCS1), is a regulator of several cytokines acting via the JAK/STAT3 pathway. Induction of SOCS1 was abrogated in LPS-stimulated Blimp-1-deficient splenic DCs as well as in BMDCs, resulting in high levels of IL-6, TNF $\alpha$  and IFN $\gamma$  secretion. The increased IL-6 expression, as well as decreased SOCS1, was reversed by lentiviral reconstitution of Blimp-1 in BMDCs placing Blimp-1 in a *let-7c*-SOCS1-regulated cytokine response in DCs.<sup>72</sup> Together, these data suggest that Blimp-1 can suppress IL-6 both directly and indirectly (Fig. 2).

In contrast to intestinal cDC2, lung DCs express little Blimp-1 expression at steady-state (Fig. 1a), possibly reflecting a lower basal activation level of lung immune cells. As such, stimulation of the lung environment with bacterial or viral triggers drives expression of Blimp-1 in lung cDC2.<sup>65</sup> Expression of the transcription factor correlates with a 'paralysed' state in these cells, as measured by lower cytokine production and a reduced ability to induce CD4 T-cell proliferation. Although the molecular targets of Blimp-1 were not identified in this study, cDC2 regulation by Blimp-1 likely contributes to the sepsis-induced immunosuppression observed in the lung upon pneumonia. Importantly, Blimp-1 expression levels in circulating cDC2 also positively correlated with the severity of secondary infection in patients.<sup>65</sup>

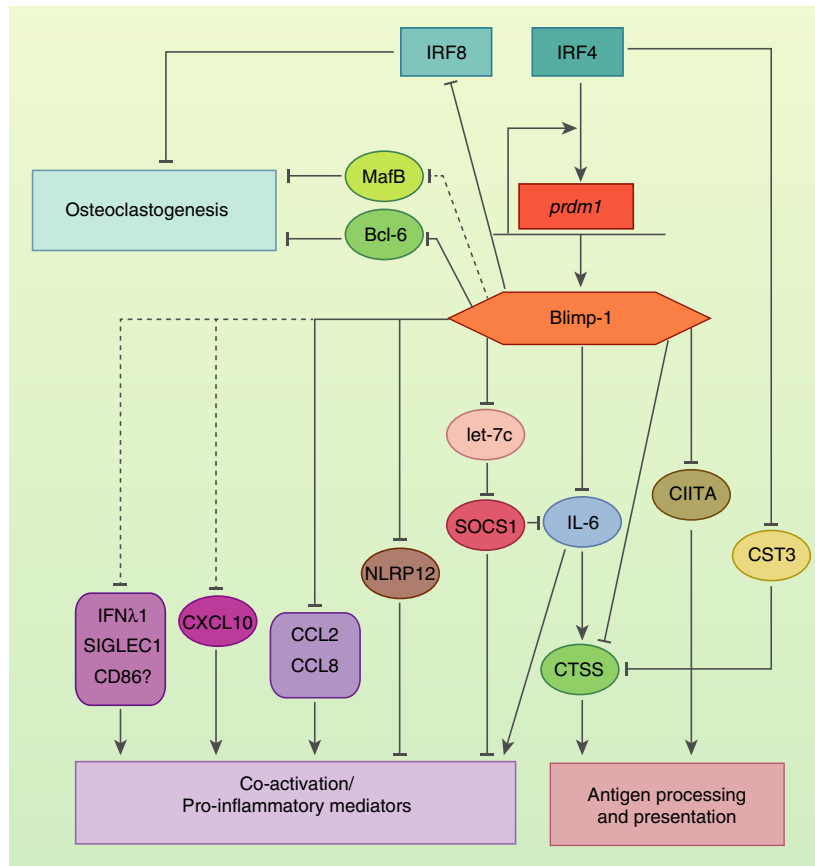
In contrast to the suggested role for Blimp-1 in negative immune regulation, Blimp-1 was also implicated in regulating immune suppression by the NLR - NLRP12 (NLR family pyrin domain containing 12).<sup>73</sup> The PRR-mediated increase in Blimp-1 expression leads to direct silencing of NLRP12 expression, enabling full activation through the NF $\kappa$ B and TNFR pathways. These findings suggest that Blimp-1 can, in the given context, remove the break from inflammatory signalling in addition to suppressing inflammation (Fig. 2). The genetic targets of Blimp-1 specifically in DCs are unknown. In-depth

phenotyping of remaining intestinal cDC2 in mice lacking Blimp-1 in DCs coupled with high-throughput single-cell gene expression and chromatin landscape assessment will reveal whether decreased abundance of cDC2 in the absence of Blimp-1 is due to activation or suppression of cDC activity, or whether Blimp-1 plays a role in DC ontogeny.

Interestingly, combined RNA- and ATAC-seq (assay for transposase-accessible chromatin sequencing) analyses identified Blimp-1 as a positive, rather than negative, upstream modulator of the IFN response during HIV infection of human GM-CSF + IL-4-cultured Mo-DCs.<sup>74</sup> ShRNA-driven inhibition of *Prdm1* resulted in defective expression of CD86 and SIGLEC1, as well as IFNL1 and CXCL10,<sup>74</sup> contrasting previous *in vitro* findings by Xiao *et al.*<sup>53</sup> The original finding that Blimp-1 potently represses IFN $\beta$  in cell lines by recruiting the G9 $\alpha$  histone methyltransferases to the IFN $\beta$  promoter,<sup>5</sup> together with these novel findings, suggests that Blimp-1-mediated positive versus negative regulation of gene expression may be highly contextual.

### Blimp-1 in antigen processing and presentation

In addition to regulation through cytokine production and costimulatory molecule expression, Blimp-1 directly interferes with antigen presentation, by influencing both antigen processing and presentation by MHC Class II. The functional importance of Blimp-1 in the regulation of antigen presentation by cDCs has received considerable attention due to its consequences for MHC-dependent systemic autoimmunity. One of the most important molecules involved in antigen presentation is cathepsin S (CTSS), which cleaves the invariant chain to permit loading into MHC Class II molecules,<sup>75</sup> and generates a pool of peptides available for presentation on MHC Class II.<sup>76,77</sup> In-depth analysis of putative causes of SLE induction in female mice harbouring Blimp-1-deficient DCs revealed heightened expression levels of CTSS in addition to IL-6.<sup>70,78</sup> Blimp-1 represses *Ctss* in cDCs directly and indirectly via the downregulation of the IL-6-STAT3 signalling pathway (Fig. 2).<sup>78,79</sup> Dysregulation of CTSS, along with CTSL expression, has been reported to modulate the pool of peptides presented to CD4 T-cells *in vitro*, either by aberrant peptide cleavage or by facilitating class II loading in a different compartment, with a potentially different peptide pool.<sup>76</sup> In fact, increased IL-6-dependent CTSS expression in Blimp-1-deficient DCs altered antigen processing and ultimately skewed differentiation of CD4 T-cells into Tfh cells bearing a diverse TCR V $\beta$  repertoire associated with autoimmunity. Adding weight to the observations in mice, patients with SLE and lupus nephritis present with increased CTSS serum levels.<sup>80</sup> GM-CSF + IL-4-cultured Mo-DCs from female SLE-risk allele carriers (rs548234) were also found to



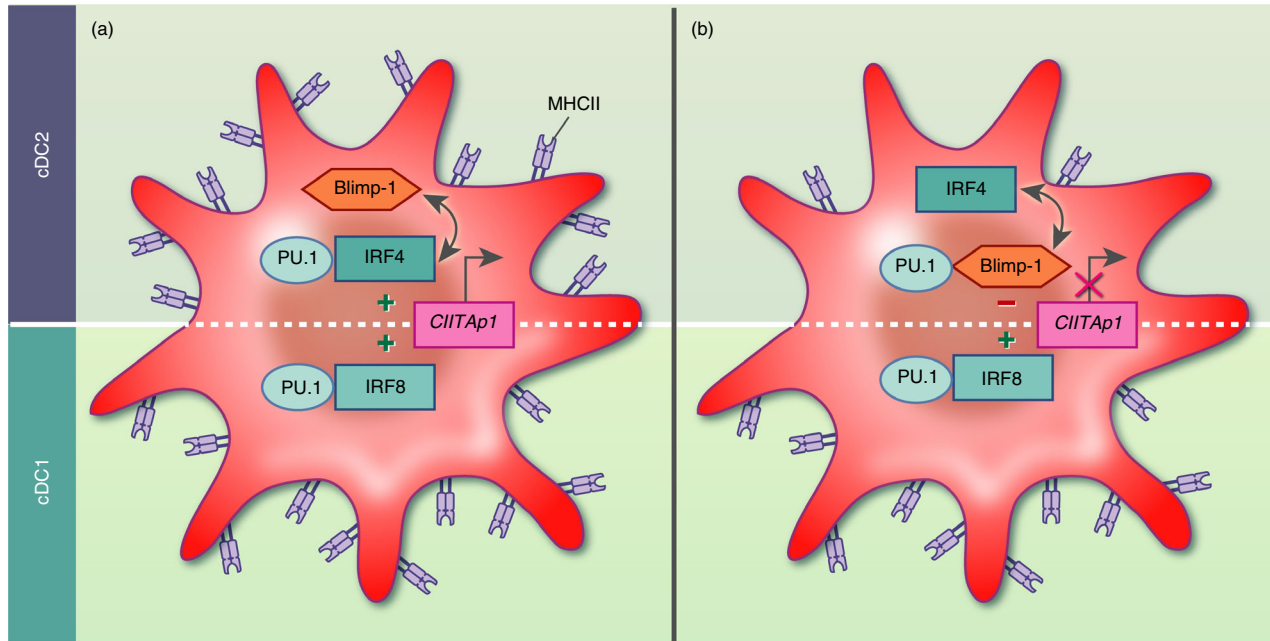
**Figure 2.** Identified B lymphocyte-induced maturation protein-1 (Blimp-1) targets in mononuclear phagocytes (MNP). Blimp-1, encoded by *Prdm1*, represses interleukin (IL)-6 directly and indirectly by regulating the levels of the microRNA (miRNA) let-7c and SOCS1.<sup>54,72</sup> It also represses cathepsin S (CTSS) directly and indirectly via repression of IL-6.<sup>78,79</sup> By downregulating CTSS and CIITA,<sup>84</sup> Blimp-1 directly influences both antigen-processing and -presentation by interfering with transcription of MHC Class II-dependent genes. Interferon regulatory factor (IRF)4 induces Blimp-1 transcription and negatively regulates CST3,<sup>50</sup> an inhibitor of CTSS, thereby allowing Blimp-1-mediated fine-tuning of the antigen-processing and -presentation machinery. Blimp-1 also directly represses CCL2 and CCL8,<sup>54,64</sup> and has been implicated to regulate CXCL10 as well as SIGLEC1, IFN $\lambda$ 1 and possibly CD86,<sup>74</sup> which are all components of a pro-inflammatory response. On the other hand, Blimp-1 can enhance activation by repressing the NF $\kappa$ B/TNFR pathway repressor NLRP12.<sup>73</sup> These regulatory mechanisms of Blimp-1 in the antigen-processing and presentation as well co-activation/pro-inflammatory mediation, are likely to be operative upon pattern recognition receptor (PRR) engagement in MNPs. Additionally, RANK-RANKL interaction (not shown) induces Blimp-1-mediated repression of anti-osteoclastogenic genes Bcl-6, IRF8 and MafB,<sup>59–61</sup> assigning a central role to Blimp-1 in osteoclast development. Solid lines indicate genes shown to be directly repressed by Blimp-1, and dotted lines indicate Blimp-1 repression by a currently unknown mechanism.

exhibit lower *Prdm1* expression, and elevated *ctss* and HLA-DR expression at steady-state.<sup>78,81</sup>

Blimp-1 also directly interferes with the expression of peptide presentation machinery by suppressing transcription of the co-activator Class II major histocompatibility complex transactivator (CIITA), which serves as a master regulator for the expression of MHC Class II genes (reviewed in Ref. [82]). Indeed, female splenic Blimp-1-deficient DCs were reported to present with constitutively increased MHC Class II expression *in vivo*.<sup>72</sup> The reduction in CIITA expression also occurs in human (GM-CSF + IL-4) Mo-DCs and murine GM-CSF-cultured BMDCs in steady-state, and upon LPS, TNF $\alpha$ , CD40L and IFN $\alpha$  stimulation, as well as infection with *Salmonella typhimurium* and Sendai virus.<sup>53,83</sup> Consistent

with the rapid induction of *Prdm1* by multiple stimuli, as discussed above, the kinetics of Blimp-1 expression in human Mo-DCs inversely correlates with CIITA expression upon DC activation, consistent with its role in B-cells during B-cell to plasma cell differentiation.<sup>84,85</sup>

CIITA expression is under the control of four independent promoters in humans (pI–pIV), and three in mice (pI, pIII and pIV). Transcription of CIITA from pI is restricted to cDCs and macrophages, while MHC Class II expression in the lymphoid lineage is primarily regulated by CIITApIII (reviewed in Ref. [86]). *In vivo* genomic footprinting analysis complemented with CHIP analysis on human LPS-stimulated Mo-DCs showed that Blimp-1 silences CIITA expression by displacing an IRF8/PU.1 complex at CIITApI during DC activation (Fig. 3). Stable



**Figure 3.** A model for B lymphocyte-induced maturation protein-1 (Blimp-1)-mediated attenuation of MHC Class II expression via silencing of the MHC Class II-transactivator CIITA in dendritic cells (DCs). (a) Interferon regulatory factor (IRF)8 and IRF4, transcription factors mutually exclusive to cDC1 and cDC2 respectively, in a complex with PU.1, facilitate promoter assembly (CIITAp1) to activate transcription of CIITA resulting in the downstream activation of MHC Class II. (b) Blimp-1 expression in cDC2 potentially silences CIITA expression by displacing IRF4. This results in the disassembly from the promoter, followed by chromatin remodelling, effectively controlling MHC Class II expression. IRF4 can directly induce Blimp-1 in response to maturation and/or activation signals.

silencing is further reinforced epigenetically by Blimp-1-mediated recruitment of the chromatin-modifying enzymes G9α and HDAC2 to the promoter, resulting in a repressed chromatin state.<sup>84</sup> Although the Ets-IRF composite element of CIITApI is able to recruit both IRF8 and IRF4 in a complex with PU.1,<sup>87</sup> Smith *et al.*<sup>84</sup> reported dominance of IRF8 in the contribution to CIITA activation. Concomitantly, B-cells utilize IRF4 and PU.1 (among others) for CIITApIII promoter activation.<sup>88,89</sup> IRF4 or IRF8 reconstitution of GM-CSF + IL-4-cultured BMDCs from IRF4-deficient DC progenitors could, however, similarly recover CIITA expression, in line with comparable expression levels of MHC Class II in both IRF8-dependent cDC1 and IRF4-dependent cDC2 in general. Because ChIP-seq analysis of LPS-treated GM-CSF-cultured BMDCs revealed that IRF4 induces *Prdm1*, transcription factors exclusive to the cDC2 subset, this argues for a more complicated incoherent feed-forward loop in transcriptional regulation of antigen presentation by cDC2, specifically.<sup>50</sup> Of note, IRF4 also negatively regulates cystatin C (CST3), which in turn inhibits the activity of CTSS,<sup>50,90</sup> suggesting an additional overlap of transcriptional targets involved in antigen presentation by IRF4 and Blimp-1 (Fig. 2). Together, these data suggest that Blimp-1 and IRF4 are part of a complicated network downstream of PRR engagement in the modulation of the MHC Class II antigen presentation pathway, with

significant relevance for the regulation of the innate-adaptive immune interface.

### Reported Blimp-1-associated polymorphisms linked to mononuclear phagocyte function

Blimp-1 has been identified as a gene contributing to IBD pathogenesis by an extensive meta-analysis of GWAS studies,<sup>91</sup> and an exome sequencing study identified variants of Blimp-1 single nucleotide polymorphisms (SNPs) that are associated with Crohn's disease. Reduced *PRDM1* expression in ileal biopsy specimens and peripheral blood mononuclear cells correlated with the Crohn's disease GWAS-associated lead risk SNP rs7746082 among the 10 identified SNPs within the *PRDM1* region.<sup>92</sup> Investigations of Blimp-1 expression in this study were narrowed to the lymphocyte lineage and, indeed, T-cell dysregulation is associated with colitis in many murine models (reviewed in Refs [34,93]). However, GWAS enriched for cell-type expression specificity of genes in IBD risk loci highlighted the strongest enrichment in DCs, suggesting that DCs are a key component of IBD pathogenesis.<sup>94</sup>

The SNPs rs548234 (Han Chinese)<sup>95</sup> and rs6568431 (European)<sup>96</sup> predispose females to the development of SLE, and are both located in the intergenic region between *PRDM1* and *ATG5*. Further analysis of the Han Chinese SNP revealed that DCs, but not B-cells, show



lower Blimp-1 expression in individuals carrying the risk allele,<sup>81</sup> while *ATG5* expression was unchanged. As expected, lower Blimp-1 expression further correlated with heightened *let-7c* miRNA and HLA-DR expression. Interestingly, the SNP induces binding of the transcriptional repressor KLF4 (kruppel-like factor 4), which is expressed at high levels in DCs, providing a mechanistic explanation for why alterations in Blimp-1 levels are specific to DCs, and cementing the finding that dysregulation of DCs, caused by low Blimp-1 expression, can lead to SLE.<sup>81</sup>

## Outlook

Taken together, a picture emerges in which Blimp-1 fulfills critical roles in the maintenance of immune homeostasis by integrating environmental triggers and imprinting context-specific function of MNPs. Despite increasing recognition of the potential of Blimp-1 as a powerful rheostat of immune activation, little is known about its *in vivo* regulation and its defined targets in the MNP system. This is mostly due to both the intrinsic heterogeneity of MNP subsets and the highly contextual expression patterns of Blimp-1. Novel technologies including single-cell RNA sequencing across tissues and immunological states will continue to pave the way for innovative approaches to modulate immune activation by harnessing Blimp-1.

## Acknowledgements

The authors thank all members of the Lahl laboratory for fruitful discussions, and Tom Fenton for critical proof-reading of the manuscript. This work was supported by a Ragnar Söderberg Foundation Fellowship in Medicine, a Lundbeck Foundation Research Fellowship, and grants from the Novo Nordisk Foundation and the Crafoord Foundation (all to KL). The Knut and Alice Wallenberg foundation, the Medical Faculty at Lund University and Region Skåne are acknowledged for generous financial support (to CFP). This work benefitted from data assembled by the ImmGen consortium.

## Conflict of interest

The authors declare no conflict of interest.

## Data Availability Statement

No new data were created for this manuscript.

## References

- 1 Keller AD, Maniatis T. Identification and characterization of a novel repressor of  $\beta$ -interferon gene expression. *Genes Dev.* 1991; 5:868–79.

- 2 Turner CA, Mack DH, Davis MM. Blimp-1, a novel zinc finger-containing protein that can drive the maturation of B lymphocytes into immunoglobulin-secreting cells. *Cell* 1994; 77:297–306.
- 3 Huang S. Blimp-1 is the murine homolog of the human transcriptional repressor PRDI-BF1. *Cell* 1994; 78:9.
- 4 Ren B, Chee KJ, Kim TH, Maniatis T. PRDI-BF1/Blimp-1 repression is mediated by corepressors of the Groucho family of proteins. *Genes Dev.* 1999; 13(1):125–37.
- 5 Györy I, Wu J, Fejér G, Seto E, Wright KL. PRDI-BF1 recruits the histone H3 methyltransferase G9a in transcriptional silencing. *Nat Immunol* 2004; 5:299–308.
- 6 Yu J, Angelin-Duclos C, Greenwood J, Liao J, Calame K. Transcriptional repression by Blimp-1 (PRDI-BF1) involves recruitment of histone deacetylase. *Mol Cell Biol.* 2000; 20(7):2592–603.
- 7 Bikoff EK, Morgan MA, Robertson EJ. An expanding job description for Blimp-1/PRDM1. *Curr Opin Genet Dev* 2009; 19:379–85.
- 8 Vincent SD, Dunn NR, Sciammas R, Shapiro-Shalef M, Davis MM, Calame K, et al. 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* 2005; 132:1315–25.
- 9 Mould A, Morgan MAJ, Li L, Bikoff EK, Robertson EJ. Blimp1/Prdm1 governs terminal differentiation of endovascular trophoblast giant cells and defines multipotent progenitors in the developing placenta. *Genes Dev* 2012; 26:2063–74.
- 10 Ohinata Y, Payer B, O'Carroll D, Ancelin K, Ono Y, Sano M, et al. Blimp1 is a critical determinant of the germ cell lineage in mice. *Nature* 2005; 436:207–13.
- 11 Magnúsdóttir E, Dietmann S, Murakami K, Günesdogan U, Tang F, Bao S, et al. A tripartite transcription factor network regulates primordial germ cell specification in mice. *Nat Cell Biol* 2013; 15(8):905–15.
- 12 Robertson EJ, Charatsi I, Joyner CJ, Koonce CH, Morgan MA, Islam A, et al. Blimp1 regulates development of the posterior forelimb, caudal pharyngeal arches, heart and sensory vibrissae in mice. *Development* 2007; 134:4335–45.
- 13 Muncan V, Heijmans J, Krasinski SD, Büller NV, Wildenberg ME, Meisner S, et al. Blimp1 regulates the transition of neonatal to adult intestinal epithelium. *Nat Commun* 2011; 2: 452
- 14 Harper J, Mould A, Andrews RM, Bikoff EK, Robertson EJ. The transcriptional repressor Blimp1/Prdm1 regulates postnatal reprogramming of intestinal enterocytes. *Proc Natl Acad Sci* 2011; 108:10585–90.
- 15 Mould AW, Morgan MAJ, Nelson AC, Bikoff EK, Robertson EJ. Blimp1/Prdm1 functions in opposition to Irf1 to maintain neonatal tolerance during postnatal intestinal maturation. *PLoS Genet.* 2015; 11:1–22.
- 16 Ahmed MI, Elias S, Mould AW, Bikoff EK, Robertson EJ. The transcriptional repressor Blimp1 is expressed in rare luminal progenitors and is essential for mammary gland development. *Development* 2016; 143:1663–73.
- 17 Elias S, Robertson EJ, Bikoff EK, Mould AW. Blimp-1/PRDM1 is a critical regulator of Type III Interferon responses in mammary epithelial cells. *Sci Rep* 2018; 8:1–11.
- 18 Wang X, Belguise K, O'Neill CF, Sanchez-Morgan N, Romagnoli M, Eddy SF, et al. RelB NF- $\kappa$ B represses Estrogen receptor expression via induction of the Zinc finger protein Blimp1. *Mol Cell Biol* 2009; 29:3832–44.
- 19 Romagnoli M, Belguise K, Yu Z, Wang X, Landesman-Bollag E, Seldin DC, et al. Epithelial-to-mesenchymal transition induced by TGF- $\beta$ 1 is mediated by blimp-1-dependent repression of BMP-5. *Cancer Res* 2012; 72:6268–78.
- 20 Horsley V, O'Carroll D, Tooze R, Ohinata Y, Saitou M, Obukhanych T, et al. Blimp1 defines a progenitor population that governs cellular input to the sebaceous gland. *Cell* 2006; 126:597–609.
- 21 Magnúsdóttir E, Kalachikov S, Mizukoshi K, Savitsky D, Ishida-Yamamoto A, Pan-teleyev AA, et al. Epidermal terminal differentiation depends on B lymphocyte-induced maturation protein-1. *Proc Natl Acad Sci USA* 2007; 104:14988–93.
- 22 Sciammas R, Shaffer AL, Schatz JH, Zhao H, Staudt LM, Singh H. Graded expression of interferon regulatory factor-4 coordinates isotype switching with plasma cell differentiation. *Immunity* 2006; 25(2):225–36.
- 23 Shapiro-Shalef M, Lin KI, McHeyzer-Williams LJ, Liao J, McHeyzer-Williams MG, Calame K. Blimp-1 is required for the formation of immunoglobulin secreting plasma cells and pre-plasma memory B cells. *Immunity* 2003; 19:607–20.
- 24 Klein U, Casola S, Cattoretti G, Shen Q, Lia M, Mo T, et al. Transcription factor IRF4 controls plasma cell differentiation and class-switch recombination. *Nat Immunol* 2006; 7:773–82.
- 25 Shaffer AL, Lin KI, Kuo TC, Yu X, Hurt EM, Rosenwald A, et al. Blimp-1 orchestrates plasma cell differentiation by extinguishing the mature B cell gene expression program. *Immunity* 2002; 17:51–62.
- 26 Lin Y, Wong K, Calame K. Repression of c-myc transcription by Blimp-1, an inducer of terminal B cell differentiation. *Science* 1997; 276:596–9.
- 27 Tunyaplin C, Shaffer AL, Angelin-Duclos CD, Yu X, Staudt LM, Calame KL. Direct repression of prdm1 by Bcl-6 inhibits plasmacytic differentiation. *J Immunol.* 2004; 173:1158–65.
- 28 Xu H, Chaudhri VK, Wu Z, Biliouris K, Dienger-Stambaugh K, Rochman Y, et al. Regulation of bifurcating B cell trajectories by mutual antagonism between transcription factors IRF4 and IRF8. *Nat Immunol* 2015; 16:1274–81.

- 29 Tellier J, Nutt SL. Plasma cells: The programming of an antibody-secreting machine. *Eur J Immunol* 2019; **49**:30–7.
- 30 Tellier J, Shi W, Minnich M, Liao Y, Crawford S, Smyth GK, et al. Blimp-1 controls plasma cell function through the regulation of immunoglobulin secretion and the unfolded protein response. *Nat Immunol* 2016; **17**:323–30.
- 31 Fu SH, Yeh LT, Chu CC, Yen BLJ, Sytwu HK. New insights into Blimp-1 in T lymphocytes: A divergent regulator of cell destiny and effector function. *J Biomed Sci* 2017; **24**:1–17.
- 32 Crotty S, Johnston RJ, Schoenberger SP. Effectors and memories: Bcl-6 and Blimp-1 in T and B lymphocyte differentiation. *Nat Immunol* 2010; **11**:114–20.
- 33 Martins GA, Cimmino L, Shapiro-Shelef M, Szabolcs M, Herron A, Magnusdottir E, et al. Transcriptional repressor Blimp-1 regulates T cell homeostasis and function. *Nat Immunol* 2006; **7**:457–65.
- 34 Cretney E, Xin A, Shi W, Minnich M, Masson F, Miasari M, et al. The transcription factors Blimp-1 and IRF4 jointly control the differentiation and function of effector regulatory T cells. *Nat Immunol* 2011; **12**:304–12.
- 35 Ogawa C, Bankoti R, Nguyen T, Hassanzadeh-Kiabi N, Nadeau S, Porritt RA, et al. Blimp-1 functions as a molecular switch to prevent inflammatory activity in Foxp3+RORγt+ regulatory T cells. *Cell Rep* 2018; **25**:19–28.e5.
- 36 Wang L, Shen E, Luo L, Rabe H, Wang Q, Yin J, et al. Control of germinal center localization and lineage stability of follicular regulatory T cells by the Blimp1 transcription factor. *Cell Rep* 2019; **29**:1848–61.
- 37 Koh B, Ulrich BJ, Nelson AS, Panangipalli G, Kharwadkar R, Wu W, et al. Bcl6 and Blimp1 reciprocally regulate ST2+ Treg-cell development in the context of allergic airway inflammation. *Journal of Allergy and Clinical Immunology* 2020. <https://doi.org/10.1016/j.jaci.2020.03.002>. [Epub ahead of print].
- 38 Vasanthakumar A, Chisanga D, Blume J, Gloury R, Britt K, Henstridge DC, et al. Sex-specific adipose tissue imprinting of regulatory T cells. *Nature* 2020; **579**:581–5.
- 39 Kallies A, Xin A, Belz GT, Nutt SL. Blimp-1 Transcription factor is required for the differentiation of effector CD8+ T cells and memory responses. *Immunity* 2009; **31**:283–95.
- 40 Rutishauser RL, Martins GA, Kalachikov S, Chandele A, Parish IA, Meffre E, et al. Transcriptional repressor Blimp-1 promotes CD8(+) T cell terminal differentiation and represses the acquisition of central memory T cell properties. *Immunity* 2009; **31**:296–308.
- 41 Ślędzka A, Vila de Mucha M, Bergerhoff K, Hotblack A, Demane DF, Ghorani E, et al. Regulatory T cells restrain interleukin-2- and Blimp-1-dependent acquisition of cytotoxic function by CD4+ T cells. *Immunity* 2020; **52**:151–66.
- 42 Shin H, Blackburn SD, Intlekofer AM, Kao C, Angelosanto JM, Reiner SL, et al. A role for the transcriptional repressor Blimp-1 in CD8+ T cell exhaustion during chronic viral infection. *Immunity* 2009; **31**:309–20.
- 43 Shin HM, Kapoor VN, Guan T, Kaech SM, Welsh RM, Berg LJ. epigenetic modifications induced by Blimp-1 regulate CD8+ T cell memory progression during acute virus infection. *Immunity* 2013; **39**:661–75.
- 44 Mackay LK, Minnich M, Kragten NAM, Liao Y, Nota B, Seillet C, et al. Hobit and Blimp1 instruct a universal transcriptional program of tissue residency in lymphocytes. *Science* 2016; **352**:459–63.
- 45 Kallies A, Carotta S, Huntington ND, Bernard NJ, Tarlinton DM, Smyth MJ, et al. A role for Blimp1 in the transcriptional network controlling natural killer cell maturation. *Blood* 2011; **117**:1869–79.
- 46 Guillemins M, Ginhoux F, Jakubczik C, Naik SH, Onai N, Schraml BU, et al. Dendritic cells, monocytes and macrophages: a unified nomenclature based on ontogeny. *Nat Rev Immunol* 2014; **14**:571–8.
- 47 Nutt SL, Chopin M. Transcriptional networks driving dendritic cell differentiation and function. *Immunity* 2020; **52**:942–56.
- 48 Chang DH, Angelin-Duclos C, Calame K. BLIMP-1: Trigger for differentiation of myeloid lineage. *Nat Immunol* 2000; **1**:169–76.
- 49 Goudot C, Coillard A, Villani AC, Gueguen P, Cros A, Sarkizova S, et al. Aryl hydrocarbon receptor controls monocyte differentiation into dendritic cells versus macrophages. *Immunity* 2017; **47**:582–96.
- 50 Vander Lugt B, Khan A, Hackney JA, et al. Transcriptional programming of dendritic cells for enhanced MHC class II antigen presentation. *Nat Immunol* 2014; **15**:161–7.
- 51 De Abrew KN, Kaminski NE, Thomas RS. An integrated genomic analysis of aryl hydrocarbon receptor-mediated inhibition of B-cell differentiation. *Toxicol Sci* 2010; **118**:454–69.
- 52 Kang B, Alvarado LJ, Kim T, Lehmann ML, Cho H, He J, et al. Commensal microbiota drive the functional diversification of colon macrophages. *Mucosal Immunol* 2020; **13**:216–29.
- 53 Xiao J, Zhang J, Li X, Dai X, Wang J, He Y, et al. Downregulation of Blimp1 inhibits the maturation of bone marrow-derived dendritic cells. *Int J Mol Med* 2019; **43**:1094–104.
- 54 Chan Y-HH, Chiang M-F, Tsai Y-CC, Su S-TT, Chen M-HH, Hou M-SS, et al. Absence of the transcriptional repressor Blimp-1 in hematopoietic lineages reveals its role in dendritic cell homeostatic development and function. *J Immunol* 2009; **183**:7039–46.
- 55 Watchmaker PB, Lahl K, Lee M, Baumjohann D, Morton J, Kim SJ, et al. Comparative transcriptional and functional profiling defines conserved programs of intestinal DC differentiation in humans and mice. *Nat Immunol* 2014; **15**:98–108.
- 56 Eisenbarth SC. Dendritic cell subsets in T cell programming: location dictates function. *Nat Rev Immunol* 2019; **19**:89–103.
- 57 Brown CC, Gudjonson H, Pritykin Y, Deep D, Lavallée VP, Mendoza A, et al. Transcriptional basis of mouse and human dendritic cell heterogeneity. *Cell* 2019; **179**:846–63.
- 58 Takayanagi H. Osteoimmunology: Shared mechanisms and crosstalk between the immune and bone systems. *Nat Rev Immunol* 2007; **7**:292–304.
- 59 Miyauchi Y, Ninomiya K, Miyamoto H, Sakamoto A, Iwasaki R, Hoshi H, et al. The Blimp1-Bcl6 axis is critical to regulate osteoclast differentiation and bone homeostasis. *J Exp Med* 2010; **207**:751–62.
- 60 Nishikawa K, Nakashima T, Hayashi M, Fukunaga T, Kato S, Kodama T, et al. Blimp1-mediated repression of negative regulators is required for osteoclast differentiation. *Proc Natl Acad Sci USA* 2010; **107**:3117–22.
- 61 Zhao B, Takami M, Yamada A, Wang X, Koga T, Hu X, et al. Interferon regulatory factor-8 regulates bone metabolism by suppressing osteoclastogenesis. *Nat Med* 2009; **15**:1066–71.
- 62 Kiyomiya H, Ariyoshi W, Okinaga T, Kaneuji T, Mitsugi S, Sakurai T, et al. IL-33 inhibits RANKL-induced osteoclast formation through the regulation of Blimp-1 and IRF-8 expression. *Biochem Biophys Res Comm* 2015; **460**:320–6.
- 63 Severa M, Coccia EM, Fitzgerald KA. Toll-like receptor-dependent and -independent Viperin gene expression and counter-regulation by PRDI-binding factor-1/BLIMP1. *J Biol Chem* 2006; **281**:26188–95.
- 64 Severa M, Islam S, Waggoner SN, et al. The transcriptional repressor BLIMP1 curbs host defenses by suppressing expression of the chemokine CCL8. *J Immunol* 2014; **192**:2291–304.
- 65 Roquilly A, McWilliam HEG, Jacqueline C, Tian Z, Cinotti R, Rimbart M, et al. Local modulation of antigen-presenting cell development after resolution of pneumonia induces long-term susceptibility to secondary infections. *Immunity* 2017; **47**:135–147.
- 66 He K, Hettinga A, Laxman Kale S, Hu S, Xie MM, Dent AL, et al. Blimp-1 is essential for Th2 cell development and allergic asthma. *J Exp Med* 2020; **217**:e20190742.
- 67 Melillo JA, Song L, Bhagat G, Blazquez AB, Plumlee CR, Lee C, et al. Dendritic cell (DC)-specific targeting reveals Stat3 as a negative regulator of DC function. *J Immunol* 2010; **184**:2638–45.
- 68 Helft J, Böttcher J, Chakravarty P, Zelenay S, Huotari J, Schraml BU, et al. GM-CSF mouse bone marrow cultures comprise a heterogeneous population of CD11c+MHCII+ macrophages and dendritic cells. *Immunity* 2015; **42**:1197–211.
- 69 Kim SJ, Goldstein J, Dorso K, et al. Expression of Blimp-1 in dendritic cells modulates the innate inflammatory response in dextran sodium sulfate-induced colitis. *Mol Med* 2014; **20**:707–19.
- 70 Kim SJ, Zou YR, Goldstein J, Reizis B, Diamond B. Tolerogenic function of Blimp-1 in dendritic cells. *J Exp Med* 2011; **208**:2193–9.
- 71 Seillet C, Rouquié N, Foulon E, Douin-Echinard V, Krust A, Chambon P, et al. Estradiol promotes functional responses in inflammatory and steady-state dendritic cells through differential requirement for activation function-1 of estrogen receptor  $\alpha$ . *J Immunol* 2013; **190**:5459–70.
- 72 Kim SJ, Gregersen PK, Diamond B. Regulation of dendritic cell activation by microRNA let-7c and BLIMP1. *J Clin Invest* 2013; **123**:823–33.
- 73 Lord CA, Savitsky D, Sitcheran R, Calame K, Wright JR, Ting JP-Y, et al. Blimp-1/PRDM1 mediates transcriptional suppression of the NLR gene NLRP12/Monarch-1. *J Immunol* 2009; **182**:2948–58.
- 74 Johnson JS, De Veaux N, Rives AW, Lahaye X, Lucas SY, Perot BP, et al. A comprehensive map of the monocyte-derived dendritic cell transcriptional network engaged upon innate sensing of HIV. *Cell Rep* 2020; **30**(3):914–31.
- 75 Shi GP, Webb AC, Foster KE, Knoll JHM, Lemere CA, Munger JS, et al. Human cathepsin S: Chromosomal localization, gene structure, and tissue distribution. *J Biol Chem* 1994; **269**:11530–6.
- 76 Hsieh C-S, deRoos P, Honey K, Beers C, Rudensky AY. A Role for Cathepsin L and Cathepsin S in peptide generation for MHC class II presentation. *J Immunol* 2002; **168**:2618–25.
- 77 Beers C, Burich A, Kleijmeer MJ, Griffith JM, Wong P, Rudensky AY. Cathepsin S Controls MHC class II-mediated antigen presentation by epithelial cells in vivo. *J Immunol* 2005; **174**:1205–12.
- 78 Kim SJ, Schätzle S, Ahmed SS, Haap W, Jang SH, Gregersen PK, et al. Increased cathepsin S in Prdm1-/- dendritic cells alters the T FH cell repertoire and contributes to lupus. *Nat Immunol* 2017; **18**:1016–24.
- 79 Kitamura H, Kamon H, Sawa SI, Park SJ, Katunuma N, Ishihara K, et al. IL-6-STAT3 controls intracellular MHC class II  $\alpha\beta$  dimer level through cathepsin S activity in Dendritic Cells. *Immunity* 2005; **23**:491–502.
- 80 Tato M, Kumar SV, Liu Y, Mulay SR, Moll S, Popper B, et al. Cathepsin S inhibition combines control of systemic and peripheral pathomechanisms of autoimmune tissue injury. *Sci Rep* 2017; **7**:1–15.

- 81 Jang SH, Chen H, Gregersen PK, Diamond B, Kim SJ. Kruppel-like factor4 regulates PRDM1 expression through binding to an autoimmune risk allele. *JCI Insight* 2017; **2**:e89569.
- 82 Wright KL, Ting JPY. Epigenetic regulation of MHC-II and CIITA genes. *Trends Immunol* 2006; **27**:405–12.
- 83 Landmann S, Mühlethaler-Mottet A, Bernasconi L, Suter T, Waldburger JM, Masternak K, *et al.* Maturation of dendritic cells is accompanied by rapid transcriptional silencing of class II transactivator (CIITA) expression. *J Exp Med* 2001; **194**:379–91.
- 84 Smith MA, Wright G, Wu J, Taylor P, Ozato K, Chen X, *et al.* Positive regulatory domain I (PRDM1) and IRF8/PU.1 counter-regulate MHC class II transactivator (CIITA) expression during dendritic cell maturation. *J Biol Chem* 2011; **286**:7893–904.
- 85 Piskurich JF, Lin KI, Lin Y, Wang Y, Ting JPY, Calame K. BLIMP-1 mediates extinction of major histocompatibility class II transactivator expression in plasma cells. *Nat Immunol* 2000; **1**:526–32.
- 86 LeibundGut-Landmann S, Waldburger JM, Krawczyk M, Otten LA, Suter T, Fontana A, *et al.* Specificity and expression of CIITA, the master regulator of MHC class II genes. *Eur J Immunol* 2004; **34**:1513–25.
- 87 Marecki S, Fenton MJ. PU.1/interferon regulatory factor interactions: Mechanisms of transcriptional regulation. *Cell Biochem Biophys*. 2000; **33**:127–48.
- 88 Yoon H, Boss JM. PU.1 binds to a distal regulatory element that is necessary for B cell-specific expression of CIITA. *J Immunol*. 2010; **184**:5018–28.
- 89 Van Der Stoep N, Quinten E, Rezende MM, Van Den Elsen PJ. E47, IRF-4, and PU.1 synergize to induce B-cell-specific activation of the class II transactivator promoter III (CIITA-PIII). *Blood* 2004; **104**:2849–57.
- 90 Pierre P, Mellman I. Developmental regulation of invariant chain proteolysis controls MHC class II trafficking in mouse dendritic cells. *Cell* 1998; **93**:1135–45.
- 91 Franke A, McGovern DPB, Barrett JC, Wang K, Radford-Smith GL, Ahmad T, *et al.* Genome-wide meta-analysis increases to 71 the number of confirmed Crohn's disease susceptibility loci. *Nat Genet* 2010; **42**:1118–25.
- 92 Ellinghaus D, Zhang H, Zeissig S, Lipinski S, Till A, Jiang T, *et al.* Association between variants of PRDM1 and NDP52 and Crohn's disease, based on exome sequencing and functional studies. *Gastroenterology* 2013; **145**:339–47.
- 93 Martins G, Calame K. Regulation and functions of Blimp-1 in T and B lymphocytes. *Annu Rev Immunol*. 2008; **26**:133–69.
- 94 Jostins L, Ripke S, Weersma RK, Duerr RH, McGovern DP, Hui KY, *et al.* Host–microbe interactions have shaped the genetic architecture of inflammatory bowel disease. *Nature* 2012; **491**:119–24.
- 95 Han JW, Zheng HF, Cui Y, Sun LD, Ye DQ, Hu Z, *et al.* Genome-wide association study in a Chinese Han population identifies nine new susceptibility loci for systemic lupus erythematosus. *Nat Genet* 2009; **41**:1234–7.
- 96 Gateva V, Sandling JK, Hom G, Taylor KE, Chung SA, Sun X, *et al.* A large-scale replication study identifies TNIP1, PRDM1, JAZF1, UHRF1BP1 and IL10 as risk loci for systemic lupus erythematosus. *Nat Genet*. 2009; **41**:1228–33.