



Revealing eukaryotic histone-modifying mechanisms through bacterial infection

Wenyang Dong^{1,2} · Melanie Anne Hamon¹

Received: 13 November 2019 / Accepted: 27 December 2019 / Published online: 4 February 2020
© Springer-Verlag GmbH Germany, part of Springer Nature 2020

Abstract

In the long co-evolution of host-pathogen interaction, bacteria have developed sophisticated strategies to manipulate host cell mechanisms and reprogram host transcription. Targeting chromatin, mainly through post-translational modification (PTM) of histone proteins, is one strategy that has been revealed over the last decade. Indeed, histone modifications play a crucial role in regulating transcription during cell type and stimulus specific responses, making them good targets during infection. Therefore, the study of host-pathogen interactions provides breakthroughs in understanding virulence mechanisms, but also in host cell mechanisms. Although chromatin is regulated by DNA methylation, noncoding RNAs, and post-translational modifications of histones, most studies have concentrated on bacteria-induced histone modifications, which will be the focus of this review. We will discuss the different mechanisms used by bacteria to induce histone PTMs, whether it is through direct targeting of pathogen effector enzymes, or indirectly through modulation of cellular signaling cascade. We will summarize the concepts we learned in cell biology from exploring bacteria-triggered histone modifications, by focusing on the signaling cascades modified by bacteria, bacterial mimics of eukaryotic enzymes, and the novel histone marks imposed upon infection.

Keywords Bacterial infection · Pathogenesis · Histone modifications · Epigenomics · Chromatin

Introduction

To achieve confinement into the tight space of cell nucleus, the genetic material of eukaryotic cells is packaged in tight association with histone proteins. This highly architected DNA-protein structure is defined as chromatin, which is made up of repeating building blocks called nucleosomes. Inside the nucleosomes, two copies of each of the core histones H2A, H2B, H3, and H4 form an octamer, around which approximately 147 base pairs of DNA are wound [1]. Outside the nucleosomes, linker histone H1 (or its isoforms) associates with this unit, keeping in place the DNA that has wrapped around the nucleosome, and forming nucleosomal arrays along the genome [2].

The state of compaction of chromatin is highly dynamic but tightly regulated. In nuclear processes requiring access to DNA, such as transcription, replication, recombination, and DNA repair, several regulatory mechanisms are involved in altering nucleosome architecture to open or close regions of the genome for appropriate outcomes. One important mechanism is chromatin remodeling, during which nucleosomes undergo recurrent structural rearrangements through DNA unwrapping and rewrapping and histone core disassembly and assembly. ATP-dependent chromatin-remodeling complexes, such as SWI/SNF and ISWI, use the energy of ATP hydrolysis to regulate chromatin architecture by repositioning or restructuring nucleosomes [3]. The second mechanism is through covalent post-translational modifications (PTMs) of chromatin components. These epigenetic marks, including the methylation on DNA and a wide array of PTMs on histones, physically regulate the accessibility of the transcriptional machinery to certain regions of genome, making nucleosomal DNA more or less permissive for transcription [4]. Post-translational modifying mechanisms frequently cross-talk with ATP-dependent chromatin-remodeling complexes and function together to regulate chromatin structure [4]. Other players, such as noncoding RNAs and miRNAs, are also involved in

This article is a contribution to the special issue on Infection-induced epigenetic changes and the pathogenesis of diseases - Guest Editor: Nicole Fischer

✉ Melanie Anne Hamon
melanie.hamon@pasteur.fr

¹ G5 Chromatine et Infection, Institut Pasteur, 75015 Paris, France

² Université de Paris, Sorbonne Paris Cité, Paris, France

regulation of chromatin structure and function, but for reasons of space, they will not be discussed in this review. It is important to note that the combination of histones and associated proteins is termed the epigenome, which is as important to the regulation of gene expression as the core genome itself, composed solely of DNA.

Histones, especially N-terminal histone tails, are subject to a variety of PTMs, including acetylation, methylation, phosphorylation, ubiquitination, sumoylation, carbonylation, and glycosylation [5, 6]. The presence and maintenance of such marks are dynamic and are attributed to the expression, localization, and activity of enzymes that mediate the addition, deletion, and reading of these marks. Enzymes creating histone marks are defined as “writers,” which utilize cofactors (mostly metabolites) to add the covalent modifications on histones. Enzymes removing histone modifications are defined as “erasers,” and the regulatory proteins that recognize and bind to modified histones are called “readers” [7]. In general, two mechanisms are employed by histone PTMs to affect chromatin remodeling. First, histone PTMs are able to alter DNA accessibility through the formation of higher-order chromatin structures, which is either loosely packaged and transcriptionally active euchromatin, or highly condensed and transcriptionally silent heterochromatin. In addition to having a role in chromatin architecture, histone PTMs play a role in the recruitment of reader proteins which mediate fundamental processes such as transcription, DNA replication, and DNA repair. The binding of reader protein to a particular histone PTM can be affected by the presence or absence of neighboring marks. Indeed, many reader proteins have more than one reader domain, which confers the ability to recognize combinations of histone marks [8].

The most studied histone modifications are those on lysine residues, which are strongly associated with distinct states of gene transcription. For instance, lysine acetylation on histone H3 and H4 and trimethylation on lysine 4 of histone 3 (H3K4me3) are linked with the promoters of transcriptionally active genes [9, 10], whereas the trimethylation on lysine 9 (H3K9me3) and lysine 27 (H3K27me3) of histone H3 commonly marks transcriptional silencing of repressed genes [11, 12]. Both the acetylation on lysine 27 (H3K27ac) and the monomethylation on lysine 4 of histone H3 (H3K4me1) are enriched in active cis-regulatory enhancer elements important for transcriptional regulation [13]. Many other histone modifications have been found to be correlated with different chromatin states and gene expression; however, the causality of some histone modifications with transcriptional regulation still needs further study [14, 15].

As a ubiquitous life form on this planet, bacteria have adapted and evolved to colonize a wide range of ecological niches. To successfully survive in the human niche, both symbiotic and pathogenic bacteria have acquired cross talk mechanisms allowing for a close interaction with host organisms or

cells [16–18]. In recent years, accumulated studies have described an intricate interplay between bacteria and the host cell epigenome [19]. Whether histone modifications induced upon infection help host cells to ensure a proper response during infection, or are actively induced in a manipulative process to alter transcription and the corresponding infection outcomes, remains to be studied case by case. Herein, we aim to summarize and discuss the importance and prevalence of histone modifications exploited by a variety of different bacteria and how this field of study has revealed new findings in chromatin biology.

Bacterial sensing triggers signal transduction-mediated histone modifications

Histone H3 acetylation and phosphorylation are highly dynamic modifications which are often downstream of signaling cascades and allow cells to adapt their transcriptional response to environmental stimuli. Physiologically, histone acetylation is well described as being a mark of active transcription [20]. The regulation of histone acetylation of lysine residues relies on two families of enzymes, histone acetyltransferases (HATs) and histone deacetylases (HDACs). By utilizing the cofactor acetyl CoA, HATs act as writers to add an acetyl group onto lysine side chains, which neutralizes the lysine’s positive charge, leading to decreased affinity between histones and DNA, and to opening of the chromatin structure. Acetylated lysines also act as scaffolds to recruit reader proteins bearing bromodomains, which in turn recruit and associate with the transcriptional machinery [21]. Histone acetylation is tightly linked to H3 phosphorylation on such residues as S10 and S28, which upon modification can recruit HATs and bromodomain proteins [22–24]. The study of H3 phosphorylation is mostly carried out in response to extracellular stimuli as several kinases, including Rsk2, JNK, and MSK1/2 from MAPK, and Ikk α from nuclear factor- κ B (NF- κ B), are reported to mediate this modification [25–28]. As bacterial factors and products are well described cellular agonists, they are often used for molecular characterization of signaling cascades and downstream histone modifications.

Lipopolysaccharide

Bacterial pathogens are sensed by pathogen recognition receptors (PRRs), including Toll-like receptors (TLRs) and Nod-like receptors (NLRs) that detect a vast array of different types of bacterial molecules, such as nucleic acids, cell wall components, and metabolites [29]. The bacterial cell wall component, lipopolysaccharides (LPSs), is the prototypical representative widely used as cellular agonist. Upon sensing of LPS, PRRs initiate an early, rapid, and nonspecific response through the activation of multiple signaling pathways,

including two major pro-inflammatory signaling cascades, NF- κ B and MAPKs [30]. The activation of these kinase-signaling cascades ultimately triggers the expression of inflammatory genes, and histone modifications such as acetylation and phosphorylation are necessary for full transcriptional activation [31, 32]. The mechanism of NF- κ B-mediated histone phosphorylation is relatively well characterized and depends on the association of I κ B kinase alpha (IKK α) with NF- κ B subunits, their transport into the nucleus where they associate to the promoters of inflammatory genes and phosphorylate histones at those loci [25, 31]. NF- κ B binding upon LPS stimulation was first shown in primary mouse macrophages at the *il-12* promoter, and this correlated with acetylation of histone H3 and H4 [33]. Further studies demonstrated that LPS stimulation leads to phosphorylation of H3S10 and phosphorylation/acetylation of H3S10K14 at multiple inflammatory genes [34, 35]. It has been suggested that S10 phosphorylation and phosphorylation/acetylation helps reposition histones upon LPS treatment in primary human dendritic cells. Such histone movement allows chromatin to adopt an open state, allowing NF- κ B to gain access to a subset of inflammatory promoters [35]. More recently, genome-wide approaches are used to identify epigenetic regulation patterns of histone phosphorylation/acetylation in LPS stimulation model. ChIP-seq analysis of H3 acetylation and phosphorylated RNA polymerase II showed a correlation between H3 acetylation and upregulation of inflammatory genes in human THP-1 cells [36]. In addition, H3S28ph rapidly accumulates at the most highly induced inflammatory genes in mouse bone marrow-derived macrophages (BMDM) upon LPS treatment and selectively promotes p300-dependent transcription of these [24]. Mechanistically, histone H3 has been identified as the substrate of JNK and MSK1/2 (kinase downstream of P38) in vitro, and phosphorylated H3 at either serine 10 or 28 will stimulate the acetylation of neighboring lysines. For example, H3S10ph promotes binding of the HAT GCN5 to histone tails resulting in an enhanced acetylation of H3K9 and H3K14, which is observed in both yeast cells and BMDM [22, 23]. Similarly, H3S28ph increases binding of the HAT p300, which enhances H3K27 acetylation and transcriptional activation in BMDM [24]. In addition, the reader protein 14-3-3 binds to H3S10ph and recruits the HAT MOF, which subsequently induces H4K16ac and release of paused Pol II polymerase from promoter proximal regions in HEK 293 cells [37]. Therefore, the notion that histone phosphorylation/acetylation contributes to the transcriptional activation of inflammatory genes was shown in a setting using bacterial products as agonists in many different cell types.

Similarly to bacterial products, bacterial infection also induces pro-inflammatory signaling pathways and histone modifications. In endothelial cells, infection with *Listeria monocytogenes* leads to a MAPK-dependent acetylation (lysine 8) of histone H4 and phosphorylation/acetylation (serine

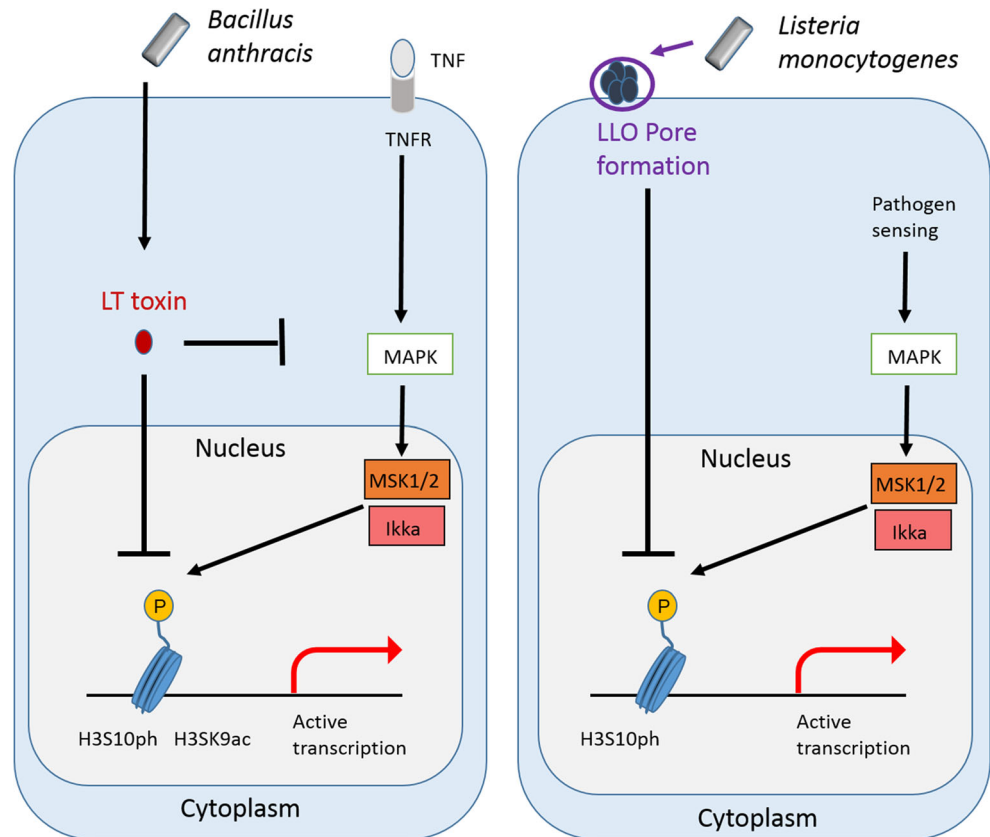
10/lysine 14) of histone H3 at *il8* promoter [38]. In lung epithelial cells, *Legionella pneumophila* has similar effects on histone modifications and the *IL8* gene expression [39]. *L. pneumophila*-mediated acetylation of histone H4 and phosphorylation/acetylation (serine 10/lysine 14) of histone H3 depends on the MAPK and IKK and correlates with increased binding of the HAT p300/CBP and decreased binding of HDAC to the *IL8* promoter.

Bacterial toxins

In theory, any bacterial stimulus activating NF- κ B and MAPKs has the potential to trigger histone phosphorylation (mostly H3S10ph and H3S28ph) and associated acetylation (Fig. 1). Accordingly, successful bacterial pathogens have evolved factors that interfere with host innate immunity by disrupting NF- κ B and MAPKs (Table 1). We will highlight two bacterial toxins that dampen host innate immune responses through inhibiting histone phosphorylation/acetylation. The first one is lethal toxin (LT) from *Bacillus anthracis*, the agent of anthrax. After LT treatment, NF- κ B is prevented from binding to the *IL8* promoter upon stimulation of cells with TNF α [40]. However, LT toxin does not affect NF- κ B activation itself, suggesting that chromatin remodeling at the *IL8* locus is being affected. Correspondingly, LT toxin treatment prevents P38-dependent phosphorylation of histone H3S10 and acetylation of H3K14 at the *IL8* locus upon stimulation with TNF α . A previously reported mechanism, which showed that endocytosed LT toxin cleaves and inactivates MAPKs to block MAPK signaling, could explain how LT toxin affects histone phosphorylation/acetylation [41].

Another toxin family inducing histone modifications is the pore-forming toxin (PFT) family (Fig. 1). Cholesterol-dependent cytolysins, produced by such pathogens as *Listeria monocytogenes*, *Streptococcus pneumoniae*, and *Clostridium perfringens*, perturb host cell membranes by binding to cholesterol and generating protein pores [61]. Upon treatment of cells with these toxins, H3S10 is dephosphorylated and H4 deacetylated [62]. These modifications correlate with downregulation of key inflammatory genes, such as *cxcl2*, *dusp4*, and *ifit3*, and occur at the promoters of those genes. A further study revealed that pore formation-induced potassium efflux is required for this process [63]. More recently, *Pseudomonas aeruginosa* was shown to induce similar potassium efflux and histone H3 dephosphorylation; however, pore formation was not induced by a toxin but by the PopB-PopD translocon generated by type III secretion system (T3SS) insertion into host cell membrane [64]. Together, these studies suggest that modulation of histone phosphorylation/acetylation through the disruption of plasma membrane integrity could be a common mechanism for bacterial pathogens to reprogram host signaling and transcription.

Fig. 1 Bacteria alter signal transduction-mediated histone modifications. Sensing of bacteria or other inflammatory signal receptors trigger the activation of kinase signaling, including MAPK and NF- κ B, which leads to the downstream MSK1/2 or I κ k α -mediated phosphorylation of histone H3. To counteract this effect, LT toxin from *Bacillus anthracis* dampens MAPK and prevents MAPK-mediated H3S10 phosphorylation, and pore-forming toxins, such as LLO from *Listeria monocytogenes*, induce dephosphorylation of histone H3



Bacteria alter the role of host enzymes to reprogram epigenome and transcription

Subversion of host signaling cascades by bacterial pathogens is a common feature, and targeting chromatin-modifying components is no exception. Interestingly, the study of host-pathogen interactions has helped to reveal the function, or new function, of chromatin-targeting proteins (Fig. 2).

Shigella flexneri OspF

The type III secreted effector protein OspF of *Shigella* is a phosphothreonine lyase mediating an enzymatic reaction called eliminylation [65, 66]. OspF, as well as another factor SpvC from *Salmonella*, is able to convert a phosphothreonine residue into a dehydrobutyrine residue and irreversibly inactivates MAPK [65]. Thus, OspF prevents MAPK phosphorylation (P38 and ERK), and thereby abrogates subsequent histone H3S10 phosphorylation at a subset of NF- κ B-regulated promoters and blocks inflammatory gene transcription (Fig. 2) [67]. OspF also controls the activity of an important chromatin reader, heterochromatin protein 1 (HP1), to repress gene expression during *Shigella* infection (Fig. 2) [68]. OspF directly interacts with HP1 γ and causes HP1 dephosphorylation on S83, through inactivating the kinase MSK1. The activity of

OspF is unique and to our knowledge, no eukaryotic homolog has yet been found.

Listeria monocytogenes

The eukaryotic protein Bromo adjacent homology domain containing 1 (BAHD1) was identified in a screen for human proteins interacting with the nuclear-targeted listerial factor LntA; however, its function in the cell was undefined. An early study suggested that BAHD1 targets insulin-like growth factor II (IGF2) transcript and its antisense transcript (IGF2AS) in HEK293 cells, and overexpression of BAHD1 induces large-scale chromatin condensation [69]. In fact, BAHD1 was shown to promote heterochromatin formation and gene repression in partnership with several repressive chromatin factors, such as HP1, HDAC1/2, KAP1, SETDB1, and SUV39H1. However, the genes regulated by BAHD1 and signals controlling BAHD1 were unknown until the finding that *L. monocytogenes* regulates the activity of BAHD1 (Fig. 2). LntA, which is targeted to chromatin, colocalizes with BAHD1 at heterochromatic regions. LntA-BAHD1 interaction alleviated the binding of BAHD1 to type III interferon (IFN) promoters, thereby upregulating their expression [70]. In agreement, interactome studies of BAHD1 showed that it co-purifies with repressive complex components such as HDACs and probably acts to repress gene transcription [71].

Table 1 Bacterial factors intercepting host MAPK and NF- κ B signaling

Bacteria	Effector	Pathway	Target
<i>Bacillus anthracis</i>	Lethal toxin	MAPK	MKKs [41]
<i>Listeria monocytogenes</i>	InlC	NF- κ B	IKK [42]
<i>Yersinia pestis</i>	YopJ	MAPK	MKKs [43]
		NF- κ B	IKK [44]
		NF- κ B	TAK1 [45]
<i>Salmonella enterica</i>	AvrA	MAPK	MAPKK [46]
	SptP	MAPK	Raf-1 [47]
	SpvC	MAPK	ERK [48]
	SpvD	NF- κ B	RelA [49]
	PipA/GogA /GtgA	NF- κ B	RelA/RelB [50]
<i>Shigella flexneri</i>	OspF	MAPK	MKKs [51]
	IpaH1.4	NF- κ B	LUBAC [52]
	IpaH2.5	NF- κ B	LUBAC [52]
	IpaH9.8	NF- κ B	IKK [53]
	OspG	NF- κ B	E2 enzyme [54]
	OspI	NF- κ B	E2 enzyme [55]
	OspZ	NF- κ B	TAB2/3 [56]
<i>Chlamydia trachomatis</i>	CT441	NF- κ B	RelA [57]
<i>Vibrio parahaemolyticus</i>	VopA	MAPK	MKKs [58]
<i>Escherichia coli</i>	NleE	NF- κ B	TAB2/3 [56, 59]
	NleC	NF- κ B	RelA [60]

L. monocytogenes was also described to hijack the deacetylase SIRT2 during infection thereby revealing a new function for this host protein (Fig. 2). SIRT2 is a nicotinamide-

adenine-dinucleotide-dependent deacetylase that was known to target tubulin for deacetylation and to localize mainly in the cytoplasm. However, upon infection, SIRT2 is relocalized

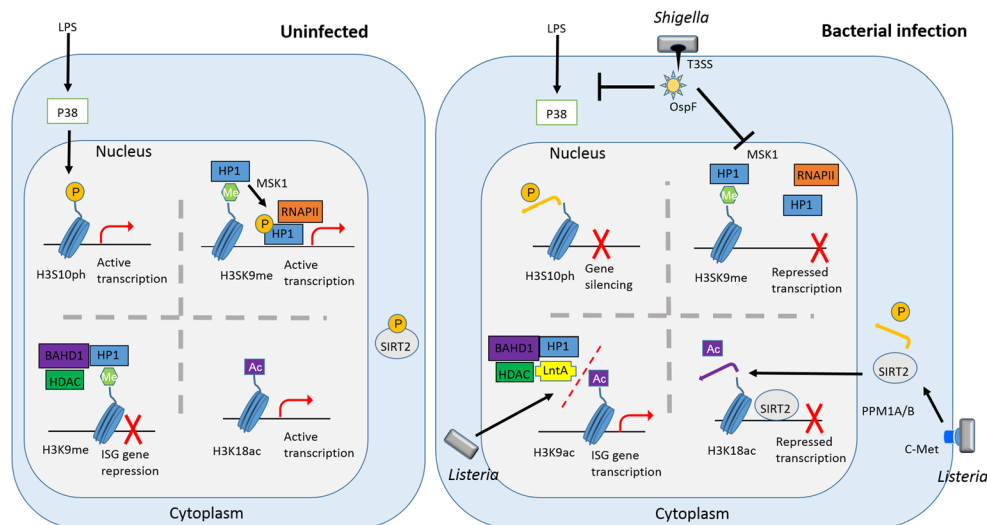


Fig. 2 Bacteria alter the role of host histone-modifying enzymes. In uninfected cells, LPS stimuli leads to P38-dependent H3S10 phosphorylation and expression of inflammatory genes; MSK1 phosphorylates HP1 at S83, which interacts with RNAPII and promotes gene transcription; BAHD1 associates with HP1 and HDAC, forms heterochromatin to suppress ISG transcription; SIRT2 with S25 phosphorylation localizes in cytoplasm. Upon *Shigella* infection, T3SS effector OspF dampens P38 signaling induced by LPS and prevents the corresponding H3S10ph. Furthermore, OspF prevents HP1 S83

phosphorylation through blocking MSK1, thereby decreases the transcription of HP1-targeted genes. During *Listeria* infection, bacterial factor LntA interacts with BAHD1 complex, alleviates the binding of BAHD1 with chromatin and restores the H3 acetylation level, resulting in the transcription of ISGs. In addition, bacterial surface protein InlB interacts with cell receptor c-Met, which induces a PPM1A/B-dependent dephosphorylation of SIRT2 at S25. SIRT2 without S25ph becomes chromatin associated, causes deacetylation of H3K18 and transcriptional repression

from the cytoplasm to the nucleus of infected cells. In the nucleus, SIRT2 is specifically targeted to chromatin where it binds to transcriptional start sites of genes repressed during infection and induces deacetylation of H3K18 [72]. SIRT2 relocalization and H3 deacetylation were shown to occur upon binding of the listerial factor internalin B (InIB), which is a surface-expressed protein. InIB binds to the host cell surface receptor c-Met and engages downstream Akt/PI3K signaling, which is required for H3K18 deacetylation [72]. This report was the first to show a role for SIRT2 at chromatin and in transcriptional regulation. Closer study revealed that SIRT2 is dephosphorylated on serine 25 during *Listeria* infection, and this dephosphorylation enhances the chromatin association of SIRT2 [73]. Infection-induced dephosphorylation of SIRT2 on S25 is mediated by the host phosphatases PPM1A and PPM1B. Importantly, blocking H3K18 deacetylation through inhibition of SIRT2, or PPM1A, and PPM1B greatly attenuates the *L. monocytogenes* load, suggesting this histone-modifying mechanism is critical for bacterial infection. These studies revealed a novel link between c-Met signaling, SIRT2, and histone modifications of H3K18 and highlight how infection can uncover new functions for host proteins.

***Mycobacterium tuberculosis* 19-kDA lipoprotein**

Infection with *Mycobacterium tuberculosis*, or activation of TLR2 signaling, leads to repression of a subset of interferon-stimulated genes, such as CIITA, which codes for the master regulator of MHC class II genes [74–76]. Importantly, histone deacetylation is involved in transcriptional repression of this locus, and 19-kDA lipoprotein is a bacterial factor required for this process. The HDAC C/EBP is recruited to the CIITA locus thereby blocking IFN- γ -induced acetylation of histones H3 and H4 [77]. Furthermore, mycobacterial infection induced the recruitment of the co-repressor Sin3A limiting expression of the CIITA-regulated genes, HLA-DR α and HLA-DR β [78]. These studies suggest that mycobacteria are able to suppress IFN- γ -dependent responses in macrophages by modulating histone acetylation. Although a mycobacterial 19-kDA lipoprotein was shown to be essential for modulating the MHC-II genes, whether this response is specific or mediated by general activation of TLR2 signaling cascades remains to be determined.

Bacteria produce new histone-modifying enzymes

In addition to targeting host chromatin modifiers, bacteria can alter the host epigenome directly through the catalytic activity of secreted bacterial proteins termed nucleomodulins (Fig. 3). The major histone modification targeted by bacteria in this manner is histone methylation. Histone methylation is deposited on lysine residues, which can be mono-, di-, or trimethylated, and arginine residues, which can be mono-, symmetrically, or

asymmetrically di-methylated [79]. Histone methylation is a mark involved in both gene activation and gene silencing depending on the site and type of methylation. For instance, trimethylated H3K4 (H3K4me3) at promoters stimulates transcription by recruiting a variety of reader proteins involved in the assembly of pre-initiation complex (PIC) containing Pol II and general transcription factors [10]. In contrast, H3K9me2/3 is associated with gene repression, as this mark recruits heterochromatin-binding protein 1 (HP1) leading to transcriptional repression and formation of heterochromatin [80]. Through the recruitment of polycomb repressive complex PRC1, H3K27me3 also contributes to the compaction of chromatin [81]. Like other histone modifications, the level of histone methylation is regulated by both writers, such as histone lysine methyltransferases (HKMTs) or protein arginine methyltransferases (PRMTs), and erasers, such as LSD demethylases or JMJC demethylases. Importantly, almost all HKMTs contain a SET domain, which is the enzymatic domain catalyzing the transfer of a methyl group from S-adenosylmethionine (SAM) to lysines [82]. Interestingly, recent studies have shown that bacteria harbor HKMT homologs containing SET domains that can directly methylate histone proteins.

***Legionella pneumophila* LegAS4**

LegAS4, a *Legionella pneumophila* type IV secretion system (T4SS) effector, localizes to the nucleus during infection and regulates the expression of ribosomal RNA genes (rDNA) in a manner depending on its SET domain (Fig. 3) [83]. The SET domain of LegAS4 has catalytic activity towards histone H3, mainly catalyzing H3K4me2. LegAS4 has a high-affinity binding to HP1 α/γ , the reader proteins recognizing H3K9me2 and enriched in transcriptionally silent rDNA locus. Through the interaction with HP1 α/γ , LegAS4 associates with the promoter and intergenic spacer regions of rDNA to promote transcription. Strikingly, bioinformatic analysis identified a large family of LegAS4-like SET domain proteins from various bacteria, suggesting that histone methylation might be a common strategy used by bacteria to modulate host transcription [83]. In fact, experimental data suggested that LegAS4-like protein BtSET, a type III secretion system (T3SS) effector from *Burkholderia thailandensis*, is also able to methylate histones. Similarly to LegAS4, BtSET promotes H3K4me2 and activates rDNA transcription [83].

***Chlamydia trachomatis* NUE and *Bacillus anthracis* BaSET**

Two other secreted bacterial methyltransferases containing SET domains have been reported, but the link between their histone-modifying ability and transcriptional regulation

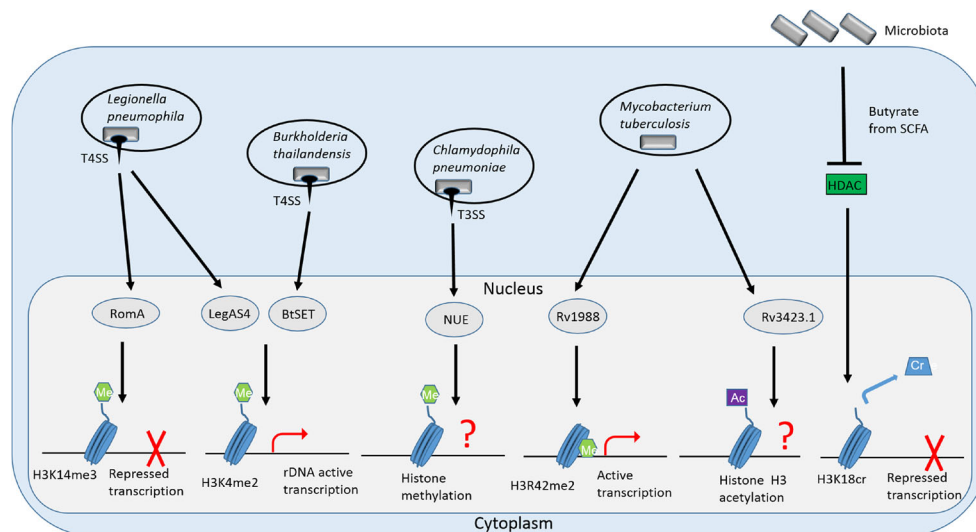


Fig. 3 Bacteria produce new histone-modifying enzymes and create new histone modifications. Through T4SS, *Legionella pneumophila* secretes RomA to induce H3K14me₃, which represses the expression of targeted genes. Another T4SS effector LegAS4 from *Legionella pneumophila*, as well as the T4SS effector BtSET from *Burkholderia thailandensis*, induces H3K4me₂ and the expression of ribosomal RNA genes (rDNA). *Chlamydia* T3SS effector NUE methylates histones, but the

target genes are unknown. Methyltransferase RV1988 secreted by *Mycobacterium tuberculosis* methylates histone H3 at residue R42, which promotes gene activation. Another *Mycobacterium*-secreted protein Rv3423.1 is acetyltransferase that enhances H3 acetylation levels during infection, but its role is not known. In addition to pathogenic bacteria, the gut microbiota is able modulate H3K18cr through the HDAC inhibitor butyrate, which is a metabolic byproduct

remains to be determined. Nuclear effector (NUE) is secreted through T3SS by *Chlamydia trachomatis*. After injection into infected cells, NUE localizes to the cell nucleus and associates with chromatin (Fig. 3). Although NUE methylates histones H2B, H3, and H4 in vitro, the sites of histone methylation by NUE, the methylation types, and regulated genes in infected cells have yet to be identified [84].

The BaSET protein from the extracellular pathogen *B. anthracis* is another example of a bacterially encoded SET domain protein targeting host histones. BaSET trimethylates histone H1 in vitro, but does not target other core histones. Furthermore, BaSET represses the expression of NF- κ B target genes in a reporter assay system [85]. Further studies will be necessary to determine whether the role of BaSET on NF- κ B genes is mediated through histone H1 methylation.

Mycobacterium tuberculosis Rv1988

Interaction studies of *Mycobacterium tuberculosis* proteins with histone H3 identified Rv1988 (Fig. 3) [86]. During infection, it is secreted, localizes to the host nucleus, and interacts with chromatin. Interestingly, Rv1988 was shown to target a non-tail histone H3 arginine, H3R42, which is a non-canonical site of modification present at the entry/exit point of DNA in the nucleosome. Infection revealed that Rv1988 induced H3R42me₂ and repressed the transcription of ROS-

related genes, which could contribute to bacterial survival in macrophages. However, the role of H3R42me₂ in gene regulation is still controversial, as studies have shown that this modification is involved in both transcriptional activation and repression. In a cell-free system, in the presence of p53 and p300, H3R42me₂ promoted transcriptional activation [87]. However, in yeast, where a lysine is present instead of arginine, H3K42me₂ caused gene repression, and this repressive effect persisted even when this lysine was replaced by arginine [88]. The study of Rv1988-mediated H3K42me₂ and its link to gene repression could therefore provide a useful tool for dissecting gene regulation by this histone modification.

Mycobacterium tuberculosis Rv3423 and RV0018

Besides histone methylation, bacteria also have the potential to target histone acetylation or phosphorylation. Rv3423.1 is an acetyltransferase secreted by *M. tuberculosis* that associates with histones during infection of macrophages, which correlates with an increase in histone acetylation (Fig. 3) [89]. In vitro biochemical assays with purified recombinant protein showed that Rv3423.1 is able to acetylate histone H3 on the K9/K14 residues, and transfection of macrophages with Rv3423.1 confirmed its co-localization with chromatin. However, although Rv3423.1 contributes to bacterial survival, the impact of Rv3423.1-mediated histone acetylation on gene expression has not been explored.

Another secreted effector Rv0018c from *M. tuberculosis* is a serine/threonine phosphatase that has been shown to dephosphorylate histones in vitro, but the evidence that Rv0018c induces histone dephosphorylation during infection is still lacking [90]. Similarly, a serine/threonine phosphatase SP-STP from extracellular pathogen *Streptococcus pyogenes* is able to dephosphorylate histone H1 in an in vitro assay [91]. Treating Detroit 562 pharyngeal cells with purified SP-STP suggested that SP-STP has an internalization mechanism to cross cell membrane and nuclear membrane. Again, whether SP-STP is able to induce histone dephosphorylation during infection has not yet been shown.

Bacteria create new histone modifications in host cells

Non-canonical targeting of histones could be a mechanism that bacteria have evolved to induce distinct modifications during infection. In fact, several bacterial effector proteins have been shown to induce modifications that have not yet been reported in eukaryotic cells (Fig. 3).

Legionella pneumophila RomA

RomA is a SET domain-containing methyltransferase secreted by the T4SS of *L. pneumophila* [92]. The other SET domain protein of *Legionella*, LegAS4, has some homology to RomA; however, its substrate preference is different. By mass spectrometry and through the probing of a large panel of antibodies, RomA was shown to mediate H3K14me3 but not H3K4me2, as LegAS4. During infection, RomA specifically trimethylates K14 of histone H3, a histone mark not previously described in mammals. The presence of H3K14 methylation was found to counteract H3K14 acetylation, leading to a loss of an active histone mark, and repression of host gene expression [92]. In fact, H3K14me3 was found to mark promoter regions of a large number of genes, including inflammatory genes, and to be essential for a productive infection. Interestingly, RomA was also shown to target other proteins besides histones suggesting additional roles for RomA besides transcriptional regulation [93].

Microbiota and histone crotonylation

In recent years, a number of new histone PTMs have been discovered or “re-discovered.” For example, histone lysines can be acylated with intermediates from metabolism, generating longer chain acylations such as crotonylation, butyrylation, and hydroxybutyrylation [94]. Those acylation modifications are similar to well-studied lysine acetylation in general, but their role in gene regulation might be distinct, due to the difference of hydrocarbon chain length and hydrophobicity or charge [95]. Among the identified non-acetyl histone

acylations, histone crotonylation is an interesting modification that has recently been the focus of several studies. Crotonyl-CoA is the crotonyl donor for histone crotonylation, which is added by HATs and removed by HDACs [96]. In a cell-free system, p300-dependent crotonylation of H3K18 (H3K18cr) induced gene expression, demonstrating a causal relationship between crotonylation and transcriptional activation [97]. In fact, the levels of H3K18cr directly affect LPS-mediated transcriptional activation of the inflammatory response [97]. In addition to LPS, more recently, a link between bacteria and lysine crotonylation has been identified. Short-chain fatty acids (SCFAs) are major products of gut bacterial fermentation, provide energy sources for the gastrointestinal tract epithelial cells, and affect multiple cellular functions including immune responses [98]. Such microbiota-derived SCFAs were shown to regulate H3K18cr in intestinal epithelial cells (Fig. 3) [99]. One of the microbiota-derived SCFAs butyrate is a well-known HDAC inhibitor. Depletion of the gut microbiota results in a decrease of butyrate, leading to a correlated reduced histone crotonylation globally in the colon. Therefore, intestinal microbiota can affect host cell gene expression through epigenomic regulation mediated by metabolism products.

Bacterial products triggered histone modifications uncover novel memory-like response

The lasting potential of bacteria-mediated histone modifications could have important consequences for either expression of inflammatory genes in cell defense or promoting bacterial survival or persistence. Although epigenetic modifications are dynamic and reversible, certain histone modifications, such as histone methylations, have a relatively longer half-life, which can even be maintained across cell division [100]. Given the fact that bacterial infections trigger histone modifications, these represent attractive candidates for mediating infection-induced memory-like responses. Such memory-like responses may determine the choice or the effectiveness of subsequent host responses in a tailor-made fashion, dependent on the history of stimuli.

Endotoxin tolerance, which occurs upon exposure to high levels of LPS, is one such example of lasting changes leading to a lack of response to a subsequent challenge. This immunosuppression phenomenon is characterized by the downregulation of the pro-inflammatory genes and is associated with poor patient prognosis [101]. A series of studies illustrate the role of epigenetic modifications in endotoxin tolerance, which is characterized by sustained levels of H3K9me2 and binding of HP1 α , reduced level of H3S10ph, and diminished binding of NF- κ B RelA to the promoter of certain inflammatory genes in LPS tolerant THP-1 cells (Fig. 4) [102, 103]. A further study suggested that another NF- κ B subunit RelB is required for endotoxin tolerance in THP-1 cells, which induces

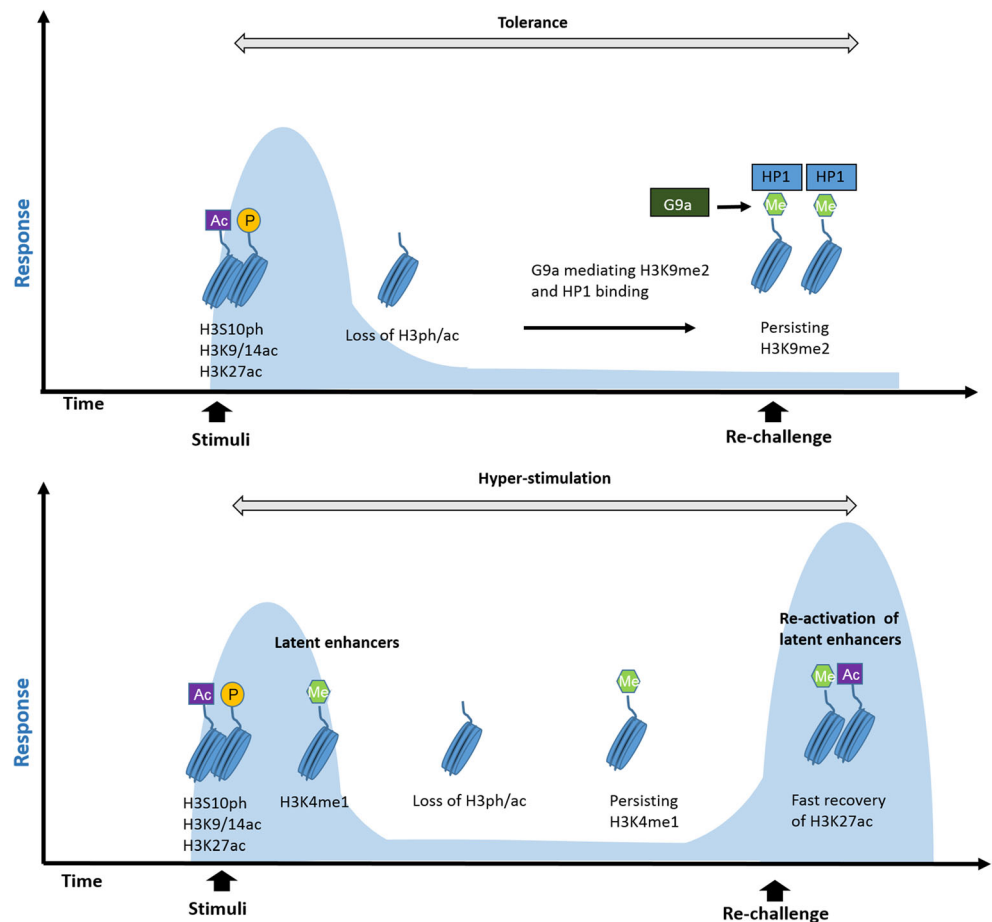
facultative heterochromatin formation by directly interacting with the H3K9 methyltransferase G9a to suppress inflammatory genes (Fig. 4) [104]. Additionally, in primary human monocyte-derived macrophages, LPS stimulation leads to the transcriptional inaction of certain genes upon LPS re-exposure, and this is correlated with the impaired accumulation of H3K27ac at the enhancers of those genes [105]. Therefore, host cells utilize histone-modifying strategies to prevent an overwhelming response upon systemic inflammation, a phenomenon observed well beyond primary exposure. The impact of bacteria on such a tolerance response has not yet been studied.

In contrast to endotoxin tolerance, exposure to low levels of LPS generates an increased response to recurrent challenges. Accumulated evidence indicates that this memory-like response of innate immunity could be conferred through epigenetic reprogramming similarly to what has been shown for tolerance [106]. Treating peritoneal macrophages with LPS leads to a p38-dependent phosphorylation of ATF7 and its dissociation from chromatin. Since ATF7 suppresses a group of immunity-related genes by recruiting G9a, the release of ATF7 induces a decrease of H3K9me2 and an increase in basal expression of target genes even upon removal [107]. Treating macrophages with β -glucan further

demonstrate the important role of epigenetics in memory-like response. β -glucan is able to induce enhanced response to secondary challenge, which is associated with changes in histone H3K4me3 [108]. Interestingly, β -glucan can rescue both the transcription and levels of H3K27ac at the enhancers of unresponsive genes in endotoxin tolerance macrophages [105].

The important role for enhancers in innate memory was further demonstrated by the discovery that the repertoire of genetic regions marked with H3K4me1 could change upon external stimulation. Indeed, stimulation of BMDM with LPS leads to sequential binding of transcriptional factors to certain regions that are completely unmarked in unstimulated conditions, enabling deposition of enhancer marks including H3K4me1 and H3K27ac (Fig. 4) [109]. After removal of the LPS stimulus, many of these enhancers return to a resting state, in parallel with loss of H3K27ac but not H3K4me1 (Fig. 4). In fact, persistent H3K4me1 at those latent enhancers endures for a long time and mediates an enhanced transcriptional response of neighboring genes upon re-stimulation. Innate memory mediated by histone marks on enhancers is observed not only in non-lymphoid macrophage cells but also in innate lymphoid cells such as natural killer (NK) cells. A recent study illustrated that NK cells isolated from mice

Fig. 4 Bacterial products induced histone modifications are involved in memory-like responses. LPS stimulation induces phosphorylation and acetylation of histone H3 at promoters, and H2K27ac at enhancers. However, dependent on the nature and intensity of primary stimulates, cells can develop a memory-like phenomenon leading to a different response from naïve cells upon re-challenge. High levels of LPS exposure induces endotoxin tolerance, which is associated with G9a-mediated H3K9me2 at promoters and the impairment of H3K27ac at enhancers of certain genes. Low levels of LPS exposure lead to transcriptional hyper-stimulation, which is linked with latent enhancers. In this novel type of enhancer, H3K4me1 and H3K27ac are deposited at unmarked regions upon primary stimulation. Although H3K27ac is a transient mark, H3K4me1 persists and enables an enhanced response to a secondary stimulus with fast recovery of H3K27ac upon re-challenge



postendotoxemia acquire memory-like features characterized by a higher production of IFN γ upon secondary LPS stimulation [110]. This memory to LPS was shown to depend on the appearance of a latent enhancer, which becomes marked with H3K4me1 at the *ifng* locus. Blocking LPS-induced H3K4me1 with a chemical methyltransferase inhibitor abolishes the memory acquired by NK cells to secondary stimulation. Therefore, the study of cellular responses to bacterial factors has uncovered a new class of enhancers, which is latent under basal conditions but amenable to subsequent stimulus. This finding changes the view that enhancers are fixed in differentiated cells and reveal that these regulatory components respond to external stimuli and maintain a transcriptional memory of passed encounters. Further studies will determine whether similar memory occurs upon infection and if bacteria are able to modify it.

Conclusions and perspectives

The study of bacteria-host interactions has provided breakthroughs in our knowledge of cell biology. Similarly, new mechanisms and players are being uncovered in the study of epigenomic regulation of gene expression. Indeed, bacteria provide privileged stimuli to induce specific responses mediated by histone modifications. Investigating the role of histone modifications, especially the new marks or novel functional types, will promote the understanding of how histone modifications influence chromatin structure and the regulatory mechanism of gene transcription. Furthermore, beyond a mechanistic role, function in a cellular process could be associated with specific sets of histone marks.

Beyond a novel understanding of cell biology mechanisms, bacteria-induced histone modifications could provide targets for host-directed therapy, a good alternative for antibiotics that target the pathogen. Indeed, manipulation of epigenomic processes is important for efficient bacterial infection, and targeting or inhibiting these may alter the outcome of infection. In fact, epigenetic therapy has been proposed and applied in multiple human diseases, including cancer, diabetic retinopathy, cardiac dysfunction, and mental diseases. Epigenetic therapy might either modulate the immune response from host cells or directly restrict bacterial infection. With the emerging classes of pharmacological agents and increased target specificity, the therapeutic potential of the infection area has been opened up, especially for chronic or antibiotic-resistant bacterial infections.

Acknowledgments We apologize to any colleagues whose work was not included in this review due to space limitations. We thank Michael Connor for critical reading of the manuscript.

Funding information Work in the Chromatin and Infection Group is supported by the Pasteur Institute and the Agence Nationale de la Recherche (ANR-EpiBActIn). Wenyang Dong is part of the Pasteur - Paris University (PPU) International PhD Program, a project which has received funding from the European Union's Horizon 2020 research and innovation programme under the Marie Skłodowska-Curie grant agreement no. 665807. Wenyang Dong is supported by the EUR G.E.N.E. (reference #ANR-17-EURE-0013) and is part of the Université de Paris IdEx #ANR-18-IDEX-0001 funded by the French Government through its "Investments for the Future" program.

Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

References

- Zhou K, Gaullier G, Luger K (2019) Nucleosome structure and dynamics are coming of age. *Nat Struct Mol Biol* 26(1):3–13
- Bednar J, Horowitz RA, Grigoryev SA, Carruthers LM, Hansen JC, Koster AJ, Woodcock CL (1998) Nucleosomes, linker DNA, and linker histone form a unique structural motif that directs the higher-order folding and compaction of chromatin. *Proc Natl Acad Sci U S A* 95(24):14173–14178
- Narlikar GJ, Sundaramoorthy R, Owen-Hughes T (2013) Mechanisms and functions of ATP-dependent chromatin-remodeling enzymes. *Cell* 154(3):490–503
- Li E (2002) Chromatin modification and epigenetic reprogramming in mammalian development. *Nat Rev Genet* 3(9):662–673
- Tan M, Luo H, Lee S, Jin F, Yang JS, Montellier E, Buchou T, Cheng Z, Rousseaux S, Rajagopal N, Lu Z, Ye Z, Zhu Q, Wysocka J, Ye Y, Khochbin S, Ren B, Zhao Y (2011) Identification of 67 histone marks and histone lysine crotonylation as a new type of histone modification. *Cell* 146(6):1016–1028
- Strahl BD, Allis CD (2000) The language of covalent histone modifications. *Nature* 403(6765):41–45
- Kouzarides T (2007) Chromatin modifications and their function. *Cell* 128(4):693–705
- Musselman CA, Lalonde ME, Cote J, Kutateladze TG (2012) Perceiving the epigenetic landscape through histone readers. *Nat Struct Mol Biol* 19(12):1218–1227
- Agalioti T, Chen G, Thanos D (2002) Deciphering the transcriptional histone acetylation code for a human gene. *Cell* 111(3):381–392
- Laubert SM, Nakayama T, Wu X, Ferris AL, Tang Z, Hughes SH, Roeder RG (2013) H3K4me3 interactions with TAF3 regulate preinitiation complex assembly and selective gene activation. *Cell* 152(5):1021–1036
- Lindroth AM, Shultis D, Jasencakova Z, Fuchs J, Johnson L, Schubert D, Patnaik D, Pradhan S, Goodrich J, Schubert I, Jenuwein T, Khorasanizadeh S, Jacobsen SE (2004) Dual histone H3 methylation marks at lysines 9 and 27 required for interaction with CHROMOMETHYLASE3. *EMBO J* 23(21):4146–4155
- Schubert D, Primavesi L, Bishopp A, Roberts G, Doonan J, Jenuwein T, Goodrich J (2006) Silencing by plant Polycomb-group genes requires dispersed trimethylation of histone H3 at lysine 27. *EMBO J* 25(19):4638–4649
- Creyghton MP, Cheng AW, Welstead GG, Kooistra T, Carey BW, Steine EJ, Hanna J, Lodato MA, Frampton GM, Sharp PA, Boyer LA, Young RA, Jaenisch R (2010) Histone H3K27ac separates

- active from poised enhancers and predicts developmental state. *Proc Natl Acad Sci U S A* 107(50):21931–21936
14. Henikoff S, Shilatifard A (2011) Histone modification: cause or cog? *Trends in genetics* : TIG 27(10):389–396
 15. Gates LA, Foulds CE, O'Malley BW (2017) Histone marks in the 'driver's seat': functional roles in steering the transcription cycle. *Trends Biochem Sci* 42(12):977–989
 16. Koul A, Herget T, Klebl B, Ullrich A (2004) Interplay between mycobacteria and host signalling pathways. *Nat Rev Microbiol* 2(3):189–202
 17. Haraga A, Ohlson MB, Miller SI (2008) Salmonellae interplay with host cells. *Nat Rev Microbiol* 6(1):53–66
 18. Lebeer S, Vanderleyden J, De Keersmaecker SC (2010) Host interactions of probiotic bacterial surface molecules: comparison with commensals and pathogens. *Nat Rev Microbiol* 8(3):171–184
 19. Bieme H, Hamon M, Cossart P (2012) Epigenetics and bacterial infections. *Cold Spring Harbor perspectives in medicine* 2(12):a010272
 20. Eberharter A, Becker PB (2002) Histone acetylation: a switch between repressive and permissive chromatin-second in review series on chromatin dynamics. *EMBO Rep* 3(3):224–229
 21. Filippakopoulos P, Picaud S, Mangos M, Keates T, Lambert J-P, Barsyte-Lovejoy D, Felletar I, Volkmer R, Müller S, Pawson T (2012) Histone recognition and large-scale structural analysis of the human bromodomain family. *Cell* 149(1):214–231
 22. Cheung P, Tanner KG, Cheung WL, Sassone-Corsi P, Denu JM, Allis CD (2000) Synergistic coupling of histone H3 phosphorylation and acetylation in response to epidermal growth factor stimulation. *Mol Cell* 5(6):905–915
 23. Lo WS, Trievel RC, Rojas JR, Duggan L, Hsu JY, Allis CD, Marmorstein R, Berger SL (2000) Phosphorylation of serine 10 in histone H3 is functionally linked in vitro and in vivo to Gcn5-mediated acetylation at lysine 14. *Mol Cell* 5(6):917–926
 24. Josefowicz SZ, Shimada M, Armache A, Li CH, Miller RM, Lin S, Yang A, Dill BD, Molina H, Park HS, Garcia BA, Taunton J, Roeder RG, Allis CD (2016) Chromatin kinases act on transcription factors and histone tails in regulation of inducible transcription. *Mol Cell* 64(2):347–361
 25. Yamamoto Y, Verma UN, Prajapati S, Kwak YT, Gaynor RB (2003) Histone H3 phosphorylation by IKK-alpha is critical for cytokine-induced gene expression. *Nature* 423(6940):655–659
 26. Dyson MH, Thomson S, Inagaki M, Goto H, Arthur SJ, Nightingale K, Iborra FJ, Mahadevan LC (2005) MAP kinase-mediated phosphorylation of distinct pools of histone H3 at S10 or S28 via mitogen- and stress-activated kinase 1/2. *J Cell Sci* 118(Pt 10):2247–2259
 27. Tiwari VK, Stadler MB, Wirbelauer C, Paro R, Schubeler D, Beisel C (2011) A chromatin-modifying function of JNK during stem cell differentiation. *Nat Genet* 44(1):94–100
 28. Sassone-Corsi P, Mizzen CA, Cheung P, Crosio C, Monaco L, Jacquot S, Hanauer A, Allis CD (1999) Requirement of Rsk-2 for epidermal growth factor-activated phosphorylation of histone H3. *Science* 285(5429):886–891
 29. Akira S, Uematsu S, Takeuchi O (2006) Pathogen recognition and innate immunity. *Cell* 124(4):783–801
 30. Arthur JS, Ley SC (2013) Mitogen-activated protein kinases in innate immunity. *Nat Rev Immunol* 13(9):679–692
 31. Anest V, Hanson JL, Cogswell PC, Steinbrecher KA, Strahl BD, Baldwin AS (2003) A nucleosomal function for I{kappa}B kinase-alpha in NF-kappaB-dependent gene expression. *Nature* 423(6940):659–663
 32. L.C. Poulsen, R.J. Edelman, S. Kruger, R. Dieguez-Hurtado, A. Shah, T.E. Stav-Noraas, A. Renzi, M. Szymanska, J. Wang, M. Ehling, R. Benedito, M. Kasprzycka, E. Baekkevold, O. Sundnes, K.S. Midwood, H. Scott, P. Collas, C.W. Siebel, R.H. Adams, G. Haraldsen, E. Sundlisaeter, J. Hol. Inhibition of endothelial NOTCH1 signaling attenuates inflammation by reducing cytokine-mediated histone acetylation at inflammatory enhancers, Arteriosclerosis, thrombosis, and vascular biology 38(4) (2018) 854–869
 33. Weinmann AS, Mitchell DM, Sanjabi S, Bradley MN, Hoffmann A, Liou H-C, Smale ST (2001) Nucleosome remodeling at the IL-12 p40 promoter is a TLR-dependent, Rel-independent event. *Nat Immunol* 2(1):51–57
 34. Rigillo G, Vilella A, Benatti C, Schaeffer L, Brunello N, Blom JMC, Zoli M, Tascetta F (2018) LPS-induced histone H3 phospho (Ser10)-acetylation (Lys14) regulates neuronal and microglial neuroinflammatory response. *Brain Behav Immun* 74: 277–290
 35. Saccani S, Pantano S, Natoli G (2002) p38-Dependent marking of inflammatory genes for increased NF-kappa B recruitment. *Nat Immunol* 3(1):69–75
 36. Iglesias MJ, Reilly SJ, Emanuelsson O, Sennblad B, Pimoradian Najafabadi M, Folkersen L, Malarstig A, Lagergren J, Eriksson P, Hamsten A, Odeberg J (2012) Combined chromatin and expression analysis reveals specific regulatory mechanisms within cytokine genes in the macrophage early immune response. *PLoS One* 7(2):e32306
 37. Zippo A, Serafini R, Rocchigiani M, Pennacchini S, Krepelova A, Oliviero S (2009) Histone crosstalk between H3S10ph and H4K16ac generates a histone code that mediates transcription elongation. *Cell* 138(6):1122–1136
 38. Schmeck B, Beermann W, van Laak V, Zahlten J, Opitz B, Witzernath M, Hocke AC, Chakraborty T, Kracht M, Rosseau S, Suttrop N, Hippenstiel S (2005) Intracellular bacteria differentially regulated endothelial cytokine release by MAPK-dependent histone modification. *J Immunol* 175(5):2843–2850
 39. B. Schmeck, J. Lorenz, D. N'Guessan P, B. Opitz, V. van Laak, J. Zahlten, H. Slevogt, M. Witzernath, A. Flieger, N. Suttrop, S. Hippenstiel. Histone acetylation and flagellin are essential for *Legionella pneumophila*-induced cytokine expression, *Journal of immunology* 181(2) (2008) 940–7
 40. Raymond B, Batsche E, Boutillon F, Wu YZ, Leduc D, Balloy V, Raoust E, Muchardt C, Goossens PL, Touqui L (2009) Anthrax lethal toxin impairs IL-8 expression in epithelial cells through inhibition of histone H3 modification. *PLoS Pathog* 5(4): e1000359
 41. Bardwell AJ, Abdollahi M, Bardwell L (2004) Anthrax lethal factor-cleavage products of MAPK (mitogen-activated protein kinase) kinases exhibit reduced binding to their cognate MAPKs. *The Biochemical journal* 378(Pt 2):569–577
 42. Gouin E, Adib-Conquy M, Balestrino D, Nahori MA, Villiers V, Colland F, Dramsi S, Dussurget O, Cossart P (2010) The *Listeria monocytogenes* InlC protein interferes with innate immune responses by targeting the I{kappa} B kinase subunit IKK{alpha}. *Proc Natl Acad Sci U S A* 107(40):17333–17338
 43. Orth K, Palmer LE, Bao ZQ, Stewart S, Rudolph AE, Bliska JB, Dixon JE (1999) Inhibition of the mitogen-activated protein kinase kinase superfamily by a *Yersinia* effector. *Science* 285(5435): 1920–1923
 44. Mukherjee S, Keitany G, Li Y, Wang Y, Ball HL, Goldsmith EJ, Orth K (2006) *Yersinia YopJ* acetylates and inhibits kinase activation by blocking phosphorylation. *Science* 312(5777):1211–1214
 45. Paquette N, Conlon J, Sweet C, Rus F, Wilson L, Pereira A, Rosadini CV, Goutagny N, Weber AN, Lane WS, Shaffer SA, Maniatis S, Fitzgerald KA, Stuart L, Silverman N (2012) Serine/threonine acetylation of TGFbeta-activated kinase (TAK1) by *Yersinia pestis YopJ* inhibits innate immune signaling. *Proc Natl Acad Sci U S A* 109(31):12710–12715
 46. Jones RM, Wu H, Wentworth C, Luo L, Collier-Hyams L, Neish AS (2008) Salmonella AvrA coordinates suppression of host

- immune and apoptotic defenses via JNK pathway blockade. *Cell Host Microbe* 3(4):233–244
47. Lin SL, Le TX, Cowen DS (2003) SptP, a *Salmonella typhimurium* type III-secreted protein, inhibits the mitogen-activated protein kinase pathway by inhibiting Raf activation. *Cell Microbiol* 5(4):267–275
 48. Mazurkiewicz P, Thomas J, Thompson JA, Liu M, Arbibe L, Sansonetti P, Holden DW (2008) SpvC is a *Salmonella* effector with phosphothreonine lyase activity on host mitogen-activated protein kinases. *Mol Microbiol* 67(6):1371–1383
 49. Rolhion N, Furniss RC, Grabe G, Ryan A, Liu M, Matthews SA, Holden DW (2016) Inhibition of nuclear transport of NF- κ B p65 by the *Salmonella* type III secretion system effector SpvD. *PLoS Pathog* 12(5):e1005653
 50. Sun H, Kamanova J, Lara-Tejero M, Galan JE (2016) A family of *Salmonella* type III secretion effector proteins selectively targets the NF- κ B signaling pathway to preserve host homeostasis. *PLoS Pathog* 12(3):e1005484
 51. Kramer RW, Slagowski NL, Eze NA, Giddings KS, Morrison MF, Siggers KA, Stambach MN, Lesser CF (2007) Yeast functional genomic screens lead to identification of a role for a bacterial effector in innate immunity regulation. *PLoS Pathog* 3(2):e21
 52. de Jong MF, Liu Z, Chen D, Alto NM (2016) *Shigella flexneri* suppresses NF- κ B activation by inhibiting linear ubiquitin chain ligation. *Nat Microbiol* 1(7):16084
 53. H. Ashida, M. Kim, M. Schmidt-Supprian, A. Ma, M. Ogawa, C. Sasakawa, A bacterial E3 ubiquitin ligase IpaH9.8 targets NEMO/IKK γ to dampen the host NF- κ B-mediated inflammatory response. *Nature cell biology* 12(1) (2010) 66–73; sup pp 1–9
 54. Kim DW, Lenzen G, Page AL, Legrain P, Sansonetti PJ, Parsot C (2005) The *Shigella flexneri* effector OspG interferes with innate immune responses by targeting ubiquitin-conjugating enzymes. *Proc Natl Acad Sci U S A* 102(39):14046–14051
 55. Sanada T, Kim M, Mimuro H, Suzuki M, Ogawa M, Oyama A, Ashida H, Kobayashi T, Koyama T, Nagai S, Shibata Y, Gohda J, Inoue J, Mizushima T, Sasakawa C (2012) The *Shigella flexneri* effector OspI deamidates UBC13 to dampen the inflammatory response. *Nature* 483(7391):623–626
 56. Zhang Y, Muhlen S, Oates CV, Pearson JS, Hartland EL (2016) Identification of a distinct substrate-binding domain in the bacterial cysteine methyltransferase effectors NleE and OspZ. *J Biol Chem* 291(38):20149–20162
 57. Lad SP, Yang G, Scott DA, Wang G, Nair P, Mathison J, Reddy VS, Li E (2007) Chlamydia CT441 is a PDZ domain-containing tail-specific protease that interferes with the NF- κ B pathway of immune response. *J Bacteriol* 189(18):6619–6625
 58. Trosky JE, Li Y, Mukherjee S, Keitany G, Ball H, Orth K (2007) VopA inhibits ATP binding by acetylating the catalytic loop of MAPK kinases. *J Biol Chem* 282(47):34299–34305
 59. Nadler C, Baruch K, Kobi S, Mills E, Haviv G, Farago M, Alkalay I, Bartfeld S, Meyer TF, Ben-Neriah Y, Rosenshine I (2010) The type III secretion effector NleE inhibits NF- κ B activation. *PLoS Pathog* 6(1):e1000743
 60. Yen H, Ooka T, Iguchi A, Hayashi T, Sugimoto N, Tobe T (2010) NleC, a type III secretion protease, compromises NF- κ B activation by targeting p65/RelA. *PLoS Pathog* 6(12):e1001231
 61. Tweten RK (2005) Cholesterol-dependent cytolysins, a family of versatile pore-forming toxins. *Infect Immun* 73(10):6199–6209
 62. Hamon MA, Batsche E, Regnault B, Tham TN, Seveau S, Muchardt C, Cossart P (2007) Histone modifications induced by a family of bacterial toxins. *Proc Natl Acad Sci U S A* 104(33):13467–13472
 63. Hamon MA, Cossart P (2011) K⁺ efflux is required for histone H3 dephosphorylation by *Listeria monocytogenes* listeriolysin O and other pore-forming toxins. *Infect Immun* 79(7):2839–2846
 64. Dortet L, Lombardi C, Cretin F, Dessen A, Filloux A (2018) Pore-forming activity of the *Pseudomonas aeruginosa* type III secretion system translocon alters the host epigenome. *Nat Microbiol* 3(3):378–386
 65. Brennan DF, Barford D (2009) Eliminylation: a post-translational modification catalyzed by phosphothreonine lyases. *Trends Biochem Sci* 34(3):108–114
 66. Li H, Xu H, Zhou Y, Zhang J, Long C, Li S, Chen S, Zhou JM, Shao F (2007) The phosphothreonine lyase activity of a bacterial type III effector family. *Science* 315(5814):1000–1003
 67. Arbibe L, Kim DW, Batsche E, Pedron T, Mateescu B, Muchardt C, Parsot C, Sansonetti PJ (2007) An injected bacterial effector targets chromatin access for transcription factor NF- κ B to alter transcription of host genes involved in immune responses. *Nat Immunol* 8(1):47–56
 68. Harouz H, Rachez C, Meijer BM, Marteyn B, Donnadieu F, Cammas F, Muchardt C, Sansonetti P, Arbibe L (2014) *Shigella flexneri* targets the HP1 gamma subcode through the phosphothreonine lyase OspF. *EMBO J* 33(22):2606–2622
 69. Biernie H, Tham TN, Batsche E, Dumay A, Leguillou M, Kerneis-Golsteyn S, Regnault B, Seeler JS, Muchardt C, Feunteun J, Cossart P (2009) Human BAHD1 promotes heterochromatic gene silencing. *Proc Natl Acad Sci U S A* 106(33):13826–13831
 70. Lebreton A, Cossart P, Biernie H (2012) Bacteria tune interferon responses by playing with chromatin. *Virulence* 3(1):87–91
 71. Lakisic G, Lebreton A, Pourpre R, Wendling O, Libertini E, Radford EJ, Le Guillou M, Champy MF, Wattenhofer-Donze M, Soubigou G, Ait-Si-Ali S, Feunteun J, Sorg T, Coppee JY, Ferguson-Smith AC, Cossart P, Biernie H (2016) Role of the BAHD1 chromatin-repressive complex in placental development and regulation of steroid metabolism. *PLoS Genet* 12(3):e1005898
 72. Eskandarian HA, Impens F, Nahori MA, Soubigou G, Coppee JY, Cossart P, Hamon MA (2013) A role for SIRT2-dependent histone H3K18 deacetylation in bacterial infection. *Science* 341(6145):1238858
 73. Pereira JM, Chevalier C, Chaze T, Gianetto Q, Impens F, Matondo M, Cossart P, Hamon MA (2018) Infection reveals a modification of SIRT2 critical for chromatin association. *Cell Rep* 23(4):1124–1137
 74. Pai RK, Pennini ME, Tobian AA, Canaday DH, Boom WH, Harding CV (2004) Prolonged toll-like receptor signaling by *Mycobacterium tuberculosis* and its 19-kilodalton lipoprotein inhibits gamma interferon-induced regulation of selected genes in macrophages. *Infect Immun* 72(11):6603–6614
 75. Gehring AJ, Dobos KM, Belisle JT, Harding CV, Boom WH (2004) *Mycobacterium tuberculosis* LprG (Rv1411c): a novel TLR-2 ligand that inhibits human macrophage class II MHC antigen processing. *J Immunol* 173(4):2660–2668
 76. Noss EH, Pai RK, Sellati TJ, Radolf JD, Belisle J, Golenbock DT, Boom WH, Harding CV (2001) Toll-like receptor 2-dependent inhibition of macrophage class II MHC expression and antigen processing by 19-kDa lipoprotein of *Mycobacterium tuberculosis*. *J Immunol* 167(2):910–918
 77. Pennini ME, Pai RK, Schultz DC, Boom WH, Harding CV (2006) *Mycobacterium tuberculosis* 19-kDa lipoprotein inhibits IFN-gamma-induced chromatin remodeling of MHC2TA by TLR2 and MAPK signaling. *J Immunol* 176(7):4323–4330
 78. Wang Y, Curry HM, Zwilling BS, Lafuse WP (2005) *Mycobacteria* inhibition of IFN-gamma induced HLA-DR gene expression by up-regulating histone deacetylation at the promoter region in human THP-1 monocytic cells. *J Immunol* 174(9):5687–5694
 79. F. Lan, Y. Shi, Epigenetic regulation: methylation of histone and non-histone proteins. *Science in China. Series C, Life sciences* 52(4) (2009) 311–322

80. Lehnertz B, Ueda Y, Derijck AA, Braunschweig U, Perez-Burgos L, Kubicek S, Chen T, Li E, Jenuwein T, Peters AH (2003) Suv39h-mediated histone H3 lysine 9 methylation directs DNA methylation to major satellite repeats at pericentric heterochromatin. *Current biology* : CB 13(14):1192–1200
81. Sanz LA, Chamberlain S, Sabourin JC, Henckel A, Magnuson T, Hugnot JP, Feil R, Arnaud P (2008) A mono-allelic bivalent chromatin domain controls tissue-specific imprinting at Grb10. *EMBO J* 27(19):2523–2532
82. Cheng X, Zhang X (2007) Structural dynamics of protein lysine methylation and demethylation. *Mutat Res* 618(1–2):102–115
83. Li T, Lu Q, Wang G, Xu H, Huang H, Cai T, Kan B, Ge J, Shao F (2013) SET-domain bacterial effectors target heterochromatin protein 1 to activate host rDNA transcription. *EMBO Rep* 14(8):733–740
84. Pennini ME, Perrinet S, Dautry-Varsat A, Subtil A (2010) Histone methylation by NUE, a novel nuclear effector of the intracellular pathogen *Chlamydia trachomatis*. *PLoS Pathog* 6(7):e1000995
85. Mujtaba S, Winer BY, Jaganathan A, Patel J, Sgobba M, Schuch R, Gupta YK, Haider S, Wang R, Fischetti VA (2013) Anthrax SET protein: a potential virulence determinant that epigenetically represses NF- κ B activation in infected macrophages. *J Biol Chem* 288(32):23458–23472
86. Yaseen I, Kaur P, Nandicoori VK, Khosla S (2015) Mycobacteria modulate host epigenetic machinery by Rv1988 methylation of a non-tail arginine of histone H3. *Nat Commun* 6:8922
87. Casadio F, Lu XD, Pollock SB, LeRoy G, Garcia BA, Muir TW, Roeder RG, Allis CD (2013) H3R42me2a is a histone modification with positive transcriptional effects. *Proc Natl Acad Sci U S A* 110(37):14894–14899
88. Hyland EM, Molina H, Poorey K, Jie C, Xie Z, Dai J, Qian J, Bekiranov S, Auble DT, Pandey A, Boeke JD (2011) An evolutionarily ‘young’ lysine residue in histone H3 attenuates transcriptional output in *Saccharomyces cerevisiae*. *Genes Dev* 25(12):1306–1319
89. L. Jose, R. Ramachandran, R. Bhagavat, R.L. Gomez, A. Chandran, S. Raghunandan, R.V. Omkumar, N. Chandra, S. Mundayoor, R.A. Kumar, Hypothetical protein Rv3423.1 of *Mycobacterium tuberculosis* is a histone acetyltransferase. *The FEBS journal* 283(2) (2016) 265–81
90. Chopra P, Singh B, Singh R, Vohra R, Koul A, Meena LS, Koduri H, Ghildiyal M, Deol P, Das TK, Tyagi AK, Singh Y (2003) Phosphoprotein phosphatase of *Mycobacterium tuberculosis* dephosphorylates serine-threonine kinases PknA and PknB. *Biochem Biophys Res Commun* 311(1):112–120
91. Agarwal S, Agarwal S, Jin H, Pancholi P, Pancholi V (2012) Serine/threonine phosphatase (SP-STP), secreted from *Streptococcus pyogenes*, is a pro-apoptotic protein. *J Biol Chem* 287(12):9147–9167
92. Rolando M, Sanulli S, Rusniok C, Gomez-Valero L, Bertholet C, Sahr T, Margueron R, Buchrieser C (2013) *Legionella pneumophila* effector RomA uniquely modifies host chromatin to repress gene expression and promote intracellular bacterial replication. *Cell Host Microbe* 13(4):395–405
93. Schuhmacher MK, Rolando M, Brohm A, Weirich S, Kudithipudi S, Buchrieser C, Jeltsch A (2018) The *Legionella pneumophila* methyltransferase RomA methylates also non-histone proteins during infection. *J Mol Biol* 430(13):1912–1925
94. Lin H, Su X, He B (2012) Protein lysine acylation and cysteine succination by intermediates of energy metabolism. *ACS Chem Biol* 7(6):947–960
95. Sabari BR, Zhang D, Allis CD, Zhao YM (2017) Metabolic regulation of gene expression through histone acylations. *Nat Rev Mol Cell Bio* 18(2):90–101
96. Wei W, Liu X, Chen J, Gao S, Lu L, Zhang H, Ding G, Wang Z, Chen Z, Shi T, Li J, Yu J, Wong J (2017) Class I histone deacetylases are major histone deacetylases: evidence for critical and broad function of histone crotonylation in transcription. *Cell Res* 27(7):898–915
97. B.R. Sabari, Z.Y. Tang, H. Huang, V. Yong-Gonzalez, H. Molina, H.E. Kong, L.Z. Dai, M. Shimada, J.R. Cross, Y.M. Zhao, R.G. Roeder, C.D. Allis, Intracellular crotonyl-CoA stimulates transcription through p300-catalyzed histone crotonylation. (vol 58, pg 203, 2015), *Mol Cell* 69(3) (2018) 533–533
98. Koh A, De Vadder F, Kovatcheva-Datchary P, Backhed F (2016) From dietary fiber to host physiology: short-chain fatty acids as key bacterial metabolites. *Cell* 165(6):1332–1345
99. Fellows R, Denizot J, Stellato C, Cuomo A, Jain P, Stoyanova E, Balazsi S, Hajnady Z, Liebert A, Kazakevych J, Blackburn H, Correa RO, Fachi JL, Sato FT, Ribeiro WR, Ferreira CM, Perea H, Spagnuolo M, Mattiuz R, Matolcsi C, Guedes J, Clark J, Veldhoen M, Bonaldi T, Vinolo MAR, Varga-Weisz P (2018) Microbiota derived short chain fatty acids promote histone crotonylation in the colon through histone deacetylases. *Nat Commun* 9(1):105
100. Simon JA, Kingston RE (2013) Occupying chromatin: Polycomb mechanisms for getting to genomic targets, stopping transcriptional traffic, and staying put. *Mol Cell* 49(5):808–824
101. Cavaille JM, Adib-Conquy M (2006) Bench-to-bedside review: endotoxin tolerance as a model of leukocyte reprogramming in sepsis. *Crit Care* 10(5)
102. Chan C, Li L, McCall CE, Yoza BK (2005) Endotoxin tolerance disrupts chromatin remodeling and NF- κ B transactivation at the IL-1 β promoter. *J Immunol* 175(1):461–468
103. El Gazzar M, Yoza BK, Hu JY, Cousart SL, McCall CE (2007) Epigenetic silencing of tumor necrosis factor alpha during endotoxin tolerance. *J Biol Chem* 282(37):26857–26864
104. Chen X, El Gazzar M, Yoza BK, McCall CE (2009) The NF- κ B factor RelB and histone H3 lysine methyltransferase G9a directly interact to generate epigenetic silencing in endotoxin tolerance. *J Biol Chem* 284(41):27857–27865
105. B. Novakovic, E. Habibi, S.Y. Wang, R.J.W. Arts, R. Davar, W. Megchelenbrink, B. Kim, T. Kuznetsova, M. Kox, J. Zwaag, F. Matarese, S.J. van Heeringen, E.M. Janssen-Megens, N. Sharifi, C. Wang, F. Keramati, V. Schoonenberg, P. Flicek, L. Clarke, P. Pickkers, S. Heath, I. Gut, M.G. Netea, J.H.A. Martens, C. Logie, H.G. Stunnenberg, Beta-Glucan reverses the epigenetic state of LPS-induced immunological tolerance. *Cell* 167(5) (2016) 1354–
106. Netea MG, Quintin J, van der Meer JW (2011) Trained immunity: a memory for innate host defense. *Cell Host Microbe* 9(5):355–361
107. Yoshida K, Maekawa T, Zhu Y, Renard-Guillet C, Chatton B, Inoue K, Uchiyama T, Ishibashi K, Yamada T, Ohno N, Shirahige K, Okada-Hatakeyama M, Ishii S (2015) The transcription factor ATF7 mediates lipopolysaccharide-induced epigenetic changes in macrophages involved in innate immunological memory. *Nat Immunol* 16(10):1034–1043
108. Quintin J, Saeed S, Martens JHA, Giamarellos-Bourboulis EJ, Ifrim DC, Logie C, Jacobs L, Jansen T, Kullberg BJ, Wijmenga C, Joosten LAB, Xavier RJ, van der Meer JWM, Stunnenberg HG, Netea MG (2012) *Candida albicans* infection affords protection against reinfection via functional reprogramming of monocytes. *Cell Host Microbe* 12(2):223–232
109. Ostuni R, Piccolo V, Barozzi I, Polletti S, Termanini A, Bonifacio S, Curina A, Prosperini E, Ghisletti S, Natoli G (2013) Latent enhancers activated by stimulation in differentiated cells. *Cell* 152(1–2):157–171
110. Rasid O, Chevalier C, Camarasa T, Fitting C, Cavaille J-M, Hamon MA (2019) H3K4me1 supports memory-like NK cells induced by systemic inflammation. *Cell Rep* 29(12):3933–3945