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Review Article

Roles of post-translational modifications of C-type lectin receptor-induced signaling cascades in innate immune responses against *Candida albicans*

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Highlights

- Risk of invasive *candida* infection and its related mortality are increasing significantly in perioperative patients.
- C-type lectin receptors are the primary pattern-recognition receptors for fungi-induced host defense and innate immune activation.
- Protein post-translational modifications are one of the core factors in host innate immune regulation.
- Post-translational modifications sites on proteins are anticipated to serve as potential targets for modulating anti-fungal immunity.

Abstract

Candida albicans (*C. albicans*), a conditional pathogenic fungus, is widespread in nature and can live in symbiosis with organisms in small quantities. When the normal microflora is imbalanced, the epithelial barrier is disrupted or the immune system becomes dysfunctional, *C. albicans* can change from commensal to pathogenic pathogen, causing both superficial and life-threatening systemic infections with no effective treatment. The morbidity and mortality of invasive *Candida* infections in perioperative patients are high due to underlying chronic diseases, immune deficiencies, and pathophysiological disorders. C-type lectin receptors (CLRs) are the main pattern-recognition receptors for fungal activation of innate immunity and host defense. Upon binding to ligands, CLRs induce multiple signal transduction cascades followed by activation of nuclear factor kappa B through spleen tyrosine kinase - and caspase recruitment domain containing protein 9-dependent pathways. Analyzing the effects of regulatory CLR-induced signaling cascades on host immune cells is critical for understanding the molecular mechanism in regulating antifungal immunity. As one of the core factors in host innate immune regulation, protein post-translational modifications regulate the strength of immune effects by modulating protein conformation, stability, affinity, subcellular localization, etc. This makes the post-translational modification sites promising as potential targets for modulating antifungal immunity. This review primarily described the study progress of post-translational modifications in controlling CLR-induced signaling cascades throughout the process of innate immunity against *C. albicans*. We aim to provide better understanding of these mechanisms and aid in the identification and development of biomarkers and drug targets for invasive candidiasis.

Keywords: *Candida albicans*, innate immunity, protein post-translational modifications, C-type lectin receptors

Introduction

The normal human body has a highly sophisticated innate and adaptive immune system that is naturally resistant to most fungal infections, with over 99% of *Candida albicans* (*C. albicans*) being cleared within the first hour of entry into the human bloodstream [1]. However, fungal infections have become a major cause of human diseases due to the increasing number

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of immune-compromised population having acquired immune deficiency syndrome, chemotherapy for tumors, organ transplants, post-immunosuppressive therapy and advanced age [2]. Hundreds of millions of patients worldwide are infected with pathogenic fungi each year, resulting in at least 1.5 million deaths per year, similar to the number of deaths due to tuberculosis [3]. *C. albicans* is the most common fungal pathogen, accounting for more than half of the invasive candidiasis [4].

In perioperative patients, various factors can increase the risk of *C. albicans* infection. First, operation compromise the immune function of body by inhibiting the activity of natural killer cells within hours of the surgery, and this inhibition can persist over time [5]. Natural killer cells are a significant component of innate immune system, play an important role in resistance to fungal infection, and kill fungi by releasing soluble cytotoxic molecules (perforin and granzyme) or activating apoptosis pathway [6, 7]. Second, perioperative pain induced by inflammatory factors can restrain immune function by interactions among the central nervous system, hypothalamic-pituitary-adrenal axis pathways, and immune system [8]. Third, critical patients undergoing surgery often need central venous indwelling catheter or peripheral venous indwelling catheter. Fungi can colonize in venous catheters, enter the blood, and cause bloodstream infections, threatening the safety of patients' lives [9, 10]. Last, longer hospitalization and intensive care unit stays undoubtedly increase the risk of infection. Fungal infection is a momentous factor for intensive care unit mortality [11].

Innate immunity plays a crucial role as the host's primary defense against invading pathogens, relying on intricate and dynamic interactions among various cellular and molecular components. Distinguishing between pathogens and self-components is a fundamental function of the innate immune system. Pattern recognition receptors (PRRs) recognize pathogen-associated molecular patterns. These PRRs are mainly expressed by myeloid macrophages including Toll-like receptors (TLRs), C-type lectin receptors (CLRs), nucleotide-binding oligomerization domain like receptors , retinoic acid-inducible gene 1-like receptors, and complement components and receptors [12]. All of these PRRs can mediate antifungal immunity, and CLRs are the most widely studied. The pathogen-associated molecular patterns mainly consist of *Candida* cell wall-associated glycans (mannans, glucans, chitin) [13].

CLRs are the main PRRs for fungi-induced activation of innate immunity and host defense [14]. The receptors dectin-1, dectin-2, dectin-3, and mincle bind to ligands, then recruit spleen tyrosine kinase (SYK) intracellularly, and are activated by an intermolecular auto-phosphorylation mechanism [15]. Activated SYK further activates dedicator of cytokinesis 2, phospholipase C-γ2, and protein kinase C-δ by phosphorylation, the latter phosphorylates caspase recruitment domain containing protein 9 (CARD9) and vesicles containing glucose transporter 1 (GLUT1). Phosphorylation of vesicles promotes GLUT1 translocation to the cytosol and thus increases glucose uptake by immune cells. SYK and CARD9 are transported from the cytosol to the cytoplasm via α-tubulin. Then, CARD9, B cell lymphoma 10, and mucosa-associated lymphoid tissue lymphoma translocation protein 1 together constitute the CBM complex. This complex ultimately leads to the activation of downstream nuclear factor kappa B (NF-κB) and mitogen-activated protein kinases [16]. These signaling pathways turn on a series of effector mechanisms, including the release of proinflammatory cytokines and phagocytosis, finally leading to fungus clearance [17].

Proteins, the essential components of living organisms, are the material basis of life and are involved in practically all cellular processes. To date, more than 50% of the proteins in human cells have been found to have different types of post-translational modifications (PTMs) [18]. PTMs can rapidly regulate various intracellular activities and expand the diversity of protein functions by affecting protein activity, stability, localization, and signal transduction under physiological and pathological conditions [19, 20], such as classical modifications including phosphorylation, ubiquitination, and methylation, as well as non-classical glycosylation, acetylation, malonylation, and succinylation [21]. During the anti-fungal process of innate immune cells, many proteins in the signaling pathway exhibit additional functions through PTMs, and these related enzymes involved in PTMs can indirectly and specifically regulate the anti-fungal ability of immune cells. Whether positively or negatively regulated, these modification-related enzymes have the potential to become targets for modulating the immune system. Therefore, this review focuses on how PTMs are involved in the innate immune response against *C. albicans*, primarily by regulating CLR-induced signaling cascades through ubiquitination, phosphorylation, acetylation, and glycosylation.
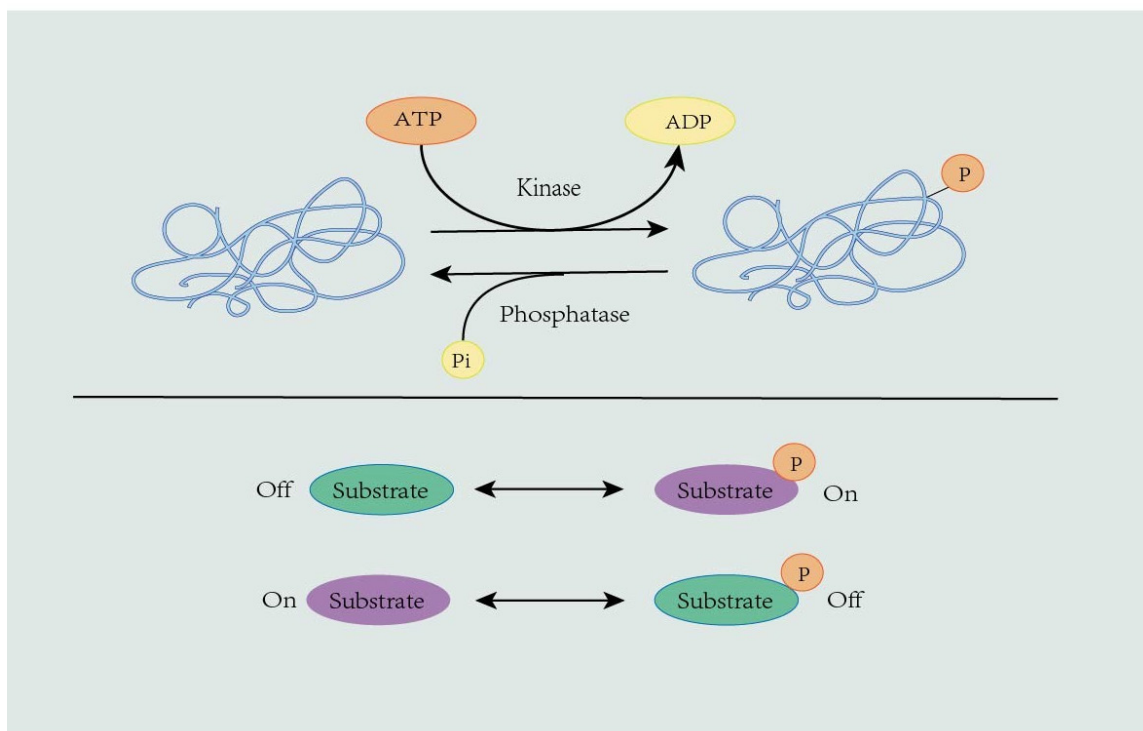
Figure 1. Phosphorylation and dephosphorylation. Phosphorylation and dephosphorylation are mediated by protein kinase and phosphatase, respectively. The phosphate group acts as a molecular switch that can be activated by phosphorylation or dephosphorylation of many substances.

Phosphorylation and dephosphorylation

In organisms, phosphorylation is the most widespread form of covalent modification in PTMs [22]. Phosphorylation plays an important regulatory role in the proper functioning of proteins by the transfer of the γ-position phosphate group of ATP or GTP to the amino acid residues of the substrate protein (mainly serine and threonine) catalyzed by phosphokinase, while the reverse process involves the removal of the corresponding phosphate group by protein kinase [23]. It is the opposing roles of these two enzymes and the energy consumption and production involved that make phosphorylation the preferred mode of regulation of many physiological activities in the body (e.g., transcriptional regulation, signal transduction, DNA damage repair) (**Figure 1**) [24].

Phosphorylation

EPH receptor B2 (EPHB2)

EPHB2 is a member of the EPH receptor family of receptor tyrosine kinase transmembrane glycoproteins. EPHB2 was recently found to directly recognize the fungal cell wall component β-glucan and to synergize with dectin-1 to activate downstream signaling. EPHB2 directly phosphorylates the downstream kinase SYK and promotes its activation. In EPHB2$^{-/-}$ mac-

rophages, fungus-stimulated phosphorylation of SYK was almost completely lost. Ephb2-deficient mice were significantly more susceptible to *C. albicans*–induced infection than the WT mice [25]. EPHB2 also phosphorylates T-cell-activated Rho GTPase-activating protein at the Y310 site, and T-cell-activated Rho GTPase-activating protein acts as an adapter to regulate upstream EPHB2 and downstream CARD9 signaling [26]. In summary, EPHB2 regulates dectin-mediated immune signaling by modulating the phosphorylation of SYK and T-cell-activated Rho GTPase-activating protein during antifungal host defense.

SYK

SYK includes two N-terminal Src Homology 2 domains, one C-terminal kinase domain and two interdomain linkers [27]. These structural domains are bound together, leaving the Src Homology 2 domain in a stable inactivation state [28]. Phosphorylation is the most common method to activate SYK, and there are 10 autophosphorylation sites in SYK [29]. CLRs engagement promotes Src-dependent phosphorylation of its immunoreceptor tyrosine-based activation motif, recruits the SHP-2 tyrosine phosphatase, and activates SYK [30, 31]. This binding leads to conformational changes and unfolding of SYK, which is subsequently activated by SYK's own phosphorylation or by

phosphorylation of its upstream kinases [29]. SYK that acquires enzymatic activity activates its downstream substrate proteins through phosphorylation or even direct interaction, and transduces its downstream signaling cascade. Activated SYK phosphorylates dedicator of cytokinesis 2 at tyrosine 985 and tyrosine 1405, promoting the recruitment and activation of Rac GTPase, thereby increasing reactive oxygen species (ROS) production required for macrophage signaling activation and bactericidal activity [32].

Protein kinase C-δ (PKC-δ)

PKC-δ is activated under dectin-SYK signaling, mediates CARD9 phosphorylation at Thr231, and is involved in CBM complex assembly and classical NF-κB regulation. PKC-δ$^{-/-}$ dendritic cells are defective in innate response to dectin-1, dectin-2, or mincle stimulation, while dendritic cells lacking PKC-α, PKC-β, or PKC-θ are immunocompetent. In addition, C. albicans significantly reduces the production of cytokines, such as tumor necrosis factor, interleukin (IL)-10, and IL-2, in PKC-δ$^{-/-}$ cells compared to wild type. Similarly, RKC-δ$^{-/-}$ mice are highly susceptible to fungal infections. Thus, PKC-δ is an important link between SYK activation and CARD9 signaling in CLR-mediated natural immunity and host protection [33]. On the other hand, PKC-δ promotes GLUT1 insertion into the cell membrane by phosphorylating vesicles bearing GLUT1, which promotes glucose uptake by neutrophils, thereby enhancing the neutrophil metabolic fitness [34].

Dephosphorylation

Steroid sulfatase (STS)

STS proteins are homologous protein phosphatases that share over-lapping functions, and are negative regulators of multiple signaling pathways [35]. For example, in T cells, STS phosphatase helps set the threshold for T cell activation by targeting Zap-70, an important kinase downstream of the T cell receptor [36]. In addition, STS-1 has been shown to control signaling downstream of glycoprotein VI-Fc receptor γ in platelets and Fcepsilon receptor in mast cells by targeting the ZAP-70 homolog SYK [37]. At 12 to 18 h after invasive *Candida* infection, the fungal burden in the kidneys of STS$^{-/-}$ mice began to decrease compared with that in wild-type mice, and a large number of leukocytes in STS$^{-/-}$ mice entered the kidneys to fight the infection, improving the survival rate of the mice. After stimulation of bone marrow-derived dendritic cells with fungal ligands, we observed no difference in other antifungal responses, such as cytokine or nitric oxide production, by STS$^{-/-}$BMDCs but enhanced ROS production. Although the signaling pathway from dectin to the initiation of the ROS response has not been fully elucidated, the involvement of SYK kinas has been identified [38]. Hyperphosphorylation of SYK in STS$^{-/-}$BMDCs was observed using a phosphor-specific antibody that recognizes the activation of SYK tyrosine phosphorylation. STS may be involved in antifungal immunity by regulating the level of SYK phosphorylation and activation, and thus ROS production [39].

Protein phosphatase 1 (PP1)

PP1 to CARD9 keeps CARD9 in a dephosphorylated and self-inhibited state, thereby inhibiting CBM complex formation and suppressing downstream NF-κB and JNK signaling. PP1 does not act directly on CARD9 but requires downstream mediation of kinase 3 (DOK3). DOK3 is a junctional molecule preferentially expressed in hematopoietic cells [40]. It acts as a specific regulator of downstream of several immune receptors, including TLR3, TLR4, and B-cell antigen receptor, during viral infection, endotoxin stimulation, and plasma cell differentiation [41-43]. In invasive fungal infections, DOK3 acts as an intermediate molecule that promotes the recruitment of PP1 to CARD9. Deletion of DOK3 enhances various antifungal effector functions of neutrophils, including phagocytosis, extracellular trap network, and pro-inflammatory cytokine production, through dephosphorylation of CARD9 by PP1, thereby increasing the fungicidal activity of neutrophils [44]. PKC-δ and PP1 keep CARD9 in a balanced activation state through phosphorylation and dephosphorylation to avoid excessive inflammatory immune response and immunosuppression (**Figure 2**).

Ubiquitination and Deubiquitination

Ubiquitin is a protein consisting of 76 amino acids and modify target proteins by mono-ubiquitination or polyubiquitination [45]. A single ubiquitin molecule is repeatedly attached to a ubiquitin lysine residue to form a ubiquitin chain. The ubiquitin molecule itself contains seven lysine residues, all of which can be ubiquitinated, and the N-terminus can also be attached to ubiquitin to form eight types of ubiquitin chains (K6, K11, K27, K29, K33, K48, K63 and MET1) [46]. Ubiquitination, an important PTM modality, is the process of ubiquitin binding to substrate molecules catalyzed by the ubiquitin-proteasome system. This process is facilitated by three enzymes: ubiquitin-activating enzyme, ubiquitin-conjugating enzyme (E2),
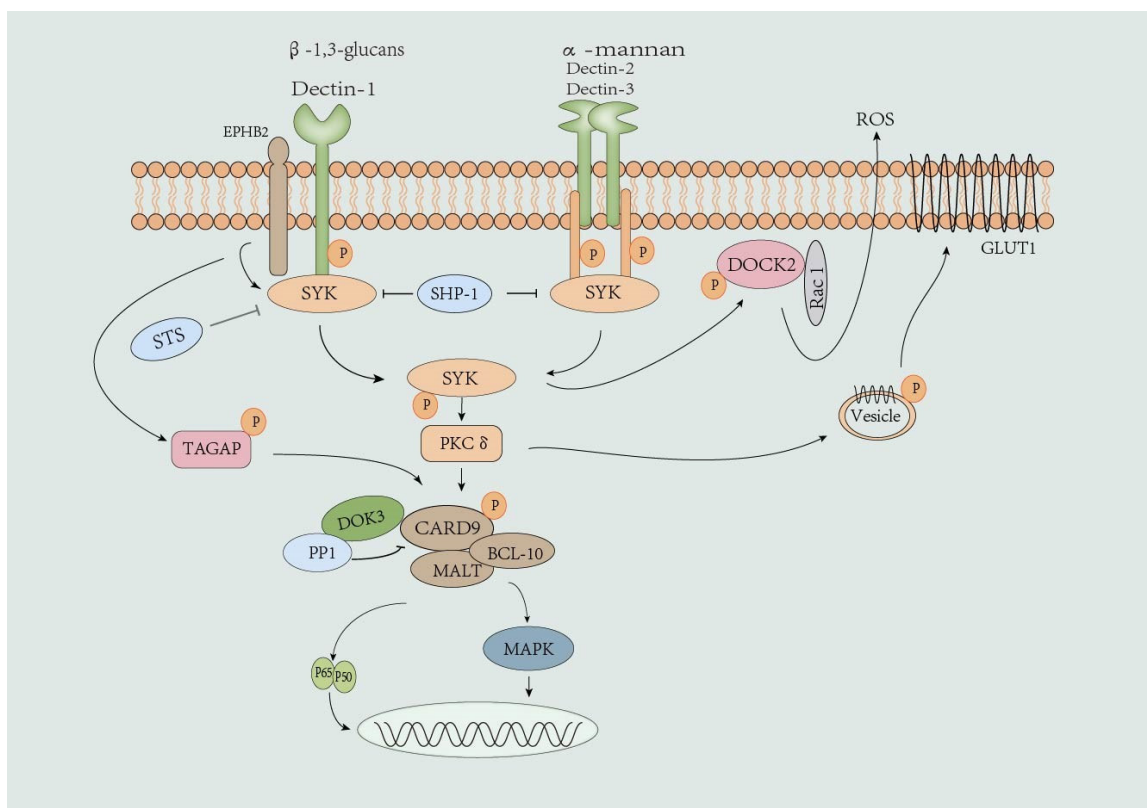
Figure 2. The role of phosphorylation and dephosphorylation in innate immunity against *C. albicans* infection. STS and SHP-1 inactivate SYK by mediating its dephosphorylation. Activated SYK stimulates DOCK2 phosphorylation and thus increases ROS production. PKC-δ activates CARD9 and vesicles containing GLUT1 by directly promoting its phosphorylation. Phosphorylation of vesicles promotes GLUT1 translocation to the cytosol and thus increases glucose uptake by immune cells. DOK3 mediates the recruitment of PP1 to CARD9, causing dephosphorylation and inactivation of CARD9. *C. albicans*, *Candida albicans*; EPHB2, EPH receptor B2; ROS, reactive oxygen species; STS, steroid sulfatase; SYK, spleen tyrosine kinase; DOCK2, dedicator of cytokinesis 2; GLUT1, glucose transporter 1; TAGAP, T-cell-activated Rho GTPase-activating protein; PKC-δ, protein kinase C-δ; CARD9, caspase recruitment domain containing protein 9; DOK3, downstream of kinase 3; PP1, protein phosphatase 1; MALT, mucosa-associated lymphoid tissue; MAPK, mitogen-activated protein kinases.

and ubiquitin ligase (E3). Ubiquitin-activating enzyme activates ubiquitin, E2 transfers ubiquitin to E3, and finally, E3 catalyzes the covalent attachment of ubiquitin to the target protein, or in some cases, E3 directly adds ubiquitin from E2 to the substrate [47]. Of these, E3 determines the specificity and precision of ubiquitin and substrate attachment, and the human genome has approximately more than 600 E3s, significantly more than 2 ubiquitin-activating enzymes and approximately 40 E2s. Thus, ubiquitination is more complex and functionally diverse than other protein modifications [48]. E3 ubiquitin ligases are divided into three families: the RING finger-type, HECT-type, and RING-between-RING [49]. At the same time, the human genome also has about 100 deubiquitinases, which are classified into 7 classes: USP, UCH, OUT, MJD, JAMM, MCPIP, and MINDY. These deubiquitinases are also highly specific and responsible for removing ubiquitin chains from proteins and other molecules (**Figure 3**) [50].

Ubiquitination

Casitas B-lineage lymphoma protein b (CBL-B)

CBL-B is one of three homologous proteins of the Cbl E3 ubiquitin ligase family that is ubiquitously expressed in all leukocyte subpopulations and negatively regulates the activation of signaling pathways, such as T-cell antigen receptor, B-cell antigen receptor, CD28, TLR4, high-affinity IgE receptor, and epidermal growth factor receptor [51-55]. During invasive *C. albicans* infection, CBL-B$^{-/-}$ mice were found to be highly resistant to disseminated candidiasis, with resistance characterized by reduced weight loss following infection, an absence of inflammatory damage, and enhanced survival. CBL-B$^{-/-}$ mice displayed enhanced fungal clearance within 24-48 hours of bloodstream infection, with a remarkable 10-fold reduction in fungal load observed in different peripheral organs when compared to control group [56]. CBL-B$^{-/-}$ bone marrow-derived myeloid mononuclear macrophages exhibited reduced ligand-mediated receptor internalization and degradation, increased dectin receptor expression, and
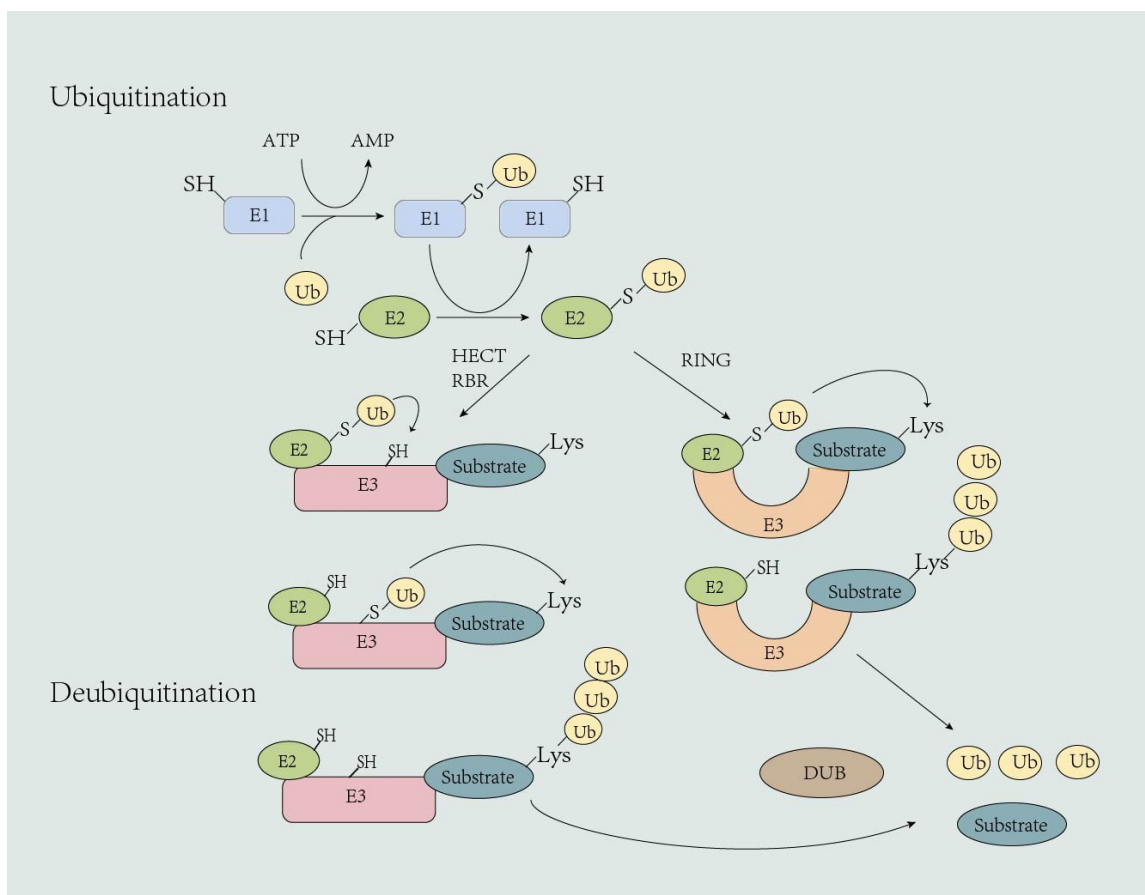
Figure 3. Ubiquitination and Deubiquitination. Ubiquitination is mainly mediated by E1, E2, E3. E1 first activates ubiquitin, then E2 transfers ubiquitin to E3, and finally E3 catalyzes the covalent binding of ubiquitin to the target protein, or E3 mediates the direct addition of ubiquitin from E2 to the substrate. De-ubiquitination is the removal process of ubiquitin from the substrate by deubiquitinating enzymes. E1, ubiquitin-activating enzyme; E2, ubiquitin-conjugating enzyme; E3, ubiquitin ligase; Ub, Ubiquitin; RBR, RING-between-RING; DUB, deubiquitinating enzyme.

elevated production of various pro-inflammatory factors and ROS compared to wild type macrophages [56]. This is because CBL-B can mediate the ubiquitination of these activated CLRs through the mutual binding of the junction protein Fc receptor γ and the tyrosine kinase SYK. Dectin-1 (K2, K27, and K34), dectin-2 (K10 and K12), and dectin-3 (K9) are the sites of ubiquitination of dectin-1, -2, -3, respectively [57, 58], and the ubiquitinated CLRs are sorted into lysosomes by the transport-essential endosomal sorting complex system for degradation, thereby negatively regulating the CLR-mediated innate immune response against fungal infections.

C- Casitas B-lineage lymphoma (C-CBL)

C-CBL, a CBL-B homologous family member, modulates intestinal inflammation by inhibiting fungal-induced non-classical NF-κB activation. Intestinal fungal-derived mannan activates C-CBL in dendritic cells via dectin-2 and dectin-3, thereby promoting C-CBL-mediated ubiquitination and degradation of RelB, a non-clas-

sical NF-κB family member. Meanwhile, the classical NF-κB family member p65 mediates the transcription of the anti-inflammatory cytokine IL-10 gene to suppress colitis, and RelB binds to p65 and inhibits p65-mediated production of IL-10. Thus, the lack of C-CBL in dendritic cells promotes mannan-induced activation of RelB, which binds to p65, thereby inhibiting p65-mediated IL-10 transcription, decreasing anti-inflammatory cytokines, exaggerating immunization, and making mice more susceptible to Dextran Sulfate Sodium Salt-induced colitis [59].

Tripartite motif containing 31 (TRIM31)

Up to now, nearly 80 TRIM family proteins have been identified in the human genome, many of which have been shown to have E3 ubiquitin ligase activity and are involved in various activities of the organism [60]. For example, TRIM31, a member of the TRIM/RBCC family and a RING finger E3 ubiquitin ligase, is involved in the regulation of viral and fungal infections, non-alcoholic fatty liver disease, hypertensive

nephropathy, ischemic brain injury, and other diseases [61-65]. TRIM31 also binds directly to nucleotide-binding oligomerization domain -like receptor thermal protein domain associated protein 3 (NLRP3), and K48-linked ubiquitination mediates protein degradation of NLRP3. TRIM31 can limit NLRP3 inflammasome activation under physiological conditions and is, therefore, expected to be a potential therapeutic target for NLRP3 inflammasome related diseases [66]. Recently, TRIM31 was found to be a key regulator of SYK activation. RIM31 interacts with SYK and catalyzes K27-linked polyubiquitination at the Lys375 and Lys517 sites of SYK. This process promotes translocation of SYK to the plasma membrane, enabling its binding to CLRs. Additionally, TRIM31 inhibits dephosphorylation of SYK by the phosphatase SHP-1, which in turn enhances SYK-mediated downstream signaling (CBM complex, inflammasome formation). In vitro TRIM31 deficient BMDCs and BMDMs significantly reduced the production of pro-inflammatory cytokines such as IL-6, tumor necrosis factor-α, IL-1β, IL-12, and IL-23. Similarly, TRIM31$^{-/-}$ mice were more susceptible to fungal infection, and demonstrated a more severe renal inflammatory response and lower survival rates than TRIM31$^{+/+}$ mice [62].

TRIM62

CARD9 is a central component of natural immune signaling against fungi mediated by CLRs, and when not activated, CARD9 is in a self-inhibited state. TRIM62 mediates polyubiquitination of CARD9 at the K27-linked at the Lys125 site of CARD9, disrupting the inhibitory state of CARD9 and enhancing the transduction of signaling pathways in which CARD9 is involved. Thus, similar with CARD9$^{-/-}$ mice, TRIM62$^{-/-}$ mice have an increased susceptibility to fungal infections [67]. Genetic sequencing of patients with inflammatory bowel disease showed that the protective CARD9 variants were not ubiquitinated by TRIM62 and suggested that the protective effect of C-terminal truncation might be mediated by the loss of TRIM62 interactions, thereby limiting the pro-inflammatory cytokine response. If the CARD9-TRIM62 interaction is blocked, the inflammatory response can be limited. The small-molecule inhibitors that block the CARD9-TRIM62 interaction have been developed and hold potential as therapeutic agents for the treatment of inflammatory bowel disease [68].

Neuronal precursor cell-expressed developmentally down-regulated 4 (NEDD4)

NEDD4 is a HECT type E3 ubiquitin ligase that has been shown to positively regulate T cell activation and proliferation [69]. Additionally, it has also been reported that Nedd4 is involved in anti-cellular bacterial clearance by promoting autophagy [70]. Nedd4$^{-/-}$ mice are highly susceptible to systemic *C. albicans* infection, which is associated with increased organ fungal load, defective inflammatory response, impaired recruitment of leukocytes to the kidney, and impaired granulocyte expression of ROS. The specific regulatory mechanism of Nedd4 is unclear. At a molecular level, Nedd4$^{-/-}$ macrophages showed reduced TGFβ-activated kinase 1 and NF-κB activity and normal SYK and PKC-δ activity during *C. albicans* infection. This indicates that NEDD4 regulates downstream of PKC-δ and upstream of TGFβ-activated kinase 1 to enhance immune cell killing against *C. albicans* [71].

Deubiquitination

Ubiquitin-proteasome system 15

The ubiquitin-specific proteases family comprises the largest number of deubiquitinating enzymes. During antifungal immunization, ubiquitin-specific protease15 removes TRIM62-mediated ubiquitination of CARD9 and inhibits the activation of CARD9, thereby inhibiting the formation of the CBM complex. Thus, the activation of CARD9 is co-regulated by ubiquitin-specific protease15 and TRIM62, so that the degree of activation is in a state of equilibrium [72].

Ovarian tumor deubiquitinase 1 (OTUD1)

OTUD1 is also an important regulator of CARD9. OTUD1 mainly removes K29, K33 and K63-linked polyubiquitination from CARD9 and promotes the formation of the CBM complex. OTUD1 deficiency reduces CARD9-mediated signaling, leading to reduced production of pro-inflammatory cytokines and chemokines. Meanwhile, OTUD1$^{-/-}$ mice have been found to have increased susceptibility to fungal infections [73]. In other studies, OTUD1 exerted tumor suppression by removing K63-linked ubiquitination on Yes-associated protein and inhibiting the degradation of P53 [74]. In addition, OTUD1 negatively regulated the RNA virus signaling pathway by targeting Smad ubiquitination regulatory factor 1 (Smurf1) (**Figure 4**) [75].

Acetylation and Deacetylation

Lysine acetylation is a conserved PTM, most commonly histone acetylation, and is usually regulated by acetyltransferase and deacetyl-
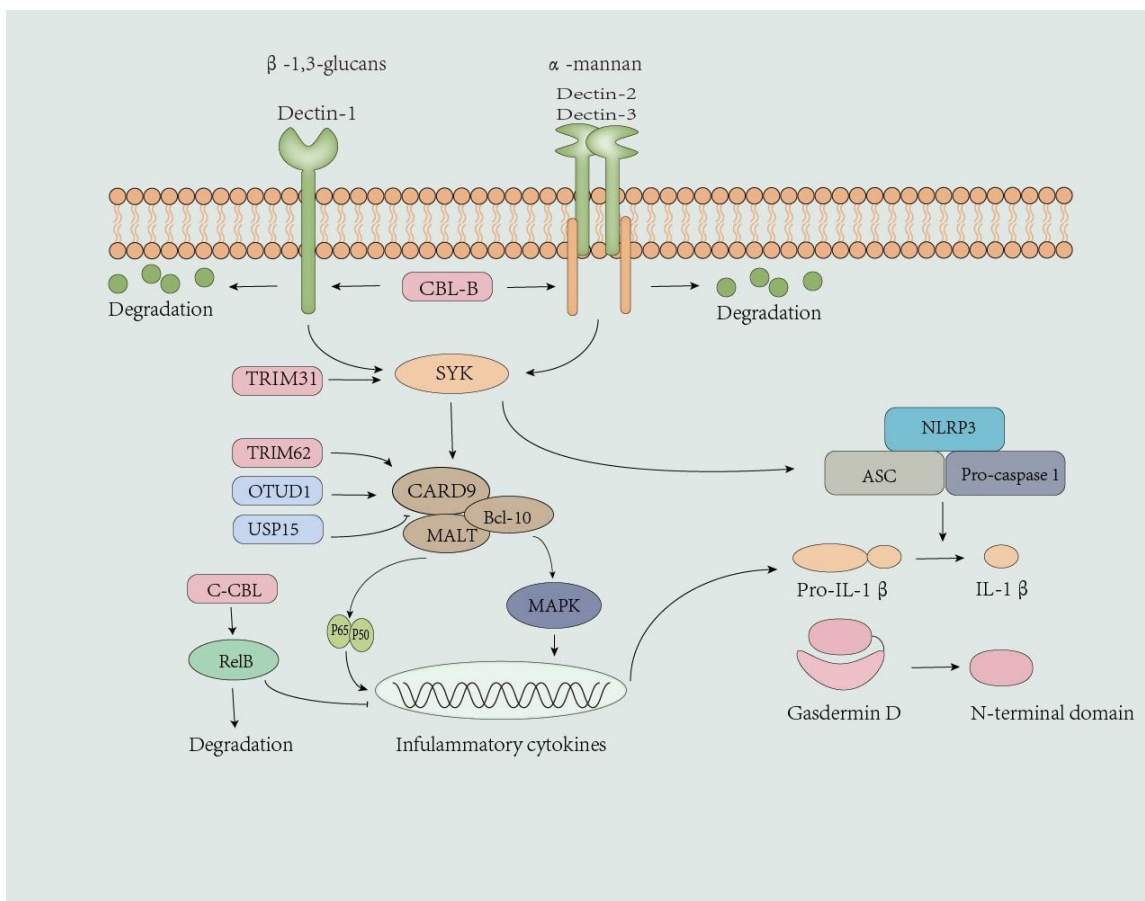
Figure 4. The role of ubiquitination and deubiquitination in innate immunity against *C. albicans* infection. Multiple E3 ubiquitin ligases are involved in the regulation of innate immunity against *C. albicans*. CBL-B and C-CBL promote the degradation of dectin-1, -2, -3 and RelB, respectively; TRIM31 facilitates SYK to immunoreceptor tyrosine-based activation motif translocation; TRIM62, OTUD1 and USP15 all act on CARD9 to maintain a balanced level of CARD9 activation through ubiquitination and deubiquitination. *C. albicans*, *Candida albicans*; CBL-B, casitas B-lineage lymphoma protein b; TRIM, tripartite motif containing; SYK, spleen tyrosine kinase; OTUD1, ovarian tumor deubiquitinase 1; CARD9, caspase recruitment domain containing protein 9; USP15, ubiquitin-specific protease 15; NLRP3, nucleotide-binding oligomerization domain -like receptor thermal protein domain associated protein 3; C-CBL, c-Casitas B-lineage lymphoma.

transferase [76]. Acetylation modifications usually occur in the structural domains of proteins, such as α-helix and β-fold, and the proteins with acetylation modifications are usually conserved, such as metabolism-related enzymes, ribosomes, and molecular chaperones [77]. With the continuous research on protein acetylation, studies have demonstrated that acetylation also plays an important regulatory role in the antifungal immune process [78, 79].

Acetylation

AP2A1 and α-tubulin N-acetyltransferase 1

AP2A1 and α-tubulin N-acetyltransferase 1 are recruited to α-tubulin via Myosin 1F (MYO1F), which promotes acetylation of α-tubulin. SYK and CARD9 molecules then activate downstream signaling pathways through acetylated tubules from the cell membrane into the cytoplasm. MYO1F is an unconventional myosin

expressed mainly in mammalian immune cells. It plays a crucial role in regulating the migration of mast cells and neutrophils [80, 81], and modulating M1 polarization by stimulating intercellular adhesion of macrophages [82]. MYO1F also plays a key role in the activation of antifungal natural immune signaling and is required for dectin-induced acetylation of α-tubulin. Following systemic *Candida* infection, MYO1F-deficient mice were more severely infected relative to wild-type mice. Administration of AGK2 or AK-1, inhibitors of the deacetylase SIRT2, promoted dectin-activated signaling and proinflammatory gene expression, and produced a protective effect in mice with systemic *Candida* infection [78].

Deacetylation

Histone deacetylase 11 (HDAC11)

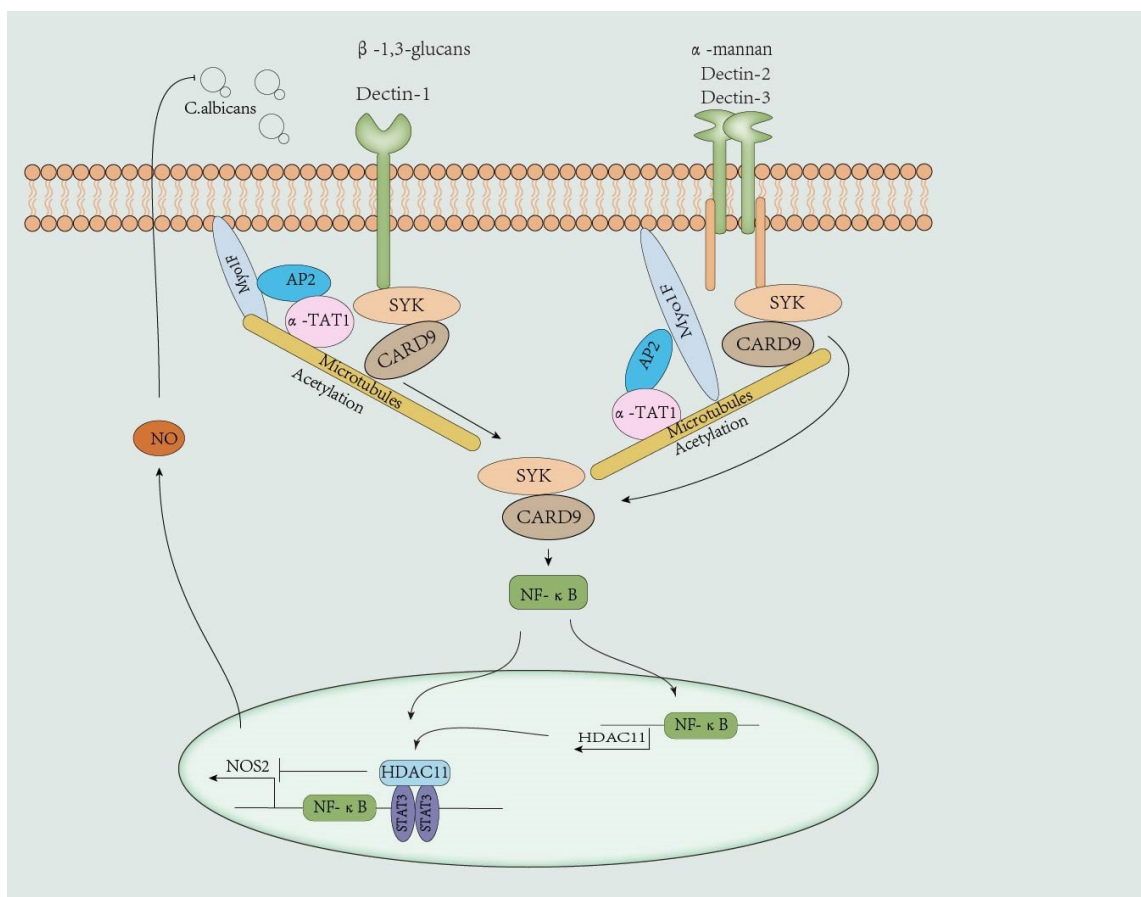HDACs are enzymes that catalyze the removal

Figure 5. The role of acetylation and deacetylation in innate immunity against *C. albicans* infection. MYO1F mediates microtubule acetylation through recruitment of Tubulin and the AP2A1/αTAT complex, while SYK and CARD9 molecules activate downstream signaling pathways through acetylated microtubules from the cell membrane into the cytoplasm. The deacetylase HDAC11 promotes fungal immune escape by interacting with the transcription factor STAT3 and using STAT3 as a scaffold to bind to the iNOS gene promoter, inhibiting iNOS expression and reducing NO production. *C. albicans*, *Candida albicans*; AP, adaptor protein; SYK, spleen tyrosine kinase; αTAT1, α-tubulin N-acetyltransferase 1; CARD9, caspase recruitment domain containing protein 9; NF-κB, nuclear factor kappa B; HDAC11, histone deacetylase 11; STAT3, signal transducer and activator of transcription 3; iNOS, inducible nitric oxide synthase.

of acetyl functional groups from the lysine residues of both histone and nonhistone proteins [83]. Based on structure and function, HDACs are classified into four groups: class I HDACs (HDAC1, 2, 3 and 8), class II HDACs (HDAC4, 5, 6, 7, 9 and 10), class III HDACs (SIRT1–SIRT7) and class IV HDAC (HDAC11) [84]. As the only class IV HDAC, HDAC11 plays a critical role in a variety of cellular events, including cell proliferation and differentiation, metabolism, and tumorigenesis [85-87]. Loss of HDAC11 increases the acetylation of histone 3, 4 at the nitric oxide synthase 2 promoter, and leads to enhanced Nos2 transcription and corresponding inducible nitric oxide synthase levels in macrophages. The transcriptional repressor of Nos2, signal transducer and activator of transcription 3, interacts with HDAC11 and acts as a scaffolding protein to facilitate the binding of HDAC11 to the Nos2 promoter. Inhibition of signal transducer and activator of transcription 3 significantly reduced the aggregation of HDAC11 on the Nos2 pro-

moter. Similarly, HDAC11 deletion reduced the abundance of signal transducer and activator of transcription 3 binding on the Nos2 promoter. Thus, fungal pathogens stimulate HDAC$^{-/-}$ mouse macrophages to produce more NO and ROS. HDAC11 inhibitor FT895 shows antifungal therapeutic effects in both *C. albicans* infected mice and human cells (**Figure 5**) [79].

Glycosylation

In the human immune system, all immunoglobulins and most complement components are glycosylated and depending on the glycosidic bond. There are two main types of glycosylation: O-linked glycosylation and N-linked glycosylation [88]. Through glycosyltransferases, sugars form glycosidic bonds with amino acid residues of various proteins to form glycoproteins, which play an important role in the regulation of protein folding and stabilization, cell growth, receptor activation, cell adhesion, and immune

Table 1. Summary of the targets and mechanisms of protein post-translational modifications in antifungal innate immunity

Enzymes	Substrate	Mechanism of Regulation	Biological Function
Phosphorylation			
EPHB2	SYK TAGAP (Tyr310)	Activation	Co-receptor; promoting SYK, TAGAP phosphorylation, and enhancing CLRs-mediated immune signaling [25, 26].
SYK	DOCK2 (Tyr985, 1405)	Activation	DOCK2 phosphorylation mediates ROS production [32].
	Vesicles with GLUT1	Transposition	Enhancing neutrophil metabolic fitness [34]
PKC-δ	CARD9 (Thr231)	Activation	Signaling between SYK and CARD9 [95].
Dephosphorylation			
STS	SYK	Inactivation	Inhibiting ROS production [39].
PP1	CARD9	Inhibited activation	Inhibiting the antifungal effect of PMN [44].
Ubiquitination			
CBL-B	Dectin-1 (Lys2, 27, 34) Dectin-2 (Lys10, 12) Dectin-3 (Lys9)	Degradation	Inhibiting dectin-1/2/3-mediated immune responses [57].
C-CBL	RelB	Degradation	Promoting IL-10 production, anti-inflammatory and immune balance[59].
TRIM31	SYK (Lys375, 517)	Transposition	Promoting the binding of SYK to CLRs and inhibiting the dephosphorylation of SYK by SHP-1 [62].
TRIM62	CARD9 (Lys125)	Activation	Dissolving the self-inhibited state of CARD9 and promoting CBM complex formation [67].
NEDD4	——	——	Enhancing the immune response [71].
De-ubiquitination			
USP15	CARD9	Inhibition	Inhibiting the activation of CARD9 [72].
OTUD1	CARD9	Activation	Promoting the formation of CBM complexes [75].
Acetylation			
αTAT1	α-tubulin	Transposition	SYK and CARD9 enter the cytoplasm and activate downstream signal pathways [78].
Deacetylation			
HDAC11	NOS2 promoter	Initiate transcription	Reducing NO, ROS generation [79].
Glycosylation			
JAGN1	——	——	Engaging in glycoprotein processing and protein transport in the endoplasmic reticulum and Golgi [91].

Note: EPHB2, erythropoietin-producing hepatocellular carcinoma receptor B2; SYK, spleen tyrosine kinase; TAGAP, T-cell-activated Rho GTPase-activating protein; CLRs, C-type lectin receptors; DOCK2, dedicator of cytokinesis 2; ROS, reactive oxygen species; GLUT1, glucose transporter 1; CARD9, caspase recruitment domain containing protein 9; PKC-δ, protein kinase C-δ; STS, steroid sulfatase; CBL-B, casitas B-lineage lymphoma protein b; C-CBL, c-casitas B-lineage lymphoma; TRIM, tripartite motif containing; NEDD4, neuronal precursor cell-expressed developmentally down-regulated 4; USP15, ubiquitin-specific protease 15; OTUD1, ovarian tumor deubiquitinase 1; αTAT1, α-tubulin N-acetyltransferase 1; HDAC11, histone deacetylase 11; JAGN1, jagunal homolog 1.

response [89, 90].

Jagunal homolog 1 (JAGN1)

The protein encoded by JAGN1 is an endoplasmic reticulum transmembrane protein that functions in the early secretory pathway and is required for neutrophil differentiation and survival. JAGN1 mutations can cause severe congenital neutropenia [91]. Following systemic *C. albicans* infection, JAGN1-deficient mice showed significant weight loss, enlarged mortality, and increased organ fungal burden. Deletion of JAGN1 alters glycoprotein processing and protein transport in the endoplasmic reticulum and Golgi by analysis of mass spectrometry data. In particular, altered glycosylation of the cell adhesion and migration molecules CD177, CD11b and CD18 was observed, as well as significant alterations in the glycosylation of neutrophil collagenase, matrix metalloproteinase-9, lactoferrin, lipocalin 2, binding bead protein,

and myeloid bacteriocin, which are associated with cytotoxic effector functions of neutrophils. Thus, deletion of JAGN1 leads to obvious glycosylation changes in key molecules involved in neutrophil migration and neutrophil cytotoxicity [92].

Discussion

Invasive *Candida* infections are a serious threat to human health, especially in perioperative immunocompromised patients. The main clinical treatment for invasive *Candida* infections is still the use of antifungal drugs. There are three main classes of antifungal drugs in common use, namely azoles, polyenes, and echinocandins [93]. These antifungal drugs usually target the ergosterol biosynthetic pathway, fungal cell membranes, or cell walls. However, the increasing toxicity and resistance of these drugs has led to unsatisfactory clinical outcomes, and the mortality rate of invasive *Candida* infections still exceeds 50% [94]. Therefore, it is necessary to explore novel therapeutic approaches to combat fungal infections.

The regression of any infection depends on the pathogenic capacity of the pathogenic bacteria and the ability of the immune system to kill it. In addition to developing new drugs to target *Candida* itself, it is possible to fight infection by modulating the immune system. Through the previous description of the regulatory role of the PTMs in the antifungal immune process, we have gained insights into the targets and mechanisms of action of the enzymes associated with PTMs in the antifungal immune process (**Table 1**). These substances play a very important role in the control of fungal infections. Therefore, modulating the progression of anti-fungal immunity through the regulation of PTMs could be a promising avenue for intervention.

In existing studies, systemic in vivo delivery of CBL-B siRNA into mice have protected them from lethal disseminated candidiasis and greatly reduced post-infection mortality and organ fungal burden. Administration of AGK2 or AK-1 inhibitors of the deacetylase SIRT2 promoted dectin-activated signaling and proinflammatory gene expression, and produced a protective effect on mice with systemic *Candida* infection [57, 79]. The above approaches interfere with enzymes and negatively regulate the immune process against *C. albicans*, by siRNA and inhibitors, thus enhancing the antibacterial capacity of the cells, as well as in mice. This suggests that the anti-inflammatory capacity of immune cells can be directly regulated by modulating the activity of these enzymes. This allows these enzymes to function as potential therapeutic targets, and the immunity of the host during invasive *Candida* infections can be enhanced by developing drugs that target these enzymes.

In conclusion, PTMs that modulate host innate immune response against *C. albicans* invasion hold great research value. However, clinical treatment for fungal infections still faces significant challenges, and more sustained and in-depth research is needed in the future.

References

[1] Lionakis MS, Lim JK, Lee CC, et al. Organ-specific innate immune responses in a mouse model of invasive candidiasis. J Innate Immun 2011;3:180-199.

[2] Bugli F, Massaro F, Buonocore F, et al. Design and Characterization of Myristoylated and Non-Myristoylated Peptides Effective against *Candida* spp. Clinical Isolates. Int J Mol Sci 2022;23:2164.

[3] Pianalto KM, Alspaugh JA. New Horizons in Antifungal Therapy. J Fungi (Basel) 2016;2(4):26.

[4] Pfaller MA, Diekema DJ. Epidemiology of invasive candidiasis: a persistent public health problem. Clin Microbiol Rev 2007;20:133-163.

[5] Page GG. Surgeryinduced immunosuppression and postoperative pain management. AACN Clin Issues 2005;16:302-309; quiz 416-418.

[6] Schmidt S, Condorelli A, Koltze A, et al. NK Cells and Their Role in Invasive Mold Infection. J Fungi (Basel) 2017;3(2):5.

[7] Schmidt S, Tramsen L, Lehrnbecher T. Natural Killer Cells in Antifungal Immunity. Front Immunol 2017;8:1623.

[8] Ben-Eliyahu S. The promotion of tumor metastasis by surgery and stress: immunological basis and implications for psychoneuroimmunology. Brain Behav Immun 2003;17 Suppl 1:S27-S36.

[9] Sato A, Nakamura I, Fujita H, et al. Peripheral venous catheter-related bloodstream infection is associated with severe complications and potential death: a retrospective observational study. BMC Infect Dis 2017;17:434.

[10] Tatsuno K, Ikeda M, Wakabayashi Y, et al. Clinical Features of Bloodstream Infections Associated with Peripheral Versus Central Venous Catheters. Infect Dis Ther 2019;8:343-352.

[11] Yang SP, Chen YY, Hsu HS, et al. A risk factor analysis of healthcare-associated fungal infections in an intensive care unit: a retrospective cohort study. BMC Infect Dis 2013;13:10.

[12] Leopold Wager CM, Hole CR, Wozniak KL, et al. Cryptococcus and Phagocytes: Complex Interactions that Influence Disease Outcome. Front Microbiol 2016;7:105.

[13] Costa-de-Oliveira S and Rodrigues AG. *Candida albicans* Antifungal Resistance and Tolerance in Bloodstream Infections: The Triad Yeast-Host-Antifungal. Microorganisms 2020;8(2):154.

[14] Lionakis MS, Drummond RA and Hohl TM. Immune responses to human fungal pathogens and therapeutic prospects. Nat Rev Immunol 2023;23:433-452.

[15] Borriello F, Zanoni I, Granucci F. Cellular and molecular mechanisms of antifungal innate immunity at epithelial barriers: The role of C-type lectin receptors. Eur J Immunol 2020;50:317-325.

[16] Roth S, Bergmann H, Jaeger M, et al. Vav Proteins Are Key Regulators of Card9 Signaling for Innate Antifungal Immunity. Cell Rep 2016;17:2572-2583.

[17] Hardison SE, Brown GD. C-type lectin receptors orchestrate antifungal immunity. Nat Immunol 2012;13:817-822.

[18] Wan L, Xu K, Chen Z, et al. Roles of Post-translational Modifications in Spinocerebellar Ataxias. Front Cell Neurosci 2018;12:290.

[19] Doyle HA, Mamula MJ. Post-translational protein modifications in antigen recognition and autoimmunity. Trends Immunol 2001;22:443-449.

[20] Karve TM, Cheema AK. Small changes huge impact: the role of protein posttranslational modifications in cellular homeostasis and disease. J Amino Acids 2011;2011:207691.

[21] Liu J, Qian C, Cao X. Post-Translational Modification Control of Innate Immunity. Immunity 2016;45:15-30.

[22] Incel A, Arribas Díez I, Wierzbicka C, et al. Selective Enrichment of Histidine Phosphorylated Peptides Using Molecularly Imprinted Polymers. Anal Chem 2021;93:3857-3866.

[23] Klement E, Medzihradszky KF. Extracellular Protein Phosphorylation, the Neglected Side of the Modification. Mol Cell Proteomics 2017;16:1-7.

[24] Kraeva N, Lestinova T, Ishemgulova A, et al. LmxM.22.0250-Encoded Dual Specificity Protein/Lipid Phosphatase Impairs Leishmania mexicana Virulence In Vitro. Pathogens 2019;8(4):241.

[25] Sun W, Wang H, Hu H, et al. Cutting Edge: EPHB2 Is a Coreceptor for Fungal Recognition and Phosphorylation of Syk in the Dectin-1 Signaling Pathway. J Immunol 2021;206:1419-1423.

[26] Chen J, He R, Sun W, et al. TAGAP instructs Th17 differentiation by bridging Dectin activation to EPHB2 signaling in innate antifungal response. Nat Commun 2020;11:1913.

[27] Long X, Yu Y, Perlaky L, et al. Stromal CYR61 Confers Resistance to Mitoxantrone via Spleen Tyrosine Kinase Activation in Human Acute Myeloid Leukaemia. Br J Haematol 2015;170:704-718.

[28] Jiang M, Liu R, Chen Y, et al. A combined experimental and computational study of Vam3, a derivative of resveratrol, and Syk interaction. Int J Mol Sci 2014;15:17188-17203.

[29] de Castro RO, Zhang J, Jamur MC, et al. Tyrosines in the carboxyl terminus regulate Syk kinase activity and function. J Biol Chem 2010;285:26674-26684.

[30] Lionakis MS, Levitz SM. Host Control of Fungal Infections: Lessons from Basic Studies and Human Cohorts. Annu Rev Immunol 2018;36:157-191.

[31] Deng Z, Ma S, Zhou H, et al. Tyrosine phosphatase SHP-2 mediates C-type lectin receptor-induced activation of the kinase Syk and anti-fungal TH17 responses. Nat Immunol 2015;16:642-652.

[32] Ma X, Tan X, Yu B, et al. DOCK2 regulates antifungal immunity by regulating RAC GTPase activity. Cell Mol Immunol 2022;19:602-618.

[33] Strasser D, Neumann K, Bergmann H, et al. Syk kinase-coupled C-type lectin receptors engage protein kinase C-delta to elicit Card9 adaptor-mediated innate immunity. Immunity 2012;36:32-42.

[34] Li DD, Jawale CV, Zhou C, et al. Fungal sensing enhances neutrophil metabolic fitness by regulating antifungal Glut1 activity. Cell Host Microbe 2022;30:530-544 e536.

[35] Carpino N, Naseem S, Frank DM, et al. Modulating Host Signaling Pathways to Promote Resistance to Infection by *Candida albicans*. Front Cell Infect Microbiol 2017;7:481.

[36] San Luis B, Sondgeroth B, Nassar N, et al. Sts-2 is a phosphatase that negatively regulates zeta-associated protein (ZAP)-70 and T cell receptor signaling pathways. J Biol Chem 2011;286:15943-15954.

[37] Reppschläger K, Gosselin J, Dangelmaier CA, et al. TULA-2 Protein Phosphatase Suppresses Activation of Syk through the GPVI Platelet Receptor for Collagen by Dephosphorylating Tyr(P)346, a Regulatory Site of Syk. J Biol Chem 2016;291:22427-22441.

[38] Kerrigan AM, Brown GD. Syk-coupled C-type lectins in immunity. Trends Immunol 2011;32:151-156.

[39] Frank D, Naseem S, Russo GL, et al. Phagocytes from Mice Lacking the Sts Phosphatases Have an Enhanced Antifungal

Response to *Candida albicans*. mBio 2018;9(4):e00782-18.

[40] Lemay S, Davidson D, Latour S, et al. Dok-3, a novel adapter molecule involved in the negative regulation of immunoreceptor signaling. Mol Cell Biol 2000;20:2743-2754.

[41] Kim SS, Lee KG, Chin CS, et al. DOK3 is required for IFN-β production by enabling TRAF3/TBK1 complex formation and IRF3 activation. J Immunol 2014;193:840-848.

[42] Peng Q, O'Loughlin JL, Humphrey MB. DOK3 negatively regulates LPS responses and endotoxin tolerance. PLoS One 2012;7:e39967.

[43] Ou X, Xu S, Li YF, et al. Adaptor protein DOK3 promotes plasma cell differentiation by regulating the expression of programmed cell death 1 ligands. Proc Natl Acad Sci U S A 2014;111:11431-11436.

[44] Loh JT, Xu S, Huo JX, et al. Dok3-protein phosphatase 1 interaction attenuates Card9 signaling and neutrophil-dependent antifungal immunity. J Clin Invest 2019;129:2717-2729.

[45] Liang W, Fang J, Zhou S, et al. The role of ubiquitin-specific peptidases in glioma progression. Biomed Pharmacother 2022;146:112585.

[46] Ebner P, Versteeg GA, Ikeda F. Ubiquitin enzymes in the regulation of immune responses. Crit Rev Biochem Mol Biol 2017;52:425-460.

[47] Varshavsky A. The Ubiquitin System, Autophagy, and Regulated Protein Degradation. Annu Rev Biochem 2017;86:123-128.

[48] Hutchins AP, Liu S, Diez D, et al. The repertoires of ubiquitinating and deubiquitinating enzymes in eukaryotic genomes. Mol Biol Evol 2013;30:1172-1187.

[49] Asmamaw MD, Liu Y, Zheng YC, et al. Skp2 in the ubiquitin-proteasome system: A comprehensive review. Med Res Rev 2020;40:1920-1949.

[50] Mevissen TET, Komander D. Mechanisms of Deubiquitinase Specificity and Regulation. Annu Rev Biochem 2017;86:159-192.

[51] Shamim M, Nanjappa SG, Singh A, et al. Cbl-b regulates antigen-induced TCR down-regulation and IFN-gamma production by effector CD8 T cells without affecting functional avidity. J Immunol 2007;179:7233-7243.

[52] Sohn HW, Gu H, Pierce SK. Cbl-b negatively regulates B cell antigen receptor signaling in mature B cells through ubiquitination of the tyrosine kinase Syk. J Exp Med 2003;197:1511-1524.

[53] Chiang YJ, Kole HK, Brown K, et al. Cbl-b regulates the CD28 dependence of T-cell activation. Nature 2000;403:216-220.

[54] Han C, Jin J, Xu S, et al. Integrin CD11b negatively regulates TLR-triggered inflammatory responses by activating Syk and promoting degradation of MyD88 and TRIF via Cbl-b. Nat Immunol 2010;11:734-742.

[55] Paolino M, Choidas A, Wallner S, et al. The E3 ligase Cbl-b and TAM receptors regulate cancer metastasis via natural killer cells. Nature 2014;507:508-512.

[56] Wirnsberger G, Zwolanek F, Asaoka T, et al. Inhibition of CBLB protects from lethal *Candida albicans* sepsis. Nat Med 2016;22:915-923.

[57] van de Veerdonk FL, Netea MG. CBLB ubiquitin ligase: a major regulator of antifungal immunity. Nat Med 2016;22:834-835.

[58] Zhu LL, Luo TM, Xu X, et al. E3 ubiquitin ligase Cbl-b negatively regulates C-type lectin receptor-mediated antifungal innate immunity. J Exp Med 2016;213:1555-1570.

[59] Duan JL, He HQ, Yu Y, et al. E3 ligase c-Cbl regulates intestinal inflammation through suppressing fungi-induced noncanonical NF-κB activation. Sci Adv 2021;7(19):eabe5171.

[60] Li Y, Wu H, Wu W, et al. Structural insights into the TRIM family of ubiquitin E3 ligases. Cell Res 2014;24:762-765.

[61] Liu B, Zhang M, Chu H, et al. The ubiquitin E3 ligase TRIM31 promotes aggregation and activation of the signaling adaptor MAVS through Lys63-linked polyubiquitination. Nat Immunol 2017;18:214-224.

[62] Wang X, Zhang H, Shao Z, et al. TRIM31 facilitates K27-linked polyubiquitination of SYK to regulate antifungal immunity. Signal Transduct Target Ther 2021;6:298.

[63] Xu M, Tan J, Dong W, et al. The E3 ubiquitin-protein ligase Trim31 alleviates non-alcoholic fatty liver disease by targeting Rhbdf2 in mouse hepatocytes. Nat Commun 2022;13:1052.

[64] Zhang J, Cao L, Wang X, et al. The E3 ubiquitin ligase TRIM31 plays a critical role in hypertensive nephropathy by promoting proteasomal degradation of MAP3K7 in the TGF-β1 signaling pathway. Cell Death Differ 2022;29:556-567.

[65] Zeng S, Zhao Z, Zheng S, et al. The E3 ubiquitin ligase TRIM31 is involved in cerebral ischemic injury by promoting degradation of TIGAR. Redox Biol 2021;45:102058.

[66] Song H, Liu B, Huai W, et al. The E3 ubiquitin ligase TRIM31 attenuates NLRP3 inflammasome activation by promoting proteasomal degradation of NLRP3. Nat Commun 2016;7:13727.

[67] Cao Z, Conway KL, Heath RJ, et al. Ubiquitin Ligase TRIM62 Regulates CARD9-Mediated Anti-fungal Immunity and Intestinal Inflammation. Immunity 2015;43:715-726.

[68] Leshchiner ES, Rush JS, Durney MA, et al. Small-molecule inhibitors directly target CARD9 and mimic its protective variant in inflammatory bowel disease. Proc Natl Acad Sci U S A 2017;114:11392-11397.

[69] Guo H, Qiao G, Ying H, et al. E3 ubiquitin ligase Cbl-b regulates Pten via Nedd4 in T cells independently of its ubiquitin ligase activity. Cell Rep 2012;1:472-482.

[70] Pei G, Buijze H, Liu H, et al. The E3 ubiquitin ligase NEDD4 enhances killing of membrane-perturbing intracellular bacteria by promoting autophagy. Autophagy 2017;13:2041-2055.

[71] Nuro-Gyina PK, Tang N, Guo H, et al. HECT E3 Ubiquitin Ligase Nedd4 Is Required for Antifungal Innate Immunity. J Immunol 2021;207:868-877.

[72] Xu W, Rush JS, Graham DB, et al. USP15 Deubiquitinates CARD9 to Downregulate C-Type Lectin Receptor-Mediated Signaling. Immunohorizons 2020;4:670-678.

[73] Chen X, Zhang H, Wang X, et al. OTUD1 Regulates Antifungal Innate Immunity through Deubiquitination of CARD9. J Immunol 2021;206:1832-1843.

[74] Piao S, Pei HZ, Huang B, et al. Ovarian tumor domain-containing protein 1 deubiquitinates and stabilizes p53. Cell Signal 2017;33:22-29.

[75] Zhang L, Liu J, Qian L, et al. Induction of OTUD1 by RNA viruses potently inhibits innate immune responses by promoting degradation of the MAVS/TRAF3/TRAF6 signalosome. PLoS Pathog 2018;14:e1007067.

[76] Ji Y, Chen Z, Cen Z, et al. A comprehensive mouse brain acetylome-the cellular-specific distribution of acetylated brain proteins. Front Cell Neurosci 2022;16:980815.

[77] Choudhary C, Weinert BT, Nishida Y, et al. The growing landscape of lysine acetylation links metabolism and cell signalling. Nat Rev Mol Cell Biol 2014;15:536-550.

[78] Sun W, Ma X, Wang H, et al. MYO1F regulates antifungal immunity by regulating acetylation of microtubules. Proc Natl Acad Sci U S A 2021;118(30):e2100230118.

[79] Wu H, Yin X, Zhao X, et al. HDAC11 negatively regulates antifungal immunity by inhibiting Nos2 expression via binding with transcriptional repressor STAT3. Redox Biol 2022;56:102461.

[80] Teixeira MM. Myo1f is critical for neutrophil migration in vivo. Blood 2018;131:1879-1880.

[81] Navinés-Ferrer A, Ainsua-Enrich E, Serrano-Candelas E, et al. Myo1f, an Unconventional Long-Tailed Myosin, Is a New Partner for the Adaptor 3BP2 Involved in Mast Cell Migration. Front Immunol 2019;10:1058.

[82] Piedra-Quintero ZL, Serrano C, Villegas-Sepúlveda N, et al. Myosin 1F Regulates M1-Polarization by Stimulating Intercellular Adhesion in Macrophages. Front Immunol 2018;9:3118.

[83] Shakespear MR, Halili MA, Irvine KM, et al. Histone deacetylases as regulators of inflammation and immunity. Trends Immunol 2011;32:335-343.

[84] Yanginlar C, Logie C. HDAC11 is a regulator of diverse immune functions. Biochim Biophys Acta Gene Regul Mech 2018;1861:54-59.

[85] Sun L, Marin de Evsikova C, Bian K, et al. Programming and Regulation of Metabolic Homeostasis by HDAC11. EBioMedicine 2018;33:157-168.

[86] Thole TM, Lodrini M, Fabian J, et al. Neuroblastoma cells depend on HDAC11 for mitotic cell cycle progression and survival. Cell Death Dis 2017;8:e2635.

[87] Byun SK, An TH, Son MJ, et al. HDAC11 Inhibits Myoblast Differentiation through Repression of MyoD-Dependent Transcription. Mol Cells 2017;40:667-676.

[88] Zhu H, Wang S, Liu D, et al. Identifying Sialylation Linkages at the Glycopeptide Level by Glycosyltransferase Labeling Assisted Mass Spectrometry (GLAMS). Anal Chem 2020;92:6297-6303.

[89] Ohtsubo K, Marth JD. Glycosylation in cellular mechanisms of health and disease. Cell 2006;126:855-867.

[90] Rudd PM, Elliott T, Cresswell P, et al. Glycosylation and the immune system. Science 2001;291:2370-2376.

[91] Boztug K, Järvinen PM, Salzer E, et al. JAGN1 deficiency causes aberrant myeloid cell homeostasis and congenital neutropenia. Nat Genet 2014;46:1021-1027.

[92] Wirnsberger G, Zwolanek F, Stadlmann J, et al. Jagunal homolog 1 is a critical regulator of neutrophil function in fungal host defense. Nat Genet 2014;46:1028-1033.

[93] Chang YL, Yu SJ, Heitman J, et al. New facets of antifungal therapy. Virulence 2017;8:222-236.

[94] Pfaller MA, Diekema DJ. Epidemiology of invasive mycoses in North America. Crit Rev Microbiol 2010;36:1-53.

[95] Li X, Cullere X, Nishi H, et al. PKC-δ activation in neutrophils promotes fungal clearance. J Leukoc Biol 2016;100:581-588.