

Review

Zoonotic Microsporidia: Host Regulation and Pathogenesis

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ABSTRACT: Microsporidia are a group of obligate intracellular fungal pathogens with extremely wide host ranges. Among these, zoonotic microsporidia such as *Encephalitozoon hellem*, *Encephalitozoon cuniculi*, and *Enterocytozoon bieneusi* can infect humans as well as other animals, causing recurrent diarrhea, hepatitis, and even death in immune-compromised individuals. The abilities of zoonotic microsporidia to regulate their hosts are essential to their survival and thriving within hosts. The manipulations of zoonotic microsporidia on hosts are employed in multiple ways, including metabolic modulation, immune suppression, signaling pathway regulations, and epigenetic modification. This review focuses on pathogen-host interactions between zoonotic microsporidia and their hosts, and compares their regulatory characteristics with those of typical fungal pathogens such as *Candida albicans*. In summary, unraveling the regulatory strategies of zoonotic microsporidia not only deepens our understanding of microsporidia pathogenesis but also provides potential targets for therapeutic intervention against these emerging pathogens. Comparative studies with typical fungal pathogens further highlight the unique and sophisticated host-manipulating mechanisms evolved by microsporidia from the fungal kingdom.

Keywords: Zoonotic microsporidia; Metabolism; Signaling pathways; Immunity; Epigenetics

1. Introduction

Microsporidia are a diverse group of obligate intracellular unicellular eukaryotic parasites. To date, approximately 220 genera and over 1700 species of microsporidia have been taxonomically identified [1,2]. These pathogens are widely distributed throughout natural ecosystems, and their host range encompasses nearly all animal groups, from invertebrates to vertebrates. Among the diverse microsporidian taxa, zoonotic microsporidia exhibit an exceptionally broad host tropism, representing a dual threat to public health security and agricultural economic sustainability [3]. These zoonotic pathogens can cause mild to severe gastrointestinal, neurological, and systemic clinical diseases, especially in those of compromised immunities such as AIDS patients, organ transplant recipients, and other immunosuppressed populations. Zoonotic microsporidia also infect domesticated animals such as cattle, swine, and poultry, resulting in



substantial production losses and economic damage to the livestock industry [4–6]. Notably, species such as *Encephalitozoon hellem*, *Encephalitozoon cuniculi*, and *Enterocytozoon bieneusi* are the most prevalent and clinically relevant zoonotic microsporidian species worldwide.

Microsporidia spores can live outside of a host cell and in a natural environment for a long time because of the rigid, chitin-rich spore wall. When chances permit, the spores could start to invade the host. Infection begins when the polar filament extrudes, injecting the infectious sporoplasm directly into the cytoplasm of a host cell [7,8]. The subsequent developmental stages—merogony (vegetative proliferation) and sporogony (spore formation)—take place within specialized parasitophorous vacuoles in the host cytoplasm, allowing the parasite to complete its life cycle [9,10]. Like other well-known fungal pathogens such as *Candida albicans* and *Cryptococcus neoformans*, microsporidia do not remain passive during intracellular infection. Instead, these obligate intracellular parasites actively reprogram host cell functions to promote their own proliferation, colonization, and immune evasion [11,12]. Recent accumulating evidence shows that this host-modulatory activity spans multiple critical cellular layers, including host metabolic homeostasis, intracellular signaling transduction, innate and adaptive immune responses, and epigenetic modification [13]. Specifically, at the metabolic level, microsporidia hijack and reprogram host carbohydrate, lipid, and protein metabolic pathways to obtain essential nutrients and energy for their own biosynthesis [14]. At the signaling level, they suppress host immune defenses by interfering with key cascades such as the mitogen-activated protein kinase (MAPK) pathway, thereby enhancing their intracellular survival [15]. At the immune level, microsporidia evade host clearance through various strategies, including escaping autophagic degradation and inhibiting host cell apoptosis [16,17]. At the epigenetic level, mechanisms such as histone post-translational modifications and DNA methylation have been implicated in microsporidia-driven remodeling of host cell fate and function [18].

Despite considerable progress in understanding microsporidia–host interactions over the past decades, few systematic reviews have specifically focused on how zoonotic microsporidia modulate host cells. Moreover, the synergistic regulatory mechanisms that coordinate different dimensions of host manipulation remain poorly understood.

This review examines how zoonotic microsporidia regulate host cells following successful infection, focusing on four core areas: metabolic reprogramming, perturbations of key signaling pathways, immune evasion strategies, and epigenetic modifications. In addition, we compare these mechanisms with the host manipulation strategies employed by other well-characterized fungal pathogens, including *Candida albicans* and *Cryptococcus neoformans*. Our goal is to systematically summarize current knowledge, consolidate existing findings on the intracellular parasitic mechanisms of zoonotic microsporidia, and provide a solid theoretical foundation for future mechanistic studies and the development of novel preventive and therapeutic approaches against microsporidiosis.

2. Metabolic Modulation

A key aspect of host manipulation begins with how these pathogens hijack fundamental metabolic pathways to secure energy and building blocks. Fungal pathogens, such as *Cryptococcus neoformans*, *Candida albicans*, and *Aspergillus fumigatus*, have the ability to regulate host metabolism to ensure their own proliferation (Figure 1A). *Candida albicans* infects host epithelial cells and regulates the cells' uptake of glucose, thereby increasing the rate of glycolysis. At the same time, the activity of the host cells' citric acid cycle decreases, creating an environment conducive to the fungus's survival [19]. In the meantime, *Cryptococcus neoformans* inhibits the autophagy process in host cells during infection, leading to abnormal intracellular lipid accumulation and the formation of lipid-rich foam cells, which provide a microenvironment conducive to the pathogen's nutrition and replication [20].

Most microsporidia only possess a tiny remnant mitochondrion (mitosome), which lost typical mitochondria cristae, organellar genome, and most canonical functions [21]. Therefore, the absence of

certain biosynthetic pathways and genes involved in the citric acid cycle results in a strong dependence on the host. Once microsporidia have successfully invaded a host cell, their own energy and material reserves are far from sufficient for normal reproduction. They must compensate for their deficiencies in nucleotide synthesis and DNA replication by stealing nucleotides from the host, and they must also steal ATP from the host to meet their own energy needs. (Figure 1B). For instance, microsporidian *Trachipleistophora hominis* has been reported to take up host purine nucleotides via the nucleotide transporter NTT to compensate for its own deficiencies in nucleotide synthesis and DNA replication [22].

As for ATP intake, the *E. hellem* sporoplasm surface protein 1 (EhSSP1) has been found to interact with the host mitochondrial outer membrane protein, the voltage-dependent anion channels (VDACs). They can mediate the tight interaction between the host mitochondria and the parasitophorous vacuole (PV). This contact may facilitate the transportation of ATP from the host to the parasite, thereby meeting the energy requirements of the microsporidian [23]. Interestingly, microsporidia can maintain the host's ATP levels at a steady state, ensuring the host cell's survival and their own proliferation [24].

So how is this achieved? Based on existing research, we know that one way is to exploit the host's autophagy. Unlike fungal pathogens such as *Cryptococcus neoformans*, which can suppress host autophagy, microsporidia, such as *Encephalitozoon cuniculi*, strongly induce autophagy in RK-13 cells, yet evade host-mediated degradation and utilize host autophagy as a nutrient source [25]. Although the mechanism by which microsporidia induce autophagy has not yet been fully elucidated, one relatively well-defined pathway involves enhancing host autophagy by increasing host ubiquitination. EhPTP4 is a polar tube protein of *Encephalitozoon hellem*. The N-terminus of the EhPTP4 protein contains a signal peptide, while the C-terminus contains a histidine-rich domain (HRD) and a nuclear localization signal (NLS). In both infected and transfected cells, EhPTP4 is secreted into the host cell nucleus. EhPTP4 significantly upregulates the expression of key genes (PDIA4, HERP, HSPA5, and Derlin3) involved in the host endoplasmic reticulum-associated degradation (ERAD) pathway and enhances the ubiquitination of host proteins, indicating that EhPTP4 functions in the regulation of host ERAD [25–27].

While metabolic modulation secures nutrient supply, the precise regulation of host signaling pathways may ensure intracellular pathogens a more non-lytic, replication-friendly environment. Therefore, the next section shifts focus to how microsporidia precisely manipulate key signaling pathways for immune evasion and survival.

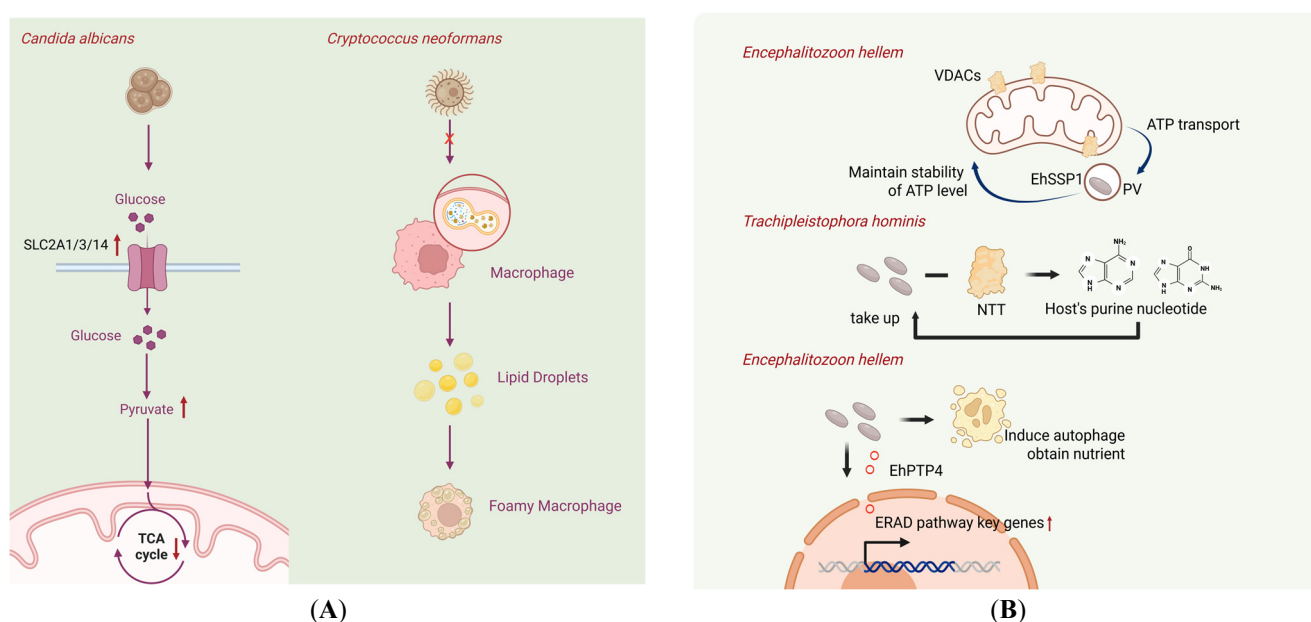


Figure 1. Modulation on host metabolism by microsporidia and other fungal pathogens. (A) *Candida albicans* infection increases glycolytic activity in epithelial cells and reduces TCA cycle activity; *Cryptococcus neoformans* infection inhibits the function of

host phagocytes, leading to abnormal intracellular accumulation of lipids. **(B)** The microsporidian *E. hellem* hijacks the host's mitochondria and steals ATP to compensate for its own energy deficit; The microsporidian *Trachipleistophora hominis* hijacks the host nucleotide transporter NTT to take up host purine nucleotides to compensate for its own defects in nucleotide synthesis and DNA replication; the microsporidian *E. hellem* secretes the host nuclear localization protein EhPTP4 to activate key genes in the host ERAD pathway (VDACs: host mitochondrial outer membrane proteins, voltage-dependent anion channels; EhSSP1: spore surface protein 1 of *E. hellem*; NTT: nucleotide transporter; EhPTP4: secretory protein of *E. hellem*, localized to the host cell nucleus).

3. Signaling Pathway Fine-Tuning

It is very impressive that pathogens such as microsporidia, despite having highly reduced genomes, still possess numerous genes encoding host-manipulating molecules, such as serpins, ricin-like proteins, and protein phosphatases [15,28–30]. To this perspective, microsporidia are similar to typical fungal pathogens such as *Cryptococcus neoformans* and *Candida albicans*, which can manipulate host signaling pathways through effector proteins and cell wall components to achieve immune evasion, intracellular colonization, and proliferation.

Different effectors targeting at different points of the signaling pathway may result in various cellular responses (Figure 2). For instance, candidalysin, the key toxin of *Candida albicans*, can activate the p38 MAPK pathway in epithelial cells, inducing the release of pro-inflammatory factors such as IL-6 and IL-8 [31]. Interestingly, candidalysin can also transactivate the EGFR-ERK pathway, precisely regulating the balance between cell damage and repair, thereby providing crucial support for hyphal invasion and tissue colonization [32]. In addition, the β -glucan in the cell wall of *Candida albicans* can activate the NF- κ B inflammatory response through the Dectin-1→Syk→CARD9 signaling axis, while its regulatory factor Dal81 can inhibit the excessive activation of NF- κ B by alkalizing the intracellular pH, forming a dynamic balance of “activation-inhibition” to achieve immune evasion [33].

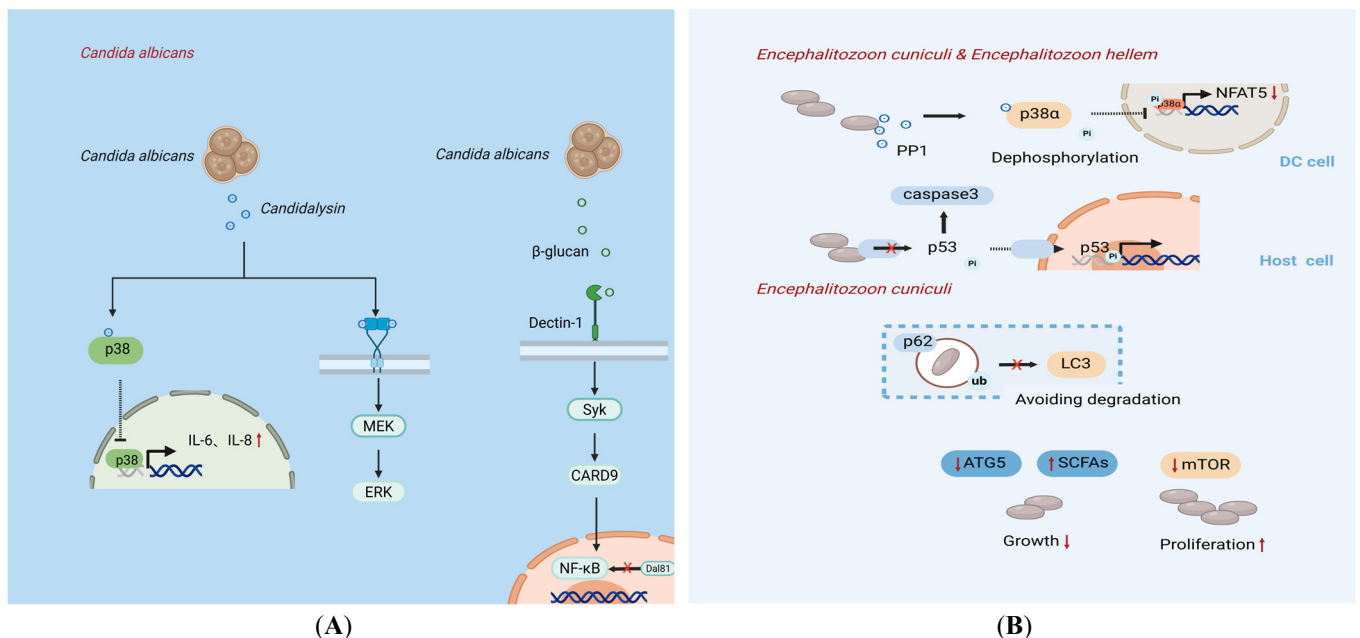


Figure 2. Fine-tuning of host signaling pathways by microsporidia and a representative fungal pathogen *Candida albicans*. **(A)** *Candida albicans* secretes candidin, which activates the p38 MAPK pathway in epithelial cells, inducing the release of pro-inflammatory factors, while simultaneously trans-activating the EGFR-ERK pathway to regulate cellular damage and repair; *Candida albicans* cell wall β -glucan activates NF- κ B via the Dectin-1–Syk–CARD9 signaling axis to initiate an inflammatory response, while its regulatory factor Dal81 inhibits excessive NF- κ B activation by alkalizing intracellular pH. **(B)** *E. hellem*/*E. cuniculi* actively secretes the serine/threonine protein phosphatase PP1, which inhibits the phosphorylation and activation of p38 α in dendritic cells (DCs) through dephosphorylation, thereby blocking the signaling of downstream transcription factors such as NFAT5; Microsporidia of the genus *Encephalitozoon* effectively inhibit the cleavage and activation of host caspase-3

and block the phosphorylation and nuclear translocation of p53, thereby suppressing p53's transcriptional regulatory function; In the early stages, *E. cuniculi* is marked by host ubiquitin and p62 to initiate heterologous autophagy, but it is able to evade degradation by autophagosomes by failing to recruit LC3, thereby hijacking the host autophagy pathway to obtain nutrients for itself. Inhibition of the upstream mTOR pathway activates autophagy and significantly promotes microsporidian proliferation, whereas knocking down the key autophagy gene ATG5 or inhibiting autophagy using short-chain fatty acids (SCFAs)—metabolites produced by the gut microbiota—significantly suppresses parasite growth.(p53:Key intracellular transcription factors;LC3:Autophagosome marker protein).

In contrast to the above typical fungal pathogens, the targeted signaling pathways of microsporidia in host cells have been revealed more recently. Furthermore, as the infection progresses and the individual's life course unfolds, it regulates various signaling pathways. The TLR-Myd88/TRAF pathway is the first one, and the TLR-signaling pathways stand out in a single-cell analysis of microsporidia (*E. cuniculi*)-infected T cells. The up-regulation of TLR2 but down-regulation of effector cytokines indicates that TLR-Myd88/TRAF pathway may be targeted and modulated [34]. The next pathway to be regulated is the MAPK pathway, which is activated by inflammatory signals. zoonotic microsporidia *E. hellem* secretes serine/threonine protein phosphatase PP1, directly targeting the p38/MAPK signaling pathway in dendritic cells (DCs), inhibiting the activation of p38 MAPK protein, suppressing downstream transcriptional factors and genes expression. Consequently, markedly lower levels of pro-inflammatory cytokines such as IL-12, IL-6, and TNF- α are expressed, and the DCs function and maturation were impaired [15].

In the later stages of the infection, zoonotic microsporidia also ensure intracellular survival by modulating the p53-mediated apoptotic pathway [35]. Members of the genus *Encephalitozoon* effectively inhibit the cleavage and activation of host caspase-3, and block the phosphorylation and nuclear translocation of p53, thereby abolishing the transcriptional regulatory function of p53. This prevents apoptosis of infected cells and provides a stable proliferative niche for the continuous propagation of microsporidia.

Besides, the autophagic pathway and the mTOR pathway are also targets of zoonotic microsporidia. *E. cuniculi* achieves efficient nutrient acquisition and proliferation by hijacking the host autophagic pathway by interfering with LC3 recruitment. As a result, *E. cuniculi* can escape degradation in autolysosomes and even supply nutrients for proliferation [25].

4. Immune Regulation

Signaling fine-tuning often serves as the upstream trigger for downstream immune modulation and therefore represents a critical battleground for intracellular pathogens. In the following section, we detail how zoonotic microsporidia reshape host immunity—from pattern recognition to T cell responses.

Both *Candida albicans* and *Cryptococcus neoformans* are facultative parasitic fungi capable of targeting host innate and adaptive immune pathways through cell wall components, secreted effector molecules, and toxins (Figure 3A). By modulating immune activation and evasion, they achieve a dynamic balance that facilitates host invasion and colonization. *Candida albicans* relies on pattern recognition receptors (PRRs), mainly Toll-like receptors (TLRs), C-type lectin receptors (CLRs), and NOD-like receptors (NLRs) [36]. Dectin-1 specifically recognizes β -glucan on the cell wall of *Candida albicans*, and upon activation initiates a series of host cellular immune responses, while acting synergistically with TLRs to regulate host antifungal immunity [37].

As the obligate intracellular parasitic fungi with highly reduced genomes and lacking typical immunogenic fungal PAMPs, microsporidia usually do not stimulate intense inflammatory responses during invasion [29,38]. The host immune responses are often suppressed during microsporidia infection alongside the maintenance of host cell survival, which is closely aligned with their intracellular parasitic requirements (Figure 3B). Innate immune suppression by microsporidia is more frequently reported. For instance, *E. hellem* targets p38 MAPK to inhibit the DCs phagocytosis, cell maturation, and antigen

presentation [15]. Microsporidia can employ effector proteins to disrupt the PRR–PAMP recognition process, inhibit nuclear translocation of the NF- κ B p65 subunit, and reduce the release of pro-inflammatory factors. IRF3 phosphorylation and the activation of the type I interferon (IFN-I) signaling pathway are also suppressed. These mechanisms prevent excessive innate immune responses and create a stable intracellular environment conducive to microsporidian proliferation [39].

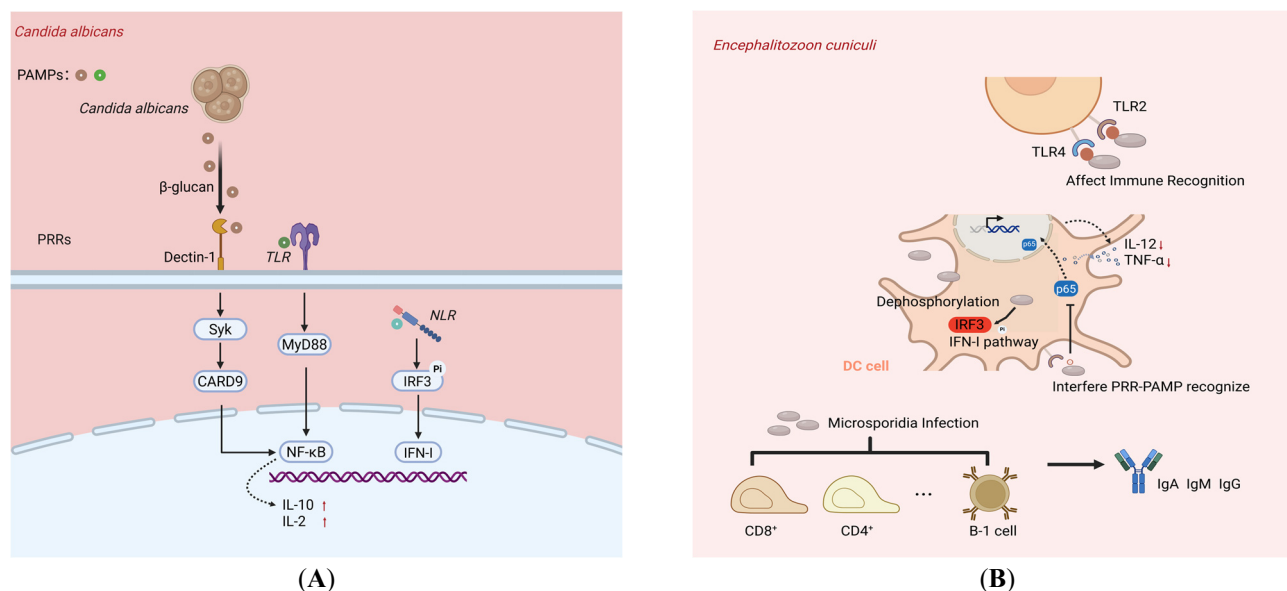


Figure 3. Immune regulations by microsporidia and a representative fungal pathogen. **(A)** *Candida albicans* is recognized by Dectin-1, Toll-like receptors (TLRs), and NOD-like receptors (NLRs) on the surface of macrophages through its pathogen-associated signaling molecules (such as β -glucan), thereby activating downstream signaling molecules including Syk, MyD88, and CARD9, which ultimately leads to upregulation of IL-2 and IL-10 expression. **(B)** Microsporidia *E. cuniculi* interferes with the TLR2/TLR4-mediated recognition by host dendritic cells, disrupting the PRR-PAMP recognition process. This leads to the dephosphorylation of p65 and the inhibition of the IRF3 and IFN-I pathways, thereby reducing the secretion of IL-12 and TNF- α , and ultimately attenuating the immune response of CD4⁺, CD8⁺ T cells, and B-1 cells. (Dectin-1: Specifically recognizes β -glucan on the cell wall of *Candida albicans* PRR-PAMP: The process by which microsporidia recognize pathogen-associated molecular patterns via host pattern recognition receptors.

The host adaptive immune system could also be targeted for regulation by zoonotic microsporidia. It's reported that microsporidia *E. cuniculi* stimulate cellular immunity, especially on the perforin and granzymes producing CD8⁺ cytotoxic T lymphocytes. In addition, other immune cells such as CD4⁺ T cells, $\gamma\delta$ T cells, and B-1 cells are also activated to participate in immune regulation [34]. In addition, microsporidia stimulate the host to produce antibodies, including IgG and IgM, to facilitate neutralization and phagocytosis [40]. Moreover, microsporidia are reported to establish a favorable parasitic immune environment by inhibiting host cell apoptosis by downregulating the expression of pro-apoptotic proteins or inhibiting the cleavage of pro-caspases [35].

5. Epigenetic Modification

Beyond immediate immune control, recent evidence reveals that microsporidia also impose long-lasting changes on host cells through epigenetic reprogramming. Epigenetics, as a heritable form of gene expression regulation independent of DNA sequence changes, serves as a key battleground for pathogens to regulate their hosts and plays a significant role in the proliferation and spread of fungal pathogens following invasion. As shown (Figure 4A), when *Cryptococcus neoformans* infects hosts, it activates and upregulates the expression of the histone demethylase LSD1, LSD1 selectively binds to the promoters of genes associated with lipid uptake (Ldlr, Cd36) and biosynthesis (Plin2), reducing the levels

of the repressive histone mark H3K9me2 and thereby activating the transcription of these genes. Concurrently, it directly inhibits host lipid autophagy (by blocking the lysosomal degradation of lipid droplets), which ultimately leads to abnormal lipid accumulation and the formation of foam macrophages [20]. *Talaromyces marneffei* primarily infects immunocompromised individuals and is an important opportunistic pathogen. *T. marneffei* infection induces elevated levels of m⁶A RNA modification, as well as lncRNAs in host cells. These modifications alter the characteristics of the host transcriptome, exacerbate damage to the host immune system via the YTHDC2-TLR2 signaling axis, and simultaneously increase the susceptibility of host cells to HIV, accelerating disease progression in co-infected patients [41,42].

Like *T. marneffei*, zoonotic microsporidia primarily infect immunocompromised individuals and are capable of modulating host epigenetic regulation. However, the mechanisms differ (Figure 4B): *T. marneffei* infection affects RNA modifications in host cells, whereas the microsporidian *E. hellem* regulates host histone ubiquitination. Microsporidian EnP1 is a nucleus-targeted effector that can modify the host cell environment. This effector contains a functional signal peptide (SP) and two NLSs, highly conserved within the genus *Microsporidium*. Following infection, EnP1 can enter the host cell nucleus, whereas the double NLS-deleted mutant (EnP1ΔNLS) localizes exclusively to the cytoplasm and fails to translocate into the nucleus. EnP1 does not directly bind RNF20, the key ubiquitin ligase responsible for H2Bub; instead, it competitively binds to H2B, thereby blocking the interaction between RNF20 and H2B and inhibiting H2Bub. Furthermore, knocking down RNF20 or overexpressing H2BK120R (which cannot be ubiquitinated) mimics the effects of EnP1, significantly promoting microsporidian proliferation [39].

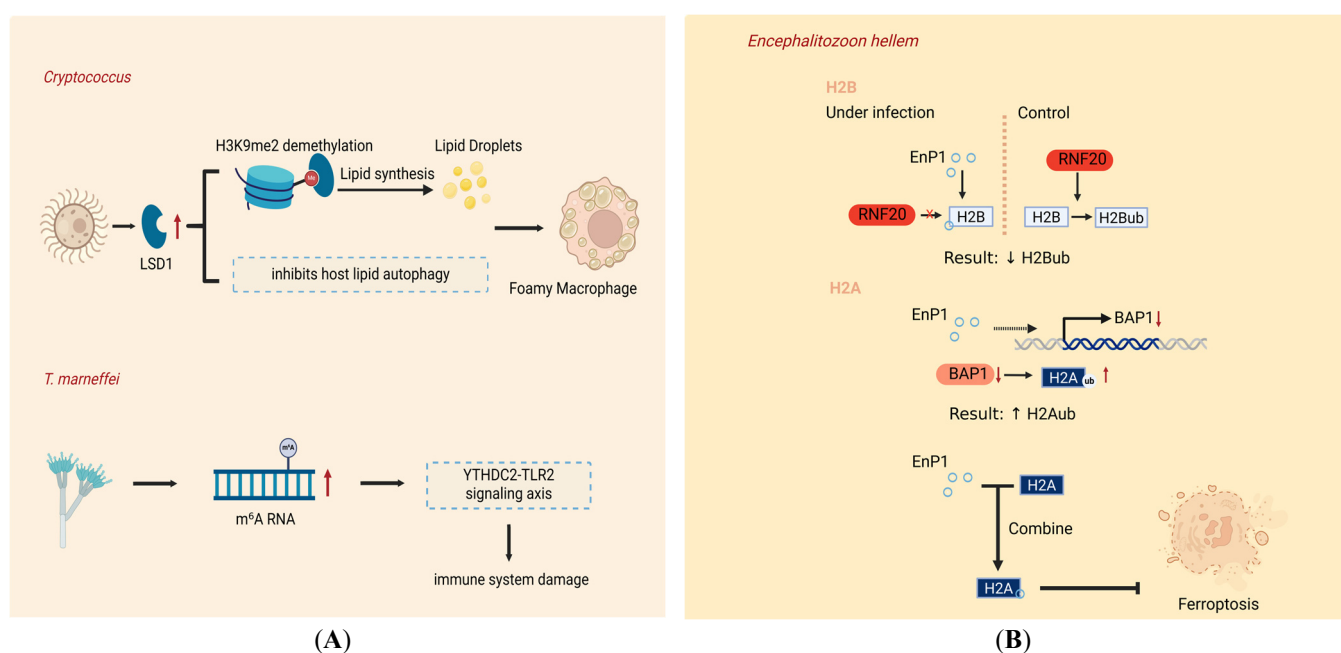


Figure 4. Epigenetic modification by microsporidia and other fungal pathogens. **(A)** When *Cryptococcus neoformans* infects a host, it activates and upregulates LSD1 expression. LSD1 selectively binds to the promoters of genes associated with lipid uptake (Ldlr, Cd36) and biosynthesis (Plin2), and reduces the levels of the repressive histone mark H3K9me2, thereby activating the transcription of these genes. Concurrently, it directly inhibits host lipid autophagy, ultimately leading to abnormal lipid accumulation and the formation of foam macrophages. Infection with *Talaromyces marneffei* induces elevated levels of m⁶A RNA modifications in host cells. This abnormal modification alters the characteristics of the host transcriptome and exacerbates damage to the host immune system via the YTHDC2-TLR2 signaling axis. **(B)** Following infection by *E. hellem*, EnP1 is secreted into the host cell nucleus, where it competitively binds to H2B, thereby blocking the interaction between RNF20 and H2B and consequently inhibiting H2Bub; Overexpression of EnP1 or microsporidian infection significantly downregulates only the mRNA and protein expression of the deubiquitinating enzyme BAP1, and since EnP1 does not directly interact with BAP1, this suggests that EnP1 elevates H2A monoubiquitin (H2Aub) levels by inhibiting BAP1 expression. (LSD1: histone demethylase;

EnP1: microsporidian capsid protein, which is also a nuclear-targeted effector protein secreted by microsporidia; BAP1: deubiquitinating enzyme, the core catalytic subunit of the PR-DUB complex).

At the same time, EnP1 also binds directly to histone H2A, and together inhibit host ferroptosis, although the mechanisms are slightly different: H2A monoubiquitination (H2Aub) is mediated by RING1A/B of the PRC1 complex, whereas deubiquitination is driven by BAP1 of the PR-DUB complex. This experimental evidence has also confirmed that EnP1 overexpression or microsporidia infection did not affect the transcription or protein levels of RING1A/B or USP16 (another H2A deubiquitinase), but significantly downregulated the mRNA and protein expression of BAP1. EnP1 does not directly interact with BAP1, indicating that EnP1 elevates H2A monoubiquitin (H2Aub) levels by suppressing BAP1 expression [17,18]. These mechanistically distinct yet functionally synergistic strategies of host epigenetic regulation employed by microsporidia and other fungal pathogens illustrate the evolutionary sophistication of microsporidia within the fungal kingdom.

Collectively, these four dimensions—metabolic, signaling, immune, and epigenetic—operate as an integrated network rather than isolated strategies.

6. Conclusions

Zoonotic microsporidia, as obligate intracellular fungal pathogens, have evolved a sophisticated, multi-layered regulatory network to reprogram host cell functions for their survival, proliferation, and immune evasion. This review systematically dissects these regulatory strategies from four interconnected dimensions: metabolic reprogramming, signaling pathway modulation, immune regulation, and epigenetic remodeling.

At the metabolic level, microsporidia exploit host ATP, amino acids, and nucleotides via mitochondrion–parasitophorous vacuole contact sites (e.g., EhSSP1-VDAC interaction), hijack the ERAD pathway (e.g., EhPTP4), and subvert host autophagy to acquire biosynthetic building blocks—contrasting sharply with canonical fungal pathogens that often suppress autophagy. At the signaling level, secreted effectors such as PP1 directly dampen p38 MAPK activation in dendritic cells, while manipulation of TLR-MyD88, p53-mediated apoptosis, and mTOR-autophagy axes ensures a non-lytic, replication-permissive intracellular niche. Immunologically, microsporidia actively suppress both innate (e.g., inhibition of DC maturation, NF- κ B, and type I IFN) and adaptive (e.g., modulation of CD8⁺ T cell cytotoxicity and antibody responses) immunity, establishing a finely tuned balance between host tolerance and parasite persistence. Epigenetically, the nuclear effector EnP1 exemplifies a novel mechanism by competitively blocking H2B monoubiquitination and downregulating BAP1 to elevate H2A ubiquitination, thereby inhibiting host ferroptosis and remodeling transcriptional programs favorable to parasite proliferation—a strategy distinct from the RNA m⁶A modification employed by other opportunistic fungi like *Talaromyces marneffe*.

Despite these advances, several critical gaps remain. First, the full repertoire of secreted effectors and their host targets is far from complete, particularly for *Enterocytozoon bieneusi*, which lacks robust *in vitro* culture systems. Second, the crosstalk among metabolic, signaling, immune, and epigenetic layers—e.g., how EnP1-mediated histone modification influences metabolic gene expression—remains poorly understood. Third, most studies rely on *Encephalitozoon* models; comparative analyses across different zoonotic genera and host species are urgently needed to delineate conserved versus species-specific regulatory principles.

Looking forward, integrating high-resolution structural biology, single-cell multi-omics, and CRISPR-based host–pathogen interaction screens will accelerate the discovery of druggable host targets and parasite-derived virulence factors. For example, cryo-electron microscopy (Cryo-EM) and cryo-electron tomography (Cryo-ET) can be used to elucidate the substrate selectivity and transport cycle of microsporidian ATP transporters, thereby guiding the design of competitive inhibitors to block the theft of host energy by microsporidians. Single-cell omics technologies can address key challenges in microsporidia

research: the spatiotemporal heterogeneity of the infection stage and the heterogeneity of the host immune response. As for CRISPR technology, it can be used to screen for genes essential for microsporidia to infect their hosts and to identify host immune pathways targeted by microsporidia effector proteins. Given that microsporidiosis primarily threatens immunocompromised populations and agricultural animals, deciphering these regulatory networks holds translational promise: host-directed therapies (e.g., modulating p38 MAPK or ferroptosis pathways) and epigenetic inhibitors could complement conventional antiparasitic drugs. Moreover, as a minimalist model of intracellular parasitism within the fungal kingdom, zoonotic microsporidia offer unique insights into the evolution of host manipulation—from metabolic dependence to epigenetic coercion—illuminating fundamental principles that may apply to other intracellular pathogens.

In conclusion, host regulatory mechanisms of zoonotic microsporidia constitute a complex, highly specialized, and evolutionarily convergent network. Unraveling this network not only deepens our understanding of microsporidia pathogenesis but also establishes a conceptual and technological foundation for next-generation interventions, ultimately strengthening public health defenses and sustaining animal husbandry against these emerging pathogens.

Author Contributions

Writing—Original Draft Preparation, L.L.-T., J.J., M.Z.; Visualization, J.J., M.Z.; Writing—Review & Editing, L.L.-T., Q.H., M.R., J.C., J.B.; Supervision, Z.Z., J.B.; Project Administration, J.B.; Funding Acquisition, J.B.

Ethics Statement

Not applicable.

Informed Consent Statement

Not applicable.

Data Availability Statement

Not applicable.

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Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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