Journal of Microbiology (2022) Vol. 60, No. 5, pp. 451–460
DOI 10.1007/s12275-022-2009-z
eISSN 1976-3794
pISSN 1225-8873

MINIREVIEW

The crosstalk between bacteria and host autophagy: host defense or bacteria offense

Lin Zheng[1], Fang Wei [1*], and Guolin Li[1,2*]

[1]*Center for Biomedical Aging, National & Local Joint Engineering Laboratory of Animal Peptide Drug Development, College of Life Sciences, Hunan Normal University, Changsha, Hunan 410081, P. R. China*
[2]*Key Laboratory of Hunan Province for Model Animal and Stem Cell Biology, School of Medicine, Hunan Normal University, Changsha, Human 410081, P. R. China*

(Received Jan 11, 2022 / Revised Mar 18, 2022 / Accepted Mar 29, 2022)

Xenophagy is a specific selective autophagy for the elimination of intracellular bacteria. Current evidence suggests that the processes for host autophagy system to recognize and eliminate invading bacteria are complex, and vary according to different pathogens. Although both ubiquitin-dependent and ubiquitin-independent autophagy exist in host to defense invading bacteria, successful pathogens have evolved diverse strategies to escape from or paralyze host autophagy system. In this review, we discuss the mechanisms of host autophagy system to recognize and eliminate intracellular pathogens and the mechanisms of different pathogens to escape from or paralyze host autophagy system, with a particular focus on the most extensively studied bacteria.

Keywords: autophagy, xenophagy, bacteria, infection, bacterial effector

Introduction

Autophagy is a conserved intracellular degradation process for the recycling of nutrients and energy, which is critical to cellular homeostasis maintenance and stress responses (Mizushima and Komatsu, 2011). Although typically low under basal conditions, cellular autophagy can be significantly upregulated under stress conditions to optimize cell survival (Simon *et al.*, 2017). According to the type of degraded cargo, autophagy can be divided into non-selective autophagy and selective autophagy (Anding and Baehrecke, 2017). Non-selective nutrient recycling process is a canonical autophagy pathway to remove multiple cellular components, while selective autophagy targets specific cargos, such as unwanted cytosolic components and damaged and/or redundant organelles (Gatica *et al.*, 2018).

Xenophagy is a specific selective autophagy for the identification and removal of intracellular bacteria, which has early been characterized as an immune response to eliminate bacteria (Sharma *et al.*, 2018). Since Rikihis first observed the formation of autophagosomes in polymorphonuclear cells of guinea pigs infected with *Rickettsia* in 1984 (Rikihis, 1984), various bacteria, including *Streptococcus*, *Mycobacterium tuberculosis* (Mtb), *Shigella flexneri*, *Salmonella* Typhimurium (*S.* Typhimurium), and *Listeria monocytogenes* (Lm), have been reported to be recognized and removed by the host autophagy system (Kohler and Roy, 2017). Although autophagy defensive strategy limits the proliferation of most intracellular pathogens, many bacteria have evolved a variety of defense strategies to escape from xenophagy through inhibiting the initiation of autophagy or the formation of autolysosomes. In this review, we discuss the mechanisms of host autophagy system to recognize and eliminate intracellular pathogens and the mechanisms of different pathogens to escape from or paralyze host autophagy system, with a particular focus on the most extensively studied bacteria.

Recognition of Invading Bacteria by Host Autophagy System

In general, host autophagy system can recognize invading bacteria through both ubiquitin-dependent and ubiquitin-independent manners. According to a prevailing view, a ubiquitin-adaptor protein-LC3 (microtubule-associated protein 1 light chain 3) model is the main ubiquitin-dependent pathway to recognize invading bacteria, in which ubiquitin-coated bacteria are recognized by adaptors and targeted by autophagy machinery. Besides, the adaptors can also target bacteria-residing vacuoles or damaged vacuolar membranes in a ubiquitin-independent manner.

Ubiquitin-adaptor-LC3 mediated xenophagy

Ubiquitination of cargos represents an important step in xenophagy (Peng *et al.*, 2017). When bacteria infect host cells, they can be recognized and labelled by ubiquitin in the cytosol and serve as autophagy targets (Linares *et al.*, 2013). Heterogeneous ubiquitin ligase E3 plays a critical role in bacterial specific ubiquitination, and SMURF1 (Smad ubiqui-

*For correspondence. (F. Wei) E-mail: weifang2019@hunnu.edu.cn; Tel.: +86-0731-88872786; (G. Li) E-mail: hnsdlgl@hunnu.edu.cn; Tel.: +86-0731-88872786

tination regulatory factor) and TRIM (tripartite motif) family proteins are the most studied E3 ligases in xenophagy (Fig. 1). SMURF1 mediates K48-linked ubiquitination of Mtb and recruitment of autophagy machinery components (adaptor NBR1 [next to BRCA1 gene 1 protein], autophagy protein LC3 and lysosomal associated membrane protein), and thus modulates Mtb replication in macrophages (Franco *et al.*, 2017). TRIM family proteins also act in autophagic lysosomes to control Mtb invasion. For instance, TRIM16 can integrate galectin- and ubiquitin-based processes, which directs the recognition of Mtb and the mobilization of the core autophagy regulators ATG16L1 (autophagy related 16 like 1), ULK1 (Unc-51 like autophagy activating kinase 1), and Beclin1 (Chauhan *et al.*, 2016).

The ubiquitinated bacteria can be recognized by a group of adaptors featuring a ubiquitin-binding domain and a LC3-interacting region motif (Wu and Li, 2019). Typical adaptors include p62/SQSTM1, optineurin (OPTN) and NDP52 (nuclear domain 10 protein 52). As the first identified mammalian selective autophagy adaptor, p62 has been reported to be involved in the xenophagy of *S.* Typhimurium, Mtb, *Staphylococcus aureus* and *Shigella flexneri* (Komatsu *et al.*, 2007; Dupont *et al.*, 2009; Watson *et al.*, 2012; Neumann *et al.*, 2016). The activity of OPTN in xenophagy is modulated by phosphorylation modification, which can enhance autophagic clearance of cytosolic *Salmonella* (Wild *et al.*, 2011). NDP52 plays a dual role in xenophagy: the targeting of pathogens to autophagosomes by its selective autophagy adaptor activity and the control of pathogen-containing autophagosome maturation by interacting with ATG8 orthologs (Verlhac *et al.*, 2015).

Ubiquitin-independent xenophagy

Ubiquitin-independent pathways also play an important role in xenophagy. Adaptors link the non-ubiquitylated cargos to the autophagy machinery by targeting bacteria residing vacuoles or damaged vacuolar membranes. The adaptors can response to a wide variety of protein-, lipid- or sugar-based signals, which include galectin, complement protein C3 and NOD (nucleotide binding oligomerization domain) proteins (Fig. 1). As a danger receptor, Galectin-8 targets damaged *Salmonella*-containing vacuoles (SCVs) by binding glycans and then initiates the upstream autophagy machinery to invading bacteria by recruiting NDP52 (Thurston *et al.*, 2012). Specifically, NDP52 forms a trimeric complex with FIP200 and SINTBAD, which are the subunit and adaptor of ULK and TBK1 complexes, respectively (Ravenhill *et al.*, 2019). However, Galectin-8-dependent recruitment of NDP52 to SCVs is transient and subsequently by ubiquitin-dependent NDP52 recruitment. The proton pump V-ATPase can also sense vacuolar damage and then recruit ATG16L1 onto bacteria-containing vacuoles to initiate LC3 lipidation (Xu *et al.*, 2019). *In vivo*, the complement protein C3 is deposited on

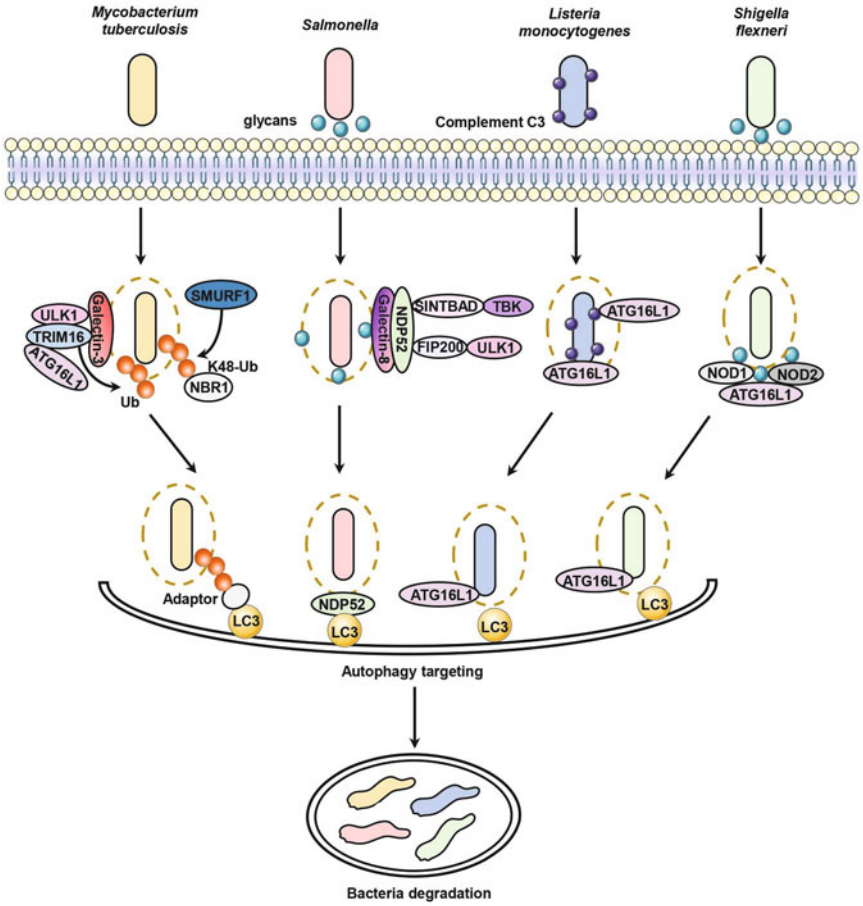

Fig. 1. Recognition of invading bacteria by host autophagy system. In the ubiquitin-adaptor protein-LC3 xenophagy model, E3 ubiquitin ligases (SMURF1 and TRIM) mediate ubiquitination of *Mycobacterium tuberculosis* (Mtb) and recruitment of autophagy machinery components (adaptors, autophagy regulators ATG16L1, ULK1 and LC3), and thus restrict Mtb invasion; In the ubiquitin-independent xenophagy models, galectin, complement protein C3 and NOD-based signals can induce the recognition of intracellular bacteria (*Salmonella*, *Listeria monocytogenes*, *Shigella flexneri*) to initiate the upstream autophagy machinery (adaptors, autophagy regulators ATG16L1 and ULK1). Ub, ubiquitin.

most invasive pathogens (such as Lm) and then carried into host cells, where it increases autophagy targeting and growth restriction through a direct interaction with ATG16L1 (Merle *et al.*, 2015; Sorbara *et al.*, 2018). NOD1 and NOD2 are also critical for recognition of intracellular bacteria through peptidoglycan detection at the site of bacterial entry in plasma membrane, where they recruit ATG16L1 to initiate xenophagy (Travassos *et al.*, 2010; Philpott *et al.*, 2014).

The Regulation of Xenophagy on the Elimination of Different Infecting Bacteria

As an important part of defense mechanism, autophagy targets a variety of bacteria, including bacteria in cytoplasm (Group A *Streptococcus*), in immature phagosomes (Mtb), or in damaged phagocytic vacuoles (*S.* Typhimurium). Owing to different intracellular pathogens adopt distinct lifestyles within host cells, the process of xenophagy also varies among different pathogens. Next, we will discuss the recent advances on the mechanisms by which xenophagy eliminates different bacteria, with a particular focus on extensively studied ones including Group A *Streptococcus* (GAS), *S.* Typhimurium and Mtb (Fig. 2).

Group A *Streptococcus* (GAS)

Group A *Streptococcus* is a clinically leading pathogen of diverse mild (pharyngitis and impetigo) and severe (toxic shock-like syndrome, acute poststreptococcal glomerulonephritis, acute rheumatic fever and rheumatic heart disease) diseases (Walker *et al.*, 2014). Tamotsu Yoshimori first found that xenophagy can act as an innate defense system against invasive GAS within nonphagocytic cells (Nakagawa *et al.*, 2004). Intracellular GAS resides in the LC3-decorated compartments during xenophagy, and the phosphorylation of LC3 by hippo kinases STK3/STK4 (serine/threonine kinase 3/4) is essential for the fusion of autophagosomes with lysosomes (Fig. 2) (Wilkinson *et al.*, 2015). In addition to LC3, Beclin1 also plays an important role in GAS-induced autophagy. Beclin1 mainly interacts with UVRAG (ultraviolet radiation resistance-associated gene protein) during GAS infection and promotes the formation of autolysosomes. However, NLRX1 (NOD-like receptor family member X1) can negatively regulate GAS-induced autophagy by interacting with the Beclin1-UVRAG complex (Fig. 2) (Nakajima *et al.*, 2017; Aikawa *et al.*, 2018). CD46 is a ubiquitous surface receptor for GAS, and the engagement of CD46-Cyt-1/GOPC-Beclin1 pathway induces autophagic degradation of GAS to restrict early pathogen infection (Fig. 2) (Cattaneo, 2004; Joubert *et al.*, 2009). Furthermore, the Rab GTPase family members play distinct roles during autophagy against GAS (Fig. 2): Rab9A is involved in GAS-containing autophagosome-like vacuole (GcAV) enlargement and lysosomal fusion (Nozawa *et al.*, 2012), Rab17, Rab23, and Rab30 are essential for GcAV formation (Haobam *et al.*, 2014; Nakajima *et al.*, 2019).

Salmonella Typhimurium

Salmonella Typhimurium is a facultative intracellular pathogen and an important cause of many host-specific diseases, such as gastroenteritis in humans (LaRock *et al.*, 2015). After invading host cells, this pathogen typically resides within a vacuolar compartment termed SCV. The fate of the SCV is

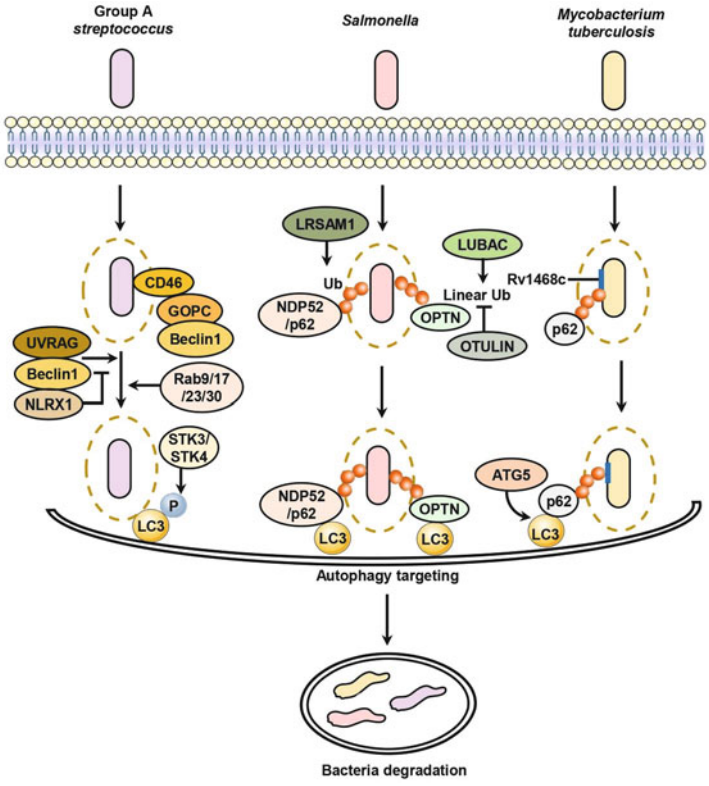

Fig. 2. The mechanisms of xenophagy clearance of three representative bacteria. STK3/STK4-LC3, Beclin1-UVRAG, CD46-GOPC-Beclin1 pathways and Rab GTPase family induce autophagic degradation of Group A *Streptococcus* to restrict pathogen infection; E3 ligases LRSAM1 and LUBAC trigger ubiquitination events and thus regulates *S.* Typhimurium-induced autophagy through the recognition of adaptors (NDP52, p62 and OPTN); ATG5 is required for Rv1468c-Ub-p62-LC3 interaction-mediated host xenophagy clearance of *Mycobacterium tuberculosis*. Ub, ubiquitin.

modified by two independent type III secretion systems (T3SS) encoded on *Salmonella* pathogenicity islands 1 and 2 (SPI-1 and SPI-2) (LaRock *et al.*, 2015), which mediates the main pathogenesis for *S.* Typhimurium to invade and colonize host cells (Haraga *et al.*, 2008). Specifically, the SPI-1 secretion system (T3SS1) functions to damage eukaryotic cell membranes and therefore enable bacterial invasion, while the SPI-2 secretion system (T3SS2) facilitates the replication of intracellular bacteria within SCVs (Casanova, 2017). Intriguingly, T3SS1 can also damage the SCVs, which has been suggested to be the main reason for autophagic capture of cytosolic *S.* Typhimurium (Birmingham *et al.*, 2006). Subsequent to the extensive membrane damage and cytosolic entry, LRSAM1 (leucine rich repeat and sterile alpha motif containing 1), LUBAC (linear ubiquitin chain assembly complex) and other host E3 ligases trigger ubiquitination events (Fig. 2). LRSAM1 detects *S.* Typhimurium via its leucine-rich repeat domain and induces ubiquitination via RING domain (Huett *et al.*, 2012). LUBAC is the only E3 ligase complex found to mediate cargos ubiquitination in a linear form, and thus regulates *S.* Typhimurium-induced autophagy initiation through OPTN (Chu *et al.*, 2020). In contrast, deubiquitinating enzymes (DUBs) can specifically hydrolyze ubiquitin chains from cargos and thus interfere with xenophagy. A typical example is OTULIN (ovarian tumor domain-containing deubiquitinase with linear linkage specificity). The depletion of OTULIN can increase the formation of linear ubiquitin coat around cytosolic *S.* Typhimurium and restrict bacterial proliferation (van Wijk *et al.*, 2017).

The most extensively studied adaptors are NDP52 and p62, and they are both required for the autophagy against *S.* Typhimurium (Fig. 2). Through recruiting to pathogen independently and targeting distinct microdomains on *S.* Typhimurium, the adaptors drive effective xenophagy cooperatively (Zheng *et al.*, 2009; Cemma *et al.*, 2011). Modulation of xenophagy and lysosomal pathway can inhibit *S.* Typhimurium infection, and small molecule xenophagy regulators have been identified to effectively curb *S.* Typhimurium survival, such as acacetin. Acacetin treatment can maintain transcription factor EB (a regulator of autophagy and lysosome biogenesis)-mediated xenophagy to reduce intracellular *S.* Typhimurium burden (Ammanathan *et al.*, 2020).

Mycobacterium tuberculosis (Mtb)

Mycobacterium tuberculosis is the main cause for serious infectious disease tuberculosis that remains a severe global public health problem (Vergne *et al.*, 2004). In addition to primarily attacking the lung, the bacteria can also attack many parts of the body including the kidney, spine, and brain. Fortunately, humans have evolved several innate immune defenses against the bacteria infection. Xenophagy is one of such innate defenses, which inhibits *mycobacterial* survival via promoting the acidification and maturation of *mycobacterial* phagosomes (Gutierrez *et al.*, 2004). Several autophagy factors (ATG5, OPTN, p62, and LC3) have been revealed to act against Mtb infection (Fig. 2). Especially, ATG5 is essential for the elimination of invading Mtb by inhibiting neutrophil-mediated immunopathology. However, loss of many other conventional autophagy genes does not affect outcome. Therefore, further researches are needed to clarify whether ATG5 restrict Mtb infection through a non-canonical autophagy pathway (Behar and Baehrecke, 2015; Kimmey *et al.*, 2015). ATG5 is also required for ubiquitin-Rv1468c interaction mediated host immune clearance of Mtb (Chai *et al.*, 2019). In addition, both OPTN and p62 are required for autophagic defense against Mtb (Zhang *et al.*, 2019).

To target autophagy, several potential reagents have been identified to improve the outcome of current antibiotic-based treatment of tuberculosis. The candidates include: (a) vitamin D receptor signaling including 1,25-dihydroxyvitamin D3, 4-phenylbutyrate and IFN-γ, which can induce the autophagy activation and/or phagosome-lysosome fusion (Yuk *et al.*, 2009; Fabri *et al.*, 2011; Rekha *et al.*, 2015). (b) Nuclear receptors (NRD1, PPARα, and ERR) and agonists that regulate the transcription and/or post-translation of autophagy genes/proteins (Chandra *et al.*, 2015; Kim *et al.*, 2017a, 2018b). (c) The mediators involved in AMPK pathway modulate autophagy activation, such as calcium-mobilizing agents, phytochemicals, resveratrol and cyclic peptides (Palucci and Delogu, 2018). (d) The inhibitors of mTOR (mammalian target of rapamycin) signaling, such as rapamycin and other analogs (sirolimus, temsirolimus, and everolimus) (Floto *et al.*, 2007; Singh and Subbian, 2018), of which everolimus presents as a good candidate drug because of the higher bioavailability and lower vascular inflammation (Cerni *et al.*, 2019). It is worth noting that mTOR inhibition interferes with phagosomal maturation but increases the replication of Mtb during HIV/Mtb co-infection, indicating that autophagy induction as a potential treatment method for tuberculosis should be handled with caution in the context of HIV/Mtb co-infection (Andersson *et al.*, 2016). (e) Other autophagy-targeting small molecules/chemicals, including gefitinib, verapamil and baicalin (Paik *et al.*, 2019).

Other bacteria

In addition to the above pathogens, a number of other intracellular pathogens can also be recognized and restricted by host autophagy system, including Lm, *Shigella flexneri*, *Acinetobacter baumannii*, *Legionella pneumophila*, *Bacillus*, and *Burkholderia* (Gomes and Dikic, 2014; Siqueira *et al.*, 2018). The mechanisms that cells use to target different bacteria for autophagic degradation has both overlaps and differences. For example, Lm can be sensed and targeted by at least two pathways, including NOD1-ATG16L1-LC3 and ubiquitin-p62-LC3 pathways (Ogawa *et al.*, 2011). Similarly, *Shigella flexneri*, an intestinal pathogen, can also be degraded through these two pathways (Travassos *et al.*, 2010). In fact, in most cases, LC3-decorated autophagosomes are formed and bring the target bacteria into lysosome for degradation (Huang and Brumell, 2014). Although in some cases, different bacteria share the same molecular events, but the autophagy responses vary among different bacterial infections. For instance, complement protein C3/ATG16L1 interaction drives autophagy-dependent elimination of invasive Lm, on the contrary, *Shigella flexneri* and *Salmonella* can use proteases to cleave bound C3 and escape C3-mediated xenophagy restriction despite the early complement C3-dependent also increases autophagy targeting (Sorbara *et al.*, 2018).

The Mechanisms of Bacteria to Escape from Xenophagy

Autophagy usually serves as a defense mechanism to limit most pathogens survival within host cells, however, some bacteria have evolved various strategies to evade autophagic recognition, to paralyze host autophagy system, and even to hijack autophagy for their proliferation. These strategies used by successful pathogens to escape from host autophagy system include the secretion of effectors, microRNAs, and lipid virulence factors. Effectors are secreted into the host cytoplasm via type I-VII secretion systems and are the common strategy to suppress autophagy initiation, autophagosome formation and/or autophagosome-lysosomes fusion. Next, we will review the represent examples of these autophagic escape mechanisms.

Effectors secreted by bacteria suppress xenophagy

SopF secreted by *Salmonella* inhibits the autophagic recognition: although *S.* Typhimurium is often used as a biological model for studying xenophagy, only a small fraction (10–20%) of intracellular *S.* Typhimurium can be targeted by autophagy in the early stages of infections (Birmingham *et al.*, 2006). SopF, a novel conserved *S.* Typhimurium T3SS effector, is identified responsible for the inhibition of *S.* Typhimurium-induced xenophagy (Cheng *et al.*, 2017). As a phosphoinositide binding effector, SopF is essential for maintaining the integrity of the nascent SCV membrane and thus increases SCV stability and inhibits autophagic capture of *S.* Typhimurium (Lau *et al.*, 2019). In addition, SopF can specifically disrupt V-ATPase-ATG16L1 axis to block the recruitment of autophagy proteins, and thereby escape from the immune recognition of host cells (Xu *et al.*, 2019) (Fig. 3). Specifically, upon SCV damage, V-ATPase recruits ATG16L1 and initiates LC3 lipidation, however, SopF catalyzes ADP-ribosylation of ATP6V0C on Gln124, which specifically disrupts recruitment of ATG16L1 by the V-ATPase and thus promotes bacterial growth. In the early stages of infection, the SopF is secreted in low levels and its autophagic inhibitory effect will be gradually enhanced with duration of infection, which answers why *S.* Typhimurium-induced autophagy is a transient process. Notably, SopF is a broad-spectrum xenophagy-specific inhibitor, but does not affect canonical autophagy. In addition, SopF and SopB (another T3SS effector) act antagonistically to regulate SCV stability (Lau *et al.*, 2019), therefore, it is suspected that SopB may counteract SopF-induced autophagy inhibition.

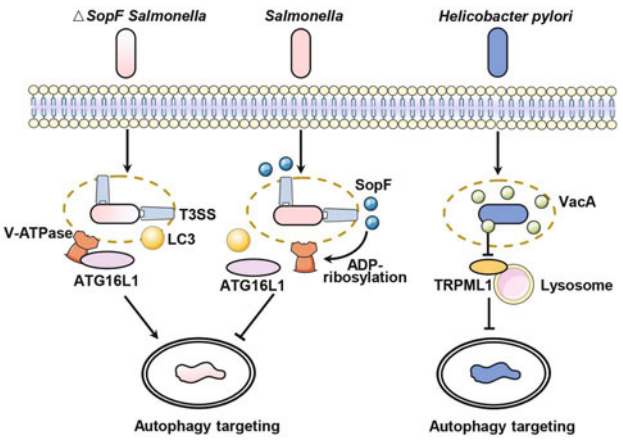

Fig. 3. The mechanisms of bacteria-secreted effectors with inhibitory effect in xenophagy. T3SS effector SopF secreted by *Salmonella* inhibits infection-induced V-ATPase-ATG16L1 axis to block autophagy; Virulence factor VacA secreted by *Helicobacter pylori* inhibits the activity of TRPML1 and impairs lysosome trafficking and autophagosomes maturation. T3SS, type III secretion system.

VacA secreted by *Helicobacter pylori* interferes with the endolysosomal trafficking and autophagosome maturation: vacuolating cytotoxin A (VacA) is a critical virulence factor of *Helicobacter pylori* (Hp), which is a gram-negative human gastric pathogen and the main risk factor of gastric cancer development (Hooi *et al.*, 2017). The effects of VacA on the host autophagy system are determined by the duration/level of exposure to the toxin (Ricci, 2016). VacA can induce autophagy as a cytoprotective effect to restrict invasive bacteria during acute Hp infection. For example, VacA promote autophagic cell death to limit cellular damage via endoplasmic reticulum stress signaling or connexin 43 accumulation in gastric epithelial cells (Terebiznik *et al.*, 2009; Yahiro *et al.*, 2012, 2015; Foegeding *et al.*, 2019). In contrast, VacA disrupts host autophagy and promotes bacterial survival during chronic Hp infection. The main reason for such a phenomenon is prolonged exposure to VacA impairs endolysosomal trafficking and autophagosome maturation, leading to the accumulation of dysfunctional lysosomes and autolysosomes (Greenfield and Jones, 2013). For example, chronic VacA exposure inhibits the activity of lysosomal calcium channel TRPML1 (transient receptor potential membrane channel mucolipin 1) to disrupt endolysosomal trafficking or induce disarmed autophagosomes (lacking of cathepsin-D) accumulation, leading to the generation of a protective intracellular reservoir for Hp survival (Fig. 3) (Raju *et al.*, 2012; Capurro *et al.*, 2019). In some cases, VacA interacts with other effectors to impair autophagy pathway. For instance, VacA promotes cytotoxin associated gene A (CagA) accumulation to disrupt endolysosomal trafficking and autophagosome maturation (Abdullah *et al.*, 2019). In addition, VacA perturbations in mitochondria disrupts cellular amino acid homeostasis, which also drives the inhibition of mTORC1 signaling and autophagic activities (Kim *et al.*, 2018a).

Other bacterial secreted effectors: in addition to the above effectors, multiple effectors secreted by bacteria are exported to the host-pathogen interface to interfere with the xenophagy machinery at different stages (Table 1). These effectors include: (a) effectors inhibiting autophagic recognition, such as SseI, SdeA, InIK, and ActA. As two DUBs, *Salmonella*-secreted SseI and *Legionella pneumophila*-secreted SdeA, can deubiquitinate cargos to inhibit the recognition of host autophagy system (Mesquita *et al.*, 2012; Sheedlo *et al.*, 2015). InIK and ActA recruit corresponding host partners to protect Lm from autophagic recognition (Yoshikawa *et al.*, 2009; Dortet *et al.*, 2011). SpeB cysteine protease degrades autophagy adaptor proteins p62, NDP52, and NBR1, thereby allowing GAS to escape autophagic recognition (Barnett *et al.*,

Table 1. Examples of bacterial effectors with inhibitory effect in xenophagy

Pathogen	Effector	Function	References
Salmonella	SopF	SopF specifically modifies V-ATPase to inhibit the autophagic recognition	Xu *et al.* (2019)
	SseI	SseL inhibits selective autophagy of cytosolic aggregates	Mesquita *et al.* (2012)
Helicobacter pylori	VacA	VacA impairs endolysosomal trafficking and autophagosome maturation	Raju *et al.* (2012) Capurro *et al.* (2019)
Listeria monocytogenes	InlK	InlK recruits major vault protein to avoid autophagy	Dortet *et al.* (2011)
	ActA	ActA recruits the Arp2/3 complex and Ena/VASP to escape from autophagic recognition	Yoshikawa *et al.* (2009)
Legionella pneumophila	SdeA	SdeA deubiquitinates Lys63-linked chains from the phagosomal surface to antagonize autophagy	Sheedlo *et al.* (2015)
	RavZ	RavZ recognizes and splits LC3-PE to avoid autophagy	Choy *et al.* (2012)
	*Lp*SPL	*Lp*SPL targets host's sphingolipid metabolism to restrain autophagy	Rolando *et al.* (2016)
Group A *Streptococcus*	SpeB	SpeB degrades p62, NDP52 and NBR1 to escape the host autophagy	Barnett *et al.* (2013)
Mycobacterium tuberculosis	SapM and PknG	SapM and PknG works in concert to arrest phagosome and autophagosome maturation	Zulauf *et al.* (2018)

2013). (b) Effectors directly interfering with key autophagy factors, such as *Legionella pneumophila*-secreted RavZ, which can induce irreversible LC3 deconjugation (Choy *et al.*, 2012). (c) Effectors impairing autophagosome maturation or blocking the fusion of autophagosome and lysosome, such as SapM and PknG, which arrest phagosome and autophagosome maturation during Mtb infection (Zulauf *et al.*, 2018). (d) Effectors modulating the host's metabolism, such as *Legionella pneumophila*-secreted *Lp*SpL, which targets the sphingolipid metabolism of host cells to exploit the host autophagy system (Rolando *et al.*, 2016).

MicroRNAs: the important mediators of bacteria to escape from xenophagy

MicroRNAs (miRNAs) are small non-coding RNA with a length of about 22 nucleotides, which can specifically regulate target gene expression via post-transcriptional mechanisms (Aredia and Scovassi, 2017). Increasing evidences suggest that various pathogens evade host autophagy system using miRNA-dependent mechanisms (Silwal *et al.*, 2020). Considering Mtb is one of the most extensively studied pathogens in xenophagy, thus, here we focus on recent researches regarding the roles of miRNAs in autophagy regulation during Mtb infection (Table 2). Several miRNAs act as autophagy inhibitors to regulate Mtb survival, such as miR-30A (Chen *et al.*, 2015), miR-27A (Liu *et al.*, 2018), miR-33 (Ouimet *et al.*, 2016), miR-1958 (Ding *et al.*, 2019), miR-129-3p (Qu *et al.*, 2019), miR-18a (Yuan *et al.*, 2020), miR-144* (Kim *et al.*, 2017b), miR-125b-5p (Liu *et al.*, 2020), miR-125a-3p (Kim *et al.*, 2015), miR-26a (Sahu *et al.*, 2017), and miR-155 (Etna *et al.*, 2018). These miRNAs inhibit integrated pathways involved in autophagy and lysosomal function by targeting the key autophagic effectors (such as Beclin1, ATG5, and LC3). Interestingly, miR-155 plays two opposite roles in regulating autophagy, depending on the host cell type and bacterial strain. Mtb infection of human dendritic cells manipulates miR-155 expression to target ATG3 for its own survival. However, miR-155 can also act as an autophagy activator to eliminate Mtb in murine macrophages by targeting Rheb, a negative regulator of autophagy (Wang *et al.*, 2013). In addition to miR-155, miR-17-5p is another autophagy inducer of xenophagy during Mtb infection by targeting Mcl-1 and STAT3 (Kumar *et al.*, 2016). In addition, most of the current studies on miRNAs

Table 2. The main microRNAs associated with xenophagy during *Mycobacterium tuberculosis* infection

miRNA	Host cell	Expersion after infection	Target gene	Autophagy	Bacterial survival	References
miR-30A	THP-1	↑	Beclin1	↓	↑	Chen *et al.* (2015)
miR-27A	PBMC from TB patients, murine macrophages, *in vivo*	↑	CACNA2D3	↓	↑	Liu *et al.* (2018)
miR-33/miR-33*	Murine macrophages, THP-1, HEK293, *in vivo*	↑	ATG5, ATG12, LAMP1 and LC3B	↓	↑	Ouimet *et al.* (2016)
miR-1958	RAW264.7	↑	ATG5	↓	↑	Ding *et al.* (2019)
miR-129-3p	RAW264.7	↑	ATG4B	↓	↑	Qu *et al.* (2019)
miR-18a	RAW264.7	↑	ATM	↓	↑	Yuan *et al.* (2020)
miR-144*/ hsa-miR-144-5p	Human monocytes and macrophages	↑	DRAM2	↓	↑	Kim *et al.* (2017b)
miR-125b-5p	PBMC from TB patients, THP-1	↑	DRAM2	↓	↑	Liu *et al.* (2020)
miR-125a-3p	Murine macrophages, *in vivo*		UVRAG	↓	↑	Kim *et al.* (2015)
miR-26a	Murine macrophages, *in vivo*	↑	KLF4	↓	↑	Sahu *et al.* (2017)
miR-155	Human dendritic cells	↑	ATG3	↓	↑	Etna *et al.* (2018)
miR-155	Murine macrophages, RAW264.7, *in vivo*	↑	Rheb	↑	↓	Wang *et al.* (2013)
miR-17-5p	Murine macrophages, RAW264.7	↓	Mcl-1, STAT3	↓	↑	Kumar *et al.* (2016)

regulating xenophagy are conducted *in vitro*. Therefore, more *in vivo* models need to be developed to evaluate the clinical value of miRNAs during bacterial infection.

The role of lipid virulence factors in xenophagy

The substance that contributes to the virulence of bacteria is called virulence factor, which is often related to the complex lipid components contained in bacteria. The lipid-rich cell wall of bacteria is a dynamic structure that participates in the transportation of nutrients and cytotoxic effectors. Thus, some certain virulence-associated lipids can help bacteria to survive long-term in host cells (Queiroz and Riley, 2017). Mannose-capped lipoarabinomannan is a *mycobacterial* cell wall component and one of the key virulence factors of Mtb, which significantly inhibits phagosomal maturation and hijacks autophagic mechanisms for bacterial survival (Shui *et al.*, 2011; Vergne *et al.*, 2014). Sulfoglycolipids (SLs) and phthiocerol dimycocerosates (PDIMs), another two major lipid virulence factors of Mtb, can also control autophagy-related pathways in host cells (Quigley *et al.*, 2017; Augenstreich *et al.*, 2019). The loss of PDIMs and SLs increases LC3 recruitment to Mtb compartments and promotes autophagy activation (Bah *et al.*, 2020). In summary, these lipid virulence factors can be used as potential therapeutic targets for restricting bacterial infection and powerful tools for further explore of xenophagy mechanism, which will help develop autophagy-based antibacterial vaccines.

Conclusions and Future Prospects

This review summarizes recent insights into the crosstalk between bacterial infection and host autophagy system. (1) For host defense, host cells use autophagy system as a defensive mechanism to eliminate intracellular bacteria through ubiquitin-dependent or independent pathways. (2) For bacteria offense, successful pathogens secrete effectors, utilize lipid virulence factors and/or regulate miRNAs expression to escape from or paralyze host autophagy system for their own survival and replication. In fact, the frequency of xenophagy is very low in most models. Therefore, it is difficult to conduct in-depth research on the mechanisms of xenophagy. In the ubiquitin-adaptor protein-LC3 model, knocking out any genes encoding ubiquitin substrate proteins, ubiquitin ligases, or adaptor proteins cannot completely inhibit xenophagy. Although the combination of immunotherapy and autophagy have been studied in clinical treatments and several autophagy-related antibacterial drugs have become the focus of research, there are many unresolved questions deserve further investigation. (1) The mechanism underlying autophagic recognition of intracellular bacteria: how does the host cells perceive the invading bacteria and specifically induce xenophagy? How ubiquitin ligases and adaptors discriminate and recognize multiple potential cargos for autophagic degradation within same cell? What are the connections between multiple redundant adaptors? Why adaptors directly recruit LC3 downstream of autophagy pathway, and how are upstream autophagy proteins activated? Does xenophagy triggered by different bacteria use the same mechanism? (2) The mechanisms of bacteria to escape from xenophagy: do all bacteria have their own unique escape mechanism? Can multiple secreted effectors synergistically assist bacteria to escape from autophagy? Does bacterial infection cause changes in the entire miRNA expression profile? Are these miRNAs synergistic or antagonistic to regulate autophagy? Therefore, a better understanding of mechanisms underlying the crosstalk between bacteria and host autophagy would help to answer the above question and provide new targets for therapeutic intervention during bacterial infection.

Acknowledgements

This work was supported by the National Natural Science Funds of China (81903138, 31871198), Cooperative Innovation Center of Engineering and New Products for Developmental Biology of Hunan Province (20134486), and the Opening Fund of The National & Local Joint Engineering Laboratory of Animal Peptide Drug Development (Hunan Normal University, National Development and Reform Commission).

Conflict of Interest

Authors declare that they are no conflicts of interest.

Author Contributions

F.W. and L.Z. wrote the manuscript, and F.W. and G.L. revised the manuscript.

References

Abdullah, M., Greenfield, L.K., Bronte-Tinkew, D., Capurro, M.I., Rizzuti, D., and Jones, N.L. 2019. VacA promotes CagA accumulation in gastric epithelial cells during *Helicobacter pylori* infection. *Sci. Rep.* **9**, 38.

Aikawa, C., Nakajima, S., Karimine, M., Nozawa, T., Minowa-Nozawa, A., Toh, H., Yamada, S., and Nakagawa, I. 2018. NLRX1 negatively regulates Group A *Streptococcus* invasion and autophagy induction by interacting with the Beclin1-UVRAG complex. *Front. Cell Infect. Microbiol.* **8**, 403.

Ammanathan, V., Mishra, P., Chavalmane, A.K., Muthusamy, S., Jadhav, V., Siddamadappa, C., and Manjithaya, R. 2020. Restriction of intracellular *Salmonella* replication by restoring TFEB-mediated xenophagy. *Autophagy* **16**, 1584–1597.

Andersson, A.M., Andersson, B., Lorell, C., Raffetseder, J., Larsson, M., and Blomgran, R. 2016. Autophagy induction targeting mTORC1 enhances *Mycobacterium tuberculosis* replication in HIV co-infected human macrophages. *Sci. Rep.* **6**, 28171.

Anding, A.L. and Baehrecke, E.H. 2017. Cleaning house: selective autophagy of organelles. *Dev. Cell* **41**, 10–22.

Aredia, F. and Scovassi, A.I. 2017. A new function for miRNAs as regulators of autophagy. *Future Med. Chem.* **9**, 25–36.

Augenstreich, J., Haanappel, E., Ferré, G., Czaplicki, G., Jolibois, F., Destainville, N., Guilhot, C., Milon, A., Astarie-Dequeker, C., and Chavent, M. 2019. The conical shape of DIM lipids promotes *Mycobacterium tuberculosis* infection of macrophages. *Proc. Natl. Acad. Sci. USA* **116**, 25649–25658.

Bah, A., Sanicas, M., Nigou, J., Guilhot, C., Astarie-Dequeker, C., and

Vergne, I. 2020. The lipid virulence factors of *Mycobacterium tuberculosis* exert multilayered control over autophagy-related pathways in infected human macrophages. *Cells* **9**, 666.

Barnett, T.C., Liebl, D., Seymour, L.M., Gillen, C.M., Lim, J.Y., Larock, C.N., Davies, M.R., Schulz, B.L., Nizet, V., Teasdale, R.D., *et al.* 2013. The globally disseminated M1T1 clone of Group A *Streptococcus* evades autophagy for intracellular replication. *Cell Host Microbe* **14**, 675–682.

Behar, S.M. and Baehrecke, E.H. 2015. Tuberculosis: autophagy is not the answer. *Nature* **528**, 482–483.

Birmingham, C.L., Smith, A.C., Bakowski, M.A., Yoshimori, T., and Brumell, J.H. 2006. Autophagy controls *Salmonella* infection in response to damage to the *Salmonella*-containing vacuole. *J. Biol. Chem.* **281**, 11374–11383.

Capurro, M.I., Greenfield, L.K., Prashar, A., Xia, S., Abdullah, M., Wong, H., Zhong, X.Z., Bertaux-Skeirik, N., Chakrabarti, J., Siddiqui, I., *et al.* 2019. VacA generates a protective intracellular reservoir for *Helicobacter pylori* that is eliminated by activation of the lysosomal calcium channel TRPML1. *Nat. Microbiol.* **4**, 1411–1423.

Casanova, J.E. 2017. Bacterial autophagy: offense and defense at the host-pathogen interface. *Cell. Mol. Gastroenterol. Hepatol.* **4**, 237–243.

Cattaneo, R. 2004. Four viruses, two bacteria, and one receptor: membrane cofactor protein (CD46) as pathogens' magnet. *J. Virol.* **78**, 4385–4388.

Cemma, M., Kim, P.K., and Brumell, J.H. 2011. The ubiquitin-binding adaptor proteins p62/SQSTM1 and NDP52 are recruited independently to bacteria-associated microdomains to target *Salmonella* to the autophagy pathway. *Autophagy* **7**, 341–345.

Cerni, S., Shafer, D., To, K., and Venketaraman, V. 2019. Investigating the role of everolimus in mTOR inhibition and autophagy promotion as a potential host-directed therapeutic target in *Mycobacterium tuberculosis* infection. *J. Clin. Med.* **8**, 232.

Chai, Q., Wang, X., Qiang, L., Zhang, Y., Ge, P., Lu, Z., Zhong, Y., Li, B., Wang, J., Zhang, L., *et al.* 2019. A *Mycobacterium tuberculosis* surface protein recruits ubiquitin to trigger host xenophagy. *Nat. Commun.* **10**, 1973.

Chandra, V., Bhagyaraj, E., Nanduri, R., Ahuja, N., and Gupta, P. 2015. NR1D1 ameliorates *Mycobacterium tuberculosis* clearance through regulation of autophagy. *Autophagy* **11**, 1987–1997.

Chauhan, S., Kumar, S., Jain, A., Ponpuak, M., Mudd, M.H., Kimura, T., Choi, S.W., Peters, R., Mandell, M., Bruun, J.A., *et al.* 2016. TRIMs and galectins globally cooperate and TRIM16 and Galectin-3 co-direct autophagy in endomembrane damage homeostasis. *Dev. Cell* **39**, 13–27.

Chen, Z., Wang, T., Liu, Z., Zhang, G., Wang, J., Feng, S., and Liang, J. 2015. Inhibition of autophagy by MiR-30A induced by *Mycobacteria tuberculosis* as a possible mechanism of immune escape in human macrophages. *Jpn. J. Infect. Dis.* **68**, 420–424.

Cheng, S., Wang, L., Liu, Q., Qi, L., Yu, K., Wang, Z., Wu, M., Liu, Y., Fu, J., Hu, M., *et al.* 2017. Identification of a novel *Salmonella* type III effector by quantitative secretome profiling. *Mol. Cell. Proteomics* **16**, 2219–2228.

Choy, A., Dancourt, J., Mugo, B., O'Connor, T.J., Isberg, R.R., Melia, T.J., and Roy, C.R. 2012. The Legionella effector RavZ inhibits host autophagy through irreversible Atg8 deconjugation. *Science* **338**, 1072–1076.

Chu, Y., Kang, Y., Yan, C., Yang, C., Zhang, T., Huo, H., and Liu, Y. 2020. LUBAC and OTULIN regulate autophagy initiation and maturation by mediating the linear ubiquitination and the stabilization of ATG13. *Autophagy* **17**, 1688–1699.

Ding, S., Qu, Y., Yang, S., Zhao, Y., and Xu, G. 2019. Novel miR-1958 promotes *Mycobacterium tuberculosis* survival in RAW264.7 cells by inhibiting autophagy via Atg5. *J. Microbiol. Biotechnol.* **29**, 989–998.

Dortet, L., Mostowy, S., Samba-Louaka, A., Gouin, E., Nahori, M.A., Wiemer, E.A.C., Dussurget, O., and Cossart, P. 2011. Recruitment of the major vault protein by InlK: a *Listeria monocytogenes* strategy to avoid autophagy. *PLoS Pathog.* **7**, e1002168.

Dupont, N., Lacas-Gervais, S., Bertout, J., Paz, I., Freche, B., Van Nhieu, G.T., van der Goot, F.G., Sansonetti, P.J., and Lafont, F. 2009. *Shigella* phagocytic vacuolar membrane remnants participate in the cellular response to pathogen invasion and are regulated by autophagy. *Cell Host Microbe* **6**, 137–149.

Etna, M.P., Sinigaglia, A., Grassi, A., Giacomini, E., Romagnoli, A., Pardini, M., Severa, M., Cruciani, M., Rizzo, F., Anastasiadou, E., *et al.* 2018. *Mycobacterium tuberculosis*-induced miR-155 subverts autophagy by targeting ATG3 in human dendritic cells. *PLoS Pathog.* **14**, e1006790.

Fabri, M., Stenger, S., Shin, D.M., Yuk, J.M., Liu, P.T., Realegeno, S., Lee, H.M., Krutzik, S.R., Schenk, M., Sieling, P.A., *et al.* 2011. Vitamin D is required for IFN-γ-mediated antimicrobial activity of human macrophages. *Sci. Transl. Med.* **3**, 104ra102.

Floto, R.A., Sarkar, S., Perlstein, E.O., Kampmann, B., Schreiber, S.L., and Rubinsztein, D.C. 2007. Small molecule enhancers of rapamycin-induced TOR inhibition promote autophagy, reduce toxicity in Huntington's disease models and enhance killing of mycobacteria by macrophages. *Autophagy* **3**, 620–622.

Foegeding, N.J., Raghunathan, K., Campbell, A.M., Kim, S.W., Lau, K.S., Kenworthy, A.K., Cover, T.L., and Ohi, M.D. 2019. Intracellular degradation of *Helicobacter pylori* VacA toxin as a determinant of gastric epithelial cell viability. *Infect. Immun.* **87**, e00783-18.

Franco, L.H., Nair, V.R., Scharn, C.R., Xavier, R.J., Torrealba, J.R., Shiloh, M.U., and Levine, B. 2017. The ubiquitin ligase Smurf1 functions in selective autophagy of *Mycobacterium tuberculosis* and anti-tuberculous host defense. *Cell Host Microbe* **21**, 59–72.

Gatica, D., Lahiri, V., and Klionsky, D.J. 2018. Cargo recognition and degradation by selective autophagy. *Nat. Cell Biol.* **20**, 233–242.

Gomes, L.C. and Dikic, I. 2014. Autophagy in antimicrobial immunity. *Mol. Cell* **54**, 224–233.

Greenfield, L.K. and Jones, N.L. 2013. Modulation of autophagy by *Helicobacter pylori* and its role in gastric carcinogenesis. *Trends Microbiol.* **21**, 602–612.

Gutierrez, M.G., Master, S.S., Singh, S.B., Taylor, G.A., Colombo, M.I., and Deretic, V. 2004. Autophagy is a defense mechanism inhibiting BCG and *Mycobacterium tuberculosis* survival in infected macrophages. *Cell* **119**, 753–766.

Haobam, B., Nozawa, T., Minowa-Nozawa, A., Tanaka, M., Oda, S., Watanabe, T., Aikawa, C., Maruyama, F., and Nakagawa, I. 2014. Rab17-mediated recycling endosomes contribute to autophagosome formation in response to Group A *Streptococcus* invasion. *Cell. Microbiol.* **16**, 1806–1821.

Haraga, A., Ohlson, M.B., and Miller, S.I. 2008. Salmonellae interplay with host cells. *Nat. Rev. Microbiol.* **6**, 53–66.

Hooi, J.K.Y., Lai, W.Y., Ng, W.K., Suen, M.M.Y., Underwood, F.E., Tanyingoh, D., Malfertheiner, P., Graham, D.Y., Wong, V.W.S., Wu, J.C.Y., *et al.* 2017. Global Prevalence of *Helicobacter pylori* infection: systematic review and meta-analysis. *Gastroenterology* **153**, 420–429.

Huang, J. and Brumell, J.H. 2014. Bacteria-autophagy interplay: a battle for survival. *Nat. Rev. Microbiol.* **12**, 101–114.

Huett, A., Heath, R.J., Begun, J., Sassi, S.O., Baxt, L.A., Vyas, J.M., Goldberg, M.B., and Xavier, R.J. 2012. The LRR and RING domain protein LRSAM1 is an E3 ligase crucial for ubiquitin-dependent autophagy of intracellular *Salmonella* Typhimurium. *Cell Host Microbe* **12**, 778–790.

Joubert, P.E., Meiffren, G., Grégoire, I.P., Pontini, G., Richetta, C., Flacher, M., Azocar, O., Vidalain, P.O., Vidal, M., Lotteau, V., *et al.* 2009. Autophagy induction by the pathogen receptor CD46. *Cell Host Microbe* **6**, 354–366.

Kim, Y.S., Lee, H.M., Kim, J.K., Yang, C.S., Kim, T.S., Jung, M., Jin, H.S., Kim, S., Jang, J., Oh, G.T., *et al.* 2017a. PPAR-α activation

mediates innate host defense through induction of TFEB and lipid catabolism. *J. Immunol.* **198**, 3283–3295.

Kim, I.J., Lee, J., Oh, S.J., Yoon, M.S., Jang, S.S., Holland, R.L., Reno, M.L., Hamad, M.N., Maeda, T., Chung, H.J., *et al.* 2018a. *Helicobacter pylori* infection modulates host cell metabolism through VacA-dependent inhibition of mTORC1. *Cell Host Microbe* **23**, 583–593.

Kim, J.K., Lee, H.M., Park, K.S., Shin, D.M., Kim, T.S., Kim, Y.S., Suh, H.W., Kim, S.Y., Kim, I.S., Kim, J.M., *et al.* 2017b. *MIR144** inhibits antimicrobial responses against *Mycobacterium tuberculosis* in human monocytes and macrophages by targeting the autophagy protein DRAM2. *Autophagy* **13**, 423–441.

Kim, S.Y., Yang, C.S., Lee, H.M., Kim, J.K., Kim, Y.S., Kim, Y.R., Kim, J.S., Kim, T.S., Yuk, J.M., Dufour, C.R., *et al.* 2018b. ESRRA (estrogen-related receptor alpha) is a key coordinator of transcriptional and post-translational activation of autophagy to promote innate host defense. *Autophagy* **14**, 152–168.

Kim, J.K., Yuk, J.M., Kim, S.Y., Kim, T.S., Jin, H.S., Yang, C.S., and Jo, E.K. 2015. MicroRNA-125a inhibits autophagy activation and antimicrobial responses during mycobacterial infection. *J. Immunol.* **194**, 5355–5365.

Kimmey, J.M., Huynh, J.P., Weiss, L.A., Park, S., Kambal, A., Debnath, J., Virgin, H.W., and Stallings, C.L. 2015. Unique role for ATG5 in neutrophil-mediated immunopathology during *M. tuberculosis* infection. *Nature* **528**, 565–569.

Kohler, L.J. and Roy, C.R. 2017. Autophagic targeting and avoidance in intracellular bacterial infections. *Curr. Opin. Microbiol.* **35**, 36–41.

Komatsu, M., Waguri, S., Koike, M., Sou, Y.S., Ueno, T., Hara, T., Mizushima, N., Iwata, J., Ezaki, J., Murata, S., *et al.* 2007. Homeostatic levels of p62 control cytoplasmic inclusion body formation in autophagy-deficient mice. *Cell* **131**, 1149–1163.

Kumar, R., Sahu, S.K., Kumar, M., Jana, K., Gupta, P., Gupta, U.D., Kundu, M., and Basu, J. 2016. MicroRNA 17-5p regulates autophagy in *Mycobacterium tuberculosis*-infected macrophages by targeting Mcl-1 and STAT3. *Cell. Microbiol.* **18**, 679–691.

LaRock, D.L., Chaudhary, A., and Miller, S.I. 2015. Salmonellae interactions with host processes. *Nat. Rev. Microbiol.* **13**, 191–205.

Lau, N., Haeberle, A.L., O'Keeffe, B.J., Latomanski, E.A., Celli, J., Newton, H.J., and Knodler, L.A. 2019. SopF, a phosphoinositide binding effector, promotes the stability of the nascent *Salmonella*-containing vacuole. *PLoS Pathog.* **15**, e1007959.

Linares, J.F., Duran, A., Yajima, T., Pasparakis, M., Moscat, J., and Diaz-Meco, M.T. 2013. K63 polyubiquitination and activation of mTOR by the p62-TRAF6 complex in nutrient-activated cells. *Mol. Cell* **51**, 283–296.

Liu, F., Chen, J., Wang, P., Li, H., Zhou, Y., Liu, H., Liu, Z., Zheng, R., Wang, L., Yang, H., *et al.* 2018. MicroRNA-27a controls the intracellular survival of *Mycobacterium tuberculosis* by regulating calcium-associated autophagy. *Nat. Commun.* **9**, 4295.

Liu, G., Wan, Q., Li, J., Hu, X., Gu, X., and Xu, S. 2020. Silencing miR-125b-5p attenuates inflammatory response and apoptosis inhibition in mycobacterium tuberculosis-infected human macrophages by targeting DNA damage-regulated autophagy modulator 2 (DRAM2). *Cell Cycle* **19**, 3182–3194.

Merle, N.S., Noe, R., Halbwachs-Mecarelli, L., Fremeaux-Bacchi, V., and Roumenina, L.T. 2015. Complement system part II: role in immunity. *Front. Immunol.* **6**, 257.

Mesquita, F.S., Thomas, M., Sachse, M., Santos, A.J., Figueira, R., and Holden, D.W. 2012. The *Salmonella* deubiquitinase SseL inhibits selective autophagy of cytosolic aggregates. *PLoS Pathog.* **8**, e1002743.

Mizushima, N. and Komatsu, M. 2011. Autophagy: renovation of cells and tissues. *Cell* **147**, 728–741.

Nakagawa, I., Amano, A., Mizushima, N., Yamamoto, A., Yamaguchi, H., Kamimoto, T., Nara, A., Funao, J., Nakata, M., Tsuda, K., *et al.* 2004. Autophagy defends cells against invading Group A *Streptococcus*. *Science* **306**, 1037–1040.

Nakajima, S., Aikawa, C., Nozawa, T., Minowa-Nozawa, A., Toh, H., and Nakagawa, I. 2017. Bcl-xL Affects Group A *Streptococcus*-induced autophagy directly, by inhibiting fusion between autophagosomes and lysosomes, and indirectly, by inhibiting bacterial internalization via interaction with Beclin 1-UVRAG. *PLoS ONE* **12**, e0170138.

Nakajima, K., Nozawa, T., Minowa-Nozawa, A., Toh, H., Yamada, S., Aikawa, C., and Nakagawa, I. 2019. RAB30 regulates PI4KB (phosphatidylinositol 4-kinase beta)-dependent autophagy against Group A *Streptococcus*. *Autophagy* **15**, 466–477.

Neumann, Y., Bruns, S.A., Rohde, M., Prajsnar, T.K., Foster, S.J., and Schmitz, I. 2016. Intracellular *Staphylococcus aureus* eludes selective autophagy by activating a host cell kinase. *Autophagy* **12**, 2069–2084.

Nozawa, T., Aikawa, C., Goda, A., Maruyama, F., Hamada, S., and Nakagawa, I. 2012. The small GTPases Rab9A and Rab23 function at distinct steps in autophagy during Group A *Streptococcus* infection. *Cell. Microbiol.* **14**, 1149–1165.

Ogawa, M., Yoshikawa, Y., Mimuro, H., Hain, T., Chakraborty, T., and Sasakawa, C. 2011. Autophagy targeting of *Listeria monocytogenes* and the bacterial countermeasure. *Autophagy* **7**, 310–314.

Ouimet, M., Koster, S., Sakowski, E., Ramkhelawon, B., van Solingen, C., Oldebeken, S., Karunakaran, D., Portal-Celhay, C., Sheedy, F.J., Ray, T.D., *et al.* 2016. *Mycobacterium tuberculosis* induces the miR-33 locus to reprogram autophagy and host lipid metabolism. *Nat. Immunol.* **17**, 677–686.

Paik, S., Kim, J.K., Chung, C., and Jo, E.K. 2019. Autophagy: a new strategy for host-directed therapy of tuberculosis. *Virulence* **10**, 448–459.

Palucci, I. and Delogu, G. 2018. Host directed therapies for tuberculosis: futures strategies for an ancient disease. *Chemotherapy* **63**, 172–180.

Peng, H., Yang, J., Li, G., You, Q., Han, W., Li, T., Gao, D., Xie, X., Lee, B.H., Du, J., *et al.* 2017. Ubiquitylation of p62/sequestosome1 activates its autophagy receptor function and controls selective autophagy upon ubiquitin stress. *Cell Res.* **27**, 657–674.

Philpott, D.J., Sorbara, M.T., Robertson, S.J., Croitoru, K., and Girardin, S.E. 2014. NOD proteins: regulators of inflammation in health and disease. *Nat. Rev. Immunol.* **14**, 9–23.

Qu, Y., Ding, S., Ma, Z., Jiang, D., Xu, X., Zhang, Y., Zhang, A., and Xu, G. 2019. MiR-129-3p favors intracellular BCG survival in RAW264.7 cells by inhibiting autophagy via Atg4b. *Cell. Immunol.* **337**, 22–32.

Queiroz, A. and Riley, L.W. 2017. Bacterial immunostat: *Mycobacterium tuberculosis* lipids and their role in the host immune response. *Rev. Soc. Bras. Med. Trop.* **50**, 9–18.

Quigley, J., Hughitt, V.K., Velikovsky, C.A., Mariuzza, R.A., El-Sayed, N.M., and Briken, V. 2017. The cell wall lipid PDIM contributes to phagosomal escape and host cell exit of *Mycobacterium tuberculosis*. *mBio* **8**, e00148-17.

Raju, D., Hussey, S., Ang, M., Terebiznik, M.R., Sibony, M., Galindo-Mata, E., Gupta, V., Blanke, S.R., Delgado, A., Romero-Gallo, J., *et al.* 2012. Vacuolating cytotoxin and variants in *Atg16L1* that disrupt autophagy promote *Helicobacter pylori* infection in humans. *Gastroenterology* **142**, 1160–1171.

Ravenhill, B.J., Boyle, K.B., von Muhlinen, N., Ellison, C.J., Masson, G.R., Otten, E.G., Foeglein, A., Williams, R., and Randow, F. 2019. The cargo receptor NDP52 initiates selective autophagy by recruiting the ULK complex to cytosol-invading bacteria. *Mol. Cell* **74**, 320–329.

Rekha, R.S., Rao Muvva, S.S.V., Wan, M., Raqib, R., Bergman, P., Brighenti, S., Gudmundsson, G.H., and Agerberth, B. 2015. Phenylbutyrate induces LL-37-dependent autophagy and intracellular killing of *Mycobacterium tuberculosis* in human macrophages. *Autophagy* **11**, 1688–1699.

Ricci, V. 2016. Relationship between VacA toxin and host cell autophagy in *Helicobacter pylori* infection of the human stomach: a few answers, many questions. *Toxins* **8**, 203.

Rikihis, Y. 1984. Glycogen autophagosomes in polymorphonuclear leukocytes induced by rickettsiae. *Anat. Rec.* **208**, 319–327.

Rolando, M., Escoll, P., Nora, T., Botti, J., Boitez, V., Bedia, C., Daniels, C., Abraham, G., Stogios, P.J., Skarina, T., *et al.* 2016. *Legionella pneumophila* S1P-lyase targets host sphingolipid metabolism and restrains autophagy. *Proc. Natl. Acad. Sci. USA* **113**, 1901–1906.

Sahu, S.K., Kumar, M., Chakraborty, S., Banerjee, S.K., Kumar, R., Gupta, P., Jana, K., Gupta, U.D., Ghosh, Z., Kundu, M., *et al.* 2017. MicroRNA 26a (miR-26a)/KLF4 and CREB-C/EBPβ regulate innate immune signaling, the polarization of macrophages and the trafficking of *Mycobacterium tuberculosis* to lysosomes during infection. *PLoS Pathog.* **13**, e1006410.

Sharma, V., Verma, S., Seranova, E., Sarkar, S., and Kumar, D. 2018. Selective autophagy and xenophagy in infection and disease. *Front. Cell. Dev. Biol.* **6**, 147.

Sheedlo, M.J., Qiu, J., Tan, Y., Paul, L.N., Luo, Z.Q., and Das, C. 2015. Structural basis of substrate recognition by a bacterial deubiquitinase important for dynamics of phagosome ubiquitination. *Proc. Natl. Acad. Sci. USA* **112**, 15090–15095.

Shui, W., Petzold, C.J., Redding, A., Liu, J., Pitcher, A., Sheu, L., Hsieh, T.Y., Keasling, J.D., and Bertozzi, C.R. 2011. Organelle membrane proteomics reveals differential influence of mycobacterial lipoglycans on macrophage phagosome maturation and autophagosome accumulation. *J. Proteome Res.* **10**, 339–348.

Silwal, P., Kim, Y.S., Basu, J., and Jo, E.K. 2020. The roles of microRNAs in regulation of autophagy during bacterial infection. *Semin. Cell Dev. Biol.* **101**, 51–58.

Simon, H.U., Friis, R., Tait, S.W.G., and Ryan, K.M. 2017. Retrograde signaling from autophagy modulates stress responses. *Sci. Signal* **10**, eaag2791.

Singh, P. and Subbian, S. 2018. Harnessing the mTOR pathway for tuberculosis treatment. *Front. Microbiol.* **9**, 70.

Siqueira, M.D.S., Ribeiro, R.M., and Travassos, L.H. 2018. Autophagy and its interaction with intracellular bacterial pathogens. *Front. Immunol.* **9**, 935.

Sorbara, M.T., Foerster, E.G., Tsalikis, J., Abdel-Nour, M., Mangiapane, J., Sirluck-Schroeder, I., Tattoli, I., van Dalen, R., Isenman, D.E., Rohde, J.R., *et al.* 2018. Complement C3 drives autophagy-dependent restriction of cyto-invasive bacteria. *Cell Host Microbe* **23**, 644–652.

Terebiznik, M.R., Raju, D., Vazquez, C.L., Torbricki, K., Kulkarni, R., Blanke, S.R., Yoshimori, T., Colombo, M.I., and Jones, N.L. 2009. Effect of *Helicobacter pylori*'s vacuolating cytotoxin on the autophagy pathway in gastric epithelial cells. *Autophagy* **5**, 370–379.

Thurston, T.L., Wandel, M.P., von Muhlinen, N., Foeglein, A., and Randow, F. 2012. Galectin 8 targets damaged vesicles for autophagy to defend cells against bacterial invasion. *Nature* **482**, 414–418.

Travassos, L.H., Carneiro, L.A., Ramjeet, M., Hussey, S., Kim, Y.G., Magalhães, J.G., Yuan, L., Soares, F., Chea, E., Le Bourhis, L., *et al.* 2010. Nod1 and Nod2 direct autophagy by recruiting ATG16L1 to the plasma membrane at the site of bacterial entry. *Nat. Immunol.* **11**, 55–62.

van Wijk, S.J.L., Fricke, F., Herhaus, L., Gupta, J., Hotte, K., Pampaloni, F., Grumati, P., Kaulich, M., Sou, Y.S., Komatsu, M., *et al.* 2017. Linear ubiquitination of cytosolic *Salmonella* Typhimurium activates NF-κB and restricts bacterial proliferation. *Nat. Microbiol.* **2**, 17066.

Vergne, I., Chua, J., Singh, S.B., and Deretic, V. 2004. Cell biology of *Mycobacterium tuberculosis* phagosome. *Annu. Rev. Cell Dev. Biol.* **20**, 367–394.

Vergne, I., Gilleron, M., and Nigou, J. 2014. Manipulation of the endocytic pathway and phagocyte functions by *Mycobacterium tuberculosis* lipoarabinomannan. *Front. Cell. Infect. Microbiol.* **4**, 187.

Verlhac, P., Grégoire, I.P., Azocar, O., Petkova, D.S., Baguet, J., Viret, C., and Faure, M. 2015. Autophagy receptor NDP52 regulates pathogen-containing autophagosome maturation. *Cell Host Microbe* **17**, 515–525.

Walker, M.J., Barnett, T.C., McArthur, J.D., Cole, J.N., Gillen, C.M., Henningham, A., Sriprakash, K.S., Sanderson-Smith, M.L., and Nizet, V. 2014. Disease manifestations and pathogenic mechanisms of Group A *Streptococcus*. *Clin. Microbiol. Rev.* **27**, 264–301.

Wang, J., Yang, K., Zhou, L., Minhaowu, Wu, Y., Zhu, M., Lai, X., Chen, T., Feng, L., Li, M., Huang, C., *et al.* 2013. MicroRNA-155 promotes autophagy to eliminate intracellular mycobacteria by targeting Rheb. *PLoS Pathog.* **9**, e1003697.

Watson, R.O., Manzanillo, P.S., and Cox, J.S. 2012. Extracellular *M. tuberculosis* DNA targets bacteria for autophagy by activating the host DNA-sensing pathway. *Cell* **150**, 803–815.

Wild, P., Farhan, H., McEwan, D.G., Wagner, S., Rogov, V.V., Brady, N.R., Richter, B., Korac, J., Waidmann, O., Choudhary, C., *et al.* 2011. Phosphorylation of the autophagy receptor optineurin restricts *Salmonella* growth. *Science* **333**, 228–233.

Wilkinson, D.S., Jariwala, J.S., Anderson, E., Mitra, K., Meisenhelder, J., Chang, J.T., Ideker, T., Hunter, T., Nizet, V., Dillin, A., *et al.* 2015. Phosphorylation of LC3 by the Hippo kinases STK3/STK4 is essential for autophagy. *Mol. Cell* **57**, 55–68.

Wu, Y.W. and Li, F. 2019. Bacterial interaction with host autophagy. *Virulence* **10**, 352–362.

Xu, Y., Zhou, P., Cheng, S., Lu, Q., Nowak, K., Hopp, A.K., Li, L., Shi, X., Zhou, Z., Gao, W., *et al.* 2019. A bacterial effector reveals the V-ATPase-ATG16L1 axis that initiates xenophagy. *Cell* **178**, 552–566.

Yahiro, K., Akazawa, Y., Nakano, M., Suzuki, H., Hisatune, J., Isomoto, H., Sap, J., Noda, M., Moss, J., and Hirayama, T. 2015. *Helicobacter pylori* VacA induces apoptosis by accumulation of connexin 43 in autophagic vesicles via a Rac1/ERK-dependent pathway. *Cell Death Discov.* **1**, 15035.

Yahiro, K., Satoh, M., Nakano, M., Hisatsune, J., Isomoto, H., Sap, J., Suzuki, H., Nomura, F., Noda, M., Moss, J., *et al.* 2012. Low-density lipoprotein receptor-related protein-1 (LRP1) mediates autophagy and apoptosis caused by *Helicobacter pylori* VacA. *J. Biol. Chem.* **287**, 31104–31115.

Yoshikawa, Y., Ogawa, M., Hain, T., Yoshida, M., Fukumatsu, M., Kim, M., Mimuro, H., Nakagawa, I., Yanagawa, T., Ishii, T., *et al.* 2009. *Listeria monocytogenes* ActA-mediated escape from autophagic recognition. *Nat. Cell Biol.* **11**, 1233–1240.

Yuan, Q., Chen, H., Yang, Y., Fu, Y., and Yi, Z. 2020. miR-18a promotes *Mycobacterial* survival in macrophages via inhibiting autophagy by down-regulation of ATM. *J. Cell. Mol. Med.* **24**, 2004–2012.

Yuk, J.M., Shin, D.M., Lee, H.M., Yang, C.S., Jin, H.S., Kim, K.K., Lee, Z.W., Lee, S.H., Kim, J.M., and Jo, E.K. 2009. Vitamin D3 induces autophagy in human monocytes/macrophages via cathelicidin. *Cell Host Microbe* **6**, 231–243.

Zhang, R., Varela, M., Vallentgoed, W., Forn-Cuni, G., van der Vaart, M., and Meijer, A.H. 2019. The selective autophagy receptors Optineurin and p62 are both required for zebrafish host resistance to mycobacterial infection. *PLoS Pathog.* **15**, e1007329.

Zheng, Y.T., Shahnazari, S., Brech, A., Lamark, T., Johansen, T., and Brumell, J.H. 2009. The adaptor protein p62/SQSTM1 targets invading bacteria to the autophagy pathway. *J. Immunol.* **183**, 5909–5916.

Zulauf, K.E., Sullivan, J.T., and Braunstein, M. 2018. The SecA2 pathway of *Mycobacterium tuberculosis* exports effectors that work in concert to arrest phagosome and autophagosome maturation. *PLoS Pathog.* **14**, e1007011.