Autophagy and inflammatory diseases

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Autophagy is a cellular mechanism for the sequestration and degradation of intracellular pathogens and compromised organelles, particularly damaged mitochondria. Autophagy also clears other cellular components, such as inflammasomes and cytokines, thus providing an important means of regulating inflammation. Defects in autophagy have been found in genetic association studies to confer susceptibility to several autoimmune and inflammatory disorders, particularly inflammatory bowel disease. Thus, the manipulation of autophagy in disease situations is of growing interest for therapeutic targeting; however, the involvement of autophagy in cellular homeostasis, in normal immune function and in inflammation is manifold. An appreciation of the intricacies of the contributions of this process to inflammation, and how these are altered by various immune and environmental stimuli, is essential for the understanding and interpretation of studies of inflammation and the design of therapeutics exploiting the manipulation of autophagy. This review focuses on the known roles of autophagy in the induction and maintenance of inflammation and on its role in the aetiology and regulation of inflammatory and autoimmune disorders.

Immunology and Cell Biology (2013) 91, 250–258; doi:10.1038/icb.2012.92; published online 15 January 2013

Keywords: Atg; autoimmune; autophagy; cytokine; inflammation

Many damaged cellular constituents are cleared through the process of macroautophagy, in which a nascent double-membrane autophagosome forms around protein aggregates and organelles in the first step of a process that ultimately results in lysosomal degradation and recycling of components for use by the cell. In a quiescent cell, macroautophagy occurs at a basal level to remove defective organelles, such as dysfunctional mitochondria and peroxisomes, as well as misfolded proteins in response to endoplasmic reticulum (ER) stress (reviewed elsewhere). Thus, autophagy is required for normal cell functioning and survival. Macroautophagy (hereafter referred to as autophagy) is characterised by the formation of an isolation membrane, or phagophore, which elongates around its target and fuses with itself to form a double-membrane autophagosome. This can then fuse with lysosomes to form an autolysosome, leading to the degradation of its luminal contents. This process is controlled by the products of numerous autophagy-specific genes (Atgs) and by the mammalian target of rapamycin (mTOR), a serine/threonine protein kinase that regulates cell growth, proliferation, motility and survival, gene transcription and protein synthesis. Inhibition of mTOR is essential for autophagy to initiate, and allows the translocation of a complex containing Atg17/Atg18-1-like kinase (ULK1/2), Atg3, FIP200 and Atg101 from the cytosol to the ER, a process dependent on the interaction between ULK1 and AMP-activated protein kinase (AMPK). This leads to the recruitment of the type III phosphatidylinositol-3-kinase, VPS34, in a complex with other proteins, including beclin 1, to the developing autophagosome.

Generation of phosphatidylinositol-3-phosphate by this complex is crucial for the recruitment of proteins required for initiation of autophagosome formation (Figure 1). Inhibitors of phosphatidylinositol-3-phosphate-3-kinase, including 3-methyladenine (3-MA), are commonly used to inhibit autophagy in in vitro studies, although such studies must be interpreted with caution due to other effects of the compounds used (Box 1). Autophagy regulates energy and nutrient homeostasis and has an essential role in tissue development. Autophagic activity is amplified in times of deprivation of oxygen, growth factors or nutrients, and this is essential for cell survival. Increased autophagy in hypoxic or starved cells facilitates a shift from aerobic respiration to glycolysis and provides a means by which cellular components can be hydrolysed to provide fuel for metabolism. This glycolytic shift also occurs in proliferating myeloid cells and lymphocytes and increased levels of autophagy are characteristic of activated immune cells. Although a moderate level of autophagy is required to maintain a healthy cytosolic environment, excessive autophagy can lead to autophagic cell death.

Autophagy also shapes immune responses by directly participating in immune cell function. For example, autophagy-degraded cellular components can be loaded onto MHC (major histocompatibility complex) class I and II molecules for presentation to T cells.
Figure 1 Pathways involved in autophagy regulation. Autophagy is regulated by numerous stimuli, including nutrient starvation, growth factors, cytokines, reactive oxygen species (ROS), pharmacological inhibitors and drug-induced autophagy-associated molecular patterns (DAMPs and RAMPs). Autophagosome formation is largely controlled by mTOR, inhibition of mTOR leads to the interaction between the serine/threonine protein kinase ULK1 and AMPK, which, in turn, recruits the type III PI3 kinase VPS34, in complex with other proteins, including ATG1, to the developing autophagosome. Nutrient deprivation activates AMPK and may also inhibit mTOR activation, leading to autophagosome formation. Activation of the Akt pathway by growth factors and cytokines, including IL-4, IL-13 and IL-10, leads to activation of mTOR and inhibition of autophagosome formation. Other cytokines induce autophagy. IFN-γ promotes autophagosome formation through an IRGM (IRGM) signal in a mTOR-dependent mechanism. This pathway is not well understood but does involve metabolic adaptation. This process is inhibited by IL-4 and IL-13 through a SMAD signaling transducer and activator of transcription factor 3-dependent mechanism. TNF-α, IL-1, ROS, engagement of Toll-like receptors (TLR) also induce autophagy, although these pathways are not well characterized. Autophagosomes can sequester and deliver cytosolic constituents to lysosomes for degradation and recycling. A full color version of this figure is available at the Immunology and Cell Biology journal online.

Intracellular pathogens can be killed by autophagy, including Mycobacterium tuberculosis, Candida albicans, adherent-invasive Escherichia coli and group A Streptococcus.1,2 Similarly, autophagy is involved in host-protective immune responses against infection with viruses, such as Sindbis virus, Epstein Barr virus3 and vesicular stomatitis virus.4-6 The rate of autophagy can be modulated in lymphocytes by antigen receptor stimulation and in macrophages following activation of Toll-like receptors (TLR) and pattern-recognition receptors with pathogen- and danger-associated molecular patterns.6,10-30 Furthermore, T helper type 1 (Th1) and pro-inflammatory cytokines, including interferon (IFN)-γ, tumour necrosis factor (TNF)-α, interleukin (IL)-1 and IL-23, induce autophagy,31-32 while Th2 and regulatory cytokines, including IL-4, IL-13 and IL-10, are inhibitory.33-36 Importantly, autophagy is now recognized to be a major mechanism for regulating the secretion of cytokines and chemokines, particularly in macrophages, facilitating macrophage-mediated control of cell recruitment and orchestration of immune responses.37-40 The influence of the metabolic state of immune cells on inflammatory responses is an area of growing interest, and the roles of autophagy in this process are of considerable potential importance.

STARvation-INDUCED AUTOPHAGY AND INFLAMMATION

In conditions of low cellular energy and essential amino-acid deprivation, the induction of autophagy is driven by AMPK, which is antagonised by mTOR when nutrients are sufficient.41-42 Activation of AMPK occurs through an increase in the ratio of AMP to ATP indicative of a state of oxygen deprivation, as well as phosphorylation by CaMKKβ when cytosolic Ca2+ accumulates, which occurs during amino-acid deprivation.43,44 AMPK activation stimulates pathways that correct imbalances in glucose and lipid concentrations and return energy levels to normal. When energy levels are low, AMPK halts cell growth and migration and supports cell survival by driving autophagic degradation of damaged mitochondria.45 Small molecule activators of AMPK can induce autophagic clearance of β-amyloid plaques in models of Alzheimer’s disease,46 and there is emerging evidence that AMPK is a key modulator of immune responses. It can reduce the severity of inflammation and tissue damage in colitis,25,47,48 and experimental autoimmune encephalomyelitis49 and airway inflammation in asthma.50 In addition, AMPK can drive the induction of regulatory T cells,51 the differentiation and inhibitory activity of myeloid-derived suppressor cells52 and, in macrophages, AMPK activation is a critical point at which anti-inflammatory signals converge to elicit suppressive responses. For example, IL-10 and transforming growth factor β activate AMPK in macrophages and inhibit AMPK in macrophages results in excessive production of IL-6, TNF-α and cyclooxygenase-2 in response to lipopolysaccharide.
(LPS). Conversely, AMPK suppresses LPS-induced IL-6 and TNF-α and inhibits the respiratory burst in neutrophils. Thus, activation of AMPK is predominantly anti-inflammatory and autophagy may represent one mechanism through which AMPK exerts these effects.

**AUTOPHAGY REGULATES CYTOKINE SECRETION**

As well as regulating responses to pathogens within cells, autophagy can influence immune responses in microenvironments through its role as a regulator of cytokine secretion, particularly within antigen-presenting cells. In particular, autophagy can modulate the secretion of members of the IL-1 cytokine family, IL-23 and, as a consequence, IL-17.

**Autophagy and IL-1 family cytokines**

The IL-1 cytokine family, including IL-1α, IL-1β, IL-18, IL-33, IL-36, IL-37 and IL-58, orchestrate a wide range of immune and physiological effects. In particular, IL-1α and IL-1β, which signal through the IL-1 type 1 receptor (IL-1R1), are pro-inflammatory, acting partly through the induction of cyclooxygenase-2, type 1 phosphatase A and inducible nitric oxide synthase. IL-1α and IL-1β also recruit myeloid cells, including neutrophils, to sites of inflammation. Like IL-1α and IL-1β, IL-18 promotes inflammation, stimulating TNF-γ production by natural killer cells and Th1 cells and IL-17 production by γδ T cells. IL-1α and IL-1β are produced as inactive pro-forms that are cleaved by caspase-1 to form the mature, bioactive cytokines. Caspase-1 is itself activated by an inflammasome, a large multiprotein complex that includes an intracellular sensor, such as the NOD-like receptor (NLR) NLPR3 or the DNA sensor absent in melanoma 2 (AIM2). Recently, findings have suggested that IL-1β can drive the secretion of both IL-1α and IL-23, further highlighting the importance of this cytokine in regulating inflammatory responses.

The activity of IL-1α and IL-1β is regulated by a naturally occurring IL-1 receptor antagonist (IL-1Ra) and by the decoy receptor (IL-1R1), whereas IL-18 is regulated by IL-18-binding protein. It has also been demonstrated that autophagy can regulate IL-1β, IL-1α and IL-18 at the levels of transcription, processing and secretion. This occurs through at least two distinct mechanisms (Figure 2). Firstly, autophagy suppresses TLR-induced secretion of IL-1β. IL-1α and IL-1β in macrophages and dendritic cells (DCs). Production of biologically active IL-1β typically requires two signals. The initial signal is provided by pathogen-associated molecular patterns, such as LPS, or danger-associated molecular patterns, such as HMGBl, and results in transcription of pro-IL-1. This is followed by activation of inflammasome assembly by a second stimulus, such as reactive oxygen species, mitochondrial DNA, ATP, particulates (for example, silica, alum), protein aggregates and liposomal normal rupture. Autophagy suppresses inflammasome assembly by degrading numerous endogenous stimuli, including mitochondrial DNA and reactive oxygen species, that would otherwise induce inflammasome activation and processing of pro-IL-1 into the mature cytokine. Thus, inhibition of autophagy under these conditions leads to an increase in inflammasome activation and subsequent processing of IL-1β and IL-18.

The second mechanism by which autophagy negatively influences IL-1 and IL-18 secretion is more direct. Autophagosomes can sequester and degrade inflammasome components and pro-IL-1β. In mouse DCs, induction of autophagy can prevent IL-1β secretion in response to TLR stimulation or ATP, while LPS-stimulated mouse macrophages, in the absence of an inflammasome-inducing signal, autophagosomes sequester and degrade pro-IL-1β. More recently, Shi et al. have demonstrated that activation of the NLPR3 and AIM2 inflammasomes reduces autophagy in human macrophages. In addition, inflammasome components have been observed to localise with autophagosome components, indicating that, similar to pro-IL-1β, inflammasomes are degraded within autophagosomes. These data suggest that autophagy is induced by inflammatory stimuli and acts a self-regulatory mechanism for the control of inflammatory
cytokine secretion, thereby downregulating potentially deleterious inflammatory responses.

**Autophagic regulation of the IL-23–IL-17 pathway**

Through regulating IL-17 secretion, autophagy also moderates the production of another inflammatory cytokine, IL-23. IL-23 is an IL-12 family cytokine that, synergistically with IL-21, IL-18 or IL-19, induces the differentiation and expansion of Th17 cells from naive CD4 T cells, as well as the secretion of IL-17 by γδ T cells and other innate lymphoid cells. Both IL-23 and IL-17 are closely linked with a number of autoimmune diseases, including psoriasis and multiple sclerosis, as well as asthma and ankylosing spondylitis (reviewed elsewhere). In both mouse and human macrophages and DCs, inhibition of autophagy allows excessive IL-23 secretion, whereas induction of autophagy has the opposite effect. IL-18 can drive IL-23 secretion, and this appears to be the mechanism through which autophagy exerts its effects on IL-23. Supporting this, IL-23 production in autophagy-impaired human macrophages is dependent on NF-kB signaling and is inhibited by IL-1-neutralizing antibodies. As IL-1, IL-18 and IL-23 have a major role in promoting IL-17 production by T cells, regulation of these cytokines by autophagy can affect IL-17 secretion. Indeed, supernatants from mouse DCs primed with LPS and cultured in the presence of the autophagy inhibitor 3-MA contained high levels of IL-1β and IL-23 and potently induced IL-17, IL-22 and IFN-γ secretion by γδ T cells in vivo. This may also operate in vivo, as mice lacking the autophagy protein Atg6 in myeloid cells secrete higher levels of IL-1α, IL-12p70, CXCL1 (C-X-C motif chemokine ligand 1) and IL-17 in response to infection with M. tuberculosis. These data indicate that autophagy in innate immune cells has the potential to influence T-cell polarization, suggesting an important role in the control of both inflammation and innate regulation of adaptive immune responses.

**AUTOPHAGY IN INFLAMMATORY DISEASES**

The regulation of IL-1β and IL-23 secretion by autophagy may be of critical importance in the prevention of the autoimmune diseases in which hyperactivation of this pathway is a major driver of pathology. Polymorphisms in the IL-23R locus confer susceptibility to inflammatory bowel disease (IBD), including Crohn’s disease (CD) and ulcerative colitis (UC), rheumatoid arthritis, and ankylosing spondylitis. In addition, systemic lupus erythematosus (SLE) patients produce excess IL-17 and IL-23 that may exacerbate their disease. To address this, mice challenged with streptococcal carbohydrate A (SCA) present with a strong susceptibility to lupus, the T300A polymorphism in the Atg5 gene, which produces a hypomorphic allele that severely impairs autophagic activity. Macrophages from mice with the Atg5C16T T300A mutation display uncontrolled production of IL-1β and are more susceptible to destroy sodium phosphate-induced colitis. As well as impaired macrophage functions, patients with this mutation and mice engineered to bear the same allele have striking defects in autophagy in Paneth cells of the intestinal epithelium. Paneth cells are located in the crypts of Lieberkühn in the small intestine and are specialized to produce lysozyme and antimicrobial peptides. Indeed, the accumulation of E. coli and mitochondria that would be expected in Paneth cells bearing the autophagy-compromising T300A mutation, these cells fail to secrete lysozyme and thus lysozyme is absent in the fecal microbiome of these patients, defects that are also seen when autophagy is impaired by deletion of Atg6 in mice. Thus, a deficiency in lysozyme-mediated control of intestinal microbiota may contribute to the development of pathology in patients bearing variants of Atg6 or, although this has not been clearly delineated.

Indeed, the effective limitation of symptoms and pathogens in the gut environment is critical for the prevention of IBD and particularly CD, which is illustrated by the finding that CD patients have abnormal gut microbiota profiles. Altered autophagic activity downstream of microbial sensors in the gut appears to be a key reason for the failure of IBD patients to control intestinal microbiota and prevent gut pathology. In particular, defects in autophagy-related genes permit the establishment of enteric-invasive E. coli, common in lesions in the intestinal epithelia of patients with CD. In addition, genetic linkage data have firmly established polymorphisms in the bacterial sensor and inducer of autophagy, NOD2, in susceptibility to CD. Several NOD2 variants have been identified, including a frameshift mutation, that confer susceptibility to CD. Following bacterial infection of host cells, NOD2 recruits Atg14L to the cell membrane, initiating autophagosome induction and bacterial clearance, a process that is impaired when NOD2 contains a CD-associated mutation. Similarly, the NOD2 ligand muramyl dipeptide induces autophagy and killing of
pathogenic Salmonella, both dependent on functional NOX2 and ATG5 in 11.

Although autophagic degradation of invasive bacteria is crucial for controlling bacterial infection, autophagy appears to have additional anti-inflammatory effects in the gut microbiome. Autophagy stimulated in response to NOX2 activation also controls IL-1β and IL-6 release, and peripheral blood mononuclear cells from CD patients bearing the Atg6L1 susceptibility allele secrete more of these pro-inflammatory cytokines.112,113 As well as controlling NOX2-dependent intestinal cytokine release, autophagy also modulates intestinal inflammation by promoting non-inflammatory DC-T-cell

interactions. In the intestine, DC sample antigens by extending protuberances through the epithelial cell layer, a process that itself depends on autophagy.115 These antigens are then presented on MHC class II complexes and if they are derived from commensal bacteria, elicit non-activating, self-recognition T-cell responses. NOX2-stimulated autophagy in DC results in tolerogenic presentation of commensal bacterial components on MHC class II complexes.112 Inhibiting autophagy prevents sampling and results in enhanced IL-1β and CD69 expression and downregulation of IL-10 production by DC. These changes produce pro-inflammatory DCs that stimulate T-cell proliferation.112 When T cells and DC interact, an immunological synapse is formed and must be stably maintained to result in T-cell activation.111 In a recent study, T-cell-DC interactions result in autophagosome formation in DC, which was oriented towards the synapse and destabilized the synapse. When autophagy was blocked, the immunological synapse persisted and resulted in excess activation of T cells and induction of a Th17 phenotype.111 DC from CD patients bearing the Atg6L1 T300A mutation had similarly persistent synapses.114 Thus, there are multiple points at which autophagy can influence immune responses that lead to the development and pathologies of CD and IBD. Autophagy can regulate the microbial profile in the gut and limit invasion of pathogenic bacteria. In addition, autophagy appears to promote tolerance to commensal bacteria by influencing the outcome of T-cell interactions with antigen-presenting cells and by directing the cytokine profile in the gut environment away from an excessive pro-inflammatory phenotype.88-91 Internalized DCs may participate in explaining the initial results demonstrating that miT-OR-2 inhibiting drugs are protective against intestinal inflammation, both in mouse models and in humans.83

SLE

SLE is an autoimmune-mediated autoimmune disease that can affect multiple organs and tissues, including the skin, joints, kidneys and brain. As the pathological mechanisms driving the initiation and progression of SLE are diverse and complex, the points at which autophagy influences these mechanisms are poorly defined. Nonetheless, genetic association studies have established autophagy as an important process in SLE as several mutations have been identified in autophagy-related genes that confer susceptibility to this disease, including IRGM.115,116 IRGM is required for the autophagic degradation of mycobacteria,117 and a Taiwanese study suggests that tuberculoid and SLE development are correlated.118 It has been suggested that this may not only be due to perturbations in the IL-23/IL-17 axis in patients,119 but also could be due to direct defects in autophagy pathways. In addition to IRGM, the locus containing Atg5 and the PRDM1 gene is a susceptibility locus for SLE.120,121 The PRDM1 gene encodes the plasma cell differentiation factor Blimp-1 and although variations in this locus may affect plasma cell differentiation and behaviour to influence SLE development, a more specific genetic association study has clarified a protective role for Blimp-1 against SLE and also confirmed the autophagy gene Atg15 as a SLE susceptibility locus.122

SLE is driven by the formation of immune complexes of autoreactive antibodies bound to autot antigens; many of these autoantigens are thought to be exposed to B cells when apoptotic debris fails to be cleared effectively, a process that requires autophagy in macrophages.121 Immune complexes accumulate in tissues, such as the fine capillaries of the glomerulonephritis, where they precipitate complement deposition and damaging inflammatory responses. In addition, immune complexes stimulate TLR7 and TLR9 on B cells and DCs, and, particularly in immature plasmacytid DCs, this stimulates the production of IFN-α which, in turn, activates and induces maturation of the B cells and other cells that participate in the disease process.122

Autophagy is also particularly important in T-cell development, function and homeostasis, and defects in autophagy genes may alter the activity of T cells in the context of SLE. In particular, deficiencies in the autophagy pathway cause defects in ER and induce T cells more prone to cell death.123,124 Interestingly, naïve CD4 T cells from patients with SLE have lower constitutive levels of autophagy than those from healthy donors, and these cells are also resistant to induction of autophagy by serum starvation.125 The functional relevance of these findings is not clear, although this resistance to autophagy may increase the susceptibility of lymphocytes to apoptosis, which could contribute to the accumulation of apoptotic debris that provides a source of autoantigens and drives autoimmune pathologies.125,126 Similarly, if autophagy is impaired in macrophages or DCs, this could affect the regulation of pro-inflammatory cytokine secretion and further promote pathology.

As well as its putative protective effects, autophagy may have roles in facilitating the initiation of SLE by stimulating processes that promote the activation of self-reactive B cells to produce autoantibodies. For example, autophagy is required for human neutrophil extracellular DNA trap (NET) release in response to FMA stimulation127 and in general.128 NET release allows the exposure of multiple typical B-cell nuclear autoantigens containing TLR ligands and may exacerbate disease by precipitating complement deposition and tissue damage.129 NETs activate innate immune cells and, in the presence of type I IFNs, autoreactive T cells further stimulate NET release, potentially driving ongoing disease. As well as promoting NET formation and thus autoantibody display, autophagy may aid in the activation of auto-reactive B cells once they encounter antigen.

B-cell receptor (BCR) stimulation by cognate antigens triggers autophagosome formation and antigen processing, which promotes B-cell acquisition of T-cell help.130 In autoreactive B cells, DNA-containing autoreactive BCRs internalization and recruitment of TLR9-containing endosomes to autophagosomes, a process that results in the B-cell hyper-responsiveness that is characteristic of autoreactive B cells.131 As well as enabling the induction of autoreactive body production, autophagy may promote cytokine release in response to immune complexes. TLR7 ligation induces autophagy,132 which is required for IFN-α production by plasmacytid DC in response to an ssRNA virus.133 However, the direct relevance of autophagy in modulating IFN-α secretion by plasmacytoid DC and other cells has not been assessed in the context of autoimmune.

Although the roles of autophagy in SLE disease processes are still unclear, it appears that autophagy has an overall protective effect in the disease. Therapeutic interventions that stimulate autophagy, particularly miTOR inhibitors, are of growing interest for the treatment of SLE and appear to be well tolerated by patients.
(NZB/NZW), lupus-prone mice, rapamycin prevented development of nephritis, inhibiting lymphoproliferation and MCP-1 expression in kidneys.129 Reducing autoreactivity and enhancing survival.130 Moreover, rapamycin treatment of older (NZB/NZW) female mice with established nephritis improved survival; splenomegaly was reduced and anti-nuclear antibodies were diminished, while renal function was significantly preserved compared with control mice.29 Low-dose rapamycin prevented determination of renal function in Immunoglobulin A nephropathy patients.16 and is currently being tested in a phase II trial in SLE (NCIT077919). Thus, rapamycin represents a significant therapeutic target for the treatment of SLE, but further studies on the precise mechanisms involved are essential to maximise the potential of such treatments.

Arthritis

Autoantibodies appear to promote the survival of cells that actively drive RA, whereas in osteoarthritic (OA) joints, the pro-survival effects of autoantibodies can prevent the death of cells that maintain joint structure. In RA, joint destruction is mediated primarily by TNF-α, which stimulates synovial fibroblasts producing the growth factors, chemokines, proteases and adhesion molecules that are characteristic of the arthritic joint environment. Autoantibodies in these fibroblasts are enhanced to counter acute ER stress and maintain cell survival.137 As well as stimulating fibroblasts to produce effector molecules, TNF-α potently activates murine osteoclasts to resorb the bone matrix,138 and autoantibodies are an optimal point at which osteoclast activity and bone degradation are regulated. TNF-α stimulates autoantibodies in osteoclasts, promoting their differentiation and TGF-β1 induction of autoantibody in TNF-α transgenic mice reduced osteoclast differentiation and joint damage.139 In addition to TNF-α, other factors that contribute to arthritis progression include MCP-1, IL-1β and IL-6, all of which induce MCP-1-induced proteolytic enzymes. MCP1 and MCP1 contribute to osteolysis in RA by promoting angiogenesis and osteoclastogenesis,140 and these may act via induction of autoantibodies. Conversely, another study found that the mTOR inhibitor everolimus, which induces autoantibody, inhibited osteoclast differentiation and activity and induced osteoclast apoptosis. However, treatment of RA patients with everolimus resulted in only a transient and modest improvement in clinical signs of disease.140 The data so far would suggest a negative role for autoantibodies in RA, although this is largely based on studies that focused specifically on osteoclasts. The role, if any, of autoantibodies in immune cells in the rheumatoid joint has yet to be elucidated.

In contrast to RA, autoantibodies appear to be protective against joint destruction in OA,141 and rapamycin reduces disease severity.142 Autoantibodies are increased in cartilage and in cartilage-producing chondrocytes, and inhibiting autoantibody results in similar gene expression changes to those seen in OA joints.143 Moreover, induction of autoantibody with rapamycin cleared reactive oxygen species and prevented IL-1β-dependent transcriptional changes that drive OA.144 Moreover, induction of autoantibody in arthritis may be very much disease- and context-specific and requires further study to elucidate the mechanisms at play.

Autoantibody regulation of autoantigen presentation

In autoantibody-mediated autoimmune diseases such as SLE and RA, autoantibody may facilitate antigen presentation and thus enable the switching and maturation of B cells to plasma cells that secrete pathogenic, T-cell-dependent antibody isotypes. Interestingly, blocking autoantibody may specifically prevent the presentation of modified peptides that are common autoantigens. Anti-self antibodies against citrullinated autoantigens are markers of autoimmunity and disease, particularly RA.146 Citrullination occurs in inflamed tissues147 and in antigen-presenting cells, where it has recently been found to occur in autophagosomes. In addition, peptidylarginine deiminase, which deaminates arginine to form citrulline, is found in autophagosomes in B cells, macrophages and DCs.148 DC and macrophages can present citrullinated peptides without extra stimuli, whereas B cells must receive BCR stimulation to present citrullinated peptides. The autoantibody inhibitor 3-MA blocked the presentation of citrullinated, but not unmodified, peptides.149 Thus, excessive autoantibody may potentiate autoantigen exposure and thus autoimmune disease initiation. However, in a more controlled environment, these effects may be balanced by other autoantibody-dependent effects, such as cell survival, increased clearance of apoptotic bodies and regulation of pro-inflammatory cytokine secretion.

Sepsis

Considering the systemic inflammation and cell death that characterise sepsis, it is perhaps not surprising that autoantibody markers increase in septic tissues and their expression is correlated with cell survival, both in animal models and in humans.150-152 Inhibition of autoantibody in septic mice boosts inflammatory cytokine levels and decreases mortality, probably due to the failure to correct damaged or dysfunctional mitochondria, which activate the NLRP3 inflammasome,153 Similarly, in mice in which Age1 is specifically deleted in the intestinal epithelium, LPS induces high levels of IL-1β and IL-18,154 while ICAM-1−/− mice produce more IL-1β and IL-18 in response to LPS- or caecal ligation and puncture-induced sepsis.155 Conversely, induction of autoantibody with rapamycin inhibits the release of IL-1β and IL-18 into the serum of mice infected intraperitoneally with LPS.156 and protects mice against Staphylococcus enteritidis-induced septic shock157 and against cardiac dysfunction following caecal ligation and puncture.158 Thus, sepsis may represent a condition where the control of pro-inflammatory cytokine secretion by autoantibody has a clear protective role to play.

CONCLUSIONS

It is evident that autoantibody has diverse functions and may contribute to altered cell behaviour in disease situations in a variety of ways. A thorough understanding of the effects of altering autophagic activity is therefore necessary for the design of therapeutics that aim to target this process to improve disease outcomes in patients. Altering autophagic systemically will affect all autophagy-dependent events and thus determining the balance of these effects will be most important in assessing whether therapeutic intervention of autoantibody will produce an overall positive, or negative, outcome for patients. Evidence from animal models and early clinical trials suggest that the generalised induction of autoantibody may be beneficial in the treatment of CD, some cases of SLE and in OA. The effects of modulating autoantibody have not yet been adequately tested for potential therapeutic in other diseases, although considering the conceivable protective role of autoantibody in situations of dysregulated inflammation, specifically enhancing autoantibody may be predicted to be an effective means of targeting many inflammatory and autoimmune diseases.

CONFLICT OF INTEREST

The authors declare no conflict of interest.
ACKNOWLEDGEMENTS

Support for this work was provided by Science Foundation Ireland (SFI) PI Grant (09/IN.118/1078) to KEMCH and a SHRI Strategic Research Cluster Grant (07/ SIRCH/0148/11), as part of the Immunology Research Centre.


