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Review Article

Phagocytic clearance of apoptotic, necrotic, necroptotic and pyroptotic cells

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Although millions of cells in the human body will undergo programmed cell death each day, dying cells are rarely detected under homeostatic settings *in vivo*. The swift removal of dying cells is due to the rapid recruitment of phagocytes to the site of cell death which then recognise and engulf the dying cell. Apoptotic cell clearance — the engulfment of apoptotic cells by phagocytes — is a well-defined process governed by a series of molecular factors including 'find-me', 'eat-me', 'don't eat-me' and 'good-bye' signals. However, in recent years with the rapid expansion of the cell death field, the removal of other necrotic-like cell types has drawn much attention. Depending on the type of death, dying cells employ different mechanisms to facilitate engulfment and elicit varying functional impacts on the phagocyte, from wound healing responses to inflammatory cytokine secretion. Nevertheless, despite the mechanism of death, the clearance of dying cells is a fundamental process required to prevent the uncontrolled release of pro-inflammatory mediators and inflammatory disease. This mini-review summarises the current understandings of: (i) apoptotic, necrotic, necroptotic and pyroptotic cell clearance; (ii) the functional consequences of dying cell engulfment and; (iii) the outstanding questions in the field.

Introduction

For over 50 years apoptosis has dominated basic and translational research in the cell death field, representing the most well-characterised type of cell death. However, this traditionally immuno-silent form of programmed cell death represents just one of many pathways in which a cell can program itself to die. In 2018, twelve different regulated forms of cell death were described, highlighting the significant expansion of the cell death field [1]. In particular, the discovery of necroptosis and pyroptosis provides a significant contrast with the anti-inflammatory properties of apoptosis, and the stochastic nature of primary necrosis (Figure 1). Regardless of the mechanism of death, the swift removal of dying cells by professional (i.e. macrophages) and non-professional (i.e. epithelial cells) phagocytes remains paramount to maintain physiological homeostasis. For example, the induction of cell death and removal of dying cells has a fundamental role in embryonic development [2], tissue repair [3,4] and resolution of inflammation [5,6]. However, the persistence of dead cells and rupture of the plasma membrane allows the release of intracellular contents including damage-associated molecular patterns (DAMPs) which can trigger a robust inflammatory response [7–10]. Defective clearance of dying cells is closely associated with the onset and pathogenesis of inflammatory disease such as atherosclerosis [11,12], autoimmune disorders such as systemic lupus erythematosus and rheumatoid arthritis [9,13–16], and asthma [17]. Thus, dying cells employ a variety of mechanisms to recruit, be recognised and be engulfed by phagocytes. Here, this mini-review highlights the mechanisms underpinning dying cell clearance, and discusses the functional impact of phagocytosis on the surrounding environment.

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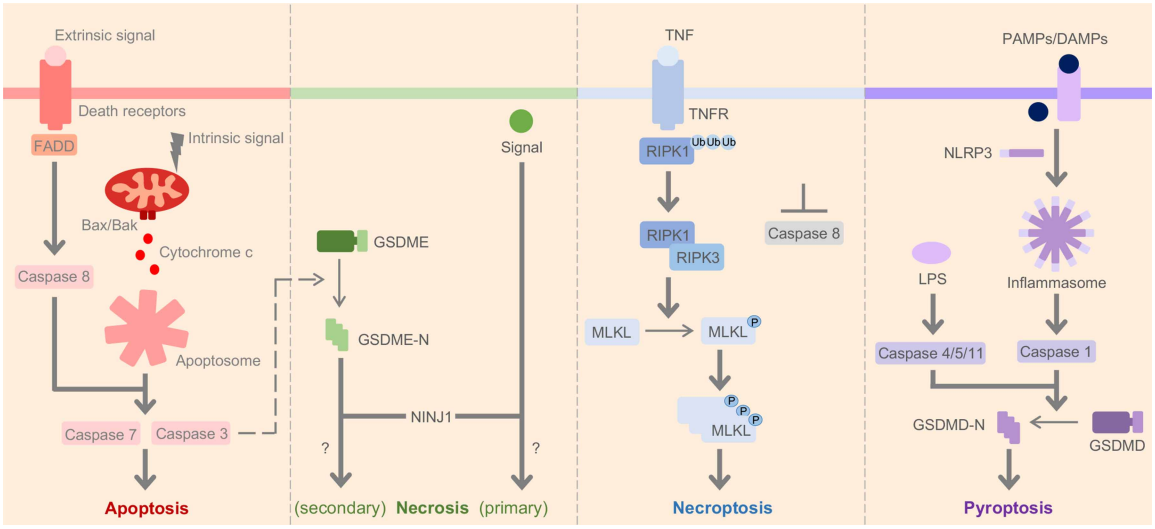

Figure 1. Schematic summary of the molecular mechanisms driving cell death.

Intrinsic and extrinsic apoptotic signals are received and converge at the activation of caspase 3/7. Caspase 3-cleaved GSMDE and NINJ1 may facilitate the progression to secondary necrosis. Primary necrosis is traditionally stochastic in nature resulting in uncontrolled membrane rupture. In the presence of caspase inhibition, TNF-α can induce necroptosis through binding the TNF-R which results in the activation of RIPK1/3 and formation MLKL pores at the membrane. Pyroptosis is mediated via either the canonical (caspase 1) or non-canonical (caspase 4/5/11) pathway which converge with the formation of GSDMD membrane pores.

Molecular mechanisms of dying cell clearance

Apoptosis

Apoptosis is largely driven by one of two pathways: the intrinsic/mitochondrial pathway and the extrinsic/receptor-mediated pathway. Both rely on the activation of the executioner caspases 3 and 7 to regulate the dismantling of the dying cell (Figure 1) [18]. Notably, cytotoxic lymphocyte killing also induces apoptosis via a Granzyme B-mediated mechanism. Similar to the induction of apoptosis, the clearance of apoptotic cells is a tightly controlled and well-studied process, and can be separated into three steps: recruitment, engagement and engulfment. Phagocytes are initially recruited to the site of apoptotic death by sensing 'find-me' signals actively released by apoptotic cells. These include soluble nucleotides such as ATP and UTP [19,20], sphinosine-1-phosphate (S1P) [21], lysophosphatidylcholine (LCP) [22] and MCP-1 [23]. Recruited phagocytes then engage with the apoptotic cell by binding either directly or via bridging molecules to an array of 'eat-me' signals exposed on the apoptotic cells outer-membrane. This includes calreticulin (CRT, both endogenous [24] and exogenous CRT secreted by phagocytes [25,26]), thrombospondin [27], ICAM3 [28], pentraxin 3 [29] and most notably, the phospholipid phosphatidylserine (PtdSer) [30]. Although normally located on the inner plasma membrane leaflet, PtdSer translocates to the outer leaflet during apoptosis through caspase 3/7 activation of the key scramblase Xrk8, and inactivation of flippases ATP11A and ATP11C which prevent PtdSer exposure on healthy cells [31–33].

Phagocytes are equipped with a diverse repertoire of engulfment receptors which engage directly with 'eat-me' signals, including TIM1/3/4 [34–36], BAI1 [37], RAGE [38], TLT2 [39], CD300b [40] and Stablin-2 [41]. Moreover, MFG-E8 and Gas6/Protein S can act as bridging molecules between PtdSer on the apoptotic cell and phagocytic intregrins and TAM receptors (Tyro, Axl, MerTK) to facilitate phagocytic-apoptotic cell engagement, respectively [42–45]. Similarly, the complement protein C1q and mannose-binding lectin can bind to exposed PtdSer and bridge with phagocytic receptors such as Megf10 and CRT/CD19 [46–49]. Apoptosis is also associated with dramatic DNA fragmentation, and a recent study identified that clusterin binding to histones exposed on the apoptotic cell surface exhibit opsoninic behaviour to aid cell clearance [50]. The efficiency of apoptotic cell clearance is also dependent on the apoptotic particle size. Recent studies have demonstrated that the fragmentation of apoptotic cells into extracellular vesicles known as apoptotic bodies

(~1–5 μm in diameter) is a tightly controlled process regulated by ROCK1 [51], PANX1 [52] and Plexin B2 [53]. As apoptotic bodies also expose the 'eat-me' signal PtdSer, the disassembly of apoptotic cells into apoptotic bodies generates numerous 'bite-sized' pieces that aid efficient engulfment by surrounding phagocytes [51,53,54]. Whether apoptotic bodies release 'find-me' signals to aid phagocytic recruitment to the initial site of cell death is unclear. Together, interactions between the phagocyte and the apoptotic fragments through these various mechanisms trigger an array of downstream signalling steps such as cytoskeletal reorganisation required to mediate phagocytosis [55].

It is important to note that 'eat-me' signal recognition can be attenuated if outcompeted by enhanced or clustered 'don't eat-me' signals such as CD47 [56,57], CD31 [58], and more recently CD24 [59] (Figure 2). The recognition of 'don't eat-me' signals negatively regulate engulfment, preventing the unnecessary clearance of healthy cells. However, cancer cells often exploit these mechanisms and up-regulate 'don't eat-me' signals to evade phagocytosis [60].

Primary and secondary necrosis

Necrosis, either primary or secondary (occurring after the completion of apoptosis), is traditionally an unregulated form of cell death largely characterised by stochastic membrane lysis (Figure 1) [61]. It was recently suggested that caspase cleavage of Gasdermin E (GSDME) may mediate the progression of apoptosis to secondary necrosis through inducing membrane lysis [62] however, results are conflicting [10,63]. Alternatively, NINJ1 may regulate necrotic-cell membrane permeabilization [10]. In contrast with apoptotic cells which tightly regulate the activation of PANX1 channels and release of 'find-me' signals such as ATP, necrotic cells may stochastically release ATP as a by-product of uncontrolled membrane permeabilization [61]. Consequently, primary necrotic cells can release significantly higher levels of ATP (compared with apoptotic cells) and may be more efficient at inducing phagocyte recruitment [64]. Thus, ATP is a necrotic 'find-me' signal [64,65] which may function in concert or independently of other necrotic 'find-me' signals including formyl-peptides [66] and chemokines [67].

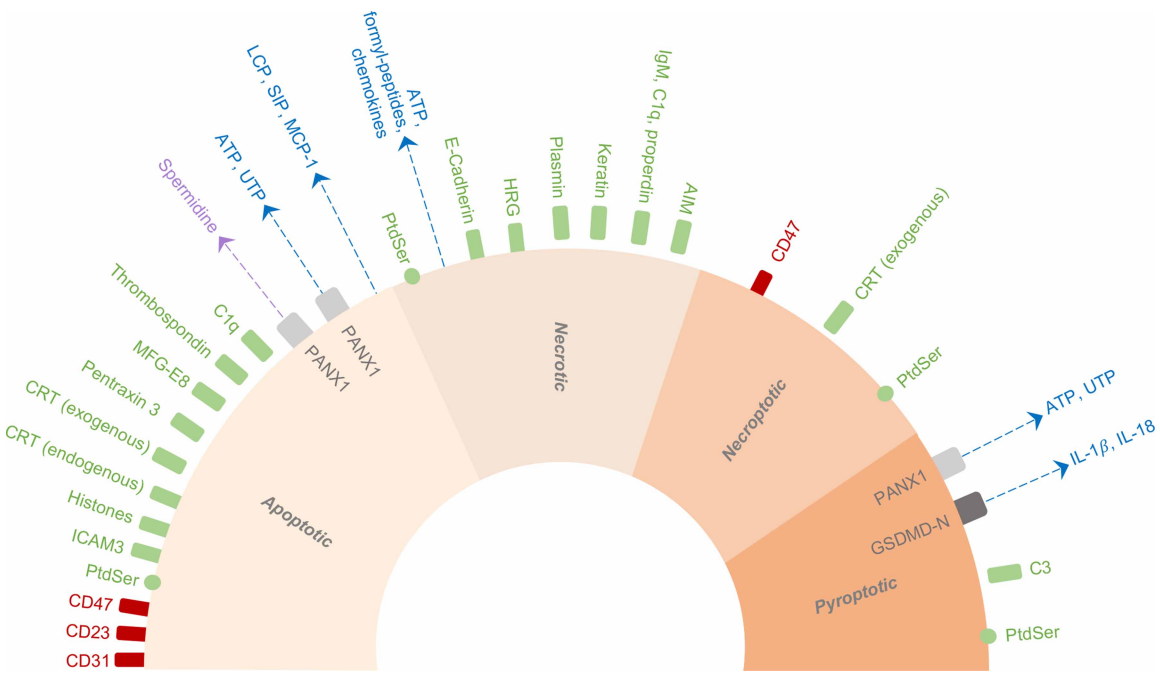

Figure 2. Molecular signals of dying cell clearance.

Schematic summary of the molecular mechanisms which facilitate apoptotic, necrotic, necroptotic and pyroptotic cell clearance. This includes the exposure of 'eat-me' (green) and 'don't eat-me' (red) signals, secretion of 'find-me' signals (blue) and release of 'good-bye' signals (purple).

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Once recruited to the site of cell death, phagocytes may internalised apoptotic and necrotic cells via different mechanisms (Figure 2) [68,69]. In comparison with the series of flippases and scramblases which regulate PtdSer exposure during apoptosis [31–33], the stochastic loss of phospholipid asymmetry by necrotic cells may result in limited or varying PtdSer exposure upon membrane lysis [64]. Although necrotic cells characteristically possess substantial Annexin V (AV) staining in flow cytometry-based assays [70], this should not be used to measure 'eat-me' signal exposure as AV will also bind PtdSer on the inner plasma membrane leaflet of necrotic cells. In fact, key phagocytic receptors such as TIM4 which mediate apoptotic cell engulfment through binding PtdSer poorly recognise necrotic cells [64]. Although necrotic cells can be cleared through PtdSer-mediated pathways [71–74], other mechanisms exist and contribute to their removal. This includes the exposure and binding of adhesion molecules such as E-cadherin [71], keratin [75], plasmin [76], as well as complement molecules including IgM, C1q and properdin [77–81] and HRG [82]. In a model of acute kidney injury, the protein AIM was also shown to label necrotic debris, interact with phagocytic KIM-1 and aid necrotic cell clearance, whereas the addition of AV to attenuate PtdSer recognition did not influence engulfment [83]. The phagocytic receptors CD14, CD36 and integrins αvβ3 may also contribute to the recognition of necrotic cell 'eat-me' signals and their timely removal [79].

Necroptosis

In comparison with the clearance of apoptotic and necrotic cells, the removal of cells dying via other cell death mechanisms such as necroptosis is only beginning to be defined. Necroptosis can be activated by the TNF pathway and is driven by the executioners RIPK1/3 and MLKL in the presence of caspase inhibition, such as during viral infections (Figure 1) [84]. Thus, many of the key regulators activated by caspase 3/7 (e.g PANX1 and Xrk8) required to facilitate clearance mechanisms (e.g. ATP release and PtdSer exposure) are not typically active during necroptosis. Although ATP release by necroptotic cells has been reported, whether this occurs prior to or as a consequence of membrane permeabilization has not been confirmed [85]. Recent findings have identified that necroptotic cells can expose PtdSer prior to membrane permeabilization, and this is dependent on the key necroptotic regulators RIPK3 and MLKL [86–88]. Therefore, the PtdSer-binding molecule MFG-E8 can recognise necroptotic cells and overexpression of the phagocytic receptor TIM4 can boost necroptotic cell clearance [88,89]. Similarly, supplementation of AV can attenuate necroptotic cell uptake [85]. In addition to PtdSer, the lipid mediator Resolvin D1 may also mediate necroptotic cell clearance by inducing phagocytic CRT secretion which can label and aid the recognition of necroptotic bone marrow-derived macrophages (BMDM) [90]. In contrasting findings, knock down or supplementation of soluble CRT was unable to alter the clearance of necroptotic fibroblasts [89] and may highlight potential cell-type specific clearance mechanisms. In line with this, as necroptotic BMDMs possess substantial expression of the 'don't eat-me' signal CD47, such cells may require additional factors, such as CRT, to mediate their efficient clearance [90]. Necroptotic cells also release small PtdSer positive extracellular vesicles akin to apoptotic bodies, termed necroptotic bodies [86,88]. Whether these necroptotic bodies also contribute to the efficiency of necroptotic cell clearance remains an unanswered question of interest.

Pyroptosis

Pyroptosis is an inflammatory form of programmed cell death triggered by the recognition of pathogen-associated molecular patterns such as bacterial LPS and DAMPs such as ATP [91]. It is initiated by either the canonical (caspase 1) or non-canonical pathway (caspase 4/5/11) which converge with activation of Gasdermin D (GSDMD) [91] (Figure 1). Similar to necroptotic cell clearance, the molecular mechanisms underpinning the removal of pyroptotic cells are still being defined (Figure 2). The clearance of pyroptotic cells is of significant interest as pyroptosis is widely implicated in inflammatory pathologies including Alzheimer's disease [92–94], liver fibrosis [95,96] and *Salmonella* infection [97,98]. Similar to the cleavage of PANX1 by caspase 3/7 during apoptosis, PANX1 is also activated by caspase 1/11 during pyroptosis and aids the release of ATP 'find-me' signals to mediate phagocytic recruitment [64,99–101]. Pyroptotic cells also secrete IL-1β and IL-18 in a cell-lysis independent manner through GSDMD pores to recruit phagocytes [102,103].

Once recruited to the site of cell death, phagocytes can engage with exposed PtdSer on the pyroptotic cell surface via bridging molecules (MFG-E8) or directly by scavenger receptors (TIM4) [64,89]. The mechanism of PtdSer exposure during pyroptosis is not dependent on caspase 1 [64] and whether it is an active or passive event remains elusive. Given that the phospholipid scramblase TMEM16F can be activated via Ca^{2+} signalling [104,105], whether such scramblases contribute to PtdSer exposure during cell death modalities without

caspase 3/7 activity, such as pyroptosis, would be of interest to determine. Nevertheless, supplementation of AV has also been shown ineffective in blocking pyroptotic cell uptake, suggesting that other factors contribute to pyroptotic cell clearance [89,99]. In line with this, complement proteins can contribute to the rapid removal of pyroptotic cells, as mice deficient in the complement protein C3 are unable to recruit phagocytes to the site of death nor clear pyroptotic cells efficiently [99]. Clearance could further be impaired by broad inhibition of scavenger receptors, suggesting that C3 may act as a bridging molecule between pyroptotic cells and phagocytic scavenger receptors to mediate clearance [99].

Functional impact of dying cell removal

The engulfment hierarchy

As the persistence of dying cells can trigger a breadth of inflammatory disease, swaying the mechanism of cell death to ensure swift, immunoprotective clearance is an exciting therapeutic potential. Moreover, understanding the engulfment hierarchy, i.e. which type of dying cells are cleared more efficiently, is of significant interest. Overall, the literature suggests that apoptotic cell clearance trumps the removal of necrotic-like cells [64,68,73,86,89,106]. As necrotic cells possess varying levels of the notable 'eat-me' signal PtdSer, phagocytic receptors may poorly recognise necrotic cells [64]. Consequently, phagocytes may require more time to engulf necrotic cells compared with their apoptotic counterparts [73]. Additionally, in comparison with apoptotic cells which rapidly bleb and fragment into apoptotic bodies, necrotic cells typically generate a single large bleb and remain as one cellular entity [107]. Given the role of dying cell fragmentation in aiding cell clearance [51,53], this may also provide a possible explanation for the inefficiency of necrotic cell clearance and the different mechanisms that contribute to their removal, compared with apoptotic cell uptake [68,73]. The clearance of apoptotic cells was also shown to be more efficient than necroptotic cell engulfment in both *in vitro* and *in vivo* settings, and also than pyroptotic cells *in vitro* [64,86,106]. However, contrasting findings have also been reported [89].

It is difficult to directly compare kinetics and phagocytic efficiencies between studies as the time post-induction of target cell death, phagocyte-to-target cell ratio and engulfment time often vary greatly. Moreover, kinetic comparison within studies must ensure equal levels of cell death to accurately compare phagocytic efficiencies. *In vitro* engulfment assays are also not representative of physiological conditions where various types of phagocytic cells (i.e. macrophages and epithelial cells) are present, and neighbouring cells may undergo different forms of cell death simultaneously (i.e. apoptosis or necrosis). Notably, competition phagocytosis assays have investigated whether apoptotic and necrotic cells could out-compete one another but results are conflicting [69,74]. Nevertheless, at a simplistic level, cells that expose 'eat-me' signals during the early stages of death (i.e. apoptosis), are expected to be cleared more rapidly [73]. Additionally, the secretion of multiple 'find-me' signals, vast number of 'eat-me' signals and significant redundancies in the phagocytic receptors which regulate apoptotic cell engulfment all strengthen the case for apoptotic cell clearance as the most efficient. However, increased interest in cell death and clearance pathways, and new findings such as the identification of PtdSer exposure prior to membrane permeabilization during necroptosis [86–88] may change our understanding of the engulfment hierarchy.

The consequence of death and dinner

Like the induction of cell death, the clearance of cells dying via different mechanisms can elicit distinct inflammatory signatures and impact the downstream immune response such as wound healing. Apoptosis is a traditionally immune-silent process which results in the direct or indirect release of anti-inflammatory mediators. For example, apoptotic cells secrete an array of anti-inflammatory factors such as IL-10, [108], TGF-β [109], and MFG-E8 [110]. Moreover, sensing of apoptotic 'find-me' signals such as S1P can both enhance cell clearance and induce phagocytic secretion of TGF-β, whilst decreasing pro-inflammatory factors like TNF-α and IL-6 [111]. Akin to 'eat-me' and 'find-me' signals, a new engulfment signal was recently described coined 'good-bye' signals. Apoptotic cells can release 'good-bye' signals in form of metabolites such as spermidine which induce anti-inflammatory gene expression in surrounding phagocytes, as well as wound healing, cytoskeletal organisation and anti-apoptotic responses [5]. The engulfment of apoptotic cells further contributes to inflammation control whereby upon uptake, phagocytes secrete anti-inflammatory factors including TGF-β [112,113] and IL-10 [114] and angiogenic factors to mediate wound healing such as VEGF [115], whilst limiting pro-inflammatory cytokine secretion [113]. Thus, not only do apoptotic cells prepare themselves for efficient clearance, but they also modulate the surrounding environment to prime phagocytes for engulfment and

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maintain anti-inflammatory conditions. As such, many studies have harnessed the anti-inflammatory properties of apoptotic cells and their clearance to combat robust inflammatory disease such as rheumatoid arthritis [5,16,116,117].

In contrast with the anti-inflammatory properties of apoptotic cells and their engulfment, necrosis is largely associated with robust inflammation. Necrotic cells undergo rapid membrane lysis and release a wide variety of intracellular contents including the pro-inflammatory cytokines IL-1α [118] and MIF [119], and DAMPs including HMGB1 [120,121], HSP70/90 [122,123] and DNA [121]. Although some studies have proposed necrotic cells to be more effective in recruiting phagocytes to the site of death through these signals [118], the pro-inflammatory consequence of necrosis likely outcompetes the benefit of rapid phagocytic recruitment. Moreover, once recruited to the site of necrosis, phagocytosis and sensing of necrotic debris can further exacerbate inflammation whereby phagocytes release pro-inflammatory cytokines such as IL-8 and TNF-α [82,124].

Necroptosis and pyroptosis are also associated with robust inflammation. Necroptotic cells release an array of pro-inflammatory mediators such as IL-8, IL-1, CXCL2 and cyclophilin A [125–127], and engulfment of necroptotic cells further exacerbates inflammation by triggering phagocytic TNF-α and MCP-1 secretion [86]. Similarly, pyroptotic cells secrete IL-1β, IL-18, TNF-α and IL-6 which can drive inflammatory disease such as liver fibrosis and arthritis [95,128]. Although the inflammatory consequence of pyroptotic cell clearance on the phagocytosing cell is unclear, engulfment of 'NLRP3 inflammasome particles' or the inflammasome-associated adaptor protein complexes 'ASC specks' can elicit phagocytic inflammatory cytokine secretion [96,129]. Altogether, as a single stimuli can elicit multiple forms of cell death, such as the induction of necrosis, apoptosis, necroptosis and potentially pyroptosis by influenza A virus [130], the functional impact of dead cell clearance in physiological settings and disease is complex and requires comprehensive investigations.

Future directions of dying cell clearance

The swift removal of dying cells is paramount to prevent disease onset and understanding the molecular mechanisms underpinning their clearance is vital. Moreover, harnessing this knowledge to develop novel therapeutics and boost dead cell clearance in inflammatory disease settings where clearance is aberrant possesses exciting clinical potential. Although many of the major molecular components contributing to efficient cell clearance have been described, there still remains a significant knowledge gap.

Exploring the mechanistic differences between targets and phagocytes

Although recent literature has shed light on how necroptotic and pyroptotic cells are recognised and phagocytosed, this literature merely represents the tip of the iceberg. Given the vast number of machineries that mediate the removal of apoptotic and necrotic cells, other factors in addition to ones currently described likely contribute to necroptotic and pyroptotic cell clearance. Moreover, the clearance of cells undergoing alternative cell death pathways such as ferroptosis, parthanatos and NETosis is poorly understood but also of significant interest. For example, the defective clearance of NETs has been observed in inflammatory disease such as respiratory distress syndrome whereby soluble components in the bronchioalveolar lavage fluid of patients could impair phagocytic NET uptake [131]. Therefore, whether known or novel engulfment receptors contribute to NET removal would be of great interest. Understanding the different mechanisms of dead cell clearance is especially important for disease settings which elicit multiple cell death pathways simultaneously and potentially require a multifaceted therapeutic strategy. Moreover, whether professional and non-professional phagocytes can recognise their targets via different modalities or receptors, or whether uptake elicits a different response is yet to be determined. The interplay between professional and non-professional phagocytes is especially interesting as upon apoptotic cell engulfment, macrophages can secrete IGF-1 and enhance the phagocytic efficiency of surrounding non-professional phagocytes [6]. Whether professional phagocytes have a superior role in the clearance of inflammatory cells (i.e. pyroptotic cells), or differences within immune cell subsets exist (i.e. M1 vs M2 macrophages), are also outstanding questions. Notably, it was reported that 'large'-DCs were more efficient in phagocytosing necrotic cells than 'small'-DCs [132]. However, whether this was due to mechanistic differences rather than restricted size and phagocytic capacity is unclear.

Clinical potential of harnessing dead cell clearance

It is well established that aberrant clearance of dying cells can trigger inflammatory disease. For example, impairment of cell clearance by clusterin or MFG-E8 deficiency results in autoimmune disease-like symptoms [14,50]. Moreover, inflammation and atherosclerotic plaque formation may in part be due to the accumulation

of apoptotic cells from impaired phagocytosis during atherosclerosis [11,12]. Therefore, boosting cell clearance represents an exciting therapeutic potential and may be achieved by exploiting various clearance mechanisms. Blocking the 'don't eat-me' signal CD47 with monoclonal antibodies during atherosclerosis has shown promise in boosting apoptotic cell clearance and reducing disease burden in mice [133]. Notably, targeting the expression of 'eat-me' signals is yet to be explored but may also represent a suitable approach. Up-regulating engulfment receptors or 'priming' macrophages to enhance engulfment likely represents the most efficient therapeutic strategy. Genetically overexpressing the phagocytic receptor BAI1 has already been shown to boost apoptotic cell engulfment *in vivo* and attenuate disease-associated inflammation in mice [134]. However, the translation of these therapeutic strategies to the clinic still remains a challenge.

Recent advances have revealed the tight molecular control underpinning the disassembly of apoptotic cells into apoptotic bodies and demonstrated the importance of this process in aiding rapid cell clearance [51,53]. Therefore, simultaneously inducing apoptosis and boosting the disassembly of apoptotic cells may provide an effective approach to enhance cell clearance in disease settings such as solid tumours where cell death and their swift removal is crucial. The antibiotic Trovafloxacin was identified as the first pharmaceutical enhancer of apoptotic body formation [52,135] and thus, Trovafloxacin or similar PANX1 inhibitors may be suitable candidates to investigate such therapeutic strategies. Furthermore, as mentioned above, whether necroptotic or pyroptotic cells also fragment into smaller vesicles (i.e. necroptotic bodies and pyroptotic bodies) which aid phagocytosis remains an unanswered question of clinical relevance.

The efficient removal of dying cells is regulated by a complex and redundant series of machineries which can elicit both pro- and anti-inflammatory effects on the phagocyte and surrounding environment. Although how phagocytes are recruited to, recognise and engulf other dying cells beyond apoptosis is still being defined, the ability to translate these findings clinically and treat inflammatory disease is an exciting prospect for the cell clearance field.

Perspectives

- Cell death and the removal of dying cells is tightly linked to a variety of inflammatory disease. Thus, understanding the molecular mechanisms responsible for phagocytic clearance and the functional impact of engulfment on the phagocyte is essential for the development of novel disease therapeutics.
- Efficient dead cell clearance can be split into three individual steps including recruitment, recognition and engulfment which are mediated by the release and exposure of 'find-me', 'eat-me', 'don't eat-me' and 'good-bye' signals.
- With the rapid expansion of the cell death field, further research is needed to understand how different cell types, and cells dying via different mechanisms are cleared, the impact this has on the phagocytosing cell and how this can be targeted therapeutically.

Competing Interests

The author declares that there are no competing interests associated with this manuscript.

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G.K.A.-S. wrote and edited the manuscript.

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Abbreviations

AIM, Apoptosis inhibitor of macrophage; ATP, Adenosine triphosphate; BAI1, Brain-specific angiogenesis inhibitor 1; DCs, Dendritic cells; FADD, Fas-associated protein with death domain; GSDMD-N, Gasdermin D N-terminal; GSDME-N, Gasdermin E N-terminal; ICAM3, Intracellular adhesion molecule 3; IGF-1, Insulin-like growth factor 1; MCP-1, Monocyte chemoattractant protein 1; MFG-E8, Milk fat globule epidermal growth factor 8; MLKL, Mixed lineage kinase domain-like; NINJ1, Nerve injury-induced protein 1; NLRP3, NLR family pyrin domain containing 3; P, Phosphorylation; PANX1, Pannexin 1; RAGE, Receptor for advanced glycation endproducts; RIPK1/3, Receptor-interacting serine/threonine-protein kinase 1/3; ROCK1, Rho-associated coiled-coil containing protein kinase 1; TNF, Tumour necrosis factor; TNFR, Tumour necrosis factor receptor; Ub, Ubiquitination; UTP, Uridine triphosphate.

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