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Phagoptosis - Cell Death By Phagocytosis - Plays Central Roles in Physiology, Host Defense and Pathology

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Abstract: Cell death by phagocytosis – termed 'phagoptosis' for short – is a form of cell death caused by the cell being phagocytosed i.e. recognised, engulfed and digested by another cell. Phagocytes eat cells that: i) expose 'eat-me' signals, ii) lose 'don't-eat-me' signals, and/or iii) bind opsonins. Live cells may express such signals as a result of cell stress, damage, activation or senescence, which can result in phagoptosis. Phagoptosis may be the most abundant form of cell death physiologically as it mediates erythrocyte turnover. It also regulates: reproduction by phagocytosis of sperm, development by removal stem cells and excess cells, and immunity by removal of activated neutrophils and T cells. Phagoptosis mediates the recognition of non-self and host defence against pathogens and cancer cells. However, in inflammatory conditions, excessive phagoptosis may kill our cells, leading to conditions such as hemophagy and neuronal loss.

Keywords: Phagocytosis, apoptosis, cell death, turnover, inflammation, clearance.

INTRODUCTION

Phagoptosis is a recently recognized form of cell death, defined as death caused by phagocytosis (recognition, engulfment and digestion) of the cell, and therefore death of the cell is prevented by inhibition of phagocytosis or phagocytic signaling [1,2].

What is the function of phagoptosis? Where phagoptosis is functional, it must be functioning to remove cells that are dysfunctional, pathological or simply not needed. There is evidence that phagoptosis functions to remove: excess cells during development in *C. elegans* [3,4], damaged cells in *C. elegans* and mammals [5-7], aged/senescent erythrocytes and neutrophils, excess activated cells as well as pathogens and cancer cells (see below). However, phagoptosis can in some conditions be dysfunctional by removing cells that are needed, for example during neuroinflammation (see below).

Although phagoptosis was only recently recognized as an important form of cell death, the history of the concept goes back to the discoverer of phagocytosis: Elie Metchnikoff [8]. Metchnikoff showed that phagocytes mediated immune defense by phagocytosing live bacteria and other pathogens, and thereby killing them. Furthermore, in 1892, he proposed that: "phagocytes eat all parts of the organism which have become weak for any reason, while ignoring parts fully capable of living." Examples of processes where this mechanism was suggested to occur included metamorphosis of the tadpole tail and removal of senescent red and white blood cells in the spleen [8].

Phagoptosis involves two cells: a cell that phagocytoses (the phagocyte) and a cell that is phagocytosed (the target cell) and thereby killed. Obviously, this form of cell death is not cell autonomous, however, this does not mean that the target cell does not play an active role in phagoptosis. In fact all host cells (in contrast to pathogens or non-self cells) must display signals on their surface in order for them to be phagocytosed by phagocytes. Phagocytes, such as macrophages, monocytes and dendritic cells, only phagocytose target cells that they contact if: i) the target cell has one or more "eat-me" signals or opsonins on its surface, and the phagocyte has the corresponding receptors, and ii) the target cell lacks "don't-eat-me" signals to which the phagocyte is responsive [1,2] (Figs. **1**, **2** and Table **1**).

EAT-ME SIGNALS, DON'T-EAT-ME SIGNALS AND OPSONINS

The most well-known eat-me signal is the exposure of phosphatidylserine on the cell surface [9,10]. In healthy, non-activated cells, the phospholipid phosphatidylserine is found within the inner leaflet but not the outer leaflet of the plasma membrane, because aminophospholipid translocases, identified as the P_4-ATPases ATP8A1 and ATP8A2 [11] or ATP11C and CDC50A [12], translocate phosphatidylserine from the outer to inner leaflet. Phosphatidylserine exposure can be caused by inhibition of phosphatidylserine translocases by calcium elevation, oxidative stress, ATP depletion [13] or caspase activation [12]. Alternatively, reversible phosphatidylserine exposure can result from calcium-activated phospholipid

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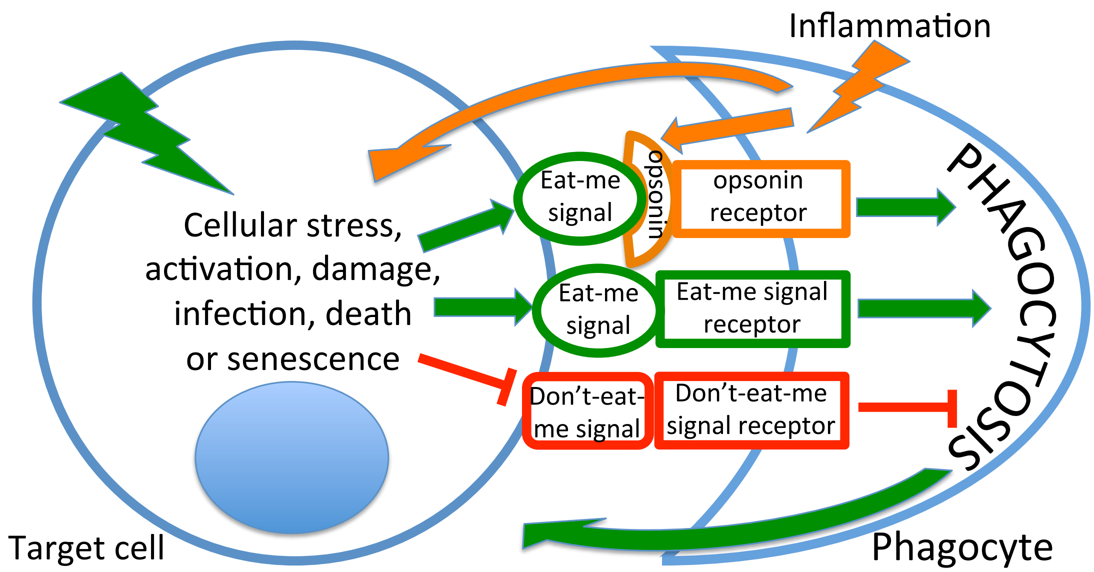

Fig. (1). Eat-me signals, don't-eat-me signals, opsonins and receptors. Cellular stress, activation, damage, infection, death or senescence may promote phagocytosis of a cell *via* cell surface exposure of eat-me signal and/or downregulation of don't-eat-me signals, which activate receptors on phagocytes that activate or inhibit phagocytosis respectively. Inflammatory activation of phagocytes enhances phagocytosis by: release of factors that stress target cells, release of opsonins, and upregulation of opsonin receptors and eat-me signal receptors.

scramblases such as TMEM16F [14]. However, irreversible phosphatidylserine exposure during apoptosis is apparently caused by caspase cleavage of XKR8 [15].

While phosphatidylserine exposure can occur on apoptotic cells, it is now clear that phosphatidylserine exposure can also occur on viable cells independent of apoptosis [16-19,23-25,79]. Phosphatidylserine exposure on viable cells may consequently lead to the phagocytosis of such cells in the presence of macrophages with phagocytic receptors activated by phosphatidylserine [12,13,16,20-22]. Phosphatidylserine exposure is sufficient to provoke phagocytosis in some cell types and conditions, but not in others [12,23,24]. Some cells may require: phosphatidylserine oxidation, or phosphatidylserine lipolysis to lysophosphatidylserine, or phosphatidylserine-binding opsonins, or other co-stimulatory eat-me signals, or the loss of don't-eat-me signals, to induce phagocytosis [21,22,25].

Exposed phosphatidylserine may be recognised by phagocytes either: i) by receptors that bind directly to phosphatidylserine such as Tim4, BAI1, Stabilin-1 and Stabilin-2, or ii) by receptors that bind indirectly to phosphatidylserine *via* opsonins that do bind directly to phosphatidylserine [10,23,26,27]. For example, MFG-E8 is an opsonin binding exposed phosphatidylserine and inducing phagocytosis *via* also binding the vitronectin receptor (an $\alpha_v\beta_{3/5}$ integrin). MerTK binds phosphatidylserine and other eat-me signals indirectly through opsonins Gas-6, Protein S, galectin-3, Tubby and Tulp1 [10]. Most of such opsonins are released by phagocytes in inflammatory conditions, for example, Annexin A1 is secreted by activated macrophages and neutrophils to bind exposed phosphatidylserine on neutrophils, and induce their phagocytosis *via* formyl peptide receptors on macrophages [26].

Opsonins are molecules that bind to the surface of cells to enhance their phagocytosis by phagocytes. Opsonins include the adapter proteins listed above that bind to eat-me signals, but also include complement factors and antibodies that induce the phagoptosis of pathogens and host cells [27].

Calreticulin is the other main eat-me signal (Fig. **2**), which induces phagocytosis of both dead and live cells *via* activation of LRP on phagocytes [28]. Calreticulin is a chaperone within the endoplasmic reticulum, but ER stress can cause exposure on the cell surface, where it induces phagocytosis of that cell [29]. Calreticulin is constitutively present on the surface of neutrophils [30], potentially contributing to their rapid turnover. And many cancer cells have calreticulin on their surface, possibly explaining why they overexpress the don't-eat-me signal CD47 to block phagoptosis [31]. Calreticulin can also bind to phosphatidylserine and complement C1q, and act as co-receptor with LRP for C1q, resulting in phagocytosis of C1q-opsonized cells [32].

Most cells protect themselves from phagocytic removal by displaying don't-eat-me signals such as CD47 on their surface, which inhibit their phagocytosis (Figs. **1**, **2**). CD47 blocks engulfment by activating the SIRPα receptor on the phagocyte, and blocking CD47 with antibodies results in phagoptosis of live cells [32-42]. Sialic acid is a derivative of neuraminic acid, found as terminal sugar on glycoproteins or glycolipids, where it may act as a don't-eat-me signal by: activation of inhibitory Siglec receptors on phagocytes, and blocking the binding of opsonising lectins and complement C3b and C1q [43-45]. Removal of sialic acid residues by

Table 1. Eat-me signals, don't-eat-me signals, opsonins and their receptors.

Eat-me signal	Receptor	Opsonin
Phosphatidylserine (PS)	Tim4, BAI1, Stabilin-1, Stabilin-2	MFG-E8, Gas6, Protein S, Annexin A1, CRT, C1q
Calreticulin (CRT)	LRP1	C1q?
Desialylated cell	CR3?	Lectins, C1q?
Don't-eat-me signal	Receptor	Opsonin
CD47	SIRPα	Thrombospondin-1
Sialylated cell	Siglec	
CD31 (PECAM-1)	CD31	
PAI-1		
Opsonin	**Receptor**	**Eat-me signal**
MFG-E8	Vitronectin receptor	Phosphatidylserine
Gas6, Protein S	MerTK	Phosphatidylserine
Annexin A1	FPR2	Phosphatidylserine
Galectin-3	MerTK	Desialylated cell
IgG antibody	Fcγ receptors	Antigen
C1q	MBL, SP-A	PS, CRT, antibodies
iC3b	CR3	Desialylated cell?

sialidase can induce phagocytosis of viable lymphoblasts or neurons [44].

PHAGOPTOSIS IN NORMAL PHYSIOLOGY AND HOST DEFENCE

Phagoptosis to Remove 'Senescent/Aged' Erythrocytes in the Blood

Phagoptosis plays multiple physiological roles in the body, including the removal of aged/senescent red blood cells (erythrocytes). 2 million erythrocytes per second are generated in human bone marrow, circulate in blood for about 120 days, and are then phagocytosed by macrophages in spleen and liver at the same rate [46,47]. This rate of cell death, mediated by a form of phagoptosis called 'erythrophagocytosis', is the highest in the body in physiological conditions, and therefore phagoptosis is the most common form of cell death in the body [1].

Old erythrocytes are phagocytosed because they expose novel antigens that are opsonised by natural antibodies (Fig. **2**). The band 3 protein changes conformation with age, probably as a result of oxidative modification, resulting in aggregates of the protein that are recognized by endogenous IgG [46,47]. Additionally, cell surface glycoproteins are desialylated

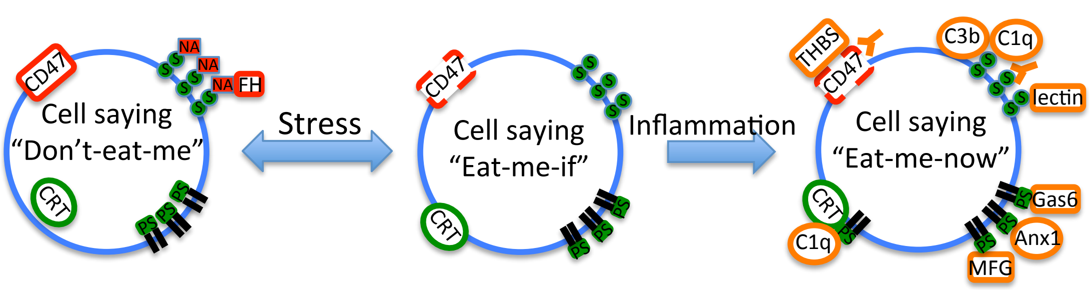

Fig. (2). Decoding the cell-eat-cell code. Healthy cells express the don't-eat-me signal CD47 and have a sialylated glycocalyx of sugar (S) chains terminated by neuraminic acid (NA), which bind complement Factor H (FH) to prevent C3b deposition. Healthy cells also lack eat-me signals phosphatidylserine (PS) and calreticulin (CRT) on their surface, because they are inside the cell. Cellular stress, activation, damage, infection or senescence may promote phagocytosis of the cell *via* exposure of PS and CRT, plus loss or altered conformation of CD47, and desialylation of the glycocalyx (causing loss of FH binding). Inflammation causes production and release of opsonins that may bind to eat-me-signals to enhance phagocytosis: exposed PS is bound by MFG-E8 (MFG), annexin A1 (Anx1), Gas6 and Protein S. C1q can bind exposed CRT and PS. Thrombospondin-1 (THBS) can bind CD47. Exposed PS and altered CD47 bind natural antibodies (Y shapes), which recruit C1q and thereby C3b. The desialylated surface binds lectins, C1q and natural antibodies.

with age, revealing novel antigens causing antibodies to bind and opsonise the aged erythrocytes [47]. Phagocytosis is blocked by the don't-eat-me signal CD47 acting on macrophage SIRPα receptors, so that deletion of CD47 results in rapid loss of erythrocytes *in vivo* [33,34]. CD47 expression is reduced with erythrocyte age, contributing to their removal [35]. CD47 on old erythrocytes may also adopt an altered conformation, enabling it to bind the opsonin thrombospondin-1 and thereby promote phagoptosis of the aged erythrocyte [36]. Eliminating macrophages increases erythrocyte survival [35], suggesting that macrophage phagocytosis of live erythrocyte mediates their turnover. Damaged, activated and *in vitro* aged erythrocytes expose phosphatidylserine and are removed by macrophages in the liver [48], but whether this contributes to physiological turnover is less clear.

Phagoptosis to Remove 'Senescent/Aged' Neutrophils in the Blood

0.5 - 1 million neutrophils per second are generated in human bone marrow, live for about one day, and are then phagocytosed by macrophages in bone marrow, liver and spleen [49]. This is by far the highest turnover and death rate of any cell in the body except the erythrocyte [1].

When isolated, neutrophils rapidly apoptose. However, overexpression of Bcl-2 in mice prevents apoptosis of isolated neutrophils, but has no effect on turnover and phagocytosis *in vivo*, so that physiological turnover of neutrophils does not appear to be mediated by apoptosis [50]. A role for apoptosis in neutrophil turnover has been claimed based on the finding that CD18 knockout mice have reduced apoptosis and much higher levels of circulating neutrophils [51], but as CD18 is part of CR3, a phagocytic receptor also required for neutrophil extravasation, this evidence is ambiguous.

Neutrophils have a circadian rhythm of entry and exit from the blood, driven by neutrophil "aging" in the circulation, causing decreased expression of CD62L and increased expression of CXCR4, which directs the "aged" neutrophils to the bone marrow, where they are phagocytosed by macrophages [49]. Usage of the terms "aged" or "senescent" in this context can be misleading as the cells are less than 24 hours old and it is not clear that they have lost function. "Aged" neutrophils in the blood do not have active caspases or phosphatidylserine exposure [49]. However, activated macrophages in tissues can cause live neutrophils to expose phosphatidylserine (not mediated by apoptosis), which induces phagoptosis *via* binding opsonins MFG-E8 and annexin A1 (AnxA1) [20]. *In vivo*, *AnxA1*$^{-/-}$ mice had reduced removal of neutrophils in the bone marrow, leading to an increased density of "senescent" neutrophils in the bone marrow [26]. Phagocytosis of "aged" neutrophils in the spleen appears to be dependent on the phagocytic receptor MerTK and its soluble phosphatidylserine-binding ligand Gas6, such that knockout of either gene increases neutrophil numbers in spleen and blood *in vivo* [52]. Thus it would appear that "aged" neutrophils are cleared *via* AnxA1 (the receptor for which is usually FPR2) in bone marrow and Gas6 and MerTK in spleen, and that "aged" neutrophils are cleared alive, rather than dead or dying, indicating that phagoptosis contributes to neutrophil turnover.

Neutrophils constitutively expose the eat-me signal calreticulin [30], and CD47 acts as a don't-eat-me signal, so that neutrophils are depleted by CD47-blocking antibodies *in vitro* [53] and *in vivo* [37]. Another don't-eat-me signal on neutrophils is plasminogen activator inhibitor-1 (PAI-1), so that knockout of the PAI-1 gene or PAI-1 blocking antibodies induced phagocytosis of live neutrophils by macrophages, reversed by adding PAI-1 [53].

If the homeostatic clearance of both erythrocytes and neutrophils is mediated by phagoptosis, then phagoptosis is several orders of magnitude more common in physiological conditions than apoptosis, necrosis and autophagic death put together [1].

Phagoptosis to Remove 'Activated' Cells and Resolve Inflammation

Viable blood cells, such as monocytes, neutrophils and lymphocytes, expose phosphatidylserine on their surface when 'activated' by detecting damage, pathogens or antigens [16-18]. This phosphatidylserine exposure on viable cells may regulate cellular fusion, trafficking and adhesion [17,18]. However, because exposure of phosphatidylserine can induce phagocytosis, it can also limit the lifetime of the activated state. For example, antigen recognition by activated T-cells causes exposure of phosphatidylserine [18] that is recognized by the macrophage Tim-4 receptor, causing phagocytosis of the activated T-cells [27]. Thus phagoptosis limits the extent of the initial adaptive response and the number of remaining memory cells [27].

Removing phosphatidylserine-exposed cells from the blood is vital because phosphatidylserine exposure on platelets or blood cells catalyses blood clotting, and thus it is important that cells exposing phosphatidylserine are cleared from the blood as rapidly as possible [54]. Turnover of both activated and aged platelets is by macrophage phagocytosis regulated by CD47 expression on platelets, so platelets lacking CD47 turnover more rapidly [55,56].

Stressed erythrocytes also expose phosphatidylserine and are cleared by stablin-1 and stablin-2 in liver, so that knockdown of these phosphatidylserine-receptors prevented phagocytosis of erythrocytes in liver and reduced removal of stressed erythrocytes *in vivo* [48].

Isolated neutrophils expose phosphatidylserine over time in culture, and this exposure can be increased on viable neutrophils by a variety of physiological stimuli thereby inducing phagocytosis by macrophages [20,57]. Activated neutrophils generate oxidized phosphatidylserine and lyso-phosphatidylserine, which potently induce macrophage phagocytosis of activated, live neutrophils *in vitro* and *in vivo* to terminate the inflammatory response [21,22,25]. Similar mechanisms may terminate the inflammatory response in the brain, where microglia can engulf any invading, activated, viable neutrophils [58].

Phagocytosis of phosphatidylserine-exposing apoptotic cells by macrophages supresses antigen presentation and promotes resolution of inflammation by stimulating production of anti-inflammatory cytokines by the phagocyte. At present it is not clear whether phagoptosis of phosphatidylserine-exposing viable cells can have similar anti-inflammatory and immune suppressive effects, but if so this would contribute to the capacity of phagoptosis to resolve inflammation.

Phagoptosis of Stem Cells and During Development

Microglia (brain macrophages) phagocytose neural precursor cells in the developing brain and thereby regulate neurogenesis [59]. Similarly, survival of hematopoietic stem cells depends on them avoiding phagocytosis by macrophages, and they upregulate the don't-eat-me signal CD47 during migration in order to reduce their phagoptosis [38,60]. Phagoptosis removes excess cells during development in *C. elegans* [3,4]. And during mammalian development, macrophages remove cells undergoing programmed cell senescence [61].

Phagoptosis in Host Defence Against Pathogens and Rejection of Non-Self

As Metchnikoff showed, phagocytosis of live pathogens by phagocytes is central to host defence against pathogens such as bacteria [62,63]. More recently it was found that neutrophils that have phagocytosed bacteria or cancer cell debris can in turn be phagocytosed alive by dendritic cells, which then present antigens derived from the bacteria or cancer cells to induce adaptive immunity [64].

Cells or tissues from one species grafted into another are rejected, partly as a result of an adaptive immune response, but also because the 'don't-eat-me' signal CD47 on the grafted cells is not recognised by SIRP-α on host macrophages, so that the grafted cells are phagocytosed by the host macrophages [65,66].

After insemination by males of the same species, large numbers of neutrophils are recruited to the uterus and phagocytose sperm, limiting fertilisation [67,68]. *In vitro* studies have indicated that viable sperm are phagocytosed to the same extent as damaged or killed sperm, but that capacitation of sperm reduces their phagocytosis [67,68].

Phagoptosis in Host Defence Against Cancer

Defence against cancer is known to be partly mediated by antibody-dependent or antibody-independent phagocytosis of live cancer cells by macrophages [31,66,69]. Phagocytosis of live cancer cells may result from the exposure of novel antigens, phosphatidylserine [54], senescence markers [40] or calreticulin [70]. However, most cancer cells overexpress CD47 to prevent such phagocytosis, but if this don't-eat-me signal is blocked by anti-CD47 antibodies then a variety of blood cancers can be cleared from the body [37,42,49,70]. However, it should be noted that while anti-CD47 antibodies induce phagocytosis of cancer cell by macrophages *in vitro*, it is unclear whether this is the only means by which such antibodies clear cancer cells *in vivo*, where complement, T cells and other mechanisms might also contribute.

In conclusion, phagoptosis has a wide variety of roles in physiology and host defence (Fig. **3**). We will next consider what is known about the contribution of phagoptosis to pathology.

PATHOLOGICAL PHAGOPTOSIS

Pathological Phagoptosis in Hemophagocytosis

Hemophagocytosis is a clinical condition characterised by macrophage engulfment of apparently live blood cells, resulting in reduced red and white cell counts (cytopenia). IFN-γ can induce hemophagocytosis during infection by stimulating macrophage phagocytosis of blood cells [71]. As macrophages consume blood cells normally, and inflammation increases their phagocytic capacity and increases phosphatidylserine exposure on leucocytes, this might explain the resulting hemophagocytosis and cytopenia.

Hemophagocytic lymphohistiocytosis (HLH) is a clinical condition characterized by macrophage phagocytosis of hematopoietic stem cells in the bone marrow, which appears to be caused by reduced CD47 expression on the stem cells, resulting in them being eaten alive by macrophages [60].

Pathological Phagoptosis in the Inflamed Brain

In inflammatory condition, microglia can phagocytose stressed-but-viable neurons, and this may contribute to neuronal loss in a variety of brain pathologies [2]. Phagoptosis of neurons by microglia can be induced by a variety of inflammatory stimuli, including lipopolysaccharide, lipoteichoic acid, Aβ, rotenone (a complex I inhibitor used to model Parkinson's Disease), TNFα and glutamate [72-82]. *In vitro*, phagoptosis is mediated by the production of oxidants from activated microglia, which induce reversible neuronal exposure of phosphatidylserine, which promotes their phagocytosis by microglia *via* MFG-E8, VNR and MerTK [72-82]. Strikingly, inhibition of the PS/MFG-E8/VNR pathway, completely prevents neuronal loss *in vitro*, and the rescued neurons are healthy, and remain viable for at least 7 days [72-82]. Furthermore, *in vivo* LPS-induced neuronal loss from the striatum is reduced in MFG-E8 knockout mice or by inhibition of the VNR or $P2Y_6$ [75,81]. Similarly, MerTK or MFG-E8 deficiency prevents long-term functional motor deficits and reduces brain atrophy after focal brain ischaemia [79]. Given that rescued neurons are healthy, phagocytosis is the cause, not the consequence, of cell death, and may contribute to neuronal loss in multiple brain pathologies [2,83].

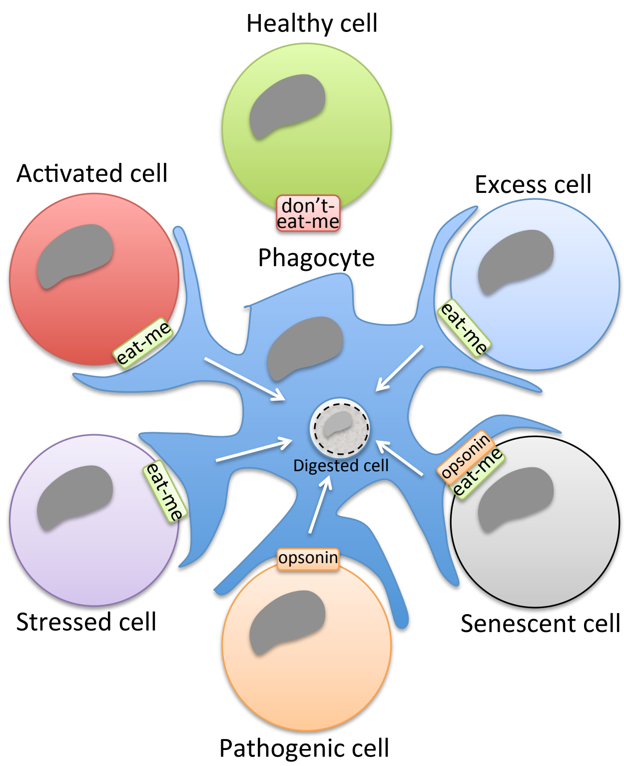

Fig. (3). Phagoptosis, i.e. cell death by phagocytosis, plays multiple roles in physiology and host defense. Phagoptosis removes excess, senescent, pathogenic, stressed and activated cells, but not otherwise healthy cells, regulated by 'eat-me' signals, 'don't-eat-me' signals and opsonins.

HOW IS PHAGOPTOSIS RELATED TO OTHER FORMS OF CELL DEATH AND CELL-IN-CELL PHENOMENA?

Phagoptosis needs to be distinguished from a variety of other forms of cell death and related processes (Table **2**). The terms "efferocytosis" and "programmed cell clearance" [84] have been used to refer to the phagocytosis of apoptotic cells by phagocytes, and therefore does not overlap in meaning with "phagoptosis". Efferocytosis and phagoptosis share many common phagocyte/target recognition systems. However, in the case of efferocytosis, inhibition of phagocytosis will not prevent target cell death, whilst phagoptosis is defined by the fact that inhibition of phagocytosis prevents target cell death. Note however, that in some conditions, the process of apoptosis, i.e. activation of Bcl-2 homologous proteins and/or caspases, is sufficient to trigger phagocytosis of the cell, but insufficient to cause death of the cell in the absence of such phagocytosis [3-7], and in such unusual conditions apoptosis and phagoptosis are coupled causes of death. The term "programmed cell removal" has been used to refer to the phagocytosis of dead, dying or viable cells by phagocytes [85], thus it does not normally cause cell death and its meaning is not synonymous with phagoptosis.

"Phagocytosis-induced cell death" has previously been defined as the cell death of a phagocyte resulting from it phagocytosing something, e.g. death of neutrophils resulting from them phagocytosing bacteria [86]. Clearly this differs from phagoptosis, in that it is the phagocyte rather than the cell that is phagocytosed that dies.

"Entosis" is a process by which one cell invades another cell, normally triggered by the invading cell detaching from extracellular matrix [87]. The invading cell may live within the invaded cell, and may later exit from the invaded cell, or may die within the invaded cell by lysomal mediated degradation or apoptosis, or may kill the invaded cell by some means [87]. But clearly entosis is not itself a form of cell death, but rather a process of cell invasion, and therefore has no obvious relation to phagoptosis. Entosis is further distinguished from phagocytosis and efferocytosis by the facts that: i) it occurs independently of phosphatidylserine recognition, and ii) actin cytoskeleton rearrangement is required in the internalised cell for entosis, rather than in the engulfing cell as is the case in efferocytosis and phagoptosis.

"Cell cannibalism" is a process described in cancer cells, defined as one cancer cell living inside another cancer cell, which may or may not result in death of one or other cell [88]. The term "emperipolesis" is more generally used to describe the phenomenon of one cell living within another. Originally the term "cell cannibalism" was used by pathologists, looking at fixed tissue sections, to describe a smaller cell inside a larger cancer cell, where the larger cell had a sickle-shaped nucleus [88]. However, more recently the term has been extended to cover tumor cells engulfing (or

Table 2. Clarification of terms used.

Name	Live Cell Process
Phagocytosis	Cellular engulfment of large particles including other cells
Entosis	One cell invading into another cell
Cell cannibalism	One cell inside another (also called Emperipolesis)
Apoptosis	Cell degradation by caspase activation and/or Bcl-2 homolog
Autophagy	Cell degradation by autophagosomes
Efferocytosis	Phagocytosis of an apoptotic cell
Programmed cell clearance	Phagocytosis of an apoptotic cell
Programmed cell removal	Phagocytosis of dead, dying or viable cells
Forms of cell death	
Phagoptosis	Cell death by phagocytosis
Apoptotic cell death	Cell death by apoptosis
Autophagic cell death	Cell death by autophagy
Necrosis	Cell death by rupture of plasma membrane
Entotic cell death	Cell death by entosis
Phagocytosis-induced cell death	Cell death of a phagocyte resulting from its phagocytosis of another cell

being invaded by) host neutrophils, lymphocytes and erythrocytes, known as 'xeno-cannibalism' [89-91]. Cannibalism has been described in bladder, breast and lung cancer, and is related with the aggressiveness of the malignancy [88]. Overlap between the meaning of terms "cell cannibalism" and "phagoptosis" is uncertain, because in cell cannibalism it is unclear whether: i) one cell entered the other by phagocytosis, entosis or some other means, ii) the outside, or inside, or neither cell will die, and iii) if the inside cell dies, whether blocking phagocytosis would prevent cell death. Recently, entosis and cell cannibalism were shown to share mechanisms, which may make them synonymous in some cases [92]. Whether cell cannibalism can result in cell death by phagoptosis in other cases is unclear, but the terms mean different things.

The means by which a phagocytosed cell dies after engulfment is not always clear and is likely to vary in different models, but in general engulfment triggers the phagocyte NADPH oxidase (PHOX) to produce superoxide and hydrogen peroxide, which go on to produce more potent oxidants within the phagosome, and fusion with lysosomes delivers cytotoxic proteases and lipases in a very acidic environment; and this is the presumed means by which cells are killed after engulfment [62,93,94]. Engulfed/invading cells may be triggered to undergo apoptosis in specific conditions, for example immune killer cells within cancer cells may undergo apoptosis as a result of granzyme B reuptake, a process termed 'emperitosis' [95].

CONCLUSION

In sum, phagoptosis is an operationally unique mode of non cell-autonomous cell death that may play crucial roles in physiological homeostasis, in particular of leukocytes. However, too little or too much phagoptosis may contribute to the pathology of diseases such as cancer and neurodegeneration.

CONFLICT OF INTEREST

The authors confirm that this article content has no conflict of interest.

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