free to move into the nucleus and switch on the expression of specific genes. The T cell antigen receptor (TCR) signaling pathway activates the IKK complex through two caspase-recruitment domain (CARD)–containing adaptor molecules, CARMA1 and BCL10. These adaptor proteins do not directly activate IKK, which implies that other molecules must be involved. As the two new studies show, MPC is one of these molecules (*3*, *4*).

Lymphomas can arise outside of the lymph nodes in the intestinal mucosa–associated lymphoid tissue (MALT) and are often associated with chromosomal translocations. One gene characteristically found at the t(11;18)(q21;q21) translocation breakpoint in these tumors encodes MPC. This finding suggests that MPC is involved in signaling pathways that promote cell proliferation or survival, especially in lymphocytes (*7*). The MPC protein contains a caspase-like domain, two immunoglobulin-like domains, and a death domain. Superficially, these domains hint at a role in programmed cell death, but previous work has shown that MPC binds to BCL10, a CARD-containing adaptor protein that mediates TCR-induced activation of NF-κB. In addition, the MALT lymphoma translocations generate a chimeric protein containing the amino-terminal portion of c-IAP2 and the carboxyl-terminal caspase-like domain of MPC that activates NF-κB, promoting tumor formation.

Using mice engineered to lack the gene encoding MPC, the Dixit, and Mak groups discovered that MPC is crucial for regulating NF-κB activity prompted by antigen receptor signaling in T and B cells. What is most notable in the MPC-deficient mice is that the abnormalities are highly selective. This finding indicates that MPC subserves only certain signals relayed through NF-κB. MPC-deficient mice are healthy during development and fertile, but manifest abnormalities in their lymphoid tissues. There are alterations in early thymocyte subpopulations and major deficits in marginal zone B cells as well as in $CD5^+$ B1 cells. Although these mice exhibit abundant $CD4^+$ and $CD8^+$ T lymphocytes, the selective nature of the abnormalities in MPC-deficient animals only becomes clear when the functions of mature lymphocytes are analyzed. Ruefli-Brasse, Dixit, and colleagues (*3*) found defective antigen receptor signaling in both B and T cells in their mice. Early tyrosine phosphorylation events were normal, but the induction of cytokines, lymphocyte proliferation, and NF-κB activation were markedly reduced. By contrast, Ruland, Mak, and co-workers (*4*) found a defect in TCR-mediated responses but little impairment in B cell responses to antigen or lipopolysaccharide. The decrease in $CD5^+$ B cells in the Ruland *et al*. mice, also seen in other mice with defective B cell receptor signaling, suggests that further examination of B cells might reveal subtle abnormalities. Both groups found abnormal immune responses and other defects in their MPC-deficient mice that resemble those in animals lacking BCL10 or CARMA1. However, MPC-deficient animals did not show any defects in the activation of NF-κB by tumor necrosis factor (TNF)–α or interleukin-1. Finally, transfection experiments showed that BCL10 operates upstream of MPC, because MPC (when fused to c-IAP2) could induce NF-κB in the absence of BCL10 but the reverse was not true. Hence, MPC may be the linchpin linking BCL10 to the IKK signaling module. Thus, MPC is selectively important for NF-κB activation through antigen receptor signaling but not cytokine signaling.

It is intriguing that MPC harbors both a caspase-like domain and a death domain. Caspases are evolutionarily conserved proteases that are key players in apoptosis. Death domains link pro-apoptotic members of the TNF receptor superfamily to caspases through adaptor proteins. CARD-containing proteins, c-IAP2, and death domains found in MPC potentially connect with several different apoptotic pathways. Although MPC has no known proteolytic activity, apoptotic signaling pathways should be carefully examined in MPC-deficient mice. However, recent evidence that caspases play key roles in cellular activation suggests that the caspase-like domain of MPC could promote activation pathways and have nothing to do with apoptosis (*8*). Domain sharing by components of lymphocyte activation/proliferation and apoptosis signaling pathways may be an evolutionary convenience, or may be a way to coregulate these processes during cell proliferation. The MPC-deficient mice offer a means to explore this question.

NF-κB occupies a central nexus in the regulation of immune response signaling. The two new studies elucidate how antigen receptors communicate with nuclear genes through NF-κB. However, key questions remain: What are the specific functions of the immunoglobulin, death, and caspase-like domains of MPC? What is the stoichiometry of the complete signaling complex involving MPC, and how does it trigger IKK activity? Are the alterations in B cell development strictly secondary to defective antigen receptor signaling? What lessons can be learned by investigating the involvement of translocated MPC in tumor formation? Finally, the MPC-deficient mice generated by the Dixit and Mak groups may unveil new drug targets for treating cancers of the immune system.

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CELL BIOLOGY

Eat Me or Die

John Savill, Chris Gregory, Chris Haslett

Apoptosis is a physiological program of cell suicide that directs engulfment and safe destruction of cell corpses by healthy neighboring cells or professional phagocytic scavengers such as macrophages (*1*). Cells dying by apoptosis provide molecular instructions for their own funeral, sometimes releasing "come hither" signals that summon scavenger cells (*2*). These phagocytes then recognize "eat me" flags, such as phosphatidylserine (PS)—a phospholipid normally limited to the inner leaflet of the plasma membrane bilayer—that is displayed prominently on the surface of dying cells. A receptor (PSR) on macrophages that recognizes PS (*3*) then orchestrates compliance with the last line of the cellular suicide note: "Don't get angry when you dispose of me." This triggers suppressive pathways—such as the release of transforming growth factor–β1 (TGF-β1)—that prevent phagocytes from mounting a proinflammatory response to the dying cells (*1*). Two new studies in this issue using mice (*4*) and worms (*5*) lacking PSR provide important insights into how PSR governs the clearance of apoptotic cells. Furthermore, one of these studies (*4*) reveals a sinister postscript to the funeral invitation—"Eat me or die."

The PSR is one of a number of receptors on phagocytic cells that have been implicated in the uptake of dying cells. In addition, there are a variety of "bridging" molecules

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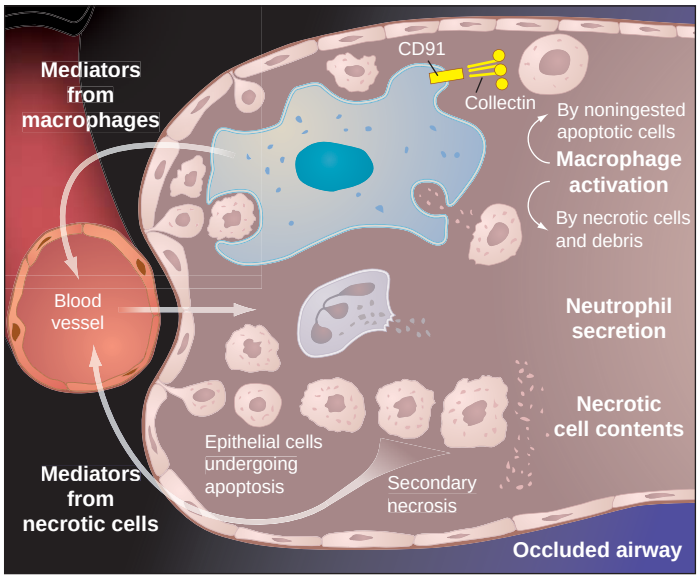
and "eat me" signals other than PS that are also important players in this process (*6*). The molecular structure of the PSR differs from that of other scavenger receptors recognizing "altered self," hinting that this receptor has additional functions. These may include oxoglutarate-dependent dioxygenase activity that is reminiscent of the peptidyl hydroxylases involved in oxygen sensing (*7, 8*). Even though it is expressed by many cell types, the PSR has proved elusive principally because the few anti-PSR antibodies available are not ideal for tracking its subcellular distribution. Indeed, some features of PSR's structure suggest that it may be a nuclear protein rather than a transmembrane receptor (*7, 8*). Reassuringly, a "knockout and rescue" experiment in the developing worm by Wang and colleagues on page 1563 of this issue (*5*) provides strong evidence that PSR is indeed the phagocytic receptor responsible for engulfment of dying cells.



Neonatal lung failure in PSR-deficient mice. During lung development in the mouse embryo, certain populations of epithelial and mesenchymal cells undergo apoptosis as part of the sculpting process that forms the airways. In mice lacking PSR, the corpses of apoptotic cells are not cleared efficiently and ultimately disintegrate. Such "secondary necrosis" releases cell contents that may directly or indirectly incite inflammation (via macrophage activation), resulting in the recruitment of neutrophils. In addition, binding of proteins called collectins by apoptotic cells may tether dying cells to the proinflammatory macrophage receptor CD91, further promoting the inflammatory cascade.

These investigators exploited a powerful and predictable model of embryonic cell deletion and clearance: that of the developing worm *Caenorhabditis elegans*. The worm has the added advantage of simplicity: Only a few proteins are involved in the uptake of dying cells, and most of those identified from mutant screens are intracellular molecules that induce cytoskeletal rearrangements in the cells engulfing corpses (*9*). The worm proteins CED-2, CED-5, CED-10, and CED-12 are structurally and functionally homologous to the mammalian signaling molecules CrkII, DOCK180, Rac guanosine triphosphatase, and ELMO, respectively. Worm embryos deficient in *psr-1* (a PSR homolog) exhibit a clear defect in the clearance of cell corpses. This defect could be rescued by overexpression of *ced-2*, *-5*, *-10*, or *-12* but not by overexpression of the *ced-6* or *-7* genes in a parallel pathway that is downstream of the scavenger receptor encoded by *ced-1*. Indeed, the clearance defect in the *psr-1* mutant was rescued not only by overexpression of wild-type *psr-1* but also by overexpression of human PSR. This finding confirms that PSR does participate in corpse engulfment, engaging a signaling pathway that is crucial for rearrangement of the cytoskeleton of phagocytic cells so that they can surround and "swallow" doomed corpses. It will be interesting to see whether PSR engages CrkII, DOCK180, and ELMO in mammalian phagocytes. The signaling pathway in which these proteins reside is also engaged by α_v integrins expressed by phagocytes. These surface receptors have been implicated in phagocytosis of apoptotic cells and TGF-β1–mediated suppression of macrophage responses.

Worms suffer few ill effects if their cell corpses are not engulfed. In contrast, the nonengulfment of cell corpses profoundly upsets higher organisms with their sophisticated innate immune systems, which present autoantigens derived from noningested apoptotic cells to the adaptive immune system. For example, mice lacking functional C1q—a component of the complement cascade that forms a bridge between macrophages and apoptotic cells (*10*)—develop multisystem autoimmune disease in response to demonstrably defective clearance of dying cells.

In contrast to the worm subjects of Wang *et al*., Li and colleagues (*4*) in their study of PSR-deficient mice on page 1560 of this issue find a dramatically different phenotype during fetal development. These animals exhibit fatal neonatal respiratory failure associated with a reduction in the number of airways formed and accumulation of noningested dying cells, cellular debris, and recruited inflammatory leukocytes in the developing lungs (see the figure). Perhaps the PSR is particularly important for clearing apoptotic cells from the developing lungs. This would be consistent with the lack of evidence of pathology in most other PSR-deficient tissues (although the authors did not undertake detailed searches for diminished apoptotic cell clearance). Indeed, different clearance mechanisms may predominate depending on the tissue—for example, C1q-deficient mice exhibit impaired clearance of apoptotic cells from the peritoneum and kidney but not from the skin. Whatever mechanisms underlie the lung phenotype of PSR-deficient mice, the new data establish PSR at the epicenter of developmental and inflammatory lung diseases.

Unexpectedly, 15% of PSR-deficient mice also exhibit hyperplastic brain malformations due to overproliferation of brain cells. This aberrant phenotype is also seen in mice lacking the Apaf-1–caspase-9–caspase-3 apoptosome pathway that normally mediates apoptotic deletion of excess cells from the developing brain. However, unlike these animals, affected brains in PSR-deficient mice show increased numbers of apoptotic cells and the recruitment of macrophages. These observations imply that in wild-type mice binding of dying cells to macrophages may trigger macrophage-mediated killing of apoptotic cells as reported in human (*11*) and worm, a phenomenon termed "grab and stab" (*9*). Alternatively, the brain malformations seen in PSR-deficient animals might reflect loss of a nonphagocytic PSR function, such as oxygen sensing.

PSR is now established as a key player in the clearance of dying cells during development (*4, 5*). Animals engineered to have only certain tissues deficient in PSR (conditional knockouts) are needed to generate viable adult mice in which the functions of PSR during inflammation and immunity can be tested. PSR and the downstream signaling pathways that it shares with other suppressive phagocytic receptors, such as the α_v integrins, may represent new therapeutic targets for treating common inflammatory diseases of the lung where leukocyte clearance by apoptosis has gone awry.

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