CellPress
OPEN ACCESS

Cell
Leading Edge

Review

Cell death in cancer

Marcus Conrad,[1,2,18,*] Andreas Strasser,[3,4,18,*] Philipp J. Jost,[5,6,7,8,18,*] Junying Yuan,[9,10,18,*] Feng Shao,[11,12,18,*] Peter Vandenabeele,[13,14,18,*] and Adam Wahida[1,5,13,14,15,16,17,18,*]

[1]Institute of Metabolism and Cell Death, Helmholtz Zentrum München, Neuherberg, Germany
[2]Translational Redox Biology, Natural School of Sciences, Technical University of Munich, Garching, Germany
[3]WEHI: Walter and Eliza Hall Institute of Medical Research, Parkville, VIC, Australia
[4]Department of Medical Biology, The University of Melbourne, Melbourne, VIC, Australia
[5]Clinical Division of Oncology, Department of Internal Medicine, Medical University of Graz, Graz, Austria
[6]Department of Internal Medicine III, Klinikum rechts der Isar, School of Medicine and Health, Technical University of Munich, München, Germany
[7]BioTechMed-Graz, Graz, Austria
[8]Austrian Comprehensive Cancer Network (ACCN), Vienna, Austria
[9]Interdisciplinary Research Center on Biology and Chemistry, Shanghai Institute of Organic Chemistry, Chinese Academy of Sciences, Shanghai, China
[10]Shanghai Key Laboratory of Aging Studies, Shanghai, China
[11]National Institute of Biological Sciences, Beijing, China
[12]New Cornerstone Science Laboratory, Shenzhen, China
[13]Molecular Signaling and Cell Death Unit, VIB-UGent Center for Inflammation Research, Flanders Institute for Biotechnology, Ghent, Belgium
[14]Department of Biomedical Molecular Biology, Ghent University, Ghent, Belgium
[15]Krantz Family Center for Cancer Research, Department of Medicine, Massachusetts General Hospital Cancer Center, Boston, MA, USA
[16]Department of Medicine, Harvard Medical School, Boston, MA, USA
[17]Department of Molecular Metabolism, Harvard T.H. Chan School of Public Health, Boston, MA, USA
[18]These authors contributed equally
*Correspondence: marcus.conrad@helmholtz-munich.de (M.C.), strasser@wehi.edu.au (A.S.), philipp.jost@medunigraz.at (P.J.J.), junying_yuan@sioc.ac.cn (J.Y.), shaofeng@nibs.ac.cn (F.S.), peter.vandenabeele@irc.vib-ugent.be (P.V.), awahida@mgh.harvard.edu (A.W.)
https://doi.org/10.1016/j.cell.2026.03.024

SUMMARY

"Evasion of cell death" is a hallmark of cancer, enabling transformed cells to withstand oncogenic and therapeutic stress. Restoring cancer cell death is an appealing strategy but requires a deep understanding of cell death programs. Over the past two decades, the cell death field has expanded from apoptosis to include necroptosis, pyroptosis, ferroptosis, and other emerging programs, reshaping cancer biology and revealing therapeutic opportunities. While apoptosis remains the primary radiation- and chemotherapy-induced cell death program, non-apoptotic programs can drive inflammatory responses and orchestrate the interplay among tumor, stroma, and immune components, influencing immunotherapy outcomes. Ferroptosis, an iron-dependent, lipid peroxidation-driven cell death modality, lacks a canonical induction signal and arises from perturbations in lipid, iron, and redox metabolism. This review presents a unified framework for understanding the roles of major cell death programs in cancer development, progression, and treatment response, as well as addressing resistance to cancer cell death and immune suppression.

"Our bodies are made of cells that live, and just as surely, of cells that must die." –S. Brenner

INTRODUCTION

Brenner's quote captures a tantalizing view of biology, in that life and death seem to be inseparably intertwined. Since the emergence of the earliest multicellular organisms, cellular demise has occurred either as a passive "accident" or, as is more amenable to therapeutic intervention, through a genetically encoded, orchestrated, and regulated process, which is key to sculpting tissues, preserving homeostasis, or defending the host against cancer and infectious agents.[1–3] As the German pathologist Rudolf Virchow famously argued in the 19th century, pathology is fundamentally cellular ("*omnis cellula e cellula*"), with disease arising from changes in cells that cause imbalances between proliferation and death.[4] Later, Elie Metchnikoff emphasized that phagocytic clearance is as critical as the act of dying itself, embedding cell death within a broader homeostatic cycle.[5]

These foundational insights from the 19th century echo more contemporary oncological concepts. In 2000, Hanahan and Weinberg introduced "evasion of apoptosis" as one of the original "hallmarks of cancer,"[6–9] later expanding its scope to "evasion of cell death" at large. Over the past quarter-century, advances in cell death biology have unveiled a diverse range of regulated cell death (RCD) modalities. It is now clear that apoptosis alone cannot account for the full complexity of cell

death in tumor progression, therapy response, or resistance. More recently discovered cell death processes, such as necroptosis, pyroptosis, or ferroptosis, have begun to reshape the original view of apoptosis as the sole targetable cell death modality, revealing a rich trove of potentially first-in-class therapeutics[10,11] (Box 1).

Necroptosis and pyroptosis represent lytic, inflammatory forms of RCD, coupling cellular demise with the release of cytokines and damage-associated molecular patterns (DAMPs) that influence antigen (including tumor antigen) availability, dendritic cell (DC) activation, and responsiveness to immune checkpoint blockade (ICB).[26–28] By contrast, ferroptosis is a mechanistically distinct process: an iron-dependent cell death driven by phospholipid peroxidation that is actively repressed under physiological conditions and lacks a classical genetically encoded trigger.[29,30] Its deregulation intersects with other cancer hallmarks, including metabolic plasticity, oxidative stress adaptation, and immune evasion, with apoptosis-refractory malignant cells and drug-tolerant persister (DTP) cells often being vulnerable to this process.[31–33]

The failure of stressed cells (e.g., those experiencing oncogene activation) to die can contribute to tumorigenesis as does unchecked cell proliferation. Cells undergoing transformation and fully malignant cells routinely (and forcibly) circumvent RCD programs to withstand oncogene-driven stress or therapy-induced injury, respectively, but also to reshape their interactions with the tumor microenvironment (TME). A deep mechanistic understanding of the different cell death events in cancer holds the opportunity for novel, efficacious therapeutic concepts, as blueprinted by the success of targeting apoptotic cell death. For decades, apoptosis was considered the only RCD pathway, with intrinsic (mitochondrial, BAX/BAK-mediated) and extrinsic (death receptor-initiated) cascades forming the molecular backbone for the removal of cells during development and in tissue homeostasis after birth, as well as therapy-induced tumor control. Landmark discoveries, including the identification of *BCL-2* as an oncogene that blocks apoptosis and promotes lymphomagenesis,[34,35] firmly established that defective apoptosis drives tumorigenesis and therapy resistance. These insights ultimately led to translation into clinical practice through the development of BH3 mimetics, in particular the BCL-2 inhibitor venetoclax, which has shown remarkable outcomes in treating chronic lymphocytic leukemia (CLL) and acute myeloid leukemia (AML).[36,37] However, emerging discoveries underscore that "cell death" is not only mediated by a single pathway but also by a network of interwoven programs whose modulation influences a panoply of aspects ranging from tumor initiation, progression, immune surveillance, and therapy response.

In this commemorative review, we integrate classical and emerging perspectives on apoptosis, necroptosis, pyroptosis, and ferroptosis in cancer. We discuss their molecular regulation, their crosstalk with other hallmarks of cancer, and their therapeutic implications within a *systems oncology* framework. The processes of cell death in cancer encompass both cell-autonomous mechanisms that directly promote the elimination of nascent neoplastic cells as well as intercellular interactions that modulate the TME. Notably, the induction of immunogenic cell death (ICD) plays a critical role in curtailing immunological tolerance, thereby enhancing anti-tumor immunity.[38] Yet, therapeutic strategies designed to intensify cancer cell killing may also compromise immune cell viability and function, potentially undermining immune-mediated cancer control.[39] Moreover, enhanced immunogenicity and its associated inflammation may, under certain conditions, create a tumor-prone environment.[38] These dualities underscore the importance of developing approaches that selectively target cancer cells while preserving, or even enhancing, immune competence, thereby preventing tumorigenesis. Ultimately, we argue that broadening our conceptual horizon to fully embrace the full spectrum of RCD processes will deepen our understanding of tumor biology, and thereby help guide the development of a new, powerful therapeutic arsenal against malignant disease.

APOPTOSIS

Evasion of apoptotic cell death, for example, due to the overexpression of BCL-2 or its anti-apoptotic relatives (e.g., BCL-XL or MCL-1) was listed as one of the original hallmarks of cancer alongside unlimited (sustained) cell proliferation and metastasis.[6–9] Since then, several additional hallmarks of cancer, including deregulated metabolism, immune evasion, and tumor-promoting inflammation, have been recognized.[8] It is important to note that the proteins encoded by some oncogenes or tumor suppressors impact not only one but several hallmarks of cancer. Pertinent to RCD, overexpression of the transcription factor MYC causes abnormal cell proliferation but at the expense of increasing the propensity of these cells to undergo apoptosis.[40] Moreover, the tumor suppressor p53 can drive apoptosis, cell cycle arrest/senescence, coordination of DNA damage repair, and other cellular responses through the transcriptional regulation of ~500 direct and many more indirect target genes (reviewed in Kastenhuber and Lowe[41]). Thus, the hallmarks of cancer are not isolated from each other but interconnected with several of them impacted by the same oncogenic driver or tumor suppressor (Figure 1).

The link between cell death and cancer was based on discoveries that inhibiting apoptosis, originally achieved by overexpressing BCL-2, can promote the development of lymphoma[34,35] and render both normal and malignant cells resistant to the cytotoxic effects of diverse anti-cancer agents.[42] Given the identification of several additional cell death programs and the recognition that their regulation is interconnected at multiple levels (see Box 2 and the review in Bedoui et al.[43]), it is timely to consider the role of cell death overall, rather than focusing solely on apoptosis, in the development, progression, and response to therapy in cancer.

Apoptosis is the first form of programmed cell death identified. In normal physiology, apoptosis plays a critical role in the removal of damaged cells or those that are no longer needed during embryonic development and in tissue homeostasis after birth.[77,78] Morphologically, apoptosis is characterized by cellular shrinkage, chromatin condensation, membrane blebbing, and the efficient removal of dying cells by efferocytosis.[79] Biochemically, it is characterized by internucleosomal DNA fragmentation.[80] The programmed removal of unwanted cells by apoptosis *in vivo* usually does not trigger inflammation due to the rapid removal of dying cells by phagocytes, a process called efferocytosis (reviewed in

Box 1. Cell-type-specific and emerging cell death modalities

Beyond these relatively linear pathways, cell death may also arise from the disruption of essential cellular processes, a phenomenon often referred to as "cellular sabotage." Such perturbations activate integrated stress responses (ISRs) and can lead to distinct cell death modalities.[12] Processes including lipid and energy metabolism, redox balance, iron and copper homeostasis, autophagy, and DNA repair are interconnected within complex metabolic networks (Box 2). When these networks are disturbed, the outcome of an ISR may range from adaptive repair with ensuing cell survival to catastrophic failure and cell death. These responses are not uniform across a cell population; instead, they produce heterogeneous outcomes, with some cells successfully adapting while others succumb.

Specific metabolic disruptions give rise to unique forms of RCD. Interference with redox, lipid, and iron metabolism can lead to ferroptosis, oxytosis, and oxeiptosis. Mitochondrial copper imbalance triggers cuproptosis, while disruption of cysteine metabolism leads to disulfidptosis. Alterations in pH homeostasis cause alkaliptosis, DNA repair failure activates poly(ADP-ribose) polymerase-1 (PARP1)-mediated parthanatos, and lysosomal zinc dysregulation induces lysozincrosis. These modalities reflect metabolic imbalances and the inability to repair damage following cellular insults and ISR activation. Cancer cells, which rely heavily on metabolic plasticity, can be more vulnerable to such sabotage than normal healthy cells. Exploiting these metabolic vulnerabilities offers promising strategies to eliminate tumor cells that resist the traditional forms of RCD.

"Cuproptosis" is a form of metabolic cell death triggered by intracellular copper accumulation.[13] Mechanistically distinct from apoptosis, necroptosis, and ferroptosis, cuproptosis involves the direct binding of copper to lipoylated components of the tricarboxylic acid (TCA) cycle, resulting in protein aggregation, loss of iron-sulfur cluster containing proteins, and ultimately proteotoxic stress. Physiologically, cuproptosis links mitochondrial metabolism with redox homeostasis, although its role in development remains unclear. It may serve as a metabolic safeguard by eliminating hyperactive or metabolically stressed cells. While not directly implicated in genome integrity yet, its dependence on mitochondrial respiration suggests a surveillance mechanism in metabolically vulnerable cells. In cancer, cells with high TCA cycle activity (e.g., certain subsets of lung cancer) may be particularly sensitive to cuproptosis. Therapeutically, copper ionophores (e.g., elesclomol) are under investigation to exploit this vulnerability. Although TP53 does not directly regulate cuproptosis, TP53-driven metabolic shifts might indirectly affect susceptibility. RT can promote cuproptosis by increasing intracellular copper accumulation and combining RT with cuproptosis inducers overcomes tumor radioresistance, while sparing normal cells.[14] These findings position cuproptosis as a compelling therapeutic target to enhance the efficacy of RT in resistant cancers.

"NETosis," or cell death associated with neutrophil extracellular trap (NET) formation, is a specialized form of programmed cell death unique to neutrophils. It is characterized by chromatin decondensation and the extrusion of web-like structures composed of DNA, histones, and granule proteins.[15] Originally identified as an anti-microbial host defense mechanism, NETosis plays a crucial role in trapping and neutralizing pathogens during acute infection or inflammation. Beyond this, NETosis has gained increasing attention in cancer biology.[16] Excessive or dysregulated NET formation was reported to contribute to a pro-thrombotic and pro-metastatic TME.[17] NETs can facilitate the adhesion of circulating tumor cells, promote endothelial disruption, and support the formation of pre-metastatic niches.[16] Accordingly, elevated NET activity is frequently observed in patients with advanced malignancies, and this has been linked to poor prognosis and therapy resistance. Therapeutically targeting NETosis is being explored to mitigate cancer-associated thrombosis and limit metastatic spread.[16] Strategies under investigation include DNase-based degradation of extracellular traps and inhibitors of NETosis that block upstream neutrophil activation.[16] These approaches may offer novel adjuncts to immunotherapy and chemotherapy, particularly in tumors with prominent inflammatory or thrombotic components.

"Disulfidptosis" is a metabolic cell death modality that is triggered by the accumulation of abnormal disulfide bonds under conditions of glucose starvation.[18] Mechanistically, it is driven by excessive disulfide stress in actin cytoskeletal proteins, resulting in the collapse of the actin network and catastrophic loss of cell integrity. Although its discovery has garnered considerable interest, evidence connecting disulfidptosis to cancer remains scarce. Given that many tumors frequently encounter fluctuating nutrient and redox stress and often rely on altered cystine/cysteine metabolism, it is conceivable that disulfidptosis may be selectively activated in malignancies with high cystine uptake or defective redox buffering.[19] At present, however, no direct functional link between disulfidptosis and tumor initiation, progression, or sensitivity to therapy has been established. Its relevance to cancer biology, therefore, remains an open question.

"Alkaliptosis" is a form of metabolic cell death characterized by intracellular alkalinization, typically mediated by aberrant activation of the Na^+/H^+ exchanger 1 (NHE1), which disrupts cellular pH homeostasis.[20] Alkaliptosis does not involve caspase activation or lipid peroxidation. Instead, it results from sustained increases in intracellular pH that destabilize essential metabolic and signaling processes.[21] Its relevance to cancer is still being defined. Preclinical studies indicate that certain tumors may be sensitized to alkaliptosis through pharmacological agents or stress conditions that perturb pH regulation. However, definitive roles in tumor initiation, progression, or therapeutic response remain to be established. Given that dysregulated metabolism and altered pH gradients are hallmarks of many cancers, alkaliptosis could represent a context-specific vulnerability. However, its mechanistic and translational significance in oncology remains unclear.

"Oxeiptosis" is a non-inflammatory, caspase-independent form of metabolic cell death triggered by high levels of ROS. KEAP1 orchestrates a PGAM5-AIFM1 signaling pathway in which KEAP1 senses oxidative stress and triggers the release of apoptosis-inducing factor (AIFM1) from mitochondria, leading to chromatin condensation and cell death without eliciting a strong inflammatory response.[22] While oxidative stress is a central feature of cancer biology, the evidence for a role of oxeiptosis in tumor initiation, progression, or therapy response remains limited. Its proposed role is more to prevent excessive inflammation or tissue damage under oxidative stress than to act as a tumor-suppressive mechanism. Nonetheless, given that cancer cells depend on redox adaptations for survival and therapy resistance, oxeiptosis may intersect with oncogenic ROS metabolism, thereby potentially influencing tumor immune system interactions or responses to ROS-inducing therapies. However, the role of oxeiptosis in cancer remains speculative and warrants further investigation.

"Parthanatos" is a distinct form of metabolic cell death triggered by the overactivation of PARP1 in response to severe DNA damage and oxidative stress.[23] Excessive PARP1 activity causes massive poly(ADP-ribose) (PAR) accumulation, NAD^+/ATP depletion, and translocation of AIFM1 from mitochondria into the nucleus, where it induces large-scale DNA fragmentation.[23] Unlike apoptosis, parthanatos is caspase-independent but closely associated with genomic instability. In cancer, PARP1 is frequently upregulated, contributing to both enhanced DNA repair capacity and therapy resistance.[24] The induction of parthanatos has been implicated as a mechanism of action for certain chemotherapeutics and ionizing radiation, particularly under conditions of overwhelming DNA damage. Moreover, pharmacological PARP inhibitors, designed initially to exploit

(Continued on next page)

Box 1. *Continued*

synthetic lethality in BRCA-deficient tumors, may also modulate parthanatos depending on dose and context.[25] Thus, parthanatos appears to play a paradoxical role in oncology. While it can serve as a tumor-suppressive mechanism when activated in cells undergoing neoplastic transformation, chronic or misregulated activation of parthanatos may promote mutagenesis and tumor progression. Its therapeutic exploitation remains an emerging area, particularly in the context of regimens that induce DNA damage and PARP-inhibitor-based strategies.

Nagata[81]). However, when cells undergo apoptosis *in vitro* in the absence of phagocytes, they undergo a process called "secondary necrosis,"[79] which leads to the release of inflammatory mediators from the dying cells (reviewed in Newton et al.[10]). This can occur within an organism when a large number of cells die by apoptosis in a short period, overwhelming the capacity of the existing phagocytes. This pro-inflammatory secondary necrosis is thought to contribute to tumor lysis syndrome, which can be fatal in patients.[82,83] To identify targets for intervention in this complication during cancer therapy, it will be fascinating to explore whether RCD forms in addition to apoptosis can also contribute to tumor lysis syndrome. For instance, a point of debate concerns whether caspase-3-activated gasdermin E (GSDME) contributes to secondary necrosis, which occurs when apoptotic cells are not cleared by efferocytosis,[84] or instead drives a transition to pyroptosis without typical apoptotic morphology. Some studies have reported delayed apoptosis associated with secondary necrosis upon GSDME loss,[85] whereas others found no such effect.[86]

Apoptosis is executed through two distinct but ultimately converging pathways[87]: the death receptor-mediated (extrinsic) and the BAX/BAK-mediated, mitochondrial (intrinsic) cascades. The intrinsic apoptotic pathway is regulated by the BCL-2 protein family, characterized by the presence of one to four BCL-2 homology (BH) domains. The BCL-2 protein family comprises three subgroups: the anti-apoptotic members (BCL-2, BCL-XL, MCL-1, BCL-W, and A1/BFL1), the pro-apoptotic BH3-only proteins (e.g., BIM, PUMA, and BID), and the multi-BH domain effectors of apoptosis, BAX, BAK, (and BOK). BAX and BAK have largely overlapping functions in mediating mitochondrial outer membrane permeabilization (MOMP) (reviewed in Newton et al.[10] and Green and Reed[88]). In surviving cells, the activation of BAX and BAK is restrained by the anti-apoptotic BCL-2 proteins. Apoptosis is initiated when developmental cues, deprivation of growth factors or nutrients, or treatment with cytotoxic agents (e.g., anti-cancer drugs) increase the levels of pro-apoptotic BH3-only proteins mediated by transcriptional and/or post-transcriptional processes. The BH3-only proteins bind with very high affinity to the anti-apoptotic BCL-2 proteins, thereby liberating BAX and BAK from their restraint, allowing them to form dimers and multimers of dimers to cause MOMP. It has also been reported that certain BH3-only proteins can directly activate BAX and BAK (reviewed in Singh et al.[89]), but other studies using cell lines in which all BH3-only proteins have been removed showed that this "direct activation" is not required for apoptosis.[90] BAX/BAK-mediated MOMP causes the release of apoptogenic factors, including cytochrome *c* and SMAC/DIABLO, from the mitochondria into the cytoplasm.[91] This causes APAF-1-assisted auto-proteolytic activation of the initiator caspase-9, which then proteolytically activates the effector caspases-3, -7, (and -6) that cleave hundreds of cellular proteins, thereby orchestrating the ordered demolition of the dying cell (reviewed in Newton et al.[10] and Nagata[81]) (Figure 1).

Death receptor-induced apoptosis is initiated by the stimulation of so-called death receptors (FAS, TNFR1, and TRAILR1/2) by their cognate ligands (FASL, tumor necrosis factor [TNF], and TRAIL) (reviewed in Newton et al.[10]). This allows the intracellular death domains of these receptors to bind through homotypic interactions with death-domain-containing adapters (FADD and TRADD). This triggers homotypic death effector domain-mediated recruitment and proximity-induced auto-proteolytic activation of caspase-8 (in primates, also caspase-10) within a so-called "death-inducing signaling complex" (DISC).[92–94] This process is regulated by FLIP long and short proteins, which are structurally related to caspase-8 (and -10) but lack proteolytic activity. The initiator caspases-8 (and -10) proteolytically activate effector caspases (-3, -7, and [-6]). In so-called type 1 cells, mostly lymphoid ones, this direct pathway is sufficient for death receptor-induced apoptosis. In contrast, in type 2 cells (hepatocytes, epithelial cells, and fibroblasts), caspase-8 mediated the proteolytic activation of the pro-apoptotic BH3-only protein BID (forming tBID), thereby engaging an intrinsic apoptosis pathway amplification loop, which is needed for effective killing.[95–97]

Extensive MOMP, rather than activation of the caspases, constitutes the "point of no return" in apoptotic cell death signaling.[98] Accordingly, only the overexpression of anti-apoptotic BCL-2 proteins[35] or the absence of pro-apoptotic BCL-2 family members, such as BIM or PUMA,[99–101] accelerates c-MYC-overexpression-induced lymphoma development, whereas the absence of the initiator caspase, caspase-9, or its activator APAF-1, downstream of MOMP fails to do so.[102] Similarly, the overexpression of BCL-2[42] or its anti-apoptotic relatives, as well as the absence of pro-apoptotic BH3-only proteins, particularly BIM and PUMA[103,104] or BAX and BAK,[77] but not the absence of caspase-9 or APAF-1,[98] renders lymphoma cells, as well as non-transformed lymphoid cells, resistant to a broad range of anti-cancer agents. PUMA plays a critical role in the killing of cells by DNA damage-inducing anti-cancer drugs (e.g., etoposide or cyclophosphamide).[104] BIM contributes to this process and is also required for the induction of apoptosis by glucocorticoids[103] or the efficacy of inhibitors of oncogenic kinases, such as the BCR-ABL inhibitor imatinib.[105] Of note, some hematological malignancies depend on a single anti-apoptotic BCL-2 protein for survival, such as MCL-1 or BCL-2 in AML[106] and MCL-1 in c-MYC-driven B cell lymphoma.[107] These discoveries prompted the development of BH3 mimetic drugs inhibiting select anti-apoptotic BCL-2 proteins to induce apoptosis in malignant cells.[108,109] The BCL-2-specific inhibitor venetoclax is effective and widely approved for the treatment of CLL,[36] and AML,[37] the first of hopefully many clinically translated successes of cell death research yet to come.

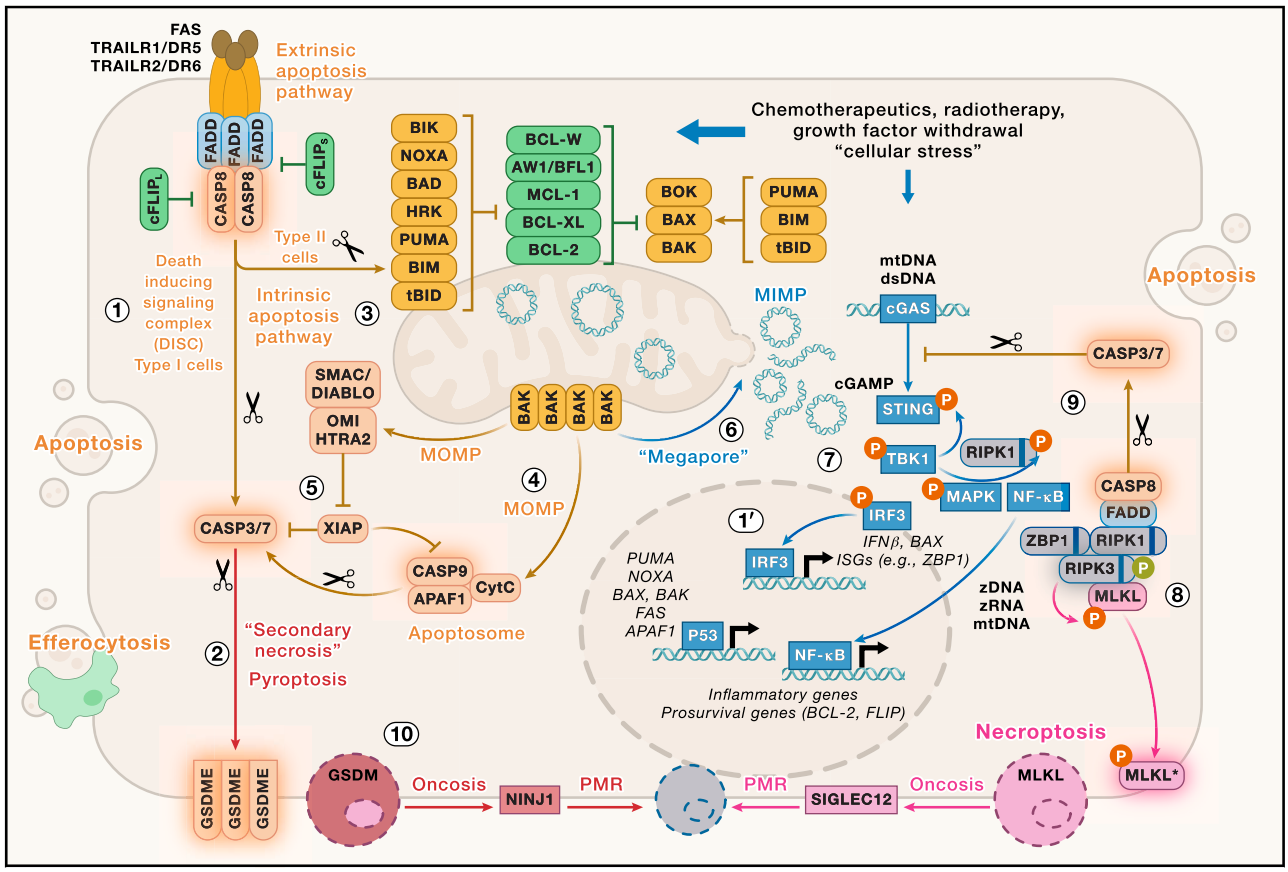

Figure 1. Extrinsic and intrinsic apoptotic pathways, and interaction with cGAS/STING pathways leading to apoptosis and necroptosis

Chemotherapeutics, RT, growth factor withdrawal, and general cellular stress conditions can activate both the extrinsic and intrinsic apoptosis pathways. (1) Binding of FASL or TRAIL to their respective death receptors (FAS, TRAILR1, or TRAILR2) initiates the formation of a DISC. In "type I" cells, the direct activation of caspase-3 and caspase-7 by caspase-8 is sufficient to induce apoptosis and does not require mitochondrial involvement through the intrinsic pathway. In contrast, "type II" cells require engagement of the mitochondrial pathway, initiated through caspase-8-mediated cleavage of BID into tBID, to undergo apoptosis. DISC formation is negatively regulated by $cFLIP_L$ (cytoplasmic FLICE-like inhibitory protein long isoform, a caspase-8-like protein lacking protease activity) and $cFLIP_S$ (a protein consisting only of death effector domains). (1′) Chemotherapeutics, RT, and "cellular stress" conditions can lead to the activation of p53, which induces gene products involved in the engagement of extrinsic and intrinsic apoptosis pathways. (2) In some cell types, caspase-3 and caspase-7 can also proteolytically activate GSDME, resulting in "secondary necrosis," which some authors classify as pyroptosis when this cell death modality is defined strictly by gasdermin activation. (3) The intrinsic apoptosis pathway is regulated by the balance between pro-apoptotic BH3-only BCL-2 family members (PUMA, BIM, BID, NOXA, BIK, BAD, and HRK), pro-survival anti-apoptotic BCL-2 family members (BCL-2, BCL-XL, MCL-1, A1/BFL-1, and BCL-W), and pro-apoptotic pore-forming BCL-2 family members (BAX, BAK, and BOK). BH3-only proteins bind and neutralize anti-apoptotic BCL-2 proteins, and some can also directly activate the pore-forming proteins BAX and BAK. (4) Once activated, BAX and BAK induce MOMP, allowing the release of cytochrome *c*. This promotes the formation of the apoptosome (cyto *c*/APAF-1/caspase-9), which subsequently activates caspase-3 and -7. Caspase-3/-7 activity results in apoptotic features like inter-nucleosomal DNA fragmentation, phosphatidylserine exposure, and membrane blebbing, preparing the cell for efferocytosis. (5) Additional apoptogenic factors released during MOMP include SMAC/DIABLO and OMI/HtrA2. These proteins inactivate XIAP, an inhibitor of activated caspase-3, -7, and -9, thereby amplifying the apoptotic cascade. (6) Sustained activation ("firing") of BAX and BAK can induce megapore formation, causing herniation of the inner mitochondrial membrane (IMM) and leading to MIMP. This results in the release of mtDNA, which is sensed by the cGAS/STING nucleic acid receptor system. cGAS/STING signaling is normally restrained by inactivating cleavage mediated by caspase-3 and caspase-7. (7) cGAMP generated by DNA-bound cGAS activates STING. Activated STING recruits and activates TANK-binding kinase 1 (TBK1), which phosphorylates STING in a positive feedback loop. TBK1 also phosphorylates and activates IRF3 and recruits RIPK1, leading to activation of MAPK and NF-κB pathways and subsequent induction of type I IFNs, ISGs, and various pro-inflammatory and pro-survival genes. (8) One IRF3-regulated gene is ZBP1, a sensor for stress-induced Z-RNA, Z-DNA, and mtDNA. ZBP1 is one of four RHIM-domain-containing proteins (ZBP1, RIPK1, RIPK3, and TRIF) that recruit and activate RIPK3 through homotypic RHIM interactions, resulting in MLKL phosphorylation and necroptosis (see B for more details). RIPK1 functions differently in mice and humans with respect to ZBP1: in human cells, RIPK1 kinase activity is required for ZBP1-mediated necroptosis, whereas in mice RIPK1 acts as an inhibitory scaffold that competes with ZBP1 for RIPK3 recruitment and activation. (9) ZBP1 can also initiate RIPK1 scaffold-dependent apoptosis. Activated caspase-3 and -7 further restrain cGAS/STING signaling via proteolytic cleavage. (10) Secondary necrosis/pyroptosis and necroptosis both cause cellular swelling (oncosis), which results in the activation of NINJ1 or SIGLEC12, respectively, leading to plasma membrane rupture (PMR).

Considerations and open questions targeting apoptosis in cancer cells

Although it has been known for 35 years that inhibiting apoptosis can promote tumorigenesis and render malignant cells resistant to anti-cancer agents, several well-established facts from this research are not being widely debated, and many critical gaps in our knowledge remain: (1) preventing apoptosis is, on its own, not potently oncogenic. Only about 5% of transgenic

Box 2. Interconnectivity of cell death modalities and immunogenic cell death

As noted above, cancer cells frequently acquire resistance to specific cell death pathways through mechanisms, such as overexpression of anti-apoptotic BCL-2 proteins,[43] activation of anti-ferroptotic adaptive stress responses,[44–46] or silencing of pro-apoptotic[47] and pro-necroptotic factors.[39] Advances in sequencing technologies, including tumor profiling and analysis of circulating tumor DNA within extracellular vesicles,[48,49] offer new opportunities for improved prognosis and precision medicine. These approaches may enable treatment strategies that either sequentially target more than one cell death modality or exploit targeting alternative cell death-unrelated processes as resistance emerges. In parallel, innovations in nanotechnology-based drug delivery promise enhanced tumor-specific targeting, minimized systemic toxicity, and improved dosing regimens, ultimately improving therapeutic outcomes[50] (Figures 3 and 4).

Cancer cells evolve within a TME composed of innate and adaptive immune cells, as well as non-immune cell types, such as fibroblasts and endothelial cells.[51] Infiltrating myeloid cells, including monocytes, macrophages, DCs, and neutrophils, can adopt either immunostimulatory or immunosuppressive roles depending on the signals they receive.[52] Through these functions, these cell populations shape adaptive immune responses and influence the efficacy of cancer therapies, particularly ICB. Central to the interplay between cell death and cancer therapy is the concept of ICD.[39] In cancer, ICD typically arises from therapy-induced stress in malignant cells, which triggers a mixture of cell death modalities due to ISRs following incomplete adaptation.[53] Therapy-induced ICD within the TME is governed by three interconnected features: adjuvanticity, antigenicity, and the pro-inflammatory context. This triforce interaction determines whether dying cancer cells can effectively stimulate an anti-cancer immune response. Adjuvanticity refers to the release of DAMPs, chemokines, IFNs, and cytokines by stressed or dying cancer cells, which serve as alarm signals that activate innate immunity. Antigenicity involves the efficient presentation of TAAs by antigen-presenting cells (APCs), such as DCs, enabling the priming of tumor antigen-specific T cells. The pro-inflammatory context of the TME is determined by the balance of pro-inflammatory (e.g., TNF, IL-1β, IFN-γ, type 1 IFNs, and chemokines) or suppressive cytokines (e.g., IL-10 and transforming growth factor β [TGF-β]) following exposure to stressed and dying cells in the TME. Different cell death modalities vary in their capacity to elicit this triforce response.[54,55]

In the TME, the immunological outcome of cell death can be tolerogenic, inflammatory, or immunogenic. Tolerogenic cell death occurs when antigen-specific responses are suppressed despite DAMP release, such as TNF, cytokines, chemokines, and IFNs. This often involves the massive release of immunosuppressive mediators, such as TGF-β and prostaglandins, by cancer cells, monocytes, and fibroblasts, which skew DC function toward regulatory T cell activation and thus immune suppression.[56] How do the distinct cell death modalities and their combinations dictate anti-tumor immunity and determine the immunogenic efficacy? Cancer immunotherapy-associated cell death serves two critical purposes: (1) an efficient elimination of malignant cells while sparing immune cells, and (2) the induction of a durable anti-tumor immune response that prevents tumor immune evasion, relapse, and metastasis. Chemotherapy- and RT-induced cellular stress within the TME generates heterogeneous biological outcomes, including cell survival, stressed cells, and RCD.[57] These responses vary depending on acquired resistance mechanisms or the silencing of cell death genes in cancer cells, ultimately resulting in a mixture of cell-fate outcomes.[10] Importantly, stress responses are not only confined to malignant cells. They also occur in TME-resident innate and adaptive immune cells and non-immune stromal and endothelial cells. This mixture of cells creates a cell death-associated TME characterized by variable adjuvanticity and a dynamic balance between pro- and anti-inflammatory signals.[39]

Adjuvanticity and the pro-inflammatory signaling from dying cancer cells depend on the various processes that drive plasma membrane permeabilization (e.g., GSDM proteolysis, MLKL phosphorylation, unrestrained lipid peroxidation, and NINJ1-mediated plasma membrane lysis[63]). The mode of cell death in each cell type, including cancer cells, immune cells, and non-immune cells, influences its adjuvanticity and inflammatory potential. For example, apoptosis in cancer cells becomes immunogenic only when accompanied by NF-κB-driven cytokine and chemokine production. ADAR1 inhibitors[58] and cGAS-STING pathway agonists[59] can enhance this immunogenicity by promoting IFN release. Pyroptosis of macrophages releases IL-1β, a potent pro-inflammatory mediator. Single-cell profiling of $CD14^+$ monocytes from patients with cryopyrin-associated periodic syndrome confirmed that lytic cell death drives pathological IL-1β secretion, redefining IL-1β as a DAMP predominantly produced by dying myeloid cells.[60]

Recently, a final cataclysmic event, viz., PMR, has been described as a kind of apotheosis of necrotic cell death. PMR is mediated by NINJ1 in secondary necrosis of apoptotic cells, pyroptosis, and perhaps ferroptosis[11,61,62,63,64,65] and SIGLEC12 in necroptosis.[66] Both molecules share amphipathic α-helical motifs that insert into the plasma membrane under conditions of oncosis-induced increased membrane tension sensed by NINJ1 or by TMPRSS4-mediated proteolytic cleavage of SIGLEC12, both facilitating the release of multiple immunostimulatory DAMPs. The convergence of multiple cell death modalities on molecularly controlled PMR positions this event as a "last message in a bottle," amplifying immune signaling after cell death.

However, the release of DAMPs alone is not sufficient to elicit a productive T cell response. A universal requirement for ICD is antigenicity. Without tumor antigen presentation and recognition, ICD cannot occur. Necroptosis and pyroptosis exhibit the highest immunogenic potential, whereas apoptosis is less effective.[26,67] This difference may relate to LMP, a late-stage event in apoptosis, or necroptosis, preceding PMR and supporting antigen processing.[68] MLKL, the key necroptosis effector, also promotes LMP,[68] potentially explaining the superior immunogenicity of necroptotic cells compared with apoptotic cells.[69] Necrotic cell death is further associated with rapid actin depolymerization and the formation of F-actin-rich filopodia on necroptotic and pyroptotic corpses. These structures are recognized by DC-associated receptor C-type lectin-domain-containing 9A (CLEC9A, also called DNGR1) receptors on DCs.[70] This triggers phagosomal rupture and antigen release, enabling cross-presentation via the major histocompatibility complex (MHC) class I pathway in cDC1 cells. Consistent with this, necrotic cell death modalities elicit strong anti-cancer immune responses in prophylactic cancer vaccination models.[70,71] By contrast, ferroptotic cells fail to do this and even impair antigen cross-presentation by DCs.[302] Excessive lipid peroxidation, a unique hallmark of ferroptosis (and oxytosis) that is not observed during apoptosis, necroptosis, and pyroptosis, may interfere with MHC-I peptide loading in DCs that engulf ferroptotic cell corpses. This reduced antigen presentation may relate to the reported role of ferroptosis in wound healing, where limiting adaptive immune activation could be beneficial.

As discussed above, chemotherapeutic and radiotherapeutic interventions in cancer elicit heterogeneous cellular responses due to the coexistence of multiple cell types within the tumor, each exhibiting distinct susceptibilities to various cell death modalities. Even genetically identical cancer cells subjected to γ-radiation *in vitro* can activate diverse cell death programs, including apoptosis or ferroptosis, and may also undergo mitotic

(Continued on next page)

Box 2. *Continued*

catastrophe or senescence.[72–74] Interconnectivity between different cell death pathways and further cellular responses (e.g., cell senescence) contributes to the heterogeneous responses of different malignant cells within an ostensibly genetically identical population after chemotherapy *in vivo*.[75] This complexity generates a dynamic network of interactions within the TME, encompassing both immunostimulatory and immunosuppressive signals[71] between dying tumor cells, senescent tumor cells, immune cells, stromal cells, and endothelial cells. Beyond these network effects, the spatial localization and metabolic states of individual cells may influence therapeutic outcomes. These observations underscore the need for advanced strategies to monitor immunologic and metabolic states before, during, and after cancer therapy. The integration of nanotechnology-based diagnostic and therapeutic platforms, commonly referred to as theranostics,[76] may offer promising avenues for personalized, precision oncology by enabling the integrated assessment and modulation of the intricate interplay between different cell death modalities and the TME.

mice overexpressing BCL-2 in their lymphocytes[110] or mice reconstituted with a BAX/BAK double-deficient hematopoietic system[111] develop lymphoma over the course of 12 months. The tumor-promoting role of inhibiting apoptosis comes to the fore only when combined with potent oncogenic drivers, such as deregulated expression of c-MYC[35] or v-ABL.[112] This suggests that defects in apoptosis contribute to tumorigenesis by preventing the cell death triggered by stressors imposed by oncogene expression. Consistent with the notion that endogenously expressed anti-apoptotic BCL-2 proteins are limiting for oncogene-induced development of hematological cancers, the removal of even one allele of *Mcl-1* can massively delay c-MYC-driven lymphomagenesis.[113] (2) The role and potential of death receptor-induced apoptosis in the development and therapy of cancer have not yet been fully explored. Defects in death receptor-induced apoptosis, i.e., mutations in the gene encoding FAS/CD95, can predispose both mice and humans to the development of certain B lymphoid malignancies.[114,115] DNA-damaging anti-cancer drugs were reported to kill lymphoid cells through autocrine/paracrine FASL>FAS signaling,[116] but later work demonstrated that death receptor signaling overall is not required for the cytotoxic effects of such drugs.[117] Given that intrinsic apoptosis and death receptor-induced apoptosis signaling synergize in lymphocyte homeostasis (i.e., are independent),[87] it would appear that therapeutic regimens that activate both pathways may be highly effective in cancer therapy, provided they are tolerable. (3) Inhibition of apoptosis is only well established to promote the development of hematological cancers. There is only scant evidence that the overexpression of anti-apoptotic BCL-2 proteins or the absence of pro-apoptotic BCL-2 family members can cause cancer in non-hematopoietic cell types.[118] Notably, the overexpression of BCL-2 in hepatocytes failed to accelerate c-MYC-driven liver cancer.[119] *What is the reason for this?* Do non-hematopoietic cells basally express sufficient levels of anti-apoptotic BCL-2 proteins to resist the stresses experienced during oncogene-induced neoplastic transformation (i.e., there is no need for mutation-induced overexpression of anti-apoptotic BCL-2 proteins or loss of pro-apoptotic BCL-2 family members)? Consistent with this idea, most solid cancer cell lines are highly resistant to BH3-mimetic drugs that inhibit only a single anti-apoptotic BCL-2 protein; in most solid cancer-derived cell lines, both BCL-XL and MCL-1 need to be inhibited for the efficient induction of apoptosis. (4) It is also noteworthy that at least certain cell types can recover from "limited MOMP" and continue to live and function.[120] It is imperative to determine whether potentially oncogenic DNA lesions emanating from limited MOMP (reviewed in Häcker and Haimovici[121]) and/or pro-inflammatory cGAS/STING signaling driven by the release of mitochondrial DNA (mtDNA) into the cytoplasm[122] may drive tumorigenesis.[123] (5) Finally, even though blocking apoptosis is broadly accepted to promote tumorigenesis, it was found that aberrant apoptosis can also drive tumorigenesis. Preventing the apoptosis of differentiated hematopoietic cells in the low-dose γ-radiation-induced thymic T lymphoma model, for example, by genetic deletion of pro-apoptotic PUMA (critical for DNA damage-induced apoptosis driven by the tumor suppressor p53[104]) ablates tumorigenesis.[124,125] Moreover, intestinal epithelial cell-specific deletion of the *Mcl-1* gene leads to the formation of intestinal cancers (that have escaped *Mcl-1* gene deletion).[126] The explanation for these findings is that apoptotic death of large numbers of differentiated cells mobilizes stem/progenitor cells, a phenomenon known as apoptosis-induced proliferation (AiP). Their rapid proliferation, to meet the needs of the depleted tissue, entails the risk of acquiring lesions during DNA replication that may be oncogenic, thus driving tumorigenesis. This scenario may underlie the development of liver cancer driven by hepatitis virus infection and the so-called "secondary cancers" in patients who had undergone cytotoxic therapy for a previous cancer. Therefore, we must now carefully evaluate not only whether inhibition of the more recently described RCD processes can promote tumorigenesis but also, conversely, test whether their excess induction may also promote cancer development.

NECROPTOSIS

Necroptosis is a regulated necrotic cell death mediated by RIPK1, RIPK3, and mixed lineage kinase domain-like protein (MLKL).[127] Necroptosis was first discovered as TNF-mediated necrosis with caspase inhibition[128,129] and, therefore, was proposed to be a "backup" cell death pathway when death receptor-induced apoptosis is inhibited.[130] The execution of necroptosis in the TNF>TNFR1 signaling pathway involves the activation of RIPK1 kinase, which promotes its binding to RIPK3 to form a necrosome in a RHIM-domain-dependent manner. Activated RIPK3, in turn, mediates the phosphorylation of MLKL and the execution of necroptosis by disrupting the integrity of the plasma membrane.[131–136] The expression of another RHIM-containing protein, Z-DNA-binding protein 1 (ZBP1) (also known as DAI/DLM-1), but not RIPK1, is induced during tumor development and can interact with RIPK3 in a RHIM-dependent manner to promote tumor necrosis.[137] The discovery that RIPK3 expression is often epigenetically silenced in cancers by hypermethylation of its promoter[138] may indicate that RIPK3 and necroptosis

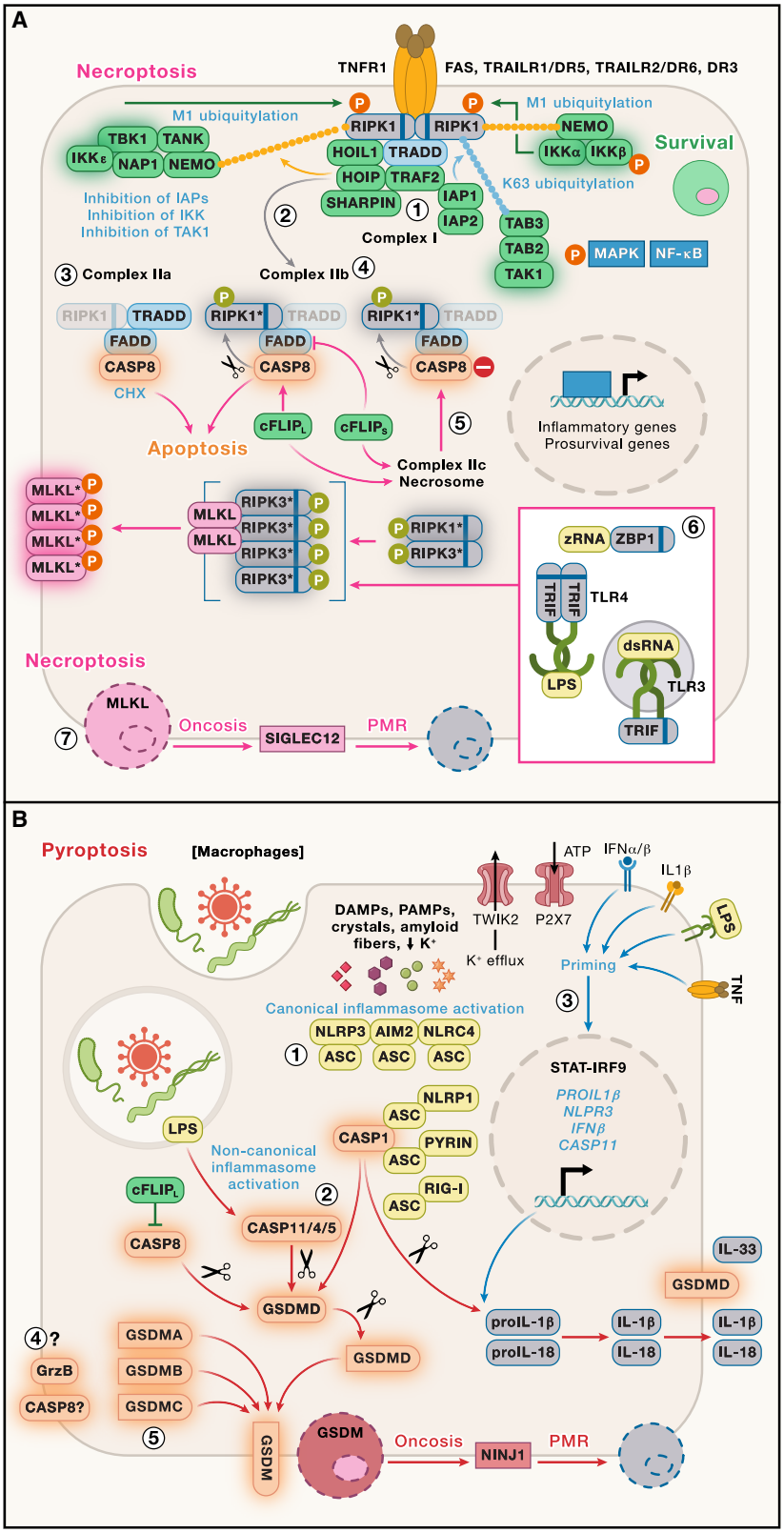

Figure 2. Induction of necroptosis and pyroptosis

(A) Similar to TNF (TNFR1), FASL (FAS), TRAIL (TRAILR1/DR5, TRAILR2/DR6), and TL1A (DR3) can induce necroptosis when caspase-8 is inhibited, and this form of cell death requires the kinase activities of RIPK1 and RIPK3. TNFR1-driven survival and cell death pathways include NF-κB activation, apoptosis, secondary necrosis, pyroptosis, and necroptosis. The outcome of TNFR1 engagement, cell survival (receptor-associated complex I) (1) or cell death (2) through apoptosis (complex IIa and complex IIb) (3) (4) or necroptosis (complex IIc or necrosome) (5) depends on the engagement of kinase-active RIPK1 (RIPK1*), the expression levels of RIPK3, and the activity or inactivity of caspase-8. (1) When TNF binds TNFR1, a plasma membrane-associated complex I (TNFR1-TRADD-RIPK1) forms, in which RIPK1 acts as a scaffold maintained in a pro-survival state through ubiquitylation and trans-phosphorylation (red "P"). K63-linked ubiquitylation by IAP1/2 creates a platform for recruiting the TAB1/TAB2/TAK1 complex, activating MAPK pathways that promote survival and inflammatory gene expression. M1-linked linear ubiquitylation by HOIL1/HOIP/SHARPIN (the LUBAC complex) recruits the NEMO/IKKα/IKKβ and TBK1 complexes (NEMO/TANK1/NAP1/TBK1/IKKε), supporting NF-κB-dependent survival and inflammation. The scaffold function of RIPK1 is regulated by multiple trans-phosphorylation events (red "P") mediated by IKK, TBK1, and related kinases. (2) Disruption of complex I survival signaling leads to the formation of a cytosolic complex II, which includes complex IIa, complex IIb, and the necrosome (sometimes referred to as complex IIc). (3) When translation is inhibited, such as with cycloheximide or defects in NF-κB signaling, complex IIa forms and induces apoptosis via FADD and caspase-8. (4) Pharmacological inhibition of IAPs, IKK, or TAK1 destabilizes RIPK1 in complex I and triggers the assembly of a cytotoxic complex II that requires RIPK1 kinase activity to induce apoptosis or necroptosis. In complex IIb, RIPK1 becomes activated and undergoes auto-phosphorylation (green "P"), enabling RIPK1* kinase-driven apoptosis through FADD and caspase-8. Activated caspase-8 can directly cleave and activate the effector caspases, caspase-3 and caspase-7, leading to the same terminal apoptotic events described for the intrinsic pathway (Figure 1). During *Yersinia* infection, TNF-mediated caspase-8 activation can also trigger pyroptosis involving GSDMD (see B). (5) In complex II, caspase-8 functions as a checkpoint controlling RIPK1* levels through proteolytic cleavage. When caspase-8 is inhibited, active RIPK1* accumulates. The RHIM domain in RIPK1* (blue stripe) promotes homotypic recruitment and auto-phosphorylation of RIPK3*. Activated RIPK3 forms large necrosome complexes, structurally reminiscent of β-amyloid assemblies, where MLKL interacts either with non-activated RIPK3 (MLKL-RIPK3 dimers in humans) or in a transient "kiss-and-run" mode (with monomeric MLKL in mice), ultimately resulting in phosphorylation of MLKL (MLKL*). Phosphorylation triggers MLKL's transition from a closed to an open conformation, driving its oligomerization (tetramers in humans and trimers in mice), membrane recruitment, insertion, and higher-order oligomer assembly. (6) Ligands for TLR3 and TLR4 recruit the RHIM-containing adaptor TRIF, which activates RIPK3* and initiates necrosome formation and MLKL activation. The nucleic acid sensor ZBP1 directly recruits and activates RIPK3 in response to Z-RNA, Z-DNA, and mtDNA. (7) MLKL* induces cellular swelling (oncosis) and PMR independently of NINJ1 but involving SIGLEC12.

(B) Pyroptosis is induced by DAMPs, PAMPs, crystals, amyloid fibers, and by decreases in intracellular potassium caused by efflux through the TWIK2 passive K^+ channel. There are two main classes of inflammasomes: canonical and non-canonical. Inflammatory caspases (caspase-1 and caspase-11 in mouse; and

(legend continued on next page)

could be involved as a tumor suppression mechanism during cancer development. Similar to inhibiting apoptosis, there might also be a selection pressure to favor cancer cells silencing *RIPK3* gene expression but not *RIPK1* gene expression.[138,139] The latter observation may indicate that RIPK3 and RIPK1 play opposing roles in cancer development (Figures 1 and 2A).

Necroptosis is highly inflammatory for multiple reasons.[26,140] Necrotic cell death can lead to the release of intracellular contents that act as DAMPs, triggering inflammasome activation and promoting inflammation. The activation of RIPK1 kinase can mediate the transcriptional induction of pro-inflammatory cytokines by promoting chromatin remodeling.[141] The preservation of RIPK1 expression in cancer cells is consistent with the notion that inflammation can play a pro-tumorigenic role in cancer development. Since RIPK1 activation is enhanced by RIPK3 deficiency,[141] the loss of RIPK3 in cancer cells may enhance RIPK1-mediated inflammation, driving tumorigenesis. Chronic activation of RIPK1 is linked to inflammatory diseases, and cancer cells can exploit RIPK1 to enhance their survival, TNF production, and immune evasion.[39] This is primarily due to RIPK1's scaffolding function, which activates nuclear factor κB (NF-κB) signaling and promotes the expression of NF-κB-driven anti-apoptotic genes, such as those encoding BCL-XL and cFLIP.[142] Moreover, sustained RIPK1-driven NF-κB activity can also trigger an immunosuppressive chemokine program, leading to reduced intra-tumoral infiltration of natural killer (NK) cells and $CD8^+$ T cells. This results in lower levels of TNF superfamily ligands in the TME, thereby contributing to resistance against ICB therapies. Additionally, RIPK1's scaffolding role can lead to sub-lethal activation of caspase-8, which then cleaves and inactivates key regulators of necroptosis, including RIPK3, CYLD (cylindromatosis lysine 63 deubiquitinase), and RIPK1 itself.[143]

Following necrosome assembly, RIPK3 phosphorylates MLKL (p-MLKL) within its pseudokinase domain (PsKD),[144] triggering MLKL's dissociation from RIPK3 and a structural transformation. This change leads to the dimerization of the MLKL C-terminal PsKDs, followed by tetramer formation. These tetramers are then trafficked to the plasma membrane, where the positively charged residues on the exposed N-terminal 4 helical bundle domain interact with phosphatidylinositol phosphates in the inner leaflet of the plasma membrane.[145–147] Inositolhexaphosphate (IP6) binding further propagates the active conformation of MLKL by displacing the auto-inhibitory brace region between the 4HBD and the PsKD.[148] Once a threshold concentration of MLKL accumulates at the plasma membrane, it is thought to form ion-conducting channels through a mechanism that remains incompletely understood. This may involve interactions with membrane proteins, such as Siglec-12, leading to osmotic imbalance, cell swelling, and ultimately plasma membrane rupture.[66,149,150] However, in contrast to the frequent silencing of the *RIPK3* gene in many cancer cell types,[139] *MLKL* gene silencing has not been reported in cancers to date.[151] This may indicate that MLKL has additional non-necroptotic functions conducive to tumorigenesis or that MLKL-mediated necroptosis does not exert a critical tumor-suppressive function. Interestingly, MLKL has been implicated in receptor internalization and degradation,[151] endosomal trafficking, extracellular vesicle formation associated with the ESCRT-III system,[152,153] autophagy,[154] lysosomal regulation,[68] and nuclear signaling,[155] in line with the presence of MLKL in the membranes of mitochondria, lysosomes, endosomes, endoplasmic reticulum, and in the nucleus.[156] However, the precise role of MLKL in cancer remains poorly understood, and it is still unclear whether these non-necroptotic functions contribute to tumor development or metastasis.[151] It must be noted that mice lacking MLKL develop normally, are viable after birth, and are not cancer-prone. This indicates that both the well-established role of MLKL in necroptotic cell killing and its additional reported roles independent of necroptosis are unlikely to be essential for preventing cancer (Figure 2A). Thus, the silencing of RIPK3 but not MLKL in cancers may reflect that loss of an unrelated necroptosis function of the RIPK3 kinase promotes tumorigenesis.

RIPK1 serves as a critical regulator of cell fate in both non-transformed and cancer cells, acting as a bifurcation point between cell survival and programmed cell death via apoptosis or necroptosis, respectively. Its scaffold function, independent of its kinase activity, promotes cell survival by activating mitogen-activated protein kinase (MAPK) and NF-κB signaling pathways, which drive the expression of pro-survival genes, thereby enabling cancer cells to evade cell death.[157] Moreover, RIPK1's scaffold function has been shown to inhibit necroptosis downstream of Toll-like receptors 3 and 4 (TLR3/4)[158] and the cytosolic DNA sensor ZBP1,[159,160] further reinforcing its role in maintaining cell survival and preventing cell death and inflammation. Targeting the scaffold function by a RIPK1-specific PROTAC degrader boosts the immunostimulatory and anti-tumor activity of radiotherapy (RT) and ICB while suppressing skin inflammation.[158]

Adenosine deaminase acting on RNA 1 (ADAR1) is another cancer-immunity-related modulator of necroptosis. ADAR1 performs A-to-I nucleotide editing by converting adenosine to inosine in double-stranded RNA (dsRNA). This editing process reduces the formation of immunogenic RNAs.[58] While ADAR1 plays a critical role in preventing severe inflammatory diseases,[58] ADAR1 expression has also emerged as a critical determinant for resistance to ICB.[161,162] Inhibition of ADAR1 results in

caspase-1, caspase-4, and caspase-5 in humans) eventually proteolytically activate GSDMD. (1) Canonical inflammasome-induced pyroptosis couples cytokine induction (e.g., pro-IL-1β) with cell death induction and GSDMD pore activation. Canonical inflammasome sensors—including NLRP3, AIM2, NLRC4, NLRP1, PYRIN, and RIG-I—recruit and activate procaspase-1 via the adaptor ASC. Caspase-1 then cleaves and activates GSDMD and pro-IL-1β, leading to pyroptosis. Potassium efflux through TWIK2 and ATP influx via P2X7 promote canonical inflammasome formation. (2) In the non-canonical pathway, cytosolic Gram-negative bacteria or LPS molecules directly activate caspase-11 (or caspase-4/-5 in humans). Caspase-11/-4/-5 subsequently cleave and activate GSDMD. During *Yersinia* infection, TNF-mediated caspase-8 activation can also induce pyroptosis through GSDMD activation. This process is negatively regulated by $cFLIP_L$. (3) Pro-IL-1β mRNA is transcriptionally induced by TLRs, TNFR1, IL-1R1, and IFNAR in a process known as "priming." GSDMD pores mediate the release of IL-1β, IL-18, and IL-33 (the latter does not require proteolytic activation and is even inhibited by caspase-1-mediated cleavage). (4) Other gasdermins are also implicated in pyroptosis: splice variants of GSDMA, granzyme B-mediated activation of GSDME, and caspase-8-mediated activation of GSDMC. (5) Pyroptosis is associated with cellular swelling (oncosis), which is sensed by NINJ1, resulting in PMR.

Z-RNA accumulation and the activation of the Z-RNA sensor ZBP1, which, in turn, promotes RIPK3-mediated necroptosis.[162,163–165] Thus, ZBP1/RIPK3-mediated necroptotic ICD might serve as a target to enhance the efficacy of cancer immunotherapies.[166,167]

Necroptosis has been described as a potent ICD modality, resulting from a tripartite interaction among antigenicity, adjuvanticity, and inflammation,[39,71,168] impacting the cancer-immunity cycle (Figure 3). A comparison between apoptotic and necroptotic cell death in the same cellular context led to the conclusion that the latter is superior in immunogenicity.[169–171] The enhanced tumor antigenicity of necroptosis might be caused by lysosomal membrane permeabilization (LMP) and concomitant release of cathepsins occurring during necroptosis,[69] possibly elicited by MLKL-mediated lysosomal rupture.[68] Necroptotic cells were shown to activate adaptive immunity by providing both antigens and inflammatory stimuli to DCs, which, in turn, stimulate $CD8^+$ T cells and anti-tumor immunity.[171,172] The induction of necroptosis, as a form of ICD, has been proposed as a strategy to promote anti-tumor immunity by providing both antigens and inflammatory signals to activate $CD8^+$ T cells.[39] Activation and maturation of DCs and consequent $CD8^+$ T cell cross-priming can be achieved by interacting with necroptotic cells in a RIPK1 and NF-κB signaling-dependent manner to initiate an immune response. The release of DAMPs by dying cells alone appears insufficient[172] (Figure 3).

Given its immunogenic nature, several pharmacological strategies are being developed to drive malignant cells into necroptosis. Caspase-8 serves as a critical checkpoint in regulating RIPK1-dependent necroptosis,[173] supporting the rationale for using caspase-8 inhibitors to promote immunogenic necroptosis.[173] Most therapeutic caspase inhibitors, such as Emricasan, target multiple caspases, thereby impacting processes in addition to necroptosis.[174] For example, extensive MOMP during apoptosis eventually results in mitochondrial inner membrane permeabilization (MIMP), leading to the release of mtDNA.[175] This mtDNA activates the cGAS-STING pathway, resulting in interferon (IFN)-β production and the induction of IFN-stimulated genes (ISGs), including the necroptosis-inducing sensor ZBP1.[176] Importantly, the cGAS-STING pathway is negatively regulated during apoptosis by executioner caspase-3, -7, (and -6) -mediated activation of the DNase CAD (caspase-activated DNase).[177,178] Therefore, the use of caspase inhibitors not only promotes mtDNA release by unleashing necroptosis but also lifts the caspase-dependent suppression of cGAS-STING signaling.[179] These insights underscore the potential of caspase inhibition to enhance ICD, thereby stimulating anti-tumor immunity.

Inhibitor of apoptosis proteins (IAPs) are E3 ligases that regulate the ubiquitylation of the RIPK1 scaffold within the TNFR1-associated complex I at the plasma membrane, promoting pro-survival and pro-inflammatory signaling.[180,181] SMAC mimetics were developed based on the IAP-binding motif (IBM) of the mitochondria-released pro-apoptotic protein SMAC/DIABLO.[182] These compounds bind to IAPs, triggering K48-linked autoubiquitylation and proteasomal degradation.[183] Combinations of SMAC mimetics (e.g., BV6, Birinapant, LCL-161, and Tolinapant) with broad-spectrum caspase inhibitors (e.g., zVAD-fmk and Emricasan) are widely used to deplete IAPs and activate RIPK1, resulting in RIPK3/MLKL-mediated necroptosis.[39,182] In preclinical studies, co-administration of Birinapant and Emricasan induced TNF/TNFR1-dependent necroptosis in AML cells, both *in vivo* in mice and in patient-derived samples.[184] The efficacy appears to depend on endogenous TNF production, either autocrine by the AML cells or paracrine from the TME.[184] This tumoricidal function of TNF recalls early clinical approaches during which TNF combined with melphalan was used for isolated limb perfusion in the treatment of soft-tissue sarcoma, aiming to avoid TNF-mediated systemic inflammatory response syndrome (SIRS).[185] Administration of SMAC mimetics, alone or in combination with caspase inhibitors, may harness this potent anti-cancer activity by leveraging elevated TNF levels in an inflammatory TME without causing unwanted TNF-induced systemic toxicity. Other members of the TNF family, such as TRAIL, have also been investigated for their cytotoxic potential in combination with SMAC mimetics (Birinapant), for example, in glioblastoma.[186] Furthermore, several chemotherapeutic agents, when combined with SMAC mimetics and caspase inhibitors, can drive cells toward necroptosis[39] (reviewed in Eggermont et al.[185]).

Necroptosis signaling also occurs downstream of the TLR3-TRIF-RIPK3 and ZBP1-RIPK3 axes, and combinations of agonists targeting these pathways are being explored therapeutically to induce ICD. Several dsRNA stabilizers (Hiltonol, BO-112, Ampligen, and Riboxxol) and mimetics (ARNAX) have been developed[39] (reviewed in Maelfait and Rehwinkel[187]). Similarly, curaxin CBL0137, a DNA-intercalating agent, has been shown to stabilize z-DNA formation, thereby inducing ZBP1-mediated necroptosis.[162] Direct ZBP1 agonists are currently under development.[187] Targeting ADAR1 with a small molecule (ZYS-1) enhances the anti-tumor effect of immunotherapy of prostate cancer.[188] However, the mechanism of action of ZYS-1 has been questioned as it did not reduce A-to-I base editing by recombinant ADAR1.[189] Other upstream agonists of ZBP1 include STING activators, which drive IFN-β-mediated induction of ZBP1 as well as lymphoma/leukemia cell intrinsic apoptosis through IFN-regulatory factor 3 (IRF3)-mediated induction of PUMA, NOXA, and BIM.[190] These agonists may also amplify the detection of z-nucleic acids generated by oxidative stress during chemotherapy.[191]

Finally, the scaffolding function of RIPK1, which mediates pro-survival signaling in many cancer cell types, can also be targeted to stimulate cell death. Recent studies have demonstrated that RIPK1-specific PROTACs synergize with immunostimulatory therapies, such as RT and ICB. Mechanistically, the depletion of RIPK1 sensitizes cancer cells to RT-induced cell death mediated by TNF and IFNs, thereby promoting ICD, triggering durable anti-tumor immune responses, and mitigating RIPK1-associated inflammation in the skin.[192] The latter is a profound side effect of many immunotherapies.[193] Collectively, these examples of direct and indirect therapeutic targeting of necroptosis illustrate a dual benefit: the direct killing of (apoptosis-resistant) tumor cells while also activating the patient's immune system to recognize and attack cancer.

Considerations and open questions targeting necroptosis in cancer cells

A primary mechanism underlying necroptosis resistance in cancer cells is the hypermethylation-mediated silencing of the

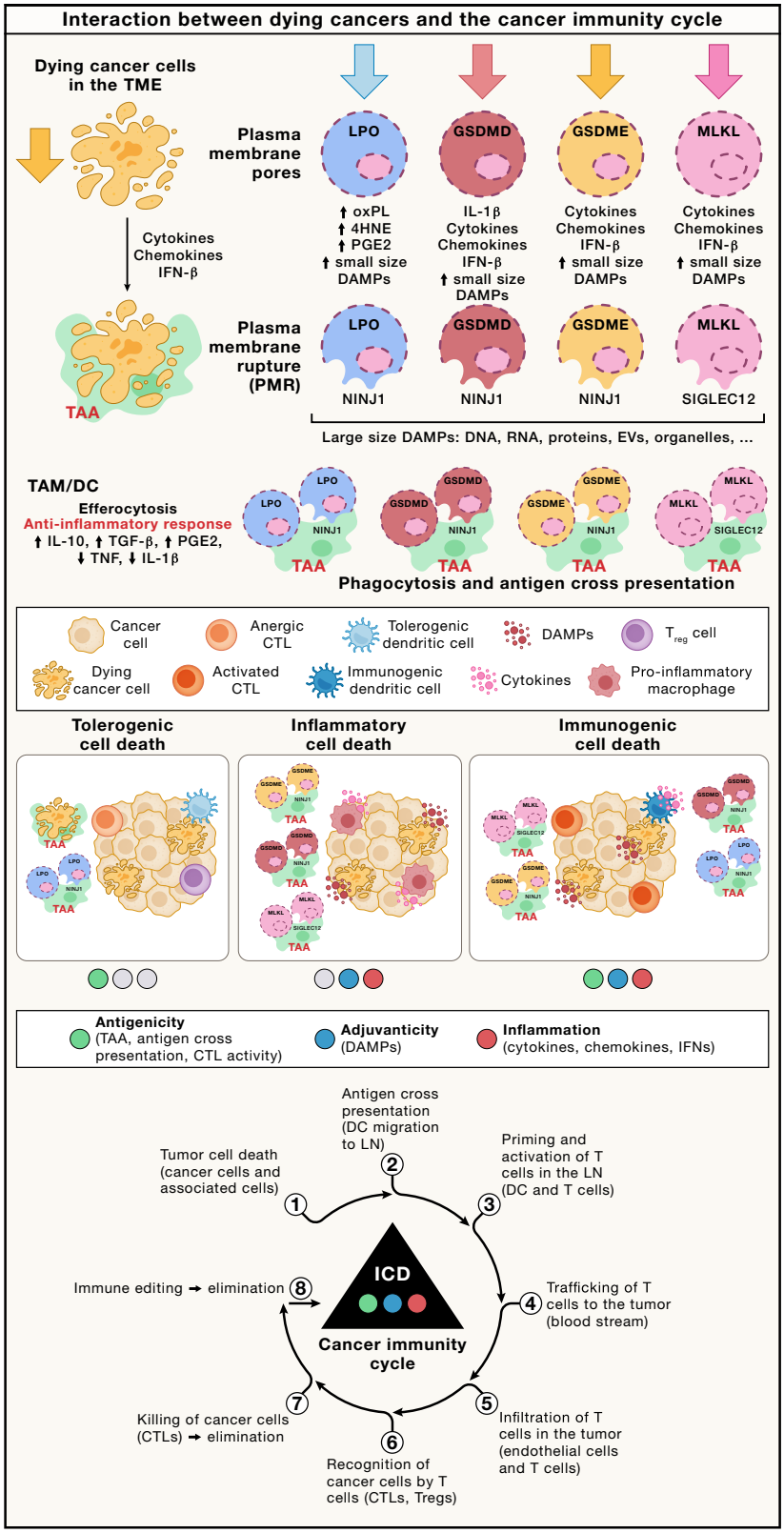

Figure 3. Interaction between cancer cell death and the cancer immunity cycle

Different cell death modalities exert differential effects on the TME. Apoptotic cancer cells are efficiently phagocytosed by TAMs and DCs (efferocytosis), thereby inducing an immunosuppressive cytokine profile in the phagocytosing cell. The secretome of ferroptotic dying cancer cells contains high levels of oxidized phospholipids (oxPLs) and PGE_2, which negatively affect antigen cross-presentation. Infammasome-driven pyroptosis is associated with high levels of IL-1β production, as well as with the release of chemokines, cytokines, IFNs, and small DAMPs, which are also released during secondary necrosis of apoptotic cells and necroptosis. Phagocytosis of dying cells by TAMs and DCs is required for the presentation of tumor-associated antigens (TAAs). The combination and balance of different cell death modalities and their interaction with the various cell types in the TME determine the outcome of cancer cell death as tolerogenic (high antigenicity, low adjuvanticity, and low inflammation), inflammatory (low antigenicity, high adjuvanticity, and high inflammation), or immunogenic (high antigenicity, high adjuvanticity, and inflammation in the cancer-immunity cycle to unleash a potent anti-tumor response. The * symbol represents an active enzyme (protease or kinase).

RIPK3 promoter, documented in several cancer cell lines and patient-derived samples.[139] This epigenetic modification prevents effective engagement of necroptosis as a backup cell death pathway when apoptosis is impaired. In contrast, *MLKL*, encoding the terminal executioner of necroptosis, generally does not exhibit widespread promoter hypermethylation in cancer, with a few exceptions, such as early-stage pancreatic adenocarcinoma,[194] colon carcinoma,[195] and cervical squamous carcinoma,[196] where the loss of MLKL expression correlates with a poor prognosis. This observation opens the possibility of exploring alternative activation mechanisms for MLKL independent of RIPK3 and of transcriptional IFN-mediated upregulation of MLKL during immune-stimulatory treatments, such as STING and TLR agonists, as well as RT.[72,197] MLKL reinforces the cGAS-STING pathway by releasing mtDNA upon necroptosis induction.[198] Further research is needed to clarify how RIPK3 is so broadly counterselected in cancer, and whether its loss is driven solely by its role in necroptosis or by other RIPK3-dependent processes, including apoptosis,[199,200] pyroptosis,[201] in which RIPK3 can also act as a signaling hub, and/or in driving the expression of certain cytokines and chemokines.[202]

Second, the caspase-8-mediated checkpoint requires the use of caspase inhibitors to sensitize cells to necroptotic death. Interestingly, in many cancers, caspase-8 is silenced, mutated, or functionally inhibited,[203] which may confer an intrinsic predisposition toward necroptosis, provided that RIPK3 and MLKL are expressed. Pharmacological inhibition of caspase-8 can enhance necroptosis sensitivity, as illustrated by studies using Emricasan.[184] To cope with such a situation of the absence of necroptosis mediators, the inducibility of necroptotic components by IFNs and inflammatory conditions[197] opens the possibility for sequential treatment strategies: first applying cellular stress or IFN-inducing therapies (e.g., chemotherapy, RT, STING agonists, and TLR agonists), followed by necroptosis-sensitizing regimens involving caspase inhibitors. Such an approach would require personalized monitoring of the tumor's initial and post-treatment status, including expression levels of RIPK3, MLKL, caspase-8, and ISGs.

Moreover, the caspase-8-mediated checkpoint represents a central regulatory node whose requirement varies across upstream signaling contexts. In TNF/TNFR1-driven necroptosis, caspase-8 functions as an obligate brake[160,204]: within cytosolic complex II, active caspase-8 continuously cleaves and inactivates RIPK1[160] and CYLD,[205] thereby dismantling nascent necrosome assembly. Consequently, pharmacological inhibition of caspase-8 using Emricasan in AML[184] is required to allow the accumulation of sufficient kinase-active RIPK1 and the subsequent activation of RIPK3/MLKL.

By contrast, in TLR3/4-TRIF-driven necroptosis, necroptotic signaling is triggered as soon as TRIF directly recruits RIPK3 via RHIM-domain homotypic interactions.[158,206] In this setting, RIPK1's scaffolding function (rather than caspase-8 per se) primarily restrains necroptosis, which is consistent with the demonstrated sensitizing effect of RIPK1-specific PROTAC degradation on TLR-driven necroptotic killing.[192] Similarly, ZBP1-RIPK3 signaling is similarly less constrained by caspase-8 activity, such as that induced by ADAR1 loss leading to interferonopathy[163] or by IFN-β-induced ZBP1 upregulation following STING activation,[162–164] which can trigger RIPK3-mediated necroptosis without obligate caspase-8 inhibition,[207,208] although caspase-8 retains the capacity to dampen signaling flux through this axis. Notably, both downstream TLR3/4 signaling and ZBP1 upregulation involve IFN-β, a cytokine that sensitizes cells to necroptosis by inducing MLKL expression,[197] whereas TNFR1-induced sensitization to necroptosis requires inhibition of caspase-8.

Collectively, this hierarchy of obligate dependence on caspase-8 inhibition during TNFR1 signaling, and of context-dependent modulation by caspase-8 during TLR/TRIF or ZBP1 activation, has direct therapeutic implications.[209] In cancers in which caspase-8 is silenced or functionally impaired, an intrinsic predisposition toward necroptosis may exist, pending whether RIPK3 and MLKL are expressed or can be induced. Sequential treatment strategies that first apply IFN-inducing therapies (e.g., chemo-RT, STING agonists, or TLR agonists) to upregulate necroptotic component expression, followed by caspase-8 inhibitor-based necroptosis sensitization, may exploit this pathway hierarchy. However, such approaches will require personalized monitoring of RIPK3, MLKL, caspase-8, and ISG expression levels.

Personalized monitoring predicting cell death modality based solely on transcriptional profiles is highly challenging because of the pleiotropy and interconnectivity of apoptotic and necroptotic pathways.[43,210] (Figure 4) A more demanding approach would involve immunohistochemical (IHC) detection of pathway-specific activation markers (e.g., caspase-3 and -7 proteolytic cleavage for apoptosis and p-MLKL for necroptosis). Notably, necroptosis should be inferred only when p-MLKL is present in the absence of caspase-3/-7 activation, as mixed phenotypes or sequential activation of apoptosis and necroptosis can occur under certain stress conditions.

A notable concern is how to prevent overshooting by necroptosis-based strategies, as conditions that promote necroptotic ICD can also trigger systemic inflammation and autoimmune reactions. One potential mitigation strategy may be the use of tumor-targeted delivery systems, such as nanoparticles, to minimize off-target effects on healthy tissues.[211] Another potential concern is tumor lysis syndrome, which can lead to SIRS and must be clinically managed.[212] In the long run, chronic exposure to necroptosis-inducing drugs could increase the risk of autoimmune complications, similar to the limitations observed with immune checkpoint inhibitors.[193] These considerations highlight the need for carefully controlled dosing, targeted delivery, and monitoring of systemic immune responses during necroptosis-based therapies.

Finally, as with any cancer therapy, the cellular stress associated with necroptosis-inducing strategies may inadvertently create a microenvironment that favors adaptation and progression of the existing tumor or lay the seeds for a secondary clonally unrelated cancer emerging years later. From the tumor's perspective, suppressing anti-tumor T cell immunity is critical for survival and metastasis.[213] The formation of a necrotic core within solid tumors is generally associated with a poor prognosis.[214] Paradoxically, necroptosis has been implicated in generating these necrotic regions under hypoxic and nutrient-deprived conditions. In mouse breast cancer models, the presence of a necrotic core correlates with increased metastasis.[214]

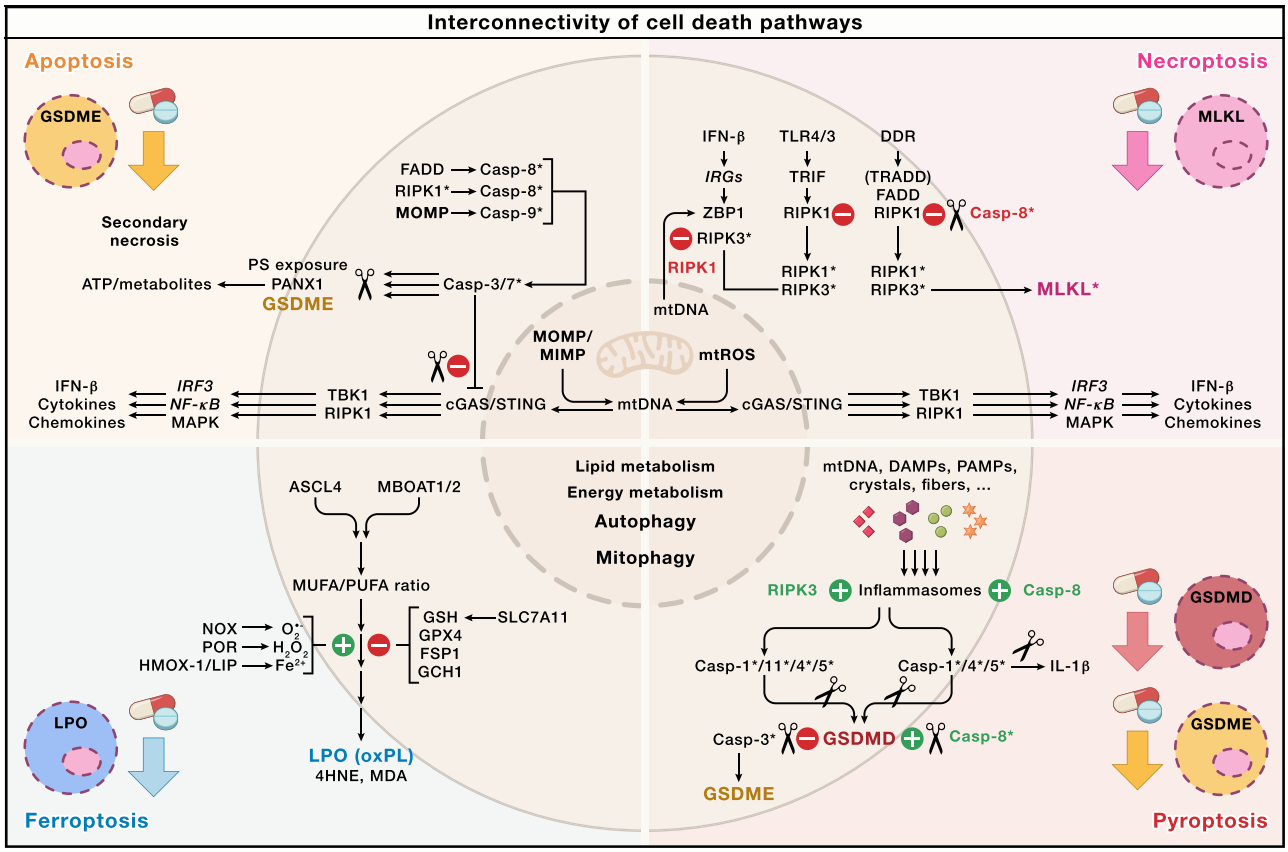

Figure 4. Interconnectivity of cell death pathways

Cell death-inducing drugs and radiotherapy engage distinct cell death pathways, clockwise, arranged in the four quarters of the circle: apoptosis, necroptosis, pyroptosis, and ferroptosis. The engagement of extrinsic apoptosis (FADD → caspase-8* and kinase-active RIPK1* → caspase-8*) and intrinsic apoptosis (MOMP → caspase-9*) results in the activation of the executioner caspases, caspase-3/-7*, leading to the proteolytic activation of substrates that mediate some typical features of apoptosis (PS exposure and active PANX1 channels) and secondary necrosis (GSDME pores) with profound intercellular consequences. Necroptosis is induced by engaging four RHIM-domain-containing proteins: the IFN-β-inducible nucleic acid sensor ZBP1, the TRIF adapter, RIPK1*, and RIPK3*. Recognition of z-shaped nucleic acids (e.g., mitDNA) by ZBP1, poly-IC, and LPS by TLR3 → TRIF and TLR4 → TRIF, respectively, and death domain receptor ligands by DDRs results in the recruitment and activation of RIPK3* (ZBP1) or RIPK1*/RIPK3 (TLRs and DDRs). Pyroptosis is executed by the proteolytic activation of GSDMD by inflammatory caspases following the engagement of the canonical (caspase-1*) and non-canonical inflammasome (caspase-4*/-5*/-11*). In the mouse, only caspase-1* (canonical inflammasome) can proteolytically activate pro-IL-1β, whereas in humans, both caspase-1* (canonical inflammasome) and caspase-4*/-5* (non-canonical inflammasome) generate active IL-1β. Ferroptosis is executed by the generation of LPO that is positively controlled by an increase in the PUFA/MUFA ratio (ACSL4), the generation of superoxide anion (NOX), hydrogen peroxide (POR), and free ferrous ions (HMOX-1 and LIP). The various cell death pathways are interconnected, and these interactions can affect the outcome of cell death in individual cancer cells or in a population of cancer cells. A common denominator of cell death-inducing drugs is their association with the generation of cellular stress (central circle in the scheme), which results in adaptive responses that alter lipid and energy metabolism, evoke MOMP/MIMP mtDNA-mediated activation of the cGAS/STING pathway, increase mtROS production, and induce autophagy and mitophagy. These stress-induced adaptive processes affect cell death versus cell survival (e.g., proliferation arrest and cell senescence) outcomes and the production of chemokines, cytokines, and IFN-β. During necroptosis, RIPK1* activation is kept in check by caspase-8*, while RIPK1 as a scaffold competes with RIPK3* recruitment and activation during ZBP1- and TLR3/4/TRIF-induced necroptosis. The executioner caspases, caspase-3*/-7* negatively regulate cGAS/STING by proteolytic activation of the DNase CAD removing cytosolic DNA, and they also proteolytically inactivate GSDMD. This dampens IFN-β responses and GSDMD-mediated pyroptosis.

Thus, therapeutic induction of necroptosis may exacerbate necrotic core formation and, within this niche, dampen $CD8^+$ T cell activity, potentially promoting the survival of circulating tumor cells. Consequently, engaging the necroptosis machinery could have unintended consequences, possibly accelerating tumorigenesis and metastasis rather than preventing them.

PYROPTOSIS

Pyroptosis is an inflammatory form of cell death characterized by cell swelling, rupture, and the release of numerous pro-inflammatory factors. Pyroptosis functions as an alarm to recruit and activate immune cells, bridging cell death with innate and adaptive immune responses. Recent studies show that pyroptosis plays multifaceted roles in cancer and anti-cancer immunity. Harnessing a pyroptotic program for the direct killing of tumor cells or the induction of effective anti-tumor immunity is emerging as a potential direct cancer cell-killing strategy and, possibly more promisingly, as a cancer immunotherapy (Figure 3).

Pyroptosis was first observed in macrophages upon exposure to bacterial toxins or infections.[215–217] Pyroptosis, named from

the Greek "pyro" (fire) and "ptosis" (falling),[218] was initially defined as a caspase-1-dependent, pro-inflammatory form of cell death,[219] but it was subsequently flooded and misconceptualized by the increased interest in apoptosis in the 1990s. Characterizations of inflammasome pathways established pyroptosis as the downstream innate effector mechanism.[220–222] A paradigm shift occurred in 2015 when gasdermin D (GSDMD) was identified as the executioner of pyroptosis.[223,224] The N-terminal domain of GSDMD or other gasdermins is sufficient to induce pyroptosis owing to their intrinsic pore-forming activity,[225–228] leading to the definition of pyroptosis as gasdermin-mediated programmed necrotic cell death.[229] Humans harbor six paralogous genes: *GSDMA*, *GSDMB*, *GSDMC*, *GSDMD*, *GSDME*, and *DFNB59* (also known as *PJVK*). Except for DFNB59,[230] a loss-of-function mutation associated with deafness, all gasdermins adopt a conserved two-domain architecture, consisting of a pore-forming domain and a C-terminal inhibitory domain.[223–226] Gasdermin activation is typically triggered by proteolytic cleavage within the interdomain linker. The thus-liberated N-terminal domain binds cell membranes and forms a large oligomeric pore.[231–233] The pores disrupt the osmotic potential, causing the cell to swell and lyse, and meanwhile it serves as a gate to release pro-inflammatory factors, including interleukin (IL)-1β as well as IL-18 in the case of macrophages[223,224,234] (Figure 2B).

In response to pathogens or danger signals,[235,236] inflammasome-activated caspase-1 (human and mouse) or caspases-4/-5 (human)/-11 (mouse) recognize an exosite in the C-terminal domain of GSDMD and cleave the interdomain linker.[237] GSDME is instead cleaved and activated by the executioner caspases-3 and -7 in response to "apoptotic" stimuli, switching non-inflammatory apoptosis to secondary necrosis, later also coined as pyroptosis.[238,239] Critical to such a switch is the presence or the expression level of GSDME in cells; indeed, numerous reports have demonstrated a high and functional expression of GSDME *in vivo* in various cell types, including skin, lung, and intestinal epithelia, as well as neurons. Several GSDMs are proteolyzed by granzymes, a family of serine proteases produced by cytotoxic T lymphocytes and NK cells.[240] Granzyme A directly processes GSDMB, whereas granzyme B can process GSDME directly or indirectly via the executioner caspases-3 and -7.[241,242] Thus, pyroptosis is an effector mechanism in cellular immunity if target cells express appropriate gasdermins. For GSDMC, recent studies suggest that mast cell-derived mast cell protease 1 (MCPT1) or lysosomal cathepsin S may mediate its proteolytic activation during helminth infection. In this context, GSDMC pores support IL-33 release from intestinal epithelial cells, thereby promoting anti-helminth type II immunity.[243,244] Caspase-8 has also been reported to activate human GSDMC.[245,246] However, it is yet to be established whether these proteases directly recognize and cleave GSDMC in the biochemical sense. No endogenous mammalian enzymes are known to cleave GSDMA. *Streptococcus pyogenes*-derived SpeB is the only reported protease capable of processing this gasdermin, thereby initiating a pyroptosis-mediated antibacterial defense.[247,248]

In anti-tumor immunity, the immune system deploys pyroptosis both directly, through cytotoxic effector cells, and indirectly, via inflammasome activation in innate immune cells.[242] In the former scenario, GSDMB expression is upregulated by IFN-γ and TNF-α released from tumor-infiltrating $CD8^+$ T cells and other immune cells, positively amplifying the tumoricidal effect.[241] Granzyme B activates caspase-3 most efficiently, which mediates pyroptotic killing of GSDME-positive cancer cells.[249] Perforin-induced plasma membrane lysis as well as death receptor ligand>death receptor (FASL>FAS)-induced apoptosis, was long thought to dominate the lymphocyte-mediated killing of target cells. However, gasdermin-mediated pyroptotic killing of cancer cells releasing various pro-inflammatory factors and danger signals to further stimulate anti-tumor immunity is now increasingly appreciated as an additional mechanism underlying the CTL- and NK-induced killing of malignant target cells.[242,250] Moreover, in the TME, DCs and macrophages sense tumor-derived signals via NLRP3 inflammasome and undergo GSDMD-mediated pore formation or pyroptosis, which amplifies the inflammation that facilitates the recruitment of cytotoxic lymphocytes.[251,252]

Pyroptosis in the TME has several immunostimulatory effects. Plasma membrane rupture of malignant cells allows the release of tumor antigens, facilitating antigen presentation by DCs and priming tumor-specific $CD8^+$ T cells by antigen cross-presentation.[253] Pyroptotic bursts promote the recruitment of additional immune cells through chemokines and danger signals, thereby converting "cold" into "hot" tumors that are more responsive to immune checkpoint inhibitor therapy.[52,54] Inflammasome activation of gasdermins in tumor-associated immune cells triggers the release of mature IL-1β and IL-18. IL-1β initiates and amplifies local inflammatory cascades, while IL-18 promotes Th1 T cell polarization and induces IFN-γ production by NK cells and T cells, collectively remodeling the TME toward tumor clearance.[254]

Pyroptosis imposes pressure on tumorigenesis, and evasion of pyroptosis is associated with the grade of malignancy. One major mechanism for the evasion of pyroptosis is transcriptional or epigenetic silencing of gasdermin genes. In various cancers, including breast, gastric, and colorectal tumors, the promoter of the *GSDME* gene is hypermethylated, leading to transcriptional repression and reduced GSDME protein expression.[255–258] The DNA methyltransferase inhibitor decitabine restores GSDME expression and can thereby sensitize certain cancer cells to undergo pyroptosis.[239] Therefore, decitabine enhances the efficacy of chemotherapy, in which pyroptosis-driven immune responses may play a key role.[259] *GSDMB* features alternative mRNA splicing, generating multiple isoforms with variable pore-forming capacities.[233,260–262] Among them, isoforms 3 and 4 retain robust pyroptotic potential, whereas isoforms 1 and 2 lack residues that are essential for pore formation and therefore cannot mediate pyroptosis. The relative expression of *GSDMB* isoforms, which varies widely across cancer cells, determines their susceptibility to granzyme A-mediated lymphocyte killing. Thus, malignant cells tend to express more of the pyroptosis-incompetent isoforms of GSDMB to evade anti-tumor immunity[233] (Figure 2B).

While pyroptosis generally inhibits tumor growth, sustained low-level pyroptotic signaling within the TME leads to chronic inflammation that may promote tumorigenesis, particularly

during the early stages of neoplasia.[254,263] For instance, IL-1β was reported to drive mesenchymal and epithelial carcinogenesis and metastasis by orchestrating a local inflammatory milieu in concert with other factors.[264] IL-1β reportedly promotes hematological malignancies, such as multiple myeloma,[265] and contributes to angiogenesis to support solid tumor progression.[266,267] Studies in mouse models indicate that IL-1β supports the development and immunosuppressive function of myeloid-derived suppressor cells (MDSCs), thus dampening effective anti-tumor immunity.[268–270] However, it must be noted that mature IL-1β release is only linked to canonical inflammasome-activated pyroptosis.[224]

Considerations and open questions targeting pyroptosis in cancer cells

Inducing pyroptosis in tumor cells or the TME may represent a promising strategy for cancer therapy. Studies in syngeneic mouse models show that enforced expression of GSDMB or GSDME in tumor cells can induce effective anti-tumor immunity.[241,249,271] Mechanistically, it was suggested that gasdermin activation in tumors promotes tumor cell phagocytosis by tumor-associated macrophages (TAMs) and thereby increases the abundance and functionality of tumor-infiltrating NK cells and $CD8^+$ T cells.[249] Moreover, treatment with inhibitors of mutant BRAF or MEK was reported to activate GSDME-dependent pyroptosis to boost anti-tumor immunity in GSDME-expressing melanoma.[272] It must, however, be noted that these agents have also been shown to induce apoptosis in melanoma cells through the induction of the BH3-only proteins BIM and PUMA.[273] Hence, pyroptosis may be secondary to apoptosis. Another study showed that pharmacological activation of the CARD8 inflammasome in human myeloid cells triggers GSDMD-mediated pyroptosis, offering a potential therapeutic avenue for AML.[274] Recently, a small-molecule agonist for GSDMD was identified, which modifies Cys191 to activate full-length GSDMD.[275] This compound effectively stimulates anti-tumor immunity in mice by inducing tumor cell pyroptosis.

However, some tumors lack sufficient gasdermin expression to trigger pyroptosis, especially since *GSDME* expression is silenced in many tumors.[276] Thus, nanoparticle-mediated delivery of active gasdermins is being developed to induce tumor cell pyroptosis. In a pioneering study, the cancer-imaging small-molecule probe phenylalanine trifluoroborate was exploited to release active GSDMA3 from the nanoparticle conjugates. This efficiently cleared tumors in mice through $CD8^+$ T cell-mediated anti-tumor immunity.[271] This provides a preclinical proof of concept for exploiting gasdermin activation in cancer immunotherapy. Similar nanomaterial-based approaches, as well as oncolytic virus-mediated delivery of activated gasdermins, are being developed.[277–280]

While pharmacologically activating gasdermins holds therapeutic promise, safety must be carefully evaluated. Aberrant activation of gasdermins in normal tissues can cause excessive inflammation and tissue damage.[239] Thus, targeted pyroptosis in tumors but not in normal tissues must be a critical factor in developing gasdermin activation-based cancer immunotherapies. Notably, granzyme B released from CAR-T cells was reported to activate GSDME in B lymphoma cells, contributing to cytokine release syndrome.[281] Thus, even the activation of gasdermins selectively in the malignant cells may cause severe toxicity that needs to be avoided or mitigated to make such proposed strategies tolerable.

Pyroptosis was initially regarded as an innate immune mechanism in myeloid cells. The identification of gasdermins has revolutionized our understanding of pyroptosis. The wide expression of gasdermins and their distinct activation mechanisms suggest the potential for pyroptosis in diverse cell types, including tumor cells. As in the cases of other forms of programmed cell death, there is a shortage of universal and reliable methods or tools to detect GSDM activation and pyroptosis *in vivo*. This direction of research efforts is of utmost importance for future studies to reveal and analyze the immunological functions of pyroptosis, including in the cases of cancer development and anti-cancer therapy. Despite the established anti-tumor effect, the precise mechanisms by which pyroptosis enhances anti-tumor immunity remain poorly defined. How pyroptosis promotes tumor-specific antigen presentation, modulates the TME, and orchestrates the recruitment of distinct immune cells requires further investigation. To be able to answer these questions, there is a need for systemic and accurate profiling the cytosolic substances and molecules in relevant cell types that can be released upon pore formation by specific GSDMs. Tumors often silence or downregulate gasdermin expression to evade immune clearance, but the mechanisms underlying this are not well understood. Various approaches have been developed to harness pyroptosis as a strategy to eliminate tumors and stimulate tumor-specific immune responses. However, therapeutic induction of pyroptosis requires the selective targeting of malignant cells while minimizing collateral damage to normal tissues. Overcoming these challenges is a prerequisite for successful clinical translation.

FERROPTOSIS

Ferroptosis is an RCD program characterized by iron-dependent, chain-propagating phospholipid peroxidation that ultimately leads to the loss of membrane integrity.[282] Unlike apoptosis, pyroptosis, or necroptosis, ferroptosis does not rely on canonical upstream "activation signals." Instead, it is continuously suppressed in living cells by metabolic surveillance systems that restrain the uncontrolled generation of peroxidized lipids.[283] When these safeguards are compromised by "cellular sabotage"[12,284]—as detailed below—for instance, by limiting cystine import, depleting glutathione (GSH), inhibiting glutathione peroxidase 4 (GPX4), or disabling parallel antioxidant circuits, such as the ferroptosis suppressor protein-1 (FSP1), cells trespass a lethal redox threshold and succumb to lethal lipid damage[285]—ferroptosis. This positions ferroptosis at the intersection of several hallmarks of cancer, including deregulated metabolism, oxidative stress adaptation, and immune evasion,[44,45] designating ferroptosis a metabolic cell death modality.[19,61] Notably, some of these features have proposed ferroptosis as a promising therapeutic opportunity in apoptosis-refractory malignancies[44,46,286] (Figure 5).

Ferroptosis is driven by the oxidative damage of polyunsaturated fatty acid (PUFA)-containing phospholipids, whereby iron

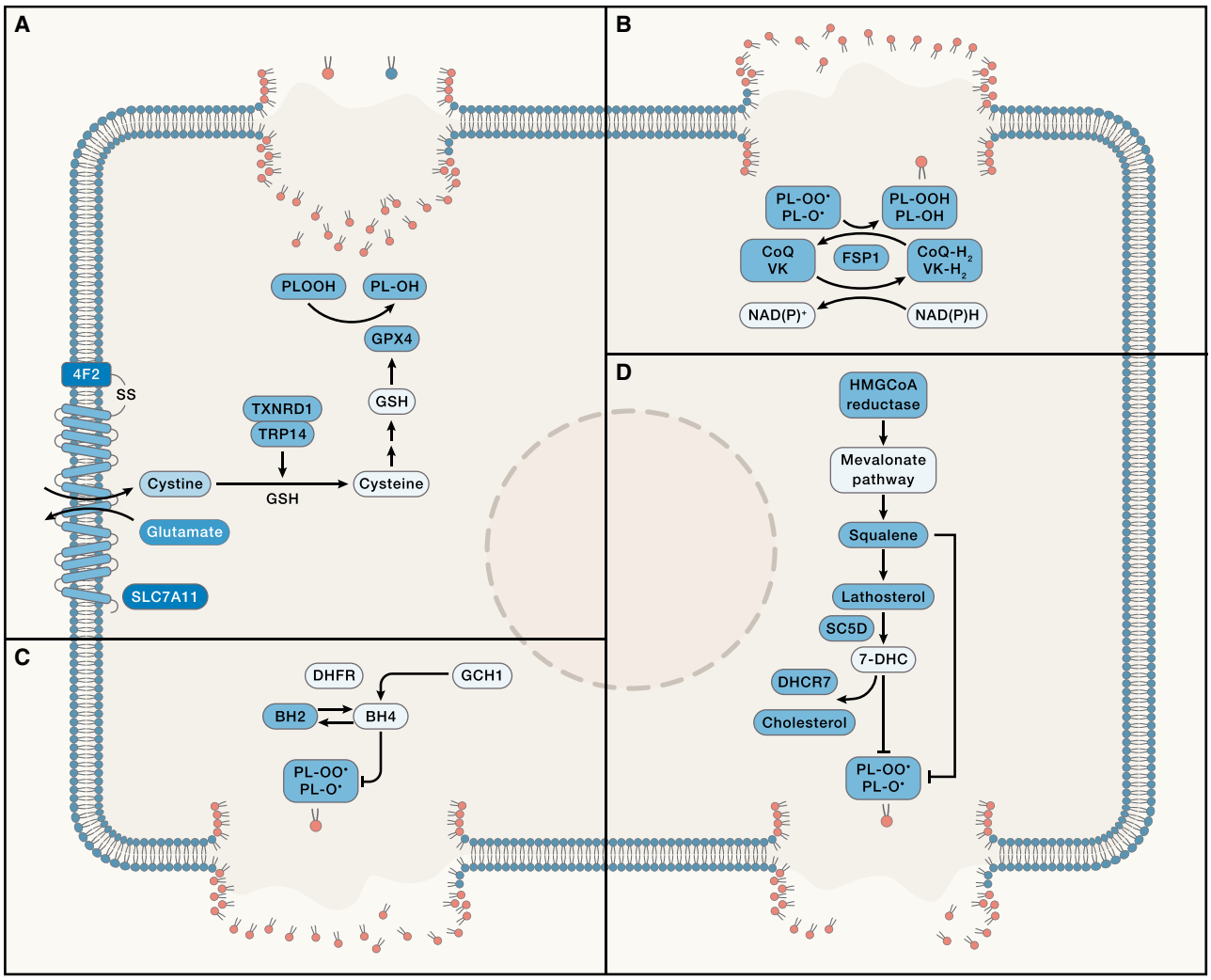

Figure 5. Induction of necroptosis and pyroptosis

(A) The x_c^--GSH-GPX4 system prevents ferroptosis caused by phospholipid peroxidation by reducing phospholipid hydroperoxides to their corresponding alcohols.

(B) The anti-ferroptotic role of the CoQ-FSP1 axis depends on its capacity to convert ubiquinone (CoQ) and VK into their hydroquinone forms, ubiquinol (CoQ-H2) and VK-H2. These reduced compounds inhibit excessive lipid peroxidation by neutralizing radicals in lipid bilayers.

(C) GCH1, responsible for producing BH4, plays a key role in the anti-ferroptotic function of the GCH1-BH4-DHFR pathway. BH4 functions as an RTA and can be regenerated by DHFR.

(D) The presence of lipophilic, naturally occurring RTAs such as squalene and 7-dehydrocholesterol (7-DHC), which are intermediates in the mevalonate pathway, affects cellular susceptibility to ferroptosis. Blocking HMG-CoA, the main enzyme in this pathway, often with statins, can lower the levels of 7-DHC and squalene, potentially making cells more prone to ferroptosis.

converts these (phospho-)lipidhydroperoxides into lipid radicals that trigger self-amplifying radical chain reactions.[287,288] This can occur via lipoxygenase enzymes or non-enzymatic reactive oxygen species (ROS)-driven reactions, with both pathways contributing, depending on the cell type. PUFA-phosphatidylethanolamines are particularly vulnerable substrates, and oxidative damage may begin in the ER[289] or lysosome[290] before spreading to the plasma membrane. At the plasma membrane, lipid oxidation increases membrane tension, opening stretch-sensitive Piezo1 and TRP ion channels.[291,292] The resulting ion imbalance, exacerbated by impaired Na^+/K^+ ATPase activity, leads to water influx, cell swelling, and eventual membrane rupture. While large polymer molecules can temporarily seal the small pores that form, they cannot prevent rupture indefinitely once oxidative damage reaches a critical level.[293,294]

Many cancer cells critically depend on the system x_c^--GSH-GPX4 axis to neutralize peroxidized phospholipids to evade ferroptosis.[295] The role of GPX4 (formerly known as phospholipid hydroperoxide glutathione peroxidase [PHGPx]) in oxidative cell death was initially identified through studies that tested nutrient dependencies in Burkitt lymphoma cells.[290] Strikingly, the same screening strategy, focused on cysteine essentiality and cell death outcomes, also uncovered a protein first hypothesized as a "putative NADH oxidase," later renamed apoptosis-inducing factor

mitochondria-associated 2 (AIFM2) and ultimately designated FSP1.[296,297] Dependency on these systems is heightened in tumors with oncogenic RAS signaling, mesenchymal plasticity, or elevated PUFA content in membranes, all of which dictate ferroptosis sensitivity.[33] These insights catalyzed the development of now widely used small molecules and a successful campaign to interrogate this unique cell death modality. The first relatively selective molecules (e.g., erastin or RAS-selective lethal 3 [RSL-3]) for targeting the cystine-glutamate antiporter system x_c^- and GPX4, respectively) were named after RAS, owing to the unique vulnerability of RAS mutant cancer cell lines to these agents.[298,299] Today, many genotypically distinct tumors, including those that are therapy-resistant and especially those that have lost apoptotic competence, are recognized as harboring a more or less pronounced non-oncogenic addiction to GPX4. This positions ferroptosis induction as an attractive strategy for overcoming the resistance of cancer cells to other forms of cell death.[286]

In terms of its immunogenicity, ferroptotic cells release DAMPs and oxidized lipid species, leukotrienes, prostaglandins, and aldehydes such as 4-hydroxynonenal (4-HNE), respectively. This is predicted to shape the TME.[300] Ferroptosis in tumor cells can elicit pro-inflammatory responses that may synergize with ICB.[301] Yet, ferroptosis occurring in $CD8^+$ T cells or antigen-presenting cells can theoretically impair anti-tumor immunity,[302] reflecting a contextual dichotomy of ferroptosis in cancer immunity (Figure 3).

Metastasis imposes a specific ferroptosis-related vulnerability: EMT-associated transcription factors (e.g., zinc finger E-box binding homeobox 1 [ZEB1]) and a PUFA-rich membrane composition sensitize mesenchymal tumor cells to ferroptosis.[303] Notably, metastatic progression often selects for clones that upregulate FSP1, GPX4, or antioxidant systems (some of them driven by nuclear factor erythroid 2-related factor 2 [NRF2]) to counteract this sensitivity, revealing ferroptosis evasion strategies during dissemination and colonization of distant tissues by malignant cells.

Conceptually, the ferroptotic process appears readily amenable to therapeutic intervention, although the gateway to clinical testing remains a major challenge. Pharmacological GPX4 inhibitors (e.g., RSL3, ML162, and ML210), SLC7A11 inhibitors (e.g., erastin and its metabolically more stable derivatives), as well as FSP1 inhibitors (e.g., iFSP1, icFSP1, viFSP1, and FSEN1), have shown preclinical efficacy in inducing ferroptosis in various cancers.[304–306] Still, concerns regarding the toxicity of targeting ferroptosis surveillance systems in healthy cells persist, as illustrated by the occurrence of early-onset dementia in hereditary GPX4 deficiency, termed spondylometaphyseal dysplasia, Sedaghatian type.[307] A combination of ferroptosis inducers with γ-radiation,[73] immune checkpoint inhibitors,[308] or metabolic modulators may circumvent therapy resistance, though the current evidence remains limited.[309] Successful ferroptosis-targeted therapies will need to account for tumor subtype, cellular redox status, plasma membrane composition, and immune cell sensitivity to minimize collateral damage to anti-tumor immunity.

Three major cooperating systems keep ferroptosis in check[282]: (1) the system x_c^--GSH-GPX4 axis, where SLC7A11-dependent cystine import fuels GSH synthesis enabling GPX4 to reduce phospholipid hydroperoxides (PL-OOH) to inert alcohols; (2) the FSP1-NAD(P)H-ubiquinone (CoQ_{10})/vitamin K (VK) axis,[296,297,310] which regenerates radical-trapping antioxidants (RTAs) (ubiquinol and VK-H_2) in membranes; and (3) the GTP cyclohydrolase 1 (GCH1)-tetrahydrobiopterin (BH4)-dihydrofolate reductase (DHFR) pathway, which supplies the endogenous RTA BH4 and regenerates it.[311,312] Each node is pharmacologically addressable, and dual-axis inhibition can be synergistic, highlighting shared robustness in lipid peroxide control. Statins lower mevalonate-derived lipophilic RTAs (i.e., squalene and 7-dehydrocholesterol), skewing membranes toward peroxidation, whereas withaferin A,[313] RSL3/ML210, imidazole ketone erastin (IKE), and GSH synthesis blockers, such as L-buthionine sulfoximine (BSO), may breach ferroptosis defenses in complementary approaches. Collectively, these findings indicate that ferroptosis sensitivity is not a binary phenotype ("1-0") but a tunable property of iron-redox-lipid metabolism ("1-10") (Figure 5).

Iron handling further shapes this landscape. Labile Fe(II) accelerates radical formation via Fenton chemistry and enhances the auto-oxidation of PUFA-rich phospholipids.[314] Cancer cells typically display an "iron addiction," upregulating their uptake (e.g., transferrin receptor [TFRC] and CD44-hyaluronan-dependent endocytosis[315]) and mobilization (ferritinophagy via nuclear receptor coactivator 4 [NCOA4][316]). This increases the propensity of cells to undergo ferroptosis while becoming increasingly reliant on anti-ferroptotic pathways as a trade-off for growth and plasticity.[317] Although iron chelators can inhibit ferroptosis at least in cultured cells, their effectiveness *in vivo* remains questionable, underscoring the need for refined maps of iron trafficking pathways within tumor environments.[314] Alternatively, mobilizing iron, for instance, that is stored in lysosomes can effectively induce ferroptosis, and this may represent a promising therapeutic strategy in particularly hard-to-target metastasizing cancer cells.[318]

Notably, ferroptosis is tightly integrated with oncogenic signaling and the transcriptional redox circuitries that tumors exploit. Hyperactivation of the KEAP1-NRF2 axis, a common genetic abnormality in lung adenocarcinoma, drives broad antioxidant and iron-handling programs, upregulates lipid detoxifying and modifying enzymes (e.g., aldo-keto reductase family 1 member C1 [AKR1C family] and stearoyl coenzyme A [CoA] reductase-1 [SCD1]), and elevates FSP1 expression.[319–322] These adaptations raise the threshold of cells to undergo ferroptosis and cause resistance to γ-irradiation and oxidation therapies. There is genetic and pharmacologic evidence that simultaneously targeting NRF2 pathway components (e.g., SCD1) and FSP1 can re-sensitize these cancer cells to ferroptosis-inducing agents.

High-risk neuroblastoma displays a distinct ferroptosis-linked transcriptional program. *MYCN* gene amplification with resultant MYCN overexpression drives iron uptake through TFRC, labile iron levels, and lipid peroxidation stress while tightening reliance on cystine uptake and GPX4.[323,324] Limiting cysteine availability (via system x_c^- blockade or its degradation via cyst(e)inase) or directly suppressing GPX4 expression or activity disrupts this redox balance and triggers ferroptosis, leading to tumor regression in preclinical models. The biology of the trace element selenium is pivotal here. GPX4 is a selenoenzyme,[325] and dedicated selenium uptake and processing by the SELENOP-LRP8 axis[323]

and PRDX6 as an intracellular selenide carrier[326–328] sustain GPX4 activity. Disrupting selenium delivery or selenoprotein synthesis disables GPX4-dependent defenses against ferroptosis in cancer cells, although tissue-specific safety windows must be carefully considered.

Membrane composition governs ferroptosis competence.[329–331] PUFA-rich phospholipid bilayers (especially ACSL4-[331] and LPCAT3-routed species[332]) are the prime substrates for peroxidation, whereas monounsaturated fatty acids (MUFAs) and certain sterol-pathway intermediates act as embedded RTAs.[333–335] Cancer cells can exploit this chemistry in both directions. For example, the accumulation of squalene or 7-dehydrocholesterol can buffer radicals to blunt ferroptosis, whereas peroxisome-derived PUFA-ether lipids heighten sensitivity. Oncotype-specific lipidomes (e.g., KRAS-mutant lung cancer, reliant on FASN and Lands cycle remodeling of membranes[336]; and CDKN2A-deleted glioblastoma,[337] which sequesters oxidizable PUFAs into lipid droplets) reveal cellular processes and pathways that can be targeted by GPX4 inhibition or metabolic co-targeting. Hypoxia adds yet another layer in metabolic targeting: HIF-2α can enrich ferroptosis-prone lipid states in clear-cell renal carcinomas,[338,339] sensitizing them to GPX4 blockade, whereas HIF-1α enhances lactate and cystine metabolism that oppose ferroptosis.[340] This indicates the need for context-matched HIF modulation in conjunction with the inducers of ferroptosis.

Beyond endogenous lipids, vitamins also regulate ferroptosis *in vivo*.[341,342] Vitamins E and K function as bona fide naturally occurring RTAs: VKORC1L1 reduces VK to its protective hydroquinone, and p53-dependent repression of VKORC1L1 can diminish this protective layer.[343] Warfarin-mediated inhibition of VK cycling, or combining IKE with dehydroascorbic acid,[344] exemplifies drug repurposing and redox-cofactor re-routing that may push resistant tumors across the ferroptosis threshold in preclinical models. These pharmacological interventions reinforce that *in vivo* ferroptosis hinges on micronutrient fluxes and compartmentalized antioxidant pools as much as on genetic cues, necessitating caution when extrapolating from *in vitro* systems.[283]

Considerations and open questions targeting ferroptosis in cancer cells

Whether ferroptosis is "immunogenic" may also have nuanced, phase-dependent answers.[345] Early ferroptotic states can impede DC maturation, antigen cross-presentation, and the efficacy of immunogenic apoptotic vaccines. This indicates that indiscriminate ferroptosis in tumor cells may dampen adaptive priming in the cancer immune cycle.[302] Conversely, activated $CD8^+$ T cells (e.g., during ICB) can promote tumor cell ferroptosis through IFN-γ-mediated repression of system x_c^- and ACSL4-coupled arachidonate metabolism,[301,346] possibly linking cytotoxic immune responses to ferroptotic killing of cancer cells. Yet, similar chemistry can injure T cells: oxidized lipid uptake via CD36 fosters T cell lipid peroxidation and dysfunction.[347,348] Moreover, broad GPX4 inhibition risks collateral damage to effector T cells, even as selective GPX4 loss in T_{reg} cells can de-repress anti-tumor responses.[349]

These push-pull dynamics extend to innate compartments: ferroptotic cancer cells also release lipid mediators, such as PGE_2. This suppresses the recruitment of cDC1 and NK cells.[350] Conversely, PMN-MDSCs undergoing ferroptosis can shed oxygenated lipids that dampen T cell responses. This can be reversed by inhibiting ferroptosis in myeloid cell subsets,[351] albeit at very early stages of tumor growth. These accelerators and brakes on ferroptosis, along with the differential sensitivity of immune and cancer cells to ferroptosis, may also prevent unwanted autoimmunity during anti-cancer responses. The spatiotemporal precision of ferroptosis, which cell types die, when, and in which niche, will therefore be critical for combining ferroptosis induction with immunotherapy[345] and possibly other types of anti-cancer therapy, such as the induction of apoptosis using BH3 mimetic drugs.[352]

Two difficult-to-treat tumor states repeatedly emerge as ferroptosis-vulnerable. First, epithelial-mesenchymal transition and hematogenous spread elevate membrane PUFA content (via ZEB1-ACSL4/FADS2/ELOVL5 programs)[33,303,353] and oxidative burden, creating GPX4 addiction. Lymphatic transit partly protects against ferroptosis due to the MUFA- and GSH-enriched lymph. This allows metastatic preconditioned clones to gain ferroptosis resistance before entering the bloodstream, an effect that can be counteracted by throttling system x_c^- or GPX4 in circulating or colonizing malignant cells.[353,354] Second, DTP cells, which evade apoptosis during targeted therapy or chemotherapy, activate an ATF4-linked stress program, remodel lipids, and thus become exquisitely dependent on GPX4.[31,32,286,314] Because drug tolerance is in theory, reversible and precedes overt clinical resistance, timed ferroptosis induction during minimal residual disease may extinguish relapse-initiating reservoirs of malignant cells.[46]

Despite a robust mechanistic understanding, bringing ferroptosis to the clinic remains ambitious. The key bottlenecks include (1) pharmacology, the potent, selective, bioavailable GPX4 or FSP1 inhibitors and drug-like system x_c^- modulators that achieve intra-tumoral exposure without triggering intolerable peroxidation in normal tissues; (2) biomarkers, the dynamic readouts of lipid peroxidation (e.g., oxidized phospholipid species), activity of ferroptosis surveillance (SLC7A11, GPX4, FSP1, and GCH1/BH4), lipidome composition (PUFA/MUFA balance and ePL content), selenium handling, and KEAP1-NRF2 wiring; (3) therapeutic window, the organ specific vulnerabilities (e.g., kidney, brain, liver, heart, and hematopoietic niches) that constrain dosing, and immune-compartment sensitivities that must be respected to preserve anti-tumor immunity; and (4) systems context, the microbiome-derived metabolites (e.g., IDA-AHR-ALDH1A3[355,356]) and hypoxia/vascular gradients that modulate ferroptosis thresholds *in vivo*. Looking forward, "rational drug combinations" may help overcome these challenges. For instance, ICB combined with ferroptosis-sensitizing redox or lipid interventions; RT with FSP1 or SCD1 inhibition in NRF2-high tumors; or DTP-targeted GPX4 inhibition in DTP cells paired with lineage-specific protectants. Crucially, the success of such approaches will rely on biomarker-informed patient selection and appropriate clinical trial design.

A clinical view on cell death induction in cancer therapy

We discussed how cancer cells avoid the activation of programmed cell death pathways and how they secure their continued survival despite the presence of cell death-inducing

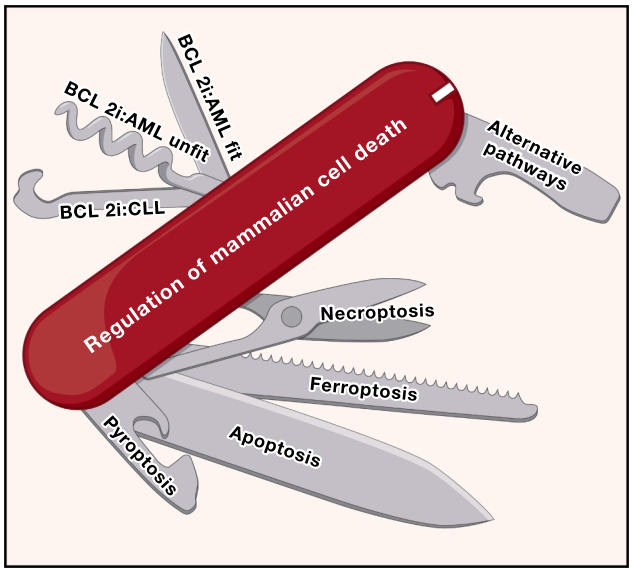

Figure 6. Swiss Army knife

This schematic uses a Swiss Army knife analogy to illustrate how mammalian cells deploy a modular set of RCD programs that can be selectively activated depending on context, stress, and genetic wiring. Different blades or tools represent major death pathways, including apoptosis, necroptosis, pyroptosis, and ferroptosis, each designed for specific biological challenges and immune responses. Additionally, these tools may also indicate alternative or backup pathways that can become active when the primary programs are blocked. In cancer therapy, selectively inhibiting anti-apoptotic BCL-2 family members (e.g., BCL-2 inhibition in CLL or in some AML cases) can be effective but may leave other blades accessible, allowing cancer cells to escape by switching to non-apoptotic death or survival strategies. Conversely, combination or context-aware methods that target multiple "tools" of the knife might overcome resistance by forcing malignant cells to also activate otherwise hidden death pathways. Overall, the figure emphasizes RCD as a versatile, adaptable system, much like a Swiss Army knife, whose full potential is realized only when its modular design is understood and used strategically.

factors.[357] Based on the hypothesis that tumor cells require ongoing protection from cell death induction, (re-)activation of cell death, either by inducing pro-death cues or by releasing the brake on cell death pathways, seemed to be a straightforward therapeutic strategy (Figure 6).

Indeed, the hallmark evasion of cell death, proposed nearly a quarter of a century ago,[6] has demonstrated its clinical relevance most strikingly in the remarkable, though unfortunately rare, success story of CLL treatment.[36] In CLL, apoptosis induction by venetoclax, a first-in-class protein-protein interface inhibitor of pro-survival BCL-2,[47,358] has demonstrated clinical efficacy and has accordingly entered first-line therapy. Yet, venetoclax is not sufficiently active as a single agent over extended periods to serve as a first-line standard. Current treatment algorithms in CLL rely on one or two combination partners in combination with venetoclax to achieve deep and durable remissions.[359] This illustrates three key points: (1) CLL depends on the continuous blockade of mitochondrial apoptosis[36]; (2) cell death induction as a therapeutic principle represents a clinical reality; and (3) resistance to single-agent venetoclax occurs frequently and is highly heterogeneous from a molecular point of view.[360–363]

Despite the success of developing a small molecule with oral bioavailability and a tolerable toxicity profile, the clinical reality in cancer subtypes other than CLL has proven more complex than initially anticipated. To date, the success seen in CLL has not been replicated in other hematopoietic cancers and certainly not in any epithelial cancer, let alone in sarcoma. The closest success story to CLL stems from AML.[364] In AML, venetoclax is indicated in combination with hypomethylating agents, such as 5-azacitidine, for newly diagnosed adult patients ineligible for intensive chemotherapy. Randomized data from the VIALE-A study showed that venetoclax plus azacytidine significantly prolonged overall survival, from 9.6 to 14.7 months.[37] First and foremost, these data illustrate that incorporating venetoclax into AML therapy has improved outcomes and is now part of standard clinical practice. However, a median extension of overall survival by 155 days in AML patients cannot be considered a transformative breakthrough,[37] especially given the relevant grade 3/4 toxicities observed in up to 40% of AML patients across multiple studies.[365]

In younger and fit patients with intermediate/adverse-risk AML, two independent trials showed that hypomethylating agents plus venetoclax as first-line treatment improved clinical outcomes such as event-free survival, transplant rates, and toxicity compared with intensive chemotherapy (NCT0517773 and NCT04801797).[366] If validated clinically in specific genetic risk groups of AML, challenging the paradigm of intensive induction chemotherapy for transplant-eligible AML patients will represent a substantial improvement. However, possibly due to a confounding cross-over effect, overall survival has not reached statistical significance in either trial to date, pending full publication and further analysis[366] (Fathi et al., abs25-8236 ASH 2025).

BCL-2 inhibition has been shown to induce apoptosis in preclinical studies in myelodysplastic syndrome (MDS) patient samples.[367,368] However, the recent phase 3 VERONA study, which randomized newly diagnosed intermediate/high-risk MDS patients to venetoclax plus azacitidine versus placebo plus azacitidine (NCT04401748), failed to achieve an overall survival benefit.[369] While complementary clinical readouts, such as reductions of bone marrow blasts or transfusion independence, represent important clinical improvements, we should acknowledge that BCL-2 inhibition alone represents a valuable but only partially effective avenue in myeloid cancers.

There is substantial activity in testing apoptosis pathway activation beyond BCL-2 inhibition in hematopoietic cancers, including the development of MCL-1 inhibitors. Several small-molecule MCL-1 inhibitors are available and are currently in phase 1/2 clinical trials for relapsed/refractory hematologic cancers (e.g., NCT02675452, NCT02992483, NCT02979366, NCT04629443, and NCT04178902). However, their development has been hampered by cardiac toxicities.[370–372] Targeting of BCL-XL represents another approach to induce mitochondrial apoptosis. Since BCL-XL plays a critical role in platelet survival, inhibiting its pro-survival function poses a clinical challenge due to imminent thrombocytopenia.[373,374] Recent advances in BCL-XL-directed protein degrader technology raise the possibility that anti-tumor activity may be dissociated from thrombocytopenia in solid or hematopoietic cancers.[375,376]

In contrast to hematopoietic cancers, outcomes reported for apoptosis induction in epithelial tumors or sarcomas have

been less encouraging. Solid tumors remain an experimental space for targeted apoptosis activation with only marginal signals of clinical benefit. One representative completed randomized phase 2 study in a solid tumor is the VERONICA trial on estrogen/progesterone receptor-positive and HER2-negative metastatic breast cancer patients, which reported negative results overall.[377] Ongoing phase 1/2 and 1b/2 studies in breast cancer (NCT03900884), small cell lung cancer (NCT04543916), and non-small cell lung cancer (NCT04274907), among many others, are primarily assessing combination strategies, but none have reported convincing phase 2 efficacy signals to date. Testing of MCL-1 inhibition in solid cancers is also progressing in phase 1 (NCT04837677 and NCT05006794), with additional data expected soon.

Given that our current clinical reality remains somewhat removed from the prospect of broadly applying cell death-inducing therapies across diverse cancer types or even across multiple cell death pathways, it may be prudent to recalibrate our expectations for this therapeutic approach. Apoptosis remains the best-studied programmed cell death pathway[378] and is the only one for which pharmaceutical-grade compounds have been approved by national medicine agencies such as the FDA and EMA. However, despite the remarkable sensitivity of malignant hematopoietic cells from both the lymphoid[379–381] and the myeloid compartments[382–384] to mitochondrial apoptosis *in vitro*, *ex vivo*, or in mouse models, many encouraging preclinical results from the laboratory have not yet translated to the patient's bedside as anticipated.

Clinical medicine rarely follows simple rules such as "patient is anemic = give iron," or in the context of cell death, "BCL-2 is overexpressed = give venetoclax." This is exemplified in the case of follicular lymphoma, where malignant cells harboring the chromosomal translocation t(14;18)(q32;q21) exhibit aberrant BCL-2 overexpression,[385] yet fail to achieve durable responses to the single-agent BCL-2 inhibitor venetoclax[386] and treatment combinations have yielded substantial toxicity,[387] unfortunately.

From the original hallmarks of cancer publication[6] to the recent summaries emphasizing the complexity of cancer cell biology,[9] the difficulty of defining one specific yet critical signaling pathway as the Achilles heel of cancer cell survival has been widely acknowledged. Still, the field may have been overly ambitious in assuming that applying leverage on one signaling pathway (apoptosis) would be sufficient to extend the overall survival of patients across diverse cancer types. Clinical relapse samples from venetoclax-treated CLL patients show that tumor cells can develop heterogeneous forms of resistance mechanisms.[360–363] The resistant clones arise through mutations in BCL-2 itself that prevent drug binding,[363] or by outgrowth of pre-existing or newly selected clones harboring alternative resistance alterations, such as upregulation of non-targeted pro-survival BCL-2 family members (e.g., MCL-1 and BCL-XL) or *TP53* mutations in CLL.[388,389]

Tumor burden and clonal heterogeneity impact the development of resistance to any therapy, and cell death-inducing agents are no exception.[9,390–392] Given the multitude of potential resistance mechanisms to venetoclax or other death-inducing agents, combinatorial treatment strategies are likely required to address tumor complexity. Evidence from CLL, arguably the "poster child" of cell death therapy, supports this principle. Reducing CLL clone size and heterogeneity is achieved by combining venetoclax with compounds, such as Bruton's tyrosine kinase inhibitors or anti-CD20 antibodies.[359] Not surprisingly, the response rates of these combinations are superior to those for venetoclax monotherapy, reinforcing the need to pair cell death-inducing agents with complementary treatment modalities.[393–397]

The concept of apoptosis-independent killing of leukemia, such as AML, has been based on the finding that RIPK3-mediated cell death serves as a leukemia-suppressive mechanism in AML,[398] and that leukemic cells readily undergo cell death and inflammation-induced differentiation in response to TNFR1-dependent signals if either TRAF2 is missing[399] or IAPs are inhibited.[184,400] Accordingly, cIAP1, cIAP2, and/or XIAP inhibitors have been developed.

Oral SMAC mimetics, such as xevinapant, are small-molecule antagonists of cIAP1/2 and XIAP.[401] The anticipated clinical utility was based, at least in part, on the central roles of cIAP1/2 and XIAP in TNF receptor 1-mediated induction of cell death upon inhibition of either of these proteins.[402,403] In addition, preclinical evidence suggested that necroptosis-inducing treatments might alert and engage the immune system as tumor cells die.[39] A phase 2 trial of IAP inhibition combined with radio-chemotherapy in squamous cell carcinoma of the head and neck reported improvements in the primary endpoint of loco-regional control as well as progression-free and overall survival.[404,405] However, the subsequent phase 3 trial failed to demonstrate any improvement in survival outcomes.[406] While there is still substantial preclinical evidence for targeting IAP in cancer, as discussed above, further translational research is needed to identify responsive cancer entities, optimal drug combinations, and the most sensitive patient subgroups.

Overall, the clinical implementation of compounds that specifically activate a defined programmed cell death signaling pathway, unlike chemo- or RT, remains currently restricted to relatively low-prevalence hematopoietic cancers, such as CLL and AML. Although the clinical development of therapies targeting apoptosis, necroptosis, and emerging modalities such as ferroptosis or pyroptosis is ongoing, achieving major breakthroughs in the near future may remain challenging. Because the evasion of cell death in cancer is a highly complex, dynamically evolving process involving longitudinal adaptation programs, it is unlikely that a single "silver bullet" will overcome this hallmark. Accordingly, a more detailed mechanistic understanding of emerging cell death modalities, more precise biomarker strategies, and the systematic testing of cell death-inducing agents as a treatment combination will be essential for expanding the therapeutic reach of cell death-based treatments across cancer entities.

CONCLUDING REMARKS: THE COMING 25 YEARS FOR CELL DEATH RESEARCH IN CANCER

We argue that over the next 25 years, cell death research is poised to transform cancer therapy by enabling the context-specific targeting of tumor cell vulnerabilities, including metastasis, drug persistence, metabolic adaptations, and cellular plasticity, through a deeper mechanistic understanding and the

therapeutic manipulation of all RCD pathways. As the field continues to expand beyond apoptosis to fully encompass necroptosis, pyroptosis, and ferroptosis, these RCD modalities will be leveraged to counteract resistance to cell death, one of the "founding" hallmarks of cancer. Importantly, apoptosis and non-apoptotic pathways closely intersect with the other core hallmarks of cancer, such as deregulated metabolism, immune evasion, and tumor-promoting inflammation, possibly pinpointing "synthetic lethality" for therapeutic intervention[407] (Figure 6).

Advances in high-throughput technologies, including single-cell profiling, spatial omics, and *in vivo* CRISPR screening, will illuminate cell type- and lineage-specific dependencies on cell death suppressive mechanisms, even within rare subpopulations like therapy-resistant persister cells and immune-evasive niches. These insights, coupled with rational drug design, will foster next-generation combination therapies that not only eradicate tumor cells but also potentially collaborate with anti-tumor immunity. Ultimately, the incorporation of cell death biology into systems oncology and personalized medicine[408] will redefine therapeutic strategy, shifting the paradigm from broadly cytotoxic approaches toward precisely targeted induction of cell death, tailored to the evolving biology of each patient's tumor.

ACKNOWLEDGMENTS

M.C. was supported by Deutsche Forschungsgemeinschaft (DFG) (CO 291/7-1, # 428858739 and the Priority Program SPP 2306 [CO 291/9-1, #461385412; CO 291/10-1, #461507177]), the DFG CRC TRR 353 (CO 291/11-1; #471011418), the German Federal Ministry of Education and Research (BMBF) VIP+ program NEUROPROTEKT (03VP04260), the BMBF program FERROPATH (01EJ2205B), and the European Research Council (ERC) under the European Union's Horizon 2020 research and innovation program (grant agreement no. GA 884754). A.S. is supported by the National Health and Medical Research Council (NHMRC), investigator grant 1020363, and a Cancer Council Victoria Grant in Aid. J.Y. was supported by the National Natural Science Foundation of China (82188101), Brain Science and Brain-like Intelligence Technology-National Science and Technology Major Project (2025ZD0214800), the Shanghai Basic Science Pioneer Project, and the Shanghai Municipal Science and Technology Major Project. F.S. was supported by the Basic Science Center Project (82388201) of the National Natural Science Foundation of China (NSFC) and the Tencent New Cornerstone Investigator Program. P.J.J. was supported by EU HORIZON-WIDERA-2023-ACCESS-02 #101160061 (ZAP Cancer), FWF #P37275, and FWF RU5659 TARGET-MPN: HE6233/15-1 Multilateral Initiatives #I6944. P.V. has received funding from Research Foundation – Flanders 3G033120, 3G0A9322, and 3G0AA824; The Excellence of Science Research Consortium3G0I5722; UGent Special Research Fund Methusalem 01M00709; Interuniversity BOF projects 01IB3920; and the Foundation against Cancer 365A08921 and 365G09122. A.W. was supported by a Walter Benjamin Fellowship by the DFG (585726918).

DECLARATION OF INTERESTS

M.C. is a co-founder and shareholder of ROSCUE Therapeutics GmbH and holds patents for some of the compounds described herein. A.S. is an employee of The Walter and Eliza Hall Institute. The Walter and Eliza Hall Institute receives milestone payments and royalties from the sale of venetoclax, part of which is distributed to current and past employees. A.S. collaborated with Servier on the development of MCL-1 inhibitors and received financial support for some of his research. F.S. is the scientific founder and shareholder of Pyrotech Therapeutics. P.J.J. is a founder, shareholder, and employee of Cycuria Therapeutics GmbH, Graz, and a shareholder of Vessel FlexCo, Graz. P.J.J. is listed on the patent application European Patent Office #EP 3 969 123 A1 and has consulting or advisory roles, received honoraria, research funding, and/or travel/accommodation expenses from Astra Zeneca, Bayer, Boehringer Ingelheim, Novartis, Pfizer, Servier, Roche, BMS and Celgene, Pierre Fabre, Janssen/Johnson&Johnson, MSD, Merck, Sanofi/Aventis, Ipsen, Amgen, Vessel FlexCo, and Cycuria Therapeutics.

REFERENCES

1. Tummers, B., and Green, D.R. (2022). The evolution of regulated cell death pathways in animals and their evasion by pathogens. Physiol. Rev. *102*, 411–454. https://doi.org/10.1152/physrev.00002.2021.
2. Oberst, A., Bender, C., and Green, D.R. (2008). Living with death: the evolution of the mitochondrial pathway of apoptosis in animals. Cell Death Differ. *15*, 1139–1146. https://doi.org/10.1038/cdd.2008.65.
3. Ameisen, J.C. (2002). On the origin, evolution, and nature of programmed cell death: a timeline of four billion years. Cell Death Differ. *9*, 367–393. https://doi.org/10.1038/sj.cdd.4400950.
4. Virchow, R., Chance, F., and Goodsir, J.; King's College London; Pathological Institute of Berlin (1860). Cellular pathology as based upon physiological and pathological histology; twenty lectures delivered in the Pathological Institute of Berlin during the months of February, March, and April, 1858 (John Churchill).
5. Metchnikoff, E. (1905). Old Age. Sci. Am. *60*, 24967–24968. https://doi.org/10.1038/scientificamerican11111905-24967supp.
6. Hanahan, D., and Weinberg, R.A. (2000). The hallmarks of cancer. Cell *100*, 57–70. https://doi.org/10.1016/S0092-8674(00)81683-9.
7. Hanahan, D., and Weinberg, R.A. (2011). Hallmarks of cancer: the next generation. Cell *144*, 646–674. https://doi.org/10.1016/j.cell.2011.02.013.
8. Hanahan, D. (2022). Hallmarks of cancer: New dimensions. Cancer Discov. *12*, 31–46. https://doi.org/10.1158/2159-8290.CD-21-1059.
9. Swanton, C., Bernard, E., Abbosh, C., André, F., Auwerx, J., Balmain, A., Bar-Sagi, D., Bernards, R., Bullman, S., DeGregori, J., et al. (2024). Embracing cancer complexity: Hallmarks of systemic disease. Cell *187*, 1589–1616. https://doi.org/10.1016/j.cell.2024.02.009.
10. Newton, K., Strasser, A., Kayagaki, N., and Dixit, V.M. (2024). Cell death. Cell *187*, 235–256. https://doi.org/10.1016/j.cell.2023.11.044.
11. Kayagaki, N., Webster, J.D., and Newton, K. (2024). Control of cell death in health and disease. Annu. Rev. Pathol. *19*, 157–180. https://doi.org/10.1146/annurev-pathmechdis-051022-014433.
12. Green, D.R. (2024). Cell death: Revisiting the roads to ruin. Dev. Cell *59*, 2523–2531. https://doi.org/10.1016/j.devcel.2024.08.008.
13. Tsvetkov, P., Coy, S., Petrova, B., Dreishpoon, M., Verma, A., Abdusamad, M., Rossen, J., Joesch-Cohen, L., Humeidi, R., Spangler, R.D., et al. (2022). Copper induces cell death by targeting lipoylated TCA cycle proteins. Science *375*, 1254–1261. https://doi.org/10.1126/science.abf0529.
14. Lei, G., Sun, M., Cheng, J., Ye, R., Lu, Z., Horbath, A., Huo, D., Wu, S., Alapati, A., Aggarwal, S., et al. (2025). Radiotherapy promotes cuproptosis and synergizes with cuproptosis inducers to overcome tumor radioresistance. Cancer Cell *43*, 1076–1092.e5. https://doi.org/10.1016/j.ccell.2025.03.031.
15. Brinkmann, V., Reichard, U., Goosmann, C., Fauler, B., Uhlemann, Y., Weiss, D.S., Weinrauch, Y., and Zychlinsky, A. (2004). Neutrophil extracellular traps kill bacteria. Science *303*, 1532–1535. https://doi.org/10.1126/science.1092385.
16. Shahzad, M.H., Rayes, R.F., Cools-Lartigue, J., and Spicer, J.D. (2025). Neutrophil extracellular traps in cancer. Nat. Rev. Cancer *26*, 104–117. https://doi.org/10.1038/s41568-025-00888-7.
17. Gupta, S., and Kaplan, M.J. (2016). The role of neutrophils and NETosis in autoimmune and renal diseases. Nat. Rev. Nephrol. *12*, 402–413. https://doi.org/10.1038/nrneph.2016.71.

18. Liu, X., Nie, L., Zhang, Y., Yan, Y., Wang, C., Colic, M., Olszewski, K., Horbath, A., Chen, X., Lei, G., et al. (2023). Actin cytoskeleton vulnerability to disulfide stress mediates disulfidptosis. Nat. Cell Biol. *25*, 404–414. https://doi.org/10.1038/s41556-023-01091-2.

19. Mao, C., Jiang, D., Koong, A.C., and Gan, B. (2026). Exploiting metabolic cell death for cancer therapy. Nat. Rev. Cancer *26*, 27–45. https://doi.org/10.1038/s41568-025-00879-8.

20. Mao, C., Wang, M., Zhuang, L., and Gan, B. (2024). Metabolic cell death in cancer: ferroptosis, cuproptosis, disulfidptosis, and beyond. Protein Cell *15*, 642–660. https://doi.org/10.1093/procel/pwae003.

21. Song, X., Zhu, S., Xie, Y., Liu, J., Sun, L., Zeng, D., Wang, P., Ma, X., Kroemer, G., Bartlett, D.L., et al. (2018). JTC801 induces pH-dependent death specifically in cancer cells and slows growth of tumors in mice. Gastroenterology *154*, 1480–1493. https://doi.org/10.1053/j.gastro.2017.12.004.

22. Holze, C., Michaudel, C., Mackowiak, C., Haas, D.A., Benda, C., Hubel, P., Pennemann, F.L., Schnepf, D., Wettmarshausen, J., Braun, M., et al. (2018). Oxeiptosis, a ROS-induced caspase-independent apoptosis-like cell-death pathway. Nat. Immunol. *19*, 130–140. https://doi.org/10.1038/s41590-017-0013-y.

23. Andrabi, S.A., Dawson, T.M., and Dawson, V.L. (2008). Mitochondrial and nuclear cross talk in cell death: parthanatos. Ann. N. Y. Acad. Sci. *1147*, 233–241. https://doi.org/10.1196/annals.1427.014.

24. Ray Chaudhuri, A., and Nussenzweig, A. (2017). The multifaceted roles of PARP1 in DNA repair and chromatin remodelling. Nat. Rev. Mol. Cell Biol. *18*, 610–621. https://doi.org/10.1038/nrm.2017.53.

25. Drew, Y., Zenke, F.T., and Curtin, N.J. (2025). DNA damage response inhibitors in cancer therapy: lessons from the past, current status and future implications. Nat. Rev. Drug Discov. *24*, 19–39. https://doi.org/10.1038/s41573-024-01060-w.

26. Pasparakis, M., and Vandenabeele, P. (2015). Necroptosis and its role in inflammation. Nature *517*, 311–320. https://doi.org/10.1038/nature14191.

27. Broz, P., and Dixit, V.M. (2016). Inflammasomes: mechanism of assembly, regulation and signalling. Nat. Rev. Immunol. *16*, 407–420. https://doi.org/10.1038/nri.2016.58.

28. Karki, R., and Kanneganti, T.-D. (2021). The "cytokine storm": molecular mechanisms and therapeutic prospects. Trends Immunol. *42*, 681–705. https://doi.org/10.1016/j.it.2021.06.001.

29. Dixon, S.J., Lemberg, K.M., Lamprecht, M.R., Skouta, R., Zaitsev, E.M., Gleason, C.E., Patel, D.N., Bauer, A.J., Cantley, A.M., Yang, W.S., et al. (2012). Ferroptosis: an iron-dependent form of nonapoptotic cell death. Cell *149*, 1060–1072. https://doi.org/10.1016/j.cell.2012.03.042.

30. Jiang, X., Stockwell, B.R., and Conrad, M. (2021). Ferroptosis: mechanisms, biology and role in disease. Nat. Rev. Mol. Cell Biol. *22*, 266–282. https://doi.org/10.1038/s41580-020-00324-8.

31. Hangauer, M.J., Viswanathan, V.S., Ryan, M.J., Bole, D., Eaton, J.K., Matov, A., Galeas, J., Dhruv, H.D., Berens, M.E., Schreiber, S.L., et al. (2017). Drug-tolerant persister cancer cells are vulnerable to GPX4 inhibition. Nature *551*, 247–250. https://doi.org/10.1038/nature24297.

32. Russo, M., Chen, M., Mariella, E., Peng, H., Rehman, S.K., Sancho, E., Sogari, A., Toh, T.S., Balaban, N.Q., Batlle, E., et al. (2024). Cancer drug-tolerant persister cells: from biological questions to clinical opportunities. Nat. Rev. Cancer *24*, 694–717. https://doi.org/10.1038/s41568-024-00737-z.

33. Viswanathan, V.S., Ryan, M.J., Dhruv, H.D., Gill, S., Eichhoff, O.M., Seashore-Ludlow, B., Kaffenberger, S.D., Eaton, J.K., Shimada, K., Aguirre, A.J., et al. (2017). Dependency of a therapy-resistant state of cancer cells on a lipid peroxidase pathway. Nature *547*, 453–457. https://doi.org/10.1038/nature23007.

34. Vaux, D.L., Cory, S., and Adams, J.M. (1988). Bcl-2 gene promotes haemopoietic cell survival and cooperates with c-myc to immortalize pre-B cells. Nature *335*, 440–442. https://doi.org/10.1038/335440a0.

35. Strasser, A., Harris, A.W., Bath, M.L., and Cory, S. (1990). Novel primitive lymphoid tumours induced in transgenic mice by cooperation between myc and bcl-2. Nature *348*, 331–333. https://doi.org/10.1038/348331a0.

36. Roberts, A.W., Davids, M.S., Pagel, J.M., Kahl, B.S., Puvvada, S.D., Gerecitano, J.F., Kipps, T.J., Anderson, M.A., Brown, J.R., Gressick, L., et al. (2016). Targeting BCL2 with venetoclax in relapsed chronic Lymphocytic leukemia. N. Engl. J. Med. *374*, 311–322. https://doi.org/10.1056/NEJMoa1513257.

37. DiNardo, C.D., Jonas, B.A., Pullarkat, V., Thirman, M.J., Garcia, J.S., Wei, A.H., Konopleva, M., Döhner, H., Letai, A., Fenaux, P., et al. (2020). Azacitidine and venetoclax in previously untreated acute myeloid leukemia. N. Engl. J. Med. *383*, 617–629. https://doi.org/10.1056/NEJMoa2012971.

38. Kroemer, G., Galassi, C., Zitvogel, L., and Galluzzi, L. (2022). Immunogenic cell stress and death. Nat. Immunol. *23*, 487–500. https://doi.org/10.1038/s41590-022-01132-2.

39. Meier, P., Legrand, A.J., Adam, D., and Silke, J. (2024). Immunogenic cell death in cancer: targeting necroptosis to induce antitumour immunity. Nat. Rev. Cancer *24*, 299–315. https://doi.org/10.1038/s41568-024-00674-x.

40. Evan, G.I., Wyllie, A.H., Gilbert, C.S., Littlewood, T.D., Land, H., Brooks, M., Waters, C.M., Penn, L.Z., and Hancock, D.C. (1992). Induction of apoptosis in fibroblasts by c-myc protein. Cell *69*, 119–128. https://doi.org/10.1016/0092-8674(92)90123-T.

41. Kastenhuber, E.R., and Lowe, S.W. (2017). Putting p53 in context. Cell *170*, 1062–1078. https://doi.org/10.1016/j.cell.2017.08.028.

42. Strasser, A., Harris, A.W., and Cory, S. (1991). bcl-2 transgene inhibits T cell death and perturbs thymic self-censorship. Cell *67*, 889–899. https://doi.org/10.1016/0092-8674(91)90362-3.

43. Bedoui, S., Herold, M.J., and Strasser, A. (2020). Emerging connectivity of programmed cell death pathways and its physiological implications. Nat. Rev. Mol. Cell Biol. *21*, 678–695. https://doi.org/10.1038/s41580-020-0270-8.

44. Nakamura, T., and Conrad, M. (2024). Exploiting ferroptosis vulnerabilities in cancer. Nat. Cell Biol. *26*, 1407–1419. https://doi.org/10.1038/s41556-024-01425-8.

45. Wahida, A., and Conrad, M. (2025). Decoding ferroptosis for cancer therapy. Nat. Rev. Cancer *25*, 910–924. https://doi.org/10.1038/s41568-025-00864-1.

46. Ubellacker, J.M., and Dixon, S.J. (2025). Prospects for ferroptosis therapies in cancer. Nat. Cancer *6*, 1326–1336. https://doi.org/10.1038/s43018-025-01037-7.

47. Diepstraten, S.T., Anderson, M.A., Czabotar, P.E., Lessene, G., Strasser, A., and Kelly, G.L. (2022). The manipulation of apoptosis for cancer therapy using BH3-mimetic drugs. Nat. Rev. Cancer *22*, 45–64. https://doi.org/10.1038/s41568-021-00407-4.

48. Tivey, A., Church, M., Rothwell, D., Dive, C., and Cook, N. (2022). Circulating tumour DNA - looking beyond the blood. Nat. Rev. Clin. Oncol. *19*, 600–612. https://doi.org/10.1038/s41571-022-00660-y.

49. Flemming, A. (2025). Gasdermin pores on extracellular vesicles spread cell death. Nat. Rev. Immunol. *25*, 75. https://doi.org/10.1038/s41577-025-01133-w.

50. Malaviya, P., Shukal, D., and Vasavada, A.R. (2019). Nanotechnology-based drug delivery, metabolism and toxicity. Curr. Drug Metab. *20*, 1167–1190. https://doi.org/10.2174/1389200221666200103091753.

51. Aliazis, K., Christofides, A., Shah, R., Yeo, Y.Y., Jiang, S., Charest, A., and Boussiotis, V.A. (2025). The tumor microenvironment's role in the response to immune checkpoint blockade. Nat. Cancer *6*, 924–937. https://doi.org/10.1038/s43018-025-00986-3.

52. Ahmed, A., and Tait, S.W.G. (2020). Targeting immunogenic cell death in cancer. Mol. Oncol. *14*, 2994–3006. https://doi.org/10.1002/1878-0261.12851.

53. Costa-Mattioli, M., and Walter, P. (2020). The integrated stress response: From mechanism to disease. Science *368*, eaat5314. https://doi.org/10.1126/science.aat5314.

54. Krysko, D.V., Garg, A.D., Kaczmarek, A., Krysko, O., Agostinis, P., and Vandenabeele, P. (2012). Immunogenic cell death and DAMPs in cancer therapy. Nat. Rev. Cancer *12*, 860–875. https://doi.org/10.1038/nrc3380.

55. Vandenabeele, P., Conrad, M., and Wahida, A. (2025). For better or worse: The impact. Apoptosis and Other forms of Cell Death in the Regulation of Inflammation in Health and Disease. Adv. Exp. Med. Biol. *1481*, 241–292.

56. Elhanani, O., Ben-Uri, R., and Keren, L. (2023). Spatial profiling technologies illuminate the tumor microenvironment. Cancer Cell *41*, 404–420. https://doi.org/10.1016/j.ccell.2023.01.010.

57. Petroni, G., Cantley, L.C., Santambrogio, L., Formenti, S.C., and Galluzzi, L. (2022). Radiotherapy as a tool to elicit clinically actionable signalling pathways in cancer. Nat. Rev. Clin. Oncol. *19*, 114–131. https://doi.org/10.1038/s41571-021-00579-w.

58. Rehwinkel, J., and Mehdipour, P. (2025). ADAR1: from basic mechanisms to inhibitors. Trends Cell Biol. *35*, 59–73. https://doi.org/10.1016/j.tcb.2024.06.006.

59. Guo, Z., Liu, T., Gao, Q., Wang, C., Wang, Q., Du, R., Liu, Y., Li, H., Huang, Y., Liu, Y., et al. (2025). Targeting the cGAS-STING pathway for cancer immunotherapy: From small-molecule agonists to advanced nanomaterials. Mol. Pharm. *22*, 7262–7284. https://doi.org/10.1021/acs.molpharmaceut.5c01263.

60. Erlich, Z., Shlomovitz, I., Edry-Botzer, L., Cohen, H., Frank, D., Wang, H., Lew, A.M., Lawlor, K.E., Zhan, Y., Vince, J.E., et al. (2019). Macrophages, rather than DCs, are responsible for inflammasome activity in the GM-CSF BMDC model. Nat. Immunol. *20*, 397–406. https://doi.org/10.1038/s41590-019-0313-5.

61. Vandenabeele, P., Conrad, M., and Wahida, A. (2025). For better or worse: the impact of necrotic cell death modalities. Adv. Exp. Med. Biol. *1481*, 241–292. https://doi.org/10.1007/978-3-031-92785-0_8.

62. Degen, M., Santos, J.C., Pluhackova, K., Cebrero, G., Ramos, S., Jankevicius, G., Hartenian, E., Guillerm, U., Mari, S.A., Kohl, B., et al. (2023). Structural basis of NINJ1-mediated plasma membrane rupture in cell death. Nature *618*, 1065–1071. https://doi.org/10.1038/s41586-023-05991-z.

63. Kayagaki, N., Kornfeld, O.S., Lee, B.L., Stowe, I.B., O'Rourke, K., Li, Q., Sandoval, W., Yan, D., Kang, J., Xu, M., et al. (2021). NINJ1 mediates plasma membrane rupture during lytic cell death. Nature *591*, 131–136. https://doi.org/10.1038/s41586-021-03218-7.

64. Kayagaki, N., Stowe, I.B., Alegre, K., Deshpande, I., Wu, S., Lin, Z., Kornfeld, O.S., Lee, B.L., Zhang, J., Liu, J., et al. (2023). Inhibiting membrane rupture with NINJ1 antibodies limits tissue injury. Nature *618*, 1072–1077. https://doi.org/10.1038/s41586-023-06191-5.

65. David, L., Borges, J.P., Hollingsworth, L.R., Volchuk, A., Jansen, I., Garlick, E., Steinberg, B.E., and Wu, H. (2024). NINJ1 mediates plasma membrane rupture by cutting and releasing membrane disks. Cell *187*, 2224–2235.e16. https://doi.org/10.1016/j.cell.2024.03.008.

66. Noh, H., Hashem, Z., Boms, E., and Najafov, A. (2026). SIGLEC12 mediates plasma membrane rupture during necroptotic cell death. Nature *649*, 460–466. https://doi.org/10.1038/s41586-025-09741-1.

67. Vanden Berghe, T., Linkermann, A., Jouan-Lanhouet, S., Walczak, H., and Vandenabeele, P. (2014). Regulated necrosis: the expanding network of non-apoptotic cell death pathways. Nat. Rev. Mol. Cell Biol. *15*, 135–147. https://doi.org/10.1038/nrm3737.

68. Liu, S., Perez, P., Sun, X., Chen, K., Fatirkhorani, R., Mammadova, J., and Wang, Z. (2024). MLKL polymerization-induced lysosomal membrane permeabilization promotes necroptosis. Cell Death Differ. *31*, 40–52. https://doi.org/10.1038/s41418-023-01237-7.

69. Vanden Berghe, T., Vanlangenakker, N., Parthoens, E., Deckers, W., Devos, M., Festjens, N., Guerin, C.J., Brunk, U.T., Declercq, W., and Vandenabeele, P. (2010). Necroptosis, necrosis and secondary necrosis converge on similar cellular disintegration features. Cell Death Differ. *17*, 922–930. https://doi.org/10.1038/cdd.2009.184.

70. Bell, E. (2009). CLEC9A: linking necrosis and immunity. Nat. Rev. Immunol. *9*, 223. https://doi.org/10.1038/nri2531.

71. Aaes, T.L., and Vandenabeele, P. (2021). The intrinsic immunogenic properties of cancer cell lines, immunogenic cell death, and how these influence host antitumor immune responses. Cell Death Differ. *28*, 843–860. https://doi.org/10.1038/s41418-020-00658-y.

72. Yang, Y., Wu, M., Cao, D., Yang, C., Jin, J., Wu, L., Hong, X., Li, W., Lu, L., Li, J., et al. (2021). ZBP1-MLKL necroptotic signaling potentiates radiation-induced antitumor immunity via intratumoral STING pathway activation. Sci. Adv. *7*, eabf6290. https://doi.org/10.1126/sciadv.abf6290.

73. Ye, L.F., Chaudhary, K.R., Zandkarimi, F., Harken, A.D., Kinslow, C.J., Upadhyayula, P.S., Dovas, A., Higgins, D.M., Tan, H., Zhang, Y., et al. (2020). Radiation-induced lipid peroxidation triggers ferroptosis and synergizes with ferroptosis inducers. ACS Chem. Biol. *15*, 469–484. https://doi.org/10.1021/acschembio.9b00939.

74. Zhang, F., Liu, T., Huang, H.-C., Zhao, Y.-Y., He, M., Yuan, W., Li, L., Li, J., Wu, D.-M., and Xu, Y. (2022). Activation of pyroptosis and ferroptosis is involved in radiation-induced intestinal injury in mice. Biochem. Biophys. Res. Commun. *631*, 102–109. https://doi.org/10.1016/j.bbrc.2022.09.073.

75. Letai, A., and de The, H. (2025). Conventional chemotherapy: millions of cures, unresolved therapeutic index. Nat. Rev. Cancer *25*, 209–218. https://doi.org/10.1038/s41568-024-00778-4.

76. Moreno-Alcántar, G., Drexler, M., and Casini, A. (2024). Assembling a new generation of radiopharmaceuticals with supramolecular theranostics. Nat. Rev. Chem. *8*, 893–914. https://doi.org/10.1038/s41570-024-00657-4.

77. Lindsten, T., Ross, A.J., King, A., Zong, W.X., Rathmell, J.C., Shiels, H.A., Ulrich, E., Waymire, K.G., Mahar, P., Frauwirth, K., et al. (2000). The combined functions of proapoptotic Bcl-2 family members bak and bax are essential for normal development of multiple tissues. Mol. Cell *6*, 1389–1399. https://doi.org/10.1016/S1097-2765(00)00136-2.

78. Ke, F.F.S., Vanyai, H.K., Cowan, A.D., Delbridge, A.R.D., Whitehead, L., Grabow, S., Czabotar, P.E., Voss, A.K., and Strasser, A. (2018). Embryogenesis and adult life in the absence of intrinsic apoptosis effectors BAX, BAK, and BOK. Cell *173*, 1217–1230.e17. https://doi.org/10.1016/j.cell.2018.04.036.

79. Kerr, J.F., Wyllie, A.H., and Currie, A.R. (1972). Apoptosis: a basic biological phenomenon with wide-ranging implications in tissue kinetics. Br. J. Cancer *26*, 239–257. https://doi.org/10.1038/bjc.1972.33.

80. Wyllie, A.H. (1980). Glucocorticoid-induced thymocyte apoptosis is associated with endogenous endonuclease activation. Nature *284*, 555–556. https://doi.org/10.1038/284555a0.

81. Nagata, S. (2018). Apoptosis and clearance of apoptotic cells. Annu. Rev. Immunol. *36*, 489–517. https://doi.org/10.1146/annurev-immunol-042617-053010.

82. Sachet, M., Liang, Y.Y., and Oehler, R. (2017). The immune response to secondary necrotic cells. Apoptosis *22*, 1189–1204. https://doi.org/10.1007/s10495-017-1413-z.

83. Bociek, R.G., and Lunning, M. (2025). Tumor lysis syndrome. N. Engl. J. Med. *393*, 1104–1116. https://doi.org/10.1056/NEJMra2300923.

84. Silva, M.T. (2010). Secondary necrosis: the natural outcome of the complete apoptotic program. FEBS Lett. *584*, 4491–4499. https://doi.org/10.1016/j.febslet.2010.10.046.

85. Lee, B.L., Mirrashidi, K.M., Stowe, I.B., Kummerfeld, S.K., Watanabe, C., Haley, B., Cuellar, T.L., Reichelt, M., and Kayagaki, N. (2018). ASC- and caspase-8-dependent apoptotic pathway diverges from the NLRC4 inflammasome in macrophages. Sci. Rep. *8*, 3788. https://doi.org/10.1038/s41598-018-21998-3.

86. Tixeira, R., Shi, B., Parkes, M.A.F., Hodge, A.L., Caruso, S., Hulett, M.D., Baxter, A.A., Phan, T.K., and Poon, I.K.H. (2018). Gasdermin E does not limit apoptotic cell disassembly by promoting early onset of secondary necrosis in jurkat T cells and THP-1 monocytes. Front. Immunol. *9*, 2842. https://doi.org/10.3389/fimmu.2018.02842.

87. Strasser, A., Harris, A.W., Huang, D.C., Krammer, P.H., and Cory, S. (1995). Bcl-2 and Fas/APO-1 regulate distinct pathways to lymphocyte apoptosis. EMBO J. *14*, 6136–6147. https://doi.org/10.1002/j.1460-2075.1995.tb00304.x.

88. Green, D.R., and Reed, J.C. (1998). Mitochondria and apoptosis. Science *281*, 1309–1312. https://doi.org/10.1126/science.281.5381.1309.

89. Singh, R., Letai, A., and Sarosiek, K. (2019). Regulation of apoptosis in health and disease: the balancing act of BCL-2 family proteins. Nat. Rev. Mol. Cell Biol. *20*, 175–193. https://doi.org/10.1038/s41580-018-0089-8.

90. O'Neill, K.L., Huang, K., Zhang, J., Chen, Y., and Luo, X. (2016). Inactivation of prosurvival Bcl-2 proteins activates Bax/Bak through the outer mitochondrial membrane. Genes Dev. *30*, 973–988. https://doi.org/10.1101/gad.276725.115.

91. Zhang, Y., Tian, L., Huang, G., Ge, X., Kong, F., Wang, P., Xu, Y., and Shi, Y. (2025). Structural basis of BAX pore formation. Science *388*, eadv4314. https://doi.org/10.1126/science.adv4314.

92. Kischkel, F.C., Hellbardt, S., Behrmann, I., Germer, M., Pawlita, M., Krammer, P.H., and Peter, M.E. (1995). Cytotoxicity-dependent APO-1 (Fas/CD95)-associated proteins form a death-inducing signaling complex (DISC) with the receptor. EMBO J. *14*, 5579–5588. https://doi.org/10.1002/j.1460-2075.1995.tb00245.x.

93. Muzio, M., Chinnaiyan, A.M., Kischkel, F.C., O'Rourke, K., Shevchenko, A., Ni, J., Scaffidi, C., Bretz, J.D., Zhang, M., Gentz, R., et al. (1996). FLICE, a novel FADD-homologous ICE/CED-3-like protease, is recruited to the CD95 (Fas/APO-1) death–inducing signaling complex. Cell *85*, 817–827. https://doi.org/10.1016/S0092-8674(00)81266-0.

94. Boldin, M.P., Goncharov, T.M., Goltsev, Y.V., and Wallach, D. (1996). Involvement of MACH, a novel MORT1/FADD-interacting protease, in Fas/APO-1- and TNF receptor-induced cell death. Cell *85*, 803–815. https://doi.org/10.1016/S0092-8674(00)81265-9.

95. Li, H., Zhu, H., Xu, C.J., and Yuan, J. (1998). Cleavage of BID by caspase 8 mediates the mitochondrial damage in the Fas pathway of apoptosis. Cell *94*, 491–501. https://doi.org/10.1016/S0092-8674(00)81590-1.

96. Luo, X., Budihardjo, I., Zou, H., Slaughter, C., and Wang, X. (1998). Bid, a Bcl2 interacting protein, mediates cytochrome c release from mitochondria in response to activation of cell surface death receptors. Cell *94*, 481–490. https://doi.org/10.1016/S0092-8674(00)81589-5.

97. Jost, P.J., Grabow, S., Gray, D., McKenzie, M.D., Nachbur, U., Huang, D.C.S., Bouillet, P., Thomas, H.E., Borner, C., Silke, J., et al. (2009). XIAP discriminates between type I and type II FAS-induced apoptosis. Nature *460*, 1035–1039. https://doi.org/10.1038/nature08229.

98. Marsden, V.S., O'Connor, L., O'Reilly, L.A., Silke, J., Metcalf, D., Ekert, P.G., Huang, D.C.S., Cecconi, F., Kuida, K., Tomaselli, K.J., et al. (2002). Apoptosis initiated by Bcl-2-regulated caspase activation independently of the cytochrome c/Apaf-1/caspase-9 apoptosome. Nature *419*, 634–637. https://doi.org/10.1038/nature01101.

99. Egle, A., Harris, A.W., Bouillet, P., and Cory, S. (2004). Bim is a suppressor of Myc-induced mouse B cell leukemia. Proc. Natl. Acad. Sci. USA *101*, 6164–6169. https://doi.org/10.1073/pnas.0401471101.

100. Hemann, M.T., Zilfou, J.T., Zhao, Z., Burgess, D.J., Hannon, G.J., and Lowe, S.W. (2004). Suppression of tumorigenesis by the p53 target PUMA. Proc. Natl. Acad. Sci. USA *101*, 9333–9338. https://doi.org/10.1073/pnas.0403286101.

101. Michalak, E.M., Jansen, E.S., Happo, L., Cragg, M.S., Tai, L., Smyth, G.K., Strasser, A., Adams, J.M., and Scott, C.L. (2009). Puma and to a lesser extent Noxa are suppressors of Myc-induced lymphomagenesis. Cell Death Differ. *16*, 684–696. https://doi.org/10.1038/cdd.2008.195.

102. Scott, C.L., Schuler, M., Marsden, V.S., Egle, A., Pellegrini, M., Nesic, D., Gerondakis, S., Nutt, S.L., Green, D.R., and Strasser, A. (2004). Apaf-1 and caspase-9 do not act as tumor suppressors in myc-induced lymphomagenesis or mouse embryo fibroblast transformation. J. Cell Biol. *164*, 89–96. https://doi.org/10.1083/jcb.200310041.

103. Bouillet, P., Metcalf, D., Huang, D.C., Tarlinton, D.M., Kay, T.W., Köntgen, F., Adams, J.M., and Strasser, A. (1999). Proapoptotic Bcl-2 relative Bim required for certain apoptotic responses, leukocyte homeostasis, and to preclude autoimmunity. Science *286*, 1735–1738. https://doi.org/10.1126/science.286.5445.1735.

104. Villunger, A., Michalak, E.M., Coultas, L., Müllauer, F., Böck, G., Ausserlechner, M.J., Adams, J.M., and Strasser, A. (2003). p53- and drug-induced apoptotic responses mediated by BH3-only proteins puma and noxa. Science *302*, 1036–1038. https://doi.org/10.1126/science.1090072.

105. Kuroda, J., Puthalakath, H., Cragg, M.S., Kelly, P.N., Bouillet, P., Huang, D.C.S., Kimura, S., Ottmann, O.G., Druker, B.J., Villunger, A., et al. (2006). Bim and Bad mediate imatinib-induced killing of Bcr/Abl+ leukemic cells, and resistance due to their loss is overcome by a BH3 mimetic. Proc. Natl. Acad. Sci. USA *103*, 14907–14912. https://doi.org/10.1073/pnas.0606176103.

106. Glaser, S.P., Lee, E.F., Trounson, E., Bouillet, P., Wei, A., Fairlie, W.D., Izon, D.J., Zuber, J., Rappaport, A.R., Herold, M.J., et al. (2012). Anti-apoptotic Mcl-1 is essential for the development and sustained growth of acute myeloid leukemia. Genes Dev. *26*, 120–125. https://doi.org/10.1101/gad.182980.111.

107. Kelly, G.L., Grabow, S., Glaser, S.P., Fitzsimmons, L., Aubrey, B.J., Okamoto, T., Valente, L.J., Robati, M., Tai, L., Fairlie, W.D., et al. (2014). Targeting of MCL-1 kills MYC-driven mouse and human lymphomas even when they bear mutations in p53. Genes Dev. *28*, 58–70. https://doi.org/10.1101/gad.232009.113.

108. Oltersdorf, T., Elmore, S.W., Shoemaker, A.R., Armstrong, R.C., Augeri, D.J., Belli, B.A., Bruncko, M., Deckwerth, T.L., Dinges, J., Hajduk, P.J., et al. (2005). An inhibitor of Bcl-2 family proteins induces regression of solid tumours. Nature *435*, 677–681. https://doi.org/10.1038/nature03579.

109. Kotschy, A., Szlavik, Z., Murray, J., Davidson, J., Maragno, A.L., Le Toumelin-Braizat, G., Chanrion, M., Kelly, G.L., Gong, J.-N., Moujalled, D.M., et al. (2016). The MCL1 inhibitor S63845 is tolerable and effective in diverse cancer models. Nature *538*, 477–482. https://doi.org/10.1038/nature19830.

110. Strasser, A., Harris, A.W., and Cory, S. (1993). E mu-bcl-2 transgene facilitates spontaneous transformation of early pre-B and immunoglobulin-secreting cells but not T cells. Oncogene *8*, 1–9.

111. Mason, K.D., Lin, A., Robb, L., Josefsson, E.C., Henley, K.J., Gray, D.H.D., Kile, B.T., Roberts, A.W., Strasser, A., Huang, D.C.S., et al. (2013). Proapoptotic Bak and Bax guard against fatal systemic and organ-specific autoimmune disease. Proc. Natl. Acad. Sci. USA *110*, 2599–2604. https://doi.org/10.1073/pnas.1215097110.

112. Vandenberg, C.J., Waring, P., Strasser, A., and Cory, S. (2014). Plasmacytomagenesis in Eμ-v-abl transgenic mice is accelerated when apoptosis is restrained. Blood *124*, 1099–1109. https://doi.org/10.1182/blood-2014-04-570770.

113. Grabow, S., Kueh, A.J., Ke, F., Vanyai, H.K., Sheikh, B.N., Dengler, M.A., Chiang, W., Eccles, S., Smyth, I.M., Jones, L.K., et al. (2018). Subtle changes in the levels of BCL-2 proteins cause severe craniofacial abnormalities. Cell Rep. *24*, 3285–3295.e4. https://doi.org/10.1016/j.celrep.2018.08.048.

114. Straus, S.E., Jaffe, E.S., Puck, J.M., Dale, J.K., Elkon, K.B., Rösen-Wolff, A., Peters, A.M., Sneller, M.C., Hallahan, C.W., Wang, J., et al. (2001). The development of lymphomas in families with autoimmune lymphoproliferative syndrome with germline Fas mutations and defective lymphocyte apoptosis. Blood *98*, 194–200. https://doi.org/10.1182/blood.V98.1.194.

115. Zheng, Y., Chen, Y., Meng, X., Zhang, L., Ma, Y., Zhou, R., Fu, S., Chen, H., Xuanyuan, X., Jiang, R., et al. (2025). FADD functions as an oncogene in Chr11q13.3-amplified head and neck squamous cell carcinoma. Cancer Res. *85*, 1909–1927. https://doi.org/10.1158/0008-5472.CAN-24-2562.

116. Friesen, C., Herr, I., Krammer, P.H., and Debatin, K.M. (1996). Involvement of the CD95 (APO-1/FAS) receptor/ligand system in drug-induced apoptosis in leukemia cells. Nat. Med. *2*, 574–577. https://doi.org/10.1038/nm0596-574.

117. Newton, K., and Strasser, A. (2000). Ionizing radiation and chemotherapeutic drugs induce apoptosis in lymphocytes in the absence of Fas or FADD/MORT1 signaling. Implications for cancer therapy. J. Exp. Med. *191*, 195–200. https://doi.org/10.1084/jem.191.1.195.

118. Pelengaris, S., Khan, M., and Evan, G.I. (2002). Suppression of Myc-induced apoptosis in beta cells exposes multiple oncogenic properties of Myc and triggers carcinogenic progression. Cell *109*, 321–334. https://doi.org/10.1016/S0092-8674(02)00738-9.

119. de La Coste, A., Mignon, A., Fabre, M., Gilbert, E., Porteu, A., Van Dyke, T., Kahn, A., and Perret, C. (1999). Paradoxical inhibition of c-myc-induced carcinogenesis by Bcl-2 in transgenic mice. Cancer Res. *59*, 5017–5022.

120. Ichim, G., Lopez, J., Ahmed, S.U., Muthalagu, N., Giampazolias, E., Delgado, M.E., Haller, M., Riley, J.S., Mason, S.M., Athineos, D., et al. (2015). Limited mitochondrial permeabilization causes DNA damage and genomic instability in the absence of cell death. Mol. Cell *57*, 860–872. https://doi.org/10.1016/j.molcel.2015.01.018.

121. Häcker, G., and Haimovici, A. (2023). Sub-lethal signals in the mitochondrial apoptosis apparatus: pernicious by-product or physiological event? Cell Death Differ. *30*, 250–257. https://doi.org/10.1038/s41418-022-01058-0.

122. McArthur, K., Whitehead, L.W., Heddleston, J.M., Li, L., Padman, B.S., Oorschot, V., Geoghegan, N.D., Chappaz, S., Davidson, S., San Chin, H., et al. (2018). BAK/BAX macropores facilitate mitochondrial herniation and mtDNA efflux during apoptosis. Science *359*, eaao6047. https://doi.org/10.1126/science.aao6047.

123. Holzgruber, J., Martins, C., Kulcsar, Z., Duplaine, A., Rasbach, E., Migayron, L., Singh, P., Statham, E., Landsberg, J., Boniface, K., et al. (2024). Type I interferon signaling induces melanoma cell-intrinsic PD-1 and its inhibition antagonizes immune checkpoint blockade. Nat. Commun. *15*, 7165. https://doi.org/10.1038/s41467-024-51496-2.

124. Michalak, E.M., Vandenberg, C.J., Delbridge, A.R.D., Wu, L., Scott, C.L., Adams, J.M., and Strasser, A. (2010). Apoptosis-promoted tumorigenesis: gamma-irradiation-induced thymic lymphomagenesis requires Puma-driven leukocyte death. Genes Dev. *24*, 1608–1613. https://doi.org/10.1101/gad.1940110.

125. Labi, V., Erlacher, M., Krumschnabel, G., Manzl, C., Tzankov, A., Pinon, J., Egle, A., and Villunger, A. (2010). Apoptosis of leukocytes triggered by acute DNA damage promotes lymphoma formation. Genes Dev. *24*, 1602–1607. https://doi.org/10.1101/gad.1940210.

126. Healy, M.E., Boege, Y., Hodder, M.C., Böhm, F., Malehmir, M., Scherr, A.-L., Jetzer, J., Chan, L.K., Parrotta, R., Jacobs, K., et al. (2020). MCL1 is required for maintenance of intestinal homeostasis and prevention of carcinogenesis in mice. Gastroenterology *159*, 183–199. https://doi.org/10.1053/j.gastro.2020.03.017.

127. Wallach, D., Kang, T.B., Dillon, C.P., and Green, D.R. (2016). Programmed necrosis in inflammation: Toward identification of the effector molecules. Science *352*, aaf2154. https://doi.org/10.1126/science.aaf2154.

128. Vercammen, D., Beyaert, R., Denecker, G., Goossens, V., Van Loo, G., Declercq, W., Grooten, J., Fiers, W., and Vandenabeele, P. (1998). Inhibition of caspases increases the sensitivity of L929 cells to necrosis mediated by tumor necrosis factor. J. Exp. Med. *187*, 1477–1485. https://doi.org/10.1084/jem.187.9.1477.

129. Degterev, A., Huang, Z., Boyce, M., Li, Y., Jagtap, P., Mizushima, N., Cuny, G.D., Mitchison, T.J., Moskowitz, M.A., and Yuan, J. (2005). Chemical inhibitor of nonapoptotic cell death with therapeutic potential for ischemic brain injury. Nat. Chem. Biol. *1*, 112–119. https://doi.org/10.1038/nchembio711.

130. Vercammen, D., Brouckaert, G., Denecker, G., Van de Craen, M., Declercq, W., Fiers, W., and Vandenabeele, P. (1998). Dual signaling of the Fas receptor: initiation of both apoptotic and necrotic cell death pathways. J. Exp. Med. *188*, 919–930. https://doi.org/10.1084/jem.188.5.919.

131. Degterev, A., Hitomi, J., Germscheid, M., Ch'en, I.L., Korkina, O., Teng, X., Abbott, D., Cuny, G.D., Yuan, C., Wagner, G., et al. (2008). Identification of RIP1 kinase as a specific cellular target of necrostatins. Nat. Chem. Biol. *4*, 313–321. https://doi.org/10.1038/nchembio.83.

132. Cho, Y.S., Challa, S., Moquin, D., Genga, R., Ray, T.D., Guildford, M., and Chan, F.K.-M. (2009). Phosphorylation-driven assembly of the RIP1-RIP3 complex regulates programmed necrosis and virus-induced inflammation. Cell *137*, 1112–1123. https://doi.org/10.1016/j.cell.2009.05.037.

133. He, S., Wang, L., Miao, L., Wang, T., Du, F., Zhao, L., and Wang, X. (2009). Receptor interacting protein kinase-3 determines cellular necrotic response to TNF-alpha. Cell *137*, 1100–1111. https://doi.org/10.1016/j.cell.2009.05.021.

134. Zhang, D.-W., Shao, J., Lin, J., Zhang, N., Lu, B.-J., Lin, S.-C., Dong, M.-Q., and Han, J. (2009). RIP3, an energy metabolism regulator that switches TNF-induced cell death from apoptosis to necrosis. Science *325*, 332–336. https://doi.org/10.1126/science.1172308.

135. Sun, L., Wang, H., Wang, Z., He, S., Chen, S., Liao, D., Wang, L., Yan, J., Liu, W., Lei, X., et al. (2012). Mixed lineage kinase domain-like protein mediates necrosis signaling downstream of RIP3 kinase. Cell *148*, 213–227. https://doi.org/10.1016/j.cell.2011.11.031.

136. Zhao, J., Jitkaew, S., Cai, Z., Choksi, S., Li, Q., Luo, J., and Liu, Z.-G. (2012). Mixed lineage kinase domain-like is a key receptor interacting protein 3 downstream component of TNF-induced necrosis. Proc. Natl. Acad. Sci. USA *109*, 5322–5327. https://doi.org/10.1073/pnas.1200012109.

137. Baik, J.Y., Liu, Z., Jiao, D., Kwon, H.J., Yan, J., Kadigamuwa, C., Choe, M., Lake, R., Kruhlak, M., Tandon, M., et al. (2021). ZBP1 not RIPK1 mediates tumor necroptosis in breast cancer. Nat. Commun. *12*, 2666. https://doi.org/10.1038/s41467-021-23004-3.

138. Koo, G.B., Morgan, M.J., Lee, D.G., Kim, W.J., Yoon, J.H., Koo, J.S., Kim, S.I., Kim, S.J., Son, M.K., Hong, S.S., et al. (2015). Methylation-dependent loss of RIP3 expression in cancer represses programmed necrosis in response to chemotherapeutics. Cell Res. *25*, 707–725. https://doi.org/10.1038/cr.2015.56.

139. Agrawal, D., Cisarova, K., Vosberg, S., Allmendinger, F., Munkhbaatar, E., Dandachi, N., Fernandez Hernandez, F.J., Tonietto, M., Jäger, V., Anton, M., et al. (2026). Aberrant methylation limits antitumoral inflammation in lung adenocarcinoma by restricting RIPK3 expression. Sci. Adv. *12*, eadz9227. https://doi.org/10.1126/sciadv.adz9227.

140. Ofengeim, D., and Yuan, J. (2013). Regulation of RIP1 kinase signalling at the crossroads of inflammation and cell death. Nat. Rev. Mol. Cell Biol. *14*, 727–736. https://doi.org/10.1038/nrm3683.

141. Li, W., Shan, B., Zou, C., Wang, H., Zhang, M.M., Zhu, H., Naito, M.G., Xu, D., Manuel, V.J., Mifflin, L., et al. (2022). Nuclear RIPK1 promotes chromatin remodeling to mediate inflammatory response. Cell Res. *32*, 621–637. https://doi.org/10.1038/s41422-022-00673-3.

142. Moriwaki, K., Balaji, S., McQuade, T., Malhotra, N., Kang, J., and Chan, F.K.-M. (2014). The necroptosis adaptor RIPK3 promotes injury-induced cytokine expression and tissue repair. Immunity *41*, 567–578. https://doi.org/10.1016/j.immuni.2014.09.016.

143. Vucur, M., Ghallab, A., Schneider, A.T., Adili, A., Cheng, M., Castoldi, M., Singer, M.T., Büttner, V., Keysberg, L.S., Küsgens, L., et al. (2023). Sublethal necroptosis signaling promotes inflammation and liver cancer.

Immunity *56*, 1578–1595.e8. https://doi.org/10.1016/j.immuni.2023.05.017.

144. Murphy, J.M., Czabotar, P.E., Hildebrand, J.M., Lucet, I.S., Zhang, J.-G., Alvarez-Diaz, S., Lewis, R., Lalaoui, N., Metcalf, D., Webb, A.I., et al. (2013). The pseudokinase MLKL mediates necroptosis via a molecular switch mechanism. Immunity *39*, 443–453. https://doi.org/10.1016/j.immuni.2013.06.018.

145. Dondelinger, Y., Declercq, W., Montessuit, S., Roelandt, R., Goncalves, A., Bruggeman, I., Hulpiau, P., Weber, K., Sehon, C.A., Marquis, R.W., et al. (2014). MLKL compromises plasma membrane integrity by binding to phosphatidylinositol phosphates. Cell Rep. *7*, 971–981. https://doi.org/10.1016/j.celrep.2014.04.026.

146. Wang, H., Sun, L., Su, L., Rizo, J., Liu, L., Wang, L.-F., Wang, F.-S., and Wang, X. (2014). Mixed lineage kinase domain-like protein MLKL causes necrotic membrane disruption upon phosphorylation by RIP3. Mol. Cell *54*, 133–146. https://doi.org/10.1016/j.molcel.2014.03.003.

147. Quarato, G., Guy, C.S., Grace, C.R., Llambi, F., Nourse, A., Rodriguez, D.A., Wakefield, R., Frase, S., Moldoveanu, T., and Green, D.R. (2016). Sequential engagement of distinct MLKL phosphatidylinositol-binding sites executes necroptosis. Mol. Cell *61*, 589–601. https://doi.org/10.1016/j.molcel.2016.01.011.

148. Dovey, C.M., Diep, J., Clarke, B.P., Hale, A.T., McNamara, D.E., Guo, H., Brown, N.W., Jr., Cao, J.Y., Grace, C.R., Gough, P.J., et al. (2018). MLKL requires the inositol phosphate code to execute necroptosis. Mol. Cell *70*, 936–948.e7. https://doi.org/10.1016/j.molcel.2018.05.010.

149. Flores-Romero, H., Ros, U., and Garcia-Saez, A.J. (2020). Pore formation in regulated cell death. EMBO J. *39*, e105753. https://doi.org/10.15252/embj.2020105753.

150. Vandenabeele, P., Bultynck, G., and Savvides, S.N. (2023). Pore-forming proteins as drivers of membrane permeabilization in cell death pathways. Nat. Rev. Mol. Cell Biol. *24*, 312–333. https://doi.org/10.1038/s41580-022-00564-w.

151. Martens, S., Bridelance, J., Roelandt, R., Vandenabeele, P., and Takahashi, N. (2021). MLKL in cancer: more than a necroptosis regulator. Cell Death Differ. *28*, 1757–1772. https://doi.org/10.1038/s41418-021-00785-0.

152. Yoon, S., Kovalenko, A., Bogdanov, K., and Wallach, D. (2017). MLKL, the protein that mediates necroptosis, also regulates endosomal trafficking and extracellular vesicle generation. Immunity *47*, 51–65.e7. https://doi.org/10.1016/j.immuni.2017.06.001.

153. Gong, Y.-N., Guy, C., Olauson, H., Becker, J.U., Yang, M., Fitzgerald, P., Linkermann, A., and Green, D.R. (2017). ESCRT-III acts downstream of MLKL to regulate necroptotic cell death and its consequences. Cell *169*, 286–300.e16. https://doi.org/10.1016/j.cell.2017.03.020.

154. Zhan, Q., Jeon, J., Li, Y., Huang, Y., Xiong, J., Wang, Q., Xu, T.-L., Li, Y., Ji, F.-H., Du, G., et al. (2022). CAMK2/CaMKII activates MLKL in short-term starvation to facilitate autophagic flux. Autophagy *18*, 726–744. https://doi.org/10.1080/15548627.2021.1954348.

155. Weber, K., Roelandt, R., Bruggeman, I., Estornes, Y., and Vandenabeele, P. (2018). Nuclear RIPK3 and MLKL contribute to cytosolic necrosome formation and necroptosis. Commun. Biol. *1*, 6. https://doi.org/10.1038/s42003-017-0007-1.

156. Wang, Y., Wei, W., Zhang, Y., Miao, J., Bao, X., and Lu, C. (2025). MLKL as an emerging machinery for modulating organelle dynamics: regulatory mechanisms, pathophysiological significance, and targeted therapeutics. Front. Pharmacol. *16*, 1512968. https://doi.org/10.3389/fphar.2025.1512968.

157. Clucas, J., and Meier, P. (2023). Roles of RIPK1 as a stress sentinel coordinating cell survival and immunogenic cell death. Nat. Rev. Mol. Cell Biol. *24*, 835–852. https://doi.org/10.1038/s41580-023-00623-w.

158. Kaiser, W.J., Sridharan, H., Huang, C., Mandal, P., Upton, J.W., Gough, P.J., Sehon, C.A., Marquis, R.W., Bertin, J., and Mocarski, E.S. (2013). Toll-like receptor 3-mediated necrosis via TRIF, RIP3, and MLKL. J. Biol. Chem. *288*, 31268–31279. https://doi.org/10.1074/jbc.M113.462341.

159. Lin, J., Kumari, S., Kim, C., Van, T.-M., Wachsmuth, L., Polykratis, A., and Pasparakis, M. (2016). RIPK1 counteracts ZBP1-mediated necroptosis to inhibit inflammation. Nature *540*, 124–128. https://doi.org/10.1038/nature20558.

160. Newton, K., Wickliffe, K.E., Maltzman, A., Dugger, D.L., Strasser, A., Pham, V.C., Lill, J.R., Roose-Girma, M., Warming, S., Solon, M., et al. (2016). RIPK1 inhibits ZBP1-driven necroptosis during development. Nature *540*, 129–133. https://doi.org/10.1038/nature20559.

161. Ishizuka, J.J., Manguso, R.T., Cheruiyot, C.K., Bi, K., Panda, A., Iracheta-Vellve, A., Miller, B.C., Du, P.P., Yates, K.B., Dubrot, J., et al. (2019). Loss of ADAR1 in tumours overcomes resistance to immune checkpoint blockade. Nature *565*, 43–48. https://doi.org/10.1038/s41586-018-0768-9.

162. Zhang, T., Yin, C., Fedorov, A., Qiao, L., Bao, H., Beknazarov, N., Wang, S., Gautam, A., Williams, R.M., Crawford, J.C., et al. (2022). ADAR1 masks the cancer immunotherapeutic promise of ZBP1-driven necroptosis. Nature *606*, 594–602. https://doi.org/10.1038/s41586-022-04753-7.

163. Jiao, H., Wachsmuth, L., Wolf, S., Lohmann, J., Nagata, M., Kaya, G.G., Oikonomou, N., Kondylis, V., Rogg, M., Diebold, M., et al. (2022). ADAR1 averts fatal type I interferon induction by ZBP1. Nature *607*, 776–783. https://doi.org/10.1038/s41586-022-04878-9.

164. de Reuver, R., Verdonck, S., Dierick, E., Nemegeer, J., Hessmann, E., Ahmad, S., Jans, M., Blancke, G., Van Nieuwerburgh, F., Botzki, A., et al. (2022). ADAR1 prevents autoinflammation by suppressing spontaneous ZBP1 activation. Nature *607*, 784–789. https://doi.org/10.1038/s41586-022-04974-w.

165. Karki, R., Sundaram, B., Sharma, B.R., Lee, S., Malireddi, R.K.S., Nguyen, L.N., Christgen, S., Zheng, M., Wang, Y., Samir, P., et al. (2021). ADAR1 restricts ZBP1-mediated immune response and PANoptosis to promote tumorigenesis. Cell Rep. *37*, 109858. https://doi.org/10.1016/j.celrep.2021.109858.

166. Wang, Q., Wu, Z., Yang, Y., Yang, C., Deng, H., Zhou, X., Xu, S., Wu, L., Hong, X., Yu, X., et al. (2025). ZBP1 pathway promotes tumor immunogenicity in the combination of anti-HER2 therapy and epigenetic therapy. Cell Rep. *44*, 116314. https://doi.org/10.1016/j.celrep.2025.116314.

167. Karki, R., Lee, S., Mall, R., Pandian, N., Wang, Y., Sharma, B.R., Malireddi, R.S., Yang, D., Trifkovic, S., Steele, J.A., et al. (2022). ZBP1-dependent inflammatory cell death, PANoptosis, and cytokine storm disrupt IFN therapeutic efficacy during coronavirus infection. Sci. Immunol. *7*, eabo6294. https://doi.org/10.1126/sciimmunol.abo6294.

168. Legrand, A.J., Konstantinou, M., Goode, E.F., and Meier, P. (2019). The diversification of cell death and immunity: Memento Mori. Mol. Cell *76*, 232–242. https://doi.org/10.1016/j.molcel.2019.09.006.

169. Van Hoecke, L., Van Lint, S., Roose, K., Van Parys, A., Vandenabeele, P., Grooten, J., Tavernier, J., De Koker, S., and Saelens, X. (2018). Treatment with mRNA coding for the necroptosis mediator MLKL induces antitumor immunity directed against neo-epitopes. Nat. Commun. *9*, 3417. https://doi.org/10.1038/s41467-018-05979-8.

170. Aaes, T.L., Verschuere, H., Kaczmarek, A., Heyndrickx, L., Wiernicki, B., Delrue, I., De Craene, B., Taminau, J., Delvaeye, T., Bertrand, M.J.M., et al. (2020). Immunodominant AH1 antigen-deficient necroptotic, but not apoptotic, Murine cancer cells induce antitumor protection. J. Immunol. *204*, 775–787. https://doi.org/10.4049/jimmunol.1900072.

171. Aaes, T.L., Kaczmarek, A., Delvaeye, T., De Craene, B., De Koker, S., Heyndrickx, L., Delrue, I., Taminau, J., Wiernicki, B., De Groote, P., et al. (2016). Vaccination with necroptotic cancer cells induces efficient anti-tumor immunity. Cell Rep. *15*, 274–287. https://doi.org/10.1016/j.celrep.2016.03.037.

172. Yatim, N., Jusforgues-Saklani, H., Orozco, S., Schulz, O., Barreira da Silva, R., Reis e Sousa, C., Green, D.R., Oberst, A., and Albert, M.L. (2015). RIPK1 and NF-kappaB signaling in dying cells determines

cross-priming of CD8(+) T cells. Science *350*, 328–334. https://doi.org/10.1126/science.aad0395.

173. Newton, K., Wickliffe, K.E., Dugger, D.L., Maltzman, A., Roose-Girma, M., Dohse, M., Kőműves, L., Webster, J.D., and Dixit, V.M. (2019). Cleavage of RIPK1 by caspase-8 is crucial for limiting apoptosis and necroptosis. Nature *574*, 428–431. https://doi.org/10.1038/s41586-019-1548-x.

174. Vringer, E., and Tait, S.W.G. (2023). Mitochondria and cell death-associated inflammation. Cell Death Differ. *30*, 304–312. https://doi.org/10.1038/s41418-022-01094-w.

175. Riley, J.S., Quarato, G., Cloix, C., Lopez, J., O'Prey, J., Pearson, M., Chapman, J., Sesaki, H., Carlin, L.M., Passos, J.F., et al. (2018). Mitochondrial inner membrane permeabilisation enables mtDNA release during apoptosis. EMBO J. *37*, e99238. https://doi.org/10.15252/embj.201899238.

176. Kelepouras, K., Saggau, J., Bonasera, D., Kiefer, C., Locci, F., Rakhsh-Khorshid, H., Grauvogel, L., Varanda, A.B., Peifer, M., Loricchio, E., et al. (2025). STING induces ZBP1-mediated necroptosis independently of TNFR1 and FADD. Nature *647*, 735–746. https://doi.org/10.1038/s41586-025-09536-4.

177. Ning, X., Wang, Y., Jing, M., Sha, M., Lv, M., Gao, P., Zhang, R., Huang, X., Feng, J.-M., and Jiang, Z. (2019). Apoptotic caspases suppress type I interferon production via the cleavage of cGAS, MAVS, and IRF3. Mol. Cell *74*, 19–31.e7. https://doi.org/10.1016/j.molcel.2019.02.013.

178. White, M.J., McArthur, K., Metcalf, D., Lane, R.M., Cambier, J.C., Herold, M.J., van Delft, M.F., Bedoui, S., Lessene, G., Ritchie, M.E., et al. (2014). Apoptotic caspases suppress mtDNA-induced STING-mediated type I IFN production. Cell *159*, 1549–1562. https://doi.org/10.1016/j.cell.2014.11.036.

179. Bock, F.J., and Tait, S.W.G. (2020). Mitochondria as multifaceted regulators of cell death. Nat. Rev. Mol. Cell Biol. *21*, 85–100. https://doi.org/10.1038/s41580-019-0173-8.

180. Moulin, M., Anderton, H., Voss, A.K., Thomas, T., Wong, W.W.-L., Bankovacki, A., Feltham, R., Chau, D., Cook, W.D., Silke, J., et al. (2012). IAPs limit activation of RIP kinases by TNF receptor 1 during development. EMBO J. *31*, 1679–1691. https://doi.org/10.1038/emboj.2012.18.

181. Annibaldi, A., Wicky John, S., Vanden Berghe, T., Swatek, K.N., Ruan, J., Liccardi, G., Bianchi, K., Elliott, P.R., Choi, S.M., Van Coillie, S., et al. (2018). Ubiquitin-mediated regulation of RIPK1 kinase activity independent of IKK and MK2. Mol. Cell *69*, 566–580.e5. https://doi.org/10.1016/j.molcel.2018.01.027.

182. Morrish, E., Brumatti, G., and Silke, J. (2020). Future therapeutic directions for Smac-Mimetics. Cells *9*, 406. https://doi.org/10.3390/cells9020406.

183. Silke, J., and Meier, P. (2013). Inhibitor of apoptosis (IAP) proteins-modulators of cell death and inflammation. Cold Spring Harb. Perspect. Biol. *5*, a008730. https://doi.org/10.1101/cshperspect.a008730.

184. Brumatti, G., Ma, C., Lalaoui, N., Nguyen, N.-Y., Navarro, M., Tanzer, M.C., Richmond, J., Ghisi, M., Salmon, J.M., Silke, N., et al. (2016). The caspase-8 inhibitor emricasan combines with the SMAC mimetic birinapant to induce necroptosis and treat acute myeloid leukemia. Sci. Transl. Med. *8*, 339ra69. https://doi.org/10.1126/scitranslmed.aad3099.

185. Eggermont, A.M., Schraffordt Koops, H., Liénard, D., Kroon, B.B., van Geel, A.N., Hoekstra, H.J., and Lejeune, F.J. (1996). Isolated limb perfusion with high-dose tumor necrosis factor-alpha in combination with interferon-gamma and melphalan for nonresectable extremity soft tissue sarcomas: a multicenter trial. J. Clin. Oncol. *14*, 2653–2665. https://doi.org/10.1200/JCO.1996.14.10.2653.

186. Lincoln, F.A., Imig, D., Boccellato, C., Juric, V., Noonan, J., Kontermann, R.E., Allgöwer, F., Murphy, B.M., and Rehm, M. (2018). Sensitization of glioblastoma cells to TRAIL-induced apoptosis by IAP- and Bcl-2 antagonism. Cell Death Dis. *9*, 1112. https://doi.org/10.1038/s41419-018-1160-2.

187. Maelfait, J., and Rehwinkel, J. (2023). The Z-nucleic acid sensor ZBP1 in health and disease. J. Exp. Med. *220*, e20221156. https://doi.org/10.1084/jem.20221156.

188. Wang, X., Li, J., Zhu, Y., Shen, H., Ding, J., Zeng, T., Min, W., Liang, S.-Q., Huang, L., Shi, Z., et al. (2025). Targeting ADAR1 with a small molecule for the treatment of prostate cancer. Nat. Cancer *6*, 474–492. https://doi.org/10.1038/s43018-025-00907-4.

189. Smoak, C.N., Gardner, E.N., Chua, R.N., and Cottrell, K.A. (2025). ZYS-1 is not an ADAR1 inhibitor. RNA *31*, 1703–1711. https://doi.org/10.1261/rna.080721.125.

190. Diepstraten, S.T., Yuan, Y., La Marca, J.E., Young, S., Chang, C., Whelan, L., Ross, A.M., Fischer, K.C., Pomilio, G., Morris, R., et al. (2024). Putting the STING back into BH3-mimetic drugs for TP53-mutant blood cancers. Cancer Cell *42*, 850–868.e9. https://doi.org/10.1016/j.ccell.2024.04.004.

191. Yang, T., Wang, G., Zhang, M., Hu, X., Li, Q., Yun, F., Xing, Y., Song, X., Zhang, H., Hu, G., et al. (2023). Triggering endogenous Z-RNA sensing for anti-tumor therapy through ZBP1-dependent necroptosis. Cell Rep. *42*, 113377. https://doi.org/10.1016/j.celrep.2023.113377.

192. Mannion, J., Gifford, V., Bellenie, B., Fernando, W., Ramos Garcia, L., Wilson, R., John, S.W., Udainiya, S., Patin, E.C., Tiu, C., et al. (2024). A RIPK1-specific PROTAC degrader achieves potent antitumor activity by enhancing immunogenic cell death. Immunity *57*, 1514–1532.e15. https://doi.org/10.1016/j.immuni.2024.04.025.

193. Jayathilaka, B., Mian, F., Franchini, F., Au-Yeung, G., and IJzerman, M. (2025). Cancer and treatment specific incidence rates of immune-related adverse events induced by immune checkpoint inhibitors: a systematic review. Br. J. Cancer *132*, 51–57. https://doi.org/10.1038/s41416-024-02887-1.

194. Colbert, L.E., Fisher, S.B., Hardy, C.W., Hall, W.A., Saka, B., Shelton, J.W., Petrova, A.V., Warren, M.D., Pantazides, B.G., Gandhi, K., et al. (2013). Pronecrotic mixed lineage kinase domain-like protein expression is a prognostic biomarker in patients with early-stage resected pancreatic adenocarcinoma. Cancer *119*, 3148–3155. https://doi.org/10.1002/cncr.28144.

195. Li, X., Guo, J., Ding, A.-P., Qi, W.-W., Zhang, P.-H., Lv, J., Qiu, W.-S., and Sun, Z.-Q. (2017). Association of mixed lineage kinase domain-like protein expression with prognosis in patients with colon cancer. Technol. Cancer Res. Treat. *16*, 428–434. https://doi.org/10.1177/1533034616655909.

196. Ruan, J., Mei, L., Zhu, Q., Shi, G., and Wang, H. (2015). Mixed lineage kinase domain-like protein is a prognostic biomarker for cervical squamous cell cancer. Int. J. Clin. Exp. Pathol. *8*, 15035–15038.

197. Sarhan, J., Liu, B.C., Muendlein, H.I., Weindel, C.G., Smirnova, I., Tang, A.Y., Ilyukha, V., Sorokin, M., Buzdin, A., Fitzgerald, K.A., et al. (2019). Constitutive interferon signaling maintains critical threshold of MLKL expression to license necroptosis. Cell Death Differ. *26*, 332–347. https://doi.org/10.1038/s41418-018-0122-7.

198. Ding, Z., Wang, R., Li, Y., and Wang, X. (2025). MLKL activates the cGAS-STING pathway by releasing mitochondrial DNA upon necroptosis induction. Mol. Cell *85*, 2610–2625.e5. https://doi.org/10.1016/j.molcel.2025.06.005.

199. Mandal, P., Berger, S.B., Pillay, S., Moriwaki, K., Huang, C., Guo, H., Lich, J.D., Finger, J., Kasparcova, V., Votta, B., et al. (2014). RIP3 induces apoptosis independent of pronecrotic kinase activity. Mol. Cell *56*, 481–495. https://doi.org/10.1016/j.molcel.2014.10.021.

200. Newton, K., Dugger, D.L., Wickliffe, K.E., Kapoor, N., de Almagro, M.C., Vucic, D., Komuves, L., Ferrando, R.E., French, D.M., Webster, J., et al. (2014). Activity of protein kinase RIPK3 determines whether cells die by necroptosis or apoptosis. Science *343*, 1357–1360. https://doi.org/10.1126/science.1249361.

201. Lawlor, K.E., Khan, N., Mildenhall, A., Gerlic, M., Croker, B.A., D'Cruz, A.A., Hall, C., Kaur Spall, S., Anderton, H., Masters, S.L., et al. (2015). RIPK3 promotes cell death and NLRP3 inflammasome activation in the

absence of MLKL. Nat. Commun. *6*, 6282. https://doi.org/10.1038/ncomms7282.

202. Alvarez-Diaz, S., Dillon, C.P., Lalaoui, N., Tanzer, M.C., Rodriguez, D.A., Lin, A., Lebois, M., Hakem, R., Josefsson, E.C., O'Reilly, L.A., et al. (2016). The pseudokinase MLKL and the kinase RIPK3 have distinct roles in autoimmune disease caused by loss of death-receptor-induced apoptosis. Immunity *45*, 513–526. https://doi.org/10.1016/j.immuni.2016.07.016.

203. Mandal, R., Barrón, J.C., Kostova, I., Becker, S., and Strebhardt, K. (2020). Caspase-8: The double-edged sword. Biochim. Biophys. Acta Rev. Cancer *1873*, 188357. https://doi.org/10.1016/j.bbcan.2020.188357.

204. Fritsch, M., Günther, S.D., Schwarzer, R., Albert, M.-C., Schorn, F., Werthenbach, J.P., Schiffmann, L.M., Stair, N., Stocks, H., Seeger, J.M., et al. (2019). Caspase-8 is the molecular switch for apoptosis, necroptosis and pyroptosis. Nature *575*, 683–687. https://doi.org/10.1038/s41586-019-1770-6.

205. O'Donnell, M.A., Perez-Jimenez, E., Oberst, A., Ng, A., Massoumi, R., Xavier, R., Green, D.R., and Ting, A.T. (2011). Caspase 8 inhibits programmed necrosis by processing CYLD. Nat. Cell Biol. *13*, 1437–1442. https://doi.org/10.1038/ncb2362.

206. He, S., Liang, Y., Shao, F., and Wang, X. (2011). Toll-like receptors activate programmed necrosis in macrophages through a receptor-interacting kinase-3-mediated pathway. Proc. Natl. Acad. Sci. USA *108*, 20054–20059. https://doi.org/10.1073/pnas.1116302108.

207. Solon, M., Ge, N., Hambro, S., Haller, S., Jiang, J., Baca, M., Preston, J., Maltzman, A., Wickliffe, K.E., Liang, Y., et al. (2024). ZBP1 and TRIF trigger lethal necroptosis in mice lacking caspase-8 and TNFR1. Cell Death Differ. *31*, 672–682. https://doi.org/10.1038/s41418-024-01286-6.

208. Yang, D., Liang, Y., Zhao, S., Ding, Y., Zhuang, Q., Shi, Q., Ai, T., Wu, S.-Q., and Han, J. (2020). ZBP1 mediates interferon-induced necroptosis. Cell. Mol. Immunol. *17*, 356–368. https://doi.org/10.1038/s41423-019-0237-x.

209. Schwarzer, R., Jiao, H., Wachsmuth, L., Tresch, A., and Pasparakis, M. (2020). FADD and caspase-8 regulate gut homeostasis and inflammation by controlling MLKL- and GSDMD-mediated death of intestinal epithelial cells. Immunity *52*, 978–993.e6. https://doi.org/10.1016/j.immuni.2020.04.002.

210. Kist, M., and Vucic, D. (2021). Cell death pathways: intricate connections and disease implications. EMBO J. *40*, e106700. https://doi.org/10.15252/embj.2020106700.

211. Zeng, Q., Ma, X., Song, Y., Chen, Q., Jiao, Q., and Zhou, L. (2022). Targeting regulated cell death in tumor nanomedicines. Theranostics *12*, 817–841. https://doi.org/10.7150/thno.67932.

212. Howard, S.C., Avagyan, A., Workeneh, B., and Pui, C.-H. (2024). Tumour lysis syndrome. Nat. Rev. Dis. Primers *10*, 58. https://doi.org/10.1038/s41572-024-00542-w.

213. Tufail, M., Jiang, C.-H., and Li, N. (2025). Immune evasion in cancer: mechanisms and cutting-edge therapeutic approaches. Signal Transduct. Target. Ther. *10*, 227. https://doi.org/10.1038/s41392-025-02280-1.

214. Richards, C.H., Mohammed, Z., Qayyum, T., Horgan, P.G., and McMillan, D.C. (2011). The prognostic value of histological tumor necrosis in solid organ malignant disease: a systematic review. Future Oncol. *7*, 1223–1235. https://doi.org/10.2217/fon.11.99.

215. Friedlander, A.M. (1986). Macrophages are sensitive to anthrax lethal toxin through an acid-dependent process. J. Biol. Chem. *261*, 7123–7126. https://doi.org/10.1016/S0021-9258(17)38364-3.

216. Zychlinsky, A., Prevost, M.C., and Sansonetti, P.J. (1992). Shigella flexneri induces apoptosis in infected macrophages. Nature *358*, 167–169. https://doi.org/10.1038/358167a0.

217. Brennan, M.A., and Cookson, B.T. (2000). Salmonella induces macrophage death by caspase-1-dependent necrosis. Mol. Microbiol. *38*, 31–40. https://doi.org/10.1046/j.1365-2958.2000.02103.x.

218. Cookson, B.T., and Brennan, M.A. (2001). Pro-inflammatory programmed cell death. Trends Microbiol. *9*, 113–114. https://doi.org/10.1016/S0966-842X(00)01936-3.

219. Bergsbaken, T., Fink, S.L., and Cookson, B.T. (2009). Pyroptosis: host cell death and inflammation. Nat. Rev. Microbiol. *7*, 99–109. https://doi.org/10.1038/nrmicro2070.

220. Martinon, F., Burns, K., and Tschopp, J. (2002). The inflammasome: a molecular platform triggering activation of inflammatory caspases and processing of proIL-beta. Mol. Cell *10*, 417–426. https://doi.org/10.1016/S1097-2765(02)00599-3.

221. Kayagaki, N., Warming, S., Lamkanfi, M., Vande Walle, L., Louie, S., Dong, J., Newton, K., Qu, Y., Liu, J., Heldens, S., et al. (2011). Non-canonical inflammasome activation targets caspase-11. Nature *479*, 117–121. https://doi.org/10.1038/nature10558.

222. Lamkanfi, M., and Dixit, V.M. (2014). Mechanisms and functions of inflammasomes. Cell *157*, 1013–1022. https://doi.org/10.1016/j.cell.2014.04.007.

223. Kayagaki, N., Stowe, I.B., Lee, B.L., O'Rourke, K., Anderson, K., Warming, S., Cuellar, T., Haley, B., Roose-Girma, M., Phung, Q.T., et al. (2015). Caspase-11 cleaves gasdermin D for non-canonical inflammasome signalling. Nature *526*, 666–671. https://doi.org/10.1038/nature15541.

224. Shi, J., Zhao, Y., Wang, K., Shi, X., Wang, Y., Huang, H., Zhuang, Y., Cai, T., Wang, F., and Shao, F. (2015). Cleavage of GSDMD by inflammatory caspases determines pyroptotic cell death. Nature *526*, 660–665. https://doi.org/10.1038/nature15514.

225. Liu, X., Zhang, Z., Ruan, J., Pan, Y., Magupalli, V.G., Wu, H., and Lieberman, J. (2016). Inflammasome-activated gasdermin D causes pyroptosis by forming membrane pores. Nature *535*, 153–158. https://doi.org/10.1038/nature18629.

226. Ding, J., Wang, K., Liu, W., She, Y., Sun, Q., Shi, J., Sun, H., Wang, D.-C., and Shao, F. (2016). Pore-forming activity and structural autoinhibition of the gasdermin family. Nature *535*, 111–116. https://doi.org/10.1038/nature18590.

227. Aglietti, R.A., Estevez, A., Gupta, A., Ramirez, M.G., Liu, P.S., Kayagaki, N., Ciferri, C., Dixit, V.M., and Dueber, E.C. (2016). GsdmD p30 elicited by caspase-11 during pyroptosis forms pores in membranes. Proc. Natl. Acad. Sci. USA *113*, 7858–7863. https://doi.org/10.1073/pnas.1607769113.

228. Sborgi, L., Rühl, S., Mulvihill, E., Pipercevic, J., Heilig, R., Stahlberg, H., Farady, C.J., Müller, D.J., Broz, P., and Hiller, S. (2016). GSDMD membrane pore formation constitutes the mechanism of pyroptotic cell death. EMBO J. *35*, 1766–1778. https://doi.org/10.15252/embj.201694696.

229. Shi, J., Gao, W., and Shao, F. (2017). Pyroptosis: Gasdermin-mediated programmed necrotic cell death. Trends Biochem. Sci. *42*, 245–254. https://doi.org/10.1016/j.tibs.2016.10.004.

230. Delmaghani, S., del Castillo, F.J., Michel, V., Leibovici, M., Aghaie, A., Ron, U., Van Laer, L., Ben-Tal, N., Van Camp, G., Weil, D., et al. (2006). Mutations in the gene encoding pejvakin, a newly identified protein of the afferent auditory pathway, cause DFNB59 auditory neuropathy. Nat. Genet. *38*, 770–778. https://doi.org/10.1038/ng1829.

231. Ruan, J., Xia, S., Liu, X., Lieberman, J., and Wu, H. (2018). Cryo-EM structure of the gasdermin A3 membrane pore. Nature *557*, 62–67. https://doi.org/10.1038/s41586-018-0058-6.

232. Xia, S., Zhang, Z., Magupalli, V.G., Pablo, J.L., Dong, Y., Vora, S.M., Wang, L., Fu, T.M., Jacobson, M.P., Greka, A., et al. (2021). Gasdermin D pore structure reveals preferential release of mature interleukin-1. Nature *593*, 607–611. https://doi.org/10.1038/s41586-021-03478-3.

233. Zhong, X., Zeng, H., Zhou, Z., Su, Y., Cheng, H., Hou, Y., She, Y., Feng, N., Wang, J., Shao, F., et al. (2023). Structural mechanisms for regulation

of GSDMB pore-forming activity. Nature *616*, 598–605. https://doi.org/10.1038/s41586-023-05872-5.

234. Heilig, R., Dick, M.S., Sborgi, L., Meunier, E., Hiller, S., and Broz, P. (2018). The Gasdermin-D pore acts as a conduit for IL-1beta secretion in mice. Eur. J. Immunol. *48*, 584–592. https://doi.org/10.1002/eji.201747404.

235. Broz, P., Pelegrín, P., and Shao, F. (2020). The gasdermins, a protein family executing cell death and inflammation. Nat. Rev. Immunol. *20*, 143–157. https://doi.org/10.1038/s41577-019-0228-2.

236. Liu, X., Xia, S., Zhang, Z., Wu, H., and Lieberman, J. (2021). Channelling inflammation: gasdermins in physiology and disease. Nat. Rev. Drug Discov. *20*, 384–405. https://doi.org/10.1038/s41573-021-00154-z.

237. Wang, K., Sun, Q., Zhong, X., Zeng, M., Zeng, H., Shi, X., Li, Z., Wang, Y., Zhao, Q., Shao, F., et al. (2020). Structural mechanism for GSDMD targeting by autoprocessed caspases in pyroptosis. Cell *180*, 941–955.e20. https://doi.org/10.1016/j.cell.2020.02.002.

238. Rogers, C., Fernandes-Alnemri, T., Mayes, L., Alnemri, D., Cingolani, G., and Alnemri, E.S. (2017). Cleavage of DFNA5 by caspase-3 during apoptosis mediates progression to secondary necrotic/pyroptotic cell death. Nat. Commun. *8*, 14128. https://doi.org/10.1038/ncomms14128.

239. Wang, Y., Gao, W., Shi, X., Ding, J., Liu, W., He, H., Wang, K., and Shao, F. (2017). Chemotherapy drugs induce pyroptosis through caspase-3 cleavage of a gasdermin. Nature *547*, 99–103. https://doi.org/10.1038/nature22393.

240. Voskoboinik, I., Whisstock, J.C., and Trapani, J.A. (2015). Perforin and granzymes: function, dysfunction and human pathology. Nat. Rev. Immunol. *15*, 388–400. https://doi.org/10.1038/nri3839.

241. Zhou, Z., He, H., Wang, K., Shi, X., Wang, Y., Su, Y., Wang, Y., Li, D., Liu, W., Zhang, Y., et al. (2020). Granzyme A from cytotoxic lymphocytes cleaves GSDMB to trigger pyroptosis in target cells. Science *368*, eaaz7548. https://doi.org/10.1126/science.aaz7548.

242. Zhang, Z., Zhang, Y., and Lieberman, J. (2021). Lighting a Fire: Can We Harness Pyroptosis to Ignite Antitumor Immunity? Cancer Immunol. Res. *9*, 2–7. https://doi.org/10.1158/2326-6066.CIR-20-0525.

243. Yang, L., He, H., Guo, X.K., Wang, J., Wang, W., Li, D., Liang, S., Shao, F., Liu, W., and Hu, X. (2024). Intraepithelial mast cells drive gasdermin C-mediated type 2 immunity. Immunity *57*, 1056–1070.e5. https://doi.org/10.1016/j.immuni.2024.03.017.

244. Pandey, S.P., Yang, D., Hedden, L., Laughlin, C.R., Wang, W., Soto, A.S., Winner, H., Medina Sanchez, L., Campana, E.E., Engl, C., et al. (2025). Gasdermin C cleavage by Cathepsin S modulates Rab7 vesicles in intestinal epithelial cells to amplify anti-helminth immunity. Immunity *58*, 2439–2455.e8. https://doi.org/10.1016/j.immuni.2025.06.018.

245. Hou, J., Zhao, R., Xia, W., Chang, C.W., You, Y., Hsu, J.M., Nie, L., Chen, Y., Wang, Y.C., Liu, C., et al. (2020). PD-L1-mediated gasdermin C expression switches apoptosis to pyroptosis in cancer cells and facilitates tumour necrosis. Nat. Cell Biol. *22*, 1264–1275. https://doi.org/10.1038/s41556-020-0575-z.

246. Zhang, J.Y., Zhou, B., Sun, R.Y., Ai, Y.L., Cheng, K., Li, F.N., Wang, B.R., Liu, F.J., Jiang, Z.H., Wang, W.J., et al. (2021). The metabolite alpha-KG induces GSDMC-dependent pyroptosis through death receptor 6-activated caspase-8. Cell Res. *31*, 980–997. https://doi.org/10.1038/s41422-021-00506-9.

247. Deng, W., Bai, Y., Deng, F., Pan, Y., Mei, S., Zheng, Z., Min, R., Wu, Z., Li, W., Miao, R., et al. (2022). Streptococcal pyrogenic exotoxin B cleaves GSDMA and triggers pyroptosis. Nature *602*, 496–502. https://doi.org/10.1038/s41586-021-04384-4.

248. LaRock, D.L., Johnson, A.F., Wilde, S., Sands, J.S., Monteiro, M.P., and LaRock, C.N. (2022). Group A Streptococcus induces GSDMA-dependent pyroptosis in keratinocytes. Nature *605*, 527–531. https://doi.org/10.1038/s41586-022-04717-x.

249. Zhang, Z., Zhang, Y., Xia, S., Kong, Q., Li, S., Liu, X., Junqueira, C., Meza-Sosa, K.F., Mok, T.M.Y., Ansara, J., et al. (2020). Gasdermin E suppresses tumour growth by activating anti-tumour immunity. Nature *579*, 415–420. https://doi.org/10.1038/s41586-020-2071-9.

250. Tsuchiya, K. (2021). Switching from apoptosis to pyroptosis: Gasdermin-elicited inflammation and antitumor immunity. Int. J. Mol. Sci. *22*, 426. https://doi.org/10.3390/ijms22010426.

251. Karki, R., Man, S.M., and Kanneganti, T.D. (2017). Inflammasomes and Cancer. Cancer Immunol. Res. *5*, 94–99. https://doi.org/10.1158/2326-6066.CIR-16-0269.

252. Sharma, B.R., and Kanneganti, T.D. (2021). NLRP3 inflammasome in cancer and metabolic diseases. Nat. Immunol. *22*, 550–559. https://doi.org/10.1038/s41590-021-00886-5.

253. Gardner, A., and Ruffell, B. (2016). Dendritic Cells and Cancer Immunity. Trends Immunol. *37*, 855–865. https://doi.org/10.1016/j.it.2016.09.006.

254. Van Gorp, H., and Lamkanfi, M. (2019). The emerging roles of inflammasome-dependent cytokines in cancer development. EMBO Rep. *20*, e47575. https://doi.org/10.15252/embr.201847575.

255. Akino, K., Toyota, M., Suzuki, H., Imai, T., Maruyama, R., Kusano, M., Nishikawa, N., Watanabe, Y., Sasaki, Y., Abe, T., et al. (2007). Identification of DFNA5 as a target of epigenetic inactivation in gastric cancer. Cancer Sci. *98*, 88–95. https://doi.org/10.1111/j.1349-7006.2006.00351.x.

256. Kim, M.S., Chang, X., Yamashita, K., Nagpal, J.K., Baek, J.H., Wu, G., Trink, B., Ratovitski, E.A., Mori, M., and Sidransky, D. (2008). Aberrant promoter methylation and tumor suppressive activity of the DFNA5 gene in colorectal carcinoma. Oncogene *27*, 3624–3634. https://doi.org/10.1038/sj.onc.1211021.

257. Yokomizo, K., Harada, Y., Kijima, K., Shinmura, K., Sakata, M., Sakuraba, K., Kitamura, Y., Shirahata, A., Goto, T., Mizukami, H., et al. (2012). Methylation of the DFNA5 gene is frequently detected in colorectal cancer. Anticancer Res. *32*, 1319–1322.

258. Wang, C.J., Tang, L., Shen, D.W., Wang, C., Yuan, Q.Y., Gao, W., Wang, Y.K., Xu, R.H., and Zhang, H. (2013). The expression and regulation of DFNA5 in human hepatocellular carcinoma DFNA5 in hepatocellular carcinoma. Mol. Biol. Rep. *40*, 6525–6531. https://doi.org/10.1007/s11033-013-2581-8.

259. Fan, J.X., Deng, R.H., Wang, H., Liu, X.H., Wang, X.N., Qin, R., Jin, X., Lei, T.R., Zheng, D., Zhou, P.H., et al. (2019). Epigenetics-Based Tumor Cells Pyroptosis for Enhancing the Immunological Effect of Chemotherapeutic Nanocarriers. Nano Lett. *19*, 8049–8058. https://doi.org/10.1021/acs.nanolett.9b03245.

260. Oltra, S.S., Colomo, S., Sin, L., Pérez-López, M., Lázaro, S., Molina-Crespo, A., Choi, K.H., Ros-Pardo, D., Martínez, L., Morales, S., et al. (2023). Distinct GSDMB protein isoforms and protease cleavage processes differentially control pyroptotic cell death and mitochondrial damage in cancer cells. Cell Death Differ. *30*, 1366–1381. https://doi.org/10.1038/s41418-023-01143-y.

261. Wang, C., Shivcharan, S., Tian, T., Wright, S., Ma, D., Chang, J., Li, K., Song, K., Xu, C., Rathinam, V.A., et al. (2023). Structural basis for GSDMB pore formation and its targeting by IpaH7.8. Nature *616*, 590–597. https://doi.org/10.1038/s41586-023-05832-z.

262. Kong, Q., Xia, S., Pan, X., Ye, K., Li, Z., Li, H., Tang, X., Sahni, N., Yi, S.S., Liu, X., et al. (2023). Alternative splicing of GSDMB modulates killer lymphocyte-triggered pyroptosis. Sci. Immunol. *8*, eadg3196. https://doi.org/10.1126/sciimmunol.adg3196.

263. Garlanda, C., and Mantovani, A. (2021). Interleukin-1 in tumor progression, therapy, and prevention. Cancer Cell *39*, 1023–1027. https://doi.org/10.1016/j.ccell.2021.04.011.

264. Mantovani, A., Dinarello, C.A., Molgora, M., and Garlanda, C. (2019). Interleukin-1 and Related Cytokines in the Regulation of Inflammation and Immunity. Immunity *50*, 778–795. https://doi.org/10.1016/j.immuni.2019.03.012.

265. de Mooij, C.E.M., Netea, M.G., van der Velden, W.J.F.M., and Blijlevens, N.M.A. (2017). Targeting the interleukin-1 pathway in patients with

hematological disorders. Blood *129*, 3155–3164. https://doi.org/10.1182/blood-2016-12-754994.

266. Carmi, Y., Dotan, S., Rider, P., Kaplanov, I., White, M.R., Baron, R., Abutbul, S., Huszar, M., Dinarello, C.A., Apte, R.N., et al. (2013). The role of IL-1beta in the early tumor cell-induced angiogenic response. J. Immunol. *190*, 3500–3509. https://doi.org/10.4049/jimmunol.1202769.

267. Voronov, E., Carmi, Y., and Apte, R.N. (2014). The role IL-1 in tumor-mediated angiogenesis. Front. Physiol. *5*, 114. https://doi.org/10.3389/fphys.2014.00114.

268. Tu, S., Bhagat, G., Cui, G., Takaishi, S., Kurt-Jones, E.A., Rickman, B., Betz, K.S., Penz-Oesterreicher, M., Bjorkdahl, O., Fox, J.G., et al. (2008). Overexpression of interleukin-1beta induces gastric inflammation and cancer and mobilizes myeloid-derived suppressor cells in mice. Cancer Cell *14*, 408–419. https://doi.org/10.1016/j.ccr.2008.10.011.

269. Elkabets, M., Ribeiro, V.S.G., Dinarello, C.A., Ostrand-Rosenberg, S., Di Santo, J.P., Apte, R.N., and Vosshenrich, C.A.J. (2010). IL-1beta regulates a novel myeloid-derived suppressor cell subset that impairs NK cell development and function. Eur. J. Immunol. *40*, 3347–3357. https://doi.org/10.1002/eji.201041037.

270. Kaplanov, I., Carmi, Y., Kornetsky, R., Shemesh, A., Shurin, G.V., Shurin, M.R., Dinarello, C.A., Voronov, E., and Apte, R.N. (2019). Blocking IL-1beta reverses the immunosuppression in mouse breast cancer and synergizes with anti-PD-1 for tumor abrogation. Proc. Natl. Acad. Sci. USA *116*, 1361–1369. https://doi.org/10.1073/pnas.1812266115.

271. Wang, Q., Wang, Y., Ding, J., Wang, C., Zhou, X., Gao, W., Huang, H., Shao, F., and Liu, Z. (2020). A bioorthogonal system reveals antitumour immune function of pyroptosis. Nature *579*, 421–426. https://doi.org/10.1038/s41586-020-2079-1.

272. Erkes, D.A., Cai, W., Sanchez, I.M., Purwin, T.J., Rogers, C., Field, C.O., Berger, A.C., Hartsough, E.J., Rodeck, U., Alnemri, E.S., et al. (2020). Mutant BRAF and MEK Inhibitors Regulate the Tumor Immune Microenvironment via Pyroptosis. Cancer Discov. *10*, 254–269. https://doi.org/10.1158/2159-8290.CD-19-0672.

273. Cragg, M.S., Jansen, E.S., Cook, M., Harris, C., Strasser, A., and Scott, C.L. (2008). Treatment of B-RAF mutant human tumor cells with a MEK inhibitor requires Bim and is enhanced by a BH3 mimetic. J. Clin. Investig. *118*, 3651–3659. https://doi.org/10.1172/JCI35437.

274. Johnson, D.C., Taabazuing, C.Y., Okondo, M.C., Chui, A.J., Rao, S.D., Brown, F.C., Reed, C., Peguero, E., de Stanchina, E., Kentsis, A., et al. (2018). DPP8/DPP9 inhibitor-induced pyroptosis for treatment of acute myeloid leukemia. Nat. Med. *24*, 1151–1156. https://doi.org/10.1038/s41591-018-0082-y.

275. Fontana, P., Du, G., Zhang, Y., Zhang, H., Vora, S.M., Hu, J.J., Shi, M., Tufan, A.B., Healy, L.B., Xia, S., et al. (2024). Small-molecule GSDMD agonism in tumors stimulates antitumor immunity without toxicity. Cell *187*, 6165–6181.e22. https://doi.org/10.1016/j.cell.2024.08.007.

276. Ibrahim, J., Op de Beeck, K., Fransen, E., Croes, L., Beyens, M., Suls, A., Vanden Berghe, W., Peeters, M., and Van Camp, G. (2019). Methylation analysis of Gasdermin E shows great promise as a biomarker for colorectal cancer. Cancer Med. *8*, 2133–2145. https://doi.org/10.1002/cam4.2103.

277. Zhao, L., Cheng, H., Tong, Z., and Cai, J. (2024). Nanoparticle-mediated cell pyroptosis: a new therapeutic strategy for inflammatory diseases and cancer. J. Nanobiotechnology *22*, 504. https://doi.org/10.1186/s12951-024-02763-3.

278. Fang, Y., Tang, Y., and Huang, B. (2023). Pyroptosis: A road to next-generation cancer immunotherapy. Semin. Immunol. *68*, 101782. https://doi.org/10.1016/j.smim.2023.101782.

279. Li, H., Lu, J., Tan, S., Jiang, T., He, X., Qiao, W., Hu, C., Dalangood, S., Lin, J., and Gui, J. (2025). In Situ Delivery of Gasdermin E mRNA Promotes Antitumor Immunity via Creatine-Elicited Type I Interferon Signaling in Monocytes. Cancer Immunol. Res. *13*, 939–956. https://doi.org/10.1158/2326-6066.CIR-24-0834.

280. Lin, J., Sun, S., Zhao, K., Gao, F., Wang, R., Li, Q., Zhou, Y., Zhang, J., Li, Y., Wang, X., et al. (2023). Oncolytic Parapoxvirus induces Gasdermin E-mediated pyroptosis and activates antitumor immunity. Nat. Commun. *14*, 224. https://doi.org/10.1038/s41467-023-35917-2.

281. Liu, Y., Fang, Y., Chen, X., Wang, Z., Liang, X., Zhang, T., Liu, M., Zhou, N., Lv, J., Tang, K., et al. (2020). Gasdermin E-mediated target cell pyroptosis by CAR T cells triggers cytokine release syndrome. Sci. Immunol. *5*, eaax7969. https://doi.org/10.1126/sciimmunol.aax7969.

282. Dixon, S.J., and Stockwell, B.R. (2019). The hallmarks of ferroptosis. Annu. Rev. Cancer Biol. *3*, 35–54. https://doi.org/10.1146/annurev-cancerbio-030518-055844.

283. Mishima, E., Nakamura, T., Doll, S., Proneth, B., Fedorova, M., Pratt, D.A., Friedmann Angeli, J.P., Dixon, S.J., Wahida, A., and Conrad, M. (2025). Recommendations for robust and reproducible research on ferroptosis. Nat. Rev. Mol. Cell Biol. *26*, 615–630. https://doi.org/10.1038/s41580-025-00843-2.

284. Green, D.R. (2019). The coming decade of cell death research: Five riddles. Cell *177*, 1094–1107. https://doi.org/10.1016/j.cell.2019.04.024.

285. Berndt, C., Alborzinia, H., Amen, V.S., Ayton, S., Barayeu, U., Bartelt, A., Bayir, H., Bebber, C.M., Birsoy, K., Böttcher, J.P., et al. (2024). Ferroptosis in health and disease. Redox Biol. *75*, 103211. https://doi.org/10.1016/j.redox.2024.103211.

286. Kalkavan, H., Chen, M.J., Crawford, J.C., Quarato, G., Fitzgerald, P., Tait, S.W.G., Goding, C.R., and Green, D.R. (2022). Sublethal cytochrome c release generates drug-tolerant persister cells. Cell *185*, 3356–3374.e22. https://doi.org/10.1016/j.cell.2022.07.025.

287. Liang, D., Minikes, A.M., and Jiang, X. (2022). Ferroptosis at the intersection of lipid metabolism and cellular signaling. Mol. Cell *82*, 2215–2227. https://doi.org/10.1016/j.molcel.2022.03.022.

288. Li, Z., Lange, M., Dixon, S.J., and Olzmann, J.A. (2024). Lipid quality control and ferroptosis: From concept to mechanism. Annu. Rev. Biochem. *93*, 499–528. https://doi.org/10.1146/annurev-biochem-052521-033527.

289. von Krusenstiern, A.N., Robson, R.N., Qian, N., Qiu, B., Hu, F., Reznik, E., Smith, N., Zandkarimi, F., Estes, V.M., Dupont, M., et al. (2023). Identification of essential sites of lipid peroxidation in ferroptosis. Nat. Chem. Biol. *19*, 719–730. https://doi.org/10.1038/s41589-022-01249-3.

290. Brielmeier, M., Béchet, J.M., Suppmann, S., Conrad, M., Laux, G., and Bornkamm, G.W. (2001). Cloning of phospholipid hydroperoxide glutathione peroxidase (PHGPx) as an anti-apoptotic and growth promoting gene of Burkitt lymphoma cells. BioFactors *14*, 179–190. https://doi.org/10.1002/biof.5520140123.

291. Hirata, Y., Cai, R., Volchuk, A., Steinberg, B.E., Saito, Y., Matsuzawa, A., Grinstein, S., and Freeman, S.A. (2023). Lipid peroxidation increases membrane tension, Piezo1 gating, and cation permeability to execute ferroptosis. Curr. Biol. *33*, 1282–1294.e5. https://doi.org/10.1016/j.cub.2023.02.060.

292. Wahida, A., and Conrad, M. (2023). Ferroptosis: Under pressure!. Curr. Biol. *33*, R269–R272. https://doi.org/10.1016/j.cub.2023.03.009.

293. Pedrera, L., Espiritu, R.A., Ros, U., Weber, J., Schmitt, A., Stroh, J., Hailfinger, S., von Karstedt, S., and García-Sáez, A.J. (2021). Ferroptotic pores induce Ca2+ fluxes and ESCRT-III activation to modulate cell death kinetics. Cell Death Differ. *28*, 1644–1657. https://doi.org/10.1038/s41418-020-00691-x.

294. Riegman, M., Sagie, L., Galed, C., Levin, T., Steinberg, N., Dixon, S.J., Wiesner, U., Bradbury, M.S., Niethammer, P., Zaritsky, A., et al. (2020). Ferroptosis occurs through an osmotic mechanism and propagates independently of cell rupture. Nat. Cell Biol. *22*, 1042–1048. https://doi.org/10.1038/s41556-020-0565-1.

295. Banjac, A., Perisic, T., Sato, H., Seiler, A., Bannai, S., Weiss, N., Kölle, P., Tschoep, K., Issels, R.D., Daniel, P.T., et al. (2008). The cystine/cysteine cycle: a redox cycle regulating susceptibility versus resistance to

cell death. Oncogene *27*, 1618–1628. https://doi.org/10.1038/sj.onc.1210796.

296. Doll, S., Freitas, F.P., Shah, R., Aldrovandi, M., da Silva, M.C., Ingold, I., Goya Grocin, A., Xavier da Silva, T.N., Panzilius, E., Scheel, C.H., et al. (2019). FSP1 is a glutathione-independent ferroptosis suppressor. Nature *575*, 693–698. https://doi.org/10.1038/s41586-019-1707-0.

297. Bersuker, K., Hendricks, J.M., Li, Z., Magtanong, L., Ford, B., Tang, P.H., Roberts, M.A., Tong, B., Maimone, T.J., Zoncu, R., et al. (2019). The CoQ oxidoreductase FSP1 acts parallel to GPX4 to inhibit ferroptosis. Nature *575*, 688–692. https://doi.org/10.1038/s41586-019-1705-2.

298. Dolma, S., Lessnick, S.L., Hahn, W.C., and Stockwell, B.R. (2003). Identification of genotype-selective antitumor agents using synthetic lethal chemical screening in engineered human tumor cells. Cancer Cell *3*, 285–296. https://doi.org/10.1016/S1535-6108(03)00050-3.

299. Conrad, M., and Wahida, A. (2025). The many paths ascending to ferroptosis. Nat. Chem. Biol. *21*, 18–19. https://doi.org/10.1038/s41589-024-01794-z.

300. Friedmann Angeli, J.P., Krysko, D.V., and Conrad, M. (2019). Ferroptosis at the crossroads of cancer-acquired drug resistance and immune evasion. Nat. Rev. Cancer *19*, 405–414. https://doi.org/10.1038/s41568-019-0149-1.

301. Liao, P., Wang, W., Wang, W., Kryczek, I., Li, X., Bian, Y., Sell, A., Wei, S., Grove, S., Johnson, J.K., et al. (2022). CD8+ T cells and fatty acids orchestrate tumor ferroptosis and immunity via ACSL4. Cancer Cell *40*, 365–378.e6. https://doi.org/10.1016/j.ccell.2022.02.003.

302. Wiernicki, B., Maschalidi, S., Pinney, J., Adjemian, S., Vanden Berghe, T., Ravichandran, K.S., and Vandenabeele, P. (2022). Cancer cells dying from ferroptosis impede dendritic cell-mediated anti-tumor immunity. Nat. Commun. *13*, 3676. https://doi.org/10.1038/s41467-022-31218-2.

303. Schwab, A., Rao, Z., Zhang, J., Gollowitzer, A., Siebenkäs, K., Bindel, N., D'Avanzo, E., van Roey, R., Hajjaj, Y., Özel, E., et al. (2024). Zeb1 mediates EMT/plasticity-associated ferroptosis sensitivity in cancer cells by regulating lipogenic enzyme expression and phospholipid composition. Nat. Cell Biol. *26*, 1470–1481. https://doi.org/10.1038/s41556-024-01464-1.

304. Nakamura, T., Hipp, C., Santos Dias Mourão, A., Borggräfe, J., Aldrovandi, M., Henkelmann, B., Wanninger, J., Mishima, E., Lytton, E., Emler, D., et al. (2023). Phase separation of FSP1 promotes ferroptosis. Nature *619*, 371–377. https://doi.org/10.1038/s41586-023-06255-6.

305. Wu, K., Vaughan, A.J., Bossowski, J.P., Hao, Y., Ziogou, A., Kim, S.M., Kim, T.H., Nakamura, M.N., Pillai, R., Mancini, M., et al. (2025). Targeting FSP1 triggers ferroptosis in lung cancer. Nature *649*, 487–495. https://doi.org/10.1038/s41586-025-09710-8.

306. Palma, M., Chaufan, M., Breuer, C.B., Müller, S., Sabatier, M., Fraser, C.S., Szylo, K.J., Yavari, M., Carmona, A., Kaur, M., et al. (2026). Lymph node environment drives FSP1 targetability in metastasizing melanoma. Nature *649*, 477–486. https://doi.org/10.1038/s41586-025-09709-1.

307. Lorenz, S.M., Wahida, A., Bostock, M.J., Seibt, T., Santos Dias Mourão, A., Levkina, A., Trümbach, D., Soudy, M., Emler, D., Rothammer, N., et al. (2026). A fin-loop-like structure in GPX4 underlies neuroprotection from ferroptosis. Cell *189*, 287–306.e35. https://doi.org/10.1016/j.cell.2025.11.014.

308. Liang, Y., Zhao, Y., Qi, Z., Li, X., and Zhao, Y. (2025). Ferroptosis: CD8+T cells' blade to destroy tumor cells or poison for self-destruction. Cell Death Discov. *11*, 128. https://doi.org/10.1038/s41420-025-02415-x.

309. Lang, X., Green, M.D., Wang, W., Yu, J., Choi, J.E., Jiang, L., Liao, P., Zhou, J., Zhang, Q., Dow, A., et al. (2019). Radiotherapy and Immunotherapy Promote Tumoral Lipid Oxidation and Ferroptosis via Synergistic Repression of SLC7A11. Cancer Discov. *9*, 1673–1685. https://doi.org/10.1158/2159-8290.CD-19-0338.

310. Mishima, E., Ito, J., Wu, Z., Nakamura, T., Wahida, A., Doll, S., Tonnus, W., Nepachalovich, P., Eggenhofer, E., Aldrovandi, M., et al. (2022). A non-canonical vitamin K cycle is a potent ferroptosis suppressor. Nature *608*, 778–783. https://doi.org/10.1038/s41586-022-05022-3.

311. Kraft, V.A.N., Bezjian, C.T., Pfeiffer, S., Ringelstetter, L., Müller, C., Zandkarimi, F., Merl-Pham, J., Bao, X., Anastasov, N., Kössl, J., et al. (2020). GTP cyclohydrolase 1/tetrahydrobiopterin counteract ferroptosis through lipid remodeling. ACS Cent. Sci. *6*, 41–53. https://doi.org/10.1021/acscentsci.9b01063.

312. Soula, M., Weber, R.A., Zilka, O., Alwaseem, H., La, K., Yen, F., Molina, H., Garcia-Bermudez, J., Pratt, D.A., and Birsoy, K. (2020). Metabolic determinants of cancer cell sensitivity to canonical ferroptosis inducers. Nat. Chem. Biol. *16*, 1351–1360. https://doi.org/10.1038/s41589-020-0613-y.

313. Hassannia, B., Wiernicki, B., Ingold, I., Qu, F., Van Herck, S., Tyurina, Y.Y., Bayır, H., Abhari, B.A., Angeli, J.P.F., Choi, S.M., et al. (2018). Nano-targeted induction of dual ferroptotic mechanisms eradicates high-risk neuroblastoma. J. Clin. Investig. *128*, 3341–3355. https://doi.org/10.1172/JCI99032.

314. Rodriguez, R., Schreiber, S.L., and Conrad, M. (2022). Persister cancer cells: Iron addiction and vulnerability to ferroptosis. Mol. Cell *82*, 728–740. https://doi.org/10.1016/j.molcel.2021.12.001.

315. Müller, S., Sindikubwabo, F., Cañeque, T., Lafon, A., Versini, A., Lombard, B., Loew, D., Wu, T.-D., Ginestier, C., Charafe-Jauffret, E., et al. (2020). CD44 regulates epigenetic plasticity by mediating iron endocytosis. Nat. Chem. *12*, 929–938. https://doi.org/10.1038/s41557-020-0513-5.

316. Mancias, J.D., Wang, X., Gygi, S.P., Harper, J.W., and Kimmelman, A.C. (2014). Quantitative proteomics identifies NCOA4 as the cargo receptor mediating ferritinophagy. Nature *509*, 105–109. https://doi.org/10.1038/nature13148.

317. Müller, S., Cañeque, T., Solier, S., and Rodriguez, R. (2025). Copper and iron orchestrate cell-state transitions in cancer and immunity. Trends Cell Biol. *35*, 105–114. https://doi.org/10.1016/j.tcb.2024.07.005.

318. Cañeque, T., Baron, L., Müller, S., Carmona, A., Colombeau, L., Versini, A., Solier, S., Gaillet, C., Sindikubwabo, F., Sampaio, J.L., et al. (2025). Activation of lysosomal iron triggers ferroptosis in cancer. Nature *642*, 492–500. https://doi.org/10.1038/s41586-025-08974-4.

319. Kim, J.W., Kim, M.-J., Han, T.-H., Lee, J.-Y., Kim, S., Kim, H., Oh, K.-J., Kim, W.K., Han, B.-S., Bae, K.-H., et al. (2023). FSP1 confers ferroptosis resistance in KEAP1 mutant non-small cell lung carcinoma in NRF2-dependent and -independent manner. Cell Death Dis. *14*, 567. https://doi.org/10.1038/s41419-023-06070-x.

320. Koppula, P., Lei, G., Zhang, Y., Yan, Y., Mao, C., Kondiparthi, L., Shi, J., Liu, X., Horbath, A., Das, M., et al. (2022). A targetable CoQ-FSP1 axis drives ferroptosis- and radiation-resistance in KEAP1 inactive lung cancers. Nat. Commun. *13*, 2206. https://doi.org/10.1038/s41467-022-29905-1.

321. Wohlhieter, C.A., Richards, A.L., Uddin, F., Hulton, C.H., Quintanal-Villalonga, À., Martin, A., de Stanchina, E., Bhanot, U., Asher, M., Shah, N.S., et al. (2020). Concurrent mutations in STK11 and KEAP1 promote ferroptosis protection and SCD1 dependence in lung cancer. Cell Rep. *33*, 108444. https://doi.org/10.1016/j.celrep.2020.108444.

322. Tesfay, L., Paul, B.T., Konstorum, A., Deng, Z., Cox, A.O., Lee, J., Furdui, C.M., Hegde, P., Torti, F.M., and Torti, S.V. (2019). Stearoyl-CoA desaturase 1 protects ovarian cancer cells from ferroptotic cell death. Cancer Res. *79*, 5355–5366. https://doi.org/10.1158/0008-5472.CAN-19-0369.

323. Alborzinia, H., Chen, Z., Yildiz, U., Freitas, F.P., Vogel, F.C.E., Varga, J.P., Batani, J., Bartenhagen, C., Schmitz, W., Büchel, G., et al. (2023). LRP8-mediated selenocysteine uptake is a targetable vulnerability in MYCN-amplified neuroblastoma. EMBO Mol. Med. *15*, e18014. https://doi.org/10.15252/emmm.202318014.

324. Alborzinia, H., Flórez, A.F., Kreth, S., Brückner, L.M., Yildiz, U., Gartlgruber, M., Odoni, D.I., Poschet, G., Garbowicz, K., Shao, C., et al. (2022). MYCN mediates cysteine addiction and sensitizes

neuroblastoma to ferroptosis. Nat. Cancer *3*, 471–485. https://doi.org/10.1038/s43018-022-00355-4.

325. Ingold, I., Berndt, C., Schmitt, S., Doll, S., Poschmann, G., Buday, K., Roveri, A., Peng, X., Porto Freitas, F., Seibt, T., et al. (2018). Selenium utilization by GPX4 is required to prevent hydroperoxide-induced ferroptosis. Cell *172*, 409–422.e21. https://doi.org/10.1016/j.cell.2017.11.048.

326. Fujita, H., Tanaka, Y.-K., Ogata, S., Suzuki, N., Kuno, S., Barayeu, U., Akaike, T., Ogra, Y., and Iwai, K. (2024). PRDX6 augments selenium utilization to limit iron toxicity and ferroptosis. Nat. Struct. Mol. Biol. *31*, 1277–1285. https://doi.org/10.1038/s41594-024-01329-z.

327. Chen, Z., Inague, A., Kaushal, K., Fazeli, G., Schilling, D., Xavier da Silva, T.N., Dos Santos, A.F., Cheytan, T., Freitas, F.P., Yildiz, U., et al. (2024). PRDX6 contributes to selenocysteine metabolism and ferroptosis resistance. Mol. Cell *84*, 4645–4659.e9. https://doi.org/10.1016/j.molcel.2024.10.027.

328. Ito, J., Nakamura, T., Toyama, T., Chen, D., Berndt, C., Poschmann, G., Mourão, A.S.D., Doll, S., Suzuki, M., Zhang, W., et al. (2024). PRDX6 dictates ferroptosis sensitivity by directing cellular selenium utilization. Mol. Cell *84*, 4629–4644.e9. https://doi.org/10.1016/j.molcel.2024.10.028.

329. Kagan, V.E., Mao, G., Qu, F., Angeli, J.P.F., Doll, S., Croix, C.S., Dar, H.H., Liu, B., Tyurin, V.A., Ritov, V.B., et al. (2017). Oxidized arachidonic and adrenic PEs navigate cells to ferroptosis. Nat. Chem. Biol. *13*, 81–90. https://doi.org/10.1038/nchembio.2238.

330. Zheng, J., and Conrad, M. (2025). Ferroptosis: when metabolism meets cell death. Physiol. Rev. *105*, 651–706. https://doi.org/10.1152/physrev.00031.2024.

331. Doll, S., Proneth, B., Tyurina, Y.Y., Panzilius, E., Kobayashi, S., Ingold, I., Irmler, M., Beckers, J., Aichler, M., Walch, A., et al. (2017). ACSL4 dictates ferroptosis sensitivity by shaping cellular lipid composition. Nat. Chem. Biol. *13*, 91–98. https://doi.org/10.1038/nchembio.2239.

332. Dixon, S.J., Winter, G.E., Musavi, L.S., Lee, E.D., Snijder, B., Rebsamen, M., Superti-Furga, G., and Stockwell, B.R. (2015). Human haploid cell genetics reveals roles for lipid metabolism genes in nonapoptotic cell death. ACS Chem. Biol. *10*, 1604–1609. https://doi.org/10.1021/acschembio.5b00245.

333. Freitas, F.P., Alborzinia, H., Dos Santos, A.F., Nepachalovich, P., Pedrera, L., Zilka, O., Inague, A., Klein, C., Aroua, N., Kaushal, K., et al. (2024). 7-Dehydrocholesterol is an endogenous suppressor of ferroptosis. Nature *626*, 401–410. https://doi.org/10.1038/s41586-023-06878-9.

334. Yamada, N., Karasawa, T., Ito, J., Yamamuro, D., Morimoto, K., Nakamura, T., Komada, T., Baatarjav, C., Saimoto, Y., Jinnouchi, Y., et al. (2024). Inhibition of 7-dehydrocholesterol reductase prevents hepatic ferroptosis under an active state of sterol synthesis. Nat. Commun. *15*, 2195. https://doi.org/10.1038/s41467-024-46386-6.

335. Li, Y., Ran, Q., Duan, Q., Jin, J., Wang, Y., Yu, L., Wang, C., Zhu, Z., Chen, X., Weng, L., et al. (2024). 7-Dehydrocholesterol dictates ferroptosis sensitivity. Nature *626*, 411–418. https://doi.org/10.1038/s41586-023-06983-9.

336. Bartolacci, C., Andreani, C., Vale, G., Berto, S., Melegari, M., Crouch, A.C., Baluya, D.L., Kemble, G., Hodges, K., Starrett, J., et al. (2022). Targeting de novo lipogenesis and the Lands cycle induces ferroptosis in KRAS-mutant lung cancer. Nat. Commun. *13*, 4327. https://doi.org/10.1038/s41467-022-31963-4.

337. Minami, J.K., Morrow, D., Bayley, N.A., Fernandez, E.G., Salinas, J.J., Tse, C., Zhu, H., Su, B., Plawat, R., Jones, A., et al. (2023). CDKN2A deletion remodels lipid metabolism to prime glioblastoma for ferroptosis. Cancer Cell *41*, 1048–1060.e9. https://doi.org/10.1016/j.ccell.2023.05.001.

338. Zou, Y., Palte, M.J., Deik, A.A., Li, H., Eaton, J.K., Wang, W., Tseng, Y.-Y., Deasy, R., Kost-Alimova, M., Dančík, V., et al. (2019). A GPX4-dependent cancer cell state underlies the clear-cell morphology and confers sensitivity to ferroptosis. Nat. Commun. *10*, 1617. https://doi.org/10.1038/s41467-019-09277-9.

339. Miess, H., Dankworth, B., Gouw, A.M., Rosenfeldt, M., Schmitz, W., Jiang, M., Saunders, B., Howell, M., Downward, J., Felsher, D.W., et al. (2018). The glutathione redox system is essential to prevent ferroptosis caused by impaired lipid metabolism in clear cell renal cell carcinoma. Oncogene *37*, 5435–5450. https://doi.org/10.1038/s41388-018-0315-z.

340. Yang, Z., Su, W., Wei, X., Qu, S., Zhao, D., Zhou, J., Wang, Y., Guan, Q., Qin, C., Xiang, J., et al. (2023). HIF-1α drives resistance to ferroptosis in solid tumors by promoting lactate production and activating SLC1A1. Cell Rep. *42*, 112945. https://doi.org/10.1016/j.celrep.2023.112945.

341. Mishima, E., Wahida, A., Seibt, T., and Conrad, M. (2023). Diverse biological functions of vitamin K: from coagulation to ferroptosis. Nat. Metab. *5*, 924–932. https://doi.org/10.1038/s42255-023-00821-y.

342. Mishima, E., and Conrad, M. (2022). Nutritional and metabolic control of ferroptosis. Annu. Rev. Nutr. *42*, 275–309. https://doi.org/10.1146/annurev-nutr-062320-114541.

343. Yang, X., Wang, Z., Zandkarimi, F., Liu, Y., Duan, S., Li, Z., Kon, N., Zhang, Z., Jiang, X., Stockwell, B.R., et al. (2023). Regulation of VKORC1L1 is critical for p53-mediated tumor suppression through vitamin K metabolism. Cell Metab. *35*, 1474–1490.e8. https://doi.org/10.1016/j.cmet.2023.06.014.

344. Ferrada, L., Barahona, M.J., Vera, M., Stockwell, B.R., and Nualart, F. (2023). Dehydroascorbic acid sensitizes cancer cells to system xc- inhibition-induced ferroptosis by promoting lipid droplet peroxidation. Cell Death Dis. *14*, 637. https://doi.org/10.1038/s41419-023-06153-9.

345. Bell, H.N., Stockwell, B.R., and Zou, W. (2024). Ironing out the role of ferroptosis in immunity. Immunity *57*, 941–956. https://doi.org/10.1016/j.immuni.2024.03.019.

346. Wang, W., Green, M., Choi, J.E., Gijón, M., Kennedy, P.D., Johnson, J.K., Liao, P., Lang, X., Kryczek, I., Sell, A., et al. (2019). CD8+ T cells regulate tumour ferroptosis during cancer immunotherapy. Nature *569*, 270–274. https://doi.org/10.1038/s41586-019-1170-y.

347. Xu, S., Chaudhary, O., Rodríguez-Morales, P., Sun, X., Chen, D., Zappasodi, R., Xu, Z., Pinto, A.F.M., Williams, A., Schulze, I., et al. (2021). Uptake of oxidized lipids by the scavenger receptor CD36 promotes lipid peroxidation and dysfunction in CD8+ T cells in tumors. Immunity *54*, 1561–1577.e7. https://doi.org/10.1016/j.immuni.2021.05.003.

348. Ma, X., Xiao, L., Liu, L., Ye, L., Su, P., Bi, E., Wang, Q., Yang, M., Qian, J., and Yi, Q. (2021). CD36-mediated ferroptosis dampens intratumoral CD8+ T cell effector function and impairs their antitumor ability. Cell Metab. *33*, 1001–1012.e5. https://doi.org/10.1016/j.cmet.2021.02.015.

349. Xu, C., Sun, S., Johnson, T., Qi, R., Zhang, S., Zhang, J., and Yang, K. (2021). The glutathione peroxidase Gpx4 prevents lipid peroxidation and ferroptosis to sustain Treg cell activation and suppression of antitumor immunity. Cell Rep. *35*, 109235. https://doi.org/10.1016/j.celrep.2021.109235.

350. Poznanski, S.M., Singh, K., Ritchie, T.M., Aguiar, J.A., Fan, I.Y., Portillo, A.L., Rojas, E.A., Vahedi, F., El-Sayes, A., Xing, S., et al. (2021). Metabolic flexibility determines human NK cell functional fate in the tumor microenvironment. Cell Metab. *33*, 1205–1220.e5. https://doi.org/10.1016/j.cmet.2021.03.023.

351. Kim, R., Hashimoto, A., Markosyan, N., Tyurin, V.A., Tyurina, Y.Y., Kar, G., Fu, S., Sehgal, M., Garcia-Gerique, L., Kossenkov, A., et al. (2022). Ferroptosis of tumour neutrophils causes immune suppression in cancer. Nature *612*, 338–346. https://doi.org/10.1038/s41586-022-05443-0.

352. Moujalled, D., Strasser, A., and Liddell, J.R. (2021). Molecular mechanisms of cell death in neurological diseases. Cell Death Differ. *28*, 2029–2044. https://doi.org/10.1038/s41418-021-00814-y.

353. Ubellacker, J.M., Tasdogan, A., Ramesh, V., Shen, B., Mitchell, E.C., Martin-Sandoval, M.S., Gu, Z., McCormick, M.L., Durham, A.B., Spitz, D.R., et al. (2020). Lymph protects metastasizing melanoma cells from ferroptosis. Nature *585*, 113–118. https://doi.org/10.1038/s41586-020-2623-z.

354. Tasdogan, A., Ubellacker, J.M., and Morrison, S.J. (2021). Redox Regulation in Cancer Cells during Metastasis. Cancer Discov. *11*, 2682–2692. https://doi.org/10.1158/2159-8290.CD-21-0558.

355. Cui, W., Guo, M., Liu, D., Xiao, P., Yang, C., Huang, H., Liang, C., Yang, Y., Fu, X., Zhang, Y., et al. (2024). Gut microbial metabolite facilitates colorectal cancer development via ferroptosis inhibition. Nat. Cell Biol. *26*, 124–137. https://doi.org/10.1038/s41556-023-01314-6.

356. Zhang, Q., Goswami, S., and Yilmaz, O. (2024). Microbial regulation of ferroptosis in cancer. Nat. Cell Biol. *26*, 41–42. https://doi.org/10.1038/s41556-023-01321-7.

357. Strasser, A., and Vaux, D.L. (2020). Cell death in the origin and treatment of cancer. Mol. Cell *78*, 1045–1054. https://doi.org/10.1016/j.molcel.2020.05.014.

358. Souers, A.J., Leverson, J.D., Boghaert, E.R., Ackler, S.L., Catron, N.D., Chen, J., Dayton, B.D., Ding, H., Enschede, S.H., Fairbrother, W.J., et al. (2013). ABT-199, a potent and selective BCL-2 inhibitor, achieves antitumor activity while sparing platelets. Nat. Med. *19*, 202–208. https://doi.org/10.1038/nm.3048.

359. NCCN Clinical Practice Guidelines in Oncology: Chronic Lymphocytic Leukemia/Small Lymphocytic Lymphoma. https://www.nccn.org/professionals/physician_gls/pdf/cll.pdf.

360. Roberts, A.W., Ma, S., Kipps, T.J., Coutre, S.E., Davids, M.S., Eichhorst, B., Hallek, M., Byrd, J.C., Humphrey, K., Zhou, L., et al. (2019). Efficacy of venetoclax in relapsed chronic lymphocytic leukemia is influenced by disease and response variables. Blood *134*, 111–122. https://doi.org/10.1182/blood.2018882555.

361. Thijssen, R., Tian, L., Anderson, M.A., Flensburg, C., Jarratt, A., Garnham, A.L., Jabbari, J.S., Peng, H., Lew, T.E., Teh, C.E., et al. (2022). Single-cell multiomics reveal the scale of multilayered adaptations enabling CLL relapse during venetoclax therapy. Blood *140*, 2127–2141. https://doi.org/10.1182/blood.2022016040.

362. Anderson, M.A., Tam, C., Lew, T.E., Juneja, S., Juneja, M., Westerman, D., Wall, M., Lade, S., Gorelik, A., Huang, D.C.S., et al. (2017). Clinicopathological features and outcomes of progression of CLL on the BCL2 inhibitor venetoclax. Blood *129*, 3362–3370. https://doi.org/10.1182/blood-2017-01-763003.

363. Blombery, P., Anderson, M.A., Gong, J.-N., Thijssen, R., Birkinshaw, R.W., Thompson, E.R., Teh, C.E., Nguyen, T., Xu, Z., Flensburg, C., et al. (2019). Acquisition of the recurrent Gly101Val mutation in BCL2 confers resistance to venetoclax in patients with progressive chronic Lymphocytic leukemia. Cancer Discov. *9*, 342–353. https://doi.org/10.1158/2159-8290.CD-18-1119.

364. Konopleva, M., Pollyea, D.A., Potluri, J., Chyla, B., Hogdal, L., Busman, T., McKeegan, E., Salem, A.H., Zhu, M., Ricker, J.L., et al. (2016). Efficacy and biological correlates of response in a phase II study of venetoclax monotherapy in patients with acute myelogenous leukemia. Cancer Discov. *6*, 1106–1117. https://doi.org/10.1158/2159-8290.CD-16-0313.

365. Cai, Q., Xiao, J., Weng, C., and Chen, H. (2025). Efficacy and safety of venetoclax plus azacitidine based regimens in the treatment of relapsed or refractory acute myeloid leukemia: a systematic review and meta-analysis. Ann. Hematol. *104*, 4931–4948. https://doi.org/10.1007/s00277-025-06643-0.

366. Lu, J., Xue, S.-L., Wang, Y., He, X.-F., Hu, X.-H., Miao, M., Zhang, Y., Tang, Z.-X., Xie, J.-D., Yang, X.-F., et al. (2025). Venetoclax and decitabine vs intensive chemotherapy as induction for young patients with newly diagnosed AML. Blood *145*, 2645–2655. https://doi.org/10.1182/blood.2024027217.

367. Jilg, S., Reidel, V., Müller-Thomas, C., König, J., Schauwecker, J., Höckendorf, U., Huberle, C., Gorka, O., Schmidt, B., Burgkart, R., et al. (2016). Blockade of BCL-2 proteins efficiently induces apoptosis in progenitor cells of high-risk myelodysplastic syndromes patients. Leukemia *30*, 112–123. https://doi.org/10.1038/leu.2015.179.

368. Bogenberger, J.M., Kornblau, S.M., Pierceall, W.E., Lena, R., Chow, D., Shi, C.X., Mantei, J., Ahmann, G., Gonzales, I.M., Choudhary, A., et al. (2014). BCL-2 family proteins as 5-Azacytidine-sensitizing targets and determinants of response in myeloid malignancies. Leukemia *28*, 1657–1665. https://doi.org/10.1038/leu.2014.44.

369. Garcia-Manero, G., Platzbecker, U., Fenaux, P., Roboz, G.J., Fong, C.Y., Hus, M., Delage, R., Brunner, A., Lee, J.-H., Turgut, M., et al. (2025). MDS-1497: Primary analysis of the randomized, phase 3 VERONA study of venetoclax with azacitidine versus placebo with azacitidine in patients with treatment-naïve, intermediate and higher-risk myelodysplastic syndromes. Clin. Lymphoma Myeloma Leuk. *25*, S644–S645. https://doi.org/10.1016/S2152-2650(25)02083-X.

370. Chien, K.S., Rodriguez-Sevilla, J.J., Alvarado, Y., Montalban-Bravo, G., Hammond, D.E., Swaminathan, M., Bazinet, A., Kimberley, J., Bodden, K., Schneider, H., et al. (2024). A phase I study of the myeloid cell leukemia 1 (MCL1) inhibitor tapotoclax (AMG 176) in patients with myelodysplastic syndromes after hypomethylating agent failure. Leuk. Res. *147*, 107602. https://doi.org/10.1016/j.leukres.2024.107602.

371. Desai, P., Lonial, S., Cashen, A., Kamdar, M., Flinn, I., O'Brien, S., Garcia, J.S., Korde, N., Moslehi, J., Wey, M., et al. (2024). A phase 1 first-in-human study of the MCL-1 inhibitor AZD5991 in patients with relapsed/refractory hematologic malignancies. Clin. Cancer Res. *30*, 4844–4855. https://doi.org/10.1158/1078-0432.CCR-24-0028.

372. Yuda, J., Will, C., Phillips, D.C., Abraham, L., Alvey, C., Avigdor, A., Buck, W., Besenhofer, L., Boghaert, E., Cheng, D., et al. (2023). Selective MCL-1 inhibitor ABBV-467 is efficacious in tumor models but is associated with cardiac troponin increases in patients. Commun. Med. *3*, 154. https://doi.org/10.1038/s43856-023-00380-z.

373. Mason, K.D., Carpinelli, M.R., Fletcher, J.I., Collinge, J.E., Hilton, A.A., Ellis, S., Kelly, P.N., Ekert, P.G., Metcalf, D., Roberts, A.W., et al. (2007). Programmed anuclear cell death delimits platelet life span. Cell *128*, 1173–1186. https://doi.org/10.1016/j.cell.2007.01.037.

374. Khan, S., Zhang, X., Lv, D., Zhang, Q., He, Y., Zhang, P., Liu, X., Thummuri, D., Yuan, Y., Wiegand, J.S., et al. (2019). A selective BCL-XL PROTAC degrader achieves safe and potent antitumor activity. Nat. Med. *25*, 1938–1947. https://doi.org/10.1038/s41591-019-0668-z.

375. Wang, Z., Skwarska, A., Poigaialwar, G., Chaudhry, S., Rodriguez-Meira, A., Sui, P., Olivier, E., Jia, Y., Gupta, V., Fiskus, W., et al. (2025). Efficacy of a novel BCL-xL degrader, DT2216, in preclinical models of JAK2-mutated post-MPN AML. Blood *146*, 341–355. https://doi.org/10.1182/blood.2024027117.

376. Mahadevan, D., Barve, M., Mahalingam, D., Parekh, J., Kurman, M., Strauss, J., Tremaine, L., Hromas, R., Sills, J., McCulloch, J., et al. (2025). First in human phase 1 study of DT2216, a selective BCL-xL degrader, in patients with relapsed/refractory solid malignancies. J. Hematol. Oncol. *18*, 98. https://doi.org/10.1186/s13045-025-01753-8.

377. Lindeman, G.J., Fernando, T.M., Bowen, R., Jerzak, K.J., Song, X., Decker, T., Boyle, F., McCune, S., Armstrong, A., Shannon, C., et al. (2022). VERONICA: Randomized phase II study of fulvestrant and venetoclax in ER-positive metastatic breast cancer post-CDK4/6 inhibitors - efficacy, safety, and biomarker results. Clin. Cancer Res. *28*, 3256–3267. https://doi.org/10.1158/1078-0432.CCR-21-3811.

378. Croce, C.M., Vaux, D., Strasser, A., Opferman, J.T., Czabotar, P.E., and Fesik, S.W. (2025). The BCL-2 protein family: from discovery to drug development. Cell Death Differ. *32*, 1369–1381. https://doi.org/10.1038/s41418-025-01481-z.

379. Opferman, J.T., Letai, A., Beard, C., Sorcinelli, M.D., Ong, C.C., and Korsmeyer, S.J. (2003). Development and maintenance of B and T lymphocytes requires antiapoptotic MCL-1. Nature *426*, 671–676. https://doi.org/10.1038/nature02067.

380. Roberts, A.W., Seymour, J.F., Brown, J.R., Wierda, W.G., Kipps, T.J., Khaw, S.L., Carney, D.A., He, S.Z., Huang, D.C.S., Xiong, H., et al. (2012). Substantial susceptibility of chronic lymphocytic leukemia to BCL2 inhibition: results of a phase I study of navitoclax in patients with relapsed or refractory disease. J. Clin. Oncol. *30*, 488–496. https://doi.org/10.1200/JCO.2011.34.7898.

381. Fernández-Marrero, Y., Spinner, S., Kaufmann, T., and Jost, P.J. (2016). Survival control of malignant lymphocytes by anti-apoptotic MCL-1. Leukemia *30*, 2152–2159. https://doi.org/10.1038/leu.2016.213.

382. Pan, R., Hogdal, L.J., Benito, J.M., Bucci, D., Han, L., Borthakur, G., Cortes, J., DeAngelo, D.J., Debose, L., Mu, H., et al. (2014). Selective BCL-2 inhibition by ABT-199 causes on-target cell death in acute myeloid leukemia. Cancer Discov. *4*, 362–375. https://doi.org/10.1158/2159-8290.CD-13-0609.

383. Opferman, J.T., Iwasaki, H., Ong, C.C., Suh, H., Mizuno, S.-I., Akashi, K., and Korsmeyer, S.J. (2005). Obligate role of anti-apoptotic MCL-1 in the survival of hematopoietic stem cells. Science *307*, 1101–1104. https://doi.org/10.1126/science.1106114.

384. Vo, T.-T., Ryan, J., Carrasco, R., Neuberg, D., Rossi, D.J., Stone, R.M., Deangelo, D.J., Frattini, M.G., and Letai, A. (2012). Relative mitochondrial priming of myeloblasts and normal HSCs determines chemotherapeutic success in AML. Cell *151*, 344–355. https://doi.org/10.1016/j.cell.2012.08.038.

385. Gaulard, P., d'Agay, M.F., Peuchmaur, M., Brousse, N., Gisselbrecht, C., Solal-Celigny, P., Diebold, J., and Mason, D.Y. (1992). Expression of the bcl-2 gene product in follicular lymphoma. Am. J. Pathol. *140*, 1089–1095.

386. Zelenetz, A.D., Salles, G., Mason, K.D., Casulo, C., Le Gouill, S., Sehn, L.H., Tilly, H., Cartron, G., Chamuleau, M.E.D., Goy, A., et al. (2019). Venetoclax plus R- or G-CHOP in non-Hodgkin lymphoma: results from the CAVALLI phase 1b trial. Blood *133*, 1964–1976. https://doi.org/10.1182/blood-2018-11-880526.

387. Portell, C.A., Jegede, O.A., Wagner-Johnston, N., Nowakowski, G.S., Fletcher, C., Cohen, J.B., Evens, A.M., Rosenstein, L.J., Craig, J.W., Reddy, N., et al. (2025). Phase II study of venetoclax added to bendamustine and obinutuzumab in patients with high-risk follicular lymphoma as front-line therapy: PrE0403. Blood Cancer J. *15*, 93. https://doi.org/10.1038/s41408-025-01300-1.

388. Brieghel, C., Aarup, K., Torp, M.H., Andersen, M.A., Yde, C.W., Tian, X., Wiestner, A., Ahn, I.E., and Niemann, C.U. (2021). Clinical outcomes in patients with multi-hit TP53 chronic Lymphocytic leukemia treated with ibrutinib. Clin. Cancer Res. *27*, 4531–4538. https://doi.org/10.1158/1078-0432.CCR-20-4890.

389. Rossi, D., Khiabanian, H., Spina, V., Ciardullo, C., Bruscaggin, A., Famà, R., Rasi, S., Monti, S., Deambrogi, C., De Paoli, L., et al. (2014). Clinical impact of small TP53 mutated subclones in chronic lymphocytic leukemia. Blood *123*, 2139–2147. https://doi.org/10.1182/blood-2013-11-539726.

390. McGranahan, N., and Swanton, C. (2017). Clonal heterogeneity and tumor evolution: Past, present, and the future. Cell *168*, 613–628. https://doi.org/10.1016/j.cell.2017.01.018.

391. Turajlic, S., Sottoriva, A., Graham, T., and Swanton, C. (2019). Resolving genetic heterogeneity in cancer. Nat. Rev. Genet. *20*, 404–416. https://doi.org/10.1038/s41576-019-0114-6.

392. Vendramin, R., Litchfield, K., and Swanton, C. (2021). Cancer evolution: Darwin and beyond. EMBO J. *40*, e108389. https://doi.org/10.15252/embj.2021108389.

393. Kater, A.P., Arslan, Ö., Demirkan, F., Herishanu, Y., Ferhanoglu, B., Diaz, M.G., Leber, B., Montillo, M., Panayiotidis, P., Rossi, D., et al. (2024). Activity of venetoclax in patients with relapsed or refractory chronic lymphocytic leukaemia: analysis of the VENICE-1 multicentre, open-label, single-arm, phase 3b trial. Lancet Oncol. *25*, 463–473. https://doi.org/10.1016/S1470-2045(24)00070-6.

394. Stilgenbauer, S., Eichhorst, B., Schetelig, J., Coutre, S., Seymour, J.F., Munir, T., Puvvada, S.D., Wendtner, C.-M., Roberts, A.W., Jurczak, W., et al. (2016). Venetoclax in relapsed or refractory chronic lymphocytic leukaemia with 17p deletion: a multicentre, open-label, phase 2 study. Lancet Oncol. *17*, 768–778. https://doi.org/10.1016/S1470-2045(16)30019-5.

395. Stilgenbauer, S., Eichhorst, B., Schetelig, J., Hillmen, P., Seymour, J.F., Coutre, S., Jurczak, W., Mulligan, S.P., Schuh, A., Assouline, S., et al. (2018). Venetoclax for patients with Chronic Lymphocytic Leukemia with 17p deletion: Results from the full population of a phase II pivotal trial. J. Clin. Oncol. *36*, 1973–1980. https://doi.org/10.1200/JCO.2017.76.6840.

396. Al-Sawaf, O., Zhang, C., Lu, T., Liao, M.Z., Panchal, A., Robrecht, S., Ching, T., Tandon, M., Fink, A.-M., Tausch, E., et al. (2021). Minimal residual disease dynamics after venetoclax-obinutuzumab treatment: Extended off-treatment follow-up from the randomized CLL14 study. J. Clin. Oncol. *39*, 4049–4060. https://doi.org/10.1200/JCO.21.01181.

397. Kater, A.P., Harrup, R., Kipps, T.J., Eichhorst, B., Owen, C.J., Assouline, S., Lamanna, N., Robak, T., de la Serna, J., Jaeger, U., et al. (2025). The MURANO study: final analysis and retreatment/crossover substudy results of VenR for patients with relapsed/refractory CLL. Blood *145*, 2733–2745. https://doi.org/10.1182/blood.2024025525.

398. Höckendorf, U., Yabal, M., Herold, T., Munkhbaatar, E., Rott, S., Jilg, S., Kauschinger, J., Magnani, G., Reisinger, F., Heuser, M., et al. (2016). RIPK3 restricts myeloid leukemogenesis by promoting cell death and differentiation of leukemia initiating cells. Cancer Cell *30*, 75–91. https://doi.org/10.1016/j.ccell.2016.06.002.

399. Höckendorf, U., Dutta, S., Kloos, A., Runtsch, M., Zötsch, C., Vosberg, S., Wang, Y., Kienreich, S., Flasch, B., Malovan, G., et al. (2025). Lymphotoxin alpha eradicates acute myeloid leukemia and simultaneously promotes healthy hematopoiesis in mice. Sci. Transl. Med. *17*, eadu3313. https://doi.org/10.1126/scitranslmed.adu3313.

400. Lalaoui, N., Hänggi, K., Brumatti, G., Chau, D., Nguyen, N.-Y.N., Vasilikos, L., Spilgies, L.M., Heckmann, D.A., Ma, C., Ghisi, M., et al. (2016). Targeting p38 or MK2 enhances the anti-leukemic activity of smac-mimetics. Cancer Cell *29*, 145–158. https://doi.org/10.1016/j.ccell.2016.01.006.

401. Ferris, R.L., Harrington, K., Schoenfeld, J.D., Tahara, M., Esdar, C., Salmio, S., Schroeder, A., and Bourhis, J. (2023). Inhibiting the inhibitors: Development of the IAP inhibitor xevinapant for the treatment of locally advanced squamous cell carcinoma of the head and neck. Cancer Treat. Rev. *113*, 102492. https://doi.org/10.1016/j.ctrv.2022.102492.

402. Vince, J.E., Wong, W.W.-L., Khan, N., Feltham, R., Chau, D., Ahmed, A.U., Benetatos, C.A., Chunduru, S.K., Condon, S.M., McKinlay, M., et al. (2007). IAP antagonists target cIAP1 to induce TNFalpha-dependent apoptosis. Cell *131*, 682–693. https://doi.org/10.1016/j.cell.2007.10.037.

403. Yabal, M., Müller, N., Adler, H., Knies, N., Groß, C.J., Damgaard, R.B., Kanegane, H., Ringelhan, M., Kaufmann, T., Heikenwälder, M., et al. (2014). XIAP restricts TNF- and RIP3-dependent cell death and inflammasome activation. Cell Rep. *7*, 1796–1808. https://doi.org/10.1016/j.celrep.2014.05.008.

404. Sun, X.-S., Tao, Y., Le Tourneau, C., Pointreau, Y., Sire, C., Kaminsky, M.-C., Coutte, A., Alfonsi, M., Boisselier, P., Martin, L., et al. (2020). Debio 1143 and high-dose cisplatin chemoradiotherapy in high-risk locoregionally advanced squamous cell carcinoma of the head and neck: a double-blind, multicentre, randomised, phase 2 study. Lancet Oncol. *21*, 1173–1187. https://doi.org/10.1016/S1470-2045(20)30327-2.

405. Tao, Y., Sun, X.-S., Pointreau, Y., Le Tourneau, C., Sire, C., Kaminsky, M.-C., Coutte, A., Alfonsi, M., Calderon, B., Boisselier, P., et al. (2023). Extended follow-up of a phase 2 trial of xevinapant plus chemoradiotherapy in high-risk locally advanced squamous cell carcinoma of the head and neck: a randomised clinical trial. Eur. J. Cancer *183*, 24–37. https://doi.org/10.1016/j.ejca.2022.12.015.

406. Bourhis, J., Licitra, L.F., Burtness, B., Psyrri, A., Haddad, R., Harrington, K., Cohen, E.E.W., Tao, Y., Tiscoski, K.A., Matitashvili, A., et al. (2025). Xevinapant or placebo plus platinum-based chemoradiotherapy in unresected locally advanced squamous cell carcinoma of the head and neck

(TrilynX): A randomized, phase III study. J. Clin. Oncol. *43*, 3209–3220. https://doi.org/10.1200/JCO-25-00272.

407. Brinkmann, K., McArthur, K., Malelang, S., Gibson, L., Tee, A., Elahee Doomun, S.N., Rowe, C.L., Arandjelovic, P., Marchingo, J.M., D'Silva, D., et al. (2025). Relative importance of the anti-apoptotic versus apoptosis-unrelated functions of MCL-1 in vivo. Science *389*, 1003–1011. https://doi.org/10.1126/science.adw1836.

408. Wahida, A., Buschhorn, L., Fröhling, S., Jost, P.J., Schneeweiss, A., Lichter, P., and Kurzrock, R. (2023). The coming decade in precision oncology: six riddles. Nat. Rev. Cancer *23*, 43–54. https://doi.org/10.1038/s41568-022-00529-3.