can swim among other animals without distressing them, thereby allowing close-up study. Li and co-workers' research now pushes the boundaries of what can be achieved: the replacement of rigid protective enclosures for electronic components by distributed electronics embedded in a soft material paves the way to a new generation of deep-sea explorers.

There is, however, more work to do before the ocean can be populated with robots of this type of design. Li and co-workers' machine is slower than previously reported underwater robots[6], and cannot withstand sizeable disturbances – it could easily be swept away by underwater currents. Its locomotor capabilities will also need to be optimized for practical applications. However, Li and colleagues' approach lays the foundations for future generations of resilient and reliable deep-sea explorers.

In the long term, one can predict avenues of research being opened up for marine biology, in which soft robots safely navigate coral reefs or underwater caves, to collect delicate specimens without damaging them. Swarms of underwater soft robots, with the ability to crawl on the seabed, anchor themselves on to specific structures, or swim over particular areas, could contribute to the development of technologies for various other applications. These might include monitoring the ocean, cleaning up and preventing sea pollution or preserving marine biodiversity. More fundamentally, they could help researchers to explore the vast uncharted depths of the oceans.

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Biochemistry

Active membrane rupture spurs a range of cell deaths

Sebastian Hiller & Petr Broz

Rupture of the plasma membrane in different forms of cell death was long thought to be a passive process. The finding that it is an active one, mediated by a specific membrane protein, reveals an unexpected feature shared by dying cells. **See p.131**

Remarkable control mechanisms exist in multicellular organisms to ensure that their cells function normally and are properly removed when necessary. These removal mechanisms include multiple forms of cell death, most of which end in rupture of the cell's plasma membrane. Until now, this breach of the cell's outer layer was generally thought to be a consequence of an uncontrolled influx of water, driven by an ionic imbalance that boosts cellular osmotic pressure. However, Kayagaki *et al.*[1] report on page 131 that this rupture is an active process, mediated by the plasma-membrane protein ninjurin-1 (NINJ1). This protein exerts its effect during the final step of many types of cell death.

Cells of multicellular organisms die in one of two main ways. Either they die passively as a consequence of damage they have incurred, or they die in a well-regulated, programmed manner as part of normal events such as development, homeostasis or the elimination of malignant and infected cells[2].

The defining feature of passive cell death (also known as necrosis) has long been thought to be the loss of integrity of the plasma membrane, which distinguishes necrosis from programmed cell death. As a consequence of plasma-membrane disruption, necrosis resembles a cellular explosion that releases a plethora of intracellular molecules, including proteins, nucleic acids and metabolites. Some of these act as danger signals, known as damage-associated molecular patterns (DAMPs), that alert neighbouring cells to the injury and thus help to induce inflammation. By contrast, apoptosis, the most-studied form of programmed cell death, preserves membrane integrity to enable immunologically 'silent', non-inflammatory removal of dead cells – a process in which they are taken up (engulfed) and dismantled by other cells as the tissue is repaired.

The past two decades have witnessed the discovery[3] of multiple forms of programmed cell death – such as pyroptosis, necroptosis and ferroptosis – that combine a programmed cellular demise with what seems to be passive disintegration of the plasma membrane. A common feature of most of these deaths is the formation of large pores, made of protein, in the plasma membrane[4]. These include, for example, the pores formed by various proteins called gasdermins, which initiate pyroptosis[5]; and the MLKL channel that is assembled in necroptotic cells[6]. Formation of either of these pores is followed by cellular swelling mediated by osmotic pressure, and then rupture of the plasma membrane (Fig. 1).

This rupture was thought to be a passive event, but Kayagaki and colleagues reveal that it is actively regulated. The authors made this discovery by analysing a group of mice with mutations at random genomic locations and examining pyroptosis of macrophage cells of the animals' immune systems. Some of the dying macrophages did not release the enzyme lactate dehydrogenase as usual, indicating that rupture of the plasma membrane was abnormal. Further investigation revealed that these mice had a mutation in the gene encoding NINJ1, which resulted in no detectable production of this protein.

NINJ1 was known to have a role in cell adhesion, but no direct ties had previously linked it to cell death[7,8]. The absence of NINJ1 prevented the dying mouse cells from releasing other large proteins, such as HMGB1, but did not block them from secreting a smaller protein, IL-1α (a member of the IL-1 family of immune-signalling molecules called cytokines), which is small enough to pass through a pore made by gasdermin D.

The lack of NINJ1 had a striking effect on macrophage shape. During pyroptosis, cells normally swell and form ballooning membrane protrusions that eventually rupture[9]. Dying cells deficient in NINJ1 also swelled and made such protrusions, but they didn't burst. Therefore, the rupture that usually occurs is clearly not caused by osmotic pressure, but instead depends on specific events that involve NINJ1. Moreover, Kayagaki and colleagues' data provide evidence that gasdermin-pore-induced cytokine release and cell swelling are distinct processes that can occur independently of plasma-membrane rupture.

Although this finding alone already provides a spectacular twist to a long-studied phenomenon, the big surprise came when the role of NINJ1 was examined in other forms of cell death. NINJ1 also mediated plasma-membrane rupture after toxin-induced cell permeabilization, on the induction of necroptosis, and even during secondary necrosis

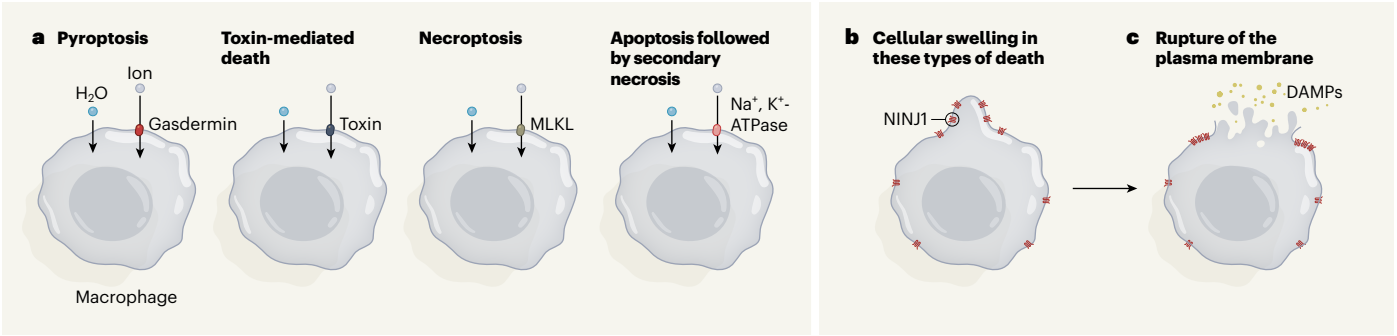

Figure 1 | Regulated rupture of the plasma membrane is an end point of multiple cell-death pathways. a, Human cells, such as macrophages, can die by a range of mechanisms, including pyroptosis, toxin-mediated death, necroptosis and apoptosis (followed by a process called secondary necrosis). A common feature of these deaths is an increase in cellular osmotic pressure, presumably arising from an ionic imbalance that drives water entry. This imbalance is triggered by ion movement through protein channels or pores, such as those formed by gasdermin proteins, toxins or MLKL channels. In apoptosis, inactivation of the Na^+, K^+-ATPase enzyme causes ion accumulation in dying cells[10]. **b**, The water entry causes cellular swelling, and bubble-like protrusions form. Kayagaki *et al.*[1] report that rupture of the plasma membrane in these types of dying cell does not occur passively, as previously thought. Instead, it is an active process that requires the protein ninjurin-1 (NINJ1). **c**, To mediate rupture of the plasma membrane, NINJ1 aggregates (oligomerizes). This rupture releases cytoplasmic content, including molecules called damage-associated molecular patterns (DAMPs), which trigger inflammation in neighbouring tissue.

of apoptotic cells. Secondary necrosis occurs if apoptotic cells are not engulfed and removed in a timely manner. Thus, NINJ1 is a common denominator at the end of many cell-death pathways.

NINJ1 is ubiquitously expressed[8], and is evolutionarily conserved, from fruit flies to humans. How might this relatively small (16 kilodaltons) protein mediate such striking effects? Its structure is predicted to contain two transmembrane helices, as well as an evolutionarily conserved extracellular helix that is needed for NINJ1 to function properly. Working out whether this helix senses a signal or serves to disrupt the membrane during cell death will require more study. Of note, this helix seems to have a mixed hydrophobic and hydrophilic (amphiphilic) character, a property similar to that of the helices found in other membrane-disrupting proteins, such as melittin or BAX.

Importantly, Kayagaki and colleagues' findings will transform cell biology in a way that goes beyond just revealing NINJ1's function. Their study underscores the enormous strength and resilience of the intact plasma membrane. It also reduces the number of events in cell biology considered to be non-specific, highlighting how stringently organisms control the fate of their cells until the very last moment of cellular existence.

Many questions remain to be answered. What signal or property is sensed by NINJ1 to activate its function in dying cells? What mechanisms, if any, exist to prevent accidental activation of NINJ1? It would be interesting to know whether NINJ1 requires other factors when mediating membrane rupture. Do other proteins with a similar function exist? And, of course, what is the structure of the membrane-rupturing entity that NINJ1 presumably forms?

Answering these questions might lead to new therapeutic strategies aimed at inhibiting NINJ1, or related proteins, that could convert necrotic death to a type of death with a less inflammatory outcome. Such treatments would thereby reduce the general level of inflammation in tissue, presumably with positive effects for chronic or acute inflammatory disorders.

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Coronavirus

Surprising effects of antibodies in severe COVID

Matteo Gentili & Nir Hacohen

Defects in the immune defences induced by the protein interferon are associated with some severe cases of COVID-19. An analysis of patients' blood samples sheds light on how antibodies might contribute to these defects. **See p.124**

Infection with the SARS-CoV-2 virus can lead to diverse outcomes, ranging from no symptoms to varying degrees of disease severity, spanning mild illness to death. What determines the degree of severity is unclear, but mounting evidence points to exacerbated and abnormal responses in the innate branch of the immune system as a main driver of major illness. Combes *et al.*[1] present a study on page 124 investigating the hallmarks of COVID-19 severity.

The authors analysed cells, including immune cells, in blood samples from 21 people with COVID-19 and 25 uninfected individuals who were either healthy or had a lung injury or breathing difficulties. They monitored gene expression during the course of the infection as patients went on to develop either what was categorized as mild–moderate COVID-19 (which required a short hospital stay without the need for intensive care or mechanical ventilation) or severe COVID-19 (requiring intensive care and mechanical ventilation). The authors found that the cells of people with