CANCER IMMUNOTHERAPY

Lipid nanodiscs give cancer a STING

Lipid nanodiscs carrying a potent STING agonist penetrate deep into solid tumours compared with gold-standard liposomes and enable long-term antitumour immunotherapy.

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The past few years have witnessed the great success of cancer immunotherapies, which harness a patient's immune system for cancer treatment. Immune checkpoint blockade therapy, chimeric antigen receptor T cell therapy, oncolytic viruses, bispecific antibodies and cancer vaccines have revolutionized cancer treatment, with many novel immunotherapy strategies currently under clinical and pre-clinical investigation[1]. However, a substantial hurdle that these immunotherapies still face is reaching the immunosuppressive areas of dense tumour tissue. As such, the cyclic GMP–AMP synthase (cGAS)–stimulator of interferon genes (STING) pathway has emerged as a promising target for cancer immunotherapy. The cGAS–STING pathway has recently been found to detect cytosolic DNA, including tumour-derived DNA, and induce innate anti-tumour immune responses[2]. Dysfunction of the cGAS–STING pathway, which results in immune suppression, is involved in many cancers. Therefore, agonist molecules that stimulate the STING pathway can be harnessed to create powerful immunotherapies. When a STING agonist is administered, it leads to the production of a variety of pro-inflammatory cytokines and chemokines, including type I interferons, which stimulate dendritic cells (DCs) to present tumour antigens to cytotoxic T cells[3].

A number of natural and synthetic STING agonists have been discovered and developed. These agonists have been tested both in preclinical models and in the clinic for cancer immunotherapy. Cyclic dinucleotides (CDNs), such as cyclic dimeric guanosine monophosphate (c-di-GMP), cyclic dimeric adenosine monophosphate (c-di-AMP) and cyclic GMP–AMP (cGAMP), are a class of STING agonists that can elicit strong immune responses. However, there are three main challenges in using CDNs for cancer immunotherapy. Natural CDNs are small hydrophilic molecules that are highly negatively charged and cannot cross cell membranes. CDNs have a non-canonical 2′,5′-phosphodiester linkage and/or a canonical 3′,5′ linkage (c[G(2′,5′)pA(3′,5′)p]) and are susceptible to enzymatic degradation, leading to low bioavailability in target tissues. Finally, delivering CDNs directly to tumour tissues is challenging as STING activation in off-target tissue induces toxicity, which leads to narrow therapeutic windows. In this issue of *Nature Materials*, Darrell Irvine and colleagues[4] tackle these limitations with a CDN-conjugated lipid nanodisc (LND) for improved cancer immunotherapy compared with gold standard spherical liposomes.

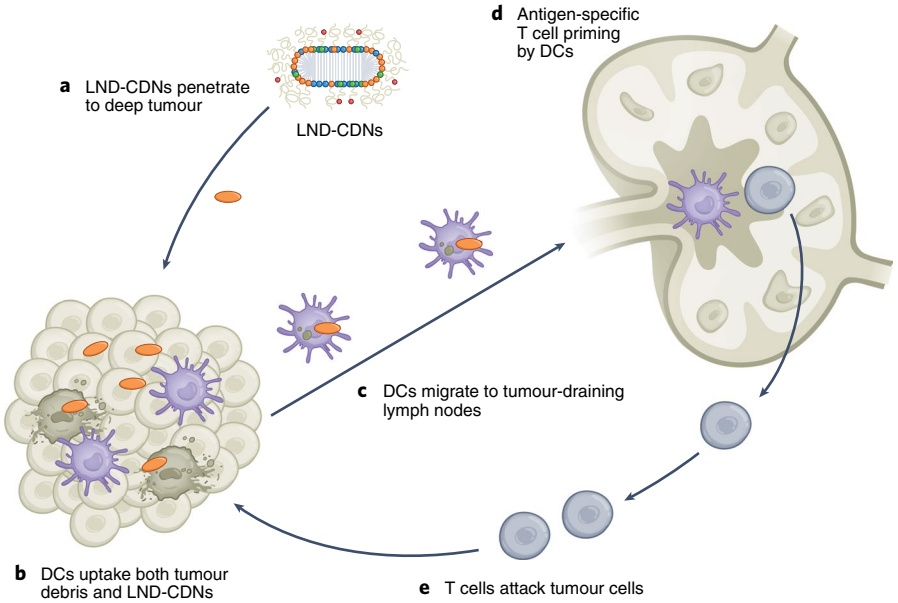

Fig. 1 | Lipid nanodiscs give cancer immunotherapy a STING. a, STING-activating CDNs were conjugated to PEGylated lipids (CDN–PEG-lipids) via a cleavable linker and incorporated into LNDs. Upon systemic administration, LND-CDNs penetrate deep into tumours and achieve substantial accumulation of the CDNs throughout the tumour tissue to reach dying tumour cells and DCs. **b**, DCs acquired tumour debris in the wave of tumour cell death following CDN-mediated vascular collapse. LND-CDNs and tumour debris are taken up simultaneously by DCs in the tumour or lymph. **c**,**d**, DCs then migrate to the tumour-draining lymph nodes (**c**) and antigen-specific T cells are primed (**d**). **e**, Antigen-specific T cells can then recognize and attack tumour cells. 'LND-CDN' in panel **a** adapted with permission from ref. [4], Springer Nature Ltd.

Nanomaterials such as lipid nanoparticles and liposomes have demonstrated great promise for drug and gene delivery for disease treatment. Nanomaterials can be engineered with tunable size, morphology and surface modifications, which are shown to affect the distribution, metabolism and excretion, and thus affect the efficacy and toxicity, of therapeutic cargo[5,6]. Previous studies have shown that liposomes and polymeric nanoparticles can help overcome biological barriers for enhanced CDN delivery[7–9]. Irvine and colleagues synthesized a CDN prodrug by linking CDNs to the polyethylene glycol (PEG)-lipid component of their LND with a peptide linker that is cleaved by an intracellular protease upon cellular uptake (Fig. 1). This protease is highly expressed by tumour cells compared with normal cells, which ensures increased CDN release in tumour cells and decreased off-target toxicity. The LND-CDNs greatly

enhanced CDN half-life compared with free CDNs. Compared with CDNs delivered using liposomes, systemic administration of LND-CDNs in mice achieved greater accumulation of the CDNs throughout the tumour tissue to reach dying tumour cells and DCs. This is likely to be due to the high aspect ratio and unique shape of LNDs, which provides them with increased flexibility that enhances tumour penetration compared with spherical liposomes. Importantly, this allows LND-CDNs and tumour debris to be taken up simultaneously by DCs in the tumour and lymph before DC migration to the tumour-draining lymph nodes, where DCs present tumour antigens to prime a long-term adaptive immune response. Thus, this therapy has the ability to induce in situ anti-tumour vaccination.

After testing a single-dose therapy in three different tumour models that use tumour cells derived from the same strain of mice, the researchers found that LND-CDNs achieved ~80% tumour rejection in the MC38 colon cancer mouse model and a ~50% increase in median survival time in the 4T1 breast cancer mouse model. However, LND-CDN treatment of STING-/- mice bearing MC38 tumours was completely ineffective, confirming the role of STING in this therapeutic approach. Surprisingly, the LND-CDNs induced only transient weight loss in mice, which recovered within a few days with no signs of major organ toxicity at the administered doses. Taken together, these mouse models confirmed the ability of LND-CDNs to induce acute tumour shrinkage while priming the adaptive immune system for a robust anti-tumour response.

These results are of broader interest to the cancer therapy field, as this lipid nanodisc platform can potentially be harnessed to deliver a range of therapeutic cargo into tumours beyond STING agonists. For example, LNDs carrying chemotherapeutics accumulating in tumours could potentially enhance antitumour efficacy while reducing toxicity; LNDs delivering imaging modalities could aid in earlier tumour diagnosis; LNDs carrying messenger RNA (mRNA) encoding cytokines, checkpoint blockades or other tumour suppressor proteins may induce better antitumour outcomes. One of the key advantages of the LND-CDN described here is that it can induce higher CDN accumulation in tumour cells due to its protease-sensitive linker and unique morphology compared with spherical liposomes. This can mitigate the potential off-target effects of systemic delivery of therapeutic cargo. It will be interesting to see how this untargeted LND and passive tumour accumulation approach compares with formulations further functionalized with active targeting ligands, such as monoclonal antibodies. Future work should also address whether CDNs adjuvant a humoral response against the PEG component of the nanodisc, which could induce an anti-PEG antibody response that results in rapid clearance of the nanodisc. Such a response could be avoided by replacing PEG with PEG alternatives such as other zwitterionic polymers[10] currently under investigation in the fields of nanomedicine and biomaterials. While further investigation is required to fully harness the potential of LNDs for cancer immunotherapy, the LND platform makes a notable stride forward in therapeutic efficacy compared with traditional spherical liposomal carriers. ❐

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Competing interests

The authors declare no competing interests.