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Innovative nanoparticle-based approaches for modulating neutrophil extracellular traps in diseases: from mechanisms to therapeutics

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Abstract

Neutrophil extracellular traps (NETs) participate in both host defense and the pathogenesis of various diseases, such as infections, thrombosis, and tumors. While they help capture and eliminate pathogens, NETs' excessive or dysregulated formation can lead to tissue damage and disease progression. Therapeutic strategies targeting NET modulation have shown potential, but challenges remain, particularly in achieving precise drug delivery and maintaining drug stability. Nanoparticle (NP)-based drug delivery systems offer innovative solutions for overcoming the limitations of conventional therapies. This review explores the biological mechanisms of NET formation, their interactions with NPs, and the therapeutic applications of NP-based drug delivery systems for modulating NETs. We discuss how NPs can be designed to either promote or inhibit NET formation and provide a comprehensive analysis of their potential in treating NET-related diseases. Additionally, we address the current challenges and future prospects for NP-based therapies in NET research, aiming to bridge the gap between nanotechnology and NET modulation for the development of novel therapeutic approaches.

Keywords Neutrophil extracellular traps (NETs), Nanoparticle (NP), Drug delivery, Neutrophils, NET modulation

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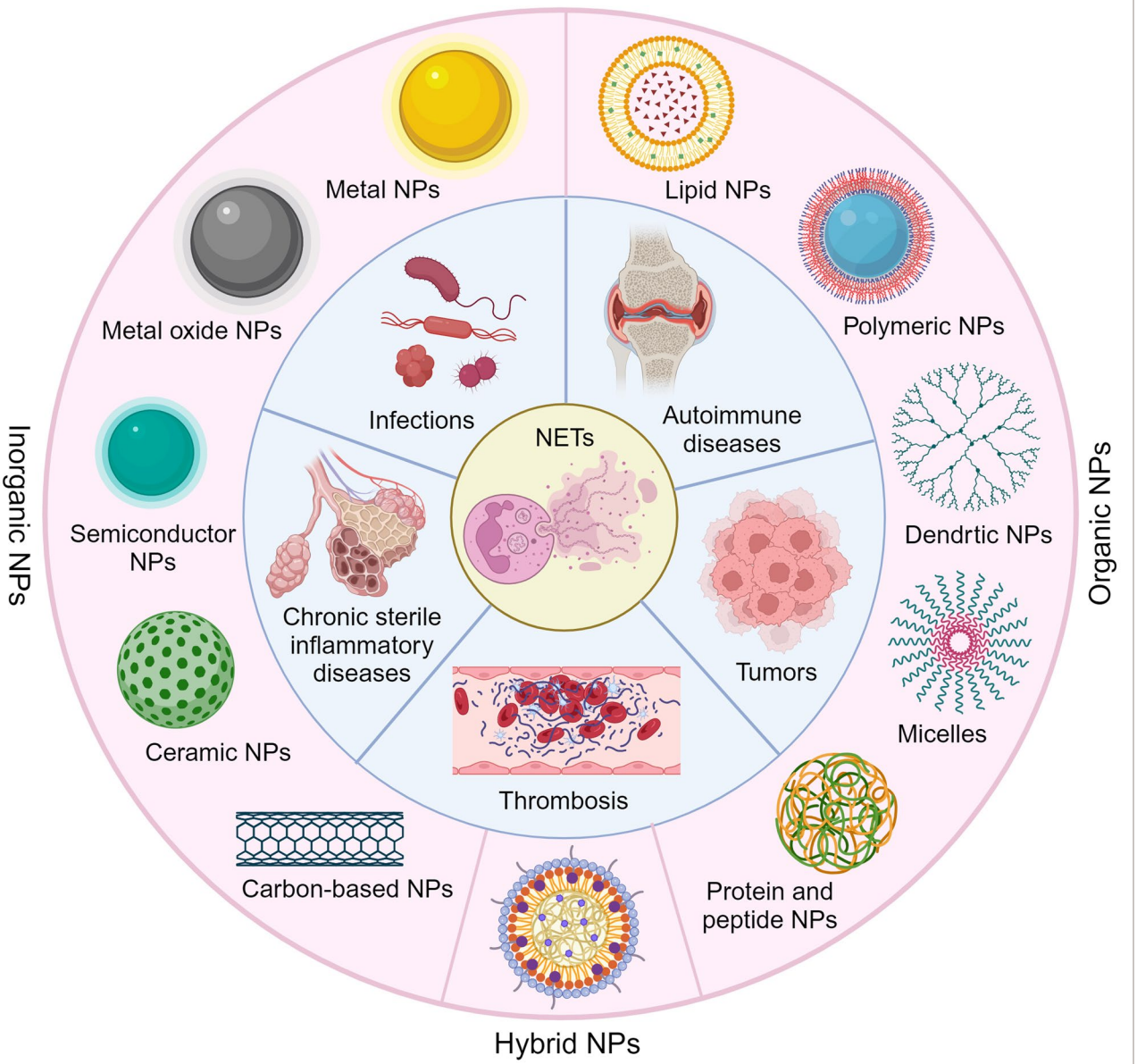
Graphical Abstract



Introduction

Neutrophils, also known as polymorphonuclear leukocytes, are the most numerous white blood cells, usually accounting for 50–70% of the total number of white blood cells in the body [1]. They are produced in the bone marrow, are one of the major members of the intrinsic immune system, and have a variety of important functions, including participation in inflammatory responses, the phagocytosis of pathogens and dead cells, and the release of killer substances. Through blood circulation, neutrophils can rapidly enter sites of infection or inflammation and eliminate foreign pathogens to protect the host [2]. As well as directly engulfing and digesting invading pathogens, neutrophils can release neutrophil extracellular trapping nets (NETs) to capture them and limit their spread [3].

The process of NET formation has been termed "NETosis," which primarily occurs through two distinct pathways: the classical pathway (also known as the suicidal NETosis pathway) and the alternative pathway (often

referred to as vital NETosis). These pathways play different roles in immune responses and disease pathology [4, 5]. In the classical pathway, pattern recognition receptors (PRRs) like Toll-like receptors (TLRs) recognize damage-associated molecular patterns (DAMPs) released by injured or dying cells and pathogen-associated molecular patterns (PAMPs) from pathogens [6–8]. This recognition triggers intracellular signaling cascades, promoting neutrophil recruitment through rolling and adhesion. At inflammation sites, Ca^{2+} in the neutrophils can be released from the endoplasmic reticulum. As calcium concentrations increase, the Ras-Raf-MEK-ERK signaling pathway is activated, leading to the activation of protein kinase C (PKC). This induces the production of reactive oxygen species (ROS) via NADPH oxidase (NOX) and the release of neutrophil elastase (NE) and myeloperoxidase (MPO) from azurophilic granules [9, 10]. These enzymes translocate to the nucleus, where NE degrades histones and MPO and peptidylarginine deiminase 4 (PAD4) citrullinate histones to promote chromatin decondensation [11]. The nuclear envelope then ruptures, expelling chromatin mixed with granulin and cytoplasmic proteins into the extracellular space as NETs, leading to neutrophil death [12]. This NET formation process takes several hours.

The alternative pathway involves activated neutrophils releasing NETs without resulting in cell death. This rapid process occurs within minutes and is usually triggered by activated platelets, bacterial peptides, TLR2, TLR4, and complement components [13, 14]. NE is translocated to the nucleus independently of NOX activity during this process, initiating chromatin decondensation. Decondensed nuclear DNA coated with antimicrobial proteins, such as NE and MPO, is packaged into vesicles and expelled through the plasma membrane while maintaining its integrity. This mechanism allows neutrophils to continue to perform functions such as phagocytosis and chemotaxis even after the release of NETs [4].

NETs play an important role in immune responses by capturing and immobilizing a variety of pathogens, preventing their proliferation and facilitating their elimination. NETs may also serve as scaffolds for various immune cell interactions, enhancing local inflammation and modulating immune responses [15, 16]. However, excessive or dysregulated NET formation can cause tissue damage and the development of a variety of diseases, including autoimmune diseases, thrombosis, and tumor metastasis. Given their dual role in host defense and disease pathogenesis, NETs could therefore serve as key targets for therapeutic intervention. Understanding the delicate balance between NETs' beneficial and detrimental effects is essential for developing effective strategies to modulate them in various clinical settings [17–19]. Nanoparticle (NP)-based delivery systems load drugs within nanoscale materials, offering significant advantages for NET-targeted therapies, including precise delivery, controlled release, enhanced drug stability, and improved bioavailability [20, 21].

This review comprehensively analyzes the application of NP-based drug delivery systems for NET modulation. NETs' formation mechanisms, biological functions, and pathological effects in various diseases are systematically investigated to elucidate NP–NET interactions. Since NETs play a crucial role in the development and progression of various diseases (e.g., infections, thrombosis, wound healing, trauma, stroke, and tumors), we highlight and summarize the therapeutic applications and mechanisms of NP-based drug delivery systems that target and modulate NETs by promoting or inhibiting their formation (Fig. 1). We also discuss the existing limitations and challenges in this field and propose future directions for research and development. Our aim was to integrate nanotechnology with NETs and provide insights for the development of novel therapeutic NPs targeting NET modulation.

Roles of NETs in various diseases

Released by neutrophils in response to various stimuli, NETs play a role in immune defense against pathogenic infections and regulate immune responses together with other immune cells [11]. Because excessive or dysregulated NET formation can trigger and amplify inflammatory responses, leading to tissue damage and a variety of diseases [22, 23], it is essential to clarify NETs' roles in different diseases to elucidate their pathogenesis and develop targeted therapeutic strategies with the overall goal of improving the prognosis of patients with NET-associated diseases.

Infections

NETs are composed of decondensed chromatin and antimicrobial proteins in a web-like structure, which acts as a physical barrier that captures and immobilizes pathogens. Antimicrobial proteins such as NE and MPO are embedded in the DNA backbone of NETs and directly eliminate pathogens by degrading their structural components and generating antimicrobial substances [14, 24]. NETs thus prevent the pathogens from spreading throughout the body, which is essential for controlling infection and limiting tissue damage [23]. NETs also act as scaffolds to enhance local immune responses by promoting the recruitment and activation of other immune cells (e.g., macrophages and dendritic cells) [25].

However, in certain infections, NETs play a deleterious role in infectious diseases [26], being capable of killing epithelial and endothelial cells, which damages tissue

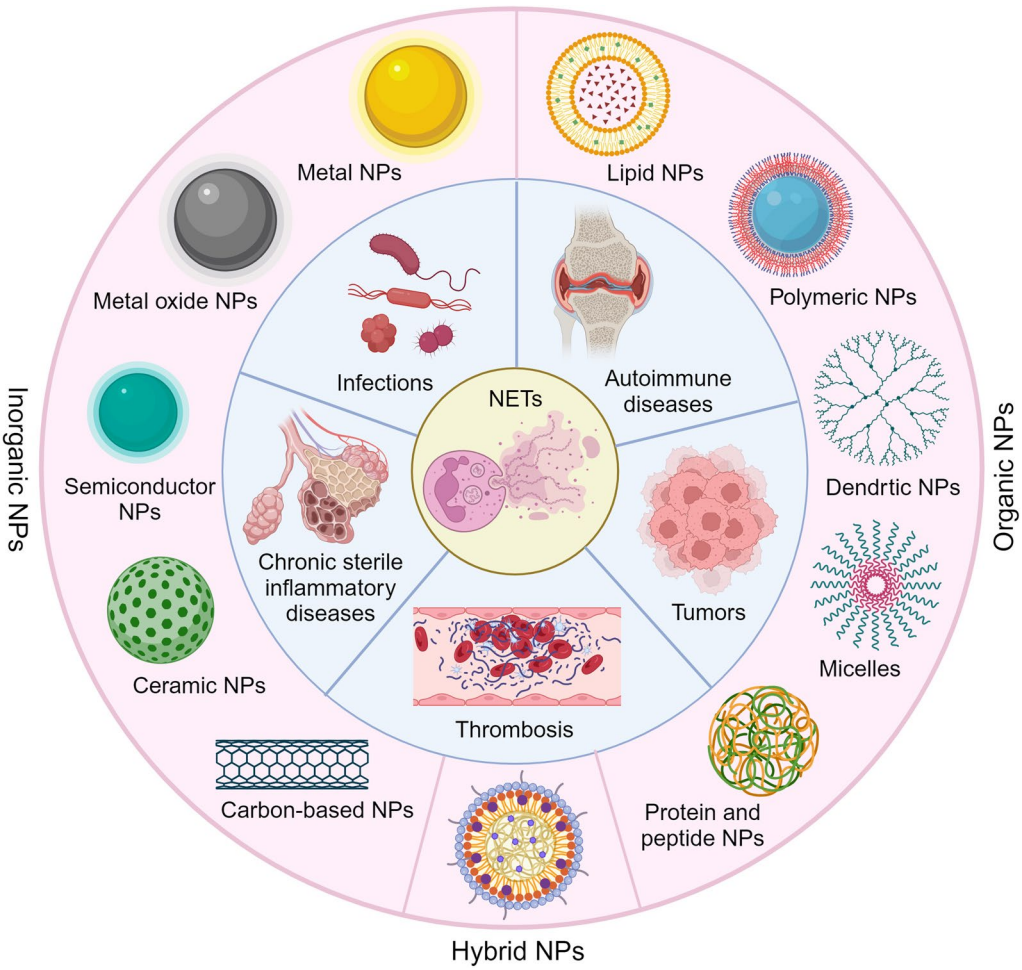


Fig. 1 Classification of NPs and their application in modulating NETs in different diseases. The classification of NPs based on composition that commonly used in biomedical applications (outer ring) and application of NPs in modulating NETs in infections, autoimmune diseases, tumors, thrombosis, and chronic sterile inflammatory diseases (inner ring). Created with BioRender.com

and organs [27]. For example, excessive NETosis damages epithelial cells in fungal infections of the lungs [16]. In bloodstream methicillin-resistant *Staphylococcus aureus* infections, NETs utilize NEs to damage hepatic vasculature. Collateral damage to host tissues can be effectively prevented by inhibiting NET production using a PAD4-deficient animal model or preventing NET formation and proteolytic activity using NE knockout [28]. In the lipopolysaccharide (LPS)-induced mouse model of acute lung injury (ALI), NETs also injure alveolar epithelial cells, mediating cytotoxic effects through the histones and MPOs involved in the destruction of lung tissue [27].

Autoimmune diseases

In autoimmune diseases such as systemic lupus erythematosus (SLE) and rheumatoid arthritis (RA), NETs can expose chromatin and cytoplasmic components, which possibly act as autoantigens to the host immune system. This leads to autoantibody production, triggering a cycle of inflammation and tissue damage. In addition, impaired NET clearance can exacerbate autoimmune diseases [29]. NETs are normally degraded by serum nucleases and macrophages in healthy individuals [30]; however, in patients with autoimmune diseases, impaired clearance results in NET accumulation, escalating their role as self-antigens and exacerbating inflammation [31]. This compromised clearance is associated with the presence of anti-NET antibodies, which inhibit NET degradation [32].

NETs' pathogenic mechanisms have been studied extensively in a number of autoimmune diseases. For example, in SLE, the presence of NETs containing modified histones and DNA stimulates antinuclear antibody production, leading to pathogenesis [33]. Larger amounts of anti-nuclear and anti-dsDNA autoantibodies in the plasma of SLE patients have been linked to higher NET

release [34]. NETs containing large amounts of citrullinated histones and other modified proteins have been found in the blood and tissues of patients with SLE; these structures not only cause systemic inflammation but also organ damage, such as to the kidneys (lupus nephritis) [17].

Similarly, NETs are found in the inflammatory environment of the joints of patients with RA; the DNA and proteins in these NETs act as DAMPs to initiate and sustain the inflammatory response [35]. NETs also release proinflammatory mediators through the production of antibodies to citrullinated proteins, which promote synovial inflammation and joint damage [36]. NETs containing citrullinated peptides are internalized by synovial fibroblast-like synoviocytes, promoting their inflammatory phenotype and the upregulation of major histocompatibility complex (MHC) class II. Once internalized, the NET peptide that causes arthritis is loaded into fibroblast-like synoviocytes MHC class II and is presented to antigen-specific T cells, leading to pathogenic autoimmune and cartilage damage [37].

NET deposition has also been found in anti-neutrophil cytoplasmic autoantibody (ANCA)-associated vasculitis; these patients produce antibodies against NET components (e.g., MPO and proteinase 3). NET deposition in inflamed kidneys and circulating MPO–DNA complexes indicates that, in small-vessel vasculitis patients, NET formation triggers vasculitis and promotes autoimmune responses against neutrophil components [38]. NETs thus play a crucial role in the pathogenesis of various autoimmune diseases by exposing autoantigens, perpetuating chronic inflammation, and contributing to tissue damage.

Tumors

NETs have a facilitating and inhibitory role in cancers [39, 40]. Some studies have shown that the components within NETs released by activated neutrophils, such as histones, NE, and ROS, may exert direct cytotoxic effects in the tumor microenvironment (TME), serving as potential anti-tumor tools [39]. However, most research indicates that NETs are associated with tumor growth, metastasis, and TME regulation processes. With respect to tumor growth, NETs form in response to stimuli from tumor cells and the TME. Neutrophil activation and NET formation can be induced by tumor-derived factors, such as granulocyte colony-stimulating factor (G-CSF), interleukin-8 (IL-8), and other chemokines [41, 42]. With respect to tumor metastasis, NETs can release various proteases such as NE and matrix metalloproteinases (MMPs) that degrade the extracellular matrix and promote tumor invasion [43] and the epithelial–mesenchymal transition (EMT) of tumor cells to increase their motility and invasiveness [44]. NETs provide a scaffold that supports the adherence and migration of circulating tumor cells (CTCs). They also capture CTCs, shielding them from immunosurveillance and mechanical stress in the bloodstream, which promotes their survival and dissemination to distant organs. In turn, CTCs stimulate neutrophils to form metastasis-supporting NETs, creating a mutually reinforcing cycle that drives metastatic progression [45, 46]. NETs also help establish pre-metastatic niches by promoting angiogenesis and increasing vascular permeability, thereby supporting metastatic cells [47]. NETs' web-like structure can act as a physical barrier that prevents immune cells from targeting tumor cells, aiding the metastatic process. NETs can also shape the TME to promote tumor growth and progression. NETs' inflammatory signals recruit other immune cells, creating a pro-tumor inflammatory environment. This regulation of the microenvironment is critical for primary tumor growth and metastatic colonization [48].

NETs have a negative impact on tumor treatment efficacy. Recent studies have shown that certain chemotherapies can induce NET formation, which may play a dual role. On the one hand, NETs can enhance the anti-tumor effect of chemotherapy by releasing cytotoxic enzymes such as cathepsin G to trap and kill tumor cells by inducing apoptosis [49]. However, they can also protect tumor cells from antitumor drugs, which can lead to therapeutic resistance [50]. Furthermore, NETs contribute to tumor radioresistance. In a mouse model of bladder cancer, NET deposition was observed in the TME of mice treated with radiotherapy, and NET inhibition improved the overall radiation response [51]. NETs have also been reported to trap chemotherapeutic drugs, such as doxorubicin (DOX), potentially reducing their efficacy in spreading and inducing apoptosis in ovarian cancer cells [52]. Martins-Cardoso et al. described how the TF/PAR2 signaling axis works by enhancing the expression of tissue factor (TF) and promoting pro-tumorigenic cytokines and the EMT-related factors that support NETs' pro-tumorigenic effects in breast cancer [53]. Research has shown that NETs activate the NF-κB/NLRP3 pathway by down-regulating lncRNA MIR503HG expression, which promotes EMT and metastasis in lung cancer [54]. In addition, Zhou et al. found that, in breast cancer, tumor cell-released autophagosomes induced PD-L1-decorated NETs, inhibiting T-cell function and promoting pulmonary metastasis [55]. These studies suggest that although NETs can enhance certain aspects of chemotherapy, they can also act as barriers to drug delivery. Therefore,

strategies that inhibit NET formation or promote NET degradation may attenuate their pro-tumorigenic effects.

Thrombosis

NETs have emerged as an important causative factor in thrombosis, which is characterized by the formation of blood clots in blood vessels that can lead to serious complications such as deep vein thrombosis (DVT), pulmonary embolism (PE), and stroke. NETs contribute to thrombosis through a variety of mechanisms involving complex interactions with platelets, coagulation factors, and the vascular endothelium [56]. NETs' web-like structure is a scaffold for platelet adhesion and aggregation as well as the accumulation of coagulation factors such as fibrinogen and von Willebrand factor (vWF) [57], thereby enhancing thrombus stability and growth, which can promote thrombosis [58]. With respect to platelet activation, histones and other NET components can directly activate platelets, leading to aggregation, which in turn amplifies thrombin generation. This process is mediated through the interaction between NET-related histones and platelet TLRs (TLR2 and TLR4), which enhances platelet adhesion and activation [59]. With respect to coagulation, histones and NE in NETs further contribute to thrombosis, as they have procoagulant properties and degrade anticoagulant proteins [60]. In addition, when TF is exposed to NETs, the extrinsic coagulation pathway is initiated, whereas factor XII (FXII) binding to NETs activates the intrinsic coagulation pathway. This dual activation enhances fibrin clot formation and stability, contributing to thrombosis [61].

Clinical studies have shown that patients with venous thrombus embolism (VTE) have elevated levels of NET biomarkers such as plasma DNA, citrullinated histone H3 (CitH3), and MPO-DNA complex [62]. In addition, NETs can activate the intrinsic coagulation pathway by providing negatively charged DNA, which activates FXII, in turn promoting thrombin generation and fibrin formation [63]. NETs' promotion of the recruitment and activation of other immune cells exacerbates the inflammatory environment, thereby mediating DVT [64]. NETs also play a critical role in arterial thrombosis, having been implicated in the formation of arterial thrombi such as myocardial infarction and stroke [65]. Research has reported that NETs interact with platelets and endothelial cells to promote atherothrombosis and plaque destabilization. Additionally, NET components, particularly proteases such as NE, can degrade the structural proteins of the blood–brain barrier (BBB), which accounts for the injury of surrounding neurons and subsequent neurological disorders [66]. This degradation enables more immune cells and harmful substances to enter the brain tissue, worsening cerebral edema and further damaging neurons. Complement C5a has been reported to induce NET formation by inhibiting mitochondrial STAT3, thereby promoting arterial thrombosis [67]. This highlights a new pathway through which NETs promote atherothrombosis. In conclusion, NETs are key players in the pathogenesis of venous and arterial thrombosis through their interactions with the coagulation system and immune cells.

Chronic sterile inflammatory diseases

Many diseases are characterized by chronic sterile inflammation, and NETs play a key role in their pathogenesis. Neutrophils release NETs in response to multiple triggers, such as cytokines, DAMPs, and environmental stressors, which can promote chronic inflammation and tissue damage [68]. Sustained stimuli often result in persistent NET formation. NETs release nuclear chromatin, ROS, and various biologically active proteins into the extracellular matrix, creating a pro-inflammatory environment that promotes and exacerbates chronic inflammation and tissue damage, particularly in sterile inflammatory disorders [69]. TLRs, particularly TLR4 and TLR9, play an important role in NET formation by recognizing DAMPs released during sterile inflammation. These receptors activate downstream signaling pathways that enhance ROS production and NET release. For example, TLR4 has been associated with NET formation in conditions such as atherosclerosis and ischemia/reperfusion injury [70, 71].

In lung diseases such as chronic obstructive pulmonary disease (COPD) and cystic fibrosis, NETs can obstruct the airways and stimulate persistent inflammation [72]. Pulmonary fibrosis is characterized by excessive tissue remodeling and fibrosis in the lungs; NETs are involved in its pathogenesis by inducing fibroblast activation and differentiation into myofibroblasts [73]. ALI or acute respiratory distress syndrome (ARDS) are severe conditions characterized by the rapid onset of widespread inflammation in the lungs. Abnormal NET formation is highly correlated with the lung mucus-plugging microenvironment and enhances the overactivation of lung macrophages [74]. Asthma is a chronic airway inflammatory disease characterized by wheezing, breathlessness, and coughing that leads to airway remodeling, with thickened walls and increased mucus production [75]. Neutrophil-derived enzymes, ROS, and NET components (DNA, histones, proteases) exacerbate inflammation, contribute to airway remodeling, and increase mucus viscosity, causing obstruction [76].

NETs also contribute to atherosclerosis and abdominal aortic aneurysm (AAA). In atherosclerosis, NET formation is induced by cholesterol crystals and initiates the transcription of genes encoding IL-6 and IL-1β

precursors in macrophages via TLR2 and TLR4, promoting myeloid cell recruitment to lesions [77]. NETs also trap cholesterol crystals and promote cholesterol deposition within the arterial wall, leading to plaque destabilization. These interactions accelerate plaque growth, increasing the risk of plaque rupture and leading to cardiovascular events such as myocardial infarction and stroke [78]. In AAA, NET components degrade the aortic wall and extracellular matrix and weaken the integrity of the vessel wall, advancing the formation and progression of the disease.

NETs also play a role in the progression of inflammatory bowel disease (IBD), such as Crohn's disease and ulcerative colitis. During the acute phase of IBD, neutrophils migrate to inflammation sites and release proteases, ROS, and pro-inflammatory cytokines, contributing to tissue damage. Excessive neutrophil infiltration can disrupt the mucosal barrier and increase intestinal permeability. NETs can exacerbate inflammation by exposing surrounding tissues to their pro-inflammatory components, damaging epithelial cells and aggravating ulceration and barrier dysfunction [79].

The normal wound-healing process involves an inflammatory response that promotes wound cleansing and tissue repair [80]. However, in diabetic patients, chronic hyperglycemia induces excessive neutrophil recruitment and NET release [81], leading to a pro-inflammatory state that sustains inflammation and causes tissue damage, delaying wound healing [82]. Overall, NETs contribute to chronic inflammatory diseases by perpetuating inflammation, promoting tissue injury, and facilitating disease progression.

In summary, NETs play diverse roles in the pathogenesis of various diseases. While essential for immune defense against infections by capturing and neutralizing pathogens, excessive or dysregulated NET formation contributes to tissue damage and exacerbates disease processes in autoimmune disorders, tumors, thrombosis, and chronic sterile inflammatory diseases. These findings highlight NETs' dual nature in acting as both protective immune structures and pathological mediators, making them important therapeutic targets in a wide range of clinical conditions.

Strategies for optimizing NPs to modulate NETs

NETs are important therapeutic targets for a variety of diseases, but the agents that are used to promote or inhibit NET formation often face challenges of non-specificity and rapid degradation in the complex environment of the human body [83]. NP-based drug delivery systems offer promising solutions to these challenges. Nanotechnology represents an innovative approach with unique characteristics and advantages, including enhanced stability, controlled drug release, and the ability to improve bioavailability. By optimizing and functionalizing NPs, it is possible to achieve targeted NET interaction, enabling more effective treatment of NET-related diseases [84].

Tuning the physicochemical properties of NPs

The interaction between NPs and NETs is complex and influenced by NPs' physicochemical properties. Manipulating these factors is essential for leveraging NPs to modulate NETs therapeutically. Key properties influencing NP–NET interactions include NP size, shape, surface charge, composition, and surface chemistry [85, 86].

Size

The size of NPs plays a critical role in their interactions with NETs. Smaller NPs can penetrate deeper into the dense DNA fiber network, then diffuse into the NET matrix and interact with the internal components. Conversely, larger NPs may be trapped at the periphery and unable to interact directly with NETs [87]. It has been reported that smaller polyphosphate NPs are more efficient in stabilizing fibrin through interactions with NET histones compared to their larger counterparts [88]. Silver NPs (AgNPs) with a size of 5 nm were reported to induce NET formation in neutrophils dependent on ROS, PAD, and NE, while the 100 nm AgNPs did not induce NETs at similar concentrations. AgNPs (5 nm) generated ROS, triggering NET formation by activating histone citrullination through PAD4 and histone cleavage through NE [89]. Furthermore, small NPs such as 10 nm nanodiamonds and 40 nm polystyrene beads could stimulate a NET-like structure, whereas larger NPs did not induce NETs [85]. In addition, LPS significantly enhanced the uptake of 10 nm gold NPs (AuNPs) by neutrophils compared to that of 40 nm and 100 nm AuNPs. The combination of AuNPs and LPS synergistically upregulated ROS modulator 1 via ERK activation, increasing mitochondrial ROS production and promoting NET release [90]. Based on the reported research, NPs less than 40 nm in size tend to penetrate the porous structure of NETs more effectively and interact with neutrophils and NET components like DNA and histones. However, NPs larger than 100 nm have limited diffusion into NETs but can form stable complexes with them for localized drug delivery.

Shape

NPs' shapes affect their surface area-to-volume ratio and hydrodynamic behavior, which in turn affect their mode of interaction with NETs [91]. For example, rod-shaped NPs are more likely to become entrapped in NETs' fibrous structures because of their higher surface

area and alignment with NET fibers, while spherical NPs may interact more uniformly [92]. This shape-dependent interaction enhances the trapping efficiency of rod-shaped NPs in NETs, potentially increasing their clearance by the immune system. Recent studies have highlighted that irregularly shaped or star-shaped NPs exhibit enhanced membrane permeability compared with spherical NPs. This is likely due to their sharp edges and increased contact points with cell membranes, which might create localized damage that triggers NETosis and affect their efficacy at NET modulation [86, 93]. For example, different shapes, such as needle-like crystals (e.g., calcium oxalate, monosodium urate) and irregularly shaped particles, enhance the formation of NET-like structures caused by neutrophil necroptosis compared to more uniform particles [94]. In contrast, spherical or amorphous particles are less effective at initiating NET formation because of their reduced ability to disrupt membranes and generate the mechanical stress required for NET induction [86].

Surface charge

The surface charge of NPs also significantly influences their interaction with NETs—which consist of negatively charged DNA strands and positively charged histones—resulting in a heterogeneous charge distribution [95, 96]. Due to electrostatic interactions, negatively charged DNA in NETs exhibits a higher affinity for positively charged NPs, enhancing their binding and interaction. For example, cationic solid lipid NPs induced significantly more NET production than neutral solid lipid NPs [97]. Conversely, negatively charged NPs may be repelled, reducing their interaction with NETs. Cationic liposomes and polymer-coated NPs exhibit strong binding to NETs, facilitating drug delivery to sites of infection or inflammation [98]. Another study compared the ability of phosphatidylcholine cationic liposomes containing stearylamine (SA liposomes) and phosphatidylcholine liposomes (PC liposomes) to form NETs. PC liposomes had -1.74 ± 0.31 mV in zeta potential and did not cause NET formation, while SA liposomes had 11.40 ± 0.44 mV but did induce NET formation due to the positively charged stearylamine-enhanced interaction with the negatively charged neutrophil membrane [99].

Composition

NP composition not only affects their biodistribution and cellular uptake but also affects innate immune responses, including neutrophil activation. Their surface composition significantly affects their interactions with cell membranes and surface receptors [100]. Some NPs with good biocompatibility and degradability, such as PLGA NPs, may reduce inflammation, neutrophil adhesion and migration, and ROS production, which could be relevant in the context of immunomodulation [101]. In contrast, some highly reactive NPs may promote neutrophil activation and NET formation. For instance, AgNPs are highly reactive and can induce ROS production in neutrophils [89]. Some metal oxide NPs can directly activate neutrophils and enhance NETosis through oxidative stress [102].

Surface chemistry

NPs' surface chemistry critically influences their interaction with the biological environment, subsequently affecting their interaction with NETs. NP surfaces are often modified to reduce the recognition and uptake of NPs by the immune system. The modification of NPs with Polyethylene Glycol (PEG), known as PEGylation, can form a steric barrier around them, preventing opsonization by serum proteins and phagocytosis by immune cells [103]. Studies reveal that PEGylated NPs tend to induce lower levels of neutrophil activation compared to their non-PEGylated counterparts [104]. In contrast, non-PEGylated silver NPs caused a dose-dependent release of NETs from neutrophils [105].

Upon their introduction into a biological system, NPs rapidly adsorb proteins and biomolecules, forming a "protein corona" that alters their physicochemical properties (e.g., size, surface charge, hydrophobicity) [106]. This corona can shield the NP surface, which potentially reduces direct interactions with NET components, thereby modifying binding affinity and uptake by NETs [107, 108]. For example, agglomerations of non-stabilized superparamagnetic iron oxide NPs (SPIONs) induced NET formation by isolated human neutrophils. In contrast, agglomeration and NET formation were reduced by the stabilization of SPIONs with biocompatible layers of either human serum albumin or dextran [109]. The protein corona therefore impacts NPs' effectiveness in targeting or modulating NETs.

To summarize, NPs' physicochemical properties influence their interactions with the biological environment, as well as their recognition and uptake by immune cells, including neutrophils; this is crucial for subsequent interactions with NETs. Designing NPs that can effectively interact with NETs for therapeutic applications—such as targeted drug delivery, inflammation control, and disease treatment—thus requires the careful consideration of these properties to enhance their efficacy and specificity.

Enhancing NP targeting specificity toward NETs

Enhancing the targeting specificity of NPs toward NETs can also promote their interactions. NPs designed and engineered to specifically target NETs can enhance therapeutic efficacy while minimizing adverse effects

[84]. Several strategies have been developed to this end, such as modifying the surface of NPs to enhance their targeting specificity toward NETs and designing pH- or ROS-responsive NPs that respond to the NET microenvironment. These approaches leverage the unique properties of NPs to target NETs.

Surface modification for targeted drug delivery

Surface modification is a key strategy for enhancing NP targeting and functionality. Modifying the surface of NPs with specific ligands, antibodies, or peptides enables them to selectively bind to NET components or neutrophil surfaces. This facilitates targeted drug delivery and increases local drug concentrations while reducing systemic side effects compared to those induced by standard systemic administration. Some neutrophil-expressed receptors, such as integrin, L-selectin, P-selectin glycoprotein ligand-1 (PSGL-1), mannose receptor, and CD11b, have been studied as targets. For example, a click reaction between the maleimide group on 1,2-Distearoyl-sn-glycero-3-phosphoethanolamine-polyethylene glycol 2000-maleimide (DSPE-PEG_{2000}-Mal) and the sulfhydryl group on cyclo (Arg-Gly-Asp-d-Phe-Cys) (cRGDfC) was used to conjugate cyclic arginine-glycine-aspartate (cRGD) peptides targeting neutrophil integrin receptors to the surface of NPs [110]. In other research, targeted delivery to neutrophils was facilitated by the electrostatic interaction between the carboxylate groups of polysialic acid and the lysine residues of L-selectin [111], which improved the delivery of therapeutic agents to NET formation sites [110]. In addition, liposomal nanocarriers modified with ROS-responsive polymers and a fibrin-binding peptide (Cys-Arg-Glu-Lys-Ala) were negatively charged, enabling their binding to positively charged histones within NETs. Additionally, these NPs delivering PAD4 inhibitor Cl-amidine suppressed NETosis, thereby neutralizing their activity and preventing further tissue damage [112].

pH- or ROS-responsive NPs

Designing NPs that respond to specific NET microenvironments, such as low pH or high ROS levels, can improve targeting precision. pH-sensitive NPs release their payload in acidic environments, which are typically associated with inflammation and infection sites where NETs are prevalent [113]. These NPs are commonly designed with acid-labile linkers (e.g., imines, acetals, and orthoesters) or pH-sensitive polymers [e.g., poly(β-amino esters)] that undergo structural breakdown or swelling under low pH conditions, leading to rapid drug release [114]. Ionizable groups like amines and carboxylic acids are protonated or deprotonated in response to pH changes, leading to alterations in solubility or charge [115]. For example, hydrazone linkers cleave in acidic environments, enabling the precise release of encapsulated drugs at target sites [116].

Similarly, ROS-sensitive NPs are designed with functional groups such as thioketal bonds, phenylboronic acid/ester, and selenium-containing linkages. These NPs respond to elevated ROS levels in NET-rich environments characterized by high oxidative stress by undergoing oxidation-induced bond cleavage or hydrophobic-to-hydrophilic transitions, causing NP disassembly and controlled drug release [117, 118]. For instance, a polyprodrug NP designed with thioketal linkers in its polymer backbone enabled ROS-responsive chain-breakage degradation and controlled drug release in tumor tissues [119]. In brief, surface modification and stimuli-responsive design of NPs enable precise drug delivery to NETs. These strategies synergistically improve therapeutic efficacy while minimizing off-target effects in NET-associated conditions.

Optimizing drug delivery strategies of NPs

To enhance therapeutic efficacy and precision in NET modulation, NPs are engineered to optimize drug delivery through strategies such as encapsulating therapeutics targeting NET components and establishing multi-drug nanoplatforms. These strategies enable comprehensive NET modulation while addressing complex pathological processes.

Encapsulation of therapeutics targeting NET components

NETs comprise DNA, histones, and various proteases. NPs can be used to deliver agents that degrade or neutralize these components, mitigating the harmful effects of excessive NET formation. Extracellular DNA can be degraded by DNase I-encapsulated NPs to dismantle the NET structure and inhibit its formation, thereby reducing its pro-inflammatory and pro-thrombotic effects [120]. Histones, exhibiting cytotoxic and pro-inflammatory effects, can be reduced using NPs encapsulating PAD4 inhibitors (e.g., Cl-amidine, GSK484), which are involved in catalyzing histone citrullination and inducing chromatin decondensation [121]. Furthermore, NE and MPO within the NETs can be degraded by protease inhibitors delivered by NPs such as sivelestat or luminol, thereby reducing tissue damage and inflammatory responses [110]. The NE inhibitor sivelestat blocks NE by competitively binding to its active site, preventing substrate hydrolysis [122], while luminol inhibits MPO by interacting with the heme group in the MPO active site and blocking its catalytic activity [123]. These nanoplatforms provide a comprehensive strategy for inhibiting NETs and hold promise for treating related diseases.

Multi-drug nanoplatforms

Loading multiple drugs with distinct therapeutic mechanisms onto a single nanoplatform enables combinatory treatments with synergistic effects. This strategy is highly suited for targeting NETs because of their complex role in different diseases. Multi-drug NPs can simultaneously deliver DNA-degrading enzymes, anti-inflammatory agents, and antioxidants to inhibit NET formation, promote degradation, and reduce inflammation. For instance, a nanoplatform was designed to deliver DNase I and methylprednisolone sodium succinate (MPS) to degrade the DNA scaffold of NETs and inhibit neutrophil cytokine release, respectively. A calcium acetate gradient method was used to encapsulate MPS into the liposomes. The liposomes were prepared with a defined lipid composition, hydrated with calcium acetate, and loaded with MPS using a remote loading technique. DNase I was conjugated to the NPs through MMP-9 cleavable peptide linkers using maleimide-thiol chemistry. The peptide linkers were functionalized with maleimide groups, and DNase I was modified to expose thiol groups. This enabled the formation of a stable covalent bond between the thiol groups on DNase I and the maleimide groups on the peptide linkers [124]. Another study reported NPs composed of luminol and alendronate for targeting anti-inflammation and calcification. The luminol was covalently conjugated to α-cyclodextrin to form the core component in the NPs, and alendronate was attached to DSPE-PEG-Mal via an amine group and then incorporated into the NP system during the nanoprecipitation process [123]. These multi-therapeutic NPs offer effective NET regulation in disease environments, though careful design is needed to ensure drug stability and efficacy.

Overall, NP-based drug delivery systems hold great potential for treating a variety of NET-related diseases. In order to achieve molecular specificity in targeting NET components, the design and manipulation of NPs incorporates several strategies. First, surface functionalization with specific ligands or peptides enables selective interaction with target components. Second, stimuli-responsive mechanisms, such as ROS- or pH-sensitive polymers, facilitate controlled drug release in inflammatory environments, enhancing specificity by limiting off-target effects. Third, encapsulated agents can degrade or neutralize NET components, including extracellular DNA, histones, NE, and MPO, effectively mitigating inflammation caused by excessive NET formation. However, because the complex composition of NETs can lead to overlapping interactions, achieving absolute molecular specificity remains challenging. Future improvements, such as multi-functionalized NPs capable of simultaneously targeting DNA, histones, and proteases with distinct mechanisms, could potentially address these limitations and enhance therapeutic precision.

Applications of NP-based NET modulation strategies

NP-based drug delivery systems represent innovative approaches to treating NET-related diseases, addressing the complex role of NETs in various conditions. The published literature includes two main strategies for modulating NETs using NPs, including promoting NET formation to boost the immune response and inhibiting NET formation to mitigate tissue damage and inflammation. Each strategy demonstrates the versatility and potential of NP-based therapies in treating NET-related disorders.

Promoting NET formation

NPs have the significant potential to enhance immune responses by promoting NET formation. Strategies to leverage NPs for this purpose include utilizing their intrinsic antimicrobial physicochemical properties, directly integrating NPs with neutrophils to exploit their natural migratory abilities, and modifying NP surfaces to enhance recognition and uptake by neutrophils. Together, these approaches aim to boost NET-mediated defense mechanisms while enabling the precise delivery of therapeutic agents to sites of infection, inflammation, or tumors (Table 1).

Utilizing NPs with antimicrobial physicochemical properties

Neutrophils are a major player in the anti-infection process. Their antimicrobial function can be achieved through direct phagocytosis and NET formation, which can capture and kill bacteria extracellularly, promoting tissue remodeling [131]. NPs with carefully designed physicochemical properties, such as the ability to generate ROS or induce immune cell activation, have emerged as promising tools for combating infections. Such NPs can enhance antimicrobial defense by inducing the formation of NETs, facilitating pathogen clearance, and creating a localized microenvironment hostile to pathogens. Biocompatible metal biomaterials are widely used in the repair of bone defects, but these implantations carry the risk of infection, often leading to delayed bone healing, implant failure, chronic pain, and even life-threatening sepsis. The development of new medical metal materials with anti-infective properties can, therefore, effectively reduce or eliminate the risk of infection [132]. A zinc-doped ferric oxyhydroxide nano-layer (PEO-FeZn) was investigated for its ability to enhance the bactericidal activity and

Table 1 Summary of NP-based drug delivery systems for promoting NET formation in various diseases

Strategies to target NETs	Diseases	NP type	NPs	Payload	Treatment mechanism	Refs.
Utilizing NPs with antimicrobial physicochemical properties	Implant-associated infections	Zinc-doped ferric oxyhydroxide	PEO-FeZn	NA	Zn ions induced ROS production and NET formation in neutrophils	[125]
	Bacterial sepsis	Carbonized polymer dots	CPDs	Curcumin	Induced NET formation and enhanced the entrapment and elimination of bacteria	[126]
Directly integrating NPs with neutrophils	Tumor	BSA	BSA-Ce6 NPs	Ce6	Generated ROS, increased NETosis, enhanced cytolytic activity of neutrophils against tumor cells	[127]
	Thrombosis	Silver	UM-NEs (Ag-UK)	Urokinase	Targeted thrombi and induced NET formation	[128]
Surface modification of NPs for neutrophil recognition and uptake	Tumor	Hybrid (mycoplasma membrane/ liposomes)	MM-LPs	Podophyllotoxin, resiquimod	Induced NET formation, suppressed tumor growth and lung metastasis of breast tumor	[129]
		Hybrid (polymer /bacterial membrane vesicles)	NPNs	Cisplatin	Hitchhiked neutrophils, delivered to tumors sites, triggered NET formation, and facilitated drug release of NPNs	[130]

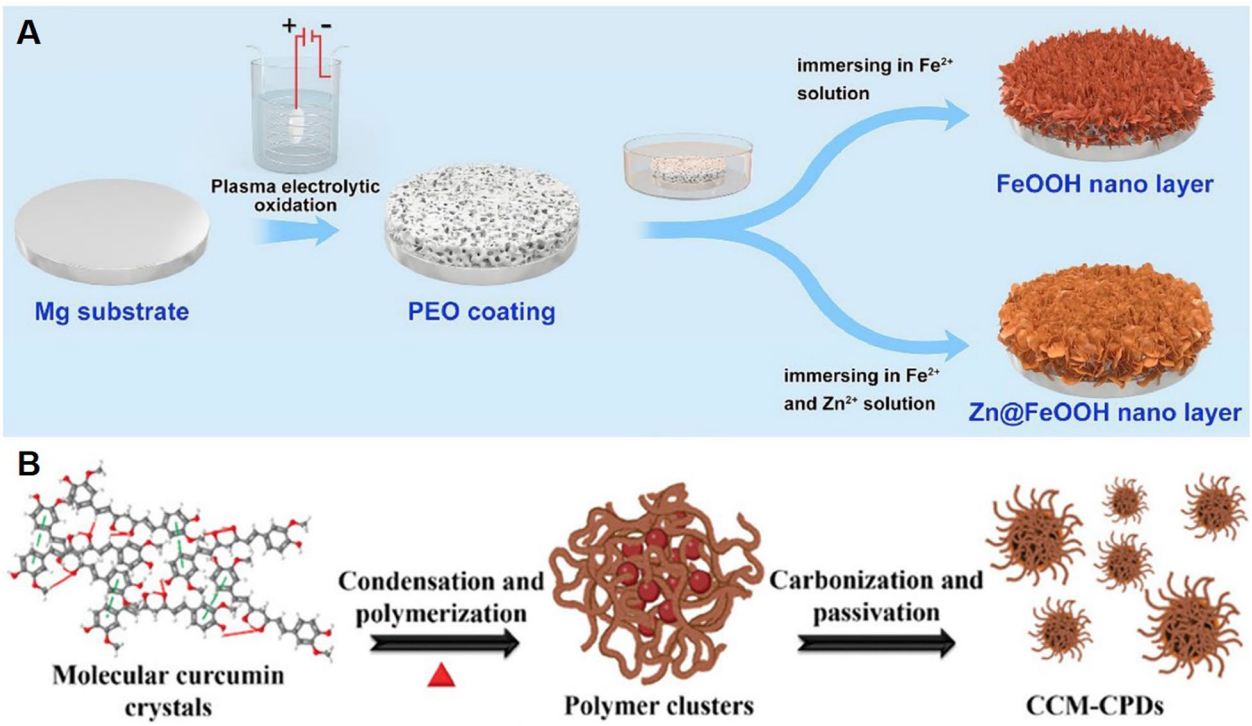


Fig. 2 Applications of NP-based drug delivery systems in promoting NET formation by utilizing NPs with antimicrobial physicochemical properties. **A** Schematic illustration for the preparation of Zn-doped FeOOH nano-layer on PEO-coated Mg alloy [125]. Copyright © 2022 Elsevier. **B** Schematic representation of the preparation and formation mechanism of CCM-CPDs via mild pyrolysis at 180 °C [126]. Copyright © 2024 Wiley-VCH GmbH

osseointegration of a magnesium alloy. These NPs were synthesized using a combination of plasma electrolytic oxidation (PEO) and immersion treatments to form a Zn-doped ferric oxyhydroxide nano-layer on a magnesium alloy (Fig. 2A). Compared to PEO and PEO-Fe, PEO-FeZn samples had a higher level of NET formation and induced higher levels of ROS in neutrophils, primarily driven by the Zn ions [125].

Sepsis is characterized by a dysregulated immune response to infection, leading to systemic inflammation, coagulation abnormalities, tissue damage, multi-organ failure, and even death [133]. Excessive neutrophil activation contributes to systemic inflammation and tissue damage through the release of ROS, proteases, and pro-inflammatory cytokines [134]. While NETs trap and kill pathogens, their components can exacerbate inflammation and tissue damage when overproduced [135]. It is therefore essential for effective sepsis treatment to develop multimodal therapies with both anti-infective and anti-inflammatory effects. For example, researchers designed carbonized polymer dots (CPDs) derived from curcumin to treat bacterial sepsis (Fig. 2B). Prepared via mild pyrolysis, these CPDs induced NET formation, enhancing bacterial entrapment and elimination. They also exhibited anti-inflammatory properties due to the phenolic hydroxyl and carbonyl groups from curcumin precursors, enhancing their therapeutic potential. In addition, they enhanced neutrophil ROS production and activated protein kinase C (PKC) and NADPH oxidase [126]. These studies demonstrate that NPs represent an innovative approach to reducing infection-related complications while maintaining immune homeostasis.

Directly integrating NPs with neutrophils

Neutrophil chemotaxis is a key immune mechanism that directs neutrophils to sites of inflammation, infection, or tumors. This natural migratory ability can be leveraged for targeted therapeutic delivery via NETs [136, 137]. Once loaded directly into or onto neutrophils, NPs can hitchhike on them and be transported to the disease site. Once neutrophils arrive at the target sites and induce NET formation, the NPs are released, delivering drugs precisely where they are needed.

This strategy was investigated in a study that reported a photosensitizer chlorin e6 (Ce6), which was nanopackaged with bovine serum albumin (BSA) to create biocompatible BSA-Ce6 NPs for integration with neutrophils. Reinfusion of the engineered neutrophils led to greater Ce6 accumulation in tumors relative to Ce6 nanoformulation. Upon near-infrared illumination, Ce6 generated ROS, inducing NET formation and enhancing neutrophil cytotoxicity against 4T1 tumor cells [127]. Another study developed enzyme-catalyzed biomotor-engineered neutrophils for targeted drug delivery and thrombolytic therapy, conjugating AgNPs with urokinase to form Ag-UK. The urease was immobilized asymmetrically onto the neutrophils' surface

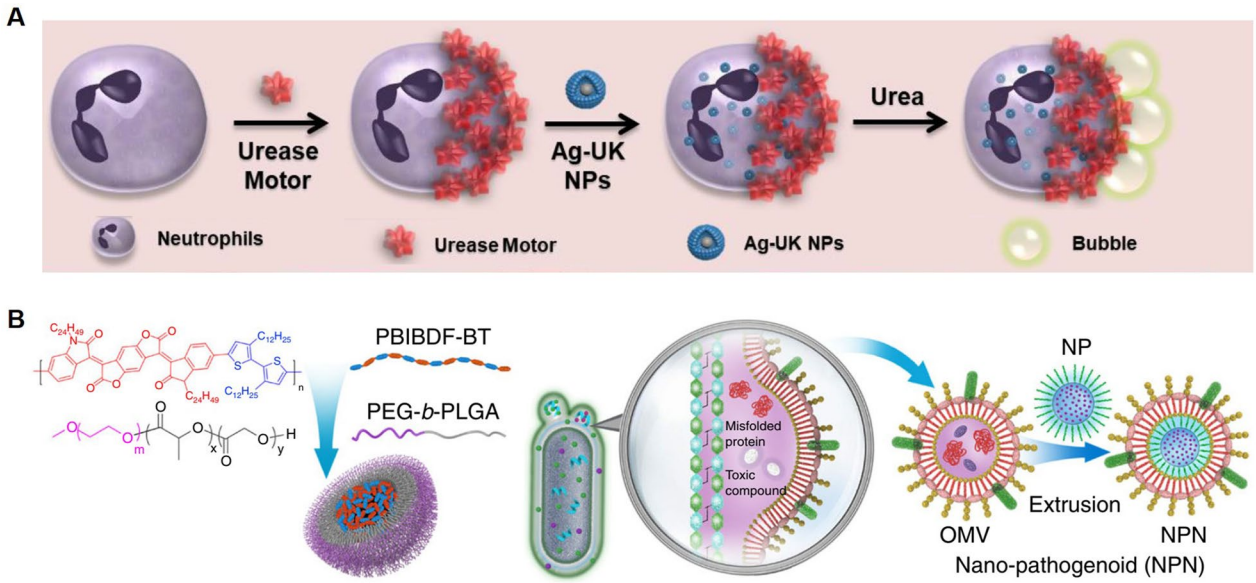


Fig. 3 Applications of NP-based drug delivery systems in promoting NET formation by directly integrating NPs with neutrophils (**A**) and modifying NPs for neutrophil recognition and uptake (**B**). **A** The synthesis route of the UM-NEs (Ag-UK) system [128]. Copyright © 2022, American Chemical Society. **B** Preparation of PEG-b-PLGA NPs encapsulating PBIBDF-BT (PBT) as a photothermal transducer (left). Preparation of NPNs by coating OMVs on NPs, which inherit PAMPs from the OMVs (right) [130]. Copyright © The Authors 2020

before loading Ag-UK (Fig. 3A). This system targeted thrombi and induced NET formation; neutrophils released Ag-UK to degrade the thrombus via urokinase, restoring vascular recanalization [128]. This approach enables precise delivery and targeting accuracy, significantly improving the local concentration and efficacy of therapies.

Surface modification of NPs for neutrophil recognition and uptake

An alternative strategy focuses on modifying NP surfaces to make them recognizable and easily phagocytosed by neutrophils. By functionalizing NPs with specific ligands, peptides, or antibodies that interact with neutrophil surface receptors, these modified NPs can selectively bind to neutrophils. Once recognized, neutrophils engulf the NPs, internalize the therapeutic agents, and deliver them to inflamed sites. On reaching their target location and forming NETs, neutrophils can release the NPs, delivering the therapeutic payload directly into the inflamed or diseased tissue [138].

Surface modifications of pathogen membranes facilitate neutrophils' recognition and engulfment. For example, a mycoplasma membrane (MM) fused with liposomes was designed to employ circulating neutrophils to transport liposome NPs, having the advantage of inflammatory cytokine-guided autonomous tumor localization. R848, a TLR7/8 agonist, was also encapsulated into NPs to activate neutrophils and increase their infiltration of tumors. NPs loaded with pycnogenol were simultaneously released from neutrophils and phagocytosed by tumor cells, suppressing tumor growth and lung metastasis in the 4T1 breast tumor model [129]. Li et al. developed chemotaxis-driven nano-pathogenoids (NPNs) to enhance the efficacy of tumor treatment post-phototherapy. The NPNs mimicked bacterial outer membrane vesicles to hitchhike on circulating neutrophils, being delivered to tumors following photothermal therapy (Fig. 3B). Upon reaching the tumor site, the inflammatory conditions triggered NET formation, thereby facilitating the release of NPNs from neutrophils, allowing the encapsulated drugs to be taken up by tumor cells [130].

Neutrophil chemotaxis and NET formation are leveraged to enhance drug delivery precision and therapeutic efficacy by utilizing NPs with antimicrobial physicochemical properties and strategies such as direct integration and surface modification. These approaches enable targeted delivery to disease sites, including infections, tumors, and inflamed tissues, while minimizing off-target effects and maximizing treatment efficiency [139]. Carefully balancing neutrophils' antimicrobial or anti-inflammatory effects with the tissue damage caused by their overactivation is crucial when designing NP-based therapeutics, as NETs' pathological and physiological roles vary across diseases [140]. In addition, designing NPs to specifically target neutrophils and induce localized NET formation at tumor sites is essential for enhanced therapeutic potential. This approach minimizes the risk of nonspecific pro-tumor effects while maximizing the anti-tumor efficacy.

Inhibiting NET formation

NETs are crucial for the immune response, but excessive or inappropriate NET formation may lead to pathologic changes. Therefore, inhibiting NET formation helps to relieve tissue damage and inflammation under some circumstances [141]. Various strategies that use NPs to inhibit NET formation have been developed to address specific diseases and conditions in which NETs play a detrimental role, such as the degradation of DNA scaffolds, the inhibition of PAD4 or NE activity, the neutralization of NET-associated components or mediators, the suppression of neutrophil recruitment and activation, and ROS scavenging (Table 2).

DNA scaffold degradation

The structural integrity of NETs is primarily maintained by extracellular DNA. Degrading this DNA scaffold effectively disrupts NET formation, reducing associated tissue damage. DNase I, a commonly used enzyme, hydrolyzes extracellular DNA, diminishing NET generation and the associated pro-inflammatory effects [25]. DNase I can be chemically conjugated to NPs via covalent bonding. This is achieved using functional groups on the NP's surface, such as maleimide or carboxyl groups, which react with thiol or carboxylic acid groups on DNase I [120, 124]. Therefore, NP-based delivery systems protect DNase I from rapid degradation in circulation and achieve controlled release at the site of inflammation using microenvironment-responsive mechanisms. For controlled release, NPs can incorporate pH-sensitive, ROS-responsive polymers or enzyme-sensitive linkers, such as MMP-cleavable peptides, to degrade inflamed tissues, enabling site-specific DNase I release and enhancing therapeutic precision [149].

NPs have been designed to encapsulate DNase I for the treatment of various diseases. For lung disease treatment, an inhalable nanoplatform comprising serum exosomes and liposomes was designed to deliver DNase I and MPS to relieve inflammation for ALI treatment. DNase I was modified to expose thiol groups, enabling the covalent bond formation with maleimide groups of DSPE-PEG in liposomes. The degradation of NETs facilitated the targeted delivery of MPS to macrophages and promoted anti-inflammatory M2 phenotype polarization [124].

Table 2 Summary of NP-based drug delivery systems for inhibiting NET formation in various diseases

Target site of NET inhibition	Diseases	NP type	NPs	Payload	Treatment mechanism	Refs.
DNA scaffold degradation	ALI	Hybrid (exosome/liposome)	MPS/D-SEL	DNase I, methylprednisolone sodium succinate	Suppressed neutrophil activation and mucus plugging microenvironment, and amplified M2 macrophage polarization	[124]
		Cellular nanovesicle	DCNV	DNase I	Reduced pro-inflammatory cytokines (TNF-α, IL-1β, and IL-6) and decreased neutrophil infiltration	[142]
		Polymer	DNase-I pMNSs	DNase I	Reduced the levels of neutrophils, NETs, extracellular DNA, reduced the activity of NE and MPO, and mitigated NETosis inflammatory response	[143]
	IBD	Polymer	DNase-NZ	DNase I, dopamine	Attenuated neutrophil infiltration and NETosis	[120]
		Polymer	ALG-SNase	Staphylococcal nuclease	Reduced proinflammatory cytokines (TNF-α, IL-1β, and IL-6), decreased neutrophil infiltration in the colon, and maintained intestinal barrier integrity	[144]
	RA	Polymer	DHY	DNase I, methotrexate	Reduced inflammatory cytokine (TNF-α, IL-6) levels and inhibited macrophages polarization to the pro-inflammatory M1 phenotype	[145]
	Ischemic stroke	Hybrid (Prussian blue/platelet membranes)	D@HPB@SPM NPs	DNase I	Scavenged ROS, relieved oxidative stress injury, and mitigated neutrophil-induced reperfusion injury	[146]
		Polymer	DNase 1@HB	DNase I	Decomposed NETs, reduced histone stability, inhibited platelet activation by blocking the binding of histones to the TLR4, disrupted the cycle of NETs-platelet mutual activation, and reduced platelet aggregation and thrombus formation	[113]

Table 2 (continued)

Target site of NET inhibition	Diseases	NP type	NPs	Payload	Treatment mechanism	Refs.
	Tumor	Polymer	5HT-NP@D/P-TC-RLA	DNase I, 5-HT/camptothecin, RLA peptide	Eliminated NETs, restrained mitochondrial biogenesis induced by NETs, and relieved hypoxic TME by reducing oxygen consumption	[147]
		Hybrid (gold/polymer)	AuPB@mPDA	DNase I	Reduced metastatic seeding and improved immune checkpoint therapy efficacy	[148]
		Organic (poly-L-lysine/paclitaxel prodrug)	mP-NPs-DNase/PTX	Paclitaxel, DNase I	Degraded NETs, reduced tumor growth and metastasis	[149]
		Mesoporous organosilica	DMMnSiO3-PEG/DOX/DNase-1	DNase I, DOX	Inhibited tumor growth using a chemotherapeutic effect and suppressed distant metastasis by disassembling NETs	[150]
		Polymer	PAAP/DNase-1	DNase I	Degraded chromatin within tumor cells, caused apoptosis and cell membrane rupture, prevented NET-induced liver metastasis	[151]
		Hybrid (cell membrane/liposomes)	C-DL	DNase I	Adhered to NETs and degraded NET structures	[152]
		Mesoporous bioactive glass	GODM-gel	DNase I	Reduced physical barriers that hinder NK cells from contacting tumor cells and improved NK cell infiltration	[153]
		Polymer	IPN	DNase I, propranolol	Block CANTs, suppressed angiogenesis, depleted MDSCs, enhanced NK cell function, prevented cytotoxic T-cell exhaustion, and improved the immune response against postsurgical tumor recurrence and metastasis	[154]

Table 2 (continued)

Target site of NET inhibition	Diseases	NP type	NPs	Payload	Treatment mechanism	Refs.
PAD4 inhibition	Ischemic stroke	Liposome	C-Lipo/CA	Cl-amidine, CREKA peptide	Inhibited NET formation, modulated the cGAS-STING pathway, reduced inflammation, improved BBB integrity, and enhanced neuronal survival in mouse model	[112]
		Polymer	T-GSK	GSK484	Prevented CitH3 citrullination and reduced neuroinflammation	[121]
NE inhibition	Atherosclerosis	Liposome	cRGD-SVT-Lipo	Sivelestat, cRGD	Reduced NE activity and inhibited NET formation	[110]
	ALI	Liposome	ICMV-Sive	Sivelestat	Inhibited NET formation, decreased the clinical signs of lung injury, reduced NE and other proinflammatory cytokines in serum	[155]
	RA	BSA	DP/BTST	Sivelestat, dexamethasone palmitate	Prevented NET production and improved RA symptoms	[156]
	Thrombosis	Hybrid (liposome/peptide)	NT-NPs, PNT-NPs	Hydroxychloroquine, α1-antitrypsin-derived peptide	Targeted and modulated activated neutrophils, inhibited NE activity, and reduced NET formation	[157]
Neutralization of NET-associated components or mediators	Tumor	Polymer	cANP	Tertiary amines	Inhibited chemotactic ability of NET-DNA and reduced cancer metastasis	[158]
	Airway inflammatory disorders	Hybrid (black phosphorus/polyglycerol-amine)	BP-PGA50	NA	Reduced the levels of cell-free DNA, NETs, and inflammatory cytokines (IL-1β, IL-6, and TNF-α) in nasal and lung tissues, inhibited TLR9 activation	[159]
	Tumor	Polymer	PMeSEA	NA	Destabilized NETs, inhibited NET formation, and reduced inflammatory cytokine (TNF-α and IL-6) levels	[160]

Table 2 (continued)

Target site of NET inhibition	Diseases	NP type	NPs	Payload	Treatment mechanism	Refs.
Inhibition of neutrophil recruitment and activation	Diabetic wound healing	Polymer	PZ	NA	Upregulated Ccl3 + macrophages, downregulated S100a9 + neutrophils, suppressed NET formation, improved angiogenesis, and accelerated the healing process of diabetic wounds	[161]
	ALI	Hybrid (neutrophil membrane/lipid)	DHA@ANeu-DDAB	Docosahexaenoic acid	Blocked neutrophil adhesion and infiltration through the β2 integrin-mediated interaction with ICAM-1/2 on the endothelium, reduced inflammatory cytokine levels	[138]
	RA	Protein	CBR NPs	Celastrol, RGD	Downregulated NF-κB pathway, reduced CitH3 expression, and induced inflammatory neutrophils apoptosis	[162]
	Thrombosis	Gold	aPSGL-1-AuNPs	Anti-PSGL-1-antibody	Targeted PSGL-1 clusters on the uropods of activated neutrophils, blocked neutrophil attachment to p-selectin on endothelial cells, activated platelets, and reduced NET formation in thrombi	[163]
	Diabetic wound healing	Hybrid (mesoporous silica/Ag)	M@M-Ag-Sil-MA	Metformin	Inhibited bacterial aggregation, shifted M1 phenotype of macrophages to anti-inflammatory M2 phenotype, inhibited NET formation, decreased the release of NE, MPO, and NETs-induced pro-inflammatory factors, and promoted fibroblast migration and endothelial cell angiogenesis in vivo	[164]
	Asthma	α-cyclodextrin	LaCD NP	Luminol	Inhibited NET formation, reduced the recruitment and activation of neutrophils, decreased MPO expression, attenuated the activation of the NLRP3 inflammasome in neutrophils, and decreased the levels of pro-inflammatory cytokines (TNF-α, IL-1β) and oxidative species (H_2O_2)	[165]

Table 2 (continued)

Target site of NET inhibition	Diseases	NP type	NPs	Payload	Treatment mechanism	Refs.
ROS scavenging	Corneal pathological fibrosis	Hybrid (quantum dots/polysaccharide)	BPQDs-OCS@SilMA	NA	Reduced ROS accumulation in neutrophils, prevented NF-κB pathway activation and NET formation, and inhibited NET-mediated M2 macrophage polarization and fibrosis	[117]
	AAA	α-cyclodextrin	LaCD NP	Luminol, alendronate	Reduced the activation and infiltration of neutrophils, decreased NET release, suppressed pro-inflammatory mediators (MPO, TNF-α, CXCL1), and decreased the levels of MMP-2 and MMP-9 in aortas	[123]
	Asthma	β-cyclodextrin	TPCN	Tempol	Inhibited NET formation, reduced pro-inflammatory cytokines (TNF-α, IL-1β, IL-6) levels and oxidative stress, decreased neutrophil infiltration, and regulated regulatory T cells/T helper 17 cells balance	[166]
	Ischemic stroke	Polymer	APTS	A151	Scavenged ROS, decreased MPO, NE, and CitH3 levels, prevented chromatin decondensation, and exposed phosphatidylserine on the neutrophil surface, leading to apoptosis rather than NETosis	[167]
		Liposome	L-Ps	Puerarin	Decreased NETs, extracellular DNA, NE, and MPO activities, and reduced oxidative stress and inflammatory responses	[168]

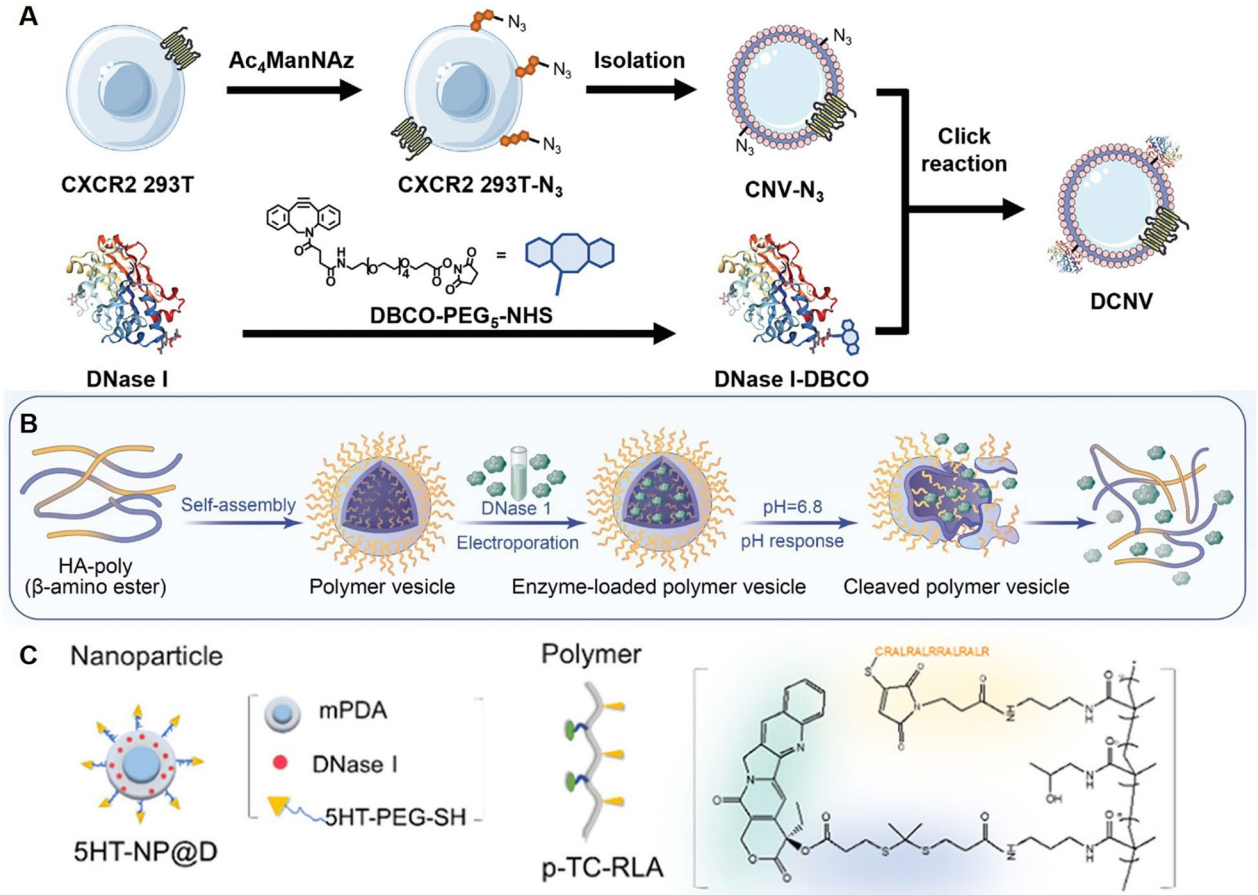


Fig. 4 Applications of NP-based drug delivery systems in inhibiting NET formation by DNA scaffold degradation. **A** Schematic illustration of the preparation of DCNV. CXCR2 293 T cells are cultured with Ac_4ManNAz to prepare azido-functionalized biomimetic nanovesicles (CNV-N_3). Subsequently, DBCO-modified DNase I is immobilized on the surface of CNV-N_3 through click chemistry to obtain DCNV [142]. Copyright © 2023 The Authors. **B** Scheme of the DNase 1@HB preparation process. The nanoplatform was formed by self-assembly of HB polymer, followed by encapsulating DNase 1 into the polymeric vesicles by electroporation. The resulted DNase 1@HB can disintegrate to release the cargo upon acid trigger [113]. Copyright © 2023 Elsevier. **C** Illustration of the construction of nanoparticle (5HT-NP@D) and polymer (P-TC-RLA) [147]. Copyright © 2024, American Chemical Society

Cell membranes overexpressing CXC motif chemokine receptor 2 (CXCR2) and mimicking neutrophil chemotaxis were conjugated with DNase I to form NPs, which degraded NETs, reduced pro-inflammatory cytokines (TNF-α, IL-1β, and IL-6), and decreased neutrophil infiltration in lung tissue (Fig. 4A) [142]. DNase-I-coated polydopamine nanospheres were also developed to reduce neutrophil levels, NETs, extracellular DNA, NE and MPO activity in the blood of septic mice, thereby mitigating the NETosis-associated inflammatory response [143].

NPs loaded with DNase I reduce NET formation and inflammation for the treatment of IBD. A DNase-I nanozyme (DNase-NZ) was fabricated using a PLGA core coated with dopamine and PEGylated polymeric NPs. DNase I was chemically conjugated to NPs via carboxylic acid, which reacted with coupling reagents on the NPs' surfaces. DNase-NZ effectively degraded NETs, reducing their accumulation and alleviating colon inflammation more efficiently than free DNase-I or mesalamine [120]. Dong et al. developed calcium alginate NPs (ALG-SNase) that encapsulated staphylococcal nuclease (SNase) to degrade intestinal NETs. SNase was used to degrade NETs' DNA backbone. ALG-SNase reduced pro-inflammatory cytokines (TNF-α, IL-1β, IL-6), decreased neutrophil infiltration, and preserved intestinal barrier integrity by increasing tight junction proteins (ZO-1, occludin) [144].

NPs have been manipulated to load DNase I to degrade NETs for RA treatment. Wang et al. developed a DNase-functionalized hydrogel (DHY) to reduce the levels of inflammatory cytokines (TNF-α, IL-6) and inhibit the polarization of macrophages to the pro-inflammatory M1 phenotype [145]. For ischemic stroke treatment,

researchers have investigated using NPs to degrade the NETs' DNA scaffold. DNase I-loaded hollow Prussian blue NPs coated with sialic acid-modified platelet membranes were designed to target neutrophils via sialic acid-L-selectin interaction by accumulating in injured brain tissue, degrading NETs, and reducing oxidative stress by scavenging ROS [146]. pH-triggered polymersome loaded with DNase 1 also showed efficacy in disrupting the thrombus skeleton structure and inhibiting platelet activation by blocking histones binding to the TLR4 (Fig. 4B) [113].

NET modulation has clinical significance for the treatment of tumors, as excessive accumulation of NETs has been found in the distant metastases foci of tumor patients [169]. DNase I-loaded NPs used to degrade NETs have been widely studied for the treatment of tumor growth and metastasis. Examples include NET-targeting polymeric NPs decorated with 5-hydroxytryptamine (5-HT) (Fig. 4C) [147], a nanoplatform comprising a plasmonic gold blackbody core and mesoporous polydopamine shell [148], a nanocarrier system consisting of paclitaxel prodrug core and poly-L-lysine-conjugated DNase I shell responsive to matrix metalloproteinase 9 (MMP-9) [149], a manganese-enriched nanosystem co-delivering DOX responsive to a lower pH and higher GSH [150], deformable protein NPs composed of a poly amino acid conjugated with PEG [151], NET-binding protein coiled-coil domain containing 25 (CCDC25) overexpressing cell membrane-derived liposomes [152], mesoporous bioactive glass NPs incorporated into a hydrogel matrix composed of oxidized starch and gelatin [153], and fibrin-alginate hydrogel incorporated with DNase I-loaded PLGA NPs and propranolol [154]. These nanomedicines not only degraded NETs but also induced an anti-tumor immune response.

PAD4 inhibition

PAD4 is a crucial enzyme in the NETosis pathway, catalyzing the conversion of arginine residues on histones into citrulline, a process known as citrullination. By citrullinating histones, PAD4 reduces their positive charge, leading to chromatin decondensation. This loosening of the tightly packed DNA allows it to spread out within the neutrophil, a necessary step for NET formation [170]. Therefore, inhibiting PAD4 activity can prevent NET formation. Small molecule PAD4 inhibitors, such as Cl-amidine and GSK484, have shown potential in reducing NETosis by blocking histone citrullination. Both can be encapsulated into NPs through physical entrapment. When these inhibitors are encapsulated in NPs, their pharmacokinetic properties improve, allowing for higher local concentrations and enhanced targeting of neutrophils at sites of inflammation [112, 121].

NPs have been developed to deliver PAD4 inhibitors for the treatment of ischemic stroke. For example, Sun et al. reported a Cys-Arg-Glu-Lys-Ala (CREKA) peptide-modified ROS-responsive liposome system targeting fibrin in microthrombi at ischemic stroke sites (Fig. 5A). The CREKA peptide bound to fibrin and fibrin-associated clots. Cl-amidine was encapsulated within the liposomal nanocarrier through physical interactions during the liposome preparation process. The nanocarriers could block the PAD4 enzyme and suppress the activation of the cGAS-STING pathway, reducing inflammation and microglial polarization toward an anti-inflammatory M2-like phenotype. This improved the integrity of the BBB by reducing vascular leakage and upregulating tight junction proteins claudin-5 and ZO-1 [112]. Additionally, GSK484-loaded ROS-responsive polymer-based nanocarriers were synthesized to prevent the citrullination of histone-3, reducing NETosis and neuroinflammation. GSK484 was encapsulated into NPs designed with ROS-sensitive diselenide (-Se-Se-) bonds within the polymer matrix, enabling the controlled release of GSK484 in response to elevated ROS levels at injury sites [121].

NE inhibition

NE is another critical enzyme and is released from neutrophil granules into the nucleus during NETosis to degrade histones. This degradation helps loosen the tightly packed chromatin and aids in the subsequent release of NETs. Along with other enzymes like PAD4, NE facilitates chromatin unraveling. Inhibiting NE activity helps to reduce chromatin decondensation and NET-associated inflammation [171]. Research has shown that NE inhibitors like sivelestat can mitigate tissue damage and inflammation in diseases in which NETs play a pathogenic role [172]. The NP-based delivery of NE inhibitors has been explored for the purposes of suppressing NET formation and reducing the detrimental effects of excessive NETosis. Sivelestat can be encapsulated into NPs or conjugated via a thioketal bond [110, 155, 156].

Sivelestat has been delivered through NPs to treat atherosclerosis, ALI, and RA. For atherosclerosis therapy, cRGD-modified sivelestat-loaded liposome was synthesized to inhibit NE activity and NET formation. Sivelestat was encapsulated into liposomes using a film dispersion method, where it was physically entrapped within the lipid bilayer. The cRGD peptide modification allowed for targeting and uptake by neutrophils [110]. An interbilayer-crosslinked multilamellar vesicle system encapsulated with sivelestat was designed to inhibit NE for the treatment of ALI. In vivo, the NPs decreased NE serum levels and pro-inflammatory cytokines, thereby reducing NET-mediated tissue damage and inflammatory responses [155]. Furthermore, using the collagen-induced

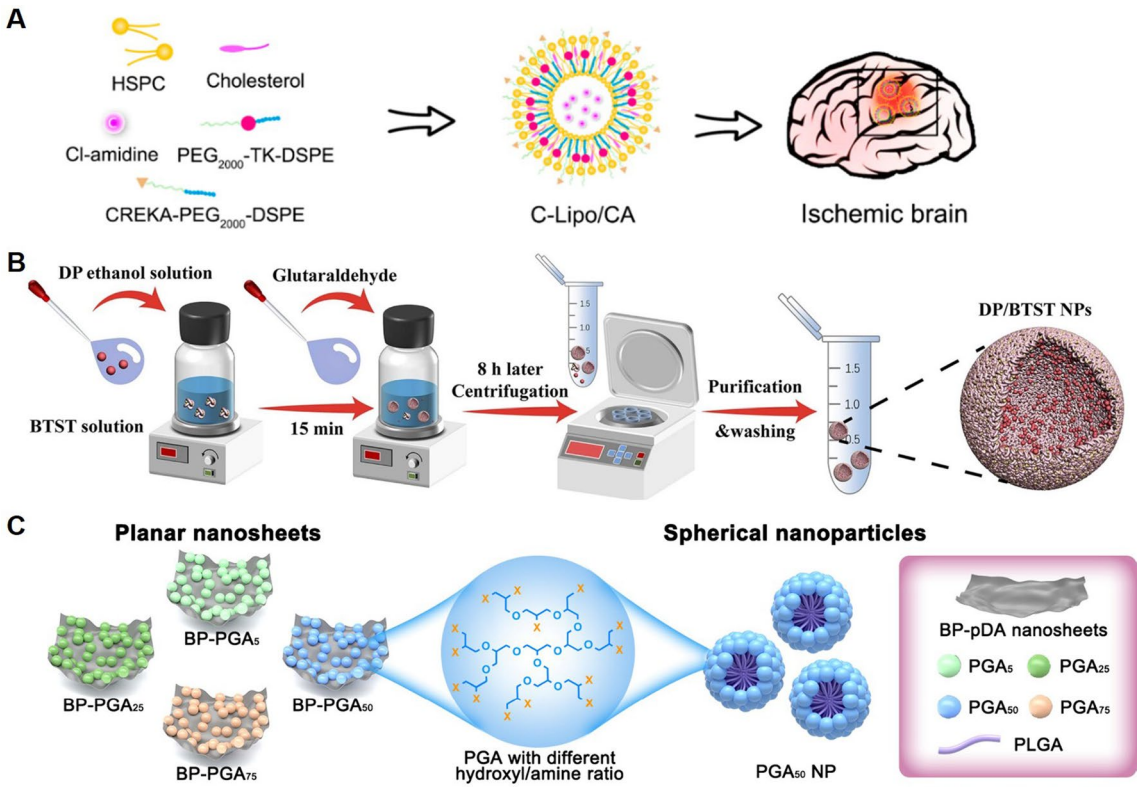


Fig. 5 Applications of NP-based drug delivery systems in inhibiting NET formation by PAD4 inhibition (**A**), NE inhibition (**B**), and neutralizing NET-associated components or mediators (**C**). **A** Preparation of Targeted ROS Stimulus-Responsive Liposomes (C-Lipo/CA) [112]. Copyright © 2023, American Chemical Society. **B** The preparation process of DP/BTST NPs [156]. Copyright © 2023 Published by Elsevier. **C** PGA-covered nanosheets with different hydroxyl/amine ratios and PGA-covered nanoparticles were fabricated [159]. Copyright © 2024, American Chemical Society

arthritis mouse model, dexamethasone palmitate-loading BSA NPs were developed for the alleviation of RA (Fig. 5B). Sivelestat was conjugated to BSA NPs via a thioketal bond, a sulfur-containing compound forming ROS-sensitive linkages. This bond ensured sivelestat's release in high ROS environments [156]. Additionally, a nanomedicine platform used α1-antitrypsin-derived peptide motif to inhibit NE activity for treating thrombosis. Hydroxychloroquine, serving as an NE inhibitor, was loaded into the NPs by way of physical encapsulation during NP assembly. By binding to NE, the NPs targeted activated neutrophils to deliver hydroxychloroquine directly to sites with abundant activated neutrophils, alleviating conditions in which excessive NETs contribute to the disease pathology, such as thrombosis and inflammation [157].

Neutralization of NET-associated components or mediators

Neutralizing NET components that contribute to tissue damage and inflammation, such as histones and proteases, can alleviate NETs' harmful effects without necessarily preventing their formation. For instance, neutralizing DNA released by neutrophils could reduce its adverse effects. During NET formation, mediators like hypochlorous acid are essential for stabilizing NET structure; inhibiting or scavenging these mediators thus interferes with the NETosis process [160].

NPs can be engineered to deliver agents that specifically neutralize NET components or inhibit protease activity. One study designed a positively charged cationic poly(aspartic acid) NP (cANP) to bind strongly to the negatively charged NET–DNA through electrostatic interactions, preventing NET–DNA from interacting with tumor cells and blocking its chemotactic function, which attracted tumor cells to metastatic sites. The NPs also competed with tumor cell surface proteins like CCDC25 for NET–DNA binding, thereby disrupting the NET-mediated signaling that promotes tumor cell migration and metastasis in mice and human metastatic models [158]. In another study, black phosphorus (BP) nanosheets functionalized with polyglycerol-50% amine (BP-PGA50) bound and removed cell-free DNA (cfDNA), a critical component inducing NET formation (Fig. 5C). By scavenging cfDNA, BP-PGA50 reduced inflammatory cytokines (IL-1β, IL-6, and TNF-α) in the nasal and

lung tissues of airway inflammatory model mice and suppressed TLR9 activation [159].

Some catalysts or mediators play important roles in NET formation, and NPs can be manipulated to interfere with this process. Wang et al. designed a type of sulfoxide-containing homopolymer with fouling-resistant and NET-inhibiting capabilities—poly(2-methacryloyloxyethyl sulfide sulfoxide) (PMeSEA)—for the treatment of tumors. The hydrating sulfoxide groups of PMeSEA inhibited protein/cell adhesion and scavenged hypochlorous acid, an oxidizing agent produced by neutrophils that plays a key role in NET stabilization and formation. In this way, PMeSEA prevented postoperative adhesions and suppressed peritoneal metastasis [160].

Inhibition of neutrophil recruitment and activation

Neutrophils' recruitment to sites of inflammation and their subsequent activation play a critical role in initiating NETosis. Inhibiting neutrophil chemotaxis and activation can reduce the number of neutrophils available to form NETs. Manipulating NPs to decrease the number of neutrophils or block neutrophil adhesion and infiltration may help to prevent excessive immune cell accumulation at inflamed sites and reduce damage caused by NET-mediated tissue injury [136].

Inhibition of neutrophil recruitment improves NET-induced immune-mediated injury. Research demonstrated that a trimethylamine N-oxide (TMAO)-derived zwitterionic hydrogel enhanced diabetic wound healing, inhibiting NET formation and coordinating the immune response by balancing the activities of macrophages and neutrophils. The hydrogel upregulated C–C motif chemokine ligand 3 (Ccl3) positive macrophages, which promote wound healing, and downregulated pro-inflammatory S100a9 + neutrophils [161]. Hybrid biomimetic nanovesicles composed of neutrophil membranes containing activated β2 integrins, fused with cationic lipid DDAB and loaded with docosahexaenoic acid (DHA), were developed to bind to intercellular adhesion molecules 1 and 2 (ICAM-1/2) on inflammatory vascular endothelium via β2 integrins. This blocked further neutrophil adhesion and infiltration, preventing NET-related inflammatory damage for the treatment of ARDS [138]. Another study reported on celastrol-loaded BSA NPs (CBR NPs) that targeted circulating neutrophils via RGD-integrin interactions, inducing apoptosis and reducing their recruitment with the aim of treating RA [162]. Star-shaped, anti-PSGL-1-antibody-coated AuNPs targeted activated neutrophils, reducing thrombosis by preventing PSGL-1 from binding to p-selectin on inflamed endothelium and activated platelets. This inhibited neutrophil recruitment and NET formation, with the star shape showing a higher affinity for PSGL-1 clusters [163].

Inhibiting neutrophil activation reduces NET release. A photocurable methacryloxylated silk fibroin hydrogel was developed to improve diabetic wound healing in orthopedic surgery. Metformin-loaded mesoporous silica NPs and AgNPs were incorporated into the hydrogel. The AgNPs ensured a sterile environment, while the metformin reduced neutrophil inflammation and promoted M2 macrophage polarization, further inhibiting NET formation [164]. Luminol-conjugated α-cyclodextrin was used to form LaCD NPs for treating asthma (Fig. 6A). LaCD NPs inhibited neutrophil recruitment and activation, decreased NET formation, and attenuated the activation of the NLRP3 inflammasome in neutrophils [165]. To summarize, inhibiting neutrophil recruitment and activation reduces NET formation and inflammation, mitigating tissue damage in diseases such as diabetic wounds, ARDS, RA, thrombosis, and asthma. NPs that target neutrophils effectively suppress immune responses and reduce immune-mediated injury.

ROS scavenging

ROS are crucial mediators of NET formation because they initiate the oxidative burst in neutrophils that triggers NETosis. Excessive ROS production not only promotes NETosis but also contributes to tissue damage [173]. NPs designed to scavenge ROS can inhibit NET formation by neutralizing oxidative stress within the microenvironment. For example, NPs loaded with antioxidants, such as luminol and Tempol, can reduce ROS levels. By diminishing oxidative stress, these NPs prevent the activation of the NETosis pathway, which ultimately reduces the harmful effects of excessive NET formation [166]. Furthermore, ROS-scavenging NPs can protect surrounding tissues from oxidative damage, providing a dual therapeutic benefit [117].

For example, with the aim of treating corneal pathological fibrosis treatment, a responsive hydrogel made of oxidized chitosan NPs loaded with black phosphorus quantum dots and grafted to silk fibroin methacrylate was used to inhibit NET formation by scavenging ROS and preventing NF-κB activation. This downregulated pro-inflammatory markers, reduced M2 macrophage polarization, and prevented macrophage-induced fibrosis [117]. In a study developing a potential treatment for AAA, LaCD NPs—comprising luminol-conjugated α-cyclodextrin and functionalized with alendronate for targeting calcified tissues—exhibited ROS scavenging and anti-inflammatory properties. These NPs inhibited NETosis by reducing neutrophil activation, suppressing pro-inflammatory mediators (MPO, TNF-α, CXCL1), and decreasing MMP-2 and MMP-9 levels in aortas [123]. Additionally, a research group developed TPCN, a cyclic oligosaccharide-derived nanotherapy for severe asthma that was based on Tempol and PBAP conjugated to

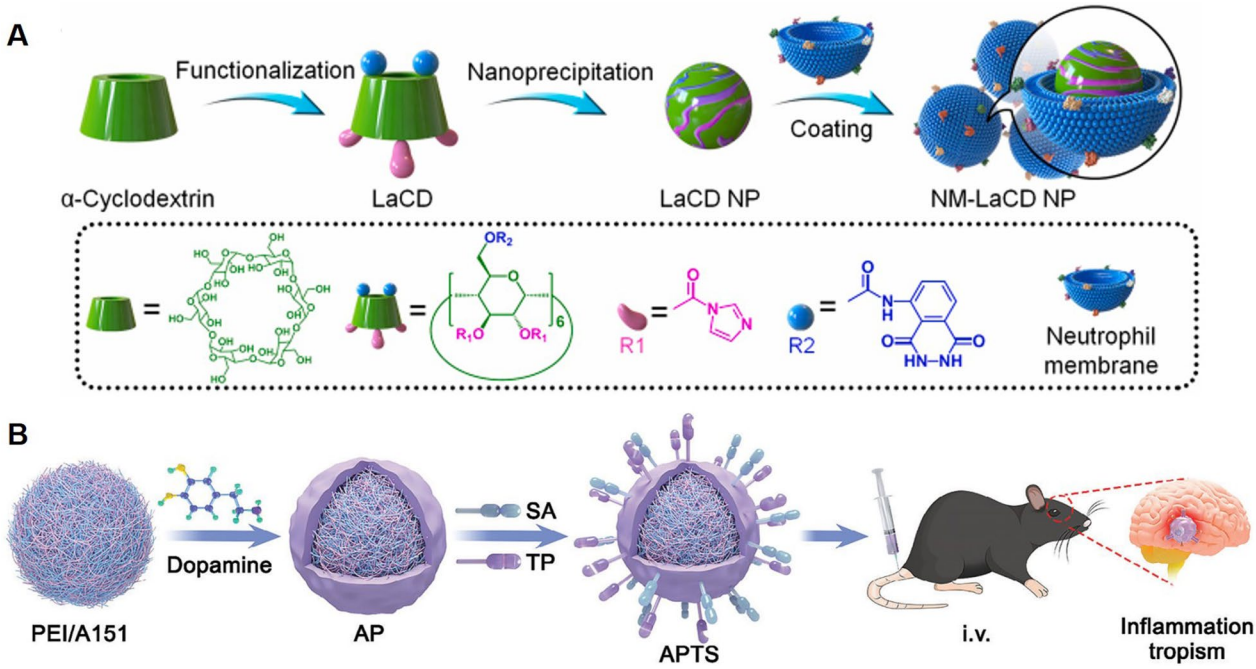


Fig. 6 Applications of NP-based drug delivery systems in inhibiting NET formation by inhibiting neutrophil recruitment and activation (**A**) and ROS scavenging (**B**). **A** Chemical structure of LaCD as well as engineering of LaCD nanoparticles (LaCD NP) and neutrophil membrane-coated LaCD NP (NM-LaCD NP) [165]. Copyright © 2023 The Authors. **B** Preparation process of APTS. AP, A151/PEI@PDA; SA, sialic acid; TP, targeting peptide; APTS, nanoplatform composed of AP with modifications of SA and TP peptide [167]. Copyright © 2024 The Authors

β-cyclodextrin. TPCN was designed to target and neutralize ROS, inhibit NET formation, and reduce pro-inflammatory cytokines and oxidative stress, thereby decreasing neutrophil infiltration and airway remodeling and restoring Treg/Th17 balance for immune homeostasis [166]. A polydopamine-coated A151/PEI nanoplatform was developed to scavenge ROS, decrease the levels of MPO, NE, and CitH3, and prevent the chromatin decondensation necessary for NET formation (Fig. 6B). Furthermore, the NPs exposed phosphatidylserine on the neutrophil surface, signaling apoptotic cell death rather than NETosis [167]. In addition, neutrophils incubated with puerarin-loaded liposomes via electrostatic interactions were designed to chemotax to the brain injury site, where they released puerarin. This reduced NETs, oxidative stress, and inflammation, thereby protecting neural cells from damage [168].

Inhibiting NET formation can effectively reduce tissue damage and inflammation in those diseases in which excessive NETs contribute to pathology. NP-based strategies, including degrading the DNA scaffold, inhibiting PAD4 or NE activity, neutralizing NET components, blocking neutrophil recruitment and activation, and scavenging ROS, have shown promise for targeting NET-related diseases and improving therapeutic outcomes.

Challenges and prospects of NP-based drug delivery systems in NET modulation

Current technological limitations of NPs

Despite the promise of NP-based drug delivery systems in targeting NETs, several technological challenges remain. One primary limitation is the complexity of designing NPs that can effectively navigate the diverse physiological environments encountered in vivo. This complexity includes difficulties ensuring NP stability, the precise targeting of NETs, and the balance between reduced neutrophil activation and NETs targeting.

NPs' stability under physiological conditions is a prerequisite to ensuring therapeutic effectiveness. NPs often encounter a range of environments within the body, including varying pH, enzymatic activity, and oxidative stress, all of which may impact their structural integrity and drug-release properties. Ensuring that NPs both remain stable long enough to reach their target and degrade or release their cargo upon reaching the intended site is a delicate balance requiring precise engineering [174]. Furthermore, because of the large molecular weight of the enzyme, how to load it with NPs while maintaining its activity is also one of nanotechnology's challenges.

The precise targeting of NETs is another challenge for NPs. While several strategies have been developed

to improve NP targeting, such as electrostatic interactions or surface modifications with ligands, their efficiency remains suboptimal. NETs are dynamic structures formed at sites of inflammation and can vary in both spatial distribution and density [175]. Additionally, NPs may face off-target effects, binding to unintended proteins or cells, which could reduce their efficacy or cause unwanted side effects. Enhanced specificity in targeting NET components like DNA, histones, or proteases is a promising way to maximize therapeutic potential while minimizing off-target toxicity.

The balance between reduced neutrophil activation and NETs targeting should also be considered. While systemically administered modified NPs can target neutrophils and inhibit their activation and recruitment to inflammatory sites, their ability to specifically target NET formation may be limited, since neutrophils would not be present at the inflammatory sites. One potential solution is to incorporate environment-responsive features into the design of NPs, such as pH- or ROS-sensitive mechanisms that enable selective activation and drug release in the inflammatory microenvironment [176]. Alternatively, NPs can be functionalized to recognize inflammation-specific markers, such as fibrin or pro-inflammatory cytokines, ensuring that therapeutic effects are concentrated at the sites of inflammation [112, 161]. These strategies allow for precise NET targeting while minimizing off-target effects and preserving the therapeutic benefits of neutrophil modulation.

Clinical challenges of NP application

Several clinical challenges must be overcome to ensure the safe and effective use of NP-based therapies in patients. These challenges include issues related to safety and toxicity, biodistribution, and regulatory hurdles that must be addressed before widespread clinical application.

Safety remains a major concern in the clinical application of NPs. Certain types of NPs, particularly those made from non-degradable materials (e.g., metal-based NPs such as gold or silver), may accumulate in organs, such as the liver, kidneys, or lungs, over time, leading to potential toxicity. These accumulations may cause chronic inflammation, organ damage, or unintended effects on the immune system [177]. Testing the toxicity of NPs often requires long-term studies to fully understand their effects.

The biodistribution of NPs is a crucial factor influencing their clinical effectiveness. After administration, NPs can be distributed to various tissues and organs, including non-target sites, which may reduce the amount of drug reaching the intended location. For instance, because of the high concentration of macrophages in the liver and spleen, many NPs are preferentially taken up by these organs, leading to reduced NP availability at the target site, including areas of NET formation [178]. Designing NPs with optimal size, charge, and surface properties to ensure effective biodistribution while minimizing unwanted accumulation in non-target tissues is critical for their clinical application.

The large-scale production of NPs with consistent size, shape, and surface characteristics is also an area of concern, affecting the reproducibility of experimental results and the scalability of clinical applications. Establishing standardized synthesis protocols and scalable manufacturing processes is crucial for the successful clinical translation of NP-based therapies [179].

Future directions and potential of NP application in NET-related diseases

NP-based therapies hold promise for targeting NETs in a wide range of diseases. With advances in research, there are several exciting future directions that could enhance the effectiveness, safety, and precision of NP-based therapies for NET-related conditions. These innovations are likely to focus on enhancing targeted delivery, exploring combination therapies, and advancing personalized nanomedicine.

Stimuli-responsive NPs allow for precise drug release in response to specific triggers in the NET microenvironment. These NPs are designed to react to particular stimuli such as pH changes, ROS, enzymatic activity, or temperature shifts, all of which are prevalent in areas of NET formation [176]. These stimuli-responsive systems facilitate localized and time-controlled drug release, reducing systemic toxicity and enhancing therapeutic efficacy.

Another exciting frontier is the integration of nanomedicine with other treatment modalities, such as gene therapy, immunotherapy, or photodynamic therapy [145]. By targeting multiple pathways simultaneously, NP-based combination therapies can enhance therapeutic efficacy and address the multifaceted nature of NET-related diseases, offering a more holistic and potent treatment strategy.

Personalized nanomedicine, treatment tailored to an individual's unique biological and genetic profile [180], is likely to play an increasing role in the future of NET-targeting therapies. As research into NET biology and patient-specific disease mechanisms advances, personalized NP-based therapies could be developed to target specific NET-related pathways that are unique to each patient. Advancements in diagnostics and biomarker discovery could also help identify the patients most likely to respond to NP-based therapies, enabling more targeted and effective treatment plans [181].

By addressing these challenges and embracing these future research directions, NP-based therapies hold the potential to revolutionize the treatment of NET-related diseases. Through advances in stimuli-responsive systems, combination therapies, and personalized approaches, the therapeutic landscape for conditions involving excessive NET formation could see transformative improvements.

Conclusion

NETs play a crucial role in both protective immune responses and pathological conditions, making them a significant target for therapeutic interventions. NP-based drug delivery systems offer innovative solutions for NET modulation, providing opportunities to enhance treatment efficacy and precision. Utilized as drug delivery vehicles to promote or inhibit NET formation, NPs have demonstrated their potential in addressing a range of NET-related diseases. However, significant challenges remain, including technological limitations, safety concerns, and the need for scalable production. Future advances in nanotechnology hold great promise for overcoming these challenges, paving the way for novel therapeutic strategies to treat complex NET-related diseases. The continued exploration of NP–NET interactions will be essential for translating these promising technologies into clinical applications.

Abbreviations

NETs	Neutrophil extracellular traps
NP	Nanoparticle
NE	Neutrophil elastase
MPO	Myeloperoxidase
PMA	Phosphomyristate
PKC	Protein kinase C
ROS	Reactive oxygen species
NOX	NADPH oxidase
PAD4	Peptidylarginine deiminase 4
TLR2	Toll-like receptor 2
TLR4	Toll-like receptor 4
LPS	Lipopolysaccharide
SLE	Systemic lupus erythematosus
RA	Rheumatoid arthritis
DAMPs	Damage-associated molecular patterns
MHC	Major histocompatibility complex
ANCA	Anti-neutrophil cytoplasmic autoantibody
TME	Tumor microenvironment
G-CSF	Granulocyte colony-stimulating factor
IL-8	Interleukin-8
MMPs	Matrix metalloproteinases
EMT	Epithelial mesenchymal transition
CTCs	Circulating tumor cells
DOX	Doxorubicin
TF	Tissue factor
DVT	Deep vein thrombosis
PE	Pulmonary embolism
vWF	Von Willebrand factor
FXII	Factor XII
VTE	Venous thrombus embolism
CitH3	Citrullinated histone H3
COPD	Chronic obstructive pulmonary disease
ALI	Acute lung injury
ARDS	Acute respiratory distress syndrome
AAA	Abdominal aortic aneurysm
IBD	Inflammatory bowel disease
PLA	Polylactic acid
PLGA	Poly(lactic-*co*-glycolic acid)
MRI	Magnetic resonance imaging
AuNPs	Gold NPs
SPIONs	Superparamagnetic iron oxide NPs
PEO	Plasma electrolytic oxidation
CPDs	Carbonized polymer dots
MPS	Methylprednisolone sodium succinate
SNase	Staphylococcal nuclease
5-HT	5-Hydroxytryptamine
MMP-9	Matrix metalloproteinase 9
cfDNA	Cell-free DNA
Ce6	Chlorin e6
BSA	Bovine serum albumin
PSGL-1	P-selectin glycoprotein ligand-1
cRGD	Cyclic arginine-glycine-aspartate
cRGDfC	Cyclo (Arg-Gly-Asp-D-Phe-Cys)
DSPE-PEG_{2000}-Mal	1,2-Distearoyl-sn-glycero-3-phosphoethanolamine-polyethylene glycol 2000-maleimide
PEG	Polyethylene Glycol
PEGylation	Polyethylene Glycolylation
CREKA	Cys-Arg-Glu-Lys-Ala
CCDC25	Coiled-coil domain containing 25
PMeSEA	Poly(2-methacryloyloxyethyl sulfide sulfoxide)
Ccl3	C–C motif chemokine ligand 3
ICAM-1/2	Intercellular adhesion molecule 1 and 2
CXCR2	CXC motif chemokine receptor 2

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HL drafted the manuscript. CW, TS and SL participated in finalizing the concept and design of the manuscript. CL and CF checked different sections of the manuscript. SL edited and polished the manuscript. YW, TL, HW, and CW revised the manuscript. All authors read and approved the final manuscript.

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Declarations

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Competing interests
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References

1. Glenn A, Armstrong CE. Physiology of red and white blood cells. Anaesth Intensive Care Med. 2019;20(3):170–4.
2. Pérez-Figueroa E, Álvarez-Carrasco P, Ortega E, Maldonado-Bernal C. Neutrophils: many ways to die. Front Immunol. 2021;12: 631821.
3. Zawrotniak M, Rapala-Kozik M. Neutrophil extracellular traps (NETs)—formation and implications. Acta Biochim Pol. 2013;60(3):277–84.
4. Yipp BG, Kubes P. NETosis: how vital is it? Blood. 2013;122(16):2784–94.
5. Vorobjeva NV, Chernyak BV. NETosis: molecular mechanisms, role in physiology and pathology. Biochemistry (Mosc). 2020;85(10):1178–90.
6. Zhou X, Jin S, Pan J, Lin Q, Yang S, Ambe PC, et al. Damage associated molecular patterns and neutrophil extracellular traps in acute pancreatitis. Front Cell Infect Microbiol. 2022;12: 927193.
7. Huang H, Tohme S, Al-Khafaji AB, Tai S, Loughran P, Chen L, et al. Damage-associated molecular pattern-activated neutrophil extracellular trap exacerbates sterile inflammatory liver injury. Hepatology. 2015;62(2):600–14.
8. Iba T, Murai M, Nagaoka I, Tabe Y. Neutrophil extracellular traps, damage-associated molecular patterns, and cell death during sepsis. Acute Med Surg. 2014;1(1):2–9.
9. Manda-Handzlik A, Stojkov D, Wachowska M, Surmiak M. Editorial: neutrophil extracellular traps: mechanistic and functional insight. Front Immunol. 2024;15:1407232.
10. Chen T, Li Y, Sun R, Hu H, Liu Y, Herrmann M, et al. Receptor-mediated NETosis on neutrophils. Front Immunol. 2021;12: 775267.
11. Papayannopoulos V. Neutrophil extracellular traps in immunity and disease. Nat Rev Immunol. 2018;18(2):134–47.
12. Fang H, Bo Y, Hao Z, Mang G, Jin J, Wang H. A promising frontier: targeting NETs for stroke treatment breakthroughs. Cell Commun Signal. 2024;22(1):238.
13. Clark SR, Ma AC, Tavener SA, McDonald B, Goodarzi Z, Kelly MM, et al. Platelet TLR4 activates neutrophil extracellular traps to ensnare bacteria in septic blood. Nat Med. 2007;13(4):463–9.
14. Yipp BG, Petri B, Salina D, Jenne CN, Scott BN, Zbytnuik LD, et al. Infection-induced NETosis is a dynamic process involving neutrophil multitasking in vivo. Nat Med. 2012;18(9):1386–93.
15. Segal AW. How neutrophils kill microbes. Annu Rev Immunol. 2005;23:197–223.
16. Branzk N, Lubojemska A, Hardison SE, Wang Q, Gutierrez MG, Brown GD, et al. Neutrophils sense microbe size and selectively release neutrophil extracellular traps in response to large pathogens. Nat Immunol. 2014;15(11):1017–25.
17. Bruschi M, Bonanni A, Petretto A, Vaglio A, Pratesi F, Santucci L, et al. Neutrophil extracellular traps profiles in patients with incident systemic lupus erythematosus and lupus nephritis. J Rheumatol. 2020;47(3):377–86.
18. Martinod K, Claessen A, Martens C, Krauel K, Velásquez Pereira LC, Witsch J, et al. NET burden in left atrial blood is associated with biomarkers of thrombosis and cardiac injury in patients with enlarged left atria. Clin Res Cardiol. 2024. https://doi.org/10.1007/s00392-024-02464-9.
19. Zhang Q, Zhang J, Gu H, Yang Y, Zhang H, Miao C. Perioperative NETosis and cancer progression: current evidence and future perspectives. Curr Oncol Rep. 2024. https://doi.org/10.1007/s11912-024-01573-y.
20. Kupor D, Felder ML, Kodikalla S, Chu X, Eniola-Adefeso O. Nanoparticle-neutrophils interactions for autoimmune regulation. Adv Drug Deliv Rev. 2024;209: 115316.
21. Chellappan DK, Yee LW, Xuan KY, Kunalan K, Rou LC, Jean LS, et al. Targeting neutrophils using novel drug delivery systems in chronic respiratory diseases. Drug Dev Res. 2020;81(4):419–36.
22. Huang J, Hong W, Wan M, Zheng L. Molecular mechanisms and therapeutic target of NETosis in diseases. MedComm (2020). 2022;3(3): e162.
23. Islam MM, Takeyama N. Role of neutrophil extracellular traps in health and disease pathophysiology: recent insights and advances. Int J Mol Sci. 2023;24(21):15805.
24. Baz AA, Hao H, Lan S, Li Z, Liu S, Chen S, et al. Neutrophil extracellular traps in bacterial infections and evasion strategies. Front Immunol. 2024;15:1357967.
25. Scozzi D, Liao F, Krupnick AS, Kreisel D, Gelman AE. The role of neutrophil extracellular traps in acute lung injury. Front Immunol. 2022;13: 953195.
26. Czaikoski PG, Mota JM, Nascimento DC, Sônego F, Castanheira FV, Melo PH, et al. Neutrophil extracellular traps induce organ damage during experimental and clinical sepsis. PLoS ONE. 2016;11(2): e0148142.
27. Saffarzadeh M, Juenemann C, Queisser MA, Lochnit G, Barreto G, Galuska SP, et al. Neutrophil extracellular traps directly induce epithelial and endothelial cell death: a predominant role of histones. PLoS ONE. 2012;7(2): e32366.
28. Kolaczkowska E, Jenne CN, Surewaard BG, Thanabalasuriar A, Lee WY, Sanz MJ, et al. Molecular mechanisms of NET formation and degradation revealed by intravital imaging in the liver vasculature. Nat Commun. 2015;6:6673.
29. Lee KH, Kronbichler A, Park DD, Park Y, Moon H, Kim H, et al. Neutrophil extracellular traps (NETs) in autoimmune diseases: a comprehensive review. Autoimmun Rev. 2017;16(11):1160–73.
30. Farrera C, Fadeel B. Macrophage clearance of neutrophil extracellular traps is a silent process. J Immunol. 2013;191(5):2647–56.
31. Wigerblad G, Kaplan MJ. Neutrophil extracellular traps in systemic autoimmune and autoinflammatory diseases. Nat Rev Immunol. 2023;23(5):274–88.
32. Sadeghi M, Dehnavi S, Jamialahmadi T, Johnston TP, Sahebkar A. Neutrophil extracellular trap: a key player in the pathogenesis of autoimmune diseases. Int Immunopharmacol. 2023;116: 109843.
33. Pisetsky DS. Unique interplay between antinuclear antibodies and nuclear molecules in the pathogenesis of systemic lupus erythematosus. Arthritis Rheumatol. 2024. https://doi.org/10.1002/art.42863.
34. van der Linden M, van den Hoogen LL, Westerlaken GHA, Fritsch-Stork RDE, van Roon JAG, Radstake T, et al. Neutrophil extracellular trap release is associated with antinuclear antibodies in systemic lupus erythematosus and anti-phospholipid syndrome. Rheumatology (Oxford). 2018;57(7):1228–34.
35. Kang N, Liu X, Haneef K, Liu W. Old and new damage-associated molecular patterns (DAMPs) in autoimmune diseases. Rheumatol Autoimmunity. 2022;2(4):185–97.
36. Khandpur R, Carmona-Rivera C, Vivekanandan-Giri A, Gizinski A, Yalavarthi S, Knight JS, et al. NETs are a source of citrullinated autoantigens and stimulate inflammatory responses in rheumatoid arthritis. Sci Transl Med. 2013;5(178):178ra40.
37. Carmona-Rivera C, Carlucci PM, Moore E, Lingampalli N, Uchtenhagen H, James E, et al. Synovial fibroblast-neutrophil interactions promote pathogenic adaptive immunity in rheumatoid arthritis. Sci Immunol. 2017;2(10):eaag3358.
38. Kessenbrock K, Krumbholz M, Schönermarck U, Back W, Gross WL, Werb Z, et al. Netting neutrophils in autoimmune small-vessel vasculitis. Nat Med. 2009;15(6):623–5.
39. Zhang Z, Niu R, Zhao L, Wang Y, Liu G. Mechanisms of neutrophil extracellular trap formation and regulation in cancers. Int J Mol Sci. 2023;24(12):10265.
40. Cristinziano L, Modestino L, Antonelli A, Marone G, Simon H-U, Varricchi G, et al. Neutrophil extracellular traps in cancer. Semin Cancer Biol. 2022;79:91–104.
41. Podaza E, Sabbione F, Risnik D, Borge M, Almejún MB, Colado A, et al. Neutrophils from chronic lymphocytic leukemia patients exhibit an increased capacity to release extracellular traps (NETs). Cancer Immunol Immunother. 2017;66(1):77–89.
42. Demers M, Krause DS, Schatzberg D, Martinod K, Voorhees JR, Fuchs TA, et al. Cancers predispose neutrophils to release extracellular DNA traps that contribute to cancer-associated thrombosis. Proc Natl Acad Sci U S A. 2012;109(32):13076–81.

43. Zhu Y, Huang Y, Ji Q, Fu S, Gu J, Tai N, et al. Interplay between extracellular matrix and neutrophils in diseases. J Immunol Res. 2021;2021:8243378.
44. Martins-Cardoso K, Almeida VH, Bagri KM, Rossi MID, Mermelstein CS, König S, et al. Neutrophil extracellular traps (NETs) promote pro-metastatic phenotype in human breast cancer cells through epithelial-mesenchymal transition. Cancers (Basel). 2020;12(6):1542.
45. Wen L, Guo L, Zhang W, Li Y, Jiang W, Di X, et al. Cooperation between the inflammation and coagulation systems promotes the survival of circulating tumor cells in renal cell carcinoma patients. Front Oncol. 2019;9:504.
46. Chen Q, Zou J, He Y, Pan Y, Yang G, Zhao H, et al. A narrative review of circulating tumor cells clusters: a key morphology of cancer cells in circulation promote hematogenous metastasis. Front Oncol. 2022;12: 944487.
47. Kaltenmeier C, Simmons RL, Tohme S, Yazdani HO. Neutrophil extracellular traps (NETs) in cancer metastasis. Cancers (Basel). 2021;13(23):6131.
48. Shaul ME, Fridlender ZG. Neutrophils as active regulators of the immune system in the tumor microenvironment. J Leukoc Biol. 2017;102(2):343–9.
49. Li Y, Wu S, Zhao Y, Dinh T, Jiang D, Selfridge JE, et al. Neutrophil extracellular traps induced by chemotherapy inhibit tumor growth in murine models of colorectal cancer. J Clin Invest. 2024;134(5): e175031.
50. Mousset A, Lecorgne E, Bourget I, Lopez P, Jenovai K, Cherfils-Vicini J, et al. Neutrophil extracellular traps formed during chemotherapy confer treatment resistance via TGF-β activation. Cancer Cell. 2023;41(4):757-75.e10.
51. Shinde-Jadhav S, Mansure JJ, Rayes RF, Marcq G, Ayoub M, Skowronski R, et al. Role of neutrophil extracellular traps in radiation resistance of invasive bladder cancer. Nat Commun. 2021;12(1):2776.
52. Tamura K, Miyato H, Kanamaru R, Sadatomo A, Takahashi K, Ohzawa H, et al. Neutrophil extracellular traps (NETs) reduce the diffusion of doxorubicin which may attenuate its ability to induce apoptosis of ovarian cancer cells. Heliyon. 2022;8(6): e09730.
53. Martins-Cardoso K, Maçao A, Souza JL, Silva AG, König S, Martins-Gonçalves R, et al. TF/PAR2 signaling axis supports the protumor effect of neutrophil extracellular traps (NETs) on human breast cancer cells. Cancers (Basel). 2023;16(1):5.
54. Wang Y, Liu F, Chen L, Fang C, Li S, Yuan S, et al. Neutrophil extracellular traps (NETs) promote non-small cell lung cancer metastasis by suppressing lncRNA MIR503HG to activate the NF-κB/NLRP3 inflammasome pathway. Front Immunol. 2022;13: 867516.
55. Zhou X, Wu C, Wang X, Pan N, Sun X, Chen B, et al. Tumor cell-released autophagosomes (TRAPs) induce PD-L1-decorated NETs that suppress T-cell function to promote breast cancer pulmonary metastasis. J Immunother Cancer. 2024;12(6): e009082.
56. Laridan E, Martinod K, De Meyer SF. Neutrophil extracellular traps in arterial and venous thrombosis. Semin Thromb Hemost. 2019;45(1):86–93.
57. Xie M, Hao Y, Feng L, Wang T, Yao M, Li H, et al. Neutrophil heterogeneity and its roles in the inflammatory network after ischemic stroke. Curr Neuropharmacol. 2023;21(3):621–50.
58. Zhou Y, Xu Z, Liu Z. Impact of neutrophil extracellular traps on thrombosis formation: new findings and future perspective. Front Cell Infect Microbiol. 2022;12: 910908.
59. Li W, Wang Z, Su C, Liao Z, Pei Y, Wang J, et al. The effect of neutrophil extracellular traps in venous thrombosis. Thromb J. 2023;21(1):67.
60. Massberg S, Grahl L, von Bruehl ML, Manukyan D, Pfeiler S, Goosmann C, et al. Reciprocal coupling of coagulation and innate immunity via neutrophil serine proteases. Nat Med. 2010;16(8):887–96.
61. Badimon L, Vilahur G. Neutrophil extracellular traps: a new source of tissue factor in atherothrombosis. Eur Heart J. 2015;36(22):1364–6.
62. Xu X, Wu Y, Xu S, Yin Y, Ageno W, De Stefano V, et al. Clinical significance of neutrophil extracellular traps biomarkers in thrombosis. Thromb J. 2022;20(1):63.
63. Rangaswamy C, Englert H, Deppermann C, Renné T. Polyanions in coagulation and thrombosis: focus on polyphosphate and neutrophils extracellular traps. Thromb Haemost. 2021;121(8):1021–30.
64. Yao M, Ma J, Wu D, Fang C, Wang Z, Guo T, et al. Neutrophil extracellular traps mediate deep vein thrombosis: from mechanism to therapy. Front Immunol. 2023;14:1198952.
65. Li C, Xing Y, Zhang Y, Hua Y, Hu J, Bai Y. Neutrophil extracellular traps exacerbate ischemic brain damage. Mol Neurobiol. 2022;59(1):643–56.
66. Allen C, Thornton P, Denes A, McColl BW, Pierozynski A, Monestier M, et al. Neutrophil cerebrovascular transmigration triggers rapid neurotoxicity through release of proteases associated with decondensed DNA. J Immunol. 2012;189(1):381–92.
67. Chen Y, Li X, Lin X, Liang H, Liu X, Zhang X, et al. Complement C5a induces the generation of neutrophil extracellular traps by inhibiting mitochondrial STAT3 to promote the development of arterial thrombosis. Thromb J. 2022;20(1):24.
68. Cui BB, Tan CY, Schorn C, Tang HH, Liu Y, Zhao Y. Neutrophil extracellular traps in sterile inflammation: the story after dying? Autoimmunity. 2012;45(8):593–6.
69. Hegde M, Girisa S, Devanarayanan TN, Alqahtani MS, Abbas M, Sethi G, et al. Network of extracellular traps in the pathogenesis of sterile chronic inflammatory diseases: role of oxidative stress and potential clinical applications. Antioxid Redox Signal. 2023. https://doi.org/10.1089/ars.2023.0329.
70. Zhang F, Li Y, Wu J, Zhang J, Cao P, Sun Z, et al. The role of extracellular traps in ischemia reperfusion injury. Front Immunol. 2022;13:1022380.
71. Qi H, Yang S, Zhang L. Neutrophil extracellular traps and endothelial dysfunction in atherosclerosis and thrombosis. Front Immunol. 2017;8:928.
72. Keir HR, Chalmers JD. Neutrophil extracellular traps in chronic lung disease: implications for pathogenesis and therapy. Eur Respir Rev. 2022;31(163): 210241.
73. Yan S, Li M, Liu B, Ma Z, Yang Q. Neutrophil extracellular traps and pulmonary fibrosis: an update. J Inflamm (Lond). 2023;20(1):2.
74. Li H, Li Y, Song C, Hu Y, Dai M, Liu B, et al. Neutrophil extracellular traps augmented alveolar macrophage pyroptosis via AIM2 inflammasome activation in LPS-induced ALI/ARDS. J Inflamm Res. 2021;14:4839–58.
75. Hough KP, Curtiss ML, Blain TJ, Liu R-M, Trevor J, Deshane JS, et al. Airway remodeling in asthma. Front Med. 2020;7:191.
76. King PT, Dousha L. Neutrophil extracellular traps and respiratory disease. J Clin Med. 2024;13(8):2390.
77. Warnatsch A, Ioannou M, Wang Q, Papayannopoulos V. Inflammation. Neutrophil extracellular traps license macrophages for cytokine production in atherosclerosis. Science. 2015;349(6245):316–20.
78. Wang Y, Wang C, Li J. Neutrophil extracellular traps: a catalyst for atherosclerosis. Mol Cell Biochem. 2024. https://doi.org/10.1007/s11010-024-04931-3.
79. Drury B, Hardisty G, Gray RD, Ho GT. Neutrophil extracellular traps in inflammatory bowel disease: pathogenic mechanisms and clinical translation. Cell Mol Gastroenterol Hepatol. 2021;12(1):321–33.
80. Landén NX, Li D, Ståhle M. Transition from inflammation to proliferation: a critical step during wound healing. Cell Mol Life Sci. 2016;73(20):3861–85.
81. Davis FM, Kimball A, Boniakowski A, Gallagher K. Dysfunctional wound healing in diabetic foot ulcers: new crossroads. Curr Diab Rep. 2018;18(1):2.
82. Wong SL, Demers M, Martinod K, Gallant M, Wang Y, Goldfine AB, et al. Diabetes primes neutrophils to undergo NETosis, which impairs wound healing. Nat Med. 2015;21(7):815–9.
83. Filipczak N, Li X, Saawant GR, Yalamarty SSK, Luther E, Torchilin VP. Antibody-modified DNase I micelles specifically recognize the neutrophil extracellular traps (NETs) and promote their degradation. J Control Release. 2023;354:109–19.
84. Rezaei N, Zadory M, Babity S, Marleau S, Brambilla D. Therapeutic applications of nanoparticles targeting neutrophil and extracellular traps. J Control Release. 2023;358:636–53.
85. Yang H, Marion TN, Liu Y, Zhang L, Cao X, Hu H, et al. Nanomaterial exposure induced neutrophil extracellular traps: a new target in inflammation and innate immunity. J Immunol Res. 2019;2019:3560180.
86. Vaseruk A, Bila G, Bilyy R. Nanoparticles for stimulation of neutrophil extracellular trap-mediated immunity. Eur J Immunol. 2024;54(4):2350582.
87. Bilyy R, Bila G, Vishchur O, Vovk V, Herrmann M. Neutrophils as main players of immune response towards nondegradable nanoparticles. Nanomaterials (Basel). 2020;10(7):1273.
88. Lovas M, Tanka-Salamon A, Beinrohr L, Voszka I, Szabó L, Molnár K, et al. Polyphosphate nanoparticles enhance the fibrin stabilization

by histones more efficiently than linear polyphosphates. PLoS ONE. 2022;17(4): e0266782.
89. Kang H, Seo J, Yang EJ, Choi IH. Silver nanoparticles induce neutrophil extracellular traps via activation of PAD and neutrophil elastase. Biomolecules. 2021;11(2):317.
90. Yang Y, Wang N, Zhu Y, Lu Y, Chen Q, Fan S, et al. Gold nanoparticles synergize with bacterial lipopolysaccharide to enhance class A scavenger receptor dependent particle uptake in neutrophils and augment neutrophil extracellular traps formation. Ecotoxicol Environ Saf. 2021;211: 111900.
91. Augustine R, Hasan A, Primavera R, Wilson RJ, Thakor AS, Kevadiya BD. Cellular uptake and retention of nanoparticles: insights on particle properties and interaction with cellular components. Mater Today Commun. 2020;25: 101692.
92. Bartneck M, Keul HA, Zwadlo-Klarwasser G, Groll J. Phagocytosis independent extracellular nanoparticle clearance by human immune cells. Nano Lett. 2010;10(1):59–63.
93. Golbek TW, Harper BJ, Harper SL, Baio JE. Shape-dependent gold nanoparticle interactions with a model cell membrane. Biointerphases. 2022;17(6): 061003.
94. Desai J, Foresto-Neto O, Honarpisheh M, Steiger S, Nakazawa D, Popper B, et al. Particles of different sizes and shapes induce neutrophil necroptosis followed by the release of neutrophil extracellular trap-like chromatin. Sci Rep. 2017;7(1):15003.
95. Fuchs TA, Brill A, Duerschmied D, Schatzberg D, Monestier M, Myers DD Jr, et al. Extracellular DNA traps promote thrombosis. Proc Natl Acad Sci U S A. 2010;107(36):15880–5.
96. Sorvillo N, Cherpokova D, Martinod K, Wagner DD. Extracellular DNA NET-works with dire consequences for health. Circ Res. 2019;125(4):470–88.
97. Hwang T-L, Aljuffali IA, Hung C-F, Chen C-H, Fang J-Y. The impact of cationic solid lipid nanoparticles on human neutrophil activation and formation of neutrophil extracellular traps (NETs). Chem Biol Interact. 2015;235:106–14.
98. Petretto E, Ong QK, Olgiati F, Mao T, Campomanes P, Stellacci F, et al. Monovalent ion-mediated charge-charge interactions drive aggregation of surface-functionalized gold nanoparticles. Nanoscale. 2022;14(40):15181–92.
99. Lotosh NY, Aliaseva SO, Malashenkova IK, Sorokoumova GM, Vasilov RG, Selischeva AA. Cationic liposomes cause ROS generation and release of neutrophil extracellular traps. Biochemistry (Moscow), Suppl Ser A Membr Cell Biol. 2019;13(1):40–9.
100. Yang H, Liu C, Yang D, Zhang H, Xi Z. Comparative study of cytotoxicity, oxidative stress and genotoxicity induced by four typical nanomaterials: the role of particle size, shape and composition. J Appl Toxicol. 2009;29(1):69–78.
101. Hou M, Wu X, Zhao Z, Deng Q, Chen Y, Yin L. Endothelial cell-targeting, ROS-ultrasensitive drug/siRNA co-delivery nanocomplexes mitigate early-stage neutrophil recruitment for the anti-inflammatory treatment of myocardial ischemia reperfusion injury. Acta Biomater. 2022;143:344–55.
102. Snoderly HT, Freshwater KA, Martinez de la Torre C, Panchal DM, Vito JN, Bennewitz MF. PEGylation of metal oxide nanoparticles modulates neutrophil extracellular trap formation. Biosensors (Basel). 2022;12(2):123.
103. Deng X, Zhao J, Liu K, Wu C, Liang F. Stealth PEGylated chitosan polyelectrolyte complex nanoparticles as drug delivery carrier. J Biomater Sci Polym Ed. 2021;32(11):1387–405.
104. Mitri N, Rahme K, Fracasso G, Ghanem E. Human blood biocompatibility and immunogenicity of scFvD2B PEGylated gold nanoparticles. Nanotechnology. 2022;33(31): 315101.
105. Wang C, Liu X, Han Z, Zhang X, Wang J, Wang K, et al. Nanosilver induces the formation of neutrophil extracellular traps in mouse neutrophil granulocytes. Ecotoxicol Environ Saf. 2019;183: 109508.
106. Yallapu MM, Chauhan N, Othman SF, Khalilzad-Sharghi V, Ebeling MC, Khan S, et al. Implications of protein corona on physico-chemical and biological properties of magnetic nanoparticles. Biomaterials. 2015;46:1–12.
107. Park SJ. Protein-nanoparticle interaction: corona formation and conformational changes in proteins on nanoparticles. Int J Nanomed. 2020;15:5783–802.
108. Kopac T. Protein corona, understanding the nanoparticle-protein interactions and future perspectives: a critical review. Int J Biol Macromol. 2021;169:290–301.
109. Bilyy R, Unterweger H, Weigel B, Dumych T, Paryzhak S, Vovk V, et al. Inert coats of magnetic nanoparticles prevent formation of occlusive intravascular co-aggregates with neutrophil extracellular traps. Front Immunol. 2018;9:2266.
110. Shi Y, Dong M, Wu Y, Gong F, Wang Z, Xue L, et al. An elastase-inhibiting, plaque-targeting and neutrophil-hitchhiking liposome against atherosclerosis. Acta Biomater. 2024;173:470–81.
111. Xu XL, Lu KJ, Zhu ML, Du YL, Zhu YF, Zhang NN, et al. Sialic acid-functionalized ph-triggered micelles for enhanced tumor tissue accumulation and active cellular internalization of orthotopic hepatocarcinoma. ACS Appl Mater Interfaces. 2018;10(38):31903–14.
112. Sun S, Lv W, Li S, Zhang Q, He W, Min Z, et al. Smart liposomal nanocarrier enhanced the treatment of ischemic stroke through neutrophil extracellular traps and cyclic guanosine monophosphate-adenosine monophosphate synthase-stimulator of interferon genes (cGAS-STING) pathway inhibition of ischemic penumbra. ACS Nano. 2023;17(18):17845–57.
113. Li R, Wang Z, Li J, Chen H, Guo X, Zhou S. Relieving thrombo-inflammation with acid-triggered polymersomes toward ischemic stroke therapy. Nano Today. 2024;54: 102114.
114. Cheng R, Wang S, Santos HA. Acid-labile chemical bonds-based nanoparticles for endosome escape and intracellular delivery. Biomed Technol. 2023;3:52–8.
115. Lim EK, Chung BH, Chung SJ. Recent advances in pH-sensitive polymeric nanoparticles for smart drug delivery in cancer therapy. Curr Drug Targets. 2018;19(4):300–17.
116. Van Driessche A, Kocere A, Everaert H, Nuhn L, Van Herck S, Griffiths G, et al. pH-sensitive hydrazone-linked doxorubicin nanogels via polymeric-activated ester scaffolds: synthesis, assembly, and in vitro and in vivo evaluation in tumor-bearing zebrafish. Chem Mater. 2018;30(23):8587–96.
117. Zhang J, Su L, Liu Z, Tang J, Zhang L, Li Z, et al. A responsive hydrogel modulates innate immune cascade fibrosis to promote ocular surface reconstruction after chemical injury. J Control Release. 2024;365:1124–38.
118. Xu Q, He C, Xiao C, Chen X. Reactive oxygen species (ROS) responsive polymers for biomedical applications. Macromol Biosci. 2016;16(5):635–46.
119. Xu X, Saw PE, Tao W, Li Y, Ji X, Bhasin S, et al. ROS-responsive polyprodrug nanoparticles for triggered drug delivery and effective cancer therapy. Adv Mater. 2017;29(33):1700141.
120. Wang CJ, Ko GR, Lee YY, Park J, Park W, Park TE, et al. Polymeric DNase-I nanozymes targeting neutrophil extracellular traps for the treatment of bowel inflammation. Nano Converg. 2024;11(1):6.
121. Mu Q, Yao K, Syeda MZ, Wan J, Cheng Q, You Z, et al. Neutrophil targeting platform reduces neutrophil extracellular traps for improved traumatic brain injury and stroke theranostics. Adv Sci (Weinh). 2024;11(21): e2308719.
122. Ocampo-Gallego JS, Pedroza-Escobar D, Caicedo-Ortega AR, Berumen-Murra MT, Novelo-Aguirre AL, de Sotelo-León RD, et al. Human neutrophil elastase inhibitors: classification, biological-synthetic sources and their relevance in related diseases. Fundam Clin Pharmacol. 2024;38(1):13–32.
123. Hu K, Zhong L, Lin W, Zhao G, Pu W, Feng Z, et al. Pathogenesis-guided rational engineering of nanotherapies for the targeted treatment of abdominal aortic aneurysm by inhibiting neutrophilic inflammation. ACS Nano. 2024;18(8):6650–72.
124. Liu C, Xi L, Liu Y, Mak JCW, Mao S, Wang Z, et al. An inhalable hybrid biomimetic nanoplatform for sequential drug release and remodeling lung immune homeostasis in acute lung injury treatment. ACS Nano. 2023;17(12):11626–44.
125. Li M, Zhang D, Peng F, Xie J, Zhang X, Qian S, et al. Zinc-doped ferric oxyhydroxide nano-layer enhances the bactericidal activity and osseointegration of a magnesium alloy through augmenting the formation of neutrophil extracellular traps. Acta Biomater. 2022;152:575–92.
126. Lin CJ, Hwang TL, Wang RYL, Nain A, Shih RH, Chang L, et al. Augmenting neutrophil extracellular traps with carbonized polymer

dots: a potential treatment for bacterial sepsis. Small. 2024;20(27): e2307210.
127. Li Y, Han Y, Su R, Liu Y, Chong G, Xu D, et al. Photosensitizer-laden neutrophils are controlled remotely for cancer immunotherapy. Cell Rep. 2020;33(11): 108499.
128. Zheng J, Qi R, Dai C, Li G, Sang M. Enzyme catalysis biomotor engineering of neutrophils for nanodrug delivery and cell-based thrombolytic therapy. ACS Nano. 2022;16(2):2330–44.
129. Cheng X, Yu P, Zhou X, Zhu J, Han Y, Zhang C, et al. Enhanced tumor homing of pathogen-mimicking liposomes driven by R848 stimulation: a new platform for synergistic oncology therapy. Acta Pharm Sin B. 2022;12(2):924–38.
130. Li M, Li S, Zhou H, Tang X, Wu Y, Jiang W, et al. Chemotaxis-driven delivery of nano-pathogenoids for complete eradication of tumors post-phototherapy. Nat Commun. 2020;11(1):1126.
131. Brinkmann V, Reichard U, Goosmann C, Fauler B, Uhlemann Y, Weiss DS, et al. Neutrophil extracellular traps kill bacteria. Science. 2004;303(5663):1532–5.
132. Zhang E, Zhao X, Hu J, Wang R, Fu S, Qin G. Antibacterial metals and alloys for potential biomedical implants. Bioact Mater. 2021;6(8):2569–612.
133. Cao M, Wang G, Xie J. Immune dysregulation in sepsis: experiences, lessons and perspectives. Cell Death Discov. 2023;9(1):465.
134. van der Poll T, Shankar-Hari M, Wiersinga WJ. The immunology of sepsis. Immunity. 2021;54(11):2450–64.
135. Zhang H, Wang Y, Qu M, Li W, Wu D, Cata JP, et al. Neutrophil, neutrophil extracellular traps and endothelial cell dysfunction in sepsis. Clin Transl Med. 2023;13(1): e1170.
136. Cesta MC, Zippoli M, Marsiglia C, Gavioli EM, Cremonesi G, Khan A, et al. Neutrophil activation and neutrophil extracellular traps (NETs) in COVID-19 ARDS and immunothrombosis. Eur J Immunol. 2023;53(1): e2250010.
137. SenGupta S, Subramanian BC, Parent CA. Getting TANned: How the tumor microenvironment drives neutrophil recruitment. J Leukoc Biol. 2019;105(3):449–62.
138. Li X, Qiao Q, Liu X, Hu Q, Yu Y, Qin X, et al. Engineered biomimetic nanovesicles based on neutrophils for hierarchical targeting therapy of acute respiratory distress syndrome. ACS Nano. 2024;18(2):1658–77.
139. Yuan S, Hu Q. Convergence of nanomedicine and neutrophils for drug delivery. Bioactive Mater. 2024;35:150–66.
140. Kruger P, Saffarzadeh M, Weber AN, Rieber N, Radsak M, von Bernuth H, et al. Neutrophils: between host defence, immune modulation, and tissue injury. PLoS Pathog. 2015;11(3): e1004651.
141. Mutua V, Gershwin LJ. A review of neutrophil extracellular traps (NETs) in disease: potential anti-NETs therapeutics. Clin Rev Allergy Immunol. 2021;61(2):194–211.
142. Du Y, Chen Y, Li F, Mao Z, Ding Y, Wang W. Genetically engineered cellular nanovesicle as targeted DNase I delivery system for the clearance of neutrophil extracellular traps in acute lung injury. Adv Sci (Weinh). 2023;10(32): e2303053.
143. Park HH, Park W, Lee YY, Kim H, Seo HS, Choi DW, et al. Bioinspired DNase-I-coated melanin-like nanospheres for modulation of infection-associated NETosis dysregulation. Adv Sci (Weinh). 2020;7(23):2001940.
144. Dong W, Liu D, Zhang T, You Q, Huang F, Wu J. Oral delivery of staphylococcal nuclease ameliorates DSS induced ulcerative colitis in mice via degrading intestinal neutrophil extracellular traps. Ecotoxicol Environ Saf. 2021;215: 112161.
145. Wang N, Ma J, Song W, Zhao C. An injectable hydrogel to disrupt neutrophil extracellular traps for treating rheumatoid arthritis. Drug Deliv. 2023;30(1):2173332.
146. Song J, Yang G, Song Y, Jiang Z, Jiang Y, Luan Y, et al. Neutrophil Hitchhiking Biomimetic Nanozymes Prime Neuroprotective Effects of Ischemic Stroke in a Tailored "Burning the Bridges" Manner. Adv Func Mater. 2024;34:2315275.
147. Kong J, Deng Y, Xu Y, Zhang P, Li L, Huang Y. A two-pronged delivery strategy disrupting positive feedback loop of neutrophil extracellular traps for metastasis suppression. ACS Nano. 2024;18(24):15432–51.
148. Chen J, Hou S, Liang Q, He W, Li R, Wang H, et al. Localized degradation of neutrophil extracellular traps by photoregulated enzyme delivery for cancer immunotherapy and metastasis suppression. ACS Nano. 2022;16(2):2585–97.
149. Yin H, Lu H, Xiong Y, Ye L, Teng C, Cao X, et al. Tumor-associated neutrophil extracellular traps regulating nanocarrier-enhanced inhibition of malignant tumor growth and distant metastasis. ACS Appl Mater Interfaces. 2021;13(50):59683–94.
150. Hao Y, Li X, Liu Y, Liu D, Zhao X, Ji S, et al. Manganese doped nanosystem for degrading neutrophil extracellular traps and improving chemotherapy efficiency to synergistically inhibit lung metastasis of breast cancer. Chem Eng J. 2023;466: 142957.
151. Zhu L, Li Z, Liu N, Sun H, Wang Y, Sun M. Dynamically deformable protein delivery strategy disassembles neutrophil extracellular traps to prevent liver metastasis. Adv Func Mater. 2021;31(42):2105089.
152. Wang Z, Chen C, Shi C, Zhao X, Gao L, Guo F, et al. Cell membrane derived liposomes loaded with DNase I target neutrophil extracellular traps which inhibits colorectal cancer liver metastases. J Control Release. 2023;357:620–9.
153. Cheng Y, Gong Y, Chen X, Zhang Q, Zhang X, He Y, et al. Injectable adhesive hemostatic gel with tumor acidity neutralizer and neutrophil extracellular traps lyase for enhancing adoptive NK cell therapy prevents post-resection recurrence of hepatocellular carcinoma. Biomaterials. 2022;284: 121506.
154. Zhou H, Zhu C, Zhao Q, Ni J, Zhang H, Yang G, et al. Wrecking neutrophil extracellular traps and antagonizing cancer-associated neurotransmitters by interpenetrating network hydrogels prevent postsurgical cancer relapse and metastases. Bioact Mater. 2024;39:14–24.
155. Okeke EB, Louttit C, Fry C, Najafabadi AH, Han K, Nemzek J, et al. Inhibition of neutrophil elastase prevents neutrophil extracellular trap formation and rescues mice from endotoxic shock. Biomaterials. 2020;238: 119836.
156. Liu M, Liu S, Liu L, Xiu J, Zhang T, Chen D, et al. Nanoparticle-inhibited neutrophil elastase prevents neutrophil extracellular trap and alleviates rheumatoid arthritis in C57BL/6 mice. Nano Today. 2023;50: 101880.
157. Cruz MA, Bohinc D, Andraska EA, Alvikas J, Raghunathan S, Masters NA, et al. Nanomedicine platform for targeting activated neutrophils and neutrophil-platelet complexes using an α(1)-antitrypsin-derived peptide motif. Nat Nanotechnol. 2022;17(9):1004–14.
158. Liang H, Du Y, Zhu C, Zhang Z, Liao G, Liu L, et al. Nanoparticulate cationic poly(amino acid)s block cancer metastases by destructing neutrophil extracellular traps. ACS Nano. 2023;17(3):2868–80.
159. Tu Z, Zhu Y, Gao W, Liu M, Wei Y, Xu C, et al. Tackling severe neutrophilic inflammation in airway disorders with functionalized nanosheets. ACS Nano. 2024;18(9):7084–97.
160. Wang J, Wang Y, Li J, Ying J, Mu Y, Zhang X, et al. Neutrophil extracellular traps-inhibiting and fouling-resistant polysulfoxides potently prevent postoperative adhesion, tumor recurrence, and metastasis. Adv Mater. 2024;36(31): e2400894.
161. Li Z, Li L, Yue M, Peng Q, Pu X, Zhou Y. Tracing immunological interaction in trimethylamine N-oxide hydrogel-derived zwitterionic microenvironment during promoted diabetic wound regeneration. Adv Mater. 2024;36: e2402738.
162. Liu S, Liu M, Xiu J, Zhang T, Zhang B, Cun D, et al. Celastrol-loaded bovine serum albumin nanoparticles target inflamed neutrophils for improved rheumatoid arthritis therapy. Acta Biomater. 2024;174:345–57.
163. Di L, Thomas A, Switala L, Kalikasingh K, Lapping S, Nayak L, et al. Surface geometry of cargo-less gold nanoparticles is a driving force for selective targeting of activated neutrophils to reduce thrombosis in antiphospholipid syndrome. Nano Lett. 2023;23(21):9690–6.
164. Mei J, Zhou J, Kong L, Dai Y, Zhang X, Song W, et al. An injectable photo-cross-linking silk hydrogel system augments diabetic wound healing in orthopaedic surgery through spatiotemporal immunomodulation. J Nanobiotechnol. 2022;20(1):232.
165. Cai J, Tao H, Liu H, Hu Y, Han S, Pu W, et al. Intrinsically bioactive and biomimetic nanoparticle-derived therapies alleviate asthma by regulating multiple pathological cells. Bioact Mater. 2023;28:12–26.
166. Li L, Ma Y, Hu Y, Wang P, Han S, Zhang X, et al. Site-specific inhibition of neutrophilic inflammation by low-dose nanotherapy for immunoregulatory treatment of asthma. Nano Today. 2023;52: 101957.
167. Yin N, Wang W, Pei F, Zhao Y, Liu C, Guo M, et al. A neutrophil hijacking nanoplatform reprograming NETosis for targeted microglia polarizing mediated ischemic stroke treatment. Adv Sci (Weinh). 2024;11(17): e2305877.

168. Liu W, Lu H, Rao X, Li X, Lu H, Li F, et al. Enhanced treatment for cerebral ischemia-reperfusion injury of puerarin loading liposomes through neutrophils-mediated targeted delivery. Nano Res. 2021;14(12):4634–43.
169. Yang L, Liu Q, Zhang X, Liu X, Zhou B, Chen J, et al. DNA of neutrophil extracellular traps promotes cancer metastasis via CCDC25. Nature. 2020;583(7814):133–8.
170. Liu X, Li T, Chen H, Yuan L, Ao H. Role and intervention of PAD4 in NETs in acute respiratory distress syndrome. Respir Res. 2024;25(1):63.
171. Singh J, Boettcher M, Dölling M, Heuer A, Hohberger B, Leppkes M, et al. Moonlighting chromatin: when DNA escapes nuclear control. Cell Death Differ. 2023;30(4):861–75.
172. Wang CL, Wang Y, Jiang QL, Zeng Y, Yao QP, Liu X, et al. DNase I and sivelestat ameliorate experimental hindlimb ischemia-reperfusion injury by eliminating neutrophil extracellular traps. J Inflamm Res. 2023;16:707–21.
173. Hallberg LAE, Barlous K, Hawkins CL. Antioxidant strategies to modulate NETosis and the release of neutrophil extracellular traps during chronic inflammation. Antioxidants (Basel). 2023;12(2):478.
174. Adelnia H, Blakey I, Little PJ, Ta HT. Poly(succinimide) nanoparticles as reservoirs for spontaneous and sustained synthesis of poly(aspartic acid) under physiological conditions: potential for vascular calcification therapy and oral drug delivery. J Mater Chem B. 2023;11(12):2650–62.
175. Datla US, Vundurthy B, Hook JS, Menon N, Razmi Bagtash H, Shihabeddin T, et al. Quantifying neutrophil extracellular trap release in a combined infection-inflammation NET-array device. Lab Chip. 2024;24(3):615–28.
176. Wang S, Hou Y. New types of magnetic nanoparticles for stimuli-responsive theranostic nanoplatforms. Adv Sci (Weinh). 2024;11(8): e2305459.
177. Najahi-Missaoui W, Arnold RD, Cummings BS. Safe nanoparticles: are we there yet? Int J Mol Sci. 2020;22(1):385.
178. Kumar M, Kulkarni P, Liu S, Chemuturi N, Shah DK. Nanoparticle biodistribution coefficients: a quantitative approach for understanding the tissue distribution of nanoparticles. Adv Drug Deliv Rev. 2023;194: 114708.
179. Agha A, Waheed W, Stiharu I, Nerguizian V, Destgeer G, Abu-Nada E, et al. A review on microfluidic-assisted nanoparticle synthesis, and their applications using multiscale simulation methods. Discov Nano. 2023;18(1):18.
180. Li H, Guan M, Zhang NN, Wang Y, Liang T, Wu H, et al. Harnessing nanomedicine for modulating microglial states in the central nervous system disorders: challenges and opportunities. Biomed Pharmacother. 2024;177: 117011.
181. Shrestha B, Tang L, Hood RL. Nanotechnology for personalized medicine. In: Gu N, editor. Nanomedicine. Singapore: Springer Nature Singapore; 2023. p. 555–603.

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