Advances in Cells

Science Publication Inc.

Editorial Open Access

Neutrophil extracellular traps in inflammatory cardiovascular disorders

Elena Butoi *

Inflammation Department, Institute of Cellular Biology and Pathology "Nicolae Simionescu", Bucharest, Romania
* Correspondence: Elena Butoi. email: elena.dragomir@icbp.ro

Received: September 01, 2025; Accepted: September 03, 2025; Published: September 05, 2025

1. Introduction

Neutrophils, key players of the innate immunity system, serve as primary effectors of both acute infection and sterile inflammation. Neutrophil extracellular traps (NETs) are released by neutrophils as part of their antimicrobial defense, helping to trap and eliminate pathogenic microorganisms [1]. However, over the last decade, NETs have emerged as a remarkable example of how the innate immune system shapes cardiovascular disease (CVD) [2]. Initially described as DNA webs that capture pathogens, NETs are now recognized as complex structures enriched in histones, proteases, and enzymes that extend far beyond antimicrobial defense [1]. In cardiovascular pathologies, NETs function as powerful amplifiers of vascular injury, thrombosis, and maladaptive remodeling, while also actively participating in inflammation processes associated with myocardial infarction, atrial fibrillation, and myocarditis. The increased understanding of NETs' roles in CVD raises both excitement and concern: NETs represent not only a novel mechanistic link between inflammation and cardiovascular pathology but also a potential therapeutic target whose modulation could reshape clinical outcomes.

2. NETs composition and formation

NETs are complex web-like DNA structures released by neutrophils in response to a wide range of stimuli. The expelled DNA, of either nuclear or mitochondrial origin, is decorated with nuclear material – citrullinated and hyperacetylated histones, and coated with antimicrobial granular proteins such as neutrophil elastase (NE), myeloperoxidase (MPO), and cathepsin G, as well as cytosolic components including cytokines and S100 proteins. Importantly, the composition and architecture of NETs vary depending on the stimulus and microenvironment.

Two main mechanisms of NET release have been described. The predominant pathway in inflammatory conditions is ***suicidal NETosis***, in which neutrophils undergo cell death following a sequence of morphological changes: nuclear envelope breakdown, chromatin decondensation, plasma membrane disruption, and eventual extrusion of DNA-based NETs into the extracellular space [1, 3]. The process of suicidal NETosis requires profound chromatin remodeling. A central step involves histone citrullination by peptidyl arginine deiminase 4 (PAD4), which neutralizes histone charge and weakens DNA–histone interaction. In parallel, NE cleaves histones, further promoting chromatin decondensation. Ultimately, the destabilized chromatin is expelled through rupture of the nuclear envelope, giving rise to extracellular DNA webs densely loaded with neutrophil effector proteins. ***Vital NETosis*** allows neutrophils to remain viable while releasing portions of nuclear or mitochondrial DNA, depending on the stimulus [4, 5]. This process sustains antimicrobial defense without immediate cell loss. The source of DNA and the local microenvironment influence NET composition, introducing the concept of NET heterogeneity. While the NET proteome appears relatively stable, the abundance of its protein constituents can vary with the stimulus [6]. Notably, when NETs arise from mitochondrial DNA, their composition and function differ, as mitochondria lack histones [7].

3. NETs as drivers of cardiovascular pathologies

Recent evidence highlights the essential role of NETs in cardiovascular disease. In acute coronary syndromes, NETs promote disease progression through multiple mechanisms, including serving as autoantigens, interacting with diverse cell types, activating inflammasomes, and accelerating atherosclerosis [2, 8, 9]. Atherosclerotic plaques provide potent triggers for NET formation, as components such as cholesterol crystals, activated platelets, and dysfunctional endothelial cells stimulate neutrophils to release NETs [8]. NETs have been detected at the luminal side of plaques, within the thrombus, and at the plaque–thrombus interface [2]. Within plaques, NETs contribute to endothelial dysfunction, lipid oxidation, and recruitment of inflammatory cells. Their proteolytic and pro-oxidative

components destabilize the fibrous cap, increasing susceptibility to rupture. Following plaque disruption, NETs catalyse atherothrombosis by activating platelets, enhancing thrombin generation, and providing a scaffold for clot formation. NETs have been detected in thrombotic lesions from human samples and experimental models, highlighting their central role in pathological clot formation [10-12]. Acting at the interface of neutrophils, platelets, and coagulation pathways, often termed immunothrombosis, NETs provide both structural and biochemical support for thrombus development [10]. Their negatively charged DNA creates a procoagulant surface, while NET-derived proteins directly activate platelets and enhance both intrinsic and extrinsic coagulation cascades. The web-like scaffold of NETs facilitates the deposition of platelets, erythrocytes, fibrin, and von Willebrand factor, thereby promoting thrombus stability. Additionally, NET-associated histones potentiate platelet aggregation and thrombin generation through interactions with fibrinogen and TLR2/TLR4, further amplifying thrombosis and impairing fibrinolysis [12]. NETs are consistently detected in human thrombi and appear particularly abundant in fresh, lytic coronary thrombi (10–30% of thrombus mass), supporting their predominant role in the early phases of atherothrombosis [11]. In coronary artery thrombi from ST-segment elevation myocardial infarction (STEMI) patients, NETs cluster around erythrocytes and associate with extracellular iron and erythrocyte fragments, indicating that NETs contribute to erythrocyte aggregation, damage, and amplification of thrombosis during plaque-related coronary events. Beyond their contribution to thrombus formation, NETs are implicated in acute coronary syndrome (ACS)–related injury. Activated neutrophils infiltrating necrotic myocardium release NETs immediately after MI, thereby exacerbating tissue damage by recruiting additional leukocytes and delaying the resolution of inflammation. We and others found that plasma NET levels are increased in patients with ACS [13, 14], and some markers of NETs have been found in acute myocardial infarction [15]. In STEMI patients, these markers were further shown to correlate with infarct size and impaired left ventricular function [16]. The burden of coronary NETs in patients with STEMI has been shown to predict ST-segment resolution and infarct size [14], although the association between NETs and major cardiovascular events is not clear. However, NETs from myocardial tissue were recently found to contribute to cardiac dysfunction and adverse outcomes in patients with heart failure with dilated cardiomyopathy, potentially through mitochondrial dysfunction of cardiomyocytes [17]. The involvement of NETs in cardiac remodeling and heart failure is also supported by studies demonstrating that NET-associated proteins can sustain fibroblast activation, chronic inflammation, and adverse ventricular remodeling [9, 18]. In animal models, neutrophil infiltration and NETosis markers were consistently detected in failing hearts [9].

4. Therapeutic strategies targeting NETs

Multiple experimental and clinical evidence identifying NETs as key contributors to cardiovascular pathology - accelerating atherosclerosis and ACS - places them as both biomarkers of cardiovascular outcomes and promising therapeutic targets. In this context, several studies have explored genetic and pharmacological strategies to inhibit NET formation or promote their clearance in CVD. Inhibition of NET formation using PAD4 inhibitors or DNase treatment reduced cardiac fibrosis and improved cardiac function in animal models of heart failure [9]. The studies using PAD4 knockout models showed that mice lacking PAD4 had significantly smaller infarcts and better preserved left ventricular function following ischemia-reperfusion injury [19], or are protected from plaque erosion [20]. Administration of PAD4-specific inhibitors such as GSK484, JBI-589, or Cl-amidine prevented the formation of NETs in plaques, reduced the number of endothelial macrophages, decreased neutrophil recruitment to the vessel wall, and reduced levels of inflammatory mediators, ultimately significantly reducing atherosclerotic plaque formation and thrombosis and decreasing the risk of myocardial infarction [20-23].

Moreover, DNase I treatment decreased the number of NETs and of inflammatory cells, and was effective in reducing the size of atherosclerotic plaques in mice [24]. In addition, administration of DNase I in a PAD4 knockout mouse model resulted in survival of endothelial cells while limiting the recruitment of neutrophils [20]. However, it remains unclear whether degradation of NETs by DNase I sufficiently neutralizes histones with procoagulant activity or instead facilitates their release, potentially increasing thrombotic risk [25]. Further investigation is required to clarify these effects.

5. Conclusion: A double-edged sword

Targeting NETs in cardiovascular disease offers substantial translational potential, as both preclinical and early clinical studies indicate that limiting NET formation or promoting their clearance can reduce thrombosis, myocardial injury, and maladaptive remodeling. However, NETs are not universally harmful; they serve as an essential arm of innate immunity, capturing and neutralizing pathogens. Broad or prolonged suppression of NET-osis could therefore weaken host defense and increase infection risk. The central challenge is to design strategies that selectively attenuate the pathological contributions of NETs to vascular inflammation, thrombosis, and tissue remodeling, while preserving their protective antimicrobial functions - a balance crucial for safe and effective clinical translation.

Acknowledgment

The author gratefully acknowledges the financial support through the European Union – NextGenerationEU and the Romanian Government through the National Recovery and Resilience Plan, Component 9 - Investment 8, HeartCure project, Financing Contract no.

760063/23.05.2023, CF93/15.11.2022, and by the Romanian Academy.

Author's Contributions

The author conducted all the research work for this study.

Competing Interests

The authors have declared that no competing interests exist.

References

1. Hidalgo A, Libby P, Soehnlein O, Aramburu IV, Papayannopoulos V, Silvestre-Roig C. Neutrophil extracellular traps: from physiology to pathology. Cardiovascular Research. 2022;118(13):2737-2753.
2. Döring Y, Libby P, Soehnlein O. Neutrophil extracellular traps participate in cardiovascular diseases: recent experimental and clinical insights. Circulation research. 2020;126(9):1228-1241.
3. Fuchs TA, Abed U, Goosmann C, Hurwitz R, Schulze I, Wahn V, et al. Novel cell death program leads to neutrophil extracellular traps. The Journal of cell biology. 2007;176(2):231-241.
4. 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. Nature medicine. 2012;18(9):1386-1393.
5. Yousefi S, Mihalache C, Kozlowski E, Schmid I, Simon H. Viable neutrophils release mitochondrial DNA to form neutrophil extracellular traps. Cell Death & Differentiation. 2009;16(11):1438-1444.
6. Wang X, Zhao J, Cai C, Tang X, Fu L, Zhang A, et al. A label-free quantitative proteomic analysis of mouse neutrophil extracellular trap formation induced by Streptococcus suis or phorbol myristate acetate (PMA). Frontiers in Immunology. 2018;9:2615.
7. Lood C, Blanco LP, Purmalek MM, Carmona-Rivera C, De Ravin SS, Smith CK, et al. Neutrophil extracellular traps enriched in oxidized mitochondrial DNA are interferogenic and contribute to lupus-like disease. Nature medicine. 2016;22(2):146-153.
8. Ji Y-M, Li T, Qin Y-H, Xiao S-Y, Lv Y-H, Dong Y, et al. Neutrophil Extracellular Traps (NETs) in Sterile Inflammatory Diseases. Journal of Inflammation Research. 2025:7989-8004.
9. Morrissey SM, Kirkland LG, Phillips TK, Levit RD, Hopke A, Jensen BC. Multifaceted roles of neutrophils in cardiac disease. Journal of Leukocyte Biology. 2025;117(4):qiaf017.
10. Kimball AS, Obi AT, Diaz JA, Henke PK. The emerging role of NETs in venous thrombosis and immunothrombosis. Frontiers in Immunology. 2016;7:236.
11. Laridan E, Martinod K, De Meyer SF, editors. Neutrophil extracellular traps in arterial and venous thrombosis. Seminars in thrombosis and hemostasis; 2019: Thieme Medical Publishers.
12. Wang Y, Luo L, Braun OÖ, Westman J, Madhi R, Herwald H, et al. Neutrophil extracellular trap-microparticle complexes enhance thrombin generation via the intrinsic pathway of coagulation in mice. Scientific reports. 2018;8(1):4020.
13. Barbu E, Mihaila AC, Gan A-M, Ciortan L, Macarie RD, Tucureanu MM, et al. The elevated inflammatory status of neutrophils is related to in-hospital complications in patients with acute coronary syndrome and has important prognosis value for diabetic patients. International Journal of Molecular Sciences. 2024;25(10):5107.
14. Ferré-Vallverdú M, Latorre AM, Fuset MP, Sánchez E, Madrid I, Ten F, et al. Neutrophil extracellular traps (NETs) in patients with STEMI. Association with percutaneous coronary intervention and antithrombotic treatments. Thrombosis Research. 2022;213:78-83.
15. Stakos DA, Kambas K, Konstantinidis T, Mitroulis I, Apostolidou E, Arelaki S, et al. Expression of functional tissue factor by neutrophil extracellular traps in culprit artery of acute myocardial infarction. European heart journal. 2015;36(22):1405-1414.
16. Mangold A, Alias S, Scherz T, Hofbauer TM, Jakowitsch J, Panzenböck A, et al. Coronary neutrophil extracellular trap burden and deoxyribonuclease activity in ST-elevation acute coronary syndrome are predictors of ST-segment resolution and infarct size. Circulation research. 2015;116(7):1182-1192.
17. Ichimura S, Misaka T, Ogawara R, Tomita Y, Anzai F, Sato Y, et al. Neutrophil extracellular traps in myocardial tissue drive cardiac dysfunction and adverse outcomes in patients with heart failure with dilated cardiomyopathy. Circulation: Heart Failure. 2024;17(6):e011057.
18. Chen W, Frangogiannis NG. Fibroblasts in post-infarction inflammation and cardiac repair. Biochimica et Biophysica Acta (BBA)-Molecular Cell Research. 2013;1833(4):945-953.
19. Savchenko AS, Borissoff JI, Martinod K, De Meyer SF, Gallant M, Erpenbeck L, et al. VWF-mediated leukocyte recruitment with chromatin decondensation by PAD4 increases myocardial ischemia/reperfusion injury in mice. Blood, The Journal of the American Society of Hematology. 2014;123(1):141-148.
20. Franck G, Mawson TL, Folco EJ, Molinaro R, Ruvkun V, Engelbertsen D, et al. Roles of PAD4 and NETosis in experimental atherosclerosis and arterial injury: implications for superficial erosion. Circulation research. 2018;123(1):33-42.
21. Knight JS, Luo W, O'Dell AA, Yalavarthi S, Zhao W, Subramanian V, et al. Peptidylarginine deiminase inhibition reduces vascular damage and modulates innate immune responses in murine models of atherosclerosis. Circulation research. 2014;114(6):947-956.
22. Molinaro R, Yu M, Sausen G, Bichsel CA, Corbo C, Folco EJ, et al. Targeted delivery of protein arginine deiminase-4 inhibitors to limit arterial intimal NETosis and preserve endothelial integrity. Cardiovascular Research. 2021;117(13):2652-2663.
23. Novotny J, Chandraratne S, Weinberger T, Philippi V, Stark K, Ehrlich A, et al. Histological comparison of arterial thrombi in mice and men and the influence of Cl-

amidine on thrombus formation. PloS one. 2018;13(1):e0190728.
24. Liu Y, Carmona-Rivera C, Moore E, Seto NL, Knight JS, Pryor M, et al. Myeloid-specific deletion of peptidylarginine deiminase 4 mitigates atherosclerosis. Frontiers in Immunology. 2018;9:1680.
25. Zalghout S, Martinod K. Therapeutic potential of DNases in immunothrombosis: promising succor or uncertain future? Journal of Thrombosis and Haemostasis. 2025;23(3):760-778.