https://doi.org/10.31925/farmacia.2025.1.3 **REVIEW**

NATRIURETIC PEPTIDES – A VALUABLE TARGET IN THE TREATMENT OF HEART FAILURE

NICOLAE PĂUN [1,2], CAMELIA NICOLAE [1,2]*, MIHAI GRIGORE [1,2], ADRIANA MIHAELA ILIESIU [1,2], VIOREL JINGA [1,2], IOAN TIBERIU NANEA [1,2], DUMITRU LUPULIASA [1], ANDREEA MADALINA MUNTEANU [1,2], GABRIELA USCOIU [1,2], MIRUNA MARIA APETROAEI [1]

[1] *"Carol Davila" University of Medicine and Pharmacy, Bucharest, Romania*
[2] *"Prof. Dr. Th. Burghele" Hospital, Bucharest, Romania*

**corresponding author: dr_camelia_nicolae@yahoo.com*

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Abstract

Heart failure (HF) continues to pose a significant worldwide health concern, marked by elevated morbidity, mortality, and economic burden. The natriuretic peptide (NP) system is particularly important in mitigating maladaptive neurohormonal activation, such as the renin-angiotensin-aldosterone system (RAAS) and the sympathetic nervous system (SNS). NPs demonstrate a variety of beneficial effects, including diuresis, natriuresis, vasodilation, and the suppression of heart hypertrophy and fibrosis. Although the NP system deteriorates in late HF, treatment approaches aimed at NPs have shown encouraging outcomes in re-establishing cardiovascular and renal homeostasis. This article examines the pathophysiological functions of NPs, the relevant signalling mechanisms, and therapeutic potential. Novel pharmacological drugs and gene therapy techniques are also discussed. Despite ongoing problems such as enzymatic degradation, reduced bioavailability and adverse reactions of new molecules, the NP system constitutes an invaluable therapeutic target. Future improvements in this field promise to enhance outcomes in HF management.

Rezumat

Insuficiența cardiacă (IC) continuă să reprezinte o problemă de sănătate publică la nivel mondial, caracterizată de o rată semnificativă de morbiditate și mortalitate. Sistemul peptidelor natriuretice (NP) este deosebit de important în atenuarea activării neurohormonale maladaptative în cazul acestei patologii. NP prezintă o multitudine de efecte biologice benefice, inclusiv diureza, natriureza, vasodilatația și suprimarea hipertrofiei cardiace și a fibrozei. Deși sistemul NP se deteriorează progresiv în IC avansată, abordările terapeutice care intervin asupra componentelor din sistemul NP au arătat rezultate promițătoare în restabilirea homeostaziei cardiovasculare și renale. Acest studiu examinează funcțiile fiziopatologice ale NP, mecanismele de semnalizare celulară relevante și potențialul terapeutic. De asemenea, sunt analizate terapiile farmacologice aprobate și noile molecule care se află în studiu. În ciuda problemelor actuale, precum degradarea enzimatică, biodisponibilitatea redusă și reacțiile adverse ale noilor molecule, sistemul NP constituie o țintă terapeutică promițătoare pentru îmbunătățirea rezultatelor terapeutice în IC

Keywords: natriuretic peptides, heart failure biomarkers, cardiovascular homeostasis, natriuretic peptides therapy

Introduction

The World Health Organisation reports that cardiovascular diseases are the leading global cause of mortality, accounting for 20.5 million deaths annually [1]. A third of all cases of coronary heart disease, cerebrovascular accidents, and rheumatic heart disease occur in people younger than 70 years old, and these conditions are mostly caused by cardiac events and strokes. Tobacco usage, alcohol abuse, sedentary lifestyles, and poor dietary habits are the main risk factors [2].

A complex and health-threatening syndrome, heart failure (HF), has been defined by considerable mortality and morbidity, low level of functioning and quality of life, along with substantial costs [3]. HF impacts around 64 million individuals globally. Consequently, efforts to mitigate its social and economic impact have emerged as a significant global public health issue. Although the incidence of HF has remained stable and appears to be decreasing in industrialised nations, its prevalence is rising due to population ageing, enhanced treatment and survival rates associated with ischemic heart disease, and the accessibility of efficient, scientifically proven therapies that extend the lifespan of HF patients [4]. The concerning inverse trend in the prevalence of various HF phenotypes may stem from a rise in conventional cardiovascular risk factors, such as high blood pressure, overweight or obese abdomens, lipid disorders, and diabetes mellitus, among relatively young persons [5]. Nonetheless, a significant proportion of patients

develop cardiac problems despite the absence of these risk factors. It is important to mention that telomere length may serve as an effective biomarker for diagnosing and monitoring the course of HF. Telomere instability and shortening are causal factors in the onset and progression of cardiovascular illnesses. Telomere shortening induces cellular senescence, subsequently leading to apoptosis, and has been recognised as a biomarker in HF development [6]. Telomere length functions as a reliable measure of the cumulative effects of both inflammation and oxidative stress experienced throughout life [7]. Therefore, systolic and diastolic HF are caused by the myocardium's ageing process, which includes telomere shortening and the buildup of senescent cells, which reduces the tissue's regenerative capacity [8].

A recent systematic analysis determined that the approximate incidence of preliminary stages of HF (stages A and B) was as high as 43% among high-risk individuals with hypertension and in men. The 7-year risk of developing symptomatic HF (stages C and D) and overall mortality were as high as 9.8% and 5.4%, respectively [9]. Patients admitted for HF have diverse phenotypes of cardiac anatomy and function, with left ventricular ejection fractions ranging from normal to significantly reduced. Notably, a reduced ejection fraction correlates with poorer outcomes, with hazards converging at a left ventricular ejection fraction of 45 - 50%. Likewise, right ventricular dysfunction correlates with elevated death rates. Left atrial enlargement and mitral regurgitation possess predictive value as well. In individuals with HF, minor or moderate functional mitral valve regurgitation is observed in 49%, whereas severe cases are present in 24%, and it is recognised to improve following treatment for acute decompensation [10]. Timely detection and treatment are thought to be essential to decelerate or potentially prevent the advancement of HF from asymptomatic to symptomatic phases. HF management requires in-depth knowledge of screening, risk assessment, diagnosis computational methods, and individualised point-of-care techniques based on established, evidence-based models [11].

Natriuretic peptides (NPs) are recognised as circulating cardiac biomarkers indicative of myocardial distension and biomechanical strain, primarily encompassing various molecules, including brain natriuretic peptide (BNP), N-terminal brain natriuretic pro-peptide (NT-proBNP), and mid-regional atrial natriuretic pro-peptide (MR-proANP), in addition to other categories of NPs, such as atrial natriuretic peptide (ANP) and C-type NP (CNP) [12]. The stretching of the heart, heightened intracardiac filling pressures, augmented intracardiac quantities, and excessive fluid intake are regarded as the primary determinants for the production and release of NPs [13]. Nonetheless, additional factors such as systemic inflammatory response, cardiac ischemia and necrosis, hypoxia, cerebral trauma, infections, and dysfunctional adipose tissues are regarded as contributing to the generation, release, elimination, and bioavailability of NPs. Alternative biomarkers, like galectin-3 and soluble suppression of tumorigenicity 2 (sST2), enhance prognostic insights alongside NPs, particularly in individuals at elevated risk of heart failure. The multiple-biomarker strategy has been evaluated; nevertheless, its economic burden and repeatability for practical application remain uncertain [14].

This article examines the valuable role of NPs in the pathogenesis and treatment of HF, emphasising their therapeutic potential as pharmaceutical targets. Additionally, we assess existing and novel therapeutic approaches to address the unmet requirements in HF treatment and enhance patient outcomes.

Natriuretic peptide system: an overview

Based on established conventional perspectives, NPs are physiological antagonists to the sympathetic nervous system (SNS) and the renin-angiotensin-aldosterone systems (RAAS). Their primary biological functions encompass diuresis, maintenance of electrolyte balance, fluid retention, blood pressure regulation, and vasodilation facilitation [15].

ANP is predominantly produced in the atria under normal conditions as a peptide (pre-prohormone) with 151 amino acids. It is subsequently stored in specialised cytoplasmic granules as a pro-hormone comprising cardiodilatin (N-terminus) and ANP (C-terminus) within its structure [16]. Corin protease, a specialised transmembrane proteolytic enzyme, cleaves ANP from its pro-hormone, releasing it into the bloodstream as an active peptide with 28 amino acids [17]. BNP (brain natriuretic peptide) is also produced as a pre-prohormone with 134 amino acids. Upon cleavage by either the corin protease or the furin endoprotease, it is retained in particular granules in a physiologically active form comprising 32 amino acids and the inert N-terminal form comprising 76 amino acids. Under physiological settings, the primary source of ANP and BNP is the atrial myocardium. When the myocardium experiences parietal stress, the ventricle becomes the primary site of their secretion [18]. CNP is synthesised in the brain, endothelial cells, and chondrocytes as pro-CNP, including 103 amino acids. This is subsequently broken by the furin protease, yielding the active form consisting of 53 amino acids, which is found in the bloodstream [19]. Urodilatin is an isoform of proANP produced in the kidneys (distal tubules) in reaction to elevated mean arterial pressure and increased blood volume. It comprises the identical 95-126 amino acids of proANP, along with the same structural ring of 17 amino acids featuring a COOH-terminus. It is subsequently transformed into the active form, including 32 amino acids with the NH_2-

terminal sequence, using a pathway distinct from ANP [16]. Urodilatin is absent in plasma; it exerts a paracrine effect at the renal level, targeting receptors in the glomeruli and collecting tubules. Consequently, the kidneys generate their own natriuretic peptide [20]. Dendroapsis NP (DNP) is the most recent NP identified. It is a 38-amino acid residue that is physically and functionally analogous to ANP and BNP; it possesses the identical 17-amino acid ring structure, although the COOH and NH2 terminal sequences are located in distinct places [21]. DNP is prevalent in numerous animal species, and although its presence in humans remains contentious (the exact gene has yet to be found), it has been immunohistochemically detected in plasma and atrial myocardium. Various forms of BNP, proBNP, and NT-proBNP were also detected in the blood, exhibiting distinct molecular weights and functions. The existence of inactive forms may elucidate the resistance to the effects of endogenous BNP, which is observed in significantly elevated amounts in the blood of patients with HF [15]. Figure 1 summarises the cellular mechanisms of action and clearance of NPs.

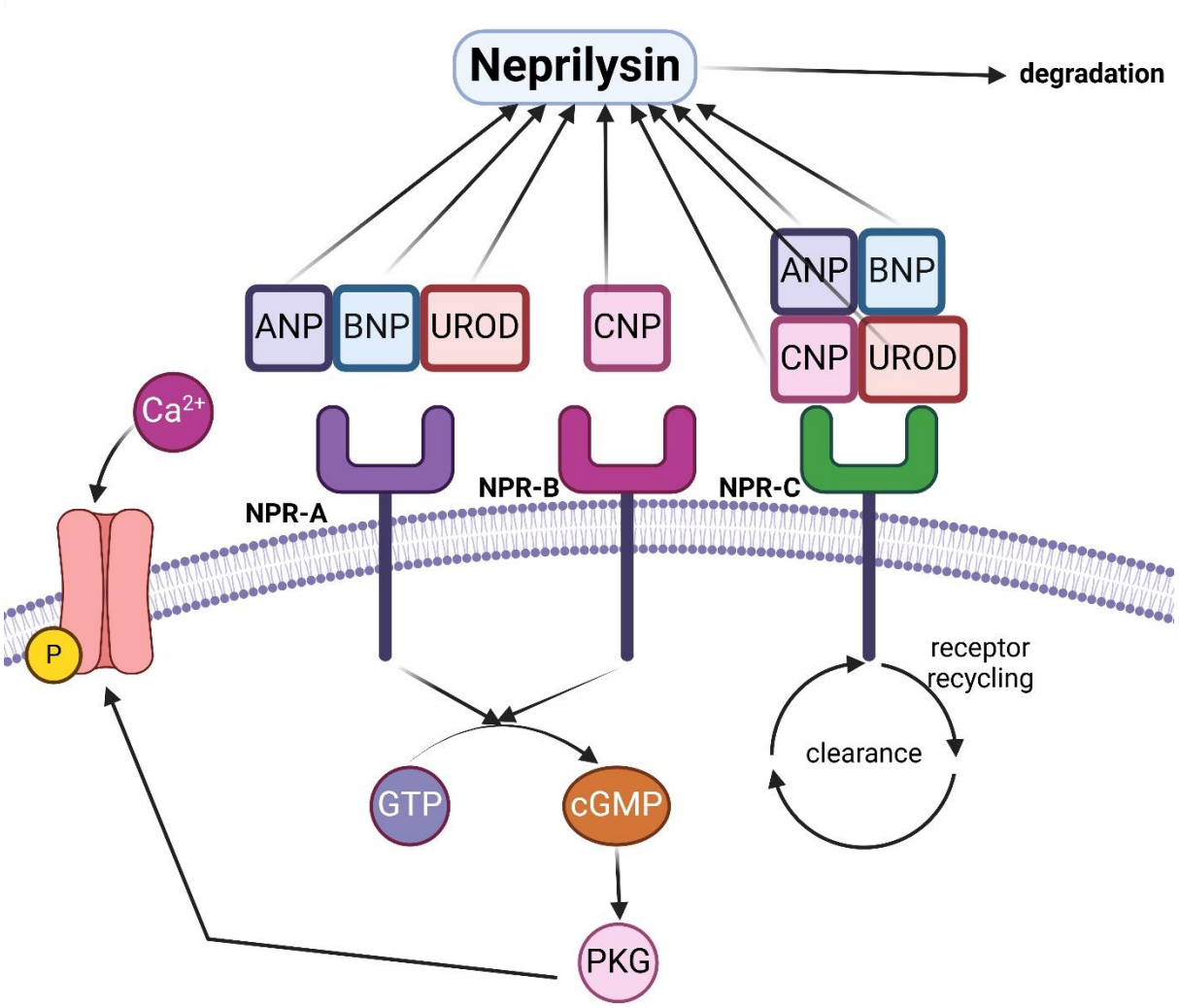

Figure 1.
The cellular mechanisms of action and clearance of NPs (created with BioRender.com)

ANP stimulates the particulate guanylyl cyclase receptor A, sometimes referred to as NP receptor A (NPR-A), leading to the synthesis of a second messenger, cyclic guanosine monophosphate (cGMP), which facilitates the various favourable physiologic effects of ANP [22, 23]. BNP possesses effects that are similarly mediated *via* the NRP-A/cGMP pathway. ANP and BNP, through NPR-A and cGMP, produce clinically significant biological effects, including natriuresis, vasodilation, inhibition of the RAAS, reducing cardiovascular myocyte enlargement and cell death, promoting vascular renewal, and preventing organ fibrosis [23, 24]. Recent studies have demonstrated beneficial metabolic effects of NPR-A/cGMP activation, encompassing lipolysis, the browning of white adipocytes, adiponectin release, and insulin production and sensitivity control, enhancing glucose absorption and promoting HDL synthesis [14, 23]. CNP serves

as the endogenous ligand for the particulate guanylyl cyclase receptor B, often referred to as NPR-B, and its activation produces cGMP as a secondary messenger. Although both NPR-A and NPR-B activation leads to cGMP synthesis, the formation of cGMP may occur in different cellular compartments, triggering distinctive effects of ANP and CNP, respectively [25]. The CNP/NPR-B/cGMP pathway possesses beneficial anti-remodelling characteristics, such as the inhibition of fibrosis and smooth muscle cell proliferation, along with the facilitation of angiogenesis. Furthermore, this route exhibits anti-inflammatory properties as well as vasodilatory effects on venous and microcirculatory systems [23]. Figure 2 illustrates the physiological effects of NP at different tissue levels.

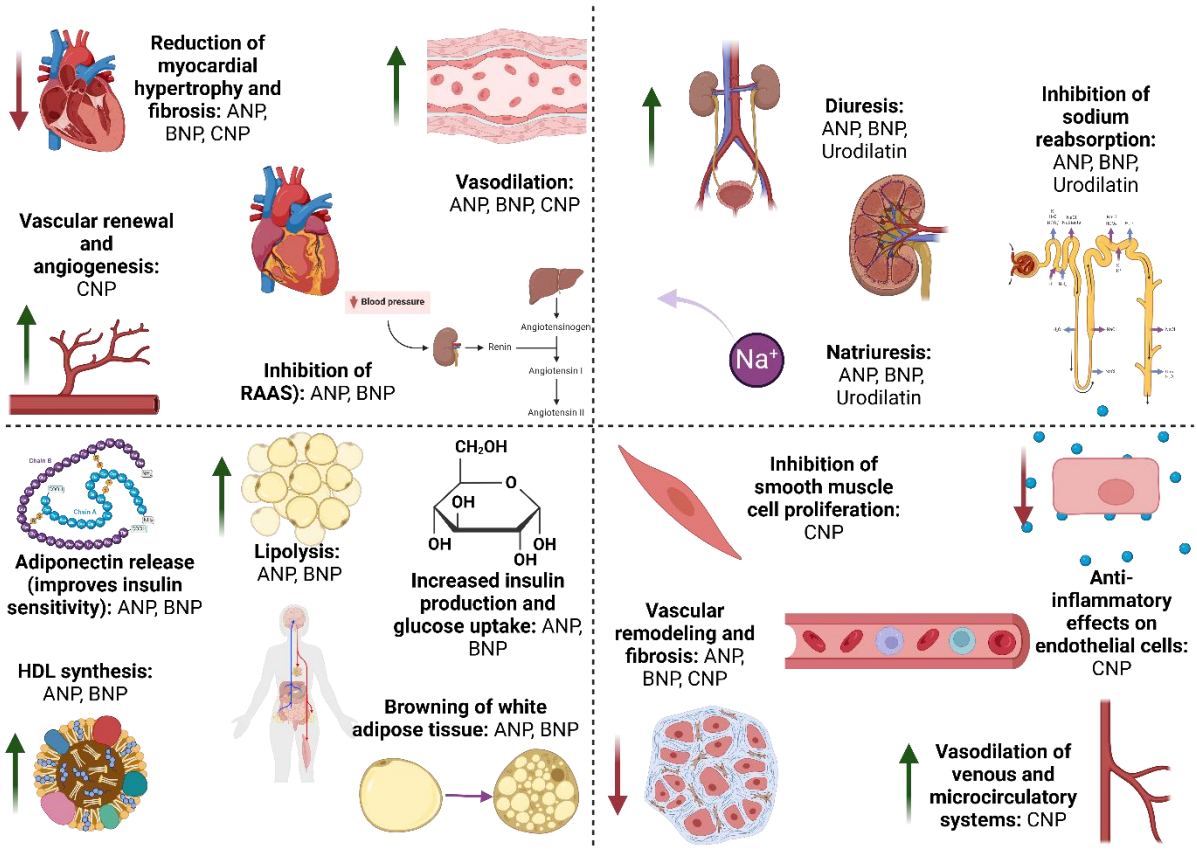

Figure 2.
NP physiological actions (created with BioRender.com)

Pathophysiology of heart failure and the role of natriuretic peptides

The pathophysiology of HF is marked by the early activation of many neurohormonal systems, specifically the SNS and RAAS, in the context of undiagnosed left ventricular systolic dysfunction. In the initial phases of HF, the SNS and the RAAS function compensatory to enhance cardiac output and promote peripheral vasoconstriction, thereby aiming to preserve circulatory homeostasis [26]. Nonetheless, the sustained stimulation of both systems is harmful and exacerbates the progression of heart failure, ultimately resulting in congestion. Alongside the traditional elements of neuroendocrine activation, additional regulatory systems are implicated, including kinins, natriuretic peptides, endothelin, erythropoietin, prostaglandins, and adrenomedullin. If the stimulation of the SNS and RAAS leads to adverse outcomes and detrimental predictive implications, triggering kinins and NP systems may have a beneficial effect [15]. In mild congestive HF, increased concentrations of ANP and BNP are thought to be essential for regulating sodium equilibrium and systemic hemodynamics. As the disease advances, the functional efficacy of the NP system diminishes, hindering the natriuretic, vasodilatory, and hormonal inhibitory actions of NPs, hence exacerbating salt retention and vasoconstriction, which adversely affects cardiac function. Consequently, there is an additional elevation in cardiac NP production, with plasma NP levels rising in direct correlation to the severity of left ventricular failure. The progression to more severe congestive heart failure is marked by the predominance of the overactive RAAS and SNS over the opposing NP system [15, 26, 27]. PreproANP and preproBNP, encoded by *NPPA* and *NPPB*, respectively, are subjected to cleavage and O-glycosylation in atrial cardiomyocytes. Endoproteolytic proteases corin and furin subsequently convert these

precursors into the physiologically relevant hormones ANP and BNP [28]. Furin serves as a cytosolic and membranal enzyme that is involved primarly in the activation of BNP, whereas corin is a membrane-bound serine protease that mainly generates ANP [29]. CNP additionally generates precursors, with proCNP undergoing further cleavage by furin. Under normal settings, ANP and BNP are predominantly synthesised and released by the atria. Under pathological situations, such as mechanical tension, there is an enhanced generation and release of ANP and BNP by ventricular cardiomyocytes. Besides mechanical stretching, several neurohormonal variables might induce the release of ANP and BNP. Alongside circulating BNP and N-terminal prohormone of brain natriuretic peptide, malfunctioning hearts also release proBNP and O-glycosylated proBNP, with the extent of glycosylation corresponding to the severity of heart failure [28].

HF is, in fact, a condition characterised by a deficiency of normal physiological function of the heart. The circulating peptides are predominantly unprocessed and inactive. Furthermore, the NPR-A receptor is desensitised in patients with HF. NP deficit is associated with many cardiometabolic disorders, for example, diminished levels of ANP and BNP in specific patient populations with hypertension and obesity [15].

The clearance of NP from circulation occurs through two mechanisms: NPR-C receptor-mediated endocytosis and neprilysin-neutral endopeptidase (NEP)-mediated proteolysis (Figure 1) [30]. In pathological conditions, elevated levels of ANP and BNP render the NPR-C receptor-mediated clearance mechanism inefficient due to saturation. In these instances, the function of NEP becomes significant [31]. Sangaralingham *et al*. posited that enhanced neprilysin-mediated degradation could be a critical mechanism underlying this ANP shortage, given that ANP is more vulnerable than BNP. Certain processes may diminish ANP production in heart failure. Micro-RNA 425 and/or 155, which inhibit ANP gene expression and synthesis, may be raised in HF. Celik *et al*. additionally identified a cis-acting ANP antisense transcript that negatively regulates ANP expression in human cardiomyocytes, which was elevated in the cardiac tissue of patients with severe HF. Consequently, in HF with increased ANP antisense transcript and ANP insufficiency, ANP antisense transcript may serve as a contributory mechanism [23].

Natriuretic peptides signalling pathways

The atrium and ventricle are the primary sites of expression and secretion of ANP and BNP by myocardial cells. Mechanical stress induces an upregulation of both in the dysfunctional heart. Many different signalling pathways and mediators can elevate ANP and BNP expression. Endothelin-1, phenylephrine, angiotensin II, cardiotrophin-1, leukaemia inhibitory factor, and neuregulin-1 are all examples of such mediators. Moreover, there are a number of intracellular signalling pathways that have been linked to the increased expression of ANP and BNP in response to external stimuli: the protein kinase C, mitogen-activated protein kinase, Janus kinase, and calcineurin-nuclear factor of activated T-cells pathways [32]. NP exert their physiological effects in various organs *via* NPR-mediated signalling. Protein kinases (PKs) dependent on cGMP, ion channels that are controlled by cGMP, and other downstream signalling cascades are activated when NP binds to the NPR-A. In cardiac myocytes and cardiac fibroblasts, all three receptors are expressed; however, under physiological conditions, NRP-B does not exhibit any activity in the ventricles [33]. ANP and BNP primarily activate NPR-A, whereas CNP serves as the principal ligand for NPR-B. NPR-C is identified as an elimination receptor that promotes nanoparticle uptake and lysosomal destruction. The longer circulation half-life is due to its low affinity for BNP [16,33]. In spite of the fact that NPR-C does not possess guanylyl cyclase function, it has been demonstrated that it interacts with a G-protein, which results in the inhibition of adenylyl cyclase and the activation of phospholipase C [34]. Atrial and ventricular cardiomyocytes, as well as pacemaker cells are inhibited by L-type calcium currents when CNP-mediated activation is present. NPR-C has also been documented to exhibit a non-NP-mediated action. CNP inhibits osteocrin's binding to NPR-C, hence averting its breakdown and promoting bone formation throughout development. There is an alternate mechanism for the clearance of NP that involves the metalloendopeptidase neprilysin [35].

NPR-A signalling is a critical pathway mediating the NP action and is mainly involved with the modulation of cardiovascular and renal functions. NPR-A is a membrane-bound receptor that functions as a guanylyl cyclase, and its ligands are ANP and BNP. Thus, NPR-A consists of an extracellular domain that is responsible for the binding of the ligand, a membrane-spanning domain and a cytoplasmic domain, which includes the kinase homology region and guanylyl cyclase domain [36]. After the binding of ANP or BNP to the extracellular domain, undergoes a conformational change that activates the intracellular guanylyl cyclase. This results in the conversion of GTP to the second messenger cGMP. Elevated levels of cGMP activate downstream effectors such as Protein Kinase G (PKG) and cGMP-specific phosphodiesterases [37]. Out of these, PKG plays a significant role in eliciting various cellular effects, including vasodilation, natriuresis, diuresis and inhibition of cardiac hypertrophy and fibrosis. Also, cGMP decreases the intracellular calcium levels in

vascular smooth muscle cells and hence decreases vascular resistance. Dysfunction of NPR-A signalling, for instance, decreased receptor numbers or defective cGMP production, is directly implicated in the pathophysiology of HF [38].

The NPR-B signalling is a highly specific pathway mainly mediated by CNP. It is involved in the maintenance of vascular function, bone growth and development, and the regulation of cellular proliferation and differentiation. NPR-B is structurally related to NPR-A and is composed of an extracellular domain that serves as a ligand binding site, a transmembrane region and an intracellular domain which contains a kinase homology domain as well as a guanylyl cyclase catalytic domain [25]. The binding of CNP to the extracellular domain of NPR-B causes a conformational change, which results in the activation of its guanylyl cyclase domain. This activation converts GTP into cGMP. The increased level of cGMP leads to the activation of downstream effectors such as PKG and other cGMP-dependent proteins to elicit various physiological responses [39]. In the vascular system, NPR-B signalling causes vasodilation, which is associated with the relaxation of vascular smooth muscle cells, and it also prevents vascular remodelling by inhibiting the proliferation and migration of vascular smooth muscle cells [15]. In the cartilage and bone, CNP-NPR-B signalling is accountable for endochondral ossification and longitudinal bone growth, and this process is regulated by chondrocyte proliferation and hypertrophy [40]. Dysfunction of NPR-B signalling, where there is a mutation in the *Npr2* gene that codes for NPR-B, results in skeletal growth disorders, including acromesomelic dysplasia, or may cause pathological vascular remodelling [41].

While both NPR-A and NPR-B are guanylyl cyclase-linked receptors, NPR-A has a higher affinity for ANP and BNP and is activated by them. In comparison, NPR-B has a lower affinity for ANP and BNP but a higher affinity for CNP and is therefore activated mainly by CNP. In contrast to NPR-A, NPR-B has a more tissue-restricted expression with lower levels of expression throughout the body but has a significant effect on the areas where it is expressed in [42].

NPR-C pathway, which was previously termed a clearance receptor, has become known to have tissue-specific signalling function in addition to being a clearance receptor. NPR-C differs from NPR-A and NPR-B because it does not have an intracellular guanylyl cyclase domain. Instead, it possesses a very short cytoplasmic domain through which signalling occurs in association with G-proteins. NPR-C binds all three major natriuretic peptides, ANP, BNP, and CNP, but it has a higher affinity for CNP. One of the classical functions of NPR-C is the clearance of natriuretic peptides from the circulation through ligand internalisation and degradation, thus regulating peptide bioavailability and homeostasis [43]. Besides clearance, NPR-C produces direct effects through G-protein coupling that results in the inhibition of adenylyl cyclase and, hence, the reduction of cAMP inside the cell. This signalling cascade has been involved in various physiological functions, including controlling vascular tone, inhibiting the proliferation of vascular smooth muscle cells, and modulating endothelial cell barrier function [25]. NPR-C activation also stimulates PI3K/Akt signalling, thus mediating anti-inflammatory response and cytoprotection from vascular injury. Also, NPR-C signalling enhances vascular permeability and engages in the angiogenesis and lymphangiogenesis processes [44]. However, the most significant function of NPR-C is to modulate the actions of natriuretic peptides by antagonising the effects of NPR-A and NPR-B that mediate their signalling through cGMP [45]. Abnormalities in NPR-C have been associated with hypertension, vascular remodelling, and HF, where the incorrect clearance or signalling of natriuretic peptides may affect the balance of action of clearance of these receptor peptides.

The inhibition of hypertrophic signalling by NPs is a key cardioprotective mechanism mediated primarily through activating the NPR-A receptor and the downstream generation of cGMP. Cardiac hypertrophy is often driven by pro-hypertrophic signalling pathways such as the RAAS, endothelin-1 (ET-1) signalling, and increased activation of calcineurin/nuclear factor of activated T-cells (NFAT) and mitogen-activated protein kinase (MAPK) pathways. Upon binding of ANP or BNP to the NPR-A receptor, intracellular guanylyl cyclase is activated, converting GTP into cGMP. Elevated cGMP levels activate PKG, which, in turn, phosphorylates and inactivates mediators of hypertrophic signalling such as transient receptor potential canonical (TRPC) channels, RhoA/Rho-kinase (ROCK), extracellular signal-regulated kinase (ERK) signalling. On the one hand, PKG inhibits TRPC-mediated calcium influx, which activates the calcineurin/NFAT pathway, thereby reducing nuclear translocation of NFAT and transcription of pro-hypertrophic genes [46]. On the other hand, PKG directly phosphorylates RhoA, leading to its inactivation and reducing cytoskeletal remodelling and the expression of hypertrophic markers [47]. Additionally, PKG interferes with ERK activation, a component of the MAPK pathway, which is often upregulated in response to hypertrophic stimuli such as angiotensin II or endothelin-1 [48].

Moreover, cGMP signalling suppresses the activity of Gαq-protein-coupled receptor (GPCR)- mediated pathways, which contribute to the pathophysiology of cardiac hypertrophy through their activation of phospholipase C, diacylglycerol, and inositol trisphosphate. This results in calcium overload and

pro-hypertrophic gene transcription [49]. In addition, the NPR-A/cGMP/PKG axis modulates the balance between hypertrophic and antihypertrophic transcription factors. For instance, PKG phosphorylates GATA4, a pro-hypertrophic transcription factor, and prevents its DNA binding activity, reducing the expression of hypertrophic genes such as atrial natriuretic factor (ANF) and BNP [50].

Approved pharmacologic interventions via the NP system

Neprilysin (NEP) is a 90 kDa membrane metallopeptidase that is zinc-dependent and possesses glycosylation domains. The highest concentrations of this protein are found in the proximal tubules, and it is also expressed in the cardiac cells, lungs, kidneys, vascular smooth muscle cells, endothelial cells, fibroblasts, neutrophils, adipocytes, testes, and cerebellum [51]. NEP contributes to the degradation of angiotensin II and catalyses the breakdown of various vasodilator peptides, including ANP and BNP, as well as bradykinin, substance P, and adrenomedullin [52]. Therefore, the relatively newly approved drug, sacubitril, inhibits the enzyme NEP, hence decreasing the degradation of circulating NP and extending their bioavailability [53].

While neprilysin inhibition elevates the levels of vasodilating peptides as anticipated, it concurrently raises the concentrations of angiotensin II and endothelin. Consequently, the inhibition of neprilysin alone exhibits minimal, if any, antihypertensive efficacy. In addition to facilitating vasodilation, NP inhibit cardiomyocyte hypertrophy and cardiac fibrosis while promoting angiogenesis. Therefore, the concurrent blockage of neprilysin and the receptor for angiotensin II exhibited synergistic effects in mitigating several pathways of pathological cardiac remodelling while enhancing perfusion and angiogenesis [52,54]. On the basis of a 20% reduction in cardiovascular death or hospitalisation for heart failure that was observed in the PARADIGM-HF trial, the FDA granted approval for the angiotensin receptor/neprilysin inhibitor (ARNI) sacubitril/valsartan for patients with HF with reduced ejection fraction in July of 2015 [55]. The PIONEER-HF trial randomised 881 stabilised HF patients to enalapril or sacubitril/valsartan to assess ARNI efficacy. Sacubitril/valsartan reduced NT-proBNP concentrations more than enalapril after 4 and 8 weeks. An exploratory study found that sacubitril/valsartan reduced HF rehospitalizations. Consequently, the TRANSITION study demonstrated that the inpatient initiation of ARNIs was both feasible and tolerable when compared to the outpatient initiation within two weeks of hospital discharge [53]. Moreover, the findings of a cross-sectional analysis indicate that in the seven years following the FDA approval of sacubitril/valsartan, the prescription prevalence of ARNI or ACEI, ARB, or ARNI at discharge rose, whereas the prescription rates of ACEI or ARB declined. The overall prescription rate of ARNI at hospital discharge was 55.4% among eligible patients by the conclusion of the trial, indicating the potential for further enhancement in ARNI prescribing practices [56].

On the other hand, the aim of developing synthetic analogues of natriuretic peptides was to replicate the cardiovascular benefits of endogenous NP. As the NP system intervenes as a compensatory mechanism, counterbalancing the negative neurohormonal effects of HF, this was an argument in favour of using exogenous NP in decompensated HF.

Carperitide, a synthetic analogue of human ANP, was among the first NPs used for therapeutic purposes. It was administered intravenously in acute decompensated left ventricular failure, but its use was associated with an increased in-hospital mortality rate [57]. This observation was reinforced by a recent study comparing carperitide with nitrates [58]. Thus, carperitide did not bring significant long-term benefits in terms of symptomatology and prognosis and did not impose itself on a large scale as a vasodilator and natriuretic in the treatment of decompensated heart failure. Carperitide was approved for clinical use only in Japan in 1995. The newest Japanese Circulation Society (JCS) guidelines for acute HF and chronic HF support intravenous carperitide as a treatment. Thus, carperitide is frequently administered in Japan. Retrospective studies indicate carperitide does not improve survival rates but instead leads to higher in-hospital mortality and hospitalisation expenses. Beginning treatment with carperitide may cause hypotension, which may worsen results. The JCS recommendations advocate low-dose continuous intravenous carperitide infusion [59].

Nesiritide is a 32-amino acid synthetic analogue of BNP, which, after the data obtained from the VMAC study [60] demonstrated favourable effects only 3 hours after the initiation of the intra-venous infusion, both clinical (decrease of dyspnoea), but especially haemodynamic effects (decrease in the pulmonary capillary pressure, increase of cardiac output and diuresis). On August 10, 2001, the FDA approved nesiritide. Nesiritide has been approved internationally in numerous countries, including the Dominican Republic, Colombia, Brazil, Canada, Slovenia, and Venezuela. Gradually, concerns were raised regarding its safety, especially its side effects. Small observational studies have reported either an increase in the rate of hospitalisation [61], the absence of any clinical benefit [62], or even a pro-arrhythmic effect with increased incidence of ventricular extrasystoles [63]. The greatest concern was caused by the induction of severe symptomatic hypotension, decreased kidney function, and especially increased mortality [64]. These led to the randomised study ASCEND-HF [65], conducted on 7141 patients with decompensated

heart failure, which demonstrated that nesiritide did not influence mortality or renal function nor decrease the rate of hospitalisation; the only benefit was the improvement of dyspnoea. However, excessive hypotension remains a secondary reaction during administration. Nesiritide is, therefore, not commonly employed but utilised only after the full exploration of all alternative therapeutic approaches to alleviate dyspnoea.

Emerging therapies and future directions

In the last two decades, a multitude of molecules that modulate the NP system have been developed for therapeutic purposes. Based on their pharmacodynamic mechanism, these molecules have been classified into several classes: synthetic natriuretic peptides, engineered chimaeras, dual vasopeptidase inhibitors, and neprilysin inhibitors.

Anaritide, a synthetic analogue of human ANP, was developed for the treatment of patients experiencing acute renal failure necessitating dialysis, acute HF, and hypotension. However, the outcomes were unsatisfactory, as the study was terminated due to the absence of positive outcomes, with numerous patients experiencing hypotension and reduced glomerular filtration rate [66]. Ularitide, a synthetic derivative of urodilatin, demonstrated in initial studies [67, 68] significant benefits *via* 24-hour continuous intravenous infusion, including reduced pulmonary capillary pressure, enhanced diuresis and natriuresis, and alleviated dyspnoea. The TRUE-AHF study, however, demonstrated no reduction in long-term mortality [69]. The application of synthetic NP is constrained by their limited clinical efficacy, significant hypotension, rapid enzymatic degradation (restricted to intravenous administration), and the absence of reduced mortality or hospitalisation rates.

Similarly, novel molecules have been developed *via* genetic engineering. Their composition includes distinct fragments from the molecular structures of two different natural NPs. Basically, a novel NP artificial molecule is consequently synthesised, integrating the effects of the two precursor molecules. CA-NP (CNAAC or Vasonatrin) consists of 27 amino acids and represents a fusion of the C-terminus amino acid ring from ANP and the N-terminus of CNP. It has shown vasodilating, diuretic, and hypotensive effects [70]. CD-NP (Cenderitide) consists of the C-terminal segment of DNP integrated into the CNP molecule, enabling activation of both NP receptors (NPR-A and NPR-B). This compound exhibits diuretic, natriuretic, and renal effects, along with antifibrotic properties, while avoiding significant hypotension, a common side effect of DNP and CNP [71]. Mutant ANP (MANP) is an isoform of ANP characterised by adding a C-terminal end comprising 12 amino acids. This modification enhances its resistance to NEP proteolytic degradation, resulting in more pronounced diuretic, natriuretic, and haemodynamic effects compared to ANP [72].

The adverse effects of synthetic (exogenous) nanoparticles have prompted the development of a novel approach to enhance the efficiency of the nanoparticle axis. Molecules have been developed to inhibit NEP, resulting in heightened endogenous NP activity and amplifying the diuretic, natriuretic, antifibrotic, and antiproliferative effects of NP. NEP inhibition not only prevents PN degradation but also inhibits the degradation of vasoconstrictor peptides inactivated by NEP, such as angiotensin II and ET-1, thereby counteracting the beneficial effects of NPs.

Dual vasopeptidase inhibitors function as dual inhibitors, simultaneously obstructing both NEP and RAAS. This elevates the levels of vasopeptides that exhibit vasodilating and cardioprotective effects (such as ANP, BNP, CNP, bradykinin, and adrenomedullin) while concurrently decreasing the levels of vasoconstrictor and profibrotic peptides, particularly Angiotensin II. All the abovementioned classes and the respective molecules are summarised in Table I.

These pharmacological agents have been evaluated in clinical trials for the treatment of cardiovascular pathologies such as HF, hypertension, and renal dysfunction. However, their approval as drugs and their use in large-scale clinical applications is still limited by the associated adverse effects or the fact that many of them have not been shown to improve long-term mortality rates.

A significant unmet demand exists for novel therapeutics in the management of HF. During the first phase of drug development, the medical benefit of a pharmaceutical remains inadequately comprehended, necessitating trial endpoints beyond mortality to inform drug development decisions [80]. However, efforts have been made to integrate new technologies into the design of HF drugs. Table II summarises some relevant new molecules that have been piloted to target the NP system.

Current preclinical research is developing a solid scientific basis for evaluating gene therapy methods. Over the past ten years, numerous molecular pathways associated with HF have been discovered, creating new targets for therapeutic interventions. Numerous targets remain unalterable through pharmaceutical means because of the previously outlined limitations; nonetheless, they are compatible with gene therapy approaches [87]. Cardiomyocytes are ideal targets for adeno-associated virus (AAV) vectors due to their terminal differentiation and non-dividing nature. Gene therapy has been implemented in numerous investigations to improve cardiac NP expression [31]. Table III summarises some recent advancements in NP system gene therapy.

Table I

New molecules derived from NP designed and tested in clinical trials but failed to be approved

Molecule	Class	Structure	Lack of approval details	Reference
Anaritide	Synthetic analogue of ANP	25 amino acids, a ring stabilised by a disulfide bond	Lack of efficacy and adverse effects, including hypotension and reduced glomerular filtration rate.	[73]
Ularitide	Synthetic analogue of urodilatin	32 amino acids, differing from ANP by the presence of an N-terminal extension of 4 amino acids	No reduction in long-term mortality (TRUE-AHF study).	[69]
CD-NP	Chimera combining DNP and CNP	Combines C-terminal segment of DNP with CNP molecule	Variability in outcomes due to multiple receptor activation, variable pharmacodynamics	[74]
CU-NP	Chimera with Urodilatin fragment	Contains NP amino acid ring, disulfide bond of CNP, and Urodilatin chains	Limited endpoint-focused studies	[75]
Mutant ANP (MANP)	Modified isoform of ANP	ANP with additional C-terminal chain (12 amino acids)	Absence of large-scale clinical trials, risks of hypotension	[76]
Thiorpan	NEP inhibitor	Propanoyl backbone with a mercapto-methyl group, a phenyl ring, and a glycine residue	Unintended and undesired haematological implications, no superior effects.	[75]
Candoxatril	NEP inhibitor	Prodrug	Counterbalanced RAAS and SNS activity; increased peripheral resistance in HF.	[77]
Sampatrilat	Dual NEP and RAAS inhibitor	Zinc-binding group inhibiting of metalloprotease activity	No superior benefits	[78]
Omapatrilat	Dual NEP and RAAS inhibitor	Heterocyclic dipeptide	High incidence of angioedema due to bradykinin accumulation.	[79]

Table II

New molecules, their target in NP system and their mechanism of action

Molecule	Target	Mechanism	Clinical implication	Reference
Allosteric enhancers	NPR-A	Enhances ANP/BNP activation by binding to novel allosteric sites on NPR-A, increasing cGMP production	Potential treatment for cardiovascular and renal disorders, allowing oral administration and extended half-life	[81]
Bis-aminotriazines (lead compound 1)	NPR-C	Acts as a potent NPR-C agonist (EC50 ~ 1 μM), promoting vasorelaxation and reducing blood pressure	Therapeutic candidate for hypertension and cardiovascular disorders	[82]
NPR-B antagonists	NPR-B	Inhibits cGMP production, preventing cross-talk with PDE3 and harmful β1-adrenoceptor signalling	Potential heart failure treatment by mitigating PDE3 inhibition and reducing adverse cardiac effects	[83]
ASB20123 (CNP analogue)	NPR-B	Combines CNP (1–22) with ghrelin (12–28) for enhanced NPR-B activation and skeletal growth	Treats growth failure and dwarfism with extended plasma half-life and cartilage-targeting properties	[84]
CNP-derived small molecules	NPR-C	Promotes vasodilation by enhancing CNP-dependent signalling	Useful in vascular disorders such as hypertension and atherosclerosis	[85]
Anti-NPR1 antibodies	NPR-A	Induces intrinsic apoptosis by downregulating *BCL-2* and mitochondrial dysfunction	Promising therapy for gastric cancer, shown to inhibit tumour growth in xenograft models	[86]

However, addressing the cardiovascular system by applying gene therapy in humans has been challenging. AAV1-mediated delivery of sarcoplasmic/endoplasmic reticulum Ca^{2+} ATPase 2a (SERCA2a) during hospitalisation or ambulatory treatment for worsening HFrEF was safe; however, it failed to improve patient outcomes in the CUPID and CUPID2b trials. Its lack of effectiveness may be attributed to the inadequate delivery of vectors to cardiac cells, as only less than 2% of cardiomyocytes within the highest dose group carry a vector. Therefore, it is necessary to enhance transduction efficiency prior to the clinical application of myocardial gene therapy to augment cardiac NP production [31].

Table III

Gene therapies for HP *via* NP system

Gene therapy technique	Target	Mechanism	Clinical implication	Reference
Epigenetic modulation of *NPPA* and *NPPB* genes	ANP and BNP genes	Epigenetic modifications (*e.g.*, histone modifications, DNA methylation, miRNAs) to regulate gene expression	Potential to treat cardiovascular diseases by restoring proper natriuretic peptide function	[88]
SERCA2a gene therapy with BNP interaction	*SERCA2a* expression	BNP is shown to inhibit *SERCA2a* expression; potential to combine BNP regulation with *SERCA2a* gene therapy	Addressing heart failure by mitigating BNP-induced downregulation of *SERCA2a*	[89]
Genetic disruption of NPR-A (*Npr1* gene)	NPRA	*Npr1* knockout in mice used to study fibrosis, targeting TGF-β/SMAD signalling pathways	Therapeutic targets to prevent fibrosis and heart dysfunction	[90]
Peptide-based vectors for gene delivery	Gene delivery systems	Cell-penetrating peptides engineered for targeted gene delivery	Enhanced delivery of gene therapies for natriuretic peptide pathways	[91]
MicroRNA-425/155 inhibition	ANP (*NPPA* gene)	Cooperative inhibition of miR-425 and miR-155 increases ANP expression, enhancing cGMP production	Promising therapeutic approach for hypertension and related cardiovascular conditions	[92]

NPs have consistently been considered appealing targets for the management of HF and hypertension. Despite the early impediments to the use of NPs due to restricted clinical efficacy, mostly stemming from the drawbacks of endogenous NPs, including rapid enzymatic degradation and adverse effects, notably significant hypotension, the therapeutic application of NPs continues to be a highly desired objective. In light of the introduction of sacubitril/valsartan, the notion of a medication possessing both NP-enhancing and RAAS-counteracting attributes is seen as highly appealing and warrants deeper investigation. However, the morbidity and mortality associated with heart failure and the challenges in managing hypertension remain significant [93]. The process of designing NPs involves the development of a substance that is not inherently present in the human body. There is an apprehension that the administration of designer peptides might turn on the body's immune system as a defence mechanism, resulting in the production of antibodies against new peptides. This could lead to the emergence of negative reactions and/or a reduction in drug efficacy. Furthermore, the efficacy of drugs may be reduced by structural modifications, irrespective of the immune response. Safety verification must be diligently conducted at every stage of the clinical trial, as it is unquestionable that adverse reactions can arise among humans even if they are not toxic in animal experiments [94].

Conclusions

NPs represent a significant therapeutic target in the treatment of HF. Their diverse functions in inhibiting the RAAS and SNS offer essential cardio-renal advantages, such as vasodilation, natriuresis, and the mitigation of heart hypertrophy and fibrosis. Innovations in treatments have demonstrated potential in utilising the endogenous NP system to re-establish cardiovascular homeostasis. Despite limitations such as quick degradation and negative effects, ongoing innovation and research highlight the significance of targeting NPs to improve morbidity, mortality, and overall outcomes for heart failure patients.

Conflict of interest

The authors declare no conflict of interest.

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