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REVIEW

Nesfatin-1: a new energy-regulating peptide with pleiotropic functions. Implications at cardiovascular level

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Abstract Nesfatin-1 is a new energy-regulating peptide widely expressed at both central and peripheral tissues with pleiotropic effects. In the last years, the study of nesfatin-1 actions and its possible implication in the development of different diseases has created a great interest among the scientific community. In this review, we will summarize nesfatin-1 main functions, focusing on its cardiovascular implications.

Keywords Nesfatin-1 · Energy homeostasis · Cardiovascular · Viability · Metabolism

Introduction

The term cardiovascular disease (CVD) comprises a wide range of pathophysiological conditions of heart, blood, and vasculature that include coronary heart disease (CHD), myocardial infarction (MI), heart failure (HF), cardiomyopathy, arrhythmia, valvular heart disease, stroke, atherosclerosis, etc. [1, 2]. Despite the great effort of the medical community during the last decades, CVDs remain the leading cause of death worldwide, increasing their prevalence every year [1]. According to the World Health Organization (WHO), although CVDs prevalence is increasing in both developed and developing countries, the percentage of premature deaths from CVDs in 2008 ranged from 4 % in high-income countries to 42 % in low-income countries, leading to growing inequalities in the occurrence and outcome of CVDs [3]. The economical difference between both scenarios implies not only that people in low- and middle-income countries often do not have the benefit of integrated primary health care programs for early detection and treatment of people with risk factors, being detected late in the course of the disease and dying younger [4], but also that the availability and promotion of cheap energy-dense food boost the poorer segments to opt for these energy-dense diets, rich in cheap vegetable oils, sugars, and trans fats [5], contributing to increase some of the main risk factors for the development of CVDs: obesity, hypertension, insulin resistance, type 2 diabetes mellitus (T2DM), and metabolic syndrome [6, 7]. Hence, due to the high prevalence of CVDs and the multiple risk factors affecting their development (most of them associated with our new way of life: industrialization, mechanized transport, urbanization, an increasingly sedentary lifestyle, and a nutritional transition to processed foods and high-calorie diets [8]), there is an urge in the understanding of the mechanisms of action of the main players involved in the development of CVDs to try to find new targets which with their therapeutic modulation could help to reduce the prevalence and mortality of CVDs.

In the last decades, numerous studies have shown an association between the adipose tissue pathological function and distribution and the higher risk to develop CVDs, mainly through the secretion of chemical mediators that act in an autocrine/paracrine/endocrine way to modulate not only cardiovascular function but also a wide range of

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biological processes. In this review, we will summarize the implication of the adipose tissue in the cardiovascular risk, and we will focus on one of its secretory products: nesfatin-1, an anorexigenic peptide with pleiotropic functions, and direct effects at cardiovascular level.

Adipose tissue and cardiovascular diseases

The association between obesity and the cardiovascular risk has been described in multiple epidemiological studies and is at present clearly accepted. As a whole, overweight/obesity is associated with numerous cardiac complications such as CHD, HF, or sudden death due to the impact of chronic excessive body fat accumulation on the cardiovascular system, which causes adaptations to maintain whole body homeostasis, including the augmentation of circulating blood volume, increased cardiac output, decreased peripheral resistance, increased stroke volume, or ventricular remodeling, which can lead to systolic and diastolic dysfunction [7, 9].

Adipose tissue distribution and cardiovascular risk

Although a high body mass index (BMI) is related to the development of CVD risk factors, nowadays there are numerous evidences supporting the idea that the regional fat accumulation is more important than the excess of adiposity *per se* when considering the risk to develop CVDs [9]. Individual's with upper abdominal, central, or android obesity (visceral fat) are at a greater risk than those with gluteofemoral, peripheral, or gynoid obesity (subcutaneous fat) due to the fact that the type of adipocytes, their endocrine function, lipolytic activity, and response to insulin and other hormones are different between both fat depots [10]. In this way, visceral fat accumulation is linked to the increase of free fatty acids (FFAs) release to the bloodstream, insulin resistance, hyperlipidemia, and endothelial dysfunction, while subcutaneous fat is associated with a protective phenotype [10]. Thus, the higher lipolytic activity and the increased release of FFAs from the visceral adipocytes are detrimental for the heart and its function. Also, the visceral adipose tissue is infiltrated by macrophages, which have a cross-talk with the adipocytes, and contribute to a low-grade inflammatory process typical of states characterized by increased fat accumulation, such as obesity [11]. Hence, the accumulation of visceral fat contributes to the development of many metabolic abnormalities and clinical outcomes such as T2DM and CVDs (Fig. 1).

Compared with other visceral fat depots, epicardial fat (located along the large coronary arteries and on the surface of ventricles and the apex of the heart) has a greater capacity for the release and uptake of FFAs, and it has been recently proposed that FFAs could diffuse bidirectionally in interstitial fluid across concentration gradients playing a significant role in the intracellular transport of FFAs from the epicardial fat to the myocardium [12]. Thus, the different fat accumulation and its activity under pathological conditions such as obesity are of a great importance to the cardiovascular system physiology.

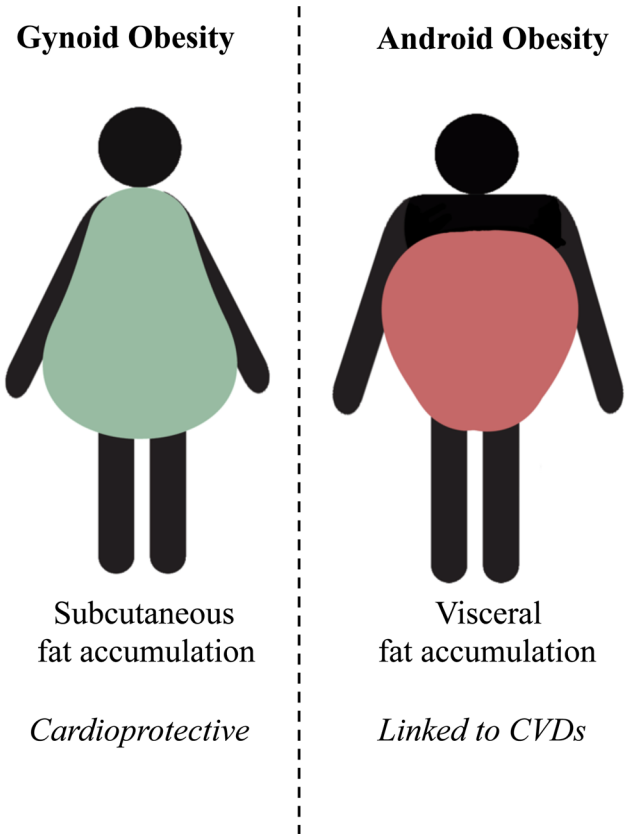

Fig. 1 Body fat distribution and cardiovascular risk

The adipose tissue as an endocrine organ

Apart from its classical function as a storage organ, white adipose tissue (WAT) has been identified as a metabolically active endocrine organ that releases chemical mediators, termed adipocytokines or adipokines, which set a communication network between the WAT and other tissues, sympathetic nervous system and brain to modulate appetite, energy balance, immunity, insulin sensitivity, angiogenesis, blood pressure, lipid metabolism, and homeostasis [10]. Obesity yields to a state of dysfunction and chronic inflammation of the WAT, mainly due to the secretion of pro-inflammatory cytokines by infiltrated macrophages, which alters the adipokines production/signaling network, having important consequences at

metabolic level that contribute to the development of CVDs (Fig. 2) [13].

Because of its anatomical position, visceral fat venous blood is drained directly to the liver through the portal vein (while venous drainage in subcutaneous fat is through systemic veins), providing direct hepatic access of adipokines secreted by visceral adipocytes that can affect hepatic functions [10]. Moreover, when compared to the subcutaneous adipocyte, the visceral fat cell has a differential production pattern of adipokines, which have a critical role in linking the metabolism with the proper heart and blood vessels homeostasis [11]. Thus, not only the amount, dysfunction and the adipokines secretion pattern of adipose tissue but also body fat distribution and the adipokines impact on other organs are important to determine the risk of CVDs development.

Nesfatin-1 as a new energy-regulating peptide with pleiotropic effects

Nesfatin-1 is a 9.5 kDa peptide with an amino acid sequence highly conserved across mammalian and non-mammalian species [14], and with a high identity in human, rat, and mouse (Fig. 3), which is indicative of its phylogenetic and functional relevance. It was identified in 2006 by Oh-I et al. as a new hypothalamic molecule implicated in the regulation of food intake through a leptin-independent melanocortin signaling system in rats [15]. Nesfatin-1 not only was found to be widely expressed in the main appetite-control hypothalamic nuclei such as the paraventricular nucleus (PVN), arcuate nucleus (ARC), supraoptic nucleus (SON), lateral hypothalamic area (LHA), zona incerta, and solitary tract (NTS) in rats and mice [15–17], but it also has a wide brain distribution in other species including swines [18], teleosts [19, 20] and amphibians [21]. Nesfatin-1 levels in the PVN in rats were found to be reduced by starvation, while it intracerebroventricular (i.c.v.) injection reduced food intake, body weight gain, and weights of subcutaneous, epididymal, and mesenteric fats in rat, launching nesfatin-1 as a potential useful target for the development of drug therapies to treat obesity [15]. The same authors who discovered nesfatin-1 had previously proposed the concept of brain-adipose axis, which refers to the fact that certain endogenous molecules and/or their receptors are expressed in the hypothalamus and in peripheral adipose tissue, function as appetite regulators in the brain, exist in the general circulation as secreted proteins, and affect adipogenesis, being some of the molecules proposed to be part of the brain-adipose axis

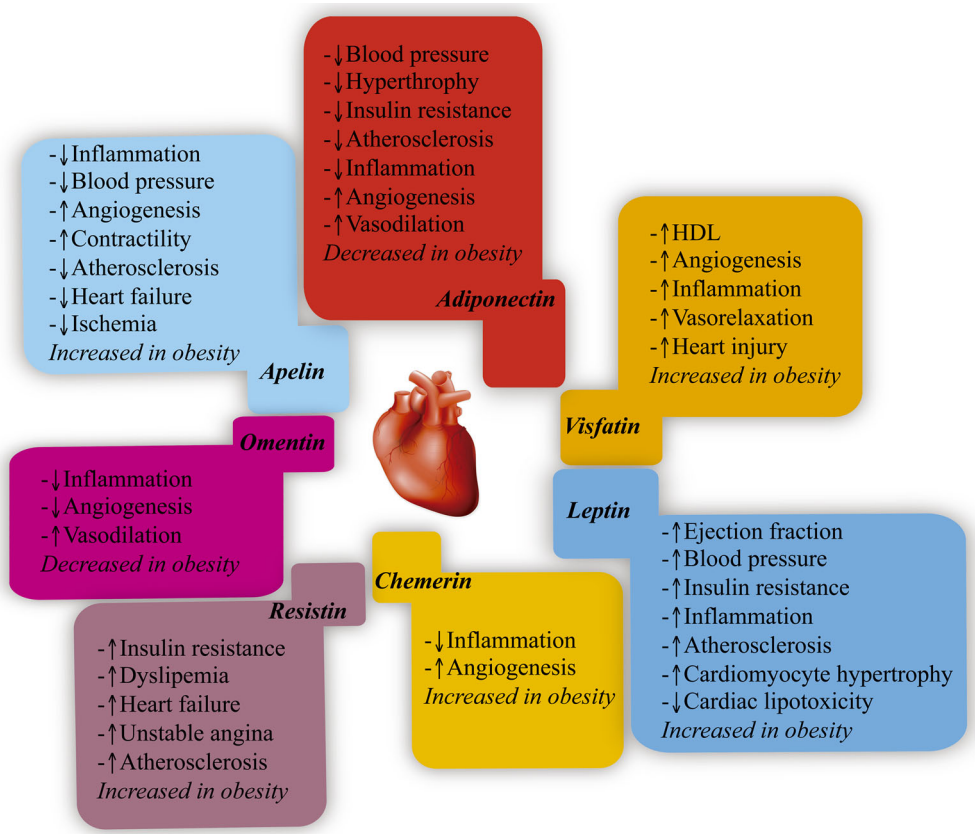

Fig. 2 Functions of different adipokines at cardiovascular level and changes in their production levels in obesity [164–166]

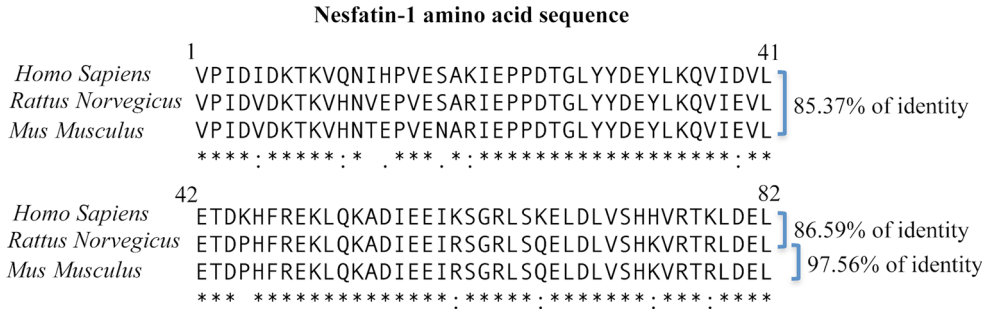

Fig. 3 Comparison of nesfatin-1 amino acid sequence in human, rat, and mouse using the multiple sequence alignment program Clustal Omega (EMBL-EBI, UK). * (asterisk): indicates positions which have a single, fully conserved residue, : (colon): indicates conservation between groups of strongly similar properties, and . (period): indicates conservation between groups of weakly similar properties

insulin, or the adipokines leptin and adiponectin [22]. According to this, Oh-I et al. demonstrated that the nesfatin-1 precursor, nucleobindin-2 (NUCB2, 47.5 kDa), was also expressed by 3T3-L1 adipocytes and that its levels in rat hypothalamic and 3T3-L1 adipocyte cells increased after treatment with troglitazone, a ligand of peroxisome proliferator-activated receptor (PPAR)-γ used in the past as an oral insulin-sensitizing anti-diabetic agent, removed from the market in 2000 because of hepatoxicity [23].

Soon after the identification of nesfatin-1 as a new hypothalamic endogenous molecule that induces anorexia [15], it was demonstrated that nesfatin-1 was able to cross the blood brain barrier without saturation in either the blood-to-brain or brain-to-blood direction, suggesting that nesfatin-1 could be produced by peripheral tissues, and that peripherally produced nesfatin-1 could exert its effects at central level and vice versa [24, 25]. Subsequently, numerous studies corroborated the anorexigenic effect of nesfatin-1 not only through its central injection but also after peripheral injection [17, 26–30]. As well, it was demonstrated the presence of nesfatin-1 in body fluids from rodents and humans such as plasma [31–33], synovial fluid [34], saliva [35] and breast milk [36], and its production by several peripheral tissues, including the adipose tissue [37], endocrine cells of the gastric mucosa (where it is co-expressed with ghrelin but in different secretion vesicles, and where its mRNA expression in rats is ten times higher than in brain) [38], β-islet cells in the pancreas (where it is co-expressed with pro-insulin and insulin) [39], Brunner glands in the duodenum, esophagus, liver, small intestine and colon [40], testis [41], ovaries and uterus [42], salivary gland [35], articular cartilage [34], lungs [43], and heart [43, 44] (Fig. 4). Apart from rodents and humans, nesfatin-1 was also found to be produced in the canine digestive tract [45], in the hypothalamus [18], adipose tissue [46] and digestive tract of pigs [47], or in plasma and milk in cows [48], as well as it was shown to have a widespread distribution in non mammals such as teleosts [19, 20, 49–51] and amphibians [21], supporting its phylogenetic and functional relevance. Although nesfatin-1 was initially found to be widely expressed at central level, it is not only just a hypothalamic molecule implicated in the regulation of food intake (indeed nowadays the stomach is considered its main source of production [52]), but also a peripherally produced peptide that has a wide range of biological effects not only at endocrine but also at autocrine/paracrine levels in both central and peripheral organs (Fig. 5) [53, 54].

From NUCB2 to nesfatin-1

Nesfatin-1 is an 82 amino acids peptide derived from the proteolytical cleavage of its precursor, NUCB2, by pro-hormone convertases (PCs) at the Lys 83-Arg 84 site [15] (Fig. 6). Although NUCB2 has other potential sites for PCs cleavage that result in the nesfatin-2 (residues 85–163) and nesfatin-3 (residues 166–396) forms, only nesfatin-1 or the full length NUCB2 have the potential role to induce satiety. Indeed, it was demonstrated that NUCB2 needs to be proteolytically processed into nesfatin-1 to be able to induce anorexia [15].

According to its secondary structure, nesfatin-1 could be divided in 3 segments: N-terminal segment (23 amino acids), a mid-segment (30 amino acids), and a C-terminal segment (29 amino acids), corresponding to amino acid residues 1–23, 24–53, and 54–82 of nesfatin-1, respectively [26] (Fig. 6). Although, there are not yet evidences proving that this process could occur in vivo, this observation led to the identification of the nesfatin-1 functional domain, located in its 30 amino acids mid-segment, which has a similar effect on the reduction of food intake as nesfatin-1 when injected both peripherally [26] and i.c.v. [55] in mice. To identify the essential core sites of the mid-segment, a homology search of the amino acids sequence of the human nesfatin-1 mid-segment against those of other molecules known to regulate appetite was performed, founding that the C-terminal region of the mid-segment contains an amino acid sequence His-Phe-Arg identical to that of the anorexigenic hormone melanocyte stimulating hormone (MSH)-α and that contains a

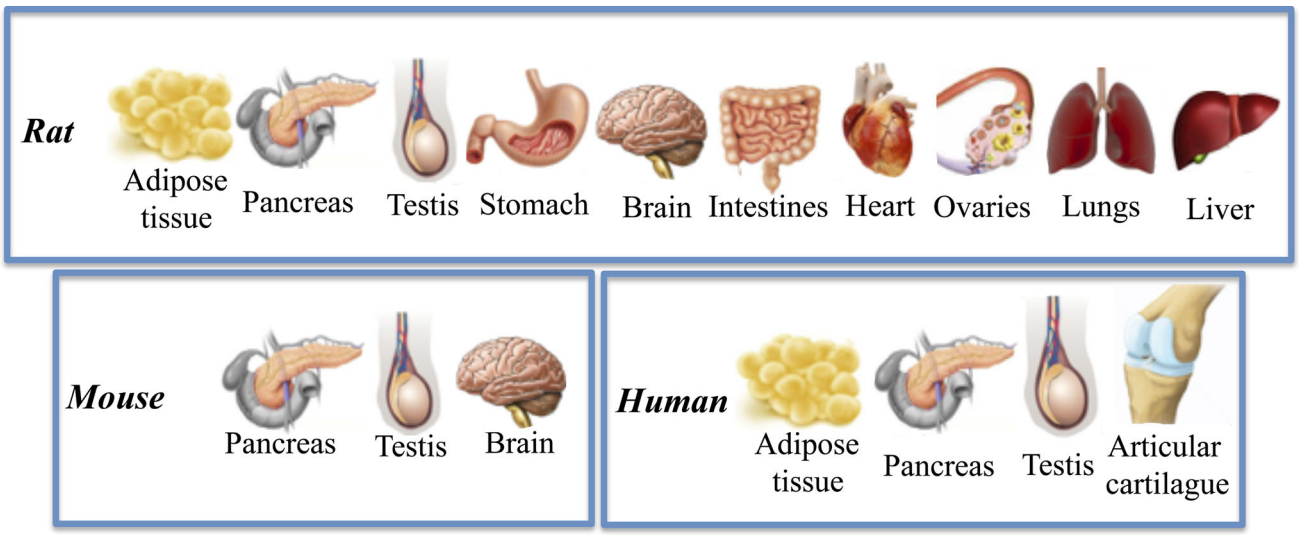

Fig. 4 Nesfatin-1 production by peripheral tissues in rat, mouse, and human

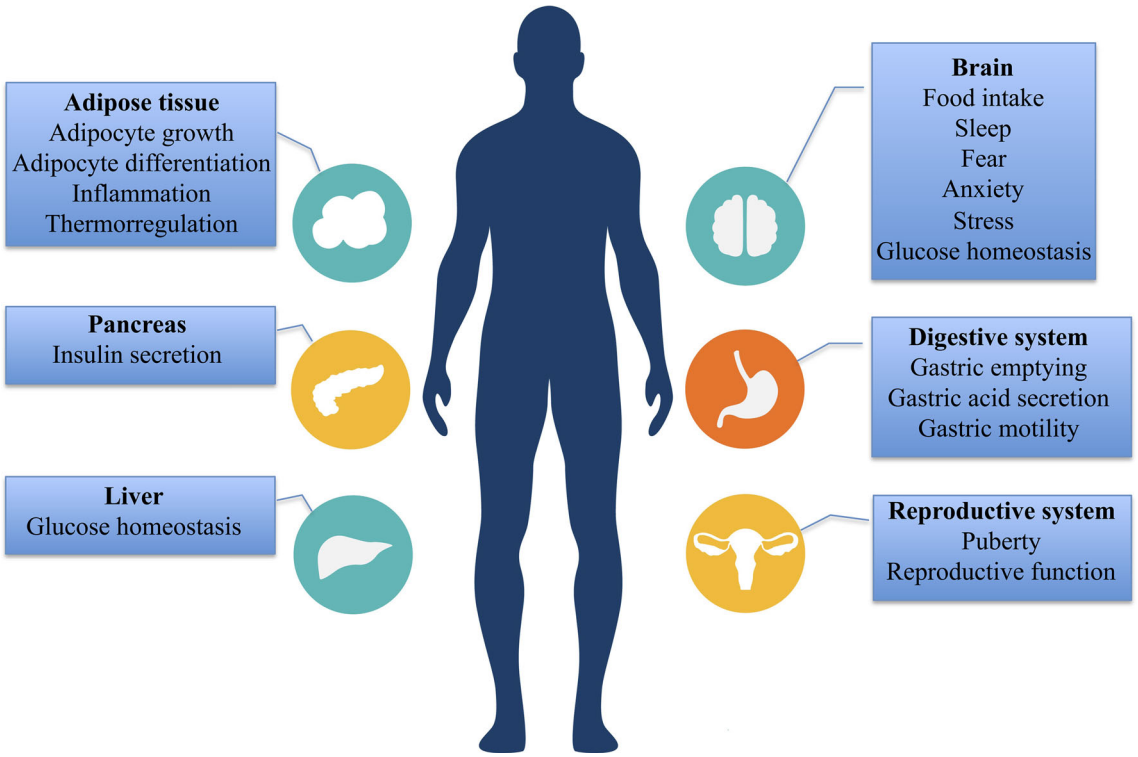

Fig. 5 Pleiotropic effects of nesfatin-1

characteristic recognition site for the MSH-α functional receptors melanocortin 3 receptor (MCR3) and MCR4 [26]. The central region of the mid-segment contains the amino acid sequence Leu-Lys-Gln-Val-Ile-Asp-Val that shows 42.9 % identity and 85.7 % similarity to an amino acid sequence in human agouti-related peptide (AgRP) (antagonist of MCR3/4); this sequence includes the site in AgRP involved in increased energy expenditure [26]. Although Oh-I et al. (2006) reported that nesfatin-1 anorexigenic function was dependent of the melanocortin system, the region of the mid-segment that shows similarity to MSH-α was not responsible for inducing anorexia, being the region of the mid-segment with similarity to AgRP essential for its anorexigenic induction [26].

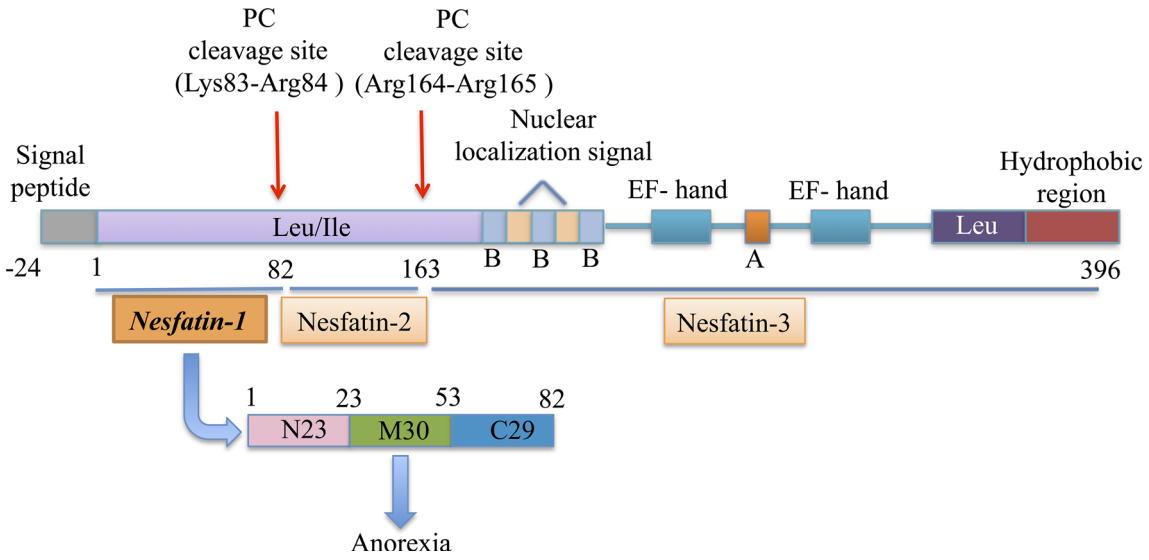

Fig. 6 Protein structure of NUCB2 and nesfatin-1. PC: prohormone convertase, A: acid amino acids, B: basic amino acids, N23: N-terminal segment of 23 amino acids, M30: mid-segment of 30 amino acids, C29: C-terminal segment of 29 amino acids [15, 26, 167].

Despite the identification of the essential core site of nesfatin-1, the mechanisms through which nesfatin-1 exerts its actions are still poorly understood. The first aim of the scientific community was to identify the possible existence of a specific receptor for nesfatin-1 to better understand its function and to have the hints to could use it as a therapeutic target. Although it was demonstrated using radiolabeled nesfatin-1 that it could bind to membrane preparations of mouse hypothalamus [56], the specific receptor for nesfatin-1 has not been identified yet.

Nesfatin-1 as an energy-regulating peptide

Nesfatin-1 has a clear effect on regulating food intake not only in mice and rats at both central and peripheral level [15, 26], but also in teleosts [20, 50]. As suggested by Shimizu et al. [22, 57], nesfatin-1 is a good candidate to be part of the brain-adipose axis, so that it was demonstrated to be expressed in hypothalamus and in both white and brown adipose tissue, and its adipose, hypothalamic, and circulating levels are modified by states of feeding and starvation, being decreased by starvation and increased after re-feeding or high-fat diet, suggesting a physiological role of nesfatin-1 in energy metabolism [15, 37]. Moreover, nesfatin-1 secretion from explants of subcutaneous adipose tissue of murine and human was found to be higher than visceral adipose tissue, suggesting a different secretion pattern of nesfatin-1 by different fat depots [37]. Nesfatin-1 is also secreted by cultured adipocytes, and its expression/secretion levels vary according to the maturation state of adipocytes although there exist contradictory results) [37, 58]. Ramanjaneya et al. have shown that 3T3-L1 preadipocytes express and secrete nesfatin-1 into the medium in increasing amounts as the adipocyte matures for 12 days [37], while Tagaya et al. describe a reduction of endogenous nesfatin-1 levels at 8 and 12 days after the induction of 3T3-L1 cells differentiation [58]. Consistent with the decrease in endogenous nesfatin-1 protein levels during 3T3-L1 adipogenesis, the stable knockdown of NUCB2 results in increased adipogenesis, while stable overexpression of NUCB2 decreased neutral lipid accumulation and reduced adipogenic gene expression [58]. Despite the contradictory results, these observations suggest that nesfatin-1 could participate in the regulation of adipogenesis [59], although more research is needed.

On the other hand, it is well established that in obesity there is a chronic low-grade inflammatory response accompanied by adipokine deregulation, which leads to chronic subclinical inflammation as well as insulin resistance [60]. In this way, nesfatin-1 expression and secretion by 3T3-L1 adipocytes and by subcutaneous adipose tissue explants have been shown to be up-regulated by not only treatment with obesity related pro-inflammatory cytokines such as interleukin-6 or tumor necrosis factor (TNF)-α, but also by anti-inflammatory drugs such as dexamethasone, suggesting a possible role for nesfatin-1 in inflammatory states associated with obesity [37].

Another important factor regarding energy metabolism regulation is the glycemic control. Nesfatin-1 stimulates glucose-induced insulin release from isolated pancreatic islets from mice [61], and its production and secretion by human and murine β-pancreatic cells have been demonstrated to be increased by high-glucose stimulation [61, 62]. These data suggest that nesfatin-1 could act as a

response factor after food intake at pancreatic level to regulate insulin secretion. Indeed, plasma nesfatin-1 levels were found to be decreased by fasting and increased by peripheral glucose injection in rats [62], indicating that nesfatin-1 circulating levels can be regulated and fluctuate by nutritional status.

In *db/db* hyperglycemic mice, intravenous injection of nesfatin-1 decreases glucose plasma levels through an insulin-signaling dependent mechanism, being its anti-hyperglycemic effect higher when co-administered with insulin [63]. This anti-hyperglycemic effect of nesfatin-1 was observed only when administered peripherally, since central injection of nesfatin-1 was found to have no effect on reducing glycemia in this study [63]. In C57BL/6J mice fed with normal or high-fat diet, peripheral injection of nesfatin-1 improves glucose tolerance and insulin sensibility (while central injection has no effect) through a mechanism that involves protein kinase B (PKB/AKT) activation and glucose transporter (GLUT)-4 translocation to the plasma membrane in skeletal muscle and adipose tissue [64], and in T2DM mice, nesfatin-1 peripheral injection induces a reduction in body weight, FFA circulating levels, glycemia, and insulin resistance and increases 5' AMP-activated protein kinase (AMPK) and acetyl-CoA carboxylase (ACC) phosphorylation in skeletal muscle, suggesting the nesfatin-1 implication in fatty acids oxidation in this tissue [65].

In the hypothalamus, nesfatin-1 has been shown to modulate the excitability of glucosensing neurons [66, 67], and nesfatin-1-producing neurons excitability was demonstrated to be affected by glucose concentrations [68, 69] and insulin [69]. Central nesfatin-1 knockdown was found to induce an increase in food intake and in liver glucose flux by increasing glucose-6-phosphatase (G6PT) and phosphoenolpyruvate carboxykinase (PEPCK) and by reducing insulin receptor, insulin receptor substrate 1 (IRS-1), and AKT phosphorylation, and to induce a decrease in glucose uptake by peripheral tissues [70].

Nesfatin-1 has also been demonstrated to increase body temperature in rats by the activation of MCR4 receptors and the brown adipose tissue non-shivering heat production [71]. Moreover, nesfatin-1 producing neurons are activated by cold, suggesting that nesfatin-1/NUCB2 may mediate thermoregulation, as well as other responses to cold [71].

In spite of the clear effect of nesfatin-1 on regulating whole body energy metabolism, studies performed to understand the nesfatin-1 role in the development of obesity and diabetes show contradictory results, possibly due to the short-life time of circulating nesfatin-1 ($\sim$ 10-20 min) [24, 25] and to the differences between studies regarding the sample size, gender, age, the different sensitivity/specificity of the methods and techniques used, the range of body weight, the different ethnic groups studied, the existence of comorbidites or even nutritional differences among subjects, so further investigation is really needed.

Nesfatin-1 levels in obesity

Basar et al. observed that in 30 Turkish patients with non non-alcoholic fatty liver disease serum nesfatin-1 levels were lower than in controls (40 age- and sex-matched healthy subjects) [72]. When analyzing the NAFLD patients and controls as a pool, they found a negative correlation between serum nesfatin-1 levels and BMI, and a statistically significant difference between obese ($n = 29$) (BMI $\geq$ 30 kg/m^2) and non-obese subjects ($n = 41$) [72]. Abaci et al. reported lower plasma nesfatin-1 levels in 37 Turkish obese children (BMI > 95th percentile) compared to 31 healthy children with BMI < 85th percentile who had similar age and gender distribution [73]. Tsuchiya et al. observed in a cohort of 43 Japanese subjects (30 non-obese; 24.5 $\pm$ 0.6 years of age; BMI = 21.1 $\pm$ 0.3 kg/m^2 and 9 obese; 32.4 $\pm$ 3.7 years of age; BMI = 37.3 $\pm$ 3.8 kg/m^2) that nesfatin-1 circulating levels were significantly lower in the obese group [74]. Tan et al. showed a negative correlation between the cerebrospinal fluid/-plasma nesfatin-1 levels and BMI, body weight and fat mass in a cohort of 18 men and 20 women from Germany, aged from 19 to 80 years and with a BMI from 16.2 to 38.1 kg/m^2, of which 14 were normal weight, 14 overweight, and 10 obese [33]. Ogiso et al. found again a negative correlation between plasma nesfatin-1 levels and BMI in a cohort of 7 Japanese women with anorexia nervosa and 8 healthy controls, founding statistically significant differences between the anorexic patients (BMI = 13.02 $\pm$ 0.3 kg/m^2) and controls (BMI = 21.57 $\pm$ 0.48 kg/m^2) [75]. Finally, Ozkan et al. observed lower plasma nesfatin-1 levels in Turkish obese patients ($n = 30$) (BMI 30–39.9 kg/m^2) and morbid obese patients ($n = 30$) (BMI > 40 kg/m2) compared to normal weight ($n = 28$) (BMI 18.5–24.9 kg/m^2) and overweight ($n = 31$) (BMI: 25–29.9 kg/m^2) subjects ranged from 18 to 70 in age and with a 50 % of representation of each sex [76].

On the contrary, there also exist a number of studies showing the opposite: Saldanha et al. described a positive correlation between plasma nesfatin-1 levels and BMI, the percentage of body fat and leptin circulating levels in a group of 15 men and 10 Brazilian women undergoing hemodialysis (age 53.2 $\pm$ 11.9 years) and their controls (10 men and 5 healthy women, age 47.9 $\pm$ 14.8 years) [77]. Ramanjaneya et al. found a positive correlation between plasma nesfatin-1 levels and BMI in 9 men and 11 English women with a BMI ranged from 21.39 to 38.10 kg/m^2 and from 28 to 58 years of age, as well as they observed higher plasma nesfatin-1 levels in mice with high-fat diet-

induced obesity [37]. Anwar et al. found in obese children and adolescents from Egypt with 9.18 ± 2.84 years of age (22 boys and 18 girls, BMI = 30.45 ± 5.93 kg/m^2) higher plasma nesfatin-1 levels than controls (24 boys and 16 healthy girls, BMI = 16.82 ± 3.17 kg/m^2), as well as a positive correlation with BMI when analyzing all together [78]. Liu et al. have described a positive correlation between plasma nesfatin-1 levels and BMI in 85 Asian subjects (30 patients with T2DM and with 61.23 ± 8.83 years of age, 25 patients with impaired glucose regulation and 59.04 ± 9.01 years of age, and 30 healthy controls with 59.60 ± 9.26 years of age) [79]. Finally, Zhao et al. also described in a cohort of 80 Chinese subjects (41 men and 39 women; 31–74 years of age, BMI = 19.83–34.21 kg/m^2) a positive correlation between plasma nesfatin-1 levels and BMI [80].

Nesfatin-1 levels in diabetes

Li et al. observed in 57 Chinese subjects that plasma nesfatin-1 levels were decreased in patients with T2DM (23 men and 24 women, 59.55 ± 1.80 years of age, BMI = 26.46 ± 0.46 kg/m^2) compared to healthy controls (13 men and 7 women, 47.30 ± 2.29 years of age, BMI = 24.96 ± 0.92 kg/m^2) or to T1DM (6 men and 4 women, 29.13 ± 3.17 years of age, BMI = 19.58 ± 0.86 kg/m^2) [32]. Aslan et al. found that serum nesfatin-1 levels were decreased in pregnant women with gestational diabetes mellitus (n = 30, 30.9 ± 4.2 years of age, BMI = 25.9 ± 3.3 kg/m^2) compared to healthy controls (n = 30, 31.0 ± 3.2 years of age, BMI = 25.7 ± 2.8 kg/m^2) and that they also correlate positively with nesfatin-1 levels in fetal cord blood [81]. Basar et al. observed in a pool of 30 Turkish patients with NAFLD and 40 age- and sex-matched healthy subjects that serum nesfatin-1 levels were negatively correlated with glycemia and that they were lower in insulin resistant patients (n = 32, homeostasis model assessment-estimated insulin resistance (HOMA-IR) ≥ 2.5) compared to the insulin-sensitive group (n = 38, HOMA-IR < 2.5) [72]. As well, Liu et al. have described in 85 Asian subjects (30 patients with T2DM and with 61.23 ± 8.83 years of age, 25 patients with impaired glucose regulation and 59.04 ± 9.01 years of age, and 30 healthy controls with 59.60 ± 9.26 years of age) that plasma nesfatin-1 levels are decreased in T2DM patients, and that when pooled, nesfatin-1 levels are negatively correlated with glycated hemoglobin (HbA_{1c}) [79].

On the contrary, Zhang et al. observed in 220 Chinese subjects divided in 74 newly diagnosed T2DM (nT2DM) patients (39 men and 35 women, 54 ± 11 years of age, BMI = 25.0 ± 3.7 kg/m^2), 73 subjects with impaired glucose tolerance (IGT) (35 men and 38 women, 54 ± 10 years of age, BMI = 24.7 ± 2.7 kg/m^2), and 73 healthy subjects (36 men and 37 women, 51 ± 7 years of age, BMI = 24.5 ± 3.6 kg/m^2) that plasma nesfatin-1 levels were increased in both nT2DM and IGT patients compared to controls [82]. Dong et al. observed using mice with T2DM induced by a combination of high-calorie diet and low-doses of Streptozotocin that plasma and gastric nesfatin-1 levels were significantly increased, while nesfatin-1 expression in neurons were decreased in hypothalamus, in the T2DM group compared to only high-calorie diet control group [65]. The authors suggest that the increased plasma nesfatin-1 levels are due to an increased secretion from gastric mucosa and that this change was originated from the reduced central nesfatin-1 in T2DM [65].

Nesfatin-1 and gastric function

Nesfatin-1 has been demonstrated to have a high expression in stomach [31, 38, 40], suggesting the potential role of this peptide in the regulation of gastric functions. Indeed, intracerebral injection of nesfatin-1 in rats inhibits gastric acid secretion induced by vagal nerve signals [83], decreases gastric emptying [27], and protects gastric mucosa from stress-induced [84] and indometacin-induced injury [85]. Central administration of nesfatin-1 in the ARC inhibits the ghrelin-responsive gastric distension excitatory neurons and stimulates ghrelin-responsive gastric distension inhibitory neurons; an effect that seems to be responsible for the nesfatin-1 reduction of gastric motility [86].

Nesfatin-1 and stress, anxiety and fear responses

Many of the main food intake regulatory peptides are involved at hypothalamic level in the regulation of the stress response necessary to the regulation of energy homeostasis and adaptation to the environment [87, 88]. Nesfatin-1 is expressed by autonomic regulatory nuclei located in the forebrain, hindbrain, and spinal cord along with other forebrain nuclei involved in stress response and cognitive function in rats [89, 90]. Nesfatin-1 producing neurons have been shown to be activated by acute exposure to restraint stress [91, 92], known to alter gut function through central modulation of autonomic outflow [93], and peripheral nesfatin-1 injection in rats protects gastric mucosa against stress-induced injury by 3.5 h of water immersion and restraint stress, decreasing gastric secretion and gastric lesions through a mechanism that involves cyclooxygenase- prostaglandin (COX-PG) and nitric oxide synthase-nitric oxide (NOS-NO) systems, the activation of vagal and sensory nerves and vanilloid receptors [84].

Nesfatin-1 i.c.v. injection in rats induces an increase in anxiety and fear-related behaviors [94]. In the same line, in obese female patients [95] and in women with anorexia nervosa [96], nesfatin-1 circulating levels are associated with elevated scores of anxiety, while in obese males it was found an inverse correlation [97]. These data suggest a relevant implication of nesfatin-1 in the regulation of mood and stress in a sex-specific way.

Nesfatin-1 and sleep regulation

Two independent studies have related nesfatin-1 to sleep regulation in rat but with contradictory results. Nesfatin-1 i.c.v. injection in rats has been described to increase both the cumulative rapid eye movement (REM) sleep quantity and the mean number of REM sleep bouts (while nesfatin-1 blockage has the opposite effect), and nesfatin-1-expressing neurons activation from the tuberal hypothalamic area (THA) are positively correlated to REM sleep amounts [98]. On the contrary, REM sleep abolition has been shown to decrease nesfatin-1/NUCB2 protein and mRNA levels in the dorsolateral hypothalamic nucleus, while sleep rebound has the opposite effect, and i.c.v. administered nesfatin-1 increases sleep fragmentation and causes a decrease in total sleep time [99]. The authors explain this discrepancy by the different doses used as well as the different timing of the i.c.v. nesfatin-1 administration [98].

Nesfatin-1 and reproductive function

Different studies have suggested that the proper nesfatin-1 signaling is necessary to the development of normal puberty and to an adequate reproductive function in different species [41, 46, 49, 100–102]. In non-obese children and adolescents, nesfatin-1 circulating levels increase with puberty progression [78]. In female rats in the puberty, central nesfatin-1 injection increases circulating levels of luteinizing hormone (LH) and follicle-stimulating hormone (FSH) in both ad libitum and fasting female rats, an effect that is not observed in adult female rats [100]. However, in another study, it was shown that nesfatin-1 at a higher dose increases circulating levels of LH and FSH in male adult rats [103], suggesting that nesfatin-1 can modulate the gonadotropic axis not only in puberty but also in adult rats.

In a study including 22 girls (mean age 6.40 ± 1.58 years) with premature thelarche (PT), defined as isolated breast development in girls before the age of 8 years without axillary and pubic hair development, accelerated growth, advanced bone maturation, or menarche, and 24 healthy pre-pubertal controls (mean age 6.12 ± 0.93 years), nesfatin-1 circulating levels were found to be higher in the PT group compared to controls, and nesfatin-1 levels did not correlate with basal LH, basal FSH, stimulated peak LH, peak FSH levels, and anthropometric variables in the PT group [104].

In women with polycystic ovary syndrome (PCOS), commonly characterized by obesity, insulin resistance, hyperandrogenemia, and hirsutism, two independent studies have shown contradictory results on nesfatin-1 plasma levels, being found decreased in one (in 30 PCOS Turkish women and 30 age- and BMI-matched controls) [101] and increased in another (in 55 PCOS and 28 healthy Turkish women matched in age) [105]. In spite of the contradictory results, these studies postulate that nesfatin-1 could be an important player in the development of this syndrome.

Nesfatin-1 effects at cardiovascular level

Nesfatin-1 is widely expressed in different murine hypothalamic nuclei implicated in the regulation of the cardiovascular function, such as the ARC [15, 106], PVN [15, 107], LHA [15, 108], SON [15, 109], dorsal motor nucleus of the vagus (DMV) [16, 110], central nucleus of amygdala (CEA) [90, 111], insular cortex (INS) [90, 112], nucleus ambiguous [90, 113], or NTS [89, 114]. This nesfatin-1 distribution at central level suggests that it could be an important player in the regulation of the cardiovascular function and led many researchers to focus their effort to find the mechanisms through which nesfatin-1 could participate in cardiovascular homeostasis. Moreover, given the relationship between the adipose tissue function/accumulation and the cardiovascular risk, and the fact that nesfatin-1 is expressed and secreted by the adipose tissue, it seems feasible that changes in nesfatin-1 levels due to changes in body fat mass could be of a great importance to the cardiovascular function, and it could be possible that nesfatin-1 is one of the players involved in the impact of obesity on CVDs.

Nesfatin-1 effects on blood pressure and heart rate

Nesfatin-1 administration into the lateral ventricle in rats has been shown to increase blood pressure at the same range as other food intake-regulating peptides with cardiovascular actions, such as prolactin or orexin, an effect that is reversed by the melanocortin receptor (a G-protein-coupled receptor) antagonist SHU9119 and the nonselective α-adrenergic antagonist phentolamine [115]. This observation suggests that nesfatin-1 could act through the MCR3/4 to activate pre-autonomous centers like the NTS to increase the sympathetic nervous system activity and the blood pressure, an effect already described for the MCR3/4 activator MSH-α [116, 117]. Thus, nesfatin-1 blood pressure regulation could be mediated by the MCR3/4

signaling, as it has already been demonstrated for its anorexigenic effect [15]. On the other hand, nesfatin-1 is co-expressed with oxytocin in the PVN [89], another food intake-regulating peptide with effects at cardiovascular level [118, 119], and induces the activation of oxytocin-producing hormones [120]. When the oxytocin receptor (another G-protein-coupled receptor) is inhibited with ornithine vasotocin, the effects of nesfatin-1 on increasing blood pressure and anorexia are reverted, as well as the anorexigenic effect of MCR3/4 signaling induced by MSH-α, suggesting that the presence of functional oxytocin receptors is needed to the hypertensive and anorexigenic action of nesfatin-1 and that the oxytocin signaling could be a downstream mediator of the melanocortin system [121]. Both MCR3/4 and oxytocin receptors are expressed on neurons of the NTS [122, 123], and it was shown that the specific microinjection of nesfatin-1 in the NTS induces the increase of blood pressure and heart rate, while injections into sites outside of the NTS did not produce any changes in cardiovascular parameters [120]. However, more studies are needed to confirm the possible role of MCR3/4 and oxytocin receptors in the NTS on the cardiovascular effects triggered by nesfatin-1.

Nesfatin-1 immunoreactivity has been detected in the nucleus ambiguous in rats [90], where premotor cardiac vagal neurons play an essential role in heart rate regulation [113]. Nesfatin-1 injection in this nucleus induces an increase in intracellular Ca^{2+} concentrations in cardiac preganglionic neurons through a signaling cascade that involves G-protein-coupled receptors activation and P/Q-type Ca^{2+} channels opening, but in this study, the authors reported a mechanism independent of MCR3/4 and oxytocin receptors [124]. As well, nesfatin-1 injection on the nucleus ambiguous decreases heart rate without any change in blood pressure [124].

The sympathetic neural network that innervates the kidneys is involved in blood pressure regulation through the renin-angiotensin system [125]. Central injection of some anorexigenic peptides such as orexin [126], neuropeptide Y (NPY) [127], or pituitary adenylate cyclase-activating polypeptide (PACAP) [128] can alter the renal sympathetic nerve activity with consequent implications on blood pressure. Likewise, i.c.v. injection of nesfatin-1 in rats increases renal sympathetic nerve activity through a mechanism dependent of MCR3/4 functional receptors and increases blood pressure without any changes in heart rate [129]. In agreement with this study, it has been probed that i.c.v. administered nesfatin-1 increases mean arterial pressure in normotensive rats and in animals subjected to hypotensive hemorrhage, as well as it induces bradycardia in normotensive and tachycardia in hemorrhaged rats, the increase of plasma catecholamine, vasopressin and renin concentrations in control animals, and potentiates the rise in arterial pressure and heart rate when are decreased by hemorrhage. These findings suggest that centrally administered nesfatin-1 enhances renal sympathetic activity and elevates vasopressin and renin concentrations to mediate its cardiovascular effects [130]. As well, i.c.v. administration of nesfatin-1 increases blood pressure and the activity of sympathetic nerves to the kidneys, liver, and WAT in an extracellular-signal-regulated kinase (ERK) 1/2-dependent manner [131]. All these findings indicate that central action of nesfatin-1 regulates the autonomic nervous system to maintain the cardiovascular function.

When nesfatin-1 is subcutaneously administered to mice, it significantly increases mean blood pressure, without affecting heart rate, through an α-adrenergic-independent but β-adrenergic-dependent mechanism [132]. The authors suggests that the blood pressure elevation induced by β-adrenergic receptor activation involves a combination of cardiac and intra-renal β-adrenergic activation, and they suggest that the central hypothalamic nesfatin-1 effect may not have a main role in the regulation of systemic blood pressure.

Regarding human studies, in a cohort of 80 Chinese subjects (41 men and 39 women; 31–74 years of age; BMI = 19.83–34.21 kg/m^2), 40 patients with hypertension and 40 age-matched healthy controls, nesfatin-1 plasma circulating levels were found to be higher in the hypertensive patients, after adjust the influence of age, sex, BMI and blood lipids, and a logistic regression analysis revealed that the plasma level of nesfatin-1 could predict the prognosis of hypertension [80]. However, in pregnant women with pre-eclampsia, characterized by hypertension, proteinuria, edema, and endothelial dysfunction, there exist contradictory results. In a study including 120 women with pre-eclampsia and 92 healthy normotensive pregnant women with the age, body mass index (BMI), and pregnancy duration similar to those of the patients, serum nesfatin-1 levels were found to be significantly decreased in women with pre-eclampsia compared to the healthy controls, being inversely correlated with the presence and severity of pre-eclampsia [133]. On the contrary, in another recent study including age-matched obese pre-eclamptic ($n = 32$), non-obese pre-eclamptic ($n = 32$), and non-obese normotensive healthy ($n = 32$) pregnant women, serum nesfatin-1 levels were increased in pre-eclamptic women, in both obese and non-obese pre-eclamptic groups [134].

Recently, it has been shown that high nesfatin-1 circulating levels may be associated with the increase of systolic and diastolic blood pressure values and heart rate in PCOS [135]. PCOS is a complex condition that affects women of reproductive age characterized by ovulatory dysfunction and androgen excess. Women with PCOS present higher prevalence of obesity, central adiposity, dyslipidemia, and

increased risk of T2DM and CVDs. PCOS is closely linked to functional derangements in adipose tissue and disturbed secretion of adipokines [136]. Increased nesfatin-1 circulating levels in these patients were suggested to induce hyperglycemia, insulin resistance, hyperandrogenism, luteinizing hormone peaks, and chronic inflammation [135].

Nesfatin-1 effects on endothelial function

Endothelial dysfunction is one of the mechanisms implicated in the development of high blood pressure [137], thus Yamawaki et al. (2012) studied the possible implication of nesfatin-1 on increasing blood pressure through the endothelial function modulation in rats [138]. The authors demonstrated that nesfatin-1 treatment of isolated mesenteric artery inhibits the relaxation induced by sodium nitroprusside through the impairment of cyclic guanosine monophosphate (cGMP) production. When sodium nitroprusside is intravenously administered induces concentration-dependent decreases in blood pressure, an effect that is also reverted by nesfatin-1 [138]. These observations suggest that nesfatin-1 may participate in the control of blood pressure by modulating the contractility of peripheral blood vessel in a direct manner.

Nesfatin-1 direct effects on heart physiology

Nesfatin-1 has been shown to be expressed at both mRNA and protein levels in murine [43, 44] and human heart tissue and in cultured cardiomyocytes at comparable concentrations to stomach or brain, its main sources of production [44]. Moreover, cultured rat cardiomyocytes can secrete nesfatin-1 [44]. Thus, the heart could be one of the main sources of nesfatin-1 and could contribute not only to an endocrine/autocrine/paracrine nesfatin-1 signaling but also to nesfatin-1 concentrations in the circulation. Apart from mammals, it was demonstrated the cardiac expression of the nesfatin-1 precursor NUCB2 in teleosts (*S. prenanti*), suggesting an important and phylogenetically conserved nesfatin-1 function at cardiac level.

Cardiac and circulating nesfatin-1 levels were found to be directly affected by diet in rats, as it also occurs in adipose tissue in mice [37, 44]. In high-fat diet-fed rats, nesfatin-1 mRNA and protein levels in heart atrium are increased when compared to normal diet-fed rats, while no differences were found in ventricle, and its cardiac levels correlate with the percentage of body fat and the circulating levels of nesfatin-1 [44]. The heart atrium is known to be a secretory tissue of peptides like the natriuretic peptides, which were initially identified as important blood pressure regulators but that recently have been related to the energy metabolism regulation [139, 140]. Thus, the atrium could respond under pathophysiological conditions to modulate the heart nesfatin-1 production and to contribute to the nesfatin-1 circulating concentrations.

In rat perfused heart, nesfatin-1 infusion affects basal cardiac performance through the induction of negative inotropism in a dose-dependent manner (0.1 pM–10 nM) and revealed by a decrease of left ventricular pressure and the maximum rate of left ventricle contraction [43]. On the contrary, nesfatin-1 induced a biphasic modulation of lusitropism, positive at lower concentrations and negative at higher concentrations. The fact that the cardiac effects of nesfatin-1 are evident from picomolar concentrations of the peptide (the same range that has been shown to induce water and food intake inhibition [15, 141, 142]) suggests that the heart has a high sensitivity to nesfatin-1 and it would be possible that the energy status could affect circulating and endogenous levels of nesfatin-1, and consequently the heart response to nesfatin-1 signaling. To note, in a recent study performed by Mazza et al. in goldfish (*C. auratus),* nesfatin-1 was found to induce a dose-dependent positive inotropism in isolated and perfused paced hearts with statistical significance from 1 pM to 10 nM, involving cAMP, protein kinase A (PKA), L-type Ca^{2+} channels (LTCCs), and SERCA2a pumps, as well as ERK1/2 and phospholamban (PLN) activity [143]. Despite the contradictory results, this finding supports the idea that the heart has a high sensitivity to nesfatin-1. Although it has been documented that nesfatin-1 can interact with G-protein-coupled receptors [16], the dependent negative inotropic and lusitropic effects of nesfatin-1 in rat heart were found to be independent of Gi/o protein-coupled receptors but dependent to the presence of functional atrial natriuretic peptide receptors (NPR-A) and nitric oxide production [43].

On the other hand, nesfatin-1 levels are decreased in heart tissue under ischemia/reperfusion injury [43]. As it occurs for different cardio depressive peptides [144, 145], nesfatin-1 has been shown to elicit cardioprotection acting as a post-conditioning agent against ischemia/reperfusion injury by inducing a significant reduction of the infarct size and a marked improvement of post-ischemic contractile function [43]. This protective effect seems to be mediated by mitochondrial potassium-dependent ATP channels and the pro-survival kinase protein kinase C (PKC)-ε [43]. In particular, PKC-ε can act on mitochondria to affect cellular survival by reducing both necrosis and apoptosis after isquemia/reperfusion injury [146, 147]. Although this finding could inspire the medical community to use nesfatin-1 as a protective post-conditioning treatment against ischemia/reperfusion injury, due to its wide range of effects in different tissues (including cardiovascular tissues), the failure in identifying the existence of a specific receptor for nesfatin-1, and the fact that it was described to have the ability to act through different types of receptors, the future

possible therapeutic application of nesfatin-1 should take into account all these aspects.

In the heart, Ca^{2+} influx via LTCCs is a multifunctional signal that triggers muscle contraction and controls action potential duration [148]. In cardiac extracts of rats subjected to restraint stress and chronic peripheral nesfatin-1 infusion, the L-type Ca^{2+} channel α1c subunit protein expression is significantly increased [149]. However, in cultured adult ventricular myocytes, nesfatin-1 decreases L-type Ca^{2+} currents by MCR4 and PKC-θ activation [150], suggesting that nesfatin-1 could modulate L-type Ca^{2+} channel activity under different pathophysiological conditions. Apart from the heart, nesfatin-1 was shown to evoke Ca^{2+} influx through activation of N-type Ca^{2+} channels within vagal afferent nodose ganglion neurons [151] and through LTCCs in β-cells of pancreatic islets in mice [152], as well as it increases the cAMP response element (CRE) reporter activity in a mouse neuroblastoma cell line [56] and activates oxytocinergic signaling in the PVN [153] through a dependent mechanism of L-type Ca^{2+} channels, suggesting that both central and peripheral effects of nesfatin-1 can be partly mediated by activation of Ca^{2+} channels, with important consequences to the cardiovascular system.

Nesfatin-1 direct effects on cardiomyocyte metabolism and viability

In cultured murine cardiomyocytes, short-term nesfatin-1 treatment has been demonstrated to induce AKT and ERK1/2 activation (an effect also observed in rat perfused hearts [43]), and the AKT substrate of 160 kDa (AS160) phosphorylation with the concomitant GLUT-4 translocation to the plasma membrane, which promotes an increase in glucose uptake [44]. However, whether or not this effect is beneficial or harmful to the heart needs to be proved. On the contrary, long-term exposures (24 h) to nesfatin-1 in neonatal rat cardiomyocytes have been shown to induce an increase in cell death through a mechanism involving the pro-survival AKT inactivation, the increase of the apoptogenic protein APOP-1 (involved in the opening of the mitochondrial permeability transition pore (mPTP) to promote apoptosis [154]) and caspase-3 activation [155].

Nesfatin-1 circulating levels in CVD

Apart from the different direct effects of nesfatin-1 in cardiac pathophysiology, under different CVDs its circulating levels are modified compared to controls, suggesting that modification in circulating nesfatin-1 levels could be involved in the development of CVDs.

In a study of 156 subjects, divided in patients with acute myocardial infarction, stable angina pectoris, and coronary artery disease without lesions (used as control), nesfatin-1 plasma levels are significant lower in those with acute myocardial infarction compared to the other groups [156], and in patients with paroxysmal supraventricular tachycardia, nesfatin-1 circulating levels are increased compared to controls [157].

Wang et al. described in a cohort of 355 non-obese T2DM patients (155 with peripheral arterial disease (PAD) and 200 without PAD) that serum nesfatin-1 levels are inversely correlated with the development and severity of PAD in T2DM patients, suggesting that serum nesfatin-1 may be utilized as a predicting biomarker for PAD disease risk [158].

In cardiac atrial tissue nesfatin-1/NUCB2 mRNA levels are significantly higher in women without coronary injury (CI) than in those with CI or in men with or without CI, suggesting a sex-dependent relationship between cardiac nesfatin-1 levels and CVD [44].

Conclusion

Nesfatin-1 is a new energy-regulating peptide widely expressed at both central and peripheral level that is involved in the regulation of different pathophysiological conditions. Although its most documented effects are food intake and energy homeostasis regulation, increasing reports suggest its implication in the development of different types of diseases, such as T2DM [32], hypertension [132], depression [159], polycystic ovary syndrome [105], sleep apnea obstructive syndrome [160], or osteoarthritis [161, 162]. Although there are a number of contradictory results regarding nesfatin-1 functions, it seems quite clear that nesfatin-1 is a pleiotropic peptide that induces anorexia, acts as an insulin coadyuvant to reduce glycemia, increases glucose utilization by peripheral tissues, and regulates gastric and cardiovascular function, and that its circulating and tissue levels are affected by nutritional status. On the contrary, there exist some gaps of knowledge to fill regarding nesfatin-1 role in anxiety, obesity, diabetes, pre-eclampsia, and PCOS pathophysiology, so that in some studies nesfatin-1 circulating levels are increased, while in others they are just the opposite, making it difficult to interpret the altered nesfatin-1 levels in these diseases, so further investigation is really needed to have a clear landscape regarding nesfatin-1 function and regulation.

Nesfatin-1 has been found to have important effects on the cardiovascular system and to modulate the cardiovascular function at different levels, being its circulating concentrations modified in some CVDs. (Figure 7). In the last years, adipokines have become promising candidates for both novel pharmacological treatment strategies and diagnostic tools, particularly at cardiovascular level [163].

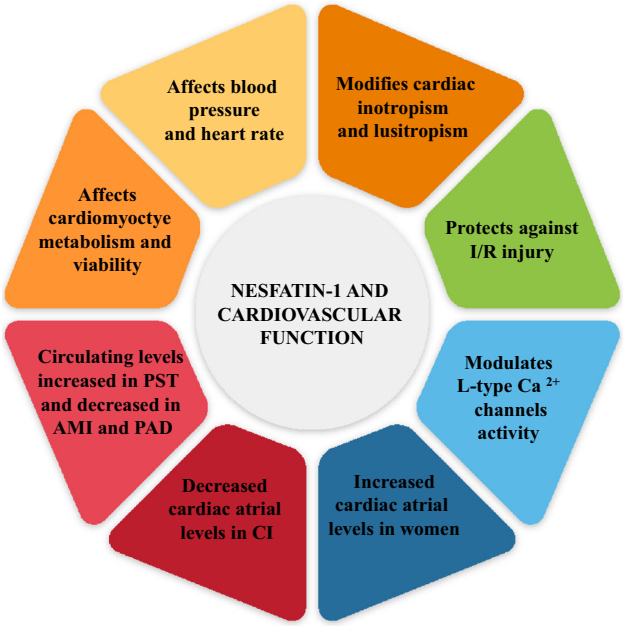

Fig. 7 Cardiovascular implications of nesfatin-1 on cardiac pathophysiology. *I/R* ischemia/reperfusion, *CI* coronary injury, *PST* paroxysmal supraventricular tachycardia, *AMI* acute myocardial infarction, *PAD* peripheral artery disease

Given the fact that nesfatin-1 is expressed and secreted by the adipose tissue, and that its secretion levels are affected by diet [37], it seems feasible that changes in nesfatin-1 levels due to changes in body fat mass could be of a great importance to the cardiovascular function, and it could be possible that nesfatin-1 is one of the players involved in the impact of obesity on CVDs. In fact, the heart seems to have a high sensitivity to nesfatin-1 [43, 143], and nesfatin-1 cardiac levels were found to be also modified by diet in rats [44]. The studies performed so far regarding nesfatin-1 function at cardiovascular level are just referred to nesfatin-1 direct actions on the heart/vasculature or to nesfatin-1 levels under different cardiovascular diseases. It would be of a great interest to elucidate if not only abdominal and subcutaneous fat (which were already proved to have different nesfatin-1 secretion in murine and humans [37]) but also epicardial and perivascular adipose tissue have a differential nesfatin-1 secretion pattern and their possible effect on cardiovascular function, at both systemic and local level.

On the other hand, nesfatin-1 is also expressed and secreted by β-pancreatic cells and it promotes insulin release and signaling, so nesfatin-1 deregulation can also affect the development of insulin resistance and T2DM, both of them well known cardiovascular risk factors. In this line, it would be of a great interest to study the possible effect of nesfatin-1 on heart performance under situations of metabolic syndrome or insulin resistance/T2DM and to describe its circulating levels in patients with coronary injury, peripheral arterial disease, or acute myocardial infarction (all of them diseases closely related to metabolic syndrome and proven to have decreased nesfatin-1 levels [44, 156, 158]) before and after treatment to determine whether or not nesfatin-1 could be used as a diagnosis tool in these diseases.

It is important to note that the possibility of considering nesfatin-1 as a therapeutic target carries the handicap of its multiple cellular signaling cascades in different organs. Despite the efforts of the scientific community to discover the specific nesfatin-1 receptor, it has not been identified yet, while different studies showed that nesfatin-1 has the ability to bind and activate different type or receptors, so it is likely that the search for the specific receptor for nesfatin-1 could lead us to a dead end, and this ability to activate different receptors could be the reason why nesfatin-1 can act at different tissues in a differentiated way. Hence, it is imperative to dig deeply in the specific mechanisms implicated in its signaling in each field of study where it was demonstrated to have important effects, such at cardiovascular, reproductive or gastric level, to help the scientific community to try to develop specific drugs to modulate nesfatin-1 effects at local level and to better understand the possible consequences of its modulation on non target organs.

Compliance with ethical standards

Conflict of interest None declared.

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