The Egyptian Journal of Hospital Medicine (April 2023) Vol. 91, Page 4725-4730

Proadrenomedullin as a Cardiac Biomarker in Prediction in Pediatric Heart Failure: Review article

Mohamed El Sayed Amer[1], Besheir Abd Alla Hassan[1],
Mohamed Milad Abdul Hamid Enbaya [2], Hanan Samir Ahmad [1]
Department of Pediatrics[1], Medical Biochemistry [2], Faculty of Medicine, Zagazig University, Egypt
***Corresponding author:** Mohamed Milad Abdul Hamid Enbaya, **Mobile:** (+20) 01288023029,
E-mail: Mohamedmiladabdulhamid@gmail.com

ABSTRACT
Background: Children's morbidity and death from heart failure continue to be major contributing factor. The evaluation of several biomarkers may serve as a valuable diagnostic aid.
Objective: To evaluate the role of Proadrenomedullin in detection of heart failure.
Conclusion: There is a high diagnostic value of measuring plasma level of proADM at admission, regarding the diagnosis in the setting of pediatric congestive heart failure, and can be used as prognostic value after treatment.
Keywords: Proadrenomedullin, Heart Failure, Pediatric.

INTRODUCTION

During the screening of a panel of peptides taken from a pheochromocytoma by a research team in Japan, they came across the regulatory peptide. They were examining the biological activity of the peptides to see if they may raise platelet cAMP levels. It originated from the adrenal medulla, the researchers named the peptide they discovered with this activity "adrenomedullin" after purifying and sequencing it. The rat gene was sequenced after the gene encoding human adrenomedullin. The first putative adrenomedullin receptor had been discovered, a new field of endocrine research had begun, and plasma adrenomedullin levels had been measured in a variety of clinical settings **(1).**

Synthesis and secretion of adrenomedullin:

One disulfide link exists between residues 16 and 21 of the 52-amino acid peptide known as human adrenomedullin, and the carboxy terminus contains an amidated tyrosine. Due to some similarities to calcitonin gene-related peptide, it has been added to the calcitonin/CGRP/amylin peptide family (CGRP). 50 amino acids make up rat adrenomedullin, which has the human peptide has 6 alterations and 2 deletions **(2).** Only one difference separating pig adrenomedullin from the human peptide occurs at position 40 (Gly for Asn). The amino acid sequences of adrenomedullin from several species are compared in figure (1). Additionally, the canine and bovine adrenomedullin sequences have been clarified **(3).**

Pre-proadrenomedullin, a more substantial precursor molecule, is used in the synthesis of adrenomedullin. A 21-amino acid N-terminal signal peptide immediately follows the proadrenomedullin N-terminal 20 peptide, or PAMP found in pre-proadrenomedullin. Additionally, it has been hypothesized that the adrenomedullin gene may produce another biologically active peptide known as adrenotensin, but this has not yet been proven **(4).**

Adrenomedullin is the name of the gene that produces pre-proadrenomedullin, and it has been identified as being on chromosome 11 at a single locus. Three introns and four exons make up the human adrenomedullin gene. The TATA, CAAT, and GC boxes can be found in the gene's 5′-flanking region (figure). Both the cAMP-regulated enhancer element and the activator protein-2 (AP-2) contain a lot of binding sites. Furthermore, Nuclear Factor-B (NF-B) sites have been found in the promoter of the adrenomedullin gene. The structure and chromosomal location of the mouse adrenomedullin gene location have also been determined **(2).**

Received: 29/11/2022
Accepted: 01/02/2023

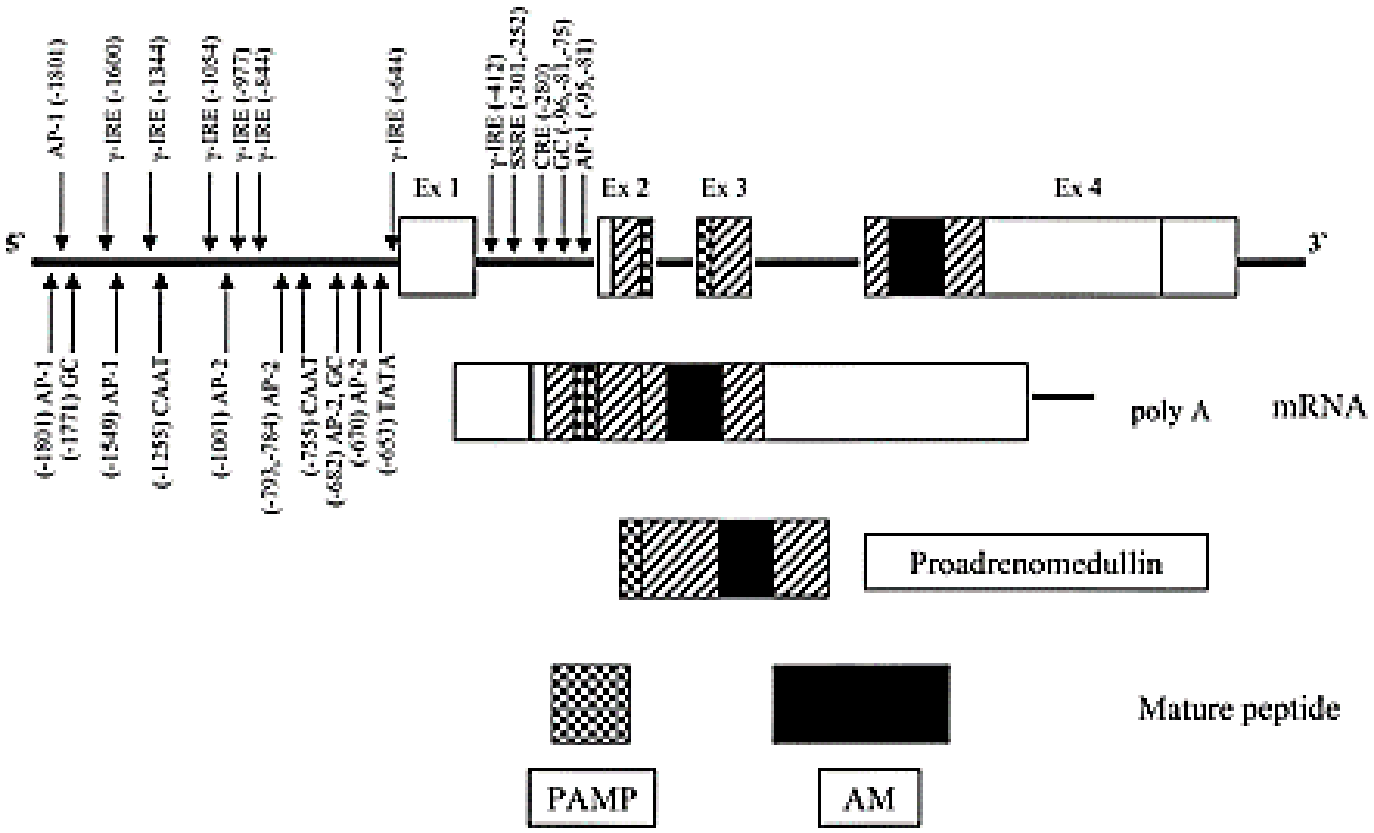

Figure (1): Schematic presentation of the adrenomedullin gene, pre-proadrenomedullin, and biosynthesis of adrenomedullin (The genomic DNA of human adrenomedullin comprises four exons and three introns, and mature adrenomedullin peptide is coded in the fourth exon. **Arrows** indicate known regulatory sequences in the 5′-noncoding and intron 1 region of the human adrenomedullin gene).

Receptors of pro-adrenomedullin:

Presently, it is thought that CGRP receptors, AM is produced by a combination of Calcitonin Receptor like Receptor (CALCRL) receptors and Receptor Activity-Modifying Proteins (RAMP2) complexes. Receptor Activity-Modifying Protein (RAMP) and Calcitonin Receptor-Like (CALCRL) interaction at the membrane is required for AM to mediate its activity, in contrast to the traditional one ligand-one receptor paradigm of receptor signaling. Cyclic AMP (cAMP) and nitric oxide are produced in the cell as a result of AM stimulating its receptor. Since they frequently have opposing effects, the production of these inside the cell may seem contradictory to some, but the timing of these effects still needs to be investigated [(5)].

Circulating pro-adrenomedullin:

Following the initial discovery of picomolar plasma pro-adrenomedullin levels, **Lewis *et al.*** [(6)] have created internal assays to assess plasma pro-adrenomedullin concentrations. The evidence according to HPLC analysis, authentic human pro-adrenomedullin coelutes with immunoreactive pro-adrenomedullin from plasma, indicating that these techniques have generally been properly validated.

The primary pro-adrenomedullin that circulates in the body is a carboxy-terminally extended peptide that undergoes enzymatic amidation to become mature pro-adrenomedullin. When preprop-adrenomedullin is digested, a glycine-extended peptide known as the intermediate form is produced. Pro-adrenomedullin with glycine extensions (iAM) has been found to have a plasma concentration of 2.7 ± 0.18 pm, with a concentration of mature pro-adrenomedullin (mAM) of 0.48 ± 0.05 pm [(7)].

Both iAM and mAM have been seen to increase in congestive heart failure; the iAM to mAM ratio does not change appreciably. It looks likely that the majority of assays used for determining plasma As far as the range of values indicated, pro-adrenomedullin are measuring either total or iAM pro-adrenomedullin. Unless additional research reveals a significant difference between mAM levels and total pro-adrenomedullin, this situation would appear to be sufficient. Pro-adrenomedullin serum-Binding Protein (AMBP-1), whose recent discovery may

affect pro-adrenomedullin bioavailability, is one aspect that might contradict this theory [8].

Rat plasma has also been used to assess pro-adrenomedullin using an antiserum that can distinguish between the rat and human forms of the hormone. Rat plasma pro-adrenomedullin levels are similar to those discovered in humans, at 3.6 ± 0.34 pm [6].

Origins of circulating pro-adrenomedullin:

Most bodily tissues generate pro-adrenomedullin. Despite the fact that the gene producing pro-adrenomedullin is highly expressed in both the zona glomerulosa and the adrenal medulla, there is substantial evidence against the adrenal being the primary source of circulating peptide. Contrary to epinephrine and norepinephrine, which have distinct trans-adrenal gradients, pro-adrenomedullin levels in adrenal venous samples are comparable to those in arterial plasma [9].

Insulin-induced hypoglycemia has been utilized to stimulate the production of medullary pro-adrenomedullin, however despite a 20-fold increase in plasma epinephrine levels, no appreciable change in the amount of circulating pro-adrenomedullin was seen. Additionally, plasma pro-adrenomedullin concentration was elevated in a patient with pheochromocytoma during a hypotensive attack remained unchanged even though epinephrine and norepinephrine concentrations significantly increased [10].

Because pro-adrenomedullin is cosecreted with catecholamines, at least by bovine chromaffin cells in culture, the adrenal medulla is unlikely to be a significant source of circulating pro-adrenomedullin. Selective arterial and venous sampling across many circulatory beds (including the heart, lungs, kidney, and adrenals) in people with a variety of cardiovascular illnesses was unable to identify a source of significant pro-adrenomedullin production. Congestive heart failure has been linked to considerable pro-adrenomedullin cardiac secretion. Nevertheless, pro-adrenomedullin plasma levels have reportedly increased concentration in several disease states, most notably cerebrovascular disease, is assumed to represent the severity of endothelial cell injury [11].

It is difficult to determine whether generally elevated levels of plasma pro-adrenomedullin show a rise in production or a decline in clearance. This problem hasn't received any particular attention. Although, there is proof that numerous cell types produce more pro-adrenomedullin during septic shock. Additionally, there is a proof of elevated pro-adrenomedullin cardiac production in congestive heart failure [6].

Metabolic clearance of pro-adrenomedullin:

The plasma half-life of pro-adrenomedullin has been reported to be 22 ± 1.6 minute with MCR of 27.4 ± 3.6 ml/kg. Investigations have been done into how plasma membrane enzymes affect pro-adrenomedullin. It looks likely that metalloproteases first break down pro-adrenomedullin to produce pro-adrenomedullins 8-52, 26-52, and 33-52, followed by an aminopeptidase action to yield pro-adrenomedullins 2-52, 27-52, and 28-52. It has been hypothesized that a significant location of pro-adrenomedullin clearance in men may be the lung [12].

Pro-adrenomedullin in other biological fluids:

Pro-adrenomedullin has been found in peripheral plasma, as well as considerable amounts in urine, milk, cerebrospinal fluid (CSF), saliva, sweat, amniotic fluid, and umbilical vein blood. Pro-adrenomedullin concentrations in the urine in healthy persons have been shown to be roughly six times greater than plasma levels [13].

The kidney is not considered to be a major location of pro-adrenomedullin excretion because there is no link between urine and plasma levels. However, it has been observed that a drop in urine pro-adrenomedullin and an increase in plasma peptide levels are linked to renal diseases, such as IgA nephropathy, and may signify inefficient excretion. However, the higher levels of urinary pro-adrenomedullin found in healthy subjects when compared to plasma may indicate that the kidney is the main source of urinary pro-adrenomedullin [14].

Pro-adrenomedullin levels in CSF are stable during pregnancy while plasma levels rise, showing independent regulation of pro-adrenomedullin in the two compartments where levels in CSF are lower than those in plasma [15].

It has been established that pro-adrenomedullin is present in murine milk, but the concentration has not yet been determined. The fact that it is actively secreted into milk is therefore unknown. There is evidence that pro-adrenomedullin is produced by the skin and kidneys and that pro-adrenomedullin concentrations in urine and sweat are much greater than those in plasma [16].

Regulation of pro-adrenomedullin gene expression and peptide synthesis in vivo:

Investigations into how different physiological changes affect the levels of plasma pro-adrenomedullin in humans and other species have been conducted. Some studies, have suggested that exercise increases plasma pro-adrenomedullin in men, with plasma pro-adrenomedullin and blood pressure are connected. Additionally, going from a low to a high altitude linked to a rise in plasma pro-adrenomedullin, which is probably connected to the patients' level of hypoxia [17].

Both hemorrhagic shock similar to how they do in humans, and endotoxic stress increase pro-adrenomedullin gene expression in blood vessels in dogs. Consistently, rat pro-adrenomedullin gene expression and

plasma levels increase in experimentally-produced sepsis. Pro-adrenomedullin concentration in the digestive system rises during period of fasting in rats [18].

Rats and women both had higher circulation pro-adrenomedullin concentrations during pregnancy. It has been claimed that the plasma concentration of pro-adrenomedullin grows progressively from the first to third trimesters, with an extra increase postpartum. All these data have been questioned due to the extremely high quantities of pro-adrenomedullin found in this study [19]. **DiIorio *et al.*** [20] showed that whereas plasma pro-adrenomedullin levels in pregnant women were on average five times greater than those in women who are not pregnant. There was no association with gestational age where plasma pro-adrenomedullin concentrations had dramatically dropped by 48 hours after delivery. An intriguing finding was that kids delivered vaginally had much higher pro-adrenomedullin levels in their umbilical cords than in infants delivered by elective Cesarean delivery [21].

By administering progesterone derivative to rats, the effects of pregnancy on plasma pro-adrenomedullin concentrations can be mimicked, indicating that the elevated pro-adrenomedullin plays a part in the cardiovascular alterations associated with pregnancy. Pregnancy has been demonstrated to greatly increase rat uterine pro-adrenomedullin mRNA suggesting that the uterus may be the source of plasma pro-adrenomedullin [22].

Investigations have also been done into the results of different endocrine interventions. Pro-adrenomedullin plasma concentrations and lung pro-adrenomedullin mRNA levels were shown to be elevated in hyperthyroid rats [23]. Pro-adrenomedullin is regulated by glucocorticoids as well replacement of glucocorticoids decreased plasma pro-adrenomedullin levels in (Primary Adrenal Insufficiency) Addison's illness patients. However, despite being a strong stimulant of glucocorticoid secretion, insulin-induced hypoglycemia had no effect on the levels of plasma pro-adrenomedullin [10].

Dexamethasone had no influence on lung mRNA levels or plasma pro-adrenomedullin in septic shock rats, but it significantly increased both in control adrenalectomized animals. Castration causes a 25-fold decrease in mRNA for the pro-adrenomedullin gene in the ventral prostate of rats, but treatment of androgen completely restores this drop [24].

Hemodialysis has been demonstrated to lower pro-adrenomedullin concentrations in individuals with renal illness. Also, pro-adrenomedullin plays a role in the control of fluid and electrolyte status [25].

It was discovered that altering the renin-angiotensin system with captopril or furosemide, as well as an infusion of ACTH, in healthy individuals had no impact on plasma pro-adrenomedullin [26]. Similarly, it has been discovered that giving rats diets rich (4%) or low (0.02%) in salt has no impact on the expression of the gene for renal pro-adrenomedullin. However, in contrast to those on a control diet, the Dahl salt-sensitive rat strain showed increased plasma and ventricular pro-adrenomedullin when fed a high-salt diet [27]. Acute or chronic alterations in salt intake in humans had no impact on the plasma levels of pro-adrenomedullin in either normotensive or hypertensive participants [28].

Healthy individuals have no impact on plasma pro-adrenomedullin. Pro-adrenomedullin levels were measured in this study by drawing blood every 30 minutes for 5 hours. The control individuals' plasma peptide levels did not appear to alter during this time, whereas the test subjects' plasma pro-adrenomedullin concentration was enhanced for only the first 60 minutes after the infusion. Within 20 minutes within 4 hours of the infusion beginning, a steady-state was reached, and levels returned to basal after 30 minutes of the infusion ending [29].

The control of pro-adrenomedullin in rats has also been studied using two pressure overload models. Hormonal-induced pressure overload using either arginine vasopressin (AVP) or angiotensin II both increased cardiac pro-adrenomedullin mRNA and peptide levels. Despite a significant rise in atrial natriuretic peptide, the surgical model had no impact on pro-adrenomedullin expression [30].

Physiological functions of pro-adrenomedullin:

AM was first discovered to be a vasodilator; some claim It is the strongest endogenous vasodilatory peptide in the body. The different model systems being employed give rise to divergent views on AM's capacity to relax vascular tone. The ability of cells to withstand oxidative stress, hypoxia damage, and angiogenesis are among AM's additional effects. AM is thought to have a beneficial effect on conditions similar to hypertension, myocardial infarction, chronic obstructive pulmonary disease, and other cardiovascular conditions, but it might impair the capacity of malignant cells to expand their blood supply and induce cell growth [31].

Proadrenomedullin in children with heart failure:

Circulating biomarkers may be crucial when patients who present with heart failure are diagnosed and treated, which improves case prognosis. Utilizing novel biomarkers in conjunction with existing ones has been shown to significantly improve prognostic and diagnostic accuracy. The complicated HF pathophysiology is often reflected by a variety of novel biomarkers, such as fibrotic remodeling, myocardial strain, or inflammation [32].

In a number of cardiovascular diseases, adrenomedullin (ADM) may have a preventive effect. In HF, resulting from ventricular wall stretching and pressure/volume overload, the ADM gene is induced in cardiac myocytes. The consequent elevated ADM levels seem to have a protective impact on the myocardium as preload and after load are decreased. ADM also prevents cell hypertrophy and proliferation, and it has been linked to decreased remodeling and fibrosis. Since ADM has a brief half-life and is swiftly eliminated from the bloodstream, nonetheless due to in vitro instability, its clinical application has been restricted for some time. Mid regional proadrenomedullin (MR-proADM), a stable pro-adrenomedullin fragment whose levels resemble those of ADM, was used to alleviate this issue [(1)].

Ebara *et al.* [(33)] reported that AM had a natriuretic effect in anesthetized dogs with an increase in renal blood flow, most likely at the physiological plasma level of the AM. AM may function defensively by working against further deterioration of heart failure as a result of the reduction of plasma AM in response to the treatment in aggravated heart failure. This is due to the AM's robust vasodilator and natriuretic activities in patients with heart disorders.

Shimosawa *et al.* [(34)] shown that PAMP reduces catecholamine release from sympathetic nerve terminals in pithed rats. The fact that plasma PAMP was elevated in heart failure patients but dropped following treatment suggests that this substance also regulates the cardiovascular system, most likely through regulating catecholamine release from the sympathetic nervous system.

Etoh *et al.* [(35)] By evaluating the levels of plasma AM and PAMP in 98 individuals with heart failure, it was possible to explore the connections between the two peptides and other clinical parameters. They came to the conclusion that plasma AM and PAMP levels were both increased and lowered in response to therapy in patients with exacerbated heart failure and in those with congestive heart failure. Additionally to their divergent biological effects, AM and PAMP's hormonal profiles in the patients looked to be different from one another, pointing to their distinct pathophysiologic functions. Thus, in patients with heart failure, the cardiovascular system is modulated by a unique dual AM and PAMP system.

Salem *et al.* [(36)] examined the application of MR-proADM plasma levels at admission for heart failure (HF) diagnosis and progression in children with CHD. In the context of juvenile HF, they discovered that evaluating high diagnostic value was provided by the on-admission level of MR-proADM in connection to clinical severity and disease progression.

In study of **Zamzam *et al.*** [(37)], pediatric plasma levels of the promising substance were measured in congestive heart failure patients. Novel diagnostic cardiac biomarker MR-proADM were examined, along with how it is related to various clinical, laboratory, and echocardiographic factors. They came to the conclusion that monitoring the diagnosis of pediatric congestive heart failure was highly specific for the plasma level of MR-proADM at the time of admission.

Sources of funding: This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.
Conflicts of interest: There are no conflicts of interest, according to the authors.

REFERENCES

1. **Jayaprasad N (2016):** Heart failure in children. Heart Views: the Official Journal of the Gulf Heart Association, 17 (3): 92-96.
2. **Okazaki T, Ogawa Y, Tamura N *et al.* (2006):** Genomic organization, expression, and chromosomal mapping of the mouse adrenomedullin gene. Genomics, 37: 395–399.
3. **Ono Y, Kojima M, Okada K *et al.* (2008):** cDNA cloning of canine adrenomedullin and its gene expression in the heart and blood vessels in endotoxin shock. Shock, 10: 243–247.
4. **Gumusel B, Chang K, Hyman A *et al.* (2005):** Adrenotensin: an ADM gene product with the opposite effects of ADM. Life Sci., 57: L87–L90.
5. **Cheng M, Wallis C, Shou Z *et al.* (2007):** Quantifying angiogenesis in VEGF-enhanced tissue-engineered bladder constructs by dynamic contrast-enhanced MRI using contrast agents of different molecular weights. J Magn Reson Imaging, 25 (1): 137–145.
6. **Lewis K, Smith W, Yandle G *et al.* (2008):** Adrenomedullin(1–52) measured in human plasma by radioimmunoassay: plasma concentration, adsorption, and storage. Clin Chem., 44: 571–577.
7. **Kitamura K, Kato J, Kawamoto M *et al.* (2008):** The intermediate form of glycine-extended adrenomedullin is the major circulating molecular form in human plasma. Biochem Biophys Res Commun., 244: 551–555.
8. **Hirayama N, Kitamura K, Imamura T *et al.* (2009):** Molecular forms of circulating adrenomedullin in patients with congestive heart failure. J Endocrinol., 160: 297–303.
9. **Kato J, Kitamura K, Eto T (2008):** Adrenomedullin in cardiovascular and renal diseases. In: Martinez A, Cuttitta F (eds). Adrenomedullin. IOS Press, Amsterdam.
10. **Ehlenz K, Koch B, Preuss P *et al.* (2007):** High levels of circulating adrenomedullin in severe illness: correlation with C-reactive protein and evidence against the adrenal medulla as site of origin. Exp Clin Endocrinol Diabetes, 105: 156–162.
11. **Hoeldtke N, Wagner K, Moore H *et al.* (2008):** Adrenomedullin concentrations in umbilical cord plasma of uncomplicated term pregnancies. Am J Obstet Gynecol., 179: 1071–1074.
12. **Lewis K, Smith W, Brennan O *et al.* (2007):** Degradation of human adrenomedullin(1–52) by plasma membrane

enzymes and identification of metabolites. Peptides, 18: 733–739.

13. **Dotsch J, Hanze J, Knufer V *et al.* (2008):** Increased urinary adrenomedullin excretion in children with urinary-tract infection. Nephrol Dial Transpl., 13: 1686–1689.
14. **Sato K, Hirata Y, Imai T *et al.* (2005):** Characterization of immunoreactive adrenomedullin in human plasma and urine. Life Sci., 57: 189–194.
15. **Nagata N, Kato J, Kitamura K *et al.* (2008):** Dissociation of adrenomedullin concentrations in plasma and cerebrospinal fluid in pregnant and non-pregnant women. Eur J Endocrinol., 139: 611–614.
16. **Jahnke D, Miller J, Martinez A *et al.* (2007):** Adrenomedullin expression in the mouse mammary gland: evidence for the mature form in milk. J Mol Endocrinol., 19: 279–289.
17. **Fujioka S, Ono Y, Kangawa K *et al.* (2009):** Plasma concentration of adrenomedullin is increased in hemorrhagic shock in dogs. Anesth Analg., 88: 326–328.
18. **Wang P, Zhou M, Ba F *et al.* (2008):** Up-regulation of a novel potent vasodilatory peptide adrenomedullin during polymicrobial sepsis. Shock, 10: 118–122.
19. **Hattori Y, Murakami Y, Atsuta H *et al.* (2008):** Glucocorticoid regulation of adrenomedullin in a rat model of endotoxic shock. Life Sci., 62: L181–L189.
20. **DiIorio R, Marinoni E, Letizia C *et al.* (2008):** Adrenomedullin, a new vasoactive peptide, is increased in preeclampsia. Hypertension, 32: 758–763.
21. **Boldt T, Luukkainen P, Fyhrquist F *et al.* (2008):** Birth stress increases adrenomedullin in the newborn. Acta Paediatr., 87: 93–94.
22. **Upton D, Austin C, Taylor M *et al.* (2007):** Expression of adrenomedullin (ADM) and its binding sites in the rat uterus: increased number of binding sites and ADM messenger ribonucleic acid in 20-day pregnant rats compared with nonpregnant rats. Endocrinology, 138: 2508–2514.
23. **Murakami Y, Hattori Y, Taniyama M *et al.* (2008):** Adrenomedullin regulation by thyroid hormone in the rat. Life Sci., 63: L337–L342.
24. **Pewitt B, Haleem R, Wang Z (2009):** Adrenomedullin gene is abundantly expressed and directly regulated by androgen in the rat ventral prostate. Endocrinology, 140: 2382–2386.
25. **Mallamaci F, Zoccali C, Parlongo S *et al.* (2008):** Plasma adrenomedullin during acute changes in intravascular volume in hemodialysis patients. Kidney Int., 54: 1697–1703.
26. **Kita T, Kitamura K, Kuwasako K *et al.* (2008):** Short-term modulation of the renin-angiotensin system does not alter plasma adrenomedullin concentration in humans. J Hypertens., 16: 2057–2062.
27. **Shimokubo T, Sakata J, Kitamura K *et al.* (2006):** Adrenomedullin: changes in circulating and cardiac tissue concentration in Dahl salt-sensitive rats on a high-salt diet. Clin Exp Hypertens., 18: 949–961.
28. **Ishimitsu T, Nishikimi T, Matsuoka H *et al.* (2006):** Behaviour of adrenomedullin during acute and chronic salt loading in normotensive and hypertensive subjects. Clin Sci Colch., 91: 293–298.
29. **Vesely L, Blankenship M, Douglass A *et al.* (2006):** Atrial natriuretic peptide increases adrenomedullin in the circulation of healthy humans. Life Sci., 59: 243–254.
30. **Kaiser M, Kahr O, Shimada Y *et al.* (2008):** Differential regulation of ventricular adrenomedullin and atrial natriuretic peptide gene expression in pressure and volume overload in the rat. Clin Sci., 94: 359–365.
31. **Hamid A, Baxter F (2005):** Adrenomedullin. Pharmacol Ther., 105 (2): 95-112.
32. **Yancy C, Jessup M, Bozkurt B *et al.* (2013):** ACCF/AHA guideline for the management of heart failure: a report of the American College of Cardiology Foundation/American Heart Association Task Force on Practice Guidelines. Circulation, 128: 240–327.
33. **Ebara T, Miura K, Okumura M *et al.* (1994):** Effect of adrenomedullin on renal hemodynamics and functions in dogs. Eur J Pharmacol., 263: 69-73.
34. **Shimosawa T, Ito Y, Ando K *et al.* (1995):** Proadrenomedullin NHz-terminal20 peptide, a new product of the adrenomedullin gene, inhibits norepinephrine overflow from nerve endings. J Clin Invest., 96: 1672-1676.
35. **Etoh T, Kato J, Takenaga M *et al.* (1999):** Differential hormonal profiles of adrenomedullin and proadrenomedullin N-terminal 20 peptide in patients with heart failure and effect of treatment on their plasma levels. Clin. Cardiol., 22: 113-117.
36. **Salem S, Saleh Y, Soliman S *et al.* (2021):** On-admission plasma levels of BNP, MR-proADM, and cTnI in pediatric heart failure: Contributions to diagnosis, prognosis, and outcome. Irish Journal of Medical Science, 191 (20): 555-562.
37. **Zamzam F, Shedeed A, Mohammed M *et al.* (2022):** Mid-regional proadrenomedullin as a diagnostic marker in pediatric heart failure. The Egyptian Journal of Hospital Medicine, 87: 1276-1280.