frontiers in Physiology

REVIEW
published: 12 December 2017
doi: 10.3389/fphys.2017.01035

Effects of Apelin on Cardiovascular Aging

Ying Zhou[1]*, *Yong Wang*[1]*, *Shubin Qiao*[2] and *Liang Yin*[3]

[1] Department of Cardiology, China-Japan Friendship Hospital, Beijing, China, [2] Department of Cardiology, Cardiovascular Institute of Fuwai Hospital, Chinese Academy of Medical Sciences, Peking Union Medical College, Beijing, China, [3] School of Science, Beijing University of Chemical Technology, Beijing, China

Apelin is the endogenous ligand of APJ, the orphan G protein-coupled receptor. The apelin–APJ signal transduction pathway is widely expressed in the cardiovascular system and is an important factor in cardiovascular homeostasis. This signal transduction pathway has long been related to diseases with high morbidity in the elderly, such as atherosclerosis, coronary atherosclerotic heart disease, hypertension, calcific aortic valve disease, heart failure and atrial fibrillation. In this review, we discuss the apelin–APJ signal transduction pathway related to age-associated cardiovascular diseases.

Keywords: Apelin, cardiovascular diseases, aging, RAAS, Atherosclerosis/CAD

OPEN ACCESS

Edited by:
Mingyi Wang,
National Institutes of Health (NIH), United States

Reviewed by:
Jingyan Han,
Boston University, United States
Daniela Carnevale,
Sapienza Università di Roma, Italy
Gianfranco Pintus,
Qatar University, Qatar

***Correspondence:**
Ying Zhou
drzhouyingzr@163.com
Yong Wang
wangyong1239117@sina.com

Specialty section:
This article was submitted to Vascular Physiology, a section of the journal Frontiers in Physiology

Received: *30 June 2017*
Accepted: *29 November 2017*
Published: *12 December 2017*

Citation:
Zhou Y, Wang Y, Qiao S and Yin L (2017) Effects of Apelin on Cardiovascular Aging. Front. Physiol. 8:1035. doi: 10.3389/fphys.2017.01035

INTRODUCTION

Apelin was discovered in 1998 as the endogenous ligand of APJ, the orphan G protein-coupled receptor (O'Dowd et al., 1993; Tatemoto et al., 1998). The gene for the APJ receptor has high sequence homology to the angiotensin receptor ATR (O'Dowd et al., 1993; Tatemoto et al., 1998). The preprotein of apelin is a 77-amino acid that is sequentially decomposed by an angiotensin-converting enzyme into four active peptides, i.e., apelin-13, apelin-12, apelin-17, and apelin-36 (Tatemoto et al., 1998; Habata et al., 1999; Hosoya et al., 2000; Lee et al., 2000); among these, the most potent peptide that has the primary active biological function is apelin-13 (Kawamata et al., 2001; Tatemoto et al., 2001).

Aging is one of the primary risk factors in cardiovascular diseases (CVDs) (Dai et al., 2012). Studies have found that the renin-angiotensin system was related to cardiovascular aging. The renin-angiotensin system is one of the major signaling pathways related to the progress of the chronic proinflammatory profile within aged arteries (Wang et al., 2014). Ang II increased markedly in the thickened intima of rats, nonhuman and human primates (Wang et al., 2003, 2005, 2007; Fu et al., 2009). The Ang II receptor, AT1, is upregulated in aged arterial walls (Wang et al., 2005, 2007, 2010).

Ang II was also found to be related to structural, functional, and molecular changes that were found in the hearts of aged animals (Groban et al., 2006; Dai et al., 2009). Ang II levels increased significantly with age in myocardial tissue. Inhibition of Ang II signaling by either angiotensin-converting enzyme inhibitor or angiotensin receptor type II inhibitor was found to slow the progress of age-related cardiovascular changes, providing evidence for the role of Ang II and the effect of RAAS inhibitor in cardiovascular disease in aged people (Basso et al., 2007). Angiotensin-converting enzyme inhibitor and angiotensin receptor type II inhibitor have been shown to inhibit myocardial fibrosis and fibrosis-related arrhythmias in aged mice (Stein et al., 2010).

Because the gene for the APJ receptor has high sequence homology to the angiotensin receptor ATR, many studies concerning apelin–APJ in age-related cardiovascular diseases have

been performed. Diseases that are prominent in the elderly, such as atherosclerosis, hypertension, coronary atherosclerotic heart diseases, heart failure, atrial fibrillation and calcific aortic valve disease (CAVD), have been associated with the apelin–APJ signaling system. This review will focus on the apelin–APJ signaling system related to age-associated cardiovascular diseases.

APELIN/APJ CELLULAR SIGNALING PATHWAYS IN THE CARDIOVASCULAR SYSTEM

A number of studies have indicated that the apelin–APJ system is a powerful factor in the cardiovascular system in addition to Angiotension II and ATR. In the cardiovascular system, apelin binds to the APJ receptor on endothelial cells, vascular smooth muscle cells, and cardiac myocytes. As a result, vasodilatation (Reaux et al., 2001) and cardiac inotropic effect are performed (Dai et al., 2006; Yu et al., 2014). Previous studies showed that apelin could inhibit cardiac fibrosis via the prevention of cardiac fibroblast activation and collagen production (Pchejetski et al., 2012).

ENDOTHELIAL CELLS

It was found that apelin could act as a vasodilator in the presence of NO and endothelium (Tatemoto et al., 2001). *In vitro* studies showed that apelin caused NO-dependent vasodilation in human mesenteric arteries (Jia et al., 2007). However, apelin-13 may conduct vasoconstriction and deteriorate hypertension in rats after harming the vascular endothelium (Han et al., 2013).

VASCULAR SMOOTH MUSCLE CELLS

Recent studies showed that apelin-13 could induce vascular smooth muscle cell (VSMC) proliferation by upregulating the expression of Cyclin D1 (Li et al., 2013a). Cui et al. found that apelin prominently reduces apoptosis of human VSMCs; apoptosis was induced by serum deprivation (Cui et al., 2010). Wang et al. determined that apelin promotes VSMC migration through a PI3K/Akt/FoxO3a/MMP-2 pathway (Wang et al., 2015).

CARDIOMYOCYTES

The cardiac inotropic effect of apelin has been found in recent studies. Apelin showed direct effects on the contractility of cardiomyocytes. Apelin significantly improved sarcomere shortening in normal and failing cardiomyocytes. One of the mechanisms may be an increased myofilament sensitivity to Ca(2+), because apelin enhanced the activity of the Na(+)/H(+) exchanger with consequent intracellular alkalinization (Farkasfalvi et al., 2007). Isolated left ventricular cardiomyocytes lacking either apelin or APJ show less sarcomeric shortening and a decreased velocity of contraction (Charo et al., 2009).

APELIN AND AGING-RELATED CARDIOVASCULAR DISEASES

Apelin and Atherosclerosis

The most important part in atherosclerotic progress is atherosclerotic plaque formation. Angiotensin had been proved to be an atherosclerosis inducer, so it is hypothesized that apelin is also a critical factor in the progress of atherosclerosis (Li et al., 2010). Pitkin SL et al. found that apelin was upregulated in human atherosclerotic coronary arteries and is also localized to the plaque, co-localizing with markers for macrophages and smooth muscle cells (Pitkin et al., 2010). Chun et al. (2008) found that apelin downregulated AS formation by inhibiting AngII actions in mice. However, Hashimoto et al. (2007) found that apelin can promote AS by mediating oxidative stress-related AS in vascular tissue. Although it is clear that apelin is an important factor for AS, it is still difficult to define whether apelin/APJ has a beneficial or harmful role in atherosclerosis. The contribution of apelin in the development of AS remains to be determined.

Apelin and Cardiac Atherosclerotic Diseases

Angiogenesis is one of the most important mechanisms of myocardial repair for cardiac atherosclerotic diseases, such as myocardial infarctions (MI) and ischemic heart diseases. The effect of apelin in angiogenesis in animal models of AMI and ischemic heart disease have been demonstrated with positive results (Li et al., 2007; Mao et al., 2011). It was reported that apelin decreased in patients with MI, and a lower apelin level was associated with downregulated myocardial angiogenesis (Li et al., 2010). Injection of apelin into the ischemic myocardium stimulated neovascularization in the peri-infarct area through paracrine activity (Tempel et al., 2012). Li et al. (2008) found that apelin-13 could promote myocardial angiogenesis, inhibit cardiac fibrosis, attenuate cardiac hypertrophy, and improve cardiac function at 14 days after myocardial infarction. Regarding the mechanism for apelin-13 promoting angiogenesis after myocardial infarction, studies explored that apelin could upregulate the expression of SDF-1a/CXCR-4 and the homing of vascular progenitor cells (Wang et al., 2013). To confirm the angiogenesis effect of apelin in the heart, a further study was performed in which murine bone marrow cells were pretreated by apelin and later delivered into myocardium. As a result, myocardial angiogenesis increased and cardiac fibrosis was attenuated (Kidoya et al., 2010).

Because myocardial angiogenesis plays an important role in cardiac function in cardiac atherosclerotic diseases, the positive effect of apelin indicates that it could be used as a myocardial protecting factor after myocardial infarction. Further clinical studies are needed to confirm this effect of apelin.

Apelin and Hypertension

Hypertension is highly related to endothelial dysfunction and arterial stiffness. In healthy individuals, age is an essential factor in arterial structure and function alteration (Azizi et al., 2013). Increases in arterial stiffness are mostly attributed to aging-induced endothelial dysfunction (Arnett et al., 1994; Blacher et al., 1999; Li et al., 2012, 2013b). NO plays an important role

in vasodilation (Laurent et al., 2001). Aging is associated with the impairment of arterial eNOS mRNA and protein expression, which contribute to increased arterial stiffness and elevated blood pressure (Csiszar et al., 2002; LeBlanc et al., 2008; Donato et al., 2009; Novella et al., 2013).

Apelin administration caused a powerful antihypertensive effect in normal and hypertensive animal models (Rowe, 1987; Katugampola et al., 2001; Napoli and Ignarro, 2001). Administration of apelin to patients causes NO-mediated arterial vasodilation with no significant effect on peripheral venous tonus (Japp et al., 2008; Quazi et al., 2009). This antihypertensive effect was blocked in the co-presence of NOS inhibitor, indicating that apelin leads to vasodilation through a mechanism associated with NO (Szokodi et al., 2002). The antihypertensive effect of apelin was inhibited, and at the same time, the eNOS phosphorylation in the endothelial cells was downregulated in APJ-deficient mice (Zhang et al., 2006). Therefore, reductions of NO expression may be associated with reduced plasma apelin levels in the elderly and may result in endothelial dysfunction and arterial stiffness. Moreover, the concentration-dependent vasodilatation effect of apelin was normal in endothelium-intact mammary arteries but disappeared after endothelial removal, indicating that the antihypertensive effect of apelin is endothelium-dependent (Charles et al., 2006; Maguire et al., 2009).

Future research about the effect of apelin in patients with hypertension should focus on the mechanism in addition to the NO pathway in order to find hidden side effects of apelin in patients with hypertension in further clinical studies.

Apelin and Heart Failure

Because it was demonstrated that apelin had a potent inotropic effect in myocardial cells, further *in vivo* studies were performed to find the effect of apelin in heart failure. Both myocardial and plasma apelin levels of heart failure patients decreased simultaneously, suggesting that the heart is a major source of circulating apelin; it plays an essential role in the maintenance of myocardial systolic function (Dalzell et al., 2015). Several studies focused on aged animals and humans with heart failure. Compared with control aged mice, apelin$^{-/-}$ mice have an increased risk of progressive left ventricular systolic dysfunction with age (Lee et al., 2005). Infusion of apelin-13 in aged apelin$^{-/-}$ mice could improve left ventricular systolic dysfunction (Ishida et al., 2004). In humans, plasma apelin levels decreased in advanced heart failure in most studies, but the studies that focused on the early stages of heart failure demonstrated that apelin levels remained normal or even increased in early stages (Chen et al., 2003; Kuba et al., 2007; Miettinen et al., 2007; Japp et al., 2010). A study from Pitkin SL et al. may explain this phenomenon. They found that apelin receptor APJ's density significantly decreased in the left ventricle of patients with dilated cardiomyopathy or ischemic heart disease compared with that in the left ventricle of control patients, but apelin peptide levels remained unchanged. The decrease in receptor density in heart failure may limit the positive inotropic actions of apelin, resulting in an initial compensatory mechanism by increasing apelin to improve myocardial contractility (Pitkin et al., 2010). Serum apelin levels were upregulated after cardiac resynchronization therapy together with an improvement in myocardial systolic function (Földes et al., 2003). The administration of apelin in patients with heart failure led to the improvement of cardiac output and vasodilatation (Chong et al., 2006).

Thus, apelin could be used as a factor that has both a cardiotonic and afterload lowering effect in heart failure patients. It seems that apelin has a similar effect to that of BNP in heart failure, so future clinical studies could be designed to compare these two factors, because Nesiritide's effect has been confirmed.

Apelin and Atrial Fibrillation (AF)

The expression of apelin in normal atrial myocardium of humans is extremely high (Miettinen et al., 2007). Compared with control subjects with sinus rhythm, patients with atrial fibrillation had significantly lower plasma apelin levels (Francia et al., 2007). Another study showed that if patients could remain in sinus rhythm, the circulating apelin level would rise subsequently as a result (Ellinor et al., 2006). Atrial fibrillation will lead to the loss of atrial systolic function and atrial tissue remodeling. It may be deduced that downregulation of atrial apelin synthesis is a result of increased atrial diastolic filling pressures in patients with atrial fibrillation. Moreover, it has been shown that apelin significantly changes atrial electrophysiology with a shortening of action potential duration that may be caused by its effects on multiple ionic currents (Cheng et al., 2013).

The morbidity of atrial fibrillation increases with age in humans. Based on the existing studies concerning apelin and atrial fibrillation, the level of apelin in patients with AF may reflect the systolic function of the atrium. Further studies could focus on the predictive effect of apelin in the morbidity of atrial fibrillation and the possibility of maintaining a sinus rhythm.

Apelin and Calcific Aortic Valve Disease

Aortic stenosis and calcific aortic valve disease (CAVD) are leading valvular heart diseases in the elderly (Kallergis et al., 2010). The prevalence of aortic stenosis is only approximately 0.2% in adults over 50 years of age but increases to 9.8% for adults over 80 years of age (Otto and Prendergast, 2014). In tissues of stenotic aortic valves, the expression levels of both mRNA and protein of apelin increased (Nishimura et al., 2014). The levels of apelin and its receptor APJ are upregulated in patients with calcified aortic valve stenosis (Peltonen et al., 2009). Apelin may be upregulated compensatorily in the development of aortic valve stenosis. APJ receptor antagonists might be beneficial in the treatment of aortic valve stenosis by suppressing angiogenesis, osteoblast activity and collagen synthesis (Peltonen et al., 2009).

CONCLUSION

CVDs are the most common causes of death in most countries of the world, and old age is a risk factor for CVDs. Studies have found that RAAS plays an important role in cardiac aging. As the newest member in the RAAS system, it has been shown that apelin can increase cardiac contractility, lower blood pressure, increase atherosclerotic plaque stability and ameliorate the harmful effects of AT1 receptor activation in the progression of aortic valve stenosis. Further clinical trials are necessary to study the application of apelin in the treatment of cardiac aging, hypertensive cardiomyopathy, and heart failure.

AUTHOR CONTRIBUTIONS

YZ is the main author of the article. YW, SQ, and LY revised the article.

FUNDING

This article was supported by the National Natural Science Foundation of China (Fund No. 81370327).

REFERENCES

Arnett, D. K., Evans, G. W., and Riley, W. A. (1994). Arterial stiffness: a new cardiovascular risk factor? *Am. J. Epidemiol.* 140, 669–682. doi: 10.1093/oxfordjournals.aje.a117315

Azizi, Y., Faghihi, M., Imani, A., Roghani, M., and Nazari, A. (2013). Post-infarct treatment with [Pyr1]-apelin-13 reduces myocardial damage through reduction of oxidative injury and nitric oxide enhancement in the rat model of myocardial infarction. *Peptides* 46, 76–82. doi: 10.1016/j.peptides.2013.05.006

Basso, N., Cini, R., Pietrelli, A., Ferder, L., Terragno, N. A., and Inserra, F. (2007). Protective effect of long-term angiotensin II inhibition. *Am. J. Physiol. Heart Circ. Physiol.* 293, H1351–H1358. doi: 10.1152/ajpheart.00393.2007

Blacher, J., Asmar, R., Djane, S., London, G. M., and Safar, M. E. (1999). Aortic pulse wave velocity as a marker of cardiovascular risk in hypertensive patients. *Hypertension* 33, 1111–1117. doi: 10.1161/01.HYP.33.5.1111

Charles, C. J., Rademaker, M. T., and Richards, A. M. (2006). Apelin-13 induces a biphasic haemodynamic response and hormonal activation in normal conscious sheep. *J. Endocrinol.* 189, 701–710. doi: 10.1677/joe.1.06804

Charo, D. N., Ho, M., Fajardo, G., Kawana, M., Kundu, R. K., Sheikh, A. Y., et al. (2009). Endogenous regulation of cardiovascular function by apelin–APJ. *Am. J. Physiol. Heart Circ. Physiol.* 297, H1904–H1913. doi: 10.1152/ajpheart.00686.2009

Chen, M. M., Ashley, E. A., Deng, D. X., Tsalenko, A., Deng, A., Tabibiazar, R., et al. (2003). Novel role for the potent endogenous inotrope apelin in human cardiac dysfunction. *Circulation* 108, 1432–1439. doi: 10.1161/01.CIR.0000091235.94914.75

Cheng, C. C., Weerateerangkul, P., Lu, Y. Y., Chen, Y. C., Lin, Y. K., Chen, S. A., et al. (2013). Apelin regulates the electrophysiological characteristics of atrial myocytes. *Eur. J. Clin. Invest.* 43, 34–40. doi: 10.1111/eci.12012

Chong, K. S., Gardner, R. S., Morton, J. J., Ashley, E. A., and McDonagh, T. A. (2006). Plasma concentrations of the novel peptide apelin are decreased in patients with chronic heart failure. *Eur. J. Heart Fail.* 8, 355–360. doi: 10.1016/j.ejheart.2005.10.007

Chun, H. J., Ali, Z. A., Kojima, Y., Kundu, R. K., Sheikh, A. Y., Agrawal, R., et al. (2008). Apelin signaling antagonizes Ang II effects in mouse models of atherosclerosis. *J. Clin. Invest.* 118, 3343–3354. doi: 10.1172/JCI34871

Csiszar, A., Ungvari, Z., Edwards, J. G., Kaminski, P., Wolin, M. S., Koller, A., et al. (2002). Aging-induced phenotypic changes and oxidative stress impair coronary arteriolar function. *Circ. Res.* 90, 1159–1166. doi: 10.1161/01.RES.0000020401.61826.EA

Cui, R. R., Mao, D. A., Yi, L., Wang, C., Zhang, X. X., Xie, H., et al. (2010). Apelin suppresses apoptosis of human vascular smooth muscle cells via APJ/PI3-K/Akt signaling pathways. *Amino Acids* 39, 1193–1200. doi: 10.1007/s00726-010-0555-x

Dai, D. F., Chen, T., Johnson, S. C., Szeto, H., and Rabinovitch, P. S. (2012). Cardiac aging: from molecular mechanisms to significance in human health and disease. *Antioxid. Redox Signal.* 16, 1492–1526. doi: 10.1089/ars.2011.4179

Dai, D. F., Santana, L. F., Vermulst, M., Tomazela, D. M., Emond, M. J., MacCoss, M. J., et al. (2009). Overexpression of catalase targeted to mitochondria attenuates murine cardiac aging. *Circulation* 119, 2789–2797. doi: 10.1161/CIRCULATIONAHA.108.822403

Dai, T., Ramirez-Correa, G., and Gao, W. D. (2006). Apelin increases contractility in failing cardiac muscle. *Eur. J. Pharmacol.* 53, 222–228. doi: 10.1016/j.ejphar.2006.09.034

Dalzell, J. R., Rocchiccioli, J. P., Weir, R. A., Jackson, C. E., Padmanabhan, N., Gardner, R. S., et al. (2015). The emerging potential of the Apelin-APJ system in heart failure. *J. Card. Fail.* 21, 489–498. doi: 10.1016/j.cardfail.2015.03.007

Donato, A. J., Gano, L. B., Eskurza, I., Silver, A. E., Gates, P. E., Jablonski, K., et al. (2009). Vascular endothelial dysfunction with aging: endothelin-1 and endothelial nitric oxide synthase. *Am. J. Physiol. Heart Circ. Physiol.* 297, H425–H432. doi: 10.1152/ajpheart.00689.2008

Ellinor, P. T., Low, A. F., and Macrae, C. A. (2006). Reduced apelin levels in lone atrial fibrillation. *Eur. Heart J.* 27, 222e226. doi: 10.1093/eurheartj/ehi648

Farkasfalvi, K., Stagg, M. A., Coppen, S. R., Siedlecka, U., Lee, J., Soppa, G. K., et al. (2007). Direct effects of apelin on cardiomyocyte contractility and electrophysiology. *Biochem. Biophys. Res. Commun.* 357, 889–895. doi: 10.1016/j.bbrc.2007.04.017

Földes, G., Horkay, F., Szokodi, I., Vuolteenaho, O., Ilves, M., Lindstedt, K. A., et al. (2003). Circulating and cardiac levels of apelin, the novel ligand of the orphan receptor APJ, in patients with heart failure. *Biochem. Biophys. Res. Commun.* 308, 480–485. doi: 10.1016/S0006-291X(03)01424-4

Francia, P., Salvati, A., Balla, C., De Paolis, P., Pagannone, E., Borro, M., et al. (2007). Cardiac resynchronization therapy increases plasma levels of the endogenous inotrope apelin. *Eur. J. Heart Fail.* 9, 306–309. doi: 10.1016/j.ejheart.2006.06.005

Fu, Z., Wang, M., Gucek, M., Wu, J., Jiang, L., Monticone, R. E., et al. (2009). Milk fat globule protein epidermal growth factor-8: a pivotal relay element within the angiotensin II and monocyte chemoattractant protein-1 signaling cascade mediating vascular smooth muscle cells invasion. *Circ. Res.* 104, 1337–1346. doi: 10.1161/CIRCRESAHA.108.187088

Groban, L., Pailes, N. A., Bennett, C. D., Carter, C. S., Chappell, M. C., Kitzman, D. W., et al. (2006). Growth hormone replacement attenuates diastolic dysfunction and cardiac angiotensin II expression in senescent rats. *J. Gerontol. A Biol. Sci. Med. Sci.* 61, 28–35. doi: 10.1093/gerona/61.1.28

Habata, Y., Fujii, R., Hosoya, M., Fukusumi, S., Kawamata, Y., Hinuma, S., et al. (1999). Apelin, the natural ligand of the orphan receptor APJ, is abundantly secreted in the colostrum. *Biochim. Biophys. Acta* 1452, 25–35. doi: 10.1016/S0167-4889(99)00114-7

Han, X., Zhang, D. L., Yin, D. X., Zhang, Q. D., and Liu, W. H. (2013). Apelin-13 deteriorates hypertension in rats after damage of the vascular endothelium by ADMA. *Can. J. Physiol Pharmacol.* 91, 708–714. doi: 10.1139/cjpp-2013-0046

Hashimoto, T., Kihara, M., Imai, N., Yoshida, S., Shimoyamada, H., Yasuzaki, H., et al. (2007). Requirement of apelin-apelin receptor system for oxidative stress-linked atherosclerosis. *Am. J. Pathol.* 171, 1705–1712. doi: 10.2353/ajpath.2007.070471

Hosoya, M., Kawamata, Y., Fukusumi, S., Fujii, R., Habata, Y., Hinuma, S., et al. (2000). Molecular and functional characteristics of APJ. Tissue distribution of mRNA and interaction with the endogenous ligand apelin. *J. Biol. Chem.* 275, 21061–21067. doi: 10.1074/jbc.M908417199

Ishida, J., Hashimoto, T., Hashimoto, Y., Nishiwaki, S., Iguchi, T., Harada, S., et al. (2004). Regulatory roles for APJ, a seven-transmembrane receptor related to angiotensin-type 1 receptor in blood pressure *in vivo*. *J. Biol. Chem.* 279, 26274–26279. doi: 10.1074/jbc.M.404149200

Japp, A. G., Cruden, N. L., Amer, D. A., Li, V. K., Goudie, E. B., Johnston, N. R., et al. (2008). Vascular effects of apelin *in vivo* in man. *J. Am. Coll. Cardiol.* 52, 908–913. doi: 10.1016/j.jacc.2008.06.013

Japp, A. G., Cruden, N. L., Barnes, G., van Gemeren, N., Mathews, J., Adamson, J., et al. (2010). Acute cardiovascular effects of apelin in humans: potential role in patients with chronic heart failure. *Circulation* 121, 1818–1827. doi: 10.1161/CIRCULATIONAHA.109.911339

Jia, Y. X., Lu, Z. F., Zhang, J., Pan, C. S., Yang, J. H., Zhao, J., et al. (2007). Apelin activates L-arginine/nitric oxide synthase/nitric oxide pathway in rat aortas. *Peptides* 28, 2023–2029. doi: 10.1016/j.peptides.2007.07.016

Kallergis, E. M., Manios, E. G., Kanoupakis, E. M., Mavrakis, H. E., Goudis, C. A., Maliaraki, N. E., et al. (2010). Effect of sinus rhythm restoration after electrical cardioversion on apelin and brain natriuretic peptide prohormone levels in patients with persistent atrial fibrillation. *Am. J. Cardiol.* 105, 90–94. doi: 10.1016/j.amjcard.2009.08.656

Katugampola, S. D., MacGuire, J. J., Mathewson, S. R., and Davenport, A. P. (2001). [125I]-(Pyr1)Apelin-13 is a novel radioligand for localizing the APJ orphan receptor in human and rat tissues with evidence for a vasoconstrictor role in man. *Br. J. Pharmacol.* 132, 1255–1260. doi: 10.1038/sj.bjp.0703939

Kawamata, Y., Habata, Y., Fukusumi, S., Hosoya, M., Fujii, R., Hinuma, S., et al. (2001). Molecular properties of apelin, tissue distribution and receptor binding. *Biochim. Biophys. Acta* 1538, 162–171. doi: 10.1016/S0167-4889(00)00143-9

Kidoya, H., Naito, H., and Takakura, N. (2010). Apelin induces enlarged and nonleaky blood vessels for functional recovery from ischemia. *Blood* 115, 3166–3174. doi: 10.1182/blood-2009-07-232306

Kuba, K., Zhang, L., Imai, Y., Arab, S., Chen, M., Maekawa, Y., et al. (2007). Impaired heart contractility in apelin gene-deficient mice associated with aging and pressure overload. *Circ. Res.* 101, e32–e42. doi: 10.1161/CIRCRESAHA.107.158659

Laurent, S., Boutouyrie, P., Asmar, R., Gautier, I., Laloux, B., Guize, L., et al. (2001). Aortic stiffness is an independent predictor of all-cause and cardiovascular mortality in hypertensive patients. *Hypertension* 37, 1236–1241. doi: 10.1161/01.HYP.37.5.1236

LeBlanc, A. J., Shipley, R. D., Kang, L. S., and Muller-Delp, J. M. (2008). Age impairs Flk-1 signaling and NO-mediated vasodilation in coronary arterioles. *Am. J. Physiol. Heart Circ. Physiol.* 295, H2280–H2288. doi: 10.1152/ajpheart.00541.2008

Lee, D. K., Cheng, R., Nguyen, T., Fan, T., Kariyawasam, A. P., Liu, Y., et al. (2000). Characterization of apelin, the ligand for the APJ receptor. *J. Neurochem.* 74, 34–41. doi: 10.1046/j.1471-4159.2000.0740034.x

Lee, D. K., Saldivia, V. R., Nguyen, T., Cheng, R., George, S. R.,and O'Dowd, B. F. (2005). Modification of the terminal residue of apelin-13 antagonizes its hypotensive action. *Endocrinology* 146, 231–236. doi: 10.1210/en.2004-0359

Li, F., Li, L. F., Qin, X. P., Pan, W. N., Feng, F., Chen, F., et al. (2007). The study of vascular smooth muscle cells proliferation stimulated by apelin-13. *Chin. Pharmacol. Bull.* 23, 949–953.

Li, F., Li, L., Qin, X., Pan, W., Feng, F., Chen, F., et al. (2008). Apelin-induced vascular smooth muscle cell proliferation: the regulation of cyclin D1. *Front Biosci.* 13, 3786–3792. doi: 10.2741/2967

Li, L., Li, L., Xie, F., Zhang, Z., Guo, Y., Tang, G., et al. (2013a). Jagged-1/Notch3 signaling transduction pathway is involved in apelin-13-induced vascular smooth muscle cells proliferation. *Acta Biochim. Biophys. Sin.* 45, 875–881. doi: 10.1093/abbs/gmt085

Li, L., Zeng, H., and Chen, J. X. (2012). Apelin-13 increases myocardial progenitor cells and improves repair postmyocardial infarction. *Am. J. Physiol. Heart Circ. Physiol.* 303, H605–H618. doi: 10.1152/ajpheart.00366.2012

Li, L., Zeng, H., Hou, X., He, X., and Chen, J. X. (2013b). Myocardial injection of apelinoverexpressing bone marrow cells improves cardiac repair via upregulation of Sirt3 after myocardial infarction. *PLoS ONE* 8:e71041. doi: 10.1371/journal.pone.0071041

Li, X., Zhang, X., Li, F., Chen, L., Li, L., Qin, X., et al. (2010). 14-3-3 mediates apelin-13-induced enhancement of adhesion of monocytes to human umbilical vein endothelial cells. *Acta Biochim. Biophys. Sin.* 42, 403–409. doi: 10.1093/abbs/gmq036

Maguire, J. J., Kleinz, M. J., Pitkin, S. L., and Davenport, A. P. (2009). [Pyr1]apelin-13 identified as the predominant apelin isoform in the human heart: vasoactive mechanisms and inotropic action in disease. *Hypertension* 54, 598–604. doi: 10.1161/HYPERTENSIONAHA.109.134619

Mao, X., Su, T., Zhang, X., Li, F., Qin, X., Liao, D., et al. (2011). Apelin-13 promote monocytes adhesion to HUVECs via PI3K signaling. *Prog. Biochem. Biophys.* 38, 1162–1170. doi: 10.3724/SP.J.1206.2011.00335

Miettinen, K. H., Magga, J., Vuolteenaho, O., Vanninen, E. J., Punnonen, K. R., Ylitalo, K., et al. (2007). Utility of plasma apelin and other indices of cardiac dysfunction in the clinical assessment of patients with dilated cardiomyopathy. *Regul. Pept.* 140, 178–184. doi: 10.1016/j.regpep.2006.12.004

Napoli, C., and Ignarro, L. J. (2001). Nitric oxide and atherosclerosis. *Nitric Oxide* 5, 88–97. doi: 10.1006/niox.2001.0337

Nishimura, R. A., Otto, C. M., Bonow, R. O., Carabello, B. A., Erwin, J. P. III., Guyton, R. A., et al. (2014). 2014 AHA/ACC guideline for the management of patients with valvular heart disease: executive summary: a report of the American College of Cardiology/American Heart Association Task Force on Practice Guidelines. *J. Am. Coll. Cardiol.* 6322, 2438–2488. doi: 10.1016/j.jacc.2014.02.537

Novella, S., Dantas, A. P., Segarra, G., Vidal-Gómez, X., Mompeón, A., Garabito, M., et al. (2013). Aging-related endothelial dysfunction in the aorta from female senescence-accelerated mice is associated with decreased nitric oxide synthase expression. *Exp. Gerontol.* 48, 1329–1337. doi: 10.1016/j.exger.2013.08.003

O'Dowd, B. F., Heiber, M., Chan, A., Heng, H. H., Tsui, L. C., Kennedy, J. L., et al. (1993). A human gene that shows identity with the gene encoding the angiotensin receptor is located on chromosome 11. *Gene* 136, 355–360. doi: 10.1016/0378-1119(93)90495-O

Otto, C. M., and Prendergast, B. (2014). Aortic-valve stenosis–from patients at risk to severe valve obstruction. *N. Engl. J. Med.* 371, 744–756. doi: 10.1056/NEJMra1313875

Pchejetski, D., Foussal, C., Alfarano, C., Lairez, O., Calise, D., Guilbeau-Frugier, C. et al. (2012). Apelin prevents cardiac fibroblast activation and collagen production through inhibition of sphingosine kinase 1. *Eur. Heart J.* 33, 2360–2369. doi: 10.1093/eurheartj/ehr389

Peltonen, T., Napankangas, J., Vuolteenaho, O., Ohtonen, P., Soini, Y., Juvonen, T., et al. (2009). Apelin and its receptor APJ in human aortic valve stenosis. *J. Heart Valve Dis.* 18, 644–652.

Pitkin, S. L., Maguire, J. J., Kuc, R. E., and Davenport, A. P. (2010). Modulation of the apelin/APJ system in heart failure and atherosclerosis in man. *Br. J. Pharmacol.* 160, 1785–1795. doi: 10.1111/j.1476-5381.2010.00821.x

Quazi, R., Palaniswamy, C., and Frishman, W. H. (2009). The emerging role of apelin in cardiovascular disease and health. *Cardiol. Rev.* 17, 283–286. doi: 10.1097/CRD.0b013e3181b3fe0d

Reaux, A., De Mota, N., Skultetyova, I., Lenkei, Z., El Messari, S., Gallatz, K., et al. (2001). Physiological role of a novel neuropeptide, apelin, and its receptor in the rat brain. *J. Neurochem.* 77, 1085–1096. doi: 10.1046/j.1471-4159.2001.00320.x

Rowe, J. W. (1987). Clinical consequences of age-related impairments in vascular compliance. *Am. J. Cardiol.* 60, 68G−71G. doi: 10.1016/0002-9149(87)90594-7

Stein, M., Boulaksil, M., Jansen, J. A., Herold, E., Noorman, M., Joles, J. A., et al. (2010). Reduction of fibrosisrelated arrhythmias by chronic renin-angiotensin-aldosterone system inhibitors in an aged mouse model. *Am. J. Physiol. Heart Circ. Physiol.* 299, H310–H321. doi: 10.1152/ajpheart.01137.2009

Szokodi, I., Tavi, P., Foldes, G., Voutilainen-Myllylä, S., Ilves, M., Tokola, H., et al. (2002). Apelin, the novel endogenous ligand of the orphan receptor APJ, regulates cardiac contractility. *Circ. Res.* 91, 434–440. doi: 10.1161/01.RES.0000033522.37861.69

Tatemoto, K., Hosoya, M., Habata, Y., Fujii, R., Kakegawa, T., Zou, M. X., et al. (1998). Isolation and characterization of a novel endogenous peptide ligand for the human APJ receptor. *Biochem. Biophys. Res. Commun.* 251, 471–476. doi: 10.1006/bbrc.1998.9489

Tatemoto, K., Takayama, K., Zou, M. X., Kumaki, I., Zhang, W., Kumano, K., et al. (2001). The novel peptide apelin lowers blood pressure via a nitric oxide-dependent mechanism. *Regul. Pept.* 99, 87–92. doi: 10.1016/S0167-0115(01)00236-1

Tempel, D., de Boer, M., van Deel, E. D., Haasdijk, R. A., Duncker, D. J., Cheng, C., et al. (2012). Apelin enhances cardiac neovascularization after myocardial infarction by recruiting aplnr+ circulating cells. *Circ. Res.* 111, 585–598. doi: 10.1161/CIRCRESAHA.111.262097

Wang, C., Wen, J., Zhou, Y., Li, L., Cui, X., Wang, J., et al. (2015). Apelin induces vascular smooth muscle cells migration via a PI3K/Akt/FoxO3a/MMP-2 pathway. *Int. J. Biochem. Cell Biol.* 69, 173–182. doi: 10.1016/j.biocel.2015.10.015

Wang, M., Jiang, L., Monticone, R. E., and Lakatta, E. G. (2014). Proinflammation: the key to arterial aging. *Trends Endocrinol. Metab.* 25, 72–79. doi: 10.1016/j.tem.2013.10.002

Wang, M., Takagi, G., Asai, K., Resuello, R. G., Natividad, F. F., Vatner, D. E., et al. (2003). Aging increases aortic MMP-2 activity and angiotensin II in nonhuman primates. *Hypertension* 41, 1308–1316. doi: 10.1161/01.HYP.0000073843.56046.45

Wang, M., Zhang, J., Jiang, L. Q., Spinetti, G., Pintus, G., Monticone, R., et al. (2007). Proinflammatory profile within the grossly normal aged human aortic wall. *Hypertension* 50, 219–227. doi: 10.1161/HYPERTENSIONAHA.107.089409

Wang, M., Zhang, J., Spinetti, G., Jiang, L. Q., Monticone, R., Zhao, D., et al. (2005). Angiotensin II activates matrix metalloproteinase type II and mimics age-associated carotid arterial remodeling in young rats. *Am. J. Pathol.* 167, 1429–1442. doi: 10.1016/S0002-9440(10)61229-1

Wang, M., Zhang, J., Walker, S. J., Dworakowski, R., Lakatta, E. G., and Shah, A. M. (2010). Involvement of NADPH oxidase in ageassociated cardiac remodeling. *J. Mol. Cell. Cardiol.* 48, 765–772. doi: 10.1016/j.yjmcc.2010.01.006

Wang, W., McKinnie, S. M., Patel, V. B., Haddad, G., Wang, Z., Zhabyeyev, P., et al. (2013). Loss of Apelin exacerbates myocardial infarction adverse remodeling and ischemia–reperfusion injury: therapeutic potential of synthetic Apelin analogues. *J. Am. Heart Assoc.* 2:e000249. doi: 10.1161/JAHA.113.000249

Yu, X. H., Tang, Z. B., Liu, L. J., Qian, H., Tang, S. L., Zhang, D. W., et al. (2014). Apelin and its receptor APJ in cardiovascular diseases. *Clin. Chim. Acta* 428, 1–8. doi: 10.1016/j.cca.2013.09.001

Zhang, J., Ren, C. X., Qi, Y. F., Lou, L. X., Chen, L., Zhang, L. K., et al. (2006). Exercise training promotes expression of apelin and APJ of cardiovascular tissues in spontaneously hypertensive rats. *Life Sci.* 79, 1153–1159. doi: 10.1016/j.lfs.2006.03.040

Conflict of Interest Statement: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.