Original Article

Potential link between microRNA-208 and cardiovascular diseases

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Background: The application of microRNAs (miRNAs) in disease diagnosis and therapy has been the center of tremendous attention over the past few years. Regarding the molecular markers involved in the pathogenesis of cardiovascular diseases, *miR-208* is closely related to myocardial hypertrophy, arrythmias, myocardial infarction, myocardial fibrosis, coronary atherosclerosis and heart failure. In this review, we focus on the regulatory mechanisms of *miR-208* and its correlation with cardiovascular diseases based on available literature.

Methods: We performed online search of medical literature published in an English Language Format through PubMed database over the past two decades, using a combination of text words and MeSH terms: "*microRNA-208*", "*miR-208*", "microRNA", "mechanism", "function" and "cardiovascular diseases". The most relevant articles were selected based on a subjective appraisal of their quality and mechanistic insight that could be relevant to the regulatory molecular pathway of *miR-208*. Full texts of the retrieved articles were accessed.

Results: In cardiac hypertrophy, overexpression of *miR-208* inhibits THAP1 and myostatin. Overload exercise may decrease *miR-208* expression, resulting in increased expression of *SOX6*, *MED13*, *SP3*, *Purβ* and *HP1β*. The *miR-208-Mef2* axis induces decompensated right ventricle hypertrophy in the pulmonary hypertension rat model. Nevertheless, *miR-208* inhibits *p21* and *NLK* in myocardial infarction. Moreover, *miR-208* regulates Cx40 and Cx43 in arrhythmias. Additionally, abnormal levels of *miR-208* suppress L-type Ca2+ channel subunits and SERCA2, affecting the handling function of Ca2+ during atrial remodeling. A strong correlation has been established between *miR-208* and cardiac fibrosis via endoglin regulation and collagen I expression. Overexpression of *miR-208b* inhibits the expression of *COL1* and *ACTA2* by inhibiting *GATA4*, hence hindering the progression of post-infarction myocardial fibrosis. With respect to vascular diseases, *miR-208* exerts its various functions by acting on proteins associated with inflammation, endothelial apoptosis, VSMC proliferation and migration, including the *PPAR*, *ACTA2*, *ROR2* and *PI3K/AKT* pathways.

Conclusions: *MiR-208* plays a pivotal role in the pathogenesis of cardiovascular diseases such as myocardial hypertrophy, myocardial infarction, arrhythmias, myocardial fibrosis and dysfunction of blood vessels. Further exploration of the potential link between *miR-208* and cardiovascular diseases would provide more valuable diagnostic and therapeutic insights.

Keywords: Cardiovascular diseases; functions; mechanisms; *miR-208*; microRNAs (miRNAs)

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Introduction

MicroRNAs (miRNAs) are endogenous small non-coding RNAs composed of approximately 22 nucleotides that directly regulate over 30% of genes. They inhibit the translation of messenger RNA (mRNA) or promote mRNA degradation either by annealing complementary sequences in the 3' untranslated regions or interacting with the coding sequence (CDS) (1,2). Moreover, miRNAs are drastically upregulated during pathological stress caused by several diseases, thus they have been investigated as potential biomarkers in cancer, autoimmune diseases, cardiovascular diseases and other diseases. For this purpose, these biomarkers have been identified in peripheral blood, saliva and urine. It is worth noting that research has detected the selective release of some premature and mature miRNAs from cells to the bloodstream via exosomes, microvesicles or protein complexes (3,4).

Among the miRNAs involved in the pathogenesis of cardiovascular diseases, *miRNA-208* (*miR-208*) is one of the significant genes linked with the pathogenesis of myocardial hypertrophy, arrythmias, myocardial infarction, myocardial fibrosis, coronary atherosclerosis and heart failure (5,6). Additionally, *miR-208* consists of two subtypes, *miR-208a* and *miR-208b*, both genes of which are located within the human chromosome 14 (7).

As cardiac-enriched miRNAs, *miR-208a* and *miR-208b* are highly chamber-specific with their host genes. To be more specific, *miR-208a* is encoded within an intron of the alpha-cardiac muscle myosin heavy chain gene (*Myh6*) and both of them are atria-specific. Meanwhile, *miR-208b* and its host gene the beta-cardiac muscle myosin heavy chain gene (*Myh7*) are ventricle-specific (7). Under physiological conditions, the expression levels of *miR-208a* and *miR-208b* are paralleled with their host gene transcription patterns. However, under pathological stress, dissociations may occur. Previous research projects have explored the molecular mechanisms regulating the expression of *miR-208* in cardiovascular diseases. In the present review, we determine the regulation mechanism of *miR-208* and the role it plays in the pathogenesis of cardiovascular diseases.

Methods

Searching methodology

We performed online search of medical literature published in an English Language Format through PubMed database over the past two decades, using a combination of text words and MeSH (Medical Subject Headings) terms: "*microRNA-208*", "*miR-208*", "microRNA", "mechanism", "function" and "cardiovascular diseases". We selected articles that featured *miR-208* in cardiovascular diseases models and patients. We then selected the most relevant articles based on a subjective appraisal of their quality and mechanistic insight that could be relevant to the regulatory molecular pathway of *miR-208*. Full texts of the retrieved articles were accessed.

Statistical analysis

No statistical analysis or comparisons, no grouping and no power calculations were conducted due to the heterogeneity of the studies. This study comprises a narrative synthesis of the available primary research studies, systematic reviews and meta-analyses in this area.

Ethical aspects

The study was conducted in accordance with the Declaration of Helsinki (as revised in 2013). The study was approved by the Ethics Committee of Shenzhen Children's Hospital (NO.: 202003802) and informed consent was waived as there were no patients involved.

Results

Molecular mechanisms involved in miR-208 regulation

THAP1 and myostatin

Thyroid hormone-associated protein 1 (THAP1) and myostatin are negative regulators of muscle growth and hypertrophy, which are both targeted by *miR-208*, expressed in the myocardial tissue, controlling heart remodeling and myocardial fibrosis by regulating the expression of β-MHC in response to various stimuli (8,9).

Thrap1 as a part of the thyroid hormone nuclear receptor complex influences gene transcription either positively or negatively (10). Data have proven that *miR-208* participates in the pre-transcription level of thyroid hormones, which modulates the expression of proteins related to cardiac hypertrophy, prevents collagen deposit and increases collagen removal (11). Furthermore, postpartum thyroid hormone (T3) signaling stimulates α-MHC while inhibiting β-MHC. Therefore, in the case of hypothyroidism or blockade of T3 biosynthesis with propylthiouracil (PTU), a decrease in the level of thyroid hormone induces

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upregulation of β-MHC, leading to a decline in cardiac performance.

Myostatin, also known as growth differentiation factor 8, is expressed in the cardiomyocytes and Purkinje fibers of the heart as an inhibitor of skeletal muscle growth (12,13). Besides, activation of the myostatin receptor inhibits the activity of AKT, a protein kinase B known as a major determinant of muscle protein synthesis and cell proliferation (12). Myostatin also inhibits myocardial proliferation and contractility, and induces fibrosis by stimulating fibroblast proliferation and survival (14). Also, it helps to maintain cardiac energy homeostasis mainly by regulating the myocardium's oxidative metabolism, and prevent pathological cardiac hypertrophy (15).

Overexpression of *miR-208a* represses *THAP1* and myostatin expression, hence contributing to pathological hypertrophy of cardiomyocytes and fibrosis (16,17).

Cyclin-dependent kinase inhibitor (P21)

Cyclin-dependent kinase inhibitor 1A (p21), a regulator of cell cycle progression at G1, is one of the targets of *miR-208*, and is upregulated by myostatin (18,19). Upregulation of the *p53-p21* pathway controls cardiomyocyte hypertrophy and apoptosis in diabetic cardiomyopathy, and is imperative in the endothelial cell senescence induced by disturbed flow (20,21). As a result, *miR-208* probably plays a major role in the development of diabetic cardiomyopathy, reduces migration and interrupts arterial repair by regulating the *p21* gene. Reduction of *miR-208* expression promotes the production of ROS (Reactive oxygen species) essentially by targeting *p21*, which then leads to cardiomyocyte apoptosis. In spite of its effects on cardiomyocytes, *miR-208* also participates in insulin-induced vascular smooth muscle cell (VSMC) proliferation via downregulation of *p21* (22).

Med13 and Mef2 axis

The complex mediator's subunit MED13 (Thrap1, thyroid hormone receptor associated protein-1) of transcription is directly targeted and negatively regulated by *miR-208a* in the heart. MED13 downregulates a series of genes involved in cellular metabolism including the *SREBP*, *RXR* and *PPARγ*, which controls gene transcription related to thyroid hormone as a negative regulator of metabolism (23-25). Data have demonstrated that overexpression of MED13 in the heart or pharmacologic inhibition of *miR-208a* increases the metabolic rate, confers resistance to obesity, enhances insulin sensitivity and decreases plasma lipid levels (26).

Another regulator of metabolism, myocyte enhancer factor 2 (*Mef2*), regulates contractile and angiogenic genes, as well as the development of the right ventricle. The *miR-208-Mef2* axis induces decompensation of the right ventricular function in a pulmonary hypertension rat model. There is a negative feedback loop through which *Mef2* inhibits *miR-208* expression by regulating its host *α-MHC* gene transcription. Consequently, the expression and activity of *Mef2* is inhibited via the *MED13/NCoR1* complex. This drives the transition from the compensating phase of right ventricular hypertrophy towards the decompensation phase as the self-limiting feedback loop is terminated (27).

SOX6, SP3, Purβ, HP1β

The sex determining region y-related transcription factor 6 (SOX6) is a member of the D sub family of the sex determining region Y (*SPY*)-box-related transcription factors. It is a *Myh*7 transcriptional repressor, which is preferentially inhibited by *miR-208b*. *SOX5* and *SOX6* both belong to the SoxD group and have a long N-terminal sequence containing a coiled-coil domain that enables them to heterodimerize with each other (28). Compared with *SOX6*, *SOX5* is preferentially repressed by *miR-208a*. Moreover, *SP3* is another *Myh*7 repressor found in skeletal muscles which can only be reduced considerably by *miR-208a* (29).

In HL-1 cells, *miR-208b* overexpression enhances fetal *Myh*7 expression by inhibiting the transcriptional repressors *SOX6* and *SP3*. Based on this fact, it can be concluded that *miR-208* overexpression supports the adaptive transcriptional switch from fast to slow contractile myosin genes in human myofibrils (30). A positive feedback loop may be present between *miR-208b* and upregulated *Myh*7 in injured cardiac tissue since fetal *Myh*7 is co-expressed with its intronic *miR-208b*.

SOX6 contains a conserved high-mobility-group DNA-binding domain, and acts as a crucial tumor-suppressor gene in the proliferation of esophageal cell carcinoma (ESCC) (31,32). Considering that *SOX6* mRNA is a direct and functional target of *miR-208*, experimental results on ESCC suggested that *miR-208* may influence the *SOX6*-mediated signaling pathway and thus play a role in promoting cell proliferation, tumorigenicity and cell cycle progression in ESCC (33). In this pathway, *miR-208* negatively regulates

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SOX6, eventually leading to downregulation of *p21*, upregulation of *cyclin D1* and phosphorylation of the *Rb* gene. However, ascertaining whether the same pathway exists in cardiomyocytes and endothelial cells requires further exploration.

High-volume swim training has been associated with decreased *miR-208* expression and increased *SOX6*, *MED13*, *SP3*, *Purβ* and *HP1β* expression levels (17). These increased molecules are all repressors of β-MHC and targets of both *miR-208a* and *miR-208b*, responsible for the regulation of cardiac metabolic and contractile adaptation via exercise. Specifically, *Purβ* (purine-rich element binding protein B) represses the expression of *ACTA2*, which stimulates myofibroblast proliferation and differentiation in fibrotic diseases (34). Likewise, *SP3* and *HP1β* both modulate histone deacetylase (HDAC) expression, which has a regulatory effect on cardiac remodeling. Class II HDACs are calcium-sensitive repressors of the *MEF2* transcription factor (35), which boosts the expression of slow myofiber genes. Since *miR-208* exerts repressive effects on *HP1β* and the latter is the corepressor of class II HDACs, the repressive influence of class II HDACs on *MEF2* could be possibly diminished under the impact of *miR-208*, hence the expression of slow muscle genes is promoted (36).

Nuclear receptors PPARβ and PPARα

The principal function of peroxisome proliferative-activated receptors (PPARs) is to regulate the transcription of several genes related to cellular metabolism. Given that both *PPARα* and *PPARβ* are modulated by *miR-208b* in skeletal muscles, *miR-208b* is probably involved in modification of the metabolic state as well (26).

PPARα is expressed primarily in the liver, intestines, brown adipose tissues, and heart, which are all organs carrying out elevated levels of fatty acid catabolism (37). PPARα is considered as a short-lived protein as it gets ubiquitinated and degraded proteasomes. Ligand binding confers protective effects on PPARα, which only lasts for a few hours before PPARα is rapidly degraded (38). Additionally, PPARα overexpression in the heart leads to metabolic cardiomyopathy as well as hepatic insulin resistance (5). PPARβ is also ubiquitinated without ligand, but the protective effect produced by ligand binding persists as long as the latter is present (39).

PPARβ increases the level of *miR-208b* by activating transcription of the *Myh7* and *Myh7b* genes, thereby leading to series of slow-twitch contractile muscle protein gene expression (40). In contrast, PPARα inhibits *Myh7*/*miR-208b* expression in muscle tissues.

It has been discovered that activation of either *PPARα* or *PPARγ* prevents proliferation and angiogenic activity in endothelial cells, while *PPARβ* exerts opposite effects under physiological conditions (41,42). Activation of the *PPARβ* signaling pathway enhances VEGF-A expression and angiogenesis in skeletal muscles and the heart via the activation of direct transcription or interaction with the *PI3K/AKT* pathway through which the capillary density increases (43,44). Its activation also prevents apoptosis of endothelial cells (45). Besides, *PPARβ* activation or overexpression inhibits inflammation through the transrepression of proinflammatory genes, whereas deletion of *PPARβ* aggravates the inflammatory state (46). Thence, this evidence points out that there may be a potential relationship between *miR-208b* and endothelial function through the regulation of *PPARs*. Withal, more studies are needed to verify this connection.

TGF-β1, Endoglin and Collagen I

It is well-known that *miR-208* is positively correlated with cardiac fibrosis, which is characterized by the myocardial collagen volume fraction in human dilated cardiomyopathy (47). Experiments carried out on rat heart cells demonstrate that *TGF-β* induces *miR-208a* expression under mechanical stretch conditions, and elevated *miR-208a* expression levels consequently encourage the expression of endoglin and stimulate collagen I formation, during which the cardiomyocytes differentiate into myofibroblasts, resulting in cardiac fibrosis (48). The discovery of specific sites complementary to *miR-208a* on the endoglin promoter sequences indicates that *miR-208a* may directly target endoglin throughout the pathogenesis of cardiac fibrosis. Fetal cardiomyocytes contain endoglin but absent adult ones do not, and it has been linked to the development vascular and other diseases (49). It is an integral membrane glycoprotein and a co-receptor of TGF-β1 and TGF-β3, and it is highly expressed on proliferating vascular endothelial cells (50). As a TGFβ coreceptor, endoglin enhances *TGF-β1* signaling by phosphorylating Smad3 (small mother against decapentaplegic protein) and binding BMP9 (bone morphogenetic protein 9) with high affinity. In this manner, endoglin stimulates the synthesis of collagen by cardiac fibroblasts, thus promotes cardiac fibrosis in pressure overload models (51).

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Connexin and GATA4

GJA5 encoding Connexin40 (Cx40), associated with chronic atrial fibrillation in humans, is a potential target of *miR-208a-3p* (52). Connexin proteins are gap junction proteins indispensable for the orderly propagation of electrical impulses throughout the heart, and are closely related with various chronic human heart conditions leading to arrythmias and sudden death. Furthermore, Cx40 is expressed restrictedly on the atria and specialized cardiomyocytes consisting of the His bundle and Purkinje fibers, which is regulated by *miR-208a* (53). Data have confirmed that Cx43 is also regulated by *miR-208* (30).

Besides, *miR-208a* directly targets the cardiac transcription factor *GATA4* by specifically targeting its 3' UTR. *GATA4* is a transcriptional cofactor expressed within the cardiac conduction system of the adult heart. It participates in transactivating serum response factor-dependent promoters, which includes Cx40. Through this molecular mechanism, *miR-208a*'s gain- and loss-of-function are associated with arrhythmias (54). In addition, *GATA4* is involved in the pathogenesis of cardiac hypertrophy, serving as a marker of myocyte hypertrophy and fibrosis, which is consistent with the cardiac hypertrophic effect induced by *miR-208* (16,55).

COL1 and ACTA2

Elevated concentration of *miR-208* in the plasma is correlated with myocardial infarction, the most evident pathogenic feature of which is the accumulation of collagen synthesized by cardiac fibroblasts, leading to post-infarction myocardial fibrosis. Research has established that *miR-208b* is downregulated in myocardial infarction rat models and its overexpression inhibits the expression of both *COL1* and *ACTA2* (the gene encoding smooth muscle α-actin) by directly targeting and inhibiting *GATA4*, which ultimately leads to the suppression of post-infarction myocardial fibrosis (56).

COL1A1 and *ACTA2* are both pro-fibrotic genes related to fibrosis, a disease characterized by excessive synthesis and accumulation of extracellular matrix (ECM) proteins in organs such as the skin, liver, lung, kidney and heart (57,58). In this pathogenic process, the myofibroblasts are responsible for producing the majority of the ECM, characterized by *de novo* expression of smooth muscle α-actin (SM α-actin) and abundant ECM, especially type I collagen (COL1A1) (59). COL1 encoded by *COL1A1* and *COL1A2*, is found increased in myocardial fibrosis and is usually used as a marker of fibrosis. Overexpression of *miR-208b* represses *COL1A1* transcription and COL1 synthesis, which in turn suppresses the progression of fibrosis.

ACTA2 is highly correlated with fibrosis in different diseases. It has been found to be upregulated at both mRNA and protein levels after myocardial infarction and repressed by *miR-208b* overexpression (60). Since SM α-actin is typically expressed in the VSMC and conducive to vascular motility and contraction, mutations in its coding gene *ACTA2* could cause various vasculopathies such as thoracic aortic aneurysm and aortic dissection, premature coronary artery disease and ischemic stroke (61). Another study centered on thoracic aortic aneurysms and dissections revealed that *ACTA2* is related to the mediation of angiotensin II (AngII) induced apoptosis, possibly by affecting the expression of *Bax* and *Bcl-2*. Knocking out *ACTA2* enhances the phonotypic modulation and apoptosis regulated by AngII in vascular smooth muscle cells, which consequently leads to a more vulnerable vascular wall and even vascular rupture (62). Therefore, overexpression of *miR-208b* probably grants a protective effect on blood vessels via *ACTA2* inhibition.

Type 1 Angiotensin II Receptor (AT1R)

Hyperthyroidism induces cardiac hypertrophy by activating Type 1 angiotensin II receptors (AT1R), whereas pharmacologic blockade of AT1R markedly inhibits the cardiac hypertrophy induced by increased levels of thyroid hormone (TH) (63). *AT1R* has a positive effect on the expression of cardiac *miR-208a*/*α-MHC* in hyperthyroidism but is irrelevant to the level of *miR-208b*/*β-MHC*, which corresponds with rising *miR-208a* and falling *miR-208b* expression in TH-induced cardiac hypertrophy. Moreover, cardiac *miR-208a* enhances obesity, and its inhibition with subsequent increase in *MED13* has been proven to be associated with attenuated weight gain despite leptin resistance (64).

Bax and PI3K/AKT Pathway

The *PI3K/AKT* pathway is a widely reported pathway conferring protection against cardiac hypertrophy and apoptosis (65). Abnormal activation of the *PI3K/AKT* signaling pathway was reported to be a negative feedback in response to abnormal stimuli (66,67). In respect of

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PI3Ks, class Ia PI3Ks are imperative for physiological hypertrophy. They emphatically adjust the heart's size via the *Akt* pathway while class Ib PI3Ks negatively influence the heart's contractile function (68).

As a serine/threonine protein kinase, AKT1 contributes to the net protein accumulation needed for cardiac hypertrophy by inhibiting the vitality of the forkhead box protein O3 (FOXO3) (69). Similarly, *Akt* signaling is also crucial in normal vascular patterning and remodeling via fine-tune control in endothelial cells (70).

In an experiment on rats, specimens transfected with Ad-anti-*miR-208* exhibited a 2-fold decrease in vascular endothelial growth factor (VEGF) mRNA levels (27). Considering that overexpression of *PI3K* and *AKT* induces transcription of VEGF and promotes formation of new blood vessels (71), the results of this experiment probably indicate an inducive effect of *miR-208* on VEGF through activation of the *PI3K/AKT* pathway in vascular homeostasis.

Phosphorylation of PI3K and AKT was significantly decreased under hypoxic conditions, but increased when the cell was transinfected with *miR-208b* mimics. This can be explained by the fact that *miR-208b* activates the *PI3K/AKT* pathway by directly downregulating Bax and further represses Bax expression after activating the *PI3K/AKT* pathway, through which *miR-208b* exerts its myocardioprotective effects (72).

SERCA2 and L-type Ca^{2+} channel subunits

Increased local (Ca^{2+} sparks) and global spontaneous Ca^{2+} released from the sarcoplasmic reticulum (Ca^{2+} waves) are associated with either chronic or paroxysmal atrial fibrillation (73,74). Studies on HL-1 myocytes and myocytes isolated from chronic atrial fibrillation (CAF) patients report that abnormal *miR-208b* expression levels repress the expression and function of L-type Ca^{2+} channel subunits (α1c and β2 encoded by *CACNA1C* and *CACNB2* genes respectively) and the sarcoplasmic reticulum-ATPase Ca^{2+} pump (SERCA2), serving as an essential mediator in Ca^{2+} handling dysfunction during atrial remodeling (30). The CAF-associated increase of *miR-208b* promotes $I_{Ca,L}$ downregulation by silencing its channel expression. Thus, *miR-208a* significantly reduces the frequency of Ca^{2+} sparks while *miR-208b* completely abolishes it. Concerning the negative effects on SERCA2 levels, both *miR-208a* and *miR-208b* remarkably reduce Ca^{2+} transient decay.

Nemo-like kinase

Nemo-like kinase (NLK), as an evolutionary conserved serine/threonine protein kinase, and an inhibitor of the *Wnt/β-catenin* signaling pathway, which plays multifunctional roles in cellular regeneration, proliferation and differentiation of cardiomyocytes (75).

NLK is a direct target of *miR-208* and its expression is suppressed by the overexpression of *miR-208* (76). It acts a maladaptive signaling effector in the heart as its deletion was shown to protect the mouse's heart from remodeling induced by pressure overload and infarction. Furthermore, NLK enhances the stability and activity of *p53* in response to DNA damage by revoking *MDM2*-mediated *p53* ubiquitination and degradation (77). Since *p53* is fundamental for cell cycle arrest, DNA repair and apoptosis induced by genotoxic and cellular stress (78,79), the relationship between *miR-208* and these pathological molecular mechanisms is probably due to the downregulation of *NLK* and the corresponding alteration incurred by *p53*. This inference has been proven by the experiment carried out on rat heart cells, which brought to the conclusion that *miR-208a* promotes the apoptosis of AngII-induced cardiomyocytes by inhibiting *NLK* (80).

ROR2 (receptor tyrosine kinase-like orphan receptor 2 gene)

The receptor tyrosine kinase-like orphan receptor 2 (ROR2) is a Wnt5a receptor since it belongs to the receptor tyrosine kinase family. It has been reported as not only a cell surface marker expressed by human mesenchymal stem cells with the potential for cartilage formation, but also a marker of mesodermal progenitors of all major cardiovascular lineages (81-83). Nonetheless, *ROR2* expression is limited to human fetal hearts from the first trimester and completely absent in adult human myocardial tissues (84).

AngII enhances VSMC proliferation and migration by augmenting *ROR2* expression and stimulating the *RhoA* signaling pathway (85). Meanwhile, knockdown of the ROR2 receptor markedly suppresses cell migration in endothelial cells, and reduces cholesterol accumulation as well as inflammatory responses in VSMCs during the progression of atherosclerosis (86,87). Based on the fact that *miR-208b* has been identified to inhibit osteosarcoma progression by directly targeting and downregulating ROR2 receptors (88), it would be reasonable to assume that *miR-208* may also be involved in vascular homeostasis via *ROR2*

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regulation. Unfortunately, there is no literature illustrating the direct relationship between *miR-208* and *ROR2* in cardiovascular diseases so far, thus further investigation on this topic is warranted.

Conclusions

In a nutshell, *miR-208* is a miRNA mainly expressed in the heart, and closely related to cardiovascular diseases. Regarding the pathogenesis of cardiac hypertrophy, overexpression of *miR-208* inhibits THAP1 and myostatin, which are both negative regulators of muscle growth and hypertrophy. Furthermore, overload exercise may decrease *miR-208* expression, resulting in increased expression of *SOX6*, *MED13*, *SP3*, *Purβ* and *HP1β*, which eventually influences cardiac metabolism and contractile adaptation. The *miR-208-Mef2* axis was found to induce decompensated right ventricle hypertrophy in the pulmonary hypertension rat model. Nevertheless, *miR-208* exerts its effects predominantly through inhibition of *p21* and *NLK* in the pathogenesis of myocardial infarction. Moreover, *miR-208* is associated with arrhythmias, through the regulation of Cx40 and Cx43. Additionally, abnormal levels of *miR-208* lead to suppression of L-type Ca^{2+} channel subunits and SERCA2, affecting the handling function of Ca^{2+} during atrial remodeling. A strong correlation has been established between *miR-208* also and cardiac fibrosis via endoglin regulation and collagen I expression. Overexpression of *miR-208b* inhibits the expression of *COL1* and *ACTA2* by inhibiting *GATA4*, hence hindering the progression of post-infarction myocardial fibrosis. With respect to vascular diseases, *miR-208* exerts its various functions by acting on proteins associated with inflammation, endothelial apoptosis, VSMC proliferation and migration, including the *PPAR*, *ACTA2*, *ROR2* and *PI3K/AKT* pathways.

In conclusion, *miR-208* plays a pivotal role in the pathogenesis of myocardial hypertrophy, myocardial infarction, arrhythmias, myocardial fibrosis and dysfunction of blood vessels, implying that it could be implemented into routine clinical practice as a valuable biomarker and therapeutic target for cardiovascular diseases. It has recently been identified as a potential plasma biomarker for coronary artery disease (89). Further exploration of the potential link between *miR-208* and cardiovascular diseases would provide more valuable diagnostic and therapeutic insights.

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Footnote

Conflicts of Interest: The authors have completed the ICMJE uniform disclosure form (available at http://dx.doi.org/10.21037/jxym-21-8). The authors have no conflicts of interest to declare.

Ethical Statement: The authors are accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved. The study was conducted in accordance with the Declaration of Helsinki (as revised in 2013). The study was approved by the Ethics Committee of Shenzhen Children's Hospital (NO.: 202003802) and informed consent was waived as there were no patients involved.

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