SCD leads to the development and progression of acute myocardial infarction through the AMPK signaling pathway

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Research article

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Abstract

Background: Acute myocardial infarction (AMI) is myocardial necrosis caused by acute coronary ischemia and hypoxia. It can be complicated by arrhythmia, shock, heart failure and other symptoms that can be life-threatening. We constructed a multi-regulator driven dysfunction module for AMI. It is intended to explore the pathogenesis and functional pathways regulation of acute myocardial infarction.

Methods: Combining differential expression analysis, co-expression analysis, and the functional enrichment analysis, we obtained a set of expression disorder modules related to AMI. Hypergeometric test was performed to calculate the potential regulatory effects of multiple factors on the module, identifying a range of non-coding RNA and transcription factors.

Results: We obtained 4551 differentially expressed genes for AMI and seven co-expression modules. These modules are primarily involved in the metabolic processes of prostaglandin transport processes, regulating DNA recombination and AMPK signal transduction. Based on this set of functional modules, we revealed 3 of 24 transcription factors (TFs) including NFKB1, MECP2 and SIRT1, 3 of 782 non-coding RNA including miR-519D-3P, TUG1 and miR-93-5p were obtained. These core regulators are thought to be involved in the progression of AMI disease. Through the AMPK signal transduction, the critical gene stearoyl-CoA desaturase (SCD) can lead to the occurrence and development of AMI.

Conclusions: In this study, we used a dysfunction module to explore the pathogenesis of multifactorial mediated AMI and provided new methods and ideas for subsequent research. It helps researchers to have a deeper understanding of its potential pathogenesis. The conclusion provides a theoretical basis for biologists to design further experiments related to AMI.

Trial registration: All analyses were based on previous study, thus no ethical approval and patient consent are required. The dataset of GSE48060 is from GEO and the accession number is PRJNA208840.

Background

Acute myocardial infarction (AMI) is the major cause of high mortality rates of cardiovascular disease in men and women. Chest compression is the common symptom in patients with acute myocardial infarction\textsuperscript{1,2}. Myocardial infarction is common in the elderly and performance is usually not. Symptoms include confusion, weakness, chest pain, difficulty breathing, and vomiting\textsuperscript{3}. AMI is a subset of the acute coronary syndrome; it can be divided into ST-segment elevation myocardial infarction and non-ST-segment elevation myocardial infarction\textsuperscript{4}. ST-segment elevation myocardial ischemia-reperfusion injury can induce no flow, lead to myocardial necrosis and apoptosis, and even poor prognosis\textsuperscript{5}. AMI caused by coronary artery disease is the guiding reason of death in both developed and developing countries. Cardiovascular disease is a critical health problem in the United States. More than 6 million people were reported having AMI each year\textsuperscript{6,7}. Plaque rupture is the guiding cause of AMI in patients with coronary heart disease. There are 6\% to 12\% of myocardial infarction patients with normal angiographic coronary
artery. Microvascular obstruction after AMI is closely related to adverse ventricular remodeling, arrhythmia and adverse clinical outcomes. Influenza vaccination in patients with coronary artery disease has been shown to cause an increase in the incidence, morbidity, and mortality of AMI. Increasing research data suggested that myocardial infarction may be closely related to a variety of other diseases.

On the one hand, according to Alpert et al, myocardial infarction is usually the conclusion of atherosclerotic coronary disease. It is associated with thrombotic coronary occlusion, which causes coronary artery occlusion and myocardial ischemia and necrosis.

On the other hand, AMI is also a critical cause of morbidity and mortality in diabetic patients. AMI can develop into cardiogenic shock and mechanical complications, and central-source shock is the guiding cause of hospital death in patients with AMI. Cardiogenic shock is a rapid progression of AMI and usually is a fatal complication. Myocardial infarction is a crucial health problem and its mortality rate is more than double that of cancer. Some studies found that more than half of cardiovascular deaths are due to AMI. Although progress has been made in the early and long-term treatment of AMI, it is still the guiding cause of high morbidity and mortality in Western countries. AMI may be a severe complication of many diseases including hypertrophic cardiomyopathy and atrial fibrillation. In the past 20 years, the short-term prognosis of patients with myocardial infarction has been steadily improved after the introduction of beta blockers, thrombolysis, and aspirin. The common treatments including systemic thrombolysis and direct percutaneous coronary intervention are used for patients with AMI. A critical reduction in mortality of AMI has been achieved through aggressive strategies in early identification and intervention. Therefore, it is imperative for medical scientists and biologists in various countries to explore the pathogenesis and treatment mechanism of AMI. In this study, based on a multifactorial mediated dysfunction module for AMI, we performed a series of comprehensive analyses. This study will explore the relevant pathogenesis of AMI. In general, our comprehensive strategy provided not only new insights to the pathogenesis of AMI, but also abundant resources and guidance for biologists to design further experiments.

**Methods**

*Differentially Expressed Genes (DEGs) Analysis*

We collected an expression microarray dataset for the AMI disease sample of numbered GSE48060 from the NCBI Gene Expression Omnibus (GEO) database. We performed DEGs analysis of two sets (normal-recurrent acute myocardial infarction, normal-no recurrent acute myocardial infarction) on the collected disease samples, and calculated using the limma package in R. Finally, based on the combination of the two sets of genes, we obtained 4551 DEGs and then constructed a related expression matrix of AMI.

*Co-Expression Analysis Identifies Relevant Functional Modules*
We used the weighted gene co-expression network analysis (WGCNA)\textsuperscript{27} to analyze the gene expression profile matrices of the 4551 DEGs, and a gene module for synergistic expression was found. We used the correlation coefficient weighting value which is the gene correlation coefficient to the power of N. Then, we calculated the correlation coefficient between any two genes. The connections between genes in the network are subject to scale-free network distribution, making the algorithm more biologically meaningful. A hierarchical clustering tree was constructed by correlation coefficients between genes. The different branches of the clustering tree indicated different gene modules and different colors indicated different modules. Seven co-expression modules were extracted, which were identified as essential dysfunctional modules of AMI.

**Functional Enrichment Analysis to Identify Dysfunction Modules**

Exploring the function of genes and their involvement in signal transductions is an effective means of studying the molecular mechanisms of disease. The functions and pathways involved in the module gene can characterize the dysfunction mechanism of the module during the AMI process. For the genes of the dysfunction module, we performed functional enrichment analysis using Clusterprofiler package\textsuperscript{28} in R language to the Go functions (\textit{p}-value cutoff = 0.01, \textit{q}valueCutoff = 0.01) and the Kyoto Encyclopedia of Genes and Genomes (KEGG) pathways (\textit{p}-value cutoff = 0.05, \textit{q}valueCutoff = 0.2). According to the functions and pathways which were involved in the module gene, it was identified as a related dysfunction module of AMI. We performed GlueGO in Cytoscape\textsuperscript{29} software to functions and pathways for each module, to build corresponding function and access networks. Next, we identified the proportion of modules participating in the corresponding functions and pathways.

**Identification of Transcription Factors and ncRNA Regulation of Modules**

All human transcription factors (TFs) target data were downloaded from the TRRUST v2 database\textsuperscript{30}, 26 interaction pairs of 24 TFs were obtained. Human ncRNA-mRNA data (score > 0.5) were downloaded in a RAID 2.0 database\textsuperscript{31}, we obtained 1239 interaction pairs involving 782 ncRNA. To identify the regulatory effects of these TFs and ncRNA on the module, we performed a pivot analysis based on these interaction data. Pivot analysis refers to the search for a driver pair with at least two pairs of modules in a target pair. According to the hypergeometric test, we calculated the significance of the interaction between the drivers and the modules. We screened TFs with a \textit{p}-value < 0.01 and ncRNA as the pivot of the essential regulatory module. A statistical analysis was applied to the pivot, and it was identified as the core pivot.

**Results**

**The DEGs In AMI**

4551 DEGs (Supplementary Table S1) was screened from DEGs analysis. We observed the expression characteristics of this group of DEGs. It should be noted that the change in SCD is higher, which may be
related to AMI directly or indirectly. Moreover, it may have essential regulatory functions in the development of the disease. While, the analysis of DEGs expression in AMI still needs further study.

**Co-Expression Behavior of Genes Associated with AMI**

To study the mechanism of action of related genes in disease samples of patients with AMI systematically, we conducted extensive analysis. Expression profiles of 4551 DEGs in AMI patient samples were constructed. Based on the co-expression network analysis with the gray module removed, we obtained the expression of 7 groups of AMI-related disorders. These 7 sets of imbalance modules were identified as dysfunction modules, the DEGs contained within these modules all had a cooperative expression behavior (Figure 1). These dysfunction modules might be involved in different functions and pathways, indicating different regulatory mechanisms that may mediate the development of AMI.

**Dysfunction Modules Characterize the Pathogenesis of AMI**

Exploring the functions and pathways involved in DEGs is an essential mean for identifying their pathogenesis. To study the possible dysfunction caused by modular gene imbalance, we performed GO functions and KEGG pathways enrichment analysis on 7 modules. We collected a wealth of GO terms, including 2677 cell composition (CC) entries, 4274 molecular functional (MF) terms, and 22975 biological processes (BP) (Figure 2A, Supplementary Table S2). Based on functional enrichment analysis, we observed that relevant functional modules tend to involve in multiple disease-related functions, such as prostaglandin, transport of isoprenoid, metabolic process, regulation of DNA recombination, and nucleobase-containing compound transport.

On the other hand, 1148 KEGG pathway enrichment results (Figure 2B) reflect that the functional module genes are mainly involved in the AMPK signal transduction, PI3K-Akt signal transduction, and T cell receptor signal transduction.

Since the functional and pathway results of modular gene enrichment were strictly related to AMI, the 7 modules were identified as dysfunction modules. From the above data, we can find that the AMPK signal transduction may be closely related to the induction of AMI. Module genes can regulate a range of functions and pathways, and module dysregulation is likely to be an essential cause of morbidity. Based on the relationships between the modules, the corresponding functional and access networks were built, and identified the proportion of modules involved in the corresponding functions and pathways (Figure 2C). This might be the dysfunctional global mechanism of AMI. The dysregulation of genes of the module can trigger dysfunction of the module, which in turn affected the functions and pathways involved, and it guided to the occurrence and progression of the disease.

**The ncRNA and TFs Driving Acute Myocardial Infarction**
The transcription and post-transcriptional regulation of genes had been recognized as the critical factors, which regulated the development of diseases, and ncRNA was considered to be an essential regulator. Accurate prediction of ncRNAs that can regulate dysfunction module genes can facilitate the in-depth exploration of the transcriptional regulatory mechanisms of AMI. The ncRNA regulators were explored according to cpRNA-based pivot analysis, which can cause dysfunction of the module. The predicted conclusion (Figure 3, Supplementary Table S3) showed that 782 ncRNA had significant regulatory effects on the module, involving 1239 ncRNA-module interaction pairs. These ncRNAs affected the development of AMI in varying degrees. The analysis found that miR-519d-3p had crucial regulatory functions in 6 dysfunction modules, which affected the progression of AMI. Both TUG1 and miR-93-5p have meaningful regulatory relationships with 4 dysfunction modules and played an important role in module dysfunction. Other ncRNA exhibit significant modulation of dysfunction modules, and had essential functions in the regulation of acute myocardial infarction. The occurrence and development of diseases were inextricably linked to the imbalance of TFs, which was reflected in the regulation of TFs to dysfunction modules. The pivot analysis was performed to the module based on the regulatory relationship of the TFs and genes. The conclusion showed that (Figure 4, Supplementary Table S4), a total of 24 TFs had crucial transcriptional regulation of the dysfunction module of AMI, which involved 26 TF-Module interaction pairs. We performed statistical analysis on the regulation of these TFs and found that NFKB1 importantly regulated 2 dysfunction modules, thereby promoting the occurrence and development of AMI. Both MECP2 and SIRT1 had regulatory functions in 1 functional module and occupied an indispensable position in the potential pathogenesis of AMI.

Discussion

Myocardial infarction is the guiding cause of high mortality in all cardiovascular diseases\textsuperscript{32}. Mortality after myocardial infarction has decreased essentially over the past few decades, while there is still important in-hospital mortality\textsuperscript{33}. Therefore, research on the pathogenesis and treatment mechanism of AMI has become a top priority. Many biologists and medical researchers have invested in the pathogenesis of AMI. They mainly focus on certain genes, some researches had achieved results in protein and related signal transductions. We combined a series of analytical methods to explore the pathogenesis of AMI. The complete expression profile of AMI disease samples was constructed for DEGs analysis, and 4551 potential pathogenic genes were screened. The increased SCD content may be the key gene for AMI. The results of the study suggested that the onset of symptoms to coronary reperfusion appears to be the strongest factor affecting thrombus in myocardial infarction\textsuperscript{34}. Co-expression analysis of the DEGs revealed that we obtained 7 co-expression modules, the genes contained in the modules were considered to have synergistic expression. Based on the results of the functional enrichment analysis, we found that 7 modules were mainly involved in response to reactive oxygen species. According to studies by many scholars, N-acetylcysteine (NAC) is an antioxidant with active oxygen scavenging properties, which has the effect of enhancing nitroglycerin\textsuperscript{35}. The enrichment of pathways revealed that functional block genes were primarily involved in the AMPK signal transduction, which might trigger AMI. This suggested that the AMPK signaling cascade was thought to be the core signal transduction that triggered
AMI. Yang et al\textsuperscript{36} accentuated the AMP-activated protein kinase (AMPK) signal transduction has key functions in intracellular adaptation to energy stress during myocardial ischemia. Inhibition of AMPK signaling by Notch1 enhances cardiac dysfunction caused by myocardial infarction\textsuperscript{36}. We then identified the TFs which were involved in these 7 dysfunction modules, TFs were obtained, and there were 26 Pivot-Module interaction pairs. NFKB1 regulated 2 dysfunction modules, thereby promoting the occurrence and development of AMI. According to Boccardi et al\textsuperscript{37}, we were able to find that nuclear factor kappa B (NFκB) was involved in various human diseases, including atherosclerosis and myocardial infarction.

Studies had shown that the -94 ins/del ATTG NFKB1 gene variant might lead to a decrease in myocardial infarction sensitivity by a potential reduction in activation of NFκB, which in turn was associated with a decrease in plasma inflammatory markers\textsuperscript{37}. Both MECP2 and SIRT1 had regulatory functions in 1 functional module and occupied an indispensable position in the potential pathogenesis of AMI. On the one hand, the anti-apoptotic effect of miR-22 was to protect myocardial infarction by targeting MECP2\textsuperscript{38} directly. On the other hand, SIRT1 was known to be a nicotinamide adenine dinucleotide-dependent histone deacetylase, which makes the heart more resistant to ischemic injury. SIRT1 may be a new promising therapeutic target for myocardial infarction\textsuperscript{39}. The expression of SIRT1 was down-regulated by many stress stimuli in the heart. These stimuli might jointly drive the pathogenesis of AMI\textsuperscript{40}. Moreover, ncRNA had been recognized as an important regulator in the development and progression of the disease. In this regard, we perform a pivot analysis based on the targeting relationship between ncRNA and genes. The predicted results showed that 782 ncRNA had important regulatory effects on the modules, involving 1239 ncRNA-Module interaction pairs. These ncRNAs affected the development of AMI in varying degrees. The results of statistical analysis revealed that miR-519d-3p had important regulatory functions in 6 dysfunction modules, which might affect the progression of AMI. Down-regulation of miR-519d-3p and over expression of HOTAIR had been reported reducing myocardial apoptosis induced by myocardial infarction or hypoxia. It can provide a potential therapeutic target for myocardial infarction\textsuperscript{41}. Both TUG1 and miR-93-5p had essential regulatory relationships with the 4 dysfunction modules, and had important functions in module dysfunction. On the one hand, TUG1 can inhibit apoptosis in hypoxia-induced injury of H9c2 cell, thereby reducing hypoxia-induced cell damage and inhibiting myocardial infarction\textsuperscript{42}. On the other hand, the data showed that the expression of miR-93-5p had a cardioprotective effect in AMI. The Adipose-derived stromal cells (ADSCs)-derived exosomes enhanced by miR-93-5p could prevent cardiac damage by inhibiting autophagy and inflammatory responses\textsuperscript{43}. The series of regulatory factors predicted by this study had a certain degree of regulation on the pathogenesis of AMI. However, except for the key factors which were above-mentioned, other unmentioned ncRNAs and TFs might have functions in the mechanism of dysregulation of AMI, which needed to be further explored.

**Conclusions**

Overall, we indicated that the core key gene, SCD, was the responsible for the development of AMI. It not only provided a new way for biologists and pharmacologists to study the effects of AMPK signaling on
myocardial infarction, but also provided a valuable reference for their subsequent treatment options.

**Abbreviations**

AMI: Acute myocardial infarction; SCD: Stearoyl-CoA desaturase; DEGs: Differentially Expressed Genes; NFkB: nuclear factor kappa B; AMPK: AMP-activated protein kinase.

**Declarations**

**Ethical approval and patient consent**

All analyses were based on previous study, thus no ethical approval and patient consent are required.

**Consent for publication**

Not applicable.

**Availability of data and material**

Not applicable.

**Competing interests**

The authors declare that they have no competing interests.

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None.

**Authors' contributions**

LW and FY contributed equally to analyze the data and write the manuscript. All authors have read and approved the manuscript.

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**References**

1. Acharya D. Predictors of outcomes in myocardial infarction and cardiogenic shock. Cardiol Rev 2018; 26: 255-266.

2. Arora G, Bittner V. Chest pain characteristics and gender in the early diagnosis of acute myocardial infarction. Curr Cardiol Rep 2015; 17: 5.
3. Sinclair D. Myocardial infarction. Considerations for geriatric patients. Can Fam Physician 1994; 40: 1172-1177.

4. Yang EH, Brilakis ES, Reeder GS, Gersh BJ. Modern management of acute myocardial infarction. Curr Probl Cardiol 2006; 31: 769-817.

5. Liu ZB, Fu XH, Wei G, Gao JL. Cytochrome c release in acute myocardial infarction predicts poor prognosis and myocardial reperfusion on contrast-enhanced magnetic resonance imaging. Coron Artery Dis 2014; 25: 66-72.

6. Mulasari AS, Balaji P, Khando T. Managing complications in acute myocardial infarction. J Assoc Physicians India 2011; 59 Suppl: 43-48.

7. Scroggins NM. Recognition and management of inferior wall myocardial infarctions and right ventricular infarcts. J Infus Nurs 2001; 24: 263-267.

8. Bounhoure JP, Ouldzen H, Carrie D, Alibelli MJ, Puel J. Myocardial infarction with "angiographically normal coronary arteries" myth or reality? Bull Acad Natl Med 2007; 191: 815-824; discussion 24-25.

9. Klem I, Kim RJ. Assessment of microvascular injury after acute myocardial infarction: the importance of the area at risk. Nat Clin Pract Cardiovasc Med 2008; 5: 756-757.

10. Hebsur S, Vakil E, Oetgen WJ, Kumar PN, Lazarous DF. Influenza and coronary artery disease: exploring a clinical association with myocardial infarction and analyzing the utility of vaccination in the prevention of myocardial infarction. Rev Cardiovasc Med 2014; 15: 168-175.

11. Alpert JS, Thygesen KA, White HD, Jaffe AS. Diagnostic and therapeutic implications of type 2 myocardial infarction: review and commentary. Am J Med 2014; 127: 105-108.

12. Jacoby RM, Nesto RW. Acute myocardial infarction in the diabetic patient: pathophysiology, clinical course, and prognosis. J Am Coll Cardiol 1992; 20: 736-744.

13. Lee SI, Lee SY, Choi CH, Park KY, Park CH. Left heart decompression in acute complicated myocardial infarction during extracorporeal membrane oxygenation. J Intensive Care Med 2017; 32: 405-408.

14. Graf T, Desch S, Eitel I, Thiele H. Acute myocardial infarction and cardiogenic shock: pharmacologic and mechanical hemodynamic support pathways. Coron Artery Dis 2015; 26: 535-544.

15. Aymong ED, Ramanathan K, Buller CE. Pathophysiology of cardiogenic shock complicating acute myocardial infarction. Med Clin North Am 2007; 91: 701-712.

16. Norton M, Letizia M, Jennrich JA. Right ventricular infarction vs. left ventricular infarction: a review of pathophysiology, medical treatment, and nursing care. Medsurg Nurs 1993; 2: 203-209, 20.

17. Pollard TJ. The acute myocardial infarction. Primary care 2000; 27: 631-49; vi.
18. Bolognese L. Changing patterns of ST-elevation myocardial infarction epidemiology. Am Heart J 2010; 160: S1-S3.

19. Gupta T, Harikrishnan P, Kolte D, Khera S, Aronow WS, Mujib M, Palaniswamy C, Sule S, Jain D, Ahmed A, Lanier GM, Cooper HA, Frishman WH, Fonarow GC, Panza JA. Outcomes of acute myocardial infarction in patients with hypertrophic cardiomyopathy. Am J Med 2015; 128: 879-887 e1.

20. Tomcsanyi J, Tako K, Sharman B. Recurrent acute myocardial infarction as a thromboembolic complication of atrial fibrillation. Orv Hetil 2016; 157: 191-193.

21. Flapan AD. Management of patients after their first myocardial infarction. BMJ 1994; 309: 1129-1134.

22. Jariwala P, Chandra S. Diagnosis and management of failed thrombolytic therapy for acute myocardial infarction. Indian Heart J 2010; 62: 21-28.

23. Bovenzi F, De Luca L, de Luca I. Which is the best reperfusion strategy for patients with high-risk myocardial infarction? Ital Heart J 2004; 5 Suppl 6: 83S-91S.

24. Mintz E. Emergency department management of acute myocardial infarction. Mt Sinai J Med 1997; 64: 258-274.

25. Barrett T, Wilhite SE, Ledoux P, Evangelista C, Kim IF, Tomashevsky M, Marshall KA, Phillippy KH, Sherman PM, Holko M, Yefanov A, Lee H, Zhang N, Robertson CL, Serova N, Davis S, Soboleva A. NCBI GEO: archive for functional genomics data sets–update. Nucleic Acids Res 2013; 41: D991-D995.

26. Ritchie ME, Phipson B, Wu D, Hu Y, Law CW, Shi W, Smyth GK. limma powers differential expression analyses for RNA-sequencing and microarray studies. Nucleic Acids Res 2015; 43: e47.

27. Langfelder P, Horvath S. WGCNA: an R package for weighted correlation network analysis. BMC Bioinf 2008; 9: 559.

28. Yu G, Wang LG, Han Y, He QY. Cluster profiler: an R package for comparing biological themes among gene clusters. Omics: J integr Biol 2012; 16: 284-287.

29. Carlin DE, Demchak B, Pratt D, Sage E, Ideker T. Network propagation in the Cytoscape cyberinfrastructure. PLoS Comput Biol 2017; 13: e1005598.

30. Han H, Cho JW, Lee S, Yun A, Kim H, Bae D, Yang S, Kim CY, Lee M, Kim E, Lee S, Kang B, Jeong D, Kim Y, Jeon HN, Jung H, Nam S, Chung M, Kim JH, Lee I. TRRUST v2: an expanded reference database of human and mouse transcriptional regulatory interactions. Nucleic Acids Res 2018; 46: D380-D6.

31. Yi Y, Zhao Y, Li C, Zhang L, Huang H, Li Y, Liu L, Hou P, Cui T, Tan P, Hu Y, Zhang T, Huang Y, Li X, Yu J, Wang D. RAID v2.0: an updated resource of RNA-associated interactions across organisms. Nucleic Acids Res 2017; 45: D115-D118.
32. Kloner RA, Dai W, Hale SL, Shi J. Approaches to Improving cardiac structure and function during and after an acute myocardial infarction: acute and chronic phases. J Cardiovasc Pharmacol Ther 2016; 21: 363-367.

33. Grisel P, Roffi M, Muller H, Keller PF. Mechanical complications of myocardial infarction. Rev Med Suisse Romande 2011; 7: 1189-1192.

34. Silvain J, Collet JP, Guedeney P, Varenne O, Nagaswami C, Maupain C, Empana JP, Boulanger C, Tafflet M, Manzo-Silberman S, Keiries M, Brugier D, Vignalles N, Weisel JW, Jouven X, Montalescot G, Spaulding C. Thrombus composition in sudden cardiac death from acute myocardial infarction. Resuscitation 2017; 113: 108-114.

35. Pasupathy S, Tavella R, Grover S, Raman B, Procter NEK, Du YT, Mahadavan G, Stafford I, Hereszty T, Holmes A, Zetz C, Arstall M, Selvanayagam J, Horowitz JD, Beltrame JF. Early use of N-acetylcysteine with nitrate therapy in patients undergoing primary percutaneous coronary intervention for ST-Segment-elevation myocardial infarction reduces myocardial infarct size (the NICAM Trial [N-acetylcysteine in Acute Myocardial Infarction]). Circulation 2017; 136: 894-903.

36. Yang H, Sun W, Quan N, Wang L, Chu D, Cates C, Liu Q, Zheng Y, Li J. Cardioprotective actions of Notch1 against myocardial infarction via LKB1-dependent AMPK signaling pathway. Biochem Pharmacol 2016; 108: 47-57.

37. Boccardi V, Rizzo MR, Marfella R, Papa M, Esposito A, Portoghese M, Paolisso G, Barbieri M. -94 ins/del ATTG NFKB1 gene variant is associated with lower susceptibility to myocardial infarction. Nutr Metab Cardiovasc Dis 2011; 21: 679-684.

38. Feng Y, Huang W, Wani M, Yu X, Ashraf M. Ischemic preconditioning potentiates the protective effect of stem cells through secretion of exosomes by targeting Mecp2 via miR-22. PLoS One 2014; 9: e88685.

39. Ding M, Lei J, Han H, Li W, Qu Y, Fu E, Wang X. SIRT1 protects against myocardial ischemia-reperfusion injury via activating eNOS in diabetic rats. Cardiovasc Diabetol 2015; 14: 143.

40. Yang G, Weng X, Zhao Y, Zhang X, Hu Y, Dai X, Liang P, Wang P, Ma L, Sun X, Hou L, Xu H, Fang M, Li Y, Jenuwein T, Xu Y, Sun A. The histone H3K9 methyltransferase SUV39H links SIRT1 repression to myocardial infarction. Nat Commun 2017; 8: 14941.

41. Zhang D, Wang B, Ma M, Yu K, Zhang Q, Zhang X. LncRNA HOTAIR protects myocardial infarction rat by sponging miR-519d-3p. J Cardiovasc Transl Res 2019; 12: 171-183.

42. Jiang N, Xia J, Jiang B, Xu Y, Li Y. TUG1 alleviates hypoxia injury by targeting miR-124 in H9c2 cells. Biomed Pharmacother 2018; 103: 1669-1677.

43. Liu J, Jiang M, Deng S, Lu J, Huang H, Zhang Y, Gong P, Shen X, Ruan H, Jin M, Wang H. MiR-93-5p-containing exosomes treatment attenuates acute myocardial infarction-induced myocardial damage. Mol
