REVIEW

MiR-92a – a key player in cardiovascular remodeling

Jochen Dutzmann, Jan-Marcus Daniel, Johann Bauersachs, Daniel G. Sedding

Dept. of Cardiology and Angiology, Hannover Medical School, Carl-Neuberg-Str. 1, 30625 Hannover, Germany

Correspondence: Daniel G. Sedding
E-mail: sedding.daniel@mh-hannover.de
Received: March 16, 2014
Published: November 17, 2014

Small non-coding, highly conserved microRNAs (miRs) play a crucial role in gene regulation, especially in post-transcriptional gene silencing, and are important for vascular homeostasis as well as during pathophysiological vascular remodeling processes. MiR-92a is known to attenuate endothelial cell proliferation, and angiogenesis. Conversely, down regulation of miR-92a improves these endothelial cell-dependent processes. We recently showed that inhibition of miR-92a also accelerates the re-endothelialization process and thus prevents neointima formation following wire-induced injury of murine femoral arteries. Thus, inhibition of miR-92a may represent a promising therapeutic strategy for the treatment of vascular diseases.

Keywords: microRNAs; cardiovascular remodeling; re-endothelialization; endothelial cells

To cite this article: Jochen Dutzmann, *et al*. MiR-92a – a key player in cardiovascular remodeling. RNA Dis 2014; 1: e371. doi: 10.14800/rd.371.

Percutaneous coronary interventions for the treatment of coronary atherosclerosis count to the most frequently performed therapeutic procedures in medicine [1]. The inevitable endovascular injury triggers a healing process of the arterial wall resulting in neointimal hyperplasia and vessel remodeling [2]. Drug-eluting stents with local delivery of anti-proliferative agents are one of the most important achievements of modern interventional cardiology. These stents sufficiently prevent vessel re-narrowing and thereby markedly reduce the rate of repeat revascularization. However, the substances currently used on drug-eluting stent platforms often impair endothelial coverage of stent struts thus initiating a chronic inflammatory process, delaying arterial healing, and increasing the risk of thrombotic events [3]. Hence, the investigation of molecular strategies targeting specifically the function of distinct cell types i.e. smooth muscle cell proliferation without affecting endothelial cell regenerative capacity are coming in the focus of novel treatment approaches. In this context, selective enhancement of endothelial regeneration after vascular injury has recently been shown to prevent neointimal lesion formation [4].

Small non-coding microRNAs (miRs) involved in post-transcriptional gene silencing are known to control several physiological and pathophysiological processes in the vascular wall [5, 6]. MiR-92a is a member of the miR-17~92a cluster comprising six mature miRs, which are involved in the regulation of cell proliferation, development, immunity and tumorigenesis. Specifically, miR-92a has been shown to inhibit EC proliferation, angiogenesis, and vascular repair by an attenuation of the expression of validated target genes, i.e. the class III histone deacetylase sirtuin (Sirt)-1, integrin α5 (Itga5), and the flow-induced atheroprotective transcription factors Krüppel-like factor (Klf)-2 and Klf4 [7-10].

In initial observations, we detected miR-92a expression primarily in EC of uninjured murine vessels. Consecutively, we evaluated the spatiotemporal expression of miR-92a in a mouse model of wire-induced injury of the mouse femoral artery and found miR-92a levels significantly up-regulated in endothelial cells adjacent to the vascular injury site [11]. Following the overexpression of miR-92a in EC in vitro, we observed a significantly reduced proliferation and migration in ECs but not in SMCs, suggesting that the functional

impact of miR-92a is predominantly restricted to EC. Neither ECs nor SMCs showed changed apoptotic rates following sole overexpression of miR-92a [11]. We then abrogated miR-92a expression following vascular injury by the use of two different strategies. Both, systemic injection of locked nucleic acid-based (LNA)-antimiRs and specific deletion of endothelial miR-92a using Tie2-Cre; miR-92a (fl/fl)-mice accelerated the re-endothelialization process. Furthermore, we found a reduced number of $CD68^+$ monocytes/ macrophages accumulating in the lesions as well as a reduced number of proliferating $Ki67^+$ SMCs within the vessel wall, and an impairment of neointima lesion development. As expected, the expression of the pro-angiogenic miR-92a target genes Sirt1 and Itga5 was strongly increased at 2 weeks after wire-induced injury[11]. Itga5 interacts with fibronectin, which represents the major component of the early extracellular matrix, and its expression has been shown to be an important prerequisite for vascular regeneration [12]. The histone deacetylase Sirt1 regulates the expression and activity of numerous pro-angiogenic proteins and has been shown to have an inhibitory effect on neointima formation [13]. Both have a critical impact on EC migration and proliferation, indicating that therapeutic inhibition of miR-92a to stimulate re-endothelialization involves at least in part the de-repression of these factors [14].

In addition to neointima formation, miR92a has been shown to promote an atheroprone EC phenotype *inter alia* by suppression of the endothelial nitric oxide synthase in response to locally disturbed blood flow [6, 9]. Consistently, specific blockade of miR-92a in $LDLR^{-/-}$ mice reduced endothelial inflammation and decreased atherosclerotic plaque burden [15]. Interestingly, circulating levels of miR-92a are significantly downregulated in patients with coronary artery disease, which might represent a compensatory mechanism. Selective analysis of these patients for vasculoprotective drug therapies revealed a trend towards lower levels of miR-92a in patients with statin therapy compared with patients without statin therapy [16, 17]. Thus, low circulating levels of miR-92a likely represent a compensatory protective mechanism that might be boosted in response to statin therapy.

Expression of miR-92a is under control of several transcription factors [18, 19]. Most of them are known to be critically involved in vascular remodeling even before miRs came to the focus of research interest [11, 20, 21]. Inhibition of their transcriptional activity or targeting the respective downstream signaling pathways have been shown to prevent neointima formation. As mentioned above and in contrast to the expression of these transcription factors, miR-92a seems to be predominantly expressed in ECs, so that clinical inhibition of miR-92a might exert higher cell specificity than inhibition of the respective transcription factors.

Regarding a possible translational approach using miR-inhibitors, a LNA-based therapy targeting miR-122 using a modified DNA phosphorothioate antisense oligonucleotide that binds miR-122 in a stable heteroduplex, thereby inhibiting its function, was demonstrated to be safe and efficiant in a first-in-man phase II trial in patients suffering from chronic hepatitis C [22]. However, miR-based therapeutics have not yet entered the stage of clinical trials for cardiovascular disorders. But given the very recent successful inhibition of miR-92a in a large-animal model of cardiac ischemia/reperfusion which resulted in a significant reduction of infarct size on the one hand and the pivotal role of miR-92a in vascular diseases on the other hand suggest that inhibition of miR-92a might emerge as a novel, promising therapeutic approach for the treatment of cardiovascular diseases [23].

Acknowledgement

This work was supported by the Deutsche Forschungsgemeinschaft (DFG, German Research Foundation) for the Cluster of Excellence REBIRTH (from Regenerative Biology to Reconstructive Therapy) to DS and JB.

References

1. Nabel EG, Braunwald E. A tale of coronary artery disease and myocardial infarction. N Engl J Med. 2012;366:54-63.
2. Costa MA, Simon DI. Molecular basis of restenosis and drug-eluting stents. Circulation. 2005;111:2257-2273.
3. Stefanini GG, Holmes DR, Jr. Drug-eluting coronary-artery stents. N Engl J Med. 2013;368:254-265.
4. Curcio A, Torella D, Indolfi C. Mechanisms of smooth muscle cell proliferation and endothelial regeneration after vascular injury and stenting: Approach to therapy. Circ J. 2011;75:1287-1296.
5. Bonauer A, Boon RA, Dimmeler S. Vascular microrna. Curr Drug Targets. 2010;11:943-949.
6. Boon RA, Hergenreider E, Dimmeler S. Atheroprotective mechanisms of shear stress-regulated microrna. Thromb Haemost. 2012;108:616-620.
7. Bonauer A, Carmona G, Iwasaki M, Mione M, Koyanagi M, Fischer A, *et al*. Microrna-92a controls angiogenesis and functional recovery of ischemic tissues in mice. Science. 2009;324:1710-1713.
8. Doebele C, Bonauer A, Fischer A, Scholz A, Reiss Y, Urbich C, *et al*. Members of the microrna-17-92 cluster exhibit a cell-intrinsic antiangiogenic function in endothelial cells. Blood. 2010;115:4944-4950.
9. Wu W, Xiao H, Laguna-Fernandez A, Villarreal G, Jr., Wang KC, Geary GG, *et al*. Flow-dependent regulation of kruppel-like factor 2 is mediated by microrna-92a. Circulation. 2011;124:633-641.

10. Fang Y, Davies PF. Site-specific microrna-92a regulation of kruppel-like factors 4 and 2 in atherosusceptible endothelium. Arterioscler Thromb Vasc Biol. 2012;32:979-987.

11. Daniel JM, Penzkofer D, Teske R, Dutzmann J, Koch A, Bielenberg W, *et al*. Inhibition of mir-92a improves re-endothelialization and prevents neointima formation following vascular injury. Cardiovasc Res. 2014;103:564-572.

12. Stenzel D, Lundkvist A, Sauvaget D, Busse M, Graupera M, van der Flier A, *et al*. Integrin-dependent and -independent functions of astrocytic fibronectin in retinal angiogenesis. Development. 2011;138:4451-4463.

13. Li L, Zhang HN, Chen HZ, Gao P, Zhu LH, Li HL, *et al*. Sirt1 acts as a modulator of neointima formation following vascular injury in mice. Circ Res. 2011;108:1180-1189.

14. Guarani V, Deflorian G, Franco CA, Kruger M, Phng LK, Bentley K, *et al*. Acetylation-dependent regulation of endothelial notch signalling by the sirt1 deacetylase. Nature. 2011;473:234-238.

15. Loyer X, Potteaux S, Vion AC, Guerin CL, Boulkroun S, Rautou PE, *et al*. Inhibition of microrna-92a prevents endothelial dysfunction and atherosclerosis in mice. Circ Res. 2014;114:434-443.

16. Fichtlscherer S, De Rosa S, Fox H, Schwietz T, Fischer A, Liebetrau C, *et al*. Circulating microrna in patients with coronary artery disease. Circ Res. 2010;107:677-684.

17. Jiang Y, Wang HY, Cao HM, Wang CY, Zhang L, Wang H, *et al*. Peripheral blood mirnas as a biomarker for chronic cardiovascular diseases. Sci Rep. 2014;4:5026.

18. Brock M, Trenkmann M, Gay RE, Michel BA, Gay S, Fischler M, *et al*. Interleukin-6 modulates the expression of the bone morphogenic protein receptor type ii through a novel stat3-microrna cluster 17/92 pathway. Circulation research. 2009;104:1184-1191.

19. Aguda BD, Kim Y, Piper-Hunter MG, Friedman A, Marsh CB. Microrna regulation of a cancer network: Consequences of the feedback loops involving mir-17-92, e2f, and myc. Proc Natl Acad Sci U S A. 2008;105:19678-19683.

20. Shi Y, Fard A, Galeo A, Hutchinson HG, Vermani P, Dodge GR, *et al*. Transcatheter delivery of c-myc antisense oligomers reduces neointimal formation in a porcine model of coronary artery balloon injury. Circulation. 1994;90:944-951.

21. Smith RC, Wills KN, Antelman D, Perlman H, Truong LN, Krasinski K, *et al*. Adenoviral constructs encoding phosphorylation-competent full-length and truncated forms of the human retinoblastoma protein inhibit myocyte proliferation and neointima formation. Circulation. 1997;96:1899-1905

22. Janssen HL, Reesink HW, Lawitz EJ, Zeuzem S, Rodriguez-Torres M, Patel K, *et al*. Treatment of hcv infection by targeting microrna. N Engl J Med. 2013;368:1685-1694.

23. Hinkel R, Penzkofer D, Zuhlke S, Fischer A, Husada W, Xu QF, *et al*. Inhibition of microrna-92a protects against ischemia/ reperfusion injury in a large-animal model. Circulation. 2013;128:1066-1075.