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Corresponding Author: Jongmin Kim

Authors: Jongmin Kim[1,*]

Institution: [1]Division of Biological Sciences, Sookmyung Women's University, Seoul, 04310, Korea,

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Title: MicroRNAs as Critical Regulators of the Endothelial to Mesenchymal Transition in Vascular Biology

Author's name: Jongmin Kim

Affiliation: Division of Biological Sciences, Sookmyung Women's University, Seoul, 04310, Korea

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Corresponding Author's Information:

TEL: 82-2-710-9553

FAX: 82-2-2077-7322

E-mail: jkim@sookmyung.ac.kr

ABSTRACT

The endothelial to mesenchymal transition (EndMT) is a newly recognized, fundamental biological process involved in development and tissue regeneration, as well as pathological processes such as the complications of diabetes, fibrosis and pulmonary arterial hypertension. The EndMT process is tightly controlled by diverse signaling networks, similar to the epithelial to mesenchymal transition. Accumulating evidence suggests that microRNAs (miRNAs) are key regulators of this network, with the capacity to target multiple messenger RNAs involved in the EndMT process as well as in the regulation of disease progression. Thus, it is highly important to understand the molecular basis of miRNA control of EndMT. This review highlights the current fund of knowledge regarding the known links between miRNAs and the EndMT process, with a focus on the mechanism that regulates associated signaling pathways and discusses the potential for the EndMT as a therapeutic target to treat many diseases.

INTRODUCTION

Endothelial cells (ECs) line the inner surfaces of the blood vessels and lymphatic vessels in the body. ECs regulate vascular function by sensing and responding to various cues, and play a key role in the maintenance of vascular homeostasis (1). ECs have the capacity to undergo a dynamic cellular phenotypic switching, termed the endothelial to mesenchymal transition (EndMT), in response to local environmental cues throughout the vascular system. Since EndMT was initially described in relation to heart development (2), many studies have demonstrated the importance of the EndMT process during development (3-6). However, an increasing number of studies have demonstrated that EndMT is closely associated with postnatal pathological processes including cancer progression (7), tissue fibrosis (8), pulmonary arterial hypertension (9), neointima formation (10, 11), vascular calcification (12) and atherosclerosis (13), as well as cerebral cavernous malformations (14). The EndMT features are similar to the extensively studied and better understood epithelial to mesenchymal transition (EMT). During EndMT, ECs lose their ability to express endothelial markers, such as vascular endothelial cadherin (VE-cadherin), platelet endothelial cell adhesion molecule (PECAM-1, also known as CD31) and von Willebrand Factor (vWF). Subsequently, ECs lose their endothelial characteristics and display mesenchymal phenotypes characterized by acquisition of a highly invasive and migratory potential and gain of expression of mesenchymal markers such as alpha smooth muscle actin (α-SMA), smooth muscle protein 22 alpha (SM22α), fibronectin, vimentin and fibroblast specific protein-1 (FSP-1) (Fig. 1) (7-15). Although the molecular mechanisms underlying EndMT are complex and still largely unclear, the EndMT has been gradually defined based on studies of EMT processes, which are better understood in terms of molecular and cellular mechanisms (16). The EndMT can be regulated by multiple extracellular cues, microRNAs (miRNAs), transcription factors and various signaling pathways in different tissues and various

pathophysiological conditions. Among the many regulators that control the EndMT process, miRNAs are emerging as key regulators of the EndMT program. MiRNAs are a class of endogenous, small non-coding RNAs containing about 22 nucleotides that play an important role in post-transcriptional regulation of gene expression, typically through direct binding to the 3'-untranslated region of messenger RNA (mRNA) (17, 18). A single miRNA has the capacity to target multiple mRNAs; thus, it is not surprising that miRNAs affect the gene regulatory network and are involved in global cellular processes, including development, differentiation, cell death and cell proliferation (17-19). A growing number of studies have revealed that several miRNAs have the capacity to regulate EndMT processes and such regulatory roles of miRNAs in EndMT suggest potential therapeutic targets to prevent and treat many vascular diseases via modulation of miRNA levels. Here, we highlight the emerging role of miRNAs during the EndMT process and discuss the potential for EndMT as a therapeutic target to treat vascular diseases.

MICRORNA REGULATION OF THE ENDOTHELIAL TO MESENCHYMAL TRANSITION

To date, a growing body of evidence shows that EndMT and EMT are primarily controlled by common signaling pathways, including transforming growth factor-beta (TGF-β) signaling, Notch signaling, and proinflammatory signaling cascades (20-22). These signaling pathways can activate or upregulate common transcription factors such as the Twist, Snail, Slug, zinc finger E-box-binding homeobox 1 (ZEB1) and ZEB2 (20-22). These transcription factors upregulate the expression of mesenchymal markers such as N-cadherin, α-SMA, SM22α, calponin, vimentin, fibronectin and FSP-1, although the precise molecular mechanisms are not fully understood. At the cellular level, these transcription factors can initiate transcriptional reprogramming and subsequently, ECs lose their apical-basal polarity

and intercellular junctions, becoming mesenchymal-like cells during EndMT (20-23). While transcriptional control of EndMT has previously been studied extensively, an understanding of post-transcriptional control in this context has recently been sought, and investigated in several studies. Emerging studies have shown that miRNAs, key regulators of post-transcriptional regulation, are potent regulators of the EndMT process via targeting of key components associated with EndMT signaling pathways (Fig. 1) (11, 24).

MICRORNAS THAT INHIBIT THE ENDOTHELIAL TO MESENCHYMAL TRANSITION

TGF-β responsive miRNAs in EndMT

Several miRNAs act to inhibit EndMT by directly targeting transcription factors or inhibiting signaling pathways associated with induction of EndMT. Among the signaling pathways that activate the EndMT process, the TGF-β signaling represents the most well-known inducer of EndMT. Several studies have revealed that TGF-β significantly downregulates the expression of several miRNAs (such as miR-200a, miR-20a, miR-29 and miR-630) leading to activation of the EndMT process (25-28). The miR-200 family is composed of five members; miR-200a, miR-200b, miR-200c, miR-141 and miR-429. Before the study of EndMT, it had been demonstrated that the miR-200 family had inhibitory effects on EMT through targeting of ZEB1 and ZEB2 (29). A recent study investigated the role of miR-200a (which is well known to inhibit the EMT process) as it relates to EndMT (25). It was demonstrated that miR-200a expression was significantly downregulated after treatment with TGF-β1 of human aortic endothelial cells (HAECs), while TGF-β1 treatment upregulated the expression of growth factor receptor-bound 2 (GRB2). Overexpression of miR-200a resulted in significant inhibition of EndMT via downregulation of FSP-1 and α-SMA and upregulation of VE-cadherin and PECAM-1. At the same time, miR-200a targets *GRB2*

mRNA, which plays a vital role in the modulation of fibrosis and regulates the expression of endothelial and mesenchymal markers including FSP-1, α-SMA, VE-cadherin and PECAM-1 (25, 30, 31). The expression of miR-20a, a member of the miR-17-92 cluster, is also regulated by TGF-β1 treatment (26). In that study, Correia *et al*. demonstrated that the expression of miR-20a was decreased during TGF-β-induced EndMT, and miR-20a overexpression inhibited EndMT induction in human umbilical vein endothelial cells (HUVECs). During TGF-β-induced EndMT, endothelial marker VE-cadherin expression decreased while expression of mesenchymal marker SM22α and mesenchymal transcription factors Snail1, Snail2 and Twist increased. These suppressive effects were partially reversed by overexpression of miR-20a. Interestingly, it was shown that transforming growth factor beta receptor I (TGFBR1, also known as ALK5), TGFBR2, and smad anchor for receptor activation (SARA, also known as ZFYVE9), which are key components of canonical TGF-β signaling, were direct targets of miR-20a. Ectopic miR-20a expression decreased the levels of TGFBR1, TGFBR2 and SARA, and resulted in inhibition of EndMT (26). In addition to the miRNAs regulating key components of TGF-β signaling during EndMT, there are miRNAs that target EndMT-inducing transcription factors activated by TGF-β, such as the Slug. The combination of TGF-β and bone morphogenetic protein-4 (BMP-4) induces EndMT via downregulation of miR-630 expression in human dermal microvascular endothelial cells. The miR-630 was shown to directly target the Slug and inhibits EndMT (27). The MiR-29 is also downregulated in TGF-β-treated human dermal microvascular endothelial cells. Alteration of miR-29 expression is closely related to induction of EndMT via regulation of endothelial and mesenchymal markers and may be involved in the upregulation of dipeptidyl peptidase-4 (DPP-4) in kidneys of diabetic mice (28).

MiRNAs in the Interaction between FGF and TGF-β Signaling in EndMT

Several studies have suggested that the fibroblast growth factor (FGF) signaling modulates TGF-β signaling via regulation of many genes in various cell types (32-34). Fafeur *et al*. showed that treatment of FGF-2 in ECs inhibits the expression of the 85-kDa TGF-β receptor subunit and attenuates the EC growth inhibitory activity of TGF-β1 (32). It has also been found that FGF-2 antagonizes TGF-β1-mediated pericyte α-SMA expression (33) and that TGF-β-induced EMT is reversed by FGF-1 through the MAPK/ERK kinase pathway (34). These studies indicate that the interaction between FGF and TGF-β signaling may play a critical role in the regulation of the EndMT. Indeed, several studies have shown that endothelial FGF signaling counteracts TGF-β-induced EndMT via regulation of the expression of miRNAs including let-7 and miR-20a (11, 24, 26). Chen *et al*. demonstrated that disruption of baseline endothelial FGF signaling by knockdown of *FRS2* (a key adaptor molecule involved in FGF signaling) decreased the level of let-7 miRNA which, in turn, increased expression of its target mRNA, *TGFβR1*. This served to produce activation of TGF-β signaling, drove EndMT progression in human umbilical artery endothelial cells (HUAEC) and, in turn, led to neointima formation (11). Another study also showed evidence to substantiate the association of FGF signaling and let-7 in the induction of EndMT by the combination of TGF-β2, IL-1β and TNF-α, demonstrating that FGFR expression decreases in TGF-β2-, IL-1β- and TNF-α-treated HUVECs and leads to EndMT via downregulation of let-7 expression (24). In addition, it was also shown that FGF-2 increases miR-20a expression and regulates TGF-β signaling in HUVECs by inhibiting TGFBR1, TGFBR2, and SARA expression via upregulated miR-20a, leading to inhibition of EndMT. These findings suggest that endothelial FGF signaling plays a critical role in maintenance of endothelial homeostasis by regulating TGF-β-induced-EndMT via modulation of miRNA expression (26). However, the correlation between FGF signaling and EndMT is still controversial. Several studies have found that IL-1β, a key inducer of

EndMT, upregulates the expression of FGF-2 through PI3K activation and, in turn, leads to EndMT of corneal ECs (35, 36), suggesting the role of FGF-2 as an inducer of EndMT in the context of inflammation caused by IL-1β. This seeming controversy may be due to endothelial heterogeneity, *i.e.*, ECs of different organs differentially respond to various stimuli (37). Thus, it is necessary to understand the molecular mechanism of EndMT in the context of endothelial heterogeneity.

Other Important miRNAs in TGF-β-induced EndMT

It has been reported that other miRNAs are also involved in TGF-β-induced EndMT. The MiR-23 negatively regulates TGF-β-induced EndMT in mouse embryonic endothelial cells (MEEC) and identified hyaluronic acid synthase 2 (Has2) as a direct target of miR-23. MiR-23 plays an essential role in cardiac valve formation by regulating Has2 expression (38). Bayoumi *et al*. showed that overexpression of miR-532 inhibits EndMT in cardiac ECs (CECs), while knockdown of the miR-532 display increased the EndMT (39). MiR-532 plays a key role in the regulation of cardiac vascularization via direct targeting of prss23 (which regulates Snail signaling to induce EndMT in HAECs), and knockdown of miR-532 inhibits CEC proliferation and cardiac vascularization after acute myocardial infarction (39). Moreover, endothelial miR-155 is upregulated by both TGF-β stimulation and hypoxic conditions in MEECs, and the combination of TGF-β stimulation and hypoxia was noted to be even more powerful in upregulation of miR-155 level and induction of the EndMT. Increased miR-155 downregulates RhoA expression, which is essential for the EndMT, and thereby functions as a negative feedback loop regulating EndMT (40). Overall, these studies may provide novel insights into the molecular mechanism(s) of miRNAs in TGF-β-driven activation of EndMT and in regulation of EndMT-mediated pathological phenotypes.

MiRNAs Crosstalk during High Glucose-driven EndMT

The molecular mechanisms of endothelial dysfunction by high glucose have been elucidated in the pathologic context of various vascular diseases (41). Emerging evidence has demonstrated that high glucose can lead to EndMT via regulation of the expression of miRNAs (such as miR-200b and miR-18a-5p), and contribute to the progression of diabetic complications (42-44). Cao *et al*. showed that high glucose led to decreased expression of miR-200b, concomitantly decreased expression of endothelial markers and increased expression of mesenchymal markers in human retinal microvascular endothelial cells. High glucose levels also upregulated the expression of TGF-β1 and its downstream mediators of EndMT, such as Snail, Smad2 and p300, a known target of miR-200b. They further confirmed the role of miR-200b in the EndMT. EndMT was observed in the retinas of wild-type diabetic mice, but the process was prevented in miR-200b transgenic diabetic mice (42). This group also showed the role of miR-200b in EndMT in mouse heart endothelial cells in the context of diabetic cardiomyopathy (43). The MiR-18a-5p also inhibited EndMT via targeting Notch2 in the context of high glucose conditions in human aortic valvular endothelial cells. MiR-18a-5p expression was downregulated and, in turn, upregulated Notch2 expression after high glucose stimulation, resulting in the promotion of EndMT. In addition, ectopic miR-18a-5p expression decreased the levels of Notch2 (which is a target of miR-18a-5p), and subsequently inhibited the EndMT. The MiR-18a-5p overexpression also attenuated myocardial fibrosis in diabetic cardiomyopathy partially by inhibiting Notch2-mediated EndMT *in vivo* (44). Most of studies on the molecular mechanisms of EndMT have focused mainly on the TGF-β-mediated signaling pathway. However, further studies are also needed to understand the molecular mechanisms associated with EndMT induced by high glucose levels, inflammation, chemokines, growth factors and other stimuli.

MICRORNAS THAT PROMOTE THE ENDOTHELIAL TO MESENCHYMAL TRANSITION

On the contrary, several miRNAs have the capacity to promote EndMT by directly targeting molecules associated with inhibition of EndMT (45-49). For example, miR-21 expression increased during TGF-β-induced EndMT in HUVECs and inhibition of miR-21 has been shown to partially prevent TGF-β-induced-EndMT. In addition, it was found that miR-21 negatively regulated phosphatase and tensin homolog (PTEN), a well-known target of miR-21, and following activation of the Akt pathway, resulted in promotion of the EndMT. *In vivo*, miR-21 was upregulated in cardiac ECs during pressure overload-induced cardiac fibrosis, and it was attenuated by inhibition of miR-21 (45). It was reported that the level(s) of several miRNAs were differentially regulated during TGF-β-induced EndMT in mouse cardiac endothelial cells (MCECs) (46). Measured by miRNA array analysis, it was found that miR-125b, let-7c, let-7g, miR-21, miR-30b and miR-195 were upregulated and miR-122a, miR-127, miR-196 and miR-375 were downregulated during TGF-β-induced EndMT. Among these miRNAs, upregulation of miR-125b expression was validated in TGF-β-induced EndMT-derived cells and control MCECs by miRNA qPCR analysis. The expression of p53, a target of miR-125b and antagonizing TGF-β-induced profibrotic responses was significantly inhibited during EndMT of MCECs, which suggested the possibility that increased miR-125b downregulates p53, thereby promoting EndMT (46). Suzuki *et al*. also demonstrated that miR-27b (a member of the miR-23/24/27 cluster), is a positive regulator of TGF-β-induced EndMT in mouse pancreatic microvascular endothelial cells (MS-1). The expression of miR-27b was upregulated by TGF-β1 treatment and knockdown of miR-27 (by the locked nucleic acid miR-27b inhibitor) showed that suppression of TGF-β-induced EndMT. It was also found that miR-27 targets Elk1, neuropilin 2, Plexin A2 and Plexin D1 in the context of TGF-β-induced EndMT of MS-1 (47).

Using miRNA array analysis, Li *et al*. found that miR-130a is upregulated in a monocrotaline pulmonary hypertension mouse model and the inhibition of miR-130a partially reversed TGF-β-induced EndMT in lung microvascular endothelial cells. It was also found that miR-130a is regulated by NF-κB, and directly targets bone morphogenetic protein receptor 2 (48). Aside from TGF-β-induced EndMT and for hypoxia-induced EndMT, one group found that miR-126a-5p was elevated in hypoxia-subjected rat pulmonary microvascular endothelial cells (RPMECs), as well as a condition of persistent pulmonary hypertension in the newborn model. Concomitantly, decreased expression of PECAM-1 and increased expression of α-SMA in the hypoxic RPMECs was observed. Finally, it was found that inhibition of miR-126a-5p ameliorates hypoxia-induced EndMT (49). The EndMT-related miRNAs and target genes are summarized in Table 1 and Figure 1.

THE ENDOTHELIAL TO MESENCHYMAL TRANSITION AS A POTENTIAL THERAPEUTIC TARGET TO TREAT VARIOUS DISEASES

Many studies have demonstrated that fibroblasts are implicated in a multitude of pathologies, and there is substantial evidence to indicate that they are the central mediator of pathological tissue remodeling (50). Several studies have demonstrated that a large proportion of fibroblasts found in damaged tissues are of endothelial origin via EndMT (7, 51). These results suggest that the EndMT is an attractive prospective target with regard to prevention and treatment of many diseases. Indeed, the EndMT plays an essential role during development (52) and can also contribute to postnatal pathologies associated with many diseases such as fibrosis, neointima formation, diabetic complications, heterotopic ossification, Kawasaki disease and pulmonary arterial hypertension (11, 24, 25, 27, 28, 38, 42-46, 48, 49, 53). Given that EndMT is closely involved in multiple diseases, the blocking of EndMT may represent a useful strategy for implementation in the treatment plans

developed to combat human diseases. In addition, the miRNAs play a key role in the maintenance of homeostasis in the entire vasculature as they have the capacity to target multiple protein-encoding genes. Therefore, imbalances in the expression of miRNAs are closely related to the pathogenesis of many diseases via abnormal regulation of their target mRNAs. Thus, a strategy based on restoration of abnormal miRNA expression could have important therapeutic value for the treatment of various diseases. Finally, given the documented close relationship between EndMT and miRNAs as it occurs in various pathologies, modulation of miRNAs involved in EndMT processes could be a new therapeutic strategy for treatment of human diseases (17, 18). The therapeutic potential of modulation of miRNA expression in regulating the EndMT process in several diseases is summarized below.

As previously discussed, ECs represent a major source of the fibroblasts found in pathological fibrotic tissues via EndMT. In this context, let-7 miRNA plays a critical role in the regulation of EndMT via targeting components of TGF-β signaling in HUVECs and HUAECs. It has also been shown that let-7 miRNA is regulated by FGF signaling (11, 24). It was demonstrated that let-7 miRNA and FGF receptor expression and phosphorylation were suppressed in diabetic condition(s). In addition, it was found that the endogenous antifibrotic peptide N-acetyl-seryl-aspartyl-lysyl-proline (AcSDKP) restored let-7 miRNA levels by means of restoration of FGF receptor expression, as well as phosphorylation to normal levels. Thereby, AcSDKP inhibited EndMT, as evidenced by increased endothelial and reduced mesenchymal marker expression and, in turn, ameliorated diabetic kidney fibrosis. This suggests that AcSDKP could be a potential therapeutic option for diabetic kidney fibrosis as an endogenous antifibrotic molecule via inhibition of EndMT (24). The same group further showed the therapeutic potential of modulation of dysregulated miRNA during EndMT in the pathogenesis of diabetic kidney fibrosis (28). They found that DPP-4

could be induced in the diabetic kidney, and miR-29 expression was inhibited in diabetic mice (which identified DPP-4 as a direct target of miR-29). Thus, they identified the potential of DPP-4 inhibitor, which is generally used to treat diabetes mellitus type 2, for restoration of dysregulated miR-29 levels. As a result, DPP-4 inhibitor linagliptin restored miR-29 levels in diabetic kidney, and also found suppression of DPP-4 activity and protein expression. Thereby, EndMT was inhibited and, in turn, kidney fibrosis was ameliorated in diabetic kidney. These results suggest that linagliptin has potential therapeutic value for the restoration of normal kidney function in diabetic patients with kidney fibrosis via inhibition of EndMT (28).

Guo *et al*. revealed the role of kallistatin in the modulation of miRNA expression levels during TGF-β-induced-EndMT in HUVECs (54). Kallistatin is an endogenous protein and has an inhibitory effect on fibrosis via inhibition of TGF-β expression (55). MiR-21 is a central activator of EndMT and fibrosis (45). They therefore attempted to find out whether kallistatin plays a key role in the association of EndMT and miR-21. It was shown that kallistatin suppressed TGF-β-induced EndMT and counteracted TGF-β-mediated miR-21 upregulation and activation of components of downstream pathways, such as Akt, NF-κB and matrix metalloproteinase 2. These results suggested that kallistatin plays a key role in protection against pathologic fibrotic diseases by suppressing miR-21 mediated activation of EndMT (54). Another recent study demonstrated the therapeutic potential of modulation of miR-483 by suppression of EndMT by means of the direct targeting of connective tissue growth factor (CTGF) in Kawasaki disease (53). They found that sera from Kawasaki disease patients led to reduction of miR-483 and induction of CTGF, as well as increased mesenchymal markers, such as α-SMA, vimentin, and FSP-1 and decreased endothelial markers such as VE-cadherin and eNOS. They identified Krüppel-like factor 4 (KLF4) as the key transacting regulator that upregulates expression of miR-483 and, in turn, directly

targets *CTGF* mRNA. In addition, it was found that atorvastatin activated the KLF4-miR-483 axis, inhibited CTGF expression, and alleviated EndMT using sera from Kawasaki disease patients. This suggests that statin could be a potential therapeutic option in Kawasaki disease, in part, through inhibition of the EndMT (53). Taken together, the current evidence indicates that the EndMT plays a key role in the pathogenesis of various diseases and that targeting of the EndMT is a novel therapeutic approach for many diseases. In addition, the restoration of dysregulated miRNA during EndMT to normal levels may be an attractive therapeutic option in the treatment of EndMT-related diseases. Thus, further studies are needed to clarify the underlying signaling mechanisms associated with EndMT and miRNA-based therapies to regulate EndMT are urgently needed.

CONCLUSIONS AND FUTURE PERSPECTIVES

MiRNAs play a key role in regulating EndMT by targeting multiple components associated with signaling pathways that regulate EndMT. As described above, many studies support that dysregulation of miRNAs in vascular ECs leads to activation of the EndMT process and contributes to the pathogenesis of human diseases. Thus, strategies that restore miRNA expression to physiological levels are attractive therapeutic opportunities for the treatment of human diseases via inhibition of EndMT. Although our current understanding of the molecular mechanisms underlying the miRNA-EndMT axis is advancing, more work is still required to better understand the complex network of miRNAs and their target mRNAs involved in the EndMT process. In addition, understanding of the regulation of the reversible biological process of EndMT is necessary for the prevention and treatment of many diseases. In conclusion, studies of miRNA involved in EndMT will provide new insights into the molecular mechanisms of a broad variety of human pathologies and the identification of potential targets that are able to inhibit EndMT, will provide effective

therapeutic drugs for the treatment of human diseases.

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CONFLICTS OF INTEREST

The authors have no conflicting interests

FIGURE LEGENDS

Figure 1. MicroRNAs involved in EndMT. Schematic representation of significant miRNAs involved in EndMT regulation. During EndMT, ECs lose the expression of endothelial markers, such as VE-cadherin, PECAM-1, Tie-1, Tie-2, and vWF. Subsequently ECs gain of mesenchymal markers, such as N-cadherin, α-SMA, SM22α, fibronectin, vimentin and FSP-1. MiRNAs can promote or inhibit the EndMT program. Solid bars denote EndMT inhibiting miRNAs, solid arrows denote EndMT promoting miRNAs.

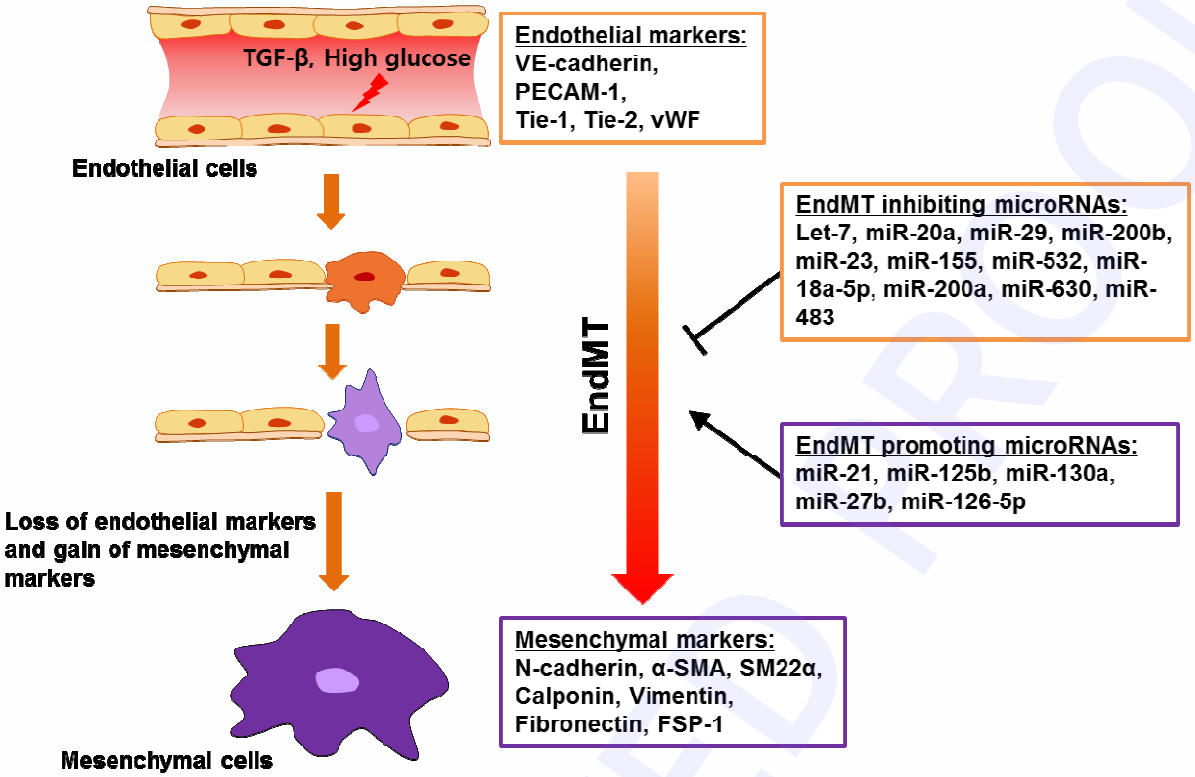

Table 1. MicroRNAs and target genes regulating the EndMT.

Endothelial Cell type	microRNA	Target	Effect on EndMT	Clinical relevance	Reference
HUAEC, HUVEC	Let-7	TGFβR1	Inhibit	neointima formation and fibrosis	11, 24
HAEC	miR-200a	GRB2	Inhibit	Cardiac fibrosis	25
HUVEC	miR-20a	TGFβR1, TGFβR2,SARA	Inhibit	Non determined	26
HD-MVEC	miR-630	Slug	Inhibit	Heterotopic ossification	27
HMVEC	miR-29	DPP-4	Inhibit	Diabetic nephropathy	28
MEEC	miR-23	Has2	Inhibit	Cardiac valve formation	38
CEC	miR-532	PRSS23	Inhibit	Acute myocardial infarction	39
MEEC	miR-155	RhoA	Inhibit	Non determined	40
HRMEC	miR-200b	Smad2, Snail	Inhibit	Diabetic retinopathy	42
MHEC	miR-200b	p300	Inhibit	Diabetic cardiomyopathy	43
HAVEC	miR-18a-5p	Notch2	Inhibit	Diabetic cardiomyopathy	44
HUVEC	miR-21	PTEN	Promote	Cardiac fibrosis	45
MCEC	miR-125b	p53	Promote	Cardiac fibrosis	46
MS-1	miR-27b	Elk1, Neuropilin 2, Plexin A2, Plexin D1	Promote	Non determined	47
LMVEC	miR-130a	BMPR2	Promote	Pulmonary arterial hypertension	48
RPMEC	miR-126-5p	Non determined	Promote	Neonatal pulmonary hypertension	49
HUVEC	miR-483	CTGF	Inhibit	Kawasaki disease	53

Abbreviations: human umbilical artery endothelial cell (HUAEC); human umbilical vein endothelial cell (HUVEC); human aortic endothelial cell (HAEC); human dermal

microvascular endothelial cell (HD-MVEC); human dermal microvascular endothelial cells (HMVEC); mouse embryonic endothelial cell (MEEC); cardiac endothelial cell (CEC); human retinal microvascular endothelial cell (HRMEC); mouse heart endothelial cell (MHEC); human aortic valvular endothelial cell (HAVEC); mouse cardiac endothelial cell (MCEC); mouse pancreatic microvascular endothelial cell (MS-1); lung microvascular endothelial cell (LMVEC); rat pulmonary microvascular endothelial cell (RPMEC).

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