Inhibitor of differentiation 1 has a helix-loop-helix (HLH) structure, belongs to a class of molecules known as the HLH trans-acting factor family, and plays an important role in advancing the cell cycle, promoting cell proliferation and inhibiting cell differentiation. Recent studies have confirmed that inhibitor of differentiation 1 plays an important role in the endothelial-mesenchymal transition of vascular endothelial cells, angiogenesis, reendothelialization after injury, and the formation and rupture of atherosclerotic plaques. An in-depth understanding of the role of inhibitor of differentiation 1 in atherosclerosis will provide new ideas and strategies for the treatment of related diseases.

Keywords: inhibitor of differentiation, endothelial-mesenchymal transition, angiogenesis, reendothelialization after injury, atherosclerosis

Introduction

Inhibitors of differentiation proteins were originally isolated from a mouse red blood cell line in 1990 and belong to the helix-loop-helix (HLH) transcription factor family. These proteins are named Id proteins because they can inhibit the binding of nuclear transcription factors to DNA (Jen et al., 1992). It was later discovered that Id proteins are negative regulators of nuclear transcription factors that are widely present in mammalian cells. To date, four Id family members (Id1-4) have been identified, which are encoded by four Id genes (Id1-4) located on different chromosomes. These genes all have a highly conserved HLH homology domain composed of two facultative \alpha-helices (Asirvatham et al., 2010). Their main function is to negatively regulate nuclear transcription factors, including suppressing the nuclear transcription of proto-oncogenes.

Atherosclerosis (AS) is the most common cause of cardiovascular disease. Atherosclerosis is driven by the accumulation of intimal lipids and chronic inflammation, and ultimately causes a series of ischemic changes in the organs due to the hardening of the blood vessel wall and the narrowing of the lumen (Falk, 2006). Early endothelial cell damage and repair, inflammation, and angiogenesis in the plaque are involved in the occurrence and development of AS plaques (Jaipersad et al., 2014).

The Molecular Structure of Id and Its Expression

Molecular structure of Id

The negative transcriptional regulatory factor Id is encoded by four Id genes located on different chromosomes, including 20p11, 2p25, 1p36, and 6p22-21, and forms four subtypes, including Id1, Id2, Id3, and Id4, which belong to the HLH transcription factor family. The HLH is composed of two relatively conserved alpha helices ~15–20 amino acids in size and a loop region with poorly conserved amino acids between them. Most HLH proteins have basic DNA-binding regions, which can bind to specific DNA sequences to exert biological effects. However, Id lacks similar DNA-binding sites and can bind to basic HLH (bHLH) family members to form a nonfunctional heterodimer, inhibit the transcription of downstream E-box or E-like sequence genes, and exert its dominant negative regulatory effect (Iwanicki and Brugge, 2009) (Fig. 1).
type II receptors, phosphorylating TGF-signaling pathway is activated by ligand binding to TGF-

GDF, and are essential in regulating the growth, develop-

ment and differentiation of cells and tissues. The TGF-β
signaling pathway is activated by ligand binding to TGF-β
type II receptors, phosphorylating TGF-β type I receptors

(ALK1 and ALK5), and then phosphorylating two R-Smad proteins, namely, Smad2/3 and Smad1/5; activated R-Smad and Smad4 form a complex in the nucleus to regulate target gene expression (Budi et al., 2017), such as that of plas-
minogen activator inhibitor-1 and Id proteins (Id1, Id2, Id3 and Id4). As one of the main members of the TGF-β family, BMP plays an important role in bone development, ho-

meostasis and diseases (Lowery and Vicki, 2018). Id1 is the direct target gene of BMP, and BMP-induced Id gene ex-

pression has been shown in different cell lines and embryonic stem cells. BMP upregulates Id1 expression without de novo protein synthesis (Korchynskyi and ten Dijke, 2002). The Smad-binding element and GC-rich region in BMP regulate the expression of the Id1 gene by binding to Smad1 and Smad5 (Gao et al., 2021). Interestingly, the vasodilatation observed in Id1 and Id3 double-knockout embryos is similar to the defective blood vessels in BMP-specific Smad1 or Smad5 knockout (Chang et al., 2020), indicating that the Id1 gene may be a key mediator of BMP activation in ECs.

As Id1 is related to the bHLH transcription factor, it is a differen-
tiation inhibitor (Zebedee and Hara, 2001). Therefore,

the early induction of Id1 may be the mechanism by

which BMP inhibits differentiation by controlling the activity

of the bHLH protein. For example, in neurodevelopment,

BMP is an important inhibitor of neuronal differentiation, and

transient induction of Id1 by BMP may reduce the stability of

the neurogenic bHLH transcription factor Mash1 (Viñals et al.,

2004). Similarly, the combination of Twist1 and
e protein (E47) inhibits BMP signal-mediated differentiation

of mesenchymal cells into osteoblasts, and Id1 induced by

BMP can relieve this inhibitory effect (Hayashi et al., 2007).
Therefore, we hypothesize that under the effects of BMP, Id1
induction helps to inhibit the bHLH transcription factor that is

involved in the differentiation of a variety of cell lines.

However, the inhibition of cell growth and partial differ-
tentiation induced by TGF-β is related to the down-
regulated expression of the gene encoding the Id protein (Huan et al., 2015). The induction of Id1 by TGF-β is de-
termined by the particular levels in the cells (Li et al., 2007).
In many epithelial cell lines, TGF-β downregulates Id1 expression (Di et al., 2006; Cao et al., 2009) and upregu-
lates Id1 expression in mouse hepatic stellate cells and human fetal lung fibroblasts (Chambers et al., 2003) (Wiercinska et al., 2006). In addition, the early induction of the Id1 gene by TGF-β depends on Smad3 but not Smad2. This effect is related to the binding of Smad3 to the upstream region of the Id1 promoter (Liang et al., 2009). ECs treated with TGF-
β exhibit a biphasic response: at low concentrations, the migration and proliferation of ECs are activated, while at high concentrations, these processes are inhibited (Gounmans et al., 2002). This effect depends on the activation of two different TGF-β receptors, ALK1 and ALK5. Low concen-
trations of TGF-β activate ALK1 signaling through Smad5, which can upregulate the expression of Id1 and increase the migration and proliferation of ECs. Under higher doses of TGF-β, Alk5 is activated, which induces PAI and may inhibit the migration and proliferation of ECs by downregulating Id1 (CelineSouilhol et al., 2018).

In short, as one of the most specific downstream target
genesis of TGF-β/BMP. Id1 can not only potentially modulate
the cellular effects of BMP signaling but also regulate Id1 to
maintain the proper response of cells to TGF-β.
Id1 Regulation of ECs and Reendothelialization

Id1 and senescence

Senescence is related to the gradual decline of cardiovascular structure and function. The increase in cardiovascular disease in aging is partly the result of the aging of vascular ECs and related vascular dysfunction. During this process, EC senescence is a pathophysiological process associated with structural and functional changes, including vascular tone disorders, increased endothelial permeability, arterial stiffness, impaired angiogenesis and vascular repair, and decreased EC mitochondrial biogenesis. Cellular senescence manifests as functional declines and low metabolism such as cell cycle arrest (Sabbatinelli et al., 2019).

It has been proposed that EC aging is mainly caused by telomere shortening (Bodnar et al., 1998). The expression of exogenous TERT has been shown to restore telomere length and extend cell lifespan (Duncan et al., 2000). Subsequent research showed that growth arrest of cultured human fetal cardiomyocytes was accompanied by an increase in p16INK4A and a decrease in Id gene expression (Ball and Levine, 2005). p16INK4A/pRb is the main mediator of cellular aging. p16INK4A-mediated senescence works through the retinoblastoma (Rb) pathway, inhibiting cyclin-dependent kinase (CDK) and leading to G1 cell cycle arrest (Anerillas et al., 2020). Id1 delays cell senescence mainly by regulating the expression of p16INK4A (Akakura et al., 2010). This finding indicates that the expression of Id1 is sufficient and necessary for the inhibition of p16INK4A. This effect is mediated at least, in part, by the inhibition of DNA binding of Ets2 transcription factors by Id1 (Ohtani et al., 2001). Although it has been suggested that Id inhibition of E protein also mediates the inhibition of p16INK4A, E47 overexpression does not upregulate the expression of endogenous p16 in 293T cells (Zheng et al., 2004). In addition, cells lacking endogenous Id1 have different senescence characteristics than ordinary cells. This phenomenon may be due to Id1 not completely inhibiting the expression of p16INK4A because other members of the Id family may also participate in cell senescence by affecting the p16INK4A/Rb pathway. For example, Id2 can bind to Rb and enhance S phase progression by weakening the growth inhibitory activity of the Rb protein (Strait et al., 2002).

BMP/Smad activation of EC migration and tube formation depends on the activation of Id1. This process may be caused by changes in the activity of transcription factors, leading to changes in the expression of metalloprotease and integrin genes (Valdimarsdotir et al., 2002). Overexpression of Id1 can also promote EC migration and tube formation, thereby simulating the effect of BMP/Smad on ECs. The upregulation of Id1 caused by endothelial injury can regulate the activation of ECs in a variety of ways (Fig. 2). Id1 can directly upregulate the expression of vascular endothelial growth factor (VEGF) and promote the proliferation and migration of ECs. The proliferation and migration of ECs can also promote the transcription of VEGF, which in turn promotes the expression of Id1, forming a positive feedback loop (Scharpfenecker et al., 2007). Another possible mechanism by which Id1 enhances ECs activation is as follows: by inhibiting bHLH transcription factor activity, E2–2 inhibits ECs proliferation, migration, and network formation by inhibiting vascular endothelial growth factor receptor 2 (VEGFR2) promoter activity, and this effect can be antagonized by Id1 (Tanaka et al., 2010). Id1−/− ECs showed increased expression of the CDK inhibitors p21 and p27 and impaired proliferation, which could be reversed by reducing the expression of E2–2 (Gadomski et al., 2020).

Id1 and reendothelialization

Endothelial injury is the main cause of AS and other cardiovascular diseases. When blood vessels are injured, endothelial progenitor cells (EPCs) are mobilized from the bone marrow into the blood, proliferate and migrate to the vascular injury site, and differentiate into mature ECs to facilitate vascular repair and endothelialization (Del Papa and Pignataro, 2018). In recent years, the potential clinical significance of EPCs has received increasing attention due to their key role in reendothelialization. The proliferation and migration of EPCs is the key mechanism of re-endothelialization after vascular injury. Past studies have shown that Id1 can be used as a marker of EPCs to track EPCs in bone marrow, blood, and tumor stroma. Id1 gene silencing can ablate bone marrow-derived EPCs and cause obvious defects in angiogenesis-mediated tumor growth (Moschetta et al., 2016). In addition, the production of EPCs in the bone marrow relies on Id1 inhibition of its target gene p21 (Ciarrocchi et al., 2007). The Id1 protein regulates the proliferation and migration of EPCs through various mechanisms and regulates the process of endothelialization (Wang et al., 2010). Id1 interacts with E2–2 to release E2–2-mediated FGFR1 and VEGFR2 expression inhibition to regulate EPC function (Yu et al., 2016). In addition, Id1 regulates the cell cycle to promote the proliferation of EPCs and induces the release of paracrine signals derived from EPCs to enhance the viability of neighboring ECs. These effects depend on the activation of Wnt2 signals (Xia et al., 2016). Percutaneous coronary intervention (PCI) is the most commonly used procedure for treating atherosclerosis and benefits most patients with AS, but there is still the risk of mechanical damage to the vascular endothelium, which will eventually lead to restenosis of the target vessel. Li et al. used a balloon injury rat model to simulate patients after PCI and found that Id1 could inhibit early reactive intimal
hyperplasia and promote the reendothelialization of injured blood vessels through NF-κB/survivin signal transduction (Li et al., 2019b). Mechanistically, Id1 affects the mobilization and proliferation of EPCs and promotes angiogenesis through NF-κB signaling. Id1 gene transfer endows human umbilical vein endothelial cells with angiogenic properties and rescues mice with ischemic limbs (Nishiyama et al., 2005).

**Id1 and EMT/Endothelial–Mesenchymal Transition**

Vascular endothelial cells (VECs) are a continuous physical barrier between the blood and the inside of the vessel wall that can maintain the stability of the vascular environment (Bischoff, 2019). Endothelial dysfunction usually refers to impaired endothelium-dependent vasodilation and increased contraction. This change is considered an early event of AS (Hong et al., 2018). Therefore, maintaining vascular endothelial function and preventing VECs damage is an important direction in the AS research field. VECs undergo endothelial–mesenchymal transition (EndMT) when stimulated by blood flow shear force, inflammatory factors, high fat, and other factors, which damages the barrier function of the endothelium (Sanchez-Duffhues et al., 2016). EndMT is a state of phenotypic transformation from ECs to mesenchymal cells in which ECs-specific markers (VE-cadherin, PECAM-1) are downregulated and mesenchymal cell-specific markers (α-SMA, Fibronectin, vimentin) are upregulated, with characteristic cell morphology and changes in proliferation, migration, and collagen synthesis ability (Nieto et al., 2016; Li et al., 2018). Recent studies have shown that EndMT plays an important role in heart and kidney fibrosis, pulmonary hypertension, cancer, and other diseases (Grande et al., 2015), and research on its role in the process of AS is intensified. Chen et al. (Evvard et al., 2016) used endothelial-specific lineage tracking and found that there were EndMT-derived ECs in AS plaques. These cells were related to the unstable plaque phenotype. Most studies have shown that EndMT is related to a reduction in Id protein expression (Moonen et al., 2010). Id1 promotes tumor metastasis and colonization by activating the EMT program in tumor cells (Castañón et al., 2017). Id1 induces the expression of mesenchymal markers (vimentin and fibronectin) by downregulating the zinc finger inhibitor protein ZBP-89 (Pillai et al., 2011).
Id1, as the most specific downstream target gene of Smad1/5, is important in antagonizing Snail (Castañón et al., 2017), Twist1 (Stankic et al., 2013a), and Slug (Asirvatham et al., 2007), which are induced by Smad2/3. TGF-β can also activate non-Smad pathways, such as ERK/MAPK, which regulates Id1 expression at the transcriptional level to facilitate the cross talk between non-Smad pathways and Smad pathways, regulate Smad2/3 phosphorylation, and ultimately regulate the EndMT process (Medici et al., 2011) (Fig. 3).

In addition, BMP, a member of the TGF-β family, promotes the migration of ECs and the formation of cardiac ducts by inducing the expression of Id1 (Jin et al., 2018). BMPs inhibit the EndMT process by activating Smad1/5 (Zhang et al., 2020). The BMPR-deficient ECs phenotype shifts to a contractile myofibroblast phenotype during actin bundle and myosin contraction, indicating that these cells undergo EndMT (Hiepen et al., 2019). LDH-198139 can inhibit BMP-Smad1/5 pathway signaling and reduce the level of Id1 protein. LDH-198139 was injected into Matrix Gla protein-null (MGP−/−) mice and was shown to prevent EndMT, cause a reduction in vascular calcification, and increase the survival rate in MGP−/− mice (Malhotra et al., 2015). In addition to the classic pathway, Wnt/β-catenin has also been reported to be involved in the regulation of EndMT (Li et al., 2019a). Id1 is a downstream target gene. Id1 knockout reduces the expression of β-catenin in the cytoplasm/nucleus and T cell factor/lymphoid enhancer factor luciferase activity and reduces expression of the Wnt target genes cyclinD1 and Survivin. Therefore, Id1 inhibits Wnt/β-catenin signaling and the EMT process (Sun et al., 2019).

In some systems, the role of Id1 in EMT seems to depend on its dominant negative effect on the E protein. Id1 binds with HEB to form a heterodimer and inhibits the binding of HEB and E-Boxes in the VE-cadherin gene promoter, thereby blocking the transcription of the VE-cadherin gene (Li et al., 2007). Cubillo et al. (2013) used chromatin immunoprecipitation to show that in complexes lacking Id1, E47 can directly bind to the endogenous VE-cadherin promoter in EndMT cells. In other cases, Id1 mediates the EMT process by interacting with cytoplasmic/membrane proteins (Zhang et al., 2007).

Studies have shown that the expression of Id1 is necessary to maintain the mesenchymal phenotype and cell viability after EndMT (Cubillo et al., 2013). The maintenance of EndMT status at the tumor metastatic colonization site depends on the Id1 target protein Twist1, while the maintenance of this cell status at the original site depends on Snail1 (Stankic et al., 2013b). Both Twist1 and Snail can drive EndMT to promote the development of atherosclerosis (Mahmoud et al., 2017). Id1-knockout colon cancer EMT cells showed increased VE-cadherin and decreased Snail and Twist, indicating that regulating Id1 may have good prospects for reversing EndMT. It is worth noting that in the early stage of TGF-β1-mediated EMT, Id1 can only inhibit the expression of ECs markers (VE-cadherin and ZO-1), but markers representing the mesenchymal phenotype (α-SMA, matrix metalloproteinase [MMP]-2, fibronectin, and integrin-linked kinase) did not change significantly (Li et al., 2007). This finding indicates that Id1 cannot induce complete EMT, which indirectly indicates that EMT is a multistep process, in which the loss of epithelial adhesion does not necessarily lead to complete mesenchymal transition.

### Id1 and Angiogenesis

There is increasing evidence that angiogenesis is a key factor in the growth and instability of AS plaques (Parma ...
During the development stage of AS, a variety of inflammatory stresses, oxidized lipids, and proteases stimulate the preexisting vasa vasorum in the plaque to generate new blood vessels, but the structure of these new blood vessels is immature and lacks the protection of the elastic layer and smooth muscle cells. Cytotoxic substances (such as oxidized lipids and oxidative stress) are produced in the plaque, causing damage to the plaque, leading to plaque rupture, and ultimately leading to death (Camare et al., 2017). Therefore, inhibiting neovascularization in the plaque is a potential therapeutic strategy for atherosclerosis.

Angiogenesis is a complex process that includes cell proliferation, migration, basement membrane degradation, and the maturation of new blood vessels (Xu et al., 2021). Early studies showed that in mouse models of cancer, in neovascularization, Id1 was necessary for normal blood vessel formation at the primary and metastatic sites of tumors (Gao et al., 2008). The blood vessels in Id1-KO animals lack the ability to branch and germinate and cannot form normal caliber lumens (Benezra et al., 2001). In ECs, Id1 can be upregulated by VECs growth factor (VEGF-A). VEGF-A is one of the most important factors that stimulates atherosclerotic plaque rupture (Wang et al., 2011). Id1 can also activate VEGF and promote angiogenesis through an autocrine pathway (Ling et al., 2005). The two factors interact to promote EPCs and mobilize hematopoietic precursors in the bone marrow. At present, Id1 is also considered to be a proangiogenic factor that is closely related to angiogenesis, in addition to VEGF, which is very important in AS-associated angiogenesis. MMPs can degrade extracellular matrix, destroy basement membrane, and affect angiogenesis in AS plaques (Chow et al., 2007). Id1 regulates the expression of MMP-14 and MMP-17, affects collagen degradation, and promotes ECs migration and angiogenesis in plaques (Chen et al., 2017).

A large number of studies have shown that different concentrations of ox-LDL play important roles in AS angiogenesis (Singh and Gautam, 2019; Liu et al., 2020). Low concentrations (<20 μg/mL) of ox-LDL induce Id1 nuclear-cytoplasmic shuttling through the PI3K pathway to promote ECs angiogenesis, thereby promoting plaque vulnerability and intravascular thrombosis (Qiu et al., 2012). High concentrations (>20 μg/mL) of ox-LDL reduce the proliferation and migration of ECs, and the level of nitric oxide accelerates AS. Id1 overexpression eliminates this inhibitory effect, which is achieved by inhibiting the nuclear translocation of P53 (Qiu et al., 2011b). In the early days, P53 was considered to be a tumor suppressor. Many studies have shown that an increase in p53 greatly promotes the development of CVD in chronic cardiovascular diseases through angiogenesis, programmed cell death, regulation of metabolism, and cell cycle arrest (Men et al., 2021). Qiu et al. (Wang Guixue et al., 2011) established a rabbit carotid artery stenosis model and found that vulnerable plaques mainly occur in the high shear stress area near the heart in the stenosis vessel. This area mainly promotes the formation of vulnerable plaques by promoting immature angiogenesis in the plaques. The Id1–p53 signaling pathway is an important signaling pathway through which shear stress regulates angiogenesis. Id1 and p53 are closely related to the regulation of angiogenesis. Id1 enhances migration and tubule formation by controlling the expression and function of p53 (Lee et al., 2009; Qiu et al., 2011a). In turn, p53 inhibits Id1 activity through its target DEC1 (Qian and Chen, 2008). In ECs, fluvastatin mediates the proangiogenic effects of statins by regulating Id1 and P53 (Pammer et al., 2004).

Interestingly, low-grade inflammation can also inhibit angiogenesis. For example, TNF-α, a classic inflammatory molecule, can transmit antiangiogenic signals through P53 to inhibit the activity of the Id1 protein (Panta et al., 2017). Truu et al. (2008) used TaqMan low-density arrays and showed that C-reactive protein could indirectly induce the protein expression of Id1 and regulate neovascularization in the inner membrane of vulnerable plaques. Recently, drugs that antagonize Id1 have shown preliminary promise in the treatment of pathological fundus angiogenesis (Wojnarowicz et al., 2019). Therefore, inhibiting Id1 is a good antiangiogenic strategy and may play an important role in the pathological process of atherosclerotic plaque instability.

Id1 and Atherosclerotic Plaque

Multiple studies have shown that Id1 is regulated by shear stress and may be an important force-sensitive factor. Zhang et al. (2018) constructed a low oscillation shear stress (OSS) model by ligating the carotid artery in ApoE−/− mice. Different shear stresses have different effects on Id1. Low shear stress (LSS 5 dyn/cm²) induced the continuous expression of Id1, but the effect of low oscillating shear stress (OSS 0.5 + 4 dyn/cm²) on Id1 was affected by time and was finally inhibited after a short activation. After Ni et al. (2010) performed gene microarray analysis, the expression of the Id1 gene in VECs was shown to be regulated by shear stress. However, there has been no further research report on the changes in endothelial function caused by the expression of Id1 under shear stress. In future studies, it will be interesting to determine whether shear stress-induced Id1 changes the functional state of ECs and contributes to subsequent plaque inflammation and instability.

Conclusions

In summary, as a dominant negative regulator, Id1 plays an important role not only in cell growth, development, cell cycle regulation, and tumor generation but also in EndMT, angiogenesis, re-endothelialization after injury, inflammation, and atherosclerosis. Id1 plays an important role in the occurrence and prevention of atherosclerotic plaque and provides a new target for the prevention and treatment of atherosclerosis in the future. However, the current role of Id1 in cardiovascular disease and its mechanism are not yet well understood, and there are still many aspects, such as its role in the formation and development of coronary heart disease and prevention and treatment, that are still unclear, and more research and discussion are needed.

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References

Akakura, S., Nochajski, P., Gao, L., Sotomayor, P., Matsui, S., and Gelman, I. (2010). Rh-dependent cellular senescence, multineurulation and susceptibility to oncogenic transformation through PKC scaffolding by SseCKS/AKAP12. Cell Cycle (Georgetown, Tex.) 9, 4656–4665.

Anerillas, C., Abdelmohsen, K., and Gorospe, M. (2020). Regulation of senescence traits by MAPKs. GeroScience 42, 397–408.

Asirvatham, A., Carey, J., and Chaudhary, J. (2007). ID1-, ID2-, and ID3-regulated gene expression in E2A positive or negative prostate cancer cells. Prostate 67, 1411–1420.

Ball, A., and Levine, F. (2005). Telomere-independent cellular senescence in human fetal cardiomyocytes. Aging Cell 4, 21–30.

Benezra, R., Raﬁi, S., and Lyden, D. (2001). The Id proteins and angiogenesis. Oncogene 20, 8334–8341.

Bischoff, J. (2019). Endothelial-to-mesenchymal transition. Circ Res 124, 1163–1165.

Bodnar, A., Ouellette, M., Frolkis, M., Holt, S., Chiu, C., Morin, G., et al. (1998). Extension of life-span by introduction of telomerase into normal human cells. Science (New York, N.Y.) 279, 349–352.

Budi, E., Duan, D., and Derynck, R. (2017). Transforming growth factor-β receptors and smads: regulatory complexity and functional versatility. Trends Cell Biol 27, 658–672.

Bischoff, J. (2019). Endothelial-to-mesenchymal transition. Circ Res 124, 1163–1165.

Bodnar, A., Ouellette, M., Frolkis, M., Holt, S., Chiu, C., Morin, G., et al. (1998). Extension of life-span by introduction of telomerase into normal human cells. Science (New York, N.Y.) 279, 349–352.

Budi, E., Duan, D., and Derynck, R. (2017). Transforming growth factor-β receptors and smads: regulatory complexity and functional versatility. Trends Cell Biol 27, 658–672.

Bischoff, J. (2019). Endothelial-to-mesenchymal transition. Circ Res 124, 1163–1165.

Bodnar, A., Ouellette, M., Frolkis, M., Holt, S., Chiu, C., Morin, G., et al. (1998). Extension of life-span by introduction of telomerase into normal human cells. Science (New York, N.Y.) 279, 349–352.

Budi, E., Duan, D., and Derynck, R. (2017). Transforming growth factor-β receptors and smads: regulatory complexity and functional versatility. Trends Cell Biol 27, 658–672.

Camaré, C., Pucelle, M., Négre, A., Salvayre, and Salvayre, R. (2017). Angiogenesis in the atherosclerotic plaque. Redox Biol 12, 18–34.

Cao, Y., Liu, X., Zhang, W., Deng, X., Zhang, H., Liu, Y., et al. (2009). TGF-beta repression of Id2 induces apoptosis in gut epithelial cells. Oncogene 28, 1089–1098.

Castañón, E., Soltermann, A., López, I., Román, M., Ecy, M., Collantes, M., et al. (2017). The inhibitor of differentiation-1 (Id1) enables lung cancer liver colonization through activation of an EMT program in tumor cells and establishment of the pre-metastatic niche. Cancer Lett 402, 43–51.

Chambers, R., Leoni, P., Kaminski, N., Laurent, G., and Heller, R. (2003). Global expression profiling of fibroblast responses to transforming growth factor-beta1 reveals the induction of differentiation-1 and provides evidence of smooth muscle cell phenotypic switching. Am J Pathol 162, 533–546.

Chang, Z., Wang, J., Jing, Z., Ma, P., Xu, Q., Nà, J., et al. (2020). Protective effects of isorhamnetin on pulmonary arterial hypertension: in vivo and in vitro studies. Phytother Res 34, 2730–2744.

Chen, Y., Zhang, K., Qiu, J., He, S., and Wang, G. (2017). Shear stress-mediated angiogenesis through Id1 relevant to atherosclerosis. MCB Mol Cell Biomech 14, 81–98.

Chow, A., Cena, C., and Schulz, R. (2007). Acute actions and novel targets of metalloproteases in the heart and vasculature. Br J Pharmacol 152, 189–205.

Ciarrocchi, A., Jankovic, V., Shaked, Y., Nolan, D., Mittal, V., Kerbel, R., et al. (2007). Id1 restrains p21 expression to control endothelial progenitor cell formation. PLoS One 2, e1338.

Cubillo, E., Diaz-Lopez, A., Cuevas, E., Moreno-Bueno, G., Peinado, H., Montes, A., et al. (2013). E47 and Id1 interplay in epithelial-mesenchymal transition. PLoS One 8, e59948.

Del Papa, N., and Pignataro, F. (2018). The role of endothelial progenitors in the repair of vascular damage in systemic sclerosis. Front Immunol 9, 1383.

Di, K., Ling, M., Tsao, S., Wong, Y., and Wang, X. (2006). Id-1 modulates senescence and TGF-beta sensitivity in prostate epithelial cells. Biol Cell 98, 523–533.

Draoui, N., P. de Zeeuw, and Carmeliet, P. (2017). Angiogenesis revisited from a metabolic perspective: role and therapeutic implications of endothelial cell metabolism. Open Biol 7, 1702119.

Duncan, E., Wadhwa, R., and Kaul, S. (2000). Senescence and immortalization of human cells. Biogerontology 1, 103–121.

Evrard, S., Lecce, L., Michelis, K., Nomura-Kitabayashi, A., Pandey, G., Purusothaman, K., et al. (2016). Endothelial to mesenchymal transition is common in atherosclerotic lesions and is associated with plaque instability. Nat Commun 7, 11853.

Falk, E. (2006). Pathogenesis of atherosclerosis. J American College Cardiol, 47, C7–C12.

Gadomski, S., Singh, S., Singh, S., Sarkar, T., Klarmann, K., Berenschot, M., et al. (2020). Id1 and Id3 maintain steady-state hematopoiesis by promoting sinusoidal endothelial cell survival and regeneration. Cell Rep 31, 107572.

Ganasssi, M., Badodi, S., Ortuste Quiroga, H., Zammit, P., Hinitis, Y., and Hughes, S. (2018). Myogenin promotes myocyte fusion to balance fibre number and size. Nat Commun 9, 4232.

Gao, D., Nolan, D.J., Mellick, A.S., Bambino, K., Mcdonnell, K., and Mittal, V. (2008). Endothelial progenitor cells control the angiogenic switch in mouse lung metastasis. Science 319, 195–198.

Gao, Y., Xin, L., Zhang, Y., Guo, X., Meng, Q., Li, Z., and Liao, Z. (2020). Technical and clinical aspects of diagnostic single-balloon enteroscopy in the first decade of use: a systematic review and meta-analysis. Gut Liver 15, 262–272.

Goumans, M., Valdimarsdottir, G., Itoh, S., Rosendahl, A., Sideras, P., and ten Dijke, P. (2002). Balancing the activation state of the endothelium via two distinct TGF-beta type 1 receptors. EMBO J 21, 1743–1753.

Grande, M., Sánchez-Laorden, B., López-Blau, C., De Frutos, C., Boutet, A., Áreválo, M., et al. (2015). Sna1l1-induced partial epithelial-to-mesenchymal transition drives renal fibrosis in mice and can be targeted to reverse established disease. Nat Med 21, 989–997.

Guixue, W., Juhui, Q., Jianjun, H., Yiming, Z., Qin, P., Tieyng, Y., et al. (2011). Id1-p53 regulates angiogenesis and participates in the mechanism of high shear stress-mediated vulnerable plaque formation. Chinese J Arteriosclerosis.

Hayashi, M., Nimura, K., Kashiwagi, K., Harada, T., Takaoka, K., Kato, H., et al. (2007). Comparative roles of Twist-1 and Id1 in transcriptional regulation by BMP signaling. J Cell Sci 120, 1350–1357.

Hiepen, C., Jatzlau, J., Hildebrandt, S., Kampfrath, B., Goktas, M., Murgai, A., et al. (2019). BMPR2 acts as a gatekeeper to protect endothelial cells from increased TGFβ responses and altered cell mechanics. PLoS Biol 17, e3000557.

Hong, L., Du, X., Li, W., Mao, Y., Sun, L., and Li, X. (2018). EndoMT: a promising and controversial field. Eur J Cell Biol 97, 493–500.

Huan, Q., Wang, Y., Yang, L., Cui, Y., Wen, J., Chen, J., and Chen, Z. (2015). Expression and function of the ID1 gene during transforming growth factor-β1-induced differentiation of human embryonic stem cells to endothelial cells. Cell Rep 17, 59–68.

Iwanicki, M., and Brugge, J. (2009). Transcriptional regulation of metastatic [Id]entity by KLF17. Genome Biol 10, 244.

Jaipersad, A., Lip, G., Silverman, S., and Shantsila, E. (2014). The role of monocytes in angiogenesis and atherosclerosis. J American College Cardiol, 63, 1–11.
et al.

Li, H., Zhao, Q., Chang, L., Wei, C., Bei, H., Yin, Y., et al.

Lee, J., Kang, M., Jang, S., Qian, T., Kim, H., Kim, C., et al. (2009). Id-1 activates Akt-mediated Wnt signaling and p27(Kip1) phosphorylation through PTEN inhibition. Oncogene 28, 824–831.

Li, H., Zhao, Q., Chang, L., Wei, C., Bei, H., Yin, Y., et al. (2019a). LncRNA MALAT1 modulates ox-LDL induced EndMT through the Wnt/β-catenin signaling pathway. Lipids Health Dis 18, 62.

Li, W., Du, D., and Li, Y. (2019b). Id-1 promotes re-endothelialization in the early phase after vascular injury through activation of NFκB/survivin signaling pathway. Drug Design Dev Ther 13, 3799–3811.

Li, Y., Lui, K., and Zhou, B. (2018). Reassessing endothelial-to-mesenchymal transition in cardiovascular diseases. Nature Reviews. Cardiology 15, 445–456.

Li, Y., Yang, J., Luo, J., Dedhar, S., and Liu, Y. (2007). Tubular epithelial cell dedifferentiation is driven by the helix-loop-helix transcriptional inhibitor Id1. J Am Soc Nephrol 18, 449–460.

Liang, Y., Brunificardi, F., and Lin, X. (2009). Smad3 mediates immediate early induction of Id1 by TGF-beta. Cell Res 19, 140–148.

Ling, M., Lau, T., Zhou, C., Chua, C., Kwok, W., Wang, Q., et al. (2005). Overexpression of Id-1 in prostate cancer cells promotes angiogenesis through the activation of vascular endothelial growth factor (VEGF). Carcinogenesis 26, 1668–1676.

Liu, H., Ma, X., Mao, Z., Shen, M., Zhu, J., and Chen, F. (2020). Circular RNA has_circ_0003204 inhibits oxLDL-induced vascular endothelial cell proliferation and angiogenesis. Cell Signal 70, 109595.

Lowery, J. W., and Vicki, R. (2018). The BMP pathway and its inhibitors in the skeleton. Physiol Rev 98, 2431–2452.

Mahmoud, M., Serbanovic-Canic, J., Feng, S., Souilhol, C., Xing, R., Hsiao, S., et al. (2017). Shear stress induces endothelial-to-mesenchymal transition via the transcription factor Snail. Sci Rep 7, 3375.

Malhotra, R., Burke, M., Martyn, T., Shakartz, H., Thayer, T., C. O’Rourke, et al. (2015). Inhibition of bone morphogeneic protein signal transduction prevents the medial vascular calcification associated with matrix Gla protein deficiency. PLoS One 10, e0117098.

Medici, D., Potenta, S., and Kalluri, R. (2011). Transforming growth factor-β2 promotes Snail-mediated endothelial-mesenchymal transition through convergence of Smad-dependent and Smad-independent signalling. Biochem J 437, 515–520.

Men, H., Cai, H., Cheng, Q., Zhou, W., Wang, X., Huang, S., et al. (2021). The regulatory roles of p53 in cardiovascular health and disease. Cell Mol Life Sci 78, 2001–2018.

Moonen, J., Krenning, G., Brinker, M., Koerts, J., M. van Luyn, and Harmsen, M. (2010). Endothelial progenitor cells give rise to pro-angiogenic smooth muscle-like progeny. Cardiovasc Res 86, 506–515.

Moschetta, M., Mishima, Y., Kawano, Y., Manier, S., Paiva, B., Palomera, L., et al. (2016). Targeting vasculosgenesis to prevent progression in multiple myeloma. Leukemia, 30, 1103–1115.

Ni, C., Qiu, H., Rezvan, A., Kwon, K., Nam, D., Son, D., et al. (2010). Discovery of novel mechano-sensitive genes in vivo using mouse carotid artery endothelium exposed to disturbed flow. Blood 116, e66–e73.

Nieto, M., Huang, R., Jackson, R., and Thiery, J. (2016). EMT: 2016. Cell 166, 21–45.

Nishiyama, K., Takaji, K., Kataoka, K., Kurihara, Y., Yoshimura, M., Kato, A., et al. (2005). Id1 gene transfer confers angiogenic property on fully differentiated endothelial cells and contributes to therapeutic angiogenesis. Circulation 112, 2840–2850.

Ohtani, N., Zebedee, Z., Huot, T., Stinson, J., Sugimoto, M., Ohashi, Y., et al. (2001). Opposing effects of Ets and Id proteins on p16INK4a expression during cellular senescence. Nature 409, 1067–1070.

Org, T., Duan, D., Ferrari, R., Montel-Hagen, A., Van Handel, B., Kerényi, M., et al. (2015). Scl binds to primed enhancers in mesoderm to regulate hematopoietic and cardiac fate divergence. EMBO J 34, 759–777.

Pammer, J., Reinisch, C., Kaun, C., Tschachler, E., and Wojta, J. (2004). Inhibitors of differentiation/DNA binding proteins Id1 and Id3 are regulated by statins in endothelial cells. Endothelium 11, 175–180.

Panta, S., Yamakuchi, M., Shimizu, T., Takenouchi, K., Oyama, Y., Koriyama, T., et al. (2017). Low grade inflammation inhibits VEGF induced HUVECs migration in p53 dependent manner. Biochem Biophys Res Commun 483, 803–809.

Parmar, L., Baghana, F., Quax, P., and de Vries, M. (2017). Plaque angiogenesis and intraplaque hemorrhage in atherosclerosis. Eur J Pharmacol 816, 107–115.

Perk, J., Iavarone, A., and Benezra, R. (2005). Id family of helix-loop-helix proteins in cancer. Nat Rev Cancer 5, 603–614.

Peverali, F.A., Ramqvist, T., Saffrich, R., Peperkok, R., Barone, M.V. and Philipson, L. (1994). Regulation of G1 progression by E2A and Id helix-loop-helix proteins. EMBO J 13, 4291–4301.

Pillai, S., Rizwani, W., Li, X., Rawal, B., Nair, S., Schell, M., et al. (2011). Id1 facilitates the growth and metastasis of non-small cell lung cancer in response to nicotinic acetylcholine receptor and epidermal growth factor receptor signaling. Mol Cell Biol 31, 3052–3067.

Qian, Y., and Chen, X. (2008). Id1 inhibitor of differentiation/DNA binding, is an effector of the p53-dependent DNA damage response pathway. J Biol Chem 283, 22410–22416.

Qiu, J., Peng, Q., Zheng, Y., Hu, J., Luo, X., Teng, Y., et al. (2012). OxLDL stimulates Id1 nucleocytoplasmic shuttling in endothelial cell angiogenesis via PI3K pathway. Biochim Biophys Acta 1821, 1361–1369.

Qiu, J., Wang, G., Hu, J., Peng, Q., and Zheng, Y. (2011a). Id1-induced inhibition of p53 facilitates endothelial cell migration and tube formation by regulating the expression of beta-integrin. Mol Cell Biochem 357, 125–133.

Qiu, J., Wang, G., Zheng, Y., Hu, J., Peng, Q., and Yin, T. (2011b). Coordination of Id1 and p53 activation by oxidized LDL regulates endothelial cell proliferation and migration. Ann Biomed Eng 39, 2869–2878.

Sabbatini, J., Prattichizzo, F., Olivieri, F., Procopio, A., Rippo, M., and Giuliani, A. (2019). Where metabolism meets senescence: focus on endothelial cells. Front Physiol 10, 1523.

Sanchez-Duffhues, G., Orlova, V., and Ten Dijke, P. (2016). In Brief: endothelial-to-mesenchymal transition. J Pathol 238, 378–380.
Sawada, S., and Littman, D.R. (1993). A heterodimer of HEB and an E12-related protein interacts with the CD4 enhancer and regulates its activity in T-cell lines. Mol Cell Biol 13, 5620.

Scharpfenecker, M., M. van Dinther, Liu, Z., R. van Bezooyen, Zhao, Q., Pukac, L., et al. (2007). BMP-9 signals via ALK1 and inhibits bFGF-induced endothelial cell proliferation and VEGF-stimulated angiogenesis. J Cell Sci 120, 964–972.

Singh, S., and Gautam, A. (2019). Upregulated LOX-1 receptor: key player of the pathogenesis of atherosclerosis. Curr Atheroscler Rep 21, 38.

Souilhol C, Martin, CHarmsen, Paul, CEvans, and GuidoKrenning. (2018). Endothelial–mesenchymal transition. Cardiovasc Res 114, 565–577.

Stankic, M., Pavlovic, S., Chin, Y., Brogi, E., and Benezra, R. (2013a). TGFβ-Id1 signaling opposes Twist1 and promotes metastatic colonization via a mesenchymal-to-epithelial transition. Cell Rep 5, 1228–1242.

Stankic, M., Pavlovic, S., Chin, Y., Brogi, E., Padua, D., Norton, L., et al. (2013b). TGF-β-Id1 signaling opposes Twist1 and promotes metastatic colonization via a mesenchymal-to-epithelial transition. Cell Rep 5, 1228–1242.

Strait, K., Dabbas, B., Hammond, E., Warnick, C., Istrup, S., and Ford, C. (2002). Cell cycle blockade and differentiation of ovarian cancer cells by the histone deacetylase inhibitor trichostatin A are associated with changes in p21, Rb, and Id proteins. Mol Cancer Ther 1, 1181–1190.

Sun, Y., Lai, X., Yu, Y., Li, J., Cao, L., Lin, W., et al. (2019). Inhibitor of DNA binding 1 (Id1) mediates stemness of colorectal cancer cells through the Id1-c-Myc-PLAC8 axis via the Wnt/β-catenin and Shh signaling pathways. Cancer Manage Res 11, 6855–6869.

Tanaka, A., Itoh, F., Nishiyama, K., Takezawa, T., Kurihara, H., Itoh, S., and Kato, M. (2010). Inhibition of endothelial cell activation by bHLH protein E2–2 and its impairment of angiogenesis. Blood 115, 4138–4147.

Turu, M., Slevin, M., Matou, S., West, D., Rodríguez, C., Luque, A., et al. (2008). C-reactive protein exerts angiogenic effects on vascular endothelial cells and modulates associated signaling pathways and gene expression. BMC Cell Biol 9, 47.

Viñals, F., Reiriz, J., Ambrosio, S., Bartrons, R., Rosa, J., and Ventura, F. (2004). BMP-2 decreases Mash1 stability by increasing Id1 expression. EMBO J 23, 3527–3537.

Wang, G., Qiu, J., Hu, J., Tang, C., and Yin, T. (2011). Id1: a novel therapeutic target for patients with atherosclerotic plaque rupture. Med Hypotheses 76, 627–628.

Wang, H., Yu, Y., Guo, R., Shi, Y., Song, M., Chen, J., et al. (2010). Inhibitor of DNA binding-1 promotes the migration and proliferation of endothelial progenitor cells in vitro. Mol Cell Biochem 335, 19–27.

Wiercinska, E., Wickert, L., Denecke, B., Said, H., Hamzavi, J., Gressner, A., et al. (2006). Id1 is a critical mediator in TGF-beta-induced transdifferentiation of rat hepatic stellate cells. Hepatology (Baltimore, Md.) 43, 1032–1041.

Wojnarowicz, P., Lima E Silva, R., Ohnaka, M., Lee, S., Chin, Y., Kulukian, A., et al. (2019). A small-molecule pan-Id antagonist inhibits pathologic ocular neovascularization. Cell Rep 29, 62–75.e7.

Wu, J., Boström, P., Sparks, L., Ye, L., Choi, J., Giang, A., et al. (2012). Beige adipocytes are a distinct type of thermogenic fat cell in mouse and human. Cell 150, 366–376.

Xia, X., Yu, Y., Zhang, L., Ma, Y., and Wang, H. (2016). Inhibitor of DNA binding 1 regulates cell cycle progression of endothelial progenitor cells through induction of Wnt2 expression. Mol Med Rep 14, 2016–2024.

Yu, Y., Liang, Y., Liu, X., Yang, H., Su, Y., Xia, X., and Wang, H. (2016). Id1 modulates endothelial progenitor cell functions through relieving the E2-2-mediated repression of FGFR1 and VEGFR2 in vitro. Mol Cell Biochem 411, 289–298.

Zebedee, Z., and Hara, E. (2001). Id proteins in cell cycle control and cellular senescence. Oncogene 20, 8317–8325.

Zhang, K., Chen, Y., Zhang, T., Huang, L., Wang, Y., Yin, T., et al. (2018). A novel role of Id1 in regulating oscillatory shear stress-mediated lipid uptake in endothelial cells. Ann Biomed Eng 46, 849–863.

Zhang, X., Ling, M., Wang, Q., Lau, C., Leung, S., Lee, T., et al. (2007). Identification of a novel inhibitor of differentiation-1 (ID-1) binding partner, caveolin-1, and its role in epithelial-mesenchymal transition and resistance to apoptosis in prostate cancer cells. J Biol Chem 282, 33284–33294.

Zhang, Y., Zhang, M., Xie, W., Wan, J., Tao, X., Liu, M., et al. (2020). Gremlin-1 is a key regulator of endothelial-to-mesenchymal transition in human pulmonary artery endothelial cells. Exp Cell Res 390, 111941.

Zhao, Z., Bo, Z., Gong, W., and Guo, Y. (2020). Inhibitor of differentiation 1 (Id1) in cancer and cancer therapy. Int J Med Sci 17, 995–1005.

Zheng, W., Wang, H., Xue, L., Zhang, Z., and Tong, T. (2004). Regulation of cellular senescence and p16 (INK4a) expression by Id1 and E47 proteins in human diploid fibroblast. J Biol Chem 279, 31524–31532.

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