Heliyon 10 (2024) e40118

Contents lists available at ScienceDirect

Heliyon

journal homepage: www.cell.com/heliyon

Review article

Endothelial-to-mesenchymal transition in the tumor microenvironment: Roles of transforming growth factor-β and matrix metalloproteins

Fei Du [a,b,c,f,1], Jing Li [d,1], Xiaolin Zhong [e], Zhuo Zhang [a,b,**], Yueshui Zhao [a,b,c,*]

[a] Laboratory of Molecular Pharmacology, Department of Pharmacology, School of Pharmacy, Southwest Medical University, Luzhou, Sichuan, China
[b] Cell Therapy & Cell Drugs of Luzhou Key Laboratory, Luzhou, Sichuan, China
[c] South Sichuan Institute of Translational Medicine, Luzhou, Sichuan, China
[d] Department of Oncology and Hematology, The Affiliated Traditional Chinese Medicine Hospital, Southwest Medical University, Luzhou, Sichuan, China
[e] Department of Gastroenterology, The Affiliated Hospital, Southwest Medical University, Luzhou, Sichuan, China
[f] Department of Pharmacy, Meishan TianFu New Area People's Hospital, Meishan, Sichuan, China

ARTICLE INFO

Keywords:
EndMT
TME
TGF-β
MMPs
Combination therapy

ABSTRACT

Cancer is a leading cause of global morbidity and mortality. Tumor cells grow in a complex microenvironment, comprising immune cells, stromal cells, and vascular cells, collaborating to support tumor growth and facilitate metastasis. Transforming growth factor-beta (TGF-β) is a multipotent factor that can not only affect fibrosis promotion but also assume distinct roles in the early and late stages of the tumor. Matrix metalloproteinases (MMPs) primarily function to degrade the extracellular matrix, a pivotal cellular player in tumor progression. Moreover, endothelial-to-mesenchymal transition (EndMT), similar to epithelial-to-mesenchymal transition, is associated with cancer progression by promoting angiogenesis, disrupting the endothelial barrier, and leading to cancer-associated fibroblasts. Recent studies have underscored the pivotal roles of TGF-β and MMPs in EndMT. This review delves into the contributions of TGF-β and MMPs, as well as their regulatory mechanisms, within the tumor microenvironment. This collective understanding offers fresh insights into the potential for combined targeted therapies in the fight against cancer.

1. Introduction

Transforming growth factor-beta (TGF-β) is an inactive dimeric cytokine produced by various cells. Its latency-associated peptide enables TGF-β to exhibit biological activity following protein hydrolysis [1,2]. TGF-β belongs to a family of structurally related

* Corresponding author. Laboratory of Molecular Pharmacology, Department of Pharmacology, School of Pharmacy, Southwest Medical University, Luzhou, Sichuan, China.
** Corresponding author. Laboratory of Molecular Pharmacology, Department of Pharmacology, School of Pharmacy, Southwest Medical University, Luzhou, Sichuan, China.
E-mail addresses: zhuozhang@swmu.edu.cn (Z. Zhang), yueshuizhao@swmu.edu.cn (Y. Zhao).
[1] These authors contributed equally to this work and share first authorship.

https://doi.org/10.1016/j.heliyon.2024.e40118
Received 28 August 2023; Received in revised form 26 October 2024; Accepted 3 November 2024
Available online 5 November 2024
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cytokines, including TGF-β1, TGF-β2, TGF-β3, bone morphogenetic protein, activin, and growth differentiation factor. These cytokines can exert diverse cellular effects on various regulatory processes within the body [3,4]. Numerous studies have demonstrated that aberrations in the TGF-β signal pathway are linked to the pathogenesis of certain human diseases [1,2,5,6]. TGF-β can induce fibrogenesis or affect the deposition of matrix proteins by regulating the expression of matrix metalloproteinases (MMPs) and tissue inhibitors of MMPs (TIMPs) [7–9]. Additionally, TGF-β can induce cell differentiation into myofibroblasts, thus modulating fibroblast phenotype and function [8,10]. Most notably, TGF-β is closely associated with cancer invasion and fibrosis in the tumor microenvironment (TME) [11].

Endothelial-to-mesenchymal transition (EndMT) was initially observed during cardiac development [12,13]. The EndMT process entails the conversion of endothelial cells (ECs) into interstitial fibroblast-like cells induced by certain inflammatory mediators (including pro-inflammatory factors and growth factors), such as interleukin-1β, tumor necrosis factor-α (TNF-α), and fetal growth factor [14,15]. Since the discovery of the EndMT process in close association with tumor progression in mouse models of melanoma and pancreatic tumors in 2007 [16], growing evidence suggests that tumors are intimately linked to the EndMT process (Table 1).

MMPs are the main proteolytic enzymes in the extracellular matrix (ECM), influencing cell migration and invasion [17]. The transcriptional levels of MMPs are typically tightly regulated during normal development. However, their regulatory mechanisms undergo alterations in cancer, particularly when MMP activity is associated with cancer progression [18]. Studies have shown that the dysregulation of MMPs can cause tumors to break through the basement membrane and extracellular matrix, invade surrounding tissues and metastasize to distant regions, or indirectly promote tumor growth, invasion and metastasis through the release of strom-related growth factors. Moreover, targeted inhibition of intracellular MMPs can effectively inhibit tumor metastasis and angiogenesis [19,20]. Currently, MMPs has become an attractive target for oncology research and anti-tumor drug development. However, TGF-β can upregulate the expression of MMPs in cancer cells (such as TIMP-2, MMP-2, and MMP-9), while the expression of MMP-2 and -9 can potentially activate TGF-β, establishing a vicious circle that promotes tumor progressio [21–23]. Fibroblast-like cells derived from EndMT may exhibit higher levels of MMPs than human fibroblasts [24]. This suggests a connection between the expression of TGF-β and certain MMPs with the process of EndMT [24,25]. This review provides a summary of the molecular mechanisms and clinical manifestations of the interactions between TGF-β, MMPs, and EndMT in the TME in recent years. It also analyzes the potential relationships among them in the TME.

2. Role of EndMT in the TME

The TME encompasses the entire in vivo milieu of tumor genesis, growth, and metastasis. This includes not only tumor cells but also various components such as fibroblasts, immune cells, blood vessels, and the ECM [48,49]. Both cellular and acellular elements of the TME collaborate to uphold tissue homeostasis and play a crucial role in impeding the proliferation of malignant cells.

ECs are specialized cells found within blood vessels throughout the body. Vascular ECs constitute a monolayer covering the inner surface of blood vessels. They serve as a barrier between the bloodstream and tissues and also participate in the body's immune functions. There is growing evidence indicating that ECs can serve as a source of myofibroblasts. This process is implicated in the fibrosis of organs such as the heart, lungs, kidneys, and intestines, as well as the formation of cancer-associated fibroblasts (CAFs; Fig. 1) [16,32,33,50,51].

Numerous factors induce EndMT. Currently, members of the TGF-β protein family are considered to be the primary inducers of EndMT, both in normal physiological processes and cancer [52,53]. Additionally, the WNT signaling pathway can influence the Smad signal transduction pathway [54]. The Notch signaling pathway triggers the proteolytic cleavage of transmembrane receptors,

Table 1
Roles of EndMT in different cancer types.

Type of cancer	Inducers	Role	References
Breast cancer	27-HC	Metastasis	[26,27]
PDAC	Acidic microenvironment	Metastasis	[28]
	TNF-α	Source of CAFs	[29]
HCC	SIRT3 and TGF-β	Angiogenesis	[30]
	Tumor-derived TGF-β	Angiogenesis	[31]
Colon cancer	Tubulin-β3 and TGF-β1/2	Source of CAFs	[32]
	Osteopontin	Source of CAFs	[33]
	TGF-β-IL6	Source of CAFs	[34]
CCM	TGF-β and BMP	Angiogenesis	[35–37]
Melanoma	NOX1 and NOX2	Metastasis and angiogenesis	[38]
	Not determined	Source of CAFs	[16,39]
	TGF-β	Metastasis and source of CAFs	[40,41]
Glioblastoma	TGF-β	Angiogenesis	[42]
Esophageal cancer	TGF-β2-IL1β	Source of CAFs	[43]
Lung cancer	TGF-β	Resistance to chemotherapy	[44,45]
	Radiation	Resistance to radiotherapy	[46,47]

27-HC, 27-hydroxy cholesterol; TNF-α, tumor necrosis factor-α; PDAC, pancreatic cancer; HCC, hepatocellular carcinoma; SIRT3, Sirtuin 3; CCM, cerebral cavernous malformation; TGF-β, transforming growth factor-β; BMP, bone morphogenetic protein; NOX, NAPDH oxidase; CAF, cancer-associated fibroblast.

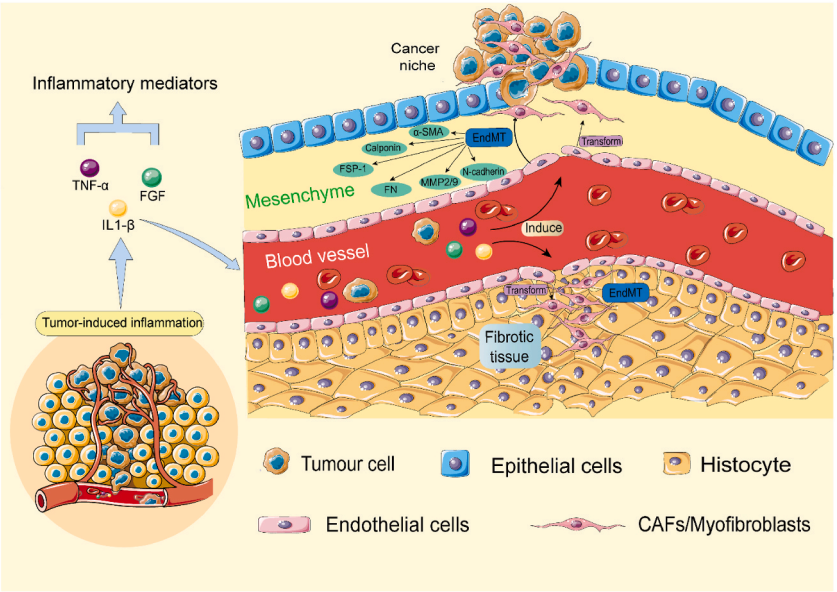

Fig. 1. Endothelial-to-mesenchymal transition (EndMT) in the tumor microenvironment (TME). Tumor-mediated inflammatory factors (such as interleukin-1-β (IL1-β), tumor necrosis factor-α (TNF-α), and fetal growth factor (FGF) can trigger the transformation of vascular endothelial cells into mesenchymal cells, promoting their conversion into CAFs or myofibroblasts. This process also upregulates the expression of fibroblast-specific proteins such as FSP-1, fibronectin (FN), calponin, α-smooth muscle actin (α-SMA), N-cadherin, MMP-2/9, and other proteins.

releasing the Notch intracellular domain (NICD) [55]. The NICD then translocates to the nucleus and binds to the DNA-binding protein CSL, subsequently influencing the EndMT process by recruiting coactivators [56–59]. As a significant marker of TME, the hypoxic environment not only plays an important role in promoting angiogenesis but also has been demonstrated to effectively induce EndMT through the hypoxia-inducible factor-dependent pathway [60–63].

The first study on the relationship between EndMT and tumors was conducted in a mouse model of melanoma and pancreatic tumor, reported in 2007 [16]. Since then, growing evidence has firmly established that EndMT is closely associated with a variety of tumors. Angiogenesis is a crucial pathway for tumor progression in the TME [64]. In recent years, the expression of major EndMT transcription factors (Slug and Snail) has been identified in the ECs of tumor vasculature in many cancers. The expression of Slug is linked to angiogenesis and tumor metastasis [65–67]. As it has been established, CAFs represent a primary component of the TME, capable of influencing the initiation and progression of tumors through the secretion of various cytokines and chemokines [68,69]. Myofibroblasts or mesenchymal cells (MCs) generated by ECs through EndMT constitute a significant source of CAFs [16,70]. Additionally, EndMT disrupts the endothelial barrier, facilitating the migration of tumor cells [40]. This underscores the crucial role of EndMT in the advancement of tumors.

3. TGF-β and EndMT in the TME

3.1. General cognition of TGF-β

TGF-β was initially isolated from virally and chemically transformed cells, exploiting its cell-transforming ability, later, it was discovered to be expressed in various tumors and cell types [71,72]. Mammals typically express three subtypes of TGF-β: TGF-β1, TGF-β2, and TGF-β3. These correspond to loci 19q13, 1q41, and 14q24 on human gene chromosomes, respectively [73,74].

Bioactive TGF-β exists as a dimer, and its intracellular signal transduction pathway involves binding to serine/threonine kinase receptors type I and II (TβRI and TβRII) on the cell surface. Notably, activation of the TβRI kinase domain is triggered by the binding of TGF-β and TβRII [75]. Within cells, phosphorylated ligands prompt the release of smad2 and smad3 signal transduction on the inner surface of the plasma membrane. Subsequently, the phosphorylated dimer Smad2/3 combines with the common Smad4 to form a heterotrimeric complex. Following this, the Smad heterotrimeric complex is translocated to the nucleus, where it regulates the expression of various target genes through interactions with other promoters [76].

The expression of Smad-6 and Smad-7 can inhibit the Smad signal transduction pathway of TGF-β, with Smad-7 also being regulated by the TGF-β signal. Mechanistically, Smad6 suppresses signal transmission by influencing the Smad1-CoSmad process and forming an inactive Smad1-Smad6 complex. However, Smad7 negatively regulates signal transduction of TGF-β superfamily members by impeding the binding and phosphorylation of Smad molecules to receptors [77].

Apart from the Smad2/3 pathway, TGF-β can activate several non-Smad signal transduction pathways. TGF-β can engage mitogen-activated protein kinase (MAPK) pathways, such as p38 MAPK, c-JUN-N terminal kinase (JNK), and extracellular signal-regulated

kinase (ERK) [78,79]. These pathways function independently of Smad but can affect the response of Smad-dependent TGF-β [78–83]. Additionally, the Wnt and Notch signals have been demonstrated to regulate the functions of TGF-β [57,58,78].

TGF-β is involved in various biological processes, encompassing cell growth, differentiation, development, and tumor progression [73,84]. Studies have shown that all cells (including most immune cells) can secrete TGF-β1, TGF-β2 is mainly secreted by epithelial cells and nerve cells, and TGF-β3 is mainly expressed in mesenchymal cells.Therefore, TGF-β has a wide range of effects. However, the mechanism of action of TGF-β often depends on the specific situation of the body [85]. In processes of growth and development, TGF-β can promote EndMT, induce epithelial-to-mesenchymal transition (EMT), and facilitate fibroblast-to-myofibroblast transition. Additionally, TGF-β has been associated with tissue development and repair [86]. In advanced cancer TME, TGF-β-induced EMT can stimulate tumor invasion, metastasis, and drug resistance in tumor cells [87,88]. EndMT, induced by TGF-β, has also been established to be connected with tumor progression. EndMT is a crucial source of CAFs in tumors [89]. Furthermore, acting as a major pro-fibrosis cytokine, TGF-β can induce various transformation processes and promote the progression of fibrosis-related diseases [8,90–92].

3.2. Role of TGF-β in EndMT

EndMT is typically instigated by inflammatory mediators, prompting the transformation of ECs into stromal fibroblast-like cells, thereby influencing disease progression [14,15]. The regulatory mechanism governing EndMT is intricate, with the most well-known induction being attributed to the TGF family. TGF-β binds both TGF-β type I receptors (TβRI) and TGF-β type II receptors (TβRII), forming polymeric complexes that subsequently activate downstream Smad-dependent pathways and other non-Smad signaling pathways (Fig. 2) [93]. In the conventional Smad-dependent signaling pathway, activation of the TGF-β receptor (TβRI) on the EC membrane leads to the binding and phosphorylation of Smad2 and Smad3. The resulting Smad2/3 phosphorylation complex interacts with Smad4, ultimately translocating to the nucleus and activating genes (such as Snail, Twist, Zeb, and Slug), which promote tumor invasion and migration (Fig. 2) [93–95]. Furthermore, certain members of the TGF-β family (TGF-β2, BMP2, and BMP4) exert effects on both Smad-dependent and non-Smad signal transduction pathways through TβRII (Fig. 2) [93]. The EndMT process often disrupts homeostasis in the TME. To confirm the occurrence of EndMT, commonly selected mesenchymal markers include α-smooth muscle actin (α-SMA), N-cadherin, calmodulin (calponin), fibroblast-specific egg protein-1 (FSP-1), fiber-linked egg whey (fibronectin), and MMPs such as MMP-2 and MMP-9 (Fig. 2) [96,97].

As established, the onset and progression of tumors are often closely associated with prolonged inflammatory stimulation [98,99]. TGF-β can induce the EndMT process in certain cancers (e.g., hepatocellular carcinoma, colon cancer, melanoma), leading to the conversion of ECs into CAFs, thereby promoting angiogenesis and tumor migration [100] (Table 1). Additionally, inflammatory factors can stimulate the EndMT process in the TME by activating WNT, Notch, and hypoxia signaling pathways [24,101,102]. Among these, the WNT pathway can collaborate with the TGF-β pathway to activate downstream Smad-dependent signaling and thus promote tumor development [101].

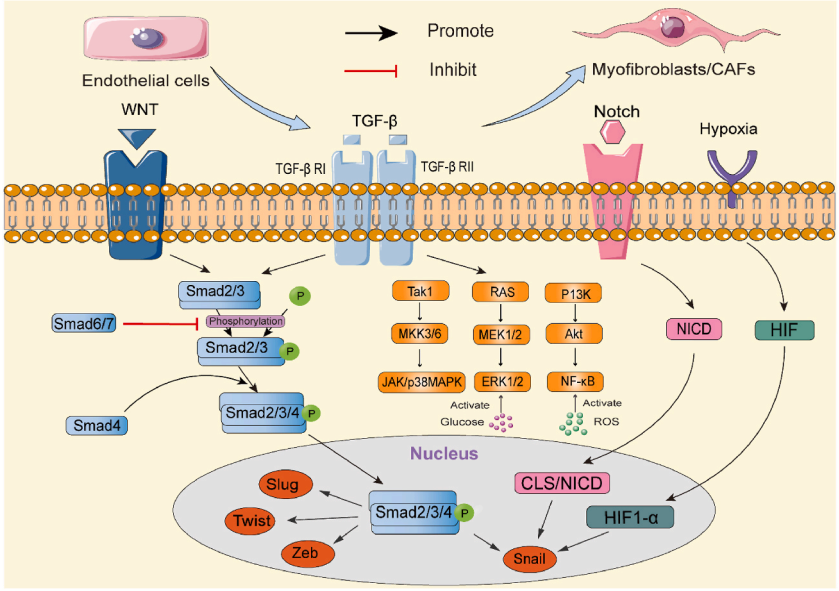

Fig. 2. Molecular pathways and downstream signaling in the process of EndMT. The molecular pathways of EndMT in endothelial cells encompass WNT, TGF-β, Notch, and hypoxia signaling pathways. EndMT induced by transforming growth factor-β involves a typical Smad2/3 pathway. Furthermore, it can activate Smad-independent pathways (Tak1, RAS, and PI3K signaling pathways). These ultimately enhance the expression of Slug, Twist, ZeB, and Snail proteins.

3.3. TGF-β in other biological processes in the TME

In the TME, TGF-β serves a multifaceted role. It not only induces EndMT but also plays a role in various biological processes, including the induction of EMT in cancer. Its function varies in different stages of cancer. During the early stages of epithelial carcinogenesis, TGF-β counters the effects of epithelial injury or stress stimulation through its antiproliferative and pro-apoptotic effects [103]. However, in the advanced stages of cancer, tumor cells manipulate TGF-β signal components through various mechanisms to diminish its growth inhibitory effects. Simultaneously, TGF-β induces the EMT process, promoting tumor cell invasion and migration, thus driving tumor progression [87,103,104].

TGF-β also interacts with immune cells. For instance, it can hinder the antigen presentation ability of dendritic cells by suppressing the expression of the MHC-II gene. Additionally, it can impede the function of natural killer cells at multiple levels (e.g., TGF-β downregulates the expression of interferon-γ and TBET genes in natural killer cells, thereby inhibiting the Th1 response) [105–108]. Human macrophage subsets can modulate the activity of TGF-β by influencing the activity of integrin αvβ8 and MMP-14 [109].

In the ECM, TGF-β also plays a role in the regulation of collagen function. For instance, endotrophin, released as a signaling molecule from collagen type VI, has been demonstrated to regulate adipose tissue fibrosis and metabolic dysfunction through the TGF-β signaling pathway (Fig. 3) [110].

4. MMPs and EndMT in the TME

4.1. General cognition of MMPs

The human MMPs family comprises 24 zinc- and calcium-dependent endopeptidases, including two duplicated genes encoding MMP-23 [111]. MMPs share similar structures, enabling them to not only degrade nearly all protein components in the ECM but also participate in a variety of biological processes under both normal and pathological conditions [112,113]. Generally, MMPs are classified into seven categories based on their substrate specificity and cutting mechanism: (1) interstitial collagenases, including MMP-1, -8, and -13; (2) gelatinases, encompassing MMP-2 and MMP-9; (3) matrix proteases, involving MMP-3, -10, and −11; (4)

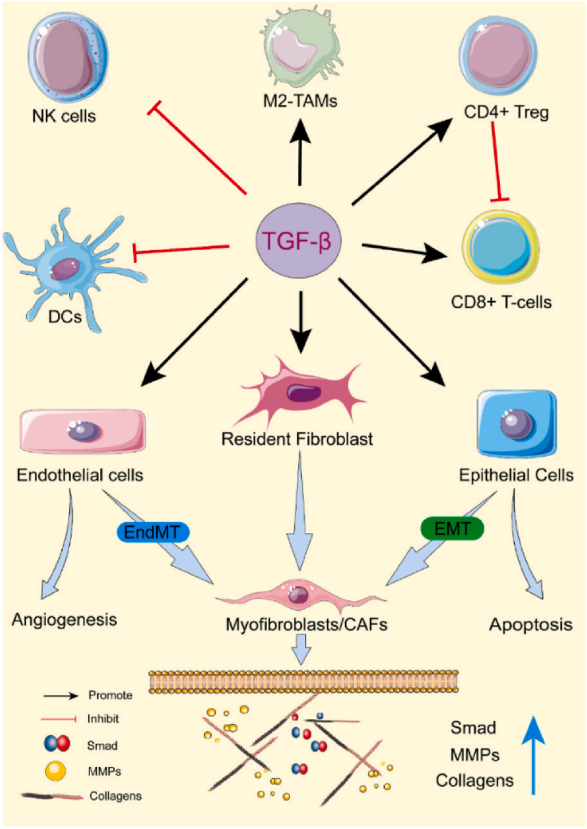

Fig. 3. TGF-β plays a crucial role in various biological processes within the TME. TGF-β inhibits the function of dendritic cells (DC) and natural killer cells while activating M2-type tumor-associated macrophages (TAMs), $CD4^+$ T cells, and $CD8^+$ T cells. Moreover, TGF-β not only directly promotes the transformation of resident fibroblasts into myofibroblasts/CAFs but also induces the transformation of endothelial cells and epithelial cells into myofibroblasts/CAFs through the EndMT and EMT processes. This leads to an increase in Smad proteins, MMPs, and collagen production.

Table 2
MMPs associated with cancers and their general role in biology.

MMPs	Biological Role	Associated with Cancer Type	References
MMP-1	Degradation of ECM; Angiogenesis	BLCA, BRCA, COAD, ESCA, HNSC, LIHC, LUAD, LUSC, STAD, THCA, UCEC	[122]
MMP-2	Tumor invasion and metastasis; Degradation of ECM; Activate TGF-β; Angiogenesis	BLCA, HNSC, KIRP, LUSC, PRAD, UCEC	[123,124]
MMP-3	Degradation of ECM; Activate proMMP-1, -3, -7, -8, -9, -13; Angiogenesis	BRCA, COAD, ESCA, HNSC, LUAD, LUSC, STAD, UCEC	[125,126]
MMP-7	Tumor invasion and metastasis; angiogenesis; degradation of ECM	BRCA, COAD, ESCA, HNSC, KICH, KIRC, KIRP, LIHC, LUAD, STAD, THCA	[127]
MMP-8	degradation of ECM	COAD, ESCA, HNSC, STAD	[128]
MMP-9	Tumor invasion and metastasis; degradation of ECM; activate TGF-β; angiogenesis	BLCA, BRCA, COAD, ESCA, HNSC, KICH, KIRC, KIRP, LIHC, LUAD, LUSC, PRAD, STAD, UCEC	[129,130]
MMP-10	Degradation of ECM	BRCA, COAD, ESCA, HNSC, LIHC, LUAD, LUSC, PRAD, STAD, THCA, UCEC	[131]
MMP-11	Tumor invasion and metastasis; degradation of ECM	BLCA, BRCA, COAD, ESCA, HNSC, KIRC, KIRP, LIHC, LUAD, LUSC, PRAD, STAD, THCA	[132,133]
MMP-12	Degradation of ECM; peptidoglycan metabolism	BRCA, COAD, ESCA, HNSC, LIHC, LUAD, LUSC, STAD, UCEC	[134]
MMP-13	degradation of ECM	BLCA, BRCA, COAD, ESCA, HNSC, KICH, LUAD, LUSC, STAD, THCA	[135]
MMP-14	Tumor invasion and metastasis; degradation of ECM; angiogenesis	BRCA, COAD, ESCA, HNSC, KIRC, KIRP, LUSC	[124,136]
MMP-15	degradation of ECM	KICH, UCEC	[137,138]
MMP-16	Degradation of ECM; activate MMP2	BLCA, HNSC, KIRC, PRAD, THCA, UCEC	[138,139]
MMP-17	Degradation of ECM	HNSC, KIRC, LUAD, LUSC, UCEC	[140]
MMP-19	Degradation of ECM	LUAD, LUSC, UCEC	[141]
MMP-20	Degradation of ECM	KIRP	[142]
MMP-23A/B	Degradation of ECM; activate MMP21	LUSC, BLCA, KICH, UCEC	[143]
MMP-24	Degradation of ECM; peptidoglycan metabolism	KICH, LUSC	[138,144]
MMP-25	Degradation of ECM; peptidoglycan metabolism	COAD, KIRC, LUSC	[140]
MMP-26	Degradation of ECM	KICH, PRAD, UCEC	[138,145]
MMP-27	Degradation of ECM	BLCA, BRCA, COAD, HNSC,	[146,147]
MMP-28	Degradation of ECM; peptidoglycan metabolism	BRCA, COAD, KICH, LUAD, LUSC, STAD, UCEC	[146]

ECM, extracellular matrix; BLCA, bladder cancer; BRCA, breast cancer; COAD, colon adenocarcinoma; ESCA, esophageal cancer; HNSC, head and neck cancer; KICH, kidney chromophobe; KIRC, renal clear cell cancer; KIRP, renal papillary cancer; LIHC, hepatocellular cancer; LUAD, lung adenocarcinoma; LUSC, lung squamous cancer; PRAD, prostate adenocarcinoma; STAD, stomach adenocarcinoma; THCA, thyroid cancer; UCEC, uterine corpus endometrial carcinoma.

matrix lysins, represented by MMP-7 and -26; (5) metalloelastases, featuring MMP-12; (6) membrane-type MMPs (MT-MMPs), which include MMP-14, -15, −16, −17, −24, and −25; and (7) other MMPs (MMP-19, -20, −23, and −28) [114] (Table 2).

Apart from membrane-type MMPs (MT-MMPs), other MMPs are typically secreted into the extracellular microenvironment by cells in an inactive form, a mechanism that safeguards cells' essential components from MMP-induced damage. Almost all MMPs possess four domains: an N-terminal propeptide, a hinge domain, a catalytic domain, and a C-terminal domain [115]. Cleavage of the propeptide, both inside and outside the cell, activates Pro-MMPs, removing the prodomain from the catalytic site and thus rendering MMPs active and capable of binding to their substrate [116,117].

TIMPs are endogenous protease inhibitors that form a binary noncovalent complex with MMPs in a 1:1 ratio. This complex regulates the activation or function of MMPs and prevents MMPs from binding to their substrates [118]. Thus far, four TIMPs (TIMP-1, TIMP-2, TIMP-3, and TIMP-4) have been identified. Although there are some differences in their specificity, each TIMP has been found to inhibit the activity of most MMPs [119–121].

4.2. TGF-β regulates MMPs through EndMT

Interestingly, based on relevant literature, when compared with the results of the EMT process, only a subset of MMPs has been reported to be associated with EndMT. The expression of MMPs (specifically, MMP-1, -2, -7, -9, -10, −11, −14, and −15) in fibroblast-like cells derived from EndMT was significantly higher than that in normal fibroblasts. In the conditioned medium obtained from these fibroblast-like cells, MMP activity was observed to be 8.2 times higher than that of normal fibroblasts, while MMP activity in cell lysates was three times lower than that in normal fibroblasts [24].

The regulatory influence of TGF-β on EndMT-related MMPs is primarily through the activation of specific domains within the MMP promoter. In the MMP promoter, there exist several trans-activating factor domains, including AP-1, NF-κB, PEA3, RARE, Sp-1, and β-catenin/Tcf-4-lef-1. The cis-acting elements in the promoter of MMP bind with these trans-acting factors to regulate MMP gene expression [18,148]. Based on the elemental composition of the cis-element site in the promoter, EndMT-related MMPs can be categorized into three groups: (1) Promoters containing both TATA-binding protein and AP-1 transcriptional activator, including MMP-1, -7, -9, and -10; (2) promoter group containing only TATA but not AP-1, such as MMP-11 and -15; (3) promoters containing

neither TATA nor AP-1, with the expression of MMPs mainly regulated by specific protein 1 (SP-1) transcription factors, including MMP-2 and -14 [18,24,148,149].

In the promoter region of the TGF-β gene, two main binding sites relate to the regulation of MMPs: TGF-β inhibitory element (TIE) and Smad binding element (SBE) [150–152]. Although TIE binding sites exist in the promoters of MMP-1, MMP-7, MMP-9, and MMP-14, TGF-β seems to regulate only the expression of MMP-1 and MMP-7 through TIE, showing no significant correlation with the TIE sequence in MMP-9 and MMP-14 promoters [152–157]. As for another binding site pertaining to TGF-β′s regulation of MMPs, SBE collaborates with other intracellular signal transduction pathways to regulate MMP expression. For instance, MMP-2 expression is induced by both Smads and the TAK1-p38 MAPK-ATF2 pathways [158,159].

TGF-β can also influence MMPs by participating in signal transduction in alternative ways. For example, the P38 signal transduction pathway contributes to the expression of MMP-2 induced by TGF-β in breast epithelial cells and pleural malignant mesothelioma [160], while the NF-κB protein signal pathway serves as the key pathway for TGF-β in inducing MMP-9 expression in cancer cells [161–164]. Additionally, MMP-14 collaborates with TGF-β to activate JNK signal transduction, thus facilitating keratinocyte migration to regulate the expression of MMP-9 [165].

4.3. MMPs and EndMT

Although the EndMT process is linked to increased expression of many MMPs, current related studies have specifically demonstrated a direct relationship between the expression of gelatinases (MMP-2 and -9) and membrane-type 1MMP (MT1-MMP) with EndMT. The specific connections between these MMPs and EndMT are summarized in Table 3 and further discussed below.

Increased expression of MMP-2 and MMP-9 has been observed in the EndMT process during tumor metastasis, inflammation-related diseases, human cytomegalovirus (HCMV) infection, and heart valve–related diseases under the influence of TGF-β [27,166–169]. Particularly in tumor migration, inducing EndMT in human microvascular EC-1 (HMEC-1) and human umbilical vein EC (HUVECs) using 27-hydroxycholesterol (27HC) significantly elevates the expression and activity of MMP-2 and MMP-9. Conversely, knocking down the STAT3 gene in HUVECs significantly inhibits EndMT [27]. Moreover, the migration of HMEC-1 through EndMT has also been correlated with the proteolytic activity of MMP-2 [170]. The ILK-MMP9-MRTF axis, consisting of integrin-linked kinase, MMP-9, and myocardin-related transcription factor, plays a crucial role in differentiating ECs into CAF-like cells during EndMT [171]. These findings indicate that not only does the EndMT process influence the expression of MMP-2 and MMP-9, but these MMPs also play a regulatory role in the occurrence of EndMT.

Regarding MT-MMPs, only MT1-MMP has been confirmed to be associated with EndMT. Depending on tissue demands or injuries, ECs may differentiate into MCs, or behave as tip cells and stem cells. The former process is referred to as EndMT, while the latter is known as EC activation. Despite their distinct differentiation pathways, they are both linked to the functions of SNAI1, SNAI2, Slug, and MMPs. The transcription factors SNAI and Slug can directly regulate the expression of MT1-MMP. Furthermore, the activities of MMP-2 and MMP-9 may also be influenced by the expression of Slug through MT1-MMP [65,172,173]. Additionally, MT1-MMP can activate latent TGF-β1 in macrophages, inducing paracrine SMAD2-mediated signal transduction and endothelial-stromal transformation in ECs. MT1-MMP-deficient macrophages have shown the ability to inhibit EndMT in co-culture with ECs [25]. This underscores the interaction between MT1-MMP and EndMT.

5. Relationship among EndMT, TGF-β, and MMPs

Currently, there is limited research on the inhibition of EndMT by targeting both TGF-β and MMPs. Most studies focus on suppressing the EndMT process and MMP expression by inhibiting TGF-β. Various clinical drugs and cytokines, such as the combination of temozolomide and perillyl alcohol (NEO212), the interaction between mitral endothelial cells and interstitial cells, Tongxinluo, curcumin, losartan, hydroxytyrosol (HT), and HT-3Os (the main plasma metabolite of HT), have been found to inhibit or block the TGF-β process. Consequently, EndMT is suppressed, leading to a decrease in related MMP expression and activity [27,166,167,174–177]. Notably, in the regulation of the microRNA (miR)-21 gene in ECs, an endogenous protein known as Kallistatin has been

Table 3
Effect of the EndMT process on MMPs in different endothelial cells.

Inducers	Cell line	Role	References
27-HC	HMEC-1 and HUVECs	Expression of MMP-2 and MMP-9 increased	[37]
IL-1β	ECs	Expression of MMP-2 and MMP-9 increased (HT-3Os protect ECs from IL-1β, downregulate MMP-2 and MMP-9)	[120]
KSHV infection	Lymphatic ECs	Expression of MT1-MMP increased	[125]
TGF-β2	HMEC-1	Expression of MMP-2 increased	[123]
HCMV and TGF-β1	HUVEC	MMP-2 expression increased and TGF-β1 was activated	[122]
TGF-β	VEC	Expression of MMP-2 increased	[119]
TGF-β1, TGFβ-3, and TNF-α	HUVEC	Expression of MMP-9 increased	[121]
TGF-β1	ECs	Expression capacity of EndMT correlates with MT1-MMP expression in Mφs	[126]

27HC, 27-hydroxy cholesterol; KSHV, Kaposi's sarcoma herpes virus; HCMV, human cytomegalovirus; TNF-α, tumor necrosis factor-α; HMEC-1, human microvascular endothelial cell-1; HUVEC, human umbilical vein endothelial cell; VEC, valvular endothelial cell; HT-3Os, HT-3 o sulfate; Mφs, macrophages.

shown to hinder TGF-β synthesis. This process inhibits the formation of reactive oxygen species mediated by TGF-β and reduces the expression and activity of NADPH oxidase. Furthermore, Kallistatin counters TGF-β-induced miR-21 and Snail1 synthesis, Akt phosphorylation, NF-κB activation, and MMP-2 synthesis and activation. These findings offer a novel approach to prevent fibrosis and cancer [176].

Some research has also demonstrated that inhibiting MMPs can affect the EndMT process. For instance, MT1-MMP-deficient macrophages have demonstrated the ability to impede EndMT in co-cultures with ECs [178]. In the microenvironment of Kaposi's sarcoma, Kaposi sarcoma herpesvirus-induced EndMT is initiated by viral proteins (vFLIP and vGPCR) through Notch pathway activation, MT1-MMP operates downstream of Notch, subsequently influencing the EndMT process [179]. Furthermore, studies have indicated that endothelial cell HUVECs infected with HCMV can activate latent TGF-β1 by stimulating MMP-2 after treatment with TGF-β1 [169]. This suggests a reciprocal regulatory relationship between certain MMPs, TGF-β, and EndMT.

6. Conclusion and perspectives

Increasing research results underscore the pivotal roles of TGF-β, MMPs, and EndMT in tumors and related diseases. In this review, we delineate the individual mechanisms and physiological processes of TGF-β, MMPs, and EndMT in the TME, while also elucidating their regulatory mechanisms and current research status in TME. The literature suggests that TGF-β, MMPs, and EndMT interact with each other to varying extents.

EndMT has always been indispensable in the context of tumors and related diseases. Presently, inhibiting the EndMT process primarily relies on regulating the TGF-β signaling pathway, and the production of MMPs has been established to be associated with TGF-β [69,180]. However, studies have proved that the expression of some MMPs will increase with the occurrence of EndMT, but only a few studies assessed the effect of MMPs on the EndMT process [25]. Although the research status indicates a certain relationship between EndMT and MMPs, the specific mechanism between MMPs and EndMT has yet to be fully elucidated.

Given the interrelated regulatory mechanisms of TGF-β, MMPs, and EndMT in the TME, they all serve as potential combined targets for chemotherapy [167,179]. Thus, concurrently targeting TGF-β and MMPs presents a highly promising research avenue for interfering with the role of EndMT in TME. In conclusion, modulating the expression of TGF-β and MMPs can impact tumor progression. However, this perspective necessitates validation through experimental studies. Future investigations will further delve into the combination of TGF-β and MMPs in tumor treatment, providing a more comprehensive understanding of its clinical benefits in cancer therapy.

CRediT authorship contribution statement

Fei Du: Writing – review & editing, Writing – original draft, Project administration. **Jing Li:** Writing – review & editing, Writing – original draft, Validation, Investigation. **Xiaolin Zhong:** Writing – review & editing. **Zhuo Zhang:** Writing – review & editing, Supervision, Investigation, Conceptualization. **Yueshui Zhao:** Writing – review & editing, Supervision, Investigation, Funding acquisition, Conceptualization.

Data availability statement

No data was used for the research described in the article.

Additional information

No additional information is available for this paper.

Funding

This work was supported by Sichuan Science and Technology Program (Nos. 2023NSFSC0728, 2022NSFSC0816, and 2022YFS0633-B4).

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Abbreviations

MMPs	matrix metalloproteinases
MT1-MMPs	membrane-type 1-matrix metalloproteinase
EndMT	endothelial-to-mesenchymal transition
TME	tumor microenvironment
TIMPs	tissue inhibitors of matrix metalloproteinases

MAPK	mitogen-activated protein kinases
JUK	c-JUN-N terminal kinase
NICD	Notch intracellular domain
27-HC	27-hydroxy cholesterol
TNF-α	tumor necrosis factor-α
SIRT3	Sirtuin3
TGF-β	transforming growth factor-β
BMP	bone morphogenetic protein
NOX	NADPH oxidase
CAFs	cancer-associated fibroblast
ECM	extracellular matrix
BLCA	bladder cancer
BRCA	breast cancer
COAD	colon adenocarcinoma
ESCA	esophageal cancer
HNSC	head and neck cancer
KICH	kidney chromophobe
KIRC	renal clear cell cancer
KIRP	renal papillary cancer
LIHC	hepatocellular cancer
LUAD	lung adenocarcinoma
LUSC	lung squamous cancer
PRAD	prostate adenocarcinoma
STAD	stomach adenocarcinoma
THCA	thyroid cancer
UCEC	uterine corpus endometrial carcinoma
KSHV	Kaposi's sarcoma herpes virus
HCMV	human cytomegalovirus
HMEC-1	human microvascular endothelial cell-1
HUVEC	human umbilical vein endothelial cell
VEC	valvular endothelial cell
HT-3Os	HT-3 o sulfate
Mφs	macrophages

References

[1] P. ten Dijke, H.M. Arthur, Extracellular control of TGFbeta signalling in vascular development and disease, Nat. Rev. Mol. Cell Biol. 8 (11) (2007) 857–869.
[2] X. Da, et al., AGGF1 therapy inhibits thoracic aortic aneurysms by enhancing integrin α7-mediated inhibition of TGF-β1 maturation and ERK1/2 signaling, Nat. Commun. 14 (1) (2023) 2265.
[3] Y. Xiang, et al., MALAT1 modulates TGF-β1-induced endothelial-to-mesenchymal transition through downregulation of miR-145, Cell. Physiol. Biochem. : International Journal of Experimental Cellular Physiology, Biochemistry, and Pharmacology 42 (1) (2017) 357–372.
[4] H. Sabbineni, A. Verma, P.R. Somanath, Isoform-specific effects of transforming growth factor β on endothelial-to-mesenchymal transition, J. Cell. Physiol. 233 (11) (2018) 8418–8428.
[5] E. Pardali, M.-J. Goumans, P. ten Dijke, Signaling by members of the TGF-beta family in vascular morphogenesis and disease, Trends Cell Biol. 20 (9) (2010) 556–567.
[6] N.R. Gough, X. Xiang, L. Mishra, TGF-B signaling in liver, pancreas, and gastrointestinal diseases and cancer, Gastroenterology 161 (2) (2021).
[7] D. Pohlers, et al., TGF-beta and fibrosis in different organs - molecular pathway imprints, Biochim. Biophys. Acta 1792 (8) (2009) 746–756.
[8] A. Biernacka, M. Dobaczewski, N.G. Frangogiannis, TGF-β signaling in fibrosis, Growth Factors 29 (5) (2011) 196–202.
[9] G. Kremastiotis, et al., Disparate effects of MMP and TIMP modulation on coronary atherosclerosis and associated myocardial fibrosis, Sci. Rep. 11 (1) (2021) 23081.
[10] Y. Wang, et al., The methyl-CpG-binding domain 2 facilitates pulmonary fibrosis by orchestrating fibroblast to myofibroblast differentiation, Eur. Respir. J. 60 (3) (2022).
[11] L. Caja, et al., TGF-B and the tissue microenvironment: relevance in fibrosis and cancer, Int. J. Mol. Sci. 19 (5) (2018).
[12] M.E. Mercado-Pimentel, R.B. Runyan, Multiple transforming growth factor-beta isoforms and receptors function during epithelial-mesenchymal cell transformation in the embryonic heart, Cells Tissues Organs 185 (1–3) (2007) 146–156.
[13] E.J. Armstrong, J. Bischoff, Heart valve development: endothelial cell signaling and differentiation, Circ. Res. 95 (5) (2004) 459–470.
[14] K. Sobierajska, et al., Endothelial cells in the tumor microenvironment, Advances in experimental medicine and biology 1234 (2020) 71–86.
[15] L. Pérez, et al., Endothelial-to-mesenchymal transition: cytokine-mediated pathways that determine endothelial fibrosis under inflammatory conditions, Cytokine Growth Factor Rev. 33 (2017) 41–54.
[16] E.M. Zeisberg, et al., Discovery of endothelial to mesenchymal transition as a source for carcinoma-associated fibroblasts, Cancer Res. 67 (21) (2007) 10123–10128.
[17] K. Kessenbrock, V. Plaks, Z. Werb, Matrix metalloproteinases: regulators of the tumor microenvironment, Cell 141 (1) (2010) 52–67.
[18] C. Yan, D.D. Boyd, Regulation of matrix metalloproteinase gene expression, J. Cell. Physiol. 211 (1) (2007) 19–26.
[19] L. Zong, et al., A review of matrix metalloproteinase-2-sensitive nanoparticles as a novel drug delivery for tumor therapy, Int. J. Biol. Macromol. 262 (Pt 2) (2024) 130043.

[20] S. Rastogi, et al., Lactate acidosis and simultaneous recruitment of TGF-β leads to alter plasticity of hypoxic cancer cells in tumor microenvironment, Pharmacology & Therapeutics 250 (2023) 108519.
[21] J.P. Annes, J.S. Munger, D.B. Rifkin, Making sense of latent TGFbeta activation, J. Cell Sci. 116 (Pt 2) (2003) 217–224.
[22] L.R. Gomes, et al., TGF-β1 modulates the homeostasis between MMPs and MMP inhibitors through p38 MAPK and ERK1/2 in highly invasive breast cancer cells, BMC Cancer 12 (2012) 26.
[23] Y. Ma, et al., Exercise training alleviates cardiac fibrosis through increasing fibroblast growth factor 21 and regulating TGF-β1-smad2/3-MMP2/9 signaling in mice with myocardial infarction, Int. J. Mol. Sci. 22 (22) (2021).
[24] S.M. Evrard, et al., Endothelial to mesenchymal transition is common in atherosclerotic lesions and is associated with plaque instability, Nat. Commun. 7 (2016) 11853.
[25] L. Alonso-Herranz, et al., Macrophages promote endothelial-to-mesenchymal transition via MT1-MMP/TGFβ1 after myocardial infarction, Elife 9 (2020).
[26] J. Zhen, et al., The 14-3-3η/GSK-3β/β-catenin complex regulates EndMT induced by 27-hydroxycholesterol in HUVECs and promotes the migration of breast cancer cells, Cell Biol. Toxicol. 37 (4) (2021) 515–529.
[27] K. Jiao, et al., 27-Hydroxycholesterol-induced EndMT acts STAT3 signaling to promote breast cancer cell migration by altering the tumor microenvironment, Cancer biology & medicine 17 (1) (2020).
[28] J. Chen, et al., A reciprocal feedback of miR-548ac/YB-1/Snail induces EndMT of HUVECs during acidity microenvironment, Cancer Cell Int. 21 (1) (2021) 692.
[29] M. Adjuto-Saccone, et al., TNF-α induces endothelial-mesenchymal transition promoting stromal development of pancreatic adenocarcinoma, Cell Death Dis. 12 (7) (2021) 649.
[30] J. Zhang, et al., miR-494 induces EndMT and promotes the development of HCC (Hepatocellular Carcinoma) by targeting SIRT3/TGF-β/SMAD signaling pathway, Sci. Rep. 9 (1) (2019) 7213.
[31] K. Zhu, et al., MiR-302c inhibits tumor growth of hepatocellular carcinoma by suppressing the endothelial-mesenchymal transition of endothelial cells, Sci. Rep. 4 (2014) 5524.
[32] M.E. Wawro, et al., Invasive colon cancer cells induce transdifferentiation of endothelium to cancer-associated fibroblasts through microtubules enriched in tubulin-β3, Int. J. Mol. Sci. 20 (1) (2018).
[33] C.-S. Fan, et al., Osteopontin-integrin engagement induces HIF-1α-TCF12-mediated endothelial-mesenchymal transition to exacerbate colorectal cancer, Oncotarget 9 (4) (2018) 4998–5015.
[34] M.E. Wawro, et al., Nonsteroidal anti-inflammatory drugs prevent vincristine-dependent cancer-associated fibroblasts formation, Int. J. Mol. Sci. 20 (8) (2019).
[35] L. Maddaluno, et al., EndMT contributes to the onset and progression of cerebral cavernous malformations, Nature 498 (7455) (2013) 492–496.
[36] Z. Zhou, et al., Cerebral cavernous malformations arise from endothelial gain of MEKK3-KLF2/4 signalling, Nature 532 (7597) (2016) 122–126.
[37] L.D. Shoemaker, et al., Evidence for endothelial-to-mesenchymal transition in human brain arteriovenous malformations, Clin. Transl. Med. 10 (2) (2020) e99.
[38] V. Platel, et al., NOX1 and NOX2: two enzymes that promote endothelial-to-mesenchymal transition induced by melanoma conditioned media, Pharmacol. Res. (2022) 106097.
[39] X. Li, et al., Disseminated melanoma cells transdifferentiate into endothelial cells in intravascular niches at metastatic sites, Cell Rep. 31 (11) (2020) 107765.
[40] I.A. Krizbai, et al., Endothelial-mesenchymal transition of brain endothelial cells: possible role during metastatic extravasation, PLoS One 10 (3) (2015) e0119655.
[41] J.H. Yeon, et al., Cancer-derived exosomes trigger endothelial to mesenchymal transition followed by the induction of cancer-associated fibroblasts, Acta Biomater. 76 (2018) 146–153.
[42] S. Krishnan, et al., Modulation of cerebral endothelial cell function by TGF-β in glioblastoma: VEGF-dependent angiogenesis versus endothelial mesenchymal transition, Oncotarget 6 (26) (2015) 22480–22495.
[43] L. Nie, et al., Endothelial-mesenchymal transition in normal human esophageal endothelial cells cocultured with esophageal adenocarcinoma cells: role of IL-1β and TGF-β2, Am. J. Physiol. Cell Physiol. 307 (9) (2014) C859–C877.
[44] S.-H. Kim, Y. Song, H.R. Seo, GSK-3β regulates the endothelial-to-mesenchymal transition via reciprocal crosstalk between NSCLC cells and HUVECs in multicellular tumor spheroid models, J. Exp. Clin. Cancer Res. : CR 38 (1) (2019) 46.
[45] K.J. Choi, et al., Endothelial-to-mesenchymal transition in anticancer therapy and normal tissue damage, Experimental & molecular medicine 52 (5) (2020) 781–792.
[46] J.-K. Nam, et al., Radiation-induced fibrotic tumor microenvironment regulates anti-tumor immune response, Cancers 13 (20) (2021).
[47] S.-H. Choi, et al., Tumour-vasculature development via endothelial-to-mesenchymal transition after radiotherapy controls CD44v6 cancer cell and macrophage polarization, Nat. Commun. 9 (1) (2018) 5108.
[48] D.F. Quail, J.A. Joyce, Microenvironmental regulation of tumor progression and metastasis, Nat. Med. 19 (11) (2013) 1423–1437.
[49] F.R. Balkwill, M. Capasso, T. Hagemann, The tumor microenvironment at a glance, J. Cell Sci. 125 (Pt 23) (2012) 5591–5596.
[50] M. Zeisberg, R. Kalluri, Cellular mechanisms of tissue fibrosis. 1. Common and organ-specific mechanisms associated with tissue fibrosis, Am. J. Physiol. Cell Physiol. 304 (3) (2013) C216–C225.
[51] G. Krenning, E.M. Zeisberg, R. Kalluri, The origin of fibroblasts and mechanism of cardiac fibrosis, J. Cell. Physiol. 225 (3) (2010) 631–637.
[52] D.R. Principe, et al., TGF-β: duality of function between tumor prevention and carcinogenesis, J. Natl. Cancer Inst. 106 (2) (2014) djt369.
[53] V.C. Garside, et al., Co-ordinating Notch, BMP, and TGF-β signaling during heart valve development, Cell. Mol. Life Sci. : CMLS 70 (16) (2013) 2899–2917.
[54] D. Rosebrock, et al., Enhanced cortical neural stem cell identity through short SMAD and WNT inhibition in human cerebral organoids facilitates emergence of outer radial glial cells, Nat. Cell Biol. 24 (6) (2022) 981–995.
[55] S. Hu, et al., NOTCH-YAP1/TEAD-DNMT1 Axis drives hepatocyte reprogramming into intrahepatic cholangiocarcinoma, Gastroenterology 163 (2) (2022) 449–465.
[56] P. Gasperini, et al., Kaposi sarcoma herpesvirus promotes endothelial-to-mesenchymal transition through Notch-dependent signaling, Cancer Res. 72 (5) (2012) 1157–1169.
[57] M. Corada, et al., The Wnt/beta-catenin pathway modulates vascular remodeling and specification by upregulating Dll4/Notch signaling, Dev. Cell 18 (6) (2010) 938–949.
[58] A.C.Y. Chang, et al., A Notch-dependent transcriptional hierarchy promotes mesenchymal transdifferentiation in the cardiac cushion, Dev. Dynam. : an official publication of the American Association of Anatomists 243 (7) (2014) 894–905.
[59] F. Lin, N. Wang, T.-C. Zhang, The role of endothelial-mesenchymal transition in development and pathological process, IUBMB Life 64 (9) (2012) 717–723.
[60] A. Toullec, et al., HIF-1α deletion in the endothelium, but not in the epithelium, protects from radiation-induced enteritis, Cellular and molecular gastroenterology and hepatology 5 (1) (2018) 15–30.
[61] H. Tang, et al., Endothelial HIF-2α contributes to severe pulmonary hypertension due to endothelial-to-mesenchymal transition. American journal of physiology, Lung cellular and molecular physiology 314 (2) (2018) L256–L275.
[62] T. Suzuki, et al., Isolation and characterization of endothelial-to-mesenchymal transition cells in pulmonary arterial hypertension. American journal of physiology, Lung cellular and molecular physiology 314 (1) (2018) L118–L126.
[63] S.-H. Choi, et al., A hypoxia-induced vascular endothelial-to-mesenchymal transition in development of radiation-induced pulmonary fibrosis, Clin. Cancer Res. : an official journal of the American Association for Cancer Research 21 (16) (2015) 3716–3726.
[64] A.U. Kabir, et al., Dual role of endothelial Myct1 in tumor angiogenesis and tumor immunity, Sci. Transl. Med. 13 (583) (2021).
[65] K.M. Welch-Reardon, N. Wu, C.C.W. Hughes, A role for partial endothelial-mesenchymal transitions in angiogenesis? Arterioscler. Thromb. Vasc. Biol. 35 (2) (2015) 303–308.
[66] A. Gu, et al., Slug is associated with tumor metastasis and angiogenesis in ovarian cancer, Reprod. Sci. 24 (2) (2017) 291–299.
[67] J. Folkman, Tumor angiogenesis: therapeutic implications, N. Engl. J. Med. 285 (21) (1971) 1182–1186.

[68] H. Murai, et al., Multiomics identifies the link between intratumor steatosis and the exhausted tumor immune microenvironment in hepatocellular carcinoma, Hepatology (Baltimore, Md 77 (1) (2023) 77–91.
[69] X. Mao, et al., Crosstalk between cancer-associated fibroblasts and immune cells in the tumor microenvironment: new findings and future perspectives, Mol. Cancer 20 (1) (2021) 131.
[70] L. Bu, et al., Biological heterogeneity and versatility of cancer-associated fibroblasts in the tumor microenvironment, Oncogene 38 (25) (2019) 4887–4901.
[71] A.B. Roberts, et al., Transforming growth factors: isolation of polypeptides from virally and chemically transformed cells by acid/ethanol extraction, Proc. Natl. Acad. Sci. U.S.A. 77 (6) (1980) 3494–3498.
[72] J.E. de Larco, G.J. Todaro, Growth factors from murine sarcoma virus-transformed cells, Proc. Natl. Acad. Sci. U.S.A. 75 (8) (1978) 4001–4005.
[73] J.F. Santibañez, M. Quintanilla, C. Bernabeu, TGF-β/TGF-β receptor system and its role in physiological and pathological conditions, Clin. Sci. (Lond.) 121 (6) (2011) 233–251.
[74] E. Meulmeester, P. Ten Dijke, The dynamic roles of TGF-β in cancer, J. Pathol. 223 (2) (2011) 205–218.
[75] L. Attisano, J.L. Wrana, Signal transduction by the TGF-beta superfamily, Science (New York, N.Y.) 296 (5573) (2002) 1646–1647.
[76] Y. Shi, J. Massagué, Mechanisms of TGF-beta signaling from cell membrane to the nucleus, Cell 113 (6) (2003) 685–700.
[77] A. Moustakas, C.-H. Heldin, Mechanisms of TGFβ-induced epithelial-mesenchymal transition, J. Clin. Med. 5 (7) (2016).
[78] X. Guo, X.-F. Wang, Signaling cross-talk between TGF-beta/BMP and other pathways, Cell Res. 19 (1) (2009) 71–88.
[79] R. Derynck, Y.E. Zhang, Smad-dependent and Smad-independent pathways in TGF-beta family signalling, Nature 425 (6958) (2003) 577–584.
[80] K. Yoshida, et al., Transforming growth factor-beta and platelet-derived growth factor signal via c-Jun N-terminal kinase-dependent Smad2/3 phosphorylation in rat hepatic stellate cells after acute liver injury, Am. J. Pathol. 166 (4) (2005) 1029–1039.
[81] M. Kretzschmar, et al., A mechanism of repression of TGFbeta/Smad signaling by oncogenic Ras, Gene Dev. 13 (7) (1999) 804–816.
[82] F. Furukawa, et al., p38 MAPK mediates fibrogenic signal through Smad3 phosphorylation in rat myofibroblasts, Hepatology 38 (4) (2003) 879–889.
[83] M. Funaba, C.M. Zimmerman, L.S. Mathews, Modulation of Smad2-mediated signaling by extracellular signal-regulated kinase, J. Biol. Chem. 277 (44) (2002) 41361–41368.
[84] A. Weiss, L. Attisano, *The TGFbeta superfamily signaling pathway*. Wiley interdisciplinary reviews, Dev. Biol. 2 (1) (2013) 47–63.
[85] C. Guido, et al., Metabolic reprogramming of cancer-associated fibroblasts by TGF-β drives tumor growth: connecting TGF-β signaling with "Warburg-like" cancer metabolism and L-lactate production, Cell Cycle 11 (16) (2012) 3019–3035.
[86] J.P. Thiery, et al., Epithelial-mesenchymal transitions in development and disease, Cell 139 (5) (2009) 871–890.
[87] Y. Tsubakihara, A. Moustakas, Epithelial-mesenchymal transition and metastasis under the control of transforming growth factor β, Int. J. Mol. Sci. 19 (11) (2018).
[88] Y. Katsuno, R. Derynck, Epithelial plasticity, epithelial-mesenchymal transition, and the TGF-β family, Dev. Cell 56 (6) (2021) 726–746.
[89] S. Potenta, E. Zeisberg, R. Kalluri, The role of endothelial-to-mesenchymal transition in cancer progression, Br. J. Cancer 99 (9) (2008) 1375–1379.
[90] X.-M. Meng, D.J. Nikolic-Paterson, H.Y. Lan, TGF-β: the master regulator of fibrosis, Nat. Rev. Nephrol. 12 (6) (2016) 325–338.
[91] C. Margadant, A. Sonnenberg, Integrin-TGF-beta crosstalk in fibrosis, cancer and wound healing, EMBO Rep. 11 (2) (2010).
[92] M. Lodyga, B. Hinz, TGF-β1 - a truly transforming growth factor in fibrosis and immunity, Semin. Cell Dev. Biol. 101 (2020) 123–139.
[93] D. Medici, S. Potenta, R. Kalluri, Transforming growth factor-β2 promotes Snail-mediated endothelial-mesenchymal transition through convergence of Smad-dependent and Smad-independent signalling, Biochem. J. 437 (3) (2011) 515–520.
[94] S. Song, et al., Foxm1 is a critical driver of TGF-β-induced EndMT in endothelial cells through Smad2/3 and binds to the Snail promoter, J. Cell. Physiol. 234 (6) (2019) 9052–9064.
[95] E. Pardali, et al., TGF-β-Induced endothelial-mesenchymal transition in fibrotic diseases, Int. J. Mol. Sci. 18 (10) (2017).
[96] X. Ma, et al., Endothelial-to-Mesenchymal transition in calcific aortic valve disease, Acta Cardiol. Sin. 36 (3) (2020) 183–194.
[97] J. Ma, et al., TGF-β-mediated endothelial to mesenchymal transition (EndMT) and the functional assessment of EndMT effectors using CRISPR/Cas9 gene editing, J. Vis. Exp. 168 (2021).
[98] K.B. Kennel, et al., Cancer-associated fibroblasts in inflammation and antitumor immunity, Clin. Cancer Res. : an Official Journal of the American Association For Cancer Research 29 (6) (2023) 1009–1016.
[99] M. Marozzi, et al., Inflammation, extracellular matrix remodeling, and proteostasis in tumor microenvironment, Int. J. Mol. Sci. 22 (15) (2021).
[100] Y. Yoshimatsu, et al., TNF-α enhances TGF-β-induced endothelial-to-mesenchymal transition via TGF-β signal augmentation, Cancer Sci. 111 (7) (2020) 2385–2399.
[101] D.M. Gonzalez, D. Medici, Signaling mechanisms of the epithelial-mesenchymal transition, Sci. Signal. 7 (344) (2014) re8.
[102] C.L. Karthika, et al., Oscillatory shear stress modulates Notch-mediated endothelial mesenchymal plasticity in cerebral arteriovenous malformations, Cell. Mol. Biol. Lett. 28 (1) (2023) 22.
[103] D. Padua, J. Massagué, Roles of TGFbeta in metastasis, Cell Res. 19 (1) (2009).
[104] B. Bierie, H.L. Moses, TGF-beta and cancer, Cytokine Growth Factor Rev. 17 (1–2) (2006) 29–40.
[105] J.F. Piskurich, et al., Identification of distinct regions of 5' flanking DNA that mediate constitutive, IFN-gamma, STAT1, and TGF-beta-regulated expression of the class II transactivator gene, Journal of immunology (Baltimore, Md. : 1950) 160 (1) (1998) 233–240.
[106] D. Nandan, N.E. Reiner, TGF-beta attenuates the class II transactivator and reveals an accessory pathway of IFN-gamma action, Journal of immunology (Baltimore, Md. : 1950) 158 (3) (1997) 1095–1101.
[107] J. Yu, et al., Pro- and antiinflammatory cytokine signaling: reciprocal antagonism regulates interferon-gamma production by human natural killer cells, Immunity 24 (5) (2006) 575–590.
[108] Y. Laouar, et al., Transforming growth factor-beta controls T helper type 1 cell development through regulation of natural killer cell interferon-gamma, Nat. Immunol. 6 (6) (2005) 600–607.
[109] A. Kelly, et al., Human monocytes and macrophages regulate immune tolerance via integrin αvβ8-mediated TGFβ activation, J. Exp. Med. 215 (11) (2018) 2725–2736.
[110] K. Sun, et al., Endotrophin triggers adipose tissue fibrosis and metabolic dysfunction, Nat. Commun. 5 (2014) 3485.
[111] X.S. Puente, et al., Human and mouse proteases: a comparative genomic approach, Nat. Rev. Genet. 4 (7) (2003) 544–558.
[112] E.S. Radisky, D.C. Radisky, Matrix metalloproteinases as breast cancer drivers and therapeutic targets, Front. Biosci. 20 (2015) 1144–1163.
[113] R. Mittal, et al., Intricate functions of matrix metalloproteinases in physiological and pathological conditions, J. Cell. Physiol. 231 (12) (2016) 2599–2621.
[114] V.J. Craig, et al., Matrix metalloproteinases as therapeutic targets for idiopathic pulmonary fibrosis, Am. J. Respir. Cell Mol. Biol. 53 (5) (2015) 585–600.
[115] A. Liacini, et al., Inhibition of interleukin-1-stimulated MAP kinases, activating protein-1 (AP-1) and nuclear factor kappa B (NF-kappa B) transcription factors down-regulates matrix metalloproteinase gene expression in articular chondrocytes, Matrix Biol. : journal of the International Society for Matrix Biology 21 (3) (2002) 251–262.
[116] G. Murphy, V. Knäuper, *Relating matrix metalloproteinase structure to function: why the "hemopexin" domain?* Matrix biology, journal of the International Society for Matrix Biology 15 (8–9) (1997) 511–518.
[117] S. Löffek, O. Schilling, C.W. Franzke, Series "matrix metalloproteinases in lung health and disease": biological role of matrix metalloproteinases: a critical balance, Eur. Respir. J. 38 (1) (2011) 191–208.
[118] G. Murphy, Matrix metalloproteinases and their inhibitors, Acta Orthop. Scand. Suppl. 266 (1995) 55–60.
[119] G. Murphy, Tissue inhibitors of metalloproteinases, Genome Biol. 12 (11) (2011) 233.
[120] M. Giannandrea, W.C. Parks, Diverse functions of matrix metalloproteinases during fibrosis, Disease models & mechanisms 7 (2) (2014) 193–203.
[121] V. Arpino, M. Brock, S.E. Gill, The role of TIMPs in regulation of extracellular matrix proteolysis, Matrix Biol. : journal of the International Society for Matrix Biology 44–46 (2015) 247–254.
[122] J. Yu, et al., Tumor-associated macrophages (TAMs) depend on MMP1 for their cancer-promoting role, Cell Death Discovery 7 (1) (2021) 343.

[123] J. Sun, et al., ELTD1 promotes invasion and metastasis by activating MMP2 in colorectal cancer, Int. J. Biol. Sci. 17 (12) (2021) 3048–3058.
[124] E. Gobin, et al., A pan-cancer perspective of matrix metalloproteases (MMP) gene expression profile and their diagnostic/prognostic potential, BMC Cancer 19 (1) (2019) 581.
[125] Z. Pan, et al., Identification of gene signatures associated with ulcerative colitis and the association with immune infiltrates in colon cancer, Front. Immunol. 14 (2023) 1086898.
[126] C. Chu, et al., MiR-519d suppresses breast cancer tumorigenesis and metastasis via targeting MMP3, Int. J. Biol. Sci. 14 (2) (2018) 228–236.
[127] Q. Zhang, et al., Interleukin-17 promotes prostate cancer via MMP7-induced epithelial-to-mesenchymal transition, Oncogene 36 (5) (2017) 687–699.
[128] K. Juurikka, et al., The role of MMP8 in cancer: a systematic review, Int. J. Mol. Sci. 20 (18) (2019).
[129] K. Jiang, et al., Exosomal ANGPTL1 attenuates colorectal cancer liver metastasis by regulating Kupffer cell secretion pattern and impeding MMP9 induced vascular leakiness, J. Exp. Clin. Cancer Res. : CR 40 (1) (2021) 21.
[130] S. Mondal, et al., Matrix metalloproteinase-9 (MMP-9) and its inhibitors in cancer: a minireview, Eur. J. Med. Chem. 194 (2020) 112260.
[131] K. Tang, et al., Elevated MMP10/13 mediated barrier disruption and NF-κB activation aggravate colitis and colon tumorigenesis in both individual or full miR-148/152 family knockout mice, Cancer Lett. 529 (2022) 53–69.
[132] G. Theocharidis, et al., Single cell transcriptomic landscape of diabetic foot ulcers, Nat. Commun. 13 (1) (2022) 181.
[133] G. Xu, et al., Exosomal miRNA-139 in cancer-associated fibroblasts inhibits gastric cancer progression by repressing MMP11 expression, Int. J. Biol. Sci. 15 (11) (2019) 2320–2329.
[134] T.-L. Huang, et al., DRP1 contributes to head and neck cancer progression and induces glycolysis through modulated FOXM1/MMP12 axis, Mol. Oncol. 16 (13) (2022) 2585–2606.
[135] D.J.H. Shih, et al., Genomic characterization of human brain metastases identifies drivers of metastatic lung adenocarcinoma, Nat. Genet. 52 (4) (2020) 371–377.
[136] G. Cui, et al., MMP14 predicts a poor prognosis in patients with colorectal cancer, Hum. Pathol. 83 (2019) 36–42.
[137] M.-H. Wu, et al., Silencing PROK2 inhibits invasion of human cervical cancer cells by targeting MMP15 expression, Int. J. Mol. Sci. 21 (17) (2020).
[138] J. Pietrzak, et al., Changes in the expression of membrane type-matrix metalloproteinases genes (MMP14, MMP15, MMP16, MMP24) during treatment and their potential impact on the survival of patients with non-small cell lung cancer (NSCLC), Biomedicine & Pharmacotherapy = Biomedecine & Pharmacotherapie 146 (2022) 112559.
[139] W. Zhang, C.J. Chen, G.L. Guo, MiR-155 promotes the proliferation and migration of breast cancer cells via targeting SOCS1 and MMP16, Eur. Rev. Med. Pharmacol. Sci. 22 (21) (2018) 7323–7332.
[140] A. Sohail, et al., MT4-(MMP17) and MT6-MMP (MMP25), A unique set of membrane-anchored matrix metalloproteinases: properties and expression in cancer, Cancer Metastasis Rev. 27 (2) (2008) 289–302.
[141] M. Müller, et al., MMP19 is upregulated during melanoma progression and increases invasion of melanoma cells, Mod. Pathol. : an Official Journal of the United States and Canadian Academy of Pathology, Inc 23 (4) (2010) 511–521.
[142] X. Chang, et al., Common variants in MMP20 at 11q22.2 predispose to 11q deletion and neuroblastoma risk, Nat. Commun. 8 (1) (2017) 569.
[143] C.A. Galea, et al., Domain structure and function of matrix metalloprotease 23 (MMP23): role in potassium channel trafficking, Cell. Mol. Life Sci. : CMLS 71 (7) (2014) 1191–1210.
[144] R.A. Okimoto, et al., Inactivation of Capicua drives cancer metastasis, Nat. Genet. 49 (1) (2017) 87–96.
[145] J. Wang, et al., Inhibition of fibroblast growth factor receptor signaling impairs metastasis of hepatocellular carcinoma, Tumour Biology : the Journal of the International Society For Oncodevelopmental Biology and Medicine 35 (11) (2014) 11005–11011.
[146] M. Liu, et al., Identification of the MMP family as therapeutic targets and prognostic biomarkers in the microenvironment of head and neck squamous cell carcinoma, J. Transl. Med. 21 (1) (2023) 208.
[147] L.A. Dan, et al., No associations of a set of SNPs in the Vascular Endothelial Growth Factor (VEGF) and Matrix Metalloproteinase (MMP) genes with survival of colorectal cancer patients, Cancer Med. 5 (9) (2016) 2221–2231.
[148] I.M. Clark, et al., The regulation of matrix metalloproteinases and their inhibitors, Int. J. Biochem. Cell Biol. 40 (6–7) (2008) 1362–1378.
[149] S. Chakraborti, et al., Regulation of matrix metalloproteinases: an overview, Mol. Cell. Biochem. 253 (1–2) (2003) 269–285.
[150] L. Zawel, et al., Human Smad3 and Smad4 are sequence-specific transcription activators, Mol. Cell 1 (4) (1998) 611–617.
[151] S. Narayan, T. Thangasamy, R. Balusu, Transforming growth factor -beta receptor signaling in cancer, Front. Biosci. : J. Vis. Literacy 10 (2005) 1135–1145.
[152] L.D. Kerr, D.B. Miller, L.M. Matrisian, TGF-beta 1 inhibition of transin/stromelysin gene expression is mediated through a Fos binding sequence, Cell 61 (2) (1990) 267–278.
[153] W. Yuan, J. Varga, Transforming growth factor-beta repression of matrix metalloproteinase-1 in dermal fibroblasts involves Smad3, J. Biol. Chem. 276 (42) (2001) 38502–38510.
[154] L.A. White, T.I. Mitchell, C.E. Brinckerhoff, Transforming growth factor beta inhibitory element in the rabbit matrix metalloproteinase-1 (collagenase-1) gene functions as a repressor of constitutive transcription, Biochim. Biophys. Acta 1490 (3) (2000) 259–268.
[155] K. Ogawa, et al., Suppression of matrix metalloproteinase-9 transcription by transforming growth factor-beta is mediated by a nuclear factor-kappaB site, Biochem. J. 381 (Pt 2) (2004) 413–422.
[156] J. Lohi, et al., Structural analysis and promoter characterization of the human membrane-type matrix metalloproteinase-1 (MT1-MMP) gene, Gene 242 (1–2) (2000) 75–86.
[157] T. Kitamura, et al., Matrix metalloproteinase 7 is required for tumor formation, but dispensable for invasion and fibrosis in SMAD4-deficient intestinal adenocarcinomas, Laboratory investigation; a journal of technical methods and pathology 89 (1) (2009).
[158] Y. Sano, et al., ATF-2 is a common nuclear target of Smad and TAK1 pathways in transforming growth factor-beta signaling, J. Biol. Chem. 274 (13) (1999) 8949–8957.
[159] E.-S. Kim, Y.-W. Sohn, A. Moon, TGF-beta-induced transcriptional activation of MMP-2 is mediated by activating transcription factor (ATF)2 in human breast epithelial cells, Cancer Lett. 252 (1) (2007) 147–156.
[160] J. Zhong, et al., ERK1/2 and p38 MAP kinase control MMP-2, MT1-MMP, and TIMP action and affect cell migration: a comparison between mesothelioma and mesothelial cells, J. Cell. Physiol. 207 (2) (2006) 540–552.
[161] X. Zhu, et al., TGF-beta1-induced PI3K/Akt/NF-kappaB/MMP9 signalling pathway is activated in Philadelphia chromosome-positive chronic myeloid leukaemia hemangioblasts, J. Biochem. 149 (4) (2011) 405–414.
[162] A. Safina, E. Vandette, A.V. Bakin, ALK5 promotes tumor angiogenesis by upregulating matrix metalloproteinase-9 in tumor cells, Oncogene 26 (17) (2007) 2407–2422.
[163] A. Safina, et al., TAK1 is required for TGF-beta 1-mediated regulation of matrix metalloproteinase-9 and metastasis, Oncogene 27 (9) (2008) 1198–1207.
[164] M. Arsura, et al., Transient activation of NF-kappaB through a TAK1/IKK kinase pathway by TGF-beta1 inhibits AP-1/SMAD signaling and apoptosis: implications in liver tumor formation, Oncogene 22 (3) (2003) 412–425.
[165] Y. Seomun, J.-T. Kim, C.-K. Joo, MMP-14 mediated MMP-9 expression is involved in TGF-beta1-induced keratinocyte migration, J. Cell. Biochem. 104 (3) (2008) 934–941.
[166] J. Wylie-Sears, R.A. Levine, J. Bischoff, Losartan inhibits endothelial-to-mesenchymal transformation in mitral valve endothelial cells by blocking transforming growth factor-β-induced phosphorylation of ERK, Biochem. Biophys. Res. Commun. 446 (4) (2014) 870–875.
[167] E. Terzuoli, et al., Targeting endothelial-to-mesenchymal transition: the protective role of hydroxytyrosol sulfate metabolite, Eur. J. Nutr. 59 (2) (2020) 517–527.
[168] C. Ciavarella, et al., Involvement of miR-30a-5p and miR-30d in endothelial to mesenchymal transition and early osteogenic commitment under inflammatory stress in HUVEC, Biomolecules 11 (2) (2021).

[169] G. Chen, et al., Human cytomegalovirus promotes the activation of TGF-β1 in human umbilical vein endothelial cells by MMP-2 after endothelial mesenchymal transition, Adv. Clin. Exp. Med. : Official Organ Wroclaw Medical University 28 (11) (2019) 1441–1450.
[170] J. Kryczka, et al., HMEC-1 adopt the mixed amoeboid-mesenchymal migration type during EndMT, Eur. J. Cell Biol. 96 (4) (2017) 289–300.
[171] W.M. Ciszewski, et al., *The ILK-MMP9-MRTF axis is crucial for EndMT differentiation of endothelial cells in a tumor microenvironment.* Biochimica et biophysica acta, Molecular cell research 1864 (12) (2017) 2283–2296.
[172] K.M. Welch-Reardon, et al., Angiogenic sprouting is regulated by endothelial cell expression of Slug, J. Cell Sci. 127 (Pt 9) (2014) 2017–2028.
[173] N. Weinstein, L. Mendoza, E.R. Álvarez-Buylla, A computational model of the endothelial to mesenchymal transition, Front. Genet. 11 (2020) 40.
[174] Y. Yin, et al., Tongxinluo attenuates myocardiac fibrosis after acute myocardial infarction in rats via inhibition of endothelial-to-mesenchymal transition, BioMed Res. Int. 2019 (2019) 6595437.
[175] N.I. Marín-Ramos, et al., NEO212, a conjugate of temozolomide and perillyl alcohol, blocks the endothelial-to-mesenchymal transition in tumor-associated brain endothelial cells in glioblastoma, Cancer Lett. 442 (2019) 170–180.
[176] Y. Guo, et al., Kallistatin inhibits TGF-β-induced endothelial-mesenchymal transition by differential regulation of microRNA-21 and eNOS expression, Exp. Cell Res. 337 (1) (2015) 103–110.
[177] X. Chen, et al., Curcumin attenuates endothelial cell fibrosis through inhibiting endothelial-interstitial transformation, Clin. Exp. Pharmacol. Physiol. 47 (7) (2020) 1182–1192.
[178] Z. Jiang, et al., MT1-MMP deficiency leads to defective ependymal cell maturation, impaired ciliogenesis, and hydrocephalus, JCI Insight 5 (9) (2020).
[179] F. Cheng, et al., KSHV-initiated notch activation leads to membrane-type-1 matrix metalloproteinase-dependent lymphatic endothelial-to-mesenchymal transition, Cell Host Microbe 10 (6) (2011) 577–590.
[180] S. Djediai, et al., MT1-MMP cooperates with TGF-β receptor-mediated signaling to trigger SNAIL and induce epithelial-to-mesenchymal-like transition in U87 glioblastoma cells, Int. J. Mol. Sci. 22 (23) (2021).