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# The Role of TGF-β and Its Receptors in Gastrointestinal Cancers

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## Abstract

Early detection of gastrointestinal tumors improves patient survival. However, patients with these tumors are typically diagnosed at an advanced stage and have poor prognosis. The incidence and mortality of gastrointestinal cancers, including esophageal, gastric, liver, colorectal, and pancreatic cancers, are increasing worldwide. Novel diagnostic and therapeutic agents are required to improve patient survival and quality of life. The tumor microenvironment, which contains nontumor cells, signaling molecules such as growth factors and cytokines, and extracellular matrix proteins, plays a critical role in cancer cell proliferation, invasion, and metastasis. Transforming growth factor beta (TGF-β) signaling has dual roles in gastrointestinal tumor development and progression as both a tumor suppressor and tumor promoter. Here, we review the dynamic roles of TGF-β and its receptors in gastrointestinal tumors and provide evidence that targeting TGF-β signaling may be an effective therapeutic strategy.

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## Introduction

Transforming growth factor beta (TGF-β) is a cytokine that participates in both physiological and pathological processes including tumorigenesis [1]. During tumor progression, TGF-β signaling regulates immune/inflammatory response and tumor microenvironment. It also regulates tumor growth, epithelial-mesenchymal transition (EMT), and cancer cell stemness depending on tumor stage and cellular context [2–4]. Malignant gastrointestinal tumors such as esophageal, gastric, liver, colorectal, and pancreatic carcinomas are a major cause of cancer-related deaths worldwide [5]. Aberrant TGF-β signaling has been associated with gastrointestinal cancer progression [6]. Several TGF-β-based therapeutics have been developed for the treatment of gastrointestinal cancers and have displayed efficacy in clinical trials [7,8]. Here, we review the roles of TGF-β and its receptors in gastrointestinal tumors and describe the evidence that targeting TGF-β signaling may be an effective therapeutic strategy.

## TGF-βs and Their Receptors

The TGF-β superfamily consists of at least 40 structurally and functionally related cytokines that are involved in various biological processes including embryonic development, extracellular matrix formation, immune regulation, inflammation, and cancer [1,9,10]. TGF-β family proteins are classified into several subtypes, including TGF-βs, activins/inhibins, and bone morphogenetic proteins (BMPs)/growth differentiation factors according to structural characteristics (Figure 1) [11].

Six TGF-β isoforms have been identified, which display variable sequence homology. TGF-β1, TGF-β2, and TGF-β3 are highly conserved and expressed in mammals [12]. TGF-β4 and TGF-β5 are predominantly expressed in birds and amphibians [13], while TGF-β6 is only expressed in fish [14]. TGF-β1 is the most abundant and ubiquitously expressed of the isoforms. TGF-β is synthesized in an inactive form (pre-proTGF-β), which contains a signal peptide, a pro region, and the mature coding region. Following removal of the signal peptide, the TGF-β dimer interacts with latency associated peptide, a protein derived from the N-terminal region of the proTGF-β, to form the small latent complex. This complex is secreted into the extracellular matrix upon binding to latent TGF-β-binding

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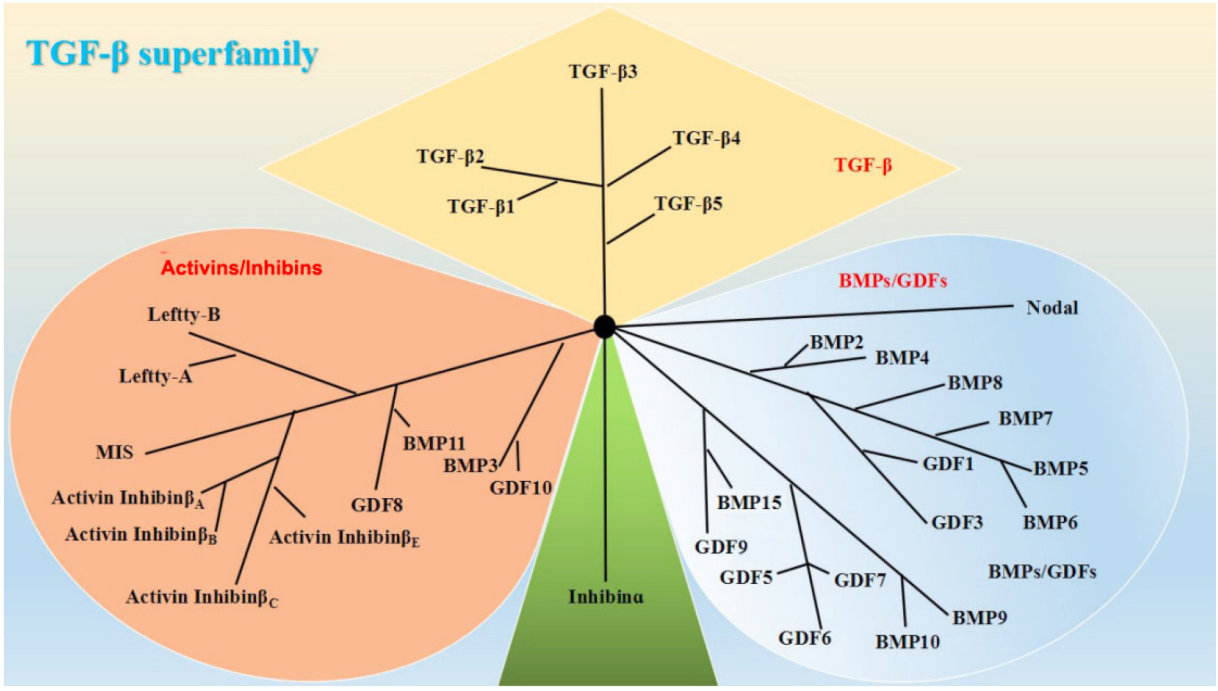


**Figure 1.** The TGF-β superfamily. Based on their structural features, the mammalian members of the TGF-β family are subdivided into (i) TGF-βs, (ii) activins/inhibins, and (iii) BMPs/growth and differentiation factors (GDFs).

protein to form the large latent complex. TGF-β is then active until it is released from the large latent complex by proteases, integrins, pH, or reactive oxygen species. Active TGF-β consists of two identical peptide chains with a molecular weight of 25 kDa [15].

TGF-β signals through TGF-β receptors (TβRs) I and II to activate downstream signaling pathways [11,16]. The TβRs are single-pass transmembrane proteins with serine/threonine kinase activity. Seven TβRIs and five different TβRIIs have been identified, which confer intracellular signaling specificity to all members of the TGF-β superfamily [1]. In the absence of ligand, TβRI and TβRII exist as monomers, homodimers, or heterodimers on the cell surface. Ligand binding promotes formation of a tetrameric complex between TβRII dimers and two TβRIs [17]. TβRIs and TβRIIs have N-terminal extracellular ligand binding domains, transmembrane segments, and C-terminal cytosolicserine/threonine kinase domains [11]. TGF-β binds specifically to the constitutively active TβRII, which activates TβRI by phosphorylating the glycine/serine-rich domain. Activated TβRI then phosphorylates downstream effectors to induce signal transduction (Figure 2) [17]. TβR activity is regulated by betaglycan, a type III TβR, and endoglin [16].

## Canonical TGF-β Signaling

Canonical TGF-β signaling is dependent upon Smad family proteins. Active TGFβI at the cell surface phosphorylates receptor-activated Smads (R-Smads). There are two sub-classes of R-Smads in which Smad2 and Smad3 mediate the TGFβ/activin pathway. Smad4 acts as a co-factor that binds to activated R-Smads to form a complex that translocates to the nucleus and regulates transcription (Figure 2) [1,18,19]. Interestingly, R-Smads have also been shown to interact with other proteins such as tripartite motif-containing 33 to regulate gene transcription [20].

TGF-β signaling is regulated through various mechanisms. Inhibitory Smad (Smad7) can inhibit TGF-β signaling by interacting with TβRI and R-Smads [21] (Figure 2). R-Smad stability is also modulated by phosphorylation [22]. Additionally, Smad ubiquitination regulatory factor (Smurf)–mediated TβRI degradation can dynamically regulate TGF-β signaling [23].

## Noncanonical TGF-β Signaling

TβRs can also activate non–Smad-dependent signaling pathways, including the mitogen-activated protein kinase (MAPK) pathway mediated by p38, c-Jun amino terminal kinase (JNK), extracellular signal-regulated kinases (ERK), nuclear factor-κB (NF-κB), Rho, and phosphatidylinositol 3-kinase (PI3K)-Akt (Figure 2) [24,25]. These non-Smad pathways might mediate signaling transduction alone or in cooperation with the canonical Smad pathway to modulate TGF-β signaling activity.

TGF-β–induced activation of ERK can phosphorylate transcription factors. Activated TGF-β receptors interact with tumor necrosis factor receptor–associated factor 6 and TGF-β-activated kinase 1 to activate multiple downstream kinases including JNK, p38 and IkB kinase. Activated JNK and p38 can phosphorylate the targeted transcription factors, while IkB kinase phosphorylates NF-κB. Activated Akt can control translation through mammalian target of rapamycin-1. These non–Smad-mediated translational and transcriptional responses cross talk with Smad-mediated transcriptional responses to contribute to either tumorigenesis or tumor suppression. Non-Smad proteins can also directly impact R-Smad activity. ERK can regulate the activity of R-Smads in a phosphorylation-dependent manner, while Akt regulates Smad3 activity by sequestering Smad3 in the cytoplasm. Furthermore, TGF-β also activates RhoA and Rho-associated protein kinase to induce actin polymerization involved in EMT process. Thus, cross talk between the canonical and noncanonical TGF-β signaling pathways contributes to tumor development (Figure 2) [25]. Cross talk has also been demonstrated

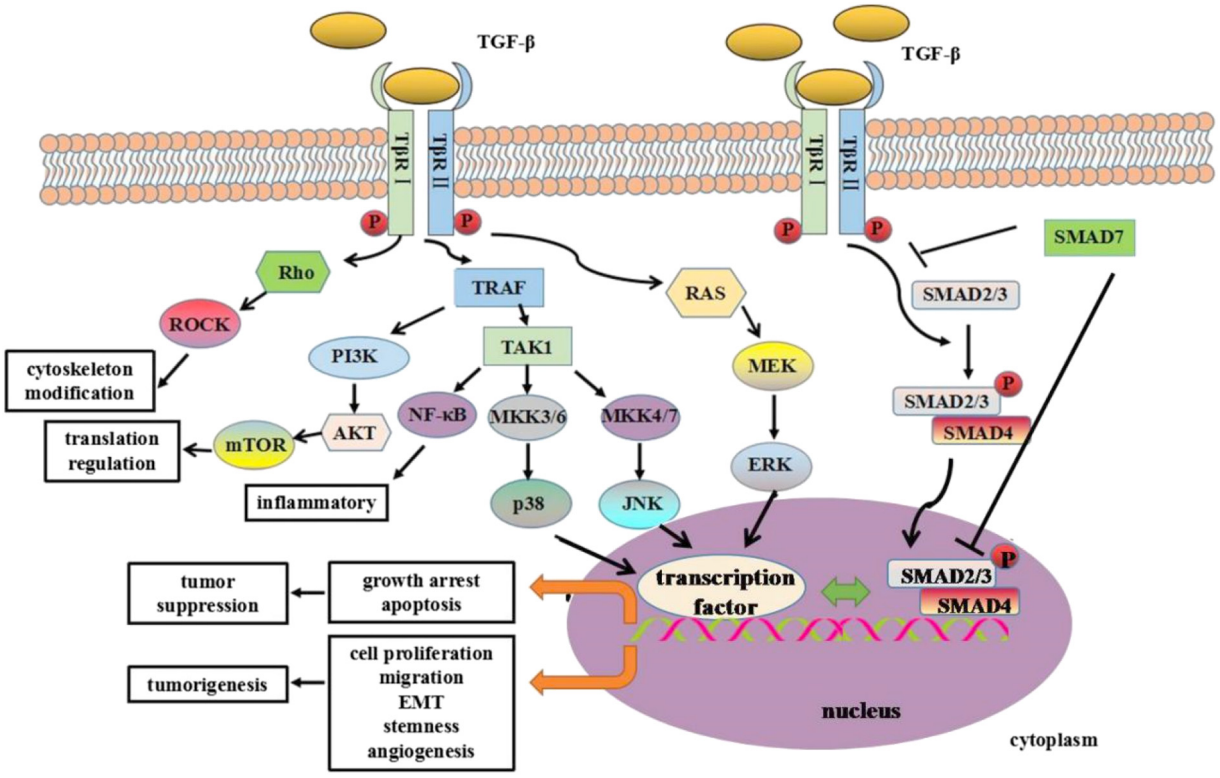


**Figure 2.** Schematic representation of TGF-β signaling as well as the role of TGF-β signaling pathway in cancer onset and progression. TGF-β activates both Smad-dependent canonical and Smad-independent noncanonical signaling pathways. TGF-β binds to TβRII which then activates TβRI. TβRI-phosphorylated Smad2/3 form complexes with Smad4, entering nucleus and regulating the transcription of different targeted genes in both early and late stages of tumor development, contributing to tumor suppression and tumorigenesis, respectively. Smad7 antagonizes TGF-β signaling through blocking Smad2/3 activation and interfering with the formation of Smad2/3/4-DNA complex. In the noncanonical signaling pathways, TGF-β receptors initiate the signal through MAPKs, PI3K, and Rho family of small GTPases etc. Activated JNK/p38/ERK either interact with SMADs or induce their individual transcriptional programs directly to affect cancer cells. Rho-activated Rho-associated protein kinase is involved in cytoskeleton modification in the process of EMT. Through PI3K-AKT pathway, TGF-β can also activate mammalian target of rapamycin to regulate protein translation. In addition, TGF-β activation of the tumor necrosis factor receptor–associated factor proteins can also induce NF-κB signaling for inflammatory response.

between the TGF-β signaling pathway and the tumor necrosis factor-α and epidermal growth factor receptor pathways [24–26].

## Role of TGF-β Signaling in Cancer

Altered TGF-β expression has been observed in several cancers [2,7,8]. Interestingly, TGF-β has dual roles in tumor progression, acting as both a tumor suppressor and tumor promoter in a stage- and context-dependent manner [4,7,8]. During tumor initiation, TGF-β signaling promotes cell cycle arrest and apoptosis, thereby acting as a tumor suppressor. In contrast, TGF-β has been shown to promote tumor cell proliferation, EMT, and stem-like behavior as well as fibrosis, inflammation, and angiogenesis during tumor progression (Figure 2) [4,7,8]. Upregulation of TGF-β expression was correlated with poor prognosis in patients with advanced-stage tumors. The accumulation of mutations in TGF-β signaling pathway components during tumor progression may contribute to the switch in TGF-β function from tumor-suppressive to tumor-promoting.

## Role of TGF-β Signal in Gastrointestinal Cancers

### *TGF-β Signal and Esophageal Cancer*

Esophageal cancer is the sixth leading cause of cancer-related death worldwide [27,28]. Esophageal squamous cell carcinoma (ESCC) and esophageal adenocarcinoma are the most common histological subtypes of esophageal cancer [29]. Risk factors for esophageal cancer include smoking, consumption of hot liquids, poor oral health, and vitamin deficiency [28]. Recent studies have provided insight into the role of TGF-β signaling in esophageal cancer. Interestingly, higher serum TGF-β levels were observed in patients with esophageal cancer compared to healthy controls, and reduced levels were observed following radiotherapy [30]. Upregulation of TGF-β and concomitant overexpression of vascular endothelial growth factor (VEGF) were correlated with tumor size in ESCC [31]. Additionally, overexpression of TGF-β and reduced TβR expression were associated with depth of invasion and pathologic stage in ESCC [32]. Smad4, but not Smad2/3, expression was inversely correlated with invasion in ESCC [33]. Smurf2-induced Smad2 degradation may also contribute to tumor development and poor prognosis in ESCC [34]. Finally, TGF-β/Smad signaling has been shown to promote EMT in ESCC through PTEN/PI3K [35,36]. Thus, TGF-β signaling plays a role in esophageal cancer progression.

MicroRNAs (miRNAs) are diagnostic and prognostic biomarkers in several cancers and have been shown to regulate TGF-β signaling in esophageal cancer [37,38]. MiR-17/20a suppressed ESCC cell migration and invasion via the TGF-β/integrin β6 subunit pathway by targeting TβR2 and Smad anchor for receptor activation for

degradation [39]. Additionally, TGF-β was shown to play a role in miR-455-3p–mediated ESCC progression [40]. MiR-655 suppressed ESCC progression by targeting zinc finger E-box binding homeobox 1and TβRII, which are required for TGF-β signaling, and suppressing EMT [41]. MiR-32 was recently found to promote ESCC metastasis through CXXC5-mediated inhibition of TGF-β signaling [42].

Cross talk between cytoskeleton-associated proteins, which are aberrantly expressed in esophageal cancer, and the TGF-β signaling pathway can promote ESCC progression. Reelin, which has a key role inneuronal migration, negatively regulates TGF-β–induced ESCC cell migration. Reelin expression was suppressed by the TGF-β pathway through the transcription factor Snail [43]. Fascin is an actin bundling protein that induces cell membrane protrusions. It regulates ESCC cell proliferation and invasion by modulating the levels of connective tissue growth factor and cysteine-rich protein 61 via the TGF-β pathway [44]. Overexpression of cysteine-rich protein 61 and connective tissue growth factor was associated with poor survival in ESCC [45]. Ezrin, a cytoskeletal cross-linking protein, also promotes ESCC cell proliferation and invasion through the TGF-β and MAPK signaling pathways [46]. Collectively, these data indicate that TGF-β signaling can promote esophageal cancer development and metastasis.

### *TGF-β Signal and Gastric Cancer*

Gastric cancer (GC) is the second leading cause of cancer-related death worldwide. It has a particularly high mortality rate in Asia, which may be linked to both social and environmental factors [47,48]. Despite early diagnosis and treatment with a combination of surgery, chemotherapy, and/or radiotherapy, the prognosis of GC patients is poor due to recurrence and distant metastasis [49]. Previous studies have demonstrated higher TGF-β levels in serum from GC patients compared to healthy controls. Elevated TGF-β levels were correlated with lymph node metastasis, worse overall survival, and poor prognosis in GC patients [50]. TGF-β was also increased in the gastric mucosa and in precancerous gastric cells [51]. Altered TGF-β signaling has been observed during GC progression. Additionally, mutations in TβRII have been implicated in gastric carcinogenesis [52]. Repression of TβRI transcription through CpG island methylation was also correlated with poor prognosis in GC patients [53]. Various mutations in the promoter regions of *TGFB1* and *TGFBR2* were associated with the riskofGC in a Chinese population [54]. In addition, an imbalance in Smad4/7 expression was associated with GC cell differentiation, metastasis, and apoptosis [55]. These data suggest TGF-β plays an important role in GC development.

EMT results in gastric epithelial cells acquiring mesenchymal characteristics and promotes stemness, invasion, and metastasis [56,57]. EMT can be induced by pathogens, stress, and hypoxia [58]. EMT is modulated by the microenvironment in gastric and other cancers [58,59]. TGF-β, Notch, and Wnt signaling can induce EMT in GC cells, thereby promoting tumor progression [58].

Several proteins regulate TGF-β-induced EMT in GC and have opposing effects. For example, ankyrin-repeat-containing, SH3-domain-containing, and proline-rich-region-containing protein 2 [60], grainyhead-like 2 [61] and metastasis suppressor protein 1 [62] suppress invasion and TGF-β–induced EMT in GC cells, whereas homeobox protein A13 [63], CC-chemokine receptor 7 [64], and homeoprotein Bapx1 [65] promote TGF-β–induced EMT. MiRNAs also regulate EMT in GC cells. MiR-381 inhibited TGF-β signaling and suppressed EMT in GC cells in part by targeting transmembrane member 16A [66]. In contrast, miR-21 enhanced TGF-β–induced EMT in GC cells by upregulating PTEN expression [67]. MiRNAs may contribute to GC progression by directly targeting components of the TGF-β signaling pathway. MiR-17-5p and miR-155 target TβRII in GC cells and are involved in proliferation and migration, while miR-424-5p, which is upregulated in GC cells, inhibits TGF-β signaling by targeting Smad3 and promotes GC cell proliferation [68–70]. Finally, miR-199a was found to target Smad4 in GC [71].

### *TGF-β Signal and Hepatocellular Carcinoma*

Hepatocellular carcinoma (HCC) is the most common malignancy worldwide [72,73]. The primary causes of HCC include toxins (e.g., alcohol and aflatoxin) and chronic hepatitis B virus or hepatitis C infection (account for approximately 80% of cases) [74]. Most patients are diagnosed at an advanced stage due to a lack of symptoms and rapid disease progression. The 5-year overall survival rate is less than 20% [75]. Surgical resection and liver transplantation are the best treatment options for HCC [76]. However, there is a high rate of recurrence and metastasis following curative hepatectomy [77]. A better understanding of the molecular mechanisms underlying HCC progression is required to identify more effective therapeutic targets.

Elevated TGF-β levels have been observed in patients with metastatic HCC compared to those with nonmetastatic disease [78,79]. Immunostaining demonstrated expression of all three TGF-β isoforms in HCC cells and in the perineoplastic stroma, while little to no expression was detected in normal liver tissue [80]. Serum TGF-β levels were associated with disease progression and poor prognosis [81]. TGF-β may be a feasible marker for the detection of early-stage HCC because it has displayed higher sensitivity than traditional markers such as alpha-fetoprotein in past studies [82]. Loss of TβRII expression was correlated with tumor progression in HCC [83]. Additionally, mutations in Smad2 and Smad4 have been observed in a small minority of HCC patients [84]. TGF-β induces alterations in the cellular composition of the tumor microenvironment, which may also play an important role in HCC progression [85,86]. A recent study stated that the combination of the expression level of c-Myc, TGF-β1, and embryonic liver fodrin (an adaptor protein of TGF-β signaling cascade) together can be used to accurately predict outcomes of patients with HCC [87].

Emerging evidence suggests TGF-β signaling has a dual or biphasic role in HCC development and progression [88,89]. TGF-β acts as a tumor suppressor in early carcinogenesis but promotes tumor progression during later stages. Consistent with these observations, reduced TβRII correlated with increased tumor size and intrahepatic metastasis in HCC patients [83], and with the risk of HCC in a mouse model [90]. Additionally, reduced TβRII staining has been observed in approximately 25% of malignant compared to adjacent nonmalignant hepatocytes [91]. TGF-β was also found to suppress HCC proliferation through activating Hippo signaling [92]. NADPH oxidase 4 may mediate the antiproliferative and proapoptotic effects of TGF-β in HCC cells [93]. On the other hand, TGF-β1 contributes to HCC invasion and metastasis by promoting fibroblast growth factor receptor 4 expression via the noncanonical ERK pathway [94]. AGO1, the main component of RNA-induced silencing complexes, may promote HCC metastasis in a TGF-β–dependent manner [95]. Finally, miRNAs may regulate EMT in HCC cells by targeting TGF-β signaling. The miR-200 family of miRNAs was shown to suppress metastasis in human HCC cells [96], and miR-125b was found to suppress EMT by targeting Smad2 and Smad4 [97].

The mechanisms underlying the dual roles of TGF-β signaling during tumor progression have not been elucidated. Previous studies have suggested that HCC cells may become resistant to the antiproliferative effects of TGF-β but maintain sensitivity to the tumor-promoting effects [98]. Importantly, upregulation of growth factor–mediated survival signals, such as MAPK/ERKs, PI3K/Akt, and NF-κB, suppressed the antiproliferative effects of TGF-β in HCC cells [99]. TGF-β promotes the production of growth factors and cytokines such as PDGF and EGF, which contribute to tumor cell proliferation and invasion [99]. PDGF links TGF-β signaling to nuclear β-catenin accumulation during HCC progression [100]. EGF promotes HCC cell survival by protecting the cells from TGF-β–induced cell death [101]. A recent study demonstrated that abrogation of the posttranscriptional regulation of c-Myc in an epigenetic modification-dependent manner promoted HCC cell resistance to antiproliferative TGF-β signaling [102].

A temporal TGF-β-specific gene expression signature was established in mouse hepatocytes that could distinguish between HCC subtypes. Tumors expressing early TGF-β–responsive genes displayed the physiological growth inhibitory effects of TGF-β, whereas tumors expressing late TGF-β–responsive genes displayed aberrant upregulation of TGF-β signaling and the antiapoptotic and invasive effects of TGF-β. The TGF-β–specific gene expression signature could predict HCC invasion and liver metastasis [103].

### *TGF-β Signal and Colorectal Cancer*

Colorectal cancer (CRC) is one of the three most common cancers worldwide [104,105]. The incidence of CRC is higher in developed countries than in less developed countries, and nutrition is thought to be a major contributing factor [104]. The 5-year survival rate is less than 20%in patients with metastatic disease compared to 70%-90% in patients with nonmetastatic disease [104]. Approximately one third of all CRC cases are inherited [106]. Highly penetrant tumor susceptibility genes account for only 3%-6% of all CRC cases. The remaining fraction of the unexplained risk can likely be attributed to a combination of low to moderately penetrant genes and polygenic mechanisms [107].

Previous studies have demonstrated increased TGFB1 expression in CRC compared to benign adenoma and noncancerous tissue [108]. Elevated TGF-β levels were observed in primary tumor tissue and in plasma from CRC patients and were correlated with metastasis and poor prognosis [109,110]. TGF-β was predominantly detected in the central regions of CRC liver metastases [111]. TGF-β induced growth arrest in moderately differentiated colon carcinomas but promoted proliferation in more aggressive tumors [112]. Reduced phosphorylation of the C-terminal region of Smad3 and activation of c-Jun NH(2)-terminal kinase–mediated phosphorylation of the linker region of Smad2/3 were observed in the human colorectal adenoma to carcinoma sequence, from which the majority of CRCs arise [113,114]. Notably, inhibition of TGF-β can prevent CRC metastasis by unleashing a cytotoxic T-cell response against cancer cells, implying that TGF-β signaling suppresses cancer recognition by the immune system [115].

Mutational inactivation of the TGF-β signaling pathway is a key cause of CRC progression [107,116]. Alterations in TGF-β signaling have been observed in nearly 50% of CRCs [107,116]. For example, mutations in TGFBR2 that abolish TGF-β signaling have been frequently detected in CRCs [107,117]. More than 80% of CRCs that display microsatellite instability harbor mutations in TGFBR2 (particularly frameshift mutations in exon 3) [116,118]. The exact mechanisms by which TGFBR2 mutations lead to CRC have not been elucidated. However, a previous study demonstrated that inactivation of TβRII induced VEGF-A expression, which enhanced the metastatic potential of CRC cells [119]. TGF-β signaling was still active in some CRCs that displayed a high level of microsatellite instability despite frameshift mutations in TGFBR2 [120]. Mutations in TGFBR1 have also been detected in CRC [121]. For example, loss of three alanine residues within a stretch of nine alanine residues in the N-terminal region of TβRI (TGFBR1*6A) was associated with an increased risk of CRC [122], though these results have not been confirmed [123]. However, a recent study pointed out an oncogenic property of TGFBR1*6A which may promote the migration and invasion of colorectal cancer cells [124].

Mutations in downstream components of the TGF-β signaling pathway may also contribute to sporadic CRC development [107]. Smad4 is the most commonly disrupted Smad family protein in various cancers. It is mutated or lost in up to one third of all CRCs. Smad4 mutation or loss of expression has been frequently observed in late-stage tumors [125–128]. Loss of Smad4 could alter BMP signaling to promote CRC metastasis through activation of Rho and Rho-associated protein kinase [129]. Mutations in Smad2 have been detected in approximately 3%-6% of CRC tumors [128,130]. Mutations were more frequently observed in early-stage tumors. Both the Smad2 and Smad4 genes are located on chromosome 18q. This region is commonly deleted in CRC owing to a loss of the long arm of chromosome 18 (loss of heterozygosity) [131]. Smad7 is also located in this region [131].

Interestingly, a low-frequency coding variant, rs3764482 (c. 83C>T; p. S28F) in Smad7, was associated with the risk of CRC in a Chinese population [132]. A large-scale meta-analysis demonstrated that several single nucleotide polymorphisms in Smad7 were associated with CRC [133]. Mutations in Smad3 were also identified and had similar frequencies to those of the Smad2 mutations in sporadic CRCs [128]. In addition to the somatic mutations described above, germline mutations in Smads including Smad4 have been observed in several patients with juvenile polyposis syndrome [134], which can develop into CRC [135]. Thus, altered TGF-β signaling plays various stage-specific roles in CRC progression with mutations in TGFBR2 and Smad as the major contributor.

### *TGF-β Signal and Pancreatic Cancer*

Pancreatic cancer is a leading cause of cancer-related death worldwide [136]. Risk factors for pancreatic cancer include tobacco use, obesity, and exposure to certain chemicals. In addition to genetic factors, age and gender are also correlated with risk of pancreatic cancer [136,137]. Pancreatic ductal adenocarcinoma (PDAC) is the major histological subtype of pancreatic cancer (90% of cases). The median survival of PDAC patients is 6 months, and the 5-year survival rate is approximately 6% [138,139].

Previous studies have demonstrated that increased TGF-β expression promotes disease progression in PDAC [140,141]. Low circulating levels of TGF-β have been associated with prolonged survival in pancreatic cancer patients [142]. Increased TβR1 and TβR2 expression has been observed in the majority of pancreatic cancer subtypes [143]. Activation of TGF-β receptor signaling in PDAC cells resulted in increased Smad3 phosphorylation and nuclear translocation, leading to inhibition of cell growth. However, it also can result in activation of Smad7, which antagonizes Smad3, leading to activation of VEGF-A, vascularization, and metastasis [144].

TGF-β appears to have dual roles in PDAC progression [139]. Overexpression of TGF-β in early-stage PDACs was associated with reduced tumor cell proliferation and improved survival [145]. Mutations in TGF-β pathway proteins may explain the stage-dependent functions of TGF-β in PDACs. Mutations in Smad4 have been observed in approximately 50% of PDACs [146,147]. Mutations in TβRII have been identified in 4%-7% of pancreatic cancers [146,148].

KRAS is mutated in nearly all PDACs and is a major driver of carcinogenesis [149]. However, mutation of KRAS alone is not sufficient for malignant transformation [150,151]. One recent study demonstrated that mutant KRAS dosage along with other oncogenic gains like Myc drives the early progression of PDAC [152]. Additional mutations in tumor suppressors such as Smad4 and CDKN2A are required for PDAC initiation [151]. Smad4 loss of function may promote KRAS-driven malignant transformation of pancreatic ductal cells [150]. In addition, inactivation of retinoblastoma 1 converts TGF-β from a tumor suppressor to a protumorigenic factor that enhances PDAC cell proliferation [153]. Rac1 may also induce a switch in TGF-β signaling by antagonizing Smad2 and Smad3 activation in PDAC cells [154]. TGF-β also regulates the interaction between tumor cells and the surrounding stroma, which can promote PDAC initiation and metastasis [2,155].

### *TGF-β–Based Therapies for Gastrointestinal Cancers*

Elevated TGF-β expression in serum and tumor tissue was correlated with tumor stage and prognosis in gastrointestinal cancers [6]. Among different gastrointestinal cancers, dysfunction of TβR(s) is derived from multiple levels including transcription, translation, and mutation. It should be noted that TGFBR(s) mutation is frequently observed in pancreatic cancers, while alternation of TβR(s) level often appears in other types of gastrointestinal cancers. Additionally, downstream components of the TGF-β signaling pathway are also dysregulated during gastrointestinal tumorigenesis. Thus, alternation from these three levels (ligand, receptor, and signal transducer) together contributes to the development of gastrointestinal cancers, and therapeutics that target the TGF-β signaling pathway may be effective for the treatment of gastrointestinal cancers. TGF-β has dual roles in gastrointestinal cancer initiation and progression. Because it functions as a tumor suppressor during early-stage carcinogenesis and as a tumor promoter during later stages, the development of TGF-β–based therapeutics is challenging. Therapeutics could target TGF-β, TβRI or TβRII, or the Smads [156]. Indeed, small molecule therapeutics, antibodies, and inhibitors against these targets are currently in clinical trials [7,156], such as antisense oligonucleotides against TGF-β or TβR to lower their synthesis, TGF-β–neutralizing monoclonal antibodies and anti-TβR monoclonal antibodies to interrupt ligand-receptor interaction, and small molecule inhibitor of TβR to prevent signaling transduction. Galunisertib, a TβRI kinase inhibitor, displayed considerable tumor suppression effects in PDAC and HCC and has entered the clinical phase II trial. Trabedersen, a TGF-β2 antisense oligodeoxynucleotide, showed certain effects in clinical trials in PDAC and CRC patients [7]. Although few drugs targeting TGF-β display efficacy in clinical trials, the future of TGF-β pathway-based strategies against gastrointestinal cancers is promising from these encouraging clinical trials [6].

## Conclusions

Existing research allows for the conclusion that dysregulation of TGF-β signaling pathway is tightly associated with the development and progression of various gastrointestinal cancers. However, there is still much to learn about the biology of TGF-β signaling under normal conditions and how its dysfunction contributes to the different types of gastrointestinal cancers. TGF-β has critical roles at different stages during gastrointestinal cancers' initiation and metastasis. Definitely, context is important to define the biological meaning of dynamic regulation of TGF-β signaling in the progression of these cancers. A better understanding of the molecular mechanisms underlying the roles of TGF-β, TβRs, and their downstream signaling pathways as well as the complicated cross talk among them during gastrointestinal cancer progression is important in order to develop more effective therapeutics for these diseases. We are still at the early stage in understanding what role TGF-β signaling plays in the development of gastrointestinal cancers. This stage is set to explore the therapeutics that target TGF-β signaling in gastrointestinal cancers, possibly in concert with other effective agents to maximize the therapeutic benefits and improve the outcome of these lethal cancers.

## Consent for Publication

We have obtained consents to publish this paper from all the participants of this study.

## Competing Interests

The authors declare that they have no competing interests.

## CRediT authorship contribution statement

**Jingwen Luo:** Writing - original draft. **Xu-Qiao Chen:** Writing - review & editing. **Ping Li:** Writing - review & editing.

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