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REVIEW

CANCER-ASSOCIATED FIBROBLASTS (CAFs): A NEW CHALLENGE FOR PATHOLOGISTS AND ONCOLOGISTS, AND A PROMISING TARGET FOR A MODERN ONCOTHERAPY

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ABSTRACT: According to the "seed and soil" theory, cancer progression is influenced by the crosstalk between tumor cells and the surrounding tumor microenvironment (TME). TME comprises several cell types, including tumor-associated macrophages (TAMs), tumor-associated neutrophils (TANs), lymphocytes, leukocytes, vascular endothelial cells, pericytes, and fibroblasts. Cancer-associated fibroblasts (CAFs) are a crucial component of the TME and contribute to tumor development and progression. CAFs are a heterogeneous population originating from different cells including resident fibroblasts, circulating fibrocytes, smooth muscle vascular cells, endothelial cells, mesothelial cells, and organ-specific cells such as hepatic stellate cells in the liver. CAFs can play a dual role in the development and progression of tumors, acting as both promoters and inhibitors of tumor growth. The acquisition of a hypermethylated state is associated with a protumorigenic phenotype. Multiple subtypes have been identified in CAFs: inflammatory (iCAFs), myofibroblastic (myCAFs), antigen-presenting (apCAFs), complement-secreting (csCAFs), and CAFs with high metabolic state (meCAFs). In this review, we will discuss the origin of CAFs, their heterogeneity, their interaction with the other components of the TME, and their role in cancer insurgence and progression. The immunohistochemical marker useful for the identification of CAF subtypes in clinical practice will be discussed. The mechanisms by which CAFs may promote cancer angiogenesis, inhibit cancer cell apoptosis, and induce epithelial-to-mesenchymal transition will be analyzed. In the final part, targeted therapies for CAFs, including their direct removal, the inhibition of CAF signaling, and CAF reprogramming will be discussed.

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Impact statement: Our work highlights the role that components of the tumor microenvironment play in tumor growth and progression. In particular, tumor-associated fibroblasts, which could be useful therapeutic targets in the future.

Key words: *cancer-associated fibroblasts; CAFs; target therapy; tumor microenvironment; TME; stromal fibroblasts.*

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INTRODUCTION

According to the "seed and soil" theory, introduced by Steven Paget in 1889 (1), the insurgence and progression of cancer are strictly related to the crosstalk between tumor cells and the peritumoral stromal cells (2). Recent studies have identified Paget's

theory as the key determinant for a better comprehension of the role played by stromal cells in cancer insurgence and development (3). Upon histological examination, the stroma surrounding the tumor presents as a mildly eosinophilic and amphophilic myxoid stroma, which may be categorized as hyalinizing fibrosis or desmoplastic reaction (4). In clinical practice, surgical pathologists easily recognize the typical aspect of the desmoplastic reaction since it is often associated with cancer insurgence and/or relapse (5, 6). Recent studies have revealed that peritumoral stromal cells express L1 cell adhesion molecule (L1CAM), which can affect cancer progression and prognosis (7). Based on recent findings, the traditional view that tumors are cell-centric has been revised. Pathologists are now encouraged to analyze the tumor microenvironment (TME) to determine its role in cancer. The TME is a complex tissue comprising various cell types, such as tumor-associated macrophages (TAMs), tumor-associated neutrophils (TANs), lymphocytes, leukocytes, vascular endothelial cells, pericytes, and fibroblasts. Among the stromal cells giving rise to the TME, peritumoral-activated fibroblasts, better known as cancer-associated fibroblasts (CAFs), play a key role in cancer insurgence, progression, invasion, and metastasis (8, 9). Here we will report the most recent data on the origin and the multiple functions of CAFs in cancer insurgence and progression, trying to allow their identification among the multiple cell types that are present in the peritumoral stroma.

FIBROBLASTS IN HEALTH AND DISEASE

After the first description in 1858 by Rudolph Virchow, who reported the presence of "spindle-shaped cells" in the connective tissue, the term "fibroblast" was subsequently proposed by Ernst Ziegler in 1895 (10).

Fibroblasts represent a very heterogeneous cell population, characterized by high plasticity, which may acquire stem/progenitor cell properties, upregulating Sca-1 (Stem Cell Antigen-1) (11). In physiology, fibroblasts support the function of multiple organs, by maintaining the architecture of extracellular matrix-rich connective tissues (12). During embryonic development, fibroblasts actively partecipate to tissue homeostasis by producing multiple factors of the extracellular matrix, creating signalig niches for stem/progenitor cells, exhibiting a marked plasticity in phenotype and cell fate (10). Moreover, fibroblasts may serve as progenitors of specialized mesenchymal cells, which may differentiate towards bone-forming osteoblasts, lipid-filled adipocytes and condrocytes (13).

Fibroblasts are mainly involved in wound healing, with multiple roles: recruitment and activation of innate immune cells, synthesis and deposition of scar tissue, remodeling of the extracellular matrix and acquisition of a contractile phenotype, leading to the fusion of the wound margins (14). Moreover, fibroblasts may promote macrophage activation, accelerating wound healing (15).

A major role for fibroblasts has been recently evidenced in the progression of the atherosclerotic plaque. In the injuried arterial wall, fibroblasts are activated to myofibroblasts, acquiring the expressionn of alpha-smooth muscle actin (alpha-SMA), promoting leukocyte recruitment, producing proinflammatory cytokines and orchestrating extracellular matrix production and remodeling (16).

CELL TYPES OF THE TUMOR MICROENVIRONMENT

The TME is formed by multiple cell types including tumor-associated neutrofils, natural killer cells, B- and T-lymphocytes, vascular endothelial cells, pericytes, myeloid-derived suppressor cells (MDSCs), dendritic cells, enteric glial cells, CAFs and TAMs (17). Among the many immune cells populating the TME, macrophages are among the most abundant, playing a cancer-promoting role. In particular, TAMs induce neoangiogenesis and promote tumor cell invasion (18). Recent evidence suggests the existence of a two-way dialogue between cancer cells and the tumor environment, with multiple cell types, molecules and physical forces acting simultaneously in the TME, profoundly affecting tumor behaviour (19). According with this view, environmental cues occurring in the TME might either drive or dampen malignant transformation and cancer progression. In the TME, CAFs play a fundamental role in ECM remodeling, in the acquisition of a motile phenotype by cancer cells leading to metastasis and favoring the insurgence of therapeutic resistance to immunotherapy (20). Understanding the communication between cells in the TME, especially between CAFs and TAMs, is crucial for developing new therapeutic strategies in oncology. CAFs and TAMs work together to produce

chemokines and cytokines that regulate communication with other cells in the TME (21).

Communication between TAMs and CAFs is not limited to cytokines and soluble factors. Non-coding RNAs (ncRNAs) and exosomes facilitate tumor-stromal and stromal-stromal cell interactions, potentially affecting the properties of both the tumor and stromal cells (22).

When cells undergo malignant transformation, the peritumoral stroma and microenvironment actively attempt to suppress and eliminate them. Consequently, tumor cells try to resist the suppressive efforts of the peritumoral stroma. During cancer progression, tumor cells may attract distant mesenchymal stem cells, inducing them to change their phenotype and function within the TME: from one attempting to suppress cancer cells to one which will support their survival and growth. Eventually, the TME modified by the cancer cells and enriched with CAFs and TAMs acquire a new ability to support cancer progression and metastasis (23, 24).

CANCER-ASSOCIATED FIBROBLAST ORIGINS

The origins of CAFs are still a topic of debate, likely due to the absence of a specific marker for them. Additionally, recent studies exploring CAFs in different types of tumors suggest that their origin can vary from one organ to another, and even within the same organ for different cancer types. The fibroblasts present in the TME can originate from various cell types, and this happens through the activation of different molecular pathways (25). Fibrocytes that circulate in the blood originate from hematopoietic stem cells and are a specific type of white blood cell that expresses both lymphocytic markers (CD45) and fibroblastic markers (Collagen type-I) (26). In a mouse model of colorectal carcinogenesis, CAFs emerge from intestinal pericriptal cells expressing the leptin receptor (Lepr) and the melanoma cell adhesion molecule (MCAM) (27). In HCC, CAFs originate from various cell types, such as fibrocytes, peritumoral fibroblasts, vascular smooth muscle cells, and mesothelial cells (28). It was discovered that hepatic stellate cells are the primary source of CAFs in HCC (29). In HCC, CAFs play a key role in the early stages of cancer development. They communicate with cancer cells, support angiogenesis, and suppress the immune system (30).

Another cell type from which CAFs may originate is the endothelium of the vessels embedded in the peritumoral stroma. Through the process known as an endothelial-to-mesenchymal transition (EndMT), endothelial cells may lose their typical markers, detach from the neighboring endothelial cells, acquire a migratory phenotype, and gain mesenchymal markers (31).

The analysis of the methylation status of CAFs in breast cancer stroma evidenced that the transition from fibroblasts to activated fibroblasts and CAFs is characterized by massive methylation, responsible for the transcriptional reprogramming of CAFs. The acquisition of a hyper-methylated state might be responsible for the acquisition of a protumorigenic phenotype in CAFs. The extensive epigenetic rewiring of CAFs could be linked to the upregulation of RUNX1, a mediator of DNA methylation. Upregulation of RUNX1 expression in breast cancer patients is associated with poor disease outcomes (32).

In peritoneal metastases, CAFs may originate from mesothelial cells, which undergo a process of mesothelial-to-mesenchymal transition (MMT). The ability of mesothelial-derived CAFs to promote adhesion, invasion, and vascularization of tumor cells is crucial for the progression of metastases in the peritoneum (33). In pancreatic ductal adenocarcinoma, CAFs may originate from pancreatic stellate cells and tumor-infiltrating mesenchymal stem cells (34). In gastric carcinoma, approximately 20% of CAFs have been reported to originate from bone marrow-derived mesenchymal stem cells (35). In breast cancer, the expression in CAFs of alpha-smooth muscle actin (alpha-SMA), the typical marker of smooth muscle cells, was associated with the secretion of osteopontin and with poor prognosis (36).

MOLECULAR PATHWAYS INVOLVED IN CAF FUNCTION

Multiple biological differences have been reported between normal fibroblasts and CAFs. CAFs exhibit higher motility and growth rate and show a stronger potential in the promotion of malignancy of cancer cells. Moreover, miRNAs are differentially expressed in CAFs and normal fibroblasts (37).

Since CAFs are characterized by a marked heterogeneity, with different CAF subtypes observed in different tumors, even the molecular pathways

involved in CAFs are different in different cancers (38). In colorectal cancer, CAFs genes related to the Wnt signaling pathway and the TGF-beta pathway are highly enriched. TGF-beta is generally considered a regulator of CAFs, whereas Wnt2 is involved in cancer growth, lymph node metastasis, venous invasion, and recurrence (39). Wtn5, a component of the beta-catenin-independent pathway, is highly expressed in CAFs. Colorectal cancers with high CAF expression of Wnt5 are characterized by aggressiveness and poor prognosis (40).

Many miRNAs are overexpressed in CAFs in the TME of colorectal cancer. These tumor-associated miRNAs may influence cancer cell spread, invasiveness, metastasis and chemioresistance (41). In colorectal cancer, CAFs may exert their role by transferring exosomes into the TME towards cancer cells, leading to increased levels of miR-92-3p in tumor cells. The release of miRNAs-rich exosomes is responsible for the activation of the Wnt/Beta-catenin pathway, enhancing the tumor cell stemness and epithelial-to-mesenchymal-transition of cancer cells (42). The TGF-beta pathway is highly activated in CAFs, TGF-beta being considered a regulator of CAF activity (39).

Multiple microRNAs, able to regulate the tumor-promoting properties of CAFs, may be aberrantly expressed in different cancers. miR-101-3p and miR-490-3p are downregulated in CAFs surrounding HCC. Since TGF-beta is a common target for both miRNAs, their decreased expression is associated with an increased expression of TGF-beta in tumor cells, associated with poor clinical outcomes. Regarding the possible causes of these two miRNAs downregulation, it has been hypothesized that infiltration in the TME of a huge amount of immunosuppressive immune cells might negatively modulate the expression of multiple miRNAs in CAFs, influencing the oncogenic signature of HCC (43).

In the TME of colorectal cancer, non-coding RNAs were identified in exosomes, probably representing the way utilized by tumor cells for exchanging information with CAFs and for crosstalk with other cell types of the TME (44). According with this study, exosome-derived non-coding RNAs might regulate the properties of the peritumoral stroma in function of tumor pregression, influencing angiogenesis, vascular permeability, tumor immunity, tumor cell metabolism and drug resistance. On the other hand, CAFs might exert their influence on cancer cells by exosome secretion. In colorectal cancer, CAFs have been shown to secrete and transfer exosomes to tumor cells, leading to increased levels of miR-92a-3p, able to activate the Wnt/Beta-catenin pathway, promoting epithelial mesenchymal transition of tumor cells and their metastatic abilities (42). According with this hypothesis, the inhibition of the exosome-related transfer of miR-92a-3p from CAFs to cancer cells might provide an intriguing modality for the prevention of metastases in colorectal cancer patients.

In recent years, a complex regulatory network has been clarified in the TME, modulating the formation, activation, phenotype, and heterogeneity of CAFs. In this CAF regulatory TME network, the following signaling pathways have been identified: TGF-beta, NOTCH, Hippo, Hedgehog, Nuclear factor kappa-beta (NF-k-beta), Janus kinase (JAK), signal transducer and activator of transcription (STAT), mitogen-activated protein kinase (MAPK), phosphoinositide 3 kinase (PI3K), hypoxia-inducible factor (HIF), heat shock transcription factor 1 (HSF1) and p53 (45).

CAFs may also exert regulatory effects on tumor cell metabolism, promoting metabolic reprogramming, dysregulating multiple molecular pathways, and inducing the expression molecules, such as CD44 in cancer cells that may influence significantly tumor progression and the insurgence of metastases (46, 47). Metabolic reprogramming mainly regards glucose, amino acid, and lipid metabolism (48).

CAF HETEROGENEITY: THE MULTIPLE PHENOTYPES OF PERITUMORAL FIBROBLASTS

Within the TME, CAFs play multiple functional roles in the initiation and progression of cancer, including the production of growth factors, signaling molecules, multiple cytokines, and exosomes, mediating crosstalk between stromal cells and cancer cells (49). CAFs are a diverse cell type with a significant amount of variability in both appearance and function. These cells play a critical role in aiding cancer cells to evade the immune system (50). CAFs are made up of multiple subtypes, each with unique characteristics and functions, and activated through different molecular signaling pathways. The following CAF subtypes have been identified: i) inflammatory CAFs, characterized by low expression of alpha-SMA and high levels of IL-6; ii) TGF-beta-dependent myofibroblastic CAFs, char-

acterized by high levels of alpha-SMA expression; iii) CAFs that suppress tumor cell growth, characterized by activation of the stroma-specific Hedgehog (Hh) (51).

Distinct subclasses of CAFs have been identified in a genetically engineered mouse model of breast cancer, with single-cell RNA sequencing. In these studies, the following CAF subclasses attributable to different origins were identified: i) CAFs originating from the peri-vascular niche; ii) CAFs originating from the mammary fat pad; iii) CAFs originating from the transformed epithelium (52).

A classification scheme of CAFs with different functional roles in the TME was defined by identifying nice CAF phenotypes through single-cell RNA sequencing of over 16,000 stromal cells from breast cancer (53). Single-cell RNA sequencing has identified three major CAF subpopulations in human non-small cell lung cancer. These subpopulations include alveolar, adventitial, and myofibroblasts. Alveolar and adventitial fibroblasts are associated with improved overall survival rates in lung adenocarcinoma, whereas myofibroblasts are associated with poor overall survival rates in lung adenocarcinoma. In contrast, the presence of myofibroblasts in lung squamous cell carcinomas did not show any correlation with prognosis. This confirms that there is a significant variability in the composition of the TME and the role of CAFs, even among tumors that develop in the same organ (54). In bladder urothelial cancer, single-cell sequencing allowed the identification of a role for inflammatory CAFs in tumor progression, which is related to poor prognosis (55).

The proposal of a classification of single cells applied to the TME, the scATOMIC, represented a new tool for a deeper understanding of the TME and for accurately subset stromal and immune cells in the TME of different cancers (56).

These data taken together clearly show that the TME of every cancer type is characterized by peculiar features, and stromal and immune cells of the different microenvironments may play different roles in cancer progression.

CAF heterogeneity is, at least in part, due to epigenetic changes occurring during their maturation, including DNA methylation and histone modifications. Loss of CAF histone methylation and gain of CAF histone acethylation has been associated to CAF activation and accelerated tumor growth. The epigenetic regulation of CAF activation and heterogeneity is mainly related to TGF-beta and to the bromodomain and extraterminal (BET) domain, an epigenetic reader that recognizes histone acetylation, activates CAF gene transcription and leads to the acquisition of a pro-tumor phenotype in CAFs. Moreover, miRNAs play a key role in the epigenetic modifications that influence gene expression in CAFs, functioning as targets and orchestrators of gene transcription in CAFs (57).

The heterogeneity and the multifaceted nature and function of CAFs has been recently confirmed in HCC, a still worrisome disease. CAFs have been dissected into four genetically distinct subsets, characterized by distinct functions in the TME of this aggressive cancer type (58). In this study, CAFs have been confirmed to represent the most prevalent cell type populating the TME in HCC, being characterized as fibroblasts in a persistent state of activation.

IMMUNOHISTOCHEMICAL MARKERS OF CAFs

A crucial point in identifying CAFs in the TME is the dependence of utilized markers on the cancer type being studied. The role of CAFs and the TME in cancer development and progression poses a significant challenge for surgical pathologists. Pathologists must identify the specific CAF markers for each cancer type rather than relying on general immunohistochemical markers. This approach is crucial in clinical practice and would help in the accurate diagnosis and treatment of cancer patients. Moreover, the panels utilized should be able to discern the various subtypes of CAFs, including protumoral, anti-tumor, proinflammatory and other CAF subtypes with different roles in carcinogenesis (59).

A panel of 12 markers has been suggested to identify CAFs in HCC, in addition to alpha-smooth muscle actin: ADAM18, ADAM32, CXCL5, FGF4, FGF5, FGF8, FGF17, FGF19, FGF23, IGFL1, IGFL2, and MMP1 (60). Vimentin and alpha-SMA are overexpressed in cervical CAFs than in normal fibroblasts from uterine leyomyomas.

Wnt2 is a marker that indicates the presence of activated CAFs. In colorectal cancer, the expression of Wnt2 in CAFs is significantly associated with various factors such as the depth of the tumor, TNM stage, vascular invasion, lymph-node metastasis, and recurrence of cancer after chemotherapy or surgery (39). Wnt5, a component of the

beta-catenin independent pathway, may be highly expressed in CAFs of colorectal cancer. A positive staining for Wnt5 in the stromal cells of colorectal cancer, particularly in CAFs, is associated with high tumor cell proliferation and migration, as well as poor prognosis (40).

A subset of CAFs, like some normal fibroblasts, may express the glial fibrillary acid protein (GFAP), a typical marker of glial cells, indicating their possible origin from the neural crest (61).

The following immunohistochemical markers have been utilized in a study aimed to characterize CAFs in the peritumoral stroma of HCC (62):

1. alpha-smooth mucle actin (alpha-SMA)
2. Desmin
3. Vimentin
4. Fibroblast activation protein (FAP)
5. Fibroblast specific protein-1 (FSP-1)

In a recent study carried out on CAFs in pancreatic cancer, the following CAF subtypes were identified, on the basis of their immunohistochemical marker expression:

1. inflammatory CAFs (iCAF), characterized by low expression of alpha-SMA and strong expression of IL-6;
2. myofibroblastic CAFs (myCAF), with strong expression of alpha-SMA and low levels of IL-6 expression
3. antigen-presenting CAFs (apCAFs) characterized by the expression of CD74;
4. complement-secreting CAFs (csCAFs) expressing C3 and C7;
5. CAFs with high metabolic state (meCAFs) with no certain specific marker known (63).

Identifying CAF subpopulations through the expression of different immunohistochemial markers represents a challenge for pathologists involved in research as well for surgical pathologists involved in clinical practice. Distinct CAF subgroups often play different roles, even opposite, just like water and fire, in carcinogenesis, so that the identification of the CAF subpopulation involved in a specific cancer will represent a breakthrough in oncology (64).

CAF FUNCTIONS IN TUMOR INSURGENCE AND PROGRESSION

CAFs play multiple, and often opposite, roles in carcinogenesis, by contributing to the deposition and remodeling of the extracellular matrix, cross-talk with other cells of the TME and with cancer cells, to the epithelial-to-mesenchymal transition of tumor cells, and influencing invasion, metastasis and therapy resistance of cancer (65).

Once activated, CAFs can secrete multiple messengers such as the multifunctional cytokine TGF-beta that, if deregulated, can promote an aggressive phenotype in cancer (66). Communication through CAF-derived exosomes is crucial in intercellular communication in the TME (67). In particular, exosomes have been shown to play a key role in crosstalk between CAFs and cancer cells (68). TGF-beta in CAF-derived exosomes promotes epithelial-to mesenchymal transition in cancer cells (69).

Moreover, CAF-secreted exosomes may promote matastasis, chemotherapy resistance and may enhance cancer cell stemness (70).

The miR-17-5p contained in exosomes derived by CAFs promotes an aggressive phenotupe in colorectal cancer by initiating a RUNX3/MYC/TGF-beta1 positive feedback loop (71). The production of Hepatocyte growth factor (HGF) by CAFs has been associated with the promotion of invasion and metastasis (72). CAF-derived Wnt signaling acivity in the TME has been shown to define cancer cell stemness in colorectal carcinoma (73).

CAFs show an abundant and stable expression of the FAP-alfa, a type II integral serine protease that plays a key role in promoting tumor growth, immunosuppression, invasion, and metastasis (74). CAFs produce the stromal cell-derived factor-1 (SDF-1), a CXC chemokine that promotes cancer cell migration and tumor growth, due to angiogenesis-dependent induction of tumor cell proliferation and inhibition of apoptotic cell death (75). CAFs are involved in promoting invasion and metastasis through the stimulation of CXCL12/CXCR4 signaling. High CXCL12 expression is associated with larger tumor size, lymphatic invasion, and poor prognosis in gastric cancer (76).

Interestingly, AMD3100 (plerixafor), a bicyclam molecule that selectively antagonizes the binding of stromal cell-derived factor-1 (SDF-1) to its receptor CXCR4, approved for the mobilization of hematopoietic stem/progenitor cells (77) might be taken into consideration as an antagonist of the promotion of invasion and metastasis exerted by CAFs through the stimulation of CXCL12/CXCR4 signaling. Moreover, CAF-derived CXCL12 enhances the immune escape of tumor cells in

bladder cancer, through the inhibition of degradation of PDL1 (78).

The activities of CAFs and tumor cells of any specific cancer type should be considered always bidirectional, with reciprocal influences. In an in vitro system, HCT-116 human colorectal cancer cells were able to induce the transformation of normal fibroblasts to CAFs. Compared to normal fibroblasts, CAFs secreted high levels of transforming growth factor-beta (TGF-beta), tumor necrosis factor-alpha (TNF-alpha), C-X-C motif chemokine ligand-2 (CXCL2), interleukin-1 beta (IL-1beta) and SDF-1 (79). In this experiment, CAFs also expressed higher levels than normal fibroblasts of fibroblast-activated protein and alpha-SMA. Colorectal cancer cell-activated CAFs stimulated tumor cell migration, which was weakened by AMD3100, an inhibitor of SDF-1, underlying the major role of CAF-derived SDF-1 in promoting metastatic behavior in colorectal cancer cells.

All these data taken together evidence that CAFs play an essential role in carcinogenesis, influencing cancer cell proliferation, stemness, invasion, migration, epithelial-to-mesenchymal transition, angiogenesis, chemoresistance, and immunosuppression, by releasing multiple chemokines and regulatory molecules in the microenvironment of tumors (80).

Since CAFs represent the most common component of the TME, CAFs play a pivotal role in cancer insurgence and progression, in tumor immunity, in the therapeutic response, and in the insurgence of drug resistance (3). It is to be noted that the TME is characterized by a marked variability among different cancer types, and the activity of CAFs may change from one tumor to another. An in-depth understanding of the crosstalk between FAFs and cancer cells in different tumor types might guide the development of new CAF-based anticancer strategies and represent a breakthrough in oncology (81).

CAFs AS TUMOR PROMOTERS

In several cancer types, CAFs have gained much attention as a pivotal component of the TME. The mechanisms by which CAFs facilitate cancer insurgence and progression are multiple and, probably, differ from one cancer type to the next. Here are the better-known mechanisms of the pro-tumoral activity of CAFs:

1. **promotion of cancer angiogenesis**. CAFs produce galectin-1, a glycan-binding protein with high angiogenic potential, which is positively associated with CD31 and vascular endothelial growth factor (VEGF) expression (82). Since cancer aggressivity and progression are highly dependent on neo-angiogenesis, CAFs appear as a possible therapeutic target in cancer (83).
2. **Induction of apoptosis in cancer cells**. CAFs have been shown to induce apoptosis of cancer cells in different cancers, regulating the invasion mode of tumors. In gastric cancer, CAFs induce apoptosis in tumor cells, through activation of caspase-8 and death receptor 4, preventing expansive invasion and facilitating CAF-led invasion. Adequate scattered cancer cell apoptosis induced by CAFs facilitates the mobilization of the adjacent cancer cells, favoring their detachment from the tumor mass and their migration away from the tumor (84).

   A CAF-induction of apoptosis of cancer cells, able to fuel oncogenic processes, has been reported in pancreatic cancer (85).
3. **Induction of epithelial-to-mesenchymal transition in cancer cells**. In breast cancer, CAFs characterized by high expression of alpha-smooth muscle actin and SDF1/CXCL12, promoted epithelial-to-mesenchymal transition through paracrine TGF-beta signaling. In particular, CAFs secreted more TGF-beta 1 than TGF-beta 2 and TGF-beta 3 and activated the TGF-beta/SMAD-signaling pathway in cancer cells (86).

   In endometrial cancer, CAFs have been shown to induce epithelial-mesenchymal transition through the secretion of multiple cytokines, including E-cadherin, N-cadherin epidermal growth factor, TGF-beta, hepatic growth factor and fibroblast growth factor (87). In bladder cancer, CAFs induce epithelial-to-mesenchymal transition through paracrine IL-6 signaling (88).

   In laryngeal squamous cell carcinoma, CAF-derived extracellular vesicles enhance the epithelial-to-mesenchymal transition of tumor cells by delivering TUC338 that upregulates CBX2 through sponging miR-8485 (89).

   Recent studies have confirmed that CAFs play a key role in the epithelial-to-mesenchymal transition process. This is because CAFs produce various matrix proteins and growth factors. Additionally, a 3-gene CAF transcriptional signature (COL1A1, COL1A2, COL3A1) has been associated with the expression of markers related to

epithelial-to-mesenchymal transition and poor prognosis (90).

The promotion of a motile phenotype in cancer cells allows them to migrate towards blood and lymphatic vessels. CAFs promote directional cancer cell migration by assembling fibronectin, producing a fibronectin-rich extracellular matrix that guides tumor cells to migrate directionally (91). In non-small cell lung cancer, CAFs promote cancer cell migration and invasion through the miR-101-3p-mediated secretion of VEGFA and activation of the AKT/eNOS pathway (92).

4. **Reshaping of the extracellular matrix of the TME, favoring the migration of motile cancer cells**.
5. **Promotion of cancer metastasis**.
6. **CAFs within the TME can promote resistance to multiple cancer therapies**, including chemotherapy, anti-angiogenesis, radiotherapy, and immunotherapy (93).
7. **Development of an immunosuppressive microenvironment in the peritumoral stroma**. It is important to note that only certain subtypes of CAFs promote tumor growth, while others may inhibit it (9).

CAFs AS TUMOR SUPPRESSORS

Recent studies have shown that CAFs not only play a major role in promoting cancer growth but also have the potential to act as tumor suppressors. In HCC, at least one population of CAFs synthesizes prolargin in the peritumoral stroma. Higher levels of prolargin are positively correlated with improved patient outcomes (94). In the same study, CAF-produced prolargin was shown to inhibit the activity of several pro-angiogenic proteins, of the hepatocyte growth factor (HGF) and fibroblast growth factor (FGF), resulting in a tumor-suppressor activity. CAFs probably represent a very plastic cell population, which may play opposite functions in cancer progression, by secreting a wide repertoire of factors that regulate cancer progression. Turning foes to friends is the goal of many novel anticancer therapies, targeting CAFs (95).

There has been a recent report on the complex relationship between CAFs and cancer stem cells (CSCs). In the case of mammary gland tumors, CSCs have been found to regulate the function of CAFs through paracrine activation of Hedgehog signaling pathway by secreting the hedgehog ligand SHH. As a result of this activation, CAFs secrete factors that promote self-renewal and expansion of CSCs. It is interesting to note that treatment with Hedgehog inhibitors reduced the expansion of CSCs, leading to a delay in tumor progression overall (96).

More recent studies carried out in pancreatic cancer, evidenced that Hedgehogh signaling is activated in CAFs, particularly in myofibroblastic CAFs. These studies confirmed the ability of Hedgehog inhibitors to reduce myCAF number in the TME, and increase the inflammatory CAFs, ending with a decrease in cytotoxic T cells and an expansion of regulatory T cells, consistent with immunosuppression (97).

CAFs AS POSSIBLE NEW ANTI-TUMOR TARGETS

Deciphering the cellular and molecular composition of the TME, as well as the intercellular interactions and crosstalks between tumor cells, stromal and immune cells is a prerequisite for a modern therapeutic approachs to cancer, targeting reprogramming of CAFs (70). Moreover, defining CAFs in the TME represents a fascinating opportunity in cancer immunotherapy and a breackthrough in clinical trials targeted to halting CAFs' protumor multiple activities (9). In short, oving to their robust function as tumor-promoters, CAFs represent, at the best of our knowledge, one of the most promising targets for oncotherapy in the near future. Recent knowledge of the multiple factors produced by CAFs and of the multiple molecular signaling pathways activated by CAFs in the TME, is at the basis of the development of novel anti-cancer strategies.

Targeted therapies for CAFs can be classified into three mechanisms. The first one involves the direct removal of CAFs by using specific molecules capable of recognizing their biomarkers, immunotherapies, and oncolytic viruses. The second mechanism is the inhibition of CAFs signaling. This objective can be achieved by targeting the extracellular matrix of the TME leading to depletion of peritumoral tumor stroma. Finally, the third mechanism is the reprogramming of CAFs.

Research is currently focusing on finding specific and exploitable biomarkers that can be used for the creation of target molecules to directly inhibit CAFs. These biomarkers differ depending on the histotype being considered. For breast cancer,

the biomarkers include CD29, FAP, FSP1, α-SMA, PDGFRb and CAV1, while for pancreatic cancer, the biomarkers are α-SMA, FAP, and PDGFRa (98, 99). For colorectal cancer, the biomarkers include FAP, MMP2, DCN, α-SMA, PDGFA, and TAGLN (100, 101). For gastric cancer, the biomarkers include IL6, CXCL12, MMP14, LOXL2, and POSTN, while for lung cancer, they include HGF, FGF7, and p-SMAD2 (102) (**Figure 1**).

FAP is one of the most expressed biomarkers in different types of tumors, and if present, it relates to a poor prognosis. However, the use of anti-FAP monoclonal antibodies (mAb), such as FAP5-DM, has shown promising results in inhibiting tumor growth and even achieving complete remission in some cases, as demonstrated in various xenograft models of cancer (103). The immunotoxin αFAP-PE38 was tested on an animal model of breast cancer. The test showed that the compound was effective in blocking tumor growth (104).

Clinical trials have been conducted on some drugs that target FAP in patients with metastatic colon cancer. Two of the drugs are Sibrotuzumab, which is a humanized monoclonal antibody that targets FAP, and Talabostat. However, these drugs were not able to progress to phase II trials (105, 106).

One approach in cancer treatment involves FAP-based DNA vaccines, which can stimulate the growth of lymphocytes that infiltrate the tumor and break down CAFs. By doing so, these vaccines can disrupt immune tolerance towards the tumor (107). Studies have shown that anti-FAP vaccines can effectively inhibit tumor growth both in vitro and in vivo during preclinical trials. However, DNA vaccines can currently only be tested on animals, and clinical trials have not yet been conducted. FAP is a key target for CAR-T cell-based therapy. A recent study has developed a radiolabeled FAP inhibitor that can detect and intercept tumor and stromal cells expressing FAP in the TME. This "probe" can help predict and monitor the efficacy of CAR-T therapies targeting FAP (108).

It has been identified that inhibiting the tumor-promoting CAF signal can be an effective therapeutic strategy. The CXCL12-CXCR4 axis is a crucial component of CAF-mediated tumorigenesis. To this end, a CXCR4 antagonist named AMD3100 has been developed. This antagonist can improve lymphocyte infiltration, reducing desmoplasia, and immunosuppression (109).

Another strategy is represented by the combination of CXCR4 antagonist (AMD3100) or BL-8040

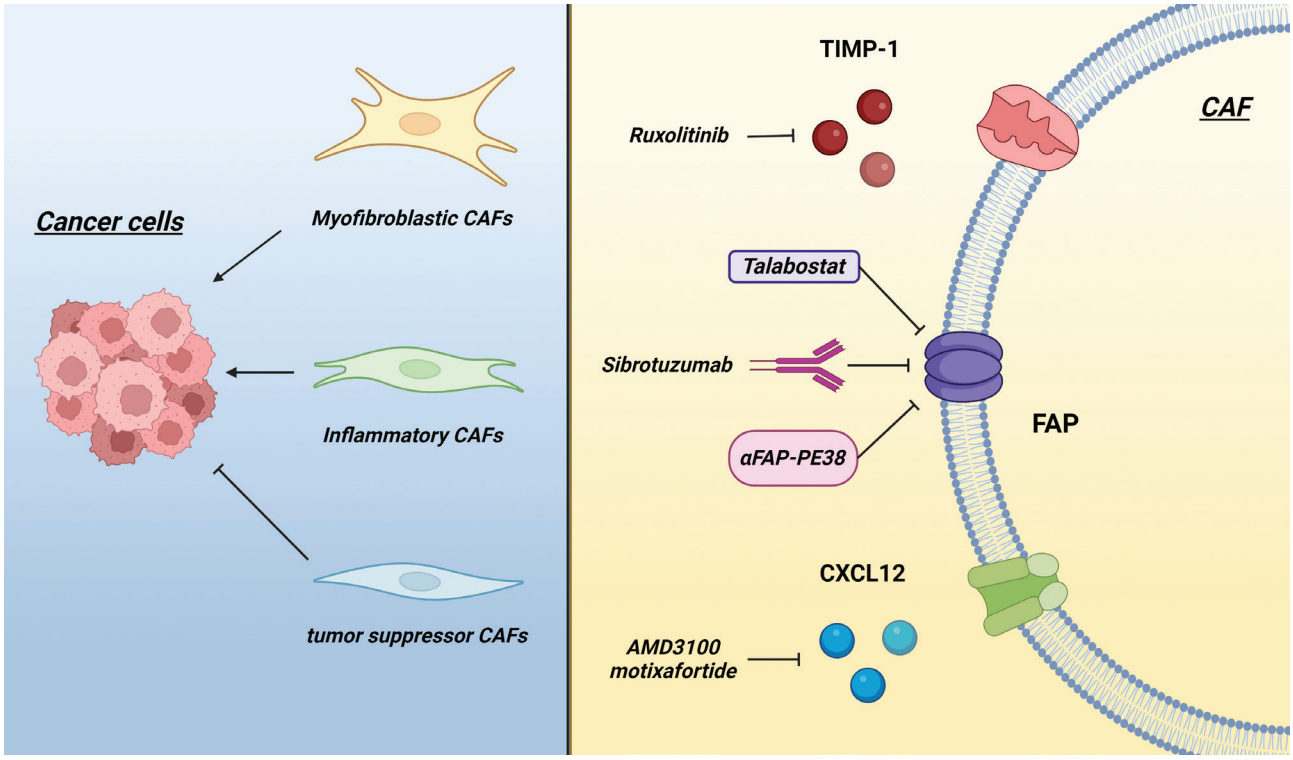

Figure 1. *Main CAFs subtypes and their therapeutic targets. Interaction of tumor cells with the main CAF subgroups: myofibroblastic and inflammatory CAFs (promoting tumor growth); tumor suppressor CAFs. FAP represents one of the main therapeutic targets (FAP inhibitors: talabostat, sibrotuzumab and αFAP-PE38). TIMP-1 (tissue inhibitor of metallo-proteinases1) and its inhibitor (ruxolitinib). CXCL12 (C-X-C motif chemokine 12) and its inhibitors (AMD3100 and motixafortide).*

(motifortide) with other antineoplastic agents, a therapy capable of slowing down the growth of PDAC, through the inhibition of CAFs (110, 111). In the Phase II study, the combination of motifafortide, pembrolizumab, and chemotherapy demonstrated efficacy in the PDAC patient population. The triplet resulted in a median progression-free survival (PFS) of 3.8 months and a median overall survival (OS) of 6.6 months (112).

A recent study investigated a combination therapy that involved using CXCR4 antagonist nanoparticles along with miR-210/KRASG12D blockade. This combination was found to be effective in breaking down the matrix components of tumors, resulting in reduced immunosuppression and slowed tumor growth. The study suggested that nanoparticles could be used as carriers for delivering CAF modulators to enhance their effectiveness (113).

The STAT3 signaling pathway plays a crucial role as it is activated by CAFs that secrete TIMP-1, leading to the hyperactivation of the pathway (114-115). A recent study found that the coactivation of the MEK and STAT3 pathways is an important mediator of neoplastic growth. To explore this further, researchers used a combination of MEK inhibitor (trametinib) and STAT3 inhibitor (ruxolitinib) in a mouse model. The results showed that the use of MEK inhibitor and STAT3 inhibitor together helped to reduce the expression of proinflammatory IL-6/CXCL1 and LRRC15-expressing myCAFs, while also enriching Ly6a/CD34-expressing CAFs that exhibit mesenchymal stem cell-like features (116). TGF-β plays a significant role in CAF activation and function. Blocking TGF-β directly, downstream signaling and receptor antagonism can reverse activated CAFs (117-119).

The "stromal switch" is a new therapeutic strategy aimed to change tumor-promoting CAFs, such as TGF-dependent myofibroblastic CAFs, into tumor-retarding fibroblasts, like Hedgehog-activating CAFs (51). One of the most important aims of this new anticancer therapy is the block of the CAFs' ability to promote actomyosin contractility in tumor cells and the remodeling of the extracellular matrix of the TME, to form the tracks utilized by cancer cells for collective migration towards blood and lymphatic vessels.

One of the goals of this approach is to change the tumor-promoting CAF phenotype, typical of a CAF subgroup with pro-invasive abilities (120) towards a tumor-suppressive one or to revert activated CAFs to a quiescent state. Therapies targeting IL-1 and TGF-β can transform CAFs. Some studies have shown that vitamin A and analogues can convert CAFs into a quiescent state. In the case of pancreatic cancer, restoring retinol levels may help neoplastic cells shift towards a quiescent phenotype. (121).

A combination of all-trans retinoic acid (ATRA) and gemcitabine has been discovered to delay the progression of pancreatic ductal adenocarcinoma (PDAC) in mouse models. This combination works by intercalating in a series of enzymatic cascades such as Wnt, Hedgehog, retinoid, and FGF (122). Furthermore, a phase Ib study conducted in pa-

Table 1. *Preclinical and clinical trials including CAFs targeting.*

STUDY	DRUG	TARGET	HISTOTYPE	
Phase II	BL-8040 (motifortide)	CXCR4	Pancreatic cancer	Bockorny B, 2020 (111)
Phase II	Talabostat	FAP	Metastatic colorectal cancer	Narra K, 2007 (106)
Phase II	Sibrotuzumab	FAP	Metastatic colorectal cancer	Hofheinz RD, 2003 (105)
Preclinical	Ruxolitinib	STAT3	Pancreatic cancer cells	Datta J, 2022 (116)
Preclinical	αFAP-PE38	FAP	Breast cancer cells	Fang J, 2016 (104)
Preclinical	AMD3100	CXCL12	Pancreatic cancer cells	Feig C, 2013 (110)
Preclinical	FAP5-DM	Serin-protease	Malignant epithelial cells	Ostermann E, 2008 (103)

tients with advanced PDAC indicated that the administration of ATRA with gemcitabine and nabp-aclitaxel is safe and well-tolerated (123) (**Table 1**).

CONCLUSIONS

Modern oncology aims at developing therapeutic strategies that target not only the tumor cells but also the cells and elements present in the TME. The most effective therapies should be combination therapies, which target not just the tumor cells but also the stromal cells, including CAFs, that play a crucial role in cancer progression, metastasis, and resistance to anticancer therapy (124). In recent years, there has been growing interest in CAFs due to their intricate interactions with cancer cells. CAFs have been identified as potential targets for the development of new therapies against aggressive tumors that involve higher levels of extracellular matrix and fibrosis, such as pancreatic cancer and HCC (124). At present, the studies related to cancer treatment are mainly in the preclinical phase and in the clinical phases 0 and 1. These studies are showing promising results in inhibiting tumor growth. However, we need to wait for the subsequent phases of clinical studies to obtain further developments, particularly in the case of more aggressive tumors.

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Ethical approval

Human studies and subjects

N/A.

Animal studies

N/A.

Authors' contributions

All the Authors listed in the manuscript have contributed to the conception and planning of this work, through the acquisition and interpretation of data here reported. All the Authors partecipated in the drafting of the manuscript and in the definitive approval of the version to be published.

Data sharing and data accessibility

Data are available upon motivated request to the Corresponding Author.

Publication ethics

Plagiarism

Authors declare no potentially overlapping publications with the content of this manuscript and all original studies are cited as appropriate.

Data falsification and fabrication

All the data correspond to the real.

REFERENCES

1. Akhtar M, Haider A, Rashid S, Al-Nabet ADMH. Paget's "Seed and Soil" theory fo cancer metastasis: an idea whose time has come. Adv Anat Patol. 2019;26(1):69-74. doi: 10.1097/PAP.0000000000000219.
2. Fidler IJ, Poste G. The "Seed and Soil" hypothesis revisited. Lancet Oncol. 2008;9(8):808. doi.org/10.1016/S1470-2045(08)70201-8.
3. Cheng B, Yu Q, Wang W. Intimate comunications within the tumor microenvironment: stromal factors function as an orchestra. J Biomed Sci. 2023;30(1):1. doi: 10.1186/s12929-022-00894-z.
4. Okuyama T, Sameshima S, Takeshita E, Mitsui T, Noro T, Ono Y, et al. Myxoid stroma is associated with postoperative relapse in patients with stage II colon cancer. BMC Cancer. 2020;20(1):842. doi: 10.1186/s12885-020-07335-w.
5. Ueno H, Shinto E, Shimazaki H, Kajiwara Y, Sueyama T, Yamamoto J, et al. Histologic categorization of desmoplastic reaction: its relevance to the colorectal cancer microenvironment and prognosis. Ann Surg Oncol. 2015;22(5):1504-12. doi: 10.1245/s10434-014-4149-9.
6. Hemandez-Ruiz E, Hemandez-Munoz I, Masferrer E, Ferrandiz-Pulido C, Andrades E, Gimeno J, et al. A myxoid fibrotic reaction pattern is associated with metastatic risk in cutaneous squamous cell carcinoma. Acta Derm Venereol. 2019;99(1):89-94.

7. Cau F, Gerosa C, Ziranu P, Pretta A, Piras M, Fraschini M, et al. L1CAM expression in the peritumoral stromal cells of the microenvironment of colorectal cancer: a new target for oncologists? Annal Res Oncol. 2023;3(4):147-54. doi: 10.48286/aro.2023.76.
8. Karagiannis GS, Poutahidis T, Erdman SE, Kirsch R, Riddell RH, Diamandis EP. Cancer-associated fibroblasts drive the progression of metastasis through both paracrine and mechanical pressure on cancer tissue. Mol Cancer Res. 2012;10(11):1403-18. doi: 10.1158/1541-7786.MCR-12-0307.
9. Zhang H. Define cancer-associated fibroblasts in the tumor microenvironment: new opportunities in cancer immunotherapy and advances in clinical trials. Mol Cancer. 2023;22(1)159. doi: 10.1186/s12943-023-01860-5.
10. Phan QM, Driskell RR. Defining the epigenetic regulation of fibroblast lineages during embryonic development. J Invest Dermatol. 2022;139:1171-81.
11. Chen G, Bracamonte-Baran W, Diny NL, Hou X, Talor MV, Fu K, et al. Sca-1+ cardiac fibroblasts promote development of heart failure. Eur J Immunol. 2018;48(9):1522-38. doi: 10.1002/eji.201847583.
12. Plikus MV, Wang X, Sinha S, Forte E, Thompson SM, Herzog EL, et al. Fiubroblasts: origins, definitions and functions in health and disease. Cell. 2021;184(15):3852-72. doi: 10.1016/j.cell.2021.06.024.
13. Junker JP. Cell Tissue Organs. 2010;191(2):105-18. doi: 10.1159/000232157.
14. Knoedler S, Broichhausen S, Guo R, Dai R, Knoedler L, Kauke-Navarro M, et al. Fibroblasts: the cellular choreographers of wound healing. Front Immunol. 2023;14: 1233800. doi.org/10.3389/fimmu.2023.1233800.
15. Ferrer RA, Saalbach A, Grünwedel M, Lohmann N, Forstreuter I, Saupe S, et al. Dermal Fibroblasts Promote Alternative Macrophage Activation Improving Impaired Wound Healing. J Invest Dermatol. 2017;137(4):941-50. doi: 10.1016/j.jid.2016.11.035.
16. Tillie RJHA, van Kuijk K, Sluimer JC. Fibroblasts in atherosclerosis: heterogeneous and plastic participants. Curr Opin Lipidol. 2020;31(5):273-8. doi: 10.1097/MOL.0000000000000700.
17. Xiao L, Wang Q, Peng H. Tumor-associated macrophages: new insights on their metabolic regulation and their influence in cancer immunotherapy. Front Immunol. 2023;14:1157291. doi: 10.3389/fimmu.2023.1157291.
18. Cassetta L, Pollard JW. A timeline of tumour-associated macrophage biology. Nat Rev Cancer. 2023;23(4):238-57. doi: 10.1038/s41568-022-00547-1.
19. Yuan S, Almagro J, Fuchs E. Beyond genetics: driving cancer with the tumour microenvironment behind the wheel. Nat Rev Cancer. 2024;24(4):274-86. doi: 10.1038/s41568-023-00660-9.
20. Zhao D, Mo Y, Neganova ME, Aleksandrova Y, Tse E, Chubarev VN, et al. Dual effects of radiotherapy on tumor microenvironment and its contribution towards the development of resistance to immunotherapy in gastrointestinal and thoracic cancers. Front Cell Dev Biol. 2023;11:1266537. doi: 10.3389/fcell.2023.1266537.
21. Raskov H, Orhan A, Gaggar S, Gögenur I. Cancer-Associated Fibroblasts and Tumor-Associated Macrophages in Cancer and Cancer Immunotherapy. Front Oncol. 2021;11:668731. doi: 10.3389/fonc.2021.668731.
22. Kovaleva O, Sorokin M, Egorova A, Petrenko A, Shelekhova K, Gratchev A. Macrophage - tumor cell interaction beyond cytokines. Front Oncol. 2023;13:1078029. doi: 10.3389/fonc.2023.1078029.
23. Zhou J, Tang Z, Gao S, Li C, Feng Y, Zhou X. Tumor-Associated Macrophages: Recent Insights and Therapies. Front Oncol. 2020;10:188. doi: 10.3389/fonc.2020.00188.
24. Wieder R. Fibroblasts as Turned Agents in Cancer Progression. Cancers. 2023;15(7):2014. doi: 10.3390/cancers15072014.
25. Ying F, Chan MSM, Lee TKW. Cancer-Associated Fibroblasts in Hepatocellular Carcinoma and Cholangiocarcinoma. Cell Mol Gastroenterol Hepatol. 2023;15(4):985-99. doi: 10.1016/j.jcmgh.2023.01.006.
26. Bucala R, Spiegel LA, Chesney J, Hogan M, Cerami A. Circulating fibrocytes define a new leukocyte subpopulation that mediates tissue repair. Mol Med. 1994;1(1):71-81.
27. Kobayashi H, Gieniec KA, Lannagan TRM, Wang T, Asai N, Mizutani Y, et al. The Origin and Contribution of Cancer-Associated Fibroblasts in Colorectal Carcinogenesis. Gastroenterology. 2022;162(3):890-906. doi: 10.1053/j.gastro.2021.11.037.
28. Zhang J, Gu C, Song Q, Zhu M, Xu Y, Xiao M, et al. Identifying cancer-associated fibroblasts as

emerging targets for hepatocellular carcinoma. Cell Biosci. 2020;10(1):127. doi: 10.1186/s13578-020-00488-y.
29. Jia W, Liang S, Cheng B, Ling C. The Role of Cancer-Associated Fibroblasts in Hepatocellular Carcinoma and the Value of Traditional Chinese Medicine Treatment. Front Oncol. 2021;11:763519. doi: 10.3389/fonc.2021.763519.
30. Kubo N, Araki K, Kuwano H, Shirabe K. Cancer-associated fibroblasts in hepatocellular carcinoma. World J Gastroenterol. 2016;22(30):6841-50. doi: 10.3748/wjg.v22.i30.6841.
31. Choi KJ, Nam JK, Kim JH, Choi SH, Lee YJ. Endothelial-to-mesenchymal transition in anticancer therapy and normal tissue damage. Exp Mol Med. 2020;52(5):781-92. doi: 10.1038/s12276-020-0439-4.
32. Halperin C, Hey J, Weichenhan D, Stein Y, Mayer S, Lutsik P, et al. Global DNA Methylation Analysis of Cancer-Associated Fibroblasts Reveals Extensive Epigenetic Rewiring Linked with RUNX1 Upregulation in Breast Cancer Stroma. Cancer Res. 2022;82(22):4139-52. doi: 10.1158/0008-5472.CAN-22-0209.
33. Rynne-Vidal A, Jiménez-Heffernan JA, Fernández-Chacón C, López-Cabrera M, Sandoval P. The Mesothelial Origin of Carcinoma Associated-Fibroblasts in Peritoneal Metastasis. Cancers. 2015;7(4):1994-2011. doi: 10.3390/cancers7040872.
34. Manoukian P, Bijlsma M, van Laarhoven H. The Cellular Origins of Cancer-Associated Fibroblasts and Their Opposing Contributions to Pancreatic Cancer Growth. Front Cell Dev Biol. 2021;9:743907. doi: 10.3389/fcell.2021.743907.
35. Huang J, Tsang WY, Li ZH, Guan XY. The Origin, Differentiation, and Functions of Cancer-Associated Fibroblasts in Gastrointestinal Cancer. Cell Mol Gastroenterol Hepatol. 2023;16(4):503-511. doi: 10.1016/j.jcmgh.2023.07.001.
36. Muchlińska A, Nagel A, Popęda M, Szade J, Niemira M, Zieliński J, et al. Alpha-smooth muscle actin-positive cancer-associated fibroblasts secreting osteopontin promote growth of luminal breast cancer. Cell Mol Biol Lett. 2022;27(1):45. doi: 10.1186/s11658-022-00351-7.
37. Hu D, Zhuo W, Gong P, Ji F, Zhang X, Chen Y, et al. Biological differences between normal and cancer-associated fibroblasts in breast cancer. Heliyon. 2023;9(9):e19803. doi: 10.1016/j.heliyon.2023.e19803.
38. Louault K, Li RR, DeClerck YA. Cancer-Associated Fibroblasts: Understanding Their Heterogeneity. Cancers. 2020;12(11):3108. doi: 10.3390/cancers12113108.
39. Aizawa T, Karasawa H, Funayama R, Shirota M, Suzuki T, Maeda S, et al. Cancer-associated fibroblasts secrete Wnt2 to promote cancer progression in colorectal cancer. Cancer Med. 2019;8(14):6370-82. doi: 10.1002/cam4.2523.
40. Hirashima T, Karasawa H, Aizawa T, Suzuki T, Yamamura A, Suzuki H, et al. Wnt5a in cancer-associated fibroblasts promotes colorectal cancer progression. Biochem Biophys Res Commun. 2021;568:37-42. doi: 10.1016/j.bbrc.2021.06.062.
41. Savardashtaki A, Shabaninejad Z, Movahedpour A, Sahebnasagh R, Mirzaei H, Hamblin MR. miRNAs derived from cancer-associated fibroblasts in colorectal cancer. Epigenomics. 2019;11(14):1627-45. doi: 10.2217/epi-2019-0110.
42. Hu JL, Wang W, Lan XL, Zeng ZC, Liang YS, Yan YR, et al. CAFs secreted exosomes promote metastasis and chemotherapy resistance by enhancing cell stemness and epithelial-mesenchymal transition in colorectal cancer. Mol Cancer. 2019;18(1):91. doi: 10.1186/s12943-019-1019-x.
43. Eun JW, Ahn HR, Baek GO, Yoon MG, Son JA, Weon JH, et al. Aberrantly Expressed MicroRNAs in Cancer-Associated Fibroblasts and Their Target Oncogenic Signatures in Hepatocellular Carcinoma. Int J Mol Sci. 2023;24(5):4272. doi: 10.3390/ijms24054272.
44. Chen X, Jia M, Ji J, Zhao Z, Zhao Y. Exosome-Derived Non-Coding RNAs in the Tumor Microenvironment of Colorectal Cancer: Possible Functions, Mechanisms and Clinical Applications. Front Oncol. 2022;12:887532. doi: 10.3389/fonc.2022.887532.
45. Fang Z, Meng Q, Xu J, Wang W, Zhang B, Liu J, et al. Signaling pathways in cancer-associated fibroblasts: recent advances and future perspectives. Cancer Commun. 2023;43(1):3-41. doi: 10.1002/cac2.12392.
46. Aimola V, Fanni D, Gerosa C, Cerrone G, Ziranu P, Pretta A, et al. Balance between the stem cell marker CD44 and CDX2 expression in colorectal cancer. Ann Res Oncol. 2022;2(2):160-6. doi: 10.48286/aro.2022.43
47. Ziranu P, Aimola V, Pretta A, Dubois M, Murru R, Liscia N, et al. New Horizons in Metastatic Colorectal Cancer: Prognostic Role of CD44

Expression. Cancers. 2023;15(4):1212. doi: 10.3390/cancers15041212.
48. Zhu Y, Li X, Wang L, Hong X, Yang J. Metabolic reprogramming and crosstalk of cancer-related fibroblasts and immune cells in the tumor microenvironment. Front Endocrinol. 2022;13:988295. doi: 10.3389/fendo.2022.988295.
49. Huang J, Tsang WY, Li ZH, Guan XY. The Origin, Differentiation, and Functions of Cancer-Associated Fibroblasts in Gastrointestinal Cancer. Cell Mol Gastroenterol Hepatol. 2023;16(4):503-11. doi: 10.1016/j.jcmgh.2023.07.001.
50. Akkız H. Emerging Role of Cancer-Associated Fibroblasts in Progression and Treatment of Hepatocellular Carcinoma. Int J Mol Sci. 2023;24(4):3941. doi: 10.3390/ijms24043941.
51. Yoshida GJ. Regulation of heterogeneous cancer-associated fibroblasts: the molecular pathology of activated signaling pathways. J Exp Clin Cancer Res. 2020;39(1):112. doi: 10.1186/s13046-020-01611-0.
52. Bartoschek M, Oskolkov N, Bocci M, Lövrot J, Larsson C, Sommarin M, et al. Spatially and functionally distinct subclasses of breast cancer-associated fibroblasts revealed by single cell RNA sequencing. Nat Commun. 2018;9(1):5150. doi: 10.1038/s41467-018-07582-3.
53. Cords L, Tietscher S, Anzeneder T, Langwieder C, Rees M, de Souza N, et al. Cancer-associated fibroblast classification in single-cell and spatial proteomics data. Nat Commun. 2023;14(1):4294. doi: 10.1038/s41467-023-39762-1.
54. Hanley CJ, Waise S, Ellis MJ, Lopez MA, Pun WY, Taylor J, et al. Single-cell analysis reveals prognostic fibroblast subpopulations linked to molecular and immunological subtypes of lung cancer. Nat Commun. 2023;14(1):387. doi: 10.1038/s41467-023-35832-6.
55. Chen Z, Zhou L, Liu L, Hou Y, Xiong M, Yang Y, et al. Single-cell RNA sequencing highlights the role of inflammatory cancer-associated fibroblasts in bladder urothelial carcinoma. Nat Commun. 2020;11(1):5077. doi: 10.1038/s41467-020-18916-5.
56. Nofech-Mozes I, Soave D, Awadalla P, Abelson S. Pan-cancer classification of single cells in the tumour microenvironment. Nat Commun. 2023;14(1):1615. doi: 10.1038/s41467-023-37353-8.
57. Kehrberg RJ, Bhyravbhatla N, Batra SK, Kumar S. Epigenetic regulation of cancer-associated fibroblast heterogeneity. Biochim Biophys Acta Rev Cancer. 2023;1878(3):188901. doi: 10.1016/j.bbcan.2023.188901.
58. Cadamuro M, Nuozzi G, Simioni P, Fabris L. The tumor microenvironment in hepatocellular carcinoma: dissecting the functions of cancer-associated fibroblasts. Hepatoma Res. 2023;9:47. doi: 10.20517/2394-5079.2023.94.
59. Nurmik M, Ullmann P, Rodriguez F, Haan S, Letellier E. In search of definitions: Cancer-associated fibroblasts and their markers. Int J Cancer. 2020;146(4):895-905. doi: 10.1002/ijc.32193.
60. Zou B, Liu X, Gong Y, Cai C, Li P, Xing S, et al. A novel 12-marker panel of cancer-associated fibroblasts involved in progression of hepatocellular carcinoma. Cancer Manag Res. 2018;10:5303-5311. doi: 10.2147/CMAR.S176152.
61. Hainfellner JA, Voigtländer T, Ströbel T, Mazal PR, Maddalena AS, Aguzzi A, et al. Fibroblasts can express glial fibrillary acidic protein (GFAP) in vivo. J Neuropathol Exp Neurol. 2001;60(5):449-61. doi: 10.1093/jnen/60.5.449.
62. Yin Z, Dong C, Jiang K, Xu Z, Li R, Guo K, et al. Heterogeneity of cancer-associated fibroblasts and roles in the progression, prognosis, and therapy of hepatocellular carcinoma. J Hematol Oncol. 2019;12(1):101. doi: 10.1186/s13045-019-0782-x.
63. Li X, Zhou J, Wang X, Li C, Ma Z, Wan Q, et al. Pancreatic cancer and fibrosis: Targeting metabolic reprogramming and crosstalk of cancer-associated fibroblasts in the tumor microenvironment. Front Immunol. 2023 Mar 22;14:1152312. doi: 10.3389/fimmu.2023.1152312.
64. Han C, Liu T, Yin R. Biomarkers for cancer-associated fibroblasts. Biomark Res. 2020;8(1):64. doi: 10.1186/s40364-020-00245-w.
65. Asif PJ, Longobardi C, Hahne M, Medema JP. The Role of Cancer-Associated Fibroblasts in Cancer Invasion and Metastasis. Cancers. 2021;13(18):4720. doi: 10.3390/cancers13184720.
66. Linares J, Marín-Jiménez JA, Badia-Ramentol J, Calon A. Determinants and Functions of CAFs Secretome During Cancer Progression and Therapy. Front Cell Dev Biol. 2021;8:621070. doi: 10.3389/fcell.2020.621070.
67. Dai J, Su Y, Zhong S, Cong L, Liu B, Yang J, et al. Exosomes: key players in cancer and potential therapeutic strategy. Signal Transduct Target Ther. 2020;5(1):145. doi: 10.1038/s41392-020-00261-0.

68. Yang X, Li Y, Zou L, Zhu Z. Role of Exosomes in Crosstalk Between Cancer-Associated Fibroblasts and Cancer Cells. Front Oncol. 2019;9:356. doi: 10.3389/fonc.2019.00356.
69. Li W, Zhang X, Wang J, Li M, Cao C, Tan J, et al. TGFβ1 in fibroblasts-derived exosomes promotes epithelial-mesenchymal transition of ovarian cancer cells. Oncotarget. 2017;8(56):96035-47. doi: 10.18632/oncotarget.21635.
70. Hu JL, Wang W, Lan XL, Zeng ZC, Liang YS, Yan YR, et al. CAFs secreted exosomes promote metastasis and chemotherapy resistance by enhancing cell stemness and epithelial-mesenchymal transition in colorectal cancer. Mol Cancer. 2019;18(1):91. doi: 10.1186/s12943-019-1019-x.
71. Zhang Y, Wang S, Lai Q, Fang Y, Wu C, Liu Y, et al. Cancer-associated fibroblasts-derived exosomal miR-17-5p promotes colorectal cancer aggressive phenotype by initiating a RUNX3/MYC/TGF-β1 positive feedback loop. Cancer Lett. 2020;491:22-35. doi: 10.1016/j.canlet.2020.07.023.
72. Ding X, Ji J, Jiang J, Cai Q, Wang C, Shi M, et al. HGF-mediated crosstalk between cancer-associated fibroblasts and MET-unamplified gastric cancer cells activates coordinated tumorigenesis and metastasis. Cell Death Dis. 2018;9(9):867. doi: 10.1038/s41419-018-0922-1.
73. Vermeulen L, De Sousa E Melo F, van der Heijden M, Cameron K, de Jong JH, Borovski T, et al. Wnt activity defines colon cancer stem cells and is regulated by the microenvironment. Nat Cell Biol. 2010;12(5):468-76. doi: 10.1038/ncb2048.
74. Xin L, Gao J, Zheng Z, Chen Y, Lv S, Zhao Z, et al. Fibroblast Activation Protein-α as a Target in the Bench-to-Bedside Diagnosis and Treatment of Tumors: A Narrative Review. Front Oncol. 2021;11:648187. doi: 10.3389/fonc.2021.648187.
75. Kollmar O, Rupertus K, Scheuer C, Junker B, Tilton B, Schilling MK, et al. Stromal cell-derived factor-1 promotes cell migration and tumor growth of colorectal metastasis. Neoplasia. 2007;9(10):862-70. doi: 10.1593/neo.07559.
76. Izumi D, Ishimoto T, Miyake K, Sugihara H, Eto K, Sawayama H, et al. CXCL12/CXCR4 activation by cancer-associated fibroblasts promotes integrin β1 clustering and invasiveness in gastric cancer. Int J Cancer. 2016;138(5):1207-19. doi: 10.1002/ijc.29864.
77. Pusic I, DiPersio JF. Update on clinical experience with AMD3100, an SDF-1/CXCL12-CXCR4 inhibitor, in mobilization of hematopoietic stem and progenitor cells. Curr Opin Hematol. 2010;17(4):319-26. doi: 10.1097/MOH.0b013e328338b7d5.
78. Zhang Z, Yu Y, Zhang Z, Li D, Liang Z, Wang L, et al. Cancer-associated fibroblasts-derived CXCL12 enhances immune escape of bladder cancer through inhibiting P62-mediated autophagic degradation of PDL1. J Exp Clin Cancer Res. 2023;42(1):316. doi: 10.1186/s13046-023-02900-0.
79. Li L, Zhai Y, Wang Y, Yang L. [Human colorectal cancer cells induce fibroblasts to secrete stromal cell-derived factor 1 (SDF-1) to stimulate cancer cell migration]. Xi Bao Yu Fen Zi Mian Yi Xue Za Zhi. 2021;37(9):821-7. Chinese. PMID: 34533130.
80. Sun H, Wang X, Wang X, Xu M, Sheng W. The role of cancer-associated fibroblasts in tumorigenesis of gastric cancer. Cell Death Dis. 2022;13(10):874. doi: 10.1038/s41419-022-05320-8.
81. Zhao Z, Li T, Sun L, Yuan Y, Zhu Y. Potential mechanisms of cancer-associated fibroblasts in therapeutic resistance. Biomed Pharmacother. 2023;166:115425. doi: 10.1016/j.biopha.2023.115425.
82. Tang D, Gao J, Wang S, Ye N, Chong Y, Huang Y, et al. Cancer-associated fibroblasts promote angiogenesis in gastric cancer through galectin-1 expression. Tumour Biol. 2016;37(2):1889-99. doi: 10.1007/s13277-015-3942-9.
83. Wang FT, Sun W, Zhang JT, Fan YZ. Cancer-associated fibroblast regulation of tumor neo-angiogenesis as a therapeutic target in cancer. Oncol Lett. 2019;17(3):3055-65. doi: 10.3892/ol.2019.9973.
84. Itoh G, Chida S, Yanagihara K, Yashiro M, Aiba N, Tanaka M. Cancer-associated fibroblasts induce cancer cell apoptosis that regulates invasion mode of tumours. Oncogene. 2017;36(31):4434-44. doi: 10.1038/onc.2017.49.
85. Pfeifer E, Burchell JM, Dazzi F, Sarker D, Beatson R. Apoptosis in the Pancreatic Cancer Tumor Microenvironment-The Double-Edged Sword of Cancer-Associated Fibroblasts. Cells. 2021;10(7):1653. doi: 10.3390/cells10071653.
86. Yu Y, Xiao CH, Tan LD, Wang QS, Li XQ, Feng YM. Cancer-associated fibroblasts induce epithelial-mesenchymal transition of breast cancer cells

through paracrine TGF-β signalling. Br J Cancer. 2014;110(3):724-32. doi: 10.1038/bjc.2013.768.
87. Wang X, Zhang W, Sun X, Lin Y, Chen W. Cancer-associated fibroblasts induce epithelial-mesenchymal transition through secreted cytokines in endometrial cancer cells. Oncol Lett. 2018;15(4):5694-702. doi: 10.3892/ol.2018.8000.
88. Goulet CR, Champagne A, Bernard G, Vandal D, Chabaud S, Pouliot F, et al. Cancer-associated fibroblasts induce epithelial-mesenchymal transition of bladder cancer cells through paracrine IL-6 signalling. BMC Cancer. 2019;19(1):137. doi: 10.1186/s12885-019-5353-6.
89. Li T, Tian L, Cao J, Liu M. Cancer-associated fibroblasts secret extracellular vesicles to support cell proliferation and epithelial-mesenchymal transition in laryngeal squamous cell carcinoma. Mol Cell Probes. 2023;72:101934. doi: 10.1016/j.mcp.2023.101934.
90. Szabo PM, Vajdi A, Kumar N, Tolstorukov MY, Chen BJ, Edwards R, et al. Cancer-associated fibroblasts are the main contributors to epithelial-to-mesenchymal signatures in the tumor microenvironment. Sci Rep. 2023;13(1):3051. doi: 10.1038/s41598-023-28480-9.
91. Erdogan B, Ao M, White LM, Means AL, Brewer BM, Yang L, et al. Cancer-associated fibroblasts promote directional cancer cell migration by aligning fibronectin. J Cell Biol. 2017;216(11):3799-816. doi: 10.1083/jcb.201704053.
92. Guo X, Chen M, Cao L, Hu Y, Li X, Zhang Q, et al. Cancer-Associated Fibroblasts Promote Migration and Invasion of Non-Small Cell Lung Cancer Cells via miR-101-3p Mediated VEGFA Secretion and AKT/eNOS Pathway. Front Cell Dev Biol. 2021;9:764151. doi: 10.3389/fcell.2021.764151.
93. Feng B, Wu J, Shen B, Jiang F, Feng J. Cancer-associated fibroblasts and resistance to anticancer therapies: status, mechanisms, and countermeasures. Cancer Cell Int. 2022;22(1):166. doi: 10.1186/s12935-022-02599-7.
94. Chiavarina B, Ronca R, Otaka Y, Sutton RB, Rezzola S, Yokobori T, et al. Fibroblast-derived prolargin is a tumor suppressor in hepatocellular carcinoma. Oncogene. 2022;41(10):1410-20. doi: 10.1038/s41388-021-02171-z.
95. Chen X, Song E. Turning foes to friends: targeting cancer-associated fibroblasts. Nat Rev Drug Discov. 2019;18(2):99-115. doi: 10.1038/s41573-018-0004-1.
96. Valenti G, Quinn HM, Heynen GJJE, Lan L, Holland JD, Vogel R, et al. Cancer Stem Cells Regulate Cancer-Associated Fibroblasts via Activation of Hedgehog Signaling in Mammary Gland Tumors. Cancer Res. 2017;77(8):2134-47. doi: 10.1158/0008-5472.CAN-15-3490.
97. Steele NG, Biffi G, Kemp SB, Zhang Y, Drouillard D, Syu L, et al. Inhibition of Hedgehog Signaling Alters Fibroblast Composition in Pancreatic Cancer. Clin Cancer Res. 2021;27(7):2023-37. doi: 10.1158/1078-0432.CCR-20-3715.
98. Öhlund D, Handly-Santana A, Biffi G, Elyada E, Almeida AS, Ponz-Sarvise M, et al. Distinct populations of inflammatory fibroblasts and myofibroblasts in pancreatic cancer. J Exp Med. 2017;214(3):579-96. doi: 10.1084/jem.20162024.
99. Elyada E, Bolisetty M, Laise P, Flynn WF, Courtois ET, Burkhart RA, et al. Cross-Species Single-Cell Analysis of Pancreatic Ductal Adenocarcinoma Reveals Antigen-Presenting Cancer-Associated Fibroblasts. Cancer Discov. 2019;9(8):1102-23. doi: 10.1158/2159-8290.CD-19-0094.
100. Zhang L, Li Z, Skrzypczynska KM, Fang Q, Zhang W, O'Brien SA, et al. Single-Cell Analyses Inform Mechanisms of Myeloid-Targeted Therapies in Colon Cancer. Cell. 2020;181(2):442-59.e29. doi: 10.1016/j.cell.2020.03.048.
101. Li H, Courtois ET, Sengupta D, Tan Y, Chen KH, Goh JJL, et al. Reference component analysis of single-cell transcriptomes elucidates cellular heterogeneity in human colorectal tumors. Nat Genet. 2017;49(5):708-18. doi: 10.1038/ng.3818. Erratum in: Nat Genet. 2018;50(12):1754. Erratum in: Nat Genet. 2023;55(1):166.
102. Hu H, Piotrowska Z, Hare PJ, Chen H, Mulvey HE, Mayfield A, et al. Three subtypes of lung cancer fibroblasts define distinct therapeutic paradigms. Cancer Cell. 2021;39(11):1531-47.e10. doi: 10.1016/j.ccell.2021.09.003.
103. Ostermann E, Garin-Chesa P, Heider KH, Kalat M, Lamche H, Puri C, et al. Effective immunoconjugate therapy in cancer models targeting a serine protease of tumor fibroblasts. Clin Cancer Res. 2008;14(14):4584-92. doi: 10.1158/1078-0432.CCR-07-5211.
104. Fang J, Xiao L, Joo KI, Liu Y, Zhang C, Liu S, et al. A potent immunotoxin targeting fibroblast activation protein for treatment of breast cancer in mice. Int J Cancer. 2016;138(4):1013-23. doi: 10.1002/ijc.29831.
105. Hofheinz RD, al-Batran SE, Hartmann F, Hartung G, Jäger D, Renner C, et al. Stromal antigen tar-

geting by a humanised monoclonal antibody: an early phase II trial of sibrotuzumab in patients with metastatic colorectal cancer. Onkologie. 2003;26(1):44-8. doi: 10.1159/000069863.
106. Narra K, Mullins SR, Lee HO, Strzemkowski-Brun B, Magalong K, Christiansen VJ, et al. Phase II trial of single agent Val-boroPro (Talabostat) inhibiting Fibroblast Activation Protein in patients with metastatic colorectal cancer. Cancer Biol Ther. 2007;6(11):1691-9. doi: 10.4161/cbt.6.11.4874.
107. Duperret EK, Trautz A, Ammons D, Perales-Puchalt A, Wise MC, Yan J, et al. Alteration of the Tumor Stroma Using a Consensus DNA Vaccine Targeting Fibroblast Activation Protein (FAP) Synergizes with Antitumor Vaccine Therapy in Mice. Clin Cancer Res. 2018;24(5):1190-201. doi: 10.1158/1078-0432.CCR-17-2033.
108. Lee IK, Noguera-Ortega E, Xiao Z, Todd L, Scholler J, Song D, et al. Monitoring Therapeutic Response to Anti-FAP CAR T Cells Using [18F] AlF-FAPI-74. Clin Cancer Res. 2022;28(24):5330-5342. doi: 10.1158/1078-0432.CCR-22-1379.
109. Timperi E, Gueguen P, Molgora M, Magagna I, Kieffer Y, Lopez-Lastra S, et al. Lipid-Associated Macrophages Are Induced by Cancer-Associated Fibroblasts and Mediate Immune Suppression in Breast Cancer. Cancer Res. 2022;82(18):3291-306. doi: 10.1158/0008-5472.CAN-22-1427.
110. Feig C, Jones JO, Kraman M, Wells RJ, Deonarine A, Chan DS, et al. Targeting CXCL12 from FAP-expressing carcinoma-associated fibroblasts synergizes with anti-PD-L1 immunotherapy in pancreatic cancer. Proc Natl Acad Sci USA. 2013;110(50):20212-7. doi: 10.1073/pnas.1320318110.
111. Bockorny B, Semenisty V, Macarulla T, Borazanci E, Wolpin BM, Stemmer SM, et al. BL-8040, a CXCR4 antagonist, in combination with pembrolizumab and chemotherapy for pancreatic cancer: the COMBAT trial. Nat Med. 2020;26(6):878-85. doi: 10.1038/s41591-020-0880-x.
112. Bockorny B, Macarulla T, Semenisty V, Borazanci E, Feliu J, Ponz-Sarvise M, et al. Motixafortide and Pembrolizumab Combined to Nanoliposomal Irinotecan, Fluorouracil, and Folinic Acid in Metastatic Pancreatic Cancer: The COMBAT/KEYNOTE-202 Trial. Clin Cancer Res. 2021;27(18):5020-7. doi: 10.1158/1078-0432.CCR-21-0929.
113. Xie Y, Hang Y, Wang Y, Sleightholm R, Prajapati DR, Bader J, et al. Stromal Modulation and Treatment of Metastatic Pancreatic Cancer with Local Intraperitoneal Triple miRNA/siRNA Nanotherapy. ACS Nano. 2020;14(1):255-71. doi: 10.1021/acsnano.9b03978.
114. Guo Z, Zhang H, Fu Y, Kuang J, Zhao B, Zhang L, et al. Cancer-associated fibroblasts induce growth and radioresistance of breast cancer cells through paracrine IL-6. Cell Death Discov. 2023;9(1):6. doi: 10.1038/s41420-023-01306-3.
115. Dittmer A, Dittmer J. A CAF-Fueled TIMP-1/CD63/ITGB1/STAT3 Feedback Loop Promotes Migration and Growth of Breast Cancer Cells. Cancers (Basel). 2022;14(20):4983. doi: 10.3390/cancers14204983.
116. Datta J, Dai X, Bianchi A, De Castro Silva I, Mehra S, Garrido VT, et al. Combined MEK and STAT3 Inhibition Uncovers Stromal Plasticity by Enriching for Cancer-Associated Fibroblasts With Mesenchymal Stem Cell-Like Features to Overcome Immunotherapy Resistance in Pancreatic Cancer. Gastroenterology. 2022;163(6):1593-612. doi: 10.1053/j.gastro.2022.07.076.
117. Pei Y, Chen L, Huang Y, Wang J, Feng J, Xu M, et al. Sequential Targeting TGF-β Signaling and KRAS Mutation Increases Therapeutic Efficacy in Pancreatic Cancer. Small. 2019;15(24):e1900631. doi: 10.1002/smll.201900631.
118. Lambies G, Miceli M, Martínez-Guillamon C, Olivera-Salguero R, Peña R, Frías CP, et al. TGFβ-Activated USP27X Deubiquitinase Regulates Cell Migration and Chemoresistance via Stabilization of Snail1. Cancer Res. 2019;79(1):33-46. doi: 10.1158/0008-5472.CAN-18-0753.
119. Huang H, Zhang Y, Gallegos V, Sorrelle N, Zaid MM, Toombs J, et al. Targeting TGFβR2-mutant tumors exposes vulnerabilities to stromal TGFβ blockade in pancreatic cancer. EMBO Mol Med. 2019;11(11):e10515. doi: 10.15252/emmm.201910515.
120. Li X, Sun Z, Peng G, Xiao Y, Guo J, Wu B, et al. Single-cell RNA sequencing reveals a pro-invasive cancer-associated fibroblast subgroup associated with poor clinical outcomes in patients with gastric cancer. Theranostics. 2022;12(2):620-38. doi: 10.7150/thno.60540.
121. Froeling FE, Feig C, Chelala C, Dobson R, Mein CE, Tuveson DA, et al. Retinoic acid-induced pancreatic stellate cell quiescence reduces paracrine Wnt-β-catenin signaling to slow tumor progression. Gastroenterol-

ogy. 2011;141(4):1486-97, 1497.e1-14. doi: 10.1053/j.gastro.2011.06.047.

122. Carapuça EF, Gemenetzidis E, Feig C, Bapiro TE, Williams MD, Wilson AS, et al. Anti-stromal treatment together with chemotherapy targets multiple signalling pathways in pancreatic adenocarcinoma. J Pathol. 2016;239(3):286-96. doi: 10.1002/path.4727.

123. Kocher HM, Basu B, Froeling FEM, Sarker D, Slater S, Carlin D, et al. Phase I clinical trial repurposing all-trans retinoic acid as a stromal targeting agent for pancreatic cancer. Nat Commun. 2020;11(1):4841. doi: 10.1038/s41467-020-18636-w.

124. Dzobo K, Dandara C. Architecture of Cancer-Associated Fibroblasts in Tumor Microenvironment: Mapping Their Origins, Heterogeneity, and Role in Cancer Therapy Resistance. OMICS. 2020;24(6):314-39. doi: 10.1089/omi.2020.0023.