

Pancreatic Ductal Adenocarcinoma: Characteristics of Tumor Microenvironment and Barriers to Treatment

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Abstract

Pancreatic ductal adenocarcinoma remains a highly aggressive disease, with a 5-year relative survival rate of 10%. Numerous barriers to treatment exist, such as dense desmoplasia, infiltration of immune suppressor cells, inhibitory cytokines, low effector T-cell infiltration, and low tumor mutational burden. These factors help form a highly suppressive tumor microenvironment unique to pancreatic ductal adenocarcinoma. This review outlines barriers to treatment of pancreatic ductal adenocarcinoma by discussing the unique characteristics of the pancreatic tumor microenvironment and the factors that contribute to making pancreatic ductal adenocarcinoma such a challenging disease to treat.

Pancreatic ductal adenocarcinoma (PDAC) is a highly aggressive disease. According to the SEER database of the National Cancer Institute, it had a 5-year relative survival rate of 10% from 2010 to 2016. The prognosis of PDAC remains dismal, with a disappointing response to cytotoxic chemoradiotherapies as well as to newer treatments such as immunotherapy. Among the numerous factors contributing to PDAC lack of response to treatment is its unique immune-suppressive micro-

environment that results in evasion from the host antitumor immune system leading to rapid progression (Murakami et al., 2019). The unique tumor microenvironment (TME) of PDAC consists of a dense fibrotic stroma comprising several different proteins, cells, growth factors, and enzymes. As the understanding of this tumor microenvironment evolves, it is becoming apparent that there is a highly complex interplay between stromal signals, the immune system, and tumor cells. At times, this interplay restrains tumor

growth and at others supports growth and metastasis (Seager, Hajal, Spill, Kamm, & Zaman, 2017; Young, Hughes, Cunningham, & Starling, 2018). The pancreatic microenvironment supports progression of disease by facilitating proliferation of cancer cells, inhibition of antitumor immunity, and induction of immunosuppressive cell proliferation and metastasis (Ren et al., 2018).

This review will improve the advanced practitioner's understanding of the factors contributing to the resistance of pancreatic ductal adenocarcinoma to treatment with an emphasis on the tumor microenvironment. Advanced practitioners will comprehend the science behind evolving therapies and the design of several clinical trials targeting barriers to the treatment of PDAC. Figure 1 shows a graphical view of the tumor microenvironment that fuels barriers to treatment.

DENSE DESMOPLASIA

In recent years, research in the treatment of PDAC has focused on the stroma in pancreatic adenocarcinoma (Rasheed, Matsui, & Maitra, 2012). Desmoplasia in PDAC involves the formation of a dense fibrotic stroma comprising extracellular matrix proteins, myofibroblastic pancreatic stellate cells, immune cells, cytokines, growth factors, and extracellular matrix metabolizing enzymes (Feig, 2012; Pandol, Edderkaoui, Gukovsky, Lugea, & Gukovskaya, 2009; Uzunparmak & Sahin, 2019). The hallmark dense stroma in PDAC is associated with poor clinical outcomes with resistance to radiation, chemotherapy, as well as immunotherapy (Rasheed et al., 2012; Tan & El-Rayes, 2018). The structural organization of the stroma is not entirely different from those in other solid tumors; however, in contrast to several other solid tumors,

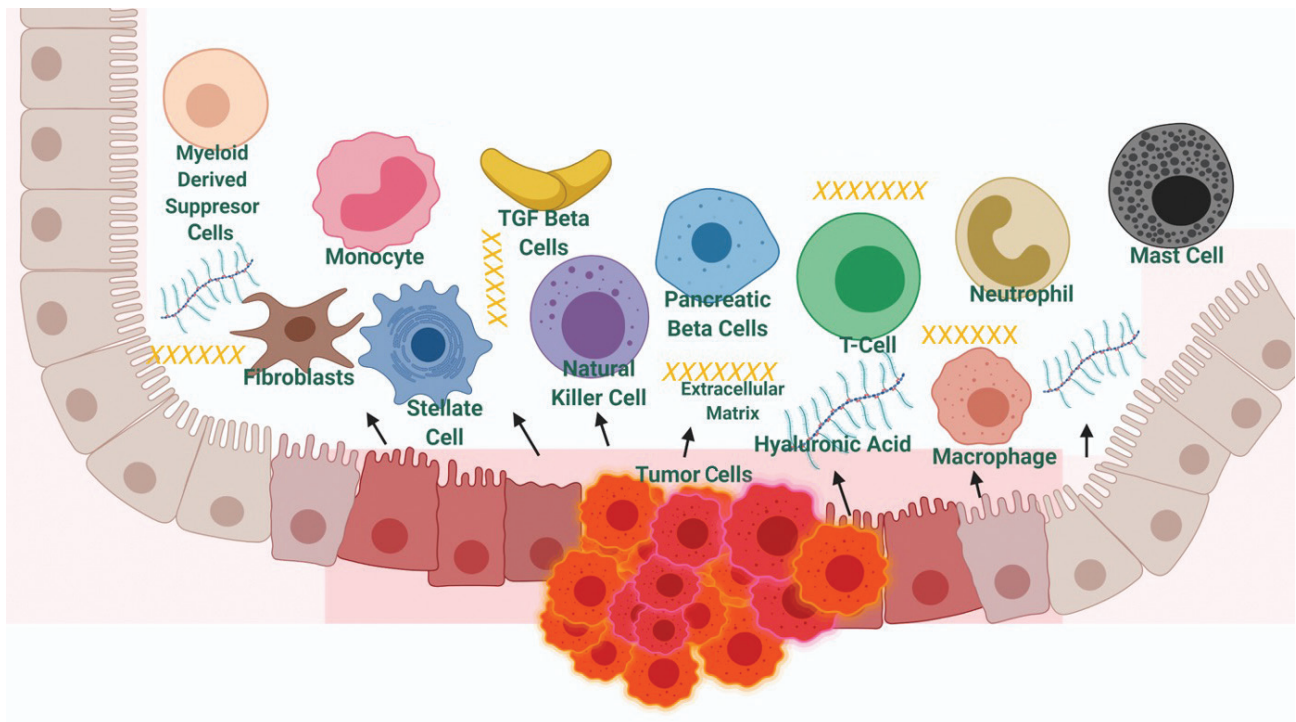


Figure 1. Pancreatic ductal adenocarcinoma barriers to treatment. The defining key players in the microenvironment of pancreatic ductal adenocarcinoma play a major role in creating barriers to treatment and igniting progression. The stroma cells are made of myeloid-derived suppressor cells, cancer fibroblasts cells, tumor macrophages, and stellate cells. Stellate cells can activate extracellular matrix and collagens necessary for stroma elements to fuel desmoplasia. Desmoplasia is the process that promotes progression, invasion, and metastasis. Desmoplasia impairs drug delivery and can cause chemotherapy resistance which prohibits treatment response. Immune cell response is inhibited in this environment, mainly due to increase of regulatory T cells known to suppress the immune cells and immunosuppressive cytokines such as transforming growth factor- β . Other cells that promote healthy immune response, such as monocytes, macrophages, neutrophils, mast cells, and natural killer cells, are not able to function properly, which confirms the need for targeted therapies designed to identify ways to activate immune cells.

stromal elements can occupy more than 80% of the total tumor volume in most pancreatic cancer cases, thereby creating a tough physical barrier (Kanat & Ertas, 2018).

This thick barrier is created by PDAC cells releasing various factors that stimulate stromal growth. The stroma then releases substances that stimulate tumor growth, metastatic spread, and drug resistance (Rasheed et al., 2012). One of these PDAC cell types is the pancreatic stellate cell (PSC). These cells play a central role by expressing growth factors and producing various stromal elements such as collagen, laminin, fibronectin, and hyaluronic acid (HA); this process is called desmoplasia (Uzunparmak & Sahin, 2019). Increased production of collagen prevents T-cell migration toward the cancer cells as a result of being trapped by a highly adhesive collagen web. The production of HA impairs the delivery of chemotherapies and immunotherapies by increasing interstitial fluid pressure and causing narrowing of the blood vessels. Therefore, desmoplasia produces a hypovascular microenvironment that impairs local drug delivery and renders tumors resistant to chemotherapies and immunotherapies (Uzunparmak & Sahin, 2019). Additionally, PSCs generate another type of cellular barrier called cancer-associated fibroblasts (CAFs). These stromal fibroblasts secrete cytokines such as interleukin-6 (IL-6) and attract IL-17-secreting CD4-positive cells, contributing to the immunosuppressive nature of the microenvironment.

INHIBITORY CYTOKINES

Within the tumor microenvironment, cytokine dynamics play an important role in potentiating the immunosuppressive milieu through modulating both inflammation and the immune response. One such proinflammatory cytokine, IL-6, is known to be elevated in patients with a diagnosis of PDAC and can contribute to *in vitro* tumor cell invasion by facilitating the proliferation of regulatory T cells (Tregs) and myeloid-derived suppressor cells (MDSCs; Razidlo, Burton, McNiven, 2018; Shadhu & Xi, 2019). Additionally, IL-6 can contribute to the transition of PSCs from a state of quiescence to activity, which in turn helps to perpetuate a proinflammatory TME (Öhlund et al., 2017; Shadhu & Xi, 2019).

Another prominent cytokine in the PDAC TME, IL-10, has been frequently associated with tumorigenesis and the ability of cancer cells to shield themselves from immunosurveillance (Batchu et al., 2018). The correlation between elevated levels of circulating IL-10 and poor patient prognosis across a multitude of cancers has lent credence to this assertion (Zhang, Wang, & He, 2016). Higher levels of IL-10 are associated with a plethora of suppressive immune cell populations, including dendritic cells, MDSCs, and Tregs (Zhang et al., 2016).

The MDSC immune suppressive effects are a result of the hyperproduction of inhibitory cytokines that decrease MDSC capacity to fully differentiate into macrophages, dendritic cells, and neutrophils (Khaled, Ammori, & Elkord, 2014). T cells, which are already present or make their way into the TME, also may not be able to mount a full immune response to the tumor cells, as they are hindered by the secretion of immunosuppressive cytokines such as IL-10 (Young et al., 2018).

INFILTRATION OF IMMUNE SUPPRESSOR CELLS

Pancreatic ductal adenocarcinomas present an extensive immune infiltrate that is essentially composed of immune cells that (under normal conditions) the body uses to reduce the immune response. These cells include MDSCs, Tregs, and macrophages. The active suppression of the immune system at the site of the tumor is a common event in the tumor development, beginning at a very early disease developmental stage. In fact, the presence of myeloid cells is associated with a worse prognosis in patients with resected disease, as are Tregs (Young et al., 2018). While MDSC levels do not always reveal a definitive association with tumor stages in pancreatic cancer patients, some studies have shown a positive correlation between MDSCs and PDAC progression (Thyagarajan et al., 2019).

The abnormal differentiation and abnormal function of myeloid cells are common in tumors. Myeloid-derived suppressor cells, prominent in the tumor microenvironment, have potent immunosuppressive activity and are immature and pathologically activated (Kumar, Patel, Tcyganov, & Gabilovich, 2016). They promote

cancer cell survival, angiogenesis, and the invasion of cancer into healthy tissue and metastasis (Kumar et al., 2016).

Another important immunosuppressive factor is the increased prevalence of Tregs in patients with PDAC (Whatcott, Posner, Von Hoff, & Han, 2012). Regulatory T cells have potent immunosuppressive activity as they express and release multiple factors into the tumor microenvironment. These factors have been shown to exhibit strong anti-inflammatory effects that contribute to suppressing autoimmunity (Ren et al., 2018).

The inflammatory cells (MDSCs, Tregs, and tumor-associated macrophages, or TAMs) that invade tumors are often localized to the tumor stroma. Tumor-associated macrophages are among the most common immune cell type to infiltrate into the tumor microenvironment. Once activated, these macrophages contribute to desmoplasia. They also interfere with the metabolism of effector T cells by releasing various growth factors, cytokines, chemokines, and enzymes into the tumor microenvironment (Ren et al., 2018; Whatcott et al., 2012). Consequently, these processes have a tremendous effect on immunosuppression and potentiation of tumor spread (Tan & El-Rayes, 2018).

LOW EFFECTOR T-CELL INFILTRATION

Effector T cell describes a group of cells that actively respond to a stimulus effecting a change. It includes CD4+, CD8+, and Treg cells which, under normal conditions, carry out cell-mediated immune responses and do so by penetrating and infiltrating tumors. In PDAC, the concentration and infiltration of these cells is typically found to be much lower. Furthermore, the T cells that have in fact penetrated the tumors have mostly lost their effector functions due to regulatory T cells provoking a dysfunctional state in these cells resembling T-cell exhaustion.

Pancreatic ductal adenocarcinoma is classically considered a nonimmunogenic tumor because very few effector T cells infiltrate these tumors. Regulatory T-cells in the area block effector T-cell division, and both macrophages and $\gamma\delta$ T cells, another type of immunosuppressive T cell, prevent effector T cells from entering the TME through mechanisms including programmed cell death protein 1 (PD-1) and programmed cell death ligand

1(PD-L1) signaling. Such escape mechanisms have been well documented in cancers with T cell-inflamed TMEs such as PDAC (Young et al., 2018). A possible explanation for the therapeutic failure of PD-1 or PD-L1 blockade therapy in PDAC is the lack of a natural infiltration of effector immune cells in the majority of PDAs (Soares et al., 2015).

LOW TUMOR MUTATIONAL BURDEN

Cancers are caused by the accumulation of somatic mutations that can result in the expression of neoantigens. Tumor mutational burden (TMB) measures the quantity of mutations found in a tumor. Tumor mutational burden levels are typically divided into three groups based on genomic testing reports: low (1–5 mutations/Mb), intermediate (6–19 mutations/Mb), and high (≥ 20 mutations/Mb). Recent reports link high TMB with response to immune checkpoint inhibitors in several cancer types (Schrock, 2019). Defining tumors that are hypermutated with an increased mutation burden and understanding the underlying mechanisms in pancreatic cancer have the potential to advance therapeutic development, particularly for immunotherapeutic strategies (Humphris et al., 2017).

High TMB tumors such as melanoma often have higher levels of neoantigens that can be recognized by the immune system. In contrast, PDAC is a “cold tumor” with low TMB and is consequently not responsive to immune checkpoint inhibitors, a situation that creates a challenge for the successful application of immunotherapy in these cancers (Vareki, 2018).

In summary, for all of the reasons mentioned above, PDAC poses a great treatment challenge leading to the failure of many otherwise-effective management modalities including immunotherapy. As our understanding of this complex situation evolves, novel combination strategies are being considered to target these elements in an attempt to maximize the chance of treatment success.

CONCLUSIONS AND FUTURE DIRECTIONS

Pancreatic ductal adenocarcinoma is a heterogeneous and complex disease with an aggressive nature and abysmal prognosis. It remains one of the most challenging cancers to treat because of its strongly immunosuppressive microenviron-

ment leading to immune evasion and rapid tumor progression. As our understanding of this disease evolves, treatment methods to overcome these obstacles are becoming apparent.

Potential regimens targeting components of the dense stroma that provide an opportunity to be studied in preclinical trials include somatostatin analogues targeting cancer-associated fibroblasts, hyaluronidase-targeting HA, and vitamin D analogs targeting PSCs. Focal adhesion kinase (FAK), which promotes tumor progression and metastasis through its effects on both cancer cells and the stromal cells of the TME (Young et al., 2018), may provide another stromal target.

Considerations in targeting the immunosuppressive tumor microenvironment include inhibition of indoleamine 2,3-dioxygenase (IDO) in combination chemotherapy. Regimens that have been promising in preclinical trials that warrant further study in clinical trials include immunotherapy along with targeted therapy against IL-6, C-X-C chemokine receptor type 2 (CXCR2), and colony-stimulating factor 1 receptor (CSF1R; Tan & El-Rayes, 2018).

Multiple additional designs have been proposed in preclinical studies to target the PDAC immunosuppressive microenvironment and increase immunotherapy efficiency, such as combinations of chemotherapy, radiation, and therapeutic vaccines. Such single or combination treatments may better target these barriers to help improve clinical outcomes for this deadly disease.

IMPLICATIONS FOR ADVANCED PRACTITIONERS

Advanced practitioners working in the area of pancreatic cancer will come away with a deeper understanding of why it can be such a challenging disease to treat. Practitioners involved in clinical trials focusing on pancreatic cancer will benefit from understanding the science behind combining different modalities to better target barriers within the pancreatic TME. On a more practical level, providers will be in a better position to educate patients curious to understand more about their clinical trial design and drug regimen. Providers will also be better able to inform colleagues and peers wishing to learn more about pancreatic TME and barriers to treatment. ●

Disclosure

The authors have no conflicts of interest to disclose.

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