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REVIEW ARTICLE

Pancreatic Cancer and Immunotherapy: Resistance Mechanisms and Proposed Solutions

Elaine Tan[1] · Bassel El-Rayes[2,3]

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Abstract

Background Pancreatic ductal adenocarcinoma (PDAC) continues to be one of the most aggressive and lethal diseases in the world. The success of immunotherapy in other types of malignancy has led to further trials to understand better the role of immunotherapy in PDAC. However, initial studies with immunotherapy, namely, the checkpoint inhibitors, in PDAC have not been met with the same outcomes. The purpose of this review is to identify and discuss the various resistance mechanisms of PDAC to immunotherapy (pancreatic stroma, genetic predisposition/epigenetics, and the immune inhibitory cells, cytokines, soluble factors, and enzymes that comprise the tumor microenvironment) and the solutions currently being studied to overcome them.

Conclusions Various preclinical and early clinical studies have shown that immunotherapy, especially checkpoint inhibitors, in PDAC may be efficacious as part of a multi-modal treatment, in combination with other therapies that target these resistance mechanisms. Several clinical trials are ongoing to explore this concept further.

Keywords Pancreatic ductal adenocarcinoma · Immunotherapy · Resistance

Introduction

Pancreatic ductal adenocarcinoma (PDAC) is a highly aggressive and lethal disease and is the seventh most common cause of cancer-related death worldwide [1]. The annual incidence of PDAC worldwide exceeds 338,000 cases, with the majority presenting as locally advanced or metastatic disease at the time of diagnosis [1, 2]. Current first line regimens for this disease include FOLFIRINOX (infusional 5 fluorouracil (FU), leucovorin, irinotecan, and oxaliplatin), which has a median survival of 11.1 months and gemcitabine with nab paclitaxel, with a median survival of 8.5 months [1, 3, 4]. Multiple other regimens have been tested in clinical trials, but unfortunately, have not shown any significant benefit and the survival rate remains at less than 5% at 5 years [5].

✉ Elaine Tan
estan@emory.edu

[1] Department of Internal Medicine, Emory University School of Medicine, 100 Woodruff Circle Suite, 327, Atlanta, GA 30322, USA

[2] Department of Hematology and Medical Oncology, Emory University, Atlanta, GA, USA

[3] Winship Cancer Institute, Emory University, 1365 Clifton Road, Atlanta, GA 30322, USA

Immunotherapies including checkpoint inhibitors are currently being evaluated in PDAC. Checkpoint inhibitors include anti-cytotoxic T lymphocyte associated antigen 4 antibody (CTLA 4) which binds to the CTLA 4 receptor on T cells and allows for costimulation between B7 on antigen presenting cells and CD28 on T cells and further T cell activation, as well as anti-programmed cell death protein 1 (PD 1) inhibitor, which prevents the interaction of programmed death ligand 1 (PD L1) on infected cells and PD 1 on T cells, natural killer cells, monocytes, and B cells, leading to increased immune activation [1, 5]. Unfortunately, clinical trials of ipilimumab, a CTLA 4 antibody, and anti-PD L1 antibodies as monotherapy have not shown clinical benefit in the majority of PDAC [6, 7].

Pancreatic Stroma

The dense stroma of PDAC creates a major source of resistance to immunotherapy. Forming more than 80% of the tumor mass, the stroma creates a physical barrier that impairs delivery of immunotherapies to cancer [8]. Although the number of tumor infiltrating T cells is increased in PDAC compared to normal pancreatic tissue, the T cells are trapped in the stroma and are prevented from interacting with the cancer

cells [9]. Hartmann et al. found that contact between T cells and collagen type I within the extracellular matrix of PDAC stroma prevents chemokine-guided migration towards the cancer cells [9]. This is due to the protein α2β1 integrin mediating the adhesion of T cells to collagen [9]. Collagen I also interacts with collagen IV and integrins on PDAC cells, allowing further proliferation and spread of the cancer [10]. Deposition of collagen I is thought to be influenced by FAK1 tyrosine kinase, as high FAK1 expression is associated with higher levels of total stromal collagen and collagen I deposition [11].

Cancer-associated fibroblasts that originate from pancreatic stellate cells (PSC) play a central role in the development and propagation of the stroma. The PSC are activated through EGFR signaling and express growth factors facilitating PDAC invasion and fibrogenesis [12, 13]. The extracellular matrix is comprised of collagen as well as elements like fibronectin, hyaluronic acid (HA), and nerve fibers that secrete factors potentiating growth of PDAC cells [13]. HA, a complex polysaccharide, increases interstitial fluid pressure, which causes narrowing of blood vessels and impaired delivery of immunotherapy into the interstitium [10]. HA has been associated with reduction in anti-tumor immune responses and activation of regulatory T cells (Tregs) [14]. Additionally, the cells in the stroma secrete other immunosuppressive factors that contribute to immunotherapy resistance. Cancer-associated fibroblasts, for example, secrete IL 6 and CXCL12 while attracting IL 17 secreting CD4+ cells.

Genetic Mutations/Epigenetic Factors

Although not extensively studied, some genetic mutations/epigenetic factors in PDAC are associated with resistance to immunotherapy. Resistance to anti-PD 1 therapy has been associated with a particulate gene signature known as innate anti-PD 1 resistance, or IPRES, in melanoma [15]. The concurrent expression of 26 genes was prevalent in 9 of 13 non responding tumors vs. 1 of 15 responding tumors treated with anti-PD 1 therapy. These genes that characterize IPRES are associated with heightened mesenchymal transition, angiogenesis, hypoxia, and immune suppressing wound healing, ultimately proposed to lead to anti-PD 1 resistance [15]. Gene signatures specific for PDAC that play a role in the resistance to anti-PD 1 therapy have not yet been identified, but are possible.

Loss of PTEN or decreased PTEN expression is thought to promote resistance to T cell-mediated immune response given that PTEN loss is associated with inferior outcomes with anti-PD 1 inhibitor therapy [16]. Decreased PTEN expression is found in PDAC and is thought to contribute to the resistance to immunotherapy by increasing PI3K levels, which led to decreased T cell infiltration in the tumor and increased expression of immunosuppressive cytokines and tumor-associated macrophages [17].

Pancreatic cancer derived exosomes contribute to genetic modulation of immune response. Exosomes can transfer miRNAs and induce mRNA expression in target cells. A study by Ding et al. revealed that the miRNA, miR 212 3p, secreted from pancreatic cancer exosomes, inhibits regulatory factor X-associated protein (RFXAP) leading to decreased MHC expression and induction of immune tolerance in dendritic cells [18]. Additionally, this miRNA may be involved in the invasion and spread of PDAC.

After exposure to immunotherapy, cancers can develop secondary resistance through loss of antigen expression, phenotypic change of anti-tumor T cells, or beta 2 microglobulin mutations leading to decreased HLA expression. While this has been observed in other malignancies, it has not been observed in PDAC given the difficulties with primary exposure to immunotherapy [19].

Immunosuppressive Tumor Microenvironment

The tumor microenvironment in PDAC is a significant barrier to successful treatment with immunotherapy. Various cells, cell receptors, cytokines/soluble factors, and enzymes create an environment that inhibits immune cells from working effectively in PDAC.

Immune Inhibitory Cells

The various cell types involved in the tumor microenvironment include regulatory T cells (Tregs), myeloid derived suppressor cells (MDSCs), and tumor associated macrophages (TAMs).

Tregs are induced by tumor-associated antigens to facilitate immune tolerance, by expressing CTLA 4 and binding to B7 ligands, subsequently inhibiting CD4 and CD8 T cell proliferation. Tregs decrease antigen presentation and produce cytokines like TGF beta and IL 10 [20]. MDSCs, also prominent in the tumor microenvironment, cause T cell apoptosis through production of reactive oxygen species, prevent protein synthesis by T cells through upregulating arginase, and impair T cell motility by downregulating L selectin. They also produce additional immunosuppressive cytokines [20]. TAMs facilitate the switch from a proinflammatory state to an anti-inflammatory state and secrete additional growth factors that potentiate tumor spread [21]. Each of these cell types correlate with advanced disease and worse outcomes in cancer [21].

Cytokines/Soluble Factors/Enzymes

Malignant and nonmalignant cells in the tumor microenvironment produce multiple cytokines and metabolites. These then interact with receptors leading to proliferation of immune suppressive cells and inhibition of immune effector cells. Examples of receptors commonly expressed in the PDAC environment include CCR2, a chemokine receptor on immunosuppressive macrophages, and CXCR2, a G protein-coupled receptor on neutrophils on MDSCs [22, 23]. Higher expression of CXCR2 at the tumor border is associated with a poorer prognosis [23]. Another cell surface receptor expressed in high levels in PDAC is CCR5, which is thought to mediate Treg activation and T cell chemotaxis [24].

In preclinical studies, high expression of colony stimulating factor 1 (CSF 1) has been associated with limited efficacy of checkpoint inhibitors [22]. By binding to the receptor CSF 1R, this cytokine leads to MDSC and TAM recruitment to the tumor microenvironment. IL 10 and TGF beta, both prominent in the tumor microenvironment, lead to increased reactive oxygen species and impaired T cells migration [21]. IL 10 further inhibits IL 12 expression leading to decreased MHC class II expression on antigen presenting cells and downregulates T cell activation, while TGF beta induces Treg differentiation and inhibits T cell and macrophage activation and proliferation [24]. Kynurenin is a prominent metabolite in the tumor microenvironment converted from tryptophan by indolamine 2,3 dioxygenase (IDO), which is highly expressed in PDAC cells. Kynurenin decreases T cell activation, stimulates Tregs, and suppresses dendritic cells [22].

The stroma is another source of cytokines, such as IL 6 and IL 17 that contribute to the immune suppressive nature of the tumor microenvironment. IL 6 facilitates downstream signaling of the JAK/STAT pathway, which leads to proliferation of immune suppressive cells, specifically MDSCs and Tregs [25]. Stimulated by tumor cells and cancer-associated fibroblasts, IL 17 secretes Th17, which further causes immune suppression and tumor progression [26].

Combating Immunotherapy Resistance

Given the multiple aspects in PDAC causing resistance to immunotherapy, a multi-targeted approach should be implemented to effectively induce an immune response. Preclinical and early clinical trials have evaluated novel multi-targeted approaches in order to overcome immunotherapy resistance in PDAC.

Targeting the Pancreatic Stroma

While the stroma facilitates various mechanisms of resistance to immunotherapy, many aspects of the stroma previously discussed are "targetable." As already discussed, cancer-associated fibroblasts comprise a large portion of the stroma and secrete several immunosuppressive cytokines. Given that the mTOR pathway is upregulated in cancer-associated fibroblasts, inhibition of this pathway has been of interest. Unfortunately, phase II trials using mTOR inhibitors in advanced PDAC were disappointing [27, 28]. Somatostatin analogue SOM230 was studied in human PDAC cells and was found to inhibit the mTOR/PI3K pathways. While this had no effect on cancer-associated fibroblast proliferation or survival, it was associated with decreased production of collagen I and IL 6 by cancer-associated fibroblasts [8]. Combining SOM230 with gemcitabine led to decreased tumor growth and chemoresistance in mice xenografted with human pancreatic cancer cells or tumor resections [8]. Given these encouraging results, it is possible that blockade with SOM230 could also abrogate immunotherapy resistance, given its downstream effects on decreasing stromal collagen and immunosuppressive cytokines.

Another targetable aspect of the stroma is hyaluronic acid (HA). Pegylated recombinant human hyaluronidase (PEGPH20) breaks down HA, causing decreased interstitial fluid pressure and increased blood vessel diameter facilitating improved drug delivery. Given its promising results in the preclinical setting, this drug is now being testing in clinical trials [29]. Recently, a phase II clinical trial compared PEGPH20 and gemcitabine/nab paclitaxel vs. gemcitabine/nab paclitaxel [14]. Median progression-free survival in the PEGPH20 group was 5.7 months compared to 5.2 months in the control group. However, when stratifying patients based on their HA levels, those with high HA levels had median progression-free survival of 9.2 months in the PEGPH20 group vs. 4.3 months in the control group ($p = 0.05$), with one complete response in the PEGPH20 group. In patients with low HA levels, there was no difference in progression-free survival between the PEGPH20 group and control group (5.3 vs. 5.6 months, $p = 0.74$) [14]. Currently, there are additional trials looking at the additive effect of PEGPH20 with chemotherapy with consideration of HA levels. Given that HA also contributes to immunotherapy resistance, it is possible that PEGPH20 or a similar intervention could enhance the effectiveness of immunotherapy, especially in tumors with high HA levels.

FAK inhibition decreases collagen deposition and fibroblast numbers and activity. Murine models have shown a synergistic effect of combining FAK inhibition with chemotherapy and immunotherapy. Combining FAK inhibition with gemcitabine led to a significant increase in median survival, while combining FAK inhibition with adoptive T cell therapy led to increased infiltration of the T cells by 4.7 times in the pancreatic tumor tissue [11]. Additionally, treatment of mice with FAK inhibitor, gemcitabine, and anti-PD 1/CTLA 4 vs. gemcitabine and anti-PD 1/CTLA 4 led to a greater than 2.5

times increase in median survival ($p < 0.01$) [11]. These results are encouraging and suggest that FAK inhibition can overcome PDAC's resistance to immunotherapy in humans; however, further clinical trials are needed.

Vitamin D receptor (VDR) expression has been noted on pancreatic stellate cells (PSC); administration of calcipotriol, a potent non hypercalcemic vitamin D analog, causes its binding to VDR which leads to suppression of PSC activity and subsequently decreased inflammation and stromal fibrosis in mouse models [30]. VDR has effects on the extracellular matrix, cytokines, and chemokines like IL 6, and CXCL12, a mediator of the T cell blockade [31]. Administration of calcipotriol concurrently with gemcitabine compared to gemcitabine alone led to significantly increased intratumoral concentrations of gemcitabine in mice with PDAC ($p < 0.05$) and a median survival of 22 days compared to 14 days ($p = 0.02$) [31]. Therefore, administration of a vitamin D analog like calcipotriol may be able to enhance the efficacy of immunotherapy by decreasing fibrosis and the stromal barrier to drug delivery, alleviating immunosuppression.

CD40, a TNF receptor, is expressed on B cells, dendritic cells, and fibroblasts; when activated, CD40 is involved in activating antigen presenting cells for T cell priming and activation [22]. CD40 agonists have been shown to cause monocyte infiltration into tumor tissue, and evaluation of the tumor stroma after treatment with CD40 agonists in one study revealed a decrease in collagen I with stromal degradation [32]. When combined with gemcitabine, median progression-free survival for 22 patients in a phase I trial was 5.2 months with median overall survival of 8.4 months. In this study, gemcitabine was given before the CD40 agonist, and the authors propose that the CD40 agonist may be more efficacious when given before gemcitabine [33]. This may be another therapy that augment immunotherapy's activity against PDAC.

Targeting Genetic Mutations

PI3K blockade is thought to affect the downstream effects of a PTEN mutation that can lead to immunotherapy resistance. Inhibition of various isoforms of PI3K has led to various effects on PDAC. PI3K gamma inhibition leads to reprogramming of tumor-associated macrophages to facilitate tumor suppression and decreases collagen synthesis by fibroblasts [34, 35]. In PDAC, combination with an anti-PD 1 agent did not lead to added benefit in preclinical models [35]. On the other hand, PI3K beta inhibition, which is thought to target cancer-associated fibroblasts, in combination with an anti-PD 1 agent, led to significantly decreased tumor growth and increased survival compared to anti-PD 1 alone in mice with melanoma [16]. Further studies will be needed to look at PDAC with PI3K beta inhibition. Additionally, reduced PTEN activity has been associated with upregulation of FAK in multiple tumor types. Potentially, FAK inhibition could be another strategy to overcome this mutation and its effects in PDAC.

Targeting the Immunosuppressive Tumor Microenvironment

Inhibition to IDO has been implemented in to combat PDAC's immunosuppressive environment. A phase II study is currently evaluating the combination of indoximod with gemcitabine/nab paclitaxel in metastatic PDAC. At time of the interim analysis, 37% of patients had demonstrated an objective response including one with complete response by RECIST criteria [36]. The results from this study are highly anticipated.

Inhibition of IL 6 alone to target the PDAC immunosuppression has not been promising; however, combination of IL 6R blockade with anti-PD L1 inhibitor in murine models with PDAC has had encouraging results. Combination therapy vs. anti-PD L1 inhibitor monotherapy led to a decrease in tumor volume ($p < 0.03$) and increased CD8+ T cell infiltration ($p < 0.01$) [25]. Additionally, this combination led to increased survival by 35% when compared to no treatment ($p = 0.001$) [25]. Given IL 6's multiple effects on the tumor microenvironment, IL 6 blockade could allow immunotherapy to be successful in patients with PDAC.

Colony stimulating factor 1 receptor (CSF 1R) is expressed prominently by monocytes, MDSCs, and macrophages. Signaling of CSF 1R has been suggested to activate tumor promoting macrophage phenotypes [22]. Blockade of CSF 1R has led to alterations in the tumor microenvironment leading to a favorable response when combined with immunotherapy in murine models. This favorable response is thought to be related to a decrease in number of TAMs, with remaining TAMs displaying decreased expression of genes related to immunosuppression as well as increased MHC class II expression. Additionally, combination of CSF 1R with anti-PD 1 blockade and chemotherapy vs. anti-PD 1 blockade and chemotherapy in a preclinical study led to a significantly decreased tumor burden ($p < 0.05$) [37]. CSF 1R with CTLA 4/anti-PD 1 vs. CTLA 4/anti-PD 1 led to decreased tumor burden as well and significantly higher number of CD4+ and CD8+ T cells [37]. Therefore, checkpoint inhibitor therapy appears to be more efficacious in conjunction with the CSF1 R blockade.

CXCR2 inhibition has also been studied as a way to combat the immune suppression in PDAC. Inhibition of CXCR2 leads to increased T cell infiltration, which may be related to decreased monocyte/macrophage tumor infiltration. Treatment with CXCR2 inhibitor in murine models leads to tissue with decreased picrosirius red staining indicating a

Table 1 Mechanisms of resistance to immunotherapy in PDAC and targeted treatment

Components	Role in immunotherapy resistance	Targeted treatment
Stroma	Creates a physical barrier decreasing delivery of immunotherapy and T cells to cancer cells	
Cancer-associated fibroblasts	Secrete immunosuppressive cytokines (IL 6 and CXCL12) and attract IL 17 secreting CD4+ cells	SOM230 decreases production of collagen I and IL 6 by cancer-associated fibroblasts; FAK inhibition decreases fibroblast numbers and activity
Pancreatic stellate cells	Express growth factors facilitating invasion and fibrogenesis	
Collagen	Prevents T cell migration to cancer cells	FAK inhibitors decrease collagen deposition; CD40 agonists decrease collagen I
Hyaluronic acid	Increases interstitial fluid pressure and blood vessel narrowing, leading to impaired delivery of immunotherapy to cancer cells	PEGPH20 breaks down hyaluronic acid
Genetics/epigenetics		
IPRES	Heightened mesenchymal transition, angiogenesis, hypoxia, impaired wound healing	
Decreased/absent PTEN expression	Increases PI3K levels, leading to decreased T cell infiltration into the tumor and increased expression of immunosuppressive cytokines and macrophages	PI3K beta inhibition leads to decreased tumor growth and increased survival with anti-PD 1 therapy
miR-212-3p	Inhibits regulatory factor X-associated protein, leading to decreased MHC expression and induction of immune tolerance in dendritic cells	
Immunosuppressive tumor microenvironment		
Regulatory T cells (Tregs)	Inhibit CD4+ and CD8+ T cell proliferation, decrease antigen presentation, produce cytokines like TGF beta and IL 10	
Myeloid-derived suppressor cells (MDSCs)	Cause T cell apoptosis, prevent T cell protein synthesis, impair T cell motility	
Tumor-associated macrophages (TAMs)	Switch environment from a proinflammatory state to an anti-inflammatory state	
CCR2 and CXCR2	Facilitate cell migration to the tumor microenvironment	CXCR2 inhibition leads to increased CD3+ T cells
CCR5	Mediates activation of Tregs and facilitates T cell migration	Anti-GITR antibody decreases CCR expression, inhibiting Treg activity
CSF 1	Recruits MDSCs and TAMs to tumor microenvironment	CSF 1R blockade leads to decreased TAMs and alteration of remaining TAMs
IL10	Inhibits IL 12 and decreases MHC class II expression on antigen presenting cells and downregulates T cell activation	
TGF beta	Induces Treg differentiation, inhibits T cell and macrophage activation and proliferation, promotes a Th2 macrophage phenotype	
IL 6	Potentiates JAK/STAT pathway, leading to proliferation of MDSCs and Tregs	Leads to decrease in tumor volume and increased CD8+ T cell infiltration with anti-PD L1 inhibitor
IL17	Secretes Th17 to cause further immunosuppression and tumor progression	
Indolamine 2,3 dioxygenase (IDO)	Causes decreased T cell activation, increased stimulation of Tregs and dendritic cell suppression	IDO inhibitors are being evaluated in clinic trials

Table 2 Summary of ongoing clinical trials investigating the role of immunotherapy in PDAC in conjunction with other therapies

Study title	Status	Patient population	Phase	ClinicalTrials.gov identifier
Phase I Study of Defactinib With Pembrolizumab and Gemcitabine in Patients With Advanced Cancer	Recruiting	Advanced pancreatic cancer	I	NCT02546531
A Phase 1/2, Open-Label, Dose-Escalation, Safety, Tolerability, and Efficacy Study of Epacadostat in Combination With a PD-1 Inhibitor and Chemotherapy in Subjects With Advanced or Metastatic Solid Tumors (ECHO-207)	Recruiting	Advanced or metastatic solid tumor	I/II	NCT03085914
A Phase I/II, Open-label, Multi-center Study of the Safety and Efficacy of BLZ945 as Single Agent and in Combination With PDR001 in Adult Patients With Advanced Solid Tumors	Recruiting	Advanced pancreatic cancer, advanced triple negative breast cancer, or recurrent glioblastoma	I/II	NCT02829723
A Phase IIb Pilot Study to Assess the Efficacy, Safety and Pharmacodynamics Effects of Pembrolizumab and BL-8040 in Patients With Metastatic Pancreatic Cancer	Recruiting	Metastatic pancreatic cancer	IIb	NCT02907099
A Phase 1a/1b Study of FPA008 in Combination With Nivolumab in Patients With Selected Advanced Cancers	Recruiting	Advanced solid tumors	Ia/b	NCT02526017
Pilot Study of Feasibility and Safety of Personalized Autologous Cluster of Differentiation 8 (CD8+) T Cell Therapy Plus Anti-PD1 Antibody in Advanced Solid Malignancies	Pending recruitment	Pancreatic adenocarcinoma, colorectal adenocarcinoma, cholangiocarcinoma, esophageal cancer, or gastric cancer with radiographic evidence of metastatic disease	I	NCT02757391
A Phase II, Multicenter, Open-label Single Arm Study to Assess the Safety and Efficacy of the Combination of BL-8040 and Pembrolizumab in Patients With Metastatic Pancreatic Cancer, the COMBAT Study	Recruiting	Metastatic pancreatic adenocarcinoma	II	NCT02826486
A Study of ARRY-382 in Combination with Pembrolizumab, a Programmed Cell Death Receptor 1 (PD-1) Antibody, for the Treatment of Patients with Advanced Solid Tumors	Recruiting	Advanced solid tumors	Ib/II	NCT02880371
A Phase 2, Multicenter Study of FOLFIRINOX Followed by Ipilimumab in Combination With Allogeneic GM-CSF Transfected Pancreatic Tumor Vaccine (GVAX) in the Treatment of Metastatic Pancreatic Cancer	Recruitment suspended	Metastatic pancreatic adenocarcinoma	II	NCT01896869
A Phase I Study of a p53 MVA Vaccine in Combination With Pembrolizumab	Recruiting	Advanced solid malignancy	I	NCT02432963
A Randomized Study of a GM-CSF Secreting Allogeneic Pancreatic Cancer Vaccine With Or Without a PD-1 Blockade Antibody (Nivolumab) for the Neoadjuvant and Adjuvant Treatment of Patients With Surgically Resectable Adenocarcinoma of the Pancreas	Recruiting	Pancreatic adenocarcinoma	I/II	NCT02451982

reduction in collagen I expression as well as an increase in CD3+ T cells [23]. A significantly longer survival is also seen with anti-PD 1 inhibitor combined with CXCR2 inhibition vs. anti-PD 1 inhibitor alone ($p = 0.037$) [23].

Interferon (IFN) alpha is a cytokine that has multiple effects on the immune system and has been studied in PDAC. The cytokine plays a role in upregulating MHC class I expression, enhancing the cytotoxic activity of NK cells and CTLs, and enhancing humoral immunity [38]. IFN alpha plays a role in tumor-associated antigen presentation to dendritic cells and the production of immune stimulating cytokines [39, 40]. A study in murine models showed that giving an agonistic anti-glucocorticoid-induced tumor necrosis factor receptor (GITR) monoclonal antibody can inhibit the activity of Tregs, which is thought to occur by decreasing CCR5 expression. Combination of these two treatments vs. interferon alpha alone led to higher infiltration of CD4+ cells ($p = 0.029$) and CD8+ cells ($p = 0.021$) effector T cells ($p = 0.0012$). Additionally, the combination treatment was associated with decreased Tregs [41]. By targeting the Tregs with agonist GITR monoclonal antibody, the interferon alpha is likely able to be more effective and allow for improved CTL infiltration.

FAK inhibition, previously mentioned, has also been shown to combat immunosuppression, specifically decreasing levels of MDSCs, Tregs, and TAMs; this may be another reason, apart from its effects on the stroma, why its combination with immunotherapy in preclinical trials has been promising [11].

Future Directions

In conclusion, PDAC has demonstrated several resistance mechanisms to immunotherapy related to its pancreatic stroma, genetic alterations, and immunosuppressive environment. Initial trials with immunotherapy have been disappointing. An increased understanding of these resistance mechanisms has led to development of targeted therapies against the immunosuppressive environment and stroma (Table 1). These therapies, in combination with immunotherapy, have led to promising results in preclinical and early clinical studies in PDAC. Several clinical trials are ongoing to explore this further (Table 2). Additionally, some of the targeted therapies have been promising in conjunction with chemotherapy, but have not been studied in conjunction with immunotherapy. Potential regimens that should be studied in preclinical trials include somatostatin analogues, hyaluronidase, vitamin D analogs, CD40 agonists, and blockade against PI3K beta and IDO in combination with immunotherapy. Regimens that have been promising in preclinical trials that warrant further study in clinical trials include immunotherapy along with targeted therapy against FAK, IL 6, CXCR2, CSF 1R, and GITR. Consideration of the individual's tumor marker and cytokine expression can determine the appropriate immune-based combination regimen, allowing for personalized treatment and potentially improved outcomes.

Compliance with Ethical Standards

Conflict of Interest The authors declare that they have no conflict of interest.

Informed Consent For this type of study, formal consent is not required.

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