Review Article

Cellular crosstalk mediating immune evasion in pancreatic cancer microenvironment

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Abstract: Pancreatic ductal adenocarcinoma (PDAC) is the fourth most common cause of death from cancer worldwide, with a poor 5-year survival rate of 6%. Immunity in PDAC patients is diminished and the associated immune evasion is an underinvestigated field. The microenvironment of pancreatic cancer is an intricate mesh-like network in which various resident cell populations are closely interacting. To understand the roles played by these cell types, we attempt to delineate the diversified components in pancreatic cancer microenvironment and their contributions in hampering immune escape. In sum, there are two tiers of force influencing the clinical outcome of patients with pancreatic cancer. The anti-tumor force includes CD8+ T cells, NK cells, M1-type macrophages, Th1 cells, and dendritic cells (DCs). The other force facilitates tumor cells to become free of attacks from immune system, including cancer cells, PSCs, M2-type tumor-associate macrophages (TAMs), myeloid-derived suppressor cells (MDSCs), Tregs, and Th2 cells. Combined therapy to break the balance between the two forces maybe a promising strategy to benefit patients with pancreatic cancer.

Keywords: Pancreatic cancer; immune evasion; immunotherapy; tumor microenvironment

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Introduction

Pancreatic ductal adenocarcinoma (PDAC) is the fourth most common cause of death in cancer worldwide, with a poor 5-year survival rate of 6%. Patients with PDAC poorly respond to chemotherapy, radiotherapy, or immunotherapy. Emerging evidence linking such poor prognosis and immune evasion compels us to explore the mechanisms behind immunosuppression in pancreatic tumor microenvironment (TME). This review introduces several potential mechanisms focusing on resident cells in pancreatic TME. We have also summarized recent published studies (1-25) in this field in Table (26-50) (http://fp.amegroups.cn/cms/apc.2019.06.04-1.pdf) (51-55).

Pancreatic cancer microenvironment

PDAC is characterized with accumulation of a desmoplastic stroma, which is composed of a plethora of cellular and acellular components, including fibroblasts, immune cells, pancreatic stellate cells (PSCs), endothelial cells, extracellular matrix, and soluble proteins such as cytokines and growth factors (56-59). These components constitute an immunosuppressive TME and up to 90% of the tumor bulk, function as an important mediator of therapy resistance and might be responsible for the failure of immunotherapy (60). While immune cells are abundant within the stroma, they mostly belong to immunosuppressive subsets such as tumor-associated macrophages (TAMs), tumor-associated

neutrophil granulocytes (TANs), regulatory T cells (Tregs), or immature myeloid cells/myeloid-derived suppressor cells (MDSCs) (61-63). These cells are recruited by tumors as an escape mechanism from immune surveillance, and they interact with other stromal components to create an immunosuppressive network (64). Conversely, anti-tumor immune cells like DCs, NK cells, and $CD8^+$ T cells are relatively few and their anti-tumor effects are generally impaired. For example, majority of $CD8^+$ T cells in pancreatic TME express high level of immune checkpoint receptors that significantly limit their cell-killing effects (65). One study has shown that adipocytes contribute to tumor microenvironment of obese pancreatic cancer patients. Hijacked adipocytes or tumor cells produce IL-1 to activate pancreas stellate cells (PSCs), and then the activated PSCs (aPSC) secrete IL-1 to recruit TANs who subsequently activate PSCs and mediate the tumor immunosuppressive microenvironment (66).

The PDAC microenvironment is glycolytic compared to surrounding tissue and associated with poor outcome in PDAC (67). Increased lactate in the extracellular environment was exported by monocarboxylate transporter 4 (MCT4) from tumor cells who obtain energy in unique metabolic ways such as aerobic glycolysis, also known as the "Warburg effect". The resultant lactate can limit T-cell effector functions, as this subset of immune cells is dependent on aerobic glycolysis (68). In contrast, macrophages and Tregs are able to use fatty acid oxidation to survive in a low-glucose environment (69,70). Additionally, the accumulated lactate at high concentration can skew macrophages towards the anti-inflammatory, tumor-promoting phenotype (M2-type) (71).

Pancreatic cancer cells

To escape attacks from the immune system, pancreatic cancer cells adaptively adjust themselves to the microenvironment around. For example, cancer cells downregulate the expression of antigen presenting molecules including major histocompatibility antigen (MHC) class I, B7-H5, and Fas receptor to diminish cell-mediated immunity and intensify the expression of Fas ligand, which induces apoptosis of activated antitumor cytotoxic T cells (12,72-75). Further, pancreatic ductal epithelium upregulates the expression of adhesion molecule L1CAM (CD171), which enriches Tregs in TME to correlate malignant progression (50,76). Soluble ULBP2 (sULBP2), a ligand of NKG2D receptor, was also upregulated in pancreatic cancer to decrease NK cytotoxicity towards tumor (4). Moreover, Hinz *et al.* found that transforming growth factor-2 (TGF-β2) induced Foxp3 (a transcription regulator) expression in pancreatic carcinoma to mediate immune privilege by suppressing proliferation of activated cytotoxic T cells, suggesting that pancreatic cancer cell may mimic Treg function in immune evasion (6). Similarly, focal adhesion kinase (FAK) was reported to correlate with high level of fibrosis and poor $CD8^+$ cytotoxic T cell infiltration (7). B7-H4 is highly expressed on pancreatic cells disregard of the presence of pancreatic cancer, and it inhibits T cell proliferation, filtration, and interleukin-2 production (77-79). It was reported that tumor-associated Tregs can stimulate macrophages to secrete IL-6, subsequently activate STAT3 which binds to the promoter of B7-H4 gene, leading to enhanced B7-H4 expression in tumor cells, antigen presenting cells (APCs), or other microenvironment-supporting cells (80-82).

It was reported that pancreatic cancer cells can secret TGF-β via activating Smads (83,84) and facilitate immature dendritic cells turning into TGF-β–secreting cells (85,86). TGF-β is a multifunctioning cytokine involved in nearly all pivotal steps of neoplasia (87). In early pancreatic tumorigenesis, TGF-β acts as a suppressor due to its growth-inhibitory effect on epithelial cells, but it appears to promote tumor progression in advanced disease (55) as it promotes the proliferation of PSCs and the recruitment of $CD4^+$ $CD25^+$ $FoxP3^+$ Tregs and also directly affects $CD8^+$ cytotoxic T lymphocytes (CTLs) (65,88). According to reports, TGFβ inhibits CTL activity and differentiation through several suppressant genes implicated in anti-tumor immune response including Granzyme B (89), which is an anti-tumor serine protease found in CTL-associated cytotoxic granules (90,91). Patients with loss of SMAD4 are appropriate target population for using TGFBR-inhibition therapy since those patients will not benefit from growth-inhibitory effects of TGF-β (55). In addition, tumor cells can also secrete IL-1 to induce intratumoral DCs to produce CCL22, which was known to recruit Treg into TME (11).

Cancer cells are also capable of transforming their metabolism in TME to escape the attacks from immune system (92). For example, increased expression of indoleamine-2,3-dioxygenase (IDO) depletes tryptophan which is an important amino acid routinely functions in immune system cells including NK cells, cytotoxic T cells, and T effector (Teff) cells in PDAC (53,93,94). MMP-9

is a 92-kDa type IV collagenase secreted by mesenchymal stem cells (MSC) that can significantly limit the cytotoxicity of NK cells *in vitro* through decreasing the expression of NKp30, NKG2D, and perforin, and inhibiting the secretion of interferon gamma (IFN-γ) and tumor necrosis factor (TNF)-α (53,95,96).

Pancreatic stellate cells (PSCs)

Increasing evidence demonstrates that the interaction between aPSCs and PDAC cells makes a difference in the development of PDAC. Through producing high level of growth factors, cytokines, chemotactic factors and excessive extracellular matrix (ECM), PSCs create desmoplasia and a glycolytic microenvironment that promote the initiation, development, invasion, metastasis, immune evasion, and chemoradiotherapy resistance of PDAC (97). In response to pancreatic injury, stress, inflammation, resident PSCs are converted into an activated myofibroblast-like phenotype to express α-smooth muscle actin (α-SMA), and synthesize ECM proteins to form fibrous tissue (98,99). The aPSCs are the main source of cancer-associated fibroblasts (CAFs). However, the difference between CAFs and aPSCs is still under debate (100).

PSCs play an important role in mediating immune escape in pancreatic cancer. The rigid ECM components (such as collagen and fibronectin) induce Rho-associated coiled-coil kinase-dependent activation of FAK1. In turn, FAK1 tyrosine kinase regulates T cell survival, antigen sensitivity, cytokine production and migration, to significantly contribute to immunosuppression (7). Known to be produced by CAFs, fibroblast activation protein-α (FAP-α) and βig-h3 disrupt anti-tumor immunity, leading to immune escape and tumor growth (43,45).

PSCs also secrete plentiful soluble cytokines that conduce to T cell exhaustion and dysfunction. FAP^+ PSCs are the only tumoral source of chemokine (C-X-C motif) ligand 12 (CXCL12, also named stromal-derived factor-1, SDF-1) that limits cytotoxic T cell trafficking, prompts macrophages' differentiation into M2-type, and recruits TANs and MDSCs to the tumor site (101). Meanwhile, CXCL12/SDF-1 bound to PDA cells inhibits T cell access, leading to reduced immune responses (40,102). A recent study showed that REG3β expressed and released by healthy cells in the peritumoral region far from microenvironment could activate CXCL12/CXCR4 signaling cascade and interfere with the intercellular communication inside the tumor mediated by extracellular vesicles, resulting in macrophage phenotype alteration and tumor cell migration (103). In PDAC, activated PSCs, TAM, Tregs and mast cells can inhibit DC activation by producing high level of immunosuppressive cytokines such as IL-10 and TGF-β (99,104,105). Similarly, interleukine-6 (IL-6), another versatile PSCs/MDSCs derived cytokine, inhibits cytotoxic T lymphocyte (CTL) anti-tumor immunity by multiple mechanisms, including impairing Teff cell trans-endothelial migration, activation of Treg cells or TAMs, and disrupting the balance of Treg/Teff activities (44,106). Other excessive PSCs-derived suppressive cytokines such as VEGF, granulocyte-macrophage colony-stimulating factor (GM-CSF), PGE2, and MCP-1, also primarily contribute to immune escape and therapeutic resistance of PDAC (107,108). Furthermore, a recent study has demonstrated that galectin-1 is also secreted by activated PSCs, and it modulates Teff cell activation, proliferation, and apoptosis, holds T cells in an anergic state, and breaks the cytokine secretion balance toward a T helper type 2 (Th2) immune response (41).

Recent studies show that CAFs are capable of attracting and sequestering $CD8^+$ T cells in the extratumoral compartment, which dampens T cells' contact with and cell-killing effect of tumor cells (109). Depletion of CAFs abolishes immune suppression (110,111), enhances anti-tumor activity of anti-CTLA-4 as well as anti-PD-L1 (40,46), but also leads to infiltration of Tregs and induction of more aggressive tumor phenotypes (46,112). PEGylated hyaluronidase-degraded hyaluronic acid increases the intratumoral delivery of chemotherapy drugs (57) and improves effective tumor infiltration by CTLs (113) but it was restricted to patients with high hyaluronic acid level in tumor (114). Cyclopamine (M-CPA)/paclitaxel (PTX) was recently reported to restrain tumor cell proliferation and increase intratumoral vasculature density without concomitant infiltration of Tregs or MDSCs (47).

Myeloid cells

Tumor-infiltrating myeloid cells including $CCR2^+$ TAM and $CXCR2^+$ TAN, known as important mediators of immune evasion (115), secrete high level of IL-10 to enhance Treg and Th2 activation and expansion, and they can be targeted by small molecule inhibitors of CCR2 (CCR2i) and CXCR2 (CXCR2i) (116-118). Combined targeting of $CCR2^+$ TAM and $CXCR2^+$ TAN has the benefit of avoiding compensatory increase of counterpart compared with either TAM or TAN targeting alone. This is expected to result in a

significantly influx of $CD8^+$ tumor-infiltrating lymphocytes with a remarkable decrease of Tregs to infiltrate (60,119).

Depending on dominant signals around, macrophages can adopt an alterable functional status (24). When stimulated by bacterial products such as lipopolysaccharide (LPS), Th1 cytokines, IFN-γ or TNF-α, macrophages become immunostimulatory (M1) with high expression of inducible nitric oxide synthase (iNOS), CD80, CD86, MHCII proteins, and TNFα to exert their tumoricidal effects (120,121). On the contrary, in response to Th2 cytokines, IL4, IL10, IL13, or immunocomplex (IC), macrophages acquire an alternatively immunosuppressive status (M2) that express arginase 1, CD206, and low amounts of MHCII (120,122). TAMs often have characteristics of both M1 and M2 and the phenotype of TAMs may change during tumor development (123). TAMs may play an M1-like phenotype role in the early stage of carcinogenesis, and gradually convert into an M2-like phenotype when tumors start to invade and metastasize (124). Along with tumor progression, Tams with M1-like phenotype, M2-like phenotype, or with co-expressed markers of both M2 and M1 macrophages exist in pancreatic TME and a higher M2:M1 ratio correlates with a poor prognosis (125,126). Although majority of TAMs tend to be M2-type, their biological activity can be redirected (127). For example, CD40 agonist, paclitaxel, low-dose γ irradiation, inhibitor of receptor-interacting protein kinase 1 (RIP1) or RIP3, or disruption of the dectin 1-galectin 9 axis are all shown to redirect macrophages from M2-type to M1-type with their respective effects, in consequential or parallel manner (22,23,25-27,128-130).

MDSCs ($CD11b^+$), the precursors of macrophage, neutrophil and dendritic cells, are regarded as another inhibitory population of myeloid cells. Accumulated MDSCs and their enhanced function are induced by soluble mediators in TME, including IFN-γ, TNF, and GM-CSF (131-136). Expansion of MDSCs was shown to be further driven by cytokines such as TGF-β, IFN-γ, G-CSF, GM-CSF, VEGF, IL-1β, IL-6, IL-10 and CCL12 in PDAC (137). There was report that cytokines inducing MDSCs via acting on a conjunct molecular pathway and the immunosuppressive activity of both tumor-induced and bone marrow-derived MDSCs are completely dependent on the C/EBPbeta transcription factor (138).

MDSCs exhibit a profound capacity to suppress T cell proliferation and activation in a dose-dependent manner and attenuate functional differentiation of tumor-specific $CD4^+$ T cells into effector TH1 cells via IL-6 production to facilitate tumor progression (106). Depletion of $CD11b^+$ MDSC cells downregulates the expression of PD-L1 in tumor cells, resulting in a significant infiltration of $CD8^+$ T cells and a decrease of immunosuppressive Tregs infiltration, leading to effective inhibition of pancreatic tumor growth in a $CD8^+$T-dependent fashion (17). Recently, neutrophil-like $CD13^{high}$ MDSCs are found to suppress anti-cancer T cell responses via expression of arginase-1 and correlate with poor prognosis of PDAC patients (139). Inhibition of cyclooxygenase-2, PGE2-mediated arginase, or phosphodiesterase-5 was shown to downregulate arginase-1 in murine MDSCs and led to an effective tumor control (19,20).

Tregs

Natural $CD4^+CD25^+$ Tregs and Foxp3-transduced $CD4^+$ T cells suppress naive T-cell proliferation *in vitro* in order to maintain immunologic tolerance (140). The forkhead transcription factor Foxp3 is a key molecular marker to identify Treg function and is the only definitive marker of $CD4^+CD25^+$ Treg (140,141). Apart from naturally occurring Tregs generated from the thymus, adaptive Tregs as negative regulators of anti-tumor cytotoxic T cells recognized, as these cells predominate in infections and tumor such as pancreatic adenocarcinoma and associate with poor prognosis in PDAC (142,143). Tumor cells recruit Tregs throughout an epithelial-to-mesenchymal (EMT) process by expressing L1CAM and secreting mediators including C-C motif chemokine ligand 2 (CCL2) and CCL22 (11,39,50). Tregs inhibit T-cell production of IFN-γ and IL-2 as well as their cytotoxic function, resulting in impediment to naturally occurring anti-tumor immunity (143).

Anti-CD25 mAb can enhance CTL responses and diminish pancreatic tumors in a $CD8^+$ T cell-dependent manner (49). However, this strategy may cause immune dysfunction since CD25 is not a specific Treg marker. Recently, low-dose CpG TLR9 agonist improved ISOCOM tumor vaccine function by breaking Treg-mediated immunosuppression (49,144).

$CD4^+$ T cells

$CD4^+$ T cells are effector helper cells that can differentiate into three major subtypes with distinct function, playing an important role in immune response via releasing dissimilar inflammatory cytokines. Th1 cells support cellular immunity by selectively producing IL-2, IFN-γ and

TNF-α (145,146). Th2 cells support humoral immunity by producing IL-4, IL-5, IL-6, Il-10 and IL-13 (147) IL-4 and IL-10 are deemed as immunosuppressive factors (108,148). Th17 cells exert a strong pro-inflammatory effect by producing IL-17, IL-21 and IL-22 (149,150). While Th1 cells are assistant to cellular response against tumor cells, Th2 cells are believed to collude with pancreatic cancer cells and a higher Th2:Th1 ratio in tumor correlates with a poorer prognosis in pancreatic cancer patients (42). Emerging evidence suggests that IL-10 and TGF-β facilitate the shift of Th1 into Th2 cell type *in vitro* (151). Another study demonstrated that pancreatic cancer cells restrict $CD4^+$ T-cell proliferation and migration, and induce IFN-γ production, supporting a role of $CD4^+$ T cells in immune evasion (152).

$CD8^+$ T cells

As the main force of tumor cell-killing immune cells, cytotoxic $CD8^+$ T cells recognize specific tumor antigens presented as peptides on MHC class I molecules (153). Unfortunately, they express high levels of immune checkpoint receptors such as PD-1 (65). Strong evidence have proved that a pre-existing anti-tumor $CD8^+$ T cell infiltration is required for therapeutic benefit from ICB and other immunotherapy (154,155). However, once memory T cells are generated, they are able to protect mice upon inoculation with other PDAC tumors since limited mutations shared among the majority of PDAC patients (34).

The prime task of the seesaw battle between tumor and body immunity is to release $CD8^+$ T cells from the suppression of PD-L1. PD-L1 (B7-H1), an important co-suppressive molecule expressed on macrophages and DCs as well as on pancreatic cancer cells, was reported as a negative regulator of T-cell responses. PD-1 interacts with its ligand PD-L1 to maintain self-tolerance and to protect against excessive tissue damage induced by immune responses through downregulating the synthesis and secretion of IL-2, IFN-γ and IL-10 by myeloid DCs and T cells, and thus functions as an immune checkpoint under physiological conditions (156-162). It has been reported that PD-L1 can be upregulated by oncogenes such as AKT and STAT3 (163,164) or by chemotherapeutics like 5-fluorouracil, gemcitabine and paclitaxel via several pathways including the JAK/STAT pathway (165). Further, IFNγ, a proinflammatory cytokine secreted by activated T and natural killer (NK) cells and a vital component of the host cancer immune system (166,167), also acts as a prime inducer of PD-L1 in tumor cells via the MEK/ERK pathway and can be inhibited via suppressing STAT1 (168-171).

With the aforementioned role of immunosuppression, blockade of the interaction between PD-1 and PD-L1 by anti-PD-1 or anti-PD-L1 already demonstrated durable efficacy of tumor suppression in both mouse tumor models and human cancer patients except PDAC (65,172-176). The explicit cause of disabled curative effect remains uncertain. It was reported that anti-PD-1 treatment motivated a compensatory increasing expression of cytotoxic T-lymphocyte-associated protein 4 (Ctla4), which is another immune checkpoint (17). Arlauckas *et al.* showed that macrophages can remove anti-PD-1 antibodies from T cells (177). Therefore, combination of anti-PD-L1 and other therapies appears logic and attractive for pancreatic cancer. As reported, combination of high-dose radiotherapy with anti-PD-L1 markedly enhanced tumor responses in PDAC cell allografts where radiotherapy induced a large amount of tumor cells sensitive to cytotoxic killing. Further, early anti-PD-L1 therapy prevented the growth of immunosuppressive cells and increased recruitment and activation of T cells (178). Analogously, depleting FAP-expressing cells, Mll1 inhibitors, CXCL12/CXC R4 pathway inhibitors, tocilizumab (anti-IL-6), CD40 agonist, STING agonists, for instance, all acted synergistically with anti-PD-L1 to significantly diminish cancer growth in a $CD8^+$T-dependent manner (22,40,44,51,65,168).

B7-H5, a new B7 ligand for receptor CD28H to deliver a costimulatory signal to the human T-cell, is downregulated in pancreatic cancer cells, which might partially cripple $CD8^+$T cells' function (52). Recently, Chen *et al.* discovered that fibrinogen-like protein 1 (FGL1), a protein that is largely limited to liver and pancreas, was a major immune inhibitory ligand of another immune checkpoint LAG-3 (179,180). FGL1 is highly produced by human cancer cells and it inhibits the activation of antigen-specific T cell as well as NK cells (179,181). While its expression appears to be downregulated in pancreas cancer (179), normal pancreatic tissues may express enough FGL1 to exert its immunosuppression. LAG-3^+ cells are frequent in $CD3^+CD8^+$ TILs (96.30%) in PDAC (182). Blockade of FGL1-LAG3 may have promising effect on pancreatic cancer. More recently, it was reported that the intrinsic capacity of intratumoral T cells to recognize tumor tissue was rare and variable, suggesting that reactivating intratumoral T cells would benefit from approaches that simultaneously increase the quality of the intratumoral

TCR repertoire (183).

NK cells

NK cells, as the first line of defense in the body, play an essential role in the innate immune system and anti-tumor immunity including pancreatic cancer (184). Evidence shows that NK cells not only kill target cancer cells directly without prior sensitization (185,186), but also bind to specific surface ligands expressed on cancer cells, such as MHC class I molecules (187). Interestingly, complete loss of MHC class I will motivate NK cells response, which called the "missing self" response (188). In addition to their ability to kill mAb-coated tumor cells via inducing antibody-dependent cellular cytotoxicity (ADCC), FcR-activated NK cells also release cytokines including IFN-γ, TNF-α, MIP-1α, IL-8, and RANTES to improve antigen presentation, stimulate the chemotaxis of T cells, and suppress tumor cell proliferation (189,190). NK cell function can be enhanced via IL-21 produced by activated $CD4^+$ T cells (191) and impaired via downregulation of specific activating surface receptors (e.g., NKG2D), cytotoxic granules (e.g., Perforin and Granzyme B), natural cytotoxicity receptors (NCR), or upregulation of MMP-9 and Igγ-1 chain C region (54,192,193). Targeting these molecular markers can partially restored NK function and retard tumor growth.

Dendritic cells

Dendritic cells (DCs) serve as specific antigen-presenting cells to pick up antigens from damaged tumor cells and then present them with the support of $CD4^+$ Th cells to $CD8^+$ T cells in the lymph nodes, a mechanism named "cross-priming" (153). Tumor-residing $CD103^+$ DC are indispensable to recruit Teff cells into the TME and establish the T cell-inflamed tumor phenotype with a large $CD8^+$ T cell infiltration (35). The establishment of non-T-cell-inflamed tumors may due to deficiencies in T cell priming for lack of cross-presenting dendritic cells, which is believed due to increased CXCL1 in tumors in a c-Myc-dependent manner (34). It was reported that increased IL-10 concentration limited antitumor cytotoxic T-cell responses and activation of NK cells during therapy by suppressing intratumoral DC production of IL-12 (28). One study showed that following three injections of the lipid-protamine-DNA (LPD) nanoparticles loaded with trap genes (IL-10 trap and CXCL12 trap), tumor growth was reduced, prolonged host survival was achieved with significantly reduced immunosuppressive cells such as M2 macrophages, MDSCs, and $PD\text{-}L1^+$ cells (28). Some agonists like CpG, ISCOM vaccines, and CD40 agonist could also enhance antigen uptake and antigen processing by DCs (33,36,37,65).

Noting that DCs carrying antigens need to migrate from the tumor to the lymph nodes to activate $CD8^+$T cells. IL-8 produced by pancreatic cancer cells prevents the journey induced by MIP-3β since DC uniformly express both IL-8 receptors CXCR1 and CXCR2 (38,194). Perhaps for this reason the quantity of DCs was significantly decreased in peripheral blood of PDAC patients (195).

CCL2, a chemokine that dramatically upregulated in pancreatic cancer cells, recruits various immunosuppressive cells such as MDSCs (196). Cooperatively, CCL2 and LCN2 in combination stimulate the generation of immunoregulatory DCs (DCreg), which have suppressive activity due to lowered expression of costimulatory molecules such as HLA-DR (39). DCreg subsequently induces immunosuppressive Treg cells, and eventually impair tumor-specific CTL function (39).

Perspectives and conclusion

Overall, pancreatic cancer immunotherapy aims to promote cytotoxic $CD8^+$ T cell recruitment to pancreatic cancer and their killing effects against cancer cells. Low infiltration of $CD8^+$ T cells is primarily due to the physical barriers of tumor stroma and inhibitory cytokines and chemokines within the TME. Lack of TILs' cytotoxicity may be due to decreased antigenicity in tumor cells, inhibition of immune checkpoints, direct effect of inhibitory cells, or effect of cytokines. It is also possible due to ineffective activation of TILs, because of function limit of DCs and poor specificity among TILs. Strategies to relieve immunosuppression include depleting inhibitory cells or suppressing their function, blocking signal transduction pathways, redirecting the relevant immune cells from pro-tumor status to anti-tumor status, relieving immune checkpoint suppression, and promoting TILs to resist cancer cells.

Although immunotherapy emerges as a promising therapeutic option for pancreatic cancer, it still has not achieved significant and satisfactory improvement on the survival of patients. Crosstalk among various cells forms an intricate network that mediates immune evasion in pancreatic cancer microenvironment. Single therapy to date seems powerless to break through the constraint of immunosuppression, leaving combination therapies a

mission and hope. In consideration of inevitable side effects, identification of specific targets and development of non-overlapping treatments may provide insights to improve efficacy and reduce adverse effects. Finally, thorough understanding of immunosuppression and enhancing of TILs' anti-tumor specificity and lethality are constant topics and avenues for conquering pancreatic cancer with immunotherapy.

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References

1. Lu C, Paschall AV, Shi H, et al. The MLL1-H3K4me3 Axis-Mediated PD-L1 Expression and Pancreatic Cancer Immune Evasion. Journal of the National Cancer Institute 2017;109:djw283.
2. Liu X, Shin N, Koblish HK, et al. Selective inhibition of IDO1 effectively regulates mediators of antitumor immunity. Blood 2010;115:3520-30.
3. Koblish HK, Hansbury MJ, Bowman KJ, et al. Hydroxyamidine inhibitors of indoleamine-2,3-dioxygenase potently suppress systemic tryptophan catabolism and the growth of IDO-expressing tumors. Mol Cancer Ther 2010;9:489-98.
4. Lin X, Huang M, Xie F, et al. Gemcitabine inhibits immune escape of pancreatic cancer by down regulating the soluble ULBP2 protein. Oncotarget 2016;7:70092-9.
5. Ludwig KF, Du W, Sorrelle NB, et al. Small-Molecule Inhibition of Axl Targets Tumor Immune Suppression and Enhances Chemotherapy in Pancreatic Cancer. Cancer Res 2018;78:246-55.
6. Hinz S, Pagerols-Raluy L, Oberg HH, et al. Foxp3 expression in pancreatic carcinoma cells as a novel mechanism of immune evasion in cancer. Cancer Res 2007;67:8344-50.
7. Jiang H, Hegde S, Knolhoff BL, et al. Targeting focal adhesion kinase renders pancreatic cancers responsive to checkpoint immunotherapy. Nat Med 2016;22:851-60.
8. Nomi T, Sho M, Akahori T, et al. Clinical significance and therapeutic potential of the programmed death-1 ligand/programmed death-1 pathway in human pancreatic cancer. Clin Cancer Res 2007;13:2151-7.
9. Liu J, Hamrouni A, Wolowiec D, et al. Plasma cells from multiple myeloma patients express B7-H1 (PD-L1) and increase expression after stimulation with IFN-γ and TLR ligands via a MyD88-, TRAF6-, and MEK-dependent pathway. Blood 2007;110:296-304.
10. Hutcheson J, Balaji U, Porembka MR, et al. Immunologic and Metabolic Features of Pancreatic Ductal Adenocarcinoma Define Prognostic Subtypes of Disease. Clin Cancer Res 2016;22:3606-17.
11. Wiedemann GM, Knott MM, Vetter VK, et al. Cancer cell-derived IL-1α induces CCL22 and the recruitment of regulatory T cells. Oncoimmunology 2016;5:e1175794.
12. Radfar S, Davrinche C, Hollande E. Serial in vivo loss and in vitro gain of Fas expression and function in human cancerous pancreatic duct cells. Int J Cancer 2005;115:214-23.
13. Miyashita T, Miki K, Kamigaki T, et al. Low-dose gemcitabine induces major histocompatibility complex class I-related chain A/B expression and enhances an antitumor innate immune response in pancreatic cancer. Clin Exp Med 2017;17:19-31.

14. Qian Y, Hong B, Shen L, et al. B7-H4 enhances oncogenicity and inhibits apoptosis in pancreatic cancer cells. Cell Tissue Res 2013;353:139-51.
15. Luedke E, Jaime-Ramirez AC, Bhave N, et al. Monoclonal antibody therapy of pancreatic cancer with cetuximab: potential for immune modulation. J Immunother 2012;35:367-73.
16. Liu WM, Fowler DW, Smith P, et al. Pre-treatment with chemotherapy can enhance the antigenicity and immunogenicity of tumours by promoting adaptive immune responses. Br J Cancer 2010;102:115-23.
17. Zhang Y, Velez-Delgado A, Mathew E, et al. Myeloid cells are required for PD-1/PD-L1 checkpoint activation and the establishment of an immunosuppressive environment in pancreatic cancer. Gut 2017;66:124-36.
18. Song J, Lee J, Kim J, et al. Pancreatic adenocarcinoma up-regulated factor (PAUF) enhances the accumulation and functional activity of myeloid-derived suppressor cells (MDSCs) in pancreatic cancer. Oncotarget 2016;7:51840-53.
19. Rodriguez PC, Hernandez CP, Quiceno D, et al. Arginase I in myeloid suppressor cells is induced by COX-2 in lung carcinoma. J Exp Med 2005;202:931-9.
20. Serafini P, Meckel K, Kelso M, et al. Phosphodiesterase-5 inhibition augments endogenous antitumor immunity by reducing myeloid-derived suppressor cell function. J Exp Med 2006;203:2691-702.
21. Stromnes IM, Brockenbrough JS, Izeradjene K, et al. Targeted depletion of an MDSC subset unmasks pancreatic ductal adenocarcinoma to adaptive immunity. Gut 2014;63:1769-81.
22. Long KB, Gladney WL, Tooker GM, et al. IFNγ and CCL2 Cooperate to Redirect Tumor-Infiltrating Monocytes to Degrade Fibrosis and Enhance Chemotherapy Efficacy in Pancreatic Carcinoma. Cancer Discov 2016;6:400-13.
23. Klug F, Prakash H, Huber PE, et al. Low-dose irradiation programs macrophage differentiation to an iNOS(+)/M1 phenotype that orchestrates effective T cell immunotherapy. Cancer Cell 2013;24:589-602.
24. Cullis J, Siolas D, Avanzi A, et al. Macropinocytosis of Nab-paclitaxel Drives Macrophage Activation in Pancreatic Cancer. Cancer Immunol Res 2017;5:182-90.
25. Wang W, Marinis JM, Beal AM, et al. RIP1 Kinase Drives Macrophage-Mediated Adaptive Immune Tolerance in Pancreatic Cancer. Cancer Cell 2018;34:757-74.e7.
26. Daley D, Mani VR, Mohan N, et al. Dectin 1 activation on macrophages by galectin 9 promotes pancreatic carcinoma and peritumoral immune tolerance. Nat Med 2017;23:556-67.
27. Seifert L, Werba G, Tiwari S, et al. The necrosome promotes pancreatic oncogenesis via CXCL1 and Mincle-induced immune suppression. Nature 2016;532:245-9.
28. Shen L, Li J, Liu Q, et al. Local Blockade of Interleukin 10 and C-X-C Motif Chemokine Ligand 12 with Nano-Delivery Promotes Antitumor Response in Murine Cancers. ACS Nano 2018;12:9830-41.
29. Germano G, Frapolli R, Belgiovine C, et al. Role of macrophage targeting in the antitumor activity of trabectedin. Cancer Cell 2013;23:249-62.
30. Schupp J, Krebs FK, Zimmer N, et al. Targeting myeloid cells in the tumor sustaining microenvironment. Cell Immunol 2017. [Epub ahead of print].
31. Di Maio M, Gridelli C, Gallo C, et al. Chemotherapy-induced neutropenia and treatment efficacy in advanced non-small-cell lung cancer: a pooled analysis of three randomised trials. Lancet Oncol 2005;6:669-77.
32. Incio J, Liu H, Suboj P, et al. Obesity-Induced Inflammation and Desmoplasia Promote Pancreatic Cancer Progression and Resistance to Chemotherapy. Cancer Discovery 2016;6:852-69.
33. Serba S, Schmidt J, Wentzensen N, et al. Transfection with CD40L induces tumour suppression by dendritic cell activation in an orthotopic mouse model of pancreatic adenocarcinoma. Gut 2008;57:344-51.
34. Horton B, Spranger S. A Tumor Cell-Intrinsic Yin-Yang Determining Immune Evasion. Immunity 2018;49:11-3.
35. Spranger S, Dai D, Horton B, et al. Tumor-Residing Batf3 Dendritic Cells Are Required for Effector T Cell Trafficking and Adoptive T Cell Therapy. Cancer Cell 2017;31:711-723.e4.
36. Drane D, Gittleson C, Boyle J, et al. ISCOMATRIX adjuvant for prophylactic and therapeutic vaccines. Expert Rev Vaccines 2007;6:761-72.
37. Krieg AM. Development of TLR9 agonists for cancer therapy. J Clin Invest 2007;117:1184-94.
38. Feijoó E, Alfaro C, Mazzolini G, et al. Dendritic cells delivered inside human carcinomas are sequestered by interleukin-8. Int J Cancer 2005;116:275-81.
39. Kudo-Saito C, Shirako H, Ohike M, et al. CCL2 is critical for immunosuppression to promote cancer metastasis. Clin Exp Metastasis 2013;30:393-405.
40. Feig C, Jones JO, Kraman M, 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;110:20212-7.

41. Martínez-Bosch N, Navarro P. Targeting Galectin-1 in pancreatic cancer: immune surveillance on guard. Oncoimmunology 2014;3:e952201.
42. De Monte L, Reni M, Tassi E, et al. Intratumor T helper type 2 cell infiltrate correlates with cancer-associated fibroblast thymic stromal lymphopoietin production and reduced survival in pancreatic cancer. J Exp Med 2011;208:469-78.
43. Goehrig D, Nigri J, Samain R, et al. Stromal protein βig-h3 reprogrammes tumour microenvironment in pancreatic cancer. Gut 2019;68:693-707.
44. Mace TA, Shakya R, Pitarresi JR, et al., IL-6 and PD-L1 antibody blockade combination therapy reduces tumour progression in murine models of pancreatic cancer. Gut 2018;67:320-32.
45. Kraman, M., et al. Suppression of antitumor immunity by stromal cells expressing fibroblast activation protein-alpha. Science 2010;330:827-30.
46. Özdemir BC, Pentcheva-Hoang T, Carstens JL, et al. Depletion of carcinoma-associated fibroblasts and fibrosis induces immunosuppression and accelerates pancreas cancer with reduced survival. Cancer Cell 2014;25:719-34.
47. Zhao J, Xiao Z, Li T, et al. Stromal Modulation Reverses Primary Resistance to Immune Checkpoint Blockade in Pancreatic Cancer. ACS Nano 2018;12:9881-93.
48. Tan MC, Goedegebuure PS, Belt BA, et al. Disruption of CCR5-dependent homing of regulatory T cells inhibits tumor growth in a murine model of pancreatic cancer. J Immunol 2009;182:1746-55.
49. Jacobs C, Duewell P, Heckelsmiller K, et al. An ISCOM vaccine combined with a TLR9 agonist breaks immune evasion mediated by regulatory T cells in an orthotopic model of pancreatic carcinoma. Int J Cancer 2011;128:897-907.
50. Grage-Griebenow E, Jerg E, Gorys A, et al. L1CAM promotes enrichment of immunosuppressive T cells in human pancreatic cancer correlating with malignant progression. Mol Oncol 2014;8:982-97.
51. Demaria O, De Gassart A, Coso S, et al. STING activation of tumor endothelial cells initiates spontaneous and therapeutic antitumor immunity. Proc Natl Acad Sci USA 2015;112:15408-13.
52. Byers JT, Paniccia A, Kaplan J, et al. Expression of the Novel Costimulatory Molecule B7-H5 in Pancreatic Cancer. Ann Surg Oncol 2015;22 Suppl 3:S1574-9.
53. Peng YP, Zhang JJ, Liang WB, et al. Elevation of MMP-9 and IDO induced by pancreatic cancer cells mediates natural killer cell dysfunction. BMC Cancer 2014;14:738.
54. Li X, Ni R, Chen J, et al. The presence of IGHG1 in human pancreatic carcinomas is associated with immune evasion mechanisms. Pancreas 2011;40:753-61.
55. Principe DR, DeCant B, Mascariñas E, et al. TGFβ Signaling in the Pancreatic Tumor Microenvironment Promotes Fibrosis and Immune Evasion to Facilitate Tumorigenesis. Cancer Res 2016;76:2525-39.
56. Olive KP, Jacobetz MA, Davidson CJ, et al. Inhibition of Hedgehog signaling enhances delivery of chemotherapy in a mouse model of pancreatic cancer. Science 2009;324:1457-61.
57. Provenzano PP, Cuevas C, Chang AE, et al. Enzymatic targeting of the stroma ablates physical barriers to treatment of pancreatic ductal adenocarcinoma. Cancer Cell 2012;21:418-29.
58. Feig C, Gopinathan A, Neesse A, et al. The pancreas cancer microenvironment. Clin Cancer Res 2012;18:4266-76.
59. Hingorani SR, Petricoin EF, Maitra A, et al. Preinvasive and invasive ductal pancreatic cancer and its early detection in the mouse. Cancer Cell 2003;4:437-50.
60. Michl P, Krug S. Overcoming immune evasion in pancreatic cancer: the combination matters. Gut 2018;67:997-9.
61. Clark CE, Hingorani SR, Mick R, et al. Dynamics of the immune reaction to pancreatic cancer from inception to invasion. Cancer Res 2007;67:9518-27.
62. Zhang Y, Yan W, Mathew E, et al. CD4+ T lymphocyte ablation prevents pancreatic carcinogenesis in mice. Cancer Immunol Res 2014;2:423-35.
63. McAllister F, Bailey JM, Alsina J, et al. Oncogenic Kras activates a hematopoietic-to-epithelial IL-17 signaling axis in preinvasive pancreatic neoplasia. Cancer Cell 2014;25:621-37.
64. Alizadeh D, Larmonier N. Chemotherapeutic targeting of cancer-induced immunosuppressive cells. Cancer Res 2014;74:2663-8.
65. Winograd R, Byrne KT, Evans RA, et al. Induction of T-cell Immunity Overcomes Complete Resistance to PD-1 and CTLA-4 Blockade and Improves Survival in Pancreatic Carcinoma. Cancer Immunol Res 2015;3:399-411.
66. Bronte V, G Tortora. Adipocytes and Neutrophils Give a Helping Hand to Pancreatic Cancers. Cancer Discovery 2016;6:821-3.
67. Baek G, Tse YF, Hu Z, et al. MCT4 defines a glycolytic subtype of pancreatic cancer with poor prognosis and unique metabolic dependencies. Cell Rep 2014;9:2233-49.
68. Chang CH, Qiu J, O'Sullivan D, et al. Metabolic

Competition in the Tumor Microenvironment Is a Driver of Cancer Progression. Cell 2015;162:1229-41.
69. Huang SC, Everts B, Ivanova Y, et al. Cell-intrinsic lysosomal lipolysis is essential for alternative activation of macrophages. Nat Immunol 2014;15:846-55.
70. Michalek RD, Gerriets VA, Jacobs SR, et al. Cutting edge: distinct glycolytic and lipid oxidative metabolic programs are essential for effector and regulatory CD4+ T cell subsets. J Immunol 2011;186:3299-303.
71. Colegio OR, Chu NQ, Szabo AL, et al. Functional polarization of tumour-associated macrophages by tumour-derived lactic acid. Nature 2014;513:559-63.
72. Pandha H, Rigg A, John J, et al. Loss of expression of antigen-presenting molecules in human pancreatic cancer and pancreatic cancer cell lines. Clin Exp Immunol 2007;148:127-35.
73. von Bernstorff W, Spanjaard RA, Chan AK, et al. Pancreatic cancer cells can evade immune surveillance via nonfunctional Fas (APO-1/CD95) receptors and aberrant expression of functional Fas ligand. Surgery 1999;125:73-84.
74. Whiteside TL. Tumor-induced death of immune cells: its mechanisms and consequences. Semin Cancer Biol 2002;12:43-50.
75. Joyce JA, Fearon DT. T cell exclusion, immune privilege, and the tumor microenvironment. Science 2015;348:74-80.
76. Geismann C, Morscheck M, Koch D, et al. Up-regulation of L1CAM in pancreatic duct cells is transforming growth factor beta1- and slug-dependent: role in malignant transformation of pancreatic cancer. Cancer Res 2009;69:4517-26.
77. Xu H, Chen X, Tao M, et al. B7-H3 and B7-H4 are independent predictors of a poor prognosis in patients with pancreatic cancer. Oncol Lett 2016;11:1841-6.
78. Sica GL, Choi IH, Zhu G, et al. B7-H4, a molecule of the B7 family, negatively regulates T cell immunity. Immunity 2003;18:849-61.
79. Miyatake T, Tringler B, Liu W, et al. B7-H4 (DD-O110) is overexpressed in high risk uterine endometrioid adenocarcinomas and inversely correlated with tumor T-cell infiltration. Gynecol Oncol 2007;106:119-27.
80. Yao Y, Ye H, Qi Z, et al. B7-H4(B7x)-Mediated Cross-talk between Glioma-Initiating Cells and Macrophages via the IL6/JAK/STAT3 Pathway Lead to Poor Prognosis in Glioma Patients. Clin Cancer Res 2016; 22:2778-90.
81. Kryczek I, Wei S, Zhu G, et al. Relationship between B7-H4, regulatory T cells, and patient outcome in human ovarian carcinoma. Cancer Res 2007;67:8900-5.
82. Chen X, Wang W, Man H, et al. Increased B7-H4 expression during esophageal squamous cell carcinogenesis is associated with IL-6/STAT3 signaling pathway activation in mice. Oncol Lett 2017;13:2207-15.
83. Fantini MC, Becker C, Monteleone G, et al. Cutting edge: TGF-beta induces a regulatory phenotype in CD4+CD25- T cells through Foxp3 induction and down-regulation of Smad7. J Immunol 2004;172:5149-53.
84. Coffer PJ, Burgering BM. Forkhead-box transcription factors and their role in the immune system. Nat Rev Immunol 2004;4:889-99.
85. Liyanage UK, Goedegebuure PS, Moore TT, et al. Increased prevalence of regulatory T cells (Treg) is induced by pancreas adenocarcinoma. J Immunother 2006;29:416-24.
86. Ghiringhelli F, Puig PE, Roux S, et al. Tumor cells convert immature myeloid dendritic cells into TGF-beta-secreting cells inducing CD4+CD25+ regulatory T cell proliferation. J Exp Med 2005;202:919-29.
87. Principe DR, Doll JA, Bauer J, et al. TGF-β: duality of function between tumor prevention and carcinogenesis. J Natl Cancer Inst 2014;106:djt369.
88. Apte MV, Haber PS, Darby SJ, et al. Pancreatic stellate cells are activated by proinflammatory cytokines: implications for pancreatic fibrogenesis. Gut 1999;44:534-41.
89. Thomas DA, Massague J. TGF-beta directly targets cytotoxic T cell functions during tumor evasion of immune surveillance. Cancer Cell 2005;8:369-80.
90. Rousalova I, Krepela E. Granzyme B-induced apoptosis in cancer cells and its regulation (review). Int J Oncol 2010;37:1361-78.
91. Cullen SP, Brunet M, Martin SJ. Granzymes in cancer and immunity. Cell Death Differ 2010;17:616-23.
92. Sahin IH, Askan G, Hu ZI, et al. Immunotherapy in pancreatic ductal adenocarcinoma: an emerging entity? Ann Oncol 2017;28:2950-61.
93. Anderson KG, Stromnes IM, Greenberg PD. Obstacles Posed by the Tumor Microenvironment to T cell Activity: A Case for Synergistic Therapies. Cancer Cell 2017;31:311-25.
94. Kalluri R. The biology and function of fibroblasts in cancer. Nat Rev Cancer 2016;16:582-98.
95. Ding Y, Xu D, Feng G., et al. Mesenchymal stem cells prevent the rejection of fully allogenic islet grafts by the immunosuppressive activity of matrix metalloproteinase-2 and -9. Diabetes 2009;58:1797-806.
96. Lee BK, Kim MJ, Jang HS, et al. A high concentration of

MMP-2/gelatinase A and MMP-9/gelatinase B reduce NK cell-mediated cytotoxicity against an oral squamous cell carcinoma cell line. In Vivo 2008;22:593-7.
97. Tang D, Wang D, Yuan Z, et al. Persistent activation of pancreatic stellate cells creates a microenvironment favorable for the malignant behavior of pancreatic ductal adenocarcinoma. Int J Cancer 2013;132:993-1003.
98. Jesnowski R, Fürst D, Ringel J, et al. Immortalization of pancreatic stellate cells as an in vitro model of pancreatic fibrosis: deactivation is induced by matrigel and N-acetylcysteine. Lab Invest 2005;85:1276-91.
99. Wehr AY, Furth EE, Sangar V et al. Analysis of the human pancreatic stellate cell secreted proteome. Pancreas 2011;40:557-66.
100. Fu Y, Liu S, Zeng S et al. The critical roles of activated stellate cells-mediated paracrine signaling, metabolism and onco-immunology in pancreatic ductal adenocarcinoma. Mol Cancer 2018;17:62.
101. Puré E, Lo A. Can Targeting Stroma Pave the Way to Enhanced Antitumor Immunity and Immunotherapy of Solid Tumors? Cancer Immunol Res 2016;4:269-78.
102. Liu Q, Liao Q, Zhao Y. Chemotherapy and tumor microenvironment of pancreatic cancer. Cancer Cell Int 2017;17:68.
103. Iovanna JL, Closa D. Factors released by the tumor far microenvironment are decisive for pancreatic adenocarcinoma development and progression. Oncoimmunology 2017;6:e1358840.
104. Kleeff J, Beckhove P, Esposito I, et al. Pancreatic cancer microenvironment. Int J Cancer 2007;121:699-705.
105. Esposito I, Menicagli M, Funel N, et al., Inflammatory cells contribute to the generation of an angiogenic phenotype in pancreatic ductal adenocarcinoma. J Clin Pathol 2004;57:630-6.
106. Tsukamoto H, Nishikata R, Senju S, et al. Myeloid-derived suppressor cells attenuate TH1 development through IL-6 production to promote tumor progression. Cancer Immunol Res 2013;1:64-76.
107. Schietinger A, Greenberg PD. Tolerance and exhaustion: defining mechanisms of T cell dysfunction. Trends Immunol 2014;35:51-60.
108. Mohammed S, Sukumaran S, Bajgain P, et al. Improving Chimeric Antigen Receptor-Modified T Cell Function by Reversing the Immunosuppressive Tumor Microenvironment of Pancreatic Cancer. Mol Ther 2017;25:249-58.
109. Ene-Obong A, Clear AJ, Watt J, et al. Activated pancreatic stellate cells sequester CD8+ T cells to reduce their infiltration of the juxtatumoral compartment of pancreatic ductal adenocarcinoma. Gastroenterology 2013;145:1121-32.
110. Heinemann V, Reni M, Ychou M, et al. Tumour-stroma interactions in pancreatic ductal adenocarcinoma: rationale and current evidence for new therapeutic strategies. Cancer Treat Rev 2014;40:118-28.
111. Zhang Y., Ertl HC. Depletion of FAP+ cells reduces immunosuppressive cells and improves metabolism and functions CD8+T cells within tumors. Oncotarget 2016;7:23282-99.
112. Lee JJ, Perera RM, Wang H, et al., Stromal response to Hedgehog signaling restrains pancreatic cancer progression. Proc Natl Acad Sci USA 2014;111:E3091-100.
113. Guo S, Contratto M, Miller G, et al. Immunotherapy in pancreatic cancer: Unleash its potential through novel combinations. World J Clin Oncol 2017;8:230-40.
114. Hingorani SR, Harris WP, Beck JT, et al. Phase Ib Study of PEGylated Recombinant Human Hyaluronidase and Gemcitabine in Patients with Advanced Pancreatic Cancer. Clin Cancer Res 2016;22:2848-54.
115. Skelton RA, Javed A, Zheng L, et al., Overcoming the resistance of pancreatic cancer to immune checkpoint inhibitors. J Surg Oncol 2017;116:55-62.
116. Gabrilovich DI, Ostrand-Rosenberg S, Bronte V. Coordinated regulation of myeloid cells by tumours. Nat Rev Immunol 2012;12:253-68.
117. Daley D, Zambirinis CP, Seifert L, et al. γδ T Cells Support Pancreatic Oncogenesis by Restraining αβ T Cell Activation. Cell 2016;166:1485-1499.e15.
118. Ochi A, Nguyen AH, Bedrosian AS, et al. MyD88 inhibition amplifies dendritic cell capacity to promote pancreatic carcinogenesis via Th2 cells. J Exp Med 2012;209:1671-87.
119. Nywening TM, Belt BA, Cullinan DR, et al. Targeting both tumour-associated CXCR2+ neutrophils and CCR2+ macrophages disrupts myeloid recruitment and improves chemotherapeutic responses in pancreatic ductal adenocarcinoma. Gut 2018;67:1112-3.
120. Mantovani A, Sozzani S, Locati M, et al. Macrophage polarization: tumor-associated macrophages as a paradigm for polarized M2 mononuclear phagocytes. Trends Immunol 2002;23:549-55.
121. Wang YC, He F, Feng F, et al. Notch signaling determines the M1 versus M2 polarization of macrophages in antitumor immune responses. Cancer Res 2010;70:4840-9.
122. Slegtenhorst BR, Dor FJ, Elkhal A, et al. Mechanisms and consequences of injury and repair in older organ

transplants. Transplantation 2014;97:1091-9.
123. Sica A, Mantovani A. Macrophage plasticity and polarization: in vivo veritas. J Clin Invest 2012;122:787-95.
124. Cui R, Yue W, Lattime EC, et al. Targeting tumor-associated macrophages to combat pancreatic cancer. Oncotarget 2016;7:50735-54.
125. Kurahara H, Shinchi H, Mataki Y, et al. Significance of M2-polarized tumor-associated macrophage in pancreatic cancer. J Surg Res 2011;167:e211-9.
126. Ino Y, Yamazaki-Itoh R, Shimada K, et al. Immune cell infiltration as an indicator of the immune microenvironment of pancreatic cancer. Br J Cancer 2013;108:914-23.
127. Beatty GL, Chiorean EG, Fishman MP, et al. CD40 agonists alter tumor stroma and show efficacy against pancreatic carcinoma in mice and humans. Science 2011;331:1612-6.
128. Ding A, Sanchez E, Nathan CF. Taxol shares the ability of bacterial lipopolysaccharide to induce tyrosine phosphorylation of microtubule-associated protein kinase. J Immunol 1993;151:5596-602.
129. Ding AH, Porteu F, Sanchez E, et al. Shared actions of endotoxin and taxol on TNF receptors and TNF release. Science 1990;248:370-2.
130. Perera PY, Mayadas TN, Takeuchi O, et al. CD11b/CD18 acts in concert with CD14 and Toll-like receptor (TLR) 4 to elicit full lipopolysaccharide and taxol-inducible gene expression. J Immunol 2001;166:574-81.
131. Parker KH, Beury DW, Ostrand-Rosenberg S. Myeloid-Derived Suppressor Cells: Critical Cells Driving Immune Suppression in the Tumor Microenvironment. Adv Cancer Res 2015;128:95-139.
132. Meyer C, Sevko A, Ramacher M, et al. Chronic inflammation promotes myeloid-derived suppressor cell activation blocking antitumor immunity in transgenic mouse melanoma model. Proc Natl Acad Sci USA 2011;108:17111-6.
133. Ostrand-Rosenberg S. Myeloid-derived suppressor cells: more mechanisms for inhibiting antitumor immunity. Cancer Immunol Immunother 2010;59:1593-600.
134. Wörmann SM, Diakopoulos KN, Lesina M, et al. The immune network in pancreatic cancer development and progression. Oncogene 2014;33:2956-67.
135. Waugh DJ, Wilson C. The interleukin-8 pathway in cancer. Clin Cancer Res 2008;14:6735-41.
136. Bayne LJ, Beatty GL, Jhala N, et al. Tumor-derived granulocyte-macrophage colony-stimulating factor regulates myeloid inflammation and T cell immunity in pancreatic cancer. Cancer Cell 2012;21:822-35.
137. Delitto D, Wallet SM, Hughes SJ. Targeting tumor tolerance: A new hope for pancreatic cancer therapy? Pharmacol Ther 2016;166:9-29.
138. Marigo I, Bosio E, Solito S, et al. Tumor-induced tolerance and immune suppression depend on the C/EBPbeta transcription factor. Immunity 2010;32:790-802.
139. Zhang J, Xu X, Shi M, et al. CD13(hi) Neutrophil-like myeloid-derived suppressor cells exert immune suppression through Arginase 1 expression in pancreatic ductal adenocarcinoma. Oncoimmunology 2017;6:e1258504.
140. Hori S, Nomura T, Sakaguchi S. Control of regulatory T cell development by the transcription factor Foxp3. Science 2003;299:1057-61.
141. Fontenot JD, Rasmussen JP, Williams LM, et al. Regulatory T cell lineage specification by the forkhead transcription factor foxp3. Immunity 2005;22:329-41.
142. Chellappa S, Hugenschmidt H, Hagness M, et al. Regulatory T cells that co-express RORγt and FOXP3 are pro-inflammatory and immunosuppressive and expand in human pancreatic cancer. Oncoimmunology 2015;5:e1102828.
143. Hiraoka N, Onozato K, Kosuge T, et al. Prevalence of FOXP3+ regulatory T cells increases during the progression of pancreatic ductal adenocarcinoma and its premalignant lesions. Clin Cancer Res 2006;12:5423-34.
144. Nicholaou T, Ebert LM, Davis ID, et al. Regulatory T-cell-mediated attenuation of T-cell responses to the NY-ESO-1 ISCOMATRIX vaccine in patients with advanced malignant melanoma. Clin Cancer Res 2009;15:2166-73.
145. Roshani R, McCarthy F, Hagemann T. Inflammatory cytokines in human pancreatic cancer. Cancer Lett 2014;345:157-63.
146. Salem ML. Estrogen, a double-edged sword: modulation of TH1- and TH2-mediated inflammations by differential regulation of TH1/TH2 cytokine production. Curr Drug Targets Inflamm Allergy 2004;3:97-104.
147. Tassi E, Gavazzi F, Albarello L, et al. Carcinoembryonic antigen-specific but not antiviral CD4+ T cell immunity is impaired in pancreatic carcinoma patients. J Immunol 2008;181:6595-603.
148. Bellone G, Smirne C, Mauri FA, et al. Cytokine expression profile in human pancreatic carcinoma cells and in surgical specimens: implications for survival. Cancer Immunol Immunother 2006;55:684-98.
149. Fietta P, Delsante G. The effector T helper cell triade. Riv Biol 2009;102:61-74.
150. Nakayamada S, Takahashi H, Kanno Y, et al. Helper

T cell diversity and plasticity. Curr Opin Immunol 2012;24:297-302.
151. Bellone G, Carbone A, Smirne C, et al. Cooperative induction of a tolerogenic dendritic cell phenotype by cytokines secreted by pancreatic carcinoma cells. J Immunol 2006;177:3448-60.
152. Fogar P, Basso D, Fadi E, et al. Pancreatic cancer alters human CD4+ T lymphocyte function: a piece in the immune evasion puzzle. Pancreas 2011;40:1131-7.
153. Duffy AG, Greten TF. Immunological off-target effects of standard treatments in gastrointestinal cancers. Ann Oncol 2014;25:24-32.
154. Gandhi L, Rodríguez-Abreu D, Gadgeel S, et al. Pembrolizumab plus Chemotherapy in Metastatic Non-Small-Cell Lung Cancer. New Eng J Med 2018;378:2078-92.
155. Tumeh PC, Harview CL, Yearley JH, et al. PD-1 blockade induces responses by inhibiting adaptive immune resistance. Nature 2014;515:568-71.
156. Munir S, Andersen GH, Met Ö, et al. HLA-restricted CTL that are specific for the immune checkpoint ligand PD-L1 occur with high frequency in cancer patients. Cancer Res 2013;73:1764-76.
157. Hayes JB, Sircy LM, Heusinkveld LE, et al. Modulation of Macrophage Inflammatory Nuclear Factor κB (NF-κB) Signaling by Intracellular Cryptococcus neoformans. J Biol Chem 2016;291:15614-27.
158. Hirano F, Kaneko K, Tamura H, et al. Blockade of B7-H1 and PD-1 by monoclonal antibodies potentiates cancer therapeutic immunity. Cancer Res 2005;65:1089-96.
159. Peng J, Hamanishi J, Matsumura N, et al. Chemotherapy Induces Programmed Cell Death-Ligand 1 Overexpression via the Nuclear Factor-κB to Foster an Immunosuppressive Tumor Microenvironment in Ovarian Cancer. Cancer Res 2015;75:5034-45.
160. Teng MW, Ngiow SF, Ribas A, et al. Classifying Cancers Based on T-cell Infiltration and PD-L1. Cancer Res 2015;75:2139-45.
161. Norde WJ, Maas F, Hobo W, et al. PD-1/PD-L1 interactions contribute to functional T-cell impairment in patients who relapse with cancer after allogeneic stem cell transplantation. Cancer Res 2011;71:5111-22.
162. Geng L, Huang D, Liu J, et al. B7-H1 up-regulated expression in human pancreatic carcinoma tissue associates with tumor progression. J Cancer Res Clin Oncol 2008;134:1021-7.
163. Marzec M, Zhang Q, Goradia A, et al. Oncogenic kinase NPM/ALK induces through STAT3 expression of immunosuppressive protein CD274 (PD-L1, B7-H1). Proc Natl Acad Sci USA 2008;105:20852-7.
164. Mittendorf EA, Philips AV, Meric-Bernstam F, et al. PD-L1 expression in triple-negative breast cancer. Cancer Immunol Res 2014;2:361-70.
165. Doi T, Ishikawa T, Okayama T, et al. The JAK/STAT pathway is involved in the upregulation of PD-L1 expression in pancreatic cancer cell lines. Oncol Rep 2017;37:1545-54.
166. Shankaran V, Ikeda H, Bruce AT, et al. IFNgamma and lymphocytes prevent primary tumour development and shape tumour immunogenicity. Nature 2001;410:1107-11.
167. Bardhan K, Paschall AV, Yang D, et al. IFNγ Induces DNA Methylation-Silenced GPR109A Expression via pSTAT1/p300 and H3K18 Acetylation in Colon Cancer. Cancer Immunol Res 2015;3:795-805.
168. Topalian SL, Drake CG, Pardoll DM. Immune checkpoint blockade: a common denominator approach to cancer therapy. Cancer Cell 2015;27:450-61.
169. Taube JM, Young GD, McMiller TL, et al. Differential Expression of Immune-Regulatory Genes Associated with PD-L1 Display in Melanoma: Implications for PD-1 Pathway Blockade. Clin Cancer Res 2015;21:3969-76.
170. Limagne E, Euvrard R, Thibaudin M, et al. Accumulation of MDSC and Th17 Cells in Patients with Metastatic Colorectal Cancer Predicts the Efficacy of a FOLFOX-Bevacizumab Drug Treatment Regimen. Cancer Res 2016;76:5241-52.
171. Spranger S, Spaapen RM, Zha Y, et al. Up-regulation of PD-L1, IDO, and T(regs) in the melanoma tumor microenvironment is driven by CD8(+) T cells. Sci Transl Med. 2013;5:200ra116.
172. Topalian SL, Hodi FS, Brahmer JR, et al. Safety, activity, and immune correlates of anti-PD-1 antibody in cancer. N Engl J Med 2012;366:2443-54.
173. Kobold S, Grassmann S, Chaloupka M, et al. Impact of a New Fusion Receptor on PD-1-Mediated Immunosuppression in Adoptive T Cell Therapy. J Natl Cancer Inst 2015;107. doi: 10.1093/jnci/djv146.
174. Lutz ER, Wu AA, Bigelow E, et al. Immunotherapy converts nonimmunogenic pancreatic tumors into immunogenic foci of immune regulation. Cancer Immunol Res 2014;2:616-31.
175. Zitvogel L, Galluzzi L, Smyth MJ, et al. Mechanism of action of conventional and targeted anticancer therapies: reinstating immunosurveillance. Immunity 2013;39:74-88.
176. Brahmer JR, Tykodi SS, Chow LQ, et al. Safety and activity of anti-PD-L1 antibody in patients with advanced

cancer. N Engl J Med 2012;366:2455-65.
177. Arlauckas SP, Garris CS, Kohler RH, et al. In vivo imaging reveals a tumor-associated macrophage-mediated resistance pathway in anti-PD-1 therapy. Sci Transl Med 2017;9. doi: 10.1126/scitranslmed.aal3604.
178. Azad A, Yin Lim S, D'Costa Z, et al. PD-L1 blockade enhances response of pancreatic ductal adenocarcinoma to radiotherapy. EMBO Mol Med 2017;9:167-80.
179. Wang J, Sanmamed MF, Datar I, et al. Fibrinogen-like Protein 1 Is a Major Immune Inhibitory Ligand of LAG-3. Cell 2019;176:334-47.e12.
180. Kim MS, Pinto SM, Getnet D, et al., A draft map of the human proteome. Nature 2014;509:575-81.
181. Anderson AC, Joller N, Kuchroo VK. Lag-3, Tim-3, and TIGIT: Co-inhibitory Receptors with Specialized Functions in Immune Regulation. Immunity 2016;44:989-1004.
182. Meng Q, Liu Z, Rangelova E, et al. Expansion of Tumor-reactive T Cells From Patients With Pancreatic Cancer. J Immunother 2016;39:81-9.
183. Scheper W, Kelderman S, Fanchi LF, et al, Low and variable tumor reactivity of the intratumoral TCR repertoire in human cancers. Nat Med 2019;25:89-94.
184. Duan X, Deng L, Chen X, et al. Clinical significance of the immunostimulatory MHC class I chain-related molecule A and NKG2D receptor on NK cells in pancreatic cancer. Med Oncol 2011;28:466-74.
185. Kiessling R, Klein E, Wigzell H. "Natural" killer cells in the mouse. I. Cytotoxic cells with specificity for mouse Moloney leukemia cells. Specificity and distribution according to genotype. Eur J Immunol 1975;5:112-7.
186. Herberman RB, Nunn ME, Lavrin DH. Natural cytotoxic reactivity of mouse lymphoid cells against syngeneic acid allogeneic tumors. I. Distribution of reactivity and specificity. Int J Cancer 1975;16:216-29.
187. Storkus WJ, Howell DN, Salter RD, et al. NK susceptibility varies inversely with target cell class I HLA antigen expression. J Immunol 1987;138:1657-9.
188. Ljunggren HG, Karre K. In search of the 'missing self': MHC molecules and NK cell recognition. Immunol Today 1990;11:237-44.
189. Somersalo K, Carpen O, Saksela E. Stimulated natural killer cells secrete factors with chemotactic activity, including NAP-1/IL-8, which supports VLA-4- and VLA-5-mediated migration of T lymphocytes. Eur J Immunol 1994;24:2957-65.
190. Bluman EM, Bartynski KJ, Avalos BR, et al. Human natural killer cells produce abundant macrophage inflammatory protein-1 alpha in response to monocyte-derived cytokines. J Clin Invest 1996;97:2722-7.
191. di Carlo E, de Totero D, Piazza T, et al. Role of IL-21 in immune-regulation and tumor immunotherapy. Cancer Immunol Immunother 2007;56:1323-34.
192. Garcia-Iglesias T, Del Toro-Arreola A, Albarran-Somoza B, et al. Low NKp30, NKp46 and NKG2D expression and reduced cytotoxic activity on NK cells in cervical cancer and precursor lesions. BMC Cancer 2009;9:186.
193. Bae S, Oh K, Kim H, et al. The effect of alloferon on the enhancement of NK cell cytotoxicity against cancer via the up-regulation of perforin/granzyme B secretion. Immunobiology 2013;218:1026-33.
194. Rossi D, Zlotnik A. The biology of chemokines and their receptors. Annu Rev Immunol 2000;18:217-42.
195. Tjomsland V, Sandström P, Spångeus A, et al. Pancreatic adenocarcinoma exerts systemic effects on the peripheral blood myeloid and plasmacytoid dendritic cells: an indicator of disease severity? BMC Cancer 2010;10:87.
196. Eruslanov E, Neuberger M, Daurkin I, et al. Circulating and tumor-infiltrating myeloid cell subsets in patients with bladder cancer. Int J Cancer 2012;130:1109-19.

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