

Cytokines and Exocrine Pancreatic Cancer: Is There a Link?

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At the beginning of the third millennium, the prognosis for patients with pancreatic cancer is still extremely poor, with a five-year survival of less than 1% [1] in spite of the availability of sophisticated diagnostic and treatment aids which have, in recent years, significantly modified the prognosis for many patients with solid tumors other than those of pancreatic origin. The failure to improve upon the therapeutical approach for this type of tumor is probably due to the biological behavior of pancreatic cancer cells which acquire several molecular and biochemical advantages in growing, spreading and escaping host control.

The rapid and uncontrolled growth which characterizes the pancreatic cancer cell cycle depends upon many factors, above all, alterations in key genes involved in controlling the cell cycle [2]. More than 90% of pancreatic tumors bear codon 12 *K-ras* point mutations. This frequency, the highest to be reported for any tumor type which has been described in the early phases of pancreatic carcinogenesis, determines the synthesis of an altered p21 protein [3, 4]. Normal p21 shifts from an active state (bound to GTP) to an inactive state (bound to GDP) via its intrinsic GTPase activity, and via its sensitivity to the activity of GAP (GTPase activating protein). The transformed p21 becomes insensitive to GAP thus leading this protein to a constitutive and permanent activation, which stimulates cell growth. Another gene frequently found to be altered in pancreatic cancer is p16 (homozygously deleted in about 40% of pancreatic carcinomas). It is an inhibitor of cyclin-dependent kinase (CDK) 4, which

promotes progression of the cell division cycle through late G1 phase to G1/S [2]. Accelerated pancreatic cancer cell growth is, however, not only due to mutations of *K-ras*, p16 or other genes involved in regulating the cell cycle, but also to an imbalance between stimulatory and inhibitory factors, mainly cytokines. Among the cytokines providing positive signals for pancreatic cancer cell growth, are EGF, IGF I, TGF α , interleukin 1 α [5-13], which originate in peri-tumoral inflammatory cells, but may also be produced by the pancreatic cancer itself thus exerting an autocrine action [9, 14, 15]. To act, all these mediators must first bind to their transmembrane receptors, the majority of which have an intrinsic tyrosine kinase activity which subsequently has a series of intracellular targets. Among these cytokines are the family of mitogen-activated protein kinases (MAPKs) and the extracellular regulated kinases (ERKs) [13, 16, 17].

It has recently been demonstrated that cytokines, TGF α in particular, and mutated *K-ras* may synergistically promote the growth of human pancreatic cells acting on similar, although distinct, signal transduction pathways [18]. This suggests that different alterations of the pancreatic cancer cell may co-operate in favoring cell growth.

A peculiar aspect of tumor cells, pancreatic cancer cells in particular, is their loss of responsiveness to growth inhibitory cytokines, such as TGF β 1 [19, 20]. Pancreatic cancer cells can escape the cell growth inhibitory effect of TGF β 1 since they may bear: 1) cell membrane receptor mutations (TGF β RII), as occurs in many colorectal

cancers [21], or 2) altered Smad proteins which are involved in the signal transduction pathway of TGFbeta1 [22, 23]. The membrane receptor TGFbetaRII, after coupling with TGFbeta1, phosphorylates and activates TGFbetaRI, which then phosphorylates Smad

proteins 2 and 3; the latter can translocate into the nucleus only after coupling with Smad 4, a protein encoded by DPC 4 (deleted in pancreatic carcinoma locus 4), a gene which is frequently deleted in pancreatic cancer (Figure 1).

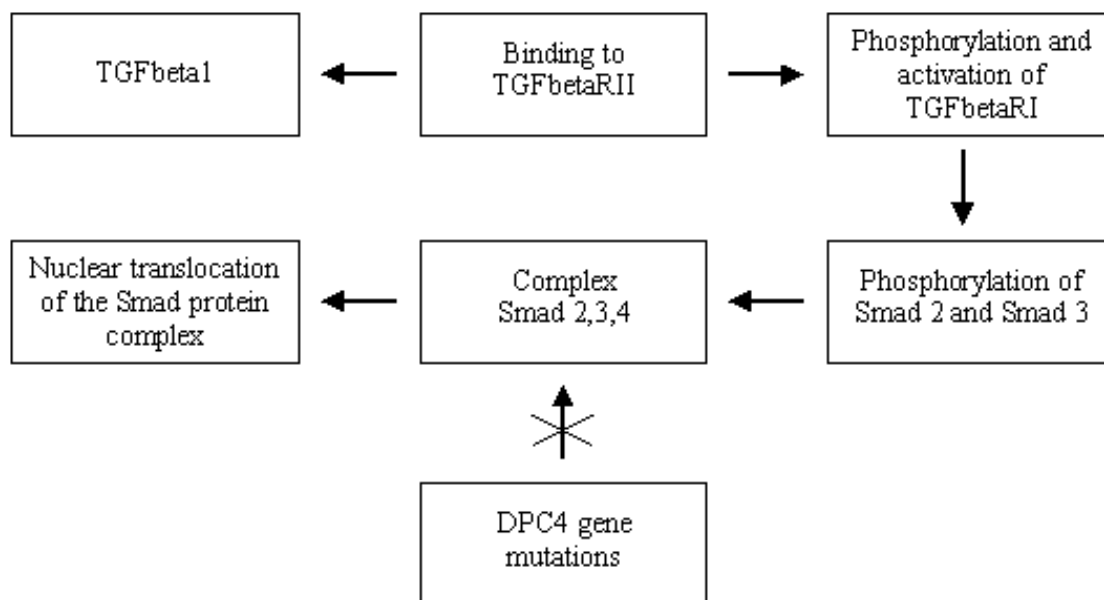


Figure 1. Schematic representation of TGFbeta1 signal transduction pathway via Smad proteins.

The devastating evolution of pancreatic cancer is not only due to the high proliferating potential of pancreatic cancer cells, but also to the ability of these cells to metastasize even when the primary tumor spread is limited. The metastatic process comprises several steps, including the detachment of metastatic cells from the primary tumor, followed by the degradation of the basement membrane and the invasion of lymphatic and/or hematic vessels (intravasation). The growth of metastatic foci in target organs, such as the liver, is preceded by the arrest of metastatic cells in the microvasculature, followed by the degradation of the basement membrane, the invasion of the target organ and the growth of the new metastatic focus (extravasation) [24]. In any of these steps, the interaction between tumor cells and the extracellular matrix plays a key role and different cytokines can enhance or diminish the adhesion of tumor cells to the ECM. They can also modify the membranal expression of ECM ligands, such as CD44 or ICAM 1 [25, 26]. Cytokines may therefore

play a role in favoring or counteracting the metastatic process in pancreatic cancer, as has also been demonstrated for other tumors [27, 28].

Each cytokine can evoke a cascade of events in inflammatory cells, including the synthesis and release of other cytokines. This phenomenon can also be observed in pancreatic cancer cells: the stimulation of PANC-1 cells with TNFalpha causes the production of IL-8 and RANTES by tumor cells [15]. In turn, it has recently been demonstrated that IL-8 renders human pancreatic cancer cells more tumorigenic and metastatic [29]. Each cytokine may thus simultaneously induce a heightening of one specific biological effect (e.g. induction of cell growth) and may trigger a series of different biological responses.

Among the biological effects evoked by cytokines, the stimulation or the inhibition of the immunological host response to tumor cells deserves consideration. The stimulation by cytokines of the host immunological

response to pancreatic cancer cell has recently been considered an aid in improving the outcome of pancreatic cancer patients [30]. In particular, it has been demonstrated in vitro that the tumor-associated transforming growth factor-beta and interleukin 10 favor a Th2-like phenotype [31], while in vivo IL-2 or IL-4 induce an anti-tumor response, even in an animal model without mature T cells (nude mice) [32]. Current research shows that cytokines play a therapeutical role in pancreatic cancer: cytokines gene transfer in

pancreatic cancer cells and treatment with anti-growth factor receptor antibodies [32-34] are now considered a potential strategy for the immune gene-therapy of pancreatic cancer. This may contribute to enhancing the efficacy of the traditional therapy given to patients with this type of neoplasia. Furthermore, tumor cell transfection with antiangiogenic cytokines genes is also considered to be of potential utility in improving pancreatic cancer treatment in the near future [35].

Table 1. Effects of cytokines on pancreatic cancer cell growth (Growth) and angiogenesis. The autocrine production by pancreatic cancer cells and the immunomodulatory (IM) effect in patients with pancreatic cancer are also indicated

Cytokines	Autocrine	Growth	Angiogenesis	IM
IL1	+	+	+	
IL2				+
IL4				+
IL8	+	+	+	+
IL10				+
BFGF			+	
VEGF			+	
EGF	+	+	+	
TNFalpha	+	+	+	
IGF-I	+	+	+	
TGF-beta1	+	=	+	
PDGF	+		+	
RANTES	+			+
MCP-1	+			+
GM-CSF	+			+

+ : stimulatory effect
 = : no effect

Abbreviations bFGF: basic fibroblast growth factor; CDK: cyclin-dependent kinase; DPC 4: deleted in pancreatic carcinoma locus 4; ECM: extra-cellular matrix; EGF: epidermal growth factor; ERKs: extracellular regulated kinases; GAP: GTPase activating protein; GDP: guanosine diphosphate; GM-CSF: granulocyte macrophage colony stimulating factor; GTP: guanosine triphosphate; ICAM 1: intercellular adhesion molecule 1; IGF I: insulin-like growth factor I; IL: interleukin; MAPKs: mitogen-activated protein kinases; MCP-1: monocyte chemo-attractant protein-1; PDGF: platelet derived growth factor; RANTES: regulated on activation, normal T cell

expressed; TGFalpha: transforming growth factor alpha; TGF-beta1: transforming growth factor beta 1; TNFalpha: tumor necrosis factor alpha; VEGF: vascular endothelial growth factor

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