Molecular pathogenesis of pancreatic ductal adenocarcinoma

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Abstract

The statistics are alarming; pancreatic ductal adenocarcinoma (PDA) will be the second leading cause of death amongst all cancers by 2020. More worrisome is that incidence is on the rise, and without more effective cancer control of this disease, the trajectory of the virtually indistinguishable rates of incidence and mortality will remain the reality for years to come. Advances in genomics are beginning to clarify the key issues about the pathogenesis of this aggressive tumour type. New insights into classic pathogenic driver genes, such as *KRAS*, *CDKN2A*, *TP53* and *SMAD4*, are portraying alternative roles for these genes beyond their function at the preneoplastic level including metastatic dissemination and chemoresistance. Clinically relevant molecular subtypes have recently emerged, which will aid oncologists in making more informed treatment decisions to improve outcomes in the future. A wealth of data surrounding these issues has been generated over the last 5 years. Below, we attempt to bring readers up to speed on recent research findings in PDA.

Keywords *CDKN2A*; driver genes; *KRAS*; pancreatic ductal adenocarcinoma; PanIN; *SMAD4*; *TP53*

Introduction

PDA has dismal prognosis with a 5-year survival rate of ~4%.[1,2] 80 to 85% of patients present with advanced disease at the time of diagnosis and with few treatment options at this stage most patients with PDA are essentially relegated to palliative care. The dismal outcomes for PDA have not changed for decades making this disease one of the deadliest tumour types in oncology. Below, we review lessons from recent findings that have contributed to an improved understanding of the pathogenesis of this disease. Much of the studies presented relate to advances in genomics as this work has recently come to fruition from enormous worldwide efforts set out nearly a decade ago.

Many unknowns regarding the fundamental nature of PDA pathogenesis remain unanswered. For example, is the universally accepted paradigm of genetic progression (*KRAS* > *CDKN2A* > *TP53* > *SMAD4*) uniformly applicable to all PDA tumours? What are the other key genetic drivers of this disease beyond these four genes? What processes or etiologies drive mutational accrual of this disease? Beyond the histological subtypes, are there specific molecular subtypes of PDA that exist? Is PDA an intrinsically aggressive tumour type or is the aggressive nature of this disease a result of late diagnosis? Why are PDA exceedingly chemoresistant? Can non-targeted based cancer therapies such as immunotherapy play a role in the treatment of PDA? If so, how do we identify the patients that will benefit from this type of therapy in a prospective manner? Answers to these questions will be of critical importance in the better understanding of the pathogenesis PDA in the future. Some key findings surrounding these topics will be discussed below. Naturally, it is difficult to cover all the topics in detail; however, please refer to the additional references provided below for a more in depth discussion of the aforementioned questions.

Classical PDA genetic drivers

PDA is thought to arise from two major precursor lesions: pancreatic intraepithelial neoplasia (PanIN) and intraductal papillary mucinous neoplasm (IPMN).[3,4] The PanIN nomenclature was first proposed in 2001, where it was suggested to classify PanINs according to a 3-tiered system (PanIN-1a/1b; PanIN-2, PanIN-3) [Figure 1]. The order of mutation of the classic four genes that compose the progression model of PDA (*KRAS* > *CDKN2A* > *TP53* > *SMAD4*) is derived from genetic analysis of PanIN where the increasing histological complexity and dysplasia approximates the extent of tumour progression. Because these genes are universally disrupted in PDA, their mutation is arguably a prerequisite for tumourigenesis. To begin, we provide an overview of their function in PDA.

KRAS

[See Figure 2] *KRAS* (*Kirsten rat sarcoma viral oncogene homologue*) is a member of the RAS superfamily of GTPases involved in a host of cellular functions such as cell growth, differentiation, metabolism, migration and apoptosis. In one form or another, all these cellular functions have gone awry in cancer and thus it is not surprising that *KRAS* mutations are amongst the most frequent in cancer.[5] Around 90% of PDAs harbour mutant *KRAS*. In a normal setting, KRAS can be turned 'on' or 'off' via GTP hydrolysis. 'On' implies GTP is bound. Hydrolysis of GTP into GDP turns the KRAS signal 'off'. In general, mutant *KRAS* is permanently left in the 'on' position unable to hydrolyse GTP efficiently, resulting in an unimpeded growth signal for the cell.

In PDA, three distinct *KRAS* mutations have been described,[6] all of which are located within the protein domain that includes the GTP binding pocket. Ninety percent of mutations are restricted to a single missense base substitution on codon 12 swapping a glycine amino acid residue for a valine (G12V) or aspartate (G12D) residue. Other notable *KRAS* mutations recurrent in PDA are located on codon 13 (G13D − 5%) and codon 61 (Q61R/H − 5%). Interestingly, patients with codon 61 substitutions have a significantly better prognosis when compared to general PDA cohort.[5] Tumour with codon 61 mutations demonstrate weaker ERK activation, a key *RAS* effector molecule, compared to other KRAS mutations suggesting this mutation is a less potent version of mutant *KRAS*.[5]

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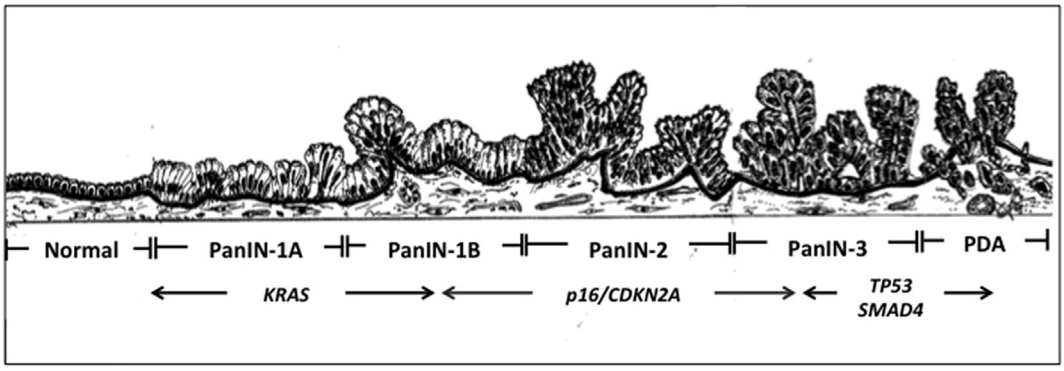

Figure 1 Histologic-genetic pancreatic intraepithelial neoplasia (PanIN) progression model for pancreatic ductal adenocarcinoma (PDA).

It is well accepted that the *KRAS* mutation is the cancer initiating event in PDA and crucial to progression of PanIN. However, to what extent do fully invasive PDAs still rely on this signal? *KRAS* mutant cell lines derived from PDA and lung adenocarcinoma tumours (where *KRAS* mutations are also common) display a variegated pattern of *KRAS* dependency.[7,8] Using RNA interference assays against *KRAS* and measuring cell growth as an experimental outcome, two classes of cell types have been established: those that completely dependent on the *KRAS* signal and those that are unaffected by its loss. Cells that do not depend on mutant *KRAS* have been shown to have hyperactive PI-3 kinase/Akt signalling, which reconciles why they no longer require the *KRAS* signal.[9] On the other hand, additional DNA copies of *KRAS* gene is correlated with increased *KRAS* dependency.[9] Genomics of PDA indicates that up to 40% of tumours show amplification of mutant *KRAS*,[10] suggesting a large subset of PDA may still depend on KRAS for tumour growth and make it an ideal therapeutic target. However, progress towards developing small molecules towards KRAS has proven quite difficult.[11] At present the term 'undruggable' is commonly associated with targeting the KRAS oncoprotein. Alterative areas exploring targeting downstream effectors of the RAS signalling

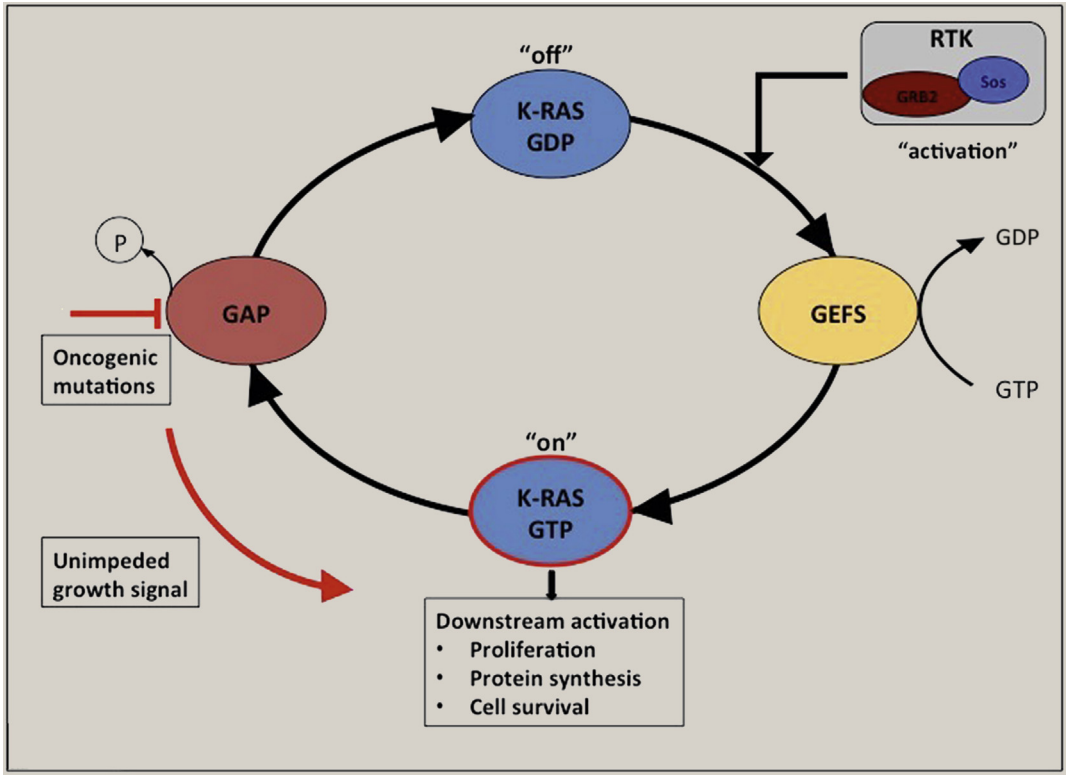

Figure 2 Following the activation of receptor tyrosine kinase (RTK), a complex comprising the activated factor, GRB2 (growth factor receptor-bound protein 2) and Sos (son of sevenless) is formed. The subsequent binding of Sos to K-RAS-GDP results in the exchange of GDP to GTP. This incites a second change, whereby the K-RAS GTP binds to downstream effectors (ie. RAF [rapidly accelerated fibrosarcoma]; PIK3 [phosphoinositide 3-kinase]), resulting in cell proliferation and prolongation of cell survival. In normal wild-type K-RAS, this signal is terminated by hydrolysis of GTP to GDP. However, under the influence of oncogenic mutations, the GTPase activity of KRAS is defective, preventing hydrolysis of GTP. As a consequence, there is a failure in the termination of signalling. (GEF = GTP exchange factor; GAP = GTPase activating proteins).