Frontiers in Bioscience Landmark

Front. Biosci. (Landmark Ed) **2024**; 29(3): 101
https://doi.org/10.31083/j.fbl2903101

Review

Molecular Classification and Pathogenesis of Pancreatic Adenocarcinoma and Targeted Therapies: A Review

Samantha M. Ruff[1], Timothy M. Pawlik[1,*]

[1]Department of Surgery, Division of Surgical Oncology, The Ohio State University Wexner Medical Center and James Comprehensive Cancer Center, Columbus, OH 43210, USA
*Correspondence: tim.pawlik@osumc.edu (Timothy M. Pawlik)
Academic Editor: Marco Falasca
Submitted: 29 October 2023 Revised: 21 December 2023 Accepted: 30 January 2024 Published: 13 March 2024

Abstract

Pancreatic adenocarcinoma (PDAC) is disease with a 5-year survival of only 12%. Many patients with PDAC present with late-stage disease and even early-stage disease can often be characterized by an aggressive tumor biology. Standard therapy for metastatic PDAC consists mainly of chemotherapy regimens like FOLFIRINOX, FOLFOX, or gemcitabine and nab-paclitaxel. Research has focused on sequencing PDAC tumors to understand better the mutational landscape and transcriptomics of PDAC with the goal to develop targeted therapies. Targeted therapies may potentially minimize the toxic risks of chemotherapy and provide a long-term survival benefit. We herein review the underlying molecular pathogenesis of PDAC, as well as the classification schema created from current sequencing data, and recent updates related to targeted therapy for PDAC.

Keywords: pancreatic cancer; targeted therapy; molecular subtypes; next generation sequencing

1. Introduction

Pancreatic adenocarcinoma (PDAC) is associated with a grave prognosis and a 5-year survival of only 12% [1]. Many patients with PDAC present with late stage disease and even early-stage disease can often be characterized by an aggressive tumor biology. The combination of surgical resection with systemic therapy traditionally has offered the best chance at long-term survival. Unfortunately, only about 15–20% of patients present with potentially resectable tumors and even after resection the incidence of recurrence can be as high as 80–85% [2].

Standard of care for metastatic PDAC consists mainly of chemotherapy regimens like FOLFIRINOX, FOLFOX, or gemcitabine and nab-paclitaxel [3]. In the setting of metastatic disease, patients are treated with upfront chemotherapy. However, chemotherapy is also given in the neoadjuvant and/or adjuvant setting based on the features of the primary tumor, concern for micrometastatic spread, and final pathology after surgery [3]. Unfortunately, these therapies are often not associated with a high response rate and have not been associated with dramatic improvements in long-term outcomes [3]. One reason for the lack of efficacy for systemic therapy is the dense desmoplastic stromal tissue that makes up the majority of the tumor microenvironment and may prevent effective delivery of drugs to tumor cells [4]. Additionally, this dense stromal tumor microenvironment is challenging to re-create in the laboratory, making it difficult to delineate which therapies will successfully translate from the bench to the bedside. As such, most research has focused on sequencing PDAC tumors to better understand the mutational landscape and transcriptomics with the goal of developing targeted therapies. Targeted therapies could potentially minimize the toxic risks of chemotherapy and provide a long-term survival benefit. We herein review the underlying molecular biology of PDAC, as well as the classification schema created from current sequencing data, and recent updates related to targeted therapy.

2. Methods

Pubmed, google scholar, and clinicaltrials.gov were utilized for all searches. Original studies, reviews, case reports, meta-analyses, and clinical trials were included and chosen based on the quality of the literature and relevance to the topic. Search words included the following terms and combinations of these terms: “targeted therapy”, “pancreatic cancer”, “pancreatic adenocarcinoma”, “pancreas cancer”, individual targeted pathways, “systemic therapy”, “molecular subtype”, “subtype”, “classification”. Data were reviewed and studies selected by the two authors. Individual data items were not collected, combined, and re-analyzed. The data was reported and interpreted in the larger context of treating PDAC. Bias of the individual studies and heterogeneity between studies was assessed by the authors and included where necessary in the manuscript.

3. Precursor Lesions in PDAC: Updated Classification System

Historically, precursor lesions for PDAC were categorized using a three-tiered classification system (low, intermediate, and high grade) that encompassed intraductal papillary mucinous neoplasms (IPMN), mucinous cystic

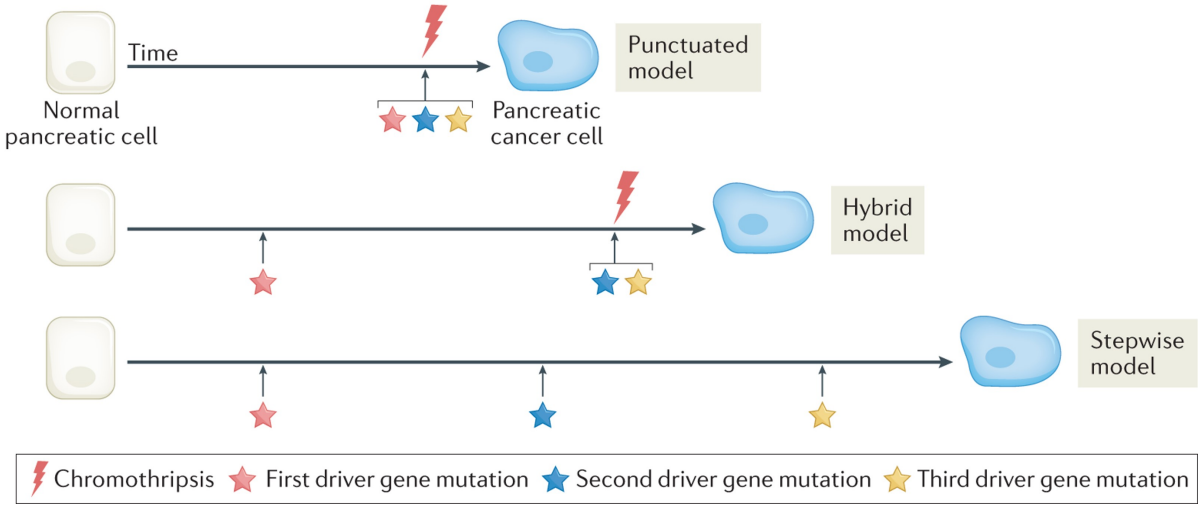

Fig. 1. The hypothetical accumulation of somatic events over time as a pancreatic cell undergoes dysplasia, including structural (chromothripsis) and simple somatic mutations under a punctuated model, stepwise model, or hybrid model. This figure was edited and reproduced with copyright permission from reference [19].

neoplasms (MCN), and pancreatic intraepithelial neoplasia (PanINs). PanINs are defined as microscopic (usually <0.5 cm) lesions while IPMN and MCN are grossly visible lesions. The goal of the three-tiered system was to guide clinical decision making and appropriately select high risk patients for surgery. However, in recent years there has been an increase in incidentally found low and intermediate grade IPMN and MCN on abdominal imaging [5]. Additionally, long term, non-operative follow-up has demonstrated that these tumors have a low risk of progressing to cancer and can likely be observed until concerning features or tumor growth develop, or if the lesion becomes symptomatic [6–8]. The third precursor lesion, PanIN, is defined as a lesion <0.5 cm and [5]. As such, a two-tiered system was more in line with clinical and practical clinical goals. Essentially the PanIN-1 (low grade) and PanIN-2 (intermediate grade) lesions were re-classified as "low grade" and the PanIN-3 (high grade) lesions were re-classified as "high grade" [5]. Additionally, the IPMN and MCN low and intermediate grade lesions were grouped together as "low grade". High grade terminology is reserved for the most advanced dysplasia, commonly classified as carcinoma *in situ* [5].

4. Suggested Molecular Pathways in PDAC Carcinogenesis

Theory of PDAC Carcinogenesis

Tumorigenesis is commonly due to a combination of somatic mutations, chromosomal rearrangements, copy number alterations, and epigenetic changes that damage the natural cell cycle and regulatory pathways. Sequencing of tumor samples has changed the understanding of disease biology and has revealed inter-tumoral and intra-tumoral heterogeneity. In turn, there has been a shift in therapeutic strategy to identify potential targets and deliver more personalized care. Based on these data, two different models of PDAC carcinogenesis have been proposed. The first is a stepwise progression through the accumulation of genetic alterations in the following order: oncogene *KRAS* (90% of patients), followed by tumor suppressors *CDKN2A* (60%), then *TP53* (80%) and *SMAD4* (40%) [9–11]. Accumulated mutations in these four genes leads to cell cycle proliferation in both human and mouse studies [12,13]. Additional studies have suggested that the early activation of *KRAS* and subsequent RAS signaling pathway is the main driver of PDAC development and concomitant mutations in the three tumor suppressor genes accelerates the development and dissemination of cancer [14–17]. This theory suggests a slow, gradual development process with a late clinical presentation of the disease.

The second theory proposes that PDAC occurs through the simultaneous knockout of genes, as opposed to a stepwise fashion, in the setting of complex chromosomal rearrangements. Essentially, several structural alterations occur in a single cell cycle on a few chromosomes that leads to rearrangement of multiple driver genes. These changes result in rapid carcinogenesis and dissemination [18]. The main difference between the two theories is whether PDAC occurs through a stepwise or punctuated progression, sometimes referred to as chromothripsis (Fig. 1, Ref. [19]). The punctuated progression theory is compatible with patients who present with sudden onset of advanced disease.

5. Molecular Classification of PDAC—Landmark Studies

Based on sequencing studies, there are several different molecular classification schema that have been proposed and adjusted over the past decade. The first study

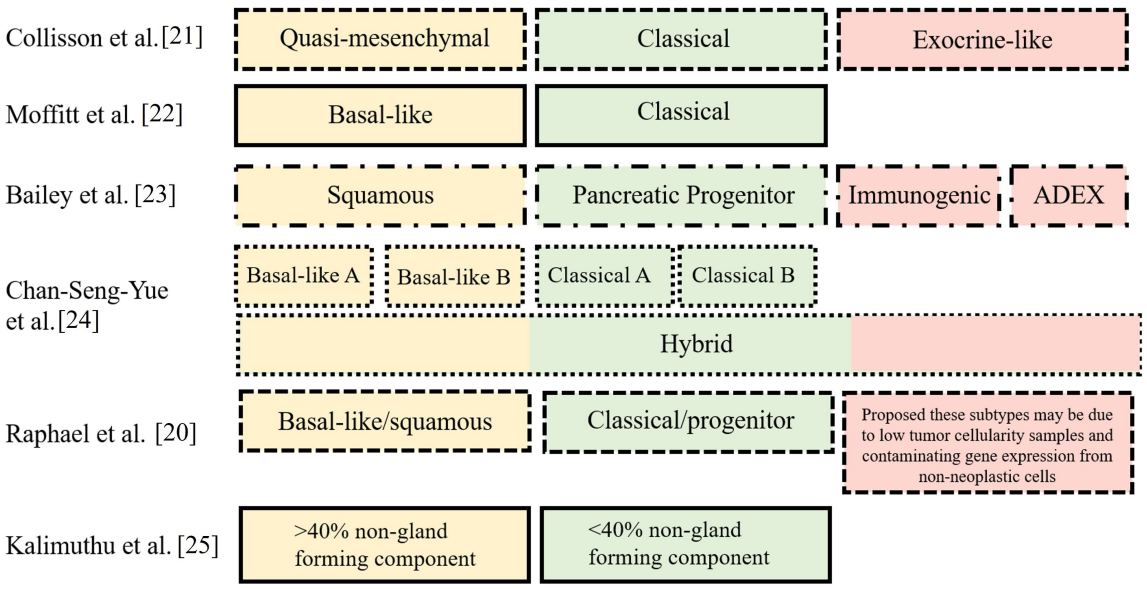

Fig. 2. Various subtypes of pancreatic adenocarcinoma based on molecular and morphologic characteristics. ADEX, aberrantly differentiated endocrine exocrine.

to comprehensively look at PDAC with whole exome sequencing was in 2008 [9]. Jones *et al*. [9] evaluated 24 PDAC tumors and proposed that the key to understand PDAC was through a core set of pathways with genetic aberrations. Most cellular pathways rely on multiple proteins to function, therefore mutations in different genes can result in disruption in the same pathways and subsequent tumorigenesis. Jones *et al*. [9] reported on 12 core cellular signaling pathways that were disrupted in most of the tumors, yet had variations in specific gene mutations among different tumors. Subsequent sequencing studies have better defined the genomic landscape of PDAC with the goal of classifying subsets of PDAC based on the genomic landscape and attempting to tailor therapy (Fig. 2, Ref. [20–25]) [20,26].

5.1 Classical versus Quasimesenchymal versus Exocrine-Like Subtypes

Collisson *et al*. [21] was the first study to categorize PDAC into subtypes based on the genomic landscape. At the time, there was a paucity of PDAC samples available so the authors combined data from primary PDAC samples into two different studies [21,27]. Based on this analysis, the authors defined three subtypes: classical, quasimesenchymal, and exocrine-like. The classical subtype had high expression of epithelial and adhesion-associated genes and was sensitive to erlotinib *in vitro*. The quasi-mesenchymal subtype was defined by high expression of mesenchymal-associated genes. These patients had worse survival outcomes compared with the other two subtypes and were more sensitive to gemcitabine on *in vitro* analysis. The exocrine-like subtype demonstrated high expression of tumor cell derived digestive enzyme genes. The exocrine-like subtype was not identified in any of the mouse or human cell lines [21]. These subtypes have been validated with additional published PDAC datasets.

5.2 Basal versus Classical Subtype

Research and treatment of PDAC is complicated by the overwhelming stromal component within the tumor. The dense stroma tissue and paucity of malignant cells makes it difficult to perform molecular analysis. Moffitt *et al*. [22] overcame this obstacle by including normal pancreas, primary PDAC, and metastatic PDAC samples in their analysis. The authors employed blind source separation and digitally separated tumor from stroma and normal tissue gene expression. Using this method, two subtypes were identified: classical and basal-like. The classical subtype was characterized by high adhesion-associated gene expression, ribosomal and epithelial gene expression, and increased GATA6 expression. This subtype overlapped with Collisson's "classical subtype". The basal-like subtype was comprised of tissue with high laminins and keratins like the basal-like subtype of bladder and breast cancers. This subtype was associated with worse outcomes.

Moffitt *et al*. [22] classified the stroma gene expression into two subtypes: normal and activated. Activated stroma was characterized by genes associated with a strong immune response and normal stoma contained markers for pancreatic stellate cells. The stroma subtypes were independently predictive of outcomes. Patients with activated stroma had worse long-term outcomes versus patients with normal stoma (median overall survival (OS): 15 months versus 24 months, respectively). Both basal and classical

subtypes were within normal and activated stroma. The normal stoma, classical subtype had the lowest hazard ratio on Cox regression, while the basal-like, activated subtype had the highest. These data suggested a cumulative effect of PDAC subtype and stoma subtype on survival.

5.3 Squamous versus Pancreatic Progenitor versus Immunogenic versus ADEX Subtypes

Bailey *et al*. [23] performed an analysis of 456 PDAC samples that used a combination of whole-genome and RNA sequencing to determine mutational mechanisms, potential genomic events, and expression profiles. The samples were initially enriched and selected for high epithelial content >40% to balance stromal gene expression. Four subtypes were defined. The squamous subtype had upregulation of *TP63ΔN* transcriptional network, hypermethylation of pancreatic endodermal cell-fate determining genes, and enrichment for *TP53* and *KDM6A* mutations. This subtype overlaps with the quasimesenchymal subtype from Collisson *et al*. [21]. Pancreatic progenitor tumors were defined by expression of genes involved in early pancreatic development (*FOXA2/3*, *PDX1*, and *MNX1*) and metabolism [23]. This subtype overlapped with the classical subtype as defined by Collisson *et al*. [21]. The aberrantly differentiated endocrine exocrine (ADEX) subtype featured upregulation of genes that regulated networks involved in *KRAS* activation and genes associated with exocrine and endocrine differentiation [23]. This subtype corresponded to Collisson's exocrine-like subtype [21]. In addition, immunogenic tumors were classified as an enrichment of genes associated with infiltrating B and T cells [23].

5.4 The Cancer Genome Atlas Comparison

Raphael *et al*. [20] compared the three molecular classification systems using The Cancer Genome Atlas. This study performed genomic, transcriptomic, and proteomic profiling of 150 PDAC samples [20]. The authors applied a clustering technique to reproduce the subtype classifications of Collisson *et al*. [21], Moffitt *et al*. [22], and Bailey *et al*. [23]. Given that the low neoplastic cellularity of PDAC made analysis challenging, Raphael *et al*. [20] divided tumors into high and low purity samples based on the malignant cellularity of the tissue. High-purity tumors were classified into two groups: the basal-like/squamous which correlated to the previously described quasi-mesenchymal (Collisson *et al*. [21]), squamous (Bailey *et al*. [23]), or basal-like group (Moffitt *et al*. [22]) and the classical group that corresponded to the previously described classical (Collisson *et al*. [21]), progenitor (Bailey *et al*. [23]), or classical group (Moffitt *et al*. [22]). The immunogenic and ADEX groups described by Bailey *et al*. [23] and the exocrine-like group reported by Collisson *et al*. [21] were only found in low tumor cellularity samples, which may have represented contaminating gene expression from non-neoplastic cells. However, these subtypes should not be entirely disregarded since there has been variation among these landmark studies, as well as the subsequent studies, in the way samples were collected, stored, and analyzed. The validity of the immunogenic, ADEX, and exocrine-like subtypes still requires further investigation.

Raphael *et al*. [20] proposed that based on these data, the two consensus subtypes should be basal-like/squamous and classical/progenitor. This proposal was further validated with subsequent long non-coding RNA sequencing, methylation analysis, and proteomics [20]. The two subtypes were predictive of overall survival following surgery in two different analyses [13,20]. These data suggest that these subtypes may provide clinically relevant information to assist with strategies related to treatment.

5.5 Basal-Like versus Hybrid versus Classical Subtypes

The underlying mechanism for the evolution of PDAC into either of these two subtypes is still unclear. Currently, basal-like tumors are associated with poor differentiation, worse survival, and resistance to therapy while classical tumors are more differentiated, have better survival, and improved response to chemotherapy [24]. However, these subtypes were primarily evaluated in patients with localized primary tumors who underwent resection and not among individuals with metastatic disease. Building on this work, Chan-Seng-Yue *et al*. [24] created a dataset of whole genomes and transcriptomes generated from purified epithelium of primary and metastatic PDAC tumors. Based on these data, a more granular classification system was proposed: basal-like A, basal-like B, hybrid, classical A, and classical B. Even within the two accepted molecular subtypes of PDAC, there was transcriptomic heterogeneity that was correlated with clinical disease manifestation. When divided into basal-like A versus B and classical A versus B signatures, metastatic disease from localized disease could be differentiated, respectively. The hybrid subtype was inconsistently classified due to multiple expression profiles. This stratification system still requires validation, but it does demonstrate the importance of including metastatic sites in PDAC molecular studies.

5.6 Chromosomal Subtypes of PDAC

Somatic structural rearrangement of chromosomes is a common mutation that causes gene disruption, activation, and/or fusions; these mutations drive carcinogenesis. Small genomic sequencing studies of PDAC demonstrated that PDAC genomes include extensive and complex chromosomal rearrangements [28,29]. In 2015, whole genome sequencing of 100 PDAC tumors revealed that alterations of chromosomal structure led to gene disruption, particularly in the genes commonly associated with PDAC [30]. Waddell *et al*. [30] classified tumors into four subtypes based on the number and location of the rearrangements: stable, locally rearranged, scattered, and unstable. Stable tumors had <50 structural rearrangements located ran-

domly throughout the genome while unstable tumors contained >200 rearrangements. Locally rearranged tumors had intra-chromosomal rearrangements clustered on only a few chromosomes, whereas scattered tumors contained 50–200 structural rearrangements present throughout the genome. Additionally, these subtypes had potential clinical relevance in patients receiving platinum-based therapy.

6. Morphological Classification of PDAC

Another potential way to classify PDAC is through morphological patterns on pathology. Kalimuthu *et al*. [25] identified morphological patterns in PDAC and subsequently proposed a new classification system. Morphological classification can be identified during pathologic slide review, as opposed to the more costly sequencing data necessary for molecular classification. Kalimuthu *et al*. [25] assessed 86 primary PDAC specimens with matched RNA sequencing data. There were four morphological patterns that were divided into two groups. Patients were classified as having either <40% or ≥40% non-gland forming components (group A and B, respectively). When matched to the sequencing data, individuals in group A were associated with the classical molecular subtype and group B were associated with basal molecular subtype. Group A patients had an improved OS compared with group B. About a quarter of the tumors were molecularly classified as classical, yet morphologically classified as group B. These data suggest that there was not complete overlap between the molecular and morphologic classification [25]. These data suggest that there may be some discrepancy between morphologic and molecular subtyping and that the two classification systems are not directly interchangeable.

7. Targeted Therapy

The goal of classifying PDAC based on underlying molecular signature is to better identify which patients may respond to targeted therapy. Understanding differences in gene expression in PDAC tumors (e.g., genes that lead to angiogenesis versus lymphangiogenesis) may provide clues to more effective combination therapies, as well as ways to improve patient selection for different therapies [31]. Unfortunately, there are little data correlating molecular subtype to existing targeted therapies.

7.1 EGFR and Anti-EGFR Strategies

Epidermal growth factor receptor (EGFR) is a transmembrane tyrosine kinase receptor (TKR) that is commonly overexpressed in PDAC. Constitutive activation of EGFR has been noted in multiple cancers and EGFR inhibitors have emerged as an effective therapy [32]. EGFR activation has been involved in pancreatic proliferation and progression. Erlotinib is an oral, selective tyrosine kinase inhibitor (TKI) that acts against EGFR. A phase III randomized trial assigned 569 patients with unresectable, locally advanced, or metastatic PDAC to receive either gemcitabine with erlotinib or a placebo [33]. Patients who received erlotinib had improved one year survival versus the placebo cohort (23% versus 17%, respectively) and longer progression free survival. Erlotinib was the first molecular-targeting agent that demonstrated a statistically significant effect among patients with PDAC. Erlotinib was subsequently studied in the CONKO-005 trial, which randomly assigned 463 patients with resected PDAC to receive adjuvant gemcitabine alone or gemcitabine with erlotinib [34]. There was no improvement in disease free or overall survival with the addition of erlotinib. In a follow up study, tumor samples from the CONKO-005 trial were sequenced [35]. A *SMAD4* genetic aberration with low mRNA expression of MAPK9 was noted to be predictive of response to erlotinib. In turn, *SMAD4* status may identify a subset of patients who would benefit from adjuvant erlotinib in early-stage PDAC.

Cetuximab is a monoclonal antibody against EGFR that is commonly used in colorectal cancer treatment [36]. Pre-clinical studies with PDAC mouse xenograft models demonstrated cetuximab efficacy [37]. However, these results did not translate to the early clinical trials. In a phase III randomized trial, 745 patients with locally advanced or metastatic PDAC were assigned to receive gemcitabine alone or gemcitabine with cetuximab [38]. There was no difference in median overall survival between the gemcitabine/cetuximab and gemcitabine alone cohorts (6.3 months versus 5.9 months, respectively).

Nimotuzumab is a monoclonal antibody against EGFR that has demonstrated better efficacy among patients with wild-type Ras PDAC. Qin *et al*. [39] randomized 92 patients with locally advanced or metastatic PDAC to receive gemcitabine with nimotuzumab or placebo. The nimotuzumab cohort had an overall survival of 10.9 months versus 8.5 months in the placebo cohort. While the differences were statistically significant, the absolute difference in survival was modest.

There may be ways to target the EGFR pathway indirectly in pancreatic cancer. For example, MASTL regulates EGFR protein stability and kinase signaling involved in the progression of PDAC [40]. In turn, this makes MASTL a potential target for pancreatic cancer treatment. Dosch *et al*. [41] reported that STAT3 is a potential biomarker of resistance to gemcitabine in PDAC. Additionally, inhibition of Src and EGFR pathways may be a mechanism to overcome gemcitabine resistance.

7.2 Ras Signaling Pathways

KRAS mutation is present in up to 98% of PDAC and often considered to be the inciting event in PDAC carcinogenesis [42]. KRAS activates several downstream oncogenic signaling pathways (e.g., PI3K/AKT/mTOR, Raf-MEK-ERK, RAL-PLD1, T1AM1-Rac). As such, targeting KRAS mediated pathways is an important area of investigation in PDAC research. Pre-clinical studies have demon-

strated some promise [43]. However, these preclinical results have not consistently translate to the clinical setting, likely due to the difficulty in re-creating the dense stromal tumor microenvironment of PDAC in pre-clinical models. As such, inhibition of downstream pathways of KRAS may exert different effects in the pre-clinical studies compared with the clinical setting.

7.3 RAS-PI3K/AKT/mTOR Pathway Inhibition

Several trials have evaluated the efficacy of inhibitors of the PI3K/AKT/mTOR pathway. Several PI3K inhibitors, including copanlisib, alpelisib, and buparlisib, have been demonstrated to be safe in phase I studies with low efficacy [44–46]. Everolimus, an mTOR inhibitor, has demonstrated some efficacy in the clinical setting. Kordes *et al*. [47] enrolled 31 patients with advanced PDAC who received capecitabine and everolimus as either first- or second-line therapy. These patients had an acceptable toxicity profile with a median overall survival of 8.9 months. However, other studies of mTOR inhibitors as a monotherapy or combination therapy have not demonstrated clinical efficacy, including other trials with everolimus [48–51]. Perifosine is an AKT inhibitor that demonstrated significant activity in the preclinical setting, but failed to demonstrate efficacy in clinical trials [52,53]. These therapies have also been tried in combination with other targeted therapies with little success [50,54–56]. Unfortunately, none of the PI3K/AKT/mTOR inhibitors have demonstrated a survival benefit in patients with PDAC.

7.4 RAS-RAF/MEK/ERK Pathway

Mutations in KRAS can lead to inappropriate activation of the MAPK pathway (Ras-Raf-MEK-ERK) and act as genetic drivers in the initiation and progression of tumors. Unfortunately, it has been challenging to target this pathway effectively in PDAC. Tipifarnib, which showed promise in preclinical studies, has failed to demonstrate efficacy in clinical trial [57–60]. Sorafenib, a TKI, demonstrated disease stability in early clinical trials, but did not improve survival when used as a monotherapy or in combination with gemcitabine [61–63]. Selumetinib, a MEK inhibitor, was well tolerated when tested in the second- or third-line setting, but has not demonstrated significant clinical activity [64,65]. The ERK inhibitor ulixertinib was tested in combination with gemcitabine and nab-paclitaxel in treatment-naïve patients with metastatic PDAC [66]. While effective (median OS 12.23 months), the study was stopped for increased adverse events. There is currently an ongoing phase I trial evaluating ulixertinib and palbociclib as a second-line therapy in patients with solid tumors, including PDAC (NCT03454035).

Based on pre-clinical evidence of a synergistic effect, trametinib (MEK inhibitor) has been tested in combination with erlotinib (EGFR inhibitor). However, this combination only demonstrated mild clinical efficacy with a median OS 7.3 months [67]. Additionally, the combination of radiation therapy, pembrolizumab, and trametinib demonstrated slightly improved OS compared with radiation therapy and gemcitabine among patients with locally recurrent PDAC (median OS: 14.9 months versus 12.8 months, respectively) [68]. A recent phase II trial of patients with advanced PDAC who progressed on chemotherapy were treated with trametinib and an oral FAK inhibitor with a median OS of 3.6 months and progression free survival of 1.6 months [69]. Trametinib may prove to be effective if the right combination therapy or subset of patients is identified, but more work is still needed to demonstrate long term clinical efficacy.

7.5 KRAS Targeted Drugs

Until now, targeting KRAS could only be done through an indirect therapeutic strategy that focused on inhibiting downstream effects, as targeting KRAS directly was not possible. However, recent research has potentially overcome this obstacle by creating a compound that can target the $KRAS^{G12c}$ mutant allele [70]. This finding has led to the development of sotorasib and adagrasib [71,72]. These inhibitors have largely had success in non-small cell lung cancer where the mutation frequency of $KRAS^{G12c}$ is 13.8%. Unfortunately, KRAS inhibitors have demonstrated less success for PDAC in which the $KRAS^{G12c}$ mutation frequency is $<1\%$ [43]. In the phase I/II trial of patients with $KRAS^{G12c}$ mutated PDAC who failed first line chemotherapy, sotorasib had some antitumor activity [73]. The KRYSTAL-1 phase I/II trial is currently evaluating adagrasib as a second line therapy in patients with advanced solid tumors and a $KRAS^{G12c}$ mutation. At this time, 21 of the 63 patients have PDAC. Adagrasib has been well tolerated and patients with PDAC had an objective response rate of 33.3%, disease control rate of 81%, median progression free survival of 5.4 months, and median overall survival of 8 months [74].

The $KRAS^{G12d}$ mutation is the most prevalent variant in PDAC, occurring in approximately 41% of KRAS mutated patients [43]. MRTX1133 is a $KRAS^{G12d}$ inhibitor that has demonstrated tumor regression in PDAC cell lines and patient derived xenografts [75]. Given the potential applicability to PDAC, work is currently underway to develop $KRAS^{G12d}$ specific inhibitors [76]. In particular, there are several ongoing trials investigating various KRAS targeted drugs in combination therapy (NCT05379985, NCT04916236, NCT04975256).

Cancer vaccination has emerged as a potential immunotherapy for patients with solid tumors by inducing specific targeted immunity. This approach has mainly been studied as a means to personalize medicine by vaccinating patients with antigens specific to their tumor to stimulate the immune system, thereby inducing targeting of cancer cells that present those same antigens. Recent work has explored mRNA vaccines that are specific to a common mu-

tation (e.g., KRAS mutated cancers) [76]. mRNA vaccines can also express multiple antigens at once, potentially allowing for a more sustained and stronger immune response [77]. A phase I trial is currently investigating a vaccine that targets KRAS-mutant non-small cell lung cancer, colorectal cancer, and PDAC (NCT03948763). There are ongoing trials evaluating dendritic cell and peptide vaccines for patients with KRAS-mutated PDAC (NCT03592888 and NCT04117087).

7.6 Claudin-18 Overexpression

Claudins are a family of transmembrane tight junction proteins that typically reside in the apical region of the cell membrane and form a paracellular barrier [78]. This barrier regulates the passage of ions between cells, maintains homeostasis, and is associated with cell signaling pathways that influence proliferation and differentiation [79]. Claudins can be modified during carcinogenesis and may be a potential therapeutic target. Claudin-18 (CLDN18) has two variants. Zolbetuximab is a monoclonal antibody to the variant CLDN18.2 that has demonstrated some efficacy against gastric, gastric-esophageal junction, and esophageal cancers based on the FAST phase II randomized trial [80]. While the variant CLDN18.2 is not expressed in normal pancreatic tissue, it has been noted to be aberrantly expressed in 60–90% of PDACs [81–83]. There is an ongoing phase II trial evaluating its use in CLDN18.2-expressing metastatic PDAC (NCT03816163) in combination with gemcitabine and nab-paclitaxel as first-line therapy.

8. Conclusions

PDAC is an aggressive disease in which surgery and chemotherapy offer the best chance at long-term survival. Unfortunately, most patients present with late-stage disease and many who present early develop recurrence or metastatic disease. There are many challenges to understand and research PDAC due to tumor heterogeneity, low neoplastic cellularity in samples making molecular analysis difficult, and the dense stromal tissue that makes up most of the tumor microenvironment. DNA and RNA sequencing has shed light on potential molecular subtypes of PDAC, but how these classification systems should guide treatment remains debated. The development of pre-clinical models that incorporate the surrounding stromal tissue and tumor microenvironment will help better identify which therapies may be effective in the clinical setting to treat PDAC.

Author Contributions

SMR and TMP have contributed to the conception, writing, and editing of this manuscript. Both authors read and approved the final manuscript. Both authors have participated sufficiently in the work and agreed to be accountable for all aspects of the work.

Ethics Approval and Consent to Participate

Not applicable.

Acknowledgment

Not applicable.

Funding

This research received no external funding.

Conflict of Interest

The authors declare no conflict of interest.

Supplementary Material

Supplementary material associated with this article can be found, in the online version, at https://doi.org/10.31083/j.fbl2903101.

References

[1] Siegel RL, Miller KD, Wagle NS, Jemal A. Cancer statistics, 2023. CA: A Cancer Journal for Clinicians. 2023; 73: 17–48.

[2] Ducreux M, Cuhna AS, Caramella C, Hollebecque A, Burtin P, Goéré D, *et al*. Cancer of the pancreas: ESMO Clinical Practice Guidelines for diagnosis, treatment and follow-up. Annals of Oncology. 2015; 26: v56–v68.

[3] Geerinckx B, Teuwen LA, Foo T, Vandamme T, Smith A, Peeters M, *et al*. Novel therapeutic strategies in pancreatic cancer: moving beyond cytotoxic chemotherapy. Expert Review of Anticancer Therapy. 2023; 23: 1237–1249.

[4] Polani F, Grierson PM, Lim KH. Stroma-targeting strategies in pancreatic cancer: Past lessons, challenges and prospects. World Journal of Gastroenterology. 2021; 27: 2105–2121.

[5] Basturk O, Hong SM, Wood LD, Adsay NV, Albores-Saavedra J, Biankin AV, *et al*. A Revised Classification System and Recommendations From the Baltimore Consensus Meeting for Neoplastic Precursor Lesions in the Pancreas. The American Journal of Surgical Pathology. 2015; 39: 1730–1741.

[6] Allen PJ, D'Angelica M, Gonen M, Jaques DP, Coit DG, Jarnagin WR, *et al*. A selective approach to the resection of cystic lesions of the pancreas: results from 539 consecutive patients. Annals of Surgery. 2006; 244: 572–582.

[7] Konstantinidis IT, Vinuela EF, Tang LH, Klimstra DS, D'Angelica MI, Dematteo RP, *et al*. Incidentally discovered pancreatic intraepithelial neoplasia: what is its clinical significance? Annals of Surgical Oncology. 2013; 20: 3643–3647.

[8] Andea A, Sarkar F, Adsay VN. Clinicopathological correlates of pancreatic intraepithelial neoplasia: a comparative analysis of 82 cases with and 152 cases without pancreatic ductal adenocarcinoma. Modern Pathology. 2003; 16: 996–1006.

[9] Jones S, Zhang X, Parsons DW, Lin JCH, Leary RJ, Angenendt P, *et al*. Core signaling pathways in human pancreatic cancers revealed by global genomic analyses. Science. 2008; 321: 1801–1806.

[10] ICGC/TCGA Pan-Cancer Analysis of Whole Genomes Consortium. Pan-cancer analysis of whole genomes. Nature. 2020; 578: 82–93.

[11] Kong B, Michalski CW, Erkan M, Friess H, Kleeff J. From tissue turnover to the cell of origin for pancreatic cancer. Nature Reviews. Gastroenterology & Hepatology. 2011; 8: 467–472.

[12] Klein WM, Hruban RH, Klein-Szanto AJP, Wilentz RE. Direct correlation between proliferative activity and dysplasia in pancreatic intraepithelial neoplasia (PanIN): additional evidence for

a recently proposed model of progression. Modern Pathology. 2002; 15: 441–447.
[13] Connor AA, Denroche RE, Jang GH, Lemire M, Zhang A, Chan-Seng-Yue M, *et al*. Integration of Genomic and Transcriptional Features in Pancreatic Cancer Reveals Increased Cell Cycle Progression in Metastases. Cancer Cell. 2019; 35: 267–282.e7.
[14] Hingorani SR, Petricoin EF, Maitra A, Rajapakse V, King C, Jacobetz MA, *et al*. Preinvasive and invasive ductal pancreatic cancer and its early detection in the mouse. Cancer Cell. 2003; 4: 437–450.
[15] Aguirre AJ, Bardeesy N, Sinha M, Lopez L, Tuveson DA, Horner J, *et al*. Activated Kras and Ink4a/Arf deficiency cooperate to produce metastatic pancreatic ductal adenocarcinoma. Genes & Development. 2003; 17: 3112–3126.
[16] Izeradjene K, Combs C, Best M, Gopinathan A, Wagner A, Grady WM, *et al*. Kras(G12D) and Smad4/Dpc4 haploinsufficiency cooperate to induce mucinous cystic neoplasms and invasive adenocarcinoma of the pancreas. Cancer Cell. 2007; 11: 229–243.
[17] Hingorani SR, Wang L, Multani AS, Combs C, Deramaudt TB, Hruban RH, *et al*. Trp53R172H and KrasG12D cooperate to promote chromosomal instability and widely metastatic pancreatic ductal adenocarcinoma in mice. Cancer Cell. 2005; 7: 469–483.
[18] Notta F, Chan-Seng-Yue M, Lemire M, Li Y, Wilson GW, Connor AA, *et al*. A renewed model of pancreatic cancer evolution based on genomic rearrangement patterns. Nature. 2016; 538: 378–382.
[19] Connor AA, Gallinger S. Pancreatic cancer evolution and heterogeneity: integrating omics and clinical data. Nature Reviews. Cancer. 2022; 22: 131–142.
[20] Cancer Genome Atlas Research Network. Electronic address: andrew_aguirre@dfci.harvard.edu, Cancer Genome Atlas Research Network. Integrated Genomic Characterization of Pancreatic Ductal Adenocarcinoma. Cancer Cell. 2017; 32: 185–203.e13.
[21] Collisson EA, Sadanandam A, Olson P, Gibb WJ, Truitt M, Gu S, *et al*. Subtypes of pancreatic ductal adenocarcinoma and their differing responses to therapy. Nature Medicine. 2011; 17: 500–503.
[22] Moffitt RA, Marayati R, Flate EL, Volmar KE, Loeza SGH, Hoadley KA, *et al*. Virtual microdissection identifies distinct tumor- and stroma-specific subtypes of pancreatic ductal adenocarcinoma. Nature Genetics. 2015; 47: 1168–1178.
[23] Bailey P, Chang DK, Nones K, Johns AL, Patch AM, Gingras MC, *et al*. Genomic analyses identify molecular subtypes of pancreatic cancer. Nature. 2016; 531: 47–52.
[24] Chan-Seng-Yue M, Kim JC, Wilson GW, Ng K, Figueroa EF, O'Kane GM, *et al*. Transcription phenotypes of pancreatic cancer are driven by genomic events during tumor evolution. Nature Genetics. 2020; 52: 231–240.
[25] N Kalimuthu S, Wilson GW, Grant RC, Seto M, O'Kane G, Vajpeyi R, *et al*. Morphological classification of pancreatic ductal adenocarcinoma that predicts molecular subtypes and correlates with clinical outcome. Gut. 2020; 69: 317–328.
[26] Biankin AV, Waddell N, Kassahn KS, Gingras MC, Muthuswamy LB, Johns AL, *et al*. Pancreatic cancer genomes reveal aberrations in axon guidance pathway genes. Nature. 2012; 491: 399–405.
[27] Badea L, Herlea V, Dima SO, Dumitrascu T, Popescu I. Combined gene expression analysis of whole-tissue and microdissected pancreatic ductal adenocarcinoma identifies genes specifically overexpressed in tumor epithelia. Hepatogastroenterology. 2008; 55: 2016–2027.
[28] Molenaar JJ, Koster J, Zwijnenburg DA, van Sluis P, Valentijn LJ, van der Ploeg I, *et al*. Sequencing of neuroblastoma identifies chromothripsis and defects in neuritogenesis genes. Nature. 2012; 483: 589–593.
[29] Campbell PJ, Yachida S, Mudie LJ, Stephens PJ, Pleasance ED, Stebbings LA, *et al*. The patterns and dynamics of genomic instability in metastatic pancreatic cancer. Nature. 2010; 467: 1109–1113.
[30] Waddell N, Pajic M, Patch AM, Chang DK, Kassahn KS, Bailey P, *et al*. Whole genomes redefine the mutational landscape of pancreatic cancer. Nature. 2015; 518: 495–501.
[31] Gore J, Imasuen-Williams IE, Conteh AM, Craven KE, Cheng M, Korc M. Combined targeting of TGF-β, EGFR and HER2 suppresses lymphangiogenesis and metastasis in a pancreatic cancer model. Cancer Letters. 2016; 379: 143–153.
[32] Vincent A, Herman J, Schulick R, Hruban RH, Goggins M. Pancreatic cancer. Lancet. 2011; 378: 607–620.
[33] Moore MJ, Goldstein D, Hamm J, Figer A, Hecht JR, Gallinger S, *et al*. Erlotinib plus gemcitabine compared with gemcitabine alone in patients with advanced pancreatic cancer: a phase III trial of the National Cancer Institute of Canada Clinical Trials Group. Journal of Clinical Oncology. 2007; 25: 1960–1966.
[34] Sinn M, Bahra M, Liersch T, Gellert K, Messmann H, Bechstein W, *et al*. CONKO-005: Adjuvant Chemotherapy With Gemcitabine Plus Erlotinib Versus Gemcitabine Alone in Patients After R0 Resection of Pancreatic Cancer: A Multicenter Randomized Phase III Trial. Journal of Clinical Oncology. 2017; 35: 3330–3337.
[35] Hoyer K, Hablesreiter R, Inoue Y, Yoshida K, Briest F, Christen F, *et al*. A genetically defined signature of responsiveness to erlotinib in early-stage pancreatic cancer patients: Results from the CONKO-005 trial. eBioMedicine. 2021; 66: 103327.
[36] Karapetis CS, Khambata-Ford S, Jonker DJ, O'Callaghan CJ, Tu D, Tebbutt NC, *et al*. K-ras mutations and benefit from cetuximab in advanced colorectal cancer. The New England Journal of Medicine. 2008; 359: 1757–1765.
[37] Overholser JP, Prewett MC, Hooper AT, Waksal HW, Hicklin DJ. Epidermal growth factor receptor blockade by antibody IMC-C225 inhibits growth of a human pancreatic carcinoma xenograft in nude mice. Cancer. 2000; 89: 74–82.
[38] Philip PA, Benedetti J, Corless CL, Wong R, O'Reilly EM, Flynn PJ, *et al*. Phase III study comparing gemcitabine plus cetuximab versus gemcitabine in patients with advanced pancreatic adenocarcinoma: Southwest Oncology Group-directed intergroup trial S0205. Journal of Clinical Oncology. 2010; 28: 3605–3610.
[39] Qin S, Li J, Bai Y, Wang Z, Chen Z, Xu R, *et al*. Nimotuzumab Plus Gemcitabine for K-Ras Wild-Type Locally Advanced or Metastatic Pancreatic Cancer. Journal of Clinical Oncology. 2023; 41: 5163–5173.
[40] Fatima I, Barman S, Uppada J, Chauhan S, Rauth S, Rachagani S, *et al*. MASTL regulates EGFR signaling to impact pancreatic cancer progression. Oncogene. 2021; 40: 5691–5704.
[41] Dosch AR, Dai X, Reyzer ML, Mehra S, Srinivasan S, Willobee BA, *et al*. Combined Src/EGFR Inhibition Targets STAT3 Signaling and Induces Stromal Remodeling to Improve Survival in Pancreatic Cancer. Molecular Cancer Research. 2020; 18: 623–631.
[42] Chen K, Zhang Y, Qian L, Wang P. Emerging strategies to target RAS signaling in human cancer therapy. Journal of Hematology & Oncology. 2021; 14: 116.
[43] Zhang J, Darman L, Hassan MS, Von Holzen U, Awasthi N. Targeting KRAS for the potential treatment of pancreatic ductal adenocarcinoma: Recent advancements provide hope (Review). Oncology Reports. 2023; 50: 206.
[44] Kim RD, Alberts SR, Peña C, Genvresse I, Ajavon-Hartmann A, Xia C, *et al*. Phase I dose-escalation study of copanlisib in combination with gemcitabine or cisplatin plus gemcitabine in patients with advanced cancer. British Journal of Cancer. 2018; 118: 462–470.

[45] Bedard PL, Tabernero J, Janku F, Wainberg ZA, Paz-Ares L, Vansteenkiste J, *et al*. A phase Ib dose-escalation study of the oral pan-PI3K inhibitor buparlisib (BKM120) in combination with the oral MEK1/2 inhibitor trametinib (GSK1120212) in patients with selected advanced solid tumors. Clinical Cancer Research. 2015; 21: 730–738.

[46] McRee AJ, Sanoff HK, Carlson C, Ivanova A, O'Neil BH. A phase I trial of mFOLFOX6 combined with the oral PI3K inhibitor BKM120 in patients with advanced refractory solid tumors. Investigational New Drugs. 2015; 33: 1225–1231.

[47] Kordes S, Klümpen HJ, Weterman MJ, Schellens JHM, Richel DJ, Wilmink JW. Phase II study of capecitabine and the oral mTOR inhibitor everolimus in patients with advanced pancreatic cancer. Cancer Chemotherapy and Pharmacology. 2015; 75: 1135–1141.

[48] Amin M, Gao F, Terrero G, Picus J, Wang-Gillam A, Suresh R, *et al*. Phase I Study of Docetaxel and Temsirolimus in Refractory Solid Tumors. American Journal of Clinical Oncology. 2021; 44: 443–448.

[49] Karavasilis V, Samantas E, Koliou GA, Kalogera-Fountzila A, Pentheroudakis G, Varthalitis I, *et al*. Gemcitabine Combined with the mTOR Inhibitor Temsirolimus in Patients with Locally Advanced or Metastatic Pancreatic Cancer. A Hellenic Cooperative Oncology Group Phase I/II Study. Targeted Oncology. 2018; 13: 715–724.

[50] Javle MM, Shroff RT, Xiong H, Varadhachary GA, Fogelman D, Reddy SA, *et al*. Inhibition of the mammalian target of rapamycin (mTOR) in advanced pancreatic cancer: results of two phase II studies. BMC Cancer. 2010; 10: 368.

[51] Wolpin BM, Hezel AF, Abrams T, Blaszkowsky LS, Meyerhardt JA, Chan JA, *et al*. Oral mTOR inhibitor everolimus in patients with gemcitabine-refractory metastatic pancreatic cancer. Journal of Clinical Oncology. 2009; 27: 193–198.

[52] Marsh RDW, Rocha Lima CM, Levy DE, Mitchell EP, Rowland KM, Jr, Benson AB, 3rd. A phase II trial of perifosine in locally advanced, unresectable, or metastatic pancreatic adenocarcinoma. American Journal of Clinical Oncology. 2007; 30: 26–31.

[53] Hedley D, Moore MJ, Hirte H, Siu L, Vincent M, Jonker D, *et al*. A phase II trial of perifosine as second line therapy for advanced pancreatic cancer. A study of the Princess Margaret Hospital [PMH] Phase II Consortium. Journal of Clinical Oncology. 2005; 23: 4166–4166.

[54] Weinberg BA, Wang H, Witkiewicz AK, Marshall JL, He AR, Vail P, *et al*. A Phase I Study of Ribociclib Plus Everolimus in Patients with Metastatic Pancreatic Adenocarcinoma Refractory to Chemotherapy. Journal of Pancreatic Cancer. 2020; 6: 45–54.

[55] Carr RM, Duma N, McCleary-Wheeler AL, Almada LL, Marks DL, Graham RP, *et al*. Targeting of the Hedgehog/GLI and mTOR pathways in advanced pancreatic cancer, a phase 1 trial of Vismodegib and Sirolimus combination. Pancreatology. 2020; 20: 1115–1122.

[56] Park H, Williams K, Trikalinos NA, Larson S, Tan B, Waqar S, *et al*. A phase I trial of temsirolimus and erlotinib in patients with refractory solid tumors. Cancer Chemotherapy and Pharmacology. 2021; 87: 337–347.

[57] Rich TA, Winter K, Safran H, Hoffman JP, Erickson B, Anne PR, *et al*. Weekly paclitaxel, gemcitabine, and external irradiation followed by randomized farnesyl transferase inhibitor R115777 for locally advanced pancreatic cancer. OncoTargets and Therapy. 2012; 5: 161–170.

[58] Van Cutsem E, van de Velde H, Karasek P, Oettle H, Vervenne WL, Szawlowski A, *et al*. Phase III trial of gemcitabine plus tipifarnib compared with gemcitabine plus placebo in advanced pancreatic cancer. Journal of Clinical Oncology. 2004; 22: 1430–1438.

[59] Macdonald JS, McCoy S, Whitehead RP, Iqbal S, Wade JL, 3rd, Giguere JK, *et al*. A phase II study of farnesyl transferase inhibitor R115777 in pancreatic cancer: a Southwest oncology group (SWOG 9924) study. Investigational New Drugs. 2005; 23: 485–487.

[60] Cohen SJ, Ho L, Ranganathan S, Abbruzzese JL, Alpaugh RK, Beard M, *et al*. Phase II and pharmacodynamic study of the farnesyltransferase inhibitor R115777 as initial therapy in patients with metastatic pancreatic adenocarcinoma. Journal of Clinical Oncology. 2003; 21: 1301–1306.

[61] Siu LL, Awada A, Takimoto CH, Piccart M, Schwartz B, Giannaris T, *et al*. Phase I trial of sorafenib and gemcitabine in advanced solid tumors with an expanded cohort in advanced pancreatic cancer. Clinical Cancer Research. 2006; 12: 144–151.

[62] Gonçalves A, Gilabert M, François E, Dahan L, Perrier H, Lamy R, *et al*. BAYPAN study: a double-blind phase III randomized trial comparing gemcitabine plus sorafenib and gemcitabine plus placebo in patients with advanced pancreatic cancer. Annals of Oncology. 2012; 23: 2799–2805.

[63] El-Khoueiry AB, Ramanathan RK, Yang DY, Zhang W, Shibata S, Wright JJ, *et al*. A randomized phase II of gemcitabine and sorafenib versus sorafenib alone in patients with metastatic pancreatic cancer. Investigational New Drugs. 2012; 30: 1175–1183.

[64] Kenney C, Kunst T, Webb S, Christina D, Jr, Arrowood C, Steinberg SM, *et al*. Phase II study of selumetinib, an orally active inhibitor of MEK1 and MEK2 kinases, in $KRAS^{G12R}$-mutant pancreatic ductal adenocarcinoma. Investigational New Drugs. 2021; 39: 821–828.

[65] Bodoky G, Timcheva C, Spigel DR, La Stella PJ, Ciuleanu TE, Pover G, *et al*. A phase II open-label randomized study to assess the efficacy and safety of selumetinib (AZD6244 [ARRY-142886]) versus capecitabine in patients with advanced or metastatic pancreatic cancer who have failed first-line gemcitabine therapy. Investigational New Drugs. 2012; 30: 1216–1223.

[66] Grierson PM, Tan B, Pedersen KS, Park H, Suresh R, Amin MA, *et al*. Phase Ib Study of Ulixertinib Plus Gemcitabine and Nab-Paclitaxel in Patients with Metastatic Pancreatic Adenocarcinoma. The Oncologist. 2023; 28: e115–e123.

[67] Ko AH, Bekaii-Saab T, Van Ziffle J, Mirzoeva OM, Joseph NM, Talasaz A, *et al*. A Multicenter, Open-Label Phase II Clinical Trial of Combined MEK plus EGFR Inhibition for Chemotherapy-Refractory Advanced Pancreatic Adenocarcinoma. Clinical Cancer Research. 2016; 22: 61–68.

[68] Zhu X, Cao Y, Liu W, Ju X, Zhao X, Jiang L, *et al*. Stereotactic body radiotherapy plus pembrolizumab and trametinib versus stereotactic body radiotherapy plus gemcitabine for locally recurrent pancreatic cancer after surgical resection: an open-label, randomised, controlled, phase 2 trial. The Lancet. Oncology. 2022; 23: e105–e115.

[69] Aung KL, McWhirter E, Welch S, Wang L, Lovell S, Stayner LA, *et al*. A phase II trial of GSK2256098 and trametinib in patients with advanced pancreatic ductal adenocarcinoma. Journal of Gastrointestinal Oncology. 2022; 13: 3216–3226.

[70] Ostrem JML, Shokat KM. Direct small-molecule inhibitors of KRAS: from structural insights to mechanism-based design. Nature Reviews. Drug Discovery. 2016; 15: 771–785.

[71] Canon J, Rex K, Saiki AY, Mohr C, Cooke K, Bagal D, *et al*. The clinical KRAS(G12C) inhibitor AMG 510 drives anti-tumour immunity. Nature. 2019; 575: 217–223.

[72] Hallin J, Engstrom LD, Hargis L, Calinisan A, Aranda R, Briere DM, *et al*. The $KRAS^{G12C}$ Inhibitor MRTX849 Provides Insight toward Therapeutic Susceptibility of KRAS-Mutant Cancers in Mouse Models and Patients. Cancer Discovery. 2020; 10: 54–71.

[73] Strickler JH, Satake H, George TJ, Yaeger R, Hollebecque A, Garrido-Laguna I, *et al*. Sotorasib in *KRAS* p.G12C-Mutated Advanced Pancreatic Cancer. The New England Journal of Medicine. 2023; 388: 33–43.

[74] Pant S, Yaeger R, Spira AI, Pelster MS, Sabari JK, Hafez N, *et al*. KRYSTAL-1: Activity and safety of adagrasib (MRTX849) in patients with advanced solid tumors harboring a KRASG12C mutation. Journal of Clinical Oncology. 2023; 41: 425082–425082.

[75] Hallin J, Bowcut V, Calinisan A, Briere DM, Hargis L, Engstrom LD, *et al*. Anti-tumor efficacy of a potent and selective non-covalent $KRAS^{G12D}$ inhibitor. Nature Medicine. 2022; 28: 2171–2182.

[76] Bannoura SF, Khan HY, Azmi AS. KRAS G12D targeted therapies for pancreatic cancer: Has the fortress been conquered? Frontiers in Oncology. 2022; 12: 1013902.

[77] Cowzer D, Zameer M, Conroy M, Kolch W, Duffy AG. Targeting KRAS in Pancreatic Cancer. Journal of Personalized Medicine. 2022; 12: 1870.

[78] Morin PJ. Claudin proteins in human cancer: promising new targets for diagnosis and therapy. Cancer Research. 2005; 65: 9603–9606.

[79] Angelow S, Ahlstrom R, Yu ASL. Biology of claudins. American Journal of Physiology. Renal Physiology. 2008; 295: F867–76.

[80] Sahin U, Türeci Ö, Manikhas G, Lordick F, Rusyn A, Vynnychenko I, *et al*. FAST: a randomised phase II study of zolbetuximab (IMAB362) plus EOX versus EOX alone for first-line treatment of advanced CLDN18.2-positive gastric and gastro-oesophageal adenocarcinoma. Annals of Oncology. 2021; 32: 609–619.

[81] Karanjawala ZE, Illei PB, Ashfaq R, Infante JR, Murphy K, Pandey A, *et al*. New markers of pancreatic cancer identified through differential gene expression analyses: claudin 18 and annexin A8. The American Journal of Surgical Pathology. 2008; 32: 188–196.

[82] Wöll S, Schlitter AM, Dhaene K, Roller M, Esposito I, Sahin U, *et al*. Claudin 18.2 is a target for IMAB362 antibody in pancreatic neoplasms. International Journal of Cancer. 2014; 134: 731–739.

[83] Lee JH, Kim KS, Kim TJ, Hong SP, Song SY, Chung JB, *et al*. Immunohistochemical analysis of claudin expression in pancreatic cystic tumors. Oncology Reports. 2011; 25: 971–978.