SWI/SNF complex alterations as a biomarker of immunotherapy efficacy in pancreatic cancer

Gregory P. Botta, … , Ryosuke Okamura, Razelle Kurzrock


BACKGROUND. Immune checkpoint inhibitors (ICIs), which have transformed the care of multiple malignancies, fail to demonstrate efficacy in pancreatic cancer. Recently, genomic biomarkers have been associated with response to ICIs: microsatellite instability high (MSI-H) and tumor mutation burden (TMB) ≥10 mutations/Mb. Some investigations suggest that alterations in Switch/Sucrose Non-Fermentable (SWI/SNF) chromatin remodeling genes may predispose to improved outcomes with immunotherapy. The current study examined a possible role for SWI/SNF complex abnormalities in pancreatic cancer responsiveness to ICIs.

METHODS. We interrogated a database of 6,831 cancer patients that had undergone next generation sequencing (NGS) in order to evaluate those with advanced pancreatic cancer, SWI/SNF alterations, and outcomes depending on immunotherapy treatment.

RESULTS. Of 6,831 cancer patients, nine had metastatic pancreatic adenocarcinoma harboring SWI/SNF chromatin remodeling gene alterations and had received ICIs: seven had an ARID1A alteration (77%); two, ARID1B (22%); three, SMARCA4 (33%); one, SMARCB1 (11%); and one, PBRM1 (11%). Three patients possessed more than one SWI/SNF complex alteration. Only three tumors were microsatellite unstable. Eight of 9 patients (89%) achieved an objective response, including a complete remission, with the two longest responses ongoing at 33+ and 36+ months. Median progression-free […]

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SWI/SNF Complex Alterations as a Biomarker of Immunotherapy Efficacy in Pancreatic Cancer

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Conflict of interest:
- Gregory Botta serves on the Advisory Board of Natera and as a consultant to TumorGen Inc. and CEND Therapeutics.
- Shumei Kato serves as a consultant for Foundation Medicine and receives speaker’s fees from Roche. Research funding from ACT Genomics, Sysmex, Konica Minolta and OmniSeq. 
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ABSTRACT

Background: Immune checkpoint inhibitors (ICIs), which have transformed the care of multiple malignancies, fail to demonstrate efficacy in pancreatic cancer. Recently, genomic biomarkers have been associated with response to ICIs: microsatellite instability high (MSI-H) and tumor mutation burden (TMB) ≥10 mutations/Mb. Some investigations suggest that alterations in Switch/Sucrose Non-Fermentable (SWI/SNF) chromatin remodeling genes may predispose to improved outcomes with immunotherapy. The current study examined a possible role for SWI/SNF complex abnormalities in pancreatic cancer responsiveness to ICIs.

Methods: We interrogated a database of 6,831 cancer patients that had undergone next generation sequencing (NGS) in order to evaluate those with advanced pancreatic cancer, SWI/SNF alterations, and outcomes depending on immunotherapy treatment.

Results: Of 6,831 cancer patients, nine had metastatic pancreatic adenocarcinoma harboring SWI/SNF chromatin remodeling gene alterations and had received ICIs: seven had an ARID1A alteration (77%); two, ARID1B (22%); three, SMARCA4 (33%); one, SMARCB1 (11%); and one, PBRM1 (11%). Three patients possessed more than one SWI/SNF complex alteration. Only three tumors were microsatellite unstable. Eight of 9 patients (89%) achieved an objective response, including a complete remission, with the two longest responses ongoing at 33+ and 36+ months. Median progression-free and overall survival was 9 and 15 months, respectively. Responses occurred even in the presence of microsatellite stability, low TMB and/or low PD-L1 expression.

Conclusions: A small subset of patients with pancreatic cancer have genomic alterations in the SWI/SNF chromatin remodeling components and these patients appear to be responsive to ICIs, suggesting the need for prospective trials.

Trial Registration: ClinicalTrials.gov NCT02478931

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INTRODUCTION

Pancreatic adenocarcinoma is currently the 3rd leading cause of cancer-related death in the United States, and over 90% of afflicted patients die from this disease (1). The only potential cure for pancreatic cancer is surgical resection; however only approximately 10-15% of patients have non-metastatic, resectable disease at diagnosis. Even after pancreaticoduodenectomy, the five-year survival is ~10% for lymph node positive disease and slightly improved (~30%) with lymph node negative disease (2). For the majority of patients presenting with metastatic disease, the two-year survival is 6.4% with a five-year survival of only 2.5%(3).

Chemotherapy in the form of FOLFIRINOX (5-fluoruracil, leucovorin, irinotecan, oxaliplatin) or gemcitabine and nab-paclitaxel are the backbone of modern metastatic pancreatic cancer therapy (4, 5). Generally, each regimen is chosen on the basis of the patient’s performance status and ability to cope with the specific side effect profiles of the two regimens. As many patients with metastatic pancreatic cancer suffer from multiple co-morbidities that develop with their disease, they are often subjected to treatment interruptions, dose reductions, and palliative single-agent therapies.

Meanwhile, the use of other targeted compounds has proven futile in pancreatic cancer, perhaps because they have been generally applied without biomarker selection (6). Specifically, small molecule inhibitors, tumor microenvironment regulators, and immunotherapy have all failed in late-phase pancreatic cancer clinical trials. While limited success (i.e. an increase of progression-free survival (PFS) of about two weeks) was described with the epidermal growth factor receptor (EGFR) tyrosine kinase inhibitor erlotinib, it is rarely used because of the paltry improvement in outcome and side effect profile (7). However, gene profiling of pancreatic cancer has uncovered that ~5% of patients with metastatic pancreatic cancer harbor a germline BRCA mutation and respond favorably to platinum based chemotherapies followed by maintenance small molecule poly(ADP-ribose) polymerase (PARP) inhibitors (8).

A modality of interest across all cancers is immune checkpoint inhibitors (ICIs). To date, six anti-programmed death receptor-1/programmed death receptor-ligand 1 (PD-1/PD-L1) agents and
One anti-cytotoxic T-lymphocyte-associated protein 4 (CTLA-4) agent are approved for a variety of cancers (9). Recently, approval of pembrolizumab has been extended to include microsatellite instability high (MSI-H)/deficient mismatch repair (dMMR) and tumor mutation burden (TMB) $\geq$10 mutations/Mb cancers regardless of tissue of origin (10–12). Even so, investigators have questioned the threshold of TMB of 10 mutations/Mb, especially for cancers such as pancreatic in which few immunotherapy responses have been documented. Furthermore, only $\sim$2% of pancreatic cancer patients have MSI-H and they are usually within clusters of Lynch syndrome families (13, 14). As such, over 98% of pancreatic cancers are deemed 'cold', having an uninflamed microenvironment incapable of spurring an immune response to checkpoint inhibition (15–18). Indeed, trial after trial has shown that immunotherapeutics have limited efficacy in advanced and metastatic pancreatic cancer when evaluating unselected patients, providing little improvement in survival beyond traditional chemotherapy (15, 19–24).

The SWI/SNF (Switch/Sucrose Non-Fermentable) complex is a sub-family of adenosine triphosphate (ATP)-dependent chromatin remodeling proteins that alter nucleosome topology and DNA access, ultimately regulating gene transcription (25) (Figure 1). Although gene alterations in individual complex members are common during carcinogenesis, their role in phenotype and exploitability for drug targeting are not well delineated. Pancreatic cancer has been found to have SWI/SNF complex alterations between 2.5% to $\sim$18% of the time by large-scale sequencing (26). Inactivating mutations, deletions, substitution or frameshift alterations, insertions, allelic loss, rearrangement, or truncation in several SWI/SNF genes, including but not limited to ARID1A, SMARCA4 and PBRM1, have been implicated in responsiveness to ICIs in a variety of cancers (27–34). This is perhaps due to lower expression of SWI/SNF complex members is associated with higher CD8+ cytotoxic T cell activity in human cancers, and their inactivation in human cancer cell lines sensitizes tumors to T cell-mediated cytotoxicity (27, 33, 35–37). To date, however, there has been no analysis as to whether this sub-population of pancreatic cancer is responsive to immunotherapy compared to clinical trials evaluating all-comers.
Herein, we examined patients with refractory metastatic pancreatic adenocarcinoma who showed alterations in one or more SWI/SNF complex chromatin remodeling genes and who received an anti-PD-L1 or anti-PD-1 agent. Our results suggest that patients harboring SWI/SNF-altered pancreatic cancer can respond to immune checkpoint inhibition.
RESULTS

**Patient Demographics:** There were 6,831 eligible cancer patients in the PREDICT database. Of these individuals, 293 (4%) patients had metastatic pancreatic cancer and, of these, 123 (42%) had had clinical-grade (tissue DNA and/or blood ctDNA) NGS performed (Figure 2). These 123 pancreatic cancer patients were further sub-stratified to those fifteen persons (12%) with definable, SWI/SNF complex alterations found by NGS. Of this cohort, nine patients had SWI/SNF complex alterations and were evaluable for checkpoint inhibitor immunotherapy. Six patients with metastatic pancreatic cancer and SWI/SNF complex alterations not treated with immunotherapy were also analyzed.

Of the nine evaluable immunotherapy-treated, SWI/SNF-altered pancreatic cancer patients, three were men (33%), six were women (67%), and the median age was 66.5 years (range, 47 to 79 years) (Table 1). The patients came from a variety of race and ethnicities: Hispanic/Latino, African American, White, Asian, and Pacific Islander. The median follow up from the date of starting immune checkpoint inhibition was 9 months and the mean was 12.7 months (range, 1 to 36 months). Of the six evaluable non-immunotherapy-treated, SWI/SNF-altered patients, three were men (50%), 3 were women (50%), and the median age was 64.5 years (range, 31 to 76 years) (Table 2).

**Next Generation Sequencing and Immunotherapeutic Intervention:** Altogether, in patients with available data, 3 of 11 SWI-SNF-altered tumors had TMB > 10 mutations/Mb and 3 of 12 patients had tumors that were microsatellite unstable (See Tables 1 and 2); 2 of 13 patients had intermediate expression of PD-L1 by IHC.

On tissue NGS of immunotherapy-treated, SWI/SNF-altered pancreatic cancer patients, seven total patients harbored an ARID1A alteration (77%); two, an ARID1B mutation (22%); three, a SMARCA4 mutation (33%); one, a SMARCB1 mutation (11%); and one, a PBRM1 mutation (11%). Three patients possessed more than one SWI/SNF complex alteration (ID #1: ARID1A, SMARCA4, SMARCB1; ID #5: ARID1A, ARID1B; ID #6: ARID1B, SMARCA4). Of note, four patients (44%) lacked
a KRAS mutation. (Details of both tissue and blood NGS are in Table 1 and Figure 3). The non-immunotherapy-treated SWI/SNF altered pancreatic cancer patients harbored five ARID1A alterations (83%) and one SMARCA4 mutation (17%), none possessing more than one SWI/SNF complex alteration. Interestingly, two patients of five evaluated patients (40%) also lacked a KRAS mutation (Table 2 and Figure 3).

TMB was evaluated in eight immunotherapy-treated SWI/SNF-altered patients (Table 1). The median of the highest TMB in each patient, represented by mutations per megabase (mutations/Mb) was 7.5 (range, 0 – 58). Three patients had TMB evaluated more than once, and the TMBs differed (8 versus 3.3; 7 versus 2; 11 versus 8.3 (all mutations/Mb)). In each of these patients, the TMB was evaluated by different laboratories and from different biopsy specimens. The majority of patients had proficient mismatch repair (MMR) proteins (N=6; 67%), but three patients had deficient MMR proteins (33%). Six patients (67%) had PD-L1 low scores (0-1%), two (22%) were intermediate (2-49%), and one was not determined (ID #5) (all IHC)(38). Seven total pancreatic cancer patients were evaluated for tumor infiltrating lymphocytes (TILs) with three immunotherapy treated patients having moderately to highly inflamed and the last having no TILs (ID #8). Those not treated with immunotherapy but with TIL data (N=3) had either no TILs or moderate TIL infiltration.

The average number of previous therapies prior to immunotherapy in the SWI/SNF-altered group was 1.7 (range, 1 - 4) with the overwhelming majority having been exposed to FOLFOX / FOLFIRINOX (N=8; 89%) and three exposed to a gemcitabine regimen (33%). Other therapies tried before immunotherapy included capecitabine, cisplatin, and olaparib. In the six patients with SWI/SNF complex alterations not receiving immunotherapy, the average number of prior therapies was similar (1.8, range 1-3) to those in the SWI/SNF-altered patients who had immunotherapy (Table 2).

When immunotherapy was initiated, pembrolizumab was given most commonly. Other immune checkpoint inhibitor (ICI) drugs included durvalumab, nivolumab, and atezolizumab.
**Efficacy:** For the nine immunotherapy-treated SWI/SNF-altered metastatic pancreatic cancer patients, at date of data cut-off, median PFS was 9 months (95% confidence interval (CI), 3.1-14.8 months) and median OS was 15 months (95% CI, uncalculatable) (Figure 4). In comparison, non-immunotherapy treated SWI/SNF-altered metastatic pancreatic cancer patients had a median PFS of 4 months (95% CI, 0-8.8 months) and a median OS of 10 months (95% CI, 0-20.8 months) (p=0.05 and 0.06, respectively, for PFS and OS difference between immunotherapy-treated and non-treated SWI/SNF-altered pancreatic cancer patients). It should be kept in mind that the small number of patients precludes robust statistical analysis.

Altogether, 8 pancreatic cancer patients (89%) achieved an objective response in the SWI/SNF-altered, immunotherapy-treated population with one achieving a CR (ID#1). In the non-immunotherapy-treated SWI/SNF-altered treated group, no patients achieved an objective response.

Three patients with SWI/SNF-altered, immunotherapy-treated patients had mismatch repair deficient disease (Table 1, ID#1, #4, and #5); two of these patients achieved a PR, and one a CR— their PFS was 15 months (for the CR), 36+ and 9+ months (for the PRs). In the two patients with mismatch repair deficient disease in whom tumor mutational burden (TMB) was assessed, it was high: 58 and 23.8 mutations/Mb. Patient ID#1 had ARID1A, SMARCA4 and SMARCB1 alterations while ID#4 had a SMARCA4 alteration and ID#5, an ARID1A and an ARID1B alteration. The remaining six patients' tumors were mismatch repair proficient. Of these patients, five achieved a PR and one was not evaluable for response (since they died of pneumonitis one month after starting therapy). The PFS in the patients attaining PR lasted 3, 4, 7+, 11, and 33+ months. The TMB in these patients ranged from 0 to 11 mutations/Mb. These patients had alterations in ARID1A (ID#2, #7, #8, and #9), PBRM1 (ID#3), ARID1B and SMARCA4 (ID#6).

Importantly, four pancreatic cancer patients in the database were treated with immunotherapy, even though they had no SWI/SNF alteration (Supplemental Table 2). The median PFS of the pancreatic cancer patients without SWI/SNF complex mutations who received immunotherapy was...
poor (median PFS = 2 months, OS = 9 months). The numbers are small, but these results are consistent with the literature.

SWI/SNF alterations in pancreatic cancer do not correlate with a better prognosis. By pooling five molecularly characterized pancreatic cancer gene datasets, we determined the prognostic impact of SWI/SNF complex alterations on overall survival (Supplemental Figure 1). There was no difference in survival between individuals with SWI/SNF-altered pancreatic cancer versus those with SWI/SNF wild-type pancreatic cancer.
Several studies have failed to show efficacy for immune checkpoint inhibition in pancreatic adenocarcinoma (39). It is postulated that the lack of responsiveness is due to a highly immunosuppressive tumor microenvironment, which makes it comparable to an immune-privileged site. In addition to PD-1 checkpoint inhibition, increased immunosuppressive cells such as T-regulatory cells (Tregs), myeloid derived suppressor cells (MDSCs), and tissue-associated macrophages (TAMs) restrict cytotoxic CD8+ and helper CD4+ T-cell anti-cancer responses (15, 16, 40–43). Furthermore, most immunotherapy clinical trials that have been deployed in pancreatic cancer have not offered biomarker-based patient selection(15, 39).

Previously, several factors have been demonstrated to correlate with responsiveness to immunotherapy, including TMB ≥10 mutations/Mb (11) and MSI-H(10). However, even with these tissue-agnostic biomarkers, there is a paucity of reports of these rare events and their correlation with immunotherapy outcomes in pancreatic cancer (13). Interestingly, in the pan-cancer setting, aberrations in chromatin remodeling genes of the SWI/SNF complex have also been shown to correlate with enhanced efficacy of immune checkpoint inhibition, although some of the data is inconsistent (27). For instance, ARID1A alterations are associated with better outcomes after immunotherapy across histologies; PBRM1 alterations correlate with responsiveness to immunotherapy in some publications but not in others; and alterations in another chromatin remodeling gene--SMARCA4--are associated with responsiveness of small cell carcinoma of the ovary, hypercalcemic type (SCCOHT), to immunotherapy (27, 35, 44).

To our knowledge, this is the first study to evaluate immunotherapy in a group of patients with advanced pancreatic cancer who harbored alterations in chromatin remodeling genes. Pancreatic cancer genomic characterization through TCGA found SWI/SNF alterations in 10% of samples (ARID1A 6% and PBRM1 4%) (45). The International Cancer Genome Consortium (ICGC) dataset found SWI/SNF alterations in 14% (46). Further, we analyzed the aggregate SWI/SNF gene alterations within pancreatic cancer patients from five genomic data sets and found an alteration rate...
of 18% (Supplemental Figure 1) (46–48). Of our entire metastatic pancreatic cancer population with NGS available (123 patients), 15 had alterations in SWI/SNF complex members (12%), which is in line with the mutation rates of these large datasets.

Altogether, of our nine treated patients, eight (89%) achieved an objective response. The longest response is ongoing past 36 months, and the best response was a CR. Median PFS and OS were 9 and 15 months, respectively. As a comparison, objective response, median PFS, and OS with traditional second-line chemotherapy in pancreatic cancer is 17%, 3.1 and 6.1 months, respectively (49). Further, first-line therapy in unselected metastatic pancreatic cancer has an average OS of approximately 11 months with either FOLFIRINOX or in contemporary gemcitabine plus nab-paclitaxel trials (5, 50, 51).

Importantly, overall, SWI/SNF complex alterations do not appear to be associated with significantly better prognostic outcomes from pooled genomic datasets (Supplemental Figure 1) (although individual SWI/SNF alterations might have prognostic significance, albeit with small numbers of patients assessed (52)). As such, the ability of immunotherapy to improve outcomes in our SWI/SNF-altered metastatic pancreatic cancer patients likely has clinical relevance. However, there are characteristics of these SWI/SNF-altered pancreatic cancers that may be relevant. For instance, microsatellite instability was seen in 3 of the 12 SWI/SNF-altered pancreatic cancers with available data in the current report, while the established rate in pancreatic cancer overall is about 2.4% (53).

If the patients within this study were evaluated for the potential use of checkpoint inhibition based on current immune biomarkers, only three would have qualified, since they had deficient mismatch repair. However, even in these types of patients, there is, up to now, a dearth of literature data specifically about the immunotherapy responsiveness of pancreatic cancer. These patients -- ID#1, ID#4 and ID#5 (Table 1) -- had MSI-H, presumably caused by the mutations in mismatch repair genes MSH2 (ID#1 and ID#5) and MSH6 (ID#4) per their molecular profile. Even so, epigenetic mechanisms such as hyper-methylation can also contribute to MSI-H; in a proteomic screen, it was
found that the ARID1A protein interacts with the mismatch repair protein MSH2, and that ARID1A protein deficiency (as occurs when ARID1A is mutated as seen in patients ID#1 and ID#5) contributes to impaired mismatch repair and a mutator phenotype (28). Of possible interest, tumors in four of our nine immunotherapy patients (44%) did not harbor a KRAS mutation. Importantly, our SWI/SNF-altered pancreatic cancer group not receiving immunotherapy similarly did not have a KRAS mutation in two of five (40%) patients with available data, underscoring the likely genomic relevance of SWI/SNF complex members in pancreatic cancer carcinogenesis. (Of note however, cBioPortal data in pancreatic cancer patients with SWI/SNF-altered genes versus not showed that KRAS was mutated in the SWI/SNF-altered group at a rate of 85.1% (N=160/188) versus SWI/SNF non-altered harboring a KRAS alteration rate of 86.2% (N = 631/732)(54, 55)). Given the fact that KRAS alterations are a hallmark driver mutation and occur in the vast majority of pancreatic cancers, whether or not the absence of KRAS mutations played a role in immune surveillance in our patients requires further investigation (56).

In terms of safety, one patient died from an immune-related adverse event (irAE) likely due to pneumonitis within one month of pembrolizumab initiation. This patient had previously been heavily pre-treated (four lines of therapy) and harbored pulmonary metastases. A second patient had a drug-induced myositis likely from the PD-L1 inhibitor atezolizumab at 17 months of treatment. Due to their ongoing partial response, the multi-disciplinary team thought it prudent to switch to the PD-1 inhibitor pembrolizumab, with ongoing good tolerance for an additional 16+ months (total PFS = 33+ months).

This study has several important limitations. First, the number of patients is small, and we did a retrospective analysis, which could be confounded by selection bias. Hence, prospective clinical trials are needed to validate the results, which should be considered preliminary. Further, not all patients had complete immune profiling. Some patients received additional agents along with their checkpoint inhibition. For instance, one patient received the MEK inhibitor trametinib with their anti-PD-1 agent; however, trametinib is not considered active in pancreatic cancer as a single agent. Another example
is a patient who had previously failed FOLFIRINOX and gemcitabine plus nab-paclitaxel, who then received pembrolizumab together with gemcitabine plus nab-paclitaxel. Another confounder was that 3 of our 9 SWI/SNF-altered tumors were MMR deficient; however, our analysis shows that five of the six MMR-proficient, SWI/SNF-altered cancers achieved a PR, while no objective responses were seen in the four patients without SWI/SNF alterations who were treated with immunotherapy (and were MMR proficient). Also, as this was not a prospective study, it was not powered to detect survival differences between groups. Finally, a variety of different laboratories provided the NGS and other immune studies; however, all laboratory tests were clinical grade.

In summary, the current analysis suggests that a subgroup of patients with pancreatic cancer and alterations in SWI/SNF complex chromatin remodeling genes, such as ARID1A, ARID1B, PBRM1, SMARCA4 and SMARCB1 can respond to immune checkpoint inhibition. Although one-third of these patients had MSI-H, the others had no mismatch repair defect and only three had a TMB ≥ 10 mutations/Mb. Further, five of the six patients (83%) with low PD-L1 by IHC achieved an objective response. Several studies have previously shown that genomic profiling can assist with patient selection for a variety of therapies (27, 57, 58). Taken together, our current data suggest that prospective studies of immune checkpoint inhibition are warranted in patients with advanced pancreatic cancer whose tumors harbor alterations in chromatin remodeling genes.

**Acknowledgements:** This work was supported in part by the Joan and Irwin Jacobs Fund and by National Cancer Institute at the National Institutes of Health [Grant No. NIH P30 CA023100 (RK) and LRP KYGF9753 (GPB), as well as the Gershenson Family, the Duarte Family, and anonymous patient donors (GPB).
METHODS

Study population & approval: Patient data was curated from the electronic medical records. The Profile Related Evidence Determining Individualized Cancer Therapy (PREDICT, NCT02478931) database of eligible patients at the Center for Personalized Cancer Therapy (University of California San Diego Moores Cancer Center), whose tissue DNA was analyzed by next-generation sequencing (NGS) was searched for patients who had clinically staged metastatic and histologically confirmed pancreatic ductal adenocarcinoma only, had completed NGS, and were treated with checkpoint inhibitor immunotherapy. The cBio Cancer Genomics Portal was analyzed for five additional pancreatic adenocarcinoma gene datasets. For survival analysis across all pancreatic cancer patients from the five available datasets (ICGC, Nature 2012; QCMG, Nature 2016; TCGA, Firehose Legacy; TCGA, PanCancer Atlas; UTSW, Nat Commun 2015), we stratified by SWI/SNF complex alteration and evaluated overall survival.

Molecular testing and other biologic markers: NGS of tissue DNA and/or blood circulating tumor DNA (ctDNA) was performed in clinical laboratory improvement amendment (CLIA) laboratories, including, most commonly, Foundation Medicine (foundationmedicine.com), Tempus (tempus.com), and University of California, San Diego (UCSD) for tissue DNA (NGS panel sizes from >180 to >400 genes); and Guardant (guardanthealth.com) (panel size ~70 genes), the latter most commonly for ctDNA NGS. Other platforms for NGS testing included Caris (carismolecularintelligence.com), Omniseq (omniseq.com), Paradigm (paradigmdx.com), Nanthealth (nanthealth.com), and Pathline (pathlinelabs.com).

Patients had their NGS report evaluated for alterations in SWI/SNF complex members focusing on ARID and SMARC family genes as well as PBRM1 (Supplemental Table 1). Only characterized SWI/SNF complex members were evaluated; all variants of unknown significance were excluded from further study.
Mismatch repair (MMR) protein proficiency was determined by expression of MLH1, MSH2, PMS2, and MSH6 IHC staining by a CLIA-licensed laboratory. Tumor mutation burden (TMB) was determined either by subtracting germline from somatic tumor sequencing when available or by computational analysis when only tumor sequencing was available. Although gathered from multiple labs and chronologic specimens, TMB were all standardized and expressed as the number of mutations per megabase (mut/Mb). All TMB analysis was prior to ICB therapy and stratified as: low (<6 mut/Mb), intermediate (6-19 mut/Mb), or high (>19 mut/Mb)\(^\text{(11)}\). Immune profiling was done per laboratory specification and at a minimum included PD-L1 determination by IHC while more expansive panels evaluated programmed death receptor 1 (PD-1), tumor infiltrating leukocytes (TILs), and the quantity of infiltrating CD8+ T-cells. PD-L1 expression was stratified on the following scale: low (0-1%), intermediate (2-49%), high (50%+) using the Dako 22C3 pharmDx qualitative immunohistochemical assay\(^\text{(38)}\).

**Statistical analysis and endpoints:** Progression-free survival (PFS) and overall survival (OS) were measured from the first date of immunotherapy until the cut-off date of 6/1/2020 (or the last time of contact). These were plotted by the Kaplan-Meier method and any patients who did not progress or were alive at the date of data cut-off (or time of last contact) were censored for PFS or OS, respectively, at that time point. Responses (partial response (PR) and complete response (CR)) were evaluated by RECIST assessment per physician. Statistical analysis was completed using the SPSS software package.

**Study Approval:** This study was performed in accordance with UCSD Institutional Review Board guidelines for data analysis and for any investigational treatments for which patients gave consent.

**Author Contributions:** GPB, KS, HP, PF, SL, and RO collected patient data, GPB, KS and RK conceived, analyzed, and wrote manuscript. All authors edited and approved manuscript.
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**Figure 1: SWI/SNF Complexes.**

The human SWI/SNF complex mediates chromatin remodeling and is composed of two sub-classes: BAF and PBAF structures. Multiple sub-units (between 8 and 14) comprise each structure with core homology between DNA repair (light blue) and Proliferation (green) sub-units. Transcriptional sub-units (orange) differentiate the two classes. The BAF complex has either SMARCA2 or SMARCA4 as a DNA repair sub-unit, but not both. DNA repair sub-units are implicated in the nucleotide excision and double-strand break repair. Each sub-unit is identified by its specific gene name, which is associated with the translated protein of the complex. BAF = BRG1 (SMARCA4)- or BRM (SMARCA2)-associated factors. PBAF = Polybromo-associated BAF.
Pancreatic cancer patients with the aforementioned tumor and treatment characteristics were extracted from the Profile-Related Evidence Determining Individual Cancer Therapy (PREDICT) database at the University of California San Diego Center for Personalized Cancer Therapy. Nine total evaluable patients were identified with SWI/SNF alterations and who received immunotherapy. Six patients with SWI/SNF alterations who did not receive immunotherapy and 4 pancreatic cancer patients without SWI/SNF alterations who did not receive immunotherapy were used as comparator arms.
Figure 3. Molecular Characteristics of Pancreatic Cancer Patients with SWI/SNF Alterations with and without Immunotherapy.

Molecular analysis of each patient (ID). Orange boxes represent point mutations, blue boxes are deletions, red boxes are insertions, green boxes are gain-of-function (GOF), and purple boxes are fusions. *KRAS* mutations are described as none (X) or the specified point mutation. Tumor mutation burden (TMB) is described by mutations per megabase (mut/Mb) or none (X). PD-L1 is stratified on the scale: low (0-1%), intermediate (2-49%), high (50%+), or none (X) using the Dako 22C3 pharmDx qualitative immunohistochemical assay of tumor cells (38). The total number of genes analyzed per patient tumor sample is specified.
Figure 4. Kaplan-Meier Estimates of Progression-free Survival and Overall Survival in Patients with Metastatic Pancreatic Ductal Adenocarcinoma Harboring SWI/SNF Complex Alterations Treated with Immunotherapy and without.

The median progression-free survival (PFS) was 9 months in the immunotherapy treated pancreatic cancer patients with SWI/SNF alterations (Panel A, blue line) versus 4 months in the patients not receiving immunotherapy (Panel A, red line) (p = 0.05 by log rank). The median overall survival (OS) was 15 months for the immunotherapy treated pancreatic cancer patients with SWI/SNF alterations (Panel B, blue line) and 10 months for the patients not receiving immunotherapy (Panel B, red line) (p = 0.06 by log rank). Tick marks indicate censored data.
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<td>FOLFIRINOX</td>
<td>Pembrolizumab</td>
<td>CR</td>
<td>15</td>
<td>15</td>
<td>_</td>
</tr>
<tr>
<td>#2 60 F</td>
<td>Paradigm</td>
<td>ARID1A E2033, CCNE1 Gain, KRAS G12V, TP53 R156P</td>
<td>8</td>
<td>Proficient</td>
<td>Guardant</td>
<td>TP53 R156P, KRAS G12V, TP53</td>
<td>Paradigm</td>
<td>Low</td>
<td>ND</td>
<td>2</td>
<td>FOLFOX gemcitabine + nab-paclitaxel</td>
<td>Pembrolizumab + Trametinib</td>
<td>PR</td>
<td>4</td>
<td>4</td>
<td>_</td>
</tr>
<tr>
<td>#3 47 F</td>
<td>Caris</td>
<td>CREBBPP Exon 16 E1058fs, PBRM1 Exon 12 Y417fs, VTCN1/M</td>
<td>7</td>
<td>Proficient</td>
<td>Guardant</td>
<td>CDK6 F164F</td>
<td>Caris</td>
<td>Low</td>
<td>TILs moderately inflamed PD-1 Very High</td>
<td>2</td>
<td>FOLFIRINOX Gemcitabine + nab-paclitaxel</td>
<td>Pembrolizumab plus nab-paclitaxel + gemcitabine</td>
<td>PR</td>
<td>11</td>
<td>21+***</td>
<td>_</td>
</tr>
<tr>
<td>#4 70 F</td>
<td>Founda</td>
<td>SMARCA4 R1135W, ERBB3 G284R, CDKN2A R68, DNMT3A G543A, TP53 R273C, KRAS G12V, CDKN2A R58, FBXW7 R505C, MSH6 2521delA</td>
<td>ND</td>
<td>Deficient</td>
<td>ND</td>
<td>ND</td>
<td>Omniseq</td>
<td>Low</td>
<td>TILs 10% - 20%</td>
<td>1</td>
<td>FOLFIRINOX</td>
<td>Durvalumab based therapy</td>
<td>PR</td>
<td>36+</td>
<td>36+</td>
<td>_</td>
</tr>
<tr>
<td>#5 68 F</td>
<td>Tempus</td>
<td>ARID1A M835fs, KRAS G12D, MEN1 R516fs, KMT2C K2797fs, NBN R466fs, FAT1 F3952fs, POT1 38-2A&gt;G, KMT2D P647fs, SLT2 N155fs, KMT2D E5011, SMAD2 R427, ARID1B R1944, RPL5 D59fs, RAD50 E723fs, MSH2 N64fs</td>
<td>23.8</td>
<td>Deficient</td>
<td>Guardant</td>
<td>Negative</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>1</td>
<td>FOLFIRINOX</td>
<td>Pembrolizumab</td>
<td>PR</td>
<td>9+</td>
<td>9+</td>
<td>_</td>
</tr>
<tr>
<td>Case # / Age / Sex</td>
<td>Tissue NGS Vendor</td>
<td>Pertinent Tissue NGS Findings</td>
<td>TMB *</td>
<td>Mismatch Repair</td>
<td>Blood NGS Vendor</td>
<td>Pertinent Blood NGS Findings</td>
<td>Immune Vendor</td>
<td>PD-L1 **</td>
<td>Immune Profile</td>
<td># Prior Lines of Therapy</td>
<td>Prior Lines of Therapy</td>
<td>Best Response</td>
<td>PFS (months)</td>
<td>OS (months)</td>
<td>Comment</td>
<td></td>
</tr>
<tr>
<td>-------------------</td>
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</tr>
<tr>
<td>#6 66 M</td>
<td>Foundation</td>
<td>ARID1B A757T, SMARCA4 S312Y, ALK V564M, IRS2 N28, H29insN, MGAl E867K, MPL I492M, NOTCH1 G269R, TSC2 A490T, ERBB3 G337F, BRCA2 S1982fs, FBXW7 L709fs, AXIN 1 R539, H534insQIVHH, BCCOR1 splice site 4531_4618+102del190, EP300 E1492, KEP splice site 1532-2A&gt;T</td>
<td>11</td>
<td>Proficient</td>
<td>ND</td>
<td>ND</td>
<td>Caris</td>
<td>PD-1 = SHPF</td>
<td>1</td>
<td>FOLFOX</td>
<td>PR</td>
<td>33+</td>
<td>33+</td>
<td>Switched from atezolizumab to pembrolizumab in setting of drug induced myositis</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Tempus</td>
<td>BRCA2 S1982fs, KEAP 1532-2A&gt;T, EP300 E1492, NBN 5615fs, BCCOR1 Y1585fsPD-L1 5%</td>
<td>8.3</td>
<td>Proficient</td>
<td>Tempus</td>
<td>ND</td>
<td>Paradigm</td>
<td>Int.</td>
<td>1</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>#7 60 M</td>
<td>Foundation</td>
<td>ARID1A Q2039fs, EZR-ERBB4 fusion (E10-E18), PIK3CA R86Q</td>
<td>1</td>
<td>Proficient</td>
<td>Tempus</td>
<td>Negative</td>
<td>Omniseq</td>
<td>Low</td>
<td>TILs</td>
<td>Highly Inflamed CD8 Moderately Infiltrating</td>
<td>1</td>
<td>FOLFIRINOX</td>
<td>Nivolumab plus gemcitabine-based therapy</td>
<td>PR</td>
<td>7+</td>
<td>7+</td>
</tr>
<tr>
<td></td>
<td>Paradigm</td>
<td>KRAS G12D, ERBB2, MGMT, TOP1,</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>#8 67 F</td>
<td>Foundation</td>
<td>ARID1A L2054fs, TP53 W53, KRAS G12D, ERBB2 R678Q</td>
<td>7</td>
<td>Proficient</td>
<td>Guarda nt</td>
<td>Negative</td>
<td>Guarda nt</td>
<td>KIT H894H</td>
<td>Emerge</td>
<td>Low</td>
<td>TILs Negative PD-1 Negative</td>
<td>4</td>
<td></td>
<td>Gemcitabine + radiation</td>
<td></td>
<td>1</td>
</tr>
<tr>
<td></td>
<td>Foundation</td>
<td>ARID1A L2054fs, TP53 W53, KRAS G12D, ERBB2 R678Q, BRCA1 truncation intron 16, CDKN1B G97fs, CDKN2A/B loss</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
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</tr>
<tr>
<td>#9 67 F</td>
<td>Foundation</td>
<td>ARID1A H1843fs, BRCA1 truncation intron 16, KRAS G12V, CDKN2A/B loss, SMAD2 intron 5, TP53 T155N</td>
<td>0</td>
<td>Proficient</td>
<td>Guarda nt</td>
<td>Negative</td>
<td>Foundation</td>
<td>KRAS G12V, TP53 T155N, ARID1A H1843fs</td>
<td>Omniseq</td>
<td>Low</td>
<td>CD8 Minimally Infiltrating, TMB 4.3 mut/Mb, pMMR</td>
<td>3</td>
<td></td>
<td>Olaparib</td>
<td>PR</td>
<td>3</td>
</tr>
</tbody>
</table>

* = mutations per megabase (mut/Mb)
** = stratified on the scale: low (0-1%), intermediate (2-49%), high (50%+) using the Dako 22C3 pharmDx qualitative immunohistochemical assay of tumor cells(38).
*** = ** means ongoing response

Abbreviations: CR = Complete Response; dMMR = deficient mismatch repair; F = female; HPF = high powered field; IE = Inevaluable; Int. = Intermediate; I/O = Immunotherapy; irAE = immune related adverse event; M = male; ND = Not done; NGS = next generation sequencing; OS = median overall survival; PD-1 = Programmed death receptor–1; PFS = Median Progression Free Survival; pMMR = proficient mismatch repair; PR = partial response; S-1 = tegafur, gimeracil, oteracil; SD = stable disease; TILs = tumor infiltrating leukocytes; TMB = tumor mutation burden
Table 2: Characteristics of Six Patients with Pancreatic Cancer Harboring SWI/SNF Complex Alterations Not Receiving Immunotherapy

<table>
<thead>
<tr>
<th>Case # / Age / Sex</th>
<th>Tissue NGS Vendor</th>
<th>Pertinent Tissue NGS Findings</th>
<th>TMB*</th>
<th>Mismatch Repair</th>
<th>Blood NGS Vendor</th>
<th>Pertinent Blood NGS Findings</th>
<th>Immune Vendor</th>
<th>PD-L1**</th>
<th>Immune Profile</th>
<th># Prior Lines of Therapy</th>
<th>Prior Lines of Therapy</th>
<th>Best Response</th>
<th>PFS (months)</th>
<th>OS (months)</th>
<th>Comment</th>
</tr>
</thead>
<tbody>
<tr>
<td>#10 65 / M</td>
<td>Foundation</td>
<td>KRASG12D, ARID1A, CDKN2A/B, loss, MLL2</td>
<td>4</td>
<td>Proficient</td>
<td>Foundation</td>
<td>KRASG12D</td>
<td>Foundation</td>
<td>0%</td>
<td>ND</td>
<td>1</td>
<td>5FU</td>
<td>PD</td>
<td>0</td>
<td>1</td>
<td></td>
</tr>
<tr>
<td>#11 76 / F</td>
<td>Foundation</td>
<td>KRASG12D, TP53, ARID1A, CDKN2A/B, SMAD4</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>Pathline</td>
<td>0%</td>
<td>TILs 0%</td>
<td>1</td>
<td>Gem/Abx</td>
<td>SD</td>
<td>4</td>
<td>10</td>
<td></td>
<td></td>
</tr>
<tr>
<td>#12 64 / M</td>
<td>Foundation</td>
<td>KRASG12D, BRD4, CDKN2A, RBM10, SMAD4, SMARCA4, TP53</td>
<td>4</td>
<td>Proficient</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>1</td>
<td>Gem/Abx</td>
<td>PD</td>
<td>2</td>
<td>2</td>
<td></td>
</tr>
<tr>
<td>#13 31 / F</td>
<td>Foundation</td>
<td>PTEN, ARID1A, CDKN2A/B, TP53</td>
<td>4</td>
<td>Proficient</td>
<td>Guardant</td>
<td>ARID1A, TP53, PTEN</td>
<td>Foundation</td>
<td>0%</td>
<td>ND</td>
<td>3</td>
<td>FOLFIRINOX, Irinotecan, 5FU</td>
<td>SD</td>
<td>6</td>
<td>11</td>
<td></td>
</tr>
<tr>
<td>#14 73 / F</td>
<td>Foundation</td>
<td>TP53, SMAD4, ARID1A</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>Emerge</td>
<td>0%</td>
<td>TILs 1-24%</td>
<td>2</td>
<td>Gem/Abx, Xeloda</td>
<td>SD</td>
<td>7</td>
<td>12</td>
<td></td>
<td></td>
</tr>
<tr>
<td>#15 62 / M</td>
<td>Foundation</td>
<td>KRASG12D, ARID1A, RB1, TP53</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>Foundation</td>
<td>0%</td>
<td>TILs 5%</td>
<td>3</td>
<td>Gem/Abx, 5-FU, Irinotecan</td>
<td>SD</td>
<td>7</td>
<td>14</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

* = mutations per megabase (mut/Mb)
** = stratified on the scale: low (0-1%), intermediate (2-49%), high (50%+) using the Dako 22C3 pharmDx qualitative immunohistochemical assay of tumor cells(38).
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