Published in final edited form as:

Cancer Immunol Res. 2020 August ; 8(8): 1075–1084. doi:10.1158/2326-6066.CIR-19-0866.

Mammalian SWI/SNF complex genomic alterations and immune checkpoint blockade in solid tumors

Sarah Abou Alaiwi, M.D.^{1,†}, Amin H. Nassar, M.D.^{1,2,†}, Wanling Xie, M.S.³, Ziad Bakouny, M.D.¹, Jacob E. Berchuck, M.D.¹, David A. Braun, M.D., Ph.D.¹, Sylvan C. Baca, M.D., Ph.D.¹, Pier Vitale Nuzzo, M.D., Ph.D.^{1,5}, Ronan Flippot, M.D.^{1,6}, Tarek H. Mouhieddine, M.D.⁴, Liam F. Spurr, B.S.⁴, Yvonne Y. Li, Ph.D.⁴, Taiwen Li, Ph.D.^{3,8}, Abdallah Flaifel, M.D.¹, John A. Steinharter¹, Claire A. Margolis, M.S.⁴, Natalie I. Vokes, M.D.⁴, Heng Du, M.D.², Sachet A. Shukla, M.S.⁴, Andrew D. Cherniack, Ph.D.⁴, Guru Sonpavde, M.D.¹, Robert I. Haddad, M.D.⁴, Mark M. Awad, M.D., Ph.D.⁹, Marios Giannakis, M.D., Ph.D.⁴, F. Stephen Hodi, M.D.¹⁰, X. Shirley Liu, Ph.D.³, Sabina Signoretti, M.D.⁷, Cigall Kadoch, Ph.D.¹¹, Matthew L. Freedman, M.D.⁴, David J. Kwiatkowski, M.D., Ph.D.², Eliezer Van Allen, M.D.⁴, Toni K. Choueiri, M.D.¹

¹Lank Center for Genitourinary Oncology, Dana-Farber Cancer Institute, Boston, Massachusetts

²Department of Medicine, Brigham and Women's Hospital, Boston, Massachusetts

³Department of Data Sciences, Dana-Farber Cancer Institute, Boston, Massachusetts

⁴Department of Medical Oncology, Dana-Farber Cancer Institute, Boston, Massachusetts

⁵Department of Internal Medicine and Medical Specialties (DIMI), School of Medicine, University of Genoa, Genoa, Italy

⁶Department of Medical Oncology, Gustave Roussy, Villejuif, France

⁷Department of Pathology, Brigham and Women's Hospital, Boston, Massachusetts

⁸State Key Laboratory of Oral Diseases, West China Hospital of Stomatology, Sichuan University, Chengdu, China

⁹Lowe Center for Thoracic Oncology, Dana-Farber Cancer Institute, Boston, Massachusetts

¹⁰Melanoma Center, Dana-Farber Cancer Institute, Boston, Massachusetts

¹¹Department of Pediatric Oncology, Dana Farber Cancer Institute and Harvard Medical School, Boston, Massachusetts

Abstract

Prior data have variably implicated the inactivation of the mammalian SWItch/Sucrose Non-Fermentable (mSWI/SNF) complex with increased tumor sensitivity to immune checkpoint inhibitors (ICIs). Herein, we examined the association between mSWI/SNF variants and clinical

Corresponding Author: Toni K. Choueiri, M.D., Lank Center for Genitourinary Oncology, Dana-Farber Cancer Institute, 450 Brookline Ave, Boston, Massachusetts, 02215 (Toni_Choueiri@DFCI.HARVARD.EDU). Tel: +1 617 632456; Fax: +1 617 6322165.

[†]these authors contributed equally to this work.

The **other authors** report no conflicts of interest.

outcomes to ICIs. We correlated somatic loss-of-function (LOF) variants in a pre-defined set of mSWI/SNF genes (*ARID1A*, *ARID1B*, *SMARCA4*, *SMARCB1*, *PBRM1*, and *ARID2*) with clinical outcomes in cancer patients treated with systemic ICIs. We identified 676 patients from Dana Farber Cancer Institute (DFCI) and 848 patients from a publicly available database from Memorial Sloan Kettering Cancer Center (MSKCC) who met the inclusion criteria. Multivariable analyses were conducted and adjusted for available baseline factors and tumor mutational burden (TMB). Median follow-up was 19.6 (17.6–22.0) months and 28.0 (25.0–29.0) months for the DFCI and MSKCC cohorts, respectively. Seven solid tumor subtypes were examined. In the DFCI cohort, LOF variants of mSWI/SNF did not predict improved overall survival (OS), time to treatment failure (TTF), or disease control rate (DCR). Only renal cell carcinoma patients with mSWI/SNF LOF showed significantly improved OS and TTF with adjusted hazard ratios (95% CI) of 0.33 (0.16–0.7) and 0.49 (0.27–0.88), respectively, and this was mostly driven by *PBRM1*. In the MSKCC cohort, where only OS was captured, LOF mSWI/SNF did not correlate with improved outcomes across any tumor subtype. We did not find a consistent association between mSWI/SNF LOF variants and improved clinical outcomes to ICIs, suggesting that mSWI/SNF variants should not be considered as biomarkers of response to ICIs.

Keywords

immune checkpoint inhibitors; biomarker; survival; response; mSWI/SNF

Introduction

Since their introduction into clinical practice, immune checkpoint inhibitors (ICIs), namely anti-PD-1/PD-L1 and anti-CTLA-4, have proven to be an effective antineoplastic drug class across several cancer subtypes (1–15). Despite the durable responses achieved with ICIs, the majority of patients do not respond, and universal determinants of clinical benefit are still lacking (16,17). Therefore, there remains an unmet need to develop predictive biomarkers essential to improve patient benefit, reduce the risk of toxicity, and advance combination strategies.

Over the past few years, evidence from both pre-clinical and clinical settings has implicated the mammalian SWItch/Sucrose Non-Fermentable (mSWI/SNF) complex as one of the players impacting response to ICIs. The mSWI/SNF complex is one of the key factors regulating gene expression and thus plays an important role in cell division, cell differentiation, and DNA replication (18,19). This complex exists in three large macromolecular complexes: BRG1/BRM-associated factor (BAF), polybromo-associated BAF (PBAF), and non-canonical BAF (ncBAF) complexes, which are collectively composed of the protein products of 29 genes (19). Whole-exome sequencing efforts have shown that 20% of human cancers harbor mutations in at least one the 29 genes involved in this complex (20). Notably, a wide range of mutation frequencies exists among the different mSWI/SNF genes, with significant variation across different cancer subtypes. These mutational patterns may be partially explained by the cell type-specific expression and function of the various subunits across cancers (20,21). For example, *SMARCA4* genomic alterations (GAs) are relatively common in non-small-cell lung cancers (NSCLCs, 11%)

(22,23), *ARID1A* in ovarian clear-cell carcinomas (~50%)(24) and urothelial carcinoma (UC, 29%)(25), *PBRM1* in renal cell carcinoma (RCC, 40%)(26), and *ARID2* in melanoma (MEL, 7%)(27).

Preclinical evidence has associated mSWI/SNF inactivation with increased CD8⁺ T-cell infiltration, enhanced sensitivity to T cell-mediated cytotoxicity, and improved tumor response to ICIs in some cancer types (28,29). A study has shown that *ARID1A*-deficient tumors in syngeneic mice demonstrate significantly increased tumor-infiltrating lymphocytes and improved response to ICIs compared to controls (29). Interestingly, analysis of The Cancer Genome Atlas (TCGA) data has linked decreased expression of multiple mSWI/SNF genes (*SMARCA4*, *PBRM1*, *ARID1A*, *ARID2*) with increased mRNA levels of stimulated 3' antisense retroviral coding sequences (SPARCS), a subclass of endogenous retroviruses that triggers innate immunity (30).

Clinically, several studies have assessed genetic correlates of response to checkpoint blockade, with a focus on the mSWI/SNF complex (Supplementary Table S1)(31–41). A salient feature of these studies is the variability of evidence in relation to whether loss of the mSWI/SNF correlates with clinical benefit in patients treated with ICIs across different lines of ICI therapy. For example, in renal cell carcinoma (RCC), loss-of-function (LOF) mutations in *PBRM1* in the post tyrosine-kinase inhibitor (TKI) setting associates with clinical benefit to the single-agent PD-1 inhibitor, nivolumab (40). This was later validated in a more comprehensive analysis of a randomized controlled trial of RCC patients treated with nivolumab vs. everolimus (42). In contrast, two other clinical trials in the frontline RCC setting did not find a significant association between *PBRM1* alterations and improved clinical outcomes in patients treated with either PD-L1-based combination therapies or atezolizumab alone (8,39,43).

Nearly all prior studies on the mSWI/SNF complex have focused on individual tumor types without studying pan-cancer relationships (40). These studies were also very heterogeneous overall in their assessments, including the examined tumor histologies, the considered line of ICI therapy, the reported patient outcomes, as well as the assessed mSWI/SNF GAs. Herein, in two independent cohorts, we systematically assessed the association between LOF variants in mSWI/SNF subunits and outcomes of cancer patients treated with ICIs, using extensive clinical phenotyping and rigorous statistical analyses across multiple cancer types.

Materials and Methods

Study design and patient cohorts

We tested our hypothesis in two independent cohorts: Dana-Farber Cancer Institute (DFCI) cohort of 676 patients and an external cohort from Memorial Sloan Kettering Cancer Center (MSKCC) of 848 patients (44). We included patients with solid tumor histologies, where ICIs were FDA-approved for therapy, who received at least one dose of an anti-PD-1/PD-L1 or anti-CTLA-4 agent in the metastatic setting, and who had next-generation targeted sequencing (NGS) of their tumor tissue performed (as described below). Included tumor histologies were MEL, NSCLC, RCC, UC, colorectal adenocarcinoma (CRC), esophago-gastric adenocarcinoma (EGC), head and neck squamous cell carcinoma (HNSCC), cancer

of unknown primary (CUP), and small-cell lung cancer (SCLC). The number of patients per each tumor histology is detailed in Table 1. Patients were excluded if they were lost to follow-up, had no measurable disease, or had clinical deterioration within one week of the first ICI dose. Tumors with missense mutations were excluded from the analysis, as we could not confidently assess functional outcomes of these mutations. The patient studies were conducted in accordance with the ethical guidelines of the Declaration of Helsinki. This study was performed after approval by the Institutional Review Board (IRB) of DFCI, and informed written consent was obtained from each subject or each subject's guardian. The MSKCC data was de-identified and publicly available. For the DFCI cohort, tissue collected encompassed primary and metastatic tumors from core biopsies and/or surgical resections. In addition to tumor tissue, patients in the MSKCC cohort also had matched normal or blood collected. Tissue specimens were collected between 2009–2018 for DFCI and between 2013–2017 for MSKCC and were stored as formalin-fixed paraffin-embedded tissue. DNA was extracted from blood after collection and was stored at -20°C if not proceeding directly to the library preparation.

Data collection

The clinical variables assessed included gender, primary tumor site, age at initiation of ICI therapy, type and class of ICI-based regimen for both cohorts. Additional clinical variables available for the DFCI cohort included lines of therapy prior to ICIs and site of lesion subjected to targeted sequencing. Patients were excluded if they were lost to follow-up, had no measurable disease, or had clinical deterioration within one week of the first ICI dose. Microsatellite instability (MSI) analysis for the DFCI CRC cohort was determined using immunohistochemistry.

Genomic analysis

Details of the tissue collection, DNA extraction, and tumor targeted sequencing using the Oncopanel/PROFILE and MSK-IMPACT for DFCI and MSKCC cohorts, respectively, were previously described in detail (panel of genes assessed in Supplementary Table S2)(45–49). Briefly, for the DFCI cohort, core biopsy and/or surgical resection specimens were reviewed by Brigham and Women's Hospital (BWH) board-certified pathologists to confirm the diagnosis, histological subtype, tumor grade, and stage. Tumor regions consisting of at least 20% tumor cells were macro-dissected from unstained slides, and DNA was isolated using the QIAamp DNA FFPE Tissue Kit (Qiagen) according to the manufacturer's instructions. DNA quantification was performed by Nanodrop and Pico-Green assays. Targeted gene sequencing was performed using an institutional analytic platform, Oncopanel, that is certified for clinical use and patient reporting under the Clinical Laboratory Improvement Amendments (CLIA) act. 200 ng genomic DNA from each sample was subject to targeted exon capture and sequencing using one of two versions of the Oncopanel assay (V2-V3) developed at BWH. The Oncopanel gene panel includes capture probes for 275–447 cancer-associated genes, as well as intronic portions of 60 genes for rearrangement detection (46). Targeted capture was performed using a solution-phase Agilent SureSelect hybrid capture kit and custom bait-sets. Sequencing libraries were prepared from captured DNA, as described in detail elsewhere (46). Paired-end sequencing was performed on an Illumina HiSeq 2500. Reads were de-multiplexed using Picard tools (<http://picard.sourceforge.net>) and aligned to

human reference genome b37 using the Burrows-Wheeler Aligner (50) (<http://bio-bwa.sourceforge.net/bwa.shtml>).

For the MSKCC cohort, a hematoxylin and eosin (H&E) stained slide was reviewed by a molecular pathology fellow and annotated for relevant specimen information including tumor type, tumor purity, and whether macrodissection of the indicated tumor region was necessary prior to nucleic extraction. Genomic DNA extraction was performed on the Chemagic STAR instrument (Hamilton) from formalin-fixed, paraffin-embedded (FFPE) tumors and patient-matched normal blood using the chemagen magnetic bead technology (PerkinElmer). DNA samples were normalized to yield 50–250 ng input and diluted in 55 µL on the Biomek FXP Laboratory Automation Workstation (Beckman Coulter) prior to shearing on the Covaris instrument (48). Sequencing libraries were prepared using the KAPA HTP protocol (KapaBiosystems, Wilmington, MA) and the Biomek FX system (Beckman Coulter, Brea, CA) through several enzymatic steps, including end repair, A-base addition, ligation of Illumina sequence adaptors, followed by PCR amplification and clean-up. Tumors and matched normal were combined in pools of 24–36 libraries for multiplexed captures using custom-designed biotinylated probes (Nimblegen). Captured DNA fragments were sequenced on an Illumina HiSeq2500 as paired-end 100-base pair reads. Reads were de-multiplexed using BCL2FASTQ version 1.8.3 (Illumina) and aligned to human reference genome b37 using the Burrows-Wheeler Aligner (50) (<http://bio-bwa.sourceforge.net/bwa.shtml>).

For both cohorts, low quality reads and duplicates were filtered and eliminated using Picard. We focused our mutational and copy number variation (CNV) analyses on the six genes most commonly altered (reported in at least 5% or more in a tumor type) within the mSWI/SNF complex: *PBRM1*, *ARID2*, *ARID1A*, *ARID1B*, *SMARCA4*, or *SMARCB1* (20). Tumor mutation burden (TMB) was defined as the number of exonic, non-synonymous base substitutions and indel mutations per megabase of genome examined.

Variant assessment

For the DFCI cohort, as the OncoPanel analysis was performed on tumor samples only without germline analysis, we excluded variants that were observed at a frequency 0.1% in the Exome Aggregation Consortium (ExAC) database (51), as they were considered likely germline variants (52). For the MSKCC cohort, germline variants were eliminated through the use of patient-matched blood DNA (48). Single-nucleotide variants (SNVs) and small insertions/deletions (indels) were analyzed using MuTect v.1.0.27200 (<https://confluence.broadinstitute.org/display/CGATools/MuTect>; accessed May 2013) and annotated using Oncotator (<http://www.broadinstitute.org/oncotator>; accessed May 2013). Loss-of-function variants were defined as nonsense mutations, frameshift insertions or deletions, splice-site variants affecting consensus nucleotides, or homozygous deletions. Tumors with missense mutations were excluded from the analysis as we could not confidently assess functional outcomes of these mutations, consistent with prior studies of NGS (42,53,54). CNVs were identified using a custom R-based tool (VisCap-Cancer)(28) that compares read-depth at all genomic regions assayed among different samples. For both cohorts, we focused on homozygous deletion CNVs in this analysis, and we excluded

heterozygous deletions, as the latter are associated with background noise in the Oncopanel and MSK-IMPACT sequencing platforms (45,55).

Statistical analysis

The clinical outcomes included overall survival (OS) for both cohorts. For the DFCI cohort, more clinical granularity in outcomes was obtained, including time to treatment failure (TTF), overall response rate (ORR), and disease control rate (DCR). OS was calculated from the date of ICI initiation to the date of death. Alive patients were censored at the date of last follow-up. TTF was calculated from the start date of ICI therapy to the start date of the next treatment or death. Patients alive and not started on next line were censored at the date of last follow up. Response was investigator-assessed. ORR was defined as complete response (CR) or partial response (PR). DCR was defined as CR, PR, or stable disease for more than 8 weeks.

The association between GAs and other patient and disease characteristics was evaluated using the Fisher's exact test for categorical variables and Kruskal-Wallis test for continuous variables. The distributions of OS and TTF were estimated with the Kaplan-Meier method along with 95% confidence intervals (95% CI), and their associations with genomic alterations were examined with the Wald chi-square test from the Cox regression, adjusted for lines of therapy prior to ICI for the DFCI cohort and type of ICI (Single vs. Combination) for the MSKCC cohort. For cohorts with significant associations, we further adjusted for TMB as a continuous variable when possible. The effects of genomic alterations on ORR and DCR were presented as odds ratios (ORs) estimated from logistic regression models, adjusted for prior lines of therapy and TMB in the DFCI cohort. All comparisons were conducted separately for each tumor histology. No multiple comparison adjustment was made given the exploratory nature. Statistical analyses were performed using SAS version 9.4 (SAS Institute, Cary, NC). Two-sided p-values are reported.

Results

Study population and patient characteristics

Of 17,046 patients with tumor sequencing at Dana-Farber Cancer Institute, we identified 676 patients who received at least one dose of ICI in the metastatic setting between June 2013 and January 2019 and met the eligibility criteria for this study (Methods, Supplementary Figure S1, Supplementary Table S3). We excluded two tumor types (SCLC and CUP) from the analysis because of the small sample size (N=2 and N=6, respectively). From the MSKCC cohort, we identified 848 patients who fit the inclusion criteria of this study across the seven included tumor types (Supplementary Table S4).

Median follow-up was 19.6 months (95% CI: 17.6 – 22.0) and 28.0 months (95% CI: 25.0–29.0) for the DFCI and MSKCC cohorts, respectively (Supplementary Table S5). The most prevalent tumors in the two cohorts, respectively, were NSCLC (DFCI: N=334, 49.4%; MSKCC: N=255, 30.1%), MEL (DFCI: N=86, 12.7%; MSKCC: N=192, 22.6%), and RCC (DFCI: N=68, 10.1%; MSKCC: N=118, 13.9%). Median age at first dose of ICI was 64 (range: 21–90) years for the DFCI cohort. The most common age group in the MSKCC

cohort was 61–70 years (N=269; 31.7%). The majority of the cohort received anti-PD-1- or anti-PD-L1-based therapy (DFCI: N=589, 87.1%; MSKCC: N=636, 75.0%), most commonly as single agents (DFCI: N=464, 68.6%; MSKCC: N=696, 82.1%)(Table 1, Supplementary Tables S3–S4).

Spectrum of genomic alterations within the mSWI/SNF complex

The most commonly altered genes in the DFCI and MSKCC cohorts (pan-cancer), respectively, were *ARID1A* (DFCI: N=89, 13%; MSKCC: N=77, 9%) and *PBRM1* (DFCI: N=70, 10%; MSKCC: N=59, 7%)(Figure 1A, Supplementary Tables S6–S10). Among the different tumor subtypes, RCC tumors harbored the highest rate of any LOF mSWI/SNF (DFCI: N=38, 56%; MSKCC: N=48, 41%). BAF complex, defined as *ARID1A* or *ARID1B*, LOF variants were more prevalent in CRC (DFCI: N=16, 46%; MSKCC: N=13, 21%) and UC (DFCI: N=22, 39%; MSKCC: N=23, 25%), whereas PBAF complex, defined as *PBRM1* or *ARID2*, LOF variants were most abundant in RCC (DFCI: N=33, 49%; MSKCC: N=41, 35%) (Figures 1B–C, Supplementary Tables S6–S10). *PBRM1* was the predominantly altered PBAF complex gene in RCC (N=33/33 and N=39/41 in the DFCI and MSKCC cohorts respectively). Among the seven histologies examined, all tumors, with the exception of HNSCC and EGC, harboring LOF variants within the mSWI/SNF complex had significantly higher TMB compared to wild-type (WT) in both cohorts ($p<0.05$; Supplementary Figure S2, Supplementary Table S11).

Treatment outcomes of patients with LOF GAs in mSWI/SNF genes

Multivariable analysis of the DFCI cohort showed significantly improved OS and DCR in CRC and RCC patients whose tumors carried LOF variants in the mSWI/SNF complex. Conversely, patients with NSCLC showed worse OS in the LOF group (Figures 2–3, Supplementary Figure S3, Table 2). OS and TTF outcomes for the overall DFCI cohort across histologies are summarized in Supplementary Table S5.

To explore whether the observed associations were driven by individual members of the mSWI/SNF complex, we analyzed clinical outcomes associated with GAs in the PBAF and BAF complexes separately (Supplementary Figures S4–S5, Supplementary Tables S12–S13). In the CRC DFCI cohort, the adjusted OS hazard ratio (HR) was 0.30 (0.10–0.89), favoring LOF GAs in mSWI/SNF genes ($p=0.03$). In terms of response, significantly higher ORR and DCR were associated with the presence of a LOF variant in any of the mSWI/SNF genes (ORR: LOF=6/18, 33% versus WT=0/17, 0%; DCR: LOF=11/18, 61% versus WT=4/17, 24%; $p=0.019$ and 0.041 , respectively). We observed similar trends for OS, TTF, ORR, and DCR for both PBAF and BAF complexes separately (Supplementary Figures S4–S6, Supplementary Tables S12–S13). TMB was significantly higher in the mSWI/SNF LOF cohort compared to the WT cohort, but the small sample size of the CRC cohort precluded TMB from being included into the multivariable analysis. A positive correlation was observed between MSI-status and mutational status of the mSWI/SNF genes in the CRC cohort. Of 18 patients with LOF mSWI/SNF, 16 (88.9%) had MSI-high tumors versus only 1 of 15 (6.7%) patients within the WT group (Supplementary Table S14).

Among RCC patients in the DFCI cohort, the mutant cohort showed significantly improved TTF and OS, with adjusted HRs of 0.49 (0.27–0.88) and 0.33 (0.16–0.7), respectively, compared to the WT cohort (Figure 2). Median TTF for the mutant cohort was 11.3 (6.9–31.6) months, and the OS was not reached (22 months–NR) compared to median TTF of 5.6 (2.6–9.2) months and median OS of 10.9 (8–21.4) months for the WT cohort (Figures 3A–B). A significantly higher DCR was observed for patients with LOF variants in the RCC cohort (LOF=27/38, 71% versus WT=12/30, 40%, $p=0.014$) compared to patients with WT variants (Table 2). After further adjusting for TMB, the OS benefit was maintained [adjusted HR 0.41 (0.18–0.91), $p=0.029$], and the adjusted HR for TTF was 0.70 (0.37–1.33, $p=0.278$). The adjusted ORs for DCR and ORR was 2.86 (0.91–8.99) and 1.81 (0.49–6.64), respectively ($p=0.073$, 0.371), after adjusting for TMB and prior lines of therapy.

To further explore these associations, we analyzed *PBRM1* LOF variants and clinical outcomes in RCC, given that *PBRM1* is the most frequently altered mSWI/SNF gene in RCC (33/38)(Figure 3C–D, Supplementary Table S10). Median TTF for *PBRM1* mutant and WT cohorts were 11.5 (6.9–NR) months and 5.6 (2.9–8.4), respectively. Median OS for *PBRM1* mutant was not reached (22–NR) compared to a median OS of 13.1 (8.3–27.5) months for the WT. We observed similar associations in terms of TTF, OS, and DCR for the mutant cohort compared to the WT cohort, as well after correcting for TMB as a continuous variable (Supplementary Table S15). We then compared OS and ORR in *PBRM1* LOF vs. WT across tertiles of TMB (Figure 3G). It was consistent that *PBRM1* LOF was associated with better clinical outcomes compared to WT. However, meaningful statistical comparisons could not be performed given the small sample size in each group. We obtained mirroring results when the analysis was restricted to metastatic RCC patients treated with single-agent ICIs at DFCI (N=28), supporting the notion that *PBRM1* LOF results in improved outcomes, but multivariate analysis could not be performed given the small cohort size (Supplementary Table S16). In contrast, patients in the NSCLC DFCI cohort with LOF variants within the mSWI/SNF showed significantly worse OS compared to WT adjusted HR =1.44 (1.06–1.96), with no significant difference in TTF [adjusted HR of 1.21 (0.92–1.59)] or ORR. After adjusting further for TMB, mSWI/SNF LOF tumor variants portended worse OS and TTF in the NSCLC cohort [adjusted HRs of 1.57 (1.13–2.17) and 1.38 (1.03–1.85), respectively] (Supplementary Table S17). Patients in the DFCI MEL, HNSCC, EGC, and UC cohorts did not show any association between mutational status of the mSWI/SNF complex or the individual subcomplexes PBAF and BAF, and either OS, TTF, or ORR (Figure 3, Supplementary Figures S3–S5).

In the MSKCC cohort (N=848 patients), we did not detect any significant associations across all tumor subtypes between LOF variants in the mSWI/SNF genes explored and OS after adjusting for the type of ICI administered (Figure 3, Supplementary Figure S7). For this analysis, we could not adjust for other clinical variables, such as lines of therapy prior to ICIs because this data was not available. Similarly, TTF and response data were not available in the MSKCC cohort.

Discussion

In this work, we evaluated the effect of genomic alterations of mSWI/SNF complex genes in patients treated with ICIs. Analysis of 676 and 848 solid tumors from patients treated with ICIs at DFCI and MSKCC, respectively, did not support the notion that loss of the mSWI/SNF complex is a pan-cancer biomarker of clinical benefit from ICIs.

Findings from the CRC cohort at DFCI highlight an association between LOF mSWI/SNF complex genes and MSI status, where 88.9% of patients with LOF mSWI/SNF had MSI-high tumors by immunohistochemistry. This is in line with previous literature correlating loss of *ARID1A* with mismatch-repair deficiency in endometrial, ovarian, and colorectal carcinomas (29,56,57). However, this creates uncertainty on whether the observed benefit is driven by the MSI status or by loss of the mSWI/SNF complex genes because it is well-established in the literature that mismatch-repair deficiency increases sensitivity to immune checkpoint blockers (58,59). An OS benefit in mSWI/SNF LOF variants was not observed in the MSKCC cohort, challenging the validity of this association.

In the DFCI RCC cohort, a consistent OS, TTF, and DCR benefit was observed in the mSWI/SNF LOF compared to WT at DFCI. The OS benefit was maintained after correcting for TMB, and these associations were mostly driven by *PBRM1* LOF variants. Conversely, no association was observed between mSWI/SNF LOF and OS benefit in the MSKCC cohort. This further adds to the variability in published literature. Some studies have suggested that RCC patients may benefit from ICIs when their tumors carry LOF mSWI/SNF complex variants because this creates an immune-responsive milieu, with increased expression of immune-stimulatory gene sets related to IL6-JAK-STAT3 signaling and TNF α signaling via NF- κ B, increased sensitization to T cell-mediated killing, and enhanced accessibility to IFN γ -inducible genes (28,40,41). In contrast, others have reported that *PBRM1* loss reduces IFN γ -STAT1 activity and promotes resistance to immunotherapy in RCC (60). One of the possible causes of these variable results in our study may be related to the different patient populations or to the different factors taken into consideration in the two independent cohorts. We corrected for prior lines of therapy in the DFCI cohort and for type of ICI in the MSKCC cohort. Another explanation could be due to the fragility of the correlation between *PBRM1* LOF and clinical benefit to immunotherapy, which can be explored through prospective clinical trials (61). Finally, the evidence supporting *PBRM1* LOF as a prognostic biomarker of benefit from ICIs is derived from patients receiving ICIs in the post VEGF-TKI-targeted therapy setting (32, 40). Given that data on prior lines of therapy from the MSKCC cohort was not available, this may partially explain the variable results.

Although no significant associations were demonstrated between mSWI/SNF complex LOF and TTF or ORR in the NSCLC cohort, these patients showed an inferior OS and TTF compared to patients whose tumors carried the WT mSWI/SNF genes in the DFCI cohort. Consistent with published data, *SMARCA4* was the most frequently mutated member of the mSWI/SNF complex in NSCLC at a rate of 12% (22). *SMARCA4*-deficient lung adenocarcinomas are associated with poor prognosis and response to platinum-based therapies, not ICIs (62,63). This may explain the worse outcomes of these patients. No

significant association was observed between mSWI/SNF complex mutations and OS in the MSKCC cohort, which may be attributed to the different patient populations.

The question of correlating genomic alterations with clinical outcomes is crucial to address in the field of immunotherapy, given the variable response rates with immune checkpoint inhibitors and the substantial toxicity these agents may have. Although this study did not provide a pan-cancer biomarker predicting response to checkpoint inhibitors, it provides evidence that loss of function of the mSWI/SNF complex in patients receiving an ICI-based therapy may not be a key player driving the response. Despite previous promising results in individual cancer cohorts (32,37,40,41), in our study, LOF variants of the mSWI/SNF complex genes were not associated with improved clinical outcomes in solid cancer patients treated with ICIs.

Our study has several limitations, given that it is retrospective in nature. First, this is a relatively select cohort of patients from two tertiary cancer centers. Second, we used targeted sequencing panels that did not assess all 29 members of the mSWI/SNF complex. However, we did assess the most frequently mutated genes within this complex (20,21). Third, we only evaluated two genotypes (LOF and WT), while excluding missense mutations and heterozygous deletions from our analysis. Nonetheless, we deemed that this type of analysis was necessary to answer our hypothesis because we could not assess with confidence the true functional impact of these genomic alterations. Fourth, this study did not investigate the possibility that driver mutations in other genes may influence clinical benefit to ICIs. For example, alterations in *TP53*, *MYC*, *BRAF*, *EGFR*, and *HER2* have previously been shown to impact clinical outcomes in patients treated with ICIs (64–68). This could be addressed in future studies by investigating how these genes interact with the mSWI/SNF complex in neural networks and impact outcomes to ICIs (69). Finally, we were unable to determine with certainty whether variants were somatic versus germline in the DFCI cohort, but we attempted to correct for that by excluding variants that were observed at a frequency >0.1% in the Exome Aggregation Consortium (ExAC) database.

In conclusion, this work provides a step forward in understanding complex and multivariable mechanisms driving tumor response to therapy, while validating and challenging simultaneously previously reported correlations from smaller studies on the association of mSWI/SNF GAs and clinical benefit from ICIs. It also highlights the intricacy of the mSWI/SNF complex and its disease-specific function, advocating for further efforts to discern the biology of mSWI/SNF complex and its interaction with immune checkpoint blockade.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgements

We thank all of the research assistants who assisted with this study, the OncoPanel study team and the patients. This work was supported by the National Institutes of Health under award number R01CA234018 (X.S.L.) and by the Dana-Farber/Harvard Cancer Center Kidney SPORE and Program, the Kohlberg Chair at Harvard Medical School and the Trust Family, Michael Brigham, and Loker Pinard Funds for Kidney Cancer Research at DFCI (T.K.C.).

Financial support. Research reported in this publication was partly supported by the National Institutes of Health under award number **R01CA234018** awarded to XSL. TKC is supported in part by the Dana-Farber/Harvard Cancer Center Kidney SPORE and Program, the Kohlberg Chair at Harvard Medical School and the Trust Family, Michael Brigham, and Loker Pinard Funds for Kidney Cancer Research at DFCI.

Conflict of Interest Disclosure. TKC reported receiving institutional and personal funds from Analysis Group, AstraZeneca, Alexion, Bayer, Bristol Myers-Squibb/ER Squibb and sons LLC, Cerulean, Eisai, Foundation Medicine Inc., Exelixis, Ipsen, Tracoon, Genentech, Roche, Roche Products Limited, F. Hoffmann-La Roche, GlaxoSmithKline, Lilly, Merck, Novartis, Peloton, Pfizer, Prometheus Labs, Corvus, Calithera, Sanofi/Aventis, Takeda; reported receiving honoraria from the Analysis Group, AstraZeneca, Alexion, Sanofi/Aventis, Bayer, Bristol Myers-Squibb/ER Squibb and sons LLC, Cerulean, Eisai, Foundation Medicine Inc., Exelixis, Genentech, Roche, Roche Products Limited, F. Hoffmann-La Roche, GlaxoSmithKline, Heron Therapeutics, Lilly, Merck, Novartis, Peloton, Pfizer, EMD Serono, Prometheus Labs, Corvus, Ipsen, Up-to-Date, NCCN, Analysis Group, NCCN, Michael J. Hennessy (MJH) Associates, Inc (Healthcare Communications Company with several brands such as OnClive, PeerView and PER), L-path, Kidney Cancer Journal, Clinical Care Options, Platform Q, Navinata Healthcare, Harborside Press, American Society of Medical Oncology, NEJM, Lancet Oncology; having a consulting or advisory role at Analysis Group, AstraZeneca, Alexion, Sanofi/Aventis, Bayer, Bristol Myers-Squibb/ER Squibb and sons LLC, Cerulean, Eisai, Foundation Medicine Inc., Exelixis, Genentech, Heron Therapeutics, Lilly, Roche, GlaxoSmithKline, Merck, Novartis, Peloton, Pfizer, EMD Serono, Prometheus Labs, Corvus, Ipsen, Up-to-Date, NCCN. No speaker's bureau. No leadership or employment in for-profit companies. Other present or past leadership roles: Director of GU Oncology Division at Dana-Farber and past President of medical Staff at Dana-Farber, member of NCCN Kidney panel and the GU Steering Committee, past chairman of the Kidney Cancer Association Medical and Scientific Steering Committee. Patents, royalties or other intellectual properties: International Patent Application No. PCT/US2018/12209, entitled "PBRM1 Biomarkers Predictive of Anti-Immune Checkpoint Response," filed January 3, 2018, claiming priority to U.S. Provisional Patent Application No. 62/445,094, filed January 11, 2017; International Patent Application No. PCT/US2018/058430, entitled "Biomarkers of Clinical Response and Benefit to Immune Checkpoint Inhibitor Therapy," filed October 31, 2018, claiming priority to U.S. Provisional Patent Application No. 62/581,175, filed November 3, 2017. Travel, accommodations, expenses, in relation to consulting, advisory roles, or honoraria. Medical writing and editorial assistance support may have been funded by Communications companies funded by pharmaceutical companies (ClinicalThinking, Envision Pharma Group, Fishawack Group of Companies, Health Interactions, and Parexel, others). The institution (Dana-Farber Cancer Institute) may have received additional independent funding of drug companies or/and royalties potentially involved in research around the subject matter. CV provided upon request for scope of clinical practice and research. **GS** is on the advisory board for BMS, Exelixis, Bayer, Sanofi, Pfizer, Novartis, Eisai, Janssen, Amgen, Astrazeneca, Merck, Genentech, EMD Serono, Astellas/Agensys; receives research support to institution from Astrazeneca, Bayer, Amgen, Boehringer-Ingelheim, Janssen, Merck, Sanofi, Pfizer; is author for Uptodate; is on the steering committee for Astrazeneca, BMS, Astellas, Debiopharm, Bavarian Nordic; and is a speaker for Onclive; Research to Practice; Physician Education Resource (PER). **RIH** is a consultant to Merck, GSK, BMS, Genentech, Bayer, Pfizer, immunomic, nonabiotix, ISA, glenamrk. Astra Zeneca and receives research support from Pfizer, Genentech. Merck, bms, kura, astra Zeneca. Gsk. **EMV** reports serving as an advisor or consultant for Tango Therapeutics, Genome Medical, Invitae, Illumina, Foresite Capital, Dynamo; received research support from Novartis, BMS; has equity in Tango Therapeutics, Genome Medical, Syapse, Microsoft; receives travel reimbursement from Roche/Genentech and holds institutional patents filed on ERCC2 mutations and chemotherapy response, chromatin mutations and immunotherapy response, and methods for clinical interpretation. **MG** receives research funding from Bristol Myers-Squibb and Merck. **DJK** reports serving as a consultant for Novartis. **CK** is a Scientific Founder, fiduciary Board of Directors member, Scientific Advisory Board member, shareholder and consultant for Foghorn Therapeutics, Inc.

References

1. Antonia SJ, Lopez-Martin JA, Bendell J, Ott PA, Taylor M, Eder JP, et al. Nivolumab alone and nivolumab plus ipilimumab in recurrent small-cell lung cancer (CheckMate 032): a multicentre, open-label, phase 1/2 trial. *Lancet Oncol.* 2016;17(7):883–95. [PubMed: 27269741]
2. Balar AV, Galsky MD, Rosenberg JE, Powles T, Petrylak DP, Bellmunt J, et al. Atezolizumab as first-line treatment in cisplatin-ineligible patients with locally advanced and metastatic urothelial carcinoma: a single-arm, multicentre, phase 2 trial. *Lancet.* 2017;389(10064):67–76. [PubMed: 27939400]
3. El-Khoueiry AB, Sangro B, Yau T, Crocenzi TS, Kudo M, Hsu C, et al. Nivolumab in patients with advanced hepatocellular carcinoma (CheckMate 040): an open-label, non-comparative, phase 1/2 dose escalation and expansion trial. *Lancet.* 2017;389(10088):2492–502. [PubMed: 28434648]
4. Garon EB, Rizvi NA, Hui R, Leigh N, Balmanoukian AS, Eder JP, et al. Pembrolizumab for the treatment of non-small-cell lung cancer. *N Engl J Med.* 2015;372(21):2018–28. [PubMed: 25891174]

5. Hodi FS, Chiarion-Sileni V, Gonzalez R, Grob JJ, Rutkowski P, Cowey CL, et al. Nivolumab plus ipilimumab or nivolumab alone versus ipilimumab alone in advanced melanoma (CheckMate 067): 4-year outcomes of a multicentre, randomised, phase 3 trial. *Lancet Oncol.* 2018;19(11):1480–92. [PubMed: 30361170]
6. Kim ST, Cristescu R, Bass AJ, Kim KM, Odegaard JI, Kim K, et al. Comprehensive molecular characterization of clinical responses to PD-1 inhibition in metastatic gastric cancer. *Nat Med.* 2018;24(9):1449–58. [PubMed: 30013197]
7. Motzer RJ, Escudier B, McDermott DF, George S, Hammers HJ, Srinivas S, et al. Nivolumab versus Everolimus in Advanced Renal-Cell Carcinoma. *N Engl J Med.* 2015;373(19):1803–13. [PubMed: 26406148]
8. Motzer RJ, Penkov K, Haanen J, Rini B, Albiges L, Campbell MT, et al. Avelumab plus Axitinib versus Sunitinib for Advanced Renal-Cell Carcinoma. *N Engl J Med.* 2019;380(12):1103–15. [PubMed: 30779531]
9. Motzer RJ, Tannir NM, McDermott DF, Aren Frontera O, Melichar B, Choueiri TK, et al. Nivolumab plus Ipilimumab versus Sunitinib in Advanced Renal-Cell Carcinoma. *N Engl J Med.* 2018;378(14):1277–90. [PubMed: 29562145]
10. Ribas A, Wolchok JD. Cancer immunotherapy using checkpoint blockade. *Science.* 2018;359(6382):1350–5. [PubMed: 29567705]
11. Rini BI, Plimack ER, Stus V, Gafanov R, Hawkins R, Nosov D, et al. Pembrolizumab plus Axitinib versus Sunitinib for Advanced Renal-Cell Carcinoma. *N Engl J Med.* 2019;380(12):1116–27. [PubMed: 30779529]
12. Rosenberg JE, Hoffman-Censits J, Powles T, van der Heijden MS, Balar AV, Necchi A, et al. Atezolizumab in patients with locally advanced and metastatic urothelial carcinoma who have progressed following treatment with platinum-based chemotherapy: a single-arm, multicentre, phase 2 trial. *Lancet.* 2016;387(10031):1909–20. [PubMed: 26952546]
13. Topalian SL, Hodi FS, Brahmer JR, Gettinger SN, Smith DC, McDermott DF, et al. Safety, activity, and immune correlates of anti-PD-1 antibody in cancer. *N Engl J Med.* 2012;366(26):2443–54. [PubMed: 22658127]
14. Topalian SL, Hodi FS, Brahmer JR, Gettinger SN, Smith DC, McDermott DF, et al. Five-Year Survival and Correlates Among Patients With Advanced Melanoma, Renal Cell Carcinoma, or Non-Small Cell Lung Cancer Treated With Nivolumab. *JAMA Oncol.* 2019.
15. Wolchok JD, Chiarion-Sileni V, Gonzalez R, Rutkowski P, Grob JJ, Cowey CL, et al. Overall Survival with Combined Nivolumab and Ipilimumab in Advanced Melanoma. *N Engl J Med.* 2017;377(14):1345–56. [PubMed: 28889792]
16. Gibney GT, Weiner LM, Atkins MB. Predictive biomarkers for checkpoint inhibitor-based immunotherapy. *Lancet Oncol.* 2016;17(12):e542–e51. [PubMed: 27924752]
17. Keenan TE, Burke KP, Van Allen EM. Genomic correlates of response to immune checkpoint blockade. *Nat Med.* 2019;25(3):389–402. [PubMed: 30842677]
18. Valencia AM, Kadoch C. Chromatin regulatory mechanisms and therapeutic opportunities in cancer. *Nat Cell Biol.* 2019;21(2):152–61. [PubMed: 30602726]
19. Mashtalir N, D'Avino AR, Michel BC, Luo J, Pan J, Otto JE, et al. Modular Organization and Assembly of SWI/SNF Family Chromatin Remodeling Complexes. *Cell.* 2018;175(5):1272–88 e20. [PubMed: 30343899]
20. Kadoch C, Hargreaves DC, Hodges C, Elias L, Ho L, Ranish J, et al. Proteomic and bioinformatic analysis of mammalian SWI/SNF complexes identifies extensive roles in human malignancy. *Nat Genet.* 2013;45(6):592–601. [PubMed: 23644491]
21. Hodges C, Kirkland JG, Crabtree GR. The Many Roles of BAF (mSWI/SNF) and PBAF Complexes in Cancer. *Cold Spring Harb Perspect Med.* 2016;6(8).
22. Imielinski M, Berger AH, Hammerman PS, Hernandez B, Pugh TJ, Hodis E, et al. Mapping the hallmarks of lung adenocarcinoma with massively parallel sequencing. *Cell.* 2012;150(6):1107–20. [PubMed: 22980975]
23. Rizvi NA, Hellmann MD, Snyder A, Kvistborg P, Makarov V, Havel JJ, et al. Cancer immunology. Mutational landscape determines sensitivity to PD-1 blockade in non-small cell lung cancer. *Science.* 2015;348(6230):124–8. [PubMed: 25765070]

24. Jones S, Wang TL, Shih Ie M, Mao TL, Nakayama K, Roden R, et al. Frequent mutations of chromatin remodeling gene ARID1A in ovarian clear cell carcinoma. *Science*. 2010;330(6001):228–31. [PubMed: 20826764]
25. Gui Y, Guo G, Huang Y, Hu X, Tang A, Gao S, et al. Frequent mutations of chromatin remodeling genes in transitional cell carcinoma of the bladder. *Nat Genet*. 2011;43(9):875–8. [PubMed: 21822268]
26. Varela I, Tarpey P, Raine K, Huang D, Ong CK, Stephens P, et al. Exome sequencing identifies frequent mutation of the SWI/SNF complex gene PBRM1 in renal carcinoma. *Nature*. 2011;469(7331):539–42. [PubMed: 21248752]
27. Hodis E, Watson IR, Kryukov GV, Arolt ST, Imielinski M, Theurillat JP, et al. A landscape of driver mutations in melanoma. *Cell*. 2012;150(2):251–63. [PubMed: 22817889]
28. Pan D, Kobayashi A, Jiang P, Ferrari de Andrade L, Tay RE, Luoma AM, et al. A major chromatin regulator determines resistance of tumor cells to T cell-mediated killing. *Science*. 2018;359(6377):770–5. [PubMed: 29301958]
29. Shen J, Ju Z, Zhao W, Wang L, Peng Y, Ge Z, et al. ARID1A deficiency promotes mutability and potentiates therapeutic antitumor immunity unleashed by immune checkpoint blockade. *Nat Med*. 2018;24(5):556–62. [PubMed: 29736026]
30. Canadas I, Thummalapalli R, Kim JW, Kitajima S, Jenkins RW, Christensen CL, et al. Tumor innate immunity primed by specific interferon-stimulated endogenous retroviruses. *Nat Med*. 2018;24(8):1143–50. [PubMed: 30038220]
31. Boileve A, Carlo MI, Barthelemy P, Oudard S, Borchellini D, Voss MH, et al. Immune checkpoint inhibitors in MITF family translocation renal cell carcinomas and genetic correlates of exceptional responders. *J Immunother Cancer*. 2018;6(1):159. [PubMed: 30591082]
32. Braun D, Ishii Y, Walsh AM, Van Allen EM, Wu CJ, Shukla SA, et al. *JAMA Oncol*. 2019.
33. Choueiri TK, Albiges L, Haanen JBAG, Larkin JMG, Uemura M, Pal SK, et al. Biomarker analyses from JAVELIN Renal 101: Avelumab + axitinib (A+Ax) versus sunitinib (S) in advanced renal cell carcinoma (aRCC). *Journal of Clinical Oncology*. 2019;37(15_suppl):101–.
34. Dizman N, Bergerot PG, Bergerot CD, Hsu J, Pal SK. Duration of treatment (DOT) with targeted therapies (TT) or immunotherapy (IO) in PBRM1 mutated metastatic renal cell carcinoma (mRCC). *Journal of Clinical Oncology*. 2019;37(7_suppl):622–.
35. Hakimi AA, Ged Y, Flynn J, Hoen DR, Natale RGD, Blum KA, et al. The impact of PBRM1 mutations on overall survival in greater than 2,100 patients treated with immune checkpoint blockade (ICB). *Journal of Clinical Oncology*. 2019;37(7_suppl):666–.
36. Henon C, Blay JY, Massard C, Mir O, Bahleda R, Dumont S, et al. Long lasting major response to pembrolizumab in a thoracic malignant rhabdoid-like SMARCA4-deficient tumor. *Ann Oncol*. 2019.
37. Li L, Li M, Jiang Z, Wang X. ARID1A Mutations Are Associated with Increased Immune Activity in Gastrointestinal Cancer. *Cells*. 2019;8(7).
38. McDermott DF, Huseni MA, Atkins MB, Motzer RJ, Rini BI, Escudier B, et al. Publisher Correction: Clinical activity and molecular correlates of response to atezolizumab alone or in combination with bevacizumab versus sunitinib in renal cell carcinoma. *Nat Med*. 2018;24(12):1941.
39. McDermott DF, Huseni MA, Atkins MB, Motzer RJ, Rini BI, Escudier B, et al. Clinical activity and molecular correlates of response to atezolizumab alone or in combination with bevacizumab versus sunitinib in renal cell carcinoma. *Nat Med*. 2018;24(6):749–57. [PubMed: 29867230]
40. Miao D, Margolis CA, Gao W, Voss MH, Li W, Martini DJ, et al. Genomic correlates of response to immune checkpoint therapies in clear cell renal cell carcinoma. *Science*. 2018;359(6377):801–6. [PubMed: 29301960]
41. Miao D, Margolis CA, Vokes NI, Liu D, Taylor-Weiner A, Wankowicz SM, et al. Genomic correlates of response to immune checkpoint blockade in microsatellite-stable solid tumors. *Nat Genet*. 2018;50(9):1271–81. [PubMed: 30150660]
42. Braun DA, Ishii Y, Walsh AM, Van Allen EM, Wu CJ, Shukla SA, et al. Clinical Validation of PBRM1 Alterations as a Marker of Immune Checkpoint Inhibitor Response in Renal Cell Carcinoma. *JAMA Oncol*. 2019.

43. Choueiri TK, Albiges L, Haanen JBAG, Larkin JMG, Uemura M, Pal SK, et al. Biomarker analyses from JAVELIN Renal 101: Avelumab + axitinib (A+Ax) versus sunitinib (S) in advanced renal cell carcinoma (aRCC). *Journal of Clinical Oncology* 2019;37.
44. Samstein RM, Lee CH, Shoushtari AN, Hellmann MD, Shen R, Janjigian YY, et al. Tumor mutational load predicts survival after immunotherapy across multiple cancer types. *Nat Genet.* 2019;51(2):202–6. [PubMed: 30643254]
45. Garcia EP, Minkovsky A, Jia Y, Ducar MD, Shivdasani P, Gong X, et al. Validation of OncoPanel: A Targeted Next-Generation Sequencing Assay for the Detection of Somatic Variants in Cancer. *Arch Pathol Lab Med.* 2017;141(6):751–8. [PubMed: 28557599]
46. Sholl LM, Do K, Shivdasani P, Cerami E, Dubuc AM, Kuo FC, et al. Institutional implementation of clinical tumor profiling on an unselected cancer population. *JCI Insight.* 2016;1(19):e87062. [PubMed: 27882345]
47. Zehir A, Benayed R, Shah RH, Syed A, Middha S, Kim HR, et al. Erratum: Mutational landscape of metastatic cancer revealed from prospective clinical sequencing of 10,000 patients. *Nat Med.* 2017;23(8):1004.
48. Zehir A, Benayed R, Shah RH, Syed A, Middha S, Kim HR, et al. Mutational landscape of metastatic cancer revealed from prospective clinical sequencing of 10,000 patients. *Nat Med.* 2017;23(6):703–13. [PubMed: 28481359]
49. Cheng DT, Mitchell TN, Zehir A, Shah RH, Benayed R, Syed A, et al. Memorial Sloan Kettering-Integrated Mutation Profiling of Actionable Cancer Targets (MSK-IMPACT): A Hybridization Capture-Based Next-Generation Sequencing Clinical Assay for Solid Tumor Molecular Oncology. *J Mol Diagn.* 2015;17(3):251–64. [PubMed: 25801821]
50. Li H, Durbin R. Fast and accurate long-read alignment with Burrows-Wheeler transform. *Bioinformatics.* 2010;26(5):589–95. [PubMed: 20080505]
51. Lek M, Karczewski KJ, Minikel EV, Samocha KE, Banks E, Fennell T, et al. Analysis of protein-coding genetic variation in 60,706 humans. *Nature.* 2016;536(7616):285–91. [PubMed: 27535533]
52. Consortium APG. AACR Project GENIE: Powering Precision Medicine through an International Consortium. *Cancer Discov.* 2017;7(8):818–31. [PubMed: 28572459]
53. Itan Y, Casanova JL. Can the impact of human genetic variations be predicted? *Proc Natl Acad Sci U S A.* 2015;112(37):11426–7. [PubMed: 26351682]
54. Miosge LA, Field MA, Sontani Y, Cho V, Johnson S, Palkova A, et al. Comparison of predicted and actual consequences of missense mutations. *Proc Natl Acad Sci U S A.* 2015;112(37):E5189–98. [PubMed: 26269570]
55. Pugh TJ, Amr SS, Bowser MJ, Gowrisankar S, Hynes E, Mahanta LM, et al. VisCap: inference and visualization of germ-line copy-number variants from targeted clinical sequencing data. *Genet Med.* 2016;18(7):712–9. [PubMed: 26681316]
56. Allo G, Bernardini MQ, Wu RC, Shih Ie M, Kalloger S, Pollett A, et al. ARID1A loss correlates with mismatch repair deficiency and intact p53 expression in high-grade endometrial carcinomas. *Mod Pathol.* 2014;27(2):255–61. [PubMed: 23887303]
57. Chou A, Toon CW, Clarkson A, Sioson L, Houang M, Watson N, et al. Loss of ARID1A expression in colorectal carcinoma is strongly associated with mismatch repair deficiency. *Hum Pathol.* 2014;45(8):1697–703. [PubMed: 24925223]
58. Le DT, Durham JN, Smith KN, Wang H, Bartlett BR, Aulakh LK, et al. Mismatch repair deficiency predicts response of solid tumors to PD-1 blockade. *Science.* 2017;357(6349):409–13. [PubMed: 28596308]
59. Le DT, Uram JN, Wang H, Bartlett BR, Kemberling H, Eyring AD, et al. PD-1 Blockade in Tumors with Mismatch-Repair Deficiency. *N Engl J Med.* 2015;372(26):2509–20. [PubMed: 26028255]
60. Liu X, Kong W, Peterson C, Hoang A, Zhang X, Zhou L, et al. PBRM1 loss reduces IFN γ -STAT1 activity and promotes resistance to immunotherapy and antiangiogenic therapy in renal cell carcinoma. *J Cancer Research.* 2019;79:945–.
61. Tignanelli CJ, Napolitano LM. The Fragility Index in Randomized Clinical Trials as a Means of Optimizing Patient Care. *JAMA Surg.* 2019;154(1):74–9. [PubMed: 30422256]

62. Matsubara N, Maemondo M, Inoue A, Ishimoto O, Watanabe K, Sakakibara T, et al. Phase II study of irinotecan as a third- or fourth-line treatment for advanced non-small cell lung cancer: NJLCG0703. *Respir Investig*. 2013;51(1):28–34.
63. Bell EH, Chakraborty AR, Mo X, Liu Z, Shilo K, Kirste S, et al. SMARCA4/BRG1 Is a Novel Prognostic Biomarker Predictive of Cisplatin-Based Chemotherapy Outcomes in Resected Non-Small Cell Lung Cancer. *Clin Cancer Res*. 2016;22(10):2396–404. [PubMed: 26671993]
64. Assoun S, Theou-Anton N, Nguenang M, Cazes A, Danel C, Abbar B, et al. Association of TP53 mutations with response and longer survival under immune checkpoint inhibitors in advanced non-small-cell lung cancer. *Lung Cancer*. 2019;132:65–71. [PubMed: 31097096]
65. Dudnik E, Peled N, Nechushtan H, Wollner M, Onn A, Agbarya A, et al. BRAF Mutant Lung Cancer: Programmed Death Ligand 1 Expression, Tumor Mutational Burden, Microsatellite Instability Status, and Response to Immune Check-Point Inhibitors. *J Thorac Oncol*. 2018;13(8):1128–37. [PubMed: 29723688]
66. Griguolo G, Pascual T, Dieci MV, Guarneri V, Prat A. Interaction of host immunity with HER2-targeted treatment and tumor heterogeneity in HER2-positive breast cancer. *J Immunother Cancer*. 2019;7(1):90. [PubMed: 30922362]
67. Hastings K, Yu HA, Wei W, Sanchez-Vega F, DeVeaux M, Choi J, et al. EGFR mutation subtypes and response to immune checkpoint blockade treatment in non-small-cell lung cancer. *Ann Oncol*. 2019;30(8):1311–20. [PubMed: 31086949]
68. Kim EY, Kim A, Kim SK, Chang YS. MYC expression correlates with PD-L1 expression in non-small cell lung cancer. *Lung Cancer*. 2017;110:63–7. [PubMed: 28676221]
69. Gunther F, Wawro N, Bammann K. Neural networks for modeling gene-gene interactions in association studies. *BMC Genet*. 2009;10:87. [PubMed: 20030838]

Figure 1. Spectrum of genomic alterations in the mSWI/SNF complex across tumor histologies.

A. Frequency and variant types detected across the 6 mSWI/SNF complex genes in the DFCI (N=676) and MSKCC (N=848) cohorts. Number of patients per each gene is indicated. **B.** Frequency of detected mSWI/SNF subcomplex mutations across tumor histologies in the DFCI cohort. Number of patients for each tumor type indicated. **C.** Frequency of detected mSWI/SNF subcomplex mutations across tumor histologies in the MSKCC cohort. Number of patients for each tumor type indicated. GAs: genomic alterations.

Figure 2. Analysis of survival outcomes in patients with loss of function (LOF) mSWI/SNF and wild-type (WT) across different tumor histologies.

A. Adjusted hazard ratios (HR) for time to treatment failure in the DFCI cohort (N=676). **B.** Adjusted HRs for overall survival in the DFCI cohort. **C.** Adjusted HRs for overall survival in the MSKCC cohort (N=848). Chi-square Wald statistic. CI: confidence interval. Cohort adjustments indicated. Error bars represent SD.

Figure 3: Clinical outcomes in the renal cell carcinoma (RCC) cohort.

A. Time to treatment failure (TTF) in patients with loss of function (LOF) mSWI/SNF complex and WT genes in the DFCI cohort. **B.** Overall survival (OS) in in patients with LOF mSWI/SNF complex and WT genes in the DFCI cohort. **C.** TTF in patients with LOF and WT *PBRM1* in the DFCI cohort. **D.** OS in in patients with LOF and WT *PBRM1* in the DFCI cohort. **E.** OS in patients with LOF mSWI/SNF complex and WT genes in the MSKCC cohort. **F.** OS in patients with LOF and WT *PBRM1* in the MSKCC cohort. **G.** OS and overall response rate (ORR) across tumor mutational burden (TMB) tertiles in *PBRM1* LOF vs. WT genes in the DFCI cohort. Md.: median; NR: not reached; mos: months. (A-F) Number of patients indicated below each graph. OS and TTF estimated with the Kaplan-Meier method along with 95% CI. Chi-square Wald statistic. P-value threshold for significance was 0.05.

Table 1:

Baseline Clinical characteristics of the overall population

	DFCI Cohort		MSKCC Cohort	
	N (Median)	% (Range)	N (Median)	% (Range)
Age at ICI	64	21–90	NA	NA
Tumor type				
Colorectal adenocarcinoma	35	5.2	63	7.4
Esophagogastric Carcinoma	66	9.8	59	7.0
Head and neck Squamous Cell Carcinoma	31	4.6	68	8.0
Melanoma	86	12.7	192	22.6
Non-Small Cell Lung Carcinoma	334	49.4	255	30.1
Renal Cell Carcinoma	68	10.1	118	13.9
Urothelial Carcinoma	56	8.3	93	11.0
Site of Specimen Sequenced				
Metastatic	347	51.3	NA	NA
Primary	329	48.7	NA	NA
ICI type				
Combination	212	31.4	152	17.9
Single	464	68.6	696	82.1
ICI Class				
Anti PD-1/PD-L1	589	87.1	636	75.0
Anti CTLA-4	12	1.8	60	7.1
Anti PD-1/PD-L1 + Anti CTLA-4	75	11.1	152	17.9
Number of prior lines				
0	329	48.7	NA	NA
1	220	32.5	NA	NA
2	127	18.7	NA	NA

Table 2:

Antitumor Activity in the loss of function (LOF) and wild-type (WT) mSWI/SNF cohorts

A: Overall response rate (ORR) in the LOF and WT mSWI/SNF cohorts

	Wild Type			LOF mSWI/SNF			p-value
	Total	N	ORR (%)	Total	N	ORR (%)	
Colorectal adenocarcinoma *	17	0	0%	18	6	33.0%	0.019
Esophagogastric Carcinoma	53	9	17%	13	4	31.0%	0.267
Head and neck Squamous Cell Carcinoma	27	3	11%	4	2	50.0%	0.112
Melanoma	47	17	36%	39	13	33.0%	0.824
Non-Small Cell Lung Carcinoma	230	60	26%	104	22	21.0%	0.41
Renal Cell Carcinoma	30	5	17%	38	12	32.0%	0.259
Urothelial Carcinoma	27	6	22%	29	7	24.0%	0.99

B: Disease control rate (DCR) in the LOF and WT mSWI/SNF cohorts

	Wild Type			LOF mSWI/SNF			p-value
	Total	N	DCR (%)	Total	N	DCR (%)	
Colorectal adenocarcinoma *	17	4	24%	18	11	61%	0.0409
Esophagogastric	53	16	30%	13	7	54%	0.1921
Head and neck	27	6	22%	4	2	50%	0.2683
Melanoma	47	26	55%	39	24	62%	0.6619
Non-small cell lung carcinoma	230	126	55%	104	52	50%	0.4775
Renal cell carcinoma *	30	12	40%	38	27	71%	0.0139
Urothelial carcinoma	27	10	37%	29	13	45%	0.5965

* p-value<0.05