The FABRIC Cancer Portal: A Ranked Catalogue of Gene Selection in Tumors Over the Human Coding Genome

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ABSTRACT

Contemporary catalogues of cancer driver genes rely primarily on high mutation rates as evidence for gene selection in tumors. Here, we present The Functional Alteration Bias Recovery In Coding-regions Cancer Portal, a comprehensive catalogue of gene selection in cancer based purely on the biochemical functional effects of mutations at the protein level. Gene selection in the portal is quantified by combining genomics data with rich proteomic annotations. Genes are ranked according to the strength of evidence for selection in tumor, based on rigorous and robust statistics. The portal covers the entire human coding genome (∼18,000 protein-coding genes) across 33 cancer types and pan-cancer. It includes a selected set of cross-references to the most relevant resources providing genomics, proteomics, and cancer-related information. We showcase the portal with known and overlooked cancer genes, demonstrating the utility of the portal via its simple visual interface, which allows users to pivot between gene-centric and cancer type views. The portal is available at fabric-cancer.huji.ac.il.

Significance: A new cancer portal quantifies and presents gene selection in tumor over the entire human coding genome across 33 cancer types and pan-cancer.

Introduction

Cancer gene catalogues, annotating accumulated knowledge and evidence about the roles of genes in tumors, are an indispensable tool in cancer research and precision therapy (1, 2). One of the main goals of such catalogues is to highlight potential cancer drivers (3, 4). However, lists of candidate cancer drivers are typically limited to a few hundreds of high-confidence genes, without any information about the rest of the coding genome [e.g., 719 genes in Census (V86); ref. 3 and 299 genes in ref. 4). Moreover, gene annotations in driver catalogues are typically binary, marking whether each gene is a potential cancer driver or not, without expressing ranking, effect size, or confidence level for candidate genes. Finally, as the primary evidence used to implicate genes in cancer within existing catalogues is mutation abundance, they fail to capture low-recurrence driver genes.

Materials and Methods

The Functional Alteration Bias Recovery In Coding-regions framework

We recently developed Functional Alteration Bias Recovery In Coding-regions (FABRIC), a new method for detecting genes involved in cancer (**Fig. 1A** and **B**; ref. 5). Unlike other computational methods, which mostly consider the recurrence of mutations, FABRIC extracts signal from the molecular functional effects of mutations altering protein-coding genes. In other words, instead of looking for genes with high rates of mutations, FABRIC detects genes with mutations that are more damaging than would be expected at random, regardless of their number. The framework is, therefore, complementary to most other methods in finding genes under positive selection. We used FABRIC to assess and quantify the selection of the entire human proteome (∼18,000 protein-coding genes) across 33 cancer types and pan-cancer, by analyzing approximately 3×10^6 somatic mutations in approximately 10,000 patients with cancer from The Cancer Genome Atlas (TCGA) cohort (6). A full description of that analysis is described in our original work (5). FABRIC's summary statistics across the entire human coding genome with respect to the 33 cancer types and pan-cancer provide a wealth of information on the potential roles of human genes in tumors.

Altogether, FABRIC recovers 593 protein-coding genes exhibiting significant positive selection in pan-cancer, and only six genes showing signs of negative selection (**Fig. 2**). In many cases, FABRIC recovers well-known cancer driver genes, such as *TP53, APC*, and *KRAS*. However, it does not always converge with other methods for detecting caner drivers, especially those based on mutation rates. Unlike most methods, FABRIC considers the functional effects of mutations rather than their number. For example, the *FAT4* gene is a tumor suppressor gene playing a role in numerous cancers (7). Despite its high mutation rate (with 1,893 single-nucleotide somatic mutations in the coding region of the gene across the 33 TCGA cancer types), it is not significant by FABRIC (FDR q value = 0.11). On the other hand, FABRIC is able to recover hundreds of overlooked genes (e.g., *MICU3*, FDR q value = 4E-7; ref. 5).

The FABRIC Cancer Portal

Here, we present The FABRIC Cancer Portal, available at fabric-cancer.huji.ac.il. The goal of the new portal is to make the rich cancer results of FABRIC widely available via a simple web resource that enables quantitative and comparative analyses. In addition to easy

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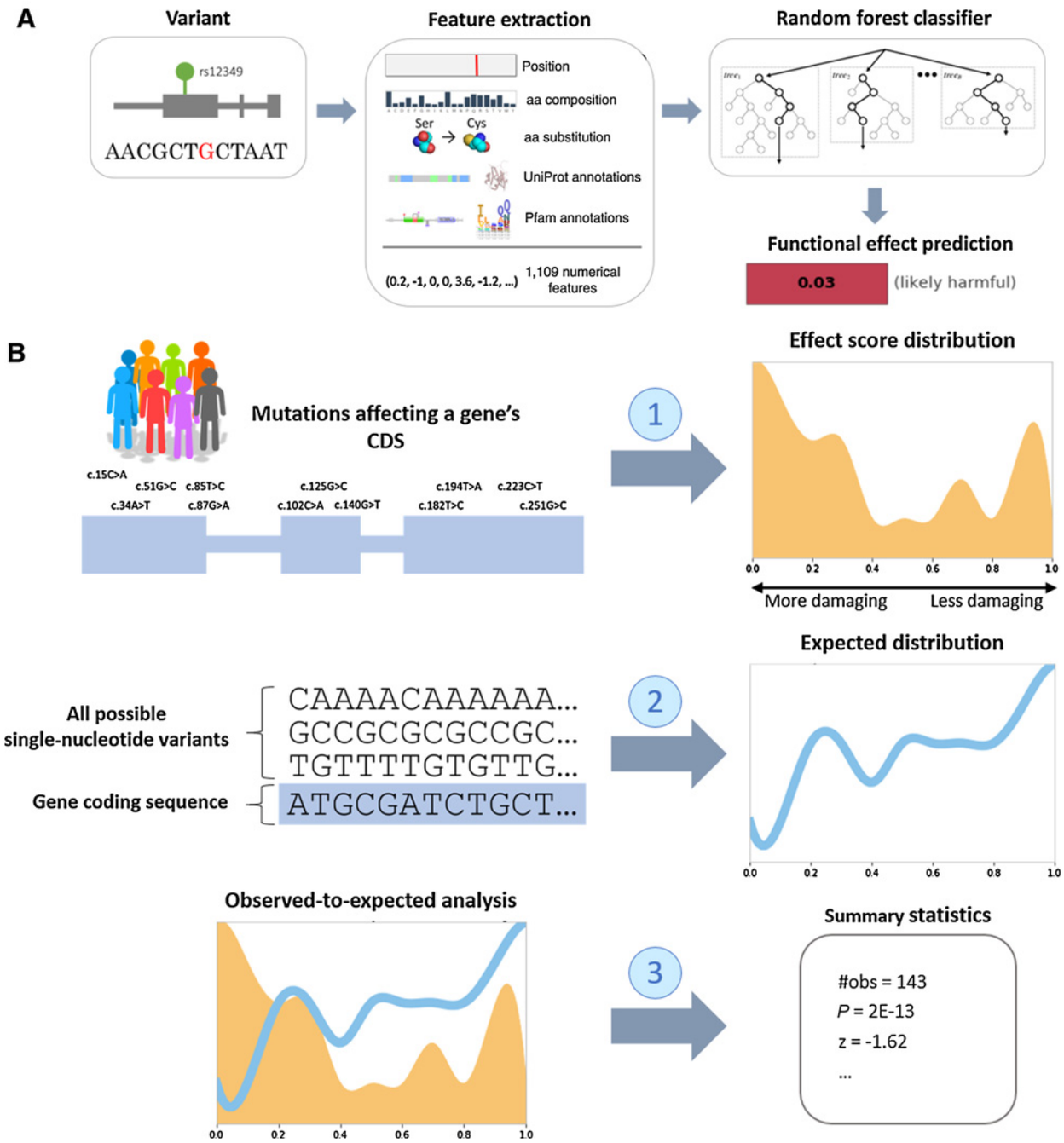


Figure 1.

The FABRIC framework. FABRIC is a framework for detecting and quantifying gene selection in coding regions. In the context of cancer, it can analyze somatic mutations to identify protein-coding genes under positive selection, which are suspected driver genes. **A,** FABRIC uses an underlying machine-learning model (called FIRM) to estimate the functional damage of genetic variants in coding regions. FIRM extracts a large set of proteomics features from a variety of sources (including UniProt and Pfam annotations). On the basis of these features, it assigns each mutation an effect score, reflecting the probability of the protein to retain its function given the mutation. **B,** The FABRIC framework consists of three steps. To assess the selection of a gene, FABRIC first collects all single-nucleotide mutations within its coding region (from a variety of samples) and uses FIRM to assign them functional effect scores. Second, FIRM is also used to record the effect scores of all possible single-nucleotide variants within the same gene, to construct a background model for the distribution of effect scores that would be expected at random. FABRIC's background model takes into account the number of observed mutations and their nucleotide substitution distribution. In the third and final step, the effect score distribution of the observed mutations is compared against the distribution of the expected effect scores to obtain the summary statistics for the analyzed gene. These summary statistics describe the significance (*P* value) and strength (*z* value) of the gene's selection, namely to what extent the observed mutations are more (or less) damaging than would be expected by the same number of random mutations.

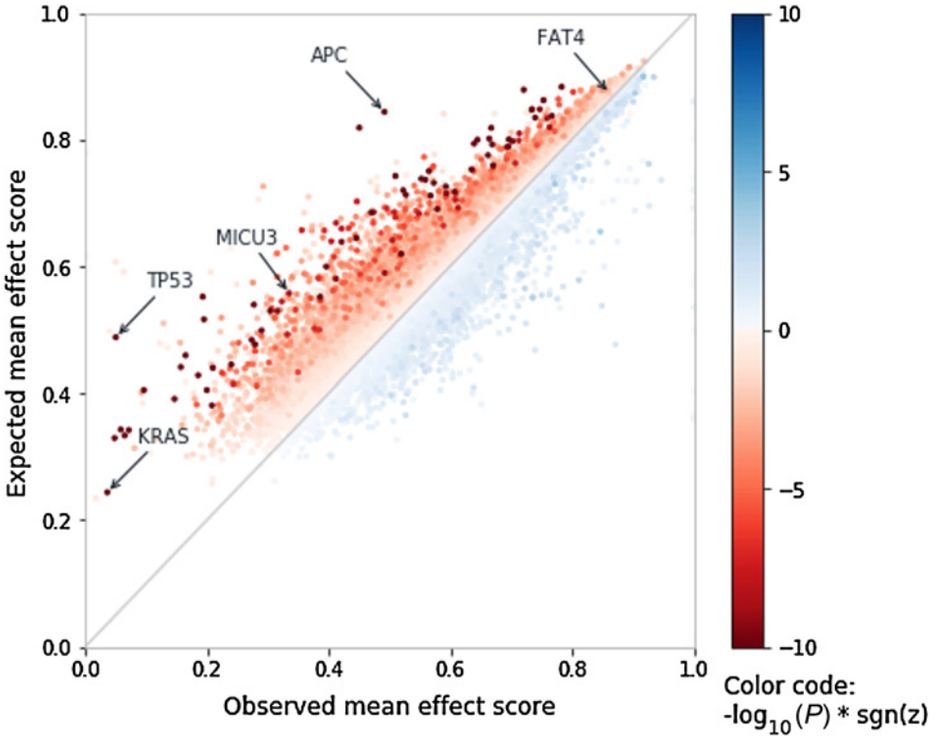


Figure 2.
FABRIC summary statistics in pan-cancer across the human coding genome. The observed and expected mean effect scores assigned by FABRIC to each of the approximately 18,000 analyzed protein-coding genes in pan-cancer. The *x*-axis marks the average effect score across all somatic mutations observed in a gene, with lower scores indicating more damaging mutations. The *y*-axis marks the mean effect score that would be expected for the same number of mutations, as calculated by FABRIC's background model. Genes are colored by their significance (log-scaled *P* values). Genes under positive selection (i.e., affected by mutations more damaging than would be expected at random, indicated by negative *z* values) are colored red, while in blue are genes under negative selection (i.e., with mutations less damaging than would be expected at random, indicated by positive *z* values).

access to downloadable data, the portal also provides quick cross-references to other genomics, proteomics, and cancer resources (**Table 1**).

Accessibility

The FABRIC Cancer Portal is available at fabric-cancer.huji.ac.il.

To learn more about its applicability, we invite the reader to browse through the tutorial (fabric-cancer.huji.ac.il/#tutorial-head) and FAQ (fabric-cancer.huji.ac.il/g/FAQ) sections of the portal.

Results

To illustrate the usability of The FABRIC Cancer Portal as a navigation and discovery tool, we present a showcase starting with adenomatosis polyposis coli (*APC*), a known cancer driver gene (**Fig. 3A**). *APC* is a classic tumor suppressor gene playing a central role in cell cycle and migration through the Wnt signaling pathway. The role of *APC* in cell division and differentiation is mediated by its direct binding to β-catenin (8).

Table 1. Cross-references in The FABRIC Cancer Portal.

Resource	URL	Purpose
GDC Data Portal (3)	https://portal.gdc.cancer.gov/	Allows access to TCGA dataset of somatic mutations analyzed by FABRIC.
Gene Cards (13)	https://www.genecards.org/	A comprehensive knowledge base of genes integrating more than 150 web sources.
UniProt (14)	https://www.uniprot.org/	Unified records of richly annotated proteins, which are the underlying entities analyzed by FABRIC.
Pfam (15)	http://pfam.xfam.org/	Presents the protein domain architecture of genes.
UCSC Genome Browser (16)	https://genome.ucsc.edu/cgi-bin/hgTracks	Displays the genomic context of genes, including rich functional annotations.
RefSeq (17)	https://www.ncbi.nlm.nih.gov/refseq/	RNA transcripts compatible with the primary isoforms of the proteins analyzed by FABRIC.
COSMIC Census (3)	https://cancer.sanger.ac.uk/census	A catalogue of genes causally implicated in cancer.
cBioPortal (2)	https://www.cbioportal.org/	Provides additional molecular profiles and clinical attributes of genes from large-scale cancer genomics.
DepMap Portal (18)	https://depmap.org/portal/	Defines gene targets for small-molecule therapeutics based on hundreds of manipulated cancer cell lines.
canSAR Black (19)	https://cansarblack.icr.ac.uk/	Integrates biological and clinical data for cancer drug discovery.

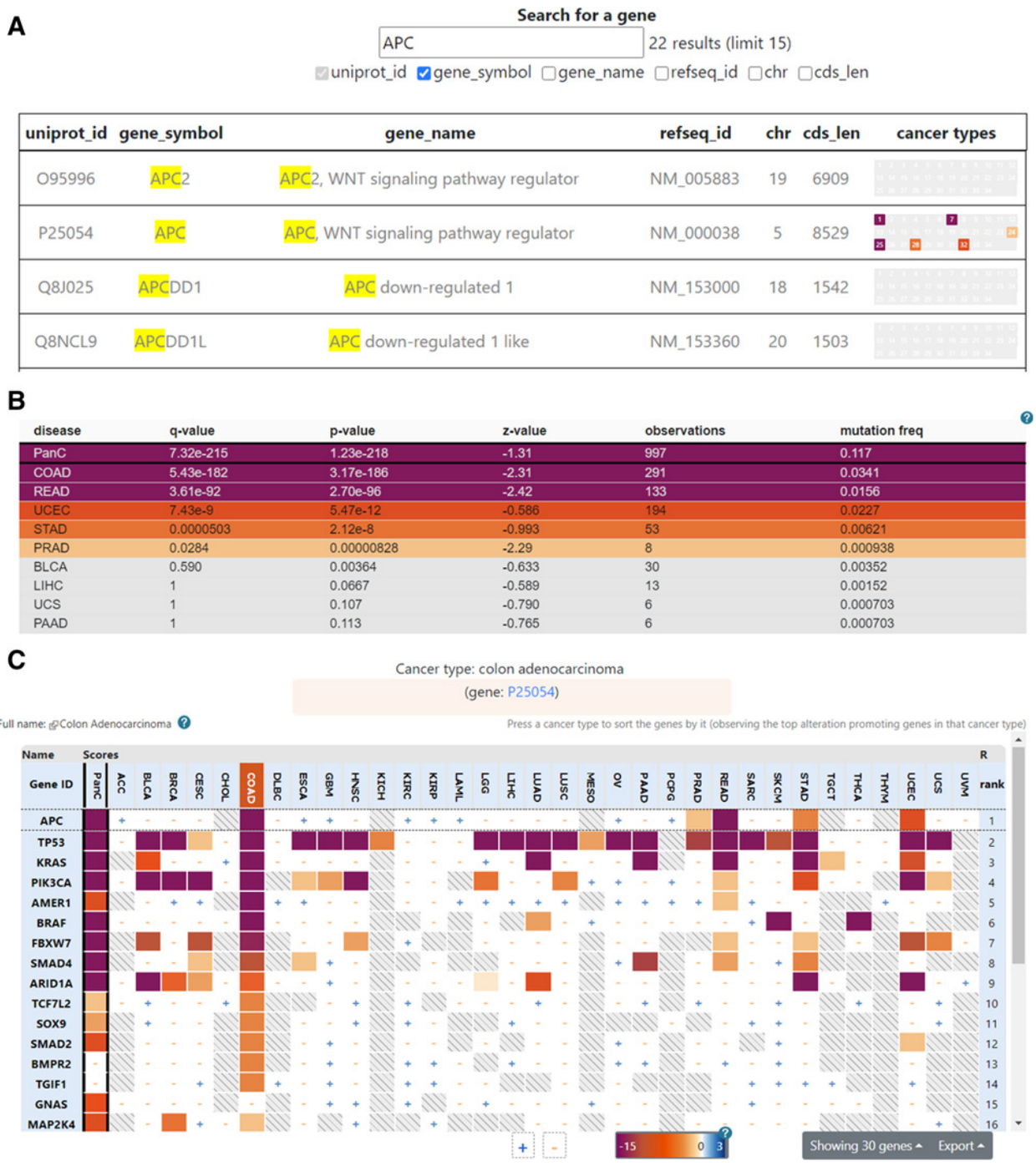
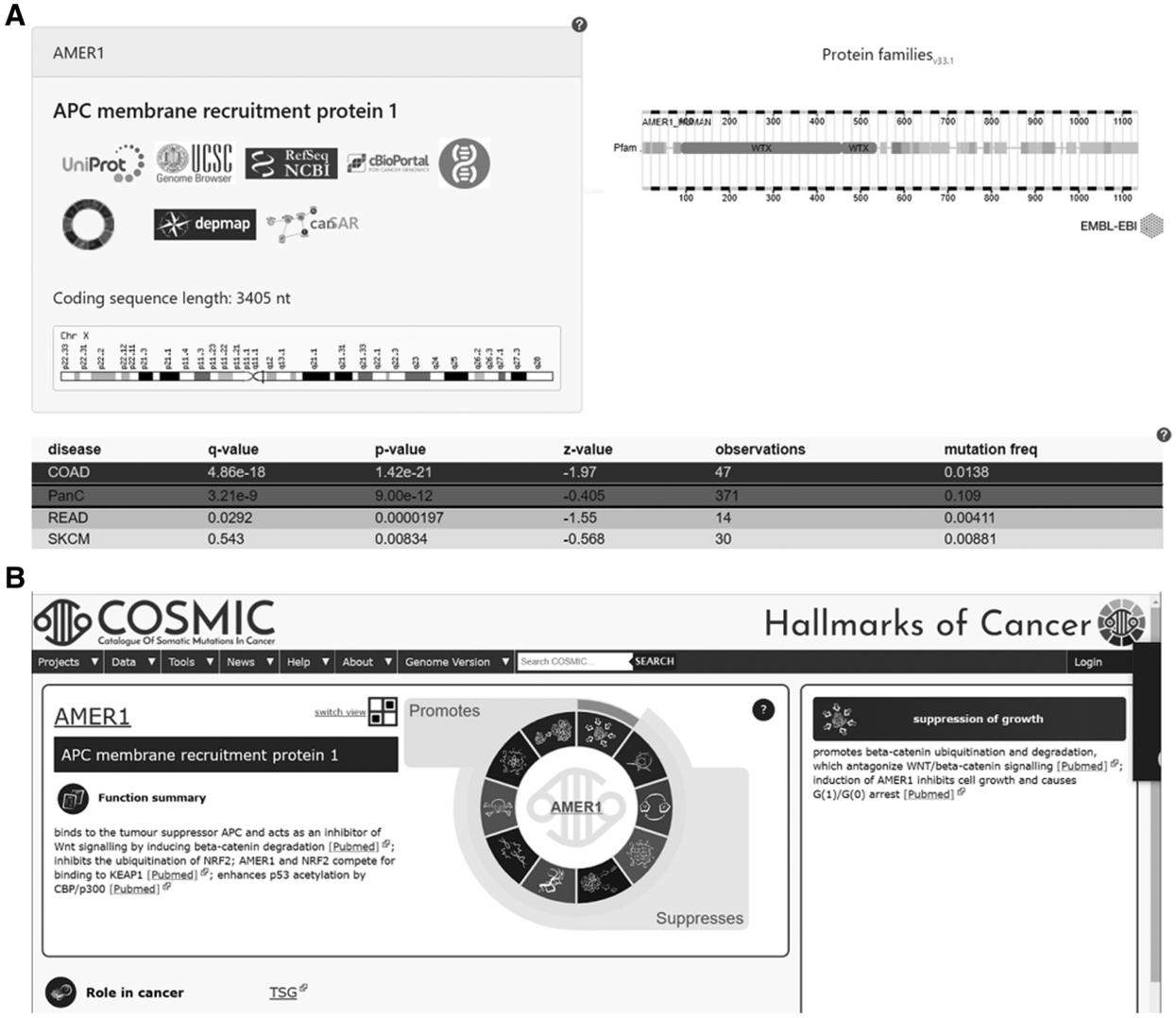


Figure 3.

Searching a gene and viewing selection patterns across cancer types (screenshots). **A,** Typing "APC" in the portal's search page (fabric-cancer.huji.ac.il/g/search). **B,** Selecting the *APC* gene links to the gene's page (fabric-cancer.huji.ac.il/g/gene/APC), with a table showing FABRIC's summary statistics for *APC* across 33 cancer types and pan-cancer. **C,** Choosing the COAD (colon adenocarcinoma) cancer type within that table switches to the primary heatmap, centered around the intersection of colon adenocarcinoma and APC (fabric-cancer.huji.ac.il/g/disease/COAD?focus=P25054). The heatmap is color coded by the significance of gene and cancer type pairs.

The FABRIC Cancer Portal allows us to observe the significance (*P* value and FDR *q* value) and effect size (*z* value) of a gene's selection across cancer types. On top of the 33 individual cancer types, it also jointly examines the somatic mutations from all cancer types aggregated, an analysis referred to as pan-cancer. We found that *APC* is specific to five cancer types (**Fig. 3B**), all of them are related to the digestive tract. In particular, the gene is under very significant and very strong positive selection in colorectal cancers, colon adenocarcinoma ($q = 5E\text{-}182$; $z = -2.31$), and rectum adenocarcinoma ($q = 3E\text{-}96$; $z = -2.42$), indicating that *APC* is a likely driver in these cancers. To a lesser extent, it also exhibits excess functional damage in uterus cancer uterine corpus endometrial carcinoma (UCEC; $q = 7E\text{-}9$; $z = -0.59$), stomach cancer ($q = 5E\text{-}5$; $z = -0.99$), and prostate cancer ($q = 0.03$; $z = -2.29$). In addition to the listed cancer types, we also observed APC's strong pan-cancer signal ($q = 7E\text{-}215$; $z = -1.31$).

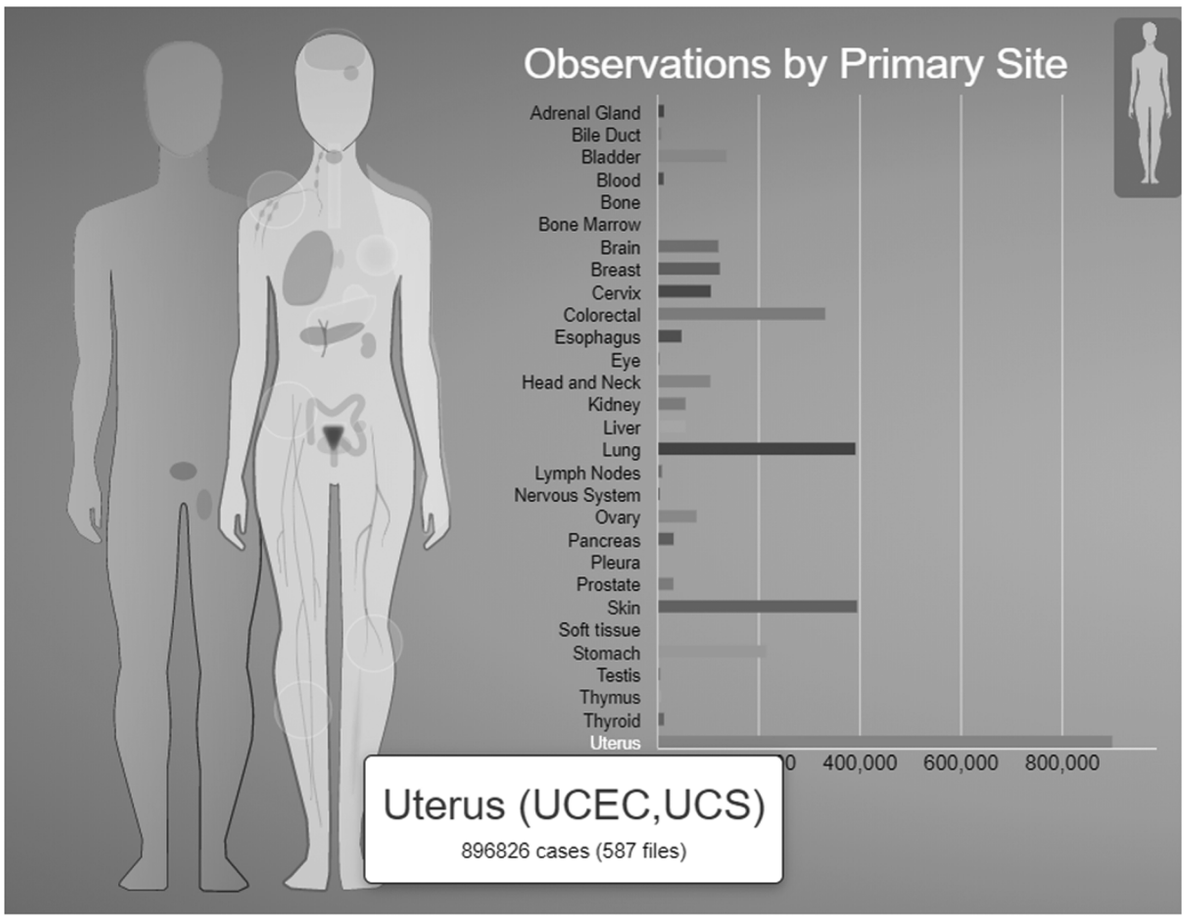
From within APC's gene page, we can choose the colon adenocarcinoma cancer type and navigate to the portal's primary heatmap. This heatmap visualizes the degree of selection across genes and cancer types, centered around the intersection of colon adenocarcinoma and *APC* (**Fig. 3C**). Having selected that particular cancer type, the heatmap ranks the genes according to their significance in colon adenocarcinoma. We observed that APC was in fact ranked first in this cancer type. The next three genes, *TP53, KRAS*, and *PIK3CA* (**Fig. 3C**), are also known cancer driver genes, which, as expected, dominate across many other cancer types and the pan-cancer analysis. The gene ranked fifth, *AMER1*, showed a different pattern. It exhibited a selection pattern that was almost exclusive to colon adenocarcinoma



disease	q-value	p-value	z-value	observations	mutation freq
COAD	4.86e-18	1.42e-21	-1.97	47	0.0138
PanC	3.21e-9	9.00e-12	-0.405	371	0.109
READ	0.0292	0.0000197	-1.55	14	0.00411
SKCM	0.543	0.00834	-0.568	30	0.00881

Figure 4.
The gene page and COSMIC cross-reference (black and white screenshots). **A,** The *AMER1* gene page includes a table at the bottom with the summary statistics of FABRIC across cancer types and, above it, the protein domain architecture of the gene according to Pfam (top, right) and cross-references to other prominent proteomics and cancer web resources with information about the gene (top, left). The gene's position overlaid on the chromatin G-bands of chromosome X appears on this panel too. **B,** Pressing the COSMIC hallmarks icon from the cross-references section links to the gene's page on COSMIC.

A



B

cancer type	q-value	p-value	z-value	observations	mutation freq
PanC	3.95e-7	1.40e-9	-0.637	88	0.0553
UCEC	0.00153	0.00000355	-0.856	28	0.0176
BLCA	0.162	0.000530	-1.47	4	0.00252
ESCA	1	0.0263	-1.74	1	0.000629
LIHC	1	0.0482	-1.74	1	0.000629
LGG	1	0.0947	-1.01	3	0.00189
KICH	1	0.112	-1.87	1	0.000629
CESC	1	0.132	-0.773	4	0.00252
LUAD	1	0.201	-0.485	7	0.00440
LUSC	1	0.226	-0.710	3	0.00189
STAD	1	0.231	-0.407	9	0.00566
SKCM	0.954	0.291	-0.487	5	0.00314
COAD	0.983	0.313	-0.323	10	0.00629
PCPG	1	0.318	1.45	1	0.000629
PAAD	1	0.437	-0.924	1	0.000629
HNSC	1	0.473	-0.379	4	0.00252
SARC	1	0.699	0.472	1	0.000629
BRCA	1	0.857	0.0973	4	0.00252
OV	1	0.997	-0.00222	1	0.000629

Figure 5.

Body parts view and the *MICU3* gene (black and white screenshots). **A,** The body parts view in The FABRIC Cancer Portal (adapted from the GDC Portal). For each primary site, the number of samples and mutations are provided. In the uterus (UCEC and UCS, as defined by the GDC Portal), for example, TCGA lists 896,826 somatic mutations from 587 samples. **B,** FABRIC's summary statistics from the gene page of *MICU3*, a gene overlooked by other cancer gene catalogues.

(q = 5E-18), with milder significance in pan-cancer (q = 3E-9) and rectum adenocarcinoma (q = 2E-5). On the basis of this selection pattern, we can infer that the role of *AMER1* in cancer is restricted to colorectal cancer.

Selecting *AMER1* in the heatmap will navigate to the gene page of *AMER1* (**Fig. 4A**). From the information on the page, we can infer that *AMER1* is tightly related to *APC*. The first indication is the gene's full name: APC membrane recruitment protein 1. Further inspection of *AMER1* can be carried out by following the available cross-references (**Table 1**). For example, pressing the Catalogue of Somatic Mutations in Cancer (COSMIC) cross-reference logo links to the *AMER1* gene page on COSMIC census (**Fig. 4B**). There, we found that *AMER1* is indeed annotated as a tumor suppressor. Like *APC*, it promotes β-catenin ubiquitination and degradation, playing a role in the inhibition of cell growth by induction of G_1–G_0-phase arrest (9). The protein family domain organization of *AMER1* according to Pfam (**Fig. 4A**) highlights numerous segments of low complexity, which often signify unstructured regions. In addition, it contains two structural WTX domains (Wilms' tumor suppressor X chromosome). This domain architecture is shared among all human paralogs of *AMER1*, namely *AMER2* and *AMER3*. Further information from GeneCards covers additional aspects of the gene, including its clinical implications, regulation, expression, and protein–protein interactions. Overall, we received confirmation that *AMER1* binds directly to *APC* and, as a result, acts to inhibit Wnt signaling by inducing β-catenin degradation. From canSAR and DepMap we observed that no approved drug targeting *AMER1* is under clinical investigation for the treatment of bowel cancers.

In addition to known cancer driver genes, such as *APC* and *AMER1*, The FABRIC Cancer Portal also lists hundreds of previously overlooked genes that are positively selected in cancer (5). One of these genes is mitochondrial calcium uptake family member 3 (*MICU3*), a gene observed to be mutationally active in UCEC. A possible entry point to UCEC (and other cancer types) is through the body parts view in the cancer types page on the portal (**Fig. 5A**). Choosing uterus as a primary site shows that *MICU3* is ranked 41st in UCEC on the heatmap.

Notably, *MICU3* is not listed as a canonical cancer driver gene by any of the major cancer catalogues (3, 4). According to FABRIC, however, it is highly significant (**Fig. 5B**). The positive selection of *MICU3* is only significant in UCEC (q = 0.0015). Nonetheless, the pan-cancer analysis is able to pick an even stronger signal (q = 4E-7), suggesting that the gene is mildly involved in other cancers as well.

Inspection of the biologic and cellular roles of *MICU3* through gene knowledge bases, such as GeneCards and UniProt, brings up relevant facts about the gene. *MICU3* is a stimulator of mitochondrial Ca^{2+} uptake through the mitochondrial calcium uniporter (MCU) complex (10). Numerous evidence links dysfunctional MCU to cancer (11), and a recent cell line study suggests that mutations in *MICU3* may lead to cell invasion by altering Ca^{2+} dynamics (12), pointing to *MICU3* as a likely cancer driver gene candidate. The strong positive selection of the gene in uterine corpus endometrial carcinoma (and potentially other cancer types), as quantified by FABRIC, provides a very strong case that *MICU3* is indeed involved in cancer.

The exemplified case studies of *APC, AMER1*, and *MICU3* demonstrate the power of The FABRIC Cancer Portal as an interactive navigation tool across genes and cancer types, promoting new discovery.

Discussion

We have introduced The FABRIC Cancer Portal, a new resource for exploring the selection patterns of human coding genes in cancer. We have showcased the use of the portal as a discovery tool, highlighting overlooked genes and providing new angles on established cancer driver genes.

FABRIC minimizes modeling assumptions and compares each gene against its own background, making it highly robust to false discoveries. The model combines genomics and proteomics data, incorporating updated high-quality proteomic annotations (including phosphorylation and other posttranslational modifications, protein domains, and secondary structure annotations). Importantly, FABRIC only analyzes coding genes, while ignoring expression data and regulatory elements of the genome (including splicing patterns).

The FABRIC Cancer Portal provides a comprehensive catalogue of the entire human coding genome across all 33 TCGA cancer types. On top of specific cancer types, the portal also provides a pan-cancer view that can capture more subtle signals that might be missed from individual cancer types due to insufficient number of observations. This is especially important in the detection of lowly recurrent genes. As demonstrated, the portal can easily pivot between gene-centric and cancer type views. Within each cancer type, FABRIC provides a clear ranking of genes by significance. From the gene view, the portal cross-references to a handful of selected external resources, providing high-quality information from genomics, proteomics, and clinical perspectives. The entire portal data and the results of specific queries are easily downloadable.

Authors' Disclosures

No disclosures were reported.

Authors' Contributions

G. Kelman: Conceptualization, software, visualization, writing-original draft, writing-review and editing. **N. Brandes:** Conceptualization, visualization, methodology, writing-original draft, writing-review and editing. **M. Linial:** Conceptualization, supervision, writing-original draft, writing-review and editing.

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