Synthetic essentiality: Targeting tumor suppressor deficiencies in cancer

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In this review, we summarize recent work exploring a novel conceptual approach termed ''synthetic essentiality'' as a means for targeting specific tumor suppressor gene deficiencies in cancer. With the aid of extensive publically available cancer genome and clinical databases, ''synthetic essentiality'' could be utilized to identify synthetic essential genes, which might be occasionally deleted in some cancers but almost always retained in the context of a specific tumor suppressor deficiency. Synthetic essentiality expands the existing concepts for therapeutic strategies, including oncogene addiction, tumor maintenance, synthetic, and collateral lethality, to provide a framework for the discovery of cancer-specific vulnerabilities. Enabled by ever-expanding large-scale genome datasets and genome-scale functional screens, the ''synthetic essentiality'' framework provides an avenue for the identification of context-specific therapeutic targets and development of patient responder hypotheses for novel and existing therapies.

Keywords:
cancer genome; cancer-specific vulnerabilities; cancer therapy; synthetic essentiality; synthetic lethality; tumor suppressor deficiency

Introduction

A decade of large-scale cancer genomics has provided a comprehensive atlas of the distribution of known oncogene and tumor suppressor gene alterations. This resource has also uncovered myriad aberrations targeting regions of unknown functional significance across many dozens of common and rare cancer types. Increased intensity in the search for recurrent dominantly acting oncogenic events in cancer can be traced to Bernard Weinstein's "oncogene addiction" concept, which describes the dependency of cancer cells on the activity of a single oncogenic protein or pathway to maintain their malignant properties (Fig. 1) [1, 2]. This concept was strengthened and experimentally validated in genetically engineered mice harboring inducible oncogenes, such as MYC and RAS [3–5]. These inducible tumor models tested the importance of oncogenic events in "tumor maintenance," that is whether a driver oncogene would remain rate limiting against the backdrop of the many other genetic alterations that accumulate during tumor development. The critical roles of these essential tumor maintenance genes have been shown to influence cancer cell growth and survival as well as the tumor microenvironment [6]. Together, these concepts, coupled with the dramatic clinical success of Gleevec (Imatinib) in chronic myeloid leukemia [7, 8], set the stage for the modern era of precision cancer therapy.

Indeed, the seminal work of Michael Stratton, Andy Futreal and colleagues, revealing that the majority of melanomas harbored activating mutations in the BRAF oncogene [9], ignited a global large-scale sequencing effort in cancer. In addition, this discovery motivated the development of BRAF inhibitors, which provided the first meaningful therapeutic progress for advanced melanoma and also catalyzed large-scale sequencing efforts of many cancer genomes. Additional impetus for comprehensive cancer profiling was generated by the work of Haber and Meyerson and their colleagues, reporting that lung cancer patients harboring mutations in the EGFR gene showed strong responsiveness to molecularly targeted agents [10, 11]. Today, while much work remains in securing a comprehensive atlas of the cancer genome, the worldwide cancer genome profiling efforts of the past decade have resulted in the discovery of new cancer genes, the development of new medicines, and improved care of cancer patients.

While successful therapeutic advances have originated from the identification of dominantly acting oncogenic mutations, the vast majority of genetic alterations have proven to be

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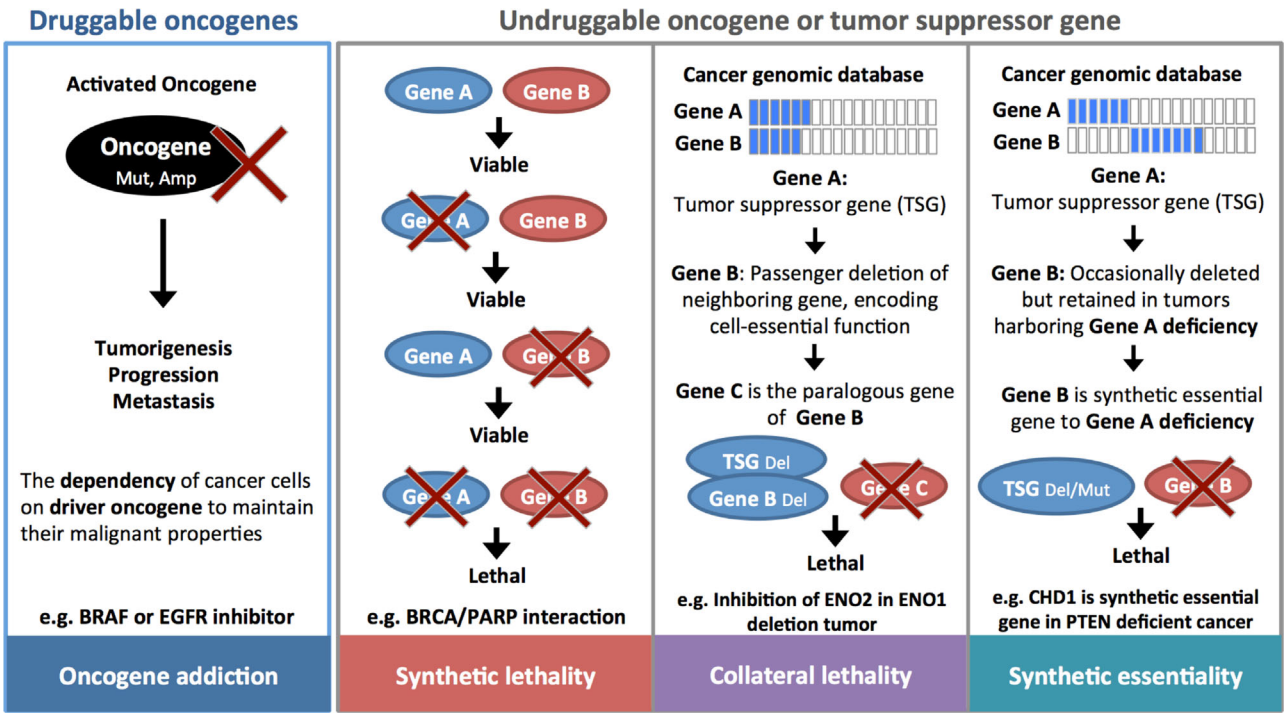

Figure 1. Schematic of conceptual approaches for the discovery of cancer-specific vulnerabilities. "Oncogene addiction" describes the dependency of cancer cells on the activity of a single oncogenic protein or pathway to maintain their malignant properties. Two genes are considered "synthetic lethal" if simultaneous inactivation of both genes causes cell death or impairs cellular fitness, whereas loss of either gene alone is viable. "Collateral lethality": a strategy for identifying cancer-specific therapeutic vulnerabilities brought about by these passenger gene deletions. "Synthetic essentiality": a new approach termed to identify synthetic essential genes, which might be deleted occasionally in some cancers but are almost always retained in the context of a specific tumor suppressor deficiency. Synthetic essentiality expands the existing concepts for therapeutic strategies, including oncogene addiction, tumor maintenance, synthetic and collateral lethality, to provide a framework for the discovery of cancer-specific vulnerabilities created by the loss of tumor suppressors. Del: deletion; Amp: Amplification; Mut: Mutation; TSG: Tumor suppressor gene.

undruggable, including activating mutations or amplifications in oncogenes such as KRAS and MYC, or neutralizing mutations or deletions in tumor suppressor genes such as PTEN and TP53. To address this challenge, an array of conceptual approaches has been designed to harness more fully the potential of the cancer genome atlas in the identification of novel therapeutic targets and to identify how such knowledge can be used to inform patient selection in clinical trials.

Synthetic lethality: Two is "worse" than one

Synthetic lethality emerged as one of the first strategies to target tumor suppressor gene alterations [12, 13]. Two genes are considered "synthetic lethal" if simultaneous inactivation of both genes causes cell death or impairs cellular fitness, whereas loss of either gene alone is viable (Fig. 1) [13]. The most celebrated example of synthetic lethality in cancer is the interaction between Poly(ADP-ribose) polymerase (PARP) and BRCA tumor suppressor genes [14]. BRCA1 and BRCA2 are important proteins in the repair of DNA double-strand breaks by homologous recombination. Mutations in these genes predispose individuals to breast, ovarian and prostate cancers [15]. BRCA-deficient cells are acutely sensitive to inhibition of PARP, which plays a key role in the repair of DNA single-strand breaks [15, 16]. PARP inhibitors were the first clinically approved drugs designed to exploit synthetic lethality to kill ovarian cancers containing BRCA mutations [14–16]. Additionally, PARP inhibitors also show promising activity in more common cancer types that possess BRCA mutations or "BRCAness," which is defined as a defect in double-strand break repair by homologous recombination repair (HRR) due to deficiencies of ATM, ATR or RAD51 genes [17]. Overall, the success of PARP inhibitors suggested the potential clinical impact of identifying additional synthetic lethal interactions in cancer.

Looking back at the history of synthetic lethality, one observes that Calvin Bridges first described this concept in 1922 in the context of his study of mutation combinations in fruit flies [18]. Hartwell, Friend and colleagues were the first to propose the use of synthetic lethality screening as a strategy to identify new anticancer drugs [12]. Over the past few decades, synthetic lethal interactions have been revealed in several model organisms including yeast, *C. elegans* and fruit flies [12, 19–21], and such studies have exposed the complexity of signal transduction and the genetic robustness of these networks in human cells. Specifically, chemical compound library screens have been performed to identify drugs capable of specifically killing cells harboring defined genetic

alterations in yeast as well as human cancer cell lines [22, 23]. But difficulties in identification of the drug targets have limited further development of these novel synthetic lethal interactions on a large scale and in a clinically relevant manner.

An important advance in the synthetic lethality arena came with the advent of RNA interference (RNAi) technology, which enabled high-throughput genetic screens using individual siRNAs or pooled lentiviral short hairpin RNA (shRNA) libraries and could be performed in human cancer cells driven by specific oncogenic mutations [24–26]. This approach proved effective in conducting large-scale loss-of-function genetic screens for the identification of synthetic lethal interactions in cancer, as well the detection of new components of cancer signaling pathways [25, 27, 28]. Two genome-wide RNAi screens performed by Elledge, Gilliland and their colleagues identified multiple synthetic lethal interactions with the *RAS* oncogenes [28, 29]. Additionally, synthetic lethal RNAi screens have been used to identify chemosensitizer loci in cancer cells [30]. Continued efforts in the identification of context specific genetic dependencies have led to an outstanding resource (Morpheus, https://software.broadinstitute.org/morpheus/ – generated by William Hahn's group), which includes parallel genome-scale shRNA screens in 216 cancer cell lines [31]. As RNAi screening hits were subjected to in-depth validation, it also became clear that additional refinements were needed to diminish the off-target effects of RNAi-dependent screening technology.

The recent development of CRISPR-Cas9 technology with its enhanced genome editing specificity [32, 33] has improved the accuracy of high throughput loss-of-function screening to uncover synthetic lethal interactions and therapeutic targets in cancer on a larger scale [34–37]. Compared to the traditional shRNA-based system, Bastiaan, et al. found that CRISPR knockout screening outperformed shRNA-based screens in identifying essential genes [38]. High-resolution CRISPR screens performed by Traver Hart, et al. are now considered the gold standard reference sets for essential and non-essential gene validation in human cells, and provide a framework by which to compare the quality of functional genetic screens [36]. Most recently, Sabatini and his group generated a gene essentiality dataset across 14 human acute myeloid leukemia cell lines, and uncovered synthetic lethal partners of oncogenic RAS by comparisons of differentially essential genes between RAS-dependent and –independent lines [39]. This study suggested a general strategy for defining mammalian gene networks and synthetic lethal interactions by exploiting genome-wide CRISPR-based screens in human cancer cells with genetic and epigenetic diversity [39]. Meanwhile, CRISPR interference (CRISPRi), a genetic perturbation technique that allows for sequence-specific repression or activation of gene expression, has also been used to search for tumor maintenance genes, tumor suppressors, and synthetic lethal interactions [40, 41].

The synthetic lethality framework has also been used to identify drug combinations in cancers using high-throughput screening of combinatorial compounds, RNAi or CRISPR-derived mutations. For example, Wong, et al. developed a technology, combinatorial genetics *en masse* (CombiGEM), which is designed to identify miRNA combinations that synergistically sensitize drug-resistant cancer cells to chemotherapy and/or inhibit cancer cell proliferation using high-order barcoded combinatorial genetic libraries [42]. Griner, et al. generated an unbiased small-molecule combination (matrix) screening for the rapid and systematic identification of synergistic, additive, and antagonistic drug combinations [43]. In addition, computational and bioinformatics approaches have contributed to the identification of even more synthetic lethal interactions [44]. A recent community computational prediction challenge launched by the DREAM consortium assessed 32 methods for ranking drug combination efficacy, four of which performed significantly better than random guessing, suggesting that computational prediction of compound-pair activity is possible [45]. Together, combined with advanced loss-of-function screening tools, the notion of targeting synthetic lethal vulnerabilities in cancer has displayed its power in illuminating novel candidate targets for the improved treatment of cancers.

Collateral lethality: The good "neighborhood"

Genomic deletions of tumor suppressor gene loci frequently result in "passenger" deletion of neighboring genes that do not play known roles in processes of malignant transformation. In 2012, we proposed the concept of "collateral lethality" as a strategy for identifying cancer-specific therapeutic vulnerabilities brought about by these passenger gene deletions (Fig. 1) [46]. Specifically, we reasoned that, while many of these neighboring genes encode cell-essential housekeeping functions, their deletion is tolerated due to the presence of functionally redundant genes residing elsewhere in the genome [47]. The inhibition of these paralogous genes would be expected to compromise cancer cells harboring the deletion, yet spare normal cells that retain intact genomes. William Hahn and colleagues proposed a similar concept, termed CYCLOPS [48]. In the collateral lethality framework, we showed that the inhibition of glycolytic gene enolase 2 (ENO2) selectively suppressed growth, survival and the tumorigenic potential of ENO1-deleted, but not ENO1-intact, glioblastoma cells [46]. In the Hahn study, an integrated analysis of genome-wide copy numbers and RNAi profiles identified 56 genes (enriched for spliceosome, proteasome, and ribosome components) as potential cancer-specific vulnerabilities associated with specific copy number losses [48]. Specifically, they identified Proteasome 26S Subunit ATPase 2 (PSMC2) as one such CYCLOPS gene; subsequent functional assays indicated that cells harboring partial PSMC2 copy number are sensitive to PSMC2 suppression.

A second example of collateral lethality was reported in pancreas cancer [49]. Deletion of the tumor suppressor gene SMAD4 is common in pancreas cancer and often results in co-deletion of a neighboring metabolism gene, malic enzyme 2 (ME2), which confers a cancer-specific metabolic vulnerability with inhibition of the paralogous isoform ME3 in ME2-deleted cells [49]. Mechanistically, cells deficient in mitochondrial malic enzymes ME2 and ME3 revealed diminished NADPH production and high levels of reactive oxygen species, consequently resulting in the dysfunction of de novo

nucleotide synthesis through the transcriptional suppression of branched-chain amino acid transaminase 2 (BCAT2) gene. Additional recent studies by Garraway and Marks and their colleagues revealed protein arginine methyltransferase 5 (PRMT5) as a therapeutic target in cancers harboring methylthioadenosine phosphorylase (MTAP) deletions, which occur as a result of deletion of the neighboring CDKN2A tumor suppressor locus across multiple cancer lineages [50, 51]. Mechanistically, MTAP deficiency leads to increased intracellular methylthioadenosine, which in turn inhibits PRMT5 activity and confers heightened susceptibility to further depletion of PRMT5. Thus, the principle of collateral lethality provides a framework for the development of therapies resulting from tumor suppressor gene deficiencies and further informs the application of such therapies in genome-annotated patient populations.

What is "synthetic essentiality"

As noted above, synthetic lethal screens have proven effective in identifying synergistic combinations. Such studies however can be constrained by the use of a limited number of cell lines, which does not capture the full genetic diversity of the human cancer type under study, and by specific in vitro screening conditions that may not recapitulate the biology of autochthonous tumors. Correspondingly, a comparison of cell lines from the Cancer Cell Line Encyclopedia (CCLE) and patient tumors from TCGA has shown a significant variation in genomic markers [52], which might be due to differing clonal selection and evolutionary paths incurred under in vivo versus in vitro conditions. These observations prompted us to ask whether the patterns of deletion in large-scale cancer genome datasets from many primary human tumors might reveal mutually exclusive events that belie a synthetic lethal interaction that occurred in the context of naturally evolving human tumors [53]. For example, examination of these genome datasets reveals that, while the PARP locus is occasionally deleted in human cancers, it is very rarely deleted in BRCA mutant tumors.

We considered the possibility of the existence of "synthetic essential" genes that are occasionally deleted in some cancers but are almost always retained in the context of a specific tumor-suppressor deficiency, reasoning that such a retained gene might be required for executing critical cancer-specific actions in the context of the given deficiency. We posited that such a synthetic essential gene could be a therapeutic target in cancers that harbor the specific tumor suppressor deficiency [53]. In addition to several known synthetic lethal interactions, such as BRCA/PARP noted above, our approach and experimental results also uncovered chromatin helicase DNA-binding factor (CHD1) as a synthetic essential gene and therapeutic target in PTEN-deficient prostate and breast cancers [53]. CHD1 is involved in the maintenance of open chromatin and cooperates with Trimethylation of histone H3 at lysine 4 (H3K4me3) to control pluripotency of murine embryonic stem cells [54]. In this study, we identified a novel PTEN pathway linking PTEN and chromatin-mediated regulation of the cancer-relevant NF-κB network. Specifically, mechanistic analyses identified a pathway comprised of PTEN → AKT → GSK3β-mediated phosphorylation and degradation of CHD1 via the ubiquitination-proteasome process (Fig. 2). In cancer, PTEN deficiency stabilizes CHD1, which in turn engages and maintains the H3K4me3 mark to activate cancer promoting gene expression including the NF-κB network, which is known to promote PCa progression (Fig. 2).

In the case of PTEN/CHD1, the synthetic essential gene CHD1 can serve as an essential downstream effector for the specific tumor suppressor gene deficiency, PTEN. This unidirectional essentiality is due to the epispastic relationship of PTEN to CHD1, that is CHD1 is essential to PTEN deficient tumors but PTEN inhibition does not cause lethality to CHD1 deficient cells. In contrast, many synthetic lethal interactions often involve two genes in parallel pathways that converge on the same essential biological process (e.g. convergence of BRCA and PARP on DNA repair processes), suggesting bidirectional essentiality of one gene to the deficiency of another gene. Thus, "synthetic essentiality" can be viewed as a subset or type of "synthetic lethality," reasoning a synthetic lethal interaction could be composed of solo or dual synthetic essentiality (Fig. 1). Additionally, "synthetic essentiality" provides an approach to identify therapeutic targets through a search for genes that are occasionally deleted in some cancers but are almost always retained in the context of a specific tumor-suppressor deficiency in human cancer genomic databases.

In cancer, the processes of mutation and selection are integral to the evolution of cancer from tumor genesis to metastasis [55]. Our approach utilizes the vast cancer genomic database with its tens of thousands of human tumor profiles that reflect the eventual outcomes of tumor evolution due to in vivo natural negative selection of loss-of-function mutations during tumor development. Beyond the BRCA/PARP and PTEN/CHD1 pairs, the mutually exclusive deletion patterns of numerous well-established synthetic lethal interactions were also observed in the TCGA cancer genomics database (Table 1). These examples provide a starting point in the search for synthetic essential targets for specific tumor suppressor gene deficiencies as well as activating oncogenic events including those that are considered "undruggable" such as KRAS and MYC.

How to identify synthetic essentiality in cancer genomes

Combined with other screening approaches or previously published data of synthetic lethal screens using RNAi, CRISPR or small compounds libraries in cancer cells as well as classic model organisms, the synthetic essentiality framework may expand and facilitate the discovery pipeline for cancers exhibiting a limited number of "tumor maintenance" oncogenic events. Another advantage of our framework comes from the vast genetic, biological and clinical information of the cancer genomic database. Gene expression profiles, protein/ phosphoprotein data, and pathology and clinical information would provide an avenue for the investigation of molecular mechanisms, biological functions and clinical

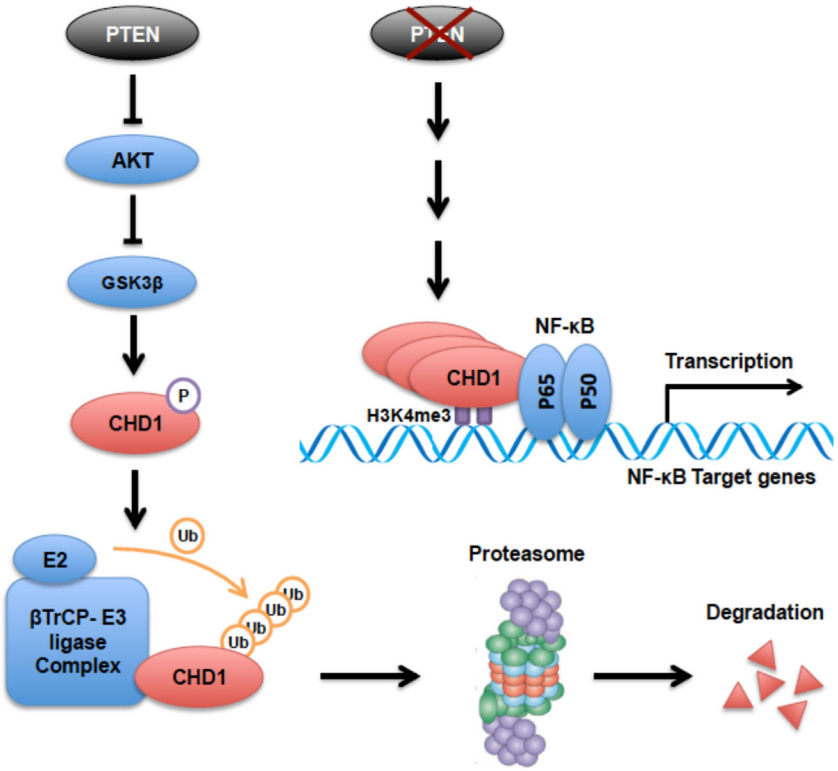

Figure 2. The PTEN-CHD1-NF-κB network. GSK3β is activated by PTEN through inhibition of AKT, and phosphorylates CHD1, which stimulates its degradation through β-TrCP mediated ubiquitination-proteasome pathway. Thus, in PTEN-deficient prostate cancer cells, accumulated CHD1 interacts with and maintains H3K4me3, followed by transcriptional activation of NF-κB downstream genes leading to prostate cancer progression.

relevance of putative synthetic essential genes, as discussed in a following section (Fig. 3).

As a first step, exploration of the large-scale publicly available cancer genomic datasets using well-established websites including cBioPortal [56, 57] (http://www.cbioportal.org) and FIREHOSE (https://gdac.broadinstitute.org) can identify genomic alterations. These tools afford investigators with the capacity to audit genomic alterations of interest across specific cancer types and evaluate mutual exclusivity patterns employing statistical methods [58]. Next, to generate a putative synthetic essential gene (Gene B) list for a given tumor suppressor gene or oncogene (Gene A) in a specific cancer type, one can download the relative cancer genomic data (for example, TCGA prostate cancer database), and calculate the log odds ratios between alterations in Gene A and deep deletions in Gene B, which indicates mutual exclusiveness (Log odds ratio $<=0$) or co-occurrence (Log odds ratio >0). Except for deletions, truncated mutations or other well-established loss-of-function mutations of Gene B can also be included in the mutual exclusiveness analysis. The gene list can then be ranked according to the odds ratio (Log odds ratio $<=0$) and the p-value of the Fisher Exact Test. In most cases, a long gene list ($N>500$ or $N>1000$) is generated, and narrowing this candidate list is a key step for further validation. Here, we highlight some methods for generating a shorter and more effective list (Fig. 3).

First, the filter of at least three occurrences of Gene B deletion should be used to guarantee the biological reproducibility. Generally speaking, a relatively lower odds ratio score and smaller p-value (typically ≤ 0.1) indicate a stronger mutually exclusive relationship between the candidate Gene B and a specific Gene A. For comparison of the odds ratios, we recommend splitting the list according to mutual exclusiveness with or without cases of overlapped alterations in Genes A and B. This is because the overlap cases may profoundly affect the odds ratio, resulting in the incomparability of mutually exclusive gene pairs in the presence or absence of overlapped alterations. The absolute number of cases with deleted Gene B can be used as another filter; due to biological duplicates and statistical significance, it may be more reliable to set three occurrences as the cutoff.

Table 1. Mutual exclusiveness of known synthetic lethal interactions

Synthetic lethal interactions		
Gene A	**Gene B**	**Mutually exclusive patterns shown in cancer genetic database**
BRCA1	PARP1/PARP2	Prostate Adenocarcinoma (TCGA, Provisional; $N=499$)
ETS (ERG/ETV1)	PARP1	Prostate Adenocarcinoma (TCGA, Provisional; $N=499$)
PTEN	PLK4	Prostate Adenocarcinoma (TCGA, Cell 2015; $N=333$)
PTEN	PARP1	Prostate Adenocarcinoma (TCGA, Cell 2015; $N=333$)
KRAS	CDK1	Pan-Lung Cancer (TCGA, Nat Genet 2016; $N=1144$)
KRAS	PLK1	Colorectal Adenocarcinoma (DFCI, Cell Reports 2016; $N=619$)
MTAP	PRMT5	Pan-Lung Cancer (TCGA, Nat Genet 2016; $N=1144$)
VHL	EPAS1 (HIF2a)	Kidney Renal Clear Cell Carcinoma (TCGA, Provisional; $N=538$)
ATM	PRKDC (DNAPK)	Skin Cutaneous Melanoma (TCGA, Provisional; $N=419$)
MYC	BRD4	Colorectal Adenocarcinoma (TCGA, Provisional; $N=633$)
MYC	MTOR	Colorectal Adenocarcinoma (TCGA, Provisional; $N=633$)
MYC	CDK1	Prostate Adenocarcinoma (TCGA, Provisional; $N=499$)

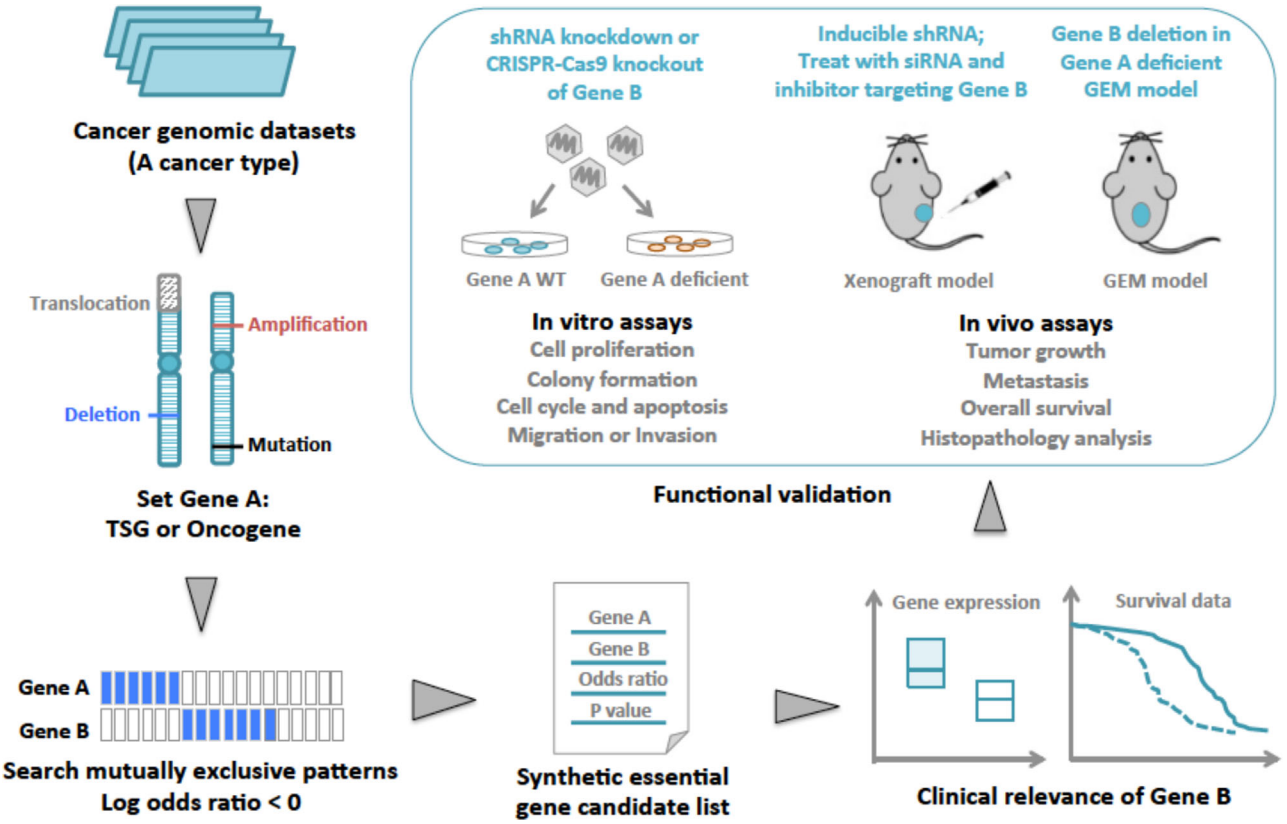

Figure 3. Pipeline for identifying synthetic essential genes. To generate a putative synthetic essential gene (Gene B) list for a given tumor suppressor gene or oncogene (Gene A) in a specific cancer type, one can download the relative cancer genomic data and search for mutually exclusive deletions (Log odds ratio <0). The candidate list can then be narrowed down according to Gene B expression and other clinical relevance. The in vitro and in vivo functional assays using isogenic cancer cell lines are needed to evaluate the anti-tumor effects of depleting a putative synthetic essential gene. TSG: tumor suppressor gene; GEM model: genetically engineered mouse model.

From the view of biological mechanism, since Gene B plays an essential role in the context of Gene A alterations, Gene B may be up-regulated or activated in a compensatory manner in tumor cells containing Gene A mutations. Therefore, expression profiles and protein levels from the same cancer database may be useful in prioritizing synthetic essential gene candidates. Moreover, these data can also inform further studies of the regulatory relationship between Genes A and B. In addition, the available clinical information in these databases can assist in uncovering the clinical relevance of putative synthetic essential genes for indicated cancer categories, including survival and cancer subtypes, which are important for the selection of potential therapeutic targets. Additional information, including whether the candidates are druggable or have existing inhibitors, can be considered as additional filters depending upon research goals.

Once a short-list has been generated, both in vitro and in vivo functional assays are needed to evaluate the anti-tumor effects of depleting a putative synthetic essential gene. Performing shRNA-mediated knockdown or CRISPR-mediated knockout of individual candidates in comparable isogenic cancer cell lines (such as a PTEN-wildtype cell line and its isogenic CRISPR-mediated PTEN knockout cell line), can be a good strategy for functional validation of cell proliferation, apoptosis, invasion and tumor growth. Another strategy is perform an in vivo CRISPR screen with pooled sgRNA targeting synthetic essential gene candidates with a large number of negative control genes; or with genome-wide CRISPR-based screens. A specific inhibitor of a given candidate could be used for cytotoxic assays as well, but off-target effects of the inhibitor may cause unexpected results in some cases. Inducible shRNA or CRISPR-Cas9 systems are good tools to study the functions of synthetic essential genes on tumor maintenance. Functional evaluations in multiple cancer cell lines with or without Gene A alterations is another strong test, but possible mutations of genes localized upstream of Gene A in a molecular pathway should be considered during data interpretation. Genetically engineered mouse cancer models provide a system for further genetic validation and phenotypic assessment that cannot be explored in human cancer cell lines in vitro including aspects of metastasis and heterotypic interactions between cancer cells and host cell of the tumor microenvironment. Beyond these genetic validation studies, pharmacological proof-of-concept using tool compounds in these models systems will be needed to further substantiate synthetic essential gene relationships that would justify full translational development.

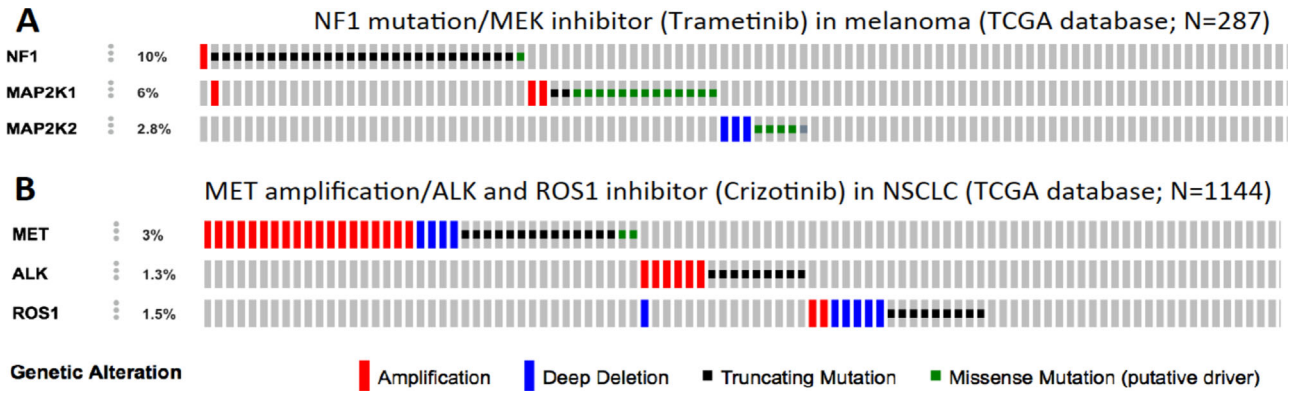

Figure 4. Mutually exclusive patterns of effective clinical treatments. **A:** Trametinib (MEK inhibitor) in melanoma containing NF1 mutation. **B:** Crizotinib (ALK and ROS1 inhibitor) in NSCLC with MET amplification.

Opportunities and challenges

Synthetic and collateral lethality concepts provide a foundation for targeting the undruggable in cancer. Along similar lines, the concept of synthetic essentiality provides another framework to identify therapeutic targets for cancers that harbor tumor suppressor gene deficiencies. Searching for synthetic essential genes might fill the gap between informative loss-of-function screening of synthetic lethality and the large-scale cancer genomics databases with clinical resources, when searching and validating therapeutic targets in cancer.

In addition, identification of synthetic essential relationships in cancer genomics may influence existing strategies of clinical cancer treatment or promising clinical trials. Beyond the use of PARP inhibitors in BRCA (BRCA/PARP) and PTEN (PTEN/PARP) deficient cancers, which has been discussed in our recent publication [53], we found several other examples of mutually exclusive deletion patterns in current clinical therapy, including Trametinib (MEK inhibitor) in melanoma containing NF1 mutation (NF1/MAPK2, Fig. 4A), Crizotinib (ALK and ROS1 inhibitor) in NSCLC with MET amplification (MET/ALK and MET/ROS1, Fig. 4B), although these mutually exclusive patterns might not be observed or shared in all cancer types. This evidence suggests that synthetic essentiality determinations may provide valuable additional information to inform on-going clinical trial designs, as well as drug repositioning in cancer therapy, thereby impacting precision medicine in oncology.

Although the screening of synthetic essential genes in cancer genomics is a powerful tool for identifying therapeutic targets and effective drug combinations, there are several factors and caveats that need to be considered in synthetic essentiality analyses of mutually exclusive deletion patterns. The mutually exclusive pattern of gene alterations is usually used to identify the components in a single signaling pathway [59], such as TP53/MDM2 and TET2/WT1 [60], thus the deletion of two tumor suppressor driver genes localizing in the same pathway may also show mutually exclusive deletion patterns. This is because an alteration in one gene of a pathway typically alleviates genetic pressure to alter another driver in the same pathway given the minimal additional selective advantage to the cancer cell. In addition, a neighboring passenger deletion of a tumor suppressor gene could yield false positive candidates, because the deleted gene might show a mutually exclusive pattern similar to a real synthetic essential gene following cancer genomic analysis. Also, in some tumor types with high stromal cell percentage, such as pancreatic cancer [61], the tumor cells are usually mixed with the stromal cells and other infiltrating immune cells, resulting in difficulties distinguishing whether the mutual exclusiveness comes from tumor cells or other cell types in the tumor microenvironment. For instance, PTEN and TP53 mutations in breast cancer stromal cells have been found to control tumor progression as well [62, 63]. Therefore, careful selection of candidates followed by in vitro and in vivo functional validation using appropriate systems is key to successfully identifying synthetic essential genes.

Conclusion

Today, publicly available large-scale cancer genomic datasets are valuable resources for scientists and clinicians, and can provide powerful weapons in the fight against cancer when linked to functional validation approaches and clinical data. Searching for synthetic essential genes in cancer genomics can provide a promising and productive starting point to discover targetable vulnerabilities and novel combinations for the development of personalized cancer therapies.

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