$TCGA_{DEPMAP}$ – Mapping Translational Dependencies and Synthetic Lethalities within The Cancer Genome Atlas.

Author(s): Xu Shi[1], Daniel Verduzco[2], Sakina Petiwala[2], Cynthia Jeffries[2], Charles Lu[2], Tifani Anton[2], Christos Gekas[1], J. Matthew Barnes[2], Soma Roy[1], Cyril Ramathal[2], Michael J. Flister[2]*, Zoltan Dezso[1]*

Affiliations:

1. AbbVie Bay Area, 1000 Gateway Boulevard, South San Francisco, CA 94080

2. AbbVie Inc., 1 North Waukegan Rd., North Chicago, IL 60064

Correspondence: Michael Flister (michael.flister@abbvie.com); Zoltan Dezso (zoltan.dezso@abbvie.com)

Statement of Significance:

The translational dependency maps described here provide novel insights into the essentiality and tolerability of target genes in human patients. Starting with 20 million data points from the Cancer Dependency Map (DEPMAP), the Cancer Genome Atlas (TCGA), the Patient-Derived Xenograft Encyclopedia (PDXE), and the Genotype-Tissue Expression (GTEx) projects, machine learning models for gene dependency and tolerability were built to predict:

- 6,581 predictive models of gene essentiality in $TCGA_{DEPMAP}$ and $PDXE_{DEPMAP}$, with corresponding prediction of gene tolerability in $GTEX_{DEPMAP}$
- 1,966 predictive models of selective gene essentiality were robustly cross-validated and accurately predict pan-cancer lineages (AUC>0.99), breast cancer sub-lineages (AUC>0.88), synthetic lethalities (SMARCA2/4, STAG1/2, EP300/CREBBP etc.), and oncogenic drivers (e.g., KRAS, BRAF, ERBB2, etc.)
- 881 synthetic lethality candidates were tested using a unique multiplexed CRISPR screen by AsCas12a across multiple cancer models, revealing multiple translatable and potentially druggable synthetic lethalities (STAG1/2, EP300/CREBBP, PAPSS1/2, etc.)

ABSTRACT

The Cancer Genome Atlas (TCGA) has yielded unprecedented genetic and molecular characterization of the cancer genome, yet the functional consequences and patient-relevance of many putative cancer drivers remain undefined. $TCGA_{DEPMAP}$ is the first hybrid map of TCGA patient dependencies that was built from 6,581 expression-based predictive models of gene essentiality across 791 cancer cell models within the Cancer Dependency Map (DEPMAP). $TCGA_{DEPMAP}$ captured well-known cancer lineage and sub-lineage dependencies (e.g., ESR1, ERBB2, etc.), genetic drivers (e.g., KRAS, BRAF, etc.), and synthetic lethalities (e.g., STAG1/2, SMARCA2/4, etc.), demonstrating the robustness of $TCGA_{DEPMAP}$ as a translational dependency map. $TCGA_{DEPMAP}$ also unveiled novel synthetic lethalities that were experimentally confirmed using multiplexed CRISPR screening across multiple cancer cell lines. Other map "extensions" were built to capture unique aspects of patient-relevant tumor dependencies, including translating gene essentiality to drug response in patient-derived xenograft (PDX) models (i.e., $PDXE_{DEPMAP}$) and predicting gene tolerability within normal tissues ($GTEX_{DEPMAP}$). Collectively, this study demonstrates how translational dependency maps can be used to leverage the rapidly expanding catalog of patient genomic datasets to identify and prioritize novel therapeutic targets with the best therapeutic indices.

INTRODUCTION

The rapid expansion of genomic technologies to characterize healthy and diseased patient populations has provided unprecedented resolution to the pathophysiological drivers of cancer and many other diseases. In 2018, TCGA completed a 10-year study of 33 tumor types across ~11,000 patients, which has broadly illuminated the genetic underpinnings of cancer (*1*). Building on the success of TCGA, multiple other initiatives have been launched to explore aspects of cancer initiation, evolution, metastasis, and response to therapy (*2–6*), with the hope that the deepening molecular characterization of cancer will improve diagnosis, treatment, and prevention. However, a critical step towards fully leveraging patient data to eradicate cancer is to assign functionality to the observations made in TCGA that translate putative tumor dependencies to life-saving therapies.

One approach to understanding tumor dependencies is through genomewide genetic and chemical perturbation datasets (e.g., DEPMAP (*7*, *8*), Project SCORE (*9*), and Connectivity Map (*10*)) that have been paired with thousands of deeply characterized cancer models (e.g., Cancer Cell Line Encyclopedia (*11*), Cancer Cell Line Factory, (*12*), and Human Cancer Models Initiative (*13*)). Multiple studies have demonstrated the ability of DEPMAP to translate gene essentially to novel therapeutic targets (*14–18*) and a broader functional understanding of tumor dependencies (*19*, *20*). Compared with TCGA, a differentiating strength of the "dependency maps" is that hypotheses can be readily tested, replicated, and refined in different contexts, whereas patient datasets are typically not amenable to functional experimentation. However, the dependency maps also pose limitations when compared to the translatability of TCGA, as homogeneous cell lines in culture dishes do not replicate the pathophysiological complexities of the intact tumor microenvironment (TME) (*21*). Further, the current experimental models do not completely recapitulate the genetic drivers that are present in the patient population (*22*) and experimental outcomes of genetic perturbation screens do not capture most aspects of disease outcome and patient survival.

To address the unique challenges posed by TCGA and DEPMAP, we generated a hybrid dependency map ($TCGA_{DEPMAP}$) that transfers predictive modelling of dependencies in the DEPMAP onto TCGA using the transcriptomic profiles of experimental models and patients, respectively. As such, $TCGA_{DEPMAP}$ leverages the experimental strengths of DEPMAP, while enabling patient-relevant translatability of TCGA. Using $TCGA_{DEPMAP}$, we were able to detect many known cancer lineage and sub-lineage dependencies (e.g., ESR1, ERBB2, etc.), genetic drivers (e.g., KRAS, BRAF, etc.), and synthetic lethalities (e.g., STAG1/2, SMARCA2/4, etc.). Finally, we built multiple map "extensions" to capture other aspects of patient-relevant tumor dependencies, including the ability to translate gene essentiality to drug response in PDX models (i.e., $PDXE_{DEPMAP}$) and the capability to predict gene tolerability within normal tissues ($GTEX_{DEPMAP}$). Collectively, this study demonstrates how translational dependency maps can be leveraged to functionalize oncogenic drivers and novel therapeutic targets in the rapidly expanding catalog of patient genomic datasets.

RESULTS

Building a translational resource for tumor dependencies: $TCGA_{DEPMAP}$

The current DEPMAP (https://depmap.org/portal/) is a compendium of genomewide CRISPR knockout screens across 791 models from the Cancer Cell Line Encyclopedia (CCLE) with genomic, epigenomic, transcriptomic, proteomic, and metabolomic annotations (*7*, *11*). Genomewide gene essentiality across the DEPMAP cell lines was estimated using CERES (*23*) and the dependency probability of each gene is calculated relative to the distribution of effect sizes for common essential and nonessential genes within each cell line (*24*). Despite the abundance of annotated features within the DEPMAP, two recent studies estimated that expression-based predictive modeling of gene essentiality outperformed all other features (e.g., somatic mutations, copy number, methylation, etc.) (*24*, *25*). An added benefit of building predictive models using transcriptomics is that it is robust and broadly captured across TCGA and many other clinically studies (*26*), as well as PDX studies (*27*, *28*). Further, expression-based predictive modeling of dependency can be extended to non-oncological studies (e.g., GTEX), which do not have somatic mutations and copy number changes (*29*). Thus, building a translational dependency map using expression-based modeling robustly predicts gene essentiality and can be broadly applied across malignant and normal tissue types alike.

To begin building translational dependency maps, expression-based predictive models of gene essentiality were generated from DEPMAP (*7*) using elastic-net regularization for feature selection and modeling, as it is suited to handle highly dimensional data with correlated variables (*30*) (**Figure 1A**). Predictive essentiality models were generated for 6,581 genes, excluding common-essential and non-essential genes (see Materials and Methods). The number of features for each predictive model ranged from 3 to 369 (median 80), and 10-fold cross-validation identified 1,966 models with the highest performance (**Figure 1B-C** and **Table S1**). ERBB2 is an example of a highly performing model with strong correlation between predicted and observed essentiality in the DepMap (R=0.75, $p<2.2e^{-16}$) (**Figure D**), which is predominantly correlated with ERBB2 and NRG1 expression (**Figure 1E**), as has been reported previously (*31*).

The expression-based predictive models of DepMap dependencies were next applied to the transcriptomic profiles of 9,596 TCGA patients, following normalization to account for differences between the expression profiles of homogenous cell lines and tumor biopsies with varying stromal content. To control for confounding stroma, the expression profiles of the DepMap cell lines and TCGA patients were transformed by contrast PCA (cPCA) to remove cPC1 and cPC2 that comprise the infiltrating immune system and other stromal cell types, respectively (*34*). As expected, the removal cPC1 and cPC2 significantly improved the alignment of the expression-based dependency models when applied to TCGA patient transcriptomes (**Figure 1F** and **1G**).

Characteristics of $TCGA_{DEPMAP}$ and comparison with the $PDXE_{DEPMAP}$ and $GTEX_{DEPMAP}$

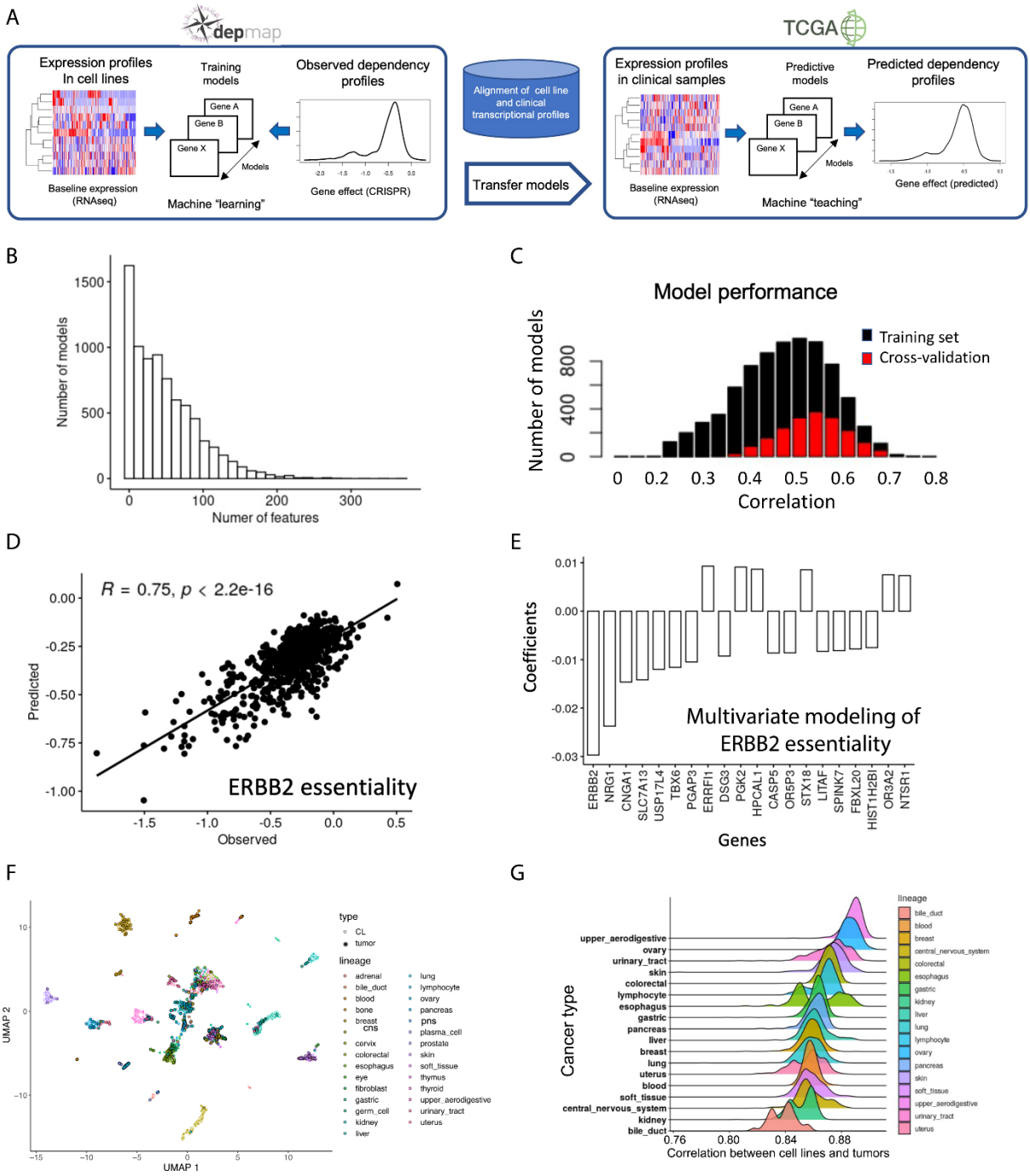

Figure 1. Building a translational dependency map ($TCGA_{DEPMAP}$). (**A**) Schematic of expression-based gene essentiality models built from the DEPMAP and applied to $TCGA_{DEPMAP}$. (**B**) Distribution of features per model and (**C**) cross-validation of models. (**D**) An example of cross-validation of the gene essentiality for ERBB2 and (**E**) the major features that predict ERBB2 essentiality. (**F**) Transcriptional normalization using cPCA is critical for proper alignment of dependency models, as indicated by strong correlation of gene essentiality models across cell lines and $TCGA_{DEPMAP}$ (**G**).

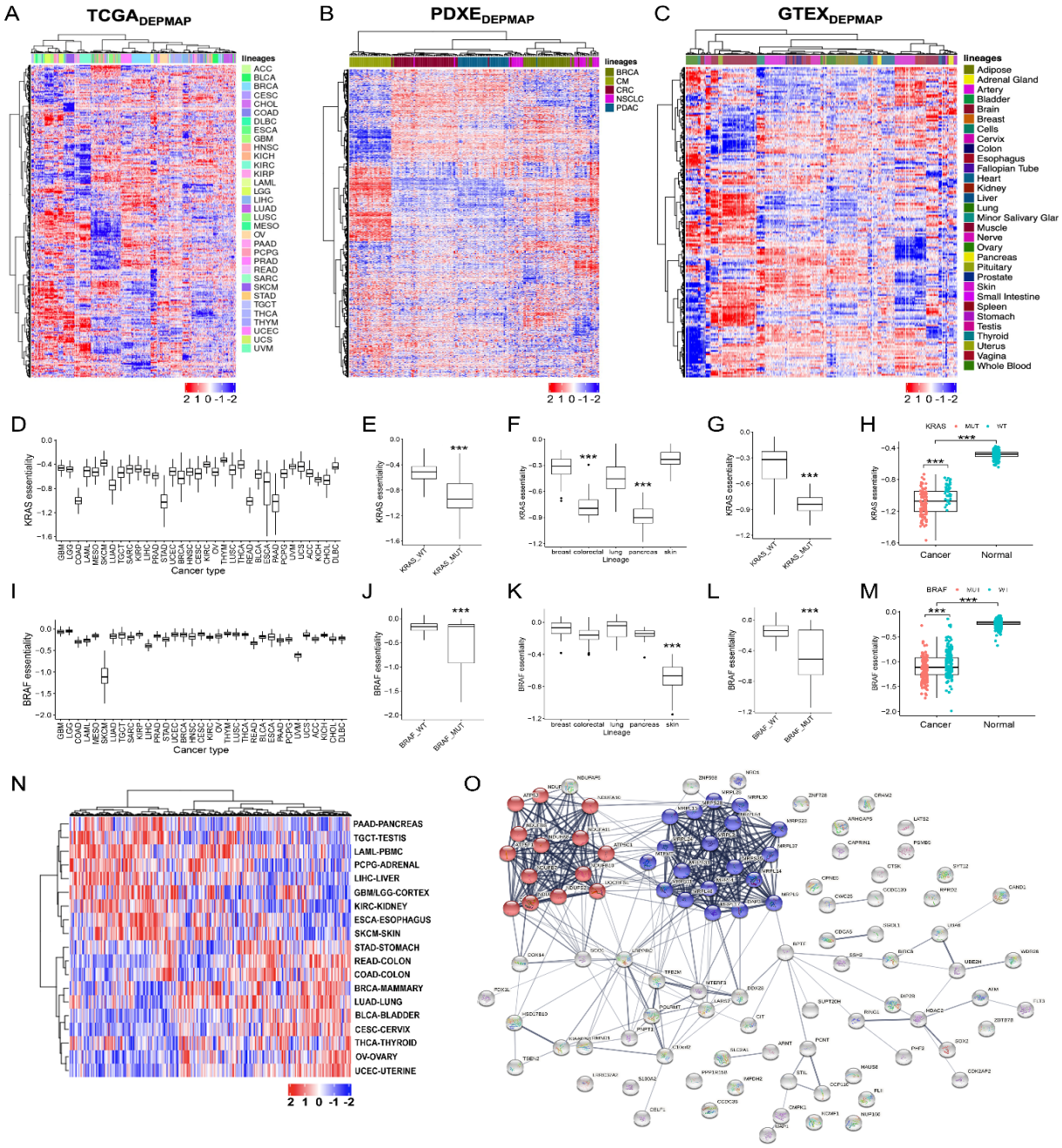

Figure 2. Characteristics of the translational dependency maps: (**A**) TCGA$_{DEPMAP}$, (**B**) PDXE$_{DEPMAP}$, and (**C**) GTEX$_{DEPMAP}$. (**D, E**) Oncogenic KRAS dependency across TCGA$_{DEPMAP}$ and (**F, G**) PDXE$_{DEPMAP}$ tissues. (**H**) KRAS essentiality is significantly higher in PAAD with GOF mutations compared with normal pancreas in GTEX$_{DEPMAP}$. (**I, J**) Oncogenic BRAF dependency across TCGA$_{DEPMAP}$ and (**K, L**) PDXE$_{DEPMAP}$ tissues. (**M**) BRAF essentiality is significantly higher in SKCM with GOF mutations compared with normal skin GTEX$_{DEPMAP}$. (**N**) Global differences between the predicted target efficacy score (TCGA$_{DEPMAP}$) and the normal tissue-of-origin tolerability score (GTEX$_{DEPMAP}$). (**O**) STRING network analysis of the top 100 LUAD targets with the greatest predicted tolerability in normal lung reveals significant connectivity ($p<1x10^{-16}$) and gene ontology enrichment oxidative phosphorylation (blue; $p=5.8x10^{-11}$) and mitochondrial translation (red; $p=2.9x10^{-20}$). ***P < 0.001, as determined by the Wilcoxon rank sum test for two group comparison and Kruskal-Wallis followed by Wilcoxon rank sum test with multiple test correction for the multi-group comparison.

The DepMap has revealed a strong relationship of gene essentiality with tissue lineage and genetic drivers across cancer cell models (*8*, *24*, *32*). Likewise, unsupervised clustering of gene essentiality within the TCGA$_{DEPMAP}$ revealed striking lineage-specific enrichments of tumor dependencies (**Figure 2A** and **Table S2**) and this was confirmed in an orthogonal

translational dependency map generated using the PDX Encyclopedia (*27*) ($PDXE_{DEPMAP}$, **Figure 2B** and **Table S3**). Intriguingly, tissue-of-origin dependencies were also observed in normal organs, as demonstrated by applying the dependency models to GTEX (*29*) ($GTEX_{DEPMAP}$, **Figure 2C** and **Table S4**), suggesting that lineage-specific biological context also contributes to gene essentiality normal tissues. Genetic drivers of gene essentiality were also observed to predict dependencies in the $TCGA_{DEPMAP}$ and $PDXE_{DEPMAP}$, including well-known druggable oncogenes such as KRAS and BRAF (**Figure 2D-M**). For example, KRAS essentiality was markedly stronger in KRAS-mutant STAD, READ, PAAD, and COAD lineages (**Figure 2D-H**), whereas BRAF essentiality was strongest in BRAF-mutant SKCM (**Figure 2I-M**). Notably, KRAS and BRAF essentialities appear to be concomitantly dependent on lineage and genetic drivers, as the normal tissues-of-origin were predicted to be significantly less affected in the $GTEX_{DEPMAP}$ compared with $TCGA_{DEPMAP}$ (**Figure 2H, M**). Likewise, similar observations were made for other oncogenic drivers that are approved therapeutic targets in cancer patients, such as ERBB2 amplified breast cancer (**Figure S1**). In contrast, there was markedly less separation in the predicted essentialities of malignant tumors and normal tissues-of-origin for molecular therapies that have yet to be successful in clinical trials (**Table S2** and **Table S4**). To refine the list of oncogenic pathways with significant differences in tumor efficacy and normal tissue-of-origin tolerability, we compared dependency ($TCGA_{DEPMAP}$) and tolerability ($GTEX_{DEPMAP}$) scores across all genes and tissues (**Figure 2N**). Pathway analysis of the strongest tumor dependencies with the least tissue-of-origin toxicity revealed enrichment of multiple oncogenic pathways and pathophysiological processes (**Table S5**), including dysregulation of oxidative phosphorylation ($p=5.8x10^{-11}$) and mitochondrial translation ($p=2.9x10^{-20}$) pathways that were enriched in LUAD compared with normal lung (**Figure 2O**). Combined, these observations suggest that predicted gene essentiality in the context of a driver mutation and correspondingly low essentiality within the normal tissue-of-origin is likely to identify efficacious drug targets with acceptable tolerability.

Predicting synthetic lethalities using translational dependency maps

In addition to illuminating lineage and oncogenic dependencies, the DEPMAP has dramatically expanded the list of potential synthetic lethalities (i.e., the loss of a gene sensitizes tumor cells to inhibition of a functional redundant gene within the same pathway) (*6*, *16*, *17*, *33*, *34*). However, one of the current limitations of DEPMAP is that the available cancer cell models do not yet fully recapitulate the genetic and molecular diversity of TCGA patients (*25*). Thus, we assessed the landscape of predicted synthetic lethalities with loss-of-function (LOF) events (damaging mutations or deletions) in the $TCGA_{DEPMAP}$ using several multivariate analyses to predict lethal partners. Unsurprisingly, the expanded repertoire of patient relevant LOF events in $TCGA_{DEPMAP}$ revealed 186 synthetic lethality candidates, in addition to the 167 candidate pairs that were readily detectable in the DEPMAP. Notably, both analyses identified known synthetic lethal pairs (e.g., STAG1/2, SMARCA2/4, EP300/CREBBP, etc.) (*35–37*) (**Figure 3A-B** and **Figure S2**), indicating that the DEPMAP and $TCGA_{DEPMAP}$ are both well-powered to detect synthetic lethal relationships with LOF events.

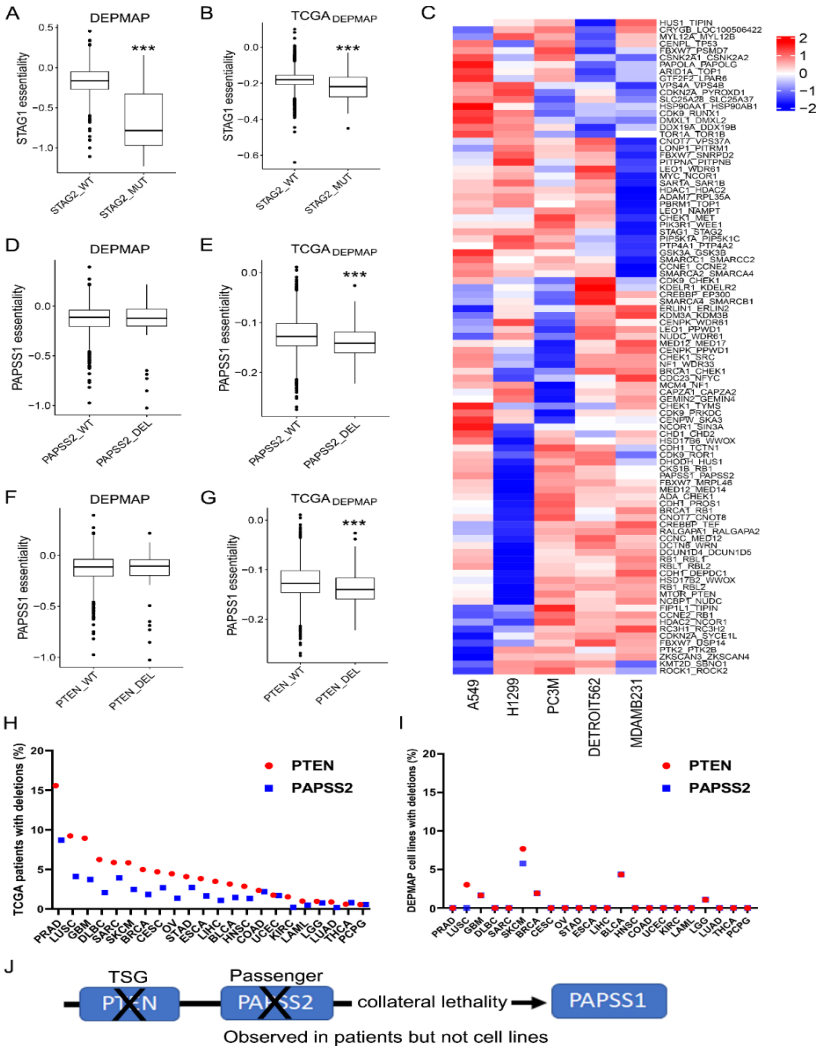

Figure 3. Predicting synthetic lethalities in the DEPMAP and TCGA$_{DEPMAP}$. STAG1 synthetic lethality with STAG2 mutation is a well-known synthetic lethality that is detected by the DEPMAP (**A**) and TCGA$_{DEPMAP}$ (**B**). (**C**) Experimental confirmation of PAPSS1/2 synthetic lethalities, as well as 89 other synthetic lethalities (Z-score < -2) that were tested by multiplexed CRISPR screening using AsCas12a. PAPSS1 is a novel synthetic lethality in the context of PAPSS2 deletion, which is not detectable in DEPMAP cell lines (**D**) and is only detectable in TCGA$_{DEPMAP}$ patient samples (**E**). Likewise, PAPSS1 is not synthetic lethal with PTEN deletion in DEPMAP cell lines (**F**) and is only detectable in TCGA$_{DEPMAP}$ patient samples (**G**). (**H**) PAPSS2 is frequently co-deleted with PTEN in patient samples; however, (**I**) PAPSS2 is rarely co-deleted with PTEN in cancer cell lines. (**J**) A schematic representation summarizing the hypothesized synthetic lethality of PAPSS1 that is driven by collateral deletion of PAPSS2 with the tumor suppressor gene (TSG), PTEN, in patients but not cell lines. ***P < 0.001, as determined by the Wilcoxon rank sum test.

Synthetic lethalities that were predicted with LOF events in DEPMAP (n=167) and TCGA$_{DEPMAP}$ (n=186) were experimentally validated by multiplexed CRISPR screening using AsCas12a (i.e., AsCpf1) (**Figure 3C**). Additionally, protein-protein interacting (PPI) members of underrepresented LOF were included (n=168), as were previously published paralog synthetic lethalities (n=360) (*6*, *16*, *17*, *33*, *34*). Of the 881 candidates, 90 synthetic lethalities were confirmed in at least one of the cell lines tested (A549, NCIH1299, PC3M, DETROIT562, and MDA-MB-231) (**Figure 3C**). As reported elsewhere (*33*, *35*, *38*), the sensitivity to synthetic lethalities varied broadly between cell models (**Figure 3C**), implicating the prevalence of unknown modifiers of synthetic lethality that manifest in different cellular contexts. The list of validated synthetic lethalities included paralog controls (e.g., HSP90AA1/B1 (*39*), DDX19A/DDX19B (*39*), HDAC1/2 (*38*, *39*), etc.), known synthetic lethalities (e.g., SMARCA2/4 (*38*, *39*), EP300/CREBBP (*37*), STAG1/2 (*36*, *38*), etc.), and novel synthetic lethality candidates that were only detectable in the TCGA$_{DEPMAP}$ (e.g., PAPSS1/2) (**Figure 3D-G**). For example, TCGA$_{DEPMAP}$ revealed appreciable incidence of deep deletion PAPSS2 (212 TCGA patients), whereas it was largely unaltered in the DepMap and therefore inaccessible for

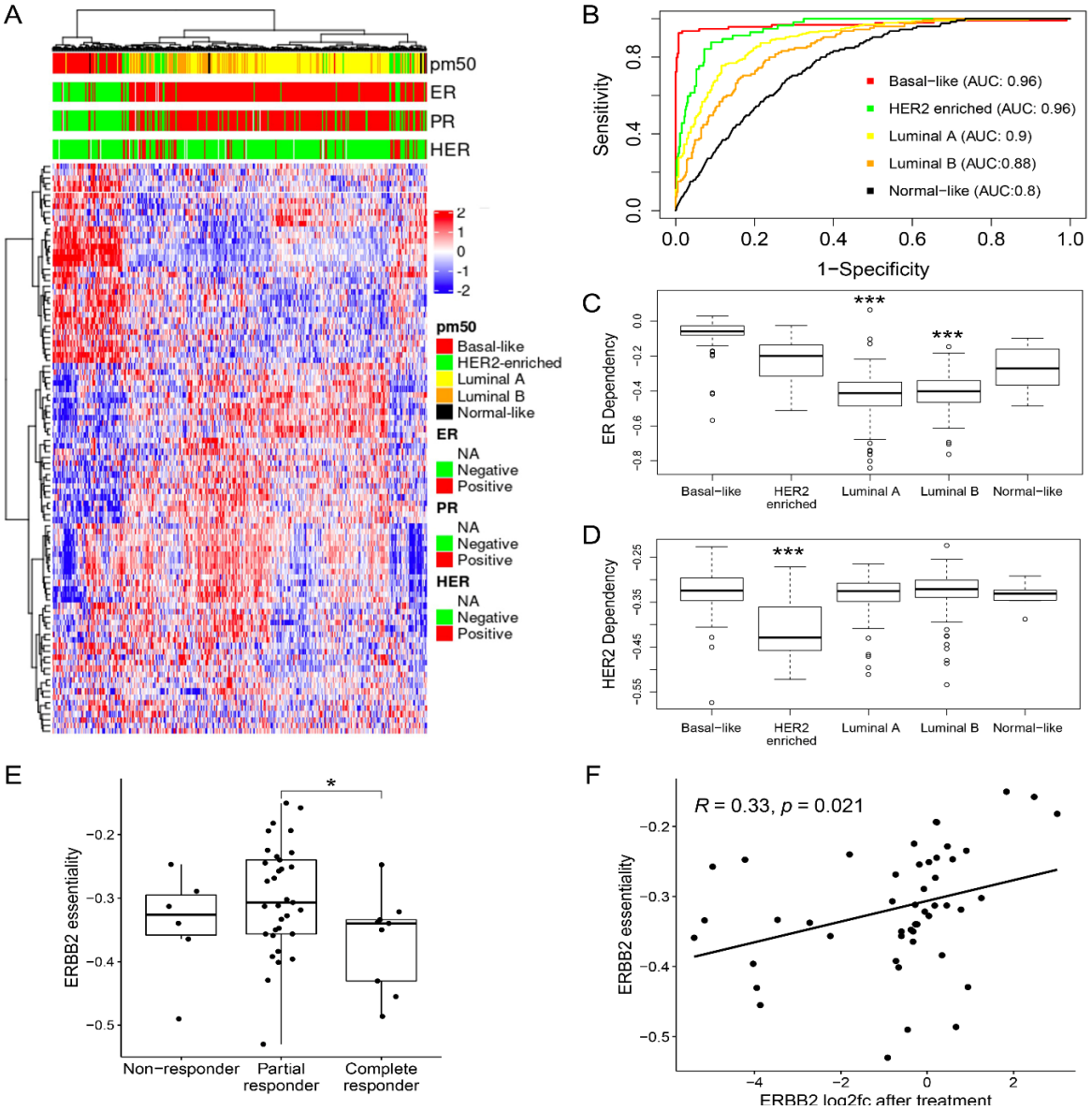

Figure 4. Translational dependency maps predict therapy response and outcome. (**A, B**) Subtyping breast cancer dependencies by the top 100 gene dependencies (DEP100). (**C**) ESR1 dependencies are strongest in ER-positive luminal breast cancer. (**D**) ERBB2 dependencies are strongest in HER2-amplified breast cancer. ERRB2 dependency predicts trastuzumab response (**E**) and correlates with ERBB2 expression change after treatment in patients (**F**). *$P < 0.05$ and ***$P < 0.001$, as determined by the Wilcoxon rank sum test for two group comparison and Kruskal-Wallis followed by Wilcoxon rank sum test with multiple test correction for the multi-group comparison. The normal-like group was excluded from the analysis because of small sample size.

detecting synthetic lethalities in cultured cancer cells (**Figure 3H-I**). Although it remains speculative as to why PAPPS2 is not frequently deleted in cultured cells (DEPMAP), it is more than likely driven in patients ($TCGA_{DEPMAP}$) by the higher prevalence of collateral deletion with the proximal tumor suppressor, PTEN (**Figure 3J**). In contrast, PTEN is predominantly silenced by LOF mutations and rarely deleted in cultured cancer cells, highlighting an example of genetic discrepancies between patient samples and cell lines that obscure synthetic lethal relationships.

Predicting drug response and outcomes using translational dependency maps

A strength of the translational tumor dependency maps is the ability to assess the impact of gene essentiality on clinically relevant phenotypes, such as molecular subtyping, therapeutic response, and patient outcomes. To evaluate the utility of $TCGA_{DEPMAP}$ for therapy-relevant patient stratification, an unsupervised clustering of the 100 most variable gene dependencies was performed using the $TCGA_{DEPMAP}$ breast cancer cohort (**Figure 4A**). The 100-dependency signature (DEP100) performed comparably to the established PAM50 signature (*40*) in classifying breast cancer subtypes (AUC close to 0.9 for most subtypes), despite only 3 overlapping genes between PAM50 and DEP100 (**Figure 4B**). As expected, dependency subtyping with DEP100 predicted significantly higher ESR1 essentiality in ER-positive tumors (**Figure 4C**) and higher ERBB2 essentiality in ERBB2-amplified tumors (**Figure 4D**). Finally, due to the limited accessibility of therapeutic response data in TCGA (*41*), we applied the predictive gene essentiality model for ERBB2 to a clinical trial of ERBB2-amplified breast cancer patients receiving trastuzumab (*42*), which revealed significantly higher predicted ERBB2 dependency in patients that responded better to trastuzumab (**Figure 4E, F**).

Another approach to assessing therapeutic drug response is through the $PDXE_{DEPMAP}$, which was built from a cohort of ~1,000 PDX models from 5 indications and treated with 62 therapies (*27*). To test the ability of gene essentiality to predict the response to corresponding targeted therapies, the change in PDX burden from baseline to experimental endpoint was correlated with target gene essentiality. This revealed that 80% of drugs (12 of 15) were significantly correlated ($p<0.05$) with the predicted essentiality of the target gene (**Table 1**). Notably, trastuzumab response in the $PDXE_{DEPMAP}$ was strongly predicted by ERBB2 dependency ($R=0.4849$, $p=0.002$), in line with the predictive power of ERBB2 dependency on trastuzumab responsiveness of in patients with ERBB2-amplified breast cancer (**Figure 4E, F**). Collectively, these data demonstrate the ability of gene essentiality to predict therapeutic response and highlight the translatability of PDX modeling to patient-relevant clinical outcomes.

Table 1. Correlation of gene essentiality and drug response in $PDXE_{DEPM}$

Drug	Treatment type	Drug Target	Predicted essentiality correlation with drug response (p-value)
erlotinib	Single target	EGFR	0.4937 (0.01213)
trastuzumab	Single target	ERBB2	0.4849 (0.002351)
BYL719	Single target	PIK3CA	0.3627 (9.881e-06)
tamoxifen	Single target	ESR1	0.2839 (0.0841)
cetuximab	Single target	EGFR	0.2293 (0.064)
figitumumab	Single target	IGF1R	0.2288 (0.1731)
HDM201	Single target	MDM2	0.1995 (0.01942)
LEE011 + everolimus	Multi-target	CDK4	0.6038 (7.59e-05)
BKM120 + binimetinib	Multi-target	PIK3CA	0.4357 (5.02e-4)
LJM716 + trastuzumab	Multi-target	ERBB2	0.4665 (3.61e-3)
CLR457	Multi-target	PIK3CA	0.2018 (0.0107)
BYL719 + LJM716	Multi-target	PIK3CA	0.215018 (0.0126)
LEE011 + encorafenib	Multi-target	CDK6	0.370181 (0.0370)
BKM120	Multi-target	PIK3CA	0.155772 (0.0425)
LEE011	Multi-target	CDK4	0.149998 (0.0495)

DISCUSSION

This study addresses the significant challenge of translating gene essentiality from cell-based dependency maps to the rapidly expanding catalogue of patient-relevant datasets. The translational dependency maps (i.e., $TCGA_{DEPMAP}$, $PDXE_{DEPMAP}$, and $GTEX_{DEPMAP}$) were built using expression-based predictive models of gene essentiality, as transcriptomic data reportedly outperforms other features (e.g., somatic mutations, copy number, etc.) in predicting dependencies (*25*, *43*). Another strength of expression-based predictive modeling is that it can be applied to the transcriptomic profiles of normal tissues (e.g., $GTEX_{DEPMAP}$) that do not have appreciable levels of the somatic alterations that are observed in malignant tissues (*29*). Moreover, transcriptomics is frequently the most widely captured dataset for many largescale clinical studies (*44*), suggesting that the methods used here will be widely applicable across many studies. Indeed, the same expression-based predictive models recapitulated a similar dependency profile in $TCGA_{DEPMAP}$ and $PDXE_{DEPMAP}$ across different cancer lineages and genetic drivers. Both maps also accurately predicted therapy-relevant molecular subtypes, therapeutic responses, and disease outcomes, whereas $GTEX_{DEPMAP}$ provided novel insights to normal tissue-of-origin tolerability. Collectively, these translational dependency maps offer clinically relevant aspects to gene essentiality that are not currently accessible in the traditional cell-based dependency maps.

Translating tumor dependencies using $TCGA_{DEPMAP}$ and $PDXE_{DEPMAP}$

A challenge of cell-based dependency maps (e.g., DEPMAP) is the inability to fully recapitulate patient genomics, therapeutic responses, and many aspects of disease outcomes and patient survival (*41*). Using $TCGA_{DEPMAP}$ to survey the landscape of translational dependencies in cancer patients, we observed a strong correlation between patient tumor type and corresponding cancer cell line lineages (**Figure 1F-G** and **Figure 2A**). A comparison of predictive dependency models across $TCGA_{DEPMAP}$ (**Figure 2A**) and $PDXE_{DEPMAP}$ (**Figure 2B**) revealed that dependencies accurately predict tumor lineages (AUC=0.75), fitting with observations made in TCGA that tissue-of-origin dominates the molecular landscape of cancer (*45*). In addition to lineage dependencies, $TCGA_{DEPMAP}$ and $PDXE_{DEPMAP}$ also demonstrated the abilities of predicted dependencies to define therapy relevant molecular subtypes, including the ER-positive luminal breast cancer (**Figure 4A-C**) and ERBB2-amplified breast cancer (**Figure 4A-B, D**). Moreover, $PDXE_{DEPMAP}$ demonstrated the ability of gene essentiality to predict the responses to corresponding targeted therapies, with a significant correlation between the predicted essentiality of the molecular target and therapeutic response in 80% of the cases (**Table 1**). Thus, translational dependency maps combined with the expanding catalogue of patient genomics (*3*, *26*) and advances in molecular therapeutics (*4*) are expected to provide novel molecular targets for patient-relevant therapies.

In addition to cancer lineage and subtype dependencies, $TCGA_{DEPMAP}$ provided novel molecular insight to the genetic drivers of gene essentiality in patient populations. Notably, well-defined oncogenic drivers of tumor dependencies (e.g., KRAS and BRAF) were highly predictable in $TCGA_{DEPMAP}$ and $PDXE_{DEPMAP}$ (**Figure 2D-M**), as would be expected based on the predictive response rates for molecular therapeutics targeting oncogenic drivers in patients (*46*). Multiple known synthetic lethalities were also detected in $TCGA_{DEPMAP}$ (STAG1/2, SMARCA2/4, and EP300/CREBBP) (*36*–*39*), as well as previously unappreciated candidate pairs that have not been reported elsewhere (e.g., PAPSS1/2). Intriguingly, PAPSS2 deletion is prevalent in the TCGA, yet deletions of this genetic loci was largely absent from DEPMAP cell lines. We hypothesize that loss of PAPSS2 is likely driven by its proximity to PTEN and is an example of collateral deletion in patient tumors (*47*). Since PTEN is largely silenced by LOF mutations in cancer cell lines and not deletion, a synthetic lethality between PAPPS1 and endogenous collateral deletion of PAPSS2 with PTEN is undetectable in cell-based dependency maps (**Figure 3F**). Thus, the observation of broader discrepancies of genetic drivers in cancer cell models compared with patient populations (*22*), highlights the benefits of translational dependency maps that closely match the diversity of patient genomic datasets (*1*, *26*).

Translating tolerability of dependencies using $TCGA_{DEPMAP}$ and $GTEX_{DEPMAP}$

As the landscape of gene essentiality continues to expand, a better understanding of the impact on normal tissues to predict tolerability is needed. To address this challenge, we applied the expression-based dependency models to GTEX, a compendium of deeply-phenotyped normal tissues collected from postmortem healthy donors (*29*). Strikingly, unsupervised clustering of the predicted gene essentialities across 17,382 normal tissues in the $GTEX_{DEPMAP}$ demonstrated a significant impact of tissue-of-origin on gene dependencies (**Figure 2C** and **Table S4**). One potential reason is that pathways required for maintaining normal tissue homeostasis are frequently co-opted for oncogenesis (*48*) and several such pathways serve as key therapeutic targets for cancer (*49*). In addition to lineage pathways, most successful cancer therapeutics are also targeted towards somatic drivers, which are absent from normal tissues and therefore likely to be more tolerable (*48*, *49*). A unique aspect of this study was the ability to systematically compare gene essentiality associated with somatic mutations in $TCGA_{DEPMAP}$ with the normal tissue-of-origin tolerability profiles in $GTEX_{DEPMAP}$. Strong oncogenic dependencies with acceptable tolerability (e.g., KRAS and BRAF) (*48*, *49*) revealed marked differences between predicted gene essentiality in malignant and normal tissues, and this therapeutic window was further widened in the presence of the oncogenic mutation (**Figure 2H**, **M**). Systematically expanding this analysis across all gene essentiality models in $TCGA_{DEPMAP}$ and $GTEX_{DEPMAP}$ revealed wide variability in the predicted tolerability windows, implicating the existence of other dependencies with strong genetic drivers that are likely to be more tolerable as therapeutic targets. However, we also recommend exercising caution when interpreting these data, as the tolerability window predicted by comparing tissue-of-origin gene essentiality between $TCGA_{DEPMAP}$ and $GTEX_{DEPMAP}$ likely does not fully capture the dose-limiting toxicities (DLT) that pose challenges to clinical drug development (*50*).

Comparison of $TCGA_{DEPMAP}$ with DeepDEP

During the completion of this study, Chiu et al (*25*) took a complimentary approach to building a translational dependency map (DeepDEP) using deep learning (DL) from the integrative genomic, epigenomic, and transcriptomic profiles of TCGA patients and DEPMAP cell lines. Here, we used elastic-net regularized regression models of expression data for predicting gene essentiality and tolerability, as these expression-based models can be applied to malignant tissue ($TCGA_{DEPMAP}$ and $PDXE_{DEPMAP}$) and nonmalignant tissues ($GTEX_{DEPMAP}$), respectively. Our rationale for using elastic-net ML is that it is suited to highly dimensional data with correlated variables (*30*) and expression-based predictive modeling of gene essentiality outperforms genomic features (e.g., somatic mutations, copy number, methylation, etc.) (*25*, *43*). Likewise, the DeepDEP authors also highlighted that a simplified DL model using expression only (Exp-DeepDEP) performed comparably well to DeepDEP (*25*), suggesting that ML and DL dependency models are both dominated by expression profiling (*25*). Intriguingly, despite the dominance of expression in the DeepDEP dependency models, it was unable to recapitulate the robust clustering of tumors and cell lines by t-SNE that was observed by expression alone (*25*). One potential explanation is the liability of DL models to overfit highly dimensional data (e.g., transcriptomics) (*51*), which was potentially compounded by the unlabeled pretraining stage of DeepDEP with TCGA expression data that included expression profiles of nonmalignant stroma (*25*). Warren et al (*52*) recently used cPCA to remove the expression signatures with the greatest intra-cluster variance to significantly improve alignment of tumors and cell lines. Notably, the alignment of $TCGA_{DEPMAP}$ and DEPMAP also significantly improved by removal of the most variant signatures (cPC1-4; i.e., stromal signatures (*52*)) before elastic net ML (**Figure S3**), confirming that contaminating tumor stromal signatures likely confound the accuracy of dependency models built from cell lines.

Due to the distinctions of ML and DL models, we sought to compare the sensitivity and specificity of dependency models from $TCGA_{DEPMAP}$ and DeepDEP. For lack of ground truth, we compared the predicted tumor dependencies of $TCGA_{DEPMAP}$ and DeepDEP by pan-cancer lineage and breast cancer subtypes, as these are clearly defined by TCGA and DEPMAP. Compared with DeepDEP, the predicted dependencies by $TCGA_{DEPMAP}$ were significantly more accurate in identifying cancer lineage (**Figure S4A**), fitting with the observations that tissue-of-origin is a key molecular driver of tumor lineage classification (*45*) and dependencies (*9*). Likewise, the precision-recall of TCGA breast cancer subtypes by $TCGA_{DEPMAP}$ was superior to the predictive accuracy of DeepDEP (**Figure S4B**). Collectively, these data suggest that the expression-based predictive models of gene essentiality and tolerability are both accurate and flexible for building translational dependency maps across malignant ($TCGA_{DEPMAP}$ and $PDXE_{DEPMAP}$) and nonmalignant ($GTEX_{DEPMAP}$) tissues.

Future considerations for translational dependency maps

The translational dependency maps presented in this study ($TCGA_{DEPMAP}$, $PDXE_{DEPMAP}$, and $GTEX_{DEPMAP}$) provide novel insights to gene essentiality and tolerability in the clinical context of patient tumors and normal tissues. The ability of these maps to accurately translate

dependencies to patients is reliant on the ability to build predictive models from cell-based mapping, which is still at the early stages (~1,000 mapped cell lines) and is expected to require 20X more data (~20,000 mapped cell lines) to fully predict gene essentiality (*7*). Further, the observations that cell-based dependencies vary between 2D and 3D settings (*53*) and are impacted by crosstalk with the TME (*54*), suggests that gene essentiality is contextual and requires models with greater relevance to intact tumors. To reach the full potential of translational dependency mapping, the catalog of patient genomic datasets will also likely require expansion to capture various stages of disease progression, including tumorigenesis (*2*), metastasis (*3*, *55*), and therapeutic resistance (*3*, *4*, *55*). Furthermore, as precision cancer clinical trials continue to expand (e.g., MSK-IMPACT) (*4*), it will be increasingly possible to refine translational dependency maps by testing outcomes of molecular therapeutics with predicted target essentiality. The utility of translational "tolerability" maps in normal tissues (e.g., $GTEX_{DEPMAP}$) remains to be fully explored and will likely benefit from further refinements to better capture aspects of DLT that impact drug development. To this end, we postulate that modeling gene tolerability could be best assessed in normal cell types by pairing CRISPR perturbations with single-cell RNA sequencing (*56*, *57*) to broadly capture the alterations of pathways required for normal tissue homeostasis. Ultimately, we postulate that predictive modeling of dependency and tolerability in patients will increase the success of drug discovery by preemptively prioritizing targets with the best therapeutic index (i.e., high dependency and tolerability).

MATERIALS AND METHODS

Gene expression normalization between tissues and cell lines

Gene expression data from multiple batches or resources are usually contaminated with batch effect and other noise. To evaluate the similarity between the gene expression data of TCGA and DEPMAP, we performed an unsupervised clustering analysis based on UMAP dimension reduction. As reported previously (*52*), homogenous cell lines (DEPMAP) and clinical samples (TCGA) do not cluster well by UMAP alignment due to contaminating transcriptional profiles of stromal and immune cells and therefore likely to impact expression-based predictive modeling of gene essentiality. To overcome this issue, expression data from DEPMAP and TCGA were quantile normalized and transformed by contrastive principal component analysis (cPCA) to identify misaligned components derived from the stromal contamination of the TCGA. The top contrastive principal components (cPC1-4) were removed, followed by multiple-batch correction to normalize the expression data by matching the corresponding clusters in TCGA and DEPMAP. An identical approach was utilized for normalization of PDXE expression data, with the slight modification that only cPC1-3 required removal, as PDX models grown in immunocompromised mice lack the adaptive immune system and typically have lower stromal contamination. For GTEX data, we utilized a different approach by combining quantile normalization and ComBat (*58*) to remove potential batch effect since GTEX data only includes normal tissues.

Expression based predictive modeling of gene essentiality

Expression-based predictive models were built for 6,581 genes (excluding pan-essential and non-essential genes) using the dependency data and normalized transcriptomic profiles of 791 DEPMAP cell lines (1Q21 release). A gene was defined as pan-essential or non-essential, as described previously (*24*). The *'glmnet'* package (*30*) was used to build elastic-net regularized regression models with balanced weights for L1 and L2 norm regularization. Models were 10-fold cross-validated to avoid over-fitting, followed by selection of the model with minimum error for each gene essentiality. Of the 6,581 expression-based predictive essentiality models for genes were attempted (excluding pan-essential and non-essential genes), of which 1,966 models cross-validated using 10-fold cross-validation (R>0.2). All the models consisted of a set of genes and the corresponding coefficients learned for the DEPMAP data. The cross-validated gene essentiality models were then applied to the normalized transcriptomes of TCGA (N = 9,596 patients), PDXE (N=191), and GTEX (N=17,382 tissues) to predict gene essentiality and tolerability within the cancer and normal tissue translational datasets, respectively.

TCGA pan-cancer lineage typing and breast cancer subtyping by $TCGA_{DEPMAP}$

The top 100 genes with the most variable dependency profiles across the TCGA pan-cancer cohort and within the breast cancer cohort (DEP100) were defined by the gene essentialities with the widest variance across the respective $TCGA_{DEPMAP}$ cohorts. A linear discriminant analysis using the lda with leave-one-out cross validation were performed in R with the CV=TRUE option in the function. Predictions for each cancer type and subtype was evaluated separately and the AUC values were determined using the function roc from the pROC (version 1.18.0) package and compared with the molecular typing and subtyping reported by the TCGA (https://www.cbioportal.org/) (*59*). An identical analysis was performed using DeepDEP gene essentiality values reported by Chiu et al (*25*) and AUC values receiver operating characteristics (ROC) were compared between $TCGA_{DEPMAP}$ and DeepDEP predictions of cancer lineages and breast cancer subtypes (**Figure S4**).

Differential dependency analysis of $TCGA_{DEPMAP}$ and $GTEX_{DEPMAP}$

TCGA and GTEX samples were matched based on the tissue-of-origin and a Student's t-test was applied to differential analysis between the dependency profiles of tumor and normal tissue of the same lineages. The t-statistic was used to characterize the dependency difference between the tumor and corresponding normal tissue with a negative t-statistic value corresponding to a higher dependency in tumor as compared with the normal tissue. The list of genes for the lung network was generated using the top 100 genes showing the largest differentiation in gene essentiality between cancer compared to normal tissue in lung based on the negative t-statistic values. Network connectivity and the gene ontology enrichment were calculated using STRING (https://string-db.org/), as described previously (*60*).

Predicting synthetic lethality relationships in the DEPMAP and $TCGA_{DEPMAP}$

Multiple approaches were used to predict synthetic lethality relationships with loss-of-function (LOF) events (defined as a predicted copy number loss or damaging mutation) in the DEPMAP and TCGA datasets. The first approach used UNCOVER (*61*) to identify essentialities that associated with LOF mutations ranging from 3% to 70% prevalence. The UNCOVER program was ran in greedy mode (UNCOVER_greedyv2.py) to identify negative association with a mutated gene sets of maximum 10 genes. To evaluate the confidence of association, we set the number of permutations as 100 to compute p-values and applied a threshold of $p<0.01$. A second approach used DAMOKLE (*62*) to identify differentially mutated subnetworks of protein-protein interacting complexes, as defined by the STRING database (https://string-db.org/) (*60*), and with a significance threshold of $p<0.05$. Gene essentiality scores from the DEPMAP and $TCGA_{DEPMAP}$ were then associated with the differentially mutated sub-networks. The Lasso regression was performed to predict the essentiality based on damaging mutation, deletion events, or a combination of both. The genes with copy number less than 1.2 were considered as deletion. We further filtered the genes with less than 5% deletion rate or 2% mutation rate in the DEPMAP and TCGA cohorts. The model was trained with lambda value selected as the lowest error by 5-fold cross-validation and the resulting models with coefficients >0.3 were further evaluated by t-test. For $TCGA_{DEPMAP}$, an additional filtering step based on the paralog information from the biomaRt (version 2.28.0) R package with the addition of synthetic lethality candidates characterized by small phylogenetic distance (with threshold of distance less than 1.5), as described previously (*63*, *64*). The final list of synthetic lethality candidates was then filtered for genes essentialities that are significantly associated ($p<0.01$) with a LOF event in the DEPMAP or $TCGA_{DEPMAP}$.

Multiplexed screening synthetic lethalities using AsCas12a (AsCpf1)

Guides were designed using the TTTV PAM for AsCas12a and synthesized into 4-guide arrays with direct repeats (DR)-1, -2, -3, and -4 preceding each guide, followed by cloning into a guide-only lentiviral vector (pRDA_052), as described previously (*38*, *39*). A double knockout construct was designed with 2 guides x 2 genes (n = 4 guides total per construct) for each pair of synthetic lethal candidates. A single knockout construct was also designed 2 guides x 1 gene + 2 non-targeting guides (n= 4 guides total per construct) for each pair of synthetic lethal candidates. A total of 500 constructs with 4 non-targeting guides were also included in the library as negative controls. The final library of synthetic lethal candidates includes 186 identified from the DEPMAP, 167 additionally identified from the $TCGA_{DEPMAP}$, 168 identified from protein-protein interaction (PPI) networks (*65*, *66*), and 360 paralog pairs nominated from the literature (*6*, *16*, *17*, *33*, *34*) (**Table S6**). Screens were performed in triplicate using A549, NCI-H1299, HSC2, MDA-MB-231, NCI-H1703, PC3M, and DETROIT562 that stably express AsCas12a, as described previously (*38*). Briefly, cells were infected at a multiplicity of infection (MOI) = 0.3 and cultured for 14 days while continuously maintaining 500X coverage, followed by DNA extraction and PCR-barcoding using the p5 Argon and p7 Kermit primers (*38*). The PCR-barcoded libraries were single-end sequenced using an Illumina HiSeq4000 (300X

cycle), followed by demultiplexing of sequencing reads (bcl2fastq, Illumina) and quantification of guide array abundance across all samples was done with a custom Perl script. Briefly, users either specify the position in a read containing the sgRNA or the flanking primer sequence (if sgRNA position is not constant in all reads). Sequences between the flanking sequences or by location were extracted and compared to a database of sgRNA for each library. Only perfectly matched sgRNA sequences were kept and used in the generation of count matrix. Normalization between all samples was done using the "TMM" method (*67*) implemented in the edgeR R Bioconductor package. Log2 fold-changes (L2FC) of guide array abundance were calculated by comparing day 14 libraries with the plasmid library using limma-voom (*68*). Genetic interactions (GI) were calculated by comparing the expected and observed L2FC of double and single knockout constructs, as described previously (*33*, *39*). Briefly, the expected L2FC for double knockout constructs is calculated as a sum (LF2C) of the individual knockout (sgRNA + Nontargeting). Synthetic lethal and buffering interactions are defined for double knockout in which the observed double knockout L2FC is significantly greater or less than that of the expected L2FC, respectively.

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REFERENCES

1. C. Hutter, J. C. Zenklusen, The Cancer Genome Atlas: Creating Lasting Value beyond Its Data. *Cell*. **173**, 283–285 (2018).
2. S. Srivastava, S. Ghosh, J. Kagan, R. Mazurchuk, The PreCancer Atlas (PCA). *Trends in Cancer*. **4**, 513–514 (2018).
3. Tracing evolution reveals new biomarkers | Nature Reviews Clinical Oncology, (available at https://www.nature.com/articles/s41571-019-0295-0).
4. A. Zehir, R. Benayed, R. H. Shah, A. Syed, S. Middha, H. R. Kim, P. Srinivasan, J. Gao, D. Chakravarty, S. M. Devlin, M. D. Hellmann, D. A. Barron, A. M. Schram, M. Hameed, S. Dogan, D. S. Ross, J. F. Hechtman, D. F. DeLair, J. Yao, D. L. Mandelker, D. T. Cheng, R. Chandramohan, A. S. Mohanty, R. N. Ptashkin, G. Jayakumaran, M. Prasad, M. H. Syed, A. B. Rema, Z. Y. Liu, K. Nafa, L. Borsu, J. Sadowska, J. Casanova, R. Bacares, I. J. Kiecka, A. Razumova, J. B. Son, L. Stewart, T. Baldi, K. A. Mullaney, H. Al-Ahmadie, E. Vakiani, A. A. Abeshouse, A. V. Penson, P. Jonsson, N. Camacho, M. T. Chang, H. H. Won, B. E. Gross, R. Kundra, Z. J. Heins, H.-W. Chen, S. Phillips, H. Zhang, J. Wang, A. Ochoa, J. Wills, M. Eubank, S. B. Thomas, S. M. Gardos, D. N. Reales, J. Galle, R. Durany, R. Cambria, W. Abida, A. Cercek, D. R. Feldman, M. M. Gounder, A. A. Hakimi, J. J. Harding, G. Iyer, Y. Y. Janjigian, E. J. Jordan, C. M. Kelly, M. A. Lowery, L. G. T. Morris, A. M. Omuro, N. Raj, P. Razavi, A. N. Shoushtari, N. Shukla, T. E. Soumerai, A. M. Varghese, R. Yaeger, J. Coleman, B. Bochner, G. J. Riely, L. B. Saltz, H. I. Scher, P. J. Sabbatini, M. E. Robson, D. S. Klimstra, B. S. Taylor, J. Baselga, N. Schultz, D. M. Hyman, M. E. Arcila, D. B. Solit, M. Ladanyi, M. F. Berger, Mutational landscape of metastatic cancer revealed from prospective clinical sequencing of 10,000 patients. *Nat Med*. **23**, 703–713 (2017).
5. J. van de Haar, L. R. Hoes, P. Roepman, M. P. Lolkema, H. M. W. Verheul, H. Gelderblom, A. J. de Langen, E. F. Smit, E. Cuppen, L. F. A. Wessels, E. E. Voest, Limited evolution of the actionable metastatic cancer genome under therapeutic pressure. *Nat Med*. **27**, 1553–1563 (2021).
6. C. J. Lord, N. Quinn, C. J. Ryan, Integrative analysis of large-scale loss-of-function screens identifies robust cancer-associated genetic interactions. *Elife*. **9**, e58925 (2020).

7. J. S. Boehm, M. J. Garnett, D. J. Adams, H. E. Francies, T. R. Golub, W. C. Hahn, F. Iorio, J. M. McFarland, L. Parts, F. Vazquez, Cancer research needs a better map. *Nature*. **589**, 514–516 (2021).
8. A. Tsherniak, F. Vazquez, P. G. Montgomery, B. A. Weir, G. Kryukov, G. S. Cowley, S. Gill, W. F. Harrington, S. Pantel, J. M. Krill-Burger, R. M. Meyers, L. Ali, A. Goodale, Y. Lee, G. Jiang, J. Hsiao, W. F. J. Gerath, S. Howell, E. Merkel, M. Ghandi, L. A. Garraway, D. E. Root, T. R. Golub, J. S. Boehm, W. C. Hahn, Defining a Cancer Dependency Map. *Cell*. **170**, 564-576.e16 (2017).
9. F. M. Behan, F. Iorio, G. Picco, E. Gonçalves, C. M. Beaver, G. Migliardi, R. Santos, Y. Rao, F. Sassi, M. Pinnelli, R. Ansari, S. Harper, D. A. Jackson, R. McRae, R. Pooley, P. Wilkinson, D. van der Meer, D. Dow, C. Buser-Doepner, A. Bertotti, L. Trusolino, E. A. Stronach, J. Saez-Rodriguez, K. Yusa, M. J. Garnett, Prioritization of cancer therapeutic targets using CRISPR-Cas9 screens. *Nature*. **568**, 511–516 (2019).
10. A. Subramanian, R. Narayan, S. M. Corsello, D. D. Peck, T. E. Natoli, X. Lu, J. Gould, J. F. Davis, A. A. Tubelli, J. K. Asiedu, D. L. Lahr, J. E. Hirschman, Z. Liu, M. Donahue, B. Julian, M. Khan, D. Wadden, I. C. Smith, D. Lam, A. Liberzon, C. Toder, M. Bagul, M. Orzechowski, O. M. Enache, F. Piccioni, S. A. Johnson, N. J. Lyons, A. H. Berger, A. F. Shamji, A. N. Brooks, A. Vrcic, C. Flynn, J. Rosains, D. Y. Takeda, R. Hu, D. Davison, J. Lamb, K. Ardlie, L. Hogstrom, P. Greenside, N. S. Gray, P. A. Clemons, S. Silver, X. Wu, W.-N. Zhao, W. Read-Button, X. Wu, S. J. Haggarty, L. V. Ronco, J. S. Boehm, S. L. Schreiber, J. G. Doench, J. A. Bittker, D. E. Root, B. Wong, T. R. Golub, A Next Generation Connectivity Map: L1000 Platform and the First 1,000,000 Profiles. *Cell*. **171**, 1437-1452.e17 (2017).
11. Next-generation characterization of the Cancer Cell Line Encyclopedia | Nature, (available at https://www.nature.com/articles/s41586-019-1186-3).
12. Y.-Y. Tseng, M. Al-Jazrawe, R. Deasy, P. Keskula, G. Johnson, A. Hong, P. Chatterji, F. Vasquez, A. Bass, B. V. Hare, D. Sandak, K. Ligon, J. Boehm, Abstract 3453: Cancer Cell Line Factory: A systematic approach to create next-generation cancer model at scale. *Cancer Res*. **80**, 3453–3453 (2020).
13. D. S. Gerhard, Abstract IA16: The International Human Cancer Models Initiative is generating models annotated with clinical and molecular data. *Cancer Res*. **80**, IA16–IA16 (2020).
14. R. Gillani, B. K. A. Seong, J. Crowdis, J. R. Conway, N. V. Dharia, S. Alimohamed, B. J. Haas, K. Han, J. Park, F. Dietlein, M. X. He, A. Imamovic, C. Ma, M. C. Bassik, J. S. Boehm, F. Vazquez, A. Gusev, D. Liu, K. A. Janeway, J. M. McFarland, K. Stegmaier, E. M. V. Allen, Gene Fusions Create Partner and Collateral Dependencies Essential to Cancer Cell Survival. *Cancer Res*. **81**, 3971–3984 (2021).
15. N. V. Dharia, G. Kugener, L. M. Guenther, C. F. Malone, A. D. Durbin, A. L. Hong, T. P. Howard, P. Bandopadhayay, C. S. Wechsler, I. Fung, A. C. Warren, J. M. Dempster, J. M. Krill-Burger, B. R. Paolella, P. Moh, N. Jha, A. Tang, P. Montgomery, J. S. Boehm, W. C. Hahn, C. W. M. Roberts, J. M. McFarland, A. Tsherniak, T. R. Golub, F. Vazquez, K.

Stegmaier, A first-generation pediatric cancer dependency map. *Nat Genet*. **53**, 529–538 (2021).

16. J. E. Neggers, B. R. Paolella, A. Asfaw, M. V. Rothberg, T. A. Skipper, A. Yang, R. L. Kalekar, J. M. Krill-Burger, N. V. Dharia, G. Kugener, J. Kalfon, C. Yuan, N. Dumont, A. Gonzalez, M. Abdusamad, Y. Y. Li, L. F. Spurr, W. W. Wu, A. D. Durbin, B. M. Wolpin, F. Piccioni, D. E. Root, J. S. Boehm, A. D. Cherniack, A. Tsherniak, A. L. Hong, W. C. Hahn, K. Stegmaier, T. R. Golub, F. Vazquez, A. J. Aguirre, Synthetic Lethal Interaction between the ESCRT Paralog Enzymes VPS4A and VPS4B in Cancers Harboring Loss of Chromosome 18q or 16q. *Cell Reports*. **33** (2020), doi:10.1016/j.celrep.2020.108493.
17. E. M. Chan, T. Shibue, J. M. McFarland, B. Gaeta, M. Ghandi, N. Dumont, A. Gonzalez, J. S. McPartlan, T. Li, Y. Zhang, J. Bin Liu, J.-B. Lazaro, P. Gu, C. G. Piett, A. Apffel, S. O. Ali, R. Deasy, P. Keskula, R. W. S. Ng, E. A. Roberts, E. Reznichenko, L. Leung, M. Alimova, M. Schenone, M. Islam, Y. E. Maruvka, Y. Liu, J. Roper, S. Raghavan, M. Giannakis, Y.-Y. Tseng, Z. D. Nagel, A. D'Andrea, D. E. Root, J. S. Boehm, G. Getz, S. Chang, T. R. Golub, A. Tsherniak, F. Vazquez, A. J. Bass, WRN helicase is a synthetic lethal target in microsatellite unstable cancers. *Nature*. **568**, 551–556 (2019).
18. Selective Modulation of a Pan-Essential Protein as a Therapeutic Strategy in Cancer | Cancer Discovery, (available at https://cancerdiscovery.aacrjournals.org/content/11/9/2282).
19. J. M. McFarland, Z. V. Ho, G. Kugener, J. M. Dempster, P. G. Montgomery, J. G. Bryan, J. M. Krill-Burger, T. M. Green, F. Vazquez, J. S. Boehm, T. R. Golub, W. C. Hahn, D. E. Root, A. Tsherniak, Improved estimation of cancer dependencies from large-scale RNAi screens using model-based normalization and data integration. *Nat Commun*. **9**, 4610 (2018).
20. J. M. Dempster, C. Pacini, S. Pantel, F. M. Behan, T. Green, J. Krill-Burger, C. M. Beaver, S. T. Younger, V. Zhivich, H. Najgebauer, F. Allen, E. Gonçalves, R. Shepherd, J. G. Doench, K. Yusa, F. Vazquez, L. Parts, J. S. Boehm, T. R. Golub, W. C. Hahn, D. E. Root, M. J. Garnett, A. Tsherniak, F. Iorio, Agreement between two large pan-cancer CRISPR-Cas9 gene dependency data sets. *Nat Commun*. **10**, 5817 (2019).
21. M. J. Flister, C. Bergom, Genetic Modifiers of the Breast Tumor Microenvironment. *Trends Cancer*. **4**, 429–444 (2018).
22. J. G. Tate, S. Bamford, H. C. Jubb, Z. Sondka, D. M. Beare, N. Bindal, H. Boutselakis, C. G. Cole, C. Creatore, E. Dawson, P. Fish, B. Harsha, C. Hathaway, S. C. Jupe, C. Y. Kok, K. Noble, L. Ponting, C. C. Ramshaw, C. E. Rye, H. E. Speedy, R. Stefancsik, S. L. Thompson, S. Wang, S. Ward, P. J. Campbell, S. A. Forbes, COSMIC: the Catalogue Of Somatic Mutations In Cancer. *Nucleic Acids Res*. **47**, D941–D947 (2019).
23. R. M. Meyers, J. G. Bryan, J. M. McFarland, B. A. Weir, A. E. Sizemore, H. Xu, N. V. Dharia, P. G. Montgomery, G. S. Cowley, S. Pantel, A. Goodale, Y. Lee, L. D. Ali, G. Jiang, R. Lubonja, W. F. Harrington, M. Strickland, T. Wu, D. C. Hawes, V. A. Zhivich, M. R. Wyatt, Z. Kalani, J. J. Chang, M. Okamoto, K. Stegmaier, T. R. Golub, J. S. Boehm, F. Vazquez, D. E. Root, W. C. Hahn, A. Tsherniak, Computational correction of copy number

effect improves specificity of CRISPR–Cas9 essentiality screens in cancer cells. *Nat Genet*. **49**, 1779–1784 (2017).

24. J. M. Dempster, J. Rossen, M. Kazachkova, J. Pan, G. Kugener, D. E. Root, A. Tsherniak, “Extracting Biological Insights from the Project Achilles Genome-Scale CRISPR Screens in Cancer Cell Lines” (2019), p. 720243.
25. Y.-C. Chiu, S. Zheng, L.-J. Wang, B. S. Iskra, M. K. Rao, P. J. Houghton, Y. Huang, Y. Chen, Predicting and characterizing a cancer dependency map of tumors with deep learning. *Sci Adv*. **7**, eabh1275 (2021).
26. P. J. Campbell, G. Getz, J. O. Korbel, J. M. Stuart, J. L. Jennings, L. D. Stein, M. D. Perry, H. K. Nahal-Bose, B. F. F. Ouellette, C. H. Li, E. Rheinbay, G. P. Nielsen, D. C. Sgroi, C.-L. Wu, W. C. Faquin, V. Deshpande, P. C. Boutros, A. J. Lazar, K. A. Hoadley, D. N. Louis, L. J. Dursi, C. K. Yung, M. H. Bailey, G. Saksena, K. M. Raine, I. Buchhalter, K. Kleinheinz, M. Schlesner, J. Zhang, W. Wang, D. A. Wheeler, L. Ding, J. T. Simpson, B. D. O’Connor, S. Yakneen, K. Ellrott, N. Miyoshi, A. P. Butler, R. Royo, S. I. Shorser, M. Vazquez, T. Rausch, G. Tiao, S. M. Waszak, B. Rodriguez-Martin, S. Shringarpure, D.-Y. Wu, G. M. Demidov, O. Delaneau, S. Hayashi, S. Imoto, N. Habermann, A. V. Segre, E. Garrison, A. Cafferkey, E. G. Alvarez, J. M. Heredia-Genestar, F. Muyas, O. Drechsel, A. L. Bruzos, J. Temes, J. Zamora, A. Baez-Ortega, H.-L. Kim, R. J. Mashl, K. Ye, A. DiBiase, K. Huang, I. Letunic, M. D. McLellan, S. J. Newhouse, T. Shmaya, S. Kumar, D. C. Wedge, M. H. Wright, V. D. Yellapantula, M. Gerstein, E. Khurana, T. Marques-Bonet, A. Navarro, C. D. Bustamante, R. Siebert, H. Nakagawa, D. F. Easton, S. Ossowski, J. M. C. Tubio, F. M. De La Vega, X. Estivill, D. Yuen, G. L. Mihaiescu, L. Omberg, V. Ferretti, R. Sabarinathan, O. Pich, A. Gonzalez-Perez, A. Taylor-Weiner, M. W. Fittall, J. Demeulemeester, M. Tarabichi, N. D. Roberts, P. Van Loo, I. Cortés-Ciriano, L. Urban, P. Park, B. Zhu, E. Pitkänen, Y. Li, N. Saini, L. J. Klimczak, J. Weischenfeldt, N. Sidiropoulos, L. B. Alexandrov, R. Rabionet, G. Escaramis, M. Bosio, A. Z. Holik, H. Susak, A. Prasad, S. Erkek, C. Calabrese, B. Raeder, E. Harrington, S. Mayes, D. Turner, S. Juul, S. A. Roberts, L. Song, R. Koster, L. Mirabello, X. Hua, T. J. Tanskanen, M. Tojo, J. Chen, L. A. Aaltonen, G. Rätsch, R. F. Schwarz, A. J. Butte, A. Brazma, S. J. Chanock, N. Chatterjee, O. Stegle, O. Harismendy, G. S. Bova, D. A. Gordenin, D. Haan, L. Sieverling, L. Feuerbach, D. Chalmers, Y. Joly, B. Knoppers, F. Molnár-Gábor, M. Phillips, A. Thorogood, D. Townend, M. Goldman, N. A. Fonseca, Q. Xiang, B. Craft, E. Piñeiro-Yáñez, A. Muñoz, R. Petryszak, A. Füllgrabe, F. Al-Shahrour, M. Keays, D. Haussler, J. Weinstein, W. Huber, A. Valencia, I. Papatheodorou, J. Zhu, Y. Fan, D. Torrents, M. Bieg, K. Chen, Z. Chong, K. Cibulskis, R. Eils, R. S. Fulton, J. L. Gelpi, S. Gonzalez, I. G. Gut, F. Hach, M. Heinold, T. Hu, V. Huang, B. Hutter, N. Jäger, J. Jung, Y. Kumar, C. Lalansingh, I. Leshchiner, D. Livitz, E. Z. Ma, Y. E. Maruvka, A. Milovanovic, M. M. Nielsen, N. Paramasivam, J. S. Pedersen, M. Puiggròs, S. C. Sahinalp, I. Sarrafi, C. Stewart, M. D. Stobbe, J. A. Wala, J. Wang, M. Wendl, J. Werner, Z. Wu, H. Xue, T. N. Yamaguchi, V. Yellapantula, B. N. Davis-Dusenbery, R. L. Grossman, Y. Kim, M. C. Heinold, J. Hinton, D. R. Jones, A. Menzies, L. Stebbings, J. M. Hess, M. Rosenberg, A. J. Dunford, M. Gupta, M. Imielinski, M. Meyerson, R. Beroukhim, J. Reimand, P. Dhingra, F. Favero, S. Dentro, J. Wintersinger, V. Rudneva,

J. W. Park, E. P. Hong, S. G. Heo, A. Kahles, K.-V. Lehmann, C. M. Soulette, Y. Shiraishi, F. Liu, Y. He, D. Demircioğlu, N. R. Davidson, L. Greger, S. Li, D. Liu, S. G. Stark, F. Zhang, S. B. Amin, P. Bailey, A. Chateigner, M. Frenkel-Morgenstern, Y. Hou, M. R. Huska, H. Kilpinen, F. C. Lamaze, C. Li, X. Li, X. Li, X. Liu, M. G. Marin, J. Markowski, T. Nandi, A. I. Ojesina, Q. Pan-Hammarström, P. J. Park, C. S. Pedamallu, H. Su, P. Tan, B. T. Teh, J. Wang, H. Xiong, C. Ye, C. Yung, X. Zhang, L. Zheng, S. Zhu, P. Awadalla, C. J. Creighton, K. Wu, H. Yang, J. Göke, Z. Zhang, A. N. Brooks, M. W. Fittall, I. Martincorena, C. Rubio-Perez, M. Juul, S. Schumacher, O. Shapira, D. Tamborero, L. Mularoni, H. Hornshøj, J. Deu-Pons, F. Muiños, J. Bertl, Q. Guo, A. Gonzalez-Perez, Q. Xiang, The ICGC/TCGA Pan-Cancer Analysis of Whole Genomes Consortium, Pan-cancer analysis of whole genomes. *Nature*. **578**, 82–93 (2020).

27. H. Gao, J. M. Korn, S. Ferretti, J. E. Monahan, Y. Wang, M. Singh, C. Zhang, C. Schnell, G. Yang, Y. Zhang, O. A. Balbin, S. Barbe, H. Cai, F. Casey, S. Chatterjee, D. Y. Chiang, S. Chuai, S. M. Cogan, S. D. Collins, E. Dammassa, N. Ebel, M. Embry, J. Green, A. Kauffmann, C. Kowal, R. J. Leary, J. Lehar, Y. Liang, A. Loo, E. Lorenzana, E. Robert McDonald, M. E. McLaughlin, J. Merkin, R. Meyer, T. L. Naylor, M. Patawaran, A. Reddy, C. Röelli, D. A. Ruddy, F. Salangsang, F. Santacroce, A. P. Singh, Y. Tang, W. Tinetto, S. Tobler, R. Velazquez, K. Venkatesan, F. Von Arx, H. Q. Wang, Z. Wang, M. Wiesmann, D. Wyss, F. Xu, H. Bitter, P. Atadja, E. Lees, F. Hofmann, E. Li, N. Keen, R. Cozens, M. R. Jensen, N. K. Pryer, J. A. Williams, W. R. Sellers, High-throughput screening using patient-derived tumor xenografts to predict clinical trial drug response. *Nat Med*. **21**, 1318–1325 (2015).
28. Y. Sun, N. Yang, F. E. Utama, S. S. Udhane, J. Zhang, A. R. Peck, A. Yanac, K. Duffey, J. F. Langenheim, V. Udhane, G. Xia, J. F. Peterson, J. M. Jorns, M. T. Nevalainen, R. Rouet, P. Schofield, D. Christ, C. J. Ormandy, A. L. Rosenberg, I. Chervoneva, S.-W. Tsaih, M. J. Flister, S. Y. Fuchs, K.-U. Wagner, H. Rui, NSG-Pro mouse model for uncovering resistance mechanisms and unique vulnerabilities in human luminal breast cancers. *Sci Adv*. **7**, eabc8145 (2021).
29. THE GTEX CONSORTIUM, The GTEx Consortium atlas of genetic regulatory effects across human tissues. *Science*. **369**, 1318–1330 (2020).
30. R. Tibshirani, J. Bien, J. Friedman, T. Hastie, N. Simon, J. Taylor, R. J. Tibshirani, Strong rules for discarding predictors in lasso-type problems. *Journal of the Royal Statistical Society: Series B (Statistical Methodology)*. **74**, 245–266 (2012).
31. S. Britsch, The neuregulin-I/ErbB signaling system in development and disease. *Adv Anat Embryol Cell Biol*. **190**, 1–65 (2007).
32. K. Shimada, J. A. Bachman, J. L. Muhlich, T. J. Mitchison, shinyDepMap, a tool to identify targetable cancer genes and their functional connections from Cancer Dependency Map data. *Elife*. **10**, e57116 (2021).
33. P. C. R. Parrish, J. D. Thomas, A. M. Gabel, S. Kamlapurkar, R. K. Bradley, A. H. Berger, Discovery of synthetic lethal and tumor suppressor paralog pairs in the human genome. *Cell Rep*. **36**, 109597 (2021).

34. B. Zhang, C. Tang, Y. Yao, X. Chen, C. Zhou, Z. Wei, F. Xing, L. Chen, X. Cai, Z. Zhang, S. Sun, Q. Liu, The tumor therapy landscape of synthetic lethality. *Nat Commun*. **12**, 1275 (2021).
35. B. Adamson, T. M. Norman, M. Jost, M. Y. Cho, J. K. Nuñez, Y. Chen, J. E. Villalta, L. A. Gilbert, M. A. Horlbeck, M. Y. Hein, R. A. Pak, A. N. Gray, C. A. Gross, A. Dixit, O. Parnas, A. Regev, J. S. Weissman, A Multiplexed Single-Cell CRISPR Screening Platform Enables Systematic Dissection of the Unfolded Protein Response. *Cell*. **167**, 1867-1882.e21 (2016).
36. P. van der Lelij, S. Lieb, J. Jude, G. Wutz, C. P. Santos, K. Falkenberg, A. Schlattl, J. Ban, R. Schwentner, T. Hoffmann, H. Kovar, F. X. Real, T. Waldman, M. A. Pearson, N. Kraut, J.-M. Peters, J. Zuber, M. Petronczki, Synthetic lethality between the cohesin subunits STAG1 and STAG2 in diverse cancer contexts. *eLife*. **6**, e26980 (2017).
37. H. Ogiwara, M. Sasaki, T. Mitachi, T. Oike, S. Higuchi, Y. Tominaga, T. Kohno, Targeting p300 Addiction in CBP-Deficient Cancers Causes Synthetic Lethality by Apoptotic Cell Death due to Abrogation of MYC Expression. *Cancer Discov*. **6**, 430–445 (2016).
38. P. C. DeWeirdt, K. R. Sanson, A. K. Sangree, M. Hegde, R. E. Hanna, M. N. Feeley, A. L. Griffith, T. Teng, S. M. Borys, C. Strand, J. K. Joung, B. P. Kleinstiver, X. Pan, A. Huang, J. G. Doench, Optimization of AsCas12a for combinatorial genetic screens in human cells. *Nat Biotechnol*. **39**, 94–104 (2021).
39. M. Dede, M. McLaughlin, E. Kim, T. Hart, Multiplex enCas12a screens detect functional buffering among paralogs otherwise masked in monogenic Cas9 knockout screens. *Genome Biol*. **21**, 262 (2020).
40. S. K. Chia, V. H. Bramwell, D. Tu, L. E. Shepherd, S. Jiang, T. Vickery, E. Mardis, S. Leung, K. Ung, K. I. Pritchard, J. S. Parker, P. S. Bernard, C. M. Perou, M. J. Ellis, T. O. Nielsen, A 50-gene intrinsic subtype classifier for prognosis and prediction of benefit from adjuvant tamoxifen. *Clin Cancer Res*. **18**, 4465–4472 (2012).
41. J. Liu, T. Lichtenberg, K. A. Hoadley, L. M. Poisson, A. J. Lazar, A. D. Cherniack, A. J. Kovatich, C. C. Benz, D. A. Levine, A. V. Lee, L. Omberg, D. M. Wolf, C. D. Shriver, V. Thorsson, S. J. Caesar-Johnson, J. A. Demchok, I. Felau, M. Kasapi, M. L. Ferguson, C. M. Hutter, H. J. Sofia, R. Tarnuzzer, Z. Wang, L. Yang, J. C. Zenklusen, J. (Julia) Zhang, S. Chudamani, J. Liu, L. Lolla, R. Naresh, T. Pihl, Q. Sun, Y. Wan, Y. Wu, J. Cho, T. DeFreitas, S. Frazer, N. Gehlenborg, G. Getz, D. I. Heiman, J. Kim, M. S. Lawrence, P. Lin, S. Meier, M. S. Noble, G. Saksena, D. Voet, H. Zhang, B. Bernard, N. Chambwe, V. Dhankani, T. Knijnenburg, R. Kramer, K. Leinonen, Y. Liu, M. Miller, S. Reynolds, I. Shmulevich, V. Thorsson, W. Zhang, R. Akbani, B. M. Broom, A. M. Hegde, Z. Ju, R. S. Kanchi, A. Korkut, J. Li, H. Liang, S. Ling, W. Liu, Y. Lu, G. B. Mills, K.-S. Ng, A. Rao, M. Ryan, J. Wang, J. N. Weinstein, J. Zhang, A. Abeshouse, J. Armenia, D. Chakravarty, W. K. Chatila, I. de Bruijn, J. Gao, B. E. Gross, Z. J. Heins, R. Kundra, K. La, M. Ladanyi, A. Luna, M. G. Nissan, A. Ochoa, S. M. Phillips, E. Reznik, F. Sanchez-Vega, C. Sander, N. Schultz, R. Sheridan, S. O. Sumer, Y. Sun, B. S. Taylor, J. Wang, H. Zhang, P. Anur, M. Peto, P. Spellman, C. Benz, J. M. Stuart, C. K. Wong, C. Yau, D. N. Hayes, J. S. Parker, M. D. Wilkerson, A. Ally, M. Balasundaram, R. Bowlby, D. Brooks, R. Carlsen, E. Chuah,

N. Dhalla, R. Holt, S. J. M. Jones, K. Kasaian, D. Lee, Y. Ma, M. A. Marra, M. Mayo, R. A. Moore, A. J. Mungall, K. Mungall, A. G. Robertson, S. Sadeghi, J. E. Schein, P. Sipahimalani, A. Tam, N. Thiessen, K. Tse, T. Wong, A. C. Berger, R. Beroukhim, A. D. Cherniack, C. Cibulskis, S. B. Gabriel, G. F. Gao, G. Ha, M. Meyerson, S. E. Schumacher, J. Shih, M. H. Kucherlapati, R. S. Kucherlapati, S. Baylin, L. Cope, L. Danilova, M. S. Bootwalla, P. H. Lai, D. T. Maglinte, D. J. Van Den Berg, D. J. Weisenberger, J. T. Auman, S. Balu, T. Bodenheimer, C. Fan, K. A. Hoadley, A. P. Hoyle, S. R. Jefferys, C. D. Jones, S. Meng, P. A. Mieczkowski, L. E. Mose, A. H. Perou, C. M. Perou, J. Roach, Y. Shi, J. V. Simons, T. Skelly, M. G. Soloway, D. Tan, U. Veluvolu, H. Fan, T. Hinoue, P. W. Laird, H. Shen, W. Zhou, M. Bellair, K. Chang, K. Covington, C. J. Creighton, H. Dinh, H. Doddapaneni, L. A. Donehower, J. Drummond, R. A. Gibbs, R. Glenn, W. Hale, Y. Han, J. Hu, V. Korchina, S. Lee, L. Lewis, W. Li, X. Liu, M. Morgan, D. Morton, D. Muzny, J. Santibanez, M. Sheth, E. Shinbro, L. Wang, M. Wang, D. A. Wheeler, L. Xi, F. Zhao, J. Hess, E. L. Appelbaum, M. Bailey, M. G. Cordes, L. Ding, C. C. Fronick, L. A. Fulton, R. S. Fulton, C. Kandoth, E. R. Mardis, M. D. McLellan, C. A. Miller, H. K. Schmidt, R. K. Wilson, D. Crain, E. Curley, J. Gardner, K. Lau, D. Mallery, S. Morris, J. Paulauskis, R. Penny, C. Shelton, T. Shelton, M. Sherman, E. Thompson, P. Yena, J. Bowen, J. M. Gastier-Foster, M. Gerken, K. M. Leraas, T. M. Lichtenberg, N. C. Ramirez, L. Wise, E. Zmuda, N. Corcoran, T. Costello, C. Hovens, A. L. Carvalho, A. C. de Carvalho, J. H. Fregnani, A. Longatto-Filho, R. M. Reis, C. Scapulatempo-Neto, H. C. S. Silveira, D. O. Vidal, A. Burnette, J. Eschbacher, B. Hermes, A. Noss, R. Singh, M. L. Anderson, P. D. Castro, M. Ittmann, D. Huntsman, B. Kohl, X. Le, R. Thorp, C. Andry, E. R. Duffy, V. Lyadov, O. Paklina, G. Setdikova, A. Shabunin, M. Tavobilov, C. McPherson, R. Warnick, R. Berkowitz, D. Cramer, C. Feltmate, N. Horowitz, A. Kibel, M. Muto, C. P. Raut, A. Malykh, J. S. Barnholtz-Sloan, W. Barrett, K. Devine, J. Fulop, Q. T. Ostrom, K. Shimmel, Y. Wolinsky, A. E. Sloan, A. De Rose, F. Giuliante, M. Goodman, B. Y. Karlan, C. H. Hagedorn, J. Eckman, J. Harr, J. Myers, K. Tucker, L. A. Zach, B. Deyarmin, H. Hu, L. Kvecher, C. Larson, R. J. Mural, S. Somiari, A. Vicha, T. Zelinka, J. Bennett, M. Iacocca, B. Rabeno, P. Swanson, M. Latour, L. Lacombe, B. Têtu, A. Bergeron, M. McGraw, S. M. Staugaitis, J. Chabot, H. Hibshoosh, A. Sepulveda, T. Su, T. Wang, O. Potapova, O. Voronina, L. Desjardins, O. Mariani, S. Roman-Roman, X. Sastre, M.-H. Stern, F. Cheng, S. Signoretti, A. Berchuck, D. Bigner, E. Lipp, J. Marks, S. McCall, R. McLendon, A. Secord, A. Sharp, M. Behera, D. J. Brat, A. Chen, K. Delman, S. Force, F. Khuri, K. Magliocca, S. Maithel, J. J. Olson, T. Owonikoko, A. Pickens, S. Ramalingam, D. M. Shin, G. Sica, E. G. Van Meir, H. Zhang, W. Eijckenboom, A. Gillis, E. Korpershoek, L. Looijenga, W. Oosterhuis, H. Stoop, K. E. van Kessel, E. C. Zwarthoff, C. Calatozzolo, L. Cuppini, S. Cuzzubbo, F. DiMeco, G. Finocchiaro, L. Mattei, A. Perin, B. Pollo, C. Chen, J. Houck, P. Lohavanichbutr, A. Hartmann, C. Stoehr, R. Stoehr, H. Taubert, S. Wach, B. Wullich, W. Kycler, D. Murawa, M. Wiznerowicz, K. Chung, W. J. Edenfield, J. Martin, E. Baudin, G. Bubley, R. Bueno, A. De Rienzo, W. G. Richards, S. Kalkanis, T. Mikkelsen, H. Noushmehr, L. Scarpace, N. Girard, M. Aymerich, E. Campo, E. Giné, A. L. Guillermo, N. Van Bang, P. T. Hanh, B. D. Phu, Y. Tang, H. Colman, K. Evason, P. R. Dottino, J. A.

Martignetti, H. Gabra, H. Juhl, T. Akeredolu, S. Stepa, D. Hoon, K. Ahn, K. J. Kang, F. Beuschlein, A. Breggia, M. Birrer, D. Bell, M. Borad, A. H. Bryce, E. Castle, V. Chandan, J. Cheville, J. A. Copland, M. Farnell, T. Flotte, N. Giama, T. Ho, M. Kendrick, J.-P. Kocher, K. Kopp, C. Moser, D. Nagorney, D. O'Brien, B. P. O'Neill, T. Patel, G. Petersen, F. Que, M. Rivera, L. Roberts, R. Smallridge, T. Smyrk, M. Stanton, R. H. Thompson, M. Torbenson, J. D. Yang, L. Zhang, F. Brimo, J. A. Ajani, A. M. Angulo Gonzalez, C. Behrens, J. Bondaruk, R. Broaddus, B. Czerniak, B. Esmaeli, J. Fujimoto, J. Gershenwald, C. Guo, A. J. Lazar, C. Logothetis, F. Meric-Bernstam, C. Moran, L. Ramondetta, D. Rice, A. Sood, P. Tamboli, T. Thompson, P. Troncoso, A. Tsao, I. Wistuba, C. Carter, L. Haydu, P. Hersey, V. Jakrot, H. Kakavand, R. Kefford, K. Lee, G. Long, G. Mann, M. Quinn, R. Saw, R. Scolyer, K. Shannon, A. Spillane, J. Stretch, M. Synott, J. Thompson, J. Wilmott, H. Al-Ahmadie, T. A. Chan, R. Ghossein, A. Gopalan, D. A. Levine, V. Reuter, S. Singer, B. Singh, N. V. Tien, T. Broudy, C. Mirsaidi, P. Nair, P. Drwiega, J. Miller, J. Smith, H. Zaren, J.-W. Park, N. P. Hung, E. Kebebew, W. M. Linehan, A. R. Metwalli, K. Pacak, P. A. Pinto, M. Schiffman, L. S. Schmidt, C. D. Vocke, N. Wentzensen, R. Worrell, H. Yang, M. Moncrieff, C. Goparaju, J. Melamed, H. Pass, N. Botnariuc, I. Caraman, M. Cernat, I. Chemencedji, A. Clipca, S. Doruc, G. Gorincioi, S. Mura, M. Pirtac, I. Stancul, D. Tcaciuc, M. Albert, I. Alexopoulou, A. Arnaout, J. Bartlett, J. Engel, S. Gilbert, J. Parfitt, H. Sekhon, G. Thomas, D. M. Rassl, R. C. Rintoul, C. Bifulco, R. Tamakawa, W. Urba, N. Hayward, H. Timmers, A. Antenucci, F. Facciolo, G. Grazi, M. Marino, R. Merola, R. de Krijger, A.-P. Gimenez-Roqueplo, A. Piché, S. Chevalier, G. McKercher, K. Birsoy, G. Barnett, C. Brewer, C. Farver, T. Naska, N. A. Pennell, D. Raymond, C. Schilero, K. Smolenski, F. Williams, C. Morrison, J. A. Borgia, M. J. Liptay, M. Pool, C. W. Seder, K. Junker, L. Omberg, M. Dinkin, G. Manikhas, D. Alvaro, M. C. Bragazzi, V. Cardinale, G. Carpino, E. Gaudio, D. Chesla, S. Cottingham, M. Dubina, F. Moiseenko, R. Dhanasekaran, K.-F. Becker, K.-P. Janssen, J. Slotta-Huspenina, M. H. Abdel-Rahman, D. Aziz, S. Bell, C. M. Cebulla, A. Davis, R. Duell, J. B. Elder, J. Hilty, B. Kumar, J. Lang, N. L. Lehman, R. Mandt, P. Nguyen, R. Pilarski, K. Rai, L. Schoenfield, K. Senecal, P. Wakely, P. Hansen, R. Lechan, J. Powers, A. Tischler, W. E. Grizzle, K. C. Sexton, A. Kastl, J. Henderson, S. Porten, J. Waldmann, M. Fassnacht, S. L. Asa, D. Schadendorf, M. Couce, M. Graefen, H. Huland, G. Sauter, T. Schlomm, R. Simon, P. Tennstedt, O. Olabode, M. Nelson, O. Bathe, P. R. Carroll, J. M. Chan, P. Disaia, P. Glenn, R. K. Kelley, C. N. Landen, J. Phillips, M. Prados, J. Simko, K. Smith-McCune, S. VandenBerg, K. Roggin, A. Fehrenbach, A. Kendler, S. Sifri, R. Steele, A. Jimeno, F. Carey, I. Forgie, M. Mannelli, M. Carney, B. Hernandez, B. Campos, C. Herold-Mende, C. Jungk, A. Unterberg, A. von Deimling, A. Bossler, J. Galbraith, L. Jacobus, M. Knudson, T. Knutson, D. Ma, M. Milhem, R. Sigmund, A. K. Godwin, R. Madan, H. G. Rosenthal, C. Adebamowo, S. N. Adebamowo, A. Boussioutas, D. Beer, T. Giordano, A.-M. Mes-Masson, F. Saad, T. Bocklage, L. Landrum, R. Mannel, K. Moore, K. Moxley, R. Postier, J. Walker, R. Zuna, M. Feldman, F. Valdivieso, R. Dhir, J. Luketich, E. M. Mora Pinero, M. Quintero-Aguilo, J. Carlotti Carlos Gilberto, J. S. Dos Santos, R. Kemp, A. Sankarankuty, D. Tirapelli, J. Catto, K. Agnew, E. Swisher, J. Creaney, B. Robinson, C. S. Shelley, E. M. Godwin, S. Kendall, C. Shipman, C. Bradford,

T. Carey, A. Haddad, J. Moyer, L. Peterson, M. Prince, L. Rozek, G. Wolf, R. Bowman, K. M. Fong, I. Yang, R. Korst, W. K. Rathmell, J. L. Fantacone-Campbell, J. A. Hooke, A. J. Kovatich, C. D. Shriver, J. DiPersio, B. Drake, R. Govindan, S. Heath, T. Ley, B. Van Tine, P. Westervelt, M. A. Rubin, J. I. Lee, N. D. Aredes, A. Mariamidze, H. Hu, An Integrated TCGA Pan-Cancer Clinical Data Resource to Drive High-Quality Survival Outcome Analytics. *Cell*. **173**, 400-416.e11 (2018).

42. V. Varadan, H. Gilmore, K. L. S. Miskimen, D. Tuck, S. Parsai, A. Awadallah, I. E. Krop, E. P. Winer, V. Bossuyt, G. Somlo, M. M. Abu-Khalaf, M. A. Fenton, W. Sikov, L. N. Harris, Immune Signatures Following Single Dose Trastuzumab Predict Pathologic Response to PreoperativeTrastuzumab and Chemotherapy in HER2-Positive Early Breast Cancer. *Clin Cancer Res*. **22**, 3249–3259 (2016).
43. J. M. Dempster, J. M. Krill-Burger, J. M. McFarland, A. Warren, J. S. Boehm, F. Vazquez, W. C. Hahn, T. R. Golub, A. Tsherniak, “Gene expression has more power for predicting in vitro cancer cell vulnerabilities than genomics” (2020), p. 2020.02.21.959627, (available at https://www.biorxiv.org/content/10.1101/2020.02.21.959627v3).
44. M. Cieślik, A. M. Chinnaiyan, Cancer transcriptome profiling at the juncture of clinical translation. *Nat Rev Genet*. **19**, 93–109 (2018).
45. K. A. Hoadley, C. Yau, T. Hinoue, D. M. Wolf, A. J. Lazar, E. Drill, R. Shen, A. M. Taylor, A. D. Cherniack, V. Thorsson, R. Akbani, R. Bowlby, C. K. Wong, M. Wiznerowicz, F. Sanchez-Vega, A. G. Robertson, B. G. Schneider, M. S. Lawrence, H. Noushmehr, T. M. Malta, Cancer Genome Atlas Network, J. M. Stuart, C. C. Benz, P. W. Laird, Cell-of-Origin Patterns Dominate the Molecular Classification of 10,000 Tumors from 33 Types of Cancer. *Cell*. **173**, 291-304.e6 (2018).
46. Synthetic lethality as an engine for cancer drug target discovery | Nature Reviews Drug Discovery, (available at https://www.nature.com/articles/s41573-019-0046-z).
47. F. L. Muller, E. A. Aquilanti, R. A. DePinho, Collateral Lethality: A new therapeutic strategy in oncology. *Trends Cancer*. **1**, 161–173 (2015).
48. C. M. Croce, Oncogenes and Cancer. *New England Journal of Medicine*. **358**, 502–511 (2008).
49. C. V. Dang, E. P. Reddy, K. M. Shokat, L. Soucek, Drugging the ‘undruggable’ cancer targets. *Nat Rev Cancer*. **17**, 502–508 (2017).
50. H. H. Wong, S. Halford, Dose-limiting toxicity and maximum tolerated dose: still fit for purpose? *The Lancet Oncology*. **16**, 1287–1288 (2015).
51. Y.-C. Chiu, H.-I. H. Chen, T. Zhang, S. Zhang, A. Gorthi, L.-J. Wang, Y. Huang, Y. Chen, Predicting drug response of tumors from integrated genomic profiles by deep neural networks. *BMC Med Genomics*. **12**, 18 (2019).
52. A. Warren, Y. Chen, A. Jones, T. Shibue, W. C. Hahn, J. S. Boehm, F. Vazquez, A. Tsherniak, J. M. McFarland, Global computational alignment of tumor and cell line transcriptional profiles. *Nat Commun*. **12**, 22 (2021).
53. CRISPR screens in cancer spheroids identify 3D growth-specific vulnerabilities | Nature, (available at https://www.nature.com/articles/s41586-020-2099-x?proof=t).

54. K. A. Lawson, C. M. Sousa, X. Zhang, E. Kim, R. Akthar, J. J. Caumanns, Y. Yao, N. Mikolajewicz, C. Ross, K. R. Brown, A. A. Zid, Z. P. Fan, S. Hui, J. A. Krall, D. M. Simons, C. J. Slater, V. De Jesus, L. Tang, R. Singh, J. E. Goldford, S. Martin, Q. Huang, E. A. Francis, A. Habsid, R. Climie, D. Tieu, J. Wei, R. Li, A. H. Y. Tong, M. Aregger, K. S. Chan, H. Han, X. Wang, P. Mero, J. H. Brumell, A. Finelli, L. Ailles, G. Bader, G. A. Smolen, G. A. Kingsbury, T. Hart, C. Kung, J. Moffat, Functional genomic landscape of cancer-intrinsic evasion of killing by T cells. *Nature*. **586**, 120–126 (2020).
55. D. R. Robinson, Y.-M. Wu, R. J. Lonigro, P. Vats, E. Cobain, J. Everett, X. Cao, E. Rabban, C. Kumar-Sinha, V. Raymond, S. Schuetze, A. Alva, J. Siddiqui, R. Chugh, F. Worden, M. M. Zalupski, J. Innis, R. J. Mody, S. A. Tomlins, D. Lucas, L. H. Baker, N. Ramnath, A. F. Schott, D. F. Hayes, J. Vijai, K. Offit, E. M. Stoffel, J. S. Roberts, D. C. Smith, L. P. Kunju, M. Talpaz, M. Cieślik, A. M. Chinnaiyan, Integrative clinical genomics of metastatic cancer. *Nature*. **548**, 297–303 (2017).
56. J. M. Replogle, T. M. Norman, A. Xu, J. A. Hussmann, J. Chen, J. Z. Cogan, E. J. Meer, J. M. Terry, D. P. Riordan, N. Srinivas, I. T. Fiddes, J. G. Arthur, L. J. Alvarado, K. A. Pfeiffer, T. S. Mikkelsen, J. S. Weissman, B. Adamson, Combinatorial single-cell CRISPR screens by direct guide RNA capture and targeted sequencing. *Nat Biotechnol*. **38**, 954–961 (2020).
57. P. Datlinger, A. F. Rendeiro, C. Schmidl, T. Krausgruber, P. Traxler, J. Klughammer, L. C. Schuster, A. Kuchler, D. Alpar, C. Bock, Pooled CRISPR screening with single-cell transcriptome readout. *Nat Methods*. **14**, 297–301 (2017).
58. Y. Zhang, G. Parmigiani, W. E. Johnson, ComBat-seq: batch effect adjustment for RNA-seq count data. *NAR Genomics and Bioinformatics*. **2** (2020), doi:10.1093/nargab/lqaa078.
59. D. C. Koboldt, R. S. Fulton, M. D. McLellan, H. Schmidt, J. Kalicki-Veizer, J. F. McMichael, L. L. Fulton, D. J. Dooling, L. Ding, E. R. Mardis, R. K. Wilson, A. Ally, M. Balasundaram, Y. S. N. Butterfield, R. Carlsen, C. Carter, A. Chu, E. Chuah, H.-J. E. Chun, R. J. N. Coope, N. Dhalla, R. Guin, C. Hirst, M. Hirst, R. A. Holt, D. Lee, H. I. Li, M. Mayo, R. A. Moore, A. J. Mungall, E. Pleasance, A. Gordon Robertson, J. E. Schein, A. Shafiei, P. Sipahimalani, J. R. Slobodan, D. Stoll, A. Tam, N. Thiessen, R. J. Varhol, N. Wye, T. Zeng, Y. Zhao, I. Birol, S. J. M. Jones, M. A. Marra, A. D. Cherniack, G. Saksena, R. C. Onofrio, N. H. Pho, S. L. Carter, S. E. Schumacher, B. Tabak, B. Hernandez, J. Gentry, H. Nguyen, A. Crenshaw, K. Ardlie, R. Beroukhim, W. Winckler, G. Getz, S. B. Gabriel, M. Meyerson, L. Chin, P. J. Park, R. Kucherlapati, K. A. Hoadley, J. Todd Auman, C. Fan, Y. J. Turman, Y. Shi, L. Li, M. D. Topal, X. He, H.-H. Chao, A. Prat, G. O. Silva, M. D. Iglesia, W. Zhao, J. Usary, J. S. Berg, M. Adams, J. Booker, J. Wu, A. Gulabani, T. Bodenheimer, A. P. Hoyle, J. V. Simons, M. G. Soloway, L. E. Mose, S. R. Jefferys, S. Balu, J. S. Parker, D. Neil Hayes, C. M. Perou, S. Malik, S. Mahurkar, H. Shen, D. J. Weisenberger, T. Triche Jr, P. H. Lai, M. S. Bootwalla, D. T. Maglinte, B. P. Berman, D. J. Van Den Berg, S. B. Baylin, P. W. Laird, C. J. Creighton, L. A. Donehower, G. Getz, M. Noble, D. Voet, G. Saksena, N. Gehlenborg, D. DiCara, J. Zhang, H. Zhang, C.-J. Wu, S. Yingchun Liu, M. S. Lawrence, L. Zou, A. Sivachenko, P. Lin, P. Stojanov, R. Jing, J. Cho, R. Sinha, R. W. Park, M.-D. Nazaire, J. Robinson, H. Thorvaldsdottir, J. Mesirov, P. J. Park, L. Chin, S. Reynolds, R.

B. Kreisberg, B. Bernard, R. Bressler, T. Erkkila, J. Lin, V. Thorsson, W. Zhang, I. Shmulevich, G. Ciriello, N. Weinhold, N. Schultz, J. Gao, E. Cerami, B. Gross, A. Jacobsen, R. Sinha, B. Arman Aksoy, Y. Antipin, B. Reva, R. Shen, B. S. Taylor, M. Ladanyi, C. Sander, P. Anur, P. T. Spellman, Y. Lu, W. Liu, R. R. G. Verhaak, G. B. Mills, R. Akbani, N. Zhang, B. M. Broom, T. D. Casasent, C. Wakefield, A. K. Unruh, K. Baggerly, K. Coombes, J. N. Weinstein, D. Haussler, C. C. Benz, J. M. Stuart, S. C. Benz, J. Zhu, C. C. Szeto, G. K. Scott, C. Yau, E. O. Paull, D. Carlin, C. Wong, A. Sokolov, J. Thusberg, S. Mooney, S. Ng, T. C. Goldstein, K. Ellrott, M. Grifford, C. Wilks, S. Ma, B. Craft, C. Yan, Y. Hu, D. Meerzaman, J. M. Gastier-Foster, J. Bowen, N. C. Ramirez, A. D. Black, R. E. Pyatt, P. White, E. J. Zmuda, J. Frick, T. M. Lichtenberg, R. Brookens, M. M. George, M. A. Gerken, H. A. Harper, K. M. Leraas, L. J. Wise, T. R. Tabler, C. McAllister, T. Barr, M. Hart-Kothari, K. Tarvin, C. Saller, G. Sandusky, C. Mitchell, M. V. Iacocca, J. Brown, B. Rabeno, C. Czerwinski, N. Petrelli, O. Dolzhansky, M. Abramov, O. Voronina, O. Potapova, J. R. Marks, W. M. Suchorska, D. Murawa, W. Kycler, M. Ibbs, K. Korski, A. Spychała, P. Murawa, J. J. Brzeziński, H. Perz, R. Łaźniak, M. Teresiak, H. Tatka, E. Leporowska, M. Bogusz-Czerniewicz, J. Malicki, A. Mackiewicz, M. Wiznerowicz, X. Van Le, B. Kohl, N. Viet Tien, R. Thorp, N. Van Bang, H. Sussman, B. Duc Phu, R. Hajek, N. Phi Hung, T. Viet The Phuong, H. Quyet Thang, K. Zaki Khan, R. Penny, D. Mallery, E. Curley, C. Shelton, P. Yena, J. N. Ingle, F. J. Couch, W. L. Lingle, T. A. King, A. Maria Gonzalez-Angulo, G. B. Mills, M. D. Dyer, S. Liu, X. Meng, M. Patangan, The Cancer Genome Atlas Network, Genome sequencing centres: Washington University in St Louis, Genome characterization centres: BC Cancer Agency, Broad Institute, Brigham & Women's Hospital & Harvard Medical School, C. H. University of North Carolina, University of Southern California/Johns Hopkins, Genome data analysis: Baylor College of Medicine, Institute for Systems Biology, Memorial Sloan-Kettering Cancer Center, Oregon Health & Science University, The University of Texas MD Anderson Cancer Center, S. C. I. University of California, NCI, Biospecimen core resource: Nationwide Children's Hospital Biospecimen Core Resource, Tissue source sites: ABS-IUPUI, Christiana, Cureline, Duke University Medical Center, The Greater Poland Cancer Centre, ILSBio, International Genomics Consortium, Mayo Clinic, MSKCC, MD Anderson Cancer Center, Comprehensive molecular portraits of human breast tumours. *Nature*. **490**, 61–70 (2012).

60. D. Szklarczyk, A. L. Gable, K. C. Nastou, D. Lyon, R. Kirsch, S. Pyysalo, N. T. Doncheva, M. Legeay, T. Fang, P. Bork, L. J. Jensen, C. von Mering, The STRING database in 2021: customizable protein-protein networks, and functional characterization of user-uploaded gene/measurement sets. *Nucleic Acids Res*. **49**, D605–D612 (2021).
61. Y. Deng, S. Luo, C. Deng, T. Luo, W. Yin, H. Zhang, Y. Zhang, X. Zhang, Y. Lan, Y. Ping, Y. Xiao, X. Li, Identifying mutual exclusivity across cancer genomes: computational approaches to discover genetic interaction and reveal tumor vulnerability. *Briefings in Bioinformatics*. **20**, 254–266 (2019).
62. M. C. Hajkarim, E. Upfal, F. Vandin, Differentially mutated subnetworks discovery. *Algorithms Mol Biol*. **14**, 10 (2019).

63. J. W. Kim, O. B. Botvinnik, O. Abudayyeh, C. Birger, J. Rosenbluh, Y. Shrestha, M. E. Abazeed, P. S. Hammerman, D. DiCara, D. J. Konieczkowski, C. M. Johannessen, A. Liberzon, A. R. Alizad-Rahvar, G. Alexe, A. Aguirre, M. Ghandi, H. Greulich, F. Vazquez, B. A. Weir, E. M. Van Allen, A. Tsherniak, D. D. Shao, T. I. Zack, M. Noble, G. Getz, R. Beroukhim, L. A. Garraway, M. Ardakani, C. Romualdi, G. Sales, D. A. Barbie, J. S. Boehm, W. C. Hahn, J. P. Mesirov, P. Tamayo, Characterizing genomic alterations in cancer by complementary functional associations. *Nat Biotechnol*. **34**, 539–546 (2016).
64. J. S. Lee, A. Das, L. Jerby-Arnon, R. Arafeh, N. Auslander, M. Davidson, L. McGarry, D. James, A. Amzallag, S. G. Park, K. Cheng, W. Robinson, D. Atias, C. Stossel, E. Buzhor, G. Stein, J. J. Waterfall, P. S. Meltzer, T. Golan, S. Hannenhalli, E. Gottlieb, C. H. Benes, Y. Samuels, E. Shanks, E. Ruppin, Harnessing synthetic lethality to predict the response to cancer treatment. *Nat Commun*. **9**, 2546 (2018).
65. D. Szklarczyk, A. L. Gable, D. Lyon, A. Junge, S. Wyder, J. Huerta-Cepas, M. Simonovic, N. T. Doncheva, J. H. Morris, P. Bork, L. J. Jensen, C. von Mering, STRING v11: protein–protein association networks with increased coverage, supporting functional discovery in genome-wide experimental datasets. *Nucleic Acids Research*. **47**, D607–D613 (2019).
66. E. L. Huttlin, L. Ting, R. J. Bruckner, F. Gebreab, M. P. Gygi, J. Szpyt, S. Tam, G. Zarraga, G. Colby, K. Baltier, R. Dong, V. Guarani, L. P. Vaites, A. Ordureau, R. Rad, B. K. Erickson, M. Wühr, J. Chick, B. Zhai, D. Kolippakkam, J. Mintseris, R. A. Obar, T. Harris, S. Artavanis-Tsakonas, M. E. Sowa, P. D. Camilli, J. A. Paulo, J. W. Harper, S. P. Gygi, The BioPlex Network: A Systematic Exploration of the Human Interactome. *Cell*. **162**, 425–440 (2015).
67. M. D. Robinson, A. Oshlack, A scaling normalization method for differential expression analysis of RNA-seq data. *Genome Biol*. **11**, R25 (2010).
68. C. W. Law, Y. Chen, W. Shi, G. K. Smyth, voom: precision weights unlock linear model analysis tools for RNA-seq read counts. *Genome Biology*. **15**, R29 (2014).