LEGENDS TO SUPPLEMENTAL FIGURES

**Supplemental Figure S1**
Schematic depicting the analysis of PI3K SL genes for the shRNA screen performed in the indicated isogenic cell lines. EdgeR was first used to determine the *lethality score* for each shRNA in HCT116 PI3K WT and Mut cells. Next RIGER was used to determine for each gene the PI3K *synthetic lethality score*, based on the delta (difference) between SL score in PI3K Mut and WT cells (see Materials and Methods). Two separate graphs generated with EdgeR represent the lethality score in PI3K WT and PI3K Mut cell lines (the red dots represent genes with a significant FDR in the EdgeR analysis for lethality in PI3K WT and Mut).

**Supplemental Figure S2**
Schematic depicting the meta-analysis including data from the screen on the isogenic cell lines HCT116, analysis of the Achilles dataset and analysis of the COLT-Cancer dataset (see text for details). For each dataset, the number of genes analyzed, the number of hits resulting from the analysis (synthetic lethal genes), and the % enrichment of the PI3K.AKT.MTOR.Activation.Pathway is shown.

**Supplemental Figure S3**
(A) (B) (C) Cell death assays were performed in HCT116 and DLD1 cells of the indicated genotypes after treatment with the indicated drug and concentration for 48-72h, as in Fig. 3C. CHX: cycloheximide, LTM: lactimidomycin; BEZ235: NVP-BEZ235
(D) HCT116 PI3K Mut and WT cells were transduced with pHAGE-pLnd10-mirE encoding EIF1AX shRNA and treated with or without doxycycline to induce expression of the shRNA. Western blot analysis after 72h of treatment is shown (see also Fig. 3F).
(E) Example of quantification of MCA assay. HCT116 PI3K Mut and WT cells were mixed in a 1 Mut:3 WT ratio and cultured in the presence of DMSO or CHX (50 nM) for 5 days. The percentage of Mut and WT cells in the mixed cell populations treated with DMSO or CHX is shown as determined by FACS at the end of the assay. CHX: cycloheximide

**Supplemental Figure S4**

(A) qPCR for the indicated mRNAs on DLD1 cells with the indicated genotype treated with DMSO, Torin2 (20 nM) or Rapamycin (20 nM) for 48h prior to harvesting the cells.

(B) Western Blot analysis for the indicated proteins on HCT116 and DLD1 cells with the indicated genotype.

(C) Cell death assays were performed in HCT116 and DLD1 cells of the indicated genotypes after treatment with the indicated drug and concentration for 48h, as in Fig. 4. Bort: Bortezomib; CLL: clasto-Lactacystin

(D) (E) Cell death assays were performed on HCT116 and DLD1 cells of the indicated genotypes after treatment with the indicated drug and concentration for 48h, as in Fig. 3C. CLL: clasto-Lactacystin

(F) MCA assays were performed by using the indicated drug and concentration as in Fig. 3A. The percentage of Mut cells at the end of the assay, relative to the treatment with DMSO is shown.
LEGENDS TO SUPPLEMENTARY TABLES

Supplemental Table S1
RNAi screen for PI3K synthetic lethality in HCT116 PI3K Mut and PI3K WT cells. This table contains both the genome-wide primary screen data and the secondary screen data performed utilizing a focused RNAi library (1045 genes).

Supplemental Table S2
Table S2A contains the PI3K.AKT.MTOR.Activation.Pathway signature containing genes implicated in PI3K/AKT/mTOR Pathway from REACTOME database (Table S2A). Table S2B contains the results of the Fisher exact test for the enrichment of this gene set among the PI3K synthetic lethal gene lists derived from the analyses (analysis of the screen in HCT116 isogenic lines; analysis of Achilles dataset; analysis of COLT-Cancer dataset; combined analysis of the three datasets for PI3K synthetic lethality) or KRAS (analysis of Achilles dataset) (Table S2B).

Supplemental Table S3
GSEA analysis (method: classic) on the PI3K synthetic lethal screen performed in HCT116 PI3K Mut and WT cell lines. The table contains the output of GSEA (NAME: pathway name; SIZE: gene set size; ES: GSEA enrichment score; NES: GSEA normalized enrichment score). Please refer to http://www.broadinstitute.org/gsea/index.jsp for more details on the output of GSEA analysis.

Supplemental Table S4
Analysis of synthetic lethality with PI3K oncogenic mutations in the Achilles RNAi dataset. The SL Lethal score is shown as well as the p-value (Wilcoxon method) in Table S4A. Table S4B shows the list of cancer cell lines from the Achilles dataset utilized in this analysis. Table S4C shows the GSEA analysis for the enrichment of the indicated pathways in the analysis of the Achilles dataset.
**Supplemental Table S5**
Analysis of synthetic lethality with PI3K oncogenic mutations in the COLT-Cancer dataset. The SL Lethal score is shown as well as the p-value (Wilcoxon method) in Table S5A. Table S5B shows the list of cancer cell lines from the Cancer-COLT dataset utilized in this analysis. Table S5C shows the GSEA analysis for the enrichment of the indicated pathways in the analysis of the Cancer-COLT dataset.

**Supplemental Table S6**
Table S8 contains the analysis of synthetic lethality for KRAS oncogenic mutations in the Achilles dataset (KRAS Mut and WT cell lines were compared, Supplemental Table S6A). The synthetic lethality score is shown as well as the p-value (Wilcoxon method). This table also contains the output of GSEA analysis for the indicated pathways (Supplemental Table S6B).

**Supplemental Table S7**
Combined analyses of the screen in the isogenic cell line HCT116 and the screen datasets of the Achilles and COLT-Cancer datasets. The combined p-value was obtained using the Fisher method.

**Supplemental Table S8**
GSEA Analysis on the genes ranked by the combined p-value calculated for the synthetic lethality with PI3K mutations. Table S8A contains the GSEA analysis on the genes ranked by the combined p-value (Table S7). The table contains the output of GSEA (NAME: pathway name; SIZE: gene set size; ES: GSEA enrichment score; NES: GSEA normalized enrichment score). Please refer to http://www.broadinstitute.org/gsea/index.jsp for more details. Table S8B contains the Fisher exact test analysis for the enrichment of the indicated pathways within the 10% of the gene list ranked by the combined p-value. Right-tail p-value is shown.
**Supplemental Table S9**
Differential analysis of gene expression between PTEN-/- and PTEN WT cell lines from GEO dataset GDS2446 (Kim et al., 2007). Table S9A contains the gene list ranked by the p-value of the differential expression between PTEN -/- and PTEN +/+ cells. Supplemental Table S9B contains the GSEA analysis of the KEGG_PROTEASOME Pathway among the genes ranked by differential expression between PTEN -/- and PTEN +/+ cells shown in Table S9A.

**Supplemental Table S10**
TCGA dataset of colorectal cancer patients with PI3K SL signature score derived from RNAseq gene expression data. This table includes information about histological type, age, stage and survival. No information on the genetic status (mutation/copy number changes) of PI3K Pathway was available for most of these patients.

**Supplemental Table S11**
TCGA dataset of colorectal cancer patients with genetic status (mutation/copy number changes, see Materials and Methods) of PI3K Pathway (hyperactive or not). The PI3K SL signature score is derived from microarray gene expression data since no RNAseq was available for these patients. This table includes information about histological type, age, stage and survival.