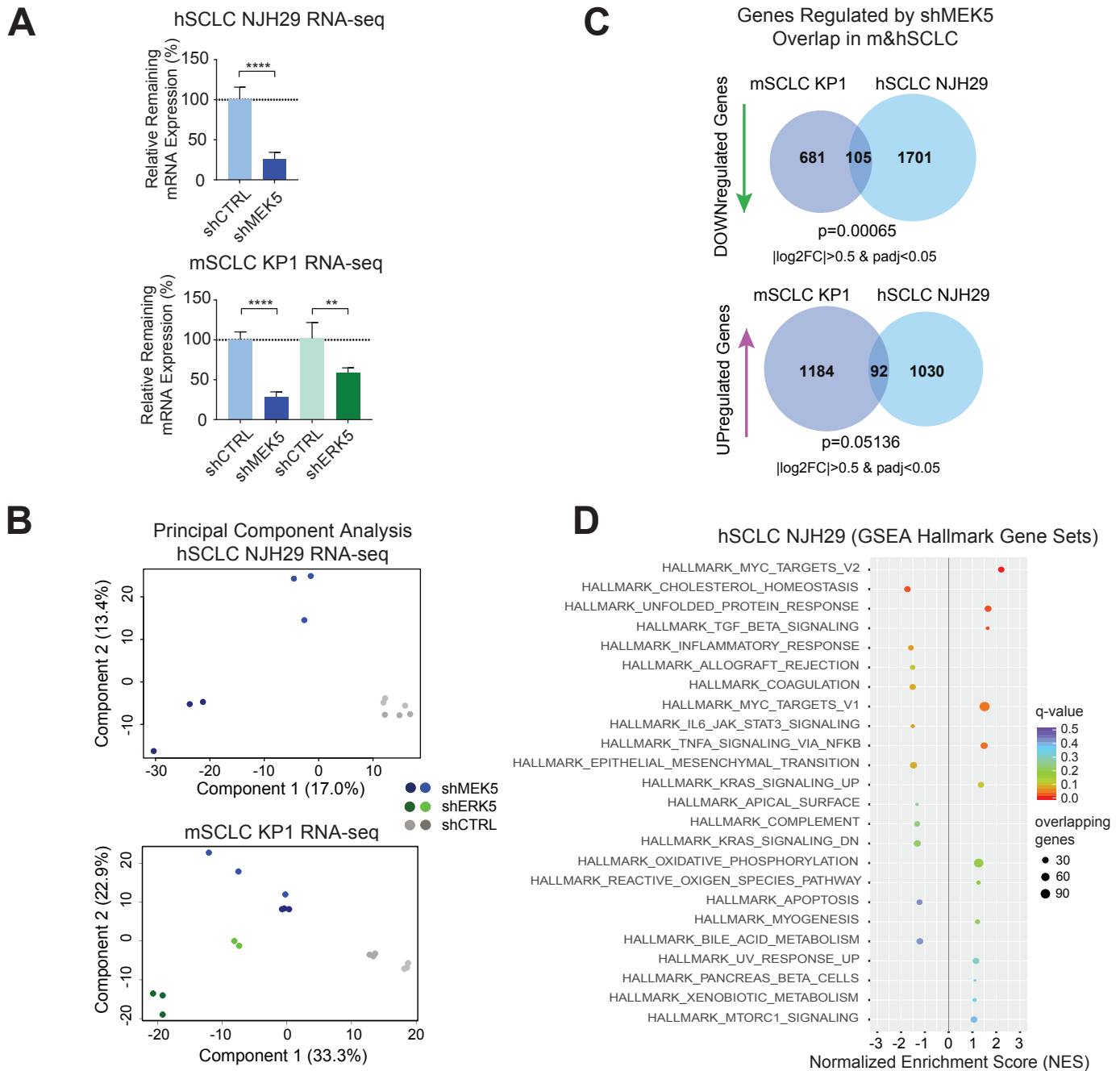


# Figure S3



**Figure S3: Analysis of human and mouse SCLC cells shows similar transcriptomic changes upon reduction of the MEK5-ERK5 axis**

**(A)** Relative remaining mRNA levels of MEK5 in hSCLC NJH29 cells (top) and both MEK5 and ERK5 after knockdowns in mSCLC KP1 cells (bottom), before subjection to RNA-seq analysis; mRNA levels for each gene are normalized to the average expression level of that gene in the shCTRLs (shGFP and shSCR samples), with two independent shRNAs per group and n=2-3 independent replicates per hairpin; \*\* signifies  $p<0.01$  and \*\*\*\*,  $p<0.0001$  (t-test).

**(B)** Principal Component Analysis of hSCLC NJH29 cells (top), and mSCLC KP1 cells (bottom), RNA-seq data clusters shMEK5 and shERK5 separate from shCTRLs.

**(C)** Genes downregulated (top) by shMEK5 in mSCLC KP1 (top left) and hSCLC NJH29 (top right) cells overlap significantly, while the overlap between genes upregulated (bottom) by shMEK5 in mSCLC KP1 (bottom left) and hSCLC NJH29 (bottom right) is just short of significance; significance is measured by the hypergeometric test; only genes with adjusted p-value  $<0.05$ , and  $|\log_2\text{FoldChange}|>0.5$  were included in these analyses.

**(D)** Hallmarks Gene Set Enrichment Analysis (GSEA) gene sets significantly enriched or disenriched when the MEK5 is knocked down in hSCLC NJH29 cells compared to shCTRLs; only enriched sets with q-values  $<0.5$  are shown.

# Figure S4

**A**

Cell Cycle  
Gene Set

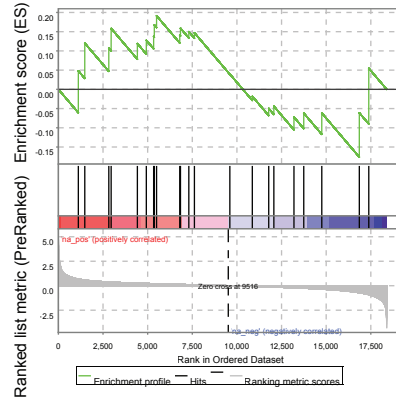
CCNA1  
CCNB1  
CCND1  
CCND2  
CCND3  
CCNE1  
CCNH  
CDC25A  
CDK1  
CDK2  
CDK4  
CDK6  
CDK7  
CDKN1A  
CDKN1B  
CDKN2A  
CDKN2B  
CDKN2C  
CDKN2D  
E2F1  
RB1  
RBL1  
TFDP1

NE  
Gene Set

BEX1  
ASCL1  
INSM1  
CHGA  
TAGLN3  
KIF5C  
CRMP1  
SCG3  
SYT4  
RTN1  
MYT1  
SYP  
KIF1A  
TMSB15A  
SYN1  
SYT11  
RUNDC3A  
TFF3  
CHGB  
FAM57B  
SH3GL2  
BSN  
SEZ6  
TMSB15B  
CELF3  
UCHL1  
CALCA  
PCSK1  
SNAP25  
ENO2  
PROP1

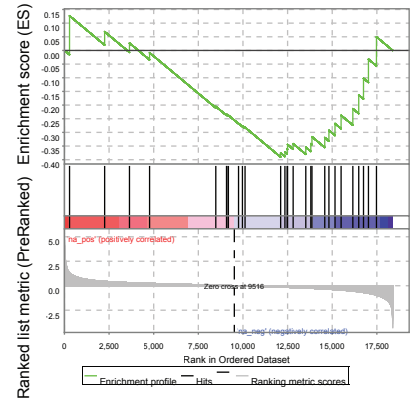
**B**

hSCLC NJH29 GSEA  
Cell Cycle Genes Enrichment Plot



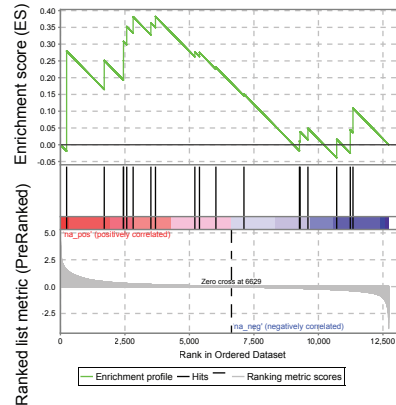
NES = 0.543, FDR = 0.967

hSCLC NJH29 GSEA  
NE Genes Enrichment Plot



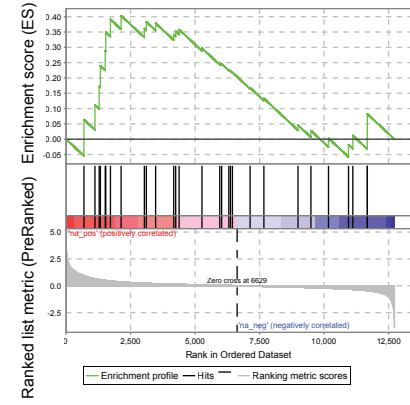
NES = -1.115, FDR = 0.296

mSCLC KP1 GSEA  
Cell Cycle Genes Enrichment Plot



NES = 0.896, FDR = 0.632

mSCLC KP1 GSEA  
NE Genes Enrichment Plot



NES = 1.030, FDR = 0.434

**Figure S4: MEK5-ERK5 axis role in SCLC cell viability is via control of pathways other than cell cycle or neuroendocrine identity programs**

**(A)** Cell Cycle and Neuroendocrine (NE) Gene Sets used to assay enrichment of terms through the GSEA (Gene Set Enrichment Analysis) platform.

**(B)** Cell Cycle Genes (left) and NE Genes (right) Enrichment plots from GSEA analysis with gene set lists from (A) on data from hSCLC NJH29 RNA-seq after MEK5 knockdown and DESeq2 analysis compared to shCTRLs (top), and mSCLC KP1 RNA-seq after MEK5-ERK5 axis knockdown and DESeq2 analysis compared to shCTRLs (bottom); normalized enrichment score (NES) and FDR p-value from GSEA analysis marked under each plot.