

Expanded View Figures

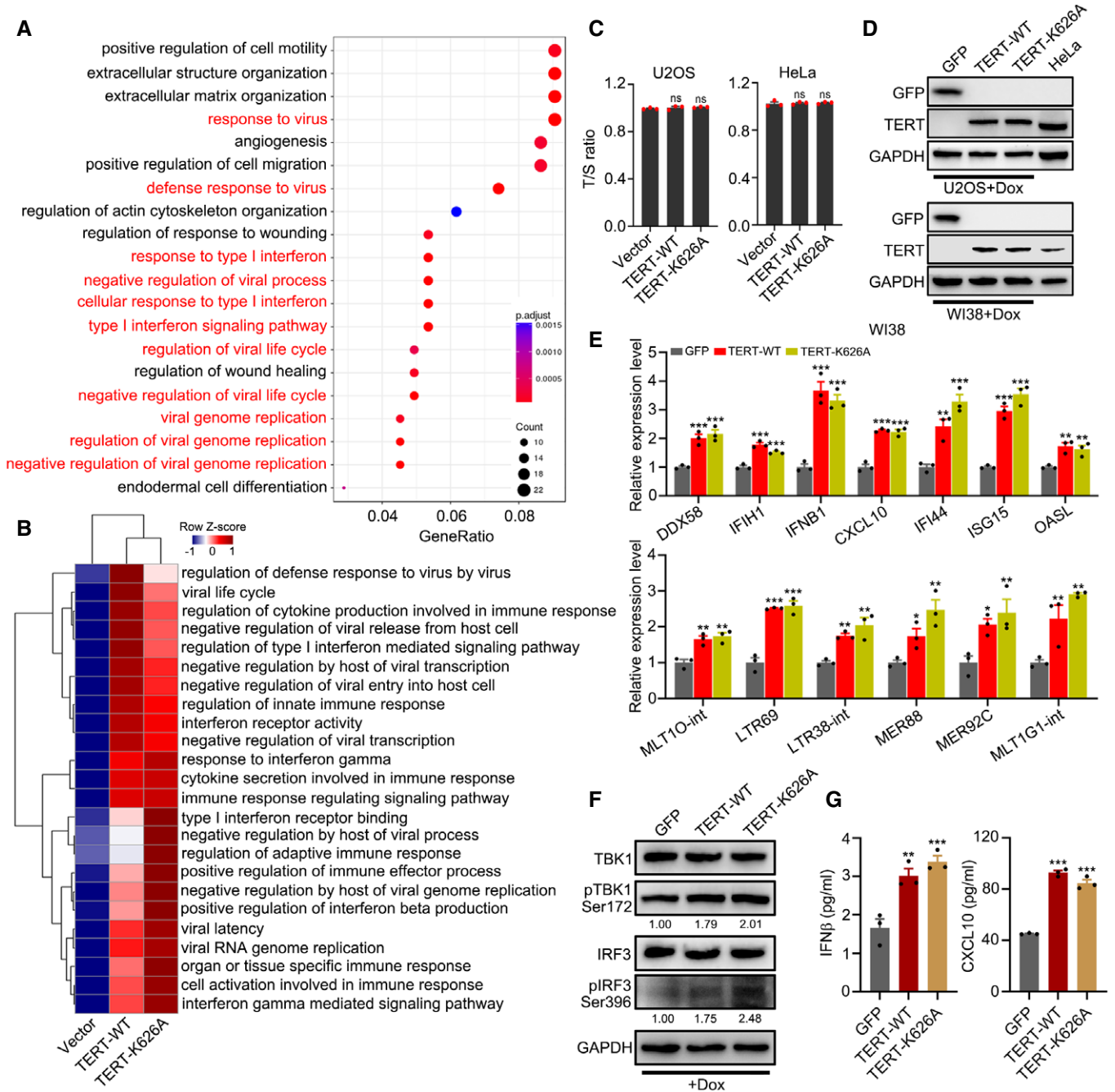


Figure 1.

Figure EV1. Ectopic expression of TERT triggers interferon response.

- A Enriched biological processes (BP) with TERT ectopic expression. GO was analysed for upregulated genes by TERT compared to those in the empty vector control. GO terms in red are biological processes related to interferon response.
- B ssGSEA of GO terms related to interferon response in U2OS cells ectopically expressing TERT-WT or TERT-K626A.
- C T/S ratio of U2OS and HeLa cells with transient transfection of TERT-WT or TERT-K626A.
- D Western blot analysis of GFP, TERT and GAPDH levels in U2OS (upper) and WI38 (lower) cells ectopically expressing TERT-WT or TERT-K626A using Dox-inducible system to induce expression of TERT at the levels comparable to that of HeLa cells. GFP as negative control.
- E–G RT–qPCR analysis of interferon-related genes and TA-ERVs (E), western blot analysis of TERT, TBK1, pTBK1, IRF3, pIRF3, and GAPDH levels (F), and ELISA of IFN β and CXCL10 in culture supernatant (G) in WI38 cells ectopically expressing TERT-WT or TERT-K626A using Dox-inducible system. GFP as negative control. Relative pTBK1 and pIRF3 protein levels were quantified with ImageJ software and normalized to GAPDH, as indicated at the bottom of the blot (F).

Data information: Data represent mean \pm SEM of three biological replicates (C, E and G). * $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$, ns, not significant, using one-way ANOVA with Fisher's LSD tests (C, E and G).

Source data are available online for this figure.

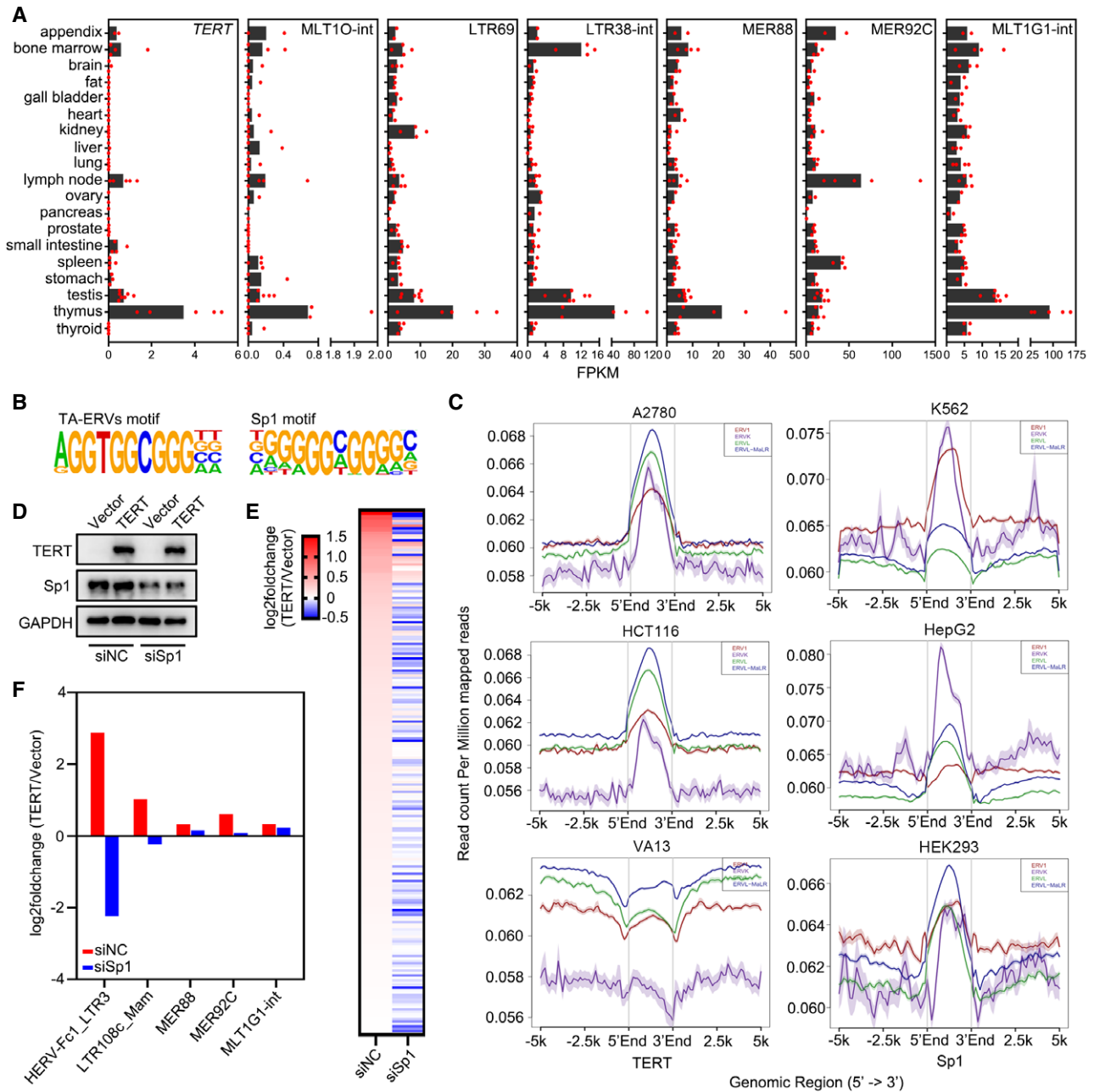


Figure EV2. TERT activates ERVs via interaction with Sp1.

A Expression of *TERT* and ERVs in human tissues using RNA-seq data from the GEO database. Data represent average expression levels of *TERT* and ERVs. Appendix, $n = 2$. Bone marrow, $n = 4$. Brain, $n = 3$. Fat, $n = 3$. Gall bladder, $n = 3$. Heart, $n = 3$. Kidney, $n = 4$. Liver, $n = 3$. Lung, $n = 4$. Lymph node, $n = 5$. Ovary, $n = 2$. Pancreas, $n = 2$. Prostate, $n = 4$. Small intestine, $n = 4$. Spleen, $n = 4$. Stomach, $n = 3$. Testis, $n = 7$. Thymus, $n = 5$. Thyroid, $n = 4$.

B Motif analysis using the sequences of representative TA-ERVs.

C ChIP-seq analysis for TERT and Sp1 from the GEO database. Average binding profiles of TERT on ERV1, ERVK, ERVL, and ERVL-MaLR in A2780 and HCT116 cells, VA13 cells as the negative control (left); and Sp1 binding profiles in K562, HepG2, and HEK293 cells (right).

D-F Western blot analysis of TERT and Sp1 expression (D), log₂ fold change of ERVs plotted as heat map (E), and log₂ fold change of representative ERVs (F) in siNC or siSp1 transfected U2OS cells expressing TERT versus control vector.

Source data are available online for this figure.

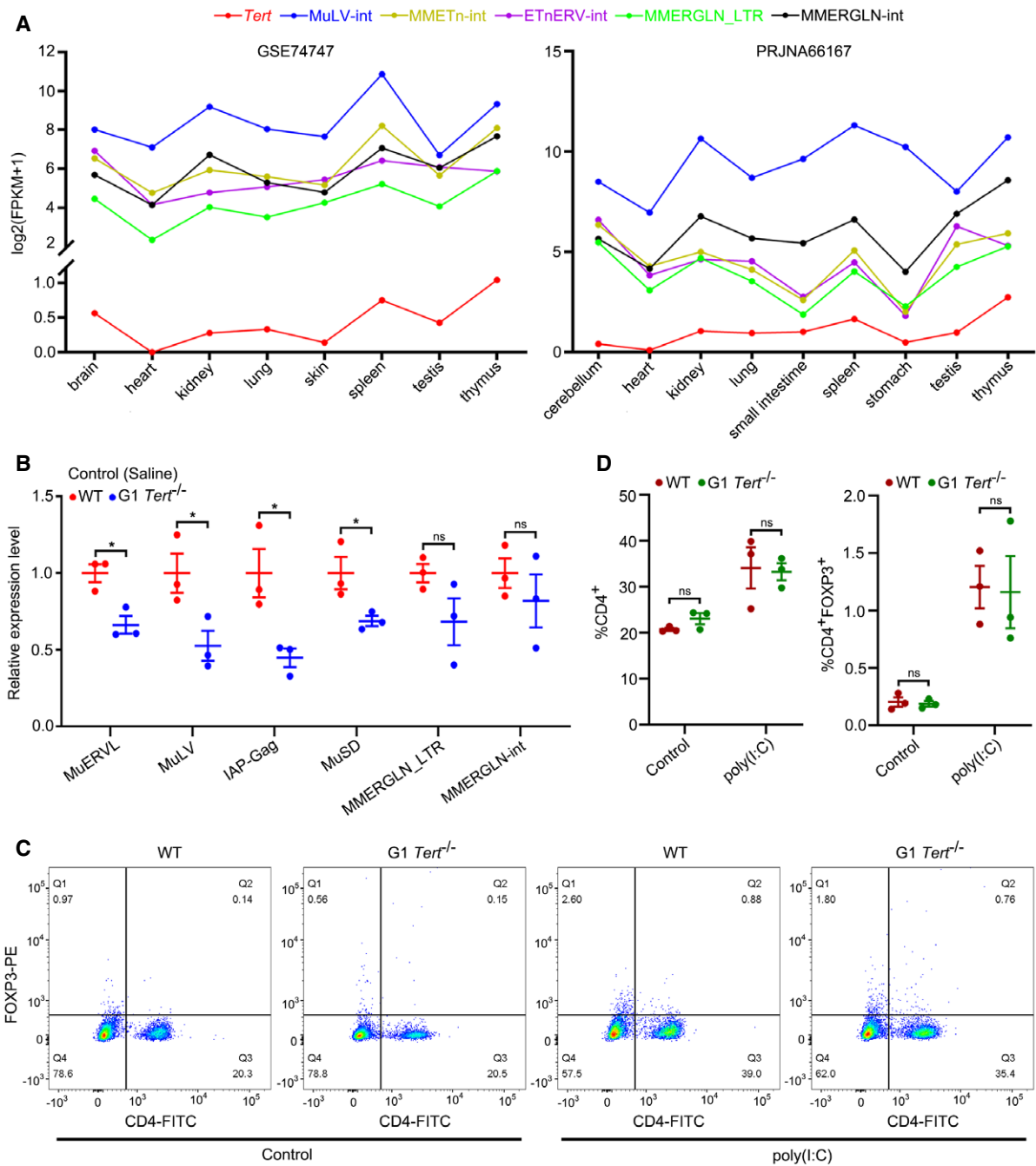


Figure EV3. Expression of *Tert* and ERVs in mice.

A Expression analysis of *Tert* and ERVs in mouse tissues using the GEO.

B RT-qPCR analysis of ERV expression in livers of WT and *G1 Tert*^{-/-} mice treated with saline (control). *n* = 3 mice per group.

C, D Representative flow cytometry images (C) and statistical analyses (D) of CD4⁺ and CD4⁺FOXP3⁺ cells among mouse peripheral blood mononuclear cells (PBMCs). *n* = 3 mice per group.

Data information: Data represent mean ± SEM. **P* < 0.05, ns, not significant. Unpaired two-tailed Student's *t*-tests were used for (B), and two-way ANOVA with Fisher's LSD tests was used for (D).

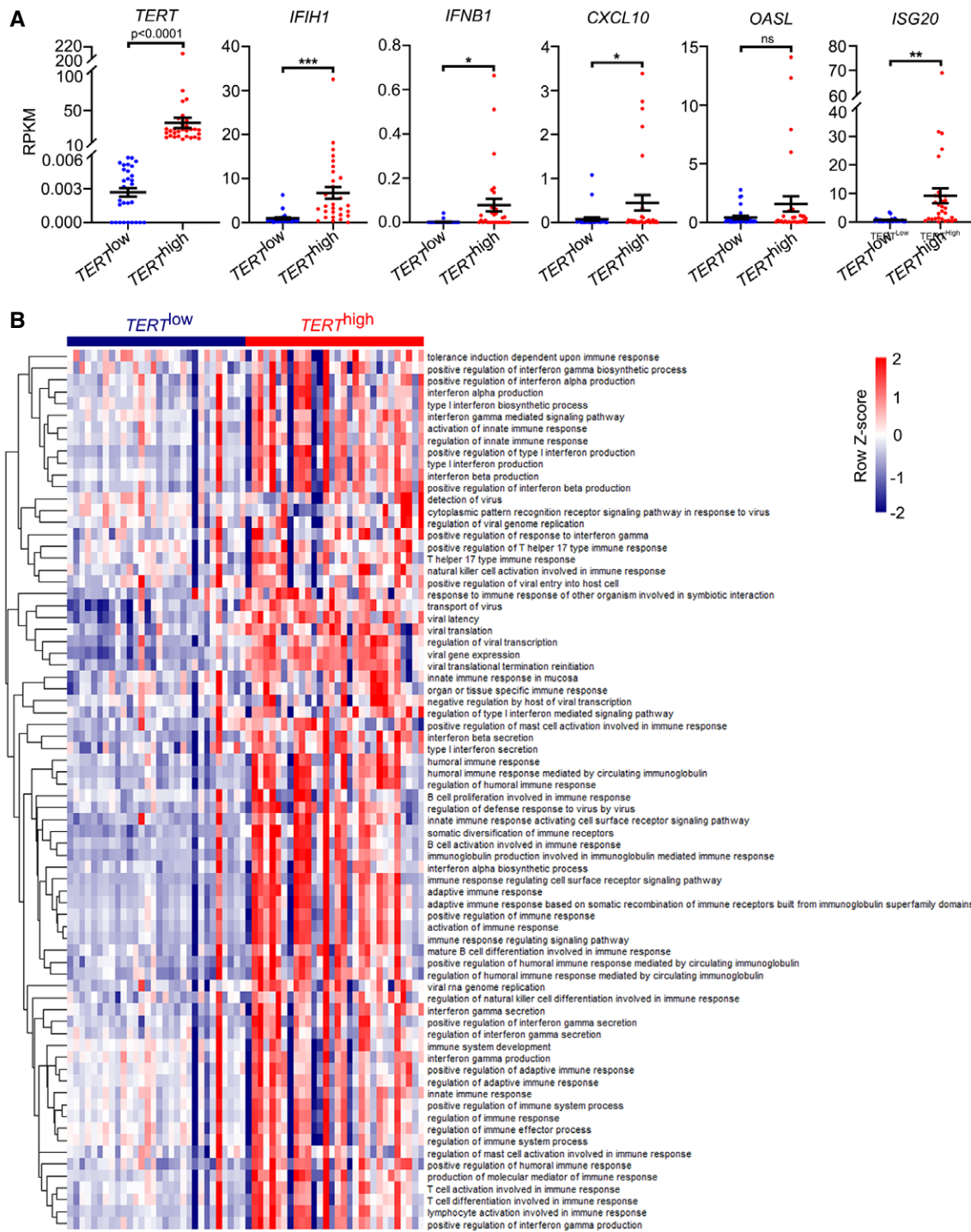


Figure EV4. Analysis of gene expression and ssGSEA in the Cancer Cell Line Encyclopaedia (CCLE).

A CCLE reads per kilobase million (RPKM) values of *TERT*, *IFIH1*, *IFNB1*, *CXCL10*, *ISG20*, and *OASL* in cancer cell lines grouped in $TERT^{high}$ and $TERT^{low}$ groups ($n = 30$ each). Data represent mean \pm SEM. $*P < 0.05$, $**P < 0.01$, $***P < 0.001$, ns, not significant, using unpaired two-tailed Student's *t* test.

B ssGSEA of GO terms related to interferon response in $TERT^{high}$ and $TERT^{low}$ cancer cell lines ($n = 30$, respectively) across CCLE.

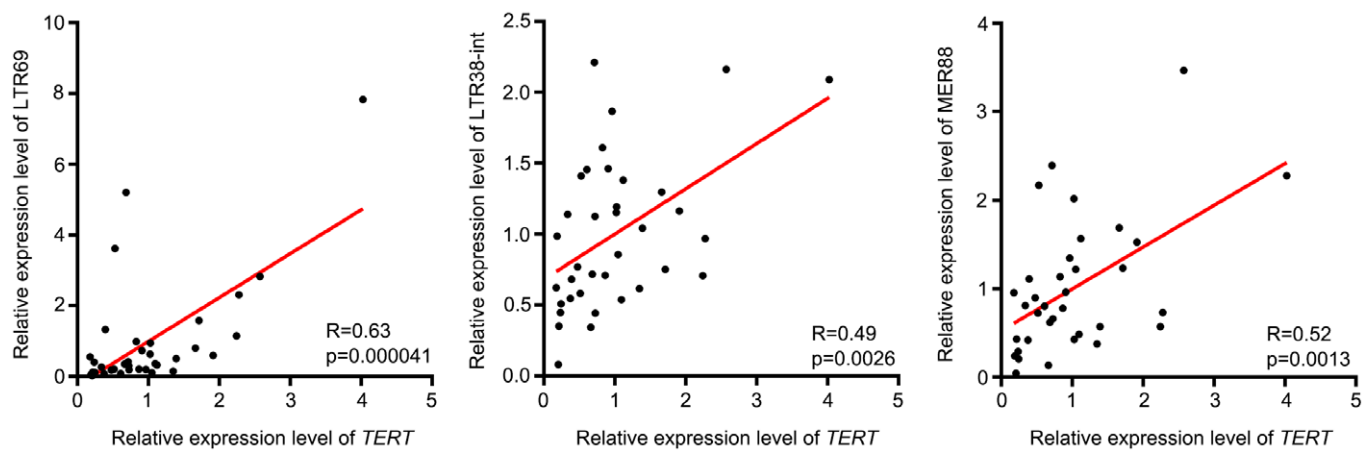


Figure EV5. Regression analysis of the correlation between *TERT* and LTR69, LTR38-int and MER88 expression in colon tumours.

The expression of *TERT*, and LTR69, LTR38-int and MER88 were determined using RT-qPCR. $n = 36$.