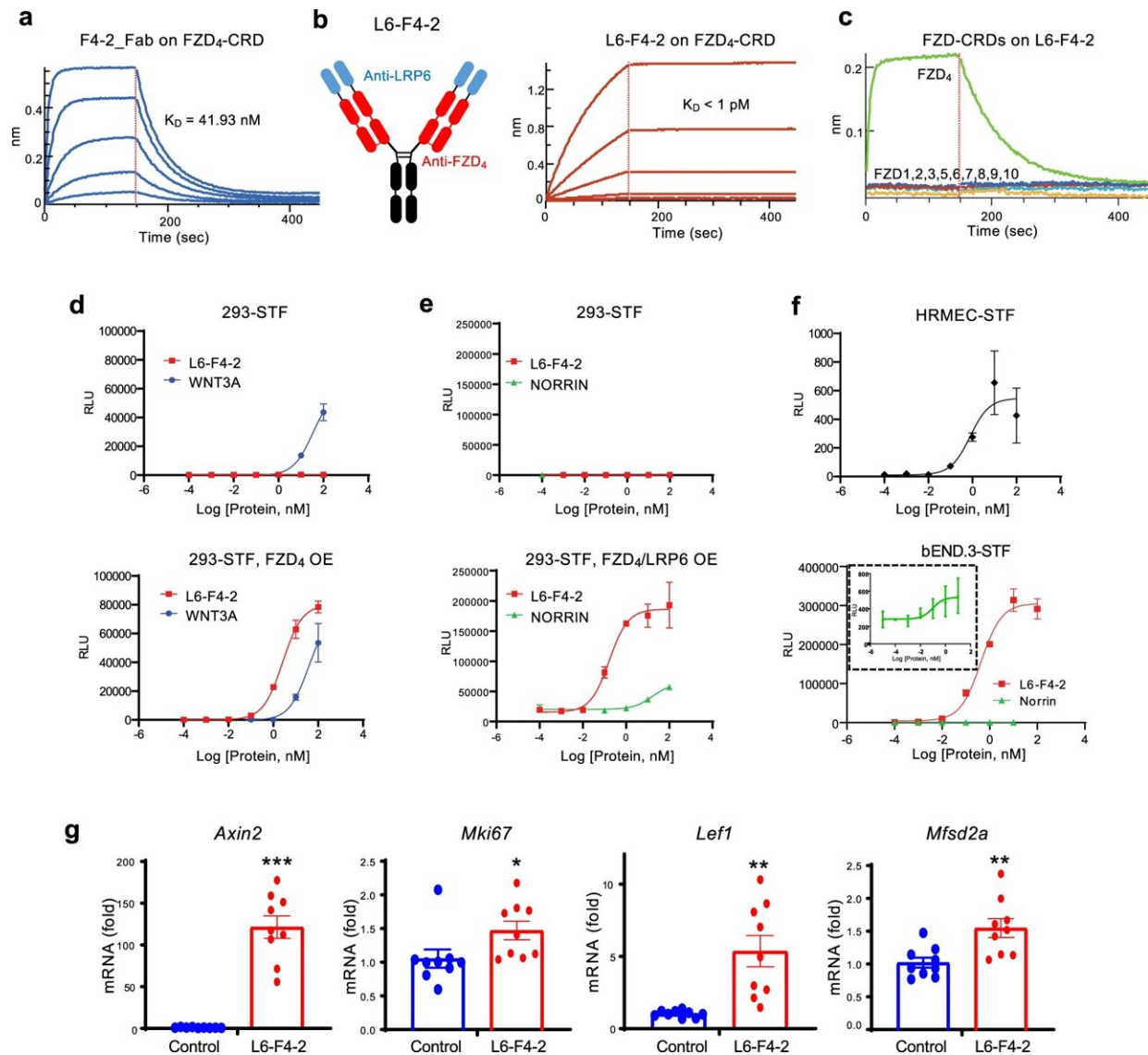


Restoring the blood-brain barrier using a novel WNT signaling pathway

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Characterization of the monoFZD₄-specific WNT/β-catenin signaling surrogate, L6-F4-2. **a** Binding affinity of the recombinant F4-2_Fab molecule to FZD₄

CRD measured by BLI assay. Dotted lines indicate the global fits generated by using a 1:1 Langmuir binding model. **b** Schematic of L6-F4-2 and the binding affinity of the L6-F4-2 molecule to the FZD₄ CRD measured by BLI assay. Dotted lines indicate the global fits generated by using a 1:1 Langmuir binding model. **c** The binding specificity of L6-F4-2 against all ten FZD CRDs was examined by BLI assay. **d** Dose-dependent STF activity of L6-F4-2 or WNT3A in 293STF cells (top) or FZD₄-transfected 293STF cells (bottom, OE = overexpression). **e** Dose-dependent STF activity of L6-F4-2 or Norrin in 293STF cells (top) or cells transfected with both FZD₄ and LRP6 (bottom, OE = overexpression). **f** Dose-dependent STF activities of L6-F4-2 in HRMEC cells (top) and bEnd.3 cells (bottom). Inset box in the bEnd.3-STF graph is an enlarged plot of the Norrin response. **g** Quantitative RT-PCR of *Axin2*, *Mki67*, *Lef1* and *Mfsd2a* gene expression in bEnd.3 cells. mRNA expression values were normalized by *Actb* gene expression. Results are from three independent experiments. Graphs are shown as mean \pm SEM; **p*

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