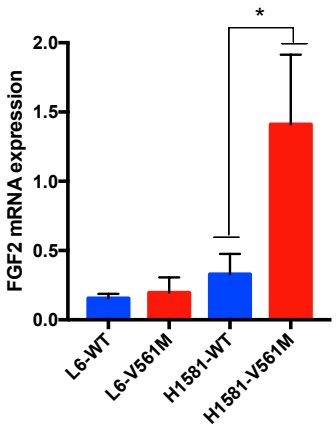
Figure S1: qPCR of FGF2 mRNA expression in H1581 and L6 cells



For end-point qPCR, RNA was isolated from L6-WT, L6-V51M, H1581-WT and H1581-V561M cells using the Qiagen RNeasy mini kit. Contaminating genomic DNA was removed with recombinant DNase I, which was then inhibited using EDTA and heat. cDNA was generated using the BioRad iScript cDNA synthesis kit. qPCR was performed on the BioRad CFX96 Touch Real-Time PCR system using the BioRad iTaq SYBR Green mix. Primers were as follows:
L6 GAPDH F: 5'-GATGGTGAAGGTCGGTGTGA-3';
L6 GAPDH R: 5'-TTGAGGTCAATGAAGGGGTC-3';
H1581 GAPDH F: 5'-GAAGGTGAAGGTCGGAGTCA-3';
H1581 GAPDH R: 5'-TTGAGGTCAATGAAGGGGTC-3';
L6 FGF2 F: 5'-GTGTGTGCGAACCGGTACCT-3';
L6 FGF2 R: 5'-GCTCTTAGCAGACATTGGAAG-3';
H1581 FGF2 F: 5'-GTGTGTGCTAACCGTTACCT-3';
H1581 FGF2 R: 5'-GCTCTTAGCAGACATTGGAAG-3'

Table S1 and S2 Proliferation after various timepoints

	L6 WT			L6 V561M				
	Initial cell count	Final cell count	% Proliferation	Initial Cell Count	Final Cell Count	% Proliferation	Fold-increase in V561M cells	P-value
72 h	5000	7000	140	5000	15000	300	2	0.03
	9000	18000	200	6000	26000	433.33		
	5000	8000	160	6000	17000	283.33		
96 h	8000	26000	325	4000	43000	1075	4.4	0.00004
	4000	23000	575	6000	61000	1016.67		
	5000	18000	360	5000	50000	1000		
120 h	5000	47000	940	5000	89000	1780	2.5	0.03
	4000	30000	750	7000	130000	1857.14		
	5000	46000	920	4000	110000	2750		
168 h	5000	350000	7000	3000	660000	22000	2.7	0.03
	5000	200000	4000	5000	790000	15800		
	4000	340000	8500	4000	630000	15750		

	H1581-WT			H1581-V561M				
	Initial cell count	Final cell count	% Proliferation	Initial Cell Count	Final Cell Count	% Proliferation	Fold-increase in V561M cells	P-value
72 h	5000	13000	260	6000	44000	733.33	3	0.003
	7000	12000	171.43	5000	34000	680		
	4000	13000	325	4000	32000	800		
96 h	5000	24000	480	5000	46000	920	2	0.0003
	8000	35000	437.5	4000	40000	1000		
	6000	30000	500	7000	67000	957.14		
120 h	9000	85000	944.44	6000	120000	2000	2	0.04
	6000	110000	1833.33	5000	150000	3000		
	6000	77000	1283.33	5000	140000	2800		
168 h	6000	330000	5500	5000	570000	11400	2.75	0.01
	7000	240000	3428.57	6000	1000000	16666.67		
	4000	260000	6500	5000	720000	14400		

Table S1 and S2 Proliferation after various timepoints
Cells were starved for 12 h, then counted using the Countess automated cell counter. Approximately 5000 cells were plated in each well of a 6-well plate. Actual cell counts for each well were recorded, and cells were incubated for 72h-168h before trypsinization and counting using the Countess automated cell counter. % cell proliferation was calculated for each well based on initial and final cell counts.

Figure S2: STAT3 knockdown sensitizes H1581-V561M cells to AZD4547 treatment.

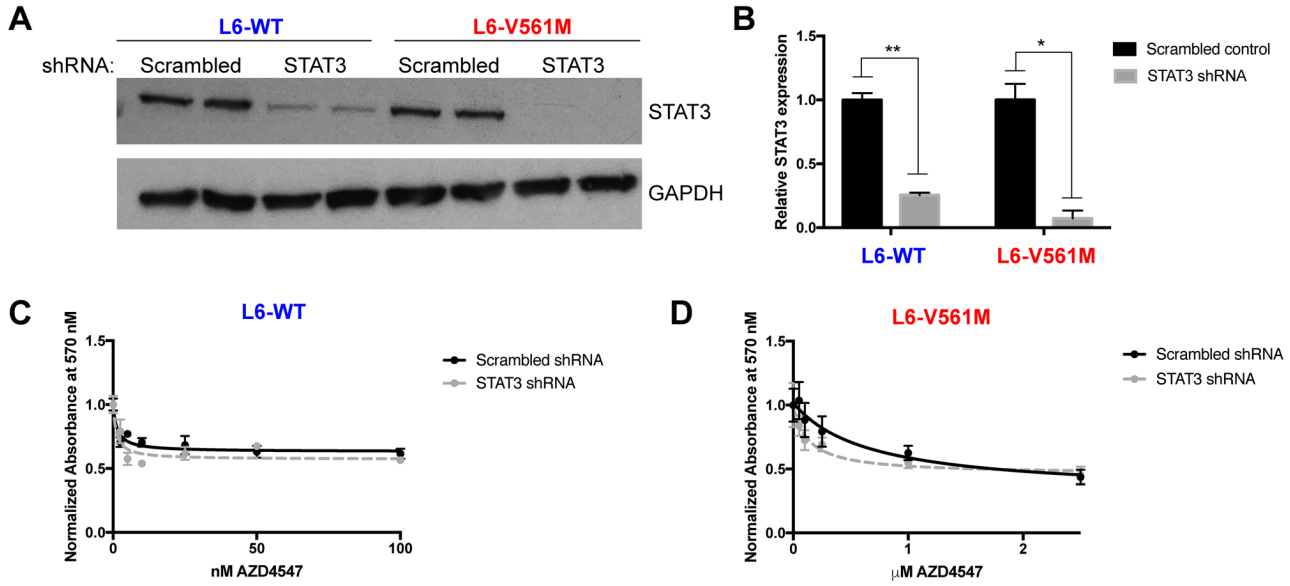


Figure S2: STAT3 knockdown sensitizes H1581-V561M cells to AZD4547 treatment.
A L6-WT and L6-V561M cells were infected with virus containing shRNA targeting STAT3 or a non-targeting scrambled control. Lysates were harvested and immunoblots of two replicate lysates were performed as described in 2A. **B** STAT3 expression levels were quantified using ImageJ and normalized to a GAPDH loading control and plotted using GraphPad Prism. **C, D** IC_{50} curves for AZD4547 treatment of L6-WT and L6-V561M cells treated with scrambled control or STAT3 targeting shRNA were generated as described for Figure 1. Significance values: *<0.05; **<0.01, ***<0.001.

Table S3: AZD4547 sensitivity of L6 shRNA-treated cells

	IC_{50} [scrambled] (nM)	IC_{50} [shRNA *STAT3*] (nM)	Fold-change
L6-WT	1.4 ± 0.6	1.2 ± 0.7	Within error
L6-V561M	700 ± 400 (0.7 μM)	140 ± 50	↓5
Fold-change	500	117	--