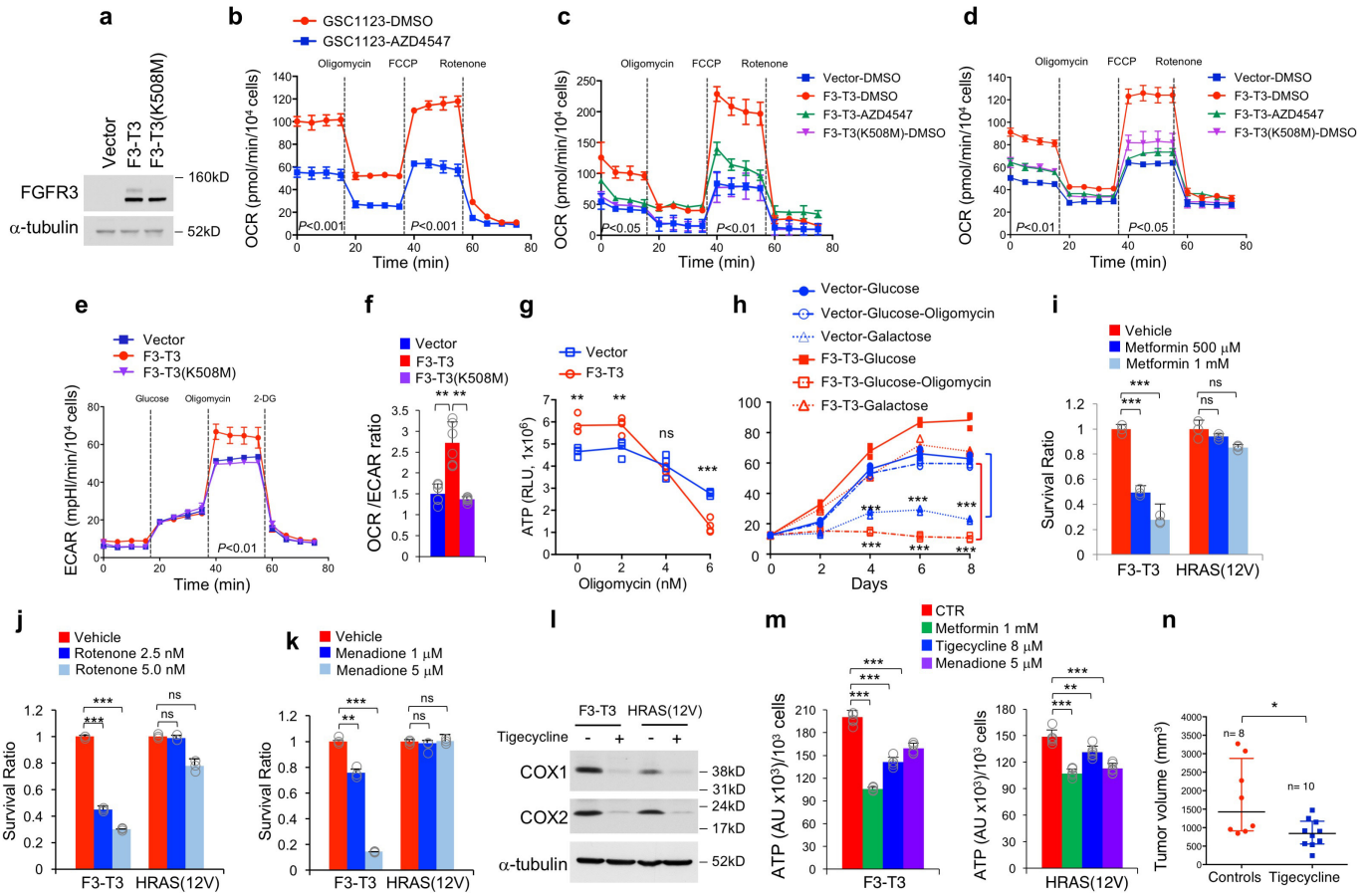

Extended Data Figure 2 | F3–T3 induces sensitivity to inhibitors of mitochondrial metabolism. a, Immunoblot analysis using the FGFR3 antibody in human astrocytes expressing vector, F3–T3 or F3–T3(K508M). α-Tubulin is shown as a loading control. Experiment was repeated five times with similar results. **b**, OCR of GSC1123 cells expressing F3–T3 in the presence or absence of AZD4547. Data are mean ± s.d. ($n = 6$ technical replicates) of one representative experiment out of two independent experiments. $P < 0.001$ for rate 1–4 and 9–12, two-tailed t-test with unequal variance. **c**, OCR of RPE cells expressing F3–T3, F3–T3(K508M) or the empty vector in the presence or absence of AZD4547. Data are mean ± s.d. ($n = 3$ technical replicates) of one representative experiment out of three independent experiments performed in triplicate with similar results. $P < 0.05$ for rate 1–4; $P < 0.001$ for rate 9–12; two-tailed t-test with unequal variance. **d**, OCR of U251 cells expressing F3–T3, F3–T3(K508M) or the empty vector in the presence or absence of AZD4547. Data are mean ± s.d. ($n = 3$ technical replicates) of one representative experiment out of two independent experiments performed in triplicate with similar results. $P < 0.01$ for rate 1–4; $P < 0.001$ for rate 9–12; two-tailed t-test with unequal variance. **e**, ECAR of human astrocytes expressing F3–T3, F3–T3(K508M) or the empty vector. Data are mean ± s.d. ($n = 3$ technical replicates) of one representative experiment out of two independent experiments performed in triplicate with similar results. $P < 0.01$ for rate 9–12; two-tailed t-test with unequal variance. **f**, Ratio between OCR (rate 4) and ECAR (rate 8) in human astrocytes expressing F3–T3, F3–T3(K508M) or vector. Data are mean ± s.d. ($n = 6$ replicates) of two independent experiments each performed in triplicate. $P < 0.01$; two-tailed t-test with unequal variance. **g**, Quantification of ATP production in human astrocytes expressing F3–T3 or vector following treatment with the indicated concentrations of oligomycin for 72 h. Data are independent technical replicates ($n = 4$) and means (connecting lines) of one representative experiment out of two independent experiments performed with similar results. $^{**}P < 0.01$; $^{***}P < 0.001$; two tailed t-test with unequal variance. **h**, Time-course analysis of cellular growth of human astrocytes expressing F3–T3 or vector cultured in the presence of glucose (25 mM) or galactose (25 mM) with or without oligomycin (100 nM). Data are independent technical replicates ($n = 3$) of one representative experiment out of two independent experiments performed with similar results. $^{***}P < 0.001$; two-tailed t-test with unequal variance. **i–k**, Survival ratio of F3–T3;shTrp53 and HRAS(12V);shTrp53 mGSCs treated for 72 h with vehicle or metformin (**i**), rotenone (**j**) or menadione (**k**) at the indicated concentrations. Data are mean ± s.d. ($n = 3$ technical replicates) of one representative experiment out of two independent experiments performed with similar results. $^{**}P < 0.01$; $^{***}P < 0.001$; two-tailed t-test with unequal variance. **l**, Western blot analysis of COX1 and COX2 proteins in F3–T3;shTrp53 and HRAS(12V);shTrp53 mGSCs treated with vehicle or tigecycline at a concentration of 8 μM for 72 h. α-Tubulin is shown as a loading control. Experiment was independently repeated twice with similar results. **m**, Quantification of cellular ATP in F3–T3;shTrp53 (left) and HRAS(12V);shTrp53 (right) mGSCs treated with vehicle or metformin (1 mM), tigecycline (8 μM) or menadione (5 μM) for 16 h. Data are mean ± s.d. of one experiment ($n = 6$ technical replicates). $^{**}P < 0.01$; $^{***}P < 0.001$; two-tailed t-test with unequal variance. **n**, Quantification of tumour volume of F3–T3;shTrp53 mGSCs in control and tigecycline-treated mice. Data are tumour volumes (median with interquartile range) at day 6 of treatment, a time when all mice were still in the study; $n = 8$ for control (median = 1,427 mm^3) and $n = 10$ for tigecycline-treated mice (median = 843.4 mm^3). $^{*}P < 0.05$; two-sided Mann–Whitney U-test. Molecular weights are indicated in immunoblots.