Supplementary materials

Fructose treatment induces metabolic reprogramming in liver cancer cells, promoting aggressiveness and platin therapy failure

Table S1. Primers used in the study

Gene	Forward	Reverse
khk-a (3a exon)	5´- tattctgtggacctacgcta	5´- ccgcaccatcctatactatg
khk-c (3c exon)	5´- catgttgctgacttcctgg	5´- tgcatcatcaacaactccaa
rps18	5´- gaggatgaggtggaacgtgt	5´- agaagtgacgcagccctcta

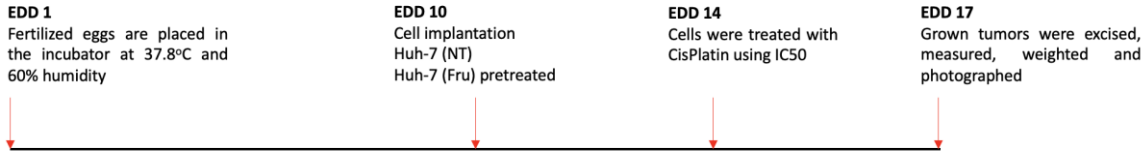
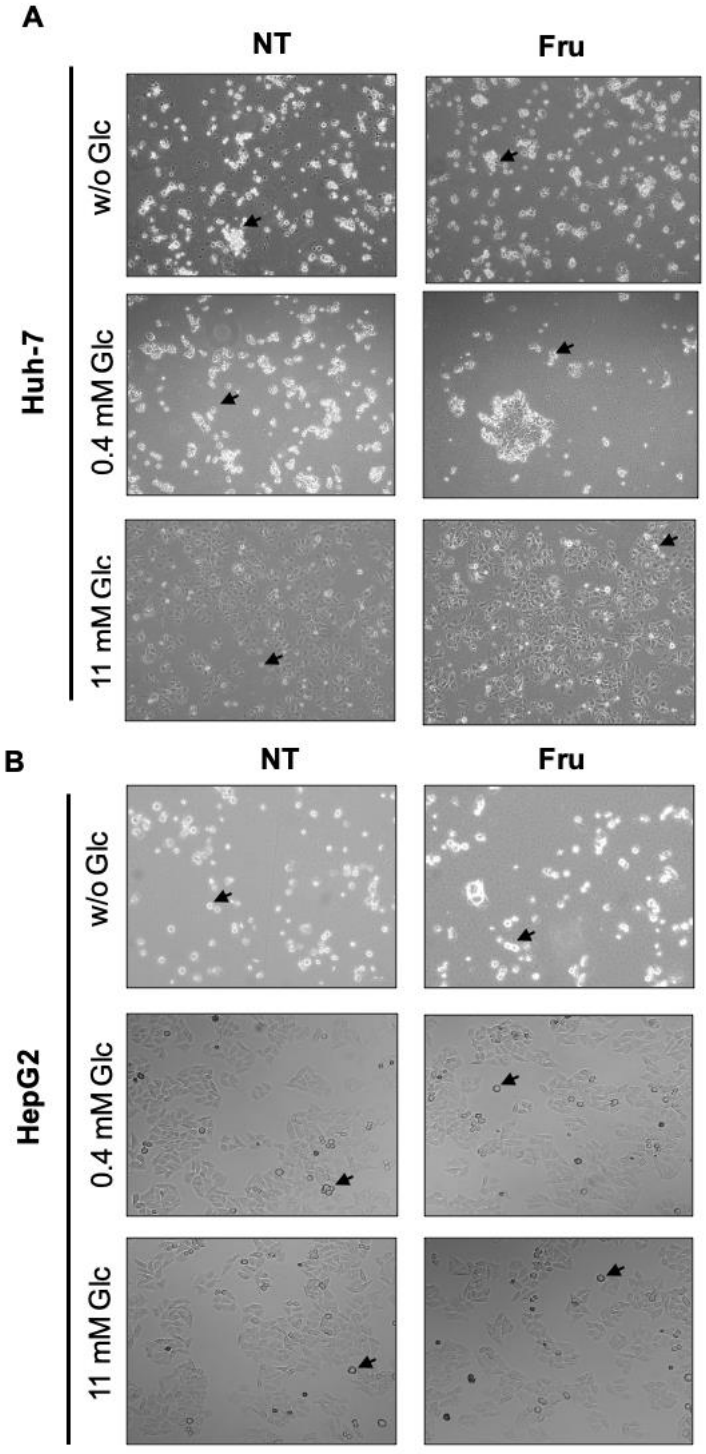


Figure S1. Chorioallantoic Membrane (CAM) model in chick embryo. Followed protocol for in vivo studies with CAM model and chemoresistance properties of Huh-7 cells treated with High Glc media (11 mM) and fructose (1mM) five days before the grafting. On Embryonic Development Day 14 (EDD14), tumors from NT or fructose-treated cells were treated with PBS, fructose (Fru, 1mM), Cisplatin (CDDP, 100µM) or CDDP + Fru (100 µM and 1 mM). The treatments finished on EDD17.


A
NT
Fru
w/o Glc
0.4 mM Glc
11 mM Glc
Huh-7
B
NT
Fru
w/o Glc
0.4 mM Glc
11 mM Glc
HepG2

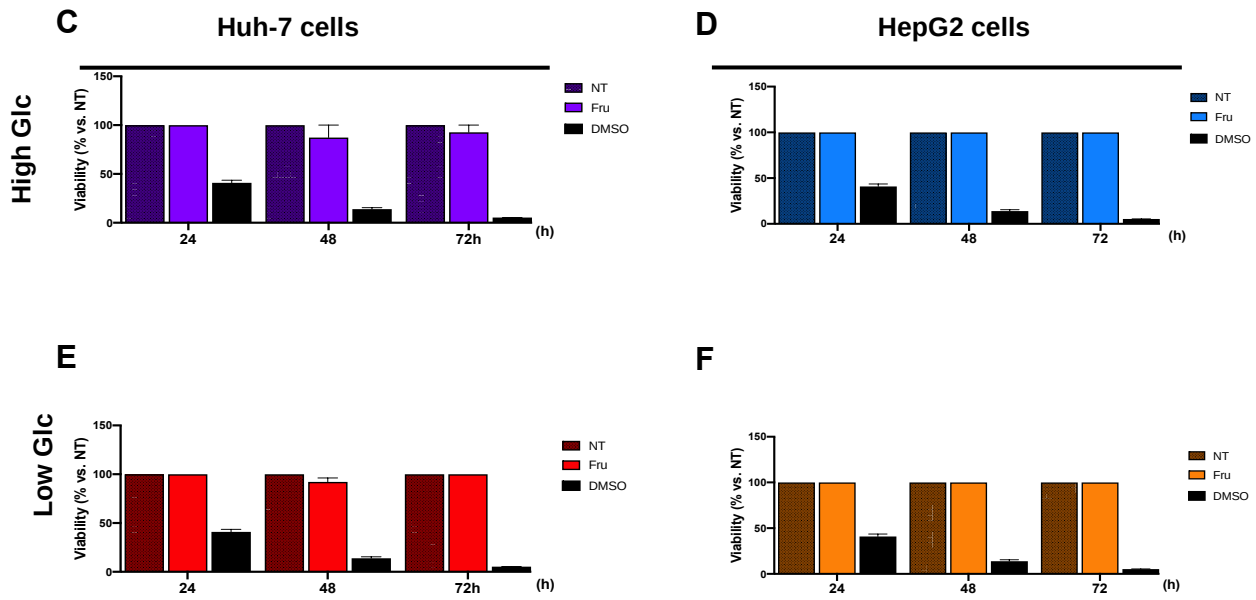


Figure S2. Cells in a glucose-free medium change their morphology and reduce their metabolism. (**A**) Huh-7 cell line morphology treated in a DMEM Glucose-free media at 200×; (**B**) HepG2 cell line morphology before the Glycolytic Rate Assay at 200×, Black arrows show dead cells; (**C**) Viability of Huh-7 and (**D**) HepG2 cells treated in a High Glc media; (**E**) Viability of Huh-7 and (**F**) HepG2 cells treated in a Low Glc media. Treatments: High Glucose: DMEM complete media with Glucose (11 mM); Low Glucose: DMEM complete media with Glucose (0.4 mM); w/o Glc: MEM complete media without Glucose; Fructose 1 mM for 48 h. DMSO (30%) was used as a positive control for viability.

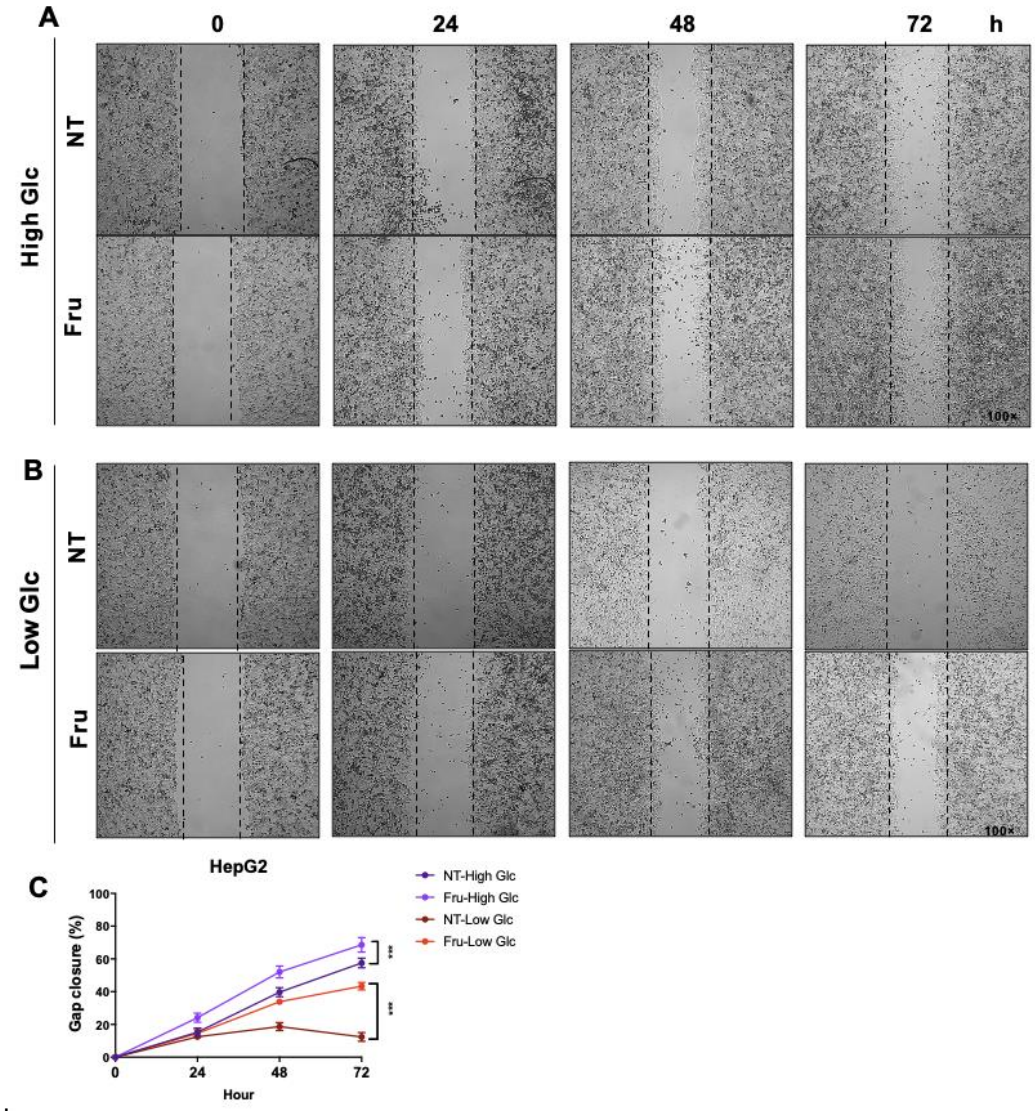


Figure S3. Metabolic rewiring sustains the proliferation and survival of liver cancer cells. (**A**) Wound-healing assay to address HepG2 cells migration with High Glc media; (**B**) Wound-healing assay to address HepG2 cells migration with Low Glc media; (**C**) Quantification of the rate of gap closure. Treatments: we used 1 mM of fructose in DMEM complete media with High Glc (11mM) or Low Glc (0.4 mM). Images are representative of at least three independent experiments. * $p \leq 0.05$, *** $p \leq 0.001$, **** $p \leq 0.0001$.