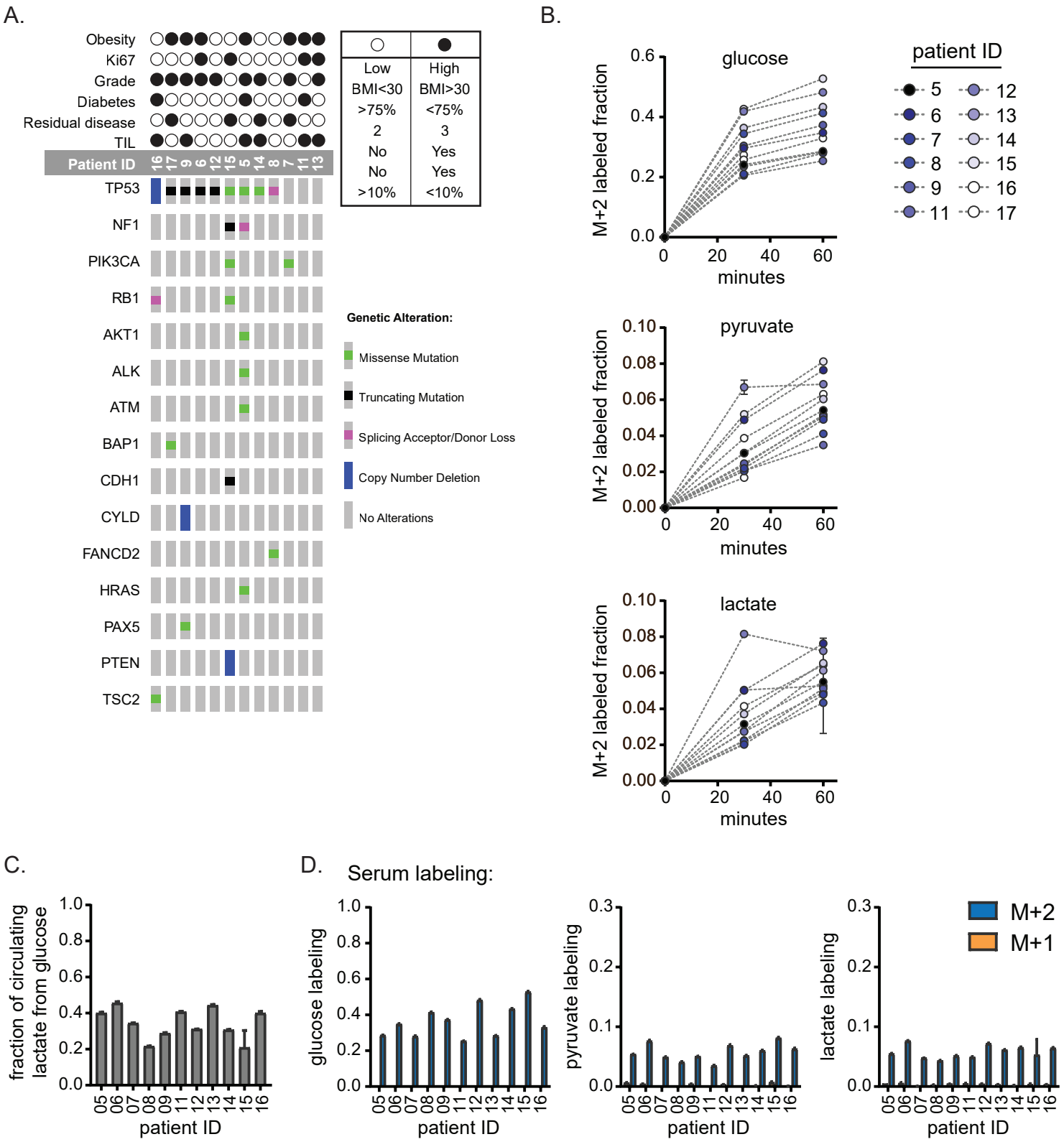

Figure S1. Patient clinical data and serum metabolite labeling related to Figure 1. A) Summary of individual patient clinical data classifications and Oncoprint depicting tumor driver variants obtained from whole exome sequencing. See also Table S1. B) Individual patient M+2 serum labeling of glucose, pyruvate and lactate from [1,2-^{13}C]glucose infusion (mean ± SEM, n = 3 technical replicates). C) Fractional carbon labeling of circulating lactate normalized to that of circulating glucose from individual TNBC patients after 1 h of infusion (mean ± SEM, n = 3; data for patient 17 are missing at this time-point due to insufficient serum collection). D) Isotope labeling patterns of serum glucose, pyruvate and lactate from [1,2-^{13}C]glucose (mean ± SEM, n = 3).

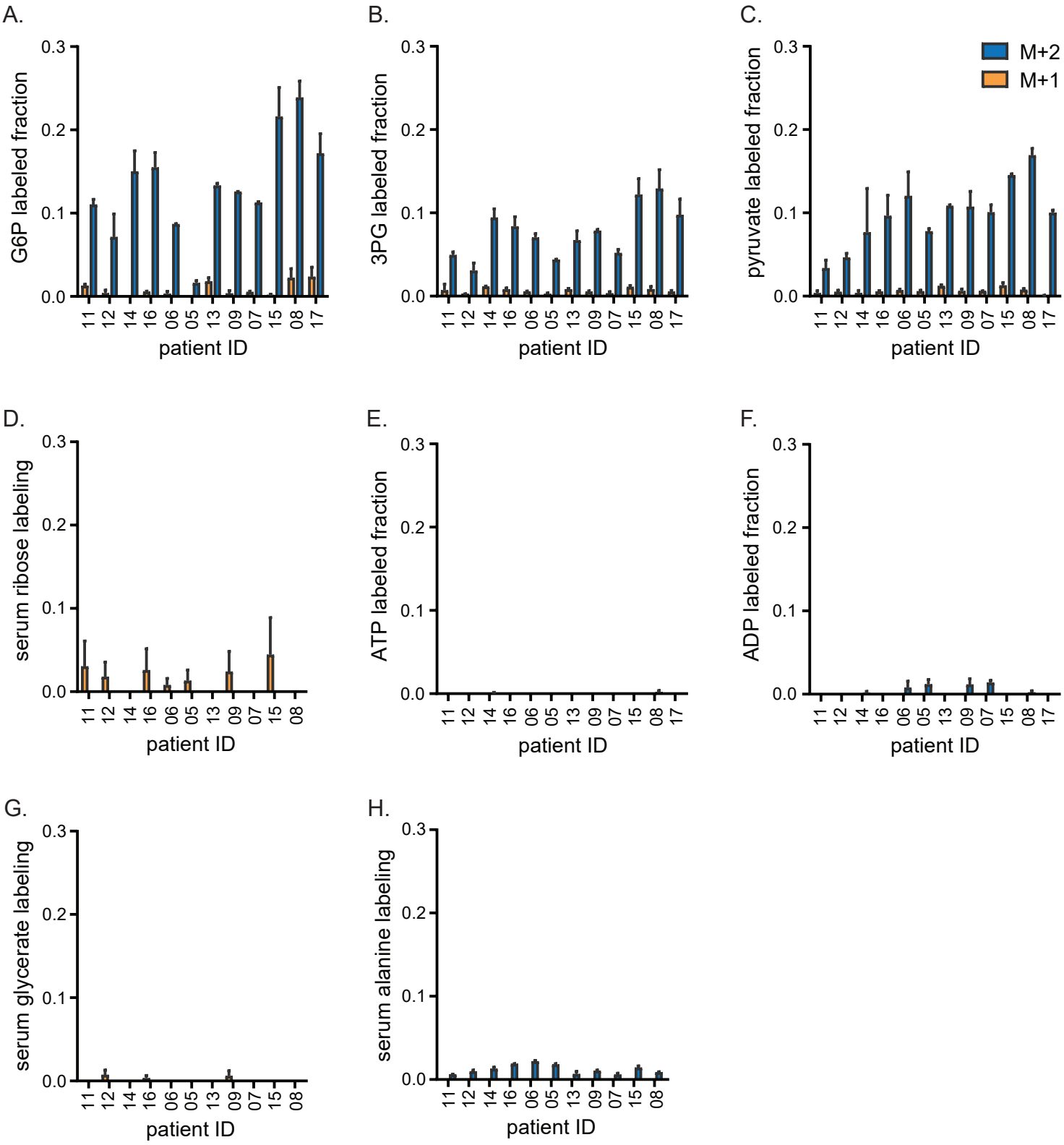

Figure S2. Tumor and serum metabolite labeling data related to Figure 2. A-C) Individual patient isotope labeling in TNBCs of A) glucose-6-phosphate (G6P), B) 3-phosphoglyerate (3PG), and C) pyruvate (mean ± SEM, n = 2 biopsy samples per patient). D) Individual patient serum isotope labeling of ribose (mean ± SEM, n = 3 technical replicates). E-F) Individual patient isotope labeling in TNBCs of E) ATP and F) ADP (mean ± SEM, n = 2 biopsy samples per patient). G-H) Individual patient serum isotope labeling of G) glycerate, and H) alanine (mean ± SEM, n = 3 technical replicates).

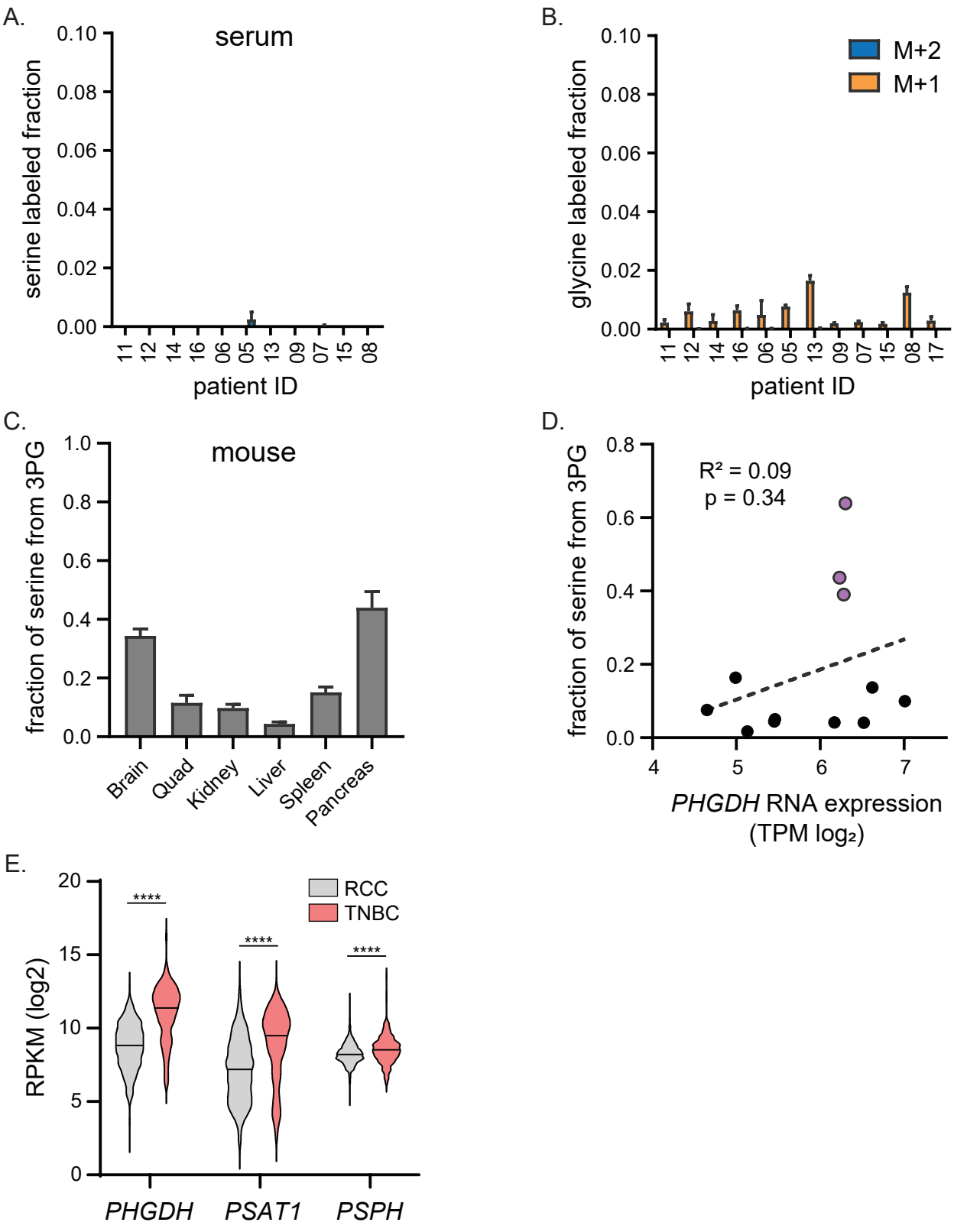

Figure S3. Metabolite labeling and gene expression data related to Figure 4. A) Individual patient serum serine isotope labeling (mean ± SEM, n = 3 technical replicates). B) Individual patient labeling of glycine in TNBCs (mean ± SEM, n = 2 biopsy samples per patient). C) Fractional carbon serine labeling normalized to that of 3PG in indicated tissues from C57BL/6 mice after infusion with [U-^{13}C]glucose for 2.5 h (mean ± SEM, n = 3 for spleen, 4 for other tissues). See also Table S5. D) RNA expression of *PHGDH* of individual TNBC biopsies compared with estimated *de novo* serine production (mean, n = 2 biopsy samples per patient). Purple dots represent same patients as purple bars in Figure 4D. E) Violin plots of serine biosynthetic enzyme expression in indicated tumors analyzed from TCGA data (Student's t-test). **** = $p < 0.0001$.

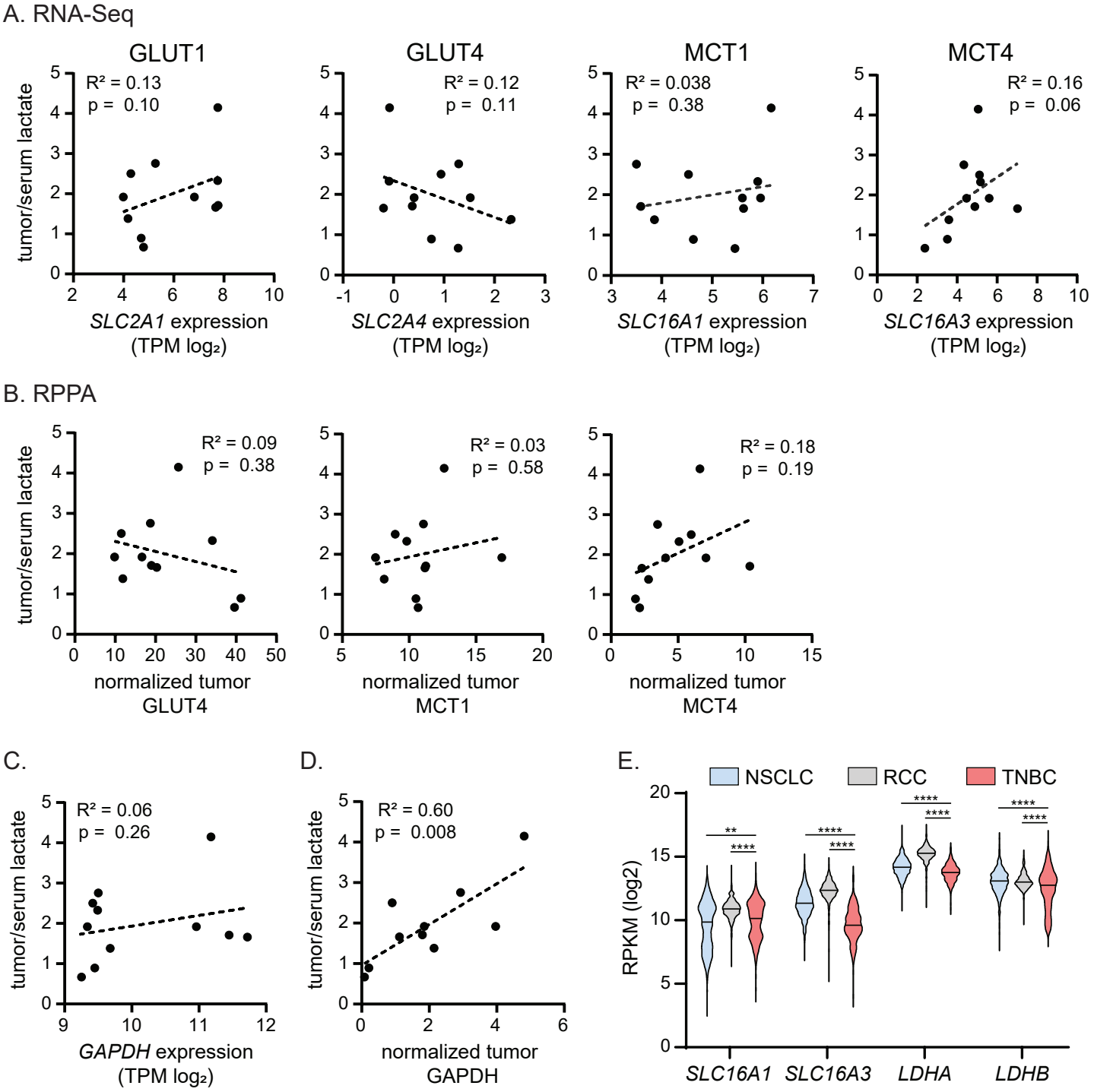

Figure S4. Gene and protein expression data related to Figure 5. A-B) TNBC expression of indicated lactate (MCT) and glucose (GLUT) transporters do not correlate with tumor-to-serum lactate labeling ratio at the level of A) RNA (via RNA-Seq) or B) protein (via RPPA). C-D) Correlation of tumor-to-serum lactate labeling in TNBC with expression of glyceraldehyde-3-phosphate dehydrogenase enzyme (GAPDH) at the level of C) RNA (via RNA-Seq) or D) protein (via RPPA). E) Violin plots of relevant lactate transporters (SLC) or dehydrogenases (LDH) in indicated tumors analyzed from TCGA data (Student's t-test). ** = $p < 0.01$, **** = $p < 0.0001$.

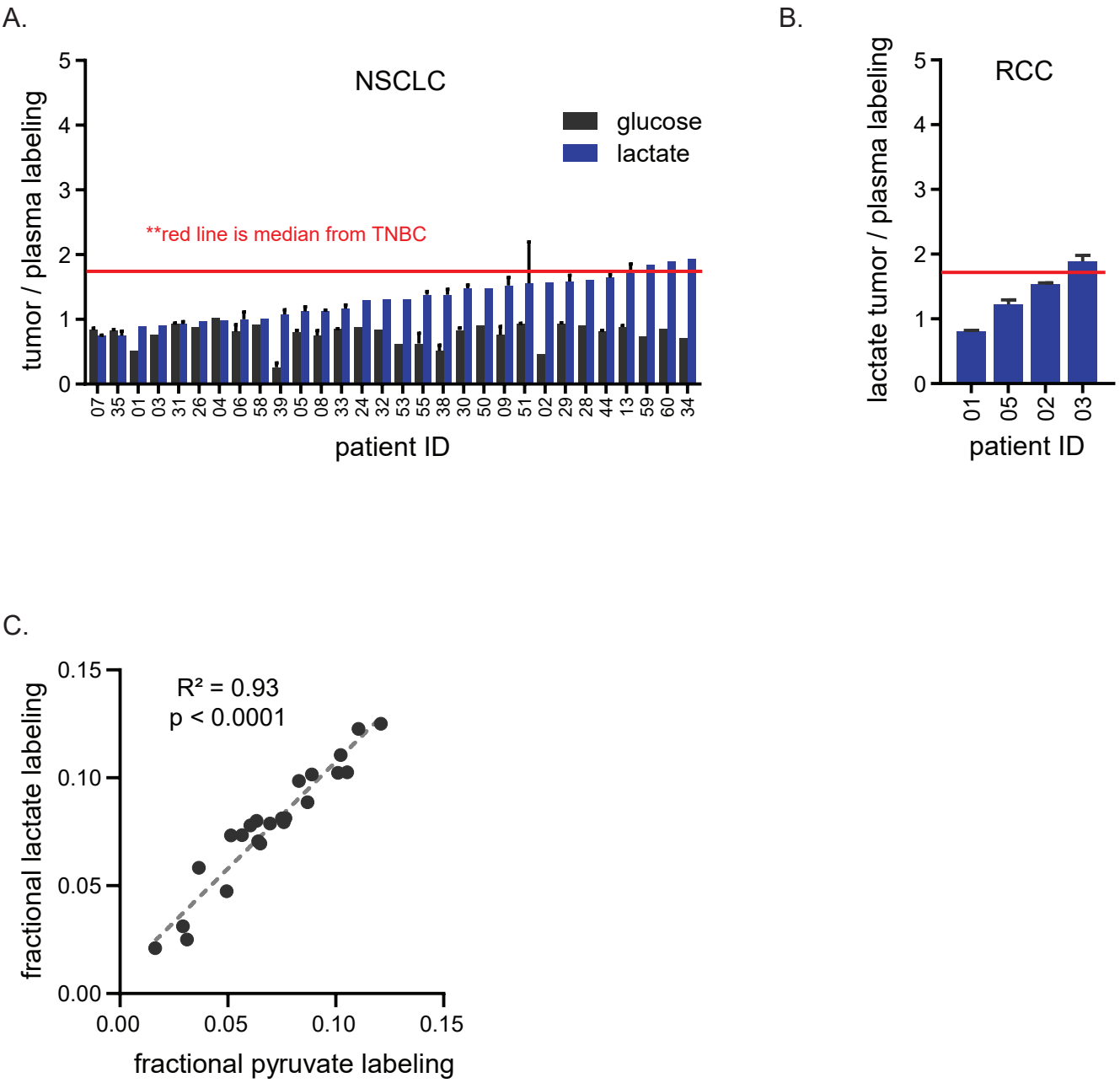

Figure S5. Tumor and serum metabolite labeling data related to Figure 5. A-B) Fractional carbon labeling of tumor glucose and/or lactate to that of plasma for A) NSCLC patients or B) RCC patients undergoing [U-^{13}C]glucose infusion (mean ± SEM, NSCLC: n ranges between one and six individual tumor fragments per patient; RCC:, n = 3 tumor samples per patient). Patient RCC4 data are missing due to poor plasma detection of labeled lactate. C) Fractional carbon labeling of tumor pyruvate and lactate are highly correlated in TNBC.

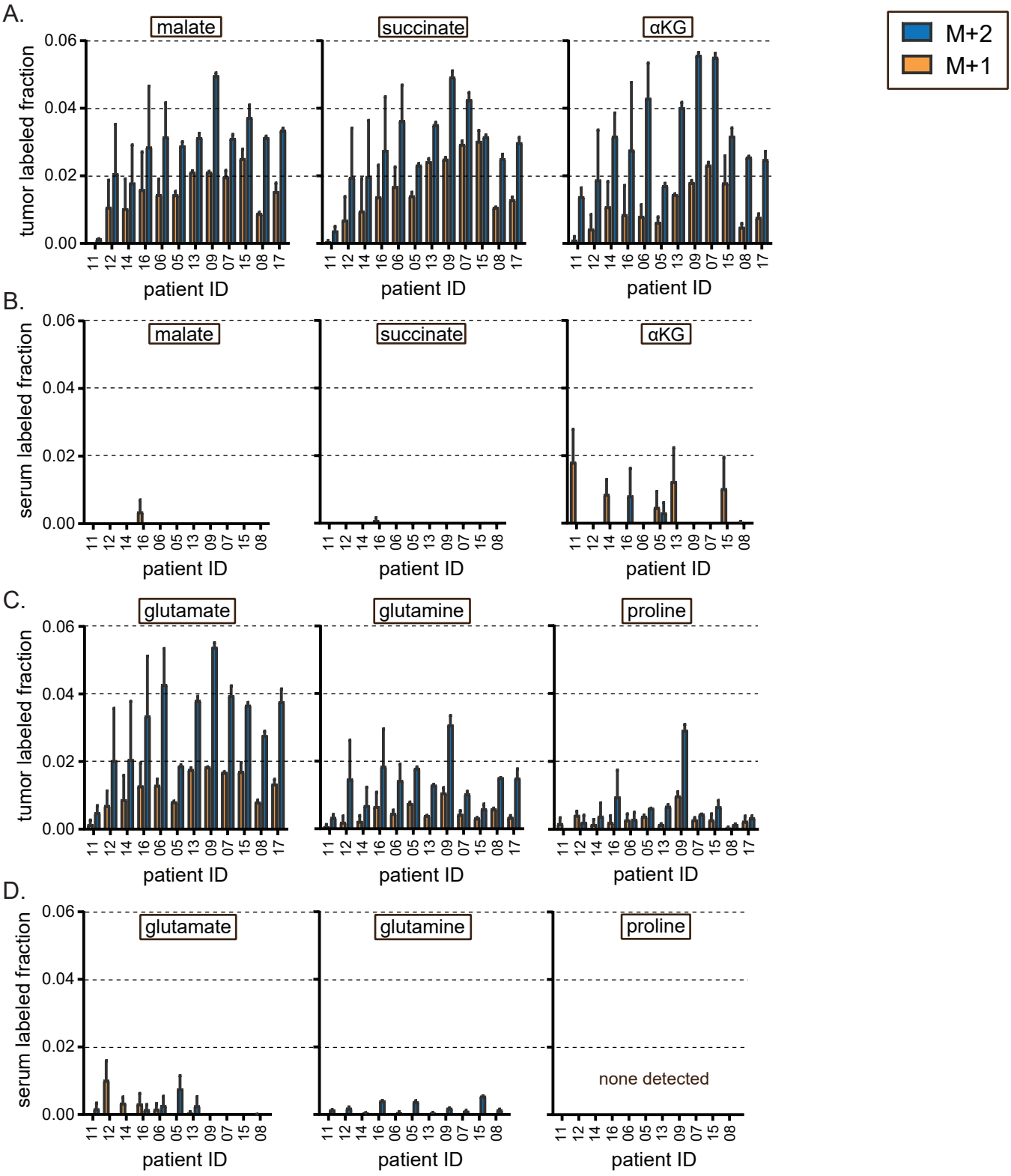

Figure S6. TCA and amino acid metabolite labeling data related to Figure 6. A-B) Individual patient labeling of malate, succinate, and αKG in A) TNBCs, and B) serum at time of biopsy from [1,2-^{13}C]glucose (mean ± SEM, n = 2 biopsies per patient, 3 technical replicates for serum). C-D) Individual patient labeling of glutamate, glutamine, and proline in C) TNBCs, and D) serum (mean ± SEM, n = 2 biopsies per patient, 3 technical replicates for serum). E) Individual patient labeling of aspartate and asparagine in TNBCs (mean ± SEM, n = 2 biopsy samples per patient).

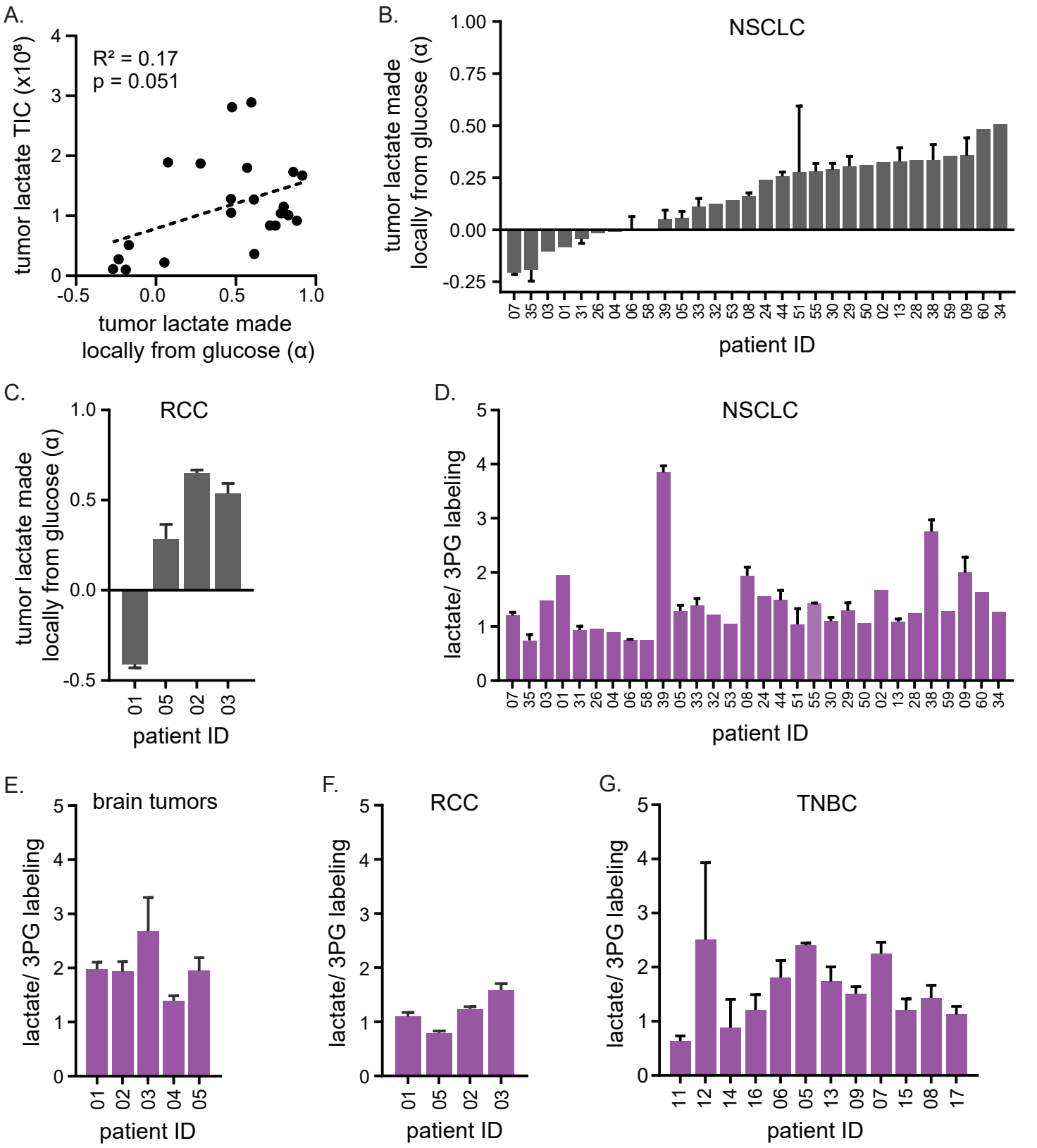

Figure S7. Additional metabolite data and calculations related to Figure 7. A) Correlation between lactate concentration and fraction of tumor lactate produced locally from glucose (α) in TNBCs. TIC = total ion count, which is linearly proportional to concentration. B-C) Fraction of tumor lactate produced locally from glucose (α) in B) NSCLC and C) RCC (mean ± SEM, NSCLC: n ranges between one and six individual tumor fragments per patient; RCC: n = 3 tumor samples per patient). D-G) Lactate/3PG labeling ratios in D) NSCLC, E) brain tumors, F) RCC, or G) TNBCs (mean ± SEM, NSCLC: n ranges between one and six individual tumor fragments per patient; brain tumors, RCC: n = 3 tumor samples per patient; TNBC: n = 2 biopsy samples per patient). In line with published work, lactate/3PG is calculated using the highest labeled form of lactate and 3PG observed in each study (M+3 in NSCLC, brain tumors, and RCC; M+2 in TNBC).