

Fig. S1

Fig. S1. Liver relative mRNA abundance of (A) phosphoenolpyruvate carboxykinase (*pck*) and (B) glucose-6-phosphatase (*g6pc*) paralogues in the control and alanine-infused rainbow trout. Data were normalized by the reference gene β -actin. The mean + s.e.m. are represented (N=5-7). Filled circles represent individual data points. Data were analyzed using two-tailed t-test and means significantly different from control are indicated by an asterisk ($p < 0.05$).

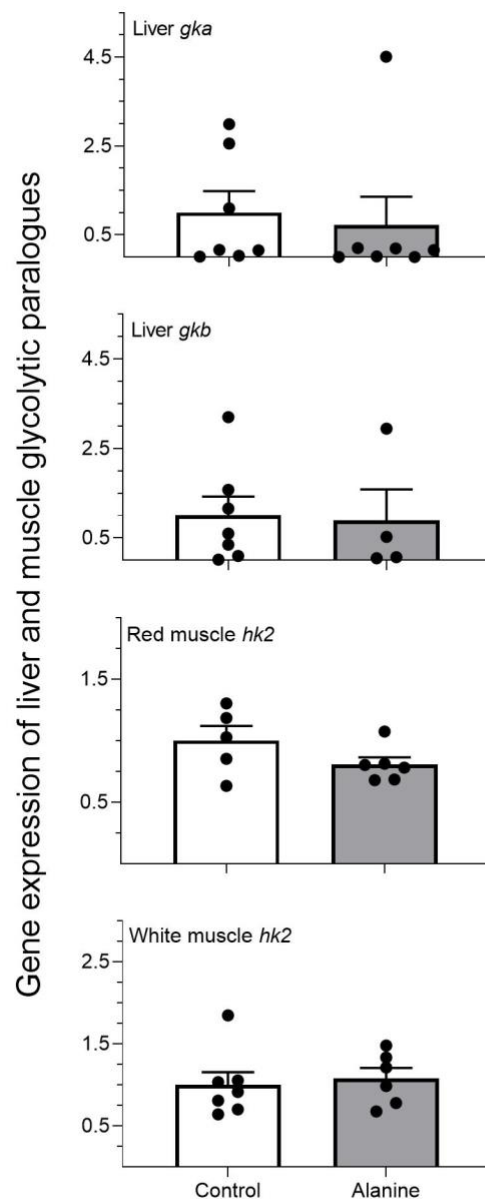


Fig. S2

Fig. S2. Relative mRNA abundance of glucokinase (*gk*) in the liver and hexokinase 2 (*hk2*) in the red and white muscle in the control and alanine-infused rainbow trout. Data were normalized by the reference gene β -actin for the liver and *ef1a* for red and white muscle. The mean + s.e.m. are represented (N=4-7). Filled circles represent individual data points. Data were analyzed using two-tailed t-test. Alanine had no effect on the measured glycolytic mRNA transcript abundance ($p>0.05$).

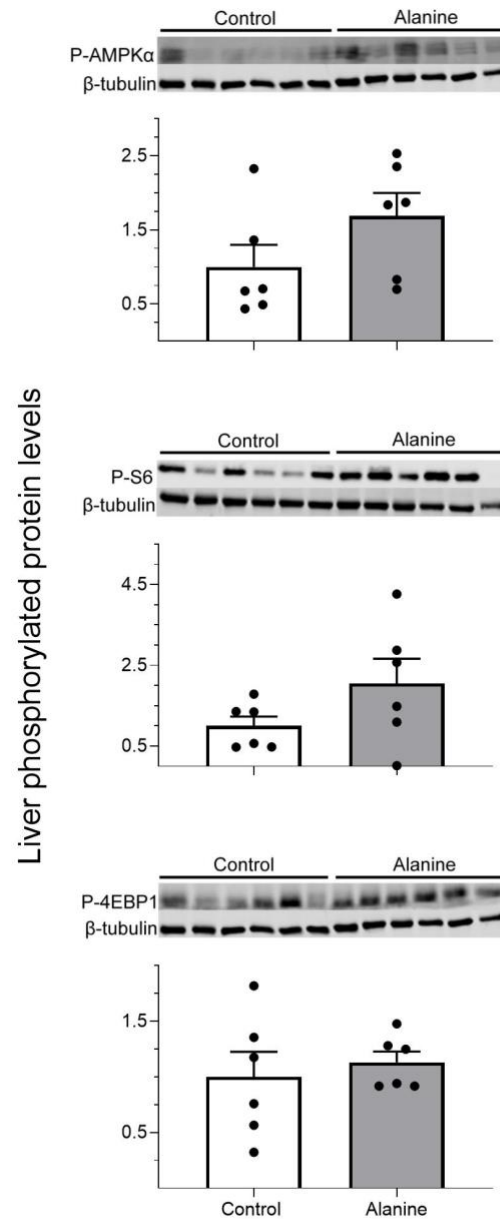


Fig. S3

Fig. S3. Liver relative abundance of phosphorylated AMPKα (at T172), ribosomal protein S6 (S6; at S235/236) and eukaryotic translation initiation factor 4E binding protein 1 (4EBP1; at T37/46) in the control and the alanine-infused groups. Data were normalized by β-tubulin and are represented as fold changes relative to the control group. The western blot of each phosphorylated protein is shown on top of its figure. The mean + s.e.m. are represented (N=6). Filled circles represent individual data points. Data were analyzed using two-tailed t-test. Alanine had no effect on the phosphorylated level of these proteins in the liver ($p > 0.05$).

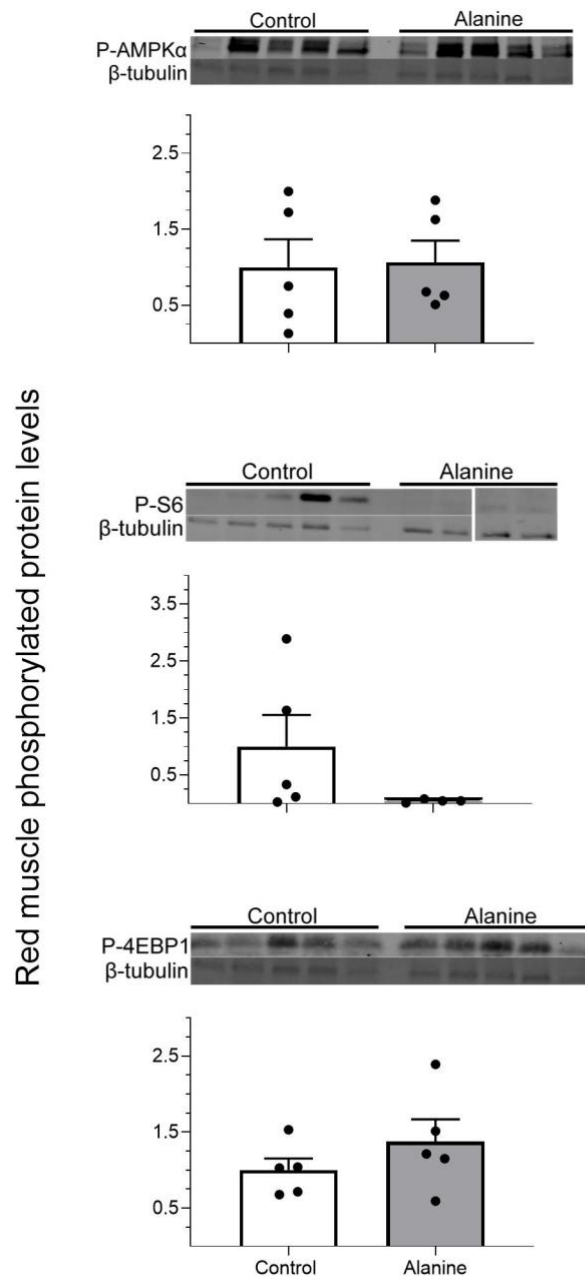


Fig. S4

Fig. S4. Red muscle relative levels of phosphorylated AMPKα (at T172), S6 (at S235/236) and 4EBP1 (at T37/46) in the control and the alanine-infused groups. Data were normalized by β-tubulin and are represented as fold changes relative to the control group. The western blot of each phosphorylated protein is shown on top of its figure. The mean + s.e.m. are represented (N=4-5). Filled circles represent individual data points. Data were analyzed using two-tailed t-test. Alanine had no effect on the phosphorylated level of these proteins in the red muscle ($p > 0.05$). A white space indicates the removal of a lane (outlier).

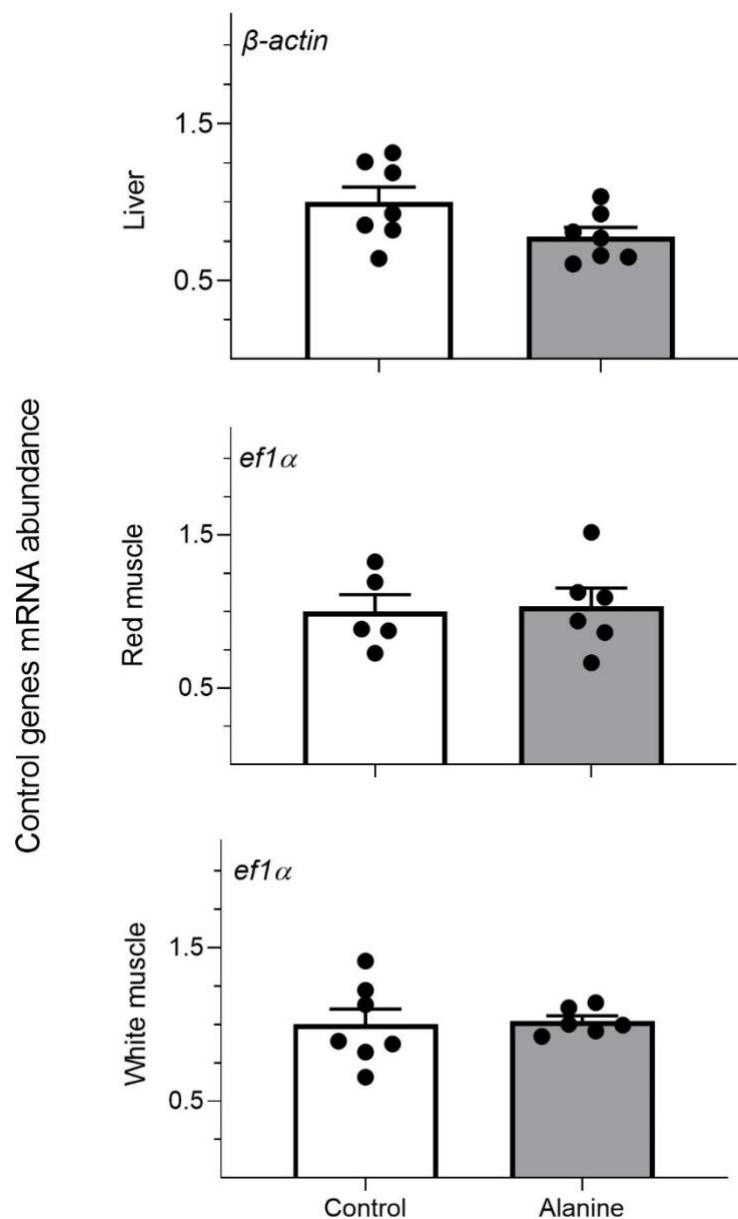


Fig. S5

Fig. S5. Relative mRNA abundance of the control genes β -actin in the liver and elongation factor 1 α ($ef1\alpha$) in the muscle of the control and alanine-infused rainbow trout. Data points are relative to the control group for each gene. The mean + s.e.m. are represented (N=5-7). Filled circles represent individual data points. Data were analyzed using two-tailed t-test. Alanine had no effect on the measured mRNA transcript abundance of the control genes ($p>0.05$).

Table S1. Primer pair conditions used for mRNA quantification by real-time RT PCR.

mRNA	Primer sequence (5' to 3')	Annealing temperature °C	Efficiency %, R ²	Reference
<i>pck1</i>	F: ACAGGGTGAGGCAGATGTAGG R: CTAGTCTGTGGAGGTCTAAGGGC	55	91.9, 0.995	Marandel et al., 2015
<i>pck2a</i>	F: ACAATGAGATGATGTGACTGCA R: TGCTCCATCACCTACAACCT	55	90.4, 0.997	Marandel et al., 2015
<i>pck2b</i>	F: AGTAGGAGCAGGGACAGGAT R: CCGTTCAGCAAAGGTTAGGC	55	102.8, 0.989	Marandel et al., 2019
<i>g6pca</i>	F: GATGGCTTGACGTTCTCCT R: AGATCCAGGAGAGTCCTCC	55	91.9, 0.995	Marandel et al., 2015
<i>g6pcb1a</i>	F: GCAAGGTCCAAAGATCAGGC R: GCCAATGTGAGATGTGATGGG	59	105.9, 0.975	Marandel et al., 2015
<i>g6pcb1b</i>	F: GCTACAGTGCTCTCCTTCTG R: TCACCCCATAGCCCTGAAA	55	91.6, 0.997	Marandel et al., 2015
<i>g6pcb2a</i>	F: ATCGGACAATACACACAGAACT R: CAACTGATCTATAGCTGCTGCCT	54	91.3, 0.994	Marandel et al., 2015
<i>g6pcb2b</i>	F: CCTCTGCTCTTCTGACGTAG R: TGTCCATGGCTGCTCTCTAG	55	92.3, 0.985	Marandel et al., 2015
<i>gka</i>	F: CTGCCCACCTACGTCTGT R: GTCATGGCGTCCTCAGAGAT	54	96.3, 0.993	Marandel et al., 2015
<i>gkb</i>	F: TCTGTGCTAGAGACAGCCC R: CATTTTGACGCTGGACTCCT	57	90.9, 0.993	Marandel et al., 2015
<i>hk2</i>	F: TGAAAAGGGACATGCAGAGA R: GGCCCTAAAAGCAAGGAAA	58	92.3-96.1, 0.992-0.988	Designed
<i>glut4a</i>	F: CATCTTTGCAGTGCTCCTTG	56	106.1-101.1, 0.997-0.982	Liu et al., 2017
<i>glut4b</i>	R: CAGCTCTGTACTCTGCTTGC F: TCGGCTTTGGCTTCCAATATG	56	101.4-106.2, 0.995-0.996	Liu et al., 2017
<i>β-actin</i>	R: GTTTGCTGAAGGTGTTGGAG F: AGAGCTACGAGCTGCCTGAC R: GTGTTGGCGTACAGGTCCTT	60	90.4, 0.996	Moltesen et al., 2016
<i>ef1α</i>	F: CACATCGCCTGCAAGTTT R: GAAGCTCTCCACACACATGG	58	106.6-110.1, 0.985-0.988	Designed

F and R represent forward and reverse primer sequences, respectively. Primer sequences for *hk2* and *ef1α* was designed using Primer 3 algorithm. The efficiency and R² values are presented for both the red and white muscle *glut4a*, *glut4b*, *hk2* and *ef1α*. The reference genes for the liver and muscle are *β-actin* and *ef1α*, respectively.