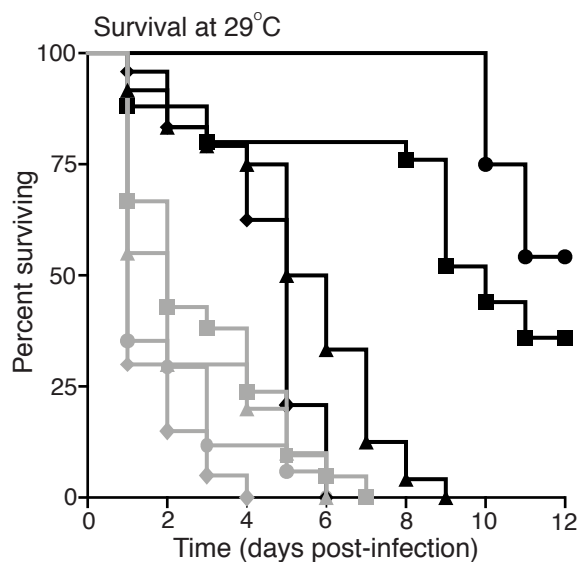
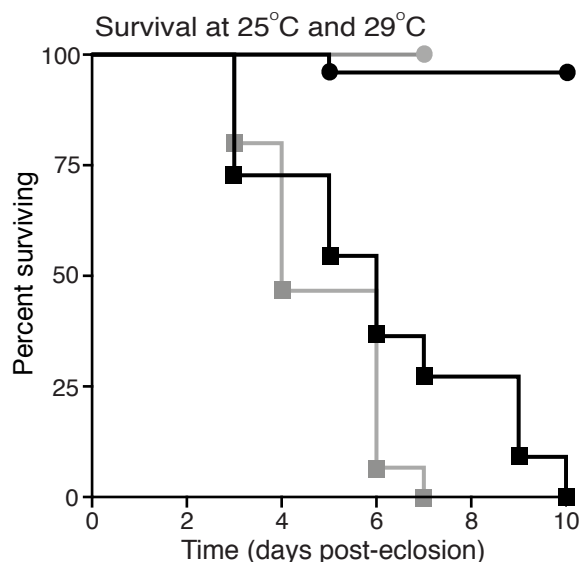


**A**



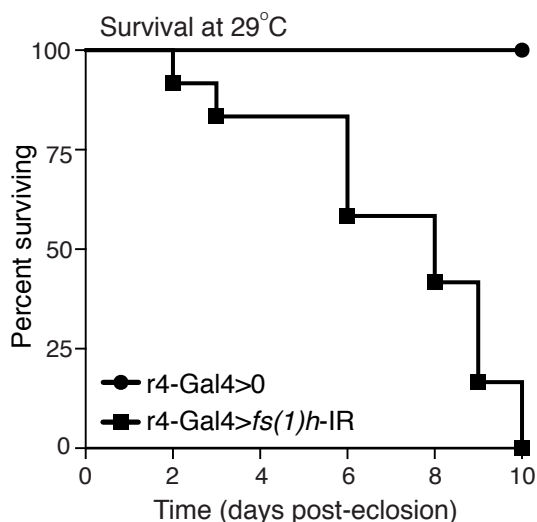
- c564>0 Uninjected
- c564>0 PBS
- ▲ c564>0 *L monocytogenes*
- ◆ c564>0 *F novicida*
- c564>fs(1)h-IR Uninjected
- c564>fs(1)h-IR PBS
- ▲ c564>fs(1)h-IR *L monocytogenes*
- ◆ c564>fs(1)h-IR *F novicida*

**B**



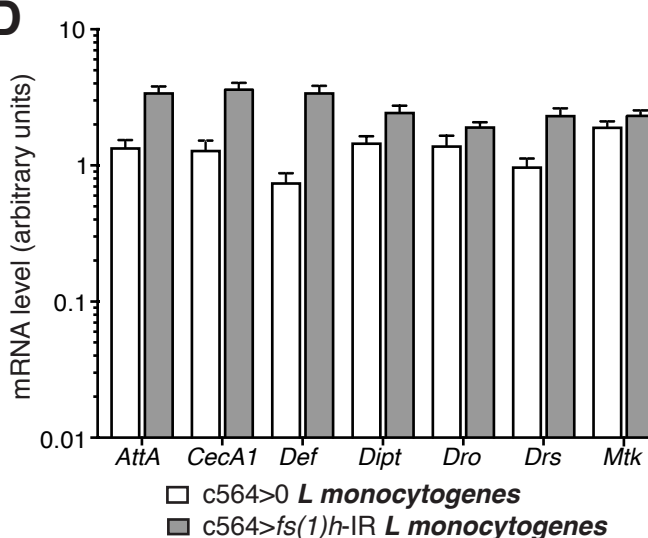
- c564>0 (25°C)
- c564>fs(1)h-IR II (25°C)
- c564>0 (29°C)
- c564>fs(1)h-IR II (29°C)

**C**



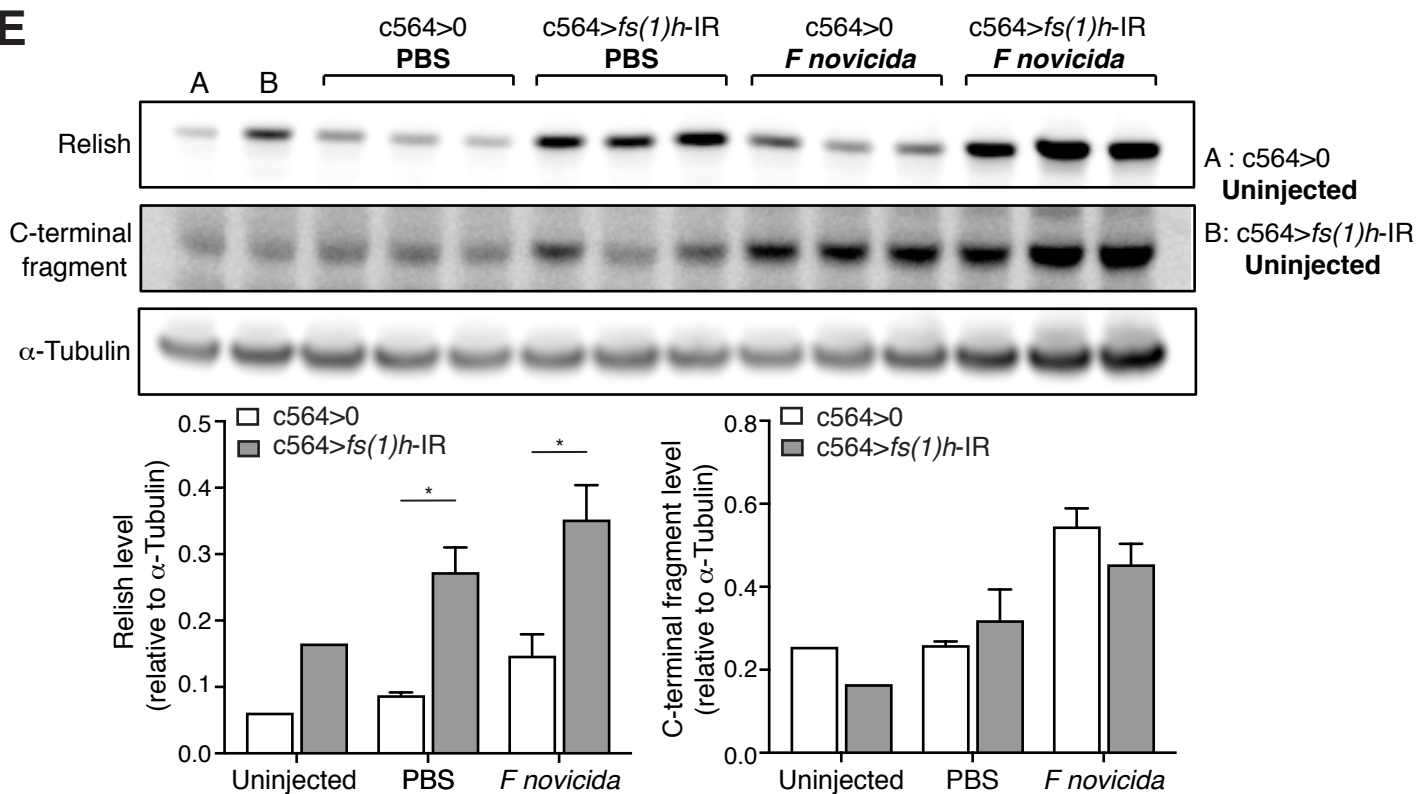
- r4-Gal4>0
- r4-Gal4>fs(1)h-IR

**D**

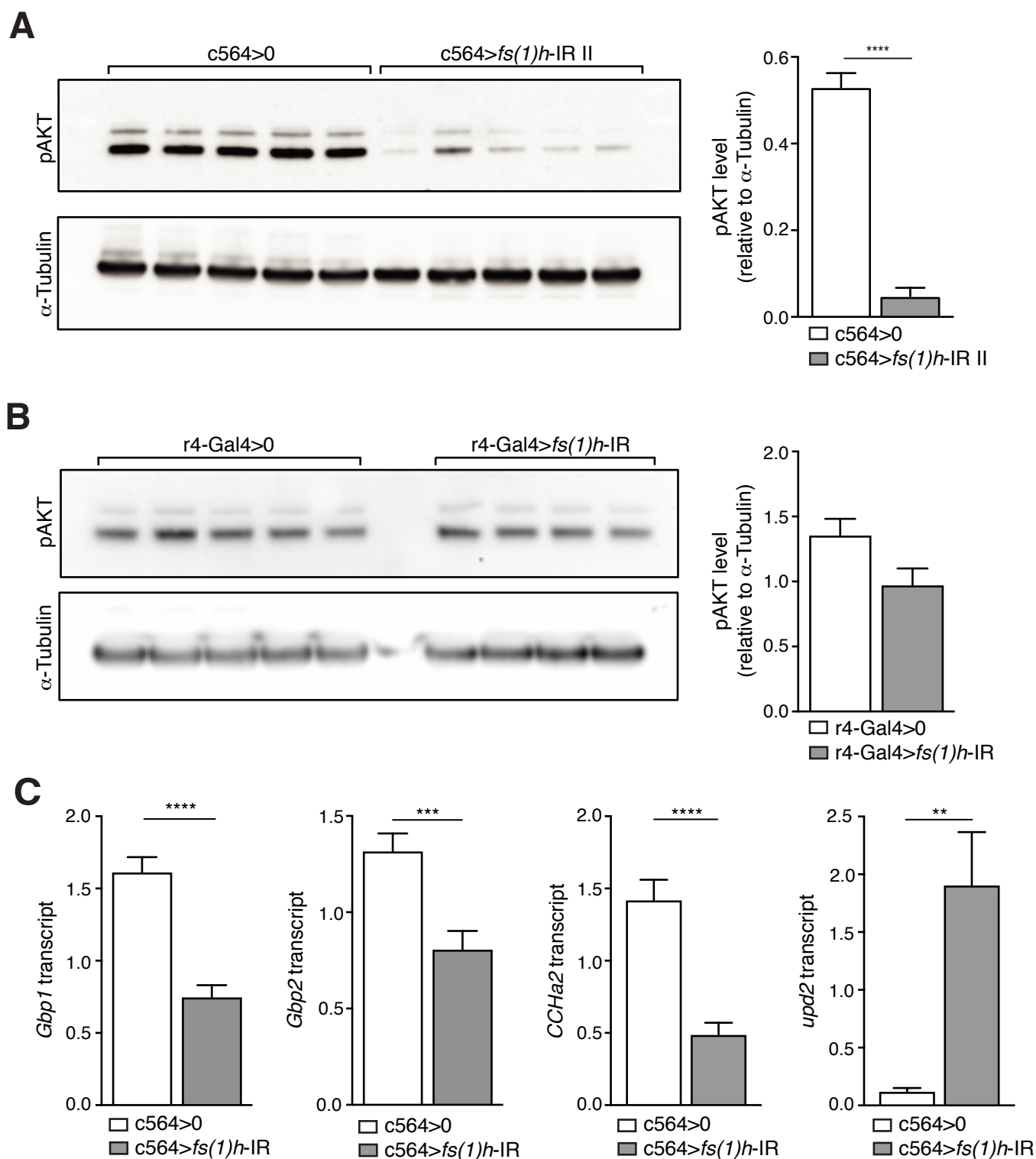


- c564>0 *L monocytogenes*
- c564>fs(1)h-IR *L monocytogenes*

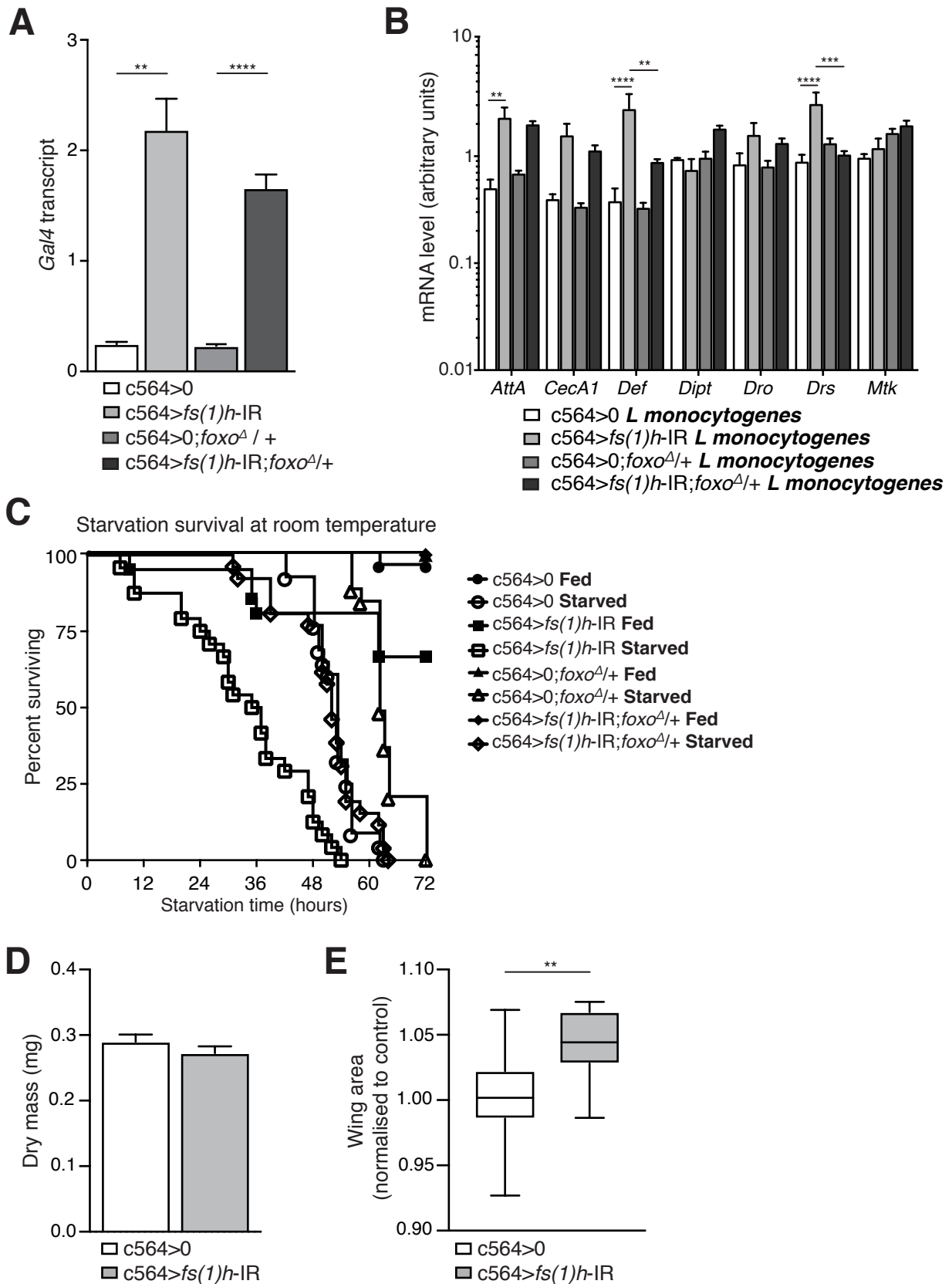
**E**



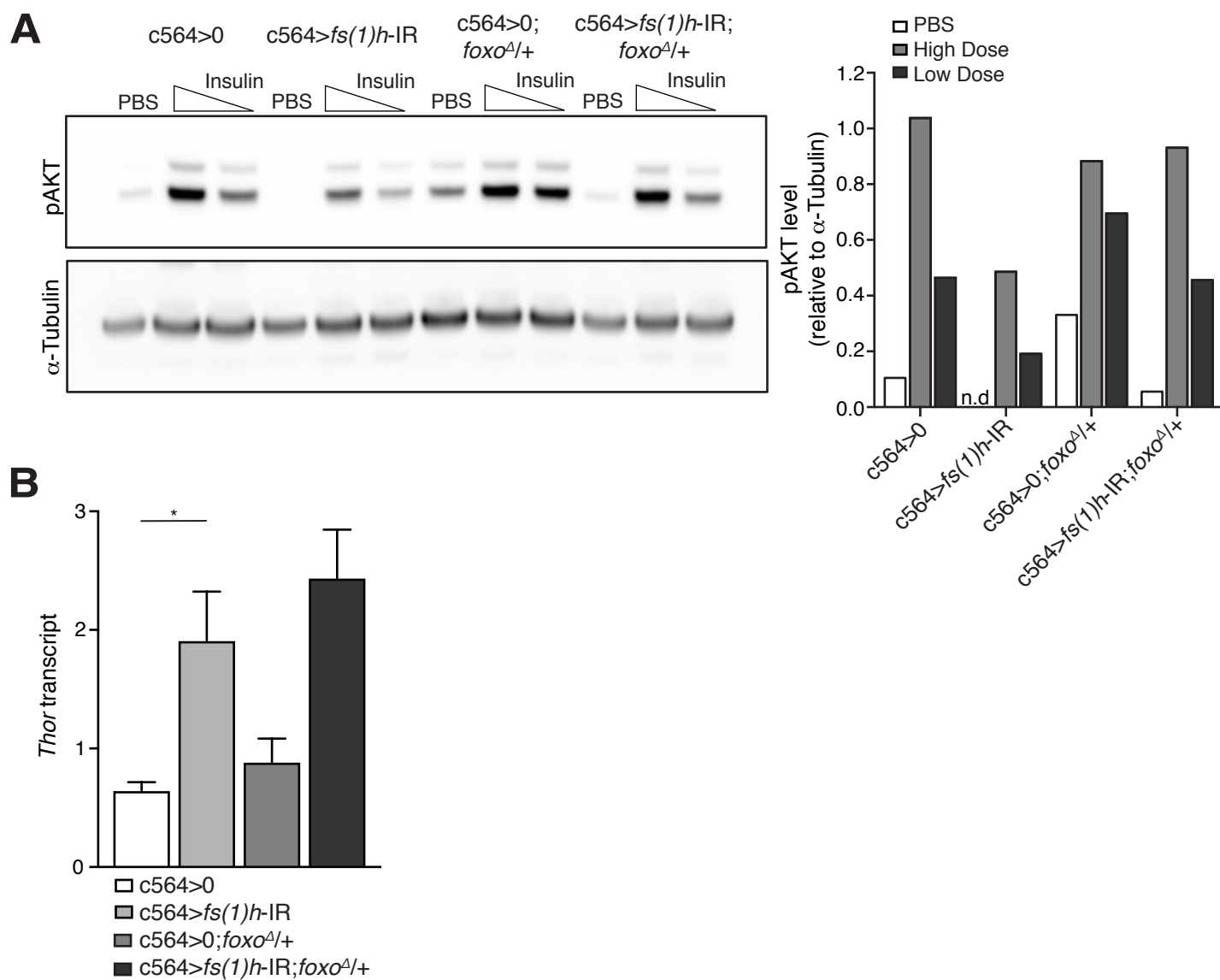
**Figure S1.** (A) Survival of fat body knockdown of *fs(1)h* (KK108662) compared to controls, either uninfected, PBS injected, or infected with *L monocytogenes* or *F novicida*. (B) Survival of flies with fat body knockdown of *fs(1)h* using a different RNAi line (GD51227) compared to driver-only controls. (C) Survival of flies with *fs(1)h* knocked down in fat body under the control of r4-Gal4. (D) Expression of antimicrobial peptide (AMP) genes in *Listeria monocytogenes*-infected 5-7 day old male control (c564>0) and *fs(1)h* fat body knockdown (c564>*fs(1)h-IR*) flies. Values shown as mean + SEM. (E) Relish protein levels in *fs(1)h* knockdowns and control animals, uninfected, PBS-injected, or infected with *Francisella novicida*.



**Figure S2.** (A) Western blot analysis of AKT phosphorylation in control and *fs(1)h* (GD51227) fat body knockdown flies. Values represented as intensity relative to  $\alpha$ -Tubulin, shown as mean+SEM, genotypes were compared using unpaired two-tailed t-test (\*\*\*\* $p$ <0.0001). (B) Western blot analysis of AKT phosphorylation in flies with *fs(1)h* knockdown driven by r4-Gal4. Values represented as intensity relative to  $\alpha$ -Tubulin. (C) Expression of insulin-regulatory secreted factors in isolated fat body from control and *fs(1)h* fat body knockdown flies, shown as mean + SEM. Normalised to expression of  $\alpha$ -tubulin as a loading control.



**Figure S3.** (A) *Gal4* expression in *fs(1)h* knockdowns and controls, wild-type or heterozygous for *foxo*. Data shown as mean + SEM; genotypes were compared using unpaired two-tailed t-test (\*\* $p < 0.01$ , \*\*\*\* $p < 0.0001$ ). (B) Expression of antimicrobial peptide (AMP) genes in *Listeria monocytogenes*-infected 5-7 day old male control (*c564>0*) and *fs(1)h* fat body knockdown (*c564>fs(1)h-IR*) flies, either wild-type or heterozygous for *foxo*. Values shown as mean + SEM. (C) Starvation survival of control and *fs(1)h* fat body knockdown flies, either wild-type or heterozygous for *foxo*. Carried out at room temperature on 5-7 day old flies. (D) Dry mass per fly of control and *fs(1)h* fat body knockdown animals. Values shown are mean + SEM. (E) Wing areas of control and *fs(1)h* fat body knockdown animals, normalised to the mean of the controls. The center line represents the median of the measurement; the extent of the box, the interquartile range; the whiskers, the furthest possible outliers. \*\* $p < 0.01$  by Wilcoxon rank-sum test.



**Figure S4.** (A) Western blot analysis of AKT phosphorylation following injection with PBS or a high (320 pg/fly) or low (64 pg/fly) dose of human insulin. Values relative to  $\alpha$ -Tubulin, shown as mean. (B) Expression of *Thor* in control and *fs(1)h* knockdown flies, either heterozygous or wild-type for *foxo*.

Table S1: qRT-PCR primers used in this study:

<b>Gene</b>	<b>Left</b>	<b>Right</b>
<i>AttA</i>	CACAATGTGGTGGGTCAGG	GGCACCATGACCAGCATT
<i>Bmm</i>	GTCTCCTCTGCGATTTGCCAT	CTGAAGGGACCCAGGGAGTA
<i>CCHa2</i>	TGCTACTGGTCGTTATCTGCAC	TAGGCCTGGCATCCCTTT
<i>CecA1</i>	TCTTCGTTTTTCGTCGCTCTC	CTTGTTGAGCGATTCCCAGT
<i>CG10702</i>	GGACTCCCTGCAGACCAA	AGACCGATGGGTTTCGTTTCT
<i>conv</i>	TTAGGGGTATGCGTTTCCTC	GACCAATATCCGTGAGCACAT
<i>Def</i>	TTCTCGTGGCTATCGCTTTT	GGAGAGTAGGTCGCATGTGG
<i>Dipt</i>	ACCGCAGTACCCACTCAATC	CCCAAGTGCTGTCCATATCC
<i>Dro</i>	CCATCGAGGATCACCTGACT	CTTAGGCGGGCAGAATG
<i>Drs</i>	GTACTIONTTTCGCCCTCTTCG	CTTGCACACACGACGACAG
<i>foxo</i>	ACTTTCCCGTGGGCAATC	CGCCGTAGCTCATCTCCT
<i>Gal4</i>	GCAGCATTCTGGAACAAAGA	GGACAATTGGATCTCCCAAG
<i>Gbp1</i>	CAGCACAGAACAGATCACCAG	GCAGCACAGTGGTGTTTTCA
<i>Gbp2</i>	AAACGGCGGACATGGATA	TGGAAAACGACCGAAGACA
<i>Hnf4</i>	AGGAAATGTTGTTGGGAGGA	GGGTGGGACTCTGGTAGTCA
<i>Hsl</i>	CTTGGAATACTTGAGGGGTTG	AGATTTGATGCAGTTCTTTGAGC
<i>ImpL2</i>	GCCGATACCTTCGTGTATCC	TTTCCGTCGTCAATCCAATAG
<i>Mtk</i>	TCTTGAGCGATTTTTCTGG	TCTGCCAGCACTGATGTAGC
<i>Plin1</i>	GCGTTCTATGGTAGCCTTCAG	GCGTCCGGATAGAAAGCTG
<i>Plin2</i>	GCAGAATGGCAAGAGTTCTGA	ACTGTGTGTAGGACTGGATCCTC
<i>Sdr</i>	GCAGCACTTTTCCCTTAAACA	GGGCGAAGTTGTGTCGTATT
<i>Thor</i>	CAGGAAGGTTGTCATCTCGGA	GGAGTGGTGGAGTAGAGGGTT
<i>upd2</i>	CGGAACATCACGATGAGCGAAT	TCGGCAGGAACTTGTACTCG
<i><math>\alpha</math>-Tubulin</i>	GATTCCGGTGACGGTGAG	GATGCACACCTATCCGCTCT