Supplemental Figure 1: Western blot analysis of phosphorylated AKT at Ser473 (#4060) and total AKT (#2920, Cell Signaling Technology, Beverly, MA). An aliquot of the sarcoplasmic fraction was prepared for SDS-PAGE and 21μg of protein was separated using 4-12% Bis-Tris gels (Life Technologies, Carlsbad, CA) then transferred to nitrocellulose membranes. Membranes were blocked in 5% bovine serum albumin, incubated overnight in primary antibodies (1:1000 in blocking buffer) then incubated with secondary fluorescent antibodies (1:5000 in blocking buffer) followed by infrared imaging and densitometry (Licor Biosciences, Lincoln, NE). Data from each group were analyzed with 2-way ANOVA (Study Day x Time) with repeated measures and multiple comparisons by Sidak adjustment. *p<0.05 vs. same time point between study days. Data are mean±SEM with n=8-10 per group.