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Supplemental Information

SIRT3 Deacetylates Mitochondrial 3-Hydroxy-3-Methylglutaryl CoA Synthase 2 and Regulates Ketone Body Production

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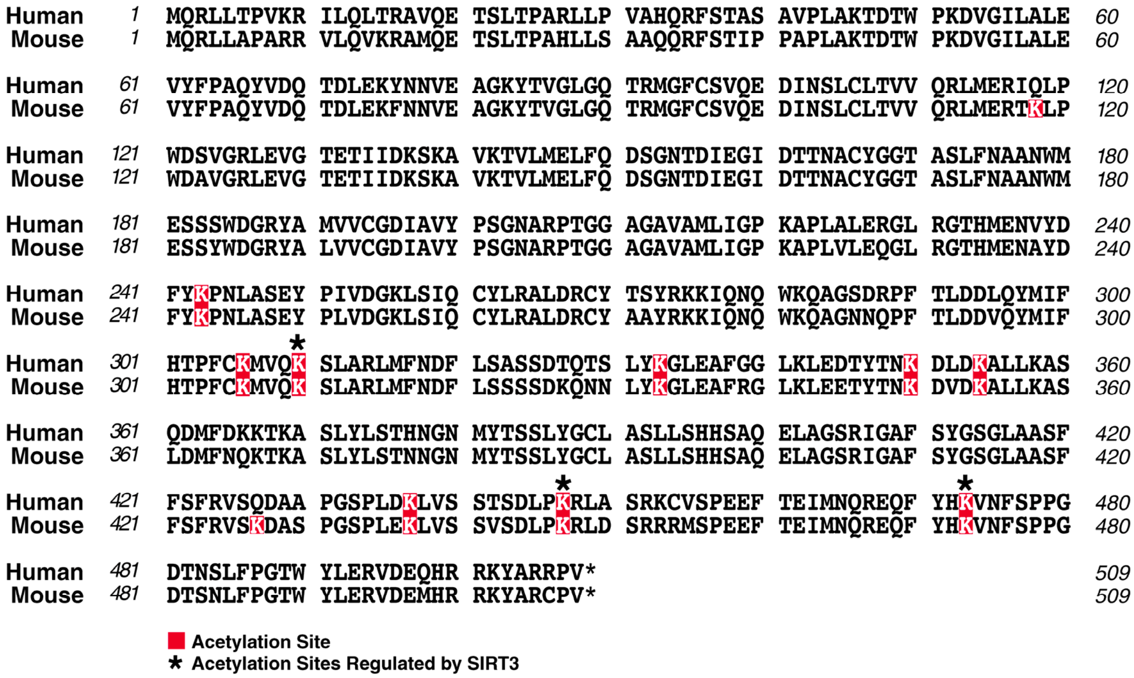

Figure S1, related to Figure 2. Human and Murine HMGCS2 Homology

Analysis of the acetylated lysines of murine mitochondrial HMGCS2 and human HMGCS2. The three acetylation sites that are studies here (K310, K447, and K473) are indicated by an asterisk.

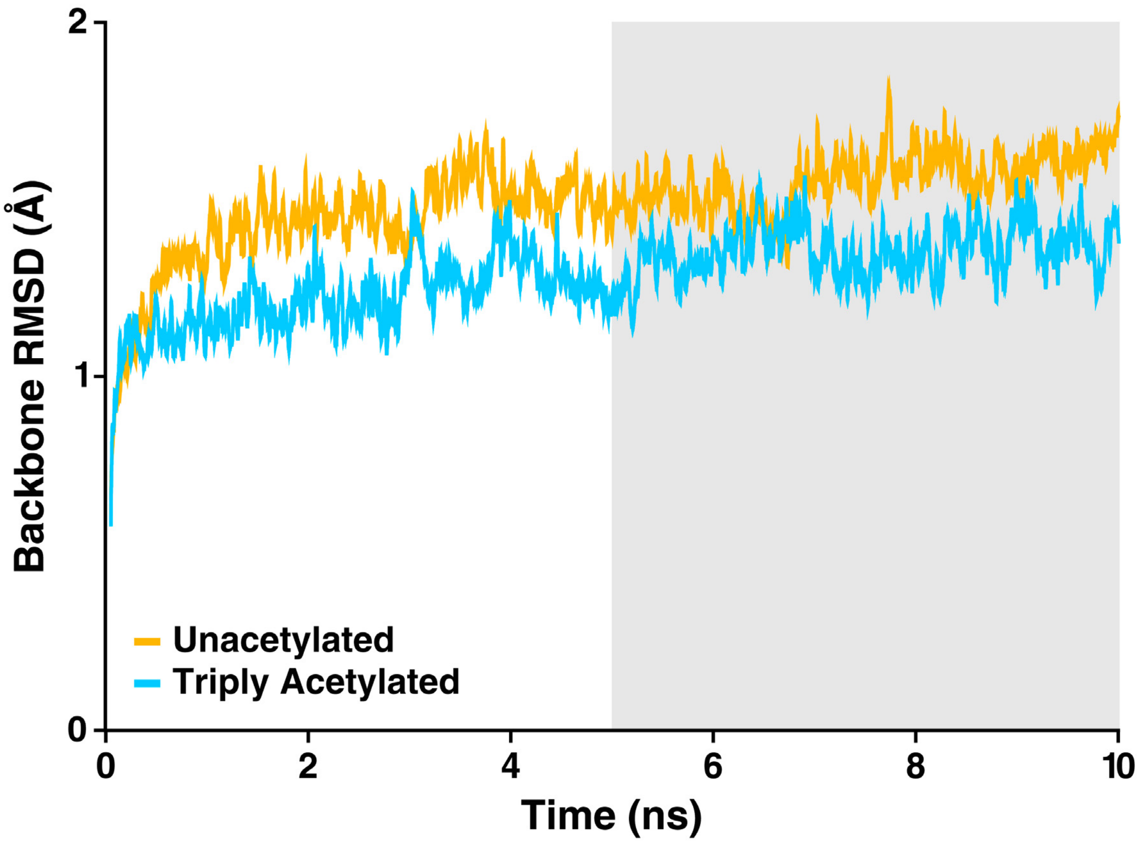

Figure S2, related to Figure 3. Backbone Root-Mean-Squared Deviation (RMSD) Was Stable over the Last 5 ns of a 10-ns Simulation

Statistical analyses of molecular dynamic simulations averaged over the last 5 ns of a 10-ns simulation (gray); data shown for the three acetylated states (unacetylated and triply acetylated) as a function of time (ns).

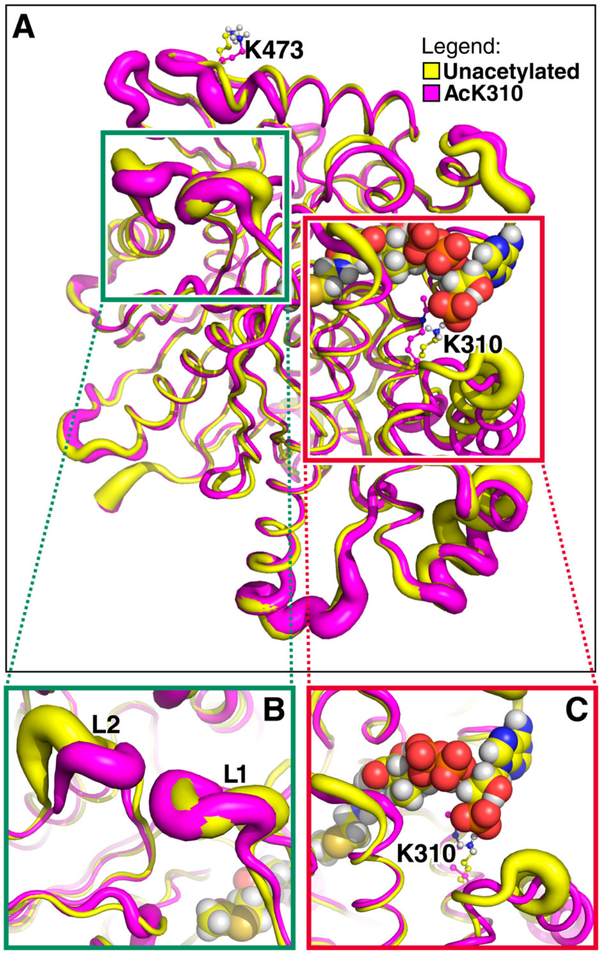

Figure S3, related to Figure 4. Molecular Dynamic Simulation of Unacetylated and Singly Acetylated (K310) HMGCS2

(A) Overview of the entire structure for the unacetylated (yellow) and singly acetylated K310 (magenta) mouse HMGCS2. Acetyl-CoA in the model is shown using spheres, whereas K310, K447, K473 and their acetylated forms are shown as balls and sticks. The tubes represent the average backbone conformation of the protein over the last 5 ns of five independent simulations, and the widths of the tubes represent the fluctuations from the average conformation (i.e., thicker lines indicate increased conformational flexibility).

(B) Closer view of the region around a distal portion of the active site [the green box in (A)]. The two loops showing significant conformational and dynamical changes are labeled L1 (residues 242–251) and L2 (residues 131–140).

(C) Closer view of the region around K310 [the red box in (A)].

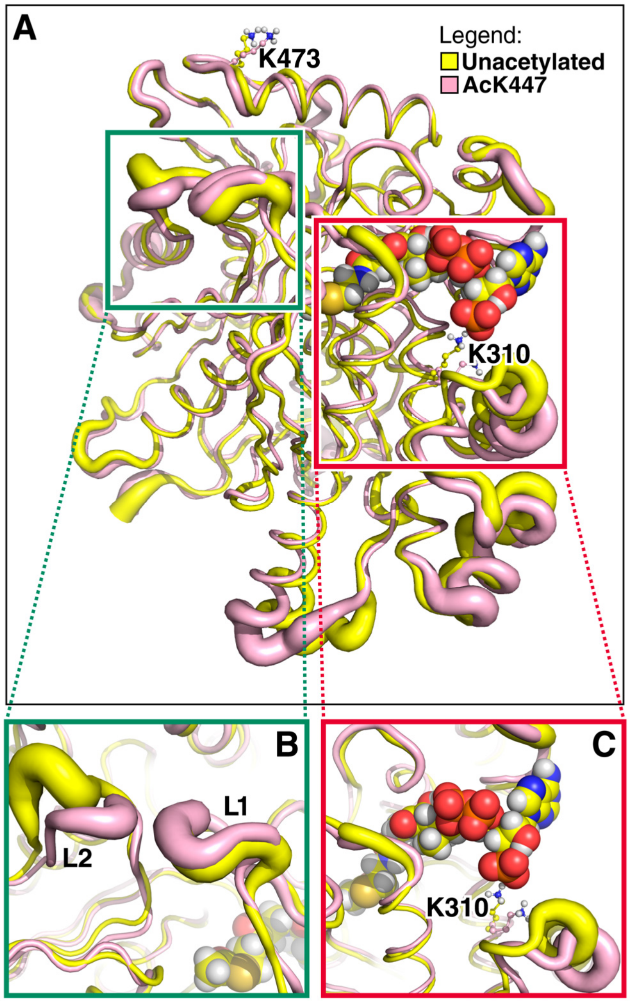

Figure S4, related to Figure 4. Molecular Dynamic Simulation of Unacetylated and Singly Acetylated (K447) HMGCS2

(A) Overview of the entire structure for the unacetylated (yellow) and singly acetylated K447 (pink) mouse HMGCS2. Acetyl-CoA in the model is shown using spheres, whereas K310, K447, K473 and their acetylated forms are shown as balls and sticks. The tubes represent the average backbone conformation of the protein over the last 5 ns of five independent simulations, and the widths of the tubes represent the fluctuations from the average conformation (i.e., thicker lines indicate increased conformational flexibility).

(B) Closer view of the region around a distal portion of the active site [the green box in (A)]. The two loops showing significant conformational and dynamical changes are labeled L1 (residues 242–251) and L2 (residues 131–140).

(C) Closer view of the region around K310 [the red box in (A)].

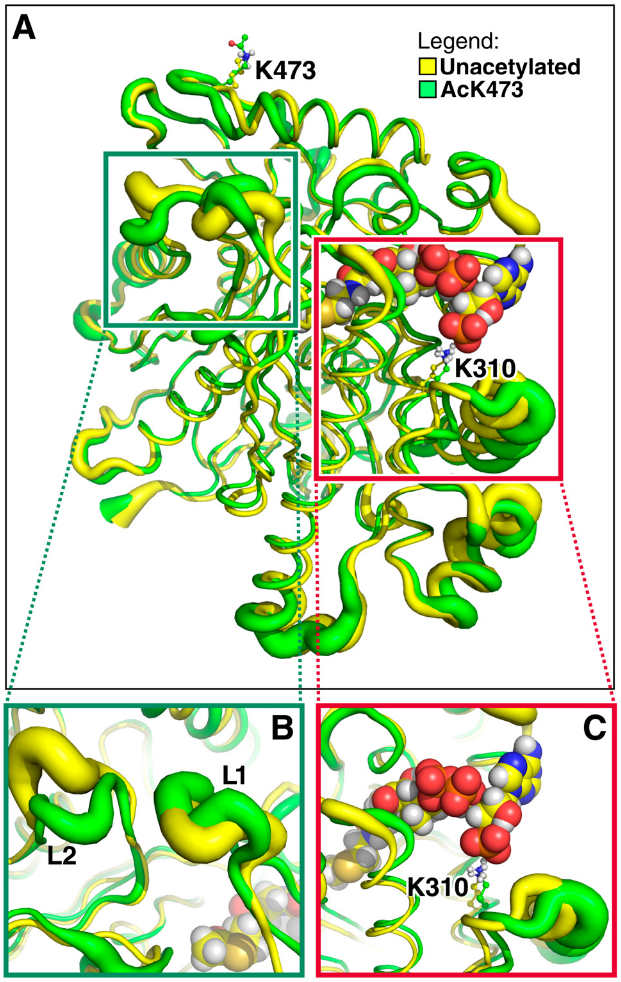

Figure S5, related to Figure 4. Molecular Dynamic Simulation of Unacetylated and Singly Acetylated (K473) HMGCS2

(A) Overview of the entire structure for the unacetylated (yellow) and singly acetylated K473 (lime) mouse HMGCS2. Acetyl-CoA in the model is shown using spheres, whereas K310, K447, K473 and their acetylated forms are shown as balls and sticks. The tubes represent the average backbone conformation of the protein over the last 5 ns of five independent simulations, and the widths of the tubes represent the fluctuations from the average conformation (i.e., thicker lines indicate increased conformational flexibility).

(B) Closer view of the region around a distal portion of the active site [the green box in (A)]. The two loops showing significant conformational and dynamical changes are labeled L1 (residues 242–251) and L2 (residues 131–140).

(C) Closer view of the region around K310 [the red box in (A)].

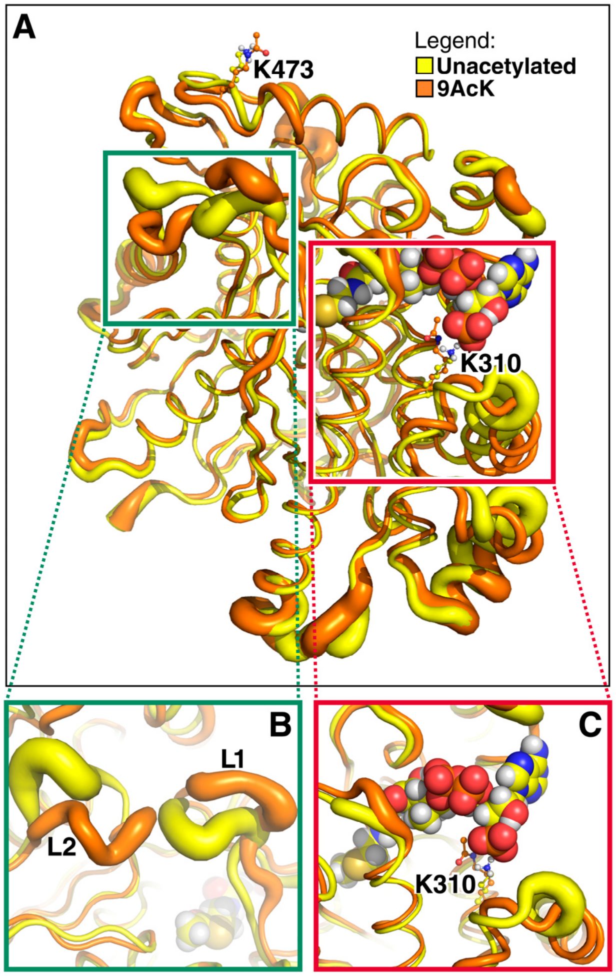

Figure S6, related to Figure 4. Molecular Dynamic Simulation of Unacetylated and Fully Acetylated (Nine Acetylated Lysines) HMGCS2

(A) Overview of the entire structure for the unacetylated (yellow) and fully acetylated (orange) mouse HMGCS2. Acetyl-CoA in the model is shown using spheres, whereas K310, K447, K473 and their acetylated forms are shown as balls and sticks. The tubes represent the average backbone conformation of the protein over the last 5 ns of five independent simulations, and the widths of the tubes represent the fluctuations from the average conformation (i.e., thicker lines indicate increased conformational flexibility).

(B) Closer view of the region around a distal portion of the active site [the green box in (A)]. The two loops showing significant conformational and dynamical changes are labeled L1 (residues 242–251) and L2 (residues 131–140).

(C) Closer view of the region around K310 [the red box in (A)].