For in vitro assays intact mitochondria were isolated, and citrate synthase activity measured from female third instar
wandering larvae

2 Set the plate reader at 412nm on a kinetic program: Duration 1.5 minute; Interval 10 seconds

3 Transfer 93 uL of isolated mitochondrial to a 96 well plate to make a final concentration of 2.0ug/mL

4 Add in 1uL of Acetyl CoA (30mM) and 1uL of DTNB (10mM)

5 Follow the absorbance of the reaction mixture for 1.5 minutes to measure the baseline reaction, endogenous levels of thiol or deacetylase activity

6 Add 5uL of 10mM oxaloacetate to each well to initiate the reaction. In order to start the reaction in all well simultaneously as possible, use multichannel pipette.

7 Shake the plate for 10 second before reading absorbance. Activity was measure at 412nm (molar extinction coefficients for DTNM were 13.6 L mol⁻¹ cm⁻¹)