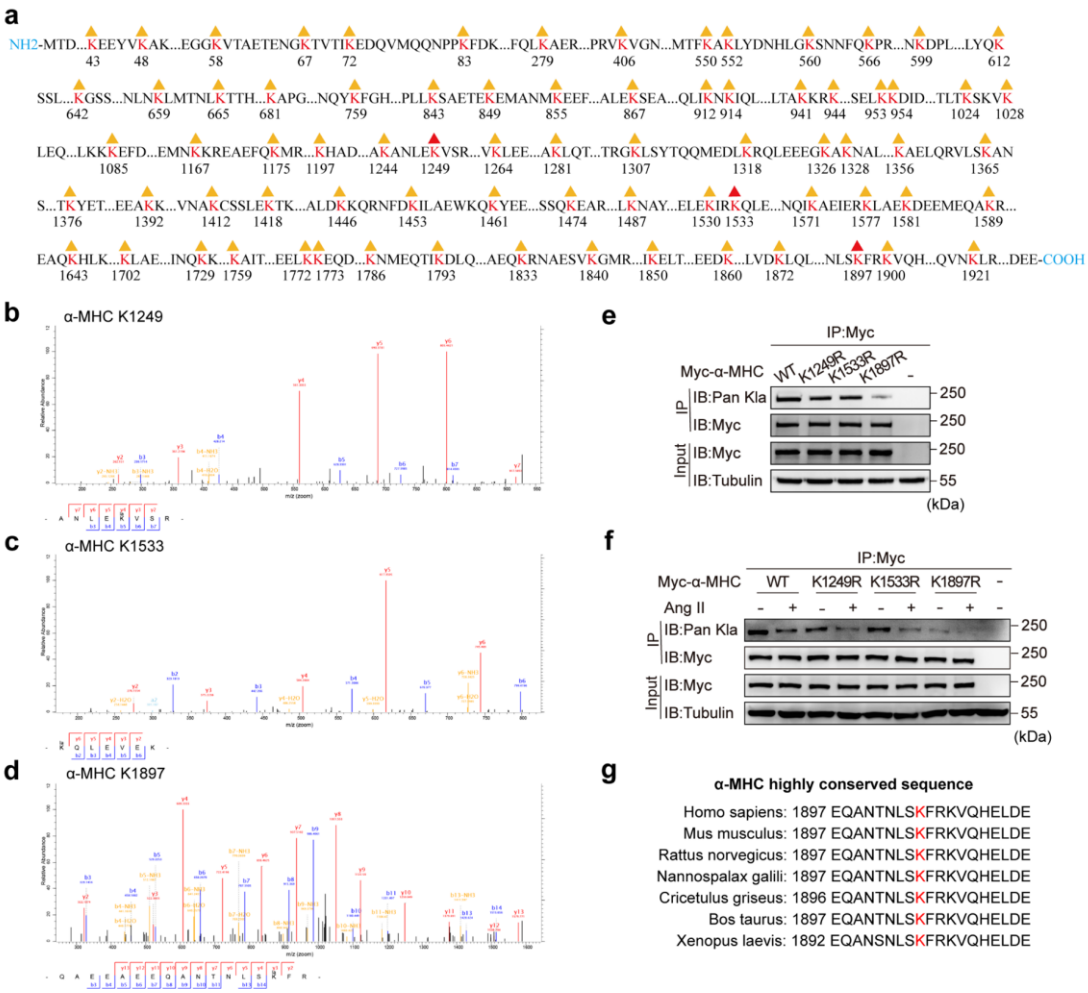

Supplementary information, Fig. S2 α-MHC K1897 is the main lactylation sites of α-MHC under physiological and Ang II treatment conditions.

a Mass spectroscopy revealed potential lactylation sites of α-MHC. **b, c, d** Lactylation sites of α-MHC identified by mass spectrometry: K1249 (**b**), K1533 (**c**) and K1897 (**d**). **e, f** Lactylation of K1897, K1533, and K1249 sites on the α-MHC under physiological conditions and Ang II treatment conditions were determined by IP analysis. (**e**) HEK293T cells were transfected with plasmids expressing Myc-α-MHC WT, Myc-α-MHC K1249R, Myc-α-MHC K1533R and Myc-α-MHC K1897R. Equal amounts of lysates were prepared for IP with anti-Myc magnetic beads, followed by detection of Pan Kla. (**f**) HEK293T cells were transfected with plasmids expressing Myc-α-MHC WT, Myc-α-MHC K1249R, Myc-α-MHC K1533R and Myc-α-MHC K1897R without or with Ang II treatment. Equal amounts of lysates were prepared for IP with anti-Myc magnetic beads, followed by detection of Pan Kla. **g** Lactylated lysine residues were highlighted (red) in alignment of sequences surrounding K1897 in α-MHC homologs from diverse species.