Supplementary information

SIRT1 deacetylates the cardiac transcription factor Nkx2.5 and inhibits its transcriptional activity

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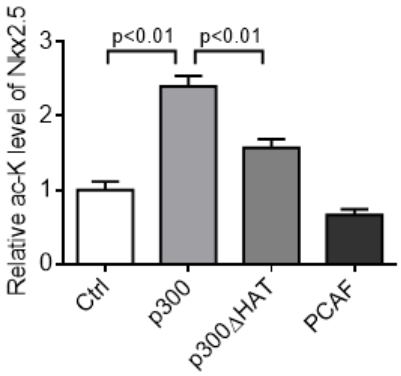
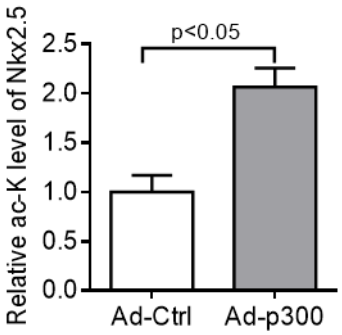
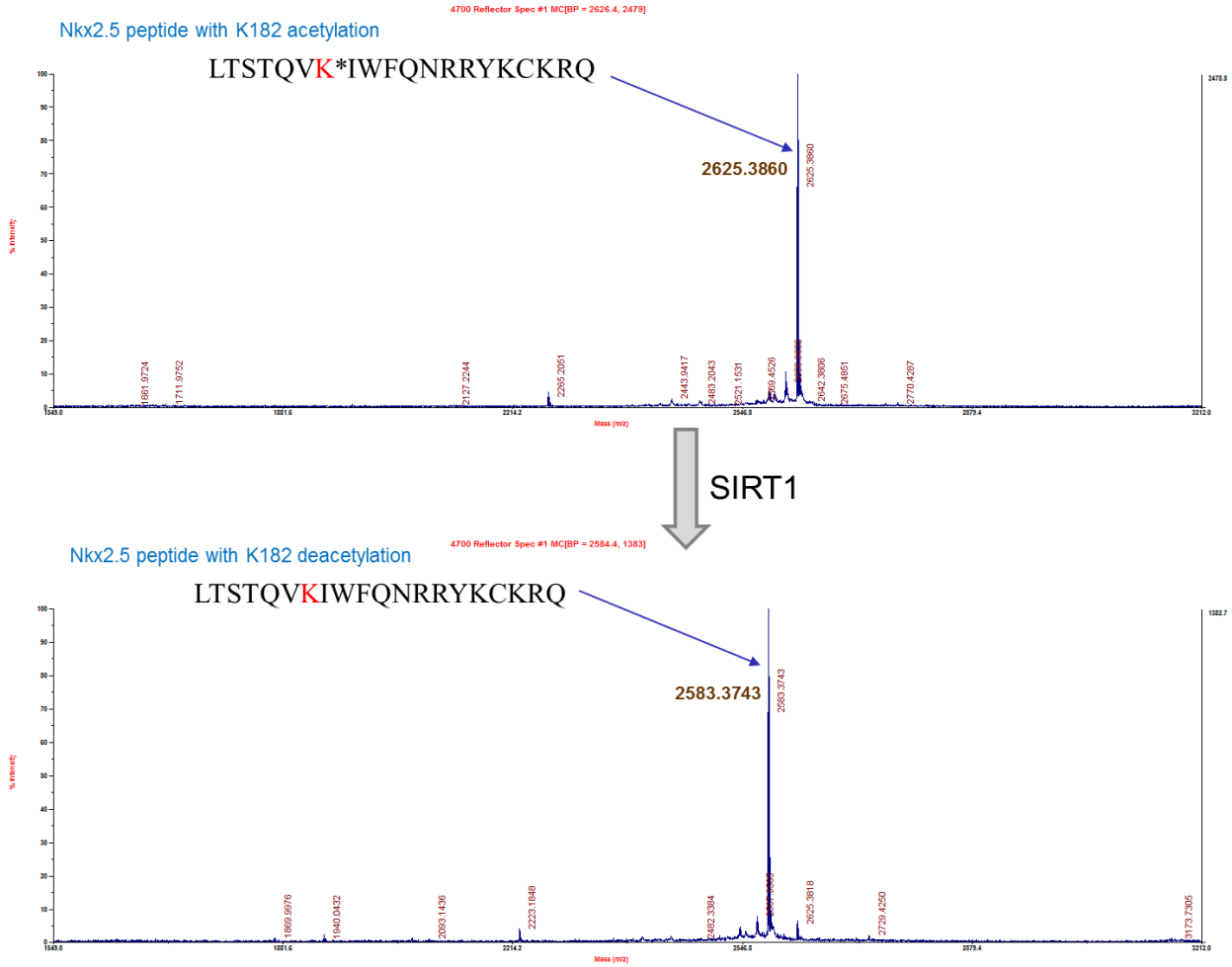


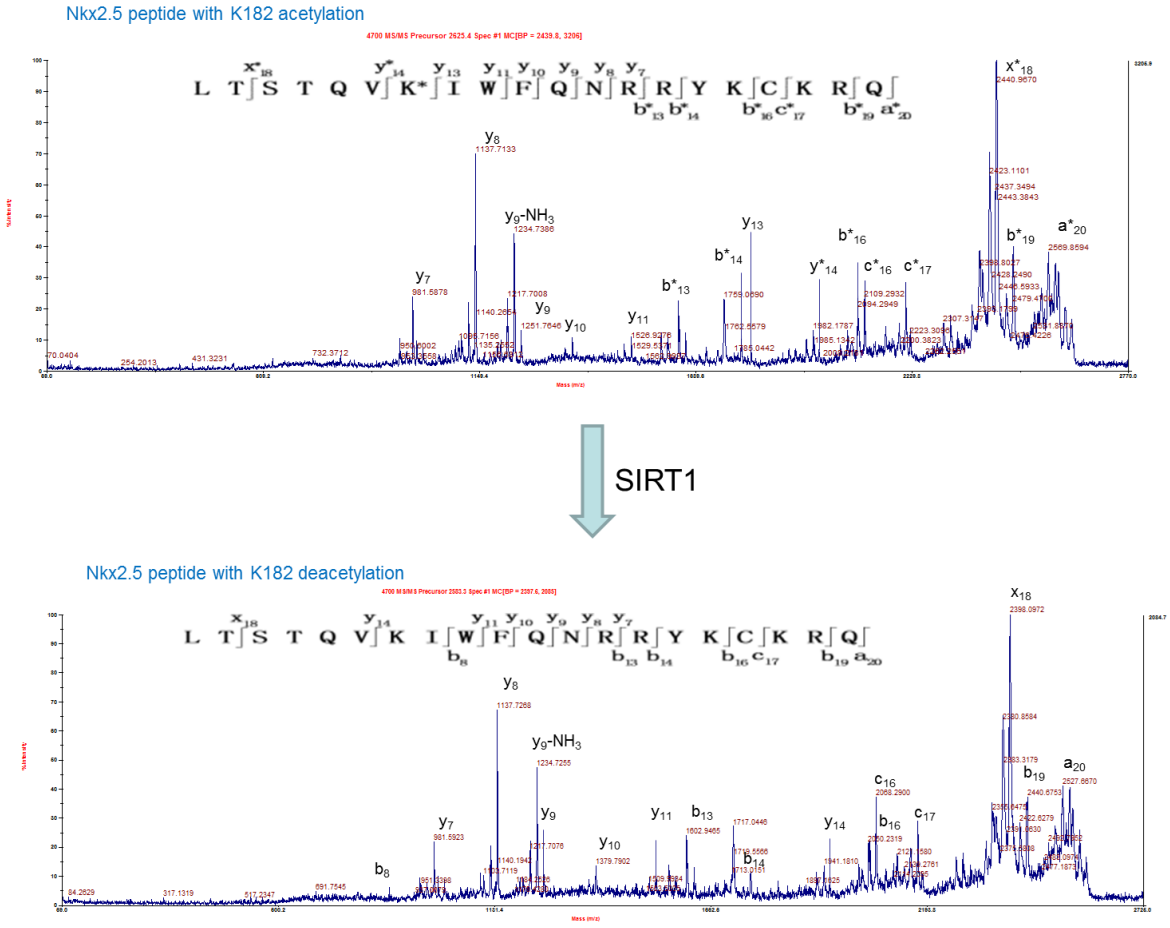
Supplementary Figure 1 Relative acetylated lysine (ac-K) level of Nkx2.5 (related to Figure 3b).



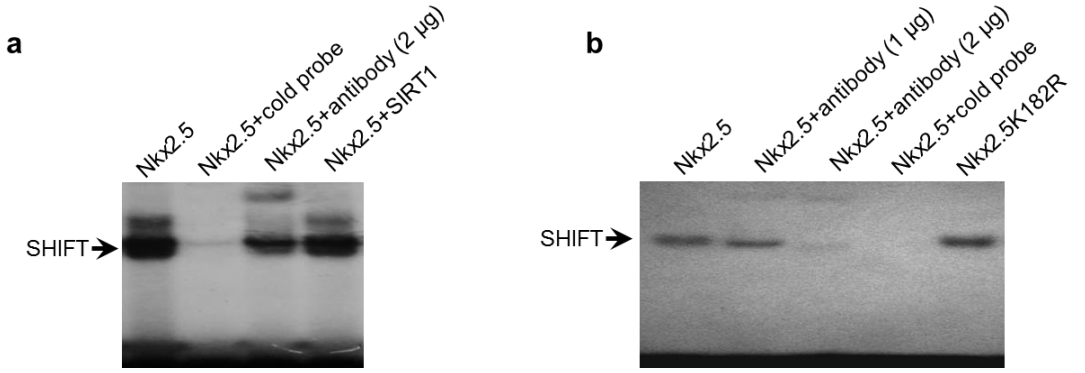
Supplementary Figure 2 Relative acetylated lysine (ac-K) level of Nkx2.5 (related to Figure 3d).



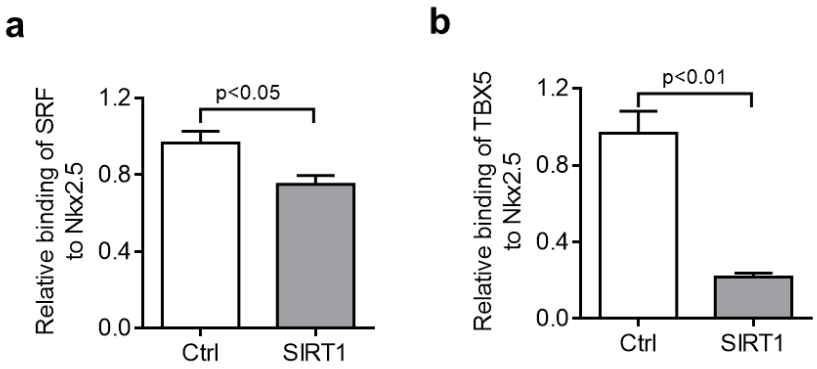
Supplementary Figure 3 Tandem MS analysis of the Nkx2.5 peptides containing acetylated K182 before and after the *in vitro* deacetylation assay.



Supplementary Figure 4 Tandem MS (MS/MS) analysis of the Nkx2.5 peptides containing acetylated K182 before and after the *in vitro* deacetylation assay.



Supplementary Figure 5 SIRT1 or K182R does not affect the DNA-binding ability of Nkx2.5.
(**a**) SIRT1 did not affect the DNA-binding ability of Nkx2.5. HEK293A cells were transfected with Nkx2.5 alone or together with SIRT1. Nuclear proteins were extracted and EMSA was performed with the Nkx2.5 binding element of the *ANF* promoter. Super-shift assay was performed with 2 μg anti-Nkx2.5 antibody.
(**b**) The DNA binding activity of Nkx2.5-K182R mutant is similar to that of WT-Nkx2.5. HEK293A cells were transfected with Nkx2.5, Nkx2.5 mutant (K182R) or control plasmids. Nuclear proteins were extracted, and EMSA was performed with Nkx2.5 binding element of the *ANF* promoter. Super-shift assay was performed with 1 or 2 μg anti-Nkx2.5 antibody.



Supplementary Figure 6 SIRT1 inhibits Nkx2.5 binding to its co-factors (related to Figure 7).
(**a**) Relative binding of SRF to Nkx2.5 in the presence/absence of SIRT1 (related to Figure 7a).
(**b**) Relative binding of TBX5 to Nkx2.5 in the presence/absence of SIRT1 (related to Figure 7b).