Effect of the acylation of TEAD4 on its interaction with co-activators YAP and TAZ

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Legend to the supplementary figures

Supplementary figure S1. Global view of the X-ray structure for myristoylated human hTEAD4$^{217-434}$ in complex with YAP$^{60-100}$. The coordinates have been deposited in the PDB databank (PDB access code = 5OAQ). hTEAD4$^{217-434}$ and hYAP$^{60-100}$ are represented by green and orange ribbons, respectively. The covalently bound myristate moiety is in purple. hTEAD4$^{217-434}$ has a similar structure to hTEAD1 and mTEAD4 in previously published structures [1, 2]. hYAP$^{60-100}$ contacts hTEAD4$^{217-434}$ via two distinct secondary structure elements (α-helix and Ω-loop) as already described [1, 2].

Supplementary figure S2. Stability of the immobilized TEAD4 proteins in SPR. The filled circles at cycle 0 are the calculated R_{max}^{theo} (see text for explanation). These values vary

between experiments, because different levels of TEAD4 have been immobilized. At the beginning and the end of every dose range experiment with a peptide to determine its K_d, a measurement with a saturating concentration of TAZ^{14-56} (1 µM) to estimate the stability of the immobilized TEAD4 protein was conducted. The filled circles represent the signal measured during these cycles. These experiments were performed at 10 and 25°C. The signal measured at 25°C (red filled circles) during these test cycles with Non-Acyl-TEAD4 and Cys367Ser decreased more rapidly than the signal measured with Acyl-TEAD4. This suggests a higher instability of the non-acylated proteins. At 10°C (blue filled circles), all 3 proteins were more stable, and the decrease in signal over the course of an experiment was largely reduced.

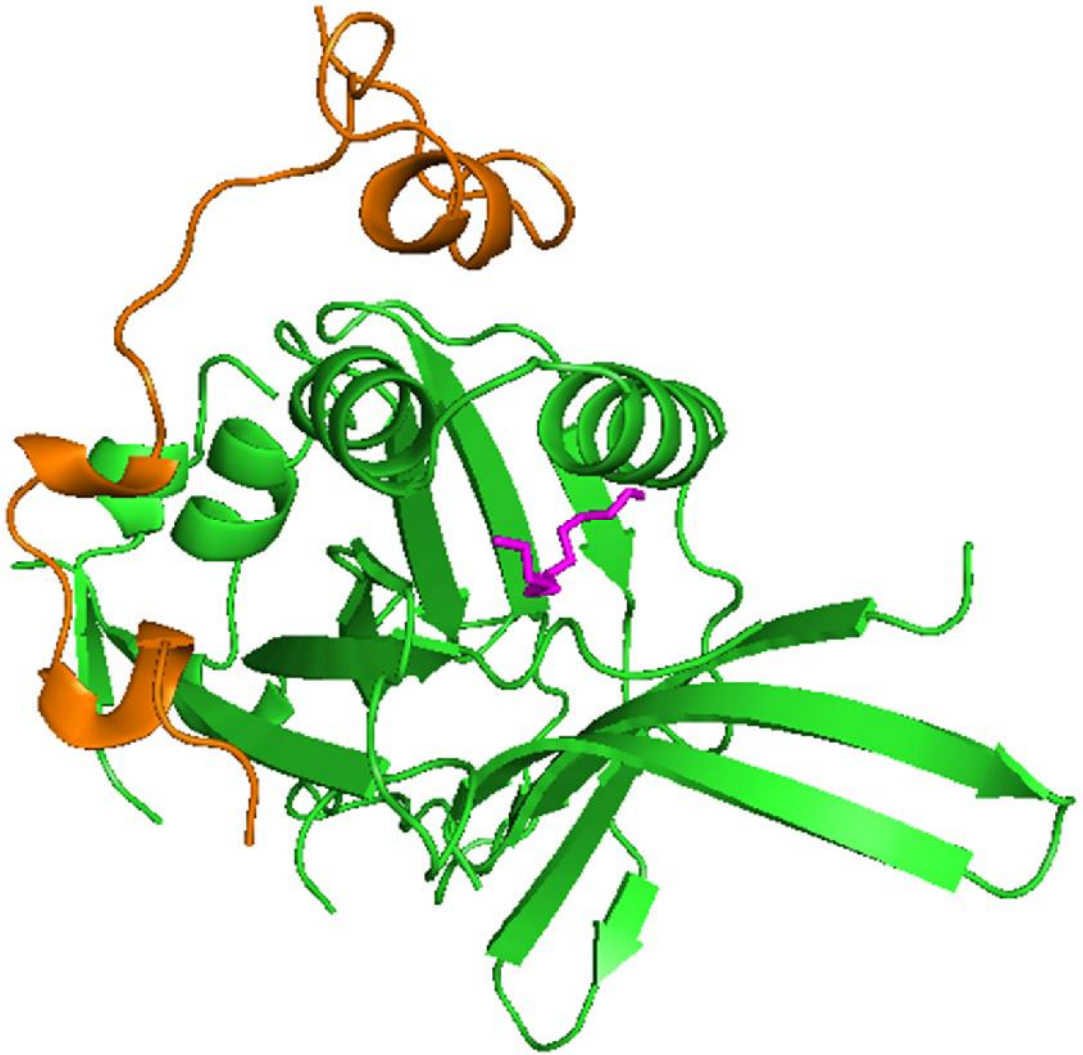
Supplementary figure S3. Thermal shift assay of refolded and non-refolded Cys367Ser. Ref-Cys367Ser (**A**) and non-refolded Cys367Ser (Non-Ref-Cys367Ser, **B**) possess a His_6-tag at their N-terminus in addition to an Avi-tag. The thermograms were obtained in a fluorescence-based thermal denaturation assay in the presence of 1-2 µM protein. The melting temperatures (T_m) were obtained by plotting the first derivative of the fluorescence emission (F) as a function of the temperature (-dF/dT). The curve minimum corresponds to T_m. The indicated T_m have been measured for the experiments depicted on the figure. These T_m are slightly different from the T_m presented on Table 2 because these later are the average values obtained from several independent measurements.

References

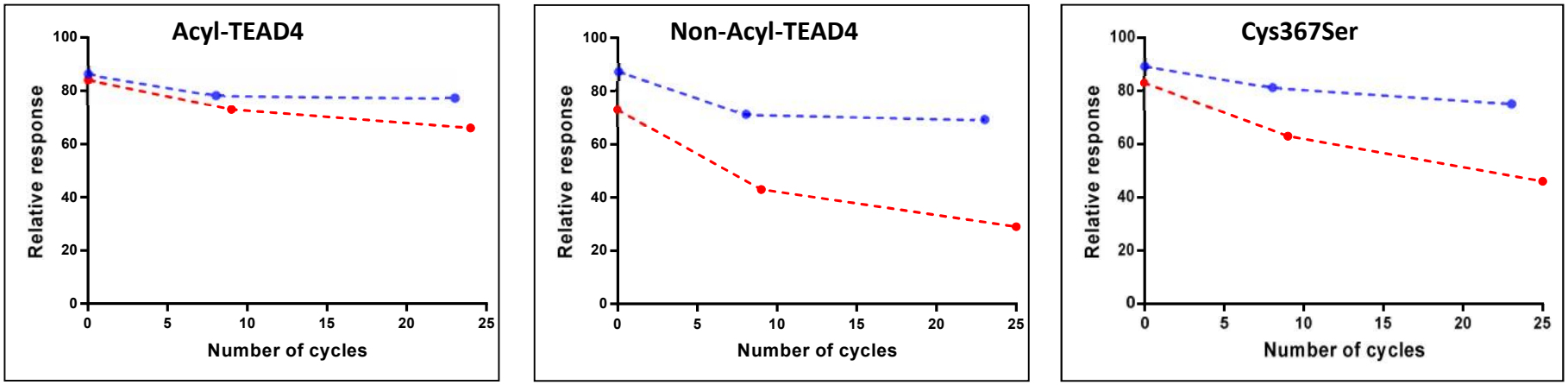
[1] Chen L, Loh PG, Song H. Structural and functional insights into the TEAD-YAP complex in the Hippo signaling pathway. Protein Cell. 2010;1:1073-83.

[2] Li Z, Zhao B, Wang P, Chen F, Dong Z, Yang H, et al. Structural insights into the YAP and TEAD complex. Genes & Dev. 2010;24:235-40.

Supplementary Fig. S1



Supplementary Fig. S2



SPR conducted at 10°C (●) or 25°C (●)

Supplementary Fig. S3