

Preferential domain orientation in HMGB2 protein revealed by NMR spectroscopy

○Jun-ichi Uewaki¹, Naoko Utsunomiya-Tate², and Shin-ichi Tate^{1,3}

¹ Dept. Mathematical and Life Sciences, School of Science, Hiroshima University,

² Research Institute of Pharmaceutical Sciences, Musashino University, ³ SENTAN/JST

High mobility group protein B2 (HMGB2) is a ubiquitously distributing DNA binding protein. HMGB2 bends double-stranded DNA (dsDNA) through its binding, thus its functions as a nucleosome chaperon. HMGB2 contains two DNA binding domains; A and B domain at N- and C-terminal parts, respectively. These two domains are linked by 10-residue linker. The N-terminal side of the linker contains YVPP sequence. In looking at the homologue protein HMGB1, this YVPP part in the linker is fixed through the CH- π interactions to the third helix, which may somehow restrict the direction of the linker. Based on the sequence conservation, this CH- π inter-locking interaction should be conserved in HMGB2.

The site-directed mutations to the conserved Pro residues (P80G/P81G) in the YVPP sequence have diminished the CH- π interaction that fixes the linker orientation. In measuring the anisotropic spin interactions for the aligned HMGB2 protein against the magnetic field, we found that the relative orientation between A and B domains has changed from that for the wild-type in Figs. (a) and (b); the observed rotation angle difference was 46 degrees. The present results may suggest the inter-domain linker, particularly the YXPP conserved sequence, should have a role to define the domain orientation in HMGB2, which presumably facilitates HMGB2 binding to dsDNA.

In the presentation, the functional significance of the domain orientation controlled by the short linker in HMGB2 will be discussed.

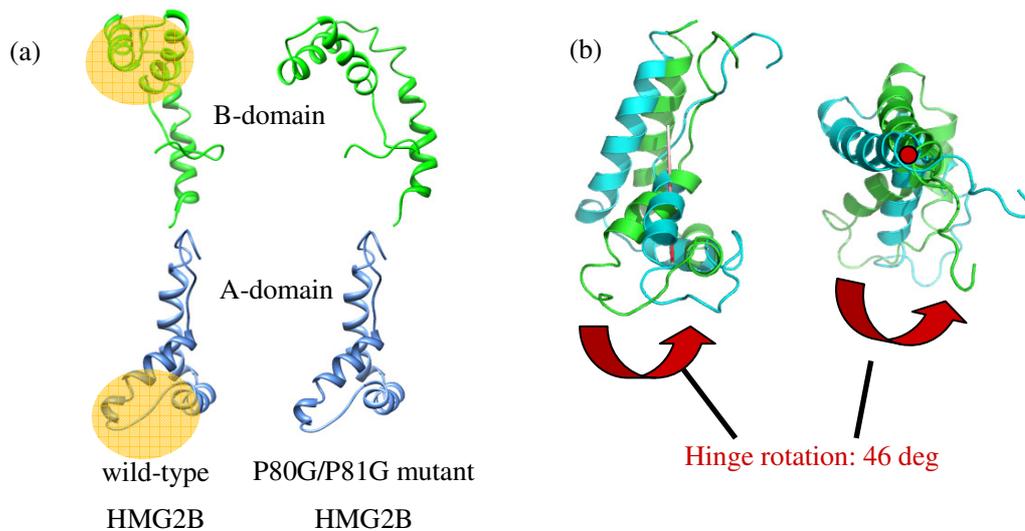


Fig. Relative domain orientations in wt-HMGB2 and P80G/P81G linker mutant

DNA interaction regions of HMGB2 are shown in circles. (a) Comparing the relative domain orientation in wild-type HMGB2 and P80G/P81G. (b) The rotation angle difference was 46 degrees.