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ROLE OF Mg^{2+} IONS IN DNA HYDROLYSIS BY *ECORV*, STUDIED BY 3D-RISM AND MD

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The role of Mg^{2+} ions in DNA hydrolysis by homodimeric restriction enzyme *EcoRV* was elucidated based on the three-dimensional reference interaction site model (3D-RISM-KH) theory and the molecular dynamics (MD) simulation. From an analysis of the spatial distribution of Mg^{2+} in an active site using 3D-RISM-KH, we identified a new position for Mg^{2+} in the X-ray *EcoRV*-DNA complex structure (1RVB), which turns out to play a crucial role in the reaction. We refer to the position as site IV[†]. Site IV[†] is almost the same position as that of a Ca^{2+} ion in the superimposed active-site structure of X-ray *PvuII*-DNA complex (1F00). The 3D-RISM-KH was also used to locate the position of water molecules including the water nucleophile at the active site. MD simulations were carried out with the initial structure having two Mg^{2+} ions at site IV[†] and at site I*, experimentally identified by Horton et al., to find a stable complex structure in which rearrangement of the DNA fragment occurred to orient the scissile bond direction toward the water nucleophile. The equilibrium active-site structure of *EcoRV*-DNA complex obtained in MD simulation was similar to the superimposed structure of X-ray *BamHI*-DNA complex (2BAM). In the active-site structure, two metal ions have the same position as that of 2BAM, and the scissile phosphate is twisted to orient the scissile bond toward the water nucleophile as is the case in 2BAM. We propose the equilibrium active-site structure obtained in this study as a precursor of the hydrolysis reaction of *EcoRV*.

Biography

Masayuki Irisa has completed a Doctoral program from Kyoto University without degree and got his PhD by the way of dissertation from Osaka University. He is an Associate Professor in Department of Bioscience and Bioinformatics, Faculty of Computer Science and Systems Engineering, Kyushu Institute of Technology, which belongs to a national university corporation, Japan. He has been serving as a Teacher and a Researcher in the Kyushu Institute of Technology for 20 years.

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