How limitless antibodies are generated by V(D)J recombination

Wei Yang[1], Xuemin Chen[1], Min-sung Kim[2], Martin Gellert[1]

[1]National Institutes of Health, USA, Bethesda, United States of America; [2]Pohang University, South Korea; wei.yang@nih.gov

Each of us has the potential to make trillions and more different antibodies and antigen receptors even though our genome contains only 3.2 billion base pairs! The antibody diversity is created by stochastic recombination of gene segments (V, D and J) that encode antibody heavy and light chains, and the process is known as V(D)J recombination. To initiate V(D)J recombination, the RAG1/2 recombinase cleaves DNA at a pair of recombination signal sequences (RSSs), the 12- and 23-RSS (12/23-RSS). DNA double strand cleavage is achieved in two consecutive steps, hydrolysis and strand transfer resulting in a DNA hairpin, in a single active site. Using X-ray crystallography and cryoEM, we have determined how two RSS DNAs are paired, nicked and completely cleaved at atomic resolution [1-3]. Both the protein and DNA undergo large conformational changes, and the active site of RAG1 re-arranges for DNA nicking and hairpin formation. Although RAG belongs to the RNH-type transposase family, RAG-catalyzed transposition is inhibited in developing lymphocytes. We show that by efficiently catalyzing the disintegration reaction that reverses the strand transfer, RAG avoids DNA transposition and consequent genome instability. The structures also rationalize many RAG mutations that cause immune deficiency in humans.

Keywords: RAG1-RAG2; 12-RSS; 23-RSS; antigen receptor, site-specific recombination