s8a.m6.p1 **Crystal structures of DNAs damaged by methoxylation reveal the reason why pyrimidine transition and purine transition occur during replication.** A. Takénaka*, T. Chatake, T. Hikima, T. Hossain, A. Ono[1], Y. Ueno[2] and A. Matsuda[2] *Graduate School of Bioscience and Biotechnology, Tokyo Institute of Technology, Yokohama 226-8501, Japan,* [1]*Graduate School of Science, Tokyo Metropolitan University, Hachioji, Tokyo 192-0364, Japan, and* [2]*Graduate School of Pharmaceutical Sciences, Hokkaido University, Sapporo 060-0812, Japan.*atakenak@bio.titech.ac.jp*
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In all organisms, the very high accuracy of DNA replication is achieved by using the Watson-Crick pairs between adenine(A) and thymine(T) residues and between guanine(G) and cytosine(C) residues as an absolute principle. However, oxy-amines such as hydroxyamine and methoxyamine disturb this rule and act as potent mutagens, causing nucleotide transitions from one purine to another purine, or from one pyrimidine to another pyrimidine. These chemicals predominantly attack and modify the exocyclic amino groups of nucleic acid bases. To understand the structural basis of mutation mechanism, it is necessary to clarify whether the modified bases can form Watson-Crick like pairs with non-Watson-Crick bases. Four kinds of DNA dodecamers containing 2'-deoxy-N^6-methoxyadenosine(mo^6A) and 2'-deoxy-N^4-methoxycytidine(mo^4C), with the sequences d(CGCGmo^6AATCCGCG), d(CGCGmo^6AATTCG CG), d(CGCAAATTmo^4CGCG), and d(CGCGAA TTmo^4CGCG) were synthesized and their crystal structures determined.

In these four dodecamers, the methoxy groups have no significant effects on the overall *B*-form duplex conformations. The mo^6A bases form Watson-Crick-like pairs with C as well as T of the opposite strands. To form hydrogen bonds for these pairings, the modified base must be in the amino form for T and in the imino form for C. In the same way, the mo^4C base can also present two alternate faces, the amino form for G and the imino form for A, so that mo^4C forms a Watson-Crick-like pair with A as well as G. An interesting finding in d(CGCGAATTmo^4CGCG) is that one of the two mo^4C bases, in the amino form, forms a regular Watson-Crick base pair with G, but the other wobbled in the imino form. The two faces found for mo^6A and mo^4C are ascribed to tautomerism between the amino and the imino forms, induced by the counter bases. In the other words, the methoxylation makes A possible to mimic G and makes C to mimic T, by which mis-incorporation is allowed in DNA replication. Based on these results, possible gene transition mechanisms are proposed.

s8a.m6.p2 **X-ray analysis of a DNA dodecamer containing 5-formyluracil.** M. Tsunoda, N. Karino [1], Y. Ueno[1], A. Matsuda[1], and A. Takénaka* *Graduate School of Bioscience and Biotechnology, Tokyo Institute of Technology, Yokohama 226-8501, Japan and* [1]*Graduate School of Pharmaceutical Sciences, Hokkaido University, Sapporo 060-0812, Japan.*atakenak@bio.titech.ac.jp*
Keywords: X-ray structure, 5-formyluracil, DNA.

Reactive oxygen radicals give damage on DNA, in which thymine bases are chemically modified by ionizing radiation to form 5-formyluracil (f^5U) through 5-methyl oxidation. Incorporation of f^5U into a synthesized DNA strand was observed for both templates, A and G, by *in vitro* DNA polymerization. In order to investigate how f^5U is incorporated for the template G and whether it still has an ability to form a canonical Watson-Crick base pair with A, a DNA dodecamer with the sequence d(CGCGAATXCGCG) containing f^5U at X was synthesized, and its crystal structure has been determined.

Single crystals obtained under two different conditions (cacodylate buffer containing spermine, 2-methyl-2,4-pentanediol and monovalent cation with or without divalent cation) were used for X-ray data collections with synchrotron radiation at 100K. The two data sets were processed separately, though their cell parameters showed no significant differences. Initial phases were derived by the molecular replacement method using the structure of the original dodecamer d(CGCGAATTCGCG). The two structures for the different data sets were refined independently with the program CNS.

The dodecamers form essentially *B*-form duplexes though including ribose with 3-end conformation locally. The formyl groups of f^5U bases adopt a *syn* conformation to the C^4 atom around the C^5-C^5M bond. In the crystal obtained from a solution containing no barium ion, however, the formyl group of one f^5U residue is disordered with *anti* conformation. Between the two crystals, the primary waters in the major and minor grooves are found at the same positions, but the others are differed. In both crystals, f^5U forms a pair with A on the opposite strand in a Watson-Crick fashion. The present X-ray structures indicate that f^5U derived from modification of thymine can form a canonical base pair with A. This is one of the reasons why f^5U is incorporated into newly synthesized DNA instead of T.