Design of intelligent nucleobases and DNA HOMO mapping

Isao Saito
Department of Synthetic Chemistry and Biological Chemistry, Kyoto University, Kyoto 606-8501, Japan

ABSTRACT
We have designed a new nucleobase, benzodeazaadenine (BD\textsubscript{A}) that has a stronger charge transport ability than guanine and is not destroyed during charge transport process. By incorporating this new nucleobase into DNA, we demonstrated a protocol for real DNA nano-wire that is far superior to natural DNA. We also demonstrated that the selectivity for the interaction of Mn(II) ion with guanine N7 in G runs is a HOMO-controlled process, and as a consequence, the selectivity for G-metal ion interactions obtained by \textsuperscript{15}N-NMR studies would directly reflect the HOMO distribution of G-containing sequences in B-DNA.

INTRODUCTION
The incorporation of artificial nucleobases into oligonucleotides has become increasingly important in molecular biology, genetics, molecular medicine and DNA microarray technology. For example, fluorescent DNA bases have been widely used as fluorescent marker for detecting nucleic acids or monitoring changes in their structure and dynamics in various contexts.

For the past several years we have designed many different types of intriguing molecules that are useful for chemical genomics and molecular biology. In the first part of my talk I would like to overview such molecules developed in our laboratory. Then, recent work on the design of DNA nano-wire using artificial nucleobases which is on-going in our laboratory will be presented. Finally, a new method for mapping HOMO of duplex DNA based on \textsuperscript{15}N-NMR data is described.

RESULTS and DISCUSSION
Design of DNA nano-wire containing benzodeazaadenine (BD\textsubscript{A}) as an artificial base
Guanine radical cation (hole) produced by one-electron oxidation of DNA is known to migrate to remote GG sites through DNA \pi-stacks. Water and/or oxygen can trap holes eventually producing guanine-damaged sites. Since hole migration through DNA \pi-stack always competes with hole trapping, destruction of guanine base is inevitable in charge transport process through natural DNA (1).

We have designed a new nucleobase, benzodeazaadenine (X = BD\textsubscript{A}) that has a stronger hole transport ability than guanine and is not destroyed during hole transport process. By incorporating BD\textsubscript{A} in duplex DNA, we have demonstrated a protocol for real DNA nano-wire that is quite stable and has a higher charge transport ability than natural DNA. Evaluation of hole migration efficiency
through these artificial DNA nano-wires was estimated by $G_b/G_a$ as depicted in Fig 1.

![Diagram of DNA nano-wires](image)

Figure 1. Evaluation of hole transport efficiency through duplex DNA containing $^{38}$A (°™ P U) and hole-generating nucleobase U* (°™ P U) upon photoirradiation. Hole transport efficiency was obtained by band intensity ratio $G_b/G_a$ of distal GGG and proximal GGG.

Mapping of HOMO of Duplex DNA

The highest occupied molecular orbital (HOMO) of organic molecules plays an important role in chemical reactions by interacting with LUMO of reactant molecules. We proposed for the first time an experimental method for mapping HOMO of B-DNA using Co(II) ion and benzoyl peroxide in combination with ab initio MO calculations of G-rich sequences and suggested an important but unestablished binding force, the interaction of the HOMO of duplex DNA with LUMO of DNA binders such as metal ions, drugs and proteins (2).

In the present study we examined the interaction of G runs with Mn(II) and Co(II) ions by means of $^{15}$N-NMR using ODNs containing $^{15}$N-enriched G at N7 at specific site of GG and GGG. As increasing the concentration of Mn(II), broadening of the signals of 5'G of GG and the middle G of GGG occurred selectively as shown in Fig 2. These selectivities were in good agreement with HOMO distributions in G runs obtained by recent high level theoretical calculations (3). The interaction of electron-deficient metal ions with N7 of electron-rich G in G runs is likely a HOMO-controlled process, and as a consequence, the selectivity of the G–metal ion interactions obtained by the $^{15}$N-NMR study would directly reflect the HOMO distribution in G-containing sequences.

![Diagram of GGG run interactions](image)

Figure 2. Change of linewidth observed in $^{15}$N-NMR (35 mM sodium phosphate, pH 7.0) of GGG-containing self-complementary duplexes by changing manganese chloride concentration. $r = [\text{Mn}^{2+}] / [\text{phosphate}]$.

![Diagram of HOMO distribution](image)

Figure 3. Line broadening of $^{15}$N-labeled G-N7 in G runs by Mn(II) as compared with calculated HOMO distributions

REFERENCES

