ACS Catalysis

Research Article

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Development of DNA-Based Hybrid Catalysts through Direct Ligand Incorporation: Toward Understanding of DNA-Based Asymmetric Catalysis

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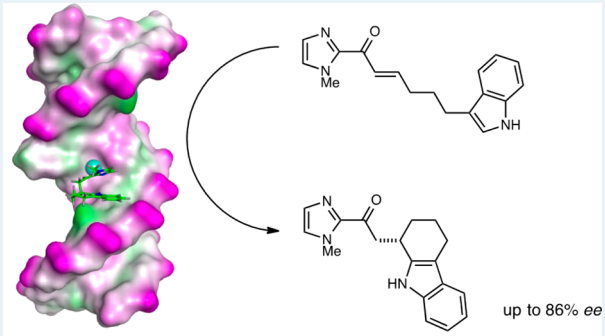
S *Supporting Information*

ABSTRACT: We have developed a DNA-based hybrid catalyst containing an intrastrand bipyridine ligand through direct ligand incorporation and successfully performed asymmetric intramolecular Friedel–Crafts alkylations. This is the first report on the DNA hybrid catalyst system where an intrastrand ligand is covalently introduced into the phosphate backbone. We have generated a series of active site to investigate the structural details of DNA hybrid catalysts and demonstrated that catalytic properties of DNA hybrid catalysts are governed by the disposition of the metal-binding site in the DNA duplex, the size of catalytic cavity, and the composition of nucleobases in the catalytic pocket.



DNA-based hybrid catalyst with intrastrand ligand

KEYWORDS: *DNA, hybrid catalyst, asymmetric catalysis, Friedel–Crafts reaction, direct ligand incorporation, intrastrand bipyridine ligand*

DNA is one of the most plentiful and naturally occurring helical polymers on Earth. Recently, this ubiquitous helical polymer has gathered attention as a chiral source for asymmetric synthesis.[1−5] In 2005, Feringa and Roelfes introduced the concept of DNA-based hybrid catalysts based on supramolecular assembly by using a copper complex of a nonchiral ligand that can bind to DNA.[6] Since then, DNA-based hybrid catalysts have been applied to various key asymmetric carbon–carbon or carbon–heteroatom bond-forming reactions.[6−17] It is evident that the enantioselectivity of the reactions originated in DNA; however, the stereo-induction mechanism is not fully understood and remains a challenge.[18−20] At this stage, there is no X-ray crystal structure that can be used to propose DNA hybrid catalysts, and NMR-based studies of DNA hybrid catalysts are restricted, because of the presence of the copper(II) complex. In this context, we have developed asymmetric intramolecular Friedel–Crafts alkylations using a DNA-based hybrid catalyst and suggested a plausible intercalation binding model that was supported by the melting temperature and the viscosity of the DNA solution in the presence of copper complexes.[21,22] However, previous studies using DNA-based hybrid catalysts based on a supramolecular assembly have provided limited information about the crucial region in DNA duplex to induce the enantioselectivity and the effect of individual bases on the catalytic performance, since it is difficult to control the location of the metal–ligand complex in the DNA. For the precise positioning of the metal–ligand complex in DNA, the covalent anchoring strategies have been applied in the form of post-synthetic modification of oligonucleotides or the introduction of modified nucleotides during solid-phase synthesis.[24−27] In this study, we devised a new DNA hybrid catalyst based on the direct incorporation of a ligand into the DNA phosphate backbone to develop a more-advanced catalytic system with an objective of understanding the structural and mechanistic features. Herein, we report on the development of new DNA hybrid catalysts with an intrastrand bipyridine ligand and its application to the asymmetric intramolecular Friedel–Crafts alkylations. The catalytic site could be readily and rationally constructed for the structural investigation of DNA hybrid catalysts. The results indicate that catalytic behavior of DNA hybrid catalyst apparently depends on the disposition of the

Received: July 27, 2014
Revised: September 29, 2014
Published: October 8, 2014

   dx.doi.org/10.1021/cs501086f | *ACS Catal.* 2014, 4, 4070−4073

binding site for metal ions in the DNA duplex and the composition of nucleobases in the vicinity of the intrastrand bipyridine ligand.

Taking synthetic efficiency into consideration, we chose the commercially available 2,2′-bipyridine-5,5′-dicarboxylic acid as an intrastrand ligand and introduced linkers with different lengths via condensation reactions (see Figure 1a).[28−30] The

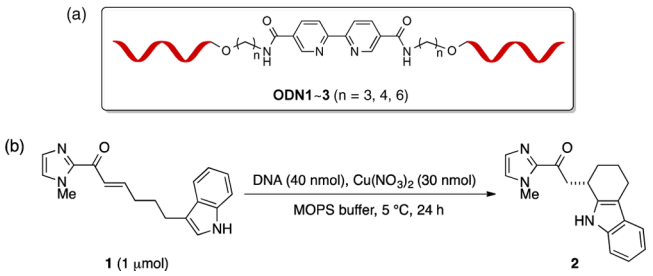


Figure 1. (a) Chemical structure of **ODN1−ODN3** with an intrastrand bipyridine ligand. (b) DNA-based intramolecular Friedel−Crafts alkylation.

phosphoramidites of bipyridine ligands were synthesized by phosphitylation and incorporated into the DNA oligonucleotides by automated solid-phase synthesis (see Scheme S1, presented in the Supporting Information). Based on our previous study, we performed the asymmetric intramolecular Friedel−Crafts alkylation of **1** with various DNA-based hybrid catalysts consisting of DNA duplexes with intrastrand bipyridine ligands and copper(II) complex in water at 5 °C for 1 day (see Figure 1b).[22] The results are summarized in Table 1.

The intrastrand bipyridine ligand was incorporated into the center of 13-mer DNA oligonucleotides 5′-d-(GCATGG^{n}**X**CACGGT)-3′, where ^{n}**X** (where n is the number of methylene groups in the linker; n = 3, 4, 6,) represents a bipyridine ligand. To investigate the effect of the size of the binding site, we altered the length of the linkers on the bipyridine ligand and hexyl, butyl, and propyl linkers were selected in this study. Complementary strands of **ODN1−ODN3** contained no base or natural bases (A, T, G, and C), and **ODN4−ODN8** were also prepared to investigate the counter base effect on the intrastrand binding ligand. As shown in Table 1, the present DNA-based hybrid catalyst provided the cyclized product with significant enantioselectivity. The absolute configuration of the product was determined by HPLC analysis using procedures described previously.[22,23] In the present reaction system, the (*S*)-enantiomer was obtained as a major product. As shown in Table 1, we found that the enantioselectivities obtained varied according to the counter base present in the complementary strand. For instance, DNA-based hybrid catalysts obtained by the combination of propyl linkers and adenine as a counter base gave the cyclized product **2** with 29% ee (entry 2 in Table 1). In the case of the absence of a base, an ee of 40% was obtained (entry 1 in Table 1). Overall, pyrimidine bases were more beneficial for the yield enantioselectivity than purine bases were. For example, the DNA-based hybrid catalysts obtained by the combination of propyl linkers and thymine as a counter base gave the cyclized product **2** in 60% yield and 63% ee (entry 3 in Table 1). Interestingly, when the complementary strand possessed cytosine as a counter base, enantioselectivity was increased dramatically to 84% ee (entry 5 in Table 1). A strong effect of the counter base located in the complementary strand on the

Table 1. Asymmetric Intramolecular Friedel−Crafts Alkylation Catalyzed by DNA-Based Hybrid Catalysts with Propyl Linkers[a]

entry	DNA sequence (with propyl linker)	ee[c,d] (%)	yield[c,e] (%)
1[b]	5'-GCATGG^{3}**X**CACGGT-3' (**ODN3**) 3'-CGTACC**dS**GTGCCA-5' (**ODN4**)	40	28
2	5'-GCATGG^{3}**X**CACGGT-3' (**ODN3**) 3'-CGTACC **A** GTGCCA-5' (**ODN5**)	29	24
3	5'-GCATGG^{3}**X**CACGGT-3' (**ODN3**) 3'-CGTACC **T** GTGCCA-5' (**ODN6**)	63	60
4	5'-GCATGG^{3}**X**CACGGT-3' (**ODN3**) 3'-CGTACC **G** GTGCCA-5' (**ODN7**)	17	20
5	5'-GCATGG^{3}**X**CACGGT-3' (**ODN3**) 3'-CGTACC **C** GTGCCA-5' (**ODN8**)	**84**	71
6	5'-GCATG**A**^{3}**X****T**ACGGT-3' (**ODN9**) 3'-CGTAC**T** **C** **A**TGCCA-5' (**ODN10**)	18	43
7	5'-GCATGGGCAC^{3}**X**GT-3' (**ODN11**) 3'-CGTACCCGTG **C** CA-5' (**ODN12**)	−6	29
8	5'-GCATGG^{3}**X**CACGGTT 3'-TTTCGTACC **C** GTGCCAT (**ODN13**)	86	80

[a]Experiments were carried out with 3.3 mM of a tethered indole substrate, 0.13 mM of DNA, 0.1 mM of $Cu(NO_3)_2$ at 5 °C in 20 mM MOPS buffer (pH 6.5) for 1 day. [b]5′-*O*-Dimethoxytrityl-1′,2′-dideoxyribose-3′-[(2-cyanoethyl)-(*N*,*N*-diisopropyl)]- phosphoramidite was used as a dSpacer (dS). [c]Results represent the average value of two experiments. The yields and enantioselectivity were determined by chiral HPLC analysis. [d]Reproducible within ±5%. [e]Reproducible within ±10%.

dx.doi.org/10.1021/cs501086f | *ACS Catal.* 2014, 4, 4070−4073

enantioselectivity of the product was demonstrated, with cytosine being the best counter base for this reaction. To investigate the effect by the size of the binding site, we altered the length of the linkers on the bipyridine ligand and carried out the asymmetric intramolecular Friedel−Crafts alkylation (see Table S2 in the Supporting Information). Although it is difficult to verify the influence by the size of the binding site for DNA hybrid catalysts including abasic site, A or G as a counter base, because of their low enantioselectivities, DNA hybrid catalysts containing pyrimidine bases (T or C) as a counter base showed the significance of the size of the binding site; DNA hybrid catalysts having propyl linkers gave rise to higher enantioselectivities than DNA hybrid catalysts having hexyl or butyl linkers. For instance, DNA hybrid catalyst based on cytosine as a counter base, when the length of the linkers of the bipyridine ligand was reduced from hexyl (n = 6) to propyl (n = 3) groups, the ee value increased from 63% ee to 84% ee, respectively (entry 5 in Table S2 in the Supporting Information and entry 5 in Table 1). For further study of the effect of sequence on enantioselectivity, next, we focused on the neighboring bases of the intrastrand bipyridine ligand. The change of the neighboring bases from −**G**^{3}X**C**− to −**A**^{3}X**T**− led to a significant drop in enantioselectivity (18% ee) (entry 6 in Table 1).

This result showed that a neighboring G−C pair is more suitable for the present reaction, compared with an A−T pair. The position of the binding ligand on the DNA duplex was investigated. When the intrastrand ligand was located away from the center of the DNA duplex, the ee value was largely decreased (−6% ee, entry 7 in Table 1). This informed us that the location of the intrastrand ligand in the center of the DNA duplex is important for inducing the enantioselectivity. Finally, using optimized DNA sequences, a hairpin-type hybrid catalyst (**ODN13**) was prepared and examined in the present reaction. The hairpin-type hybrid catalyst gave the product with 86% ee and confirmed the potential of this DNA hybrid catalyst (entry 8 in Table 1). These results indicated clearly that the perturbation of the catalytic site affects the selectivity of the reactions significantly. Based on the results obtained in this study, the following points regarding the DNA hybrid catalytic system should be considered: (i) the counter base of the ligand, (ii) the size of the binding (catalytic) pocket, (iii) the neighboring bases of the ligand, and (iv) the disposition of the ligand. These factors are considered to have a direct effect on the binding of the metal complex and on the formation of the DNA-based hybrid catalyst.[31−33] In turn, this will affect the enantioselectivity of the product. Based on these observations, a suitable DNA duplex was determined for the intramolecular Friedel−Crafts alkylation of substrate **1** and, thus, a molecular model of the DNA-based hybrid catalyst was constructed. An oligonucleotide with the sequence 5′-d-(GCATGG^3**X**CACGGT)-3′/5′-d(ACCGTGCCCATGC)-3′ (**ODN3**/**ODN8**) was selected as a receptor for the copper(II) complex during molecular modeling. We utilized circular dichroism (CD) spectroscopy to investigate the overall duplex conformation of DNA hybrid catalyst and confirmed that the DNA duplex formed by **ODN3**/**ODN8** in the presence of the copper(II) complex remained the typical B-DNA structure, despite the incorporation of intrasrand bipyridine ligand (see Figures S2 and S3 in the Supporting Information). In this context, the molecular mechanics optimization of the DNA hybrid catalyst consisting of **ODN3**/**ODN8** and the copper(II) complex was performed (Figure S5 in the Supporting Information). Subsequently, the energy minimization of the molecular assembly of product **2** and the DNA hybrid catalyst was performed by docking the most-stable conformer of the product manually into the DNA sequences. Considering that the most-stable conformer of the product is assumed to be similar to the transition state of intramolecular cyclization, this conformer was selected to bind the DNA catalytic pocket.

As mentioned above, in the present study, an (*S*)-enantiomer was obtained as a major product. The lowest-energy conformer of the (*S*)-enantiomer was determined based on a dihedral angle (O1C5C4N1 = −25.4°), considering the coordination of the carbonyl oxygen and imidazole nitrogen to the copper(II). The complex between the stable conformer and the DNA hybrid catalyst was minimized using the AMBER force field in the presence of a 10 Å layer of water and counterions. Consequently, we found that the (*S*)-enantiomer bound snugly to *the major groove of the duplex* when it formed a complex with the copper complex and resided inside the DNA pocket (see Figure 2). The DNA onto which the (*S*)-enantiomer complex

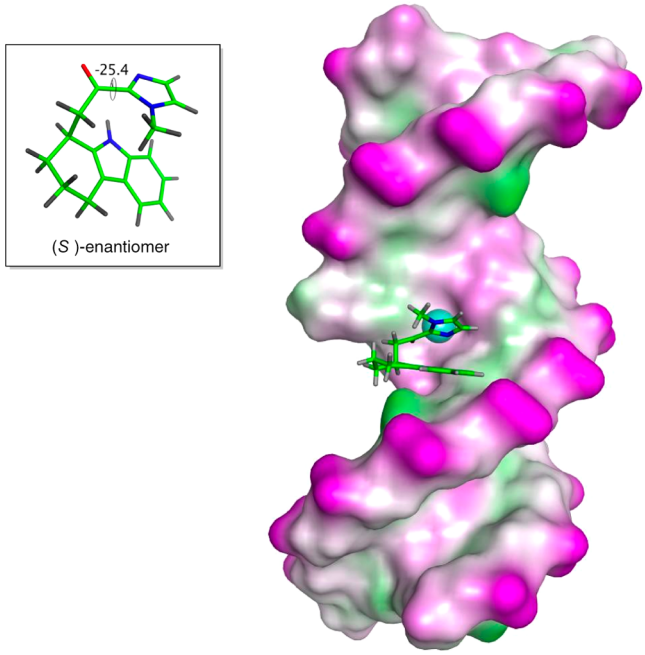


Figure 2. Energy-minimized complex of the (*S*)-enantiomer and copper(II)−**ODN3**/**ODN8**. The color of molecular surface represents the lipophilicity (pink denotes hydrophilic, white denotes neutral, and light green denotes lipophilic).

was docked afforded lower energy structures than did the DNA onto which the (*R*)-enantiomer complex was docked (see Figure S6 in the Supporting Information). These results are in agreement with the experimental findings. In addition, it is worthy of note that the energy-minimized complex of the (*S*)-enantiomer with copper(II)−DNA suggests the major groove as a binding site. In the previous study, we presumed that the narrow minor groove of the DNA duplex might be more suitable to provide a chiral environment, compared with the spacious major groove.[22] Still, we cannot exclude the minor groove as a catalytic site, considering the elongation by intercalation ligand, and further studies are underway to clarify the stereoinduction mechanism of DNA-based asymmetric synthesis.

dx.doi.org/10.1021/cs501086f | *ACS Catal.* 2014, 4, 4070−4073

In conclusion, we have developed a new type of DNA hybrid catalysts with an intrastrand bipyridine ligand via direct incorporation into the DNA phosphate backbone. The present DNA hybrid catalysts were optimized by the rational construction of the active site and identified as an efficient catalyst for asymmetric intramolecular Friedel–Crafts alkylations. We have demonstrated that the catalytic abilities of DNA hybrid catalysts are dependent on the disposition of the binding site for metal ions in the DNA duplex and the combination of nucleobases in the catalytic site. In addition, we found that the combination of the copper(II) ion, an intrastrand bipyridine ligand, and cytosine as a counter base plays an important role in affording high enantioselectivity for asymmetric intramolecular Friedel–Crafts alkylations. We believe that the present study advances the understanding of DNA hybrid catalysts considerably and provides valuable information for rational tailoring of DNA-based hybrid catalysts. We are currently developing fine-tuned DNA hybrid catalysts based on the results from this study.

■ ASSOCIATED CONTENT

Supporting Information

Synthetic routes of intrastrand bipyridine ligands, characterization data of new compounds, HPLC data, ESI-TOF-Mass data and CD spectra of oligonucleotides. This material is available free of charge via the Internet at http://pubs.acs.org.

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Notes

The authors declare no competing financial interest.

■ ACKNOWLEDGMENTS

We express our sincere thanks for the CREST grant from the Japan Science and Technology Corporation (JST), grants from the WPI program (iCeMS, Kyoto University), and for the global COE program from the Ministry of Education, Culture, Sports, Science and Technology (MEXT), Japan.

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