Supporting Information

Sensitive analysis of DNA methyltransferase based on a hairpin-shaped DNAzyme

Tian Tian, ^{†+} Heng Xiao, ^{†+} Yuelin Long[†], Xiaoe Zhang[†], Shaoru Wang, ^{†*} Xiang Zhou^{*† ¶}, Songmei Liu, ^{$\overline{+}$} Xin Zhou^{$\overline{+}}$ </sup>

[†] College of Chemistry and Molecular Sciences, Key Laboratory of Biomedical Polymers of Ministry of Education, Wuhan University, Wuhan, Hubei, 430072, P. R. of China.,

¶ State Key Laboratory of Natural and Biomimetic Drugs, Peking University, Beijing, China.

⁺ Zhongnan Hospital, Wuhan University, Wuhan, Hubei 430072, P. R. of China



DNA methylation plays important roles in a variety of biological processes, and their aberrant profiles have been found in many diseases. Herein, the analysis of DNA methylation and MTase (methyltransferase) activity is pretty important in epigenetic study. We developed an effective strategy for sensitive analysis of MTase activity based on a hairpin shaped DNAzyme. Dam MTase has been chose as the model, and 8-17 DNAzyme amplicon has been adopted and found to be very effective in such analysis. With the newly developed platform, a sensitive and convenient detection of MTase activity could be achieved.

Contents List

1.1 General information.

1.2 Table S1

1.3 Figure S1

1.4 Figure S2

Electronic Supplementary Material (ESI) for Chemical Communications This journal is C The Royal Society of Chemistry 2012

1.1 General information: MgCl₂, Tris base and NaCl were purchased from Acros and the oligonucleotides were purchased from Invitrogen Technology(Shanghai, China). DNA adenine methylation(Dam) MTase, DpnI and S-adenosylmethionine (SAM) were purchased from New England Biolabs (NEB) Inc. Fluorescence was measured by LS55 Perkin Elmer. All measurements were performed at room temperature.

Oligomer	Sequence(from 5'to 3')
Dam F1	AGAAGAG ATGGATCAAG
	TAACCAATGTGCAGA <i>CTTGATC</i>
	CATCTCTTCTCCGAGCCGGTCG
	AAATAGTGGGTG
Dam F2	GAAGAG ATGGATCAAG
	TAACCAATGTGCAGACTTGATC
	CATCTCTTCTCCGAGCCGGTCG
	AAATAGTGGGTG
Dam F3	AAGAG ATGGATCAAG
	TAACCAATGTGCAGACTTGATC
	CAT CTCTT CTCCGAGCCGGTCG
	AAATAGTGGGTG
Dam F4	AGAG ATGGATCAAG
	TAACCAATGTGCAGACTTGATC
	CAT CTCT TCTCCGAGCCGGTCG
	AAATAGTGGGTG
Dam F5	GAG ATGGATCAAG
	TAACCAATGTGCAGACTTGATC
	CATCTCTTCTCCGAGCCGGTCG
	AAATAGTGGGTG
MB substrate	/FAM/-
	CCACCACATTCAAATTCACCAACTATrAGGAAG-
	AGATGTTACGAGGCGGTGGTGG-/BHQ/

1.2 Table S1 Sequences of DNA probes used in methylation analysis.

Electronic Supplementary Material (ESI) for Chemical Communications This journal is O The Royal Society of Chemistry 2012

1.3 Figure S1



Figure S1 Preliminary dynamic assay of fluorescence response of different hairpin-shaped DNA probes. The concentration for DNA probe is 1.0 μ M and concentration of Dam MTase are 200 U/mL. The methylation buffer contained 50 mM of NaCl, 10 mM of tris-HCl (pH7.5), 10 mM of MgCl₂. The following cleavage reaction was conducted in a buffer containing 5 mM of NaCl, 1 mM of tris-HCl (pH7.5), 1.5 mM of MgCl₂ and 0.1 μ M of MB substrate. The curves were obtained with a kinetic mode on a fluorescence spectrameter.

Electronic Supplementary Material (ESI) for Chemical Communications This journal is C The Royal Society of Chemistry 2012

1.4 Figure S2



Figure S2 Time-dependent fluorescence response of different concentrations of hairpin-shaped DNA probe and Mg^{2+} . The methylation buffer contained 50 mM of NaCl, 10 mM of tris-HCl (pH7.5), 10 mM of MgCl₂, 200 U/mL of Dam Mtase, 400U/mL of DpnI and a varing concentrations of Dam F3 probe(the concentrations were specified in the curves). The following cleavage reaction was conducted in a buffer containing 5 mM of NaCl, 1 mM of tris-HCl (pH7.5), 0.1 μ M of MB substrate and a varing concentrations of MgCl₂(the concentrations were specified in the curves). The curves were obtained with a kinetic mode on a fluorescence spectrameter.