

# Searching for a New Paradigm in Metal-DNA Interactions: Selection and Characterization of Novel Metal-specific DNAzymes

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One of the most significant discoveries in bioinorganic chemistry is that DNA can be enzymes (and thus called DNAzymes) and almost all DNAzymes are metalloenzymes under physiological conditions. While much information has been obtained on metal-binding sites in proteins, similar information in nucleic acids is missing. To search for such a paradigm in metal-DNA interactions that help catalyze diverse ranges of reactions, we sought to use combinatorial biology technique called *in vitro* selection to obtain new DNAzymes from a large DNA library that are specific for metal ions such as Pb(II), Co(II), Zn(II), As(III), As(V), Fe(II), Fe(III), Hg(II) and U(VI) (1,2). By introducing a negative selection strategy into the *in vitro* selection process, a DNAzyme specific for Co(II) has also been developed and characterized, which may prove useful as a Co(II) spectroscopic handles to provide direct information about the ligand binding pocket (3). In order to select for DNAzyme systems that possess both high activity and increased structural stability, a Zn(II)-specific DNAzymes that is active at 90 °C has been selected (4). Further selections and characterization of the DNAzymes (5) continue to demonstrate the adaptability of DNAzymes in forming metal specific ligand binding sites that will provide fundamental information to fill the current void of structure/function information on DNA-metal interactions. Practical applications of those DNAzymes as fluorescent and colorimetric biosensors for metal ions have also been demonstrated (6-8).

Sequence Alignment of Co<sup>2+</sup>-dependent DNAzymes

**Clone 18 5'-TCTC TTGTATTAGCTACACTGTTAGTGGATCGGGTCTAATCTCG GTG-3'**  
**Clone 11 5'-TCTC TTGTATTAGCTACACTGTTAGTGCATCGTTTTAATCTCG GTG-3'**

1) Li, J. *et al.*, (2000) *Nucleic Acids Res.* 28, 481; 2) Lu, Y., (2002) *Chem. Eur. J.* 8, 4588; 3) Bruesehoff, P.J. *et al.*, (2002) *Combin. Chem. High Throughput Screening* 5, 327; 4) Nelson, K.E. *et al.*, *J. Mol. Evol.*, in press; 5) Brown, A.K., *et al.*, (2003) *Biochemistry*, 42, 7152; 6) Li, J. & Lu, Y. (2000) *J. Am. Chem. Soc.*, 122, 10466. 7) Liu, J. & Lu, Y., (2003) *J. Am. Chem. Soc.*, 125, 6642; 8) Liu, J. & Lu, Y., (2004) *J. Am. Chem. Soc.* 126, 12298.