

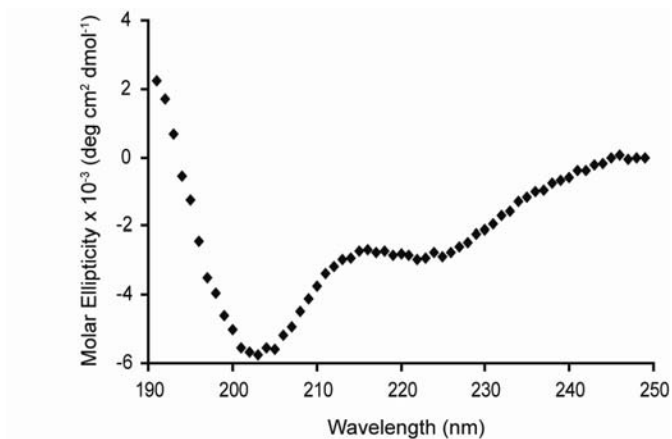
Selection and Evolution of Enzymes  
from a Partially Randomized Non-Catalytic Scaffold

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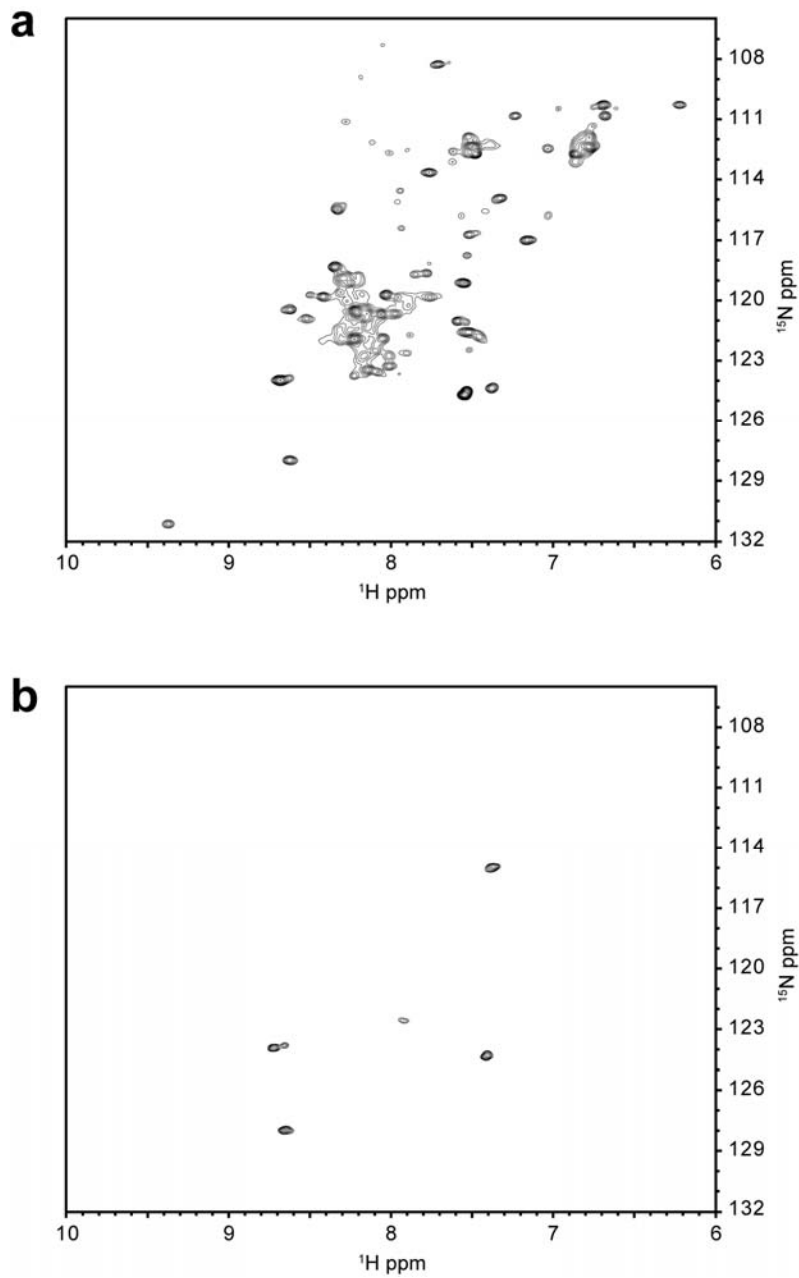
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This PDF file includes:

Supplementary Figures 1 and 2  
Sequences of oligonucleotides, DNA library, selected clones



Supplementary Figure 1 | Far UV-CD spectrum of ligase #6



**Supplementary Figure 2 | Two-dimensional  $^1\text{H}$ - $^{15}\text{N}$ -HSQC NMR of ligase #6.**

**a**, Uniformly  $^{15}\text{N}$ -labelled protein. **b**, Selectively  $^{15}\text{N}$ -cysteine labelled protein.

**Oligonucleotides** (DNA unless stated otherwise)

BS47, 5'-TTCTAATACGACTCACTATAGGAGACTCTTT

BS48, 5'-AAAGAGTCTCCTATAGTGAGTCGTATTAGAA

BS50, 5'-pTTTTTTTTTTTTTTTTTTTTTCCCAGATCCAGACATTC

BS56, 5'-CATATGGGAATGTCTGGATCTGGGAAAAAAAAAAAAAAAAAAAA  
GAGTCTCCGCGAACGTTAGACAGTGTGACTTCGTCATGCTATTCA

BS3long, 5'-

TCTAATACGACTCACTATAGGGACAATTACTATTTACAATTACAATGGACT

BS24RXR2, 5'-

TTAATAGCCGGTGCCAGATCCAGACATTCCCATAGAACCGCCATGATGATG

XL-PSO, X(tagccggtg)AAA AAA AAA AAA AAA ZZ ACC P

Synthesized at the W. M. Keck Facility at Yale University with the following reagents from Glen Research (Sterling, VA): X=psoralen C6, lower case=2'OMe, Z=spacer 9, P=puromycin, stretch of A's and ACC was DNA.

HO-substrate (PC-biotin), 5'-PC-UCACACUGUCUAACGUUCGC

PC biotin = PC biotin phosphoramidite (Glen Research, Sterling, VA), all nucleotides are RNA

splint RNA, 5'-GAGUCUCCGCGAACGU, all nucleotides are RNA

BS63, 5'-AGGATTATAGCATATGGACTACAAGGACGACGACG

BS65, 5'-ATGTTTCAGACCTCGAG TTAATAGCCGGTGCCAGAT

BS68, 5'-

AGGATTATAGCATATGGACTACAAGGACGACGACGACAAGGGCGGAAAGCA  
CATCTGC

BS70, 5'-

TCATTCAGACCTCGAGTTAATAGCCGGTGCCAGATCCAGACATTCCCATAGA  
ACCGCCATGATGATGGTGGTGGTGGTACTACCTCCTCCTGCACGGCTTCC

HO-substrate (11mer), 5'-CUAACGUUCGC, all nucleotides are RNA

**DNA library sequence**

5'-

TCTAATACGACTCACTATAGGGACAATTACTATTTACAATTACAATGGACTAC  
AAGGACGACGACGACAAGGGCGGAAAGCACATCTGCGCCATCTGTGGAGAT

N<sub>36</sub>

TCCTGTGAGGGCTGTAAAGGCTTCTTCAAGCGCACCGTGAGAAAGGACCTGA  
CCTACACCTGTTCGGGACAACAAGGATTGT N<sub>27</sub>

TGCCAGTACTGTAGGTACCAGAAGGCCCTCGCCATGGGCATGAAAAGGGAGG  
CCGTGCAGGAAGAGGTAGGTAGTCACCACCACCATCATCATGGCGGTTCTAT  
GGGAATGTCTGGATCTGGCACCGGCTATTAA

**Sequences of selected clones from round 17**

b5,

MDYKDDDDKSGKHICAICGDYIPEEDSHRDGDSCEGCKGFSKRTVRKDLTYTCR  
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f9,

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MSGSGTGY

e3,

MDYKDDDDKGGKHICAICGDVVDTADAKTQYDSCGGCKGIPKRTERKELTYTC  
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g9,

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GMSGSGTGY

e5,

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MSGSGTGY

h4,

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e7,

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b1,

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