

Supplementary Information for MS #2003-10-10586, A 1.7-Kilobase Single-Stranded DNA that Folds into a Nanoscale Octahedron, by W. M. Shih, J. D. Quispe & G. F. Joyce

Supplementary Table 1. Sequences of the heavy and light chains and the oligodeoxynucleotides used to assemble the heavy chain. An initial version of the heavy chain, without any unpaired thymidine residues at the vertices, was constructed in four parts by PCR assembly from oligodeoxynucleotides X48-71, A0-1, B0-1, C0-1, D0-1, E0-1, F0-1, and G0-1. The parts then were assembled using unique internal restriction sites. This construct was prone to significant aggregation during folding (data not shown). A revised version containing two unpaired thymidines in all four strands at all six vertices was constructed in two parts by overlap extension PCR, employing fragments that were amplified using the initial construct as a template and oligodeoxynucleotides X226-251 as primers. The two fragments then were assembled using a unique internal restriction site.

Heavy chain: 5'-GGCGATATCGATACATGGACGGAGAATACGAGCAACGGACCACTTT
TGTGGTGGAGCCATCAGGTTTCGCGCCACCATTGGTCGCATAGAGCGTGGATCTTCGT
TTTCGAGCATGGACGCGGTAGCCGACGCATCCTGGCGCGTTGCACTTATCCGGACTA
GATGCGCTGATCTGGGACGTCGATTTGCCGACGCCAAGGAGCGCCGTGACGCATCT
CGCTGGTTTTCCATGGTGGTGGCGCCAAGTCGCTACTAGTCGTCGGCTTGGGGGACGC
ATCAGCTGTAGGTTACGCAGAGCTGGTTTTCCATGTGGCCGTGGAGGTTACAGCGCG
TTGGTCCCCCTTCGTGAAGTGGACATTTTTGTCCTGGCGTCTGGTACCTATGCCTGGC
ATTTTTGCCACCTGATGCAAGATTTCCGCCGAGCCGTTTCATAAACCTTCATCAACCCG
GCCTTTTGGCGCTGTAGATGTACCTCTATGGAGTCTTCGGCGATTGCGGGCATCAAC
AGCCATGACTTCACGCCTTGGTGCTTTTGCATCGTGACGTGGATTGCTGGCGCTAGG
TGCCCGCTTCGGCGCGCCGGTTCACCTACAGTTCAGAGCGAGCCGCTTTTGCAGAGTCC
TTCTGGAGAGGAGTGTAGAGTCGCGCCGTTTCTTGCATCAGGACTTGCTACCAGTCG
ATTGGACTTCACGTTATCGACGTGGTGAGGATCCTCACCTGGTTGCTATCAGAGGCA
CCTGCTCCTGCAGGAGCACCGGATAAGTGCTTGGAGGGAGCTCGCCGTAAACTGTC
GTCAACCGGAGGTTTTCTACTCTAGACGCCTCTCTACGAGGACTTCCCTCCTTCCTG
ATGGCTCCTGCCGATCATGCATTCGGCCTGTGCTGCTTCGCCCCGCCAAGGTGTAAA
GTCAGCCATGTTGACGGTTTTCCGCCTAGCTGGCGCAATCTACATCACGACGGGGCG

TTTGTTTCGGTGGCCTCTCGAGAGGCCTGGAACCGACTTCTCCGTCCTGTTAAAGCTTT
AACACCGATCCGGATGTTGGCGGCGCGGGTTCATCAAGGTTCATAAACGGCCGCTTT
TGCGAGACTCTATGGAGGTAGATGTACAGCCGCCGCCTTGTGGGGCGCTCTGGTTAA
ACCTAGACATGATGCCGGTTTTCCGCAACGCTGTCAACCTCTAACGCCACAGCCCCA
CTTCCCGGGCGCTAGAGCCATCTGGCTGCGCGTCCCTGCTTTTGCACCCTGCCGCAT
GGTCCTGGCGGTCCAGCCCCGGGTTCATCCGGATCGGACAGGGCCTGTTGTCGTAGGA
CCGAACATTGGCAGAGTGGATATTTTTATCCTGCACTGGGAATTCGTAGCCTGTGGT
TTTCCACACCACTGGCAGAATTGCCTCGTGGGACGGCGCGCCAGAGCCATCTAGCTG
CTTTTGCATGGACCTGGCGGACCAGCGCGCAGGGACGAGGCTTGCCGAGCGCGAGA
CGCATCACGGAGCGCCTTGGAGGTTTTCTACTAGTCGCTACTTGGTGCGCCACCAG
CTCGGCTTCGCGCCGAGATCCTGCATCTATGGTCGCAATGGACGTTTTTCGTGGATGC
CGACGCTACCTGCACCATGCCGGCGCGTTTTCTGCCAGTGGACAACCTGATTCTAGTC
CCGGACTCTGCCTTGCAGCACACCATGTATCGCTGAGGTCTGCC-3'

Light chain 1: 5'-CGGCACCGTTGCTCGTATTCTCCGTGGCCGAATGCATGAT-3'

Light chain 2: 5'-ATAGCAACCACCCAGATCAGCGCATCTAGTGGTGCCTCTG-3'

Light chain 3: 5'-CCAGACGCCACCAATCGACTGGTAGCAAGTGGCATAGGTA-3'

Light chain 4: 5'-GTCGGTTCACCTACGACAACAGGCCCTGTGGACGGAGAA-3'

Light chain 5: 5'-TCCCAGTGCACCGGGACTAGAATCAGTTGTGGCTACGAAT-3'

Oligodeoxynucleotides used to assemble the heavy chain:

X48: 5'-GGCGGCGGCTTCGAGCAACGGACCACTTTTGTGGTGGAGCCATCAGGCGCG
CCACCATTGGTCGCATAGAGCGTGGATCT-3'

A0: 5'-GCGCGCCACCATTGGTCGCATAGAGCGTGGATCTTCGTTTTCGAGCATGGAC
GCGGTAGCCGACGCATCCTGGCGCGC-3'

X49: 5'-CCCAGATCAGCGCATCTAGTCCGGATAAGTGCCGCGCCAGGATGCGTCGGC
TACCGCGTCCATGC-3'

X50: 5'-ACTAGATGCGCTGATCTGGGACGTCGATGCCGACGCCAAGGAGCGCCGTGA
CGCATCTCGC-3'

B0: 5'-GGCCGACGCCAAGGAGCGCCGTGACGCATCTCGCTGGTTTTCCATGGTGGTG
CGCCAAGTCGCTACTAGTCGTCGGCC-3'

X51: 5'-GCTCTGCGTGAACCTACAGCTGATGCGTCCCCCGCCGACGACTAGTAGCGAC
TTGGCGCACCACCA-3'

C0: 5'-GGGGGGACGCATCAGCTGTAGGTTACGCAGAGCTGGTTTTCCATGTGGCCG
TGGAGGTTACAGCGCGTTGGTCCCCCC-3'

X52: 5'-GGCATAGGTACCAGACGCCAGGACAAAAATGTCCACTTCACGGGGGGACCA
ACGCGCTGAACCTCCACGGCCACA-3'

X53: 5'-TGGCGTCTGGTACCTATGCCTGGCATTTTTGCCACCTGATGCAAGATCGCCG
AGCCGTTCATAAACCTTCATCAACCCG-3'

D0: 5'-CCCGCCGAGCCGTTCATAAACCTTCATCAACCCGGCCTTTTGGCGCTGTAGAT
GTACCTCTATGGAGTCTTCGGCGGG-3'

X54: 5'-CCAAGGCGTGAAGTCATGGCTGTTGATGCCCGCTCGCCGAAGACTCCATAG
AGGTACATCTACAGC-3'

E0: 5'-GGCGGGCATCAACAGCCATGACTTCACGCCTTGGTGCTTTTGCATCGTGACGT
GGATTGCTGGCGCTAGGTGCCCCGCC-3'

X55: 5'-GCTCGCTCTGAACTGTAGTGAACCGGCGCGCCGGCGGGCACCTAGCGCCAG
CAATCCACGTCACGA-3'

F0: 5'-GCGGCGCGCCGGTTCCTACTACAGTTCAGAGCGAGCCGCTTTTGCAGATCCTTCT
GGAGAGGAGTGTAGAGTCGCGCCGC-3'

X56: 5'-CCAATCGACTGGTAGCAAGTCCTGATGCAAGACGGCGCGACTCTACACTCCT
CTCCAGAAGGACT-3'

X57: 5'-GGTGCCTCTGATAGCAACCAGGTGAGGATCCTCACCACGTCGATCGTGAAG
TCCAATCGACTGGTAGCAAGT-3'

X58: 5'-TGGTTGCTATCAGAGGCACCTGCTCCTGCAGGAGCACCGGATAAGTGCGGA
GGGAGCTCGCCGTAAACTGTCGTCAACCGG-3'

F1: 5'-GGGAGGGAGCTCGCCGTAAACTGTCGTCAACCGGAGGTTTTCTACTCTAGA
CGCCTCTCTACGAGGACTTCCCTCCC-3'

X59: 5'-GGCCGAATGCATGATCGGCAGGAGCCATCAGGGGAGGGAAGTCCTCGTAGA
GAGGCGTCTAGAGT-3'

X60: 5'-TGCCGATCATGCATTCGGCCTGTGCTGCCGCCCCGCCAAGGTGTAAAGTCAG
CCATGTTGA-3'

E1: 5'-CCGCCCCGCCAAGGTGTAAAGTCAGCCATGTTGACGGTTTTCCGCCTAGCTGG
CGCAATCTACATCACGACGGGGCGG-3'

X61: 5'-GGACGGAGAAGTCGGTTCAGGCCTCTCGAGAGGCCACCGAACACGCCCCG
TCGTGATGTAGATTGCGCCAGCTAGG-3'

X62: 5'-TGGAACCGACTTCTCCGTCCTGTTAAAGCTTTAACACCGATCCGGATGGGCG
GCGCGGGTTCATCAAGGTTTCATAAACGGC-3'

D1: 5'-GGGCGGCGCGGGTTCATCAAGGTTTCATAAACGGCCGCTTTTGCGAGACTCTA
TGGAGGTAGATGTACAGCCGCCGCCC-3'

X63: 5'-GCATCATGTCTAGGTTTAACCAGAGCGCCCCACGGCGGCGGCTGTACATCTA
CCTCCATAGAGTCT-3'

C1: 5'-GGGGGGGCGCTCTGGTTAAACCTAGACATGATGCCGGTTTTCCGCAACGCTG
TCAACCTCTAACGCCACAGCCCCCCC-3'

X64: 5'-GGGACGCGCAGCCAGATGGCTCTAGCGCCCGGGGTGGGGCTGTGGCGTTAG
AGGTTGACAGCGTTG-3'

G0: 5'-AGGGTGGCGCTAGAGCCATCTGGCTGCGCGTCCCTGCTTTTGCACCCTGCCGC
ATGGTCCTGGCGGTCCAGCCACCCT-3'

X65: 5'-CCTACGACAACAGGCCCTGTCCGATCCGGATGCCCGGGCTGGACCGCCAGG
ACCATGCGGCAGGG-3'

X66: 5'-GGCTACGAATTCCCAGTGCAGGATAAAAATATCCACTCTGCCTGTTTCGGTCC
TACGACAACAGGCCCTGT-3'

X67: 5'-TGCACTGGGAATTCGTAGCCTGTGGTTTTCCACACCACTGGCAGAAGCCTCG
TGGGACGGCGCGCCAGAGCCATCTAGC-3'

G1: 5'-ACCCTCGTGGGACGGCGCGCCAGAGCCATCTAGCTGCTTTTGCATGGACCTG
GCGGACCAGCGCGCAGGGACGAGGGT-3'

X68: 5'-CCAAGGCGCTCCGTGATGCGTCTCGCGCTCGGCGCCTCGTCCCTGCGCGCTG
GTCCGCCAGGTCCA-3'

B1: 5'-GGCCGAGCGCGAGACGCATCACGGAGCGCCTTGGAGGTTTTCTACTAGTCG
CTACTTGGTGCGCCACCAGCTCGGCC-3'

X69: 5'-CCATTGCGACCATAGATGCAGGATCTCGGCGCGGCCGAGCTGGTGGCGCAC
CAAGTAGCGACTAGT-3'

A1: 5'-GCGCGCCGAGATCCTGCATCTATGGTCGCAATGGACGTTTTCTGTGGATGCCG
ACGCTACCTGCACCATGCCGGCGCGC-3'

X70: 5'-CCGGGACTAGAATCAGTTGTCCACTGGCAGAACGCGCCGGCATGGTGCAGG
TAGCGTCGGCATCC-3'

X71: 5'-GGCGGCGGCTATTCTCCGTCCATGAAAACATGGTGTGCTGCGGCAGAGTCC
GGGACTAGAATCAGTTGT-3'

X226: 5'-GGCGATATCGATACATGGACGGAGAATACGAGCAACGGACCACTTTTG-3'

X227: 5'-GGCAGACCTCAGCGATACATGGTGTGCTGCGGCAGAGT-3'

X228: 5'-GCGACCAATGGTGGCGCGAACCTGATGGCTCCACCACA-3'

X229: 5'-CCGACGCATCCTGGCGCGTTGCACTTATCCGGACTAGA-3'

X230: 5'-GCGCTCCTTGGCGTCCGCAAATCGACGTCCCAGATCAG-3'

X231: 5'-TCGCTACTAGTCGTCGGCTTGGGGGACGCATCAGCTGT-3'

X232: 5'-AAAAATGTCCACTTCACGAAGGGGGACCAACGCGCTGA-3'

X233: 5'-TTGCCACCTGATGCAAGATTTGCGCCGAGCCGTTTCATAA-3'

X234: 5'-ATGGCTGTTGATGCCCCGAATCGCCGAAGACTCCATAG-3'

X235: 5'-CTGGCGCTAGGTGCCCCGCTTCGGCGCGCCGGTTCACTA-3'

X236: 5'-GCAAGTCCTGATGCAAGAAACGGCGCGACTCTACACTC-3'

X237: 5'-AGTCGATTGGACTTCACGTTATCGACGTGGTGAGGATC-3'

X238: 5'-TTACGGCGAGCTCCCTCCAAGCACTTATCCGGTGCTCC-3'

X239: 5'-CTACGAGGACTTCCCTCCTTCCTGATGGCTCCTGCCGA-3'

X240: 5'-TTACACCTTGGCGGGGCGAAGCAGCACAGGCCGAATGC-3'

X241: 5'-CTACATCACGACGGGGCGTTTGTTCGGTGGCCTCTCGA-3'

X242: 5'-TGATGAACCCGCGCCGCCAACATCCGGATCGGTGTTAA-3'

X243: 5'-AGATGTACAGCCGCCGCCTTGTGGGGCGCTCTGGTTAA-3'

X244: 5'-ATGGCTCTAGCGCCCGGGAAGTGGGGCTGTGGCGTTAG-3'

X245: 5'-CTGGCGGTCCAGCCCGGGTTCATCCGGATCGGACAGGG-3'

X246: 5'-AAAAATATCCACTCTGCCAATGTTTCGGTCCTACGACAA-3'

X247: 5'-TCCACACCACTGGCAGAATTGCCTCGTGGGACGGCGCG-3'

X248: 5'-ATGCGTCTCGCGCTCGGCAAGCCTCGTCCCTGCGCGCT-3'

X249: 5'-GTGCGCCACCAGCTCGGCTTCGCGCCGAGATCCTGCAT-3'

X250: 5'-AGTTGTCCACTGGCAGAAAACGCGCCGGCATGGTGCAG-3'

X251: 5'-CTAGTCCCGGACTCTGCCTTGCAGCACACCATGTTTTTC-3'

Supplementary Figure 1. Projections of the model and corresponding class averages for a reconstruction performed imposing octahedral symmetry with an angular interval of 5 degrees. For each comparison, the projection is shown on the left (numbered c0–c62) and the class average is shown on the right (listing the number of particles used to create that image).

Supplementary Figure 2. Three orientations of a map obtained from a reconstruction performed on images collected at 1.0 μm defocus. For the first dataset (described in the main text), in which paired images were collected at 0.6 and 2.0 μm defocus, the 0.6 μm defocus data could not be used for reconstruction because of poor contrast. A second set of paired images was collected from an independent grid at 1.0 and 2.0 μm defocus. For this second data set, the 1.0 μm defocus images had sufficient contrast for reconstruction. 773 particle images taken at 1.0 μm defocus were used for this reconstruction, for which octahedral symmetry was imposed using a 3 degree angular interval, producing a map with a resolution of 30 Å. The resulting map closely matches the one obtained from the first data set, indicating that radiation damage was not an issue.



