

# **SUPPLEMENTARY INFORMATION FOR**

## **Translation of DNA Signals into Polymer Assembly Instructions**

**Shiping Liao & Nadrian C. Seeman\***

**Department of Chemistry**

**New York University,  
New York, NY 10003, USA.**

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### **Experimental Methods**

This section contains experimental details for procedures that are standard (*S1*), but which have been modified slightly in the work described here.

*Device Assembly.* The strands were designed using the program *SEQUIN* (*S2*). The component strands of diamonds I, II, III, IV and V (Figure 1b) were dissolved separately to a concentration of 1  $\mu$ M in a solution containing 40 mM Tris, pH 8.0, 20 mM acetic acid, 2 mM EDTA and 12.5 mM magnesium acetate (TAEMg), and the mixtures were heated to 90 °C and cooled slowly to 4 °C. Diamonds II and III were then combined, as were diamonds IV and V; these mixtures were heated to 48 °C and cooled slowly to 4 °C. In the following, diamond I is termed U1, complex II+III is termed U2 and complex IV+V is termed

U3. The component strands of PX1, PX2, DX1, DX2, DX3, DX4, DX5, DX6, DX7, DS1, and DS2 (Figure 1c) were dissolved separately to a concentration of 2  $\mu$ M in the same solution. These mixtures were heated to 90 °C (5 min), and cooled as follows: 65 (30 min), 45°C (30 min), 37°C (30 min), 4°C (30 min). U1, PX1, U2, PX2, and U3 were combined, heated to 40 °C and cooled slowly to 4 °C.

*Setting the Device State.* Unset strands were added to the solution and the solution was kept at 20 °C 3 hours; the solution was treated with streptavidin beads at 20 °C for 30 minutes to remove the set-strand/unset-strand duplexes. At this point the set strands for the target JX<sub>2</sub> or PX states were added to the solution and kept at 20 °C for 3 hours to establish the device conformation.

*Preparing and Purifying the Assembly Complex.* DS1 and DX complexes 1-7 were added to the solution. The mixture was heated with the following thermo-cycling protocol: 35 °C (10 min), 33 °C (10 min), 30 °C (10 min), 25 °C (10 min), 20 °C (10 min) for 15 cycles. The solution was treated with magnetic streptavidin beads at 20 °C 30 minutes to bind devices with an intact left-hand side; failed assemblies are washed away and the solution containing the beads is replaced with a new solution containing 40 mM Tris, pH 8.0, 20 mM acetic acid, 2 mM EDTA, 12.5 mM magnesium acetate, 50 mM potassium acetate and 1 mM DTT. Ten units of Sma I (NEB) were added and the solution was incubated at 20 °C for 1 hour to release the target assembly, and the beads then removed from the solution. DS2 was then added, and the solution was incubated at 20 °C for 2 hr, followed by magnetic streptavidin bead treatment at 20 °C for 30 minutes only those with an intact right-hand side should be bound. The solution was replaced with a new solution containing 40 mM Tris, pH 8.0, 20 mM acetic acid, 2 mM EDTA, 12.5 mM magnesium acetate, 50 mM potassium acetate and 1 mM DTT.

Ten units of Apa I (NEB) were added, and the solution was incubated at 20 °C for 1 hour and the magnetic beads were removed.

*Ligation and Analysis.* The solution was brought to 1 mM in ATP and 10 units of T4 polynucleotide ligase (USB) were added. The ligation proceeded at 16 °C for 16 hours. Following ligation, the solution was heated at 90 °C for 5 minutes, and the ligation products were purified using 6% denaturing PAGE. The ligation products were sequenced to establish the correct assembly. A few missed or unknown bases are noted in the experimental sequencing, but these are far from the ligation points, and likely represent errors in the sequencing procedure.

## REFERENCES

- S1. N.C. Seeman, *Current Protocols in Nucleic Acid Chemistry*, Unit 12.1, John Wiley & Sons, New York (2002).
- S2. N.C. Seeman, *J. Biomol. Struct. & Dyns.* **8**, 573 (1990).

## LEGEND TO FIGURES S1-S3

The components used in this work are shown and the strand numbering is indicated. The coloring is used to aid the eye in distinguishing all strands, and has no further significance. Strands used twice in the same physical unit are labeled with an asterisk.

## LEGEND TO FIGS. S4-S7.

These figures show the sequencing traces of the complements to the products in device states PX-PX, PX-JX<sub>2</sub>, JX<sub>2</sub>-PX and JX<sub>2</sub>-JX<sub>2</sub>, respectively.

## The Sequences of the DNA Strands Used Here (See Figs. S1-S3 for Numbering).

Strand 1:

5'.GATCGGTGCTCATGTGTATGGACGCTACGGGACCGCAGTACGGCACGTTGCTAT  
CGCTCAATGC.3'

Strand 2(2\*):

5'.GTACTAGCAGTTTTCGCGGTACACCGTACAGCATTGGTGCATGGCACCG  
TGCACATGGTCGTCG.3'

Strand 3(3\*):

5'.CGTCGTATCTGACGCATGGGACGATGATGGACCTACGATCCTAGATTGGACTG  
TTGACCTGTGACCGCGCTGCTAGTACGTCTGTCATC.3'

Strand 4(4\*):

5'.GAGATGCCGAGGCCGTACCCATGCGTCTTTAGATAACGACGGATGACAG  
AC.3'

Strand 5(5\*):

5'.AACGTGGTCAACAGTGGTGC.3'

Strand 6(6\*):

5'.ATCGTGGCATTACCACCATC.3'

Strand 7:

5'.GTCCAGACGGTAGTCTGCTTCAGCCTGGTAATGCCTGACGGCGCTGGCGATCT  
CCGTT.3'

Strand 8(8\*):

5'.GTGCCGCGCATTTTAGTTGGCTACCAATCTACGTTTGATCGTAGGTGGC  
TGAAGCAGACTACCGTCTGGAC.3'

Strand 9:

5'.CTACACCCGTAGCGTGGCGT.3'

Strand 10(10\*):

5'.GACAATGACGGGCAGTATCCTGTAGACGCCTGCCATGCACTGCTGTACGGACGT  
TGCACCTGAGCCAATTGCGCGGCACGGATGTGCGC.3'

Strand 11:

5'.GATCCACGTGCAGCTATCGGGTGGATACTGCCTTTCGTCATTGTCGCGCACA  
TCC.3'

Strand 12:

5'.GTCATAGTCAGTGTACGAACGACTGCGTACACGGACAGCGCAGGACTGTCTA  
GCTCCAGTGGGATC.3'

Strand 13:

5'.CTACACCTGCGCTGTGGCGT.3'

Strand 14:

5'.TAGCACACTGGCGTACGACAGTGGATACTGCCTTTCGTCATTGTCGCGCACA  
TCC.3'

Strand 15:

5'.GACCAGACGGTAGTCTGCTTCAGCCTGGTAATGCCTGACGGCGCTGGCGATCT  
CTAGGC.3'

Strand 16:

5'.GTACTAGCAGTTTCGCGGTACACCGTACAGCATTGGCATGGCACCCG  
ACTATGTCGATC.3'

Strand 17:

5'.GTAACATGATTAGCGGATCGACATAGTCGGACAGCGCAGGACACGCTGGCTACC  
ATACATCAGTCAGTGTTCG.3'

Strand 18:

5'.GTATGGTAGGCCAGCGTGTGGATACTGCCTTTTCGTCATTGTCGCGCACATCC.3'  
,

Strand 19:

5'.GAGTCGACGGTAGTCTGCTTCAGCCTGGTAATGCCTGACGGCGCTGGCGATCT  
CCGACC.3'

Strand 20:

5'.GATCGGTGCTCATGTGTATGGACGCTACGGGACCGCAGTACGGCACGTTGCTAT  
CGCTTATGTCTCGAATC.3'

Strand 21:

5'.GACGGTAGTCTGCTTCAGCCTGGTAATGCCTGACGGCGCTGGCGATCTCGAA  
G.3'

Strand 22:

5'.GACACGCATCAGTGTACGAACGACTGCGTACACGGACAGCGCAGGACTGTCTA  
GCTCCAGTGGGATC.3'

Strand 23:

5'.GACGGTAGTCTGCTTCAGCCTGGTAATGCCTGACGGCGCTGGCGATCTGAAT  
G.3'

Strand 24:

5'.AGCGCATTACTGCCAACGTTAGTCTCGATAACCTGTAAGTATCGCGTGGTCGGTA  
CTTTGTACCTACACCGCGATCCAGA.3'

Strand 25:

5'.GTCGTGCAGTATGGTATTTTACCACTCAGTCAGGACGACCGTGCACGCCCTCACAT  
CTGGCAACGAGTGTACG.3'

Strand 26:

5'.CGTGGCCTGACATGCGCTCGAACACTGACTGAT.3'

Strand 27:

5'.ATCACTATGACGATTGGACCATCGTTGACTTA.3'

Strand 28:

5'.CAGGTTGGCGTAGACTAATCAGCAC.3'

Strand 29:

5'.GCTAACTGTGTGAGATCGACGCACG.3'

Strand 30:

5'.CAGGTTATCGAAGACTAGCATGAGC.3'

Strand 31:

5'.AGCCGTACTGTGAGGGCGTCGCACG.3'

Strand 32:

5'.TATTACTGCCAACGGACGGCTCGATTACCTGTAAGTGTGCGTGGTCGGTACTTT  
TGTACCTACACCGCGACTTCGA.3'

Strand 33:

5'.GTCGTGCAGTATGGTATTTACCACACAGAACGACCGTGCCTGCCACTACATC  
GAACAACGAGTGTACG.3'

Strand 34:

5'.CGTGGTCTGTGATAACTGCATTGAGCGA.3'

Strand 35:

5'.ATCTGCGTGTCAAGTCGGACCATCGTTGACTTA.3'

Strand 36:

5'.CAGGTAGGCAGGCCGTCGCCAGACT.3'

Strand 37:

5'.TCGTAGCTTGTAGTATCGACGCACG.3'

Strand 38:

5'.CAGGTAATCGAGCCGTCGGTACTGG.3'

Strand 39:

5'.GGCCAAGTTGTAGTGGCAGCGCACG.3'

Unset Strand for Strand 28:

Biotin.5'.GTGCTGATTAGTCTACGCCAACCTG.3'

Unset Strand for Strand 29:

Biotin.5'.CGTGCCTCGATCTCACACAGTTAGC.3'

Unset Strand for Strand 30:

Biotin.5'.GCTCATGCTAGTCTTCGATAACCTG.3'

Unset Strand for Strand 31:

Biotin.5'.CGTGCGACGCCCTCACAGTACGGCT.3'

Unset Strand for Strand 36:

Biotin.5'.AGTCTGGCGACGGCCTGCCTACCTG.3'

Unset Strand for Strand 37:

Biotin.5'.CGTGCCTCGATACTACAAGCTACGA.3'

Unset Strand for Strand 38:

Biotin.5'.CCAGTACCGACGGCTCGATTACCTG.3'

Unset Strand for Strand 39:

Biotin.5'.CGTGCCTGCCACTACAACTTGGCC.3'

Strand 40:

5'.GTAGTCGATGTACCACCATGCAGTCTTTGACTGCATGGACAGCGCAGGACGAT  
CAGCCAAGCTACGGTCG.3'

Strand 41:

5'.CTACACCTGCGCTGTGGCGT.3'

Strand 42:

5'.GTAGCTTGGCTGATCGTGGCCTATA CGCACGTACTGACGGCTATCACGCGGTA  
TGCCATCATG.3'

Strand 43:

5'.PO<sub>4</sub>.CATGATGGCATACCGCGTGATGCCGTAGCGTCAGTACGTGCGTATAGGCCCTGTA  
GACGCCTGGTACATCGACTAC.3'

Strand 44:

5'.GCACGAACAGTAGATGCGCTAACGAGATTGCACACATAGTTGCGTCACCGATCA  
TCCAGTCGTC.3'

Strand 45:

5'.GACTCGACGACTGGATGATCGGACAGCCGTCTAGCTGGCCGCTTGTGCGTTAC  
CGTATGCAGGACGTACATGCACCACTGCCTA.3'

Strand 46:

5'.AGCGGCCAGCTAGACGGCTGTGGCGTAGCCATGCTATCACGCTGATGGTCGGC  
ATTGACTACACCTGCATACGGTAACGCGA  
CA.3'

Strand 47:

5'.AGTGGTGCATGTACGTGGCACTCATTACTGGCAAGGTACTAGGTCCATTG  
CTCAGTTATC.3'

Strand 48:

5'.PO<sub>4</sub>.GATAACTGAGCGAATGGACCTAGTACCTGCCAAGTAATGAGTGCCTGTAG  
TCAATGCCGACCACATCAGCGTATGCGATGGCTACGCCGACGCAACTATGTGTGCA  
ATCTGCTTAGCGCATCTACTGTTCGTGC.3'

Strand 49:5'.

GCACGAACAGTAGATGCGCGGAGCAGTGATGGTCGGCATTGACTACACCGATCAT  
CCAGTCGTC.3'

Strand 50:

5'.GACTCGACGACTGGATGATCGGACAGCCGCTAGCTGGCCTATTGTCGCGTTAC  
CGTATGCAGGACGTACATGCCGGATTAAGCG.3'

Strand 51:

5'.ATAGGCCAGCTAGACGGCTGTGGGCACTCATTACTGGCAAGATTGCACACATA  
GTTGCGTCACCTGCATACGGTAACGCGACA.3'

Strand 52:

5'.AATCCGGCGATGTACGTGGCGTAGCCATGCTATCGTTGGTACTAGGTCCATTG  
CTCAGTTATC.3'

Strand 53:

5'.PO<sub>4</sub>.GATAACTGAGCGAATGGACCTAGTACCAACGATAGCATGGCTACGCCGTGAC  
GCAACTATGTGTGCAATCTGCCAAGTAATGAGTGCCTGTAGTCAATGCCGACCA  
TCACTGCTCCGCGATCTACTGTTCGTGC.3'

Strand 54:

5'.GCACGAACAGTAGATGCGCGGAGCAGTGATAAGCGGCAGACACTACACCGATCA  
TCCAGTCAAC.3'

Strand 55:

5'.PO<sub>4</sub>.TGTTAGCGCCTGACGCAACTATGTGTGCAATCTGCCAAGTAATTGGTGC  
CTGAGTGTCTGCCGCTTACTGCTCCGCGATCTACTGTTCGTGC.3'

Strand 56:

5'.TGGTCGTTGACTGGATGATCGGACAGCCGCTAGCTGGCCTATTGTCGCGTTAC  
CGTATGCAGGACGTACATGCCGGATTCATTG.3'

Strand 57:

5'.ATAGGCCAGCTAGACGGCTGTGGGACCAATTACTGGCAAGATTGCACACATA  
GTTGCGTCACCTGCATACGGTAACGCGACA.3'

Strand 58:

5'.AATCCGGCGATGTACGTGGCGCTAACCA.3'

Strand 59:

5'.GCACGAACAGTAGATGCGCTAACGAGATTGCACACATAGTTGCGTCACCGATCA  
TCCGTTGAAC.3'

Strand 60:

5'.PO<sub>4</sub>.ATGAGTGCCCTGTAGTCATGCCGAGCCTCAGCGTGATAGCATGGCTACGC  
CTGACGCAACTATGTGTGCAATCTGCTTAGCGCATCTACTGTTCGTGC.3'

Strand 61:

5'.TGGTCGTTAACGGATGATCGGACAGCCGCTAGCTGGCCGCTTGTGCGCGTTAC  
CGTATGCAGGACGTACATGCACCAGACTCG.3'

Strand 62:

5'.AGCGGCCAGCTAGACGGCTGTGGCGTAGCCATGCTATCACGCTGAGGCTCGGC  
ATTGACTACACCTGCATACGGTAACGCGACA.3'

Strand 63:

5'.TCTGGTGCATGTACGTGGGCACTCAT.3'

Strand 64:

5'.GCACACATGAGCACGATATTAGCGCAGCGTGAGTCATAGTTGCGTCACCGATCA  
TCAGCATCCA.3'

Strand 65:

5'.PO<sub>4</sub>.GAATCGACCCTGTAGTCATCTACTGCTATACTTGACCGCATGGCTACGCC  
TGACGCAACTATGACTCACGCTGCGCTAATATCGTGCATGTGTGC.3'

Strand 66:

5'.TGGACTGGATGCTGATCGGACAGCCGCTAGCTGGCCGCTTGTGCGCGTTAC  
CGTATGCAGGACGTACATGCACCAGACATTG.3'

Strand 67:

5'.AGCGGCCAGCTAGACGGCTGTGGCGTAGCCATGCGGTCAAGTATAAGCAGTAGAT  
TGCACATACACCTGCATACGGTAACGCGACA.3'

Strand 68:

5'.TCTGGTGCATGTACGTGGGCGATTG.3'

Strand 69:

5'.GCACAATGCGGGTGGATAATGTGAGTGATATGACCATAGTTGCGTCACCGATCAT  
CCGGCATCA.3'

Strand 70:

5'.PO<sub>4</sub>.GAGCGTGCCTGTAGTACCACTGCCAACAGAGGTCGATAGCATGGCTACGC  
CTGACGCAACTATGGTCATATCACTCACATTATCCACCCGATTGTGC.3'

Strand 71:

5'.TGGACTGATGCCGGATGATCGGACAGCCGTCTAGCTGGCCGCTTGTGCGCTTAC  
CGTATGCAGGACGTACATCGCACCAGACTTCG.3'

Strand 72:

5'.AGCGGCCAGCTAGACGGCTGTGGCGTAGCCATGCTATCGACCTCTGTTGGCAGT  
GGTACTACACCTGCATACGTAACGCGACA.3'

Strand 73:

5'.TCTGGTGCATGTACGTGGGCACGCTC.3'

Strand 74:

5'.CGACTCACGTACTGCCTACGATCACCCGGGATCGCACCCTTT.Biotin.TTT.Bio  
tin.TGACGGTGCATCCC GG GTGATCGTAGT.3'

Strand 75:

5'.CGCTAATCATGTTACCAGCTATCTACATCGACCGCTCAGCCTGTGATGCTGTC  
AGTCACCATACGCTGCTAGTCCACTGTAC.3'

Strand 76:

5'.GCAGTACGTGAGTCGGTACAGTGGACTAGCAGCGTATGGTGACAGCATCACA  
CAGGCTGAGCGGTGATGTAGATAGCTG.3'

Strand 77:

5'.ACGTTGGATTGGGCCCTGATCGTAGTGTGTT.Biotin.TTT.Biotin.TCACTACGATCAG  
GGCCAATCCAACGTCGCTAATCATGTTAC.3'

Strand 78:

5'.GTACAGTGGACTAGCAGCGTATGGTGAUTGACAGCATCACACAGGCTGAGCGGT  
CGATGTAGATAGCTGGTAACATGATTAGCG.3'

Strand 79:

5'.CAGCTATCTACATCGACCGCTCAGCCTGTGATGCTGTCAGTCACCATAACGCTG  
CTAGTCCACTGTACGATTGAGACATAAG.3'

Experimental Sequences of Ligation Products:

PX1-PX2:

5'.TGTTAGGCCTGACGCAACTATGTGTGCAATCTGCCAAGTAATTGGTGCCTGT  
AGTGTCTGCCGCTTATCACTGCTCCGCGCATCTACTGTTCGTGCATAACTGAGCG  
AATGGACCTAGTACCTTGCCAAGTAATGAGTGCCTGTAGTCAATGCCGACCATCA  
GCGTGATAGCATGGCTACGCCGACGCAACTATGTGTGCAATCTGCTTAGCGCATT  
TACTGTTCGTGCATGATGGCATACCGCGTGATAGCCGTCAGTACGTGCGTATAGG  
CCCTGTAGACGCCTGGTACATCGACTAC.3'

PX1-JX<sub>2</sub>:

5'.ATGAGTGCCTGTAGTCATGCCGAGCCTCAGCGTGATAGCATGGCTACGCCCTG  
ACGCAACTATGTGTGCAATCTGCTTAGCGCATTACTGTTCGTGCATAACTGAGC  
GAATGGACCTAGTACCTTGCCAAGTAATGAGTGCCTGTAGTCAATGCCGACCATC  
AGCGTGATAGCATGGCTACGCCGACGCAACTATGTGTGCAATCTGCTTAGCGCATT  
TACTGTTCGTGCATGATGGCATACCGCGTGATAGCCGTCAGTACGTGCGTATAGG  
GCCCTGTAGACGCCTGGTACATCGACTAC.3'

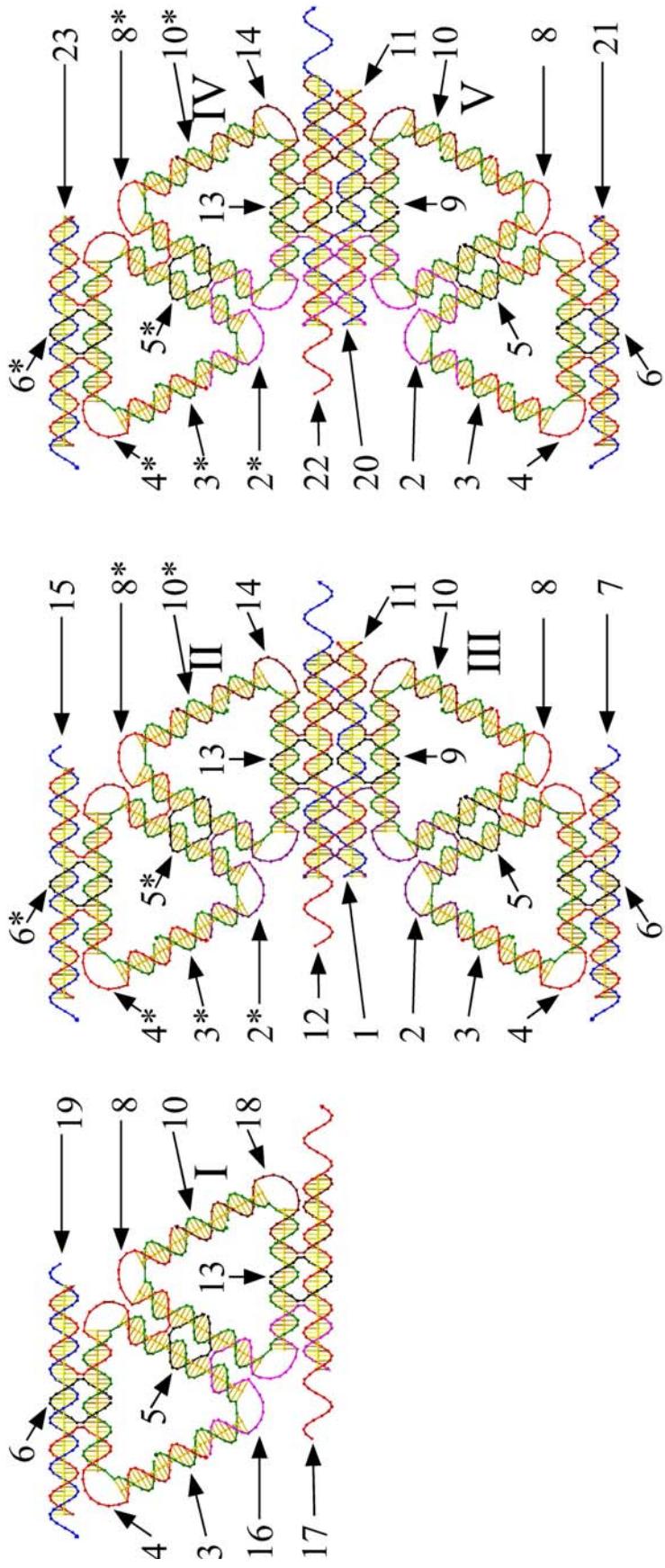
JX<sub>2</sub>1-PX2:

5'.GAGCGTGCCCTGTAGTACCACTGCCAACAGAGGTCGATAGCATGGCTACGCCCTG  
ACGCAACTATGGTCATATCACTCACATTATCCACCCGCATTGTGCATACTGAGC  
GAATGGACCTAGTACCAACGATAGCATGGCTACGCCGACGCAACTATGTGTGCAA  
TCTTGCCAAGTAATGAGTGCCTGTAGTCATGCCGACCATCACTGCTCCGCGCAT  
TACTGTTCGTGCATGATGGCATACCGCGTGATAGCCGTCAGTACGTGCGTATAGG  
CCCTGTAGACGCCTGGTACATCGACTAC.3'

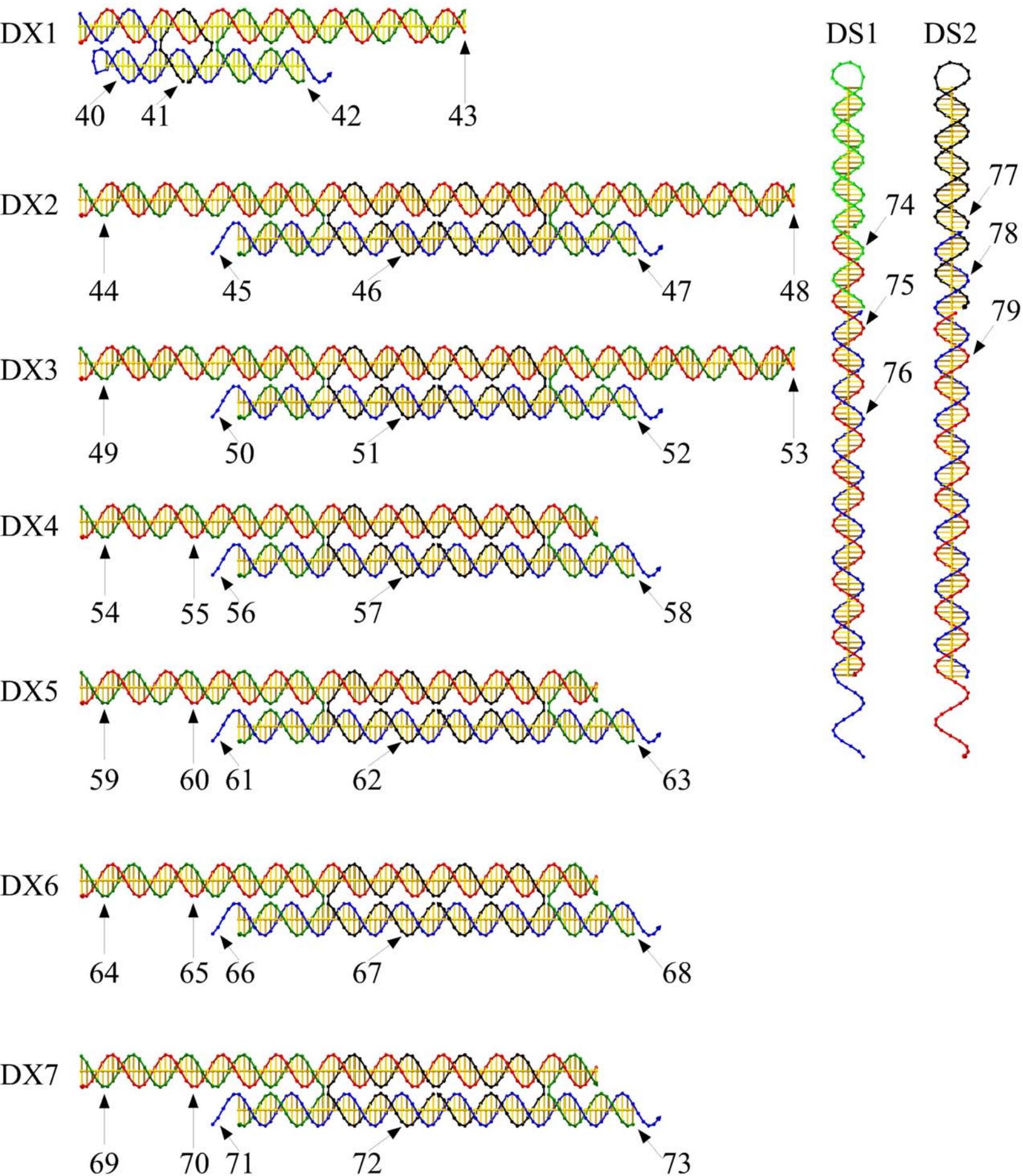
JX<sub>2</sub>1-JX<sub>2</sub>:

5'.GAATCGACCCCTGTAGTGCAATCTACTGCTATACTTGACCGCATGGCTACGCCCTGA  
CGCAACTATGACTCACGCTGCCATAATATCGTGCATGTGTGCATAACTGAGCG  
AATGGACCTAGTACCAACGATAGCATGGCTACGCCGACGCAACTATGTGTGCAA  
CTTGCCAAGTAATGAGTGCCTGTAGTCATGCCGACCATCACTGCTCCGCGCATC  
TACTGTTCGTGCATGATGGCATACCGCGTGATAGCCGTCAGTACGTGCGTATAGG  
CCCTGTAGACGCCTGGTACATCGACTAC.3'

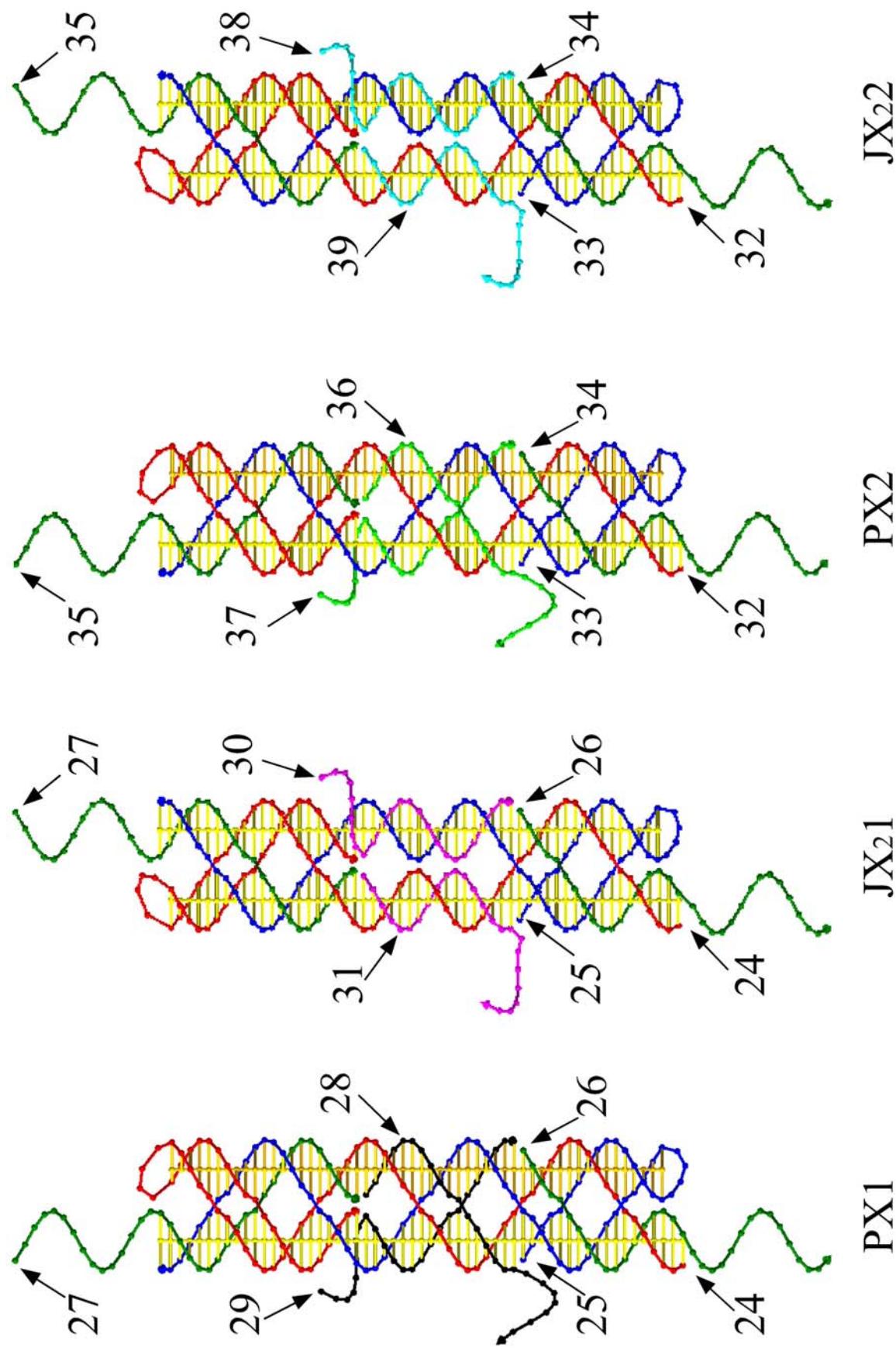
# Strand Numbering for Diamond Components

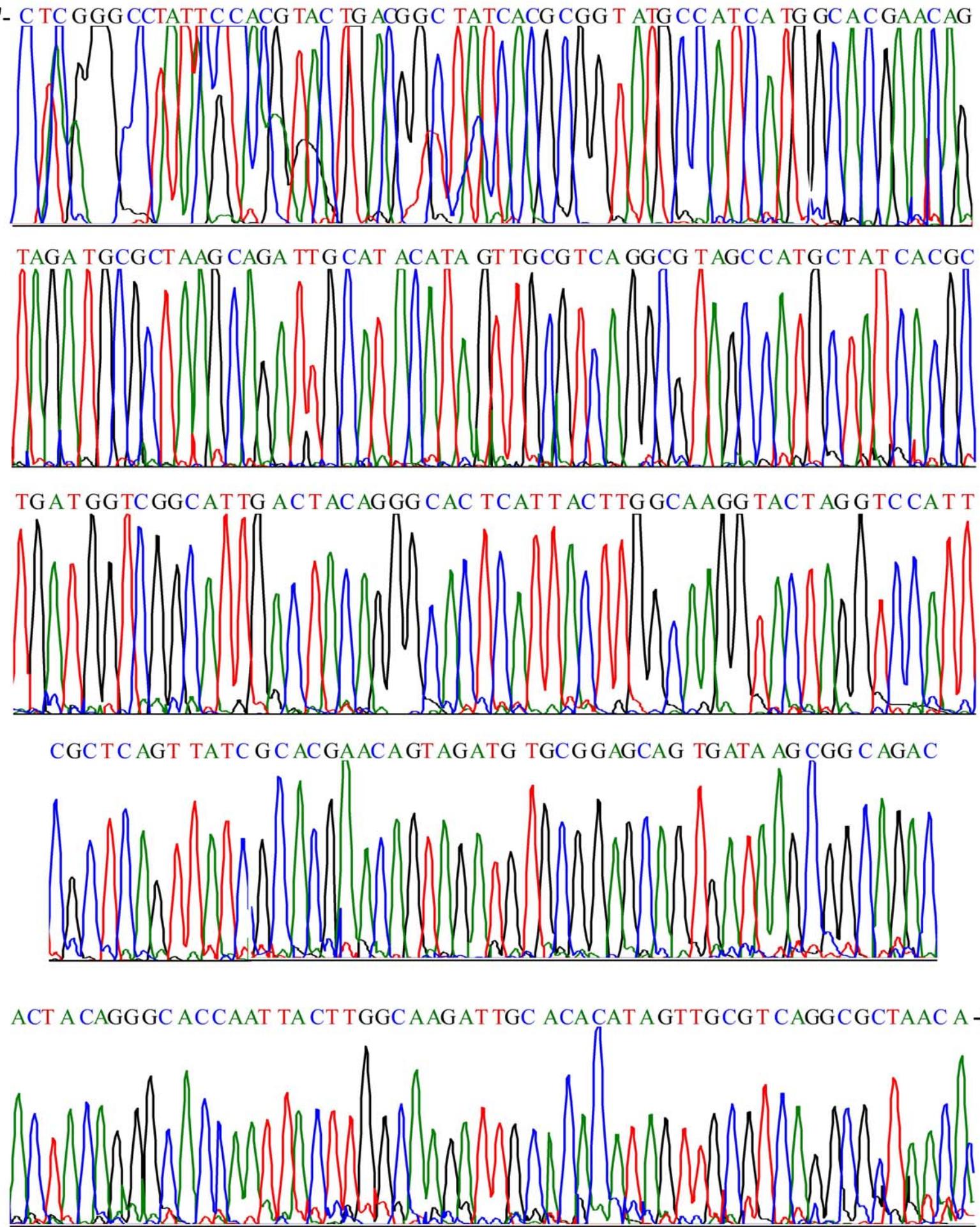


# Strand Numbering for DX and DS Components

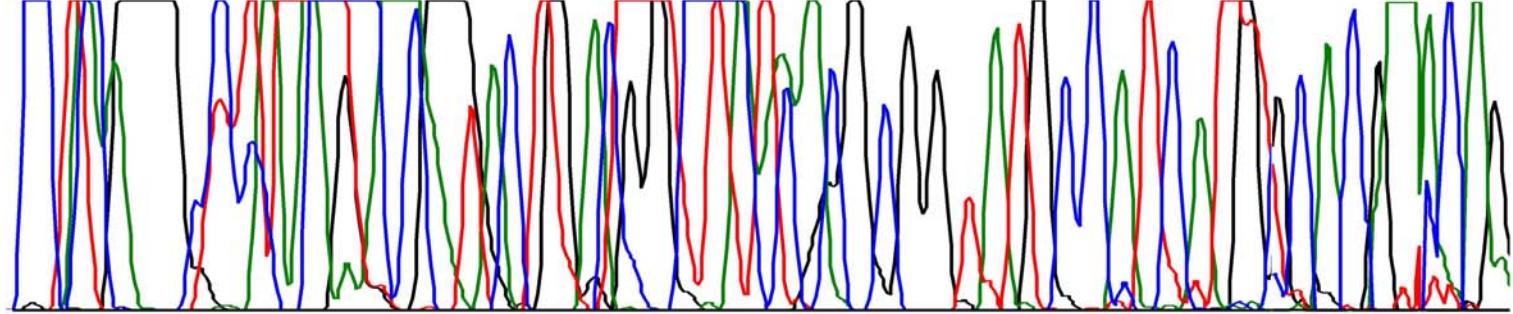


# Strand Numbering for Devices (in Both States)

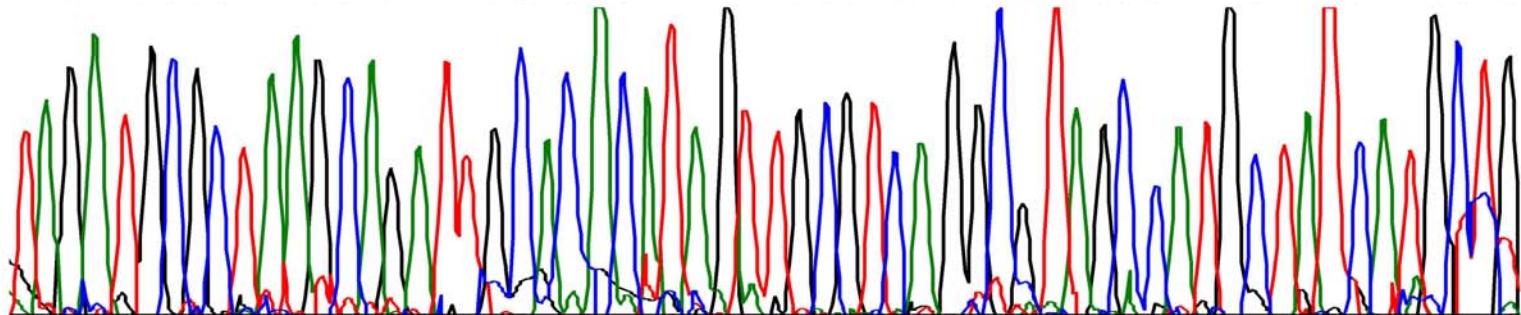




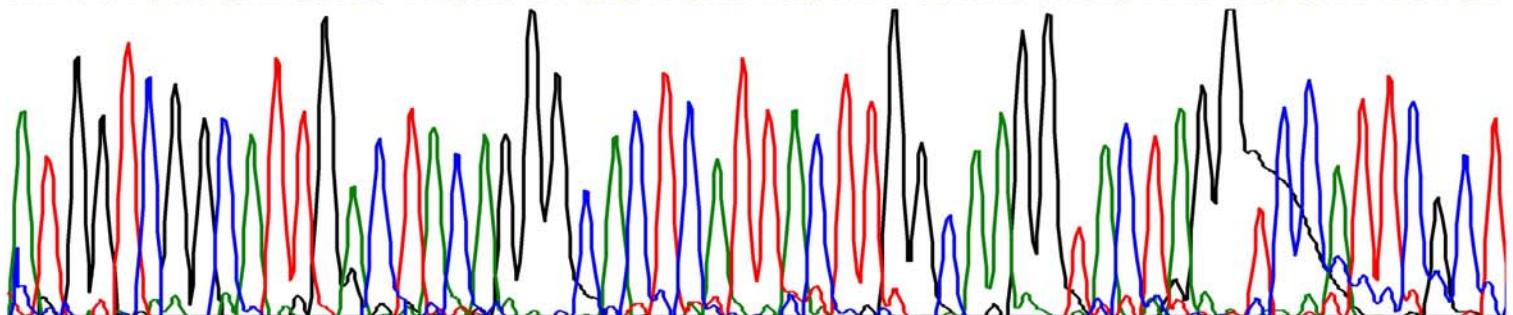
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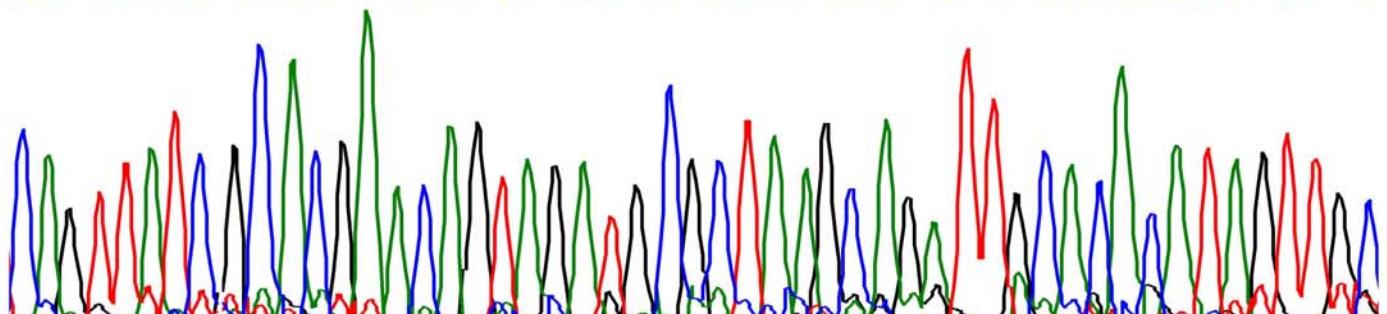
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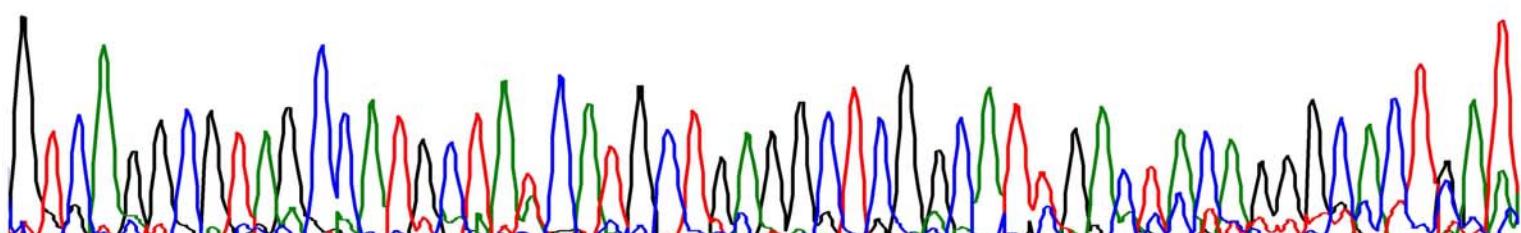
A T GG TCGGC ATTGAC T ACAGGGCAC TC AT TAC TTGGC AAGGT ACT AGG TCC CAT TCGCT



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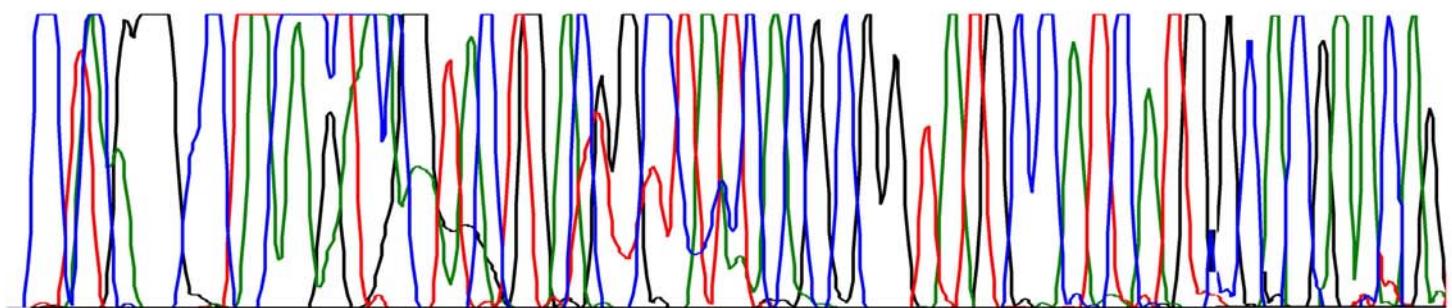


GTCAGGGCG TAG CCATGCTATC ATGCTGAGG CTCGGCA TT GACTACAGGGCA CTGA T -3'

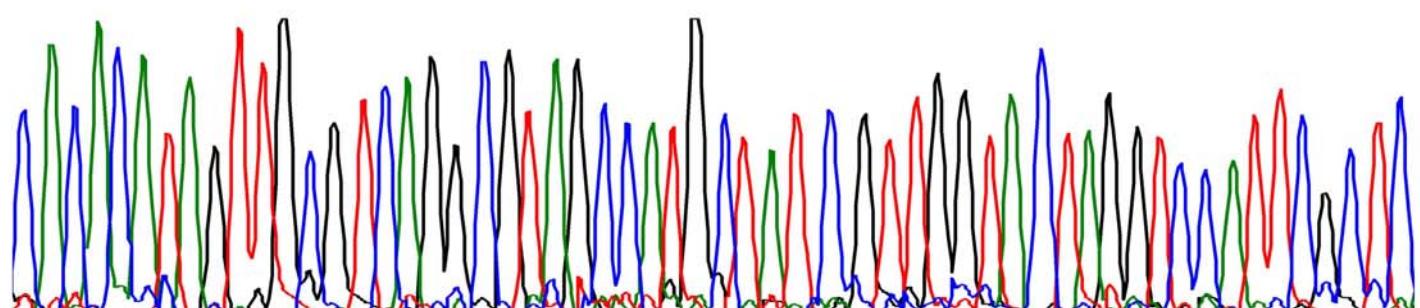


Sequencing result for the complementary strand of the PX-JX2 ligation product

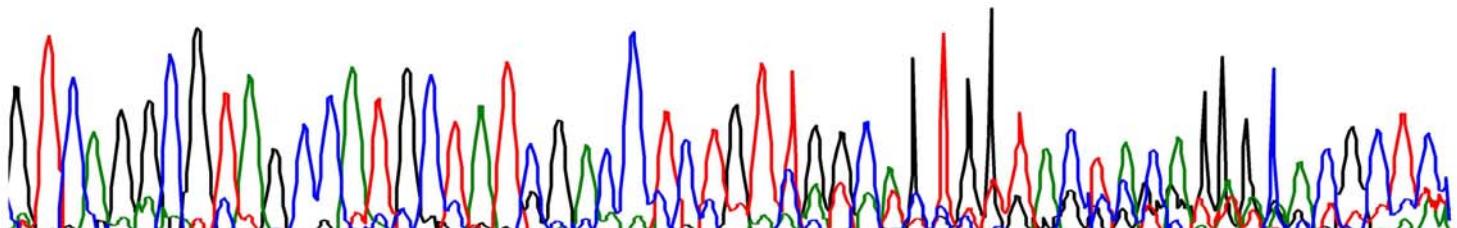
5'- C T N GGG CCT NT C T C A C G T A C T G A C G G C T A T C A C G C G G T A T G C C A T C A T G G C A C G A A C A G



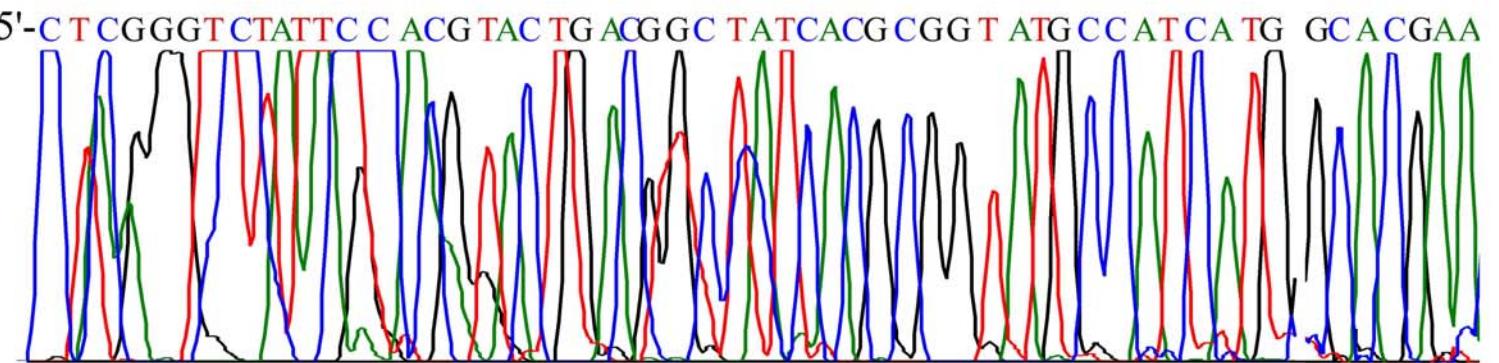
C A C A C A T A G T T G C G T C A G G C G T A G C C A T G C T A T C G T T G G T A C T A G G T C C A T T C G C T C



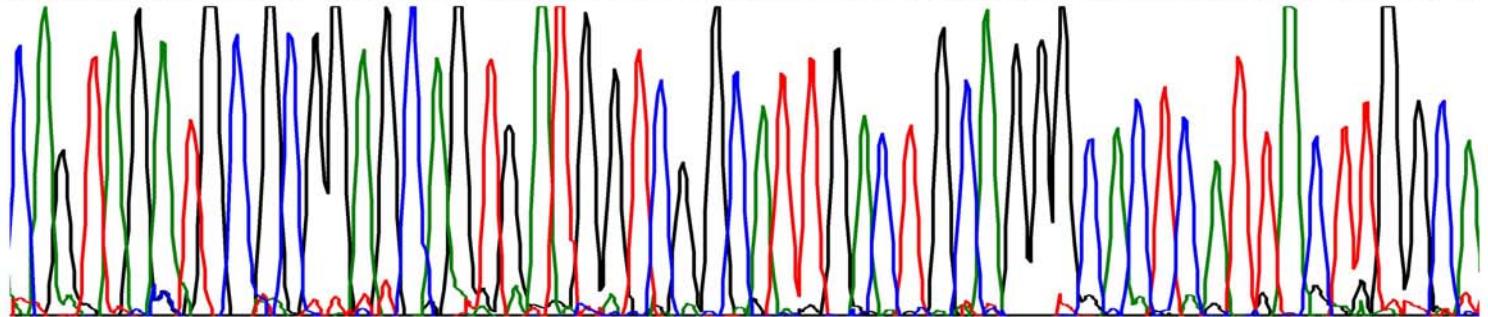
G T C A G G C G T A G C C A T G C T A T C G A C C T C T G T T G G C A G T G G T A C T A C A G G G C A C G C T C -3'



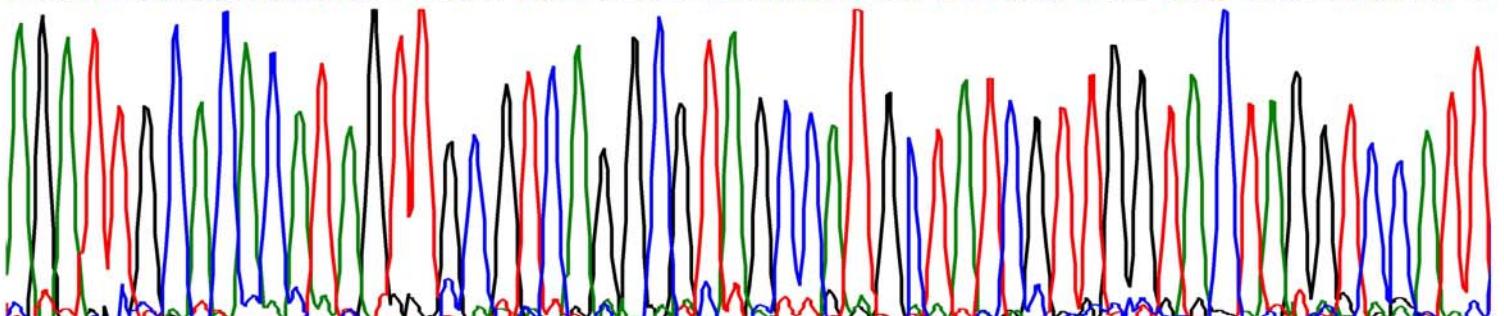
Sequencing result for the complementary strand of the JX2-PX ligation product



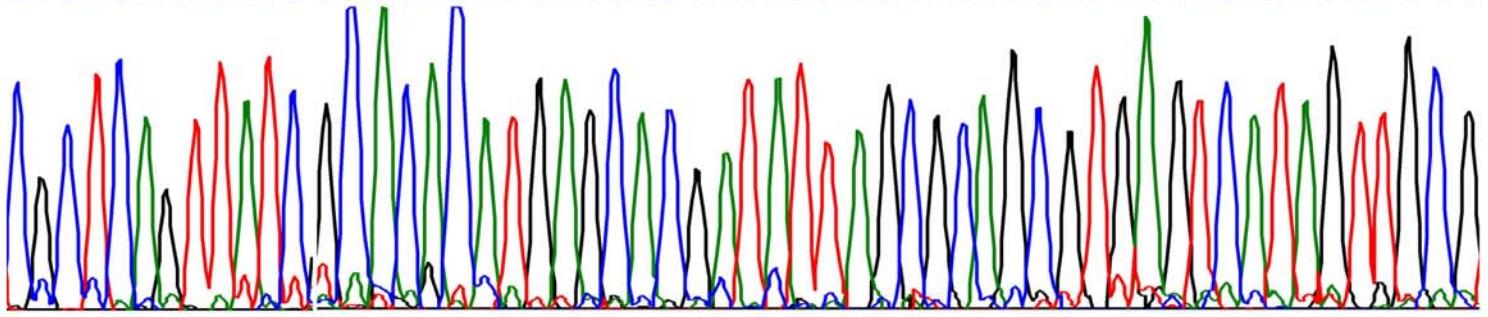
CAGTAGATG C GCGGAGCAG TGATGGTCGGCATTGACT GCAGGGCAC TCATTACTTGGCA



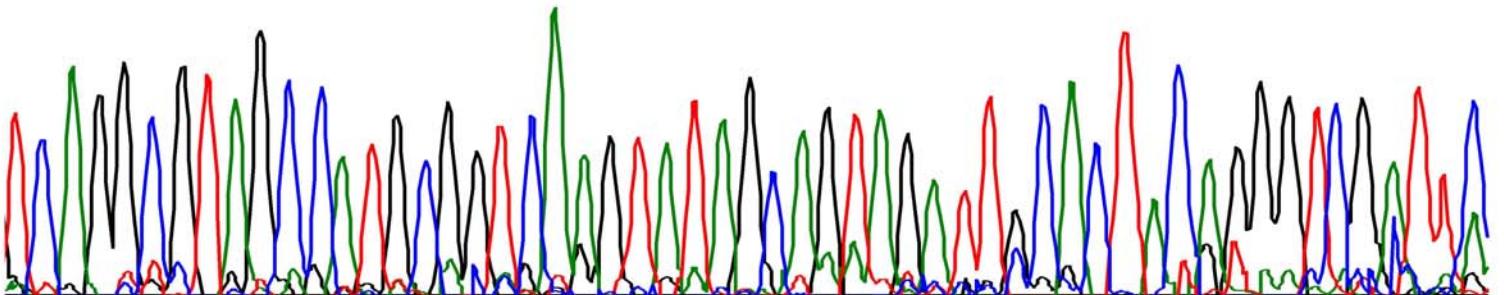
AGATTGCACACA TAGTTGCG TCAGGCGTAGCCATGCTATCGTTGGTAC TAGGTCCATT



CGCTCAGTTATCGCACACATGAGCACGATATTA GCGCAGCGTGA GTCATAGTTGCG



TCAGG CGTAGC CATGCGGTCAAGTATAGCAGTAGATTGCACTACAGGGTCGATTTC-3'



Sequencing result for the complementary strand of the JX2-JX2 ligation product.