

Single-Stranded Templates as Railroad Tracks for Hierarchical Assembly of DNA Origami

Supporting Information

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I. Materials and Instrumentation:

A. Materials

Acetic acid, boric acid, EDTA, urea, magnesium chloride, GelRed, tris(hydroxymethyl)aminomethane (Tris), D(+) glucose, 2-β-mercaptoethanol, were purchased from Aldrich. Nucleoside (1000 Å)-derivatized LCAACPG solid support with loading densities of 25-40 μmol/g, Sephadex G-25 (super fine DNA grade), and reagents for automated DNA synthesis were used as purchased from BioAutomation. Acrylamide (40%)/bis-acrylamide 19:1 solution and agarose were purchased from BioShop. All staple strands used for the assembly of origami were purchased from Bioneer. The scaffold M13mp18 single-stranded was purchased from New England Biolabs. AFM cantilevers were purchased from Asylum Research (model AC160TS) and RubyRed mica were ordered from Electron Microscopy Sciences. TBE buffer is composed of 90 mM Tris and boric acid and 1.1 mM EDTA, with a pH of ~8.3. TAMg buffer is composed of 45 mM Tris and 12.5 mM MgCl₂ with a pH of ~7.8 adjusted by glacial acetic acid. 1xTAE is composed of 45 mM Tris and 1 mM EDTA, with pH adjusted to 8.0 using glacial acetic acid. 1xOK buffer is composed of 50 mM Tris-HCl, 10 mM MgCl₂, 5 mM dithiothreitol (DTT), with a pH of 7.5. 1xQL buffer is composed of 66 mM Tris-HCl, 10 mM MgCl₂, 1 mM DTT, 1 mM ATP, 7.5% w/v PEG6000, with a pH of 7.6, and was made in-house as a 2x concentrate. 1xALK is composed of 30 mM NaOH and 1 mM EDTA. 1xSDA buffer is composed of 40 mM Tris, 10 mM MgCl₂, 50 mM NaCl, 5 mM DTT, 100 μg/mL bovine serum albumin (BSA), and 500 μM of each dNTP. 0.5xSSC buffer is composed of 75 mM NaCl and 7.5 mM sodium citrate, with a pH of 7.0.

Kits for Optikinase and Quick, T4 and T7 DNA ligase were purchased from New England Biolabs.

A MyTaq™ HS Red PCR kit was purchased from CedarLane Laboratories. QIAquick Gel

Extraction and PCR purification kits from Qiagen were used for extraction or cleanup of PCR products. FastDigest® XbaI and EcoRI were purchased from Life Technologies. rSAP (Shrimp Alkaline Phosphatase) was purchased from New England Biolabs. Streptavidin Magnesphere® Paramagnetic Particles were purchased from Promega.

DH5α (Subcloning Efficiency Competent Cells) were purchased from Life Technologies. LB media is composed of 2.5 g BioTryptone, 2.5 g NaCl and 1.25 g Yeast Extract. PureLink Quick Plasmid DNA Mini/Maxi Prep Kits from Qiagen were used to isolate the plasmid.

B. Instrumentation

Standard automated oligonucleotide solid-phase synthesis was carried on a BioAutomation MerMade MM6 DNA synthesizer. UV-Vis quantifications were performed with a NanoDrop Lite Spectrophotometer. Polyacrylamide gel electrophoresis (PAGE) was carried out on a 20 x 20 cm vertical Hoefer 600 electrophoresis unit. Agarose gel electrophoresis (AGE) was performed on an Owl Mini gel electrophoresis unit. Thermal anneals, polymerase chain reaction (PCR), and enzymatic digestions were conducted using an Eppendorf Mastercycler Pro 96 well thermocycler. AFM was performed with a MultiMode™ MM8 SPM connected to a Nanoscope™ controller, from the Digital Instruments Veeco Metrology Group. The plasmids were sequenced using Sanger methods at McGill University Genome Center and Innovation Quebec.

II. Sequential growth of dsKL[10]

A. DNA synthesis

The sequences of each building block were generated by CANADA version 2.0 (available online at <http://ls11-www.cs.uni-dortmund.de/molcomp/downloads/>), a program that intends to minimize undesired secondary interactions, and idt DNA (Table S1). DNA synthesis was carried on a on a BioAutomation MerMade MM6 DNA synthesizer at 1 μ mole scale. Deprotection and cleavage from the solid support was achieved through the addition of concentrated ammonium hydroxide (55°C, 14 hours). Crude products were then purified via polyacrylamide gel electrophoresis under denaturing conditions (4M urea). Following PAGE, the gel was visualized by UV light over a fluorescent TLC plate. The product was rapidly excised, then crushed and incubated in 11 mL of autoclaved water at 65°C overnight. Size exclusion chromatography (Sephadex G-25) was used to desalt the samples. Finally, strands were quantified via a NanoDrop Lite Spectrophotometer (OD260) and using IDT's extinction coefficient at 260.

B. Temporal growth

Every strand having an internal 5'-terminus was phosphorylated by OptiKinase. The concentration of the strands was adjusted to 10 μ M, with a 1xOptikinase buffer, 2.5 mM ATP, and 0.1 U/ μ L of OptiKinase. The mixture was incubated at 37°C for 30 min, then at 75°C for 10 min to inactivate the enzyme. It is worthy to mention that the efficiency of Optikinase is around 60% according to the manufacturer. KL[10] was synthesized following the Sequential Growth procedure by Hamblin *et. al.*¹ It includes in-situ ligation, isolation of the dsKL[10] by native AGE, PCR enrichment and the separation of the non-nicked dsKL[10] via denaturing AGE. In PCR enrichment, PrimerL2eco and PrimerK1xba were used for dsKL[10]; no additional restriction site was added through PCR.

Table S1. Duplex Seed Sequences for KL[10]

Name	Sequence (5' -> 3')	Length (bp)
K1pxba	AATTAAGATAGGCGCGGCTCTAGAGCGATATAATCTG GCTGCGCTTGAAACAACGGAAGGTCATGCTTTAGGA	73
K2pxba	TGACCTTCCGTTGTTTCAAGCGCAGCCAGATTATATCG CTCTAGAGCCGCGCCTATCTTAATT	63
K1	AATCTGGCTGCGCTTGAAACAACGGAAGGTCATGCTT TAGGA	43
K2	TGACCTTCCGTTGTTTCAAGCGCAGCCAGATTTCTTCT GATC	42
L1	ATCAAACCAAAGTTCAGCAACAGGCCGTTAAGGATCA GAAGA	42
L2	CTTAACGGCCTGTTGCTGAACTTTGGTTTGATTCCTAA AGCA	42
L1peco	ATCAAACCAAAGTTCAGCAACAGGCCGTTAAGGATCA GAAGACGTAGTCCGAATTCACCTGCAA	64
L2peco	TTGCAGGTGAATTCGGACTACGTCTTCTGATCCTTAAC GGCCTGTTGCTGAACTTTGGTTTGATTCCTAAAGCA	74

III. Transformation protocol of pUC19-KL[10]

First, dsKL[10] was amplified by PCR with annealing temperature 1-2 degree lower than that used in Sequential Growth. 1µg of dsKL[10] and 1µg of pUC19 were digested using FD EcoRI and FD XbaI (LifeTechnologies) for 1.5 hr at 37°C and purified using 2.5% and 1.2% (w/v) native agarose gel, respectively. The band from the gel was excised and the product was extracted by ethanol precipitation. Then 40ng of the digested backbone were ligated into 80ng of the digested plasmid using NEB T4 Ligase. We followed the manufacturer's manual for overnight or 10 min ligation.

Finally, we transformed our insert into DH5 α cells. Around 70 ng of ligated product was gently added to 50 μ L aliquot of DH5 α cells previously placed on ice. Then, the cells were heat shocked for 30 s at 42°C after 30 min incubation time. Lysogeny broth (LB) media was later added and the mixture was shaken at 225 rpm for 1 hour at 37°C. The transformation was plated on ampicillin-agar and incubated overnight at 37°C. Few colonies were picked from the ligation plate and each one was used to inoculate 3 mL of LB media containing ampicillin (100 μ g/mL) at 225 rpm and 37°C. PureLink Quick Plasmid DNA Mini Prep Kit was used to isolate the plasmid containing our insert.

IV. Sequencing of pUC19-KL[10]

In order to examine the sequence of our backbone within the plasmid, we submitted our samples for Sanger Sequencing to McGill University Genome Center and Innovation Quebec.

The observed sequence was the following:

NNNNNNNcgatataaatctggctgcgcttgaNNNacggaaggtcatgctttNNNNNNcaaaccaaagttcagcaacaggccg
ttaaggatcagaagaaatctggctgcgcttgaacaacggaaggtcatgctttaggaatcaaaccaaagttcagcaacaggccgtaaggat
cagaagaaatctggctgcgcttgaacaacggaaggtcatgctttaggaatcaaaccaaagttcagcaacaggccgtaaggatcagaag
aaatctggctgcgcttgaacaacggaaggtcatgctttaggaatcaaaccaaagttcagcaacaggccgtaaggatcagaagaatctg
gctgcgcttgaacaacggaaggtcatgctttaggaatcaaaccaaagttcagcaacaggccgtaaggatcagaagacgtagtccgaatt
c

The red color represents the K block and the blue color represents the building block L.

The expected sequence is:

5'-

caggctgactctagagcgatataaatctggctgcgcttgaacaacggaaggtcatgctttaggaatcaaaccaaagttcagcaacaggccgt
taaggatcagaagaaatctggctgcgcttgaacaacggaaggtcatgctttaggaatcaaaccaaagttcagcaacaggccgtaaggat
cagaagaaatctggctgcgcttgaacaacggaaggtcatgctttaggaatcaaaccaaagttcagcaacaggccgtaaggatcagaag
aaatctggctgcgcttgaacaacggaaggtcatgctttaggaatcaaaccaaagttcagcaacaggccgtaaggatcagaagaatctg

gctgcgcttgaaacaacggaaggatcatgcttaggaatcaaaccaaggtcagcaacaggccgtaaggatcagaagacgtagtccgaatt
cactggc-3'

After determining the plasmid with perfect backbone sequence we wanted to improve further the yield of our product. We used 50ng of this plasmid from Miniprep solution to transform 50μL of DH5α, by means of standard protocol. The next day, we picked a single colony using a sterile pipette tip and dropped it into 4.5 mL of LB with ampicillin to inoculate the media for 6 hrs at 225rpm, 37°C. Then we poured the media into 300mL of LB with ampicillin to inoculate further overnight. Finally, we used Maxiprep (Qiagen) to prepare a bulk quantity of the plasmid from the rest of the media. The yield was 850 ng/μL.

V. Magnetic beads separation of ssKL[10]

To isolate dsKL[10] from pUC19-KL[10], two primers that bind the flanking region of ds[10] were designed: Primer pUC19-KL10-For (caggtcgactctagagcgatat) and primer pUC19-KL10-Rev (gccagtgaattcggactacg). Throughout this manuscript, when designing a new primer, we avoided using any sequence included within the repeating pattern of dsKL[10] since it can introduce non-specific binding. We followed the standard strategy for designing primers and synthesized them using DNA synthesizer. Optimal PCR conditions intend to improve the yield of the desired product and minimize the amount of contaminants. This is achieved by: (i) using the gradient function on the thermal cycler to find out the best temperature, (ii) varying the amounts of pUC19-KL[10] and primers. Table S2 summarizes the final quantities we used to extract ds[10] from pUC19-KL[10]. Note that the extension of the primers occurred at 63°C.

Table S2. Optimized PCR conditions to extract dsKL[10] from pUC19-KL[10]

	Volume (μL)
pUC19-KL[10] (20ng)	0.2
pUC19-KL10-For (0.5 μ M)	0.5
pUC19-KL10-Rev (0.5 μ M)	0.5
Autoclaved H ₂ O	18.8
MyTaq 2 \times mix	20

Before starting the conversion of dsKL[10] to its single-stranded analogue ssKL[10], the PCR products were purified via PCR purification kit. Figure S1 lane 1 displays the successful isolation of dsKL[10] from the plasmid. In order to use our scaffolds in building higher-order DNA nanoarchitectures, we employed PCR followed by magnetic beads separation to convert double-stranded backbones to their single-stranded correspondents. To allow the binding of the double-stranded product to streptavidin-coated magnetic beads, we biotinylated the 5' end of the antisense strand (the sense that will not be used in future experiments) by using a biotinylated reverse primer. However, prior to the incubation of the beads with dsKL[10], we pre-treated them with 30 mM NaOH solution for 2 hours. This step is supposed to cleave the weak interaction between biotin/streptavidin on the beads, hence minimizing byproducts. Later, dsKL[10] was added to the beads in 0.5 \times SSC buffer at pH=7 for 2 hours (gently inverted to maximize binding). The single-stranded scaffold was obtained upon denaturing dsKL[10] with a 20 mM solution of NaOH for 10 min. It is worthy to mention that both the concentration of the alkaline solution and the duration of incubation play a key role in determining the yield and the purity of ssKL[10]. Incubation for a shorter time results in a low yield of ssKL[10], whereas keeping the mixture in contact with NaOH for a longer time can break the remaining weak biotin-streptavidin interaction. Similarly, using a concentration higher than 30 mM might have a greater impact on biotin/ streptavidin interaction. The supernatant containing the released ssKL[10] product was finally recovered by ethanol

precipitation. Figure S1 lane 2 shows a discrete band between 200 and 300 bp that corresponds to ssKL[10]. Despite pre-treating the beads with NaOH, the non-penetrating band appearing in lane 2 is attributed to streptavidin proteins attached to 1-4 ssKL[10] or dsKL[10].

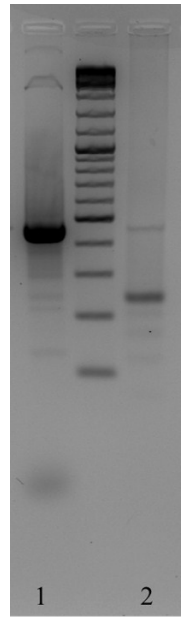


Figure S1. 2.5% AGE in TAE showing ds-KL[10] after PCR amplification (lane 1) and ss-KL[10] after magnetic beads separation.

VI. Synthesis of dsKL[20] and ssKL[20]

A. Synthesis of dsKL[20]

Adapted from the Golden Gate assembly, type II restriction enzyme BsmBI was used to help linking 2 fragments of dsKL[10]. Because dsKL[10] was already inserted into pUC19, we had to design 4 primers (Primer K1p-pUC19-For, PrimerL2p-In-Rev, PrimerK1p-In-For and PrimerL2p-pUC19-Rev) that introduce the corresponding complementary regions into dsKL[10] to synthesize dsKL[20]. As such, each primer was extended by 5 unique bases and a restriction site for the enzyme BsmBI (Figure S2). We made sure that all of the sticky ends are unique and do not overlap

with each other besides than their own compatible ends. To generate the left fragment M, primer K1p-pUC19-For and primer L2p-In-Rev were used (In stands for the unique bases added to this primer allowing M to hybridize N). In a separate reaction mixture, primer K1p-In-For and primer L2p-pUC19-Rev were used to produce the right fragment N.

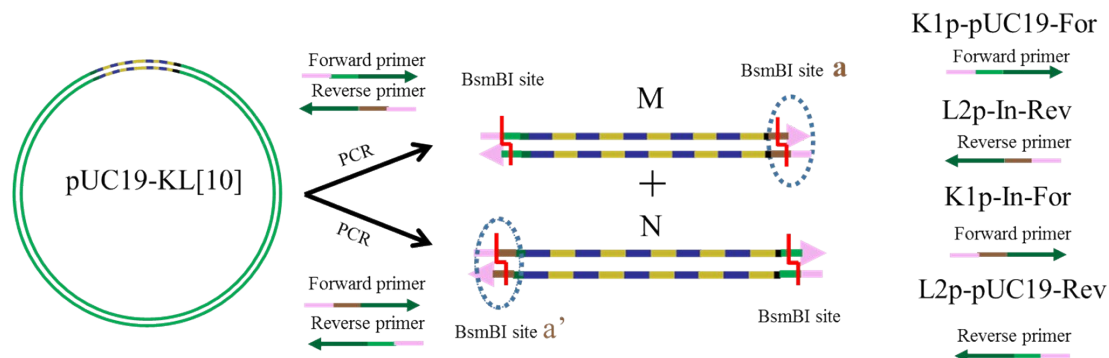


Figure S2. Scheme showing the generation of the two DNA fragments M and N via two PCR reactions.

dsKL[10] left fragment: M

TGCGTCCGTCTCGCGCGCAGGTCGACTCTAGAGCGATATaatctggctgcgcttgaaacaacggaag
gtcatgctttaggaatcaaaccaaagttcagcaacaggccgtaaggatcagaagaaatctggctgcgcttgaaacaacggaaggtcatgct
ttaggaatcaaaccaaagttcagcaacaggccgtaaggatcagaagaaatctggctgcgcttgaaacaacggaaggtcatgctttaggaatcaaacca
aaacaaagttcagcaacaggccgtaaggatcagaagaaatctggctgcgcttgaaacaacggaaggtcatgctttaggaatcaaacca
aagttcagcaacaggccgtaaggatcagaagaaatctggctgcgcttgaaacaacggaaggtcatgctttaggaatcaaaccaaagttca
gcaacaggccgtaaggatcagaagaCGTAGTCCGAATTCACCTGGCGGAGACGGACGCA

PrimeK1p-pUC19-For

TGCGTCCGTCTCGCGCGCAGGTCGACTCTAGAGCGATAT

PrimerL2p-In-Rev

TGCGTCCGTCTCCGCCAGTGAATTCGGACTACG

dsKL[10] right fragment: N

TGCGTCCGTCTCGTGGCCAGGTCGACTCTAGAGCGATATaatctggctgcgcttgaacaacggaaggtcatgcttaggaatcaaaccaaagttcagcaacaggccgtaaggatcagaagaaatctggctgcgcttgaacaacggaaggtcatgcttaggaatcaaaccaaagttcagcaacaggccgtaaggatcagaagaaatctggctgcgcttgaacaacggaaggtcatgcttaggaatcaaaccaaagttcagcaacaggccgtaaggatcagaagaaatctggctgcgcttgaacaacggaaggtcatgcttaggaatcaaaccaaagttcagcaacaggccgtaaggatcagaagaCGTAGTCCGAATTCAGTGGCTCCCGGAGACGGACGCA

PrimerK1p-In-For

TGCGTCCGTCTCGTGGCCAGGTCGACTCTAGAGCGATAT

PrimerL2p-pUC19-Rev

TGCGTCCGTCTCCGGGAGCCAGTGAATTCGGACTACG

The PCR conditions are listed in Table S3. The reaction were carried on at 63°C annealing temperature.

Table S3. PCR conditions to generate M and N fragments

Left-500mer-Insert (or dsKL[10]-M)	Volume (μL)	Right-500mer-Insert (or dsKL[10]-N)	Volume (μL)
pUC19-KL[10] (20 ng)	0.461	pUC19-KL[10] (20 ng)	0.461
Primer ApF (0.625 μM)	1.100	Primer AiF (0.625 μM)	1.362
Primer BiR (0.625 μM)	1.623	Primer BpR (0.625 μM)	0.767
Autoclave H ₂ O	16.82	Autoclave H ₂ O	17.41
MyTaq 2×mix	20	MyTaq 2×mix	20

* Primer KpF = PrimerK1p-pUC19-For; Primer L= PrimerL2p-In-Rev...so on

Following PCR purification of M and N, each backbone was digested with BsmBI for 16 hours at 37°C. The enzyme was then heated and inactivated for 20 min at 65°C. PCR purification kit was used to purify the two fragments and pUC19 prior to ligation. Table S4 summarizes the conditions

employed for digestion. Later, digested pUC19 plasmid was mixed with M and N scaffolds in the presence of T7 ligase for 2 hours at room temperature. Table S5 summarizes the ligation conditions.

Table S4. Digestion conditions of pUC19 and M and N fragments with BsmBI

Volume (μL)	pUC19 (1μg)	M (500ng)	N (500ng)
DNA (Insert OR Plasmid)	14.12	3.75	4.06
10X Tango Buffer	2	2	2
DTT (10mM)	2	2	2
dH ₂ O	0.88	11.25	10.94
BsmBI	1	1	1

Table S5. Ligation conditions of pUC19 and M and N fragments with T7 ligase

pUC19 (100ng)	2.84 (35.2 ng/μL)
ds[10]-M Insert (18.5 ng)	0.877 (21.1 ng/μL)
ds[10]-N Insert (18.5 ng)	0.894 (20.7 ng/μL)
2X T7 Ligase Buffer	5
T7 Ligase	0.25
dH ₂ O	0.139

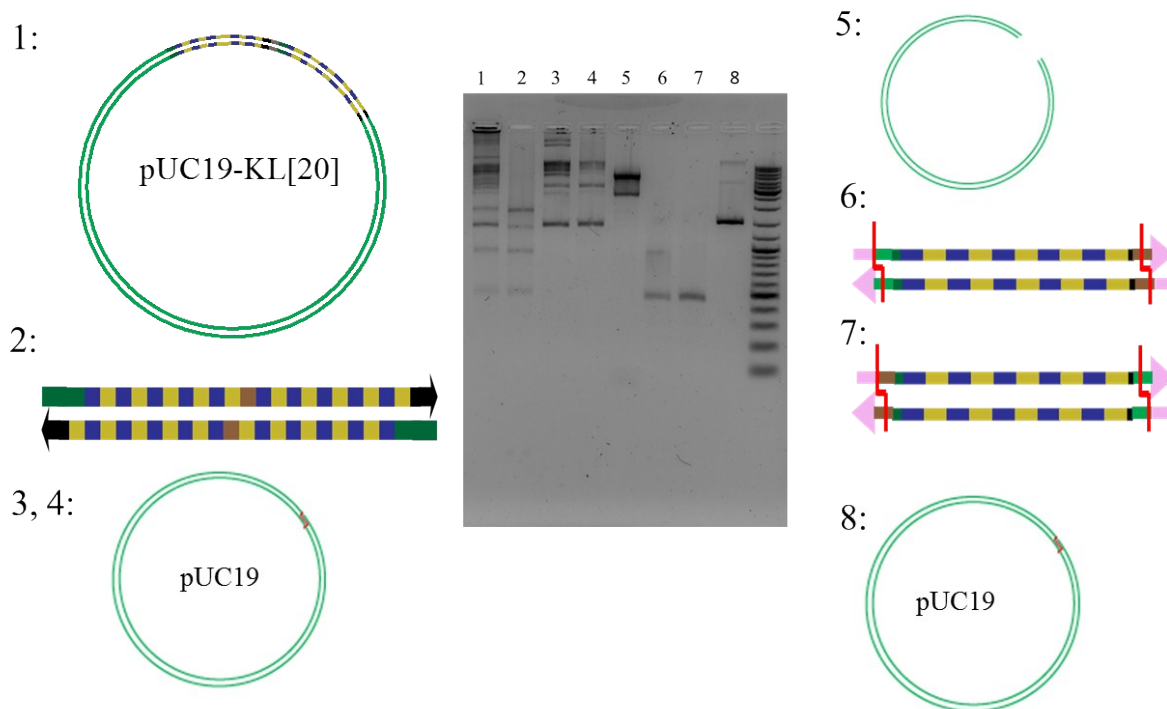


Figure S3. Native 2.5% AGE in TAE buffer displaying the comparison between the presence of ligase (Lanes 1 to 4) versus its absence (lanes 5 to 7). Lane 1: pUC19 + M + N, lane 2: M + N, lane 3: pUC19, lane 4: pUC19 (not purified), lane 5: pUC19 (not ligated), lane 6: M, lane 7: N and lane 8: pUC19 (not digested, nor ligated). Note that the all samples were digested except the ones in lane 8.

B. Transformation of dsKL[20] into pUC19

The ligation reaction (10 μ L) was used without further purification for transforming 90 μ L of MAX Efficiency DH5 α cells (LifeTechnologies), following the standard protocol provided. Positive and negative controls consisting of pUC19 digested and pUC19 intact were transformed into the cells to compare with our product. Subsequent to picking the colonies from the ligation plates, each colony was inoculated overnight in a separate 3 mL of LB media containing ampicillin (100 μ g/mL) at 225 rpm and 37°C. Finally, the plasmid was isolated using PureLink Quick Plasmid DNA Mini Prep Kit (eluted with 30 μ L of EB) and the backbone dsKL[20] was screened

via BsmBI. Table S6 displays one example in which colony 1 was screened for dsKL[20]. The mixture was incubated at 37°C for 1h.

Table S6. Digestion conditions of pUC19-[KL]20 with BsmBI

	Colony 1
DNA (50ng)	1.088
10x Tango Buffer	1
DTT (10mM)	1
dH ₂ O	6.41
Esp3I (0.5U)	0.5

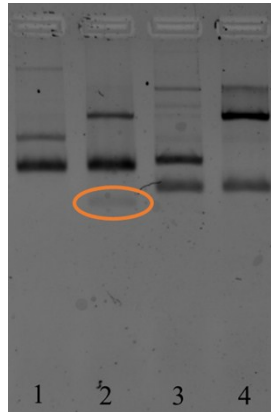


Figure S4. native AGE 2.5 % in TAE buffer showing one colony containing KL[20] (lanes 1 and 2) and another one that does not (lanes 3 and 4). Lanes 1 and 3: colonies were not treated with BsmBI, lanes 2 and 4: colonies were treated with BsmBI.

C. Sequencing of pUC19-KL[20]

The same procedure for sequencing pUC19-KL[10] was followed to test the sequence of pUC19-KL[20]. Primers pUC19-KL20BB-FOR (ataagggcgacacggaaatg) and pUC19-KL20BB-REV (atgcccttcccaacagtt) were used to perform the experiment. Note that every primer is able to sequence about 500bp from each end of the insert region. The samples were submitted for Sanger

Sequencing to McGill University Genome Center and Innovation Quebec. From combining the two sequences given by the center, we found that our scaffold contains 1 substitution (highlighted in green) with respect to our theoretical expectations.

The observed sequence was:

cgtatcacgaggcccttcgtctcgcgcgcaggtcgactctagagcgatat
aatcaaaccaaagttcagcaacaggccgtaaggatcagaagaaatcaaa
ccaaagttcagcaacaggccgtaaggatcagaagaaatcaaaccaaagttcagca
tcagcaacaggccgtaaggatcagaagaaatcaaaccaaagttcagca
caggccgtaaggatcagaagaaatcaaaccaaagttcagcaacaggcc
gtaaggatcagaagacgtagtccgaattcactggcgcgactctagagcgatat
ctttaggaatcaaaccaaagttcagcaacaggccgtaaggatcagaaga
atcaaaccaaagttcagcaacaggccgtaaggatcagaagaaatcaaac
caaagttcagcaacaggccgtaaggatcagaagaaatcaaaccaaagtt
cagcaacaggccgtaaggatcagaagaaatcaaaccaaagttcagcaac
aggccg' ■taaggatcagaagacgtagtccgaattcactggcgcgactctagagcgatat
gtcacagcttgctgtaagc

D. Conversion of dsKL[20] to ssKL[20]

We followed the same protocol described in section V to generate ss[20] using Primer pUC19-KL[20]BB-PCR-FOR (cgtatcacgaggcccttc) and 5' biotinylated Primer pUC19-KL[20]BB-PCR-REV (gcttacagacaagctgtgac). We incubated the double-stranded backbone for 3 hours instead of 2 in 0.5×SSC buffer, while gently inverting the mixture, to maximize binding. Because dsKL[20] is longer than dsKL[10], increasing the incubation time is expected to improve the binding of biotinylated dsKL[20] to the beads. Figure S5 lane 2 shows a discrete band around 500 bp that corresponds to ssKL[20]. It is noteworthy that improving the yield of ssKL[20] using this method is challenging. As described in the previous section, we attempted to increase the concentration of NaOH to enhance the separation of the double helix. However, an intense non-penetrating band was observed in the gel indicating the cleavage of biotin-streptavidin bond.

Alternatively, we optimized the conditions for using Lambda Exonuclease to generate ssKL[20] since it is faster and results in less byproducts (Figure S5). Table S7 summarizes the optimal conditions used to isolate ssKL[20] through Lambda Exonuclease method. Note that the same primers were used during PCR step before adding the exonuclease. In this case, the 5' end of the reverse primer was phosphorylated to facilitate the digestion of the antisense strand. The mixture was incubated at 37°C for 40 min then the enzyme was deactivated at 75°C for 15 min.

Table S7. Lambda Exonuclease digestion of dsKL[20]

dsKL[20] (180 ng/ μ L)	8.3 μ L (60 ng/ μ L)
10x Lambda Exo Buffer	2.5 μ L (1 \times)
λ Exo (5000 Units/mL)	7.5 μ l (1.5 U/ μ L)
dH ₂ O	6.7 μ L

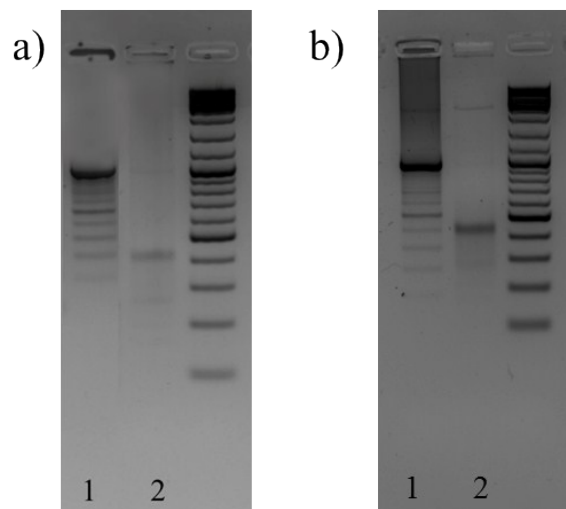


Figure S5. 2.5% AGE gel in 1 \times TAE buffer displaying the mobility shift between dsKL[20] and ssKL[20]. (a) via magnetic beads separation and (b) via Lambda Exonuclease technique. Lane 1: dsKL[20] and lane 2: ssKL[20].

VII. Preparation of the 3-tile system and related control experiments

A. Tile assembly

The assembly of DNA tiles was based on the method reported by Rothmund.² The long circular single-stranded viral scaffold M13mp18 was folded into rectangular tile with the aids of multiple short staple single-stranded DNA. The sequences of staple strands required for different tiles are listed in final section.

DNA tiles were assembled in one-pot annealing at 1 nM M13mp18 scaffold and 10 nM each staple strands in 1xTAMg buffer (45 mM Tris, 20 mM acetic acid, 12.5 mM $\text{MgCl}_2 \cdot 6\text{H}_2\text{O}$ at pH ~8.0). Then, 500 μL samples were heated to and held at 95°C for 5 min and slowly annealed to 20°C (-1°C/min). To remove excess staple strands, the samples were purified with 100kDa Amicon centrifugal filters (Millipore). First, 500 μL samples were centrifuged at 6000 rpm at 4°C for 5 mins. Then, 400 μL 1xTAMg was added and the samples were centrifuged at 5000 rpm at 4°C for 5 mins. This filtration step was repeated two more time. Approximately 50-100 μL samples were recovered, which can be stored at 4°C up to a week before use.

To determine the concentrations of DNA tiles, absorbance at 260 nm was measured by Biotek Synergy HT plate reader. The extinction coefficient of different DNA tiles can be approximated by equation (1)³

$$\varepsilon = 6700\text{ds} + 10000\text{ss} \quad (1)$$

where ds is the number of double stranded base pairs and ss is the number of single-stranded base. Using Beer-Lambert's law ($A_{260 \text{ nm}} = \varepsilon bc$, $b = 1 \text{ cm}$), the concentrations of DNA tiles can be calculated.

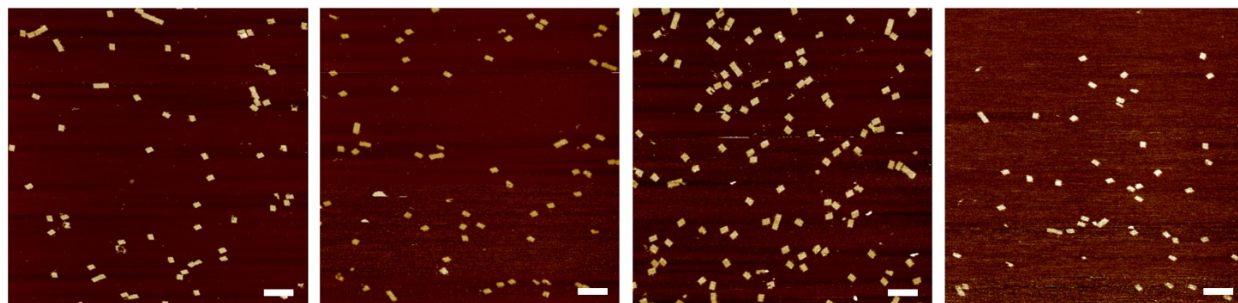


Figure S6. AFM micrographs showing the assembly of individual origami tiles, scale bar 500 nm.

B. AGE and AFM characterization of the 3-tile system

We first attempted to increase a rigidity of single-stranded backbone, ss[10], by hybridizing K blocks of the backbones with its complements (K^*). Briefly, ssKL[10] was mixed with single-stranded K^* at 1:5 ratio in 1xTAMg, and the samples were annealed from 56°C to 20°C over 1 h. To construct 3-tile system, equimolar amounts of tile X, Y and Z (0.2 nM each) were mixed in 1xTAMg, which will give final tile concentration of 0.6 nM. Then, ssKL[10]/ K^* solution was added in 5 equimolar amount with respect to total tile concentration, i.e., 3 nM, before annealing from 44°C to 20°C over 45 min. Agarose gel electrophoresis (AGE) was used to characterize the products by mixing 20 μ L samples with 4 μ L 6X loading dye then running on 1% AGE at 80 V for 2.5 h. Lane 5 of Figure S7 shows a strong band correlated to the trimers, compared to a faint band representing individual tiles. Please see refer to Figure 2 on the main text for more quantitative analysis by AFM.

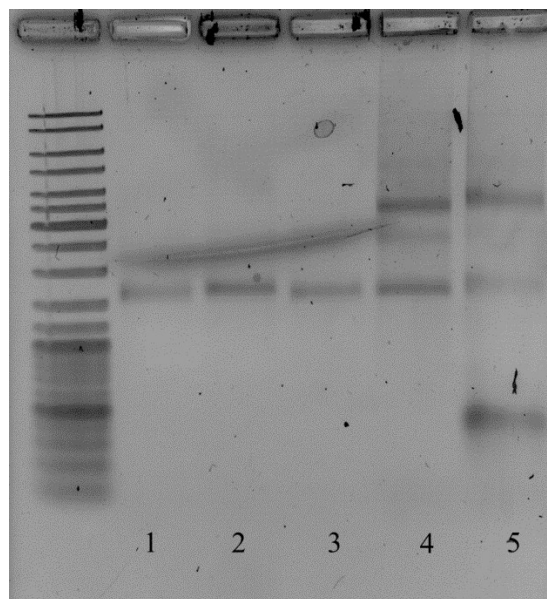


Figure S7. AGE 0.7% in TAMg. Lanes 1, 2 and 3 represent the individual tiles X, Y and Z. Lane 4: X + Y + Z and lane 5: X + Y + Z with ssKL[10] where the upper band represents the trimers, the middle band is attributed to individual tiles hybridized to ssKL[10]/K* and the lower band represents excess of ssKL[10]/K* .

To maximize the cohesion strength between DNA tiles, an assembly of DNA tiles containing 10 sticky ends (X10, Y10, Z10) was performed. In the absence of the backbone, there were tile monomer, band smearing, and non-penetrating materials, which were likely to be aggregations of DNA tiles (lane 4, Figure S8). Addition of the backbone did not improve the trimer yield as expected, even at higher backbone concentration (lane 5-7). Atomic force microscopy (AFM) revealed that tile aggregates were the major product. This is likely due to strong cohesion between tiles and various possible connections between tiles (e.g., linear dimer, staircase dimer), which can easily result in aggregation.

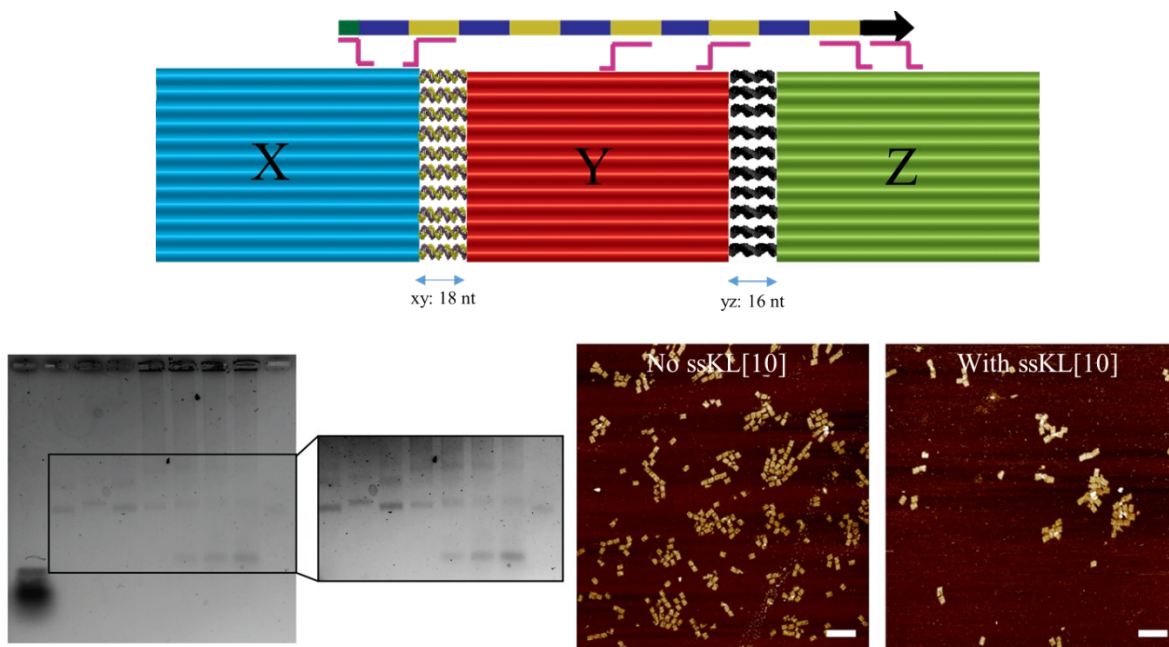


Figure S8. Top: Scheme showing 10 complementary extensions between X/Y tiles and Y/Z tiles. Bottom Left: 1% AGE in \times TAMg. Lane 1: X10, lane 2: Y10, lane 3: Z10, lane 4: X10+Y10+Z10, lane 5: (X10+Y10+Z10) + 5 equiv (ssKL[10]/K^{*}), lane 6: (X10+Y10+Z10) + 10 equiv (ssKL[10]/K^{*}) and lane 7: (X10+Y10+Z10) + 20 equiv (ssKL[10]/K^{*}). Right: AFM micrographs displaying the aggregation of three tiles in the absence and presence of ssKL[10].

Since the backbone is shown to improve the trimer yield as observed by AFM, we then followed the yield of trimers with respect to the concentration of ssKL[10] scaffolds. An efficient binding of the tiles to the backbone will, in theory, directly translate to more efficient alignment of the tiles on the backbone. Therefore, the titration of ssKL[10]/K^{*} to the mixture X+Y+Z was carried out. The gel mobility decreased with increasing backbone concentration. However, the gel mobility became unchanged at 5 equivalences of the backbone with respect to total tile concentration. Thus, we decided to choose 5 equivalences of the backbone for all experiments.

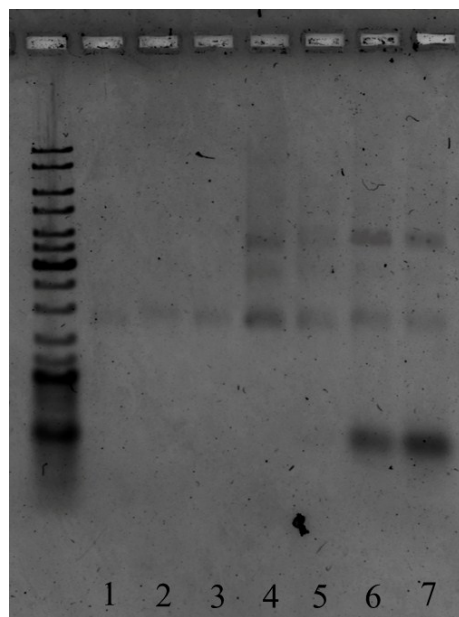


Figure S9. Titration of backbone to 3-tile system. Lane 1: X, lane 2: Y, lane 3: Z, lane 4: X + Y + Z without ssKL[10]/K^{*}, lane 5: X + Y + Z with 1 equiv. ssKL[10]/K^{*}, lane 6: X + Y + Z with 5 equiv. ssKL[10]/K^{*} and lane 7: X + Y + Z with 10 equiv. ssKL[10]/K^{*}.

As one-pot assembly of 3-tile systems, which involved mixing together X, Y, Z and ssKL[10]/K^{*}, was used in all previous experiment, we then investigated whether an order of addition of the tiles can improve the product formation. For example, X can bind first to the backbone and this preorganization may direct the binding of another two tiles to the backbone. As such, an assembly was performed in stepwise fashion: 1) annealing one of the tiles with the backbone from 44°C to 20°C, 2) adding the second tile and annealing from 44°C to 20°C, and 3) adding the third tile and annealing from 44°C to 20°C. In Figure S10, the stepwise assembly did not significantly improve the product formation (lane 6-9 VS lane 5). An exception was lane 8, which gave higher percentage of tile trimer, compared to other stepwise additions. The preorganization of the middle tile Y on the backbone seemed to be important for trimer formation. However, one-pot assembly showed significantly lower percentages of tile monomer and tile dimer.

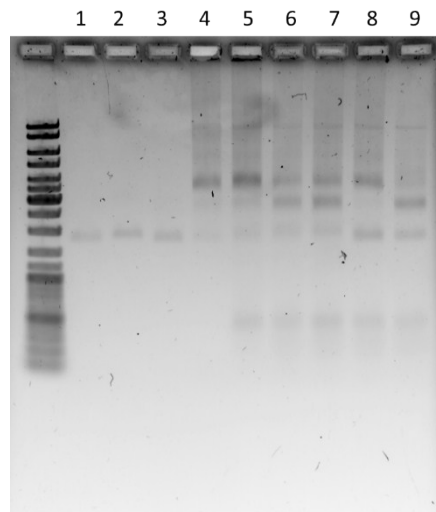


Figure S10. Step of tile additions. Lane 1: X, lane 2: Y, lane 3: Z, lane 4: X+Y+Z, lane 5: (X+Y+Z) + (ssKL[10]/K*), lane 6: (X+(ssKL[10]/K*) + Z + Y, lane 7: (Z+(ssKL[10]/K*) + X + Y, lane 8: (Y+(ssKL[10]/K*) + X + Z and lane 9: (X+(ssKL[10]/K*) + Y + Z.

Subsequent to the AGE experiments showing that the stepwise assembly starting by tiles Y and the backbone, followed by the addition of X and Z was effective, we carried on AFM measurements to study the amount of trimers with respect to monomers and dimers (Figure S11). Interestingly, the sequential addition of the tiles to ssKL[10] did not improve the yield of trimers compared to the one-pot strategy. We think that the one-pot assembly provides more binding sites to the backbone (3 tiles binding the scaffold simultaneously), hence it amplifies the effect of ssKL[10] on the construction of higher-order architectures.

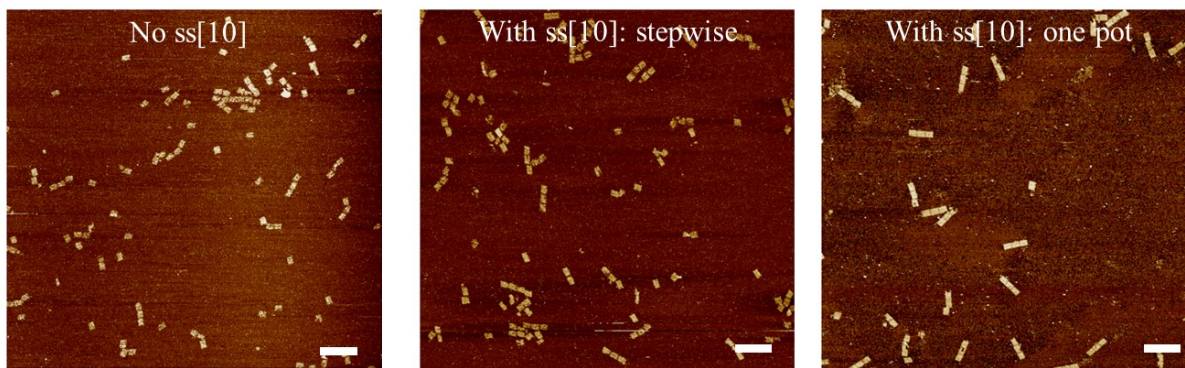


Figure S11. AFM micrographs displaying the stepwise assembly in the absence and presence of ssKL[10] versus one-pot assembly, scale bar 500 nm.

VIII. Construction of the railroad track and related control experiments

A. AGE characterization of the railroad track

The one-pot assembly of the 5-tile system follows the same protocol as the 3-tile system. ssKL[20] was first rigidified by hybridizing either K or L blocks of the backbones with their complements (K^* or L^* , respectively). We first attempted to hybridize one scaffold to tiles A, B, C, D and E, then examined the railroad system. Briefly, ssKL[20] was mixed with single-stranded K^* or L^* at 1:10 ratio in 1xTAMg, and the samples were annealed from 56°C to 20°C over 1 h. To construct 5-tile system, equimolar amounts of tiles A, B, C, D and E (0.2 nM each) were mixed in 1xTAMg, which will give final tile concentration of 1 nM. Then, ssKL[20]/ K^* and/or ssKL[20]/ L^* solutions were added each in 5 equimolar amount with respect to total tile concentration before annealing from 44°C to 20°C over 4 hours. AGE (Figure S12) was used to characterize the products by mixing 20 μ L samples with 4 μ L 6X loading dye then running on 0.7% AGE at 80 V for 2.5 h. Lane 9 corresponds to the nanostructure with ssKL[20]/ K^* only and lane 8 to the railroad track in the presence of ssKL[20]/ K^* and ssKL[20]/ L^* . The results validate the AFM data shown in Figure 3 where 2 scaffolds were needed to enhance the formation yield of pentamers. Compared to lane

7, the railroad track system helped minimize other contaminants such as dimers, trimers and so on.

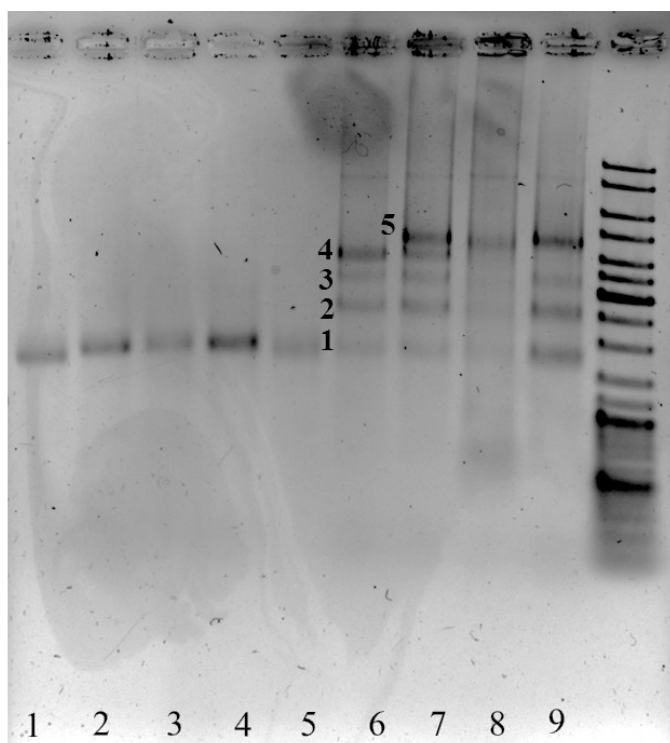


Figure S12. 1% AGE in 1×TAMg. Lane 1: A, lane 2: B, lane 3: C, lane 4: D, lane 5: E, lane 6: A + B + C + D, lane 7: A + B + C + D + E, lane 8: A + B + C + D + E + ssKL[20]/K* + ssKL[20]/L* and lane 9: A + B + C + D + E + ssKL[20]/K*.

In an attempt to further increase the yield of pentamers, we only added ssKL[20]/K* to the 5 tiles during 44 to 20°C cycle. Then, we incubated the mixture with ssKL[20]/L* at room temperature for 2 hours. The AGE gel in Figure S13 shows no significant enhancement in the yield of pentamers. Again, we believe that both backbones are needed to better organize the pentamers by minimizing the non-desired interactions and maximizing the hybridizing sites between tiles and backbones.

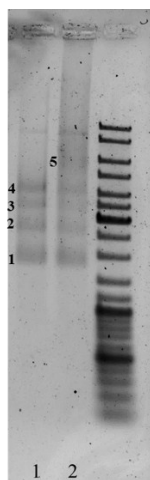


Figure S13. 1% AGE in 1×TAMg. lane 1: A + B + C + D + E and lane 2: (A + B + C + D + E + ssKL[20]/K*) at 44 to 20°C + ssKL[20]/L* at RT.

B. AFM characterization of the railroad track

We examined the ability of ssKL[20] to organize pentamers and improve their yields at room temperature. Thus, we annealed/cooled the mixture containing A, B, C, D and E from 44°C to 20°C over 4 hours first, then added ssKL[20]/K* and ssKL[20]/L* at room temperature. Figure S14 displays a mixture of individual tiles, trimers, tetramers, pentamers and other misassembled nanostructures. Compared to the simultaneous addition of ssKL[20] to the tiles, the railroad track does not seem to improve the yield of pentamers significantly. Similar to the 3-tile system, we suppose that the interactions holding the pre-formed higher-order structures are difficult to rearrange even after the addition of the backbones. As such, it is critical to add all the strands at the same time in order to promote the assembly of pentamers.

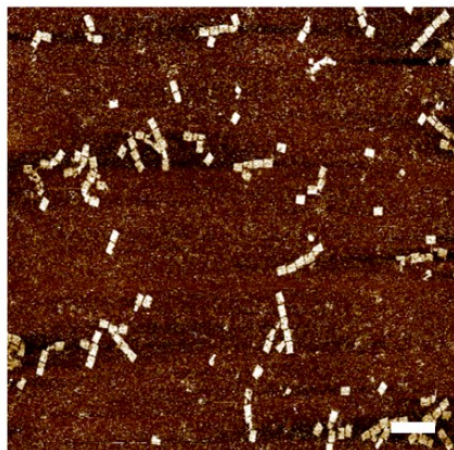


Figure S14. AFM micrograph displaying the assembly of 5 tiles with the 2 backbones at RT, scale bar 500 nm.

Next, we examined the organization of the 5 tiles using the same sticky ends between two tiles (with 16 bp complementary domains instead of 10). For example, the sequence of the 5 overhangs between tiles C and D is the same but different than the one between D and E. Similar to the 3-tiles system, we aimed at further decreasing the number of strands used and at increasing symmetry. However, the tiles did not arrange correctly and they aggregated instead (Figure S15). Furthermore, we increased the number of sticky ends, using the same sequences between the tiles, from 5 to 10. Figure S15 demonstrates the formation of aggregates on the mica surface. Thus, we decided to use 5 unique sticky ends to link the tiles.

It is noteworthy that in order to further grow the 1D tracks, a longer backbone is required. We are currently working on generating custom made DNA scaffolds with repetitive sequences of 4000 base pairs in length. Theoretically, higher-order assemblies of 1D and 2D DNA origami should be produced when these three main rules are respected: (i) the hybridization length between the backbone and each origami tile must be larger than the length of the lateral cohesion between the tiles; (ii) the pre-assembled individual origami tiles and the backbone must be annealed in a single

reaction mixture for a least 4 hours and (iii) the ratio of backbone to the origami tiles is critical to avoid secondary interactions and the growth of unwanted products.

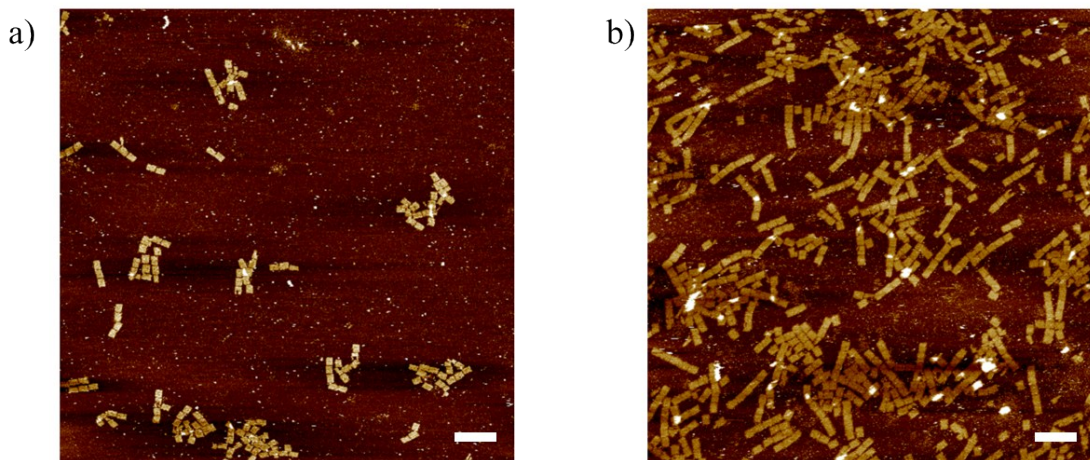


Figure S15. AFM micrographs depicting the formation of aggregates between the tiles when using (a) 5 sticky ends and (b) 10 sticky ends, scale bar 500 nm.

IX. Additional AFM micrographs

We present in this section additional AFM images that correspond to the railroad track system with ssKL[20]/K* and ssKL[20]/L* (Figure S17) and 3 tiles system with ssKL[10]/K*(Figure S16). The corresponding control experiments in the absence of the backbones are described in figures S18 and S19. These images were part of the data acquired to perform the statistical analysis on these nanostructures.

To perform the quantification on these images, we manually counted the number of trimers and pentamers versus all the other byproducts. For the 5-tiles system, the aim of our strategy was to improve the organization of pentamers by minimizing the non-desired interactions between the tiles and ss[20]. Interestingly, 5 tiles were successfully arranged by ss[10]/K* and ss[20]/L*

scaffolds up to 66% (69% if we considered not well aligned pentamers) compared to 19% (30% including all types of pentamers) in the absence of any backbone. Accordingly, the percentage of tetramers decreased from 31% to 10% and the amount of trimers was reduced from 10% to 7%. We have also found that the yield of monomers was reduced from 12% (in the absence of backbones) to 4% upon addition of ssKL[10]/K* and ssKL[10]/L*. The results suggest that the addition of 2 scaffolds is highly advantageous to assure the organization of the tiles in 1-D. By using the railroad track system, we think that applications requiring larger areas than a single origami can be achievable.

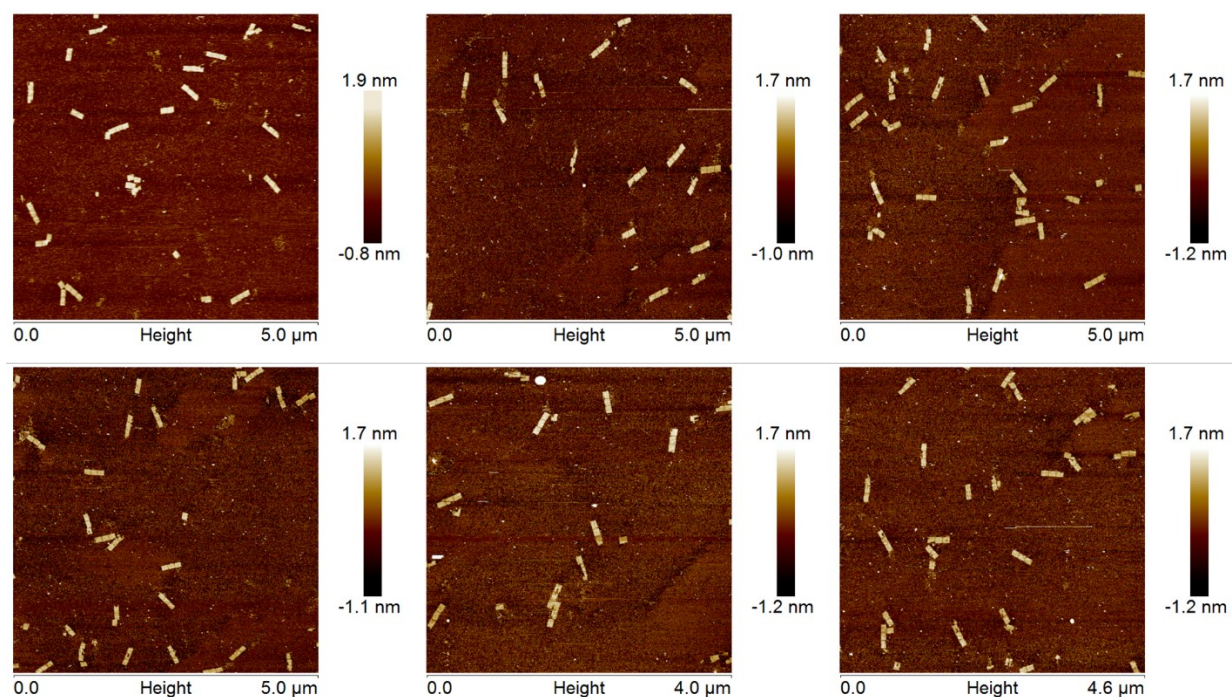


Figure S16. 3-tiles system

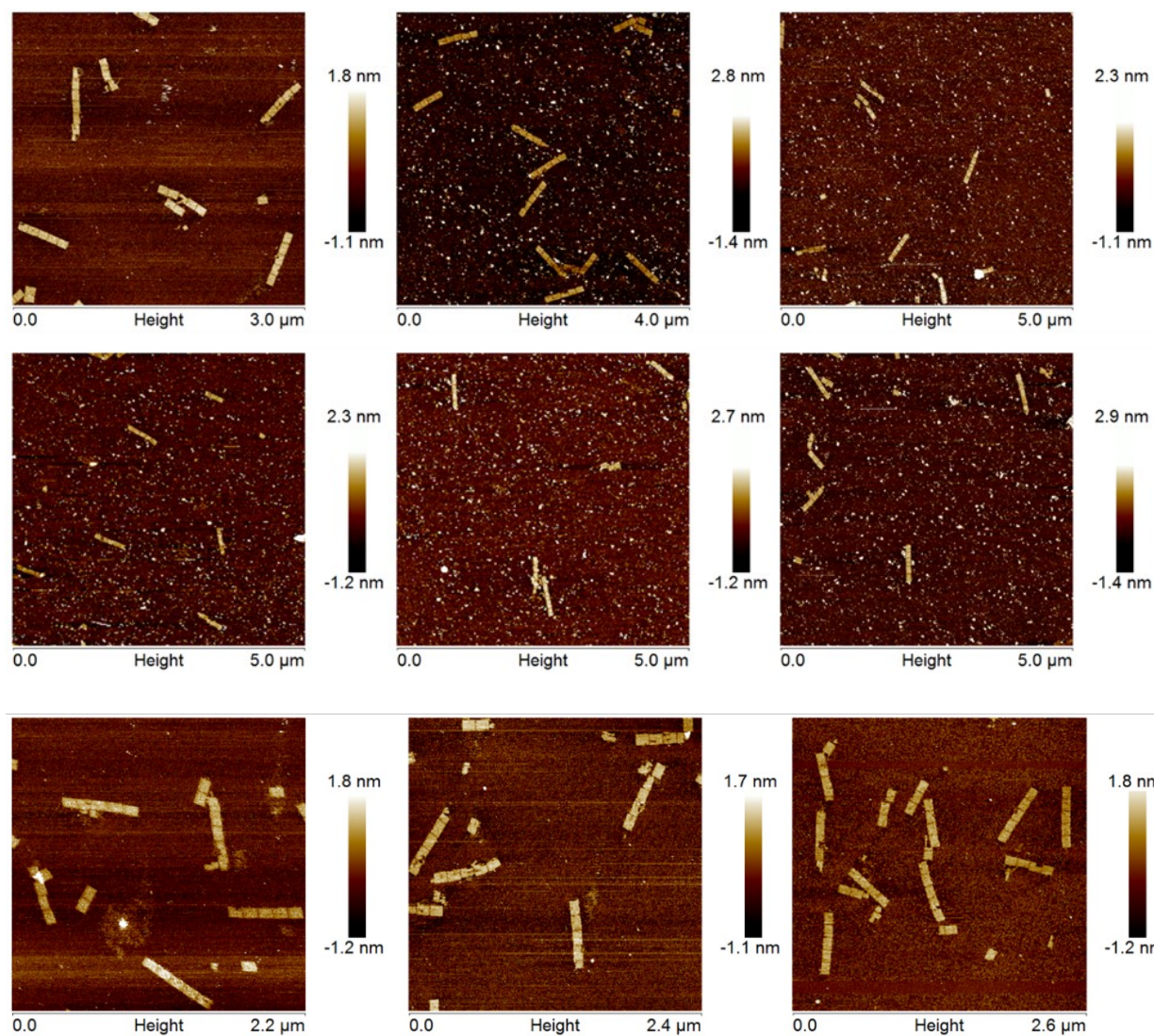


Figure S17. Railroad track system.

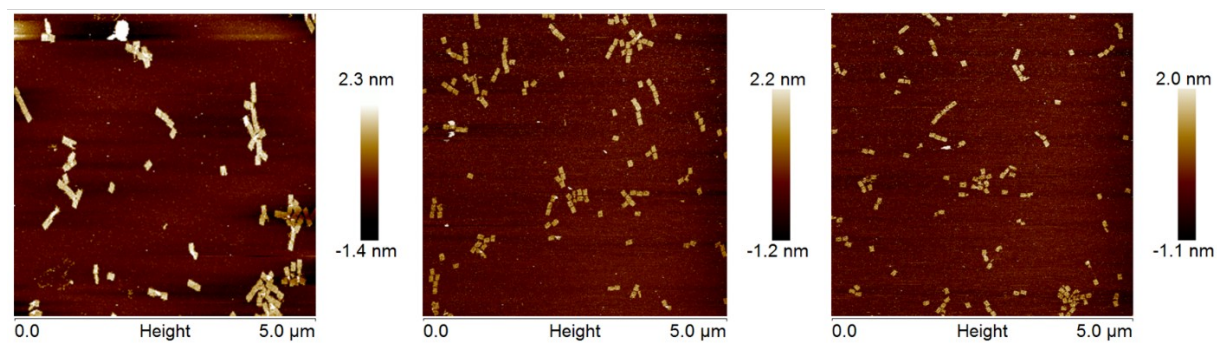


Figure S18. 3-tiles system in the absence of ssKL[10]/K*.

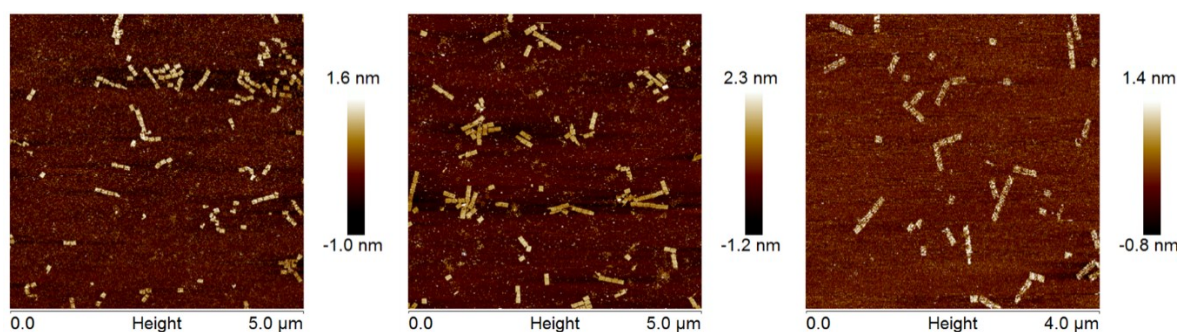


Figure S19. 5-tiles system in the absence of ssKL[10]/K* and ssKL[10]/L*.

X. Origami sequences

Tables S8-S10 show lists of modified staple strands that replaced unmodified strands of the same number for generating different origami tiles. In general, the three-digit numbers or the numbers following the first letter indicate the position of the corresponding modified staple strands on origami tile. Other nomenclatures are described below.

Staple strands that hybridize between the backbone and the origami tiles:

- 1) Strands starting with letters X, Y and Z are staples that have single-stranded extension either on their 3' or 5' end to provide the hybridization between the backbone and origami tiles X, Y and Z (Figure S20).
- 2) Strands starting with letters A, B, C, D and E are staples that have single-stranded extension either on their 3' or 5' end to provide the hybridization between the backbone and origami tiles A, B, C, D and E (the same strategy as shown in Figure S20). In addition, the 10b abbreviation indicates that these staples are only used in the 5-tile system containing 10-base overhangs (see Table S10).

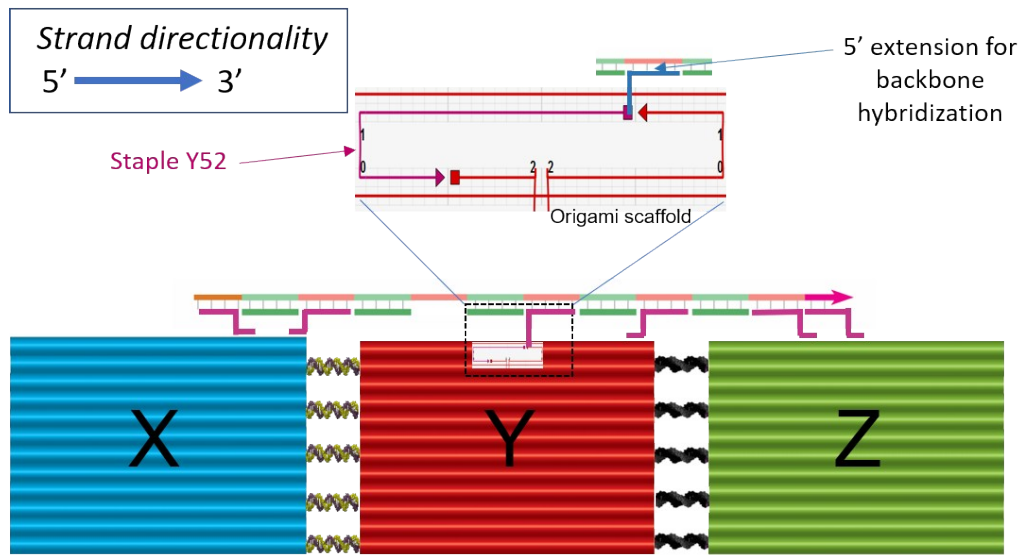


Figure S20. Scheme illustrating the staple-strand extension for hybridization of origami tiles to the backbone. The extension is highlighted in blue.

Staples strands that have an internal hairpin:

- 3) Strands starting with the letter H have an internal hairpin sequence (TCCTCTTTTGAGGAACAAGTTTTCTTGT). See Figure S21.

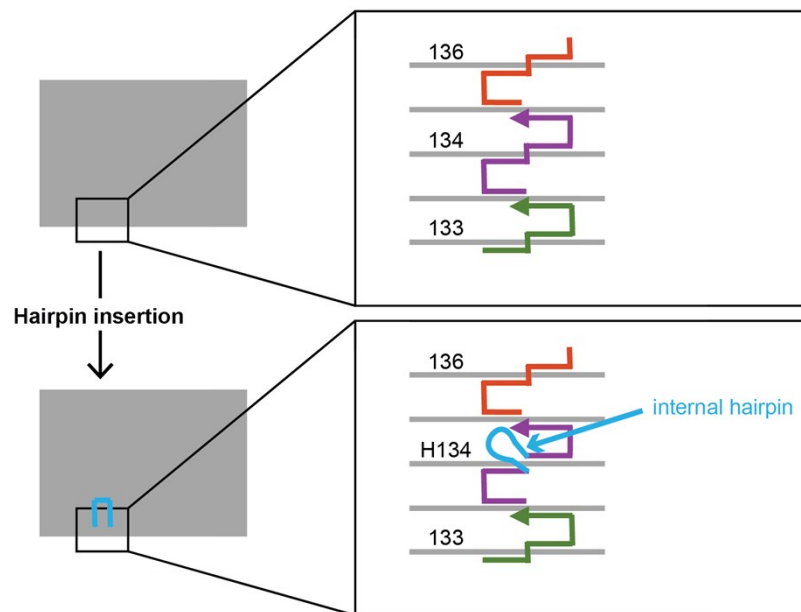


Figure S21. Incorporation of hairpins on origami tiles

Staple strands for lateral cohesion between origami tiles:

- 4) Staples in the DNA tile's corners (100, 111, 205 and 216) have a portion of their 5' ends removed. This creates a set of strands starting with letter M. The purpose is to remove T4 spacer and to allow the extension of their neighboring staple strands for lateral cohesion between origami tiles. (i.e., M216 allows us to extend the 3' end of staple 215; see Figure S22).

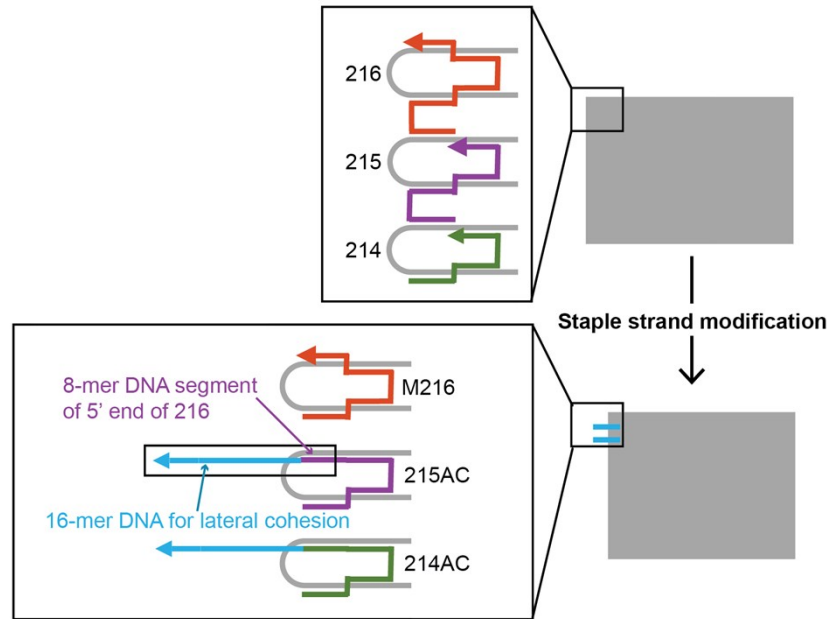


Figure S22. Scheme illustrating the modification of the strands involved in the lateral cohesion between the origami tiles. A portion of 5' end of staple 216 was removed to allow the 3' extension of staple 215, resulting in M216 (and 215AC).

- 5) A set of staples called 'S' strands have 12-bases deletion (8 bases of M13-binding segment and T4 spacer) on their 5' ends. This allows us to extend the length of their neighboring staple strands for lateral cohesion (i.e., S103 allows us to extend the 3' end of staple 102; see Figure S23).

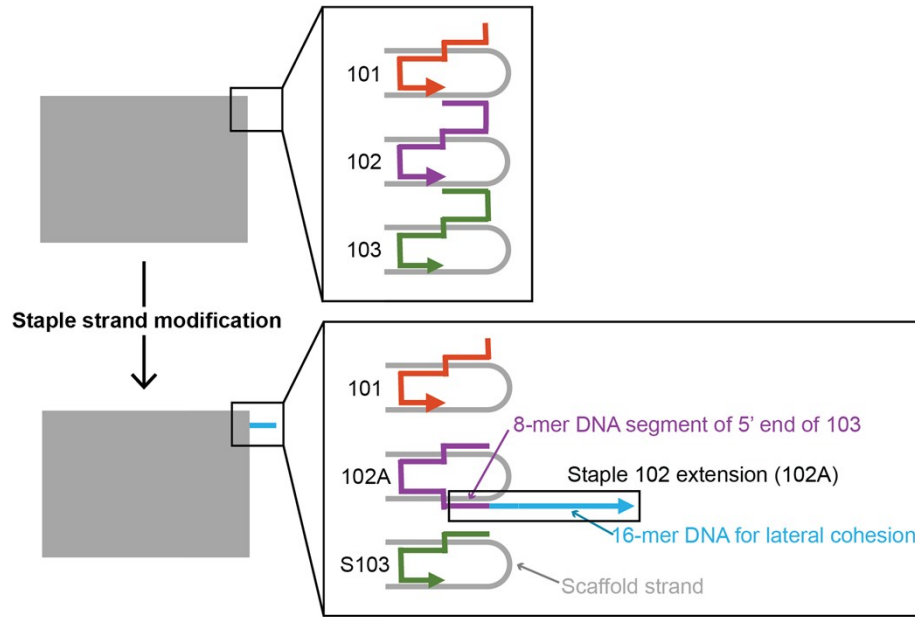


Figure S23. Scheme illustrating the modification of the strands involved in the lateral cohesion between the origami tiles. A portion of 5' end of staple 103 was removed to allow the 3' extension of staple 102, resulting in S103 (and 102A).

- 6) Strands starting with three-digit number followed by one or two letters are staples that provide lateral cohesion between pairs of origami tiles (for example, 102A in Figure S23 and 214BC in Figure S24). Two modifications are required for these lateral cohesion staple strands (see the transformation of staple 102 to staple 102A in Figure S23):
 - a. Remove 12 bases on the 5' end,
 - b. Extend the 3' end for an additional 24 bases—8 bases bind to the M13 backbone and 16 bases overhang are for lateral cohesion.

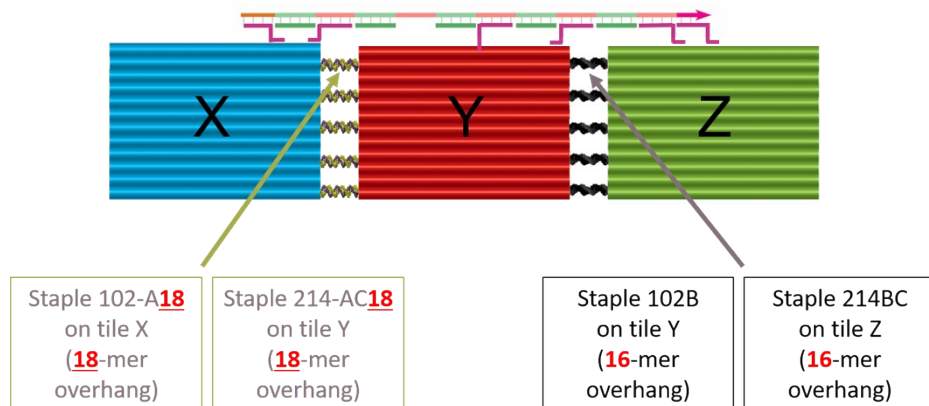


Figure S24. Example of staple strands for lateral cohesion between origami tiles X, Y and Z.

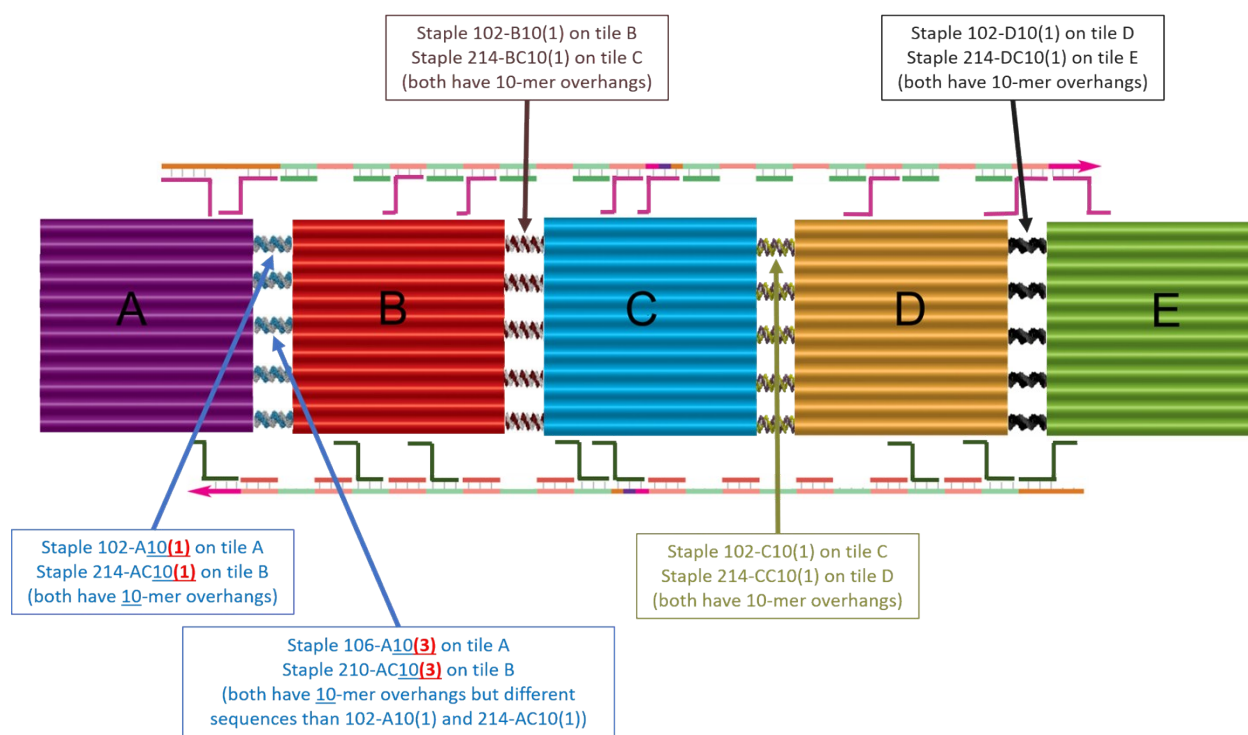


Figure S25. Example of staple strands for lateral cohesion between origami tiles A, B, C, D and E.

Additional notes:

- a. A and its complementary AC connect tile X and Y or tile A and B (Figures S24 and S25).
- b. B and its complementary BC connect tile Y and Z or tile B and C (Figures S24 and S25).
- c. C and its complementary CC connect tile C and D (Figures S25).
- d. D and its complementary DC connect tile D and E (Figures S25).
- e. The abbreviations A18 and AC18 indicate that the overhangs for lateral cohesion are 18 bases long, rather than 16 bases long (Figure S24).
- f. The abbreviations 10 indicate that the overhangs for lateral cohesion are 10 bases long, rather than 16 bases long, and all have unique sequences (Figure S25).

Table S8. Modification of staple strands in 3-tile system. 5 stands for 5 sticky ends between the tiles and 10 stands for 10 sticky ends linking the tiles.

Tile	Staple strand modifications
X	X54, X100 H134, H136, H137, H139, H158, H160 102-A18, S103, 104-A18, S105, 106-A18, S107, 108-A18, S109, 110-A18, M111
Y	Y52, Y132 H134, H136, H137, H139, H158, H160 102B, S103, 10B, S105, 106B, S107, 108B, S109, 110B, M111 M205, 206-AC18, S207, 208-AC18, S209, 210-AC18, S211, 212-AC18, S213, 214-AC18, S215
Z	Z178, Z202 H134, H136, H137, H139, H158, H160 M205, 206BC, S207, 208BC, S209, 210BC, S211, 212BC, S213, 214BC, S215
X10	X54, X100 H134, H136, H137, H139, H158, H160 101-A18, 102-A18, 103-A18, 104-A18, 105-A18, 106-A18, 107-A18, 108-A18, 109-A18, 110-A18, M111

Tile	Staple strand modifications
Y10	Y52, Y132 H134, H136, H137, H139, H158, H160 101B, 102B, 103B, 104B, 105B, 106B, 107B, 108B, 109B, 110B, M111 M205, 206-AC18, 207-AC18, 208-AC18, 209-AC18, 210-AC18, 211-AC18, 212-AC18, 213-AC18, 214-AC18, 215AC-18, M216
Z10	Z178, Z202 H134, H136, H137, H139, H158, H160 M205, 206BC, 207BC, 208BC, 209BC, 210BC, 211BC, 212BC, 213BC, 214BC, 215BC, M216

Table S9. Modification of staple strands in 5-tile system. The same sequences were employed in each sticky end linking the same tiles.

Tile	Staple strand modifications
A	A78, A100, A111 102A, S103, 104A, S105, 106A, S107, 108A, S109, 110A, M111
B	B52, B132, B159, B73 102B, S103, 104B, S105, 106B, S107, 108B, S109, 110B, M111 M205, 206AC, S207, 208AC, S209, 210AC, S211, 212AC, S213, 214AC, S215
C	C132, C156, C135, C113 102C, S103, 104C, S105, 106C, S107, 108C, S109, 110C, M111 M205, 206BC, S207, 208BC, S209, 210BC, S211, 212BC, S213, 214BC, S215
D	D52, D180, D135, D197 102D, S103, 104D, S105, 106D, S107, 108D, S109, 110D, M111 M205, 206CC, S207, 208CC, S209, 210CC, S211, 212CC, S213, 214CC, S215
E	E216, E205 206DC, S207, 208DC, S209, 210DC, S211, 212DC, S213, 214DC, S215
A10	A78, A100, A111 101A, 102A, 103A, 104A, 105A, 106A, 107A, 108A, 109A, 110A, M111
B10	B52, B132, B159, B73 M100, 101B, 102B, 103B, 104B, 105B, 106B, 107B, 108B, 109B, 110B, M111 M205, 206AC, 207AC, 208AC, 209AC, 210AC, 211AC, 212AC, 213AC, 214AC, 215AC, M216
C10	C132, C156, C135, C113 M100, 101C, 102C, 103C, 104C, 105C, 106C, 107C, 108C, 109C, 110C, M111 M205, 206BC, 207BC, 208BC, 209BC, 210BC, 211BC, 212BC, 213BC, 214BC, 215BC, M216
D10	D52, D180, D135, D197 M100, 101D, 102D, 103D, 104D, 105D, 106D, 107D, 108D, 109D, 110D, M111 M205, 206CC, 207CC, 208CC, 209CC, 210CC, 211CC, 212CC, 213CC, 214CC, 215CC, M216

Tile	Staple strand modifications
E10	E216, E205 206DC, 207DC, 208DC, 209DC, 210DC, 211DC, 212DC, 213DC, 214DC, 215DC, M216

Table S10. Modification of staple strands in 5-tile system. Each overhang (10 bases) is unique in this case.

Tile	Staple strand modifications
A5(10)u	100, 101, 102A10(1), S103, 104A10(2), S105, 106A10(3), S107, 108A10(4), S109, 110A10(5), S111
B5(10)u	B52, B132, B159, B73 100, 101, 102B10(1), S103, 104B10(2), S105, 106B10(3), S107, 108B10(4), S109, 110B10(5), S111 205, 206AC10(5), S207, 208AC10(4), S209, 210AC10(3), S211, 212AC10(2), S213, 214AC10(1), S215, 216
C5(10)u	C132-10b, C156-10b, C135-10b, C113-10b 100, 101, 102C10(1), S103, 104C10(2), S105, 106C10(3), S107, 108C10(4), S109, 110C10(5), S111 205, 206BC10(5), S207, 208BC10(4), S209, 210BC10(3), S211, 212BC10(2), S213, 214BC10(1), S215, 216
D5(10)u	D1-10b, D76-10b, D97-10b, D135-10b 100, 101, 102D10(1), S103, 104D10(2), S105, 106D10(3), S107, 108D10(4), S109, 110D10(5), S111 205, 206CC10(5), S207, 208CC10(4), S209, 210CC10(3), S211, 212CC10(2), S213, 214CC10(1), S215, 216
E5(10)u	E183-10b, E202-10b 205, 206DC10(5), S207, 208DC10(4), S209, 210DC10(3), S211, 212DC10(2), S213, 214DC10(1), S215, 216

Unmodified staple strands (1-216)

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1      CAAGCCCAATAGGAACCCATGTACAAACAGTT
2      AATGCCCCGTAACAGTGCCCGTATCTCCCTCA
3      TGCCTTGACTGCCTATTTGGAACAGGGATAG
4      GAGCCGCCCCACCAACCGGAACCGCGACGGAAA
5      AACCAGAGACCTCAGAACCGCCAGGGGTCAG
6      TTATTCATAGGGAAGGTAAATATTCATTCAGT
7      CATAACCCGAGGCATAGTAAGAGCTTTTAAAG
8      ATTGAGGGTAAAGGTGAATTATCAATCACCGG
9      AAAAGTAATATCTTACCGAAGCCCTTCCAGAG

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10 GCAATAGCGCAGATAGCCGAACAATTCAACCG
11 CCTAATTTACGCTAACGAGCGTCTAATCAATA
12 TCTTACCAGCCAGTTACAAAATAAATGAAATA
13 ATCGGCTGCGAGCATGTAGAAACCTATCATAT
14 CTAATTTATCTTTCCTTATCATTCATCCTGAA
15 GCGTTATAGAAAAAGCCTGTTTAGAAGGCCGG
16 GCTCATTTTCGCATTAAATTTTGTAGCTTAGA
17 AATTACTACAAATTCTTACCAGTAATCCCATC
18 TTAAGACGTTGAAAACATAGCGATAACAGTAC
19 TAGAATCCCTGAGAAGAGTCAATAGGAATCAT
20 CTTTACACAGATGAATATACAGTAAACAATT
21 TTTAACGTTTCGGGAGAAACAATAATTTCCCT
22 CGACAATAAGTATTAGACTTTACAATACCGA
23 GGATTTAGCGTATTAAATCCTTTGTTTTCAGG
24 ACGAACCAAAACATCGCCATTAAATGGTGGTT
25 GAACGTGGCGAGAAAGGAAGGGAACAACTAT
26 TAGCCCTACCAGCAGAAGATAAAAACATTTGA
27 CGGCCTTGCTGGTAATATCCAGAACGAACTGA
28 CTCAGAGCCACCACCCTCATTTTCCTATTATT
29 CTGAAACAGGTAATAAGTTTAAACCCCTCAGA
30 AGTGTACTTGAAAGTATTAAGAGGCCGCCACC
31 GCCACCACTCTTTTCATAATCAAACCGTCACC
32 GTTTGCCACCTCAGAGCCGCCACCGATACAGG
33 GACTTGAGAGACAAAAGGGCGACAAGTTACCA
34 AGCGCCAACCATTTGGGAATTAGATTATTAGC
35 GAAGGAAAATAAGAGCAAGAAACAACAGCCAT
36 GCCCAATACCGAGGAAACGCAATAGGTTTACC
37 ATTATTTAACCCAGCTACAATTTTCAAGAACG
38 TATTTTGCTCCCAATCCAAATAAGTGAGTTAA
39 GGTATTAAGAACAAGAAAAATAATTAAAGCCA
40 TAAGTCCTACCAAGTACCGCACTCTTAGTTGC
41 ACGCTCAAAATAAGAATAAACACCGTGAATTT
42 AGGCGTTACAGTAGGGCTTAATTGACAATAGA
43 ATCAAAATCGTCGCTATTAATTAACGGATTCG
44 CTGTAAATCATAGGTCTGAGAGACGATAAATA

45 CCTGATTGAAAGAAATTGCGTAGACCCGAACG
46 ACAGAAATCTTTGAATACCAAGTTCCTTGCTT
47 TTATTAATGCCGTCAATAGATAATCAGAGGTG
48 AGATTAGATTTAAAAGTTTGAGTACACGTAAA
49 AGGCGGTCATTAGTCTTTAATGCGCAATATTA
50 GAATGGCTAGTATTAACACCGCCTCAACTAAT
51 CCGCCAGCCATTGCAACAGGAAAAATATTTTT
52 CCCTCAGAACCGCCACCCTCAGAACTGAGACT
53 CCTCAAGAATACATGGCTTTTGATAGAACCAC
54 TAAGCGTCGAAGGATTAGGATTAGTACCGCCA
55 CACCAGAGTTCGGTCATAGCCCCCGCCAGCAA
56 TCGGCATTCCGCCGCCAGCATTGACGTTCCAG
57 AATCACCAAATAGAAAATTCATATATAACGGA
58 TCACAATCGTAGCACCATTACCATCGTTTTCA
59 ATACCCAAGATAACCCACAAGAATAAACGATT
60 ATCAGAGAAAGAACTGGCATGATTTTATTTTG
61 TTTTGTTTAAGCCTTAAATCAAGAATCGAGAA
62 AGGTTTTGAACGTCAAAAATGAAAGCGCTAAT
63 CAAGCAAGACGCGCCTGTTTATCAAGAATCGC
64 AATGCAGACCGTTTTTATTTTCATCTTGCGGG
65 CATATTTAGAAATACCGACCGTGTTACCTTTT
66 AATGGTTTACAACGCCAACATGTAGTTCAGCT
67 TAACCTCCATATGTGAGTGAATAAACAAAATC
68 AAATCAATGGCTTAGGTTGGGTACTAAATTT
69 GCGCAGAGATATCAAAATTATTTGACATTATC
70 AACCTACCGGAATTATTCATTTCCAGTACAT
71 ATTTTGCCTCTTTAGGAGCACTAAGCAACAGT
72 CTAAAATAGAACAAAGAAACCACCAGGGTTAG
73 GCCACGCTATACGTGGCACAGACAACGCTCAT
74 GCGTAAGAGAGAGCCAGCAGCAAAAAGGTTAT
75 GGAAATACCTACATTTTGACGCTCACCTGAAA
76 TATCACCGTACTCAGGAGGTTTAGCGGGGTTT
77 TGCTCAGTCAGTCTCTGAATTTACCAGGAGGT
78 GGAAAGCGACCAGGCGGATAAGTGAATAGGTG
79 TGAGGCAGGCGTCAGACTGTAGCGTAGCAAGG

80 TGCCTTTAGTCAGACGATTGGCCTGCCAGAAT
81 CCGGAAACACACCACGGAATAAGTAAGACTCC
82 ACGCAAAGGTCACCAATGAAACCAATCAAGTT
83 TTATTACGGTCAGAGGGTAATTGAATAGCAGC
84 TGAACAAACAGTATGTTAGCAAATAAAAGAA
85 CTTTACAGTTAGCGAACCTCCCGACGTAGGAA
86 GAGGCGTTAGAGAATAACATAAAAGAACACCC
87 TCATTACCCGACAATAAAACAACATATTTAGGC
88 CCAGACGAGCGCCCAATAGCAAGCAAGAACGC
89 AGAGGCATAATTTTCATCTTCTGACTATAACTA
90 TTTTAGTTTTTCGAGCCAGTAATAAATTCTGT
91 TATGTAAACCTTTTTTAATGGAAAAATTACCT
92 TTGAATTATGCTGATGCAATCCACAAATATA
93 GAGCAAAAACCTTCTGAATAATGGAAGAAGGAG
94 TGGATTATGAAGATGATGAAACAAAATTCAT
95 CGGAATTATTGAAAGGAATTGAGGTGAAAAAT
96 ATCAACAGTCATCATATTCCTGATTGATTGTT
97 CTAAAGCAAGATAGAACCCTTCTGAATCGTCT
98 GCCAACAGTCACCTTGCTGAACCTGTTGGCAA
99 GAAATGGATTATTTACATTGGCAGACATTCTG
100 TTTTATAAGTATAGCCCGGCCGTCGAG
101 AGGGTTGATTTTATAAATCCTCATTAAATGATATTC
102 ACAAACAATTTTAATCAGTAGCGACAGATCGATAGC
103 AGCACCGTTTTTTAAAGGTGGCAACATAGTAGAAAA
104 TACATACATTTTGACGGGAGAATTAACACAGGGAA
105 GCGCATTATTTTGCTTATCCGGTATTCTAAATCAGA
106 TATAGAAGTTTTTCGACAAAAGGTAAAGTAGAGAATA
107 TAAAGTACTTTTCGCGAGAAAACCTTTTATCGCAAG
108 ACAAAGAATTTTATTAATTACATTTAACACATCAAG
109 AAAACAAATTTTTTCATCAATATAATCCTATCAGAT
110 GATGGCAATTTTAATCAATATCTGGTCACAAATATC
111 AAACCTCTTTTACCAGTAATAAAAGGGATTCACCAGTCACACGTTTT
112 CCGAAATCCGAAAATCCTGTTTGAAGCCGGAA
113 CCAGCAGGGGCAAAATCCCTTATAAAGCCGGC
114 GCATAAAGTTCCACACAACATACGAAGCGCCA

115 GCTCACAATGTAAAGCCTGGGGTGGGTTTGCC
116 TTCGCCATTGCCGGAACACAGGCATTAAATCA
117 GCTTCTGGTCAGGCTGCGCAACTGTGTTATCC
118 GTTAAAATTTTAACCAATAGGAACCCGGCACC
119 AGACAGTCATTCAAAAGGGTGAGAAGCTATAT
120 AGGTAAAGAAATCACCATCAATATAATATTTT
121 TTTCATTTGGTCAATAACCTGTTTATATCGCG
122 TCGCAAATGGGGCGCGAGCTGAAATAATGTGT
123 TTTTAATTGCCCCGAAAGACTTCAAAACACTAT
124 AAGAGGAACGAGCTTCAAAGCGAAGATACATT
125 GGAATTACTCGTTTACCAGACGACAAAAGATT
126 GAATAAGGACGTAACAAAGCTGCTCTAAAACA
127 CCAAATCACTTGCCCTGACGAGAACGCCAAAA
128 CTCATCTTGAGGCAAAAGAATACAGTGAATTT
129 AAACGAAATGACCCCCAGCGATTATTCATTAC
130 CTTAAACATCAGCTTGCTTTCGAGCGTAACAC
131 TCGGTTTAGCTTGATACCGATAGTCCAACCTA
132 TGAGTTTCGTCACCAGTACAACTTAATTGTA
133 CCCCATTAGAGCTTGACGGGGAAATCAAAA
134 GAATAGCCGCAAGCGGTCCACGCTCCTAATGA
135 GAGTTGCACGAGATAGGGTTGAGTAAGGGAGC
136 GTGAGCTAGTTTCCTGTGTGAAATTTGGGAAG
137 TCATAGCTACTCACATTAATTGCGCCCTGAGA
138 GGCGATCGCACTCCAGCCAGCTTTGCCATCAA
139 GAAGATCGGTGCGGGCCTCTTCGCAATCATGG
140 AAATAATTTTAAATTGTAAACGTTGATATTCA
141 GCAAATATCGCGTCTGGCCTTCCTGGCCTCAG
142 ACCGTTCTAAATGCAATGCCTGAGAGGTGGCA
143 TATATTTTAGCTGATAAATTAATGTTGTATAA
144 TCAATTCTTTAGTTTGACCATTACCAGACCG
145 CGAGTAGAACTAATAGTAGTAGCAAACCCTCA
146 GAAGCAAAAAAGCGGATTGCATCAGATAAAAA
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148 CCAAAATATAATGCAGATACATAAACACCAGA
149 CATTCAACGCGAGAGGCTTTTGCATATTATAG

150 ACGAGTAGTGACAAGAACCGGATATACCAAGC
151 AGTAATCTTAAATTGGGCTTGAGAGAATACCA
152 GCGAAACATGCCACTACGAAGGCATGCGCCGA
153 ATACGTAAAAGTACAACGGAGATTTTCATCAAG
154 CAATGACACTCCAAAAGGAGCCTTACAACGCC
155 AAAAAAGGACAACCATCGCCACGCGGGTAAA
156 TGTAGCATTCCACAGACAGCCCTCATCTCAA
157 GTAAAGCACTAAATCGGAACCCTAGTTGTTCC
158 AGTTTGGAGCCCTTCACCGCCTGGTTGCGCTC
159 AGCTGATTACAAGAGTCCACTATTGAGGTGCC
160 ACTGCCCCGCCGAGCTCGAATTCGTTATTACGC
161 CCCGGGTACTTTCCAGTCGGGAAACGGGCAAC
162 CAGCTGGCGGACGACGACAGTATCGTAGCCAG
163 GTTTGAGGGAAAGGGGGATGTGCTAGAGGATC
164 CTTTCATCCCCAAAAACAGGAAGACCGGAGAG
165 AGAAAAGCAACATTAAATGTGAGCATCTGCCA
166 GGTAGCTAGGATAAAAATTTTAGTTAACATC
167 CAACGCAATTTTTGAGAGATCTACTGATAATC
168 CAATAAATACAGTTGATTCCCAATTTAGAGAG
169 TCCATATACATACAGGCAAGGCAACTTTATTT
170 TACCTTTAAGGTCTTTACCCTGACAAAGAAGT
171 CAAAAATCATTGCTCCTTTTGATAAGTTTCAT
172 TTTGCCAGATCAGTTGAGATTTAGTGGTTTAA
173 AAAGATTTCAGGGGTAATAGTAAACCATAAAT
174 TTTCAACTATAGGCTGGCTGACCTTGTATCAT
175 CCAGGCGCTTAATCATTGTGAATTACAGGTAG
176 CGCCTGATGGAAGTTTCCATTAAACATAACCG
177 TTTCATGAAAATTGTGTCGAAATCTGTACAGA
178 ATATATTCTTTTTTCACGTTGAAAATAGTTAG
179 AATAATAAGGTCGCTGAGGCTTGCAAAGACTT
180 CGTAACGATCTAAAGTTTTGTCGTGAATTGCG
181 ACCCAAATCAAGTTTTTTGGGGTCAAAGAACG
182 TGGACTCCCTTTTCACCAGTGAGACCTGTCGT
183 TGGTTTTTAACGTCAAAGGGCGAAGAACCATC
184 GCCAGCTGCCTGCAGGTCGACTCTGCAAGGCG

185 CTTGCATGCATTAATGAATCGGCCCCGCCAGGG
186 ATTAAGTTCGCATCGTAACCGTGCGAGTAACA
187 TAGATGGGGGGTAACGCCAGGGTTGTGCCAAG
188 ACCCGTCGTCATATGTACCCCGGTAAAGGCTA
189 CATGTCAAGATTCTCCGTGGGAACCGTTGGTG
190 TCAGGTCACTTTTGCGGGAGAAGCAGAATTAG
191 CTGTAATATTGCCTGAGAGTCTGGAAAAGTAG
192 CAAAATTAAAGTACGGTGTCTGGAAGAGGTCA
193 TGCAACTAAGCAATAAAGCCTCAGTTATGACC
194 TTTTGGCGCAGAAAACGAGAATGAATGTTTAG
195 AAACAGTTGATGGCTTAGAGCTTATTTAAATA
196 ACTGGATAACGGAACAACATTATTACCTTATG
197 ACGAACTAGCGTCCAATACTGCGGAATGCTTT
198 CGATTTTAGAGGACAGATGAACGGCGCGACCT
199 CTTTGAAAAGAACTGGCTCATTATTTAATAAA
200 GCTCCATGAGAGGCTTTGAGGACTAGGGAGTT
201 ACGGCTACTTACTTAGCCGGAACGCTGACCAA
202 AAAGGCCGAAAGGAACAATAAGCTTTCCAG
203 GAGAATAGCTTTTGGGGATCGTCGGGTAGCA
204 ACGTTAGTAAATGAATTTTCTGTAAGCGGAGT
205 TTTTCGATGGCCCACTACGTAAACCGTC
206 TATCAGGGTTTTCGGTTTGCCTATTGGGAACGCGCG
207 GGGAGAGGTTTTTGTAACGACGGCCATTCCCAGT
208 CACGACGTTTTTGTAATGGGATAGGTCAAAACGGCG
209 GATTGACCTTTTGATGAACGGTAATCGTAGCAAACA
210 AGAGAATCTTTTGGTTGTACCAAAAACAAGCATAAA
211 GCTAAATCTTTTCTGTAGCTCAACATGTATTGCTGA
212 ATATAATGTTTTATTGAATCCCCCTCAAATCGTCA
213 TAAATATTTTTTGAAGAAAAATCTACGACCAGTCA
214 GGACGTTGTTTTTCATAAGGGAACCGAAAGGCGCAG
215 ACGGTCAATTTTGACAGCATCGGAACGAACCCCTCAG
216 CAGCGAAATTTTAACTTTCAACAGTTTCTGGGATTTTGCTAAACTTTT

Modified staple strands

X54 TAAGCGTCGAAGGATTAGGATTAGTACCGCCATTATATCGCTCTAGAGTCGACCTGTTTTTTTTTT
X100 ATCCTTAACGGCCTGTTGCTGTTTTTATAAGTATAGCCCGGCCGTCGAG
Y52 CCTTAACGGCCTGTTGCTGAATTCCCTCAGAACCGCCACCCTCAGAACTGAGACT
Y132 TTTTTTTTTTTGTTGCTGAACCTTTGGTTTGATTTTTTGAGTTTCGTCACCAGTACAACTTAATTGTA
Z178 ATATATTCTTTTTTCACGTTGAAATTTAGCTAACAAAGACGCCTGCCATTTTTTTTTTT
Z202 AAAGGCCGAAAGGAACAATAAGCTTTCCAGTATCTGATCCTTAACGGCCTGTT

A78 GGAAAGCGACCAGGCGGATAAGTGTTAAAGGGCCTCGTGATACG
A100 ATATCGCTCTAGAGTCGTTTTTATAAGTATAGCCCGGCCGTCGAG
A111 ACCAGTAATAAAAGGGATTACCAGTCACACGTTTTTTAGTGAATTCGGACTACG
B52 CTTCTGATCCTTAACGGCCTTTCCCTCAGAACCGCCACCCTCAGAACTGAGACT
B73 GCCACGCTATACGTGGCACAGACAACGCTCATTTAAGCATGACCTTCCGTTGTTT
B132 TTAACGGCCTGTTGCTGAACCTATTGAGTTTCGTCACCAGTACAACTTAATTGTA
B159 AGCTGATTACAAGAGTCCACTATTGAGGTGCCTTCCTTCCGTTGTTTCAAGCGCA
C113 CCAGCAGGGGCAAAATCCCTTATAAAGCCGGCTTTTCGCTCTAGAGTCGACCTGGC
C132 ATCGCTCTAGAGTCGACCTGGTTTGAGTTTCGTCACCAGTACAACTTAATTGTA
C135 GAGTTGCACGAGATAGGGTTGAGTAAGGGAGCTTTCCTTCCGTTGTTTCAAGCGCA
C156 GACTACGTCTTCTGATCCTTTTTGTAGCATTCCACAGACAGCCCTCATCTCCAA
D52 GTTGCTGAACCTTTGGTTTGATTTTCCCTCAGAACCGCCACCCTCAGAACTGAGACT
D135 GAGTTGCACGAGATAGGGTTGAGTAAGGGAGCTTTTTCAAGCGCAGCCAGATT
D180 ACGGCCTGTTGCTGAACCTTTGTTTCGTAACGATCTAAAGTTTTGTCGTGAATTGCG
D97 CTAAAGCAAGATAGAACCCTTCTGAATCGTCTTTTTCCGTTGTTTCAAGCGCAGC
E205 CTAGAGTCGACCTGCGCGGATTTTTTCGATGGCCCACTACGTAAACCGTC
E216 AACTTTCAACAGTTTCTGGGATTTTGCTAAACTTTTTCTCCGGGAGCCAGTGAATTC

C113-10b CCAGCAGGGGCAAAATCCCTTATAAAGCCGGCTTATTATATCGCTCTAGAGTCGA
C132-10b CTAGAGTCGACCTGGCCAGTGTGTTGAGTTTCGTCACCAGTACAACTTAATTGTA
C135-10b GAGTTGCACGAGATAGGGTTGAGTAAGGGAGCTTAGCATGACCTTCCGTTGTTTC
C156-10b CGTCTTCTGATCCTTAACGGCTTTGTAGCATTCCACAGACAGCCCTCATCTCCAA
D1-10b ACGGCCTGTTGCTGAACCTTTGTTCAAGCCCAATAGGAACCCATGTACAAACAGTT
D76-10b TTCTGATCCTTAACGGCCTGTATTATCACCCTACTCAGGAGGTTTAGCGGGGTTT
D97-10b CTAAAGCAAGATAGAACCCTTCTGAATCGTCTTTTAAAGCATGACCTTCCGTTGT
D135-10b GAGTTGCACGAGATAGGGTTGAGTAAGGGAGCTAGACCTTCCGTTGTTTCAAGCG

E183-10b TGGTTTTTAACGTCAAAGGGCGAAGAACCATCTTTCGACCTGCGCGCGAGACGAA
E202-10b AAAGGCCGAAAGGAACAATAAGCTTTCCAGTTGCTTACAGACAAGCTGTGACC

M100 TATAAGTATAGCCCGGCCGTCGAG
M111 ACCAGTAATAAAAGGGATTACACAGTCACACGTTTT
M205 CGATGGCCCACTACGTAAACCGTC
M216 AACTTTCAACAGTTTCTGGGATTTTGCTAACTTTT

101-A18 AGGGTTGAATAAATCCTCATTAATGATATTCACAAACAACGCTCTCTCAAGTAGAAT
102-A18 AATCAGTAGCGACAGATCGATAGCAGCACCGTCGCTCTCTCAAGTAGAAT
103-A18 TAAAGGTGGCAACATAGTAGAAAATACATACACGCTCTCTCAAGTAGAAT
104-A18 GACGGGAGAATTAACCTACAGGGAAGCGCATTACGCTCTCTCAAGTAGAAT
105-A18 GCTTATCCGGTATTCTAAATCAGATATAGAAGCGCTCTCTCAAGTAGAAT
106-A18 CGACAAAAGGTAAAGTAGAGAATATAAAGTACCGCTCTCTCAAGTAGAAT
107-A18 CGCGAGAAAACCTTTTTATCGCAAGACAAAGAACGCTCTCTCAAGTAGAAT
108-A18 ATTAATTACATTTAACACATCAAGAAAACAAACGCTCTCTCAAGTAGAAT
109-A18 TTCATCAATATAATCCTATCAGATGATGGCAACGCTCTCTCAAGTAGAAT
110-A18 AATCAATATCTGGTCACAAATATCAAACCCTCCGCTCTCTCAAGTAGAAT

206-AC18 CGGTTTGCGTATTGGGAACGCGCGGGGAGAGGATTCTACTTGAGAGAGCG
207-AC18 TGTA AACGACGGCCATTCCCAGTCACGACGTATTCTACTTGAGAGAGCG
208-AC18 GTAATGGGATAGGTCAAACGGCGGATTGACCATTCTACTTGAGAGAGCG
209-AC18 GATGAACGGTAATCGTAGCAAACAAGAGAATCATTCTACTTGAGAGAGCG
210-AC18 GGTTGTACCAAAAACAAGCATAAAGCTAAATCATTCTACTTGAGAGAGCG
211-AC18 CTGTAGCTCAACATGTATTGCTGAATATAATGATTCTACTTGAGAGAGCG
212-AC18 CATTGAATCCCCCTCAAATCGTCATAAATATTATTCTACTTGAGAGAGCG
213-AC18 GGAAGAAAAATCTACGACCAGTCAGGACGTTGATTCTACTTGAGAGAGCG
214-AC18 TCATAAGGGAACCGAAAGGCGCAGACGGTCAAATTCTACTTGAGAGAGCG
215-AC18 GACAGCATCGGAACGAACCCTCAGCAGCGAAAATTCTACTTGAGAGAGCG

101A AGGGTTGAATAAATCCTCATTAATGATATTCACAAACAATTACTTGAGAGAGCG
102A AATCAGTAGCGACAGATCGATAGCAGCACCGTATTACTTGAGAGAGCG
103A TAAAGGTGGCAACATAGTAGAAAATACATACAATTACTTGAGAGAGCG
104A GACGGGAGAATTAACCTACAGGGAAGCGCATTAATTACTTGAGAGAGCG

105A GCTTATCCGGTATTCTAAATCAGATATAGAAGATTACTTGAGAGAGCG
 106A CGACAAAAGGTAAAGTAGAGAATATAAAGTACATTACTTGAGAGAGCG
 107A CGCGAGAAAACCTTTTATCGCAAGACAAAGAAATTACTTGAGAGAGCG
 108A ATTAATTACATTTAACACATCAAGAAAACAAAATTACTTGAGAGAGCG
 109A TTCATCAATATAATCCTATCAGATGATGGCAAATTACTTGAGAGAGCG
 110A AATCAATATCTGGTCACAAATATCAAACCCTCATTACTTGAGAGAGCG

 206AC CGGTTTGCGTATTGGGAACGCGCGGGGAGAGGCGTCTCTCAAGTAAT
 207AC TGTA AACGACGGCCATTCCCAGTCACGACGTCGCTCTCTCAAGTAAT
 208AC GTAATGGGATAGGTCAAAACGGCGGATTGACCCGCTCTCTCAAGTAAT
 209AC GATGAACGGTAATCGTAGCAAACAAGAGAATCCGCTCTCTCAAGTAAT
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 214AC TCATAAGGGAACCGAAAGGCGCAGACGGTCAACGCTCTCTCAAGTAAT
 215AC GACAGCATCGGAACGAACCCTCAGCAGCGAAACGCTCTCTCAAGTAAT

 101B AGGGTTGAATAAATCCTCATTAATGATATTCACAAACAAAGCGTCCATTGAGTTA
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 103B TAAAGGTGGCAACATAGTAGAAAATACATACAAGCGTCCATTGAGTTA
 104B GACGGGAGAATTA ACTACAGGGAAGCGCATTAAAGCGTCCATTGAGTTA
 105B GCTTATCCGGTATTCTAAATCAGATATAGAAGAGCGTCCATTGAGTTA
 106B CGACAAAAGGTAAAGTAGAGAATATAAAGTACAGCGTCCATTGAGTTA
 107B CGCGAGAAAACCTTTTATCGCAAGACAAAGAAAGCGTCCATTGAGTTA
 108B ATTAATTACATTTAACACATCAAGAAAACAAAAGCGTCCATTGAGTTA
 109B TTCATCAATATAATCCTATCAGATGATGGCAAAGCGTCCATTGAGTTA
 110B AATCAATATCTGGTCACAAATATCAAACCCTCAGCGTCCATTGAGTTA

 206BC CGGTTTGCGTATTGGGAACGCGCGGGGAGAGGTA ACTCAATGGACGCT
 207BC TGTA AACGACGGCCATTCCCAGTCACGACGTTA ACTCAATGGACGCT
 208BC GTAATGGGATAGGTCAAAACGGCGGATTGACCTAACTCAATGGACGCT
 209BC GATGAACGGTAATCGTAGCAAACAAGAGAATCTAACTCAATGGACGCT
 210BC GGTTGTACCAAAAACAAGCATAAAGCTAAATCTAACTCAATGGACGCT

211BC CTGTAGCTCAACATGTATTGCTGAATATAATGTAACCTCAATGGACGCT
 212BC CATTGAATCCCCCTCAAATCGTCATAAATATTTAACTCAATGGACGCT
 213BC GGAAGAAAAATCTACGACCAGTCAGGACGTTGTAACCTCAATGGACGCT
 214BC TCATAAGGGAACCGAAAGGCGCAGACGGTCAATAACTCAATGGACGCT
 215BC GACAGCATCGGAACGAACCCTCAGCAGCGAAATAACTCAATGGACGCT

 101C AGGGTTGAATAAATCCTCATTAATGATATTCACAAACAACACCACAAGACCACTC
 102C AATCAGTAGCGACAGATCGATAGCAGCACCGTCACCACAAGACCACTC
 103C TAAAGGTGGCAACATAGTAGAAAATACATACACACCACAAGACCACTC
 104C GACGGGAGAATTAACCTACAGGGAAGCGCATTACACCACAAGACCACTC
 105C GCTTATCCGGTATTCTAAATCAGATATAGAAGCACCACAAGACCACTC
 106C CGACAAAAGGTAAAGTAGAGAATATAAAGTACCACCACAAGACCACTC
 107C CGCGAGAAAACCTTTTATCGCAAGACAAAGAACACCACAAGACCACTC
 108C ATTAATTACATTTAACACATCAAGAAAACAACACCACAAGACCACTC
 109C TTCATCAATATAATCCTATCAGATGATGGCAACACCACAAGACCACTC
 110C AATCAATATCTGGTCACAAATATCAAACCCTCCACCACAAGACCACTC

 206CC CGGTTTGCGTATTGGGAACGCGCGGGGAGAGGGAGTGGTCTTGTGGTG
 207CC TGTA AACGACGGCCATTCCCAGTCACGACGTGAGTGGTCTTGTGGTG
 208CC GTAATGGGATAGGTCAAAACGGCGGATTGACCGAGTGGTCTTGTGGTG
 209CC GATGAACGGTAATCGTAGCAAACAAGAGAATCGAGTGGTCTTGTGGTG
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 213CC GGAAGAAAAATCTACGACCAGTCAGGACGTTGGAGTGGTCTTGTGGTG
 214CC TCATAAGGGAACCGAAAGGCGCAGACGGTCAAGAGTGGTCTTGTGGTG
 215CC GACAGCATCGGAACGAACCCTCAGCAGCGAAAGAGTGGTCTTGTGGTG

 101D AGGGTTGAATAAATCCTCATTAATGATATTCACAAACAATTACTACTGAGACATT
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206DC CGGTTTGCGTATTGGGAACGCGCGGGGAGAGGAATGTCTCAGTAGTAA
207DC TGTA AACGACGGCCATTCCCAGTCACGACGTAATGTCTCAGTAGTAA
208DC GTAATGGGATAGGTCAAAACGGCGGATTGACCAATGTCTCAGTAGTAA
209DC GATGAACGGTAATCGTAGCAAACAAGAGAATCAATGTCTCAGTAGTAA
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211DC CTGTAGCTCAACATGTATTGCTGAATATAATGAATGTCTCAGTAGTAA
212DC CATTGAATCCCCCTCAAATCGTCATAAATATTAATGTCTCAGTAGTAA
213DC GGAAGAAAAATCTACGACCAGTCAGGACGTTGAATGTCTCAGTAGTAA
214DC TCATAAGGGAACCGAAAGGCGCAGACGGTCAAAATGTCTCAGTAGTAA
215DC GACAGCATCGGAACGAACCCTCAGCAGCGAAAAATGTCTCAGTAGTAA

102A10(1) AATCAGTAGCGACAGATCGATAGCAGCACCGTATTACTTGAG
104A10(2) GACGGGAGAATTA ACTACAGGGAAGCGCATTACGCTGCTGAG
106A10(3) CGACAAAAGGTAAAGTAGAGAATATAAAGTACGATCAGTCGG
108A10(4) ATTAATTACATTTAACACATCAAGAAAACAAACGTACGTCCA
110A10(5) AATCAATATCTGGTCACAAATATCAAACCCTCCAGAAAGCAT

214AC10(1) TCATAAGGGAACCGAAAGGCGCAGACGGTCAACTCAAGTAAT
212AC10(2) CATTGAATCCCCCTCAAATCGTCATAAATATTCTCAGCAGCG
210AC10(3) GGTTGTACCAAAAACAAGCATAAAGCTAAATCCCGACTGATC
208AC10(4) GTAATGGGATAGGTCAAAACGGCGGATTGACCTGGACGTACG
206AC10(5) CGGTTTGCGTATTGGGAACGCGCGGGGAGAGGATGCTTTCTG

102B10(1) AATCAGTAGCGACAGATCGATAGCAGCACCGTAGCGTCCATT
104B10(2) GACGGGAGAATTA ACTACAGGGAAGCGCATTACTTCGGCGGAG
106B10(3) CGACAAAAGGTAAAGTAGAGAATATAAAGTACAGTATCTTAT
108B10(4) ATTAATTACATTTAACACATCAAGAAAACAAAGAACCTCATC
110B10(5) AATCAATATCTGGTCACAAATATCAAACCCTCGTTCGTACGC

214BC10(1) TCATAAGGGAACCGAAAGGCGCAGACGGTCAAAATGGACGCT
212BC10(2) CATTGAATCCCCCTCAAATCGTCATAAAATATTCTCGCCGAAG
210BC10(3) GGTTGTACCAAAAAACAAGCATAAAGCTAAATCATAAGATACT
208BC10(4) GTAATGGGATAGGTCAAAACGGCGGATTGACCGATGAGGTTC
206BC10(5) CGGTTTGCGTATTGGGAACGCGCGGGGAGAGGGCGTACGAAC

102C10(1) AATCAGTAGCGACAGATCGATAGCAGCACCGTCACCACAAGA
104C10(2) GACGGGAGAATTAACACTACAGGGAAGCGCATTACTATGTCTCC
106C10(3) CGACAAAAGGTAAAGTAGAGAATATAAAGTACGACAACGAGT
108C10(4) ATTAATTACATTTAACACATCAAGAAAACAAAGTAAGCCATC
110C10(5) AATCAATATCTGGTCACAAATATCAAACCCTCTGGCACGTCA

214CC10(1) TCATAAGGGAACCGAAAGGCGCAGACGGTCAATCTTGTGGTG
212CC10(2) CATTGAATCCCCCTCAAATCGTCATAAAATATTGGAGACATAG
210CC10(3) GGTTGTACCAAAAAACAAGCATAAAGCTAAATCACTCGTTGTC
208CC10(4) GTAATGGGATAGGTCAAAACGGCGGATTGACCGATGGCTTAC
206CC10(5) CGGTTTGCGTATTGGGAACGCGCGGGGAGAGGTGACGTGCCA

102D10(1) AATCAGTAGCGACAGATCGATAGCAGCACCGTTTACTACTGA
104D10(2) GACGGGAGAATTAACACTACAGGGAAGCGCATTAAGTCTGCCGA
106D10(3) CGACAAAAGGTAAAGTAGAGAATATAAAGTACCACATTTCGAG
108D10(4) ATTAATTACATTTAACACATCAAGAAAACAAATAGTATTCCA
110D10(5) AATCAATATCTGGTCACAAATATCAAACCCTCCTTGGCACAT

214DC10(1) TCATAAGGGAACCGAAAGGCGCAGACGGTCAATCAGTAGTAA
212DC10(2) CATTGAATCCCCCTCAAATCGTCATAAAATATTTCCGCAGACT
210DC10(3) GGTTGTACCAAAAAACAAGCATAAAGCTAAATCCTCGAATGTG
208DC10(4) GTAATGGGATAGGTCAAAACGGCGGATTGACCTGGAATACTA
206DC10(5) CGGTTTGCGTATTGGGAACGCGCGGGGAGAGGATGTGCCAAG

S102 AATCAGTAGCGACAGATCGATAGC
S103 TAAAGGTGGCAACATAGTAGAAAA
S104 GACGGGAGAATTAACACTACAGGGAA
S105 GCTTATCCGGTATTCTAAATCAGA

S106 CGACAAAAGGTAAAGTAGAGAATA
 S107 CGCGAGAAAACCTTTTATCGCAAG
 S108 ATTAATTACATTTAACACATCAAG
 S109 TTCATCAATATAATCCTATCAGAT
 S110 AATCAATATCTGGTCACAAATATC
 S111 ACCAGTAATAAAAGGGATTACACCAGTCACACG

 S207 TGTAACGACGGCCATTCCCAGT
 S208 GTAATGGGATAGGTCAAAACGGCG
 S209 GATGAACGGTAATCGTAGCAAACA
 S210 GGTTGTACCAAAAACAAGCATAAA
 S211 CTGTAGCTCAACATGTATTGCTGA
 S212 CATTGAATCCCCCTCAAATCGTCA
 S213 GGAAGAAAAATCTACGACCAGTCA
 S214 TCATAAGGGAACCGAAAGGCGCAG
 S215 GACAGCATCGGAACGAACCCTCAG
 S216 AACTTTCAACAGTTTCTGGGATTTTGCTAAACTTTT

 H134 GAATAGCCGCAAGCGTCTCTTTGAGGAACAAGTTTCTTGTCCACGCTCCTAATGA
 H136 GTGAGCTAGTTTCTGTCTCTTTGAGGAACAAGTTTCTTGTGTGAAATTTGGGAAG
 H137 TCATAGCTACTCACATTCCTCTTTGAGGAACAAGTTTCTTGTGAATTGCGCCCTGAGA
 H139 GAAGATCGGTGCGGGCTCTCTTTGAGGAACAAGTTTCTTGTCTCTTCGCAATCATGG
 H158 AGTTTGAGCCCTTCATCCTCTTTGAGGAACAAGTTTCTTGTCCGCCTGGTTGCGCTC
 H160 ACTGCCCGCCGAGCTCTCCTCTTTGAGGAACAAGTTTCTTGTGAATTCGTTATTACGC

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