

Gene-encoding DNA origami for mammalian cell expression

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1. Materials and Methods

1.1 Scaffold cloning

Plasmids encoding for custom scaffolds were created via standard cloning techniques. All plasmids, with the exception of sc_EGFP3, were created via Golden gate assembly using either Esp3I (NEB cat. no. R0734), or BsaI-HF@v2 (NEB cat. no. R3733), together with T4 DNA ligase (NEB cat. no. M0202). For each plasmid, appropriate cut sites were introduced with PCR, and the assembly was conducted as per manufacturer's protocol.

The exception, sc_EGFP3, was assembled via digestion ligation, and compatible enzyme cut sites were added to the fragment of interest by means of PCR. Enzymes EcoNI and PacI (NEB cat. no. R0521 and R0547 respectively) were used to digest fragments of interest, and then ligated using NEB T4 DNA ligase, as above.

Touchdown PCR with primers for all constructs are detailed in Table S1. In all cases, PCR products were confirmed by agarose gel electrophoresis (AGE), bands were excised, and fragments were extracted (Qiagen QIAquick Gel Extraction Kit) as per manufacturer's protocol.

Table S1 | Primer sequences used for the construction of plasmids for custom scaffold production.

Plasmid	Template	Primer	Sequence 5' - 3'
sc_EGFP1	Addgene plasmid #126854	FWD	CTGGATGGTCTCCgtgacattaagcgcggcggggtg
		REV	CTGGATGGTCTCCaatgagtgagcaaaaggccagca
	Addgene plasmid #13031	FWD	CTGGATGGTCTCCcattgatatacgcgttgacattga
		REV	CTGGATGGTCTCCtcacattccgcctcagaagccat
sc_EGFP2	Addgene plasmid #126854	FWD	CACTGACGTCTCTgtgacattaagcgcggcggggtg
		REV	CACTGACGTCTCTaatgagtgagcaaaaggccagca
	Addgene plasmid #13031	FWD	CACTGACGTCTCTcattgcctcagaagccataga
		REV	CACTGACGTCTCTtcacagatatacgcggttgacattga
sc_EGFP3	Addgene plasmid #105530	FWD	TTAATTAACattaagcgcggcggggtg
		REV	CCTAATTAAGGgtgagcaaaaggccagcaaa
sc_EGFP4	sc_EGFP3	FWD	CACTGACGTCTCTcattatgctctaggaagatcgga
		REV	CGTGATCGTCTCTaatgtaagggtgagcaaaaggcca
sc_EGFP5	sc_EGFP3	FWD	CACTGACGTCTCTcattgccttaattaacattaagcgc
		REV	CACTGACGTCTCTaatgcgccatgctacttatctacg
sc_EGFP6	sc_EGFP3	FWD	AGATGGCGTCTCCttagggcctcagtgagcgcgagcg
		REV	AGATGGCGTCTCCaggggcctcagtgagcgcgagcg
		FWD	AGATGGCGTCTCCcccttaattaacattaagcgcg
		REV	AGATGGCGTCTCCcctaatTAAGGgtgagcaaaagg
sc_mCherry5	sc_EGFP5	FWD	TGGCTACGTCTCGgtgattgatccaatcaacctctg
		REV	TGGCTACGTCTCGaatgcctgacacctgtggaga
	Addgene plasmid #127813	FWD	TGGCTACGTCTCGcattattcgccacctggtgag
		REV	TGGCTACGTCTCGtaccgccctcactgtacagct

1.2 Scaffold production

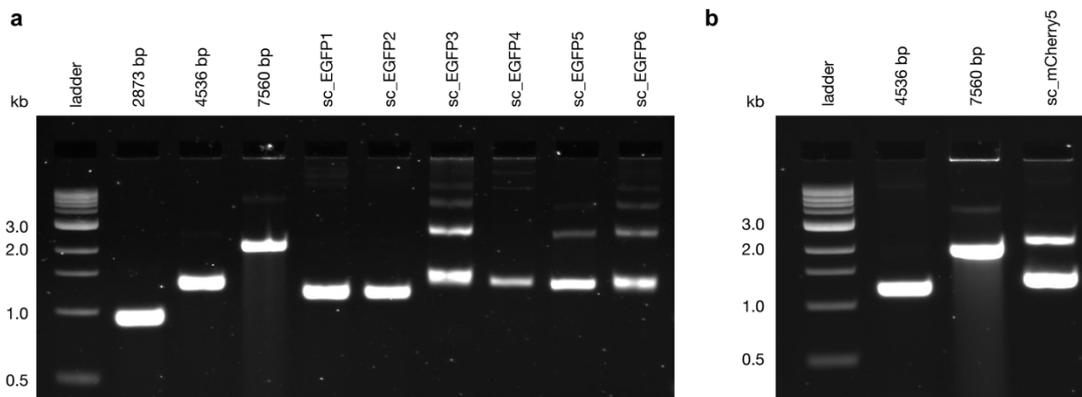


Fig. S1 | AGE showing all custom scaffolds created in this study. ssDNA markers at 2873 bp, 4535 bp and 7560 bp were used as molecular weight markers, while 'ladder' depicts NEB 1 kb dsDNA ladder.

1.3 DNA origami folding

Table S2 | Folding conditions summary for each of the structures and scaffolds used.

Scaffold	Structure	Program	10x Folding Buffer	v(scaffold, 100 nM), μL^*	v(staples, 100 μM), μL^*
sc_EGFP1	20HB-ext	3	FoB10	10	8
	20HB-int	1	FoB10	10	8
	20HB-ext-W	1	FoB15	10	8
	20HB-int-W	1	FoB15	10	8
	32HB	1	FoB20	10	8
	12HB	2	FoB10	10	8
	20HB LS	3	FoB15	10	8
	20HB LP	1	FoB20	10	8
	20HB LPv2	1	FoB20	10	8
sc_EGFP2	20HB Circ	3	FoB10	10	8
	20HB	1	FoB10	10	8
	20HB LPv2	3	FoB15	10	8
sc_EGFP3	20HB	3	FoB10	10	8
	20HB	5	FoB20	2.5	8 [‡]
	20HB	3	FoB15	10	8
sc_EGFP4	20HB	2	FoB15	10	8
	20HB	3	FoB15	4	14
	20HB loop	5	FoB10	4	8
	20HB loop LPv2	4	FoB15	4	8
	16HB_2	7	FoB12.5	10	8
	16HB_3	7	FoB25	10	8
	16HB_4	7	FoB7.5	10	8
sc_EGFP5	20HB	2	FoB15	4	14
	20HB LPv2	6	FoB10	4	8
sc_EGFP6	20HB	1	FoB10	10	8
	16HB_1	1	FoB10	10	8

*For 20 μL folding reaction, where necessary volume supplemented to 20 μL total with ddH₂O.

‡Staples used here were at 500 μM concentration.

Table S3 | Folding programs used for the folding reactions detailed in Table S2.

Program	1. Denaturation time		2. Temperature ramp	3. Storage temperature
	30 s	15 min		
1	-	65 °C	60–44 °C, at 1 °C/1 h	20 °C
2	-	65 °C	60–44 °C, at 1 °C/2 h	20 °C
3	70 °C	65 °C	60–35 °C, at 1 °C/1 h	20 °C
4	70 °C	65 °C	60–35 °C, at 1 °C/2 h	20 °C
5	70 °C	65 °C	60–35 °C, at 1 °C/4 h	20 °C
6	70 °C	65 °C	60–30 °C, at 1 °C/1 h	20 °C
7	-	65 °C	60–44 °C, at 1 °C/4 h	20 °C

2. Supplemental Data

2.1 Characterisation and supporting information

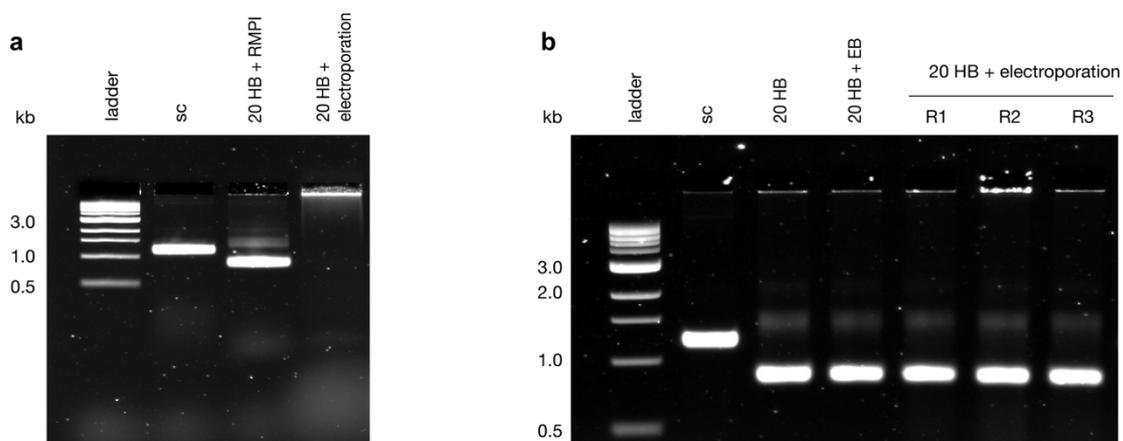


Fig. S2 | 20 HB structure integrity after electroporation. **a**, Structural integrity of the 20HB was maintained when diluted in RPMI 1640 media but demonstrated complete aggregation after electroporation in a traditional cuvette. **b**, 20HBs were stable in both the electroporation buffer (EB, buffer R in kit) and after electroporation using the Neon™ transfection system. R1-R3 depicts 3 independent replicates. 'Sc' is the corresponding sc_EGFP1 ssDNA scaffold strand, while 'ladder' depicts NEB 1 kb dsDNA ladder.

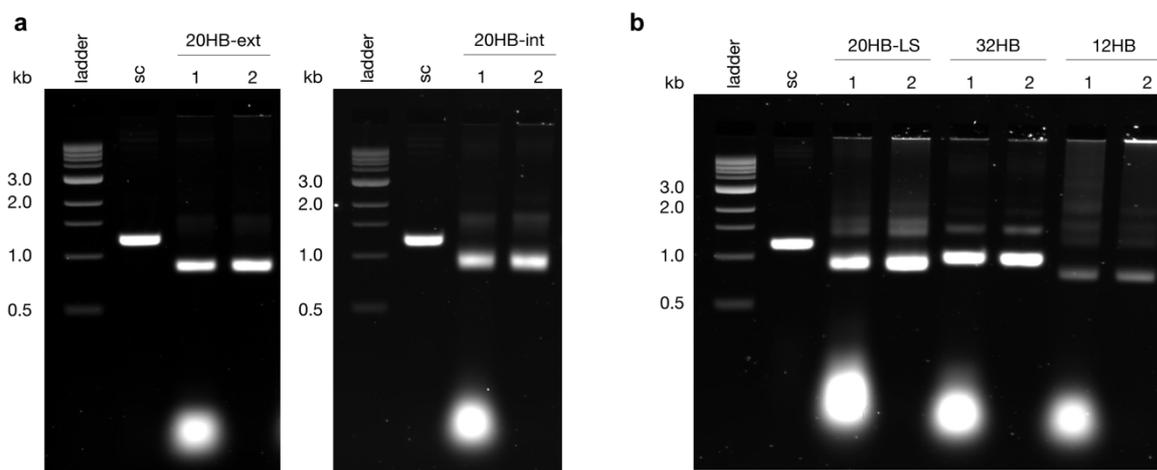


Fig. S3 | Purified DNA origami objects based on sc_EGFP1 scaffold. **a**, Crude and PEG-purified DNA origami objects are given in lane 1 and 2 for 20HB-ext and 20HB-int respectively. **b**, Crude and PEG-purified DNA origami objects are given in lane 1 and 2 for 20HB-LS, 32HB and 12HB respectively. The corresponding sc_EGFP1 ssDNA scaffold strand is given by 'sc', while 'ladder' depicts NEB 1 kb dsDNA ladder.

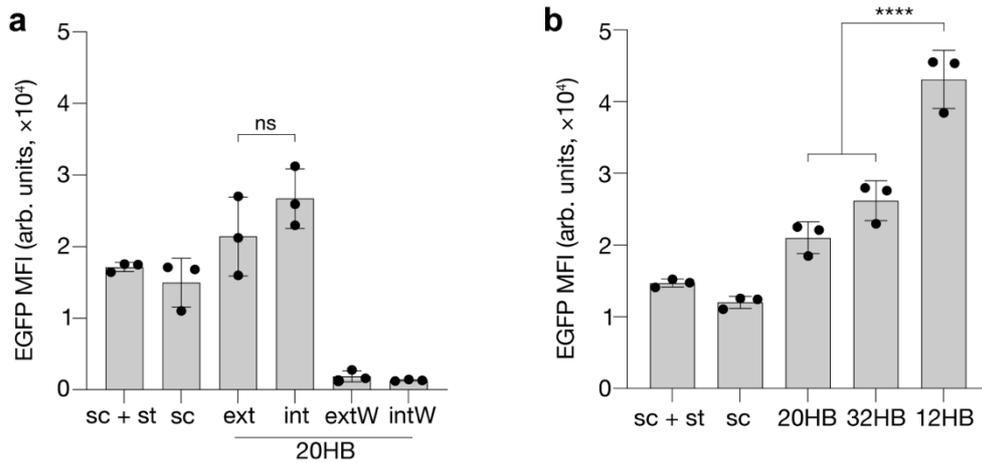


Fig. S4 | Mean fluorescence intensity of EGFP depending on gene orientation. **a**, HEK293T cells transfected with either 20HB-ext or 20HB-int demonstrated no significant difference in the EGFP MFI. **b**, HEK293T cells transfected with either 20HB(-ext) or 32HB demonstrated no statistically significant difference in EGFP MFI. Transfection with 12HB demonstrated significantly higher EGFP MFI compared to both 20HB(-ext) and 32HB. Data was collected quantified using flow cytometry and are presented as mean \pm s.d. for $n = 3$ biologically independent experiments, individual data points are overlaid. Statistical analysis in was performed using one-way ANOVA with Tukey's multiple comparison (**** $p \leq 0.0001$, ns $p > 0.05$).

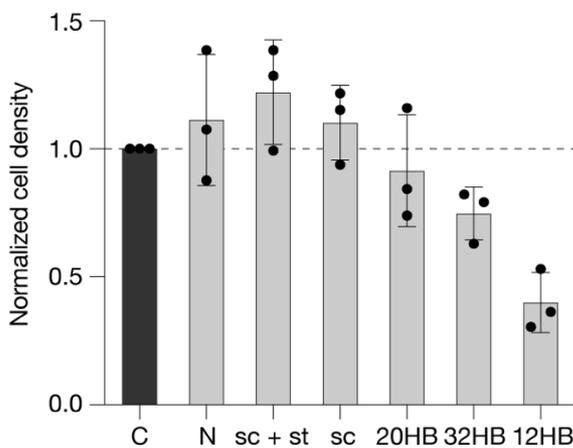


Fig. S5 | Cell density after electroporation of HEK293T cells. Single cell density was counted via AttuneNxt Flow Cytometer 48 h after electroporation. 'C' represents untreated control, 'N' represents control electroporated with blank buffer. Cell densities were normalized against control C. The data is presented as mean \pm s.d. with individual data points overlaid, the data given is from $n = 3$ biologically independent experiments.

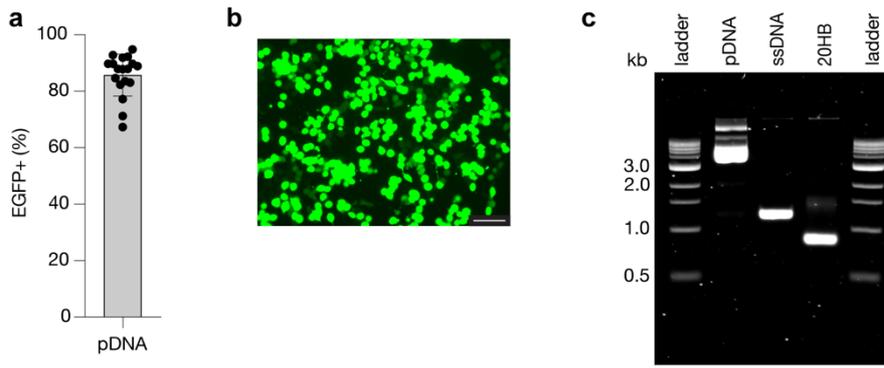


Fig. S6 | Electroporation in HEK293T cells using plasmid EGFP-encoding plasmid DNA (pDNA). **a**, Transfection efficiency in HEK293T cells, with representative epifluorescent image given in **b**. The data is presented as mean \pm s.d. with individual data points overlaid, the data given is from $n = 18$ biologically independent experiments, where similar results were observed each time. Scale bar is $100 \mu\text{m}$. Plasmid DNA was used as a positive control, however cannot be directly compared to the electroporation of ssDNA and folded DNA origami objects due to differences in electrophoretic mobility, as demonstrated in **c**. Plasmid DNA is given as pDNA, and the corresponding EGFP-encoding ssDNA scaffold strand and 20HB origami object is given as ssDNA and 20HB respectively, while 'ladder' depicts NEB 1 kb dsDNA ladder.

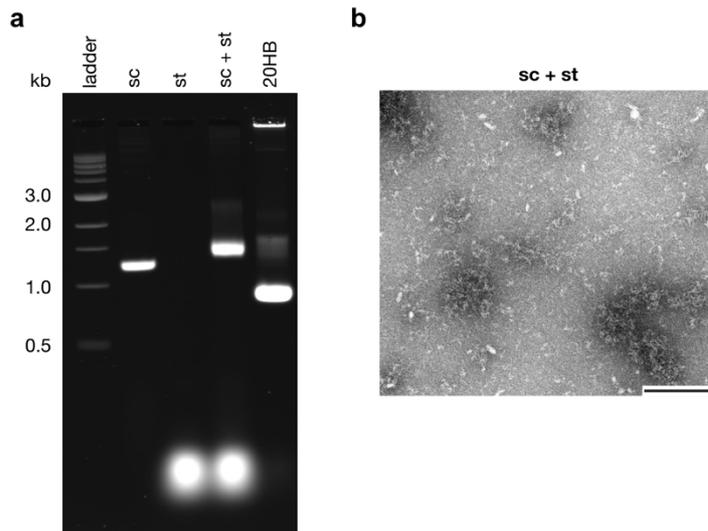


Fig. S7 | Staples associate with scaffold after mixing. **a**, Staples and scaffold were mixed ('sc + st') and incubated at rt for 10 min, while 20HB represents the corresponding purified folded object. A slight shift in the 'sc + st' band compared to scaffold only ('sc') demonstrates some staple association has occurred, but no structure has folded. 'st' is the corresponding mixture of staples, and 'ladder' depicts NEB 1 kb dsDNA ladder. **b**, Representative negative stain TEM image of the corresponding sc + st mixture. Scale bar 100 nm .

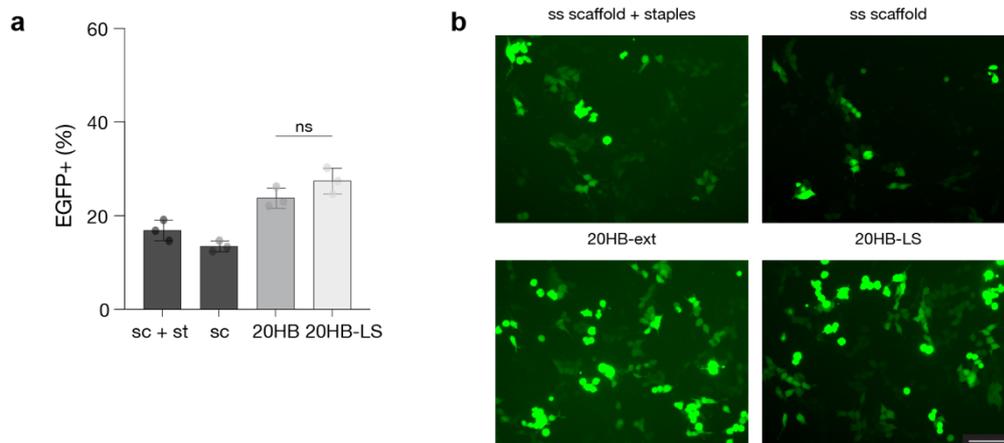


Fig. S8 | Electroporation of 20HB designs with different staple routings. 20HB with typical staple routings, as tested previously (20HB-ext, average staple length of ~ 42 bases) was compared to a 20HB design with longer staples and fewer staple crossovers (20HB-LS, average staple length of ~ 57 bases). **a**, Quantification of EGFP positive cells by flow cytometry is given in **a**. The data is presented as mean \pm s.d. with individual data points overlaid. One-way ANOVA with Tukey's multiple comparison was performed to test statistically significant differences between samples. Both 20HB-ext and 20HB-LS demonstrated statistically significant increases in EGFP expression compared to ss scaffold + staples ($p < 0.05$, $p < 0.01$ respectively) and ss scaffold only ($p < 0.01$, $p < 0.001$ respectively). No statistically significant difference was observed between 20HB-ext and 20HB-LS (ns). Representative epifluorescent microscopy images demonstrating EGFP expression are given in **b**. The data in both **a** and **b** is representative of $n = 3$ biologically independent experiments, where similar results were observed each time. Scale bar is 100 μ m.

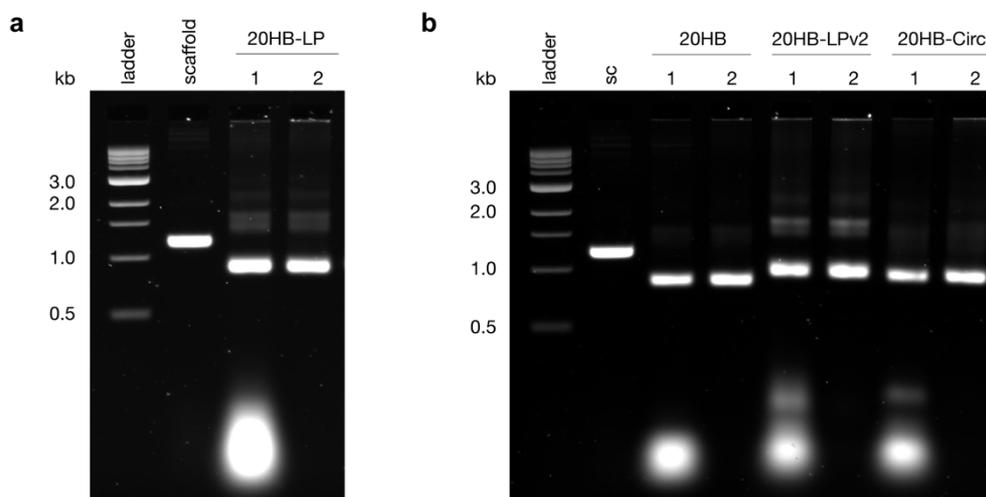


Fig. S9 | Purified DNA origami objects based on sc_EGFP1 scaffold with alternative staple designs. **a**, Crude and PEG-purified DNA origami objects are given in lane 1 and 2, respectively, for 20HB-LP. **b**, Crude and PEG-purified DNA origami objects are given in lane 1 and 2, respectively, for 20HB, 20HB-LPv2 and 20HB-Circ. The corresponding sc_EGFP1 ssDNA scaffold strand is given by 'sc', while 'ladder' depicts NEB 1 kb dsDNA ladder.

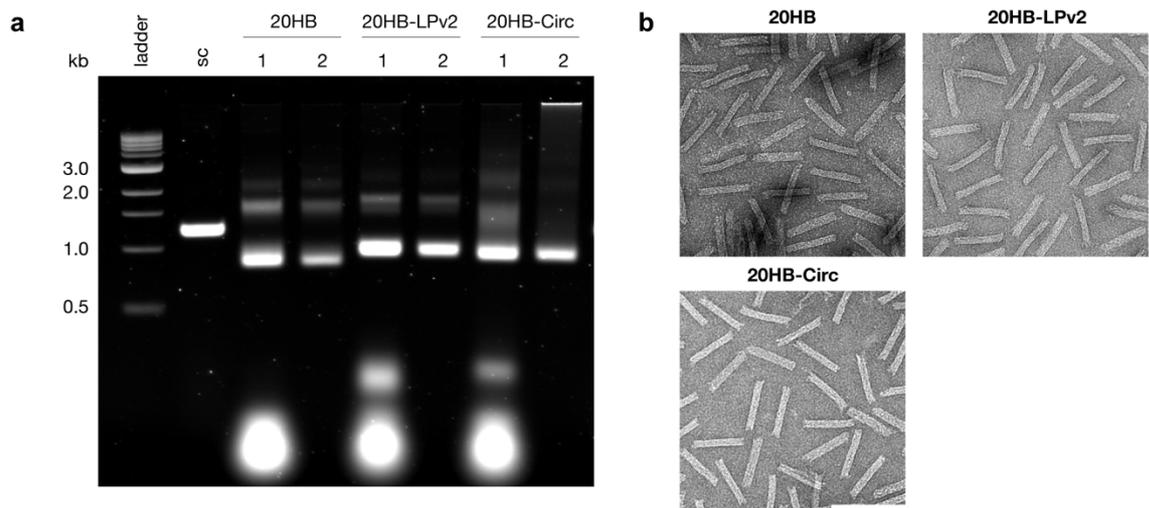


Fig. S10 | Purified DNA origami objects based on sc_EGFP2 scaffold. **a**, Crude and PEG-purified DNA origami objects are given in lane 1 and 2 for 20HB, 20HB-LPv2 and 20HB-Circ. The corresponding sc_EGFP2 ssDNA scaffold strand is given by 'sc', while 'ladder' depicts NEB 1 kb dsDNA ladder. **b**, Representative negative stain TEM images of the corresponding purified DNA origami objects. Scale bar 100 nm.

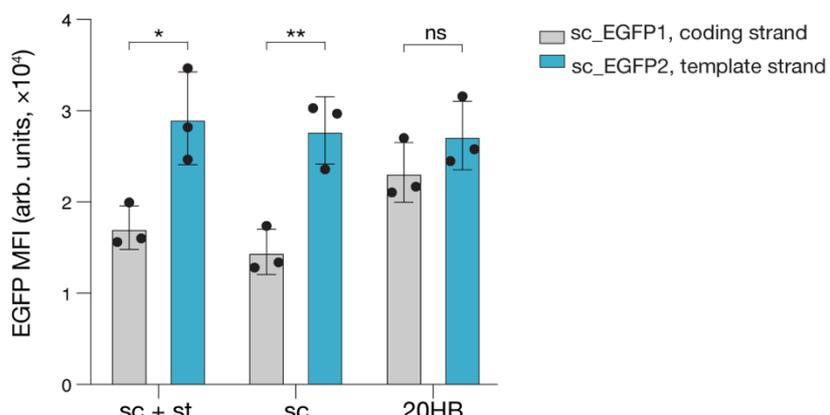


Fig. S11 | Mean fluorescence intensity of EGFP depending on scaffold orientation. HEK293T cells transfected with either sc_EGFP2 scaffold + staple mixture, or scaffold only, demonstrated significantly higher EGFP MFI than that of the sc_EGFP1 counterpart. No significant difference in the transfection efficiency from 20HB structures was observed. Data was collected quantified using flow cytometry and are presented as mean \pm s.d. for $n = 3$ biologically independent experiments, individual data points are overlaid. Statistical analysis in was performed using Student's t-test (* $p \leq 0.05$, ** $p \leq 0.01$, *** $p \leq 0.001$, ns $p > 0.05$).

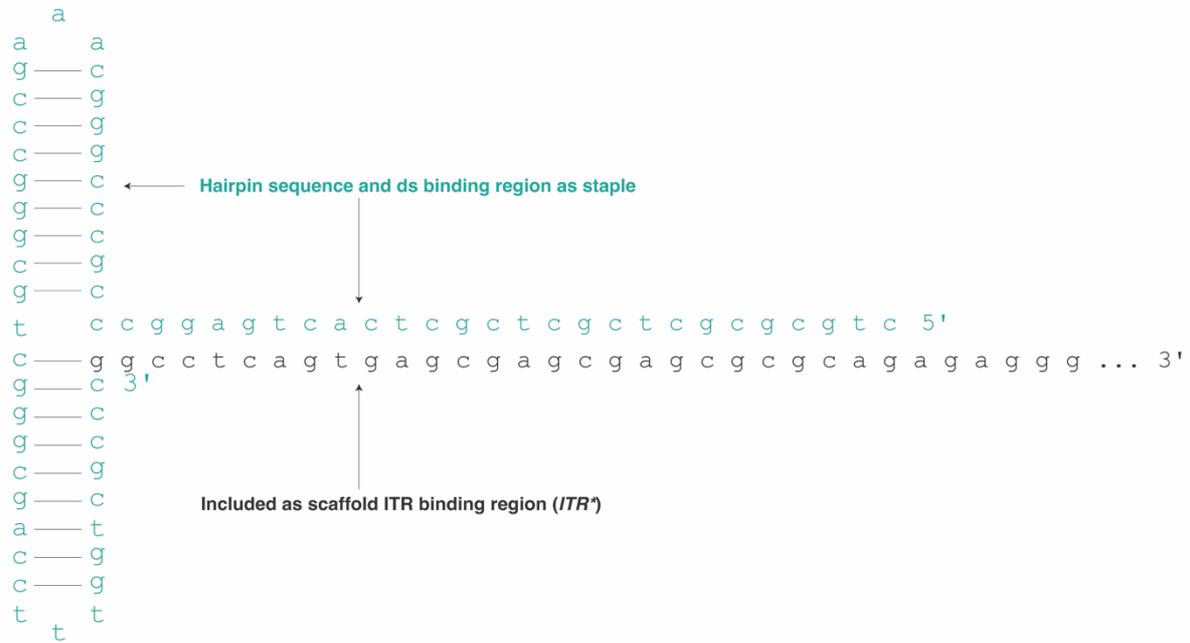


Fig. S12 | Exemplary schematic of ITR design for sc_EGFP6 scaffold. ITR hairpin and complementary binding sequence (green) was removed from the scaffold sequence, to be included as a separate staple. The sequence remaining in the scaffold (black) is given as the ITR binding region (*ITR**).

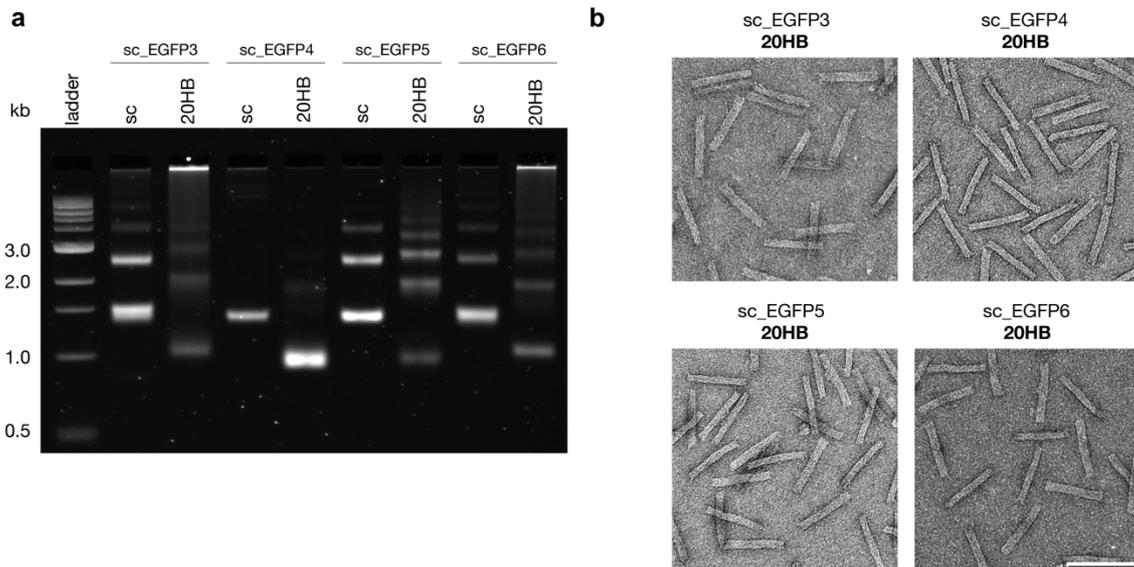


Fig. S13 | Purified DNA origami objects based on alternative scaffolds sc_EGFP3, sc_EGFP4, sc_EGFP5 and sc_EGFP6. **a**, Gel-purified origami objects. For each scaffold, the first lane ('sc') depicts the scaffold only MW control, while the second lane depicts the folded 20HB object. Higher order bands can be seen some of the scaffold samples, and in folded origami samples. In **a** 'ladder' depicts NEB 1 kb dsDNA ladder. **b**, Representative negative stain TEM images corresponding to each of the samples. Scale bar is 100 nm.

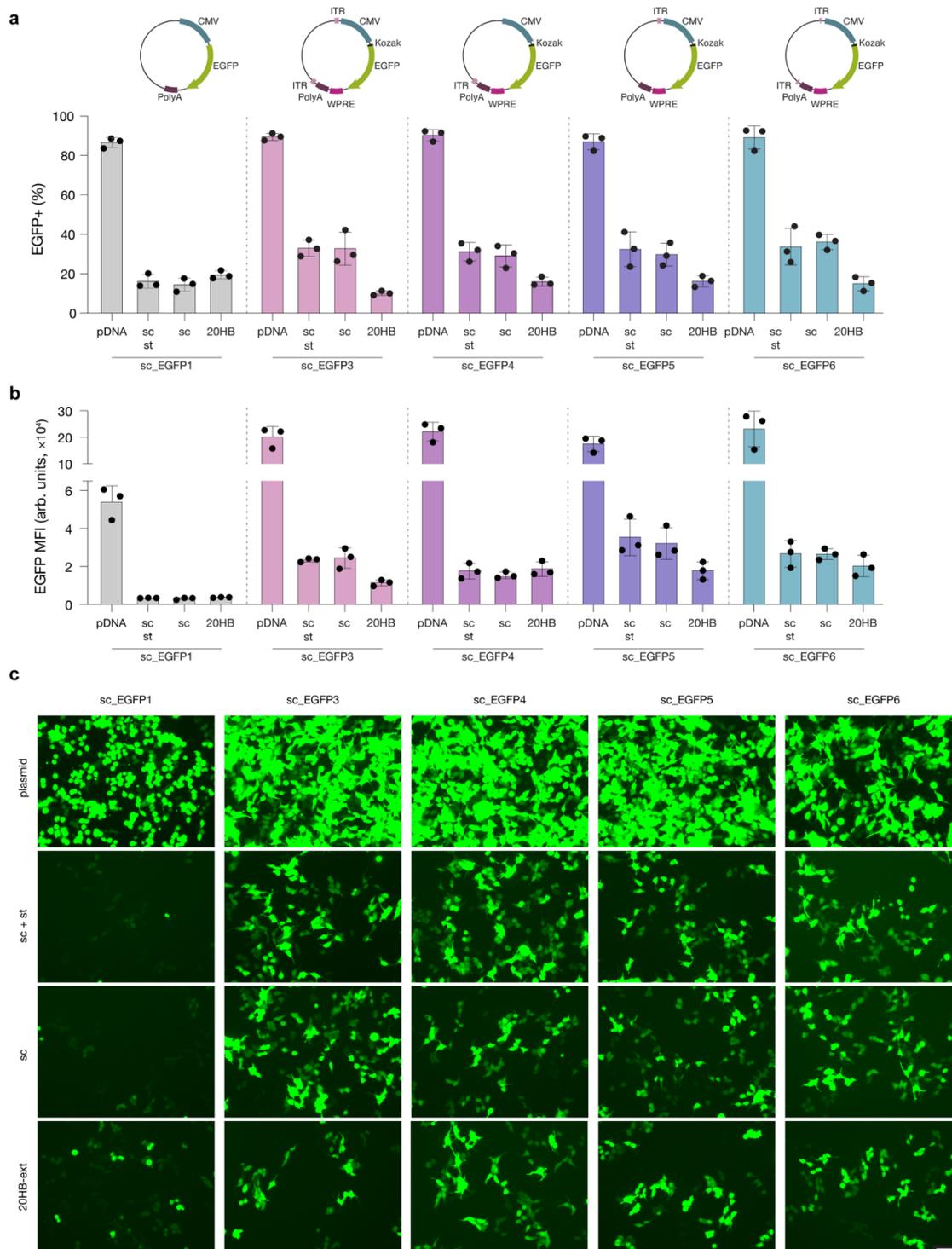


Fig. S14 | Screening expression from different scaffold sequence designs. To improve expression of ssDNA several sequence features, encoding for chimeric intron, kozak, WPRE, and ITRs were incorporated in the scaffold. Scaffolds with several ITR sequence derivatives were also investigated. Each ss scaffold DNA was expressed and folded into a 20HB. **a**, Plasmid/scaffold design schematics, CMV promoter (pink), kozak sequence (black), EGFP (green), WPRE (yellow), polyA (blue) and ITRs (purple) are shown. The ITR components are present in full at both upstream and downstream of the expression cassette (sc_EGFP3), only downstream (sc_EGFP4) and only upstream (sc_EGFP5). The final design (sc_EGFP6) has been constructed so that the only D-D' and A-A' ITR sequences are present on the scaffold strand, and the hairpin B-B', C-C', and corresponding A-A' sequence was supplied on staples strands. Plasmid, ss scaffold + staples (sc + st), ss scaffold (sc) and 20HB origami with the various sequences were electroporated into HEK293T cells and quantified by flow cytometry, looking at population of EGFP positive cells **a**, and mean fluorescent intensity (arb. units) **b**. Data is presented as mean \pm s.d. with

individual data points overlaid. Representative epifluorescent microscopy images for each of the conditions is given in **c**. All data **a–c** is representative of $n = 3$ biologically independent experiments, where similar results were observed each time. Scale bar is $100 \mu\text{m}$.

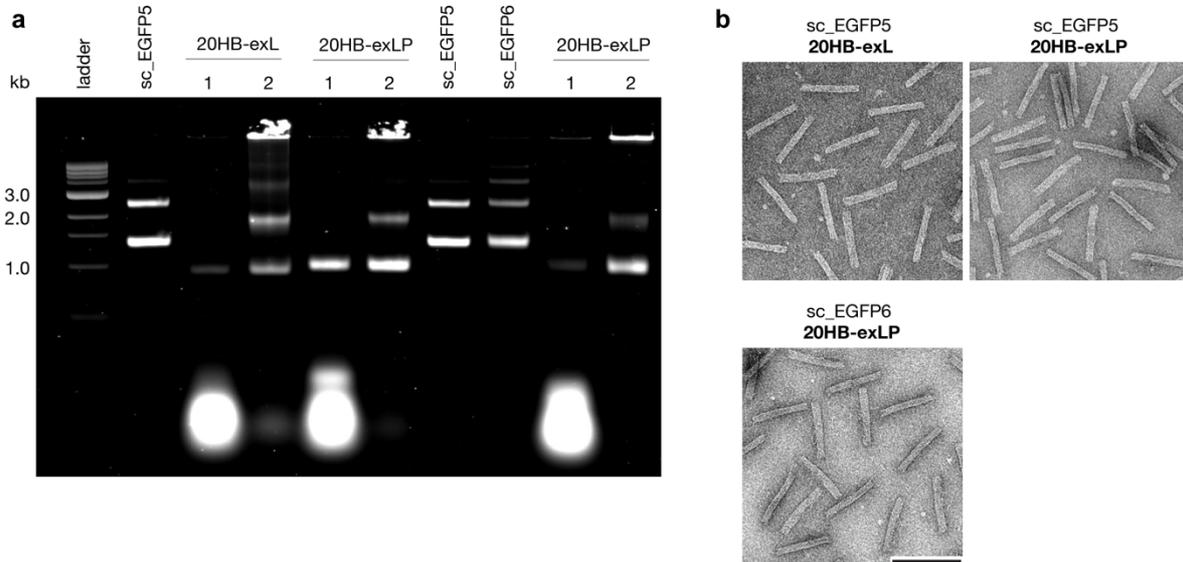


Fig. S15 | Purified DNA origami objects based on alternative scaffolds sc_EGFP5 and sc_EGFP6 with external loops and long promoter staple. **a**, Crude and PEG-purified DNA origami objects are given in lane 1 and 2 respectively for 20HB-exL, 20HB-exLP (sc_EGFP5) and 20HB-exLP (sc_EGFP6). Respective ssDNA scaffolds are included as MW controls, while 'ladder' depicts NEB 1 kb dsDNA ladder. **b**, Representative negative stain TEM images corresponding to each of the samples. Scale bar is 100 nm .

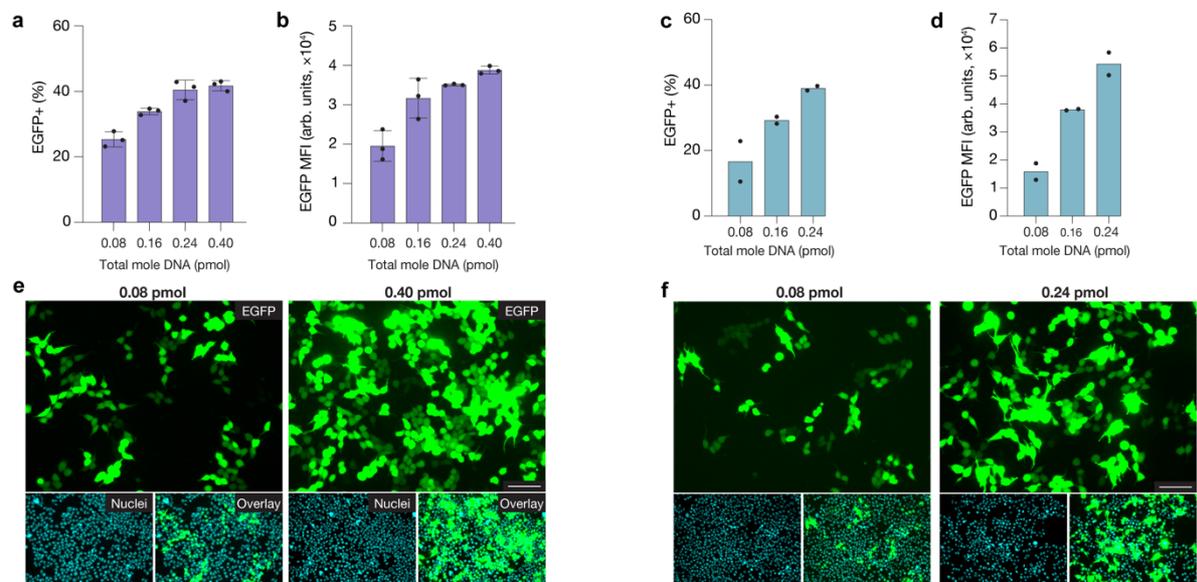


Fig. S16 | Titration of 20HB achieves high transfection efficiency. Titrating the 20HB-exLP folded from the sc_EGFP5 (**a** and **b**) and sc_EGFP6 (**c** and **d**) scaffolds increased both the transfection efficiency and MFI, respectively. Data is presented as mean \pm s.d. (**a**, **b**) or mean (**c**, **d**) with individual data points overlaid. **e** and **f**, Representative epifluorescent microscopy images demonstrated the expression of EGFP by successfully transfected cells. For each of the conditions, EGFP expression (green), cell nuclei (blue) and the overlay are given. The images are representative of one of $n = 3$ and $n = 2$ biologically independent experiments for **e** and **f** respectively; similar results were observed each time. Scale bar $100 \mu\text{m}$.

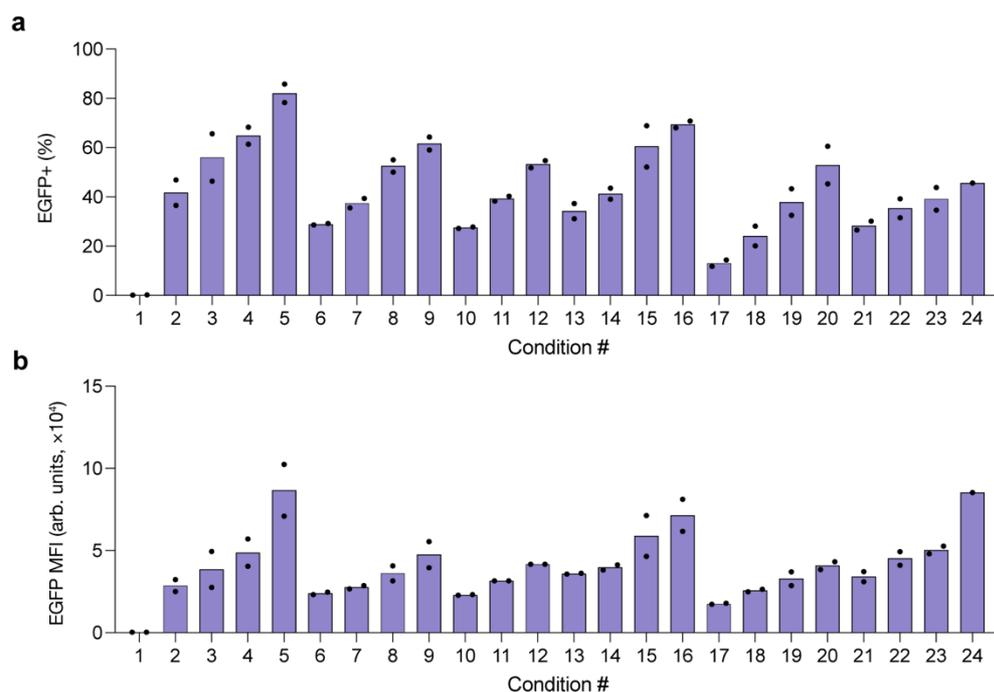


Fig. S17 | Electroporation condition screen in HEK293T cells. Electroporation conditions were screened to optimise delivery of 20HB DNA origami objects. 20HB-exLP sample from scaffold sc_EGFP5 was used in all conditions. Numbers refer to programs detailed in Table S4. Data is presented as mean with individual data points overlaid representing biologically independent experiments.

Table S4 | Electroporation conditions screened as featured in Fig S17.

Sample	Pulse voltage (V)	Pulse width (ms)	Pulse no.
1	-	-	-
2	1400	20	1
3	1500	20	1
4	1600	20	1
5	1700	20	1
6	1100	30	1
7	1200	30	1
8	1300	30	1
9	1400	30	1
10	1000	40	1
11	1100	40	1
12	1200	40	1
13	1100	20	2
14	1200	20	2
15	1300	20	2
16	1400	20	2
17	850	30	2
18	950	30	2
19	1050	30	2
20	1150	30	2
21	1300	10	3
22	1400	10	3
23	1500	10	3
24	1600	10	3

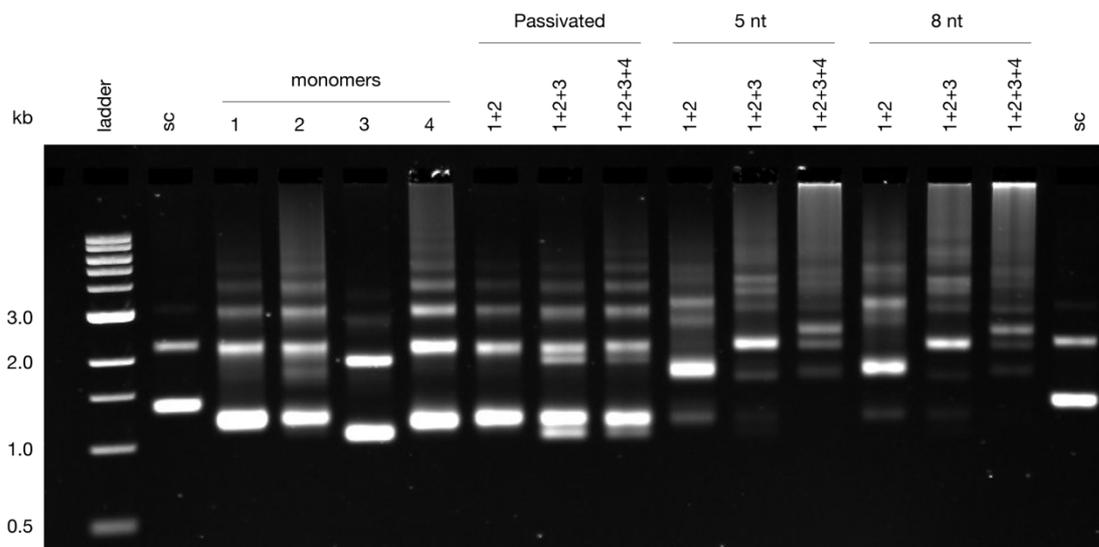


Fig. S18 | AGE of purified DNA origami objects and assembly products. Lanes labelled 1 – 4 demonstrate purified monomer samples. Lanes 1+2, 1+2+3 and 1+2+3+4 demonstrate the monomer addition to create dimer, trimer, and tetramer higher-order structures respectively. No assembly can be observed in the passivated samples. sc_EGFP5 was used as MW control, while 'ladder' depicts NEB 1 kb dsDNA ladder.

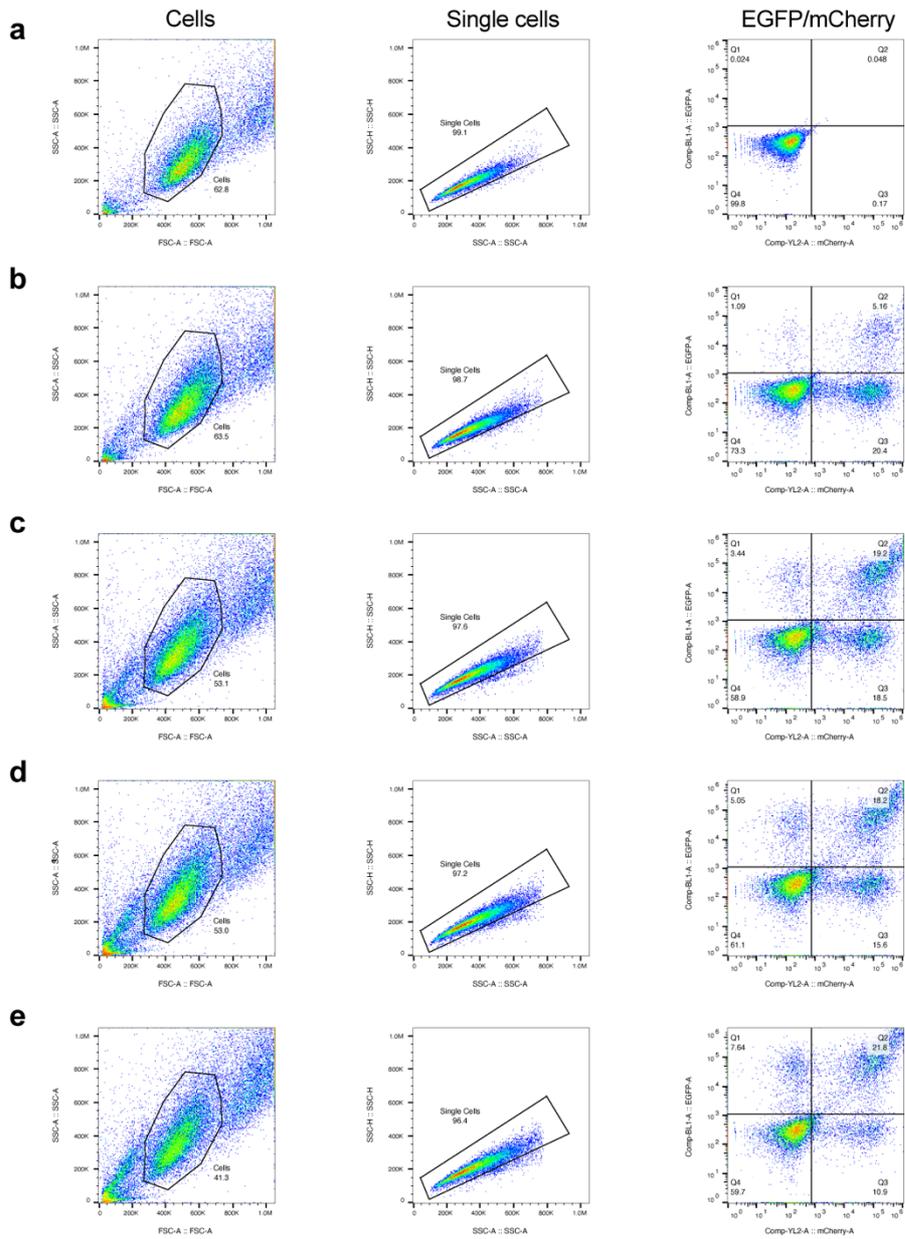


Fig. S19 | Exemplary flow cytometry gating. From left to right: cell populations were first gated on forward scatter-area (FSC-A) versus side scatter-area (SSC-A), gate 'cells'; single cells were selected by gating SSC-A versus side scatter-height (SSC-H), gate 'single cells'; cells were then assessed for EGFP and/or mCherry expression. In this example, cell populations were investigated for expression of both mCherry (x-axis) and EGFP (y-axis) expression, where cells can be either EGFP+/mCherry- (Q1), EGFP+/mCherry+ (Q2), EGFP-/mCherry+ (Q3) or EGFP-/mCherry- (Q4). **a** gives representative gating for untreated cells, **b** demonstrates delivery of individual passivated mChery and EGFP origami objects at a 1:1 ratio mChery:EGFP. Delivery of objects assembled with 8 nt overhangs at mChery:EGFP ratios of 1:1, 1:2 and 1:3 are given in **c–e**, respectively.

2.2 Scaffold sequences and origami design

2.2.1 sc_EGFP1

Scaffold sequence for sc_EGFP1:

cgcggtgacattgattattgactagttattaatagtaaatcaattacggggcattagttcatagcccatatattggagttccggttacataacttacggt
aaatggccccgctggctgaccgccaacgacccccgccattgacgtcaataatgacgtatgttcccataagtaacgccaatagggactttcca
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Corresponding DNA origami designs and staple lists:

2.2.1.1 sc_EGFP1 20HB-ext and 20HB-ext-W

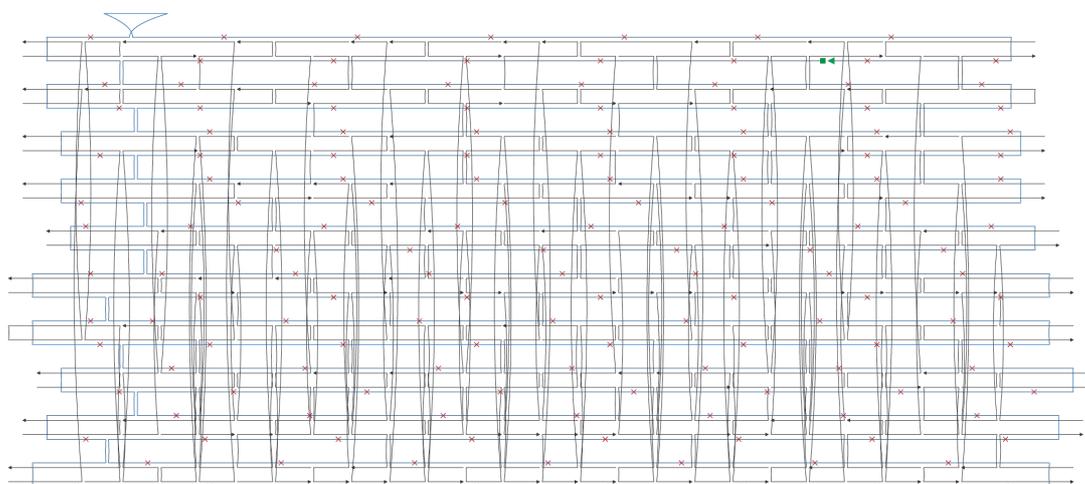


Fig. S20 | Scaffold routing and staple design for sc_EGFP1 20HB-ext and 20HB-ext-W. Scaffold routing is shown in blue, staples are given in black, breakpoint for addition of sequence is marked by a green square (5') and arrow (3'). Design was prepared using caDNAo v0.1.

Table S5 | Individual staple sequences for sc_EGFP1 20HB-ext.

Name	Sequence 5' - 3'
core_1	GGAAGCTCCCTCGTGCGGCGGGCCGGGCGACAACCACCG
core_2	GGACTATAAAGATACGGAACCTCCGGAAGGCTGCAAGCA
core_3	TCCGCCCCCTGACGAATAATCGTGCACCCGGGGTCT
core_4	GCGTTTTTAATTCGCGTTAAATTTTATAAATAGCAGAA
core_5	ATTTACCAAGTCCCTATTGGCGTTGACGTC
core_6	ATTGATTACACTTGATGTAAGTACTGCGATGTAC
core_7	AAGGCCAGTAATGCCAGGCGGGCCCAAGTAGGGGTCTCCC
core_8	CGTCATTATTGACGTGTATTGATTTTGGTGCCAAAACAACTCCCATTACTATG
core_9	TCAATGGAGTAAGTTATGTAACGCCAGGCGTT
core_10	CGTAAATACTCCACCCCGTGAGACCGTACACGCCTACC
core_11	CAGTTTACGGCTATGAACTAATGAGTGGCGAA
core_12	ATATGATACTATTAATAACTAGTCAGCATCAC
core_13	GGGGCGTATAGCGAATAAGCCAGTAAGCAGAGCGGCCGTAAACTCCGCCTCC
core_14	GCTATCCACGCCCATTCAGTGGGAAAAAAG
core_15	CCATGGTAACTTGGCTTTTCTACAAGTATTCTTGCCC
core_16	TGGGGCGGAGTTGTTACACGCTGACCGTCGTGTACCGCGA
core_17	GCGTCAAAATGGGGTGGAGACTTGAAATCCATTGACGTTTTTGTAAAATGCC
core_18	GCCAGAGAGCTCTGCTGAACAGCATCAGTGGCCGAGCG
core_19	AGTCGTATGTGGAACGGTCATTGACGCGTATATCAATGA
core_20	TATAGTGAATCCAGAAGTTTTGGAAGCTTCGTCGTT
core_21	CGCCGGACGACATTTTATCCGGAACATATTGGGCGGTCAGCCAGCTCTCCT
core_22	CCGTTTACGTCGCCGCCCCATTTCCGGTGGTT
core_23	CCTTGCTACCATCTCGTGGGTTCCCTTTGATC
core_24	CGAGGGTTGGGCACCACCCCGTTATATAGACGCGCAG
core_25	AGCGGCTGAACCTTATGGTCTGACGAGTTGCCTTCAGCA
core_26	TGTGGCTTACCTTGATGCCGTTGGGGCAAAGTGTATAATCGGA
core_27	TGTAATCCAGCTTGTAGCTTGCCGCTCCAGATTCATCCA
core_28	GCCCCAGGCCGTCCTCGATGTTGTGGCACAGTCCGATCAGAGCTTG
core_29	ATGTTCCGTCCTCCTTGAAGTCGGTGGTCAACCGGAAGG
core_30	GGGTGTTTACGCTCGATGCGGTTACCCAGG
core_31	CAGCACGGGGCCGTCTAGATTAACGATCAAGGGCGCTA
core_32	TCGGGCAGCTCGAACTTACCTCGGCGGGGAAGAAGTCTGTTGCCGAAATCAA
core_33	ACTGGGTGCTCAGGTAGTATGATCTGGTATGACGCTGCG
core_34	GGCTGGCAACTAGAAGGCGGATCTCACCGGGTGGTG
core_35	ATGCCGAGAGTGGTTGCTATTAATGTGCTGCTTGATGGATATCTGCAG

core_36	CCGGCGGCGCTGCTATTGTCTTCCCAATCCTAGCGGTCGCTTCATT
core_37	GGTCAAGAAGCACTACACTCATGGTTACTA
core_38	ATTTTATTAGGAAAGGCCGATTTGTGTGTCAGCCGTAAGATAGTTGCCCTGGTAG
core_39	CGATGCAGATCAGCGAGCTCTAGGCGAGCTGCAGCCAG
core_40	CCAGAATAATAGAATAGGGCCCTCGCCGATGGGGTCTGCAAGCACTG
core_41	ACCACACCCCCAGCATGGTCACGAACCTCCAGTCTTTGCT
core_42	TCACCCTAATCAAGTTTTATTGAACTCATAACGGGGTCTG
core_43	CTATCAGGGCGATGGCATGAGCGGGGAATAA
core_44	AACGTCAAAAATAACAAAACAATATATG
core_45	TTTGGACAAGAGTCTTCCCGATCTTTTAC
core_46	AATCGGCAAAATCCCTTTGTTAATCTTACCG
core_47	TTTGAATGTATTTAGAAGGGCGAAGGTGAGTATTAGCTCCAATAAACCACGCTG
core_48	GGGTTCCGCGCACATCACTATTAAGTGTATGACATGATCCAGAAGT
core_49	AAAGTGCCAAAAATCGACGCTCAAGTCAGAGCCCCGTATTTACCAGATCTCA
core_50	ACCTAAATTGTAAGCTTGAGTGTGGCGTCACAGCTCCGATCCAGT
core_51	GCGTTTCTGAGAATAAGAACGGAAGGGAAGAAAGCG
core_52	CTGTGACTAAACCGTACGGGGAAAGCCGGCG
core_53	ATACGGGATCTAAAGTGACGCTCATAATTTTCGTGACTAATACGTAGATGTAAGTGCATTTACC
core_54	CCCCATGTGACACTGAATGACACCTACTCAGACAATGAACGTGG
core_55	GTTCCCACTTGTACAGCTGTCCTGCCACCCACCCCAAAGGAGC
core_56	CATCGTGGCTTTAAATTTTAAAGGATTTTG
core_57	TAGATAACTAATTCTGTTGAATAGCATTAGTTCCGACCCTGCCGCTTACCCT
core_58	TGACTCCACTTGTGGCAGATGAACCTCAGGG
core_59	GATCTGTGACACCAGGAGGGCCAGGGCACGGGC
core_60	ATATATGAGCCAGTGTATGTGGTCCGGGGTGTGTGCGC
core_61	TTAAAAATGCACACTGGGCATGGCGGACTTGTCTTGTAG
core_62	GGCAAACAACGGAAATCTTACTGTGTGAACCAACCCCTAAAGGGAGCCACAGTGG
core_63	CAATGGGCTCTTCTTAAAGAGTTGGGAGGGCACTCGCCCTCGCCCT
core_64	TCCAGCTCTATTTCTTTATCAGCTTCCGGTCCGAGGCTATTTCTCT
core_65	AGTGGTCGCATTTAGTTGTGCAACGAGAAAGTGGACTCC
core_66	TGTTCCAGGGGCGCTGGCAAGTGTCCCCCTTGCTCGTCC
core_67	TAGACCGAGATAGGGGTTAATATACCCACTCAATGTCAACGTCAATA
core_68	TCAGGGTTATTGTCTCCCACTACCATGCCATAAGTAAGGACCCACGCTTGAAGTGTGTAGT
core_69	TGCTTTTGCAAAAAAGATACATATCCCCCT
core_70	TAATACCGCGATGTATTTGTTAACCATAGGC
core_71	TTGGCCGCAACAGATGAGTGGCACCTTCCAG
core_72	AAAAGCGGCTCAACCATCTCAGCGCAGATTACCTCCCTCAAACC
core_73	GGCGAGTTCCGGCGACCAGTTACCAGAAGATCTCTAGTTATGCCAAAACCGCATCA
core_74	AGGCACCTAAGTCATTTGGGTGAGACAAATAGACCCGACA
core_75	AATGCTTATCCTCGCCACGCCGTAGGTGAGGATGCCCTCTGCTGGT
core_76	AAAACCTATCCAGTTCGCCACATCAAAGAACGTAACC
polyT_1	TTTTTGGAAACCGTAAAAAGGCCGCTTGTCTGGTGAGCACTGTTGAGACGTTAAG
polyT_2	TTTTTTCGCTGGCGCTTTCTCATTCTCCCGCGGTGTTTCAATATTTTGGG
polyT_3	TTTTTGTATCTCAGTTCGGTTAGCTCA
polyT_4	CAAGCTTGAAGTCCCATAAGGTCAATTTTTTTTTTGTACTGGGCACAAAAGGCCATTTTT
polyT_5	TTTTTTCGAGCTCGGTACGTACATGAGATTATCAAATTTTT
polyT_6	TTTTTTTTTTGGGCTGTGTGCTTATCTGCGCTCTGCTGATTTTT
polyT_7	TTTTTGTCTCTGGACGTCAGTTAATCTAGATCC
polyT_8	TTTTTTTTTTTGAACCGCTGCGCCTTATCGGCCATGATATAGACGTTACGCTTGAATGA
polyT_9	TTTTTCGCTTCTCGTTGGGGCAGGACCACTACAGGTTAATGTC
polyT_10	TTTTTCTTGAGTCCAACCCGGGGCACGGGGACTTCTGC
polyT_11	TTTTTTCAGAAGCCATAGAGCCACCGCATCCGCCGCGC
polyT_12	TTTTTGCAGCAGCCACTGGTAGAGGTGCCGTAGAATAAGACACGACTTATCGCCACTGTTTTT
polyT_13	TTTTTCAATAGGCCGAACATTCCGCCTTTTT
polyT_14	TTTTTTTTCGGGGCGAAAACTCCATTGGAAAAACGTTCTTTTT
polyT_15	TTTTTGAAGTGGTGGCCGTCACAGGATTAGCAGAGTTTTTTTTTTCGAGGTATGTATTCGGGAATTTTT
polyT_16	TAATGGCAGCACTGCATACGATACGGTAGCTCTTGGAAAGTCCCGGTCGTTGCTCCAAGTTTTT
polyT_17	GTGCTCATTCAAGGAATCAGCTCATTTTTTAACTTTTT
polyT_18	TGTCACGCAGAGTAAGCAGGGCGGTTGCCGTGCTCCTTGAAGAAGATGGTGCCTTTTT
polyT_19	TTTTTGTGTTGCCATTGTGTGATCGTTTTT
polyT_20	TTTTTACAGTATTTGGTACCCGGCTACACTAGAAGATTTTT
polyT_21	ATCTGGCCTTGTCCGGTAACTATCGTTTTTTT
polyT_22	CCAGTGTGCGGCTAGGTGGCAGAACCCCGGTTCCAGCTTTTT
polyT_23	TAGTTCGACGCTTCCGGCGCCGTTACTAGTGGATCTTTTT
polyT_24	TTTTTAGGATCTTACAGTTTGCGCAACTTTTT
polyT_25	TTTTTAGCCAGTTACCGCTGTAGTTTTT
polyT_26	C TTCGGAACCTACAGAGTCTTTTTTTT

Table S6 | Individual staple sequences for sc_EGFP1 20HB-ext-W.

Name	Sequence 5' - 3'
core_1	tGGAAGCTCCCTCGTGCttGGCGGGCCttGGGCGActtAACACCGt
core_2	tGGACTATAAAGATACttGGAACCTCttGGAAGGcttTGCAAGCAt
core_3	tTCCGCCCCCTGACGttAATAATCttGTGCACCCttGGGGTCTt
core_4	tCGTTTTTttAATTCGCGTAAATttTATAAAttAGCAGAA
core_5	tATTTACcttAAGTCCCTATTGGCGttTTGACGTct
core_6	tATTGATTtCACTTGATGTACTGcttGATGTACT
core_7	tAAGGCCAGttTAATGCCAGGCGGGCCttCAAGTAGGttGGTCTCCct
core_8	tCGTCATTATTGACGTttGTAtttTGATTTTGGTGCCAAAACAACTCCCAttTACTATGt
core_9	tTCAATGGAttGTAAGTTATGTAACGcttCAGGCGTTt
core_10	tCGTAAATACTCCACcttCCCGTGAGttACCGTACACGCCCTACct
core_11	tCAGTTTACTttGGCTATGAACTAATGAttGTGGCGAA
core_12	tATATGATtCTATTAATAACTAGTcttAGCATCAct
core_13	tGGGGGCGTttATAGCGAttATAAGCCAGTAAGCAttGAGCGGCCcttGTAAACTTttCCGCCTCct
core_14	tGCTATCCACGCCCACTttCAAGTGGGttAAAAAAGt
core_15	tCCATGGTtACTTGGCttTTTTCTACTtAACTGATttTCTTGCCct
core_16	tTGGGGCGGAGTTGTTAttCACGCTGAttCCGTCGTGttTACCGCGAt
core_17	tCGCTCAAAttAATGGGGTGGAGACTTGGAAATCttCATTGACGttTTTTTGTttAAAATGCCt
core_18	tGCCAGAGAGCTCTGcttTGAACAGcttATCAGTgttGCCGAGCGt
core_19	tAGTCGTAttGTGGAACGttGTCATTGAttCGCGttTATATCAATGAt
core_20	tTATAGTGAAttAATTCAttGAAGTTTTttGGAAGCTttTCGTGCTTt
core_21	tCGCCGGAttCGACATttTTGATCCttGGAACATAttTTGGGCGGTTCAGCCAttGCTCTCCTt
core_22	tCCGTTTACGTGCGCCttGCCCATTTTtCGGTGGTTt
core_23	tCCTTGCTCACCATCTcttGTGGGTTcttCTTTGATct
core_24	tCGAGGGTttTGGGCACCACCCCGttTTATATAGttACGCGCAGt
core_25	tAGCGGCTGttAACTTTAttGGTCTGAttCGAGTTGcttCTTCAGCAt
core_26	tTGTGGCTttTCACCTTGATGCCGTTttGGGGCAAAttAGTGTTAttAATCGGAt
core_27	tTGTACTCCAGCTTGTttAGCTTGCCttGCTCCAGAttTCATCCA
core_28	tGCCCCAGGttCCGTCTCGATGTTGttGGCACAGTttCTCCGATCttAGAGCTTGt
core_29	tATGTTGCCGTCCCTCCTTGAAGTcttGGTGGTCAttCCGGAAGGt
core_30	tGGGTGTTttTCAGCTCGATGCGGTTtCACCAGGt
core_31	tCAGCACGGGGCCGCTcttTAGATTAAttACGATCAAttGGGCGCTAt
core_32	tTCGGGCAGttCTCGAACTTACCTCGGCGCGGGttAAGAAGTcttTGTTGCCGttAAATCAAt
core_33	tACTGGGTGCTCAGGttAGTGATcttTGGTATGttACGCTGCGt
core_34	tGGCTGGCAACTAGAAttGGCGGAtttCTCACCGttGGTGGTGt
core_35	tATGCCGAGttAGTGGTTGttCTATTAAttGTGCTGCTttTGATGGATATCTGCAGt
core_36	tCCGGCGGcttGCCCTGCTATTGCTTCCCAATCctTAGCGGCTttGCTTCATTt
core_37	tGGTCAAGttAAGCACTAttCACTCATGGTTAttCTAt
core_38	tATTTTATTAGGAAAGGttCCGATTTttGTTGTCAGttCCGTAAGAttTAGTTGCCttCTGGTAGt
core_39	tCGATGCAttGATCAGCGAGCTTAGttGCGAGCTGttCAGCCAGt
core_40	tCCAGAATAttATAGAATAGGGCCCTcttGCCGATGgttGGTCTGcttAAGCACTGt
core_41	tACCACACcttCCCAGCAttGGTCACGAActCCAGttCTTTGCTt
core_42	tTCACCTAATCAAGTTttTTATTGAAttCTACAttGGGGGTCGt
core_43	tCTATCAGGGCGATGGcttATGAGCGttGGGAATAAt
core_44	tAACGTCAAttAAAATAAttCAAAAACAttATATATGt
core_45	tTTTGAACAAGAGTcttTCCCCGAttTCTTTTACT
core_46	tAATCGGCAAAATCCcttTTTGTTAAttTCTTACCGt
core_47	tTTTGAATGTATTAGAttAGGGCGAAttGGTGAGTAttTTAGCTcttCAATAAACTtCACGCTGt
core_48	tGGGTTCCGCGCACAttCACTATTAttAGTGATGttACATGATcttCAGAAGTt
core_49	tAAAGTGCCttAAAAATCGACGCTCAAGTCAGAGttCCCCGAttTTTCCACAttGATCTCA
core_50	tACCTAAATTGTAAGCttTTGAGTGTttGGCGTCAttCAGCTCCGttATCCAGTt
core_51	tGCGTTTcttCTGAGAAAttAAGAACGttGAAGGGAAGAAAGCGt
core_52	tCTGTGACTttAAACCGTttACGGGGAAAGCCGGCGt
core_53	tATACGGGAttTCTAAAGTttGACGCTCAttTAATTTCCttTGACTAATACGTAGATGTACTGcttATTACCT
core_54	tCCCCATGttTGACACTttGAATGACACCTACTCAGACAAttGttAACGTGGt
core_55	tGTTCCCActtCTTGACAttGCTGTCTTtGCCCCACCCCACTtAAAGGAGct
core_56	tCATCGTGGttCTTTAAAAttTTTTAAAttGGATTTTt
core_57	tTAGATAACttTAATTCTttGTTGAATAttGCATTTAttGTTCCGACCCTGCCGCTTACcttCTt
core_58	tTGAATCCcttACTTGTGGttCAGATGAACTTCAGGGt
core_59	tGATCTGTcttGACCAGGAttGGGCCAGGGCACGGGct
core_60	tATATATGAttGCCAGTgttTCATGTGGTGGGGTttGTGTGCGCct
core_61	tTTAAAAAttGACACACTGttGGCATGGCGGACTTtGttTCTTGATGt
core_62	tGGCAAACAttACGGAAttCTTACTGttGTGAACAttACCCTAAAGGGAGCCcttACAGTGGt
core_63	tCAATGGGcttTCTTCCTTttAAAGAGTttGGGAGGGttCACttTCGCCCTCGCCCTt

core_64	tTCCAGCTCttATTTTCGttTTTATCAGttCTTCGGTCttCGAGGCTttATTTCTCt
core_65	tAGTGGTCGttCATTTAGGttTTGTGCAAAtCGAGAAAGttTGGACTCCt
core_66	tTGTTCAGttGGGCGCTGGCAAGTttTCCCCCTttGCTCGTCCt
core_67	tTAGACCGAGATAGGttGTTAATAtttACCCACTCttAATGTCAAttCGTCAATAt
core_68	tTCAGGGTTATTGTCTCttCCACTAChCATGCCAtttAAGTAAGttGACCCACGttCTTGAAGTttGTTGTAGTt
core_69	tTGCTTTttGCAAAAAAtGATACATAtttCCCCCTt
core_70	tTAATACCGttCGATGtAtttTGTAAAtCCATAGGct
core_71	tTTGGCCGctCAACAGAtttGAGTGGCACCTTCCAGt
core_72	tAAAAGCGGttCTCAACctAtCTCAGctGCAGATTttACCTCCctTCAAACct
core_73	tGGCGAGTTttCGGCGACTtCAGTTACctAGAAGATCttTCTAGTTAttTGCCAAAACCGCATCAAt
core_74	tAGGCACCTttAAGTCATttTGGGTGAGttACAAATAGttACCCGACAt
core_75	tAATGCTTAttTCCTCGctCACGCCGTAGGTCAGttGATGCCCTttCTGCTGGTt
core_76	tAAAACtCttATCCAGTTttCGCCACAtttCAAAGAAttCGTAACct
polyT_1	tttttGGAACCGTAAAAAGCCGCTTGTGcttGTGAGCAAAtCTGTTGAGttACGTTAAGt
polyT_2	tttttCGTGGCGCTTTCTCAAtttTCTCCctGGCGGTGttTTTCAATAttTTTTGGGt
polyT_3	ttttGTATCTCAGTTCCGGttTAGCTCAAt
polyT_4	tCAAGTTGttAAAAGTCCCATAGGTCAttttttttGtACTGGGCAttCAAAGGCCAttttt
polyT_5	tttttCGAGCTCGGTACTtGTCATGAGATTATCAAAAttttt
polyT_6	tttttttCTGGGCTGTGTGttCTTttATCTGCGCTCTGCTGAttttt
polyT_7	tttttGCTCCTGGACGttCAGTTAATttCTAGATCCt
polyT_8	tttttttCCGACCGCTGCGCCTTAttTCGGCCATGATATAGACGttTCAGCTttGCAATGAt
polyT_9	tttttCGCTTCTCGTTGGGGttCAGGACctCTACAGGttTTAATGTCt
polyT_10	tttttCTTGTAGTCCAACCCGGttGGCACGGGGGAttCTTCTGct
polyT_11	tttttTCAGAAGCCATAGAGCCCACCGCATcttCGCCGCGct
polyT_12	tttttGCAGCAGCCACTGGTAttGAGGTGCCGtAttGAAttTAAGACACGACTTATCGCCACTGtttt
polyT_13	tttttCAATAGGCCGAttACATTCCGCCttttt
polyT_14	tttttTTCGGGGCGAAAACtCttCATTGGAAAACGTTCCtttt
polyT_15	tttttGAAGTGGTGGCCttGTCttACAGGATTAGCAGAGtttttttCGAGGTATGTAttTTCGGGAAttttt
polyT_16	tTAAAttTGGCAGCACTGCAttTACGATACttGGTAGCTCttTGGAAAAGTCCCgttGGTCGTTGCTCCAAGttttt
polyT_17	tGTGCTCAttTCAAGGAttATCAGCTCAttTTTTTAACttttt
polyT_18	tTGTACGcttAGAGTAAGttCAGGGCGGttTTGCCGTCGTCCTTGAAGAAGATGGTGCttttt
polyT_19	tttttGTTGTTGCCATTGttTGTGATCGttttt
polyT_20	tttttACAGTATTTGGTttACctCGGCTACACTAGAAGAtttt
polyT_21	tATCTGGCCttTTGttTCCGGTAACTATCGTttttt
polyT_22	tCCAGTGCttGCCGTAGGTGGCAAttGAACCCCCCGTTTCCAGCttttt
polyT_23	tTAGTTCGcttAGCCTTCGttGCGGCCGTTACTAGTGGATCttttt
polyT_24	tttttAGGATCTTCActtAGTTTTCGCGAACttttt
polyT_25	tttttAGCCAGTTACTtCGCTGTAGttttt
polyT_26	tCTTCGGAAttCTACAGAGTTCTTttttt

2.2.1.2 sc_EGFP1 20HB-int and 20HB-int-W

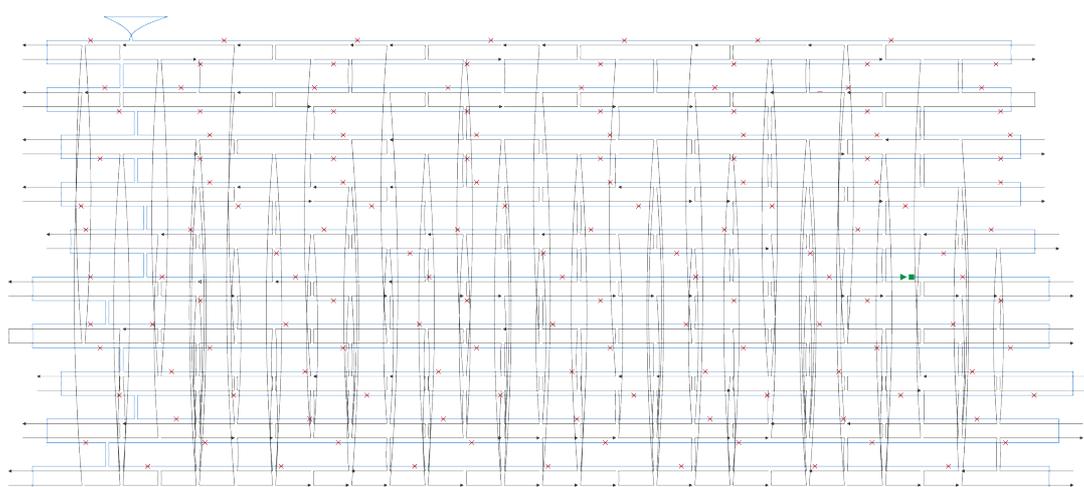


Fig. S21 | Scaffold routing and staple design for sc_EGFP1 20HB-int and 20HB-int-W. Scaffold routing is shown in blue, staples are given in black, breakpoint for addition of sequence is marked by a green square (5') and arrow (3'). Design was prepared using caDNAo v0.1.

Table S7 | Individual staple sequences for sc_EGFP1 20HB-int.

Name	Sequence 5' - 3'
core_1	GGCAAATCCCTTATAGAGCAAATTAATTTCCCAATCC
core_2	AGTCAATTTTTAACAAAGGGAACCAAGCTCCACCCC
core_3	GTGCCACCTAAATTGGAAGCATTGATGGATAGGAAAG
core_4	CACATTTCACTTGGAAATCCCCGATGGTAATACTTGTG
core_5	ACAGGAAGAGATCCAGTTCGATTGGTCT
core_6	TACTCTTCAAAGTGCTCATCATTGCACTGC
core_7	ATATTTGAGACCGAGTTGCTCTTACTCAACCATTAATTG
core_8	CCCACTGATCTTCATCAATGTTGTGCAAAAAGCGGTTAGCTCCTGTAACCCA
core_9	CCGCTGTTGGCAAATGCCGCAAACAATAGGC
core_10	GGCGAAAACCTCTCAAGGCCGAGCTCGTCGTTTGGTATG
core_11	TTCTTCGGGACACGGAAATGTTGAAAATTTTT
core_12	GAACTTTACTTTTTCAATATTATTTAAGCGTT
core_13	CCGCGCCACCGTAAGGTTTCGCCAGTTAATACTCCAGATATAGAATAGTTCACCT
core_14	ACTCATGGTTATGGCAGGAAAACGACCTACTC
core_15	CATGCCATCATAGCAATTTTATTATCTGCACGTAGGTG
core_16	GGTTCCTAACGATCAACCTGACTCGCGCTTCTCAGCAGCA
core_17	CAGCTCCCCGATCGTTGTCAGAAGTAAGTTGGATCTTATGCCCCACTGGGTCTC
core_18	TGCCATTGCTACAGGGCAATGATGCTCGTCTGCCGTCC
core_19	GAAGCTAGGAGTGGCATCAATACGTTATTGTCTCATGA
core_20	TTGCCGGCGAAGGGATCAGCGGCCATGATGCCCTTC
core_21	ATAGTTGGGCGAGTGCCTGCTCTCGTGACCAGCGTTTCTGGGTAATCAAAA
core_22	GTAGATAACTACGATGCTTCATTTGCTGTCC
core_23	GACCCACGCTCACCGGGTTTGCGCATTTCCCTC
core_24	AGTATATTCTGGCCCCAGTGCTCATCGTGGAGAATGAC
core_25	ATCCTTTTATCTTGAAGGGCCCTCAGCTTGCGAATTCCA
core_26	CTGAAGCAACTACGGGTACACTACCCCTGGAAGAAGTCGACGTCA
core_27	TTCGGAAAAAGAGTTTGACAGTTTGGGGGTGAACTCCAG
core_28	GGTAGTCCGGTGCTACAGAGTTCTTCCGACCCCTGGACGTCTATGGGA
core_29	TTGATCCGGCAAACAAACCACCGCAATCTAACTGCACGC
core_30	GCCACTGGGTGGTTTTTTTGTGTTGCAAGCA
core_31	ACGACTTATCGCCACTCTCATATCGAAGTTGGGCCATT
core_32	GGTAAGACCGCGCAGAAAAAAGGATCTCAACATGAGATGCTTGTGAGCTCTA
core_33	AACATCGTCTTGAGCGGTGTAAGCTCGAAGTCCATA
core_34	TCTGCGCTCTCCTGTTGAAGTTCCGCGAACCAATG
core_35	TCTCAGTTTCCAACCCGTTCTTCTTATCAAAAACAATAAACAGCCAGC
core_36	GGTCGTTCCAATGAGTGAGCAAAGGCCAGCAACGCGGATGCGGTTCC
core_37	CTATAAACACCCATTGTGCTGCTTCATGCT
core_38	AATCGACGCTCAAGTCGGCGTTAAGCCTTCGGGTCAGGGCAGGACCATCCCCCT
core_39	GACGAGCTTACCGGATACCTGTGACGAGGTTCGGCGAG
core_40	GCTGGCGTGGGAAGCGTGGCGCTTTGGCAGCATGTGGCGGAAATTA
core_41	TATGAACTGCGTATATGCTCCAAGCTGGGCTCTGCGCCT
core_42	GATGTAAGTCCAAGTGGAGCTCTGAGTGGGTACTTTCA
core_43	ATAGGGGGCGTACTTGACGCCTAGAGTCGTA
core_44	GCCATTTAGAGTTGTGCTCGGTATAAGGGC
core_45	GTCCATAAGGTCATTGATTTGGCACACTG
core_46	CAAAACCGCATCACCTGAGTCAACTCCTCGC
core_47	TTGCGTCAATGGGGCGCCGTCATTGCCAGGGCGAAGATGGTAGTGGTATGTAGG
core_48	TTTGGAAAGTCCCGTACTGGGGATGAACTGCGGGTCTCGATGT
core_49	GTGCCAAAAATTTTTGTAAAATTCGCGTTATACTCAGCGCCGTAGACAAT
core_50	ACAAACTCCCATTGAACTGCCAAGCATCGCACCAGGGTTGATGCC
core_51	TACTAGTGTGGTGCACATAATGGGTGTTGGGCGGTC
core_52	GAGGGTGGGACGTCAACATACGTCAATTATTG
core_53	CCTCGCCCGCATTTAGGACAGTGGAGTAAGTAATGCTTTTCTGTGACTGGTGGTCCCGGCG
core_54	TTGTAGTTCCTTCTTTTCCATAGGCTCCGCCCCCTACGTCAA
core_55	GTGCCCCGCTCACGCAGGAACCGTAAAAAGGCCGCGTTAGCCAGGC
core_56	TCCTCCTTGCCGTTTAGGCACAGGAAGGCAC
core_57	CGTTGGGGCGGCTGACAGTAAGCCTTATATGAATAGACCGAGATAGGGTTGTT
core_58	TGTGATCCCCGTCGTTAATCAGTGAGGCA
core_59	CCGGCGGCGCTTACCAATGAGTAAACTTGGTC
core_60	GTGACACTTTATCAGAGGATCTTACCTAGGCAGATTA
core_61	TCGAGGCTGCCGAGCGTTAAGGGATTTTGGTGAAGATCC
core_62	ATTGCTTCGATAAGCAGCACTGCATACACTTATGGAAAGTCCCTATTAGAGGTG
core_63	GCATCTTTTCTCTAGTACCGCATCCAGGGCGCCATTTTCGTTTCATCC
core_64	ACGGGAGGGGTACGTTCTGCTGGTGGCTCCTGCCGCATCACAAA
core_65	ATTAGCAGCGCCTTCTGCCGTCGTGGGCGGGCCAGGCGG

core_66	GTAGGAAATACCGTAAGTTATGTAAAAGGCCTGTAGGTA
core_67	CTAATACGTAGATGTCGTCGAATGCGCCAGTGTTATCAGGGGATAATA
core_68	AGACCTCCCACCGTACGCATATGACGCCGTAGGCATGGCGGGGCCGGGTGGCCTCAGTTACC
core_69	TGGTACCCTATAGTCCGCCATCGAAATC
core_70	TCGCCGAGAGCGGCGGGTGGAGCCGAAAA
core_71	CGGACTTGAAGCTCCCGCAAACCCGACAGGA
core_72	TCCTTGAAACGGGCAGAGTGATCCCAGAATTGTCACGTGTTATC
core_73	CACCTCGGTACGGGTCTAGATTAGCGATGCAAACGTTGTATAATTCTTACTGT
core_74	CATGCCGAGCTTGCCGGGATCCGATACGACATGTTAAATC
core_75	CTTTGACAACCGGAAATGAAGTTTTAAATCTGGTAGCGTAACAGG
core_76	CCTTCCAACCATCTCCACGCTGAAGCGATGATATGGGC
polyT_1	TTTTTAAAAATAAACAAATAGGGGTTCCGCGGCGGATACCCTTGCTCGGGTCAAG
polyT_2	TTTTTTATTAAGAAGCTGGAGGAACAAGCGGGCGTAGCCAGAGGCAGTT
polyT_3	TTTTTGCGAAAAACCGTCTACTCCAAC
polyT_4	TCCAGTCTAAGTCATTCTGAGAATAGTTTTTTTTTTTGTATGCCGCATGTATTTAGTTTT
polyT_5	TTTTTATCCGCCTCCAGGGGAGGGGCAAACAACCTTTTT
polyT_6	TTTTTCGTAACCATCAGACAATGTCACATTCCGCCTTTTT
polyT_7	TTTTTCAGTGAACGAAGTTGTAGTAAGTAGAA
polyT_8	TTTTTGGTCGAGGTGCCGTAAAATTTGGTATCTGCGCTCTGCCTATCTTGTCCGG
polyT_9	TTTTTCGTTACGCCGACCGGTGTGCAGTTGCCGCGTAATTG
polyT_10	TTTTTCTAAAGGGAGCCCCACCAGCGTTTGAAGAAC
polyT_11	TTTTTATACTAGTCAATAATCAATGTCAACAATGACCC
polyT_12	TTTTTCCGGCGAACGTGGCGACGTAATAACTCGATGATTTAGAGCTTGACGGGGAAAGTTTT
polyT_13	TTTTTTGATGTACTGCATTACTATTTTT
polyT_14	TTTTTATGGGCACCACCCCGGTCCAGCTCGACCAGTTTTT
polyT_15	TTTTTAAGTGTAGCGGTTACGAAAGGAAGGGAAGATTTTTTTTTTAAGCGAAAGGAGAGTCCACTTTTT
polyT_16	CACGTGGTCGGGGTAGTCTTTGCTCCAGCATTACATGATCCCCGGGCGATGGCCCACTATTTTT
polyT_17	CGTCGCCGTGAACAGACCGCTATCCACGCCATTTTTT
polyT_18	GAAGTCGATATAGACGTATCCGGTTTTGATCTTTTCTACGGGGTCTGACGCTTTTTT
polyT_19	TTTTTTGTGCCCCAGGATGAACCCCTTTTT
polyT_20	TTTTTCCCGCCGCGCTTTGGGCGCGTAACCACCATTTTTT
polyT_21	GTGCTCAGAGTGCACATAATCGGAACCTTTTT
polyT_22	GTAGTGGTCAGCGATCTGTCTTAATCAAGTTTTTTGGTTTTT
polyT_23	TTGTGGCTAACTCACGCAGAAGTGGTCCGTGCAACTTTTTTT
polyT_24	TTTTTAGATGGCTGGCTGTACTCCAGCTTTTTT
polyT_25	TTTTTTCAGAAGCCAGTCAAAGGTTTTT
polyT_26	TAGAGCCCCTAGGGCGCTGGCTTTTT

Table S8 | Individual staple sequences for sc_EGFP1 20HB-int-W.

Name	Sequence 5' - 3'
core_1	tGGCAAATCCCTTATAttGAGCAAAAtTTAATTTtCCCAATCct
core_2	tAGCTATTTTTTAACtAAAGGGAAtCCAAGCTtCCCACCCct
core_3	tGTGCCACCTAAATTGtGAAGCAtTtTGATGGATtAGGAAAGt
core_4	tCACATTTctACTTGGAAATCCCCGtATGGTAATtACTTGTgt
core_5	tACAGGAAtGAGATCCAGTTCGATtTCGGTCCct
core_6	tTACTCTTctAAAGTGCTCATCATTtGCACTGct
core_7	tATATTTGAtGACCGAGTTGCTCTGtACTCAACCtATTAATTGt
core_8	tCCCAACTGATCTTCAAtTCAtATGTTGTGCAAAAAAGCGTTAGCTCcttGTAACCCAt
core_9	tCCGCTGTTtGGCAAAATGCCGCAAAAtCAATAGGct
core_10	tGGCGAAAACCTCAAtGGCCGCGAGtCTCGTCGTTTGGTATgt
core_11	tTCTTCGgtGACACGGAAATGTTGAtAAATTTTTt
core_12	tGAACTTTAttCTTTTTCAATATTATTtTAAGCGTTt
core_13	tCCGCGCCAtCCGTAAGtGTTCCGAGTTAATtCTCCAGATtATAGAATtGTTACCCt
core_14	tACTCATGGTTATGGCAttGGAAAACGtACCTACTct
core_15	tCATGCCAttCATAGCAAtATTTTATTtATCTGCAAtCGTAGGTgt
core_16	tGGTCCCAACGATCAAtCCTGACTctGCGCTTcttCAGCAGCAAt
core_17	tCAGTCCtCCGATCGTTGTGAGAAGTAAGTTtGGATCTTAtTGCCCCACtTGGGTCTct
core_18	tTGCCATTGCTACAGGtGCAATGATtGCTCGTcttTGCCGTCCct
core_19	tGAAGCTAGtGAGTGGCAtTCAATACGtGTTATTGTCTCATGAt
core_20	tTTGCCGgtCGGAAGgtGATCAGCGtGCCATGAtTGCCCTTct
core_21	tATAGTTgtGGCGAGtGCCTGCTtCTCGTGCAAtCCAGCGTTTCTGGGTtAATCAAAAt
core_22	tGTAGATAACTACGAttGCTTCATTtTGCTGTCCct
core_23	tGACCCACGCTCACCGgtGTTGCGcttATTTCCCTct
core_24	tAGTATAtTCTGGCCCCAGTGCTtCATCGTGGtAGAAATGACt
core_25	tATCCTTTTtATCTTGAAtGGGCCcttCAGCTTGTtGAATTCcAt
core_26	tCTGAAGcttAACTACGGCTACACTAtCCCCCTGGtAAGAAGTcttGACGTCAAt

core_27	tTTCGGAAAAAGAGTtTGACAGTtTGGGGGTGtAACTCCAGt
core_28	tGGTAGCTCtCGGTGCTACAGAGTtTCCGACCTtCTGGACGtTCTATGGGAt
core_29	tTTGATCCGGCAAACAAACCACCGtCAATCTAAttCTGCACGct
core_30	tGCCACTGtGGTGGTTTTTTGTTTTGCAAGCAt
core_31	tACGACTTATCGCCACtTCTCATAtTCGAACTtTGGGCCATTt
core_32	tGGTAAGACTtCGCGCAGAAAAAAGGATCTCAAttCATGAGATtGCTTGTCGtAGCTCTAt
core_33	tAACTATCGTCTTGAGtCGGTGtAtAGCTCGAttACTCCATAt
core_34	tTCGTGCGCTCTCCTGtTTGAAGTtTCGCCGAttACCAATGt
core_35	tTCTCAGTtTCCAACCCtGTTCTTtTATCAAAAttCAATAAACCAGCCAGct
core_36	tGGTCGTTctCAATGAGTGAGCAAAAGGCCAGctAACGCGGAttTGCGGTtCt
core_37	tCTATAAAAtCACCCATTtGTGCTGCTTCATtGCTt
core_38	tAATCGACGCTCAAGTtGCGGTTtAGCCTTCGtGGTCAGGGtCAGGACCAAtTCCCCCTt
core_39	tGACGAGctTTACCGGATACCTGTCtAGCGAGGtTTCGGCAGt
core_40	tGCTGGCGTtGGGAAGCGTGGCGCTtTGGCAGCAAtTGTGGCGGtAAATTAAt
core_41	tTATGAACTtGCGtTATAAtGCTCCAAGCTGGGCTtCTGCGCCTt
core_42	tGATGTAActGCCAAGTtGAGCTCTGtAGTGGGtTACTTTCAAt
core_43	tATAGGGGCGTACTTtGtACGCCAttGAGTCGtAt
core_44	tGCCATTTAtGAGTTGtGCTCGGtAtTAAGGGct
core_45	tGTCCATAAGGTCAttTGATTTTtGtGCACACTGt
core_46	tCAAAACCGCATCACctTGAGTCAAttCTCCTCGct
core_47	tTTGCGTCAATGGGGCGtCCGTCATTtGCCAGGGctGAAGATGtGTAGTGGTtATGTAGGt
core_48	tTTTGGAAAGTCCCGTtGTAActGGGtGATGAActtCGCGGGTctTCGATGt
core_49	tGTGCCAAAtAATTTTTGTTAAAtTCGCGTtATACTCAAttGCGGCGTtAGACAAtt
core_50	tACAAACTCCCATTGAttACTGCCAAttGCATCGctACCAGGGtTGTATGCCt
core_51	tTACTAGTtGTGGTGCAttCATAATGtGGTCGTTGGGCGGct
core_52	tGAGGGTGGtGACGTCAttACATACGTCATTATTGt
core_53	tCCTCGCCctGCATTTAGtGACAGTGGtAGTAAGTAttATGCTTTTCTGTGACTGGTtGAGTtCCCGGCGt
core_54	tTTGTAGTtTCCCTTctTTTTCCATAGGCTCCGCCCCCTtACGTCAt
core_55	tGTCGCCctGTCACGctAGGAACCGTAAAAAGCCCGGtTtAGCCAGGct
core_56	tCCTCCTTtGCCGTTtAtGGCACAGtGAAGGCAt
core_57	tCGTTGGGtCGGCTGAtCAGTAAGctCTTATAtGAATAGACCGAGATAGGGTtTt
core_58	tTGTGATctCCCGTCGtTCTTAATCAGTtGAGGCAAt
core_59	tCCGGCGGctGCTTACCAttATGAGTAAACTTGGTct
core_60	tGTGACACTtTTATCAGtAGGATCTTACCTAGtGCAGATTAt
core_61	tTCGAGGCTtGCCGAGCGtTTAAGGGATTTGGTtGAAGATCCt
core_62	tATTGTCTtCGATAAGctAGCACTGctATACACTtATGGAAAGTCCCTATTtAGAGGTGt
core_63	tGCATCTTtTCTCTAGtACCAGCATctCAGGGCGtCCctATTTCTGTTCTATCCct
core_64	tACGGGAGGtGGTCAGctTCTGCTGtGTGCGCTctCTGCCGctATCACAAAt
core_65	tATTAGCAGtCGCCTTctTGCCGTCGtTGGGCGGctCCAGGCGGt
core_66	tGTAGGAAAtTACCGTAAGTTATGtAAAAGGCCtTGTAGGTAAt
core_67	tCTAATACGTAGATGtTCGTCAATGtCGCCAGTtTTATCAGGtGGATAATAAt
core_68	tAGACCTCCACCGTACTtGCATATGtACGCGTAtGGCATGtCGGGCCGtGGTGGCCTtCAGTTACct
core_69	tTGGTCActCCTATAGtTCCGCCAttCGAAATct
core_70	tTCGCCGAtGAGCGGctGGGTGGAGtCCCCAAAAAt
core_71	tCGGACTTtAAGCTCCctGCGAAACCCGACAGGAt
core_72	tTCTTGAAttACGGGCAAtGAGTGATctCCAGAAttTGTCACGtTGTATCTt
core_73	tCACCTCGGtTCAGGGtCTAGATtGCGATGCAtAACGTTGtATAATTCTCTTACTGtTt
core_74	tCATGCCAtGCTTCCGtGGATCCGAttACGACATtGTTAAATct
core_75	tCTTGACAttACCGCAAtAATGAAGTTTTAAAttCTGGTAGctGTAACAGGt
core_76	tCCTTCAAtACCATCTctCACGCTGAtAGCGATGAtTATGGGct
polyT_1	ttttAAAAATAAACAAATAGGGGTCCGCGtGCGGATACtCCTTGCTctGGGTCAAGt
polyT_2	ttttTATTAAGAACGTGGAtGGAACAAtGCGGGCGtTAGCCAGAtGGCAGTt
polyT_3	ttttGCGAAAAACCGTCAAtCTCCAAt
polyT_4	tTCCAGTCTtAAGTCACTGAGAATAGtttttttTGTATGCGGctATGTATTTAGtttt
polyT_5	ttttTATCCGCCTCCAAtGGGGGAGGGGCAACAACtttt
polyT_6	ttttttCGTGAACCATCAAtGACtAATGTACATTCGCCctttt
polyT_7	ttttCAGTGAACGAAtGTTGTAGTtAACTAGAAt
polyT_8	ttttttGGTCGAGGTGCCGTAAtATTTGGTATCTGCGCTCTGtCCTATCTtTGTCGGGt
polyT_9	ttttCGTTCAGCCGACCGtGTGTGCActGTTGCCGtCGTAATTGt
polyT_10	ttttCTAAAGGGAGCCCCctACCAGGCGTTTTtGAAGAAct
polyT_11	ttttATAACTAGTCAATAATCAATGTCAACtAATGACCCt
polyT_12	ttttCCGGCGAACGTGGCGAttCGTAAATACTctGATtGATTTAGAGCTTGACGGGAAAGtttt
polyT_13	ttttTGATGTAActATTACTATtAtttt
polyT_14	ttttATGGGCACCACCCCGGtTCCAGCTCGACCAGGtttt
polyT_15	ttttAAGTGTAGCGGtTACTtGAAAGGAAGGGAAGAttttttAAGCGAAAGGAtGAGTCCACtttt
polyT_16	tCACTGTTGGTCGGGGTAgTtCTTTGCTtCCCAGCATtTACATGATCCCCctGGGCGATGGCCCCACTAtttt
polyT_17	tCGTCGCGGtTGAACAGtACCAGTtACCAGCCCCAtttt
polyT_18	tGAAGTCGAtTATAGACGtTATCCGGTtTTTGTATCTTTCTACGGGGTCTGACGCTtttt
polyT_19	ttttTGTGCCCCAGGAttGAACCCCTttt

polyT_20	tttttCCCGCCGCGCTTtTGGttGCGCGTAACCACCACAtttt
polyT_21	tGTGCTCAGttAGTttGCACTAAATCGGAACtttt
polyT_22	tGTAGTGGTttCAGCGATCTGTCTttTAATCAAGTTTTTGGttttt
polyT_23	tTTGTGGCTttAACTCAGGttCAGAAGTGGTCCTGCAACTTtttt
polyT_24	tttttAGATGGCTGGCttTGTACTCCAGCTtttt
polyT_25	tttttTCAGAAGCCAttGTCAAAGGttttt
polyT_26	tTAGAGCCctTAGGGCGCTGGCtttt

2.2.1.3 sc_EGFP1 32HB

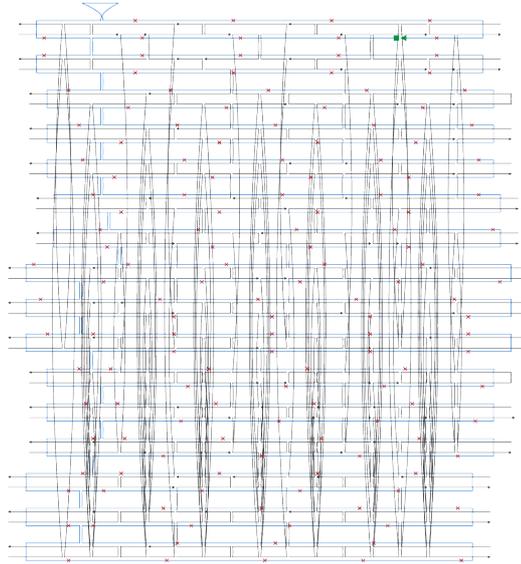


Fig. S22 | Scaffold routing and staple design for sc_EGFP1 32HB. Scaffold routing is shown in blue, staples are given in black, breakpoint for addition of sequence is marked by a green square (5') and arrow (3'). Design was prepared using caDNAo v0.1.

Table S9 | Individual staple sequences for sc_EGFP1 32HB.

Name	Sequence 5' - 3'
core_1	GAACCGTACCACTACGTGAACCATGCCGTAAA
core_2	ACCGTAAGTTATGTACTGCATTTACCGTCATTGACGTCAATAGGGGGCCAGGCG
core_3	GGCGTTACCAAAAAGGTTTTGTTACCTAAATGCACTAAAAGCCATAGAGCCCACC
core_4	GCGTACTTCATAAAGGTCATGTACTTTGGTGCCTGCTTAATGACTCCTATTATTG
core_5	CAAGTAGCGAAAACGGGCGGGGATTACTATTAATAACTTTCCATAG
core_6	TACGTAGATGTACTGCATGGGGTGATATGAGTGGGAGGGTGTGAAGTGCACCC
core_7	AATAGCGAGAGATTATTATGGGAATCAACGCGTATATCAA
core_8	ACCGCATCACCATGGTCAAACCGAAAAATGACAATGATACGCAAAATTTACCA
core_9	TGGGTTCTCTAGTTAGAGTGAGTCAGATTTATTGGTATG
core_10	CTCTGATGGATATCTGCAGAATTGGGGCGGATAGTTGCCTCAGTGAG
core_11	CCAGCACATTATCCGCGTTAATAATCATTGGGCACTGCA
core_12	CTGGCGGCTCGCCCTTGCTCACTGGGCCAGCCGATCGT
core_13	GCTTGGGTCTCCCTATCCAGAGAAGTGTGAGTTTTAAAGTGCC
core_14	TTGTGGCCGTTTACGTGTGGCATATCAAGG
core_15	GCAGCTTGCCGGTGGTCCCCTGTGCCATTGAGCGCAGCCACTCTACTCATA
core_16	GCAGATGGTGCAAAAGGTTACCTGCTTGTCCGCCATG
core_17	AACTTCAGTGGCGGACTTGAAGAAGTCGTGCCTCGATGC
core_18	GGTCAGCTTGCCGTAGCGCCGTCTCGTCTCAGCAATCTTTTACAAGGGAAT
core_19	ATGGTGGCTCCTGGACGTAGCCAACTTCACCGAGTTACGGTGAGTACCGGCGTC
core_20	TGCCGTCTGTGAGAATATGGCAAAAACGTTGGTCAAGGTCCTCATTTTATTAG
core_21	TCCTTGAATGTTGTAGTTGTACTGGGCGGAC
core_22	CTCGGCGGATCTTGAAGTTCACGTGTTCTG
core_23	GTTGTGGCGTCGATGCCCTTCAGTGCTTCAT
core_24	CGTTCTTCAGGGTGTCCGCCCTCGTTCCGGGCA
core_25	CTTGATGCTGTGACTATGATCCCTGTCAGCCAGCTCGA
core_26	GGGCAGCATCACGAACTCCAGCAGGGCAACA
core_27	CTTCTCGTTGGGGTCCCTTCCAGCTTCGGGGCGAAGC

core_28	TGATCGCGTGGGTGCTCAGGTAGTGGTTGTCATATAGACTTACTGTCAGCTCCTT
core_29	TCGTCCATGCCGAGAGCACAGTCAATACGGGTTCCAGCAT
core_30	AGGCTGATGTCCTGCCACCCACATTCCAAGGGCGATCCCCGAA
core_31	ATGACACTACTCAGCGTAACCACTCTTCTAAAATAAA
core_32	AGAATAGACTGGCAACTAGAAGGTGATCCCTGCTTTTC
core_33	CACCCCCCAACTGATCATAATACCATCGTGGCCATGTT
core_34	CCCCTTGCTCAGCGAGCTCTAGCAGTACTT
core_35	GGGCGCTGATCAGGGTTATTCTATATTTTCGTAATTGTT
core_36	CCACACCGCTTGACGGGGAAAGGGACTCCA
core_37	GCCTCAGATCGGAACCCTAAAGGTATCAGG
core_38	GAAAGGAAGGGAAGATTTGGAACGATAGGGTCTCCATA
core_39	CACTATTAAGAAGCTCCGGCGAATATTTAGATTTTCAACCGTCGTGCTGCAACT
core_40	AGGGCGAAAAACCGTCGAGCCCCCGCACATTCACGGAAA
core_41	TGAGTGTCAAGAAGACTCAGCGATCCTTTGGAAGTCCCCTTGATTGGGCATA
core_42	TATAAATCTGACGCTGTTACCAAAAAACAACCCATTTGCGTCAAT
core_43	AAATCCCTGTAATTGGTCGTTGGGCGGTCA
core_44	AATCGGCAACGTCAAGCTCCGCCCCCTGACGAGCATCATGACCCC
core_45	TAACCAATAGGGATTTTAAAGTATGAGACTTTATATAGACCTCCCACCGAGCTCGAAGGGCCG
core_46	TCATTTTTAATCAATGCATACGTCAATTATTGTGCCAAG
core_47	AAATCAGCGCGATGGCAAAAGCCGCGTTGCTGGCGTTTAGTCAAT
core_48	ATACATATTAGACCGAAAGAGTCCGACGCTCAAGTCAGAGGTGCAGACGCGGAA
core_49	TGTTGAGATGCAATTAAGGCACGGGGAGGGACCATGTAATTCTC
core_50	AGTGCTCGTTTGGCCGGTCTGGCAGGGTGGTCGGGGTAGCGGCTGAAGC
core_51	ACTTTAAACGATGTAAGAAGTGGTCTAGATAACGCCTACCGACTCCCATTGACGTCA
core_52	AAGCGTTATGCCATATAGCAGAACAGATGG
core_53	AAGCTAGGGCCGACGGTCCGGTCAGGGTGGTCACGAGGGCATCTCGAGTTCCGCA
core_54	AACGTTGTAACAGCTCCCCTTACTAGTGGATCCGTACACTACGATACAAACTTGCAAATAG
core_55	CTACAGGCGCGCCACCCGTAAGAGGCGGGCAGGGGCCGCTCGCCGATGGGG
core_56	CTCCATCCTTACCGCAAGCATTTGCAAGTG
core_57	AAACCAGCTGGCCCGCTCTGCTGGAAATCCCCGTGAGT
core_58	CTTACCATCCAGCCGGGTACCAACCAGGATGGGCACCAC
core_59	GTCTGACACAGTGAAGAAAGTCCGGCATATGATACACTTGATGTACAGTCAAT
core_60	AATCAATCTGGTCAATTGACTAATGGGCAGTTTACCCTAAATACTCC
core_61	TCTTTTCGGCCATTTTATGGGCTATGAACTAACAAAAAT
core_62	TACGGGGTCAAAAGAATTGAATGCGTGGCGATAGCGGTACGCTGCGACAATGCGATCCAGTT
core_63	TCACGTTAAGGCCGAGGGTTCCGGATTTAGACGCGCTTAATGTC
polyT_1	CTCTTGCCCTCAACCAGTACAGCCTGGTAGTGGTCGGCGAGCTGCACGCTGTTTTT
polyT_2	TTTTTCCAGCAAAAGGCCAGTGAGTGACGTTAAATATCTTCACCTAGTTTTT
polyT_3	TTTTTGGCGTTTCCCCTGGGACTATAATGCTACAGTGTCCAGAAGCGAA
polyT_4	TTTTTTGGAAGTCCCTATTACCCATT
polyT_5	TTTTTCTCTCCTGTTCCGACCAAGCTCCCAGGATCTAGTTCTTGAAGTTTTTT
polyT_6	TTTTTCTGTCGCTTTCTCTCAAGCAGCAGATTACGTTTTT
polyT_7	TTTTTCAGTAAGCAGGCCATTGATGTTTTTTTTTTTACTGCCAAAGACGTCAATTTTT
polyT_8	TTTTTCGCTTTCTCATAGGCCCCACCGCTGGTAGCGGTTTTT
polyT_9	TTTTTCACGCTGAACCTTTGATAAGCTTTTT
polyT_10	TTTTTCAGTTCGGTGTAGAAAGAGTTGGTAGCTCTTTTTTT
polyT_11	TTTTTCTTGAAGAAGCCCTCGCCGATTTTT
polyT_12	TTTTTGGGCTGTGTGCACGACCCAGGATGTACTGCACGCCGTAGTTCGCTCCAAGCTTTTT
polyT_13	TTTTTCCGTCCTCGATGTGAGAATTAGTTTTTT
polyT_14	TTTTTCGACCGCTGCGCCTTATCTTTGCTCACCAGCTTGCTCATGGT
polyT_15	TTTTTAGAATAGGGCCCTCTATTTAGGTGACACTATTTTTT
polyT_16	TTTTTCGGTAAGACACGACTTGGGAGTGGCACGGTAACT
polyT_17	AATCCTCGCGTTTCTCCGAGTTGGCTTCATTCAGCTTTTTT
polyT_18	TTTTTCTGCTATTGTCTTCCGCATCCCCAGCATGCTTTTT
polyT_19	TTTTTAGCACTGGTAACAGGAGCGGGCCTAGAAAGGAC
polyT_20	TTTTTTTTGGGGTCGAGGTCACCCTAATCAAGTTTTTTTT
polyT_21	TTTTTATGTAGGCGGAGATACCATTTTT
polyT_22	TTTTTGGTGGCCTAACTAAGGATTAGCAGAGCGAGGTTTTT
polyT_23	CGGGTCTCATGAGCGGGCACCTATTCTTTGAAATGCCAGGCGGGCCCCGCTTACCGGATACTTTTT
polyT_24	TGTAAGCGCAAAATGCCCGCAGACCCACTTTTT
polyT_25	TTTTTAAACAGGAAGGTTAATTTTTTTTTTTTTTTTGTAAAATTCCGGCAAAAGGTTTTT
polyT_26	TTTTTAGTATTTGGTATCAGTATCGCCACTGGCAGCTTTTTT
polyT_27	TGCAACTCTCAAGGATCAGTCTATTTTATCCAGTTGTTACGACATCTTCGGGAAGCGTGGTTTTT
polyT_28	TTTTTGTATGCGGCGAGGGTGTAGCAATTTTT
polyT_29	TTTTTGTACCTTCGAAATCGTCTTGAGTCCAACCTTTTT
polyT_30	TGTTATCATGCACCCCGCTTACGCTTTTTT
polyT_31	AGTCATTTCTGTGGCGGGTCTTTGTAGTTGCCGCTGTTTTT
polyT_32	GTAAGTTAGTAAGTAGCGGCCAGTGACGCTGTAGGTATCTTTTTT
polyT_33	TTTTTCCGGTCCCAACGCGCCCTCG

polyT_34	TTTTTGATCCGGCAAACAAAGGGTCTGCTGAAGCCATTTTT
polyT_35	TTTTTGCTCACCGGCTCCGTATTAA
polyT_36	TTTTTTGGTTTTTTTTGTTTGGTCCACTAGAAGAACTTTTT
polyT_37	TTTTTATCCTTTTAAATTCTATCCAC
polyT_38	TTTTTCGCAGAAAAAATCGTGCGTTTTT

2.2.1.4 sc_EGFP1 12HB

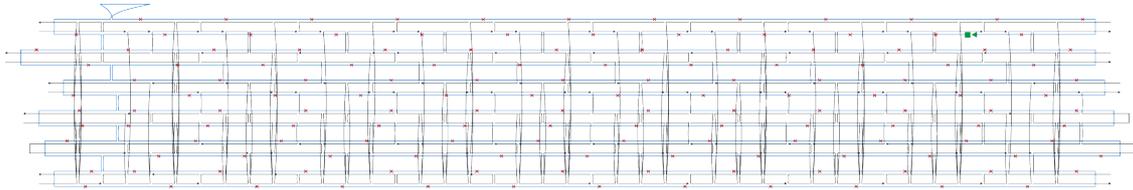


Fig. S23 | Scaffold routing and staple design for sc_EGFP1 12HB. Scaffold routing is shown in blue, staples are given in black, breakpoint for addition of sequence is marked by a green square (5') and arrow (3'). Design was prepared using caDNAo v0.1.

Table S10 | Individual staple sequences for sc_EGFP1 12HB.

Name	Sequence 5' - 3'
core_1	TGCACGAAGTCCTCCGATCGTTGTTAAAATTC
core_2	CGTTCGCTCCAAGCTGGGCTGTGGTACTGCC
core_3	GTGTAGGTGCGAGTGTTATCACTCGCCACCTA
core_4	CACGCTGTAGGTATCTCAGTTCGCTCCACCC
core_5	TCATAGCTCTGCATAATTCTCTTTAGGGGTT
core_6	CCTTCGGGAAGCGTGGCGCTTTCATTGGCGT
core_7	CTTTCTCTAAGATGCTTTTCTGATATTTGA
core_8	GCCGCTTACCGGATACCTGTCCGCTTGACGT
core_9	CGACCCTAACCAAGTCATTCTGTTATCAGG
core_10	CTCCCTCGTGCCTCTCCTGTTCTCAGCCA
core_11	CCTGGAAGCGACCGAGTTGCTCTACTCTTC
core_12	TATAAAGATACCAGGCGTTTCCCTGTAACGC
core_13	GACAGGACCGGGATAATACCGCGCTAAGGGC
core_14	TCAAGTCAGAGGTGGCGAAACCCACTAATGA
core_15	ATCGACGCTAAAAGTGCTCATACACAGGAAG
core_16	CCCCCTGACGAGCATCACAAAAACTAGTC
core_17	AGGCTCCGGGGGCGAAAACCTCTCA
core_18	CCGCGTTGCTGGCGTTTTTCCATTCAATGAG
core_19	TCATTGACGTCAATAGTAGGTGCAGATGAACTTCAGGGTCAGCTTGGGCGGGCCCCGCGCAGA
core_20	CCGTAGGTACGAGGGTGGGCCAGGCATTTA
core_21	GGCATTGCGCCCTCGCCCTCGCCGGCCATAAGTTTGATCTATGGCTTC
core_22	ACACGCTGGAAGCACTGCACGCCCTAGATT
core_23	AACTTGTGGCCGTTTACGTGCGCATACGTA
core_24	GTCCAGCTCGTGCTGCTTCATGTAGAGTGAT
core_25	CGACCAGGATGGGCACCACCCGAACCGCATCATGAGATAGTTTG
core_26	GTGAACAGTAGCCTTCGGGCATGGCAGGACC
core_27	CTCCTCGCCCTTGCTCACCATCTCCGCTATCGATCCTTT
core_28	CGAGCGGGTCCTTGAAGAAGATGGTCTTTGC
core_29	CCGCCAGTGTGATGGATATCTGCAGGTGGAGATCAATCT
core_30	GAATTCCTCACCTCGGCGCGGGTTAGTGGTT
core_31	AGCACACTGGCGGCCGTTACTAGTGCCAAAACCTGACAG
core_32	TGGATCCGATGCGGTTACCAGGGCGCCGATG
core_33	AGCTCGGTACCAAGCTTGGGTCTACGACATTCACCTATC
core_34	CCCTATAGTCTCCTTGAAGTCGGGCGAGC
core_35	TGAGTCGTATTAATTTGATAAGCGCCATTATCCATAG
core_36	CCAGTAAGGTACTCCAGCTTGTGTGGCGGA
core_37	GTAGGTCAAAGATCCGTCATGTAAGTTTACCCTAAATA
core_38	GGTGGGGACGCTCAGGATGTACTTCAATGGAAAGTCCCT
core_39	GCGGACTTGATTTTTGGTACCATGGGAACATACGTCATTA
core_40	GTGCGTTTTACCTACACGCCCGGGGGTTCGTTGGGCGG
core_41	CTTGTAGGTTTTAACTTGGAAATTTACCCTAAGTTA
core_42	TGTCGCCAACTTGGTCAAACCTCCATATATGGGCTATGA
core_43	ATGCCCTTAGTGAGGTTGGAAAGTTGATTACTATTAAT

core_44	CCCCAGGATTTCGTTCTGCGTCAAATGTCAACGCGTATA
core_45	GGCTGGCAACTAGAAAGCAATAGGCGTTACATG
core_46	GGAAAGGCACGGGGGAGAATCAAAAATTCAGCTAAGTTGGC
core_47	GACAGTGGGAGTGGCTGTTGTTCTGGTGTATGGCAGCA
core_48	GCGATTGCAATTTCCCTTTAAAGACGCAACGTGCCATCCG
core_49	CCCAGAAATAGAATGAGAAAAACAGAGTAAGAGTACTC
core_50	CTCCCCCTTGCTGTCTACGTGAACATCCAGGTATGCGG
core_51	CCCCAGCATGCCTGCTGGGGTCGAGCAGAAAGTCAATA
core_52	TCCGCCTCAGAAGCCAAACCCTAAGCAATAAAGAAGCTT
core_53	ACCACCACACCCGCCGGACGGGAAGACCCACCGTTCTTC
core_54	CTGGCAAGTGTAGCGGGAAGGGACATCTGGCTACCGCTG
core_55	GCAAAATCCCTTATAGGGCAAACATCTCAAGGGGTGGTC
core_56	GAATAGAAATTGTAAGCGTTAATATTTTGTGAGAAGT
core_57	CCGAGATAGGGTTGAGACCTTCCAGGGTCTGTAGCGGCT
core_58	GAACAAGAGTCCACTACATTTTTATGTTAAGGGAAGAAGT
core_59	ACGTGGACATGATTTAGAAAAATAAACAAA
core_60	TCCAACGTCAAAGGGCCACCTACTAAAGGATCCCTGGACG
core_61	AGGGCGATGGCCACCTGCCCAAAAATGAATTGCCGTC
core_62	CTAATCAAGTTTTTATTGTCTTATGAGTATCGAACT
core_63	AAAGCACTAAATCGGTAGAGCCGCTTAATCCAGCTCG
core_64	CCCGATTTAGAGCTTCGCTTAATCTGTCTATTGTTGCCG
core_65	TACTTTCACGAACGTGGCGAGAAAGTCACGCT
core_66	TCCTTCGCCCCCGTTTCAGCCCGACCGACTGGGGCGT
core_67	ATGGTTACGCTCGTCATTGACCGCCAAGTAGGAAAGT
core_68	ACTGTCATTGTTGCCATACTATGGTAATAGCGATGACTA
core_69	TGACTGGTGTAGTTGCAATGGGCATTGATGTACTGCCAA
core_70	AGAATAGTTCTATTAAGGCGGGCCATCCCCGTGAGTCAAA
core_71	TGCCCGCGGTCTCGGGAACCTCCATTGACGTCAATGG
core_72	CACATAGCCCAGCCACCCCGTAATCCCGTTGATTTTGG
core_73	TGGAAAAGCTCACCGAATAATCATGGGGCGGAGTTGTT
core_74	CAAACAACGCTCTGCTGAAATTTTTTAACGCACAGT
core_75	CAAGGCGACGAAATCGGCGTTAAATTTTTGTTAAATCAGCGTTAGCATCCCCCACTTGGCA
core_76	CAAGGATAAAAAAGGAACAGATGGTGACACTATAGAATAGGGCCCT
core_77	CCGGTCCAAGTGGGCTGGGCATAATGCCA
core_78	GTTTGGTTTTCTACGGGGTCAAACCTTGACAGCTCGTCCATGCCG
core_79	TTGCTACAAAACCTCACTAGGAAAGCCCGCGGCGTACGAACTCCA
core_80	CCAGTTAATTATCAACAGACAATATGTGATCGCGCTTCTCGTTGGG
core_81	GGGAAGCTCGTCTATCGTTATTGTCTCATGAGCGGATAC
core_82	TTGTTGCCTAAATTACCCACCCCTCAGGGCGGACTGGGTGCTCAGG
core_83	TCCGCCTCCCATCACCTTTTTCAATATTATTGAAGCAT
core_84	CAACTTTAAAAGTATATCCCAATCGTCGGGCAGCAGCACGGGGCCGT
core_85	GGCCGAGCGGTGCCGTGACACGGAAATGTTGAATACTCAT
core_86	GCCGGAAGTTACCAATCACCGCATGGGGTGTCTGCTGGTAGTGGTC
core_87	ATTTATCAAGGGAGCCGCAAAATGCCGCAAAAAAGGGAA
core_88	GCTCCAGTCAGCGATGTCACATTGCACGCTGCCGTCCTCGATGTTG
core_89	ATACCGCGAAGCCGCGCAGCGTTTTCTGGGTGAGCAAAA
core_90	TGCAATGTTGCCTGAGCGCGTATCTTGAAGTTCACCTTGATGCCGTGTGGCTG
core_91	CCCAGTGTGAGCAAACCGTACACGCCTAC
core_92	CAGATTAATTTACCGTATGATACACTTGAT
core_93	TGGAACGAGGCATCGCAGTTTGCCGCGCACATTTCCCCGAAAAGT
polyT_1	TTTTTACCGTAAAAGGTTGAGATCCAGTTTCGATGTATTTTT
polyT_2	TTTTTCCCGGTAAGACACGAATCGTCTTTGATCCGGCAAAAAAG
polyT_3	TTTTTCTGCTTATATAATAACTACGATACTTTTT
polyT_4	TTTTTGTAACAGGATTAGCAGAGCGAGGTATCTTATCGCGTGGTTT
polyT_5	TTTTTCCATGATATAGACGTTTCTTCTG
polyT_6	TTTTTCTTGAAGTGGTGGCCTGGCTGATCAGCGAGCTCTAGGCACGGG
polyT_7	TTTTTATTTGGTATCTGACCACCGCTTTTT
polyT_8	CTCGCCAGTTACCTTCGGAAAAAGAGTTTTTTTTTTTTGGTAGCTCTGAGTCCAATTTTT
polyT_9	TTTTTACCCACTCGTGCAGAAAGGAGCGTTTTTTTTTTTGGCTAGGGCGCTTGTCCGTTTTT
polyT_10	ATGTTGTGTTTTGTTTCGAAACTACGGCTACACTAGAAGAAGTTTTT
polyT_11	TTTTTGGGAGGCTTACAGAAAGCCCAACTGATCTTTCAGCATCTTTAGGATCT
polyT_12	TTTTTTGGTAGCGCACTGGCAGCAGCCACTGTTTTT
polyT_13	GCAAGCAGCAGCTTGCCGGTGGCGGTGCTACAGAGTTTTTT
polyT_14	CTCCCCGTTGTAGTTCAAGTGGGTTCTCTAGTTAGCCAGAGAGCTTTTT
polyT_15	TCGTTAGGACCTCCAAGGCCAGCAAAAGGCCAGGATTTTT

2.2.1.5 sc_EGFP1 20HB-LS

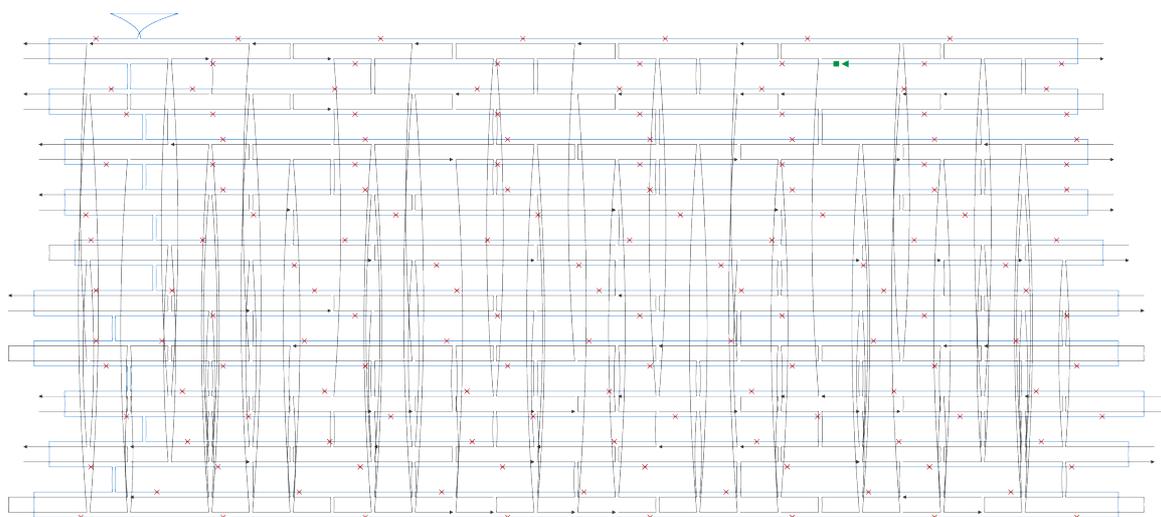


Fig. S24 | Scaffold routing and staple design for sc_EGFP1 20HB-LS. Scaffold routing is shown in blue, staples are given in black, breakpoint for addition of sequence is marked by a green square (5') and arrow (3'). Design was prepared using caDNAo v0.1.

Table S11 | Individual staple sequences for sc_EGFP1 20HB-LS.

Name	Sequence 5' - 3'
core_1	CGCGTATATCAATGAGTGAGCAACCATAGGCTTAAATTTTTGTTAACAAAAGAA
core_2	CGTAAATAACTAATGACCCCGTAAACCCGACA
core_3	CAAAACAAACTCCCATTGACGTCGGAACATAAACCCGGTTGAATACATGCCATTACCCCTAGGGAGC CCCGATTTA
core_4	TCAAACCGCGCCTACCGCCATTTGCGTCAATTTTTGTTAAGTCCCTGTAACGCGGAACCTCTCCCC TG
core_5	GCCAAAAGCTCTGCTTATATAGTCCTCGCCGGCACCTCAGAAGTGTGTGCAAAAAGCGG
core_6	GACTAATGTAAGCAGTGGGTTCCGAGTGTGGTCTGACATCCAGTCCGATCAA
core_7	AAAGTCCCTATAGTGAGTCGTATCACACTGAAATCAAT
core_8	AGTTGTTACGACATTTACTTGTGGTAGATAACGACCACGAGTGTATGGCTGGCAACTAGAAGGCACAG T
core_9	TCTAGTTATTTTCTACCACTTGATACTAGTCAATAATCAAAAATCG
core_10	TAATTTTCGAAAACCTCACGTCAATAGGGGGCGTACTTGGCA
core_11	CCCGCCCTCGCCGGACACGCTGATGGAAAGGCAACACGTCATTATCAGCCAGGCGGGCCGTTCCGAC
core_12	GGCCAGGACCCCGGTGAACAGCACCTCCCAAAAAAAG
core_13	TCATGTGGTCGGGGTAGCGGCTGAAGCACTGCACCCGATCCATCTCGAGCGCC
core_14	TTGAAGTTCACCTTGTGTACTCCAGCTTGTGCCCCAGGGGTGGTCTTTATCAGTAGTTGCCTGTGACT
core_15	CACGCTGCCGTCCTCGATGTTGTGGCGGATCCGAGGCTGGTTGTCAG
core_16	AGTGGTCGGCGAGCTGTGACACTATAGAATACCTACTCAGACAATGCGATGCAAGAGAAAGTTAGCTCC AGTCATT
core_17	GGCCGTCGCCGATGGGGGTGTTCTGCTGGTATGCGGTTACCAGGGTGTGCCCTCGAACT
core_18	TCGGGCAGCAGCACGGTCACCTCGGCGGGTCTTGTAGGTGCTGCTGGAAGCTACTAAAGTCGCCAC AT
core_19	GGGCCCTCTAGATTACTTGTACAGGCGAGTTACATGATC
core_20	GCTCGTCCGCCCCACCCACCCCCAGAATAGGGCGCTAGGGCGCTGAAGAGTCCACTATTA
core_21	AGTGGCACCTTCCAGGGTCAAGGAAGGCACAATCGGAAGTTATGGCAGCAACA
core_22	TTGTCTTCCCAATCCTCCCCCTTGTGTCTATGCCGAGAGTGATCC
core_23	ACGGGAAAGCCGGCGAACGTGGCTTTCCTCATTTTATTAGGAAAGGACAGTGGGGAGCTTGAAGTAAG TTGCTTTTC
core_24	GAAGGGAAGAAAGCGAAAGGAGCGAATGACA
core_25	ATCAAGTTTTTTGGGGGCATTTATCTTCTTTTGGGCGG
core_26	TACAGAGTTATTGAATCGAGGTGCTCTGCATAATTCTCGGGAGGGCTTACCAT
core_27	ATTGTCTCATGAGCGGATACATAGGGCGACATAAGTTAATTGGCGTTACTATGAATGGGGT
core_28	TATTTAGAAAAATAAAAAACCGTCTCAACCATTCGGTCCAGCCAGCCGGAAGG
core_29	CGCACATTTCCCGAAAGAAGTCCGCGACCCCCCATGTGTCTGCAACTTTATCCGCCTCCAGTTACC TCTTGCCC
core_30	AAAGTGCCTTTACCCTATTAATGTAAGTCCCAAGTGGG
core_31	TGTAAGCGTTAATATTGTTCCAGTACGGGAGTTCCTCAATATTAATTGTTGCCG

core_32	ATCAGCTCCTGTTGAGCAAAGGCGGGCCATTTACCG
core_33	CGGAAATCTGGTAGCCGTCGTGCCGTTTACCTTCAGGGTCAGCTTGTTGTAGT
core_34	GGGAATAATTTGAATGGAAGCTCCCTCGTGCCTCTCCTATTTACCG
core_35	GCAAAAAGGTGAGTATATCAGGGCGATGGCCCACTACGTGAACCA
core_36	CAAAAACAGGGTCCGGGACTATAAAGATACCAGCGTTATATATGGAAATGCCGCAGATTA
core_37	GGGTGAGAGTGTATGGGACTCCAACGTCAAAGGGCGAACAAATAGGGAAGGCAGCTATGACTCCACCC ATTGACG
core_38	TCTTTTACACCTAAATACGCTCAAGTCAGAGGTGGCGAATTGATTAGCGTTTCT
core_39	CCCACTCAATTCGCGTCCGCCCCCTGACGAGCATCACATGTCAAACTGATC
core_40	CCGTAAGATGGCCGCCTCACGGGTCGAGAAGATGAAGTCGCCGTCAGCTCGACCAGGA
core_41	GAGTTGCAATGCTTACTTTGATCGCCAGAGACCGCATCACCATGGTAATAGCGATTATGATA
core_42	TTTAAATCGTCGTTTAGTTCGCACTGGGTGCTCAGGTAGTGGTTGCGGCGGGTCACTCA
core_43	GAGTAAGTGGTATGGCTTATTTCGTAACCACACACCCGCCGCGCCTGCTAACTCCAGCAGGACCA
core_44	TACGATACTTACTGTCTCATACTCAGGGTTCTGCCGCTTACCGGATACCTCTTCGGGAA
core_45	TGACTCCCGGTGGTTGGGGCGGGGAGACTTGGAAATCCCGTGAGTCAATGGATGCAAGCA
core_46	TCATCCACAATAAACCTCCGATCATCAGCGAGCTCTAGCATTTAGG
core_47	TATTTGTTGGGCACCGCACGGGACGTTGCCATGTTGCCGTCTCTTGAAGTCGATGCCCTTCAGCT CG
core_48	GATCTGTCCGCGCAGCCGTACACTATCCACGCCATTGATGTAAGTACTCAGTTACGATCTCAA
core_49	ATATATGAGTGGAAAGGATAAGCCAACGTAGATGTAAGTCCAAAGTAGGTCATTGA
core_50	GAAGATCATCAGTACTGCTCAGGTCAGGGTGGTACAGAGGGTGGCCGAGCGATCTCAGCCTGAGAA T
core_51	GGGGTCTGTTTACGAGGCGTCAATTTGGAACGCAAGTGTAGCGGTACGCTGCGCAGCTCCGTAATAC CG
core_52	ACGCTCAGTAAACTTGATGGATATCTGCAGAATTCCAG
core_53	CGTTAAGCGATGTAAAGCAGAACTAGACCGAGATAGGGTTGAGTGTGTTTGAAGTGCACCC
polyT_1	TTTTTAAAAAGGCCGCGTTGCTGGCGTTTTTAAAGCCAGATCCAGTTGGATTTTG
polyT_2	TTTTTCTTTCTCATAGCTCACGGGGTTCGTTTCAATAGGTAGCTCTTGATCCGTCCTGTTG
polyT_3	TTGACGTCAATGGGCTTCGGTGCATTTTCTCCAAGCTGGGCTGTTTTT
polyT_4	TTTTTGTTCGGTGTAGGTCGGCTGTAGAAAGAGTTTTTCTTGAAGTGGTTTTTT
polyT_5	ATAAGGTCACTACTGGGTTTTTTTTTTCATAATGCCACAGGAACCGTTTTTT
polyT_6	TTTTTGTACCAAGCTTGATTATCAAAAAGGATCTTTTTTTTTTTTACCTTAGATCCCAACGTTGTTGCTTTTT
polyT_7	TTTTTGTGCACGAACCTGTCTGCTGAAGCCAGTTTTTTTTTTTACCTTCGGAAGTATCTCATTTTTT
polyT_8	TTTTTACGTAGCCTTCGAGTTTTCGTTTTTAAATCATTGGAACCTACCG
polyT_9	TTTTTTGCGCCTTATCCGGTAAGATATAGACGTTGTGGCTGCCGTAGGACCGCGA
polyT_10	ATGCCGTTCTTCTGCTTGTGCGGCGCAATGATTGGCATCGCCCTCCGTTTCAGCCCCACCGCTTTTTT
polyT_11	CAGGGCGGCAGTTAATGGCATGGCTACTAGTGGATCCGAGCTCGTTTTT
polyT_12	TTTTTGTGGGGTCTTTGCTTGTGATCGATCGTGGACATTCGAAATCCCTTATAAAT
polyT_13	TTTTTCATAGAGCCACCGCATCCCAGCATTTAATGTCTGTACGCGTGTCTATAAAAATGTCATGAG GGTCTCCC
polyT_14	TTTTTCACTGGTAACAGGATTGTAAGCACTAGGGGACTTATCGCCACTGGCAGCAGCTTTTTT
polyT_15	ATTTTTTAACCAATAGGCTTTTTTTTTTTCGAAATCGGCACCTCAGAAGCTTTTTT
polyT_16	TTTTTGAAAACCTCTCAAGGATAACGTTCTTCGGGGCTTTTTT
polyT_17	TTTTTGGCCCTAACTACGGCCAGCAGAGCGAGGTATTTTTTTTTTGTAGGCGGTGCGCGTGGCGTTTTT
polyT_18	TTTTTCATTGCTACAGGCCGCTTCTTTTTT
polyT_19	TTTTTTGGTATCTGCGCGCCCTAGAAGACCCGGTAAGACACGGAGGGGCAAACAACAGATCACTCATGC CCTAAA
polyT_20	CCAGTGCTCATCTATCGTCTTGGTCTTTTTTTTTTCAAACAGTATTTTTTT
polyT_21	GAAGTTTTGCGGCCGTGGACTTGAAGAAGTCTTCCGTCGTCTTGAAGAAGATGGTGCCTCCTGGTT TTT

2.2.1.6 sc_EGFP1 20HB-LP

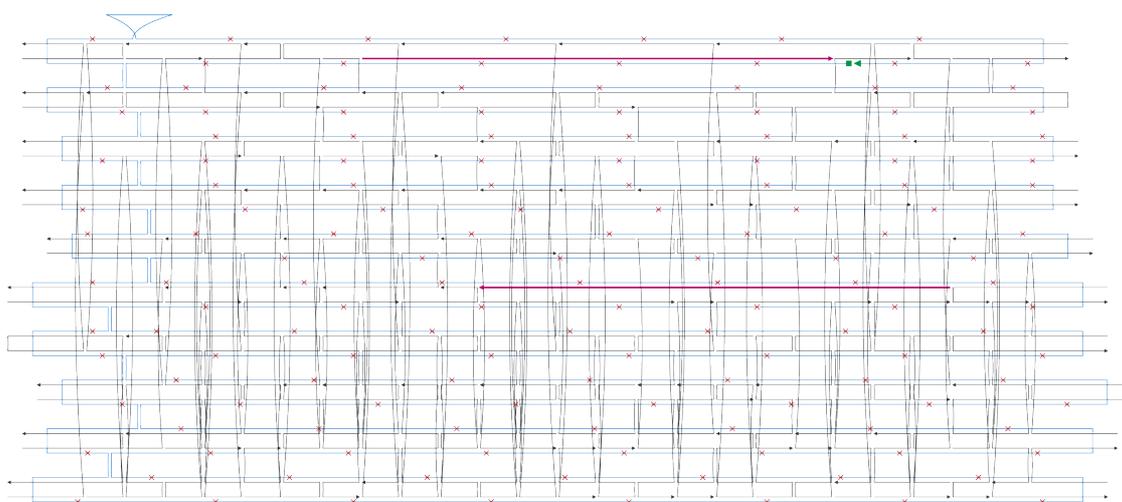


Fig. S25 | Scaffold routing and staple design for sc_EGFP1 20HB-LP. Scaffold routing is shown in blue, staples are given in black, breakpoint for addition of sequence is marked by a green square (5') and arrow (3'). Long 93-mer 'primer' sequences at the 5' promoter and 3' polyA regions flanking the EGFP gene are given in pink. Design was prepared using caDNAo v0.1.

Table S12 | Individual staple sequences for sc_EGFP1 20HB-LP.

Name	Sequence 5' - 3'
core_1	GCGTTTTTAATTCGCGTTAAATTTTATAAATAGCAGAA
core_2	CAATGGGCTCTTCCTTAAAGAGTTGGGAGGGCACTCGCCCTCGCCCTCGCCGGA
core_3	ATTTACCAAGTCCCTATTGGCGTTGACGTC
core_4	AAGGCCAGTAATGCCAGGCGGGCCCAAGTAGGGGTCTCCC
core_5	CGTCATTATTGACGTGTATTGATTTTGGTGCCAAAACAACTCCCATTACTATG
core_6	ATATGATACACTTGATGTACTGCGATGTAC
core_7	CGTCAATAGGGGGCGTATAGCGAATAAGCCAGTAAGCA
core_8	CCCGTGAGACCGTACACGCCTACCTCCAGTC
core_9	TCAAACCGCTATCCACGCCATTCAAGTGGGAAAAAAG
core_10	CGACATTTTGATCCGGAACATATTGGGCGGTGAGCCAGCTCTCCT
core_11	TGGGGCGGAGTTGTTACACGCTGACCGTCGTGTACCGCGA
core_12	GCGTCAAAATGGGGTGGAGACTTGAAATCCATTGACGTTTTTTGTTAAAATGCC
core_13	GCCAGAGAGCTCTGCTGAACAGCATCAGTGGCCGAGCG
core_14	TCTAGTTATGCCAAAACCGCATCACCATGGTAACCTGGCTTTTCTACAAGTATTCTTGCCC
core_15	TATAGTGAATCCAGAAGTTTTTGGAAAGCTTCGTCGTT
core_16	CCGTTTACGTCGCCGGCCATTTTCGGTGGTTTCAATGGA
core_17	TGGGCACCACCCGTTATATAGACGCGCAGCAGTTTACCCTAAATACTCCACC
core_18	CCTTGCTCACCATCTCGTGGGTTCTTTGATC
core_19	GAGCGGCCGTAACCTTCCGCCTCCGTTCCCA
core_20	AGCGGCTGAACCTTTATGGTCTGACGAGTTGCCTTCAGCA
core_21	GTGCTGCTTGATGGATATCTGCAGAGTCGTAT
core_22	TGTGGCTTACCTTGATGCCGTTGGGGCAAAGTGTATAATCGGA
core_23	GTTGTAGTTGTAAGTCCAGCTTGTAGCTTGCCGCTCCAGATTCATCCA
core_24	GCCCCAGGCCGCTCCTCGATGTTGTGGCACAGTCTCCGATCAGAGCTTG
core_25	ATGTTGCCGCTCCTCCTTGAAGTCGGTGGTCACCGGAAGG
core_26	GATGCCCTCTGCTGGTAGTGGTGCATTTAGG
core_27	GGGTGTTTCAGCTCGATGCCGTTCCACCAGG
core_28	CAGCACGGGGCCGCTAGATTAACGATCAAGGGCGCTA
core_29	TCGGGCAGCTCGAACTTACCTCGGCGCGGGAAGAAGTCTGTTGCCGAAATCAA
core_30	ACTGGGTGCTCAGGTAGTGATCTGGTATGACGCTGCG
core_31	GGCTGGCAACTAGAAGGCGGATCTCACGGGTGGTG
core_32	GATCAGCGAGCTCTAGGCGAGCTGCAGCCAGCGAGGGT
core_33	CTTGACAGCTCGTCCATGCCGAGAGTGGTTGCTATTAAT
core_34	CCGGCGCGGTCACGAACTCCAGTCTTTGCT
core_35	AAGCACTACACTCATGGTTACTA
core_36	CCGATTTGTTGTCAGCCGTAAGATAGTTGCCCTGGTAG
core_37	AACGTGAAAAAGCGGCTCAACCATCTCAGCGCAGATTACCTCCC

core_38	TCACCTAATCAAGTTTTATTGAACTCATACGGGGGTCG
core_39	CTATCAGGGCGATGGCATGAGCGGGGAATAA
core_40	TTTGGAAACAAGAGTCTTCCCGATCTTTTAC
core_41	TGTTCCAGGGGCGCTGGCAAGTGTAGCGGTGCTTCATT
core_42	TAGACCGAGATAGGGGTTAATATACCCACTCGTGCACCC
core_43	AATCGGCAAAATCCCTTTGTTAATCTTACCG
core_44	TCAGGGTTATTGTCTCCACTACCATGCCATAAGTAAGGACCCACGCTTGAAGT
core_45	TTTGAATGTATTTAGAAGGGCGAAGGTGAGTATTAGCTCCAATAAACCACGCTG
core_46	GGGTTCCGCGCACATCACTATTAAGTGTATGACATGATCCAGAAGT
core_47	AAAGTGCCAAAATCGACGCTCAAGTCAGAGGTGGCGAA
core_48	ACCTAAATTGTAAGCTTGAGTGTGGCGTCACAGCTCCGATCCAGT
core_49	GGAAGGCTGCAAGCAGATCTGTCGACCAGGAGGGCCAGGGCACGGGC
core_50	GCGTTTCTGAGAATAAGAACGGAAGGGAAGAAAGCGAAAGGAGC
core_51	TTTACCAGATCTCAAATGCTTATCCTCGCCACGCCGTAGGTCAG
core_52	TGCTTTTGCAAAAAGATACATATCCCCCTGGAAGCTCCCTCGTGCGGCGGGCCGGGGCGAC
core_53	CTGTGACTAAACCGTACGGGGAAAGCCGGCGCGATGCA
core_54	ATACGGGATCTAAAGTGACGCTCATAATTTCTGTGACTAATACGTAGATGTACTGCATTTACC
core_55	TAATACCGCGATGTATTTGTTAACCATAGGCTCCGCCCCCTGACGAGCATCAC
core_56	TTGGCCGCCAACAGATGAGTGGCACCTTCCAGGGTCAAG
core_57	TTGTGCAACGAGAAAAGTGGACTCCAACGTCAAAAATAACAAAAACA
core_58	CCCCATGTGACACTATAGAATAGGGCCCTCGCCGATGGGGTCCCTGCAAGCACTG
core_59	GGCGAGTTCGGCGACCAGTTACCAGAAGATC
core_60	CATCGTGGCTTTAAAAATTTAAAGATTITG
core_61	TAGATAACTAATTCTGTTGAATAGCATTTAGTTCGACCCCTGCCGCTTACCC
core_62	TATTTCTGTTTATCAGCTTCCGGTCCGAGGCTATTTCTCATTTTATTAGGAAAGG
core_63	AGGCACCTAAGTCATTTGGGTGAGACAAATAGACCCGACAGGACTATAAAGATACCAGGCGTT
core_64	TTAAAAATGCACACTGGGCATGGCGGACTTGTCTTGTAG
core_65	GGCAAACAACGGAAATCTTACTGTGTGAACCAACCTAAAGGGAGCCACAGTGG
core_66	AACCACCGTACTCCACTTGTGGCAGATGAACTTCAGGG
core_67	GGGGTCTATATGAGCCAGTGTATGTGGTGGTGGTGTGTCGCC
core_68	GTGGAACGGTCAATTGACGCGTATATCAATGA
core_69	AAAACCTCATCCAGTTCGCCACATCAAAGAACGTAACCACCACACC
polyT_1	TTTTTGAACCGTAAAAAGGCCGCGTTGCTGGTGAGCAACTGTTGAGACGTTAAG
polyT_2	TTTTTGCCTGGCGCTTCTCATTCTCCCGCGGTGTTTCAATATTTTGGG
polyT_3	TTTTTGTATCTCAGTTCCGGTTAGCTCA
polyT_4	CAAGCTTGAAAGTCCCATAAGGTCATTTTTTTTTTGTACTGGGCACAAAAGGCCATTTTT
polyT_5	TTTTTTCGAGCTCGGTACGTCATGAGATTATCAAAATTTTT
polyT_6	TTTTTTTTTTGGGCTGTGTGCTTATCTGCGCTCTGCTGATTTTT
polyT_7	TTTTTGCTCCTGGACGTCAGTTAATCTAGATCC
polyT_8	TTTTTTTTTTGACCGCTGCGCCTTATCGGCCATGATATAGACGTTTCACTTGAATGA
polyT_9	TTTTTCGCTTCTCGTTGGGGCAGGACCACTACAGGTTAATGTC
polyT_10	TTTTTCTGAGTCCAACCCGGGGCACGGGGGACTTCTGC
polyT_11	TTTTTTCAGAAGCCATAGAGCCCACCGCATCCGCCGCGC
polyT_12	TTTTTGCAGCAGCCACTGGTAGAGGTGCCGTAGAATAAGACACGACTTATCGCCACTGTTTTT
polyT_13	TTTTTCAATAGGCCGAACATTCCGCCTTTTT
polyT_14	TTTTTTTCGGGGCGAAAACCTCCATTGAAAACGTTCTTTTT
polyT_15	TTTTTGAAGTGGTGGCCGTCACAGGATTAGCAGAGTTTTTTTTTTCGAGGTATGTATTCGGGAATTTTT
polyT_16	TAATGGCAGCACTGCATACGATACGGTAGCTCTTGGAAAGTCCCGGGTCTGCTCCAAGTTTTT
polyT_17	GTGCTCATTCAAGGAATCAGCTCATTTTTTAACTTTTT
polyT_18	TGTCACGCAGAGTAAGCAGGGCGGTTGCCGTCGTCCTGAAGAAGATGGTGTCTTTTT
polyT_19	TTTTTGTGTTGCCATTGTGTGATCGTTTTT
polyT_20	TTTTTACAGTATTTGGTACCCGGCTACACTAGAAGATTTTT
polyT_21	ATCTGGCCTTGTCCGGTAACTATCGTTTTTT
polyT_22	CCAGTGCTGCCGTAGGTGGCAGAACCCCGTTCAGCTTTTT
polyT_23	TAGTTCGACGCTTCCGGCGGCCGTTACTAGTGGATCTTTTT
polyT_24	TTTTTAGGATCTTACAGTTTTCGCAACTTTTT
polyT_25	TTTTTAGCCAGTTACCGCTGTAGTTTTT
polyT_26	CTTCGGAACACTACAGAGTTCTTTTTTT
LP_1	GTAAGTTATGTAACGCGGAACTCCATATATGGGCTATGAACTAATGACCCCGTAATTGATTACTATTAATAA CTAGTCAATAATCAATGTCAA
LP_2	CCCAGCATGCCTGCTATTGTCTTCCAATCCTCCCCCTGCTGTCTGCCCCACCCACCCCCAGAATA GAATGACACCTACTCAGACAATG

2.2.1.7 sc_EGFP1 20HB-LPv2

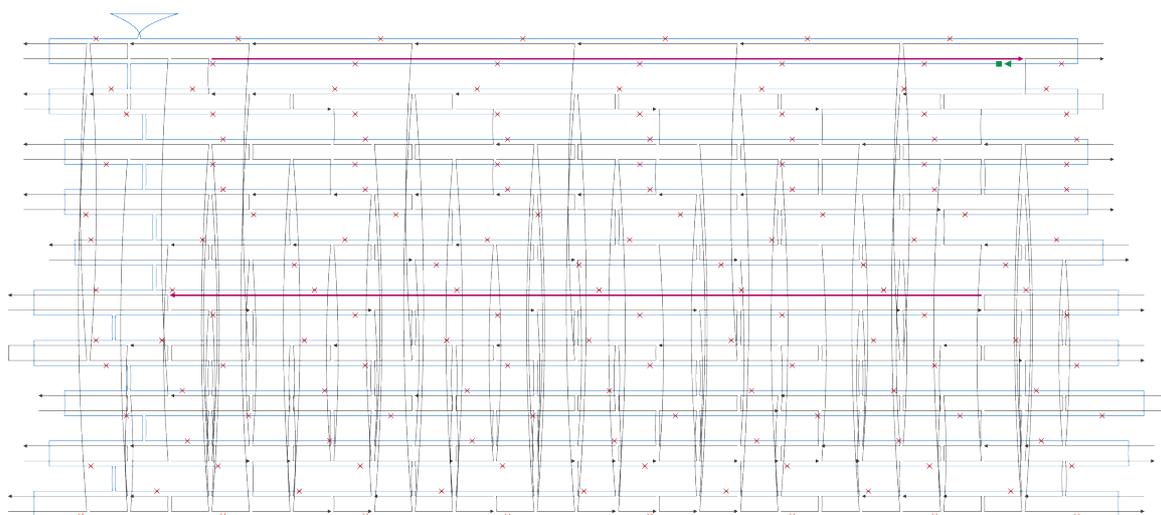


Fig. S26 | Scaffold routing and staple design for sc_EGFP1 20HB-LPv2. Scaffold routing is shown in blue, staples are given in black, breakpoint for addition of sequence is marked by a green square (5') and arrow (3'). Long 154-mer 'primer' sequences at the 5' promoter and 3' polyA regions flanking the EGFP gene are given in pink. Design was prepared using caDNAno v0.1.

Table S13 | Individual staple sequences for sc_EGFP1 20HB-LPv2.

Name	Sequence 5' - 3'
core_1	GGAACATAACGGAAATGCTGAAGCCGTCGTG
core_2	CGTCATTAATTGGCGTTACTATGCGCTTACGACATTTTGGAAAGTCCCGTTGATTCAATGGA
core_3	CGTCAATAGGGGGCGTACTTGGCTCAAACC
core_4	CCCATAAGGTCATGTATGACTAATATCCGAGCTCGGTACGCCAGTGCTAGATCC
core_5	AATGGGGTAGCTCTGCTTATATAGACTTGTGGGGCACCTGACCCACG
core_6	GGAGACTTGGAATCCCCGTGAGATATGATATGCAAGCA
core_7	CGCCTACCGCCATTTGCATCGCCTTCATCCAATCTGGCC
core_8	CCGTACACAAAACAACTCCCATTGACGTCCAGTTTACAACCACCGTGGGTGAG
core_9	GTAAGCAGTGGGTTTCGACCAGGAGTCTGACAATAAAC
core_10	ATAAGCCAGCTATCCACGCCATTGATGTACTTCATTGAGATCTCAACGATGTAAGTGTATG
core_11	TATAGTGAGTCGTATTCCTCGCCAAATCAATCAGAAGTG
core_12	TTCGGGTCAGCTTGCCGTAGGTGGCGTCAATTCGGAAAAGTCCCT
core_13	CGCCGGACACGCTGAACCTCCCAGGCAAACACGTAATACTCCACCCATTGACGTTTGGTGC
core_14	GTCGCCGTCCAGCTCTCTAGTTATTTTGTTCACTTGATGTACTGCCAAGTGGG
core_15	CACCCCGGTGAACAGCTAATTTTCGAAAAAAG
core_16	TGATGGATTGCGCTCCTGGACGTCTCGAACT
core_17	GGCATGGCACCATCTCGAGCGGCCCAAGCTTG
core_18	CTTGTCCGATGTTGTGGCGGATCAGGGTCAAGTTGTCAGGGTCCGAG
core_19	GCCATGATATAGACGTTGTGGCTCGAGGGTGTACCGCGAATCTCAGC
core_20	GTTGTAGTAGTGGTCCGGCAGCTGCAACAGAT
core_21	GTACTCCAGCTTGTGCCCCAGGAAGCACTGTTTATCAG
core_22	ATGTTGCCGGGCCGTCGCCGATGGCGAGGCTG
core_23	TCAGCTCGCTCAGGTAGTGGTTGTGACACT
core_24	CAGCACGGTCTCCTTGAAGTCGATGCCCTGTGCTGCTGCCGAGCG
core_25	ACTGGGTGATGCCGTTTACCAGGGTGTGCCAGCCTTCGCCGCCTCCTTTTAA
core_26	GTTGGGGTCTTTGCTTTGTACAATCGTGGGGGCGCTG
core_27	TTTTTTGGAAGTAAGTTGGCGAG
core_28	CGTAACCATCATTTTTTAACCAATTTGTAA
core_29	GCGATGGCCCACTACGCTCATACTGGGCGAC
core_30	TAGACCGAGAAAGCGAAAGGAGCGGGCGCTATGTCACGC
core_31	AAATCCCTTATAAATAAAGTGCCTGTTGAGATCCAGTT
core_32	TTTCAATATTATTGAAAAACCGTGCCTGCATTCCGGTCCCAGTGCTCCGTCCTC
core_33	GCATTTAGGAAGGCAAATGCCGGTAGCTC
core_34	ATTGTCTCATGAGCGGAAGAACGTCCGTAAGACCCCATGCTCACCAGGTGCTGGT
core_35	ATACATATTTACCAGCGTTTCTGGTAGC
core_36	TATTTAGAAAAATAATGTTCCAGCTCAACCAGTTCCCAACAGCCAG

core_37	ACAATAGGTGCACCCAAGTATCGCAGATT
core_38	CGCACATTTCCCCGACAAAAGAACGGCGACTCGTCGTTGTCCTGC
core_39	TTCAGCAGGTGAGTATTTGAAAGCCGGCGAACGTGGCGAGAAAG
core_40	TTCGGGGCGAAAACCTCGACGCTCAATCTTCAC
core_41	TTACTGTCAAAAACATCAGGGTTGGACTATAAAGATACCAGGCGTTTCCCCCTG
core_42	CATGCCATGGACTCCGGGAGCCCCGATTTAGAGCTTG
core_43	CGAGTTGCTTAAAAATCTTTGATCGGTCTCCCCGCATCACCATGGTAATAGCGACTGGGCA
core_44	TCTTGCCCTTACCGACCTAAATAAGGCCGCGTTGCTGGCGTTTTTCCATAGGC
core_45	TAATACCGAAAAAGGGTGAACGACTAGTGG
core_46	CTCCGATCGGAAGGCACGGGGAGGGGCAACACGCTGGCAATGAGGCCAGG
core_47	TTAGTCTTAATTCTCGATCTGTCTTGATCC
core_48	AAAAGCGGACCCTAAAAACGTCAAAGGGCGAA
core_49	TTGTGCAAGGCTGGCAACTAGAAGGCACAGTGGGTGTTCTCCAGACACGCCG
core_50	ACATGATCTGCTTTTAAATGCTTAGGTGGTTGCCAGAG
core_51	GGCGAGTTACGGGGAACAAGAGTCCACTATTA
core_52	CGATCAAATCAGCGAGCTCTAGCATTAGGTCCGGGCGAGCCGGAAGGTCATGTGG
core_53	CAGTCCGAGTCAATATATATGAACGCGCAG
core_54	GCTTCATTGAAGGGAAGATAGGGTTGAGTGT
core_55	TGGTATGATAGAATAGGGCCCTCTAGATTACCAGGGCGGAACTTTAT
core_56	ATCCAGTCTACAGGGCGCTCGTCCATGCCGAGAGTATCTGTGATCG
core_57	TAGTTGCCCTCATGGGGGAATAACTTCCCTTGAAGCTCCCTCGTGCCTCTCCC
core_58	TATTTGCTCGCCCTGCACGGGCAGCTTGCC
core_59	ATCAGTGACCGTTTACTAGGTCAGGGTGGTCA
core_60	CAGTTACCCTGTGACTTCTTTACTTTGAATGACGCTCAAGTCAGAGGTGGCGAAACCCGACA
core_61	GTAACCTTTGGGCACTCGGGGTAGCGGCTG
core_62	CTAAAGTCTGAGAATACCCACTCGGGTTCGTCGCCCCCTGACGAGCATCACAAAAATCG
core_63	AAAGAGTTGCAAAAAATTATGGCACTATCAGGGTCCGTAAGCACTAAATCGGA
core_64	GAAGATCGAAGTTTTCTTGTCTCGGACTTGAAGAAGTC
core_65	TTTTCTACTAATGCCAGGCGGGCCATTTACCGGCCAAAA
core_66	GGGGTCTTCAAGGATGGCGTCAAAATCGGCAGCAAGTGTAGCGGTC
polyT_1	TTTTTAGCAAAAGGCCAGCAAAAGGCCAGGAACCGTAAATGTAAGCGTTAATATTAGGCCGA
polyT_2	TTTTTGTCCGCTTTCTCCCTGCTTACCGGATTAGGTTGAATATGAACCA
polyT_3	TTTTTCTTCTCATAGCTCATCGGGAA
polyT_4	ACGTAGATGTACTGCCAATTTTTTTTTTGTAGGAAAGTTCAATGAGTGTTTTT
polyT_5	TTTTTCTGGCGGCCGTTAAAACTCACGTTAAGGGATTTTT
polyT_6	TTTTTTTTTTGTGTAGGTCGTAGACACTAGAAGAACAGTTTTTTT
polyT_7	TTTTTGTGCTCCTTGAAGGAAGCTAAGATTATC
polyT_8	TTTTTTTTTTTGCACGAACCCCGCCTTGATGCCGTTCTTCTGGGTGGTGGCTTACC
polyT_9	TTTTTACTCCAGCAGGACCACCGGCGGCTTTGCGC
polyT_10	TTTTTGCCTTATCCGGTAATGGCACCTTCCCTTGAAGT
polyT_11	TTTTTCGCGCTTAATGTCACATTCGCCCTCAACGCTGCGAACGTTGT
polyT_12	TTTTTAAAGACAGACTTATCCCCTAATCAAGGAGCTATCGTCTTGAGTCCAACCCGGTTTTT
polyT_13	TTTTTGTAAATCAGCCACACCCGCTTTTTT
polyT_14	TTTTTGTGCTCATCATTGGAAAGCAGAAGTTAAAAATTTTT
polyT_15	TTTTTGTAGGCGGTGCTTTCAGCCACTGGCAGCAGCTTTTTTTTTTCACTGGTAACAGGATACCTTTTTT
polyT_16	ACACGCAGTGTATCATGACTCCCCAGTTACCTGGGCGGAGTTGTGTAGGTATCTCAGTTTTTTTT
polyT_17	TACGGGATGCCATTGCTATTAATCGCTTCTCTCACCTCGGCGCGGGTCTTGATGTTGCCTTTTT
polyT_18	CGCCACATAACGTTCAATTCGCGTTAAATTTTTTTTTT
polyT_19	TTTTTCGCCAGTTAATAGGGTCACGATTTTTT
polyT_20	TTTTTCTAACTACGGCTATATTCTTGAAGTGGTGGCTTTTTT
polyT_21	ACTACGATTCATTACGCCGACCGCTTTTTT
polyT_22	ACGGGAGGCAGATGAACTTCAGCTCCAAGCTGGGCTGTTTTT
polyT_23	TGTTGCCGGAAGATGGATCTGCAGAATTCAGCACATTTTTT
polyT_24	TTTTTTTTTGGTCATGGAGTAAGTAGTTTTTTTT
polyT_25	TTTTTATTTGGTATCGCGTGGCGTTTTT
polyT_26	TGCGCTCTCAGAGCGAGGTATTTTTT
LPv2_1	TTGACGTCAATGGGCGGGGGTGGTGGGCGGTGAGCCAGGCGGGCCATTTACCGTAAGTTATGTAACGC GGAACCTCATATATGGGCTATGAACTAATGACCCCGTAATTGATTACTATTAATAACTAGTCAATAATCAAT GTCAACGCGTATA
LPv2_2	GAAGCCATAGAGCCCACCGCATCCCCAGCATGCCTGCTATTGTCTTCCAATCCTCCCCCTTGCTGTCCT GCCCCACCCACCCCGAATAGAATGACACCTACTCAGACAATGCGATGCAATTTCTCATTATTATTAG GAAAGGACAGTGG

2.2.1.8 sc_EGFP1 20HB-Circ

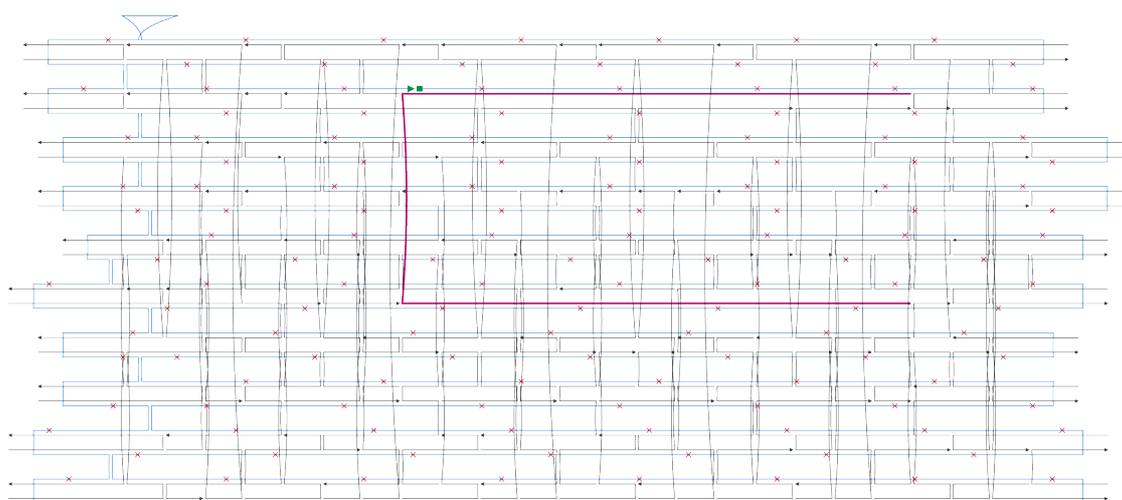


Fig. S27 | Scaffold routing and staple design for sc_EGFP1 20HB-Circ. Scaffold routing is shown in blue, staples are given in black, breakpoint for addition of sequence is marked by a green square (5') and arrow (3'). Long 200-mer 'primer' sequence linking the 5' promoter and 3' polyA regions flanking the EGFP gene is given in pink. Design was prepared using caDNAo v0.1.

Table S14 | Individual staple sequences for sc_EGFP1 20HB-Circ.

Name	Sequence 5' - 3'
core_1	TCAGAGGGGCGAGAAAGGAAGGATAAATCA
core_2	AGATACCTTAGAGCTTGACGGGGAGTGT
core_3	TACCTGTCAGGCGGGCCATTTACCTGACGTC
core_4	ATCAATGAAAAATCGACGCTCAAGCTTATCCG
core_5	TACGGCCATTTACCGTCATTGACGTCAATAGGCAAAAGG
core_6	GGGGCGTATCCATAAGGTCATGTTGGGTCTC
core_7	CTTGGCATATGATACACTTGATGTACTGCCAAATACGTAGATGTACCGATAAGC
core_8	AGTGGGCAGTTTACCGTAAATACTCCACCCAAAACCGCATCACCATTAGCCAGAAGTCGAGG
core_9	ACCGCTATCCACGCCACCGTACAACAACAGATGGGGG
core_10	GGGTGGAGACTTGGAATGGGGCGCCAGGGTAGCTGCACGAAGCATTAAAAGGG
core_11	TCTTCCGAGCTCGGTACCAAGCTACTGGGCA
core_12	CTTATATAGACCTCCATTGATGTACTGCCATTGACGTCAATGGAAAGTCCCTATTGGCGT
core_13	ATATCTGGAGTCGTATTAATTTTGCCAAGTTTCCGCT
core_14	TCACCATCAGTGGGTCTCTAGTGGTAATAGCGATGACT
core_15	TACGTGCCCCGCCATTTGCGTCAAATCCCCGTGAGTCAATACTATGGGAACATACGTCATTAT
core_16	GTGCGCTCCCTCGGCGCGGTCTACTCTCA
core_17	CTGGACGTAGCCTTCGGGCATGGTGTGATGGGGGTCTTCTATAGAAAATCCCTTGAAGAAAG
core_18	CGGACTTGCGGTTCCACAGGGTGTGAGTTCGA
core_19	AAGAAGTCGTGCTGCTTCATGTGGCCCTTGACAGGTAGTGTGATCAGATAGGGTTAAAGCCG
core_20	GTCGGGGTCTCCTTGAAGTCGATGTGATCTTC
core_21	TAGGTCAGACTCCAGCTTGTGCCCTTTCTGGG
core_22	GCACGGGCTGATATAGACGTTGTTGCCGCAA
core_23	TAGTTGTGGTGGTCACGAGGGTGGGCCAGGTGGCCGTTGTGCGGCAAGGAAGCGTCAAA
core_24	CCAGCAGGCAGCTCGCATTTTTCTGGCAAGAATTAAGACACGACTTATCGCCACTGGGTA
core_25	GTTGTGAAACAAATAGCATCTTACCGAGTTGCTCTTGCCCGCGT
core_26	CGTCGCCGATGGCTGAGAGTCCAAAGGGAG
core_27	GCTGCCGTGTCGGCCAAGCTTGCCGGTGGTGCAGATGAA
core_28	CCTCGATCCTCGCCGTACGACATTTTTGAAAA
core_29	TACTTGTAAACCATGTGATTTGTAGGATCTTAAGTGCTCATCATTGGAAAACGT
core_30	CTCTAGATCCTATAGTCAGAATCCAGCACAC
core_31	AGGTGACATGCTCAGGAAAAGTGTGTAACCCGATAATACCGCGCCACATAGCAGGGTATGGC
core_32	GCAACTAGAAAGGCACGAGCTCTGGGTGAACAGCTCCTC
core_33	GCACGGGGGAGGGGCACGCCTACGTCCAGCTCGACCA
core_34	GGCACCTTGAGTTGTGACACGCTGAACCTTG
core_35	AACCACCACCAGGAACATAGGCTCCGCCCCCGGTAA
core_36	CACCCGCTAGGGCGAACCAATAAGTAGTTCTCACGCTCGTCTTAACTTTAA
core_37	TGTAGCGGGTTTTCCCGTAAAAAGGCCGCG

core_38	GAACCTACTATTAACACCAGCCTAGCTCCTTCGGTCCT
core_39	AGGTGCCGGCCTGACTGCTGTAGGTATCTCAGTTCGGTGCTCGTGCGCTAAATCG
core_40	TGGGGTCGGGGCGAAAAACGCTCACAGTAAAGTTGGCCGCGAG
core_41	TAAAATTCCTGTGGTGGCCAGTTAATAGTTTTCTTTTA
core_42	AATTGTAATCGGCAATAGGGCCATGTACAAGGAAAG
core_43	CCACCTATTCATTCAATTGTTGCCGGGAAG
core_44	AGGGGTTGAGTTAGCAACTTTATCCGCC
core_45	AGAAAAATGGCAGCAGTGCCGTCAGCGGCTGAAGCACTGCACGCCGGGATGGGC
core_46	TATTTGAAAAGCGGTAGCCGGAAGGGCCGA
core_47	CGGATACATGAGCAAACCAAGTCATTCTGAGAATAGTGTGTGCAAAATGTATTTTTGGAACA
core_48	TATCAGGGTTGTCCAGACGGCTCCAGATTTATC
core_49	ACCGCTGTTGAGATCCGCCCTCTTCTCGTT
core_50	ACTCGTGCACCCAACCCCTTCAGGGTGCTC
core_51	TTACTTTTACCAGCGCAGGATGTCACGGGGCACCACCCC
core_52	TACTCAAACAGGAAGGCAAAAGGCTGTTGTGGTAGTG
core_53	AGATGCTTTTTCTGTGACTGGTGAGCCGATCGTTATTGTGACTCCAA
core_54	ATCCGTAAATAAGGGCGACACGGTTCTGCTT
core_55	GATCAAGGCCGCGCACAGACCGAGCGAGCTCTAGCATTTTCAGTAAGCTCGAGCGGCCGCCAG
core_56	GTCTATTAGCTCCGGTTCCCAACCAATACGG
core_57	GTGGTCCTCATGATCCCCCATGTTATGCGGCG
core_58	GCTGCAATGATACCGCACTACGACCTAATC
core_59	ATGAAGTTTTAAATCCTAGAGTAGGCCGAAGCGTTAATATCGCGGAACCTCA
core_60	AATCTAAACGAAAGGACATCACAGTGAGCAAAAGGCCA
core_61	GTATATATGTAACATATCGTCTTGAGTCCAACCTGACGAGCGGGCGCGCTTA
core_62	GAGTAAACTTGGTCTTCCATCCAAAAGAATATTTCCCGGGCGACTGCTCGATG
core_63	GACAGTTAGCGAACGTTGGCGAAACCCGACAGGACTATAAACGAACCC
core_64	CCAATGCCCGTTTCAGCCCGACCGCTGCGC
core_65	TTAATCAGTGAGGCACGCGCAGAAGTTCAGT
core_66	CTATCTCACCCCGATAGGCGTTTTCCCCTGGAAGCTCCTAGGTCGT
core_67	GCGATCTTCGCTCCAAGCTGGGCTGTGTGC
core_68	GTCTATTTCTGTTTCATCAGCAATAAGAACGTGCTCATGAGTGTCTGC
core_69	CATAGTTTAAAGCACTCTCCTGTTCCGACCCTGCCGCTAGCTCAC
core_70	CCCCGTCGTGTAGATAGAGACCCAACCGTCTTATTATT
polyT_1	TTTTCTCCTCGGGAAGCGTGGCGCTTTCTCATTACCGGAAAGTTTTACACCTAC
polyT_2	TTTTTCGAGGTATGTAGGCGGACAGGATTAGCAGAGTTTTT
polyT_3	TTGCTGGCTCAGTTTTGCAAGCAGCAGATTTTTT
polyT_4	TTTTTGCGGTCAGCCCGCTTTCTCTTTTT
polyT_5	TTTTTGAAGTGGTGGCCTAACTGCTACAGAGTTCTTTTTT
polyT_6	CCATTGATGCGATGCAATTTCTCATTITTT
polyT_7	TTTTTGTGCCAAAACAACTCGTCCCGTTGATTTTTGTTTT
polyT_8	TTTTTACAGTATTTGGTATCCTCCGGCAAACAACTTTTT
polyT_9	CCCTCGCGTTGTGGCAAAGGACAGATGGCCCACTATTTTT
polyT_10	TTTTTGCCGTAGGTGGCATCGCTTCAGGGTCAGCTTTTTTT
polyT_11	TTTTTCAGTTACCTTCGGAAAATCGTCTTGAAGAAGATGTGGCGGCC
polyT_12	TTTTTTTACCTTGATGCCGTTCAAATGTTGAATATTTTT
polyT_13	TTTTTCTTGACGGATCTCAAGAAGATTTTTT
polyT_14	CACGAACTGTTACTAGTGGAGCGCTCTGCTGAAGCTTTTT
polyT_15	GTGGGAGTTTACAGACAACGTCATGAATGAGCGGGGGTCTGGTTTTT
polyT_16	TTTTTTTTTATTAGGGGATCTTGAAGTTTTT
polyT_17	TTTTTACCGCTGGTAGCGGTGGTTTTTTTCGCGAGAGTGA
polyT_18	TGCGCGTTAATGCCAGGCGGGCTACACTAGAAGATTTTT
polyT_19	TTTTTCGTGAACCATCACTACGGGAGGGCTTACCATCTTTTT
polyT_20	TTTTTACGCGCAGAAATGGATTTTGGTCATGAGTTTTT
polyT_21	TTTTTCTATACTCTTCCACTCATGG
polyT_22	TTTTTCCTTTGATCTTTTCTACTCGGGGCGAAATGTAGTTG
polyT_23	TTTTTCTGCATAATTCTTACTGTATGCCTGTTATCTTTTTCAAATCAGGGC
polyT_24	TTTTTAAACTCACGTTAAGGTTGCCATTGCTCTGGGGTCTGACGCTCAGTGAACGTTTTT
polyT_25	TACAGGCAGCGTTAAATTTTTAGGCGCGGTCAGAGAGTTGGTAGCTTTTTT
polyT_26	TTTTTTGGCCCCAGTTTATGGCAGCATTTTT
polyT_27	TTTTTATTATCAAAAAGGATCTTACCTAGAGCGCAACGAAAGTTAAATCAGCTTCCATGCC
Circ_1	GTAAGTTATGTAACGCGGAACCTCCATATATGGGCTATGAACTAATGACCCCGTAATTGATTACTATTAATAA CTAGTCAATAATCAATGTCAACGCGTATCAGAAGCCATAGAGCCCACCGCATCCCCAGCATGCCTGCTATT GTCTTCCAATCCTCCCCCTTGTCTGCTGCCCCACCCACCCCCAGAATAGAATG

2.2.2 sc_EGFP2

Scaffold sequence for sc_EGFP2:

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Corresponding DNA origami designs and staple lists:

2.2.2.1 sc_EGFP2 20HB

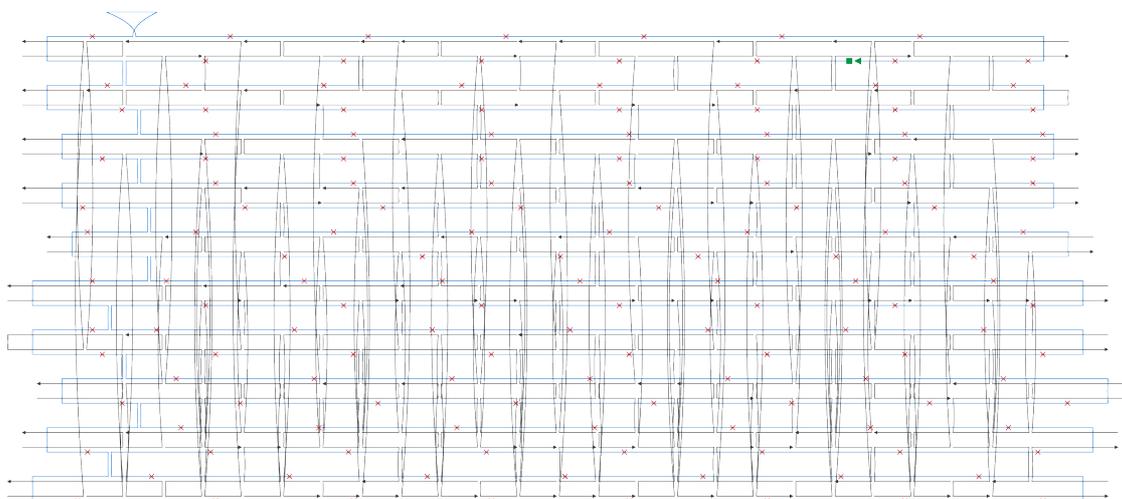


Fig. S28 | Scaffold routing and staple design for sc_EGFP2 20HB. Scaffold routing is shown in blue, staples are given in black, breakpoint for addition of sequence is marked by a green square (5') and arrow (3'). Design was prepared using caDNAo v0.1.

Table S15 | Individual staple sequences for sc_EGFP2 20HB.

Name	Sequence 5' - 3'
core_1	GAAGCTCCCTCGTGCGCTGAGTAGAAGGGCGCAAACCAC
core_2	GACTATAAAGATACCGGGTGGGGCAGGAAGTTTGCAAG
core_3	CCGCCCCCTGACGAGTGGGCTTCGTGCACACGGGGT
core_4	CGTTTTTCAAATTCGCGTTAAACCTTATAAATAGCAG
core_5	GTGTCATCCGTGCCCTTCTTGACCGACCAC
core_6	AATAGCAGAGCTCGCTGATCAGCTACCTGA
core_7	AGGCCAGCGCATGGACGAGCTGTATGCTGGAGGACCCCT
core_8	TCCCACTGTCCTTTCGTGAACATCGAGGACGGCAGCGTGCAGCTCGCCCTGGAA
core_9	CCCCTCCCTCTATTCTGGGGGGTGAGGCGTTT
core_10	AGTTGCCAGCCATCTCGACGGCCCCACAACGTCTATATC
core_11	TGCCTTCTGCAAGGGGGAGGATTGTGGCGAAA
core_12	AATGCTAGGCATGCTGGGGATGCGGCATCACA
core_13	CTATAGTGACCCCAAGCATCGACTTCAAGGTGCCCGAAGAGTAAACATCCGCCT
core_14	GCTGCCCGACAACCACCTCGACTGAGAAAAAA
core_15	GAGCAAAGTCACCTATCTTTTCTCCAAGTGGCTCTTGC
core_16	AAGAACGGCATCAAGGGGCCACCCCGCTCGGATACCGC
core_17	CAAGCAGTACCAGCAGAACACCCCATCGGGTTGTTTGTGTTTTTGGCAAAATG
core_18	GGGGCACAAGCTGGAATGAAGCATAATCAGGGGCCGAG
core_19	CGCATCGACAGTGAATCTAGAGGTCTGAGGCGAATGAG
core_20	GGTGAACCTTCTTCATGAAGTTCGGGAAGGCTCGTCG
core_21	GTGCCCTTGAACCTTCTTGATGGTGCCACATTGCATCGCATTGTCTCTCCTG
core_22	CACCCTGACCTACGGATGGCCGAAGCGGTGG
core_23	TTCTTCAAGTCCGCCAAGGACGGCTCCTTTGA
core_24	AAGTTCACGCTACCCCGACCACGTACAACCTTACGCGC
core_25	CCCATCTGCAACTTTTTGGTCTACCGAGTTATCTTCAG
core_26	ACGGTGGTCGTAACAACCTCCGCTTGACGTGCGAGTGTGTTAAATCG
core_27	TATAAGCAGAGCTCTCGAGGGCGCGGCTCCACGTTTCATC
core_28	CTGGCTAAGGCACCAAAATCAACGACTGCCATCCTCCGATTAGAGCT
core_29	CTAGAGAACCCACTGCTTACTGGACGGCCACAGCCGGAA
core_30	AAGTCTCAATTAATACGACTCACTATAGGG
core_31	CGGTTTGACTCACGGTGACGGTCAACGATCGCGGGCGC
core_32	TGGATAGGCTTGGTACCGAGCTCGGATCCACAAGGGCGATTGTTGCTTAAATC
core_33	GTTTTGGCAGTACATCAGTACATTTGGTATCACGCTG
core_34	GAGTATTTACGGTAAGGACTTTCGCTCACATGCCAC
core_35	ATTATGCCAATGGGCGTCTATTAAGGAGCTGTCCAGGAGCGCACCAT

core_36	TGACCTTAAGTAATCAATTACGGGGTCATTATGTAGCGGTGGCTTCA
core_37	AACGCCATAAAGCACATCACTCATGGTAAC
core_38	CCGCCATTGACGTCACCCCGATTTCGTTGTCATCCGTAACATAGTTGCGCTGGT
core_39	ACCGCCCGTACATCAAGTGTATCATGGGAGTACCAGCC
core_40	CATAACTTCCCCTATTGACGTC AAGGATTTCCGTGGTCCCTGGTCGAGC
core_41	CCACCACATTATTAATTGGGACTTTCCTACTTTACCATG
core_42	CATCACCCTAATCAAGTATTATTGTA CTATATGAGGAA
core_43	GTCTATCAGGGCGATGTCATGAGAAGGGAAT
core_44	CCAACGTCGAAAAATAGCAAAAAACAGGACA
core_45	AGTTTGGAAACAAGAGATTTCCCCATCTTTT
core_46	GAAATCGGCAAAATCTTTTTGTTGATCTTAC
core_47	TATTTGAATGTATTTAAAAGGGCGCTGGT GAGGGTTAGCAGCAATAATTGTTTT
core_48	AGGGGTTCCGCGCACTCCACTATATAGTGTATTACATGACGCAGAA
core_49	GAAAAGTGAAAAATCGACGCTCAAGTCAGAGGGGAAGACACTTTTACAGGATCT
core_50	CCACCTAAATTGTAAGGTTGAGTCCGGCGTTT CAGCTCCCATCCA
core_51	CAGCGTTTTCTGAGATAAAGAAAGGAAGGGAAG
core_52	TTCTGTGAAAAACCTGACGGGGAAAGCCGG
core_53	CAATACGGAATCTAAACTGACGCTGCTGAAGGCGAGAAGCGGATCACATGGTCCCAAGTAA
core_54	TCCCCAAGTACGCACGGTAAATGGCCCGCTGGCTGCGAACGT
core_55	CGGTTCCAAATGGCCCCATATATGGAGTTCGGCGTTACGAAAGGA
core_56	GGCATCGTAACTTTAACCTTTTAAGGGATTT
core_57	TGTAGATACATAATTATGTTGAAAAGCATT TCCGACCCTGCCGCTTACCGGC
core_58	CCTGACTCTCGTGACCTACGGCAAGCTGACC
core_59	GCGATCTGGCTTCAGCGCGTGTCCGGCGAGGG
core_60	GTATATATGGCTACGTTACCCGGGGTGGTGAGACCCAA
core_61	AATTA AAAAGGACGAGCTCGAGATGGTGAGCTAGTAAC
core_62	CCGGCAAAACACGGAACCTTACTACGTGAACGAACCTAAAGGGAGCATAATGA
core_63	CTAATAAAACTCTTCCAAAAAGAGACGGGAGTGCCGGCAAGCTGCC
core_64	CGTGCAGTTCTATTTGATTTATCTCCTTCGGCTTGGCAAACGACCC
core_65	TGACGTCAATATGCCATGTTGTGCGGCGAGAACGTGGACT
core_66	GTTGTTCTAGGGCGCTGGCAAGGTT CATAGCGCCTGGC
core_67	AATAGACCGAGATAGGCGTTAATTAACCCACCTATGGCTGCCCTATT
core_68	TATCAGGGTTATTGTGCGCCACTGTCATGCCAGAAGTAGAGACCCACCAAAATGGAGGTCTA
core_69	GATGCTTCCGCAAAACGGATACACCCCTG
core_70	GATAACTTTCGATGATTTTGTT CATAGGCT
core_71	AGTTGGCCAATGGGTGCGTATGTTCCCATAGT
core_72	AAAAAAGCTACTCAACTATCTCACAGCAGAACAACAGCCGTGCT
core_73	AAGGCGAGTGCGGCGGACAGTTACAAGAAGAAACATCCTGCACCCAGTCCGCCCT
core_74	TGAGGCACCCAAGTCACTG GGGTAAACAAATCCCACAG
core_75	CCAATGCTGCACGACTGGACGGCGACGTAACCTTATCGACACCCCAT
core_76	CGAAAACAGATCCAGCGCGCCACATCAA AAGCGCGTAA
polyT_1	TTTTTGAACCGTAAAAAGCCGCGTTGCTGGTGAGCAAACGCTGTTGTACAGTTA
polyT_2	TTTTTAAGCGTGGCGCTTTCTCTTTCTCTAGGCGGTTTTTCAATTTTTTG
polyT_3	TTTTTAGGTATCTCAGTTCGCATAGCT
polyT_4	TCGAGGGCTTCGTGACCGCCGCGGGTTTTTTTTTATCACTCTCGAAAAGGCCAGTTTTT
polyT_5	TTTTTCCGAGGTGAAGTTGGTCA TGAGATTATCAATTTTT
polyT_6	TTTTTAGCTGGGCTGTGGGCGTATCTGCGCTCTGCTTTTTT
polyT_7	TTTTTATCCATCACAGCCAGTTAACCTAGAT
polyT_8	TTTTTCCCGACCGCTGCGCCTAAATGGGCGGTAGGCGTGTCTGAAGTCTGCAAT
polyT_9	TTTTTTATTAGTCATCGCTATGGCAGTATGCTACAGCTTAATG
polyT_10	TTTTTGTCTTGAGTCCAACCCGGGACTTTCCACCATTGA
polyT_11	TTTTTACGCGTTGACATTGATTATTGACTAGCCCCGCCG
polyT_12	TTTTTTGGCAGCAGCCACTGGTTCGAGGTGCCGATAGGTAAGACACGACTTATCGCCACTTTTT
polyT_13	TTTTTACCAATAGGCCTCACAGATATTTTTT
polyT_14	TTTTTCTTCCGGGCGAAAAACATCATTGGA AAAACGTTTTT
polyT_15	TTTTTTGAAGTGGTGGGGTAAACAGGATTAGCAGTTTTTTTTTATAGCGAGGTATGCCTTCGGGTTTTT
polyT_16	CCTTATGGCAGCACTGACTACGATTTGGTAGCCAAGATCCGCCACTAGGTCGTTCCGCTCCATTTTT
polyT_17	AAGTGCTCTCTCAAGAAATCAGCTATTTTTTATTTTT
polyT_18	GGTGTACCTAGAGTAGTGATGCGGGCCGCCAGTGTGCTGGAATTCTGCAGATTTTT
polyT_19	TTTTTACGTTGTTGCCATCATCTACGTTTTT
polyT_20	TTTTTGAACAGTATTTGTTATACGGCTACACTAGAATTTTT
polyT_21	CCATCTGGCGCTATCCGGTAACTATCTTTTT
polyT_22	CCCCAGTGTCTATCTGCACCACACGAACCCCCGTTTCAATTTTT
polyT_23	AGTAGTTCTGGCGCCCGGCAACTACAAGACCCGCGTTTTT
polyT_24	TTTTTAAAGGATCTTCATAGTTTTCGCGATTTTT
polyT_25	TTTTTGAAGCCAGTTCACGCTGTTTTT
polyT_26	ACTTCCGGTGCTACAGAGTCTTTTT

2.2.2.2 sc_EGFP2 20HB-LPv2

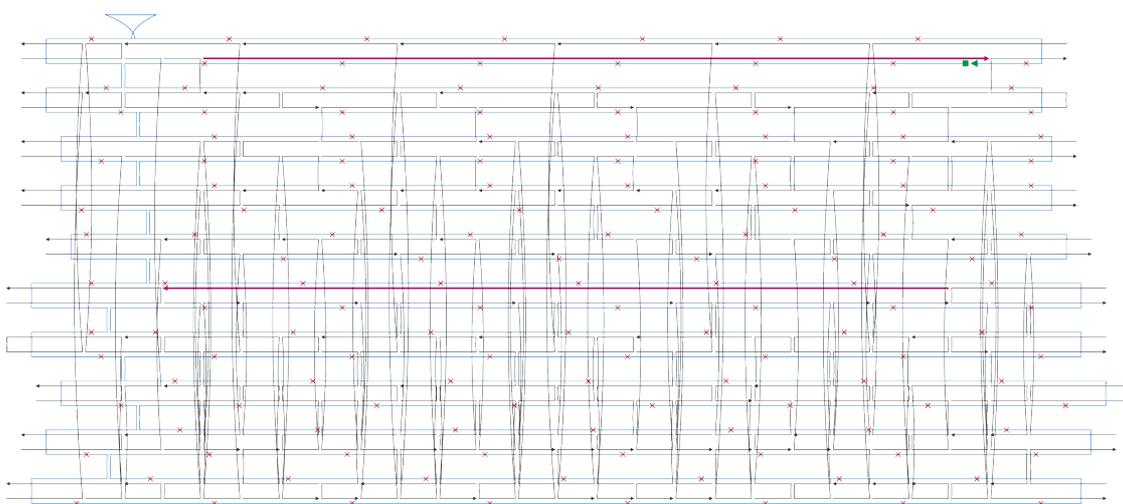


Fig. S29 | Scaffold routing and staple design for sc_EGFP2 20HB-LPv2. Scaffold routing is shown in blue, staples are given in black, breakpoint for addition of sequence is marked by a green square (5') and arrow (3'). Long 154-mer 'primer' sequences at the reverse complement sequences for the polyA (5') and promoter (3') regions, flanking the EGFP gene are given in pink. Design was prepared using caDNAo v0.1.

Table S16 | Individual staple sequences for sc_EGFP2 20HB-LPv2.

Name	Sequence 5' - 3'
core_1	GGTGCCACACACGGAAGCTGCTGAACCCGTCG
core_2	TCCCCTGTCCCTTGACCCTGGAACACAGGTGAACCTCAAGATCCGCCACAACATCCCCTCCC
core_3	GCCCTATTCTATAGTGTACCTACCGTGCT
core_4	GACCGCCGCGGGATCCGAGAAGCGCCGAGGTGAAGTGGCTACGACCTAGAT
core_5	TACCAGCAAAGCTGGAGTACAACCTCTCGTGACGAGGCACGAGACCCA
core_6	GAACACCCCATCGGCGACGGCCAATGCTAGTTTGCAAG
core_7	TCTATATCATGGCCGACACCGGCACGTTTCATCCCATCTGG
core_8	CACAACGGCAGCGTGCAGCTCGCCGACCACTGCCTTCTCAAACCACTCTGGGTG
core_9	TTCAAGGAGGACGGCGCTTACGCTTGGTCTAGCAATAA
core_10	GCATCGACGCTGCCCCGACAACCACTACCTGAGCTAGAGGAGGATCTCTTCGATGATAGTGTA
core_11	GGTGAAACCGCATCGAGCAGCACTTTAAATCACGCAGAAG
core_12	CGTACCCTGAAGTTCATCTGCACCAAGCAGCCTTCGGCCGTGCCT
core_13	GTGCCCTGGCCACCACAACAGCCCGCAAAAGTTGCCAGCCATCTGTTGTTTGCAGGACG
core_14	CCTACGGCGTGCAGTAACATCCTTTTTTTTGGAGCTCGCTGATCAGCCTCGACTG
core_15	CCGACCACATGAAGCAGCTGAAGGAGAAAAAA
core_16	TCCAGGAGAGATATCCATCACACGCTTGGTA
core_17	GCTCGAGAGTCCGCCATGCCGAATCGAGGGC
core_18	ACGCAAAAATCAACGGGACTTTCGTAACGCCTCGTTGTGCGGGGTCG
core_19	TGGGCGGTAGGCGTGTACGGTGAAGTTCAGGATACCGCCTATCTCA
core_20	GAGGTCTATGACGTCATGGGAGTAATGGGTG
core_21	TATAAGCAGAGCTCTCTGGCTAAGGTCGAGCGATTTATC
core_22	CTAGAGAACTCACGGGGATTTCCCTTGGCAG
core_23	AATTAATAAGTACATCAATGGGCAGTACGC
core_24	CGGTTTGCCCACTGCTTACTGGCTTATCGAAGGAGCTGGGGCCGAG
core_25	GTTTTGGCCGACTCACTATAGGGAGACCCAATGGCGGCCATCCGCTCCTTTTA
core_26	ATCGCTATTACCATGAATGGCCGCATCGTTAGGGCGC
core_27	AGTTTTTTAGAAGTAAGTTGACG
core_28	CGCGTAACGCTCATTTTTTAACCATTTTTGTT
core_29	GGCGATGGCCCACTATACTCATAAAGGGCG
core_30	AATAGACCAAGAAAGCGAAAGGAGCGGGCGCGGTGTACAC
core_31	CAAAATCCCTTATAAGAAAAGTGCAGTGTGAGATCCAG
core_32	TTTTTCAATATTATTGAAAAACCCAGCACTGCCTTCGGCCCCAGTGGGCACCAA
core_33	AAGCATTGAGGAAGGCAAAATGTTGGTAGC
core_34	TTATTGTCTCATGAGCTAAAGAACATCCGTAATCCCCACGCTCACCACCCCAT
core_35	GGATACAACCTTACCAGCGTTTCGCTGGTA
core_36	TGATTTTAGAAAAATGTTGTTTCTACTCAACCGGTTCCACCAGCC

core_37	AAACAAATTCGTGCACCCAACCTGACAGCAGA
core_38	CGCGCACATTTCCCATCAAAAAGTGCGGCGGCTCGTCTGGTCC
core_39	TCTTCAGCTGGTGAGAGTTTGGAAAGCCGGCGAACGTGGCGAGAA
core_40	TCTTCGGGGCGAAAACCTGACGCTGGATCTTC
core_41	TCTTACTAGCAAAAATATCAGGGGACTATAAAGATACCAGGCGTTTCCCCCTGG
core_42	GTCATGCCGTGGACTAAGGGAGCCCCGATTAGAGCT
core_43	ACCGAGTTAATTAATCCTTTGAGACACCCTCCGCCCTGAGCAAAGACCCCCAACTCTCG
core_44	GCTCTTGCATCTTACCCACCTAAAGGCCGCTTGCTGGCGTTTTTCCATAGGCT
core_45	GATAACTCAAAAACAGTGAACAGACCC
core_46	TCCTCCGAAATAGGGACTTCCATTGACGTCTTGTCTGCAATCGTGTCC
core_47	GGTTAGCTCATAATTCGCGATCTGTCTTGAT
core_48	AAAAAAGCGAACCCCTACCAACGTCAAAGGGCG
core_49	TGTTGTGCGAGTATTACGGTAAACTGCCAAAGTCTCCGGCTCCATGGACGG
core_50	TTACATGAGATGCTTCCAATGCTGCGGTGGGGGGCAC
core_51	AAGGCGAGTGACGGGGAACAAGAGTCCACTAT
core_52	AACGATCTACATCAAGTGTATCATATGCCAGTGGATAGAGCCGGAATTCACCGG
core_53	TTCAGTCCAAGTCAGTATATATTTACGCGC
core_54	TGGCTTCAAGGAAGGGGAGATAGGGTTGAGT
core_55	TTTGGTACCCCTATTGACGTCAATGACGGTAGTGATGCGGCAACTTT
core_56	CCATCCATGCTACAGCGCTGGCATTATGCCAGTACACATCTAGC
core_57	CATAGTTGCACTCATAAGGGAATCTTCCAAGCTCCCTCGTGCGCTCTCCTG
core_58	TCTATTTAGCTGCCCGGCGAGGGCGAGGGCG
core_59	TAATCAGTCAACCTGACGACGTAACGGCCAC
core_60	GACAGTTATTCGTGACATCTTTTTATTTGAACGCTCAAGTCAGAGGTGGCGAAACCCGACAG
core_61	GAGTAAACCGCTACCGGTGGTGCCCATCCT
core_62	ATCTAAATTCGTGAGATAACCCACAGGGTCCCGCCCCCTGACGAGCATCACAAAATCGA
core_63	AAAAAGAGCCGCAAAAGTTATGGGTCTATCAAGGTGCCGTAAGCACTAAATCG
core_64	AAGAAGAATGAAGTTTCTTCAATGGTGAGCAAGGGCG
core_65	TCTTTTCTGCATGGACGAGCTGTACAAGTAATCACCAG
core_66	ACGGGGTCTCAAGCCGCGTCAAAATCGGTGGCAAGTGTAGCGG
polyT_1	TTTTTGCAAAAGGCCAGCAAAAGGCCAGGAACCGTAAAAATTGTAAGCGTTAATAATAGGCC
polyT_2	TTTTTCTGTCCGCTTTCTCCCGCTTACAGGATTATGTTGAACGTGAAC
polyT_3	TTTTTCGCTTCTCATAGCTCTTCGGG
polyT_4	GCGATCACATGGTCTGCTTTTTTTTTTTGGAGTTCGTGAATGAGTGATTTTT
polyT_5	TTTTTCGACGGCAACTACGAAAACCTACGTTAAGGTTTTT
polyT_6	TTTTTTTCGGTGTAGGTTGTCTACACTAGAAGAACATTTTT
polyT_7	TTTTTCCAGTGTGCTGGCGGGAAGCTGAGATTA
polyT_8	TTTTTTGTGTGCACGAACCCCTAACAACCTCCGCCCATGATGCCACGGGCTTA
polyT_9	TTTTTCTACTTGGCAGTATGACCTAAGTTTGC
polyT_10	TTTTTCTGCGCCTTATCCGGTATGTTCCCATACAAAATG
polyT_11	TTTTTGCCGCGCTTAATGTCACAGATATACGTACGCTGGCAACGTT
polyT_12	TTTTTGGTAAAGACAGCACTTACACCTAATCAGTAACTATCGTCTTGAGTCCAACCCTTTTT
polyT_13	TTTTTTGTAAATCACACCACCCCTTTTTT
polyT_14	TTTTTAAGTGCTCATCTATTGGATAGCAGAACTTTAATTTTT
polyT_15	TTTTTATGTAGGCGGTGCATTCGCCACTGGCAGCATTTTTTTTTTGGCACTGGTAACCGGATACTTTTT
polyT_16	CTAGCCGCAAGTGTATCCTGACTCGCCAGTTAAAGAACGGCATCAGCTGTAGGTATCTCAGTTTTT
polyT_17	AATACGGGTTGCCATGTCTATTATATTAGTCCCGAGCTCGGATCCACTAGTAACGGCCGTTTTT
polyT_18	CGCGCCACAAAACGTAATAATTCGCGTTAAATTTTTTTT
polyT_19	TTTTTTTCGCCAGTTAATTGGGACTTTTTTTT
polyT_20	TTTTTGCCCTAACTACGGAGAAGTCTTGAAGTGGTGTTTTT
polyT_21	TAACCTACGTGCGTTCAGCCCCGACCGTTTTT
polyT_22	ATACGGGACTACGGCAAGCTGTCGCTCCAAGCTGGGCTTTTT
polyT_23	ATTGTTGCAATTCTGCCGCACCATCTTCTTCAAGGATTTTT
polyT_24	TTTTTGATTTTGGTCATAGAGTAAGTAGTTTTT
polyT_25	TTTTTGATTTTGGTAAAGCGTGGTTTTT
polyT_26	TCTGCGCTAGCAGAGCGAGTTTTTTT
LPv2_1	TCCTTTCTAATAAAATGAGGAAATTGCATCGCATTGTCTGAGTAGGTGTATTCTATTCTGGGGGGTGGG GTGGGGCAGGACAGCAAGGGGGAGGATTGGGAAGACAATAGCAGGCATGCTGGGGATGCGGTGGGCTC TATGGCTTCTGAGGC
LPv2_2	CGTTGACATTGATTATTGACTAGTTATTAATAGTAATCAATTACGGGGTCAATTAGTTCATAGCCCATATATGG AGTTCCGCGTTACATAAATTACGGTAAATGGCCCGCTGGCTGACCGCCCAACGACCCCCGCCATTGAC GTCAATAATGA

2.2.2.3 sc_EGFP2 20HB-Circ

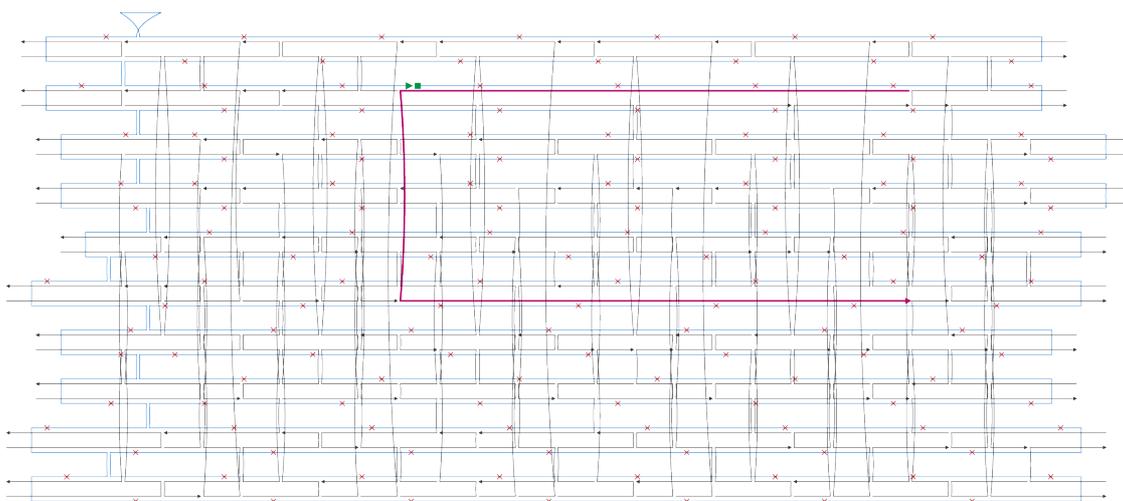


Fig. S 30 | Scaffold routing and staple design for sc_EGFP2 20HB-Circ. Scaffold routing is shown in blue, staples are given in black, breakpoint for addition of sequence is marked by a green square (5') and arrow (3'). Long 200-mer 'primer' sequence linking the reverse complement sequences for the polyA (5') to the promoter (3') region, flanking the EGFP gene is given in pink. Design was prepared using caDNAo v0.1.

Table S17 | Individual staple sequences for sc_EGFP2 20HB-Circ.

Name	Sequence 5' - 3'
core_1	CAGAGGTGTGGCGAGAAAGGAATTATAAAT
core_2	GATACCAATTTAGAGCTTGACGGTTGAGTG
core_3	ACCTGTCTCTGAGTAGGTGTCATCCTTTCC
core_4	CGAATGAGAAATCGACGCTCAAGTTTATCCGG
core_5	ACTTGTACAAGTAATCTAGAGGGCCCTATTCCAAAAGGC
core_6	TATAGTGTGACCGCCGCCGGGATGCGACACC
core_7	CACCTAAATGCTAGAGCTCGCTGATCAGCCTAGCGCGATCACATGGGGGCATCG
core_8	CGACTGTGCCTTCTAGTTGCCAGCCATCTGTCAGTCCGCCCTGAGCCTGGGGCACCCTTGG
core_9	GCTGCTGCCCGACAAGCCACAACGTC AATGGTTTCCAAG
core_10	AGCAGAACACCCCCAGAAGAACGTCAGTAACGGAGTTTGTGAAGCAAAAAAAG
core_11	TATCGCCGAGGTGAAGTTCGAGGCACTCTCG
core_12	AGTACAACCTACAAACACCACTACCTGAGCACCTGTTTGCCCCCTCCCCCGTGCCTTCCTTGAC
core_13	AGCGCACCCGCATCGAGCTGAATCCTGCTGCAGATATA
core_14	AAGTCCGCGAGGACGGCAACATCAAAGACCCCAACGAGA
core_15	TGACCTACTCATGGCCGACAAGCATCGGCGACGGCCCCGTCCTGGAAGGTGCCACTCCCCTGT
core_16	CAGATATCAGCTCGGATCCACTAAACTCT
core_17	CATCACACTGGCGGCCGCTCGAGCGTCCAGGGCTATTACGCCCCCTAAAAATCCCGGGAAGAA
core_18	ATGGTGAGCTCACTATAGGGAGACTCCAGTTC
core_19	CAAGGGCGAGGAGCTGTTACCCGACTTCTTACATCAATCAGTACATAGATAGGGGGAAAGC
core_20	GGGTGGTGACTGCTTACTGGCTTAACTGATCT
core_21	CGACGTAAAAGCAGAGCTCTCTGGCGTTTCTG
core_22	GGCGAGGGCGGTAGGCGTGTACGAATGCCGC
core_23	TCTATATACGGCCACAAGTTCAGCGTGTCCGACCACCCGTC AATGGCCAATAGAACGTCA
core_24	TACTTGGCCCCGCTCTCATTTCGCTGGCATAAATTAACACGACTTATCGCCACTGGCTGG
core_25	GGGCGTGATAAACAATCAGCATCCGACCGAGTTGCTCTTGCCCCGGC
core_26	ACGGGGAGTGGAGTACAAGAGTCTAAAGGG
core_27	TTTTGGCACAATGGGCGAGGGCGATGCCACCTACGGCA
core_28	CCAAAATCCCGTGCCGGTGAACCTTCAAGATC
core_29	GTAATGGAGTACATCATATTTCAAGGATCAAAGTGCTCATCATTGGAAAAC
core_30	CAATGACGCTGGTGAACATCTTCTCAAGGAC
core_31	CCAAGTACCATGGTGCCGAAAAGGATGTAACGGGATAATACCGGCCACATAGCTTGGTATG
core_32	TTTACGGTAAACTGCCAAGCTGGACATGAAGCAGCACG
core_33	GGACTTTCATTGACGTCTATAGGCGTGCAGTGCTTC
core_34	TGTTCCAGCATCAACTGCCCCACCCTCGT
core_35	GTAACCACCAGGAACATAGGCTCCGCCCCCCCGGTAAG
core_36	CACACCCGGCTAGGGTTAACCAATAAGTAGTTGTCACGCTCGTCGTAGAACTTT

core_37	AGTGTAGCTTTTTCCCGTAAAAAGGCCGCGT
core_38	CGGAACCCCACTATTTAAACCAGGTTAGCTCCTTCGGTC
core_39	CGAGGTGCTTGCCTGACTGTAGGTATCTCAGTTCCGGTTCGTGCGCCACTAAAT
core_40	TTTGGGGTAAGGGCGACCACGCTCGAAGTAAGTTGGCCG
core_41	GTTAAAATCATCGTGGTCGCCAGTTAATAGTGATCCTTT
core_42	TAAATTGTAATCGGCTTGACGTTAATGTCAGAGTTTCG
core_43	TGCCACCGCTTCATTTAATTGTTGCCGGA
core_44	ATAGGGGGGCGAGTCTGCAACTTTATCCG
core_45	TTAGAAAAGATAGCGGAGAACCCCATCCTGGTCGAGCTGGACGGAGCCGCTA
core_46	CATATTTGAAAAGCGCCAGCCGGAAGGGCC
core_47	AGCGGATAGGTGAGCAAACCAAGTCATTCTGAGAATAGTTTGTGCAAATGTATGTTGGAA
core_48	TTTATCAGCGTTGTCAACCGGCTCCAGATTTA
core_49	TTACCGCTGTTGAGACCAAGCTTAGTCATC
core_50	CCACTCGTGCACCCATCGAAATTTGGCAGT
core_51	TTTTACTTTTACCAGCTAACTAGTTTGACTCCCCCGACC
core_52	AGTACTCAAACAGGAAGGCAAGTGGGAGGCCATTGAC
core_53	TAAGATGCTTTTCTGTGACTGGTGCTCCGATGGTTATTTGGACTCC
core_54	CCATCCGGGAATAAGGGCGACACCATTTGACG
core_55	ACGATCAATTCGCGCATAGACCGCAAGTGTATCATATGACTTCAAGCATGCCCGAAGGCTA
core_56	CAGTCTATCAGCTCCGGTTCCAGTCAATAC
core_57	AAGTGGTCTACATGATCCCCCATGGTATGCGG
core_58	GTGCTGCAATGATACCTAACTACACCCTAA
core_59	AAATGAAGTTTTAAAAGCTAGAGTAGGCCGAAGCGTTATACGTATTGGTACCG
core_60	TCAATCTAAGCGAAAGATCACAATGAGCAAAAAGGCCAG
core_61	AAGTATATTAACATATCGTCTTGAGTCCAACCTGACGAGCGAGCGGGCCCCGCGCT
core_62	ATGAGTAAACTTGGTCTCCATCCAAAAGAACATTTCCATGCGGTTTAAATACGA
core_63	CTGACAGTCGGCGAACGGCGAAACCCGACAGGACTATAAACGAACCCC
core_64	TACCAATCCGTTCCAGCCGACCGCTGCGCC
core_65	GCTTAATCAGTGAGGCGAGCGCAGTTGTTCCA
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core_67	CAGCGATCGCTCCAAGCTGGGCTGTGTGCA
core_68	CTGTCTATTTGTTTCATCAGCAAAAAGAACGGTCTCATGTCTCCACC
core_69	TCCATAGCGTAAAGTCTCCTGTTCCGACCCTGCCGCTAGCTCACG
core_70	CTCCCCGTCGTGTAGAGCGAGACAAAACCGTAATATTA
polyT_1	TTTTTCTTCGGGAAGCGTGGCGCTTTTCTCATTACCGGATTCAAGTTTAAATGGCC
polyT_2	TTTTTAGCGAGGTATGTAGGCTAACAGGATTAGCAGTTTTT
polyT_3	TGCTGGCGGGTTTTGTTTGAAGCAGCAGTTTTT
polyT_4	TTTTTCATCGCATTGGCCTTTCTCCTTTTT
polyT_5	TTTTTTGAAGTGGTGGCCTAGGTGCTACAGAGTTCTTTTT
polyT_6	TCGCCGATGACCGCCCAACGACCCCTTTTT
polyT_7	TTTTTGACGGCAGCGTGCAGCCGCCACAACATCGAGTTTTT
polyT_8	TTTTTGAACAGTATTTGGCATGATCCGGCAAACAAATTTTT
polyT_9	CAAGCTGCAACGGGACGTCAATAGCGATGGCCCACTTTTT
polyT_10	TTTTTTCATCTGCACCACCGGAGCTGACCCTGAAGTTTTTT
polyT_11	TTTTTGCCAGTTACCTTCGGAACAGTGTGCTGGAATTCTGGACGGCAA
polyT_12	TTTTTGTGTAACAACCTCCGCCCGAAATGTTGAATTTTT
polyT_13	TTTTTCTCTTGACGGATCTCAAGAAGTTTTT
polyT_14	GACTTTCCCTACAAGACCCGCTGCGCTCTGCTGAATTTTT
polyT_15	ATGACGTACGCCCTGGCCCACTACCTAATAAAATGAGGAAATGTTTTT
polyT_16	TTTTTCGCCCAATTGACTTTCCAAAATTTTTT
polyT_17	TTTTTCCACCGCTGGTAGCGGTGGTTTTTCACGCCAGTA
polyT_18	GCTGCGCGCATGGACGAGCACGGCTACACTAGAATTTTT
polyT_19	TTTTTACGTGAACCATCGATACGGGAGGGCTTACCATTTTT
polyT_20	TTTTTATTACGCGCAGACGTGGGATTTTGGTCATGTTTT
polyT_21	TTTTTACTCATACTCTTTCACTCAT
polyT_22	TTTTTATCCTTTGATCTTTTCTCTTCGGGGCGAGTAACGGC
polyT_23	TTTTTCACTGCATAATTCTTTACTGTATGAGTGTACCTTTTTCTATCAGG
polyT_24	TTTTTCGAAAACCTCACGTTAATGTTGCCATTGTTACGGGGTCTGACGCTCAGTGGAATTTTT
polyT_25	GCTACAGGTGCGGTTAAATTTAACTTATGGCGCAAAGAGTTGGTAGTTTTT
polyT_26	TTTTTCTGGCCCCAGGTTATGGCAGTTTTT
polyT_27	TTTTTAGATTATCAAAAAGGATCTTCACCTATTGCGCAAAAATTGTTAAATCAGGGCATTAT
Circ_1	TCTATTCTGGGGGTGGGGTGGGGCAGGACAGCAAGGGGGAGGATTGGGAAGACAATAGCAGGCATGCT GGGGATGCGGTGGGCTCTATGGCTTCTGAGGCGGTTGACATTGATTATTGACTAGTTATTAATAGTAATC AATTACGGGGTCATTAGTTCATAGCCATATATGGAGTTCGCGTTACATAACTTACGGT

2.2.3 sc_EGFP3

Scaffold sequence for sc_EGFP3:

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Corresponding DNA origami designs and staple lists:

2.2.3.1 sc_EGFP3 20HB

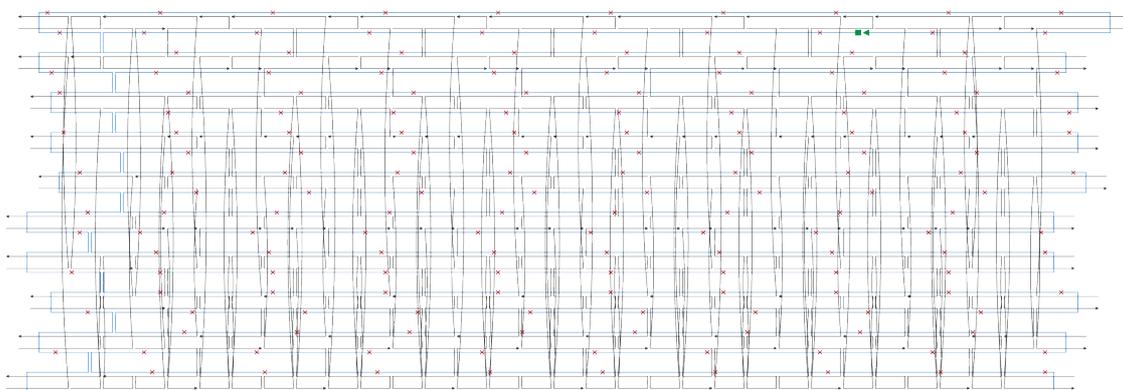


Fig. S31 | Scaffold routing and staple design for sc_EGFP3 20HB. Scaffold routing is shown in blue, staples are given in black, breakpoint for addition of sequence is marked by a green square (5') and arrow (3'). Design was prepared using caDNAo v0.1.

Table S18 | Individual staple sequences for sc_EGFP3 20HB.

Name	Sequence 5' - 3'
core_1	TAGATAAGTCAGGGCGAAAAAGGTAGAGTAA
core_2	CTACAAGACTCCAACCTTTCTACGCCATCCAGCTTGTGGCAGATGAACTTCAGGGT
core_3	GAACCCCTTAGCTAGCTTAAGGGGATGTACT
core_4	ACTCCCTGGAACAAAACTCAGCAGAAGACCAGGATGGCCAGGGCACGGGCA
core_5	CTCTGCGCAATGACCCCGTAATTATAGCGA
core_6	GAGGCCGGACCGAGATTATCAAGCAATAACCTCGCCACGCCGTAGGTCAGG
core_7	GCGACCAAAACGCGGAACTCCATAAGTAGG
core_8	CGGGCTTTCGGCAAAATTAATTATGACCCACGACACCTCATGTGGTCGGGGTA
core_9	GCCCCGGCAGCCAGGCGGGCCATGGGCATAA
core_10	GCGAGCGCTCAGCTCAAAGTATACATCTGGCGTCAGTAAGCATGGCGGACTTGA
core_11	GCAGCCTAGACGTCAATGGGCGGGATTGACGT
core_12	AAGGCCAGTTGTTAAATTACCAATGTGTAGATCGCAAGAGAAGATGGTGCGCTCC
core_13	CAAAGGTGGCGTTACTATGGGATGATACAC
core_14	CTAGAGCATGGCTACGAGTGTGAGTCAAACCGCTATCCACGCCATT
core_15	GATCTTCGCGGGTTAATCATTAAATTCGGTGT
core_16	CGAATTCCTTTGATCTGTCAAAGGCAGCATCTTATCGGGACCCCTTGCTGTCCT
core_17	TTAATAACAGTGATGGAGTTGGCCTTTCTCAT
core_18	GATTACTATGGAACGAGAGTCCACGTGAGCAATATCTACGCAGAATAGAATGACA
core_19	TATGAAGTCTCGCTCGCTCACTTCCGCCT
core_20	ATATGGGCTCATGAGATAGGGTTGAAAAAGGGTTGTAGTTGATGCAATTCCTCA
core_21	AGTTATGTAGGTCGCCGACGCCGTTCCGA
core_22	TTACCGTAGATCCTTTCCCTTAGAATACTCGGAGTGGCAGTGGGAGTGGCAC
core_23	GGGCGTCCGCCCTAGTGAGCGATCCCCCT
core_24	GGTCGTTTCAATCTTTTTTATTGAAGCAGGCCTCAAAGGCACGGGGGAGGG
core_25	TCATTATTATTAAGGGTGAGCAAACCCGACA
core_26	ACATACGTCTGACAGATTCGCGTAGCGGATAGCCCGGGCTGGCAACTAGAAGGC
core_27	TCCCTATCCAGGAACCGTAAAAA
core_28	CATTGACGTCATGGGACGCTGAATCTATTAAGTTGTTGCTGTTGAGA
core_29	TCCGTTGATTTTGGTCCAGCTCGTGGTCCTGCACGCTCGTCGGGGCG
core_30	TGGGGCGGAGTTGTTAGAACAGCTACCAGCCACTCCGGTTTTAAAAG
core_31	CCGTACAGCCTACCGCCGCTGGCTCACCTGATCCCTACGGGAT
core_32	CTAAACCAGCTCTGCAGTGGATCCAGTGAGCTCCTTCGGCGACC
core_33	ACGACCAACTTCTGCTCAGAAAACTACGAGTAAGTTGTCAACCA
core_34	GTAACCTTGATACTTCAAGCCCACCATAGTTTATGGCAGCGTAAGA
core_35	ACTCGCCTCGCCCTCGCCGGACGTGGAGAGAAGATCC
core_36	CGTTACGTCCCGTGCCAAAAACGCTCAG
core_37	GGGCACCACCCGGTTCGACATTTGATTTTGG
core_38	CTTGCTCACCATGGCGGCCATTTTTACCTA
core_39	GTGGAGAGAAAGGCAATTATATAGGTTTTAAA

core_40	GACCAATAGGTGCCTAAGGATCTGAACCTGG
core_41	TCTTCTGTCTCGAACCTGCCAGTGAGG
core_42	CTGGTAGTTTGTCTGGGCAGCAGCAAGCGAATCCAGTTCCAATCCT
core_43	TCCTCGATGCTCAGGGCGGACTGGGGTGGCAAAAACCTCCACCCCC
core_44	CCTTGATGCCATGTGATCGCGCTTGTGACGAATGCTCATCGACAATGC
core_45	GATATAGAATCCCGCGGCGGTCATGCCTGACAATACCGCGGAAAGGA
core_46	TCCAGCTTTACTTGTACAGCTCGGAGACAGCGAGTTGCTGTCAAGG
core_47	CCTTGAATAATCCAGAGGTTGAGAAAAAAGATCATTCCAGATGG
core_48	GTTACCAAGAATACCAGTCAATGATAACAATGCTTTTCAGGCAGAT
core_49	GGTAGTGGGGTCGGCGAGCTGCACGCTGCCGGCTTGGCG
core_50	GGGTCTTTGTTGTGGCGGATCTTGAAGTTCAAGTGGTCAC
core_51	AGCAGGACCGTTCTTCTGCTTGTGCGCCATGCGGCTGA
core_52	GAGAGTGCCTTGTGGCTGTTGTAGTTGTACAGAAGTCG
core_53	AAGCTTATGTGCCCCAGGATGTTGCCGTCCTTGGACGTA
core_54	AAATTTTGGTCGATGCCCTTCAGCTCGATGCGCCGTCGT
core_55	AGTCCCGGGCGGCGATGAGTTCGCCCGTGGCTGTCTTC
core_56	ATGCCCCACCGAGCCCTGTCCAGCAGCGGGGCCCCACC
core_57	ACACAGTGACAACACCACGGAATTGTCAGTGCCCTACTCA
core_58	AACGGGCCGACGCAAGGAAAGGACGATGATTTTTATTA
core_59	AACCAGGACCGCGCAGAATCCAGGTGGCAACCTTCCAGG
core_60	AAGCCATGAGGGCCGAAGGGACGTAGCAGAGCAAACAA
core_61	AGGCATTGCAGGCCGCGGGAAGGAAGGTCCACAGTCG
core_62	CAAGCGAAAAGGAGCTGACAGGTGTGCTCAACAGGCATGTGGTGC
core_63	CCAACAGACCAGTGGGGTTGCCTCGTTGGTATGGCTGAGGGTG
core_64	TTCCCCGACACCACGCCACGTCGAACTCCATCAAGGCAGCACTGC
core_65	ACAGCGAACAACCTCCTATAAATCCATGCCTGCAAAAATGCTGCTT
core_66	AGGACGCTTTATACAAGGAGGATTGGATCCCGATCGTTGCCTTCGG
core_67	GCTGGATTACGGGAAGCAATAGCATCTTTCACGTTATCACCTTGAAG
core_68	GAGTCCCGATTCTGCACCCAAC
core_69	ATTCGCCCTTAACTCCTACGTGCCGTCTATAGCATG
core_70	TAGCCATGCTCTAGGTGGGGTCAACGTGG
core_71	AATGATTAACCCGCCAGGAACCTTCCAGTTT
core_72	CCAACTCCATCACTAGTTGACGGGAAGAATAG
core_73	GTGAGCGAGCGAGCGCAAGGAAGGGCCGAAAT
core_74	CGTCGGGCGACCTTCTAGGGCGTTGTTAAA
core_75	GCTCACTGAGGCCGCGCGTAATTAATATT
core_76	CCCTAATCAAGTTTTTAAGATCCTTTTACTTTTCTTACCGCCATTGCT
core_77	GTAAAGCACTAAATCTGCTACTAAACAGGAAACGTTCTTCGTTTG
core_78	CCCCGATTTAGAGCGGGTTCCAATAAGGGAGCAGAACTCCAACG
core_79	GCGAACGTGGCGAGAGCAGAGATACTCTGCGTCAACCATGTTG
core_80	CGAAAAGGAGCGGGCGGGTCGCCCTTTATCAGTGTATGCGGTCTCTC
core_81	GTGTAGCGGTCACGCTCCGGGCAAACATATTTGTGAGTACGCCGAGT
core_82	TCAAGGATCACCAGCGTCCGCCTGGGTCTGCAAACCTCCGCCAAAACCGCATCACCATGGTA
core_83	ATTGGAAGGCAAAGGCCGAGCCGTTAAGGTGGAAAGTGACTAATACGTAGATGACTGCC
core_84	GCCACATCGACACGATTTATCAAAAGGATCGCGTCAAAAAGTCCCATAAGGTCATGTACT
core_85	CTTGCCCGTCTTTTTATACCGCGAAAATGAAACCTCCCATGCCAGGCGGGCCATTTACCGTC
core_86	TGAGAATAGGGTTATTGGCTTACTATGAGTAACGGTTCACAATAGGGGGCGTACTTGGCATA
core_87	TGTGACTGGAATGTATTCCCCGCTGCTTAATCAGTGCCTTGTGATGACTGCCAAGTGGGCAG
core_88	TGCGCAACTTGTTCCTGATCTTGCAGAAAAAACCATCAAGGTCGTTTCGCTCCAAGCTGGGC
core_89	CGTGGTGTCAACTTTATTTCTGGTATTAAGGAGGTGCCAGCTCACGCTGTAGGTATCTCAG
core_90	TCATTGAGGCCGGAAGATGCCGCAAGTGTGTAAGGGAGTCTCCCTTCGGGAAGCGTGGCGC
core_91	GAGTTACAGGCTCCAGGAAATGTTTAAATCAAGAAAGCCGCCCTGCCGTTACCGGATACCTG
core_92	AGCGGTTCTGCAATGCAATTAACCAATAGGAAGAAAGGGAAGCTCCCTCGTGCCTCTCTCT
core_93	GTCAGAATACGGGAGTCTCATGTAATTTCTGGCAAGGACTATAAAGATACCAGGCGTT
core_94	CTCATGGTGCCTGACTTAGAAAAGTAAGCGCCACCACGCTCAAGTCAGAGGTGGCGAA
polyT_1	AAAAATCGACCCGCCGCGCTTAATGTTAATTTTT
polyT_2	TTTTTGGCTCCGCCCCCTGACGAGCATCACGGCCGCGT
polyT_3	TTTTTCCCGTTACGCCGACTGTGTGCAAGTTACCATGGCCCA
polyT_4	ATGGAAAGCACCTATCCCTAAATTAATAACAGCTCGCTCCTCGAAGACGCGGAAGAGGCTTTTT
polyT_5	TTTTTCCACCCATTGACGTCATTTACCGT
polyT_6	TTTTTGTAACTATCGTCTTTCGCGTGC
polyT_7	TTTTTAAACCTGTCTTAAATACTTTTTT
polyT_8	TTTTTACGACTTATCGCCGATTTTTTTGTTTGAAGTTTTT
polyT_9	TTTTTTCACCTCGGCTGGTCTCCTTTTTT
polyT_10	TTTTTTAACAGGATTAGCAGAGCGCCGATGGGGGTGTTCTGCAGCTTG
polyT_11	TTTTTGTAAAAGGAGCAAATTTCTTTTTT
polyT_12	TTTTTGTACAGAGTTCTTGAGGGAGGGGAACGGGGCC
polyT_13	TTTTTTCGAGAGCCGGCAAAAGCAGCGTATCCACATAGCTTTTTT
polyT_14	TTTTTCACTAGAAGGACAGTATGCCTGCTATAATAAGTGGTGGCCTAACTACGGCTATTTTTT

polyT_15	TTTTTTTAAGGCTGCGCAATAGGGGATGCCATC
polyT_16	TTTTTCTGCTGAAGCCCGAACCCTTTTT
polyT_17	AAGTGCCATCAGCGATCTGTCTTTTT
polyT_18	TTTTTTTTCCCCGAATGCTGGCGTTTTTCCATTTTT
polyT_19	TTTTTTTGGTAGCTCTTAGCATTGGTATCTGCGCTTTTTT
polyT_20	CGATGTAACCCACCCGCGCCAGTTGTCGAGGTATGTAGGCGGTTTTT
polyT_21	TTTTTTTACTGTCTTCCGCGCACATTTTT
polyT_22	TTTTTTGGTAGCGGTGGGTTGCAAACAAACCACCGCTTTTT
polyT_23	AATAGTTCCGTAGGTGGCATGGCAGCAGCCACTGGTTTTT
polyT_24	CACTGCATACATAGTTAGGGTGTGCGCCCTCGAACTTTTT
polyT_25	GGGAAGCATCTCAACTTGAAATCCCCCAACCCGGTAAGACTTTTT
polyT_26	TTTTTTATTTGTTTCATGTTTCTATGCGGGTCTTGTAGTTG
polyT_27	TTTTTCAGCAGATTACTTATCCGTTTTT
polyT_28	CGCGCAGATTCGGAAAAAGAGTTTTT

2.2.4 sc_EGFP4

Scaffold sequence for sc_EGFP4:

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Corresponding DNA origami designs and staple lists:

2.2.4.1 sc_EGFP4 20HB

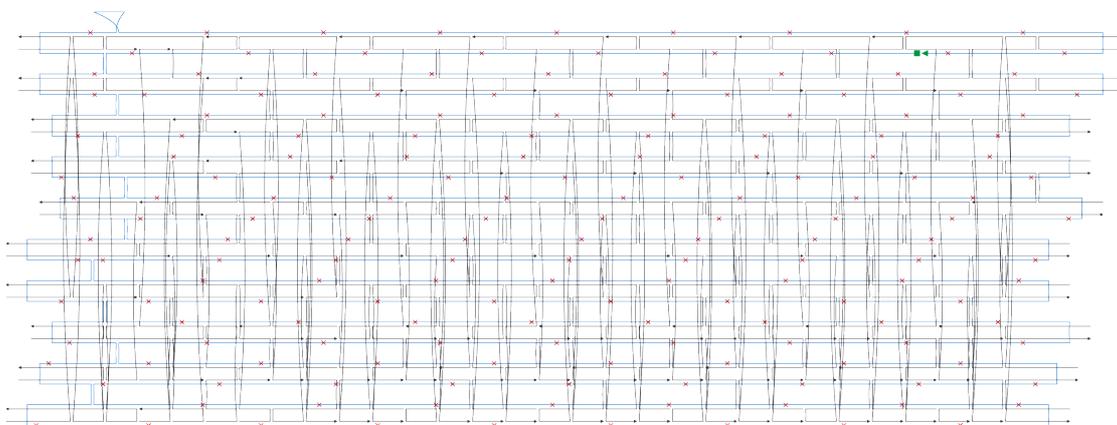


Fig. S32 | Scaffold routing and staple design for sc_EGFP4 20HB. Scaffold routing is shown in blue, staples are given in black, breakpoint for addition of sequence is marked by a green square (5') and arrow (3'). Design was prepared using caDNAo v0.1.

Table S19 | Individual staple sequences for sc_EGFP4 20HB.

Name	Sequence 5' - 3'
core_1	CAATGGAAGAAAGTCCCATAGGCTGTCTT
core_2	GAACATACATGACTAATACGTAGAGTGCTC
core_3	GGGGTCGTCTGCCAAAACCGCATACGGTTCA
core_4	TTTACCGAGTCAAACCGCTATCACCTCCCA
core_5	TATATGGTCAATGGGGTGGAGACGCGTCAAT
core_6	GTC AATAGGGGGCGTAGCTCAGAAACGCAAGAGTCTTCTCTGTCTCGTTACCGT
core_7	TGGGCATGTAAATACTCCACCCAGGTGGCG
core_8	CCAAGTAGAGTCCCTATTGGCGTCGAGCATC
core_9	TAATAGCGGTCATTATTGACGTCTGGCGTTT
core_10	TTGATGTATGGGCGGTCAGCCAGAAGGCCAG
core_11	TCCCCGTGTAAGTTATGTAACGCGCATAATGT
core_12	ACAAGCCGCAAAGTGGATGTCATGTCGCC
core_13	GGCGGCCGCTGGACACTTGTAGTCTGACTCCGCCGGAAG
core_14	CGGTGAACAGCTCCTCGTGCGCTCCGATCTGTCAACTTTA
core_15	CCGTCCAGCTCGACCACGGACTTGCAATGCTTTTGTGGCC
core_16	GACACGCTGAACTTGGTCCGGGTATATATGCCAGTTAA
core_17	GCCGTAGGTGGCATCTAGGTCATTAATAATTGCTACA
core_18	GCCCCAGCACCTCGGCGGGTCTGTGGTGGTAGCT
core_19	TTGTGGCTCCTTGAAGAAGATGGCCCTTGGCTGGTAG
core_20	GTTCTTCTTAGCCTTCGGGCATGGGGATGGGCAGCAGATT
core_21	TGTGGCGGCGTGCTGCTTCATGTGTGGCCGTTAGAAGAT
core_22	TCGGCGAGGAAGCACTGCACGCCGCCCTCGCGACGCTC
core_23	GTCCGCGAACGAGGGTGGGCCAGACTTCAGGGGGATTTT
core_24	AGGAGCAAGCATGATACAAAGGCGGCAGCAG
core_25	TCTTTCACAGGAGAAAATGAAAGCGATTGAGG
core_26	GAAGCAATACATAGTTAAGAATACCAGTCAACAGCTTGCCAGCCACCGTCGTATACGGG
core_27	TACAAGGAAATTTTGAATCCAGAGGTTGATATAGCGGGTCTGCTATTTTCGCTTTAAA
core_28	TAAAGAGACAGCAACCCGTCCCAGCAGCACT
core_29	CTCCTCAAAGCTTATTACTTGTACAGCTCGTTGATGCCTCTATTAATAATCAGTGCTTCGGGG
core_30	CGTTGCCTGACAACGGCGAGCAGTTGGCCG
core_31	CCACGCCAGAGAGTGATCCCAGCGGCGGCTACTCGATGTGTAGTTCGAGTAACTGCTGTTGA
core_32	TGCGTCAGCAAACACCCGACAACCTCGGT
core_33	GTGGGGGTAGCAGGACCATGTGATCGCGCTTGGTAGTGGTGTGCCATGAAGTTTCGTGCACC
core_34	AGGTGGTGGCAATGCACAGCCGACCCCAT
core_35	GAGCTGACGGTCTTTGCTCAGGGCGGACTGGCGGGGCCCGCTCGTCGATCTTCACTTTACC
core_36	GCAGGCGGGGTTCCCAAGCATGCCGGTCGAGGTGCCGTAACACTACGCGCAAAAA
core_37	ACAGTCGAAAGGAAGTCCGCTGCATACGGGCAATAAA
core_38	GCAAACAAGGACGTAGCAGAAGGAAGGATTTAGCAGAAGT

core_39	CTTCCAGGCCAGGTGGCAACACAGGGCCACAACATCCAG
core_40	TTTTATTAAAGGACGATGATTTCCAGTGCACAAGAGTAA
core_41	CCTACTCAAATTGTCAGTGCCCACCCAACCACGCAACGT
core_42	CCCACCCCAGCAGCGGGCAAGCCGGAAGTGGTGTCA
core_43	GGAGTGGGGCGCTGGCAAGTGTATAAATCAAAAATA
core_44	TTGTAGTTAAGGGAAGAAAGCGATGAGTGTTCATGAGCG
core_45	TATCTACGCGGGGAAAGCCGGCGAACTATTAATTATTGA
core_46	TATCGGGACCCTAAAGGGAGCCCCGGGCGAAATGTTGAAT
core_47	TCGCTCGCGTAGAGAATAGTGTATTCTGGCCC
core_48	AATCAGCTCATTTTTGCCGCGCTCTTGCCTTCTGTGATTTATCA
core_49	ATCGGCAAAATCCCTAGCGGTGAGCGCCACACTCTTACGGCCGAGC
core_50	AGACCGAGATAGGGTAAGGAGCGTCAATTGGATCACTCATTCCGCCTC
core_51	TTGGAACAAGAGTCCACGTGGCCTCAAGGACGTTGTCAGGGAAGCT
core_52	GGACTCCAACGTCAAACGATTTATCGATGTAAAAAGCGTAGTTTG
core_53	TATCAGGGCGATGGCCAGCCTAACTTACAGCAGGCGAGTGGCATCG
core_54	TTTTGTATTGGTATCCAGTGTGAGTCTCGATGCGGTTACCAGGGGTAAGACC
core_55	TTCCCGAGTTTACCAATGCCAGGCGGGCCA
core_56	GCGCACATAGGCCGAAAAACCCGACAGGACTATAAAGATAGTGGGCAAAAGTGC
core_57	GGGGTTCCATAATACCCGCTGCGCGTGAGCGAGCGAGCGC
core_58	GTATTTAGAAAAGAATACAAAATCGACGCTCAAGTCAGATTGACGTAACAAATAGGCAAAC
core_59	ATTTGAATAGTGCTCAGGCGCTAGCCAACCTCCATCACTAG
core_60	TATTGTCTGTTCCAGTTTCCATAGGCTCCGCCCCCTGATACTATGGGATACATTTTTTTGT
core_61	ATCAGGGTCGAAAACGTGAGAAAGGAATGATTAACCCGCCA
core_62	TTTTCAATAGAACGTGAACCGTAAAAAGGCCGCGTTGCAATGGGCGAGCATTTGAAAAAAA
core_63	TCTTCCTGATCCAGTGAGCTTGATAGCCATGCTCTAGG
core_64	CACGGAAAAACCGTCAAGGGTGAGCAAAAGGCCAGCAAGCGGGCCAACTCATACCTTTTCTA
core_65	AGGGCGACAACCTGATATCGGAAATTCGCCCTTAACTC
core_66	AAAATGCTGAACCATGGCGAATTCCGATCTTCTAGAGGAACTCCAAGGGAATAGAAAACTC
core_67	GTACTCAAAGCCCGGATTAAGGCTGCGCGCTTAAATCGTTAATATATGATAC
core_68	CATCCGTAGGCCTCAGTAACCACACCCCTAACCAAT
core_69	TGTCATGCGCCGAAGCAGATGGCTGGCAACTAGAAGCGGTCGCCAGATGCTTCGGCGTCA
core_70	GGTTATGGCAGAATGTCAAGGAAGGCACGGGGGAGGGGCAGAGAGGCATAATTTAGCAGAA
core_71	GAAGTAAGCCAAGGAGGAAAGGACAGTGGGAGTGGCACGGGTTCCAGTGTAAAAACGTT
core_72	GTTAGCTCCACCACGGGACAATCCCATGCAATTTCTCTCATGCTACTCCTCCGATTCTTACC
core_73	TACATGATGCCCTGTCCACCCCAGATGAATAGAATGACAAAGATCCTGTTGTGCCAAACCCACT
core_74	CTTACCAGCGGCGACTTGTAAAGTTTTGTAGTTTTCCCTTGGAAAGCTCCCTCTCCTTGCCA
core_75	CGGGAGGGGCTCCAGACTGGTGTAGCCGCGGGGGCAGATCTCGAAGACGCGGAAGCCGGGCAA
core_76	GTAGATAAAAAAGAGTAGAGAAAGCAGTTTCTATTGGTCTCCTTAAACTCATGTACCTTGATCC
core_77	ATAGTTGCTGCCGTGCTGTTGTAGTTGTAATC
core_78	TTCATCCAAACCACCCTCACCATGTAACCTTGATACTTACCTGCCATGTACTGCGGTGGT
core_79	TATCTCAGCTGGACGGCTTGTGCGCCATGATTGGATCC
core_80	AGGCACCTTGAAGCACCCACCCAGCAACTTCTGCAGGATCTGCACCATGGACGCGCA
core_81	CAGTTACAAGAAGTATCTTGAAGTTACCTCCATGCC
core_82	TGGTCTGAGGATCTCATACGTCGCTAAACCAGCTCTGCTTATATAGCACGCCACCTTTGAT
core_83	TCTAAAGTAGCGCTCTGCACGCTGCCGTCCGAACTCC
core_84	TAAATCAACGGGGTCTCCTCGCCGCGTACACGCCTACCGCCATTTTTGGAAAAGTGGAAAC
core_85	CTTTTAAAGGGTGGTCTGGGGGTGTTCTGCTCTCGTTGG
core_86	TGCTGAAGCATTGACACTTGTACTGCAACACAGGC
core_87	CCAGTTACCACCTAAACGAGTTGTAATGTTACGTGCGGGCAGCCTTT
core_88	CTTCGACTACGATATCGAACTTGTGTTGCCGTCTCC
polyT_1	TTTTTTATTAATAACTAGCTACCCGTAATTGATTACTTTTT
polyT_2	TTTTTCGCTTACCGGATACCTCTGTTCCAGCAGCCAAAATTCGC
polyT_3	CATTGACGGCTATGAACTAATGACGCTTAAGCACCCCTAATCATTTTT
polyT_4	TTTTTATTTTTGGTGCCAAAACCTGAAAAGTCCCGTTGTTTT
polyT_5	TTTTTTTCGGGAAGCGTGGCGCCGACAGTATCTGGTAACAGATTTTTT
polyT_6	GTCAGCTTGGGGCGGAGTTGTACGACATTTAAACTCCGGTCATGAGATTTTTT
polyT_7	TTTTTCGTTGGTGCAGATGAGGCACGGGCAGCTTGCTTTTT
polyT_8	TTTTTTGTAGGTATCTCCAATGGCCTAACTACGGCTTTTTT
polyT_9	TTTTTTTGTGCGGCAGCAGCAGTGTCTCAGGGAAGCGAAAAGTC
polyT_10	TTTTTGCTCCAAGCTGGGCTGTCGTATCCACATAGCGTAAATTTGAAGTGTACCCG
polyT_11	TTTTTCAATAGGGAGGGGTAGTGGTTTTT
polyT_12	TTTTTTGTCAGCCCGACCGCTCCGCAGAGCCATTAAG
polyT_13	TTTTTTTCCCAATCCTCCCTTGTCTGCTGGAGTCCCACGATCAATCTTTTACCTAGATCACGTAA
polyT_14	TTTTTCTTGAGTCCAACCCGCTGAGGCCGAGGGCGCTTATCCGGTAACTATCGTTTTTT
polyT_15	TTTTTAGTTTTTTGGTGCTATTGCTTTTT
polyT_16	TTTTTCGCCACTGGCGACCCTGCTTTTT
polyT_17	TTTTTGCAAAAACAGGAAGGCAGCGTTTCTGGGTGATTTTT
polyT_18	TTTTTTAGCAGAGCGAGTCAAGTAAAGACACGACTATTTTTT
polyT_19	CCAAGTCACTTCTGTATACCGCGCAGGTGCACGAACCCCCCTTTTT

polyT_20	TTTTTTCATTCAGCTCCCGATGAGTTCGCGCGTGGTTTTT
polyT_21	TTTTTGTTCTTGAAGTGGTGAGGCGGTGCTACAGATTTTT
polyT_22	TGACCCACCGATGCCCTTCAGTCGGTGTAGGTCGTTCTTTTT
polyT_23	TTTTTATCAAAAAGTTTGGTATGGCTTTTT
polyT_24	TTTTTACACTAGAAGTTTCTCCCTTTTT
polyT_25	TGCGCTCAATAGGTGCCTATTTCTCATAGCTCACGCTTTTT

2.2.5 sc_EGFP5

Scaffold sequence for sc_EGFP5:

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Corresponding DNA origami designs and staple lists:

2.2.5.1 sc_EGFP5 20HB

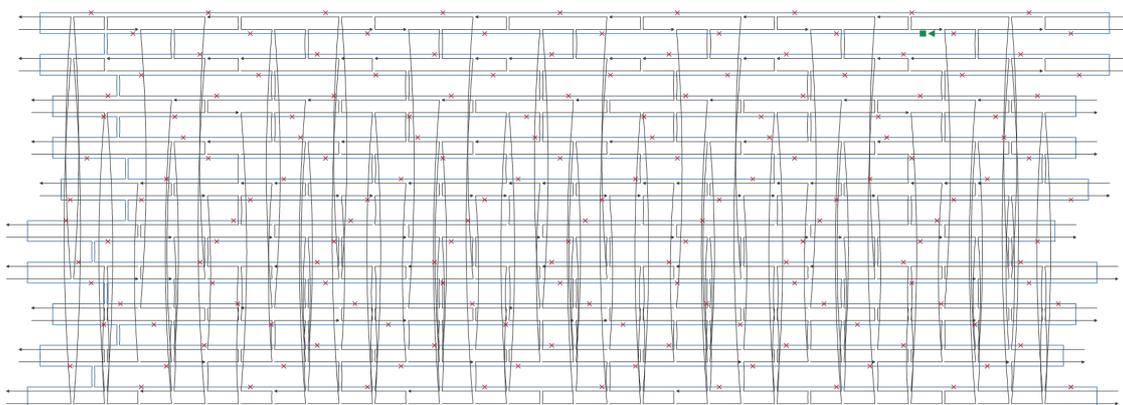


Fig. S33 | Scaffold routing and staple design for sc_EGFP5 20HB. Scaffold routing is shown in blue, staples are given in black, breakpoint for addition of sequence is marked by a green square (5') and arrow (3'). Design was prepared using caDNAo v0.1.

Table S20 | Individual staple sequences for sc_EGFP5 20HB.

Name	Sequence 5' - 3'
core_1	CAATGGAAGAAAAGTCCCATAAAGGCTGTCTT
core_2	GAACATACATGACTAATACGTAGAGTGCCCTC
core_3	GGGGTCGTCTGCCAAAACCGCATACGGTTCA
core_4	TTTACCGAGTCAAACCGCTATCACCTCCCA
core_5	TATATGGTCAATGGGGTGGAGACGCGTCAAT
core_6	GTCAATAGGGGGCGTAGCTCAGAAACGCAAGAGTCTTCTCTGTCTCGTTACCGT
core_7	TGGGCATGTAAATACTCCACCCAGGTGGCG
core_8	CCAAGTAGAGTCCCTATTGGCGTGCAGCATC
core_9	TAATAGCGGTCATTATTGACGCTGGCGTTT
core_10	TTGATGTATGGGCGGTCAGCCAGAAGGCCAG
core_11	TCCCCGTGTAAGTTATGTAACGCGCATAATGT
core_12	ACAAGCCGCAAAGTGGATGTCATGTCGCC
core_13	GGCGGCCGCTGGACACTTGTAGTCTGACTCCGCCGGAAG
core_14	CGGTGAACAGCTCCTCGTGCGCTCCGATCTGTCAACTTTA
core_15	CCGTCCAGCTCGACCACGGACTTGCAATGCTTTTGTGGCC
core_16	GACACGCTGAACTTGGTCCGGGTATATATGCCAGTTAA
core_17	GCCGTAGGTGGCATCTAGGTCATTAATAAATTGCTACA
core_18	GCCCCAGCACCTCGGCGCGGGTCTGTGGTGGTAGCT
core_19	TTGTGGCTCCTTGAAGAAGATGGCCCTTGGCTGGTAG
core_20	GTTCTTCTTAGCCTTCGGGCATGGGGATGGGCAGCAGATT
core_21	TGTGGCGGGTGTGCTTTCATGTGTGGCCGTTAGAAGAT
core_22	TCGGCGAGGAAGCACTGCACGCCGGCCCTCGCGACGCTC
core_23	GTCCGCGAACGAGGGTGGGCCAGACTTCAGGGGGATTTT
core_24	AGGAGCAAGCATGATACAAAGGCGGCAGCAG
core_25	TCTTTCACAGGAGAAAAATGAAAGCGATTGAGG
core_26	GAAGCAATACATAGTTAAGAATACCAGTCAACAGCTTGTCCAGCCACCGTCGTATACGGG
core_27	TACAAGGAAATTTTGAATCCAGAGGTTGATATAGACGGGTCTGCTATTTGCTTTAAA
core_28	TAAAGAGACAGCAACCCGTCCCAGCAGCACT
core_29	CTCCTCAAAGCTTATTACTTGTACAGCTCGTTGATGCCTCTATTAATAACAGTGCTTCGGGG
core_30	CGTTGCCTGACAACGGCGAGCAGTTGGCCG
core_31	CCACGCCAGAGAGTATCCCGCGCGGCTACTCGATGTGTAGTTCCGAGTAACTGCTGTTGA
core_32	TGCGTCAGCAAACACCCGACAACCTTCGGT
core_33	GTGGGGGTAGCAGGACCATGTGATCGCGCTTGGTAGTGGTGTGCCATGAAGTTTCGTGCACC
core_34	AGGTGGTGGCAATGCACAGCCGACCCCAT
core_35	GAGCTGACGGTCTTTGCTCAGGGCGGACTGGCGGGGGCCCGCTCGTCGATCTTCACTTTACC
core_36	GCAGGCGGGTTCCCAAGCATGCCGGTCCGAGGTGCCGTAACACTACGCGCAAAA
core_37	ACAGTCGAAAGGAAGTCCGCTGCATACGGGCAATAAA
core_38	GCAAACAAGGACGTAGCAGAAGGAAGGATTTAGCAGAAGT

core_39	CTTCCAGGCCAGGTGGCAACACAGGGCCACAACATCCAG
core_40	TTTTATTAAAGGACGATGATTTCCAGTGCACAAGAGTAA
core_41	CCTACTCAAATTGTCAGTGCCCACCCAACCACGCAACGT
core_42	CCCACCCCAGCAGCGGGCAAGCCGGAAGTGGTGTCA
core_43	GGAGTGGGGCGCTGGCAAGTGTATAAATCAAAAATA
core_44	TTGTAGTTAAGGGAAGAAAGCGATGAGTGTTCATGAGCG
core_45	TATCTACGCGGGGAAAGCCGGCGAACTATTAATTATTGA
core_46	TATCGGGACCCTAAAGGGAGCCCCGGGCGAAATGTTGAAT
core_47	TCGCTCGCGTAGAGAATAGTGTATTCTGGCCC
core_48	AATCAGCTCATTTTTGCCGCGCTCTTGCCTTGTGATTTATCA
core_49	ATCGGCAAAATCCCTAGCGGTGAGCGCCACACTCTTACGCGCGAGC
core_50	AGACCGAGATAGGGTAAGGAGCGTCATTGGATCACTCATTCCGCCTC
core_51	TTGGAACAAGAGTCCACGTGGCCTCAAGGACGTTGTCAGGGAAGCT
core_52	GGACTCCAACGTCAAACGATTTATCGATGTAAAAAGCGTAGTTTG
core_53	TATCAGGGCGATGGCCAGCACTAACTTCAGCAGGCGAGTGGCATCG
core_54	TTTTGTTATTGGTATCCAGTGTGAGTCTCGATGCGGTTACCAGGGGTAAGACC
core_55	TTCCCGAGTTTACCAATGCCAGGCGGGCCA
core_56	GCGCACATAGGCCGAAAAACCCGACAGGACTATAAAGATAGTGGGCAAAAGTGC
core_57	GGGGTTCCATAATACCCGCTGCGCGTGAGCGAGCGAGCGC
core_58	GTATTTAGAAAAGAATACAAAATCGACGCTCAAGTCAGATTGACGTAACAAATAGGCAAAC
core_59	ATTTGAATAGTGCTCAGGCGCTAGCCAACTCCATCACTAG
core_60	TATTGTCTGTTCCAGTTTCCATAGGCTCCGCCCCCTGATACTATGGGATACATTTTTTTGT
core_61	ATCAGGGTCGAAAACGTGAGAAAGGAATGATTAACCCGCCA
core_62	TTTTCAATAGAACGTGAACCGTAAAAAGGCCGCGTTGCAATGGGCGAGCATTTGAAAAAAA
core_63	TCTTCCTGATCCAGTGAGCTTGATAGCCATGCTCTAGG
core_64	CACGGAAAAACCGTCAAGGGTGAGCAAAAGGCCAGCAAGCGGGCCAACTCATACCTTTTCTA
core_65	AGGGCGACAACCTGATATCGGAAATTCGCCCTTAACTC
core_66	AAAATGCTGAACCATGGCGAATTCGATCTTCTAGAGGAACTCCAAGGGAATAGAAAACCTC
core_67	GTACTCAAAGCCCGGATTAAGGCTGCGCGCTTAAATCGTTAATATATGATAC
core_68	CATCCGTAGGCCTCAGTAACCACACCCCTAACCAAT
core_69	TGTCATGCGCCGAAGCAGATGGCTGGCAACTAGAAGCGGTCGCCAGATGCTTCGGCGTCA
core_70	GGTTATGGCAGAATGTCAAGGAAGGCACGGGGGAGGGGCAGAGAGGCATAATTTAGCAGAA
core_71	GAAGTAAGCCAAGGAGGAAAGGACAGTGGGAGTGGCACGGGTTCCAGTGTTAAAAACGTT
core_72	GTTAGCTCCACCACGGGACAATCCCATGCAATTTCTCTCATGCTACTCTCCGATTCTTACC
core_73	TACATGATGCCCTGTCCACCCCGAGATAGAATGACAAAGATCCTGTTGTGCCAAACCCACT
core_74	CTTACCAGCGGCGACTTGTAAAGTTTTGTAGTTTTCCCTTGGAAGCTCCCTCTCCTTGCCA
core_75	CGGGAGGGGCTCCAGACTGGTGAGCCGCGGGGGCAGATCTCGAAGACGCGGAAGCCGGGCAA
core_76	GTAGATAAAAAAGAGTAGAGAAAGCAGTTTCTATTGGTCTCCTTAAACTCATGTACCTTGATCC
core_77	ATAGTTGCTGCCGTGCTGTTGTAGTTGTAATC
core_78	TTCATCCAAACCACCCTCACCATGTAACCTTGATACTTACCTGCCATGTACTGCGGTGGT
core_79	TATCTCAGCTGGACGGCTTGTGCGCCATGATTGGATCC
core_80	AGGCACCTTGCAAGCACCCACCCAGCAACTTCTGCAGGATCTGCACCATGGACGCGCA
core_81	CAGTTACAAGAAGTATCTTGAAGTTCACTCCATGCC
core_82	TGGTCTGAGGATCTCATACGTCGCTAAACCAGCTCTGCTTATATAGCACGCCACCTTTGAT
core_83	TCTAAAGTAGCGGCTCTGCACGCTGCCGTCCGAACTCC
core_84	TAAATCAACGGGGTCTCCTCGCCGCGTACACGCCTACCGCCATTTTTGGAAAAGTGGAAAC
core_85	CTTTTAAAGGGTGGTCTGGGGGTGTTCTGCTCTCGTTGG
core_86	TGCTGAAGCATTGACACTTGATGTAAGTCCCAACAGGC
core_87	CCAGTTACCACCTAAACGAGTTGTAATGTTACGTGCGGGCAGCCTTT
core_88	CTTCGACTACGATATCGAACTTGATGTTGCCGTCTCC
polyT_1	TTTTTTATTAATAACTAGCTACCCGTAATTGATTACTTTTT
polyT_2	TTTTTCGCTTACCGGATACCTCTGTTCCAGCAGCCAAAATTCGC
polyT_3	CATTGACGGCTATGAACTAATGACGCTTAAGCACCCCTAATCATTTTT
polyT_4	TTTTTATTTTTGGTGCCAAAACCTGAAAAGTCCCGTTGTTTT
polyT_5	TTTTTTTCGGGAAGCGTGGCGCCGACAGTATCTGGTAACAGGATTTTTT
polyT_6	GTCAGCTTGGGGCGGAGTTGTACGACATTTAAACTCCGGTCATGAGATTTTTT
polyT_7	TTTTTCGTTGGTGCAGATGAGGCACGGGCAGCTTGCTTTTT
polyT_8	TTTTTTGTAGGTATCTCCAATGGCCTAACTACGGCTTTTTT
polyT_9	TTTTTTTGTGCGGCAGCAGCAGTGTCTCAGGGAAGCGAAAAGTC
polyT_10	TTTTTGCTCCAAGCTGGGCTGTCGTATCCACATAGCGTAAATTGAAGTGTACCCG
polyT_11	TTTTTCAATAGGGAGGGGTAGTGGTTTTT
polyT_12	TTTTTTGTCAGCCCGACCGCTCCGCAGAGCCATTAAG
polyT_13	TTTTTTTCCCAATCCTCCCTTGTCTGCTGGAGTCCCACGATCAATCTTTTACCTAGATCACGTAA
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polyT_15	TTTTTAGTTTTTTGGTGCTATTGCTTTTT
polyT_16	TTTTTCGCCACTGGCGACCCTGCTTTTT
polyT_17	TTTTTGCAAAAACAGGAAGGCAGCGTTTCTGGGTGATTTTT
polyT_18	TTTTTTAGCAGAGCGAGTCAGTAAGACACGACTATTTTTT
polyT_19	CCAAGTCATTCTGTATACCGCGCAGGTGCACGAACCCCCCTTTTT

polyT_20	TTTTTTCATTAGCTCCCGATGAGTTCGCCGTGGTTTTT
polyT_21	TTTTGTTCCTTGAAGTGGTGAGGCGGTGCTACAGATTTTT
polyT_22	TGACCCACCGATGCCCTTCAGTCGGTGTAGGTCGTTCTTTTT
polyT_23	TTTTTATCAAAAAGGTTTGGTATGGCTTTTT
polyT_24	TTTTTACACTAGAAGTTTCTCCCTTTTT
polyT_25	TGCGCTCAATAGGTGCCTATTTCTCATAGCTCAGCTTTTT

2.2.5.2 sc_EGFP5 20HB-exL

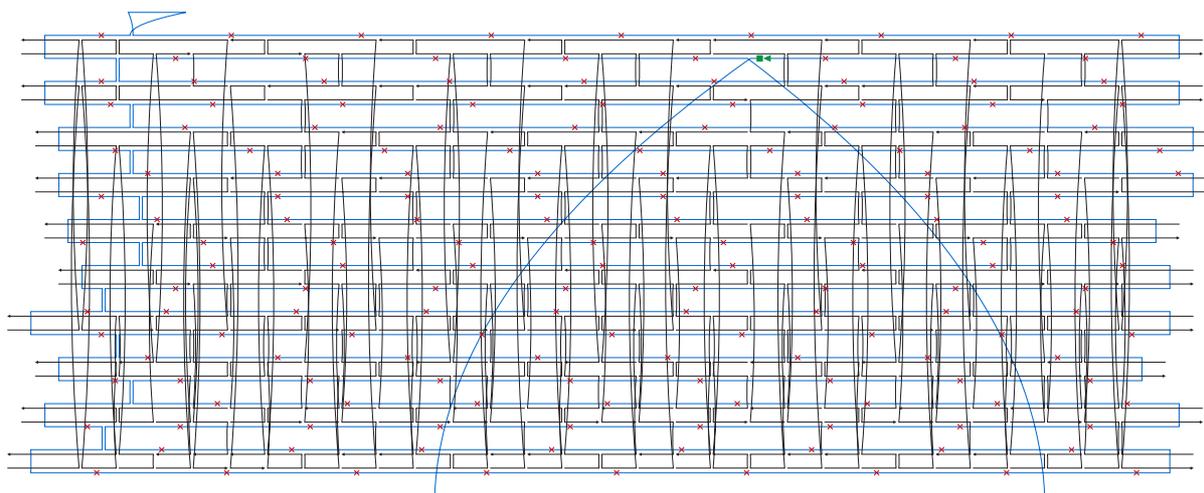


Fig. S34 | Scaffold routing and staple design for sc_EGFP5 20HB-exL. Scaffold routing is shown in blue, staples are given in black, breakpoint for addition of sequence is marked by a green square (5') and arrow (3'). Design was prepared using caDNAo v0.1.

Table S21 | Individual staple sequences for sc_EGFP5 20HB-exL.

Name	Sequence 5' - 3'
core_1	TTAACTACCTAATTAACCTCCACCCATTGACCATCACC
core_2	GCGTTTTTAGGGGGCGTACTTGGCAAGGTCAT
core_3	ATGTAACGCGGAACTCCCCTTGGAAAGTCCCGTTGATTTTGGTGCCACATTTACCTCTCAAGA
core_4	GTAAGTTAACTAATGACCCCGTATCTCAGT
core_5	AAACAAACCCATTTGCGTCAATGGTGAACCTTAGTGGTCC
core_6	AGACTTGGTATAGACCTCCCACCATCGCCCAAACCAGC
core_7	ATCCACGCATCTGACGGTTCACTCTTGTGGCAGCTCA
core_8	TAGATGTTTAAACCTGTCTTGTCTCGCCATAACTAC
core_9	TGCCAGTGCCTCACACCAGGATGCCCCAG
core_10	GTCTCGACAAGCCCAGGACACCTGATCCATAG
core_11	ACGCCGTAGCTTGCCGTAGGTGGCGTACACGC
core_12	CATGTGGTGCCGGACACGCTGAAAACCAGC
core_13	GCATGGCGTCGCCGTCCAGCTCGGACCAACT
core_14	AAGATGGTCCCCGGTGAACAGCTAACCTTGA
core_15	CGGGTCTCATGGCGGCCCTGTTTCTAT
core_16	CAGGGTGAAAGGCAAAGTGGATGGCAAGAG
core_17	AGCACGGGTTGCTCAGGGCGGACTGTCCCGG
core_18	GCTGGTAGACCATGTGATCGCGCATGCCCCA
core_19	GTCTCGGATCCCGGCGGGTACACAGTG
core_20	CACCTTGAATTACTTGTACAGCTAACGGGCC
core_21	ATGATATATGTAATCCAGAGGTTAACAGGA
core_22	ACTCCAGCTTAAGAATACCAGTCAAGCCATA
core_23	GGGGTCTGCCGTGCCGATGGGGGTGTTCTAGCACTGCTTTGCGCACAGCCGGA
core_24	CCAGCAGGTGGTCCGGCAGCTGCACGCTGCCTGCTGCTTATCGTGGTCCGGCTCC
core_25	CCGAGAGTATGTTGTGGCGGATCTTGAAGTTCCTTCGGCTTCATTCTGCTGCAA
core_26	CCAAGCTTTGCCGTTCTTCTGCTTGTCCGCCCTTGAAGGCGAGTTAGATACGG
core_27	ACAAATTTGACGTTGTGGCTGTTGAGTTGTCCTCGGCCGAAAGCGGTTGCCTG
core_28	GGTTGCGTCAGCAACACGAACTCTACAGGC
core_29	GCCACGTTGCCTGACCGTCCATGTGGTATGG
core_30	TCATAAAGAGACAGCGATTGGATCGATCAAG

core_31	AAGGAGGAGAAAATGAAATCTTTCTGTGCAA
core_32	GCGGCGATTGACAGGTGGTGGCATTCTCGTTTTAATAG
core_33	GAGGGCCGCAATAGCATGATACAAAAGGAGTCCGATCG
core_34	TCCCAATCAGTGCGAAAACCTCTATCCGC
core_35	GGAAGGGAAGAAAGCGAGCTTGACTCATGA
core_36	AGTTTGAACAAGAGTCTCGAGTTTGAGATCTTTAAAAGTTCCGCCAG
core_37	GTGGACTCCAACGTCAAGGAAGACACCCAATACGGGATGCCATTG
core_38	TCTATCAGGGCGATGGGCCATGCTCACCAGCCGGCGACCCGTCGTT
core_39	ATCACCTAATCAAGCCGCCGCGAAGGCAAATCAACCAAGTTCCCAA
core_40	TGCCGTAAGCACTAGTAGCGGTGCGACACGCGTAAGACCCCATGT
core_41	GGAGCCCCCGATTTAGAAAGGAGTCCTTTTTCACTGCAT
core_42	CAAAATCCTACCCTGCCAGCAGCCCCAGAATAGAATG
core_43	AAATCGGAGATCCTTGCGCAGACAGGGTCAGGTCAGGGTGGTCAC
core_44	ATAGGCCGCTATTAAGTCAGCCAGGCGGGC
core_45	ATTTTTGTTCCGATCATTGACGTCAATGGG
core_46	AGCGTTAATAAGTAGCTATTGGCGTACTAT
core_47	CGAAAAGTCACCTAGACCATCTGGGGCACCAGCGCTCCTGGACGTAG
core_48	AGAAAAAGGCCAGGATGTAAGTCCAAAGTGGG
core_49	AAGGATCTCTATAAATTGTTCTCGGTGTAGGTCGTTGCTCCTGCCATATATGATAGACC
core_50	CAGTTGAGGGGCCGATGATCTTTTACCCTGCCATTGACGTCAATGGGGTGGCGGGGGTC
core_51	ATGTAACCTTAACCAAAAGAACTCTCATAGCTCACGCTGTAGGTAATTGATTA
core_52	CTGATCTTAGATTTATGAACGAATCTGCTTAAAATCCCCGTGAGTCAAACCGCTGGGAACAT
core_53	CAGCATCTCGCGTTAAAAAACCCGCGCTTTCTCCCTCGGGAAGCGGGCGAATAAATCAG
core_54	GTTTCTGGTGATACCGATGAGATTCTGCAGGCCATTGATGTAAGTCCAAAACCGGTCAATGG
core_55	GTGAGCAAAATTGTACGTGAACCTCCGACCCTGCCGCTTACCGGAACGTAGATATTTGT
core_56	ATGCCGAGAGGGCTTATCCTTTTTACTTACCATGGTAATAGCGATGACTAATACGCAGTTA
core_57	AAAAAGGGCCGCGCAGGTGAGGCCCCCTGGAAGCTCCCTCGTGCG
core_58	GAAATGTTACTCCCCGCAATCTAATGGTCTCCACTGCCAAGTAGGAAAAGTCCCATATATGAT
core_59	GAATACTATGATTTCCCTAAAGCCCGACAGGACTATAAGATACCCAGCAAAATAACAAA
core_60	ATTGGAACACCCCACTGCCTGCTATTGCTGTTGAGTGTCAAAGAGGCTATG
core_61	GCCACATCTCAGACAGGGAATTCGCCCTTAACCACTATT
core_62	CTTGCCCGATTAGGAATACGTAGCCATGCTCTAAGGGCG
core_63	TGAGAATACAGGGTCATTAATTAAGGCAATGCCCACTA
core_64	TGTGACTGACAACAGAGCGTAACCACCACACTTTTTTGGCATTTC
core_65	TACTGTCTCGAGGCAGGGCGCTGGCAAGTAATCGGAA
core_66	GTAAGTAGTGCTCATCAAAGGAGCGAGTTCCGCCGTGGCAATAGGGACCTGCC
core_67	ACGTTGTTAATACCGCACCAGTGGCTGTCCAGCAGCGGGCAAGGCAGACACCTAAGCAGAAC
core_68	GTCACGCTGAGTTGCTCACACCACACGGAATTGTCAGTGCCCAACAGCTCATTTTGCCTCAA
core_69	AGCTCCGAGTCATTACAACCTCCAGGAAAGGACGATGATTTCCCGGCACCTTCGTGTATG
core_70	CATGATCTGCTTTTCTTTATACGAATCCAGGTGGCAACACAGGGCAAGGGGCAAGTGAGTAC
core_71	TTAGCTCCAATTCTCCGGGAAGAAGGGACGTAGCAGAAGGACGTGAGGCACAGATGCCATC
core_72	TTCGGTCCCAACATAGTTGTGCCAGGATGTTGCCGTC
core_73	AGATTACGCTCCATCCGACGAGCTTGCCGGTGGTGCAGAGGCGGAGTAAAAGGA
core_74	TCTACGGGTTGGGCGTAACTAGCTAGCTTAATGGCGCTT
core_75	GGTCTGACCTCATTCACTCGTGTCTTATCATGCGATGCAATTTCCCGAGCCC
core_76	GCTCAGTGCAGCAATTCGCCCTCCGGGGTAGCGGCTGA
core_77	AACTCAGCAGCTCATTTCCTAGAGCATGGCTTACCTGTC
core_78	TTAAGGGATAAAATTTTACTTTACTTATCAGGACAGTGGGAGTGACAACACC
core_79	TTTTGGTCCGTGACCCGTTTACGGACTTGAAGAAGTCCG
core_80	TATCAAAAAAGTCCCATGGCGGGTTAATCACTCTCCTG
core_81	AGGATCTTGCCACCTAAAACAGGCTTAATGAGGAAGGCACGGGGGGCAGCCA
core_82	AAATTAACCGTAAATGGGTGAGCAAAGGCAGGCGTTT
core_83	AATGAAGTTAGGGGTTAATAAGGCACGCTGCTGGCTGGCAACTAGACCGCGCA
core_84	TTTAAATTGCTGTAGTTGCTCACTGTAGTTGCCGTCGTC
core_85	AGTATAAACACTTGAACCGTAAAAAGGCCGTGGCGAAA
core_86	TGAGTAAATATTTGACATACTCTCGGGCGCTAGATCTCGAAGACGCG
core_87	ACTTGGTCTTTGTTCTGGAGAGTCCCTCGAACTTCA
polyT_1	TTTTTACGAGCATCACAGCCGGCGAACTTTTT
polyT_2	TTTTTAGCCCGACCGCTGCGCACGAACCGCTCTGCGAGATAGG
polyT_3	TTTTTTACCGTCATTGATTAATCAGTGTTTT
polyT_4	TTTTTTCGTTGAGTCCAACTTATCCAAGCAGCTGAAGCCAGTTACTTTTT
polyT_5	TTTTTGGTGCCTATCAGAAACTCAGTAAGACCAATATTTTT
polyT_6	TTTTTTCGCCACTGGCAAGTGTGGTAGCGGTGGTTTTTTTT
polyT_7	CGGTTCACTTGTGAGTCAGCGATTTGAAGCATTTATTTTT
polyT_8	TTTTTCCCTTCAGCTCGATGCTCCTTGAAGTCGATGTTTT
polyT_9	TTTTTTAGCAGAGCGAGGTATGTAGTGGTTGTCGGGCGAGCGAGGGTGGAAAGCTAGATGCAACTT
polyT_10	TTTTTGTATCCACATAGCGTAAGGCATTTCTATGGTCCGAGAG
polyT_11	TTTTTAGTTCTTGAAGTGGTGGAAAGCGAAAGGGTGTCT
polyT_12	TTTTTCCGCGGGGAAGGAGGTCGCTGGATTGAAGAGGCTATGGCAGCAATATTA

polyT_13	TTTTTCTACACCCCTTGCTGTGGGGGCCTAACTACGGTTTTT
polyT_14	TTTTTGTGGCGAGAAACCGGCAGCAGGTTTTT
polyT_15	TTTTTTGGTATCTGCCCCCGTTCTTTTT
polyT_16	TTTTTTCAGGGTATTGTGCGGGGAAAAAATCGACGCTCAAGTCAGAGGCGTTGCTGGCGGATAC
polyT_17	TTTTTCTTCGGAAAAAGCTCCTAGAAGGACAGTATTTTTT
polyT_18	AACGTTCTTCGGGTGGTTAATTGTCAGGTAGGCGGTGCTACAGTTTTT
polyT_19	TTTTTGCAGTGTTATCAAAAGCAGCTTTTT
polyT_20	TTTTTAAACAAACCACCCTATAGCTCTTGATCCGGCTTTTT
polyT_21	TGCCGGGGCCAGGGCACGGGCCACTGGTAACAGGATTTTT
polyT_22	CTGCTATGACAGTTTCTTCTGTACTGGGCATAATGCCAGGCGGGCCATTTTT
polyT_23	TTTTTAGGCACCTATCAAGTAAGTTGGCCTTTTT
polyT_24	TTTTTTTTTTGTTTGCGGTAACTATTTTT
polyT_25	CGCAGAAATGTTACGACATTGGTAAGACACGACTTATTTTT
polyT_26	ACCAATGCCGTC AATCCATAGGCTCCGCCCCCTGTTTT

2.2.5.3 sc_EGFP5 20HB-exLP

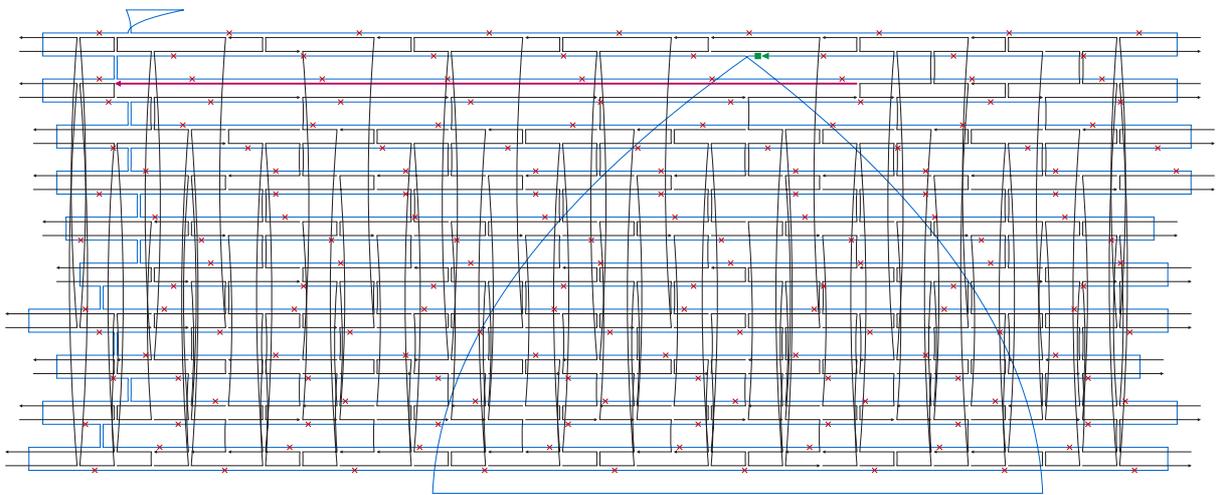


Fig. S35 | Scaffold routing and staple design for sc_EGFP5 20HB-exLP. Scaffold routing is shown in blue, staples are given in black, breakpoint for addition of sequence is marked by a green square (5') and arrow (3'). Long 154-mer 'primer' sequence at the promoter (5') region is given in pink. Design was prepared using caDNAno v0.1.

Table S22 | Individual staple sequences for sc_EGFP5 20HB-exLP.

Name	Sequence 5' – 3'
core_1	TTAACTACCTAATTAAGGGTGAGCAAAAGGCAGGCGTTT
core_2	GCGTTTTTAGGGGGCGTACTTGGCAAGGTCAT
core_3	GGCGGAGTAAAAGGATCTCAAGACAAAATCC
core_4	CCATTTGCGTCAATGGTGAACCTAGTGGTCC
core_5	TATAGACCTCCCACCATCGCCCAAACCAGC
core_6	ATCTGACGGTTCACTCTTGTGGCAGCTCA
core_7	TGCCCAGTGCCTCACACCAGGATGCCCCAG
core_8	TTAAACCTGTCTTGTCTCGCCATAACTAC
core_9	GTCTCGACAAGCCCAGGACACCTGATCCATAG
core_10	ACGCCGTAGCTTGCCGTAGGTGGCGTACACGC
core_11	CATGTGGTGCCGGACACGCTGAAAAACCAGC
core_12	GCATGGCGTCGCCGTCCAGCTCGGACCAACT
core_13	AAGATGGTCCCCGGTGAACAGCTAACCTTGA AAAATTA
core_14	CGGGTCTCATGGCGGCCGCCTGTTTCTAT
core_15	CAGGGTGAAAGGCAAAGTGGATGGCAAGAG
core_16	AGCACGGGTTGCTCAGGGCGGACTGTCCCGG
core_17	GCTGGTAGACCATGTGATCGCGCATGCCCCA
core_18	GTCCTCGGATCCCGGGCGGCTACACAGTG
core_19	CACCTTGAATTACTTGTACAGCTAACGGGCC
core_20	ATGATATATGTAATCCAGAGGTTAACCAGGA
core_21	ACTCCAGCTTAAGAATACCAGTCAAGCCATA

core_22	GGGGTCTGCCGTCGCCGATGGGGGTGTTCTAGCACTGCTTTGCGCACAGCCGGA
core_23	CCAGCAGGTGGTCCGCGAGCTGCACGCTGCCTGCTGCTTATCGTGGTCCGGCTCC
core_24	CCGAGAGTATGTTGTGGCGGATCTTGAAGTTCCTTCGGCTTCATTCTGCTGCAA
core_25	CCAAGCTTTGCCGTTCTTCTGCTTGTGCGCCCTTGAAGCGAGTTAGATACGG
core_26	ACAAATTTGACGTTGTGGCTGTTGTAGTTGTCTCGGCGAAAGCGGTTGCCTG
core_27	GGGTTGCGTCAGCAACACGAACCTCTACAGGC
core_28	GCCACGTTGCCTGACCGTCCATGTGGTATGG
core_29	TCATAAAGAGACAGCGATTGGATCGATCAAG
core_30	AAGGAGGAGAAAATGAAATCTTTCTGTGCAAA
core_31	GCGGCGATTGACAGGTGGTGGCATTCTCGTTTTAATAG
core_32	GAGGGCCGCAATAGCATGATACAAAAAGGAGTCCGATCG
core_33	TCCAATCAGTGCGAAAACCTCTATCCGC
core_34	TCCTTATCATGCGATGCAATTTCCCGAGCCC
core_35	ACTTATCAGGACAGTGGGAGTGACAACACC
core_36	CTTAATGAGGAAGGCACGGGGGGCAGCCA
core_37	GGAAGGGAAGAAAGCGAGCTTGACTCATGA
core_38	AGTTTGGAAACAAGAGTCTCGAGTTTGAGATCTTAAAAGTTCGCCAG
core_39	GTGGACTCCAACGTCAAGGAAGACACCCAATACGGGATGCCATTG
core_40	TCTATCAGGGCGATGGGCCATGCTCACCAGCCGGCGACCCGTCGTT
core_41	ATCACCTAATCAAGCCGCCGCGAAGGCAATCAACCAGTTCCTCAA
core_42	TGCCGTAAGCACTAGTAGCGGTGCGACACGCGTAAGACCCCATGT
core_43	GGAGCCCCGATTTAGAAAGGAGTCTTTTCACTGCAT
core_44	TCAAAAGAGGCTATGAACATAATGACCCCGTATCTCAT
core_45	AAATCGGAGATCCTTGCAGACAGGGTCAGGTCAGGGTGGTCAC
core_46	ATAGGCCGCTATTAATAACTAGCTAGCTTAATGGCGCTT
core_47	ATTTTTGTTTCCGATCTTCTAGAGCATGGCTTACCTGTC
core_48	AGCGTTAATAAGTAGCATGGCGGGTTAATCACTCTCCTG
core_49	CGAAAAGTCACCTAGACCATCTGGGGCACCAGCGCTCCTGGACGTAG
core_50	AGAAAAGGCCAGGATGTACTGCCAAGTGGGTAGATGT
core_51	AAGGATCTTTATAAATTGTTCTCGGTGATGGTTCGTTCCCTGCCATATATGATAGACC
core_52	TACCGCTGCCCCAGCACCCCCAGAATAGAATG
core_53	CAGTTCGAGGGCCGATGATCTTTCTACCGCTCCCATTGACGTCAATGGGGTGGAGACTTGG
core_54	ATGTAACCTTTAACCAAAAGAACTCTCATAGCTCACGCTGTAGGTAATTGATTA
core_55	CTGATCTTAGATTTATGAACGAATCTGCTTAAAATCCCCGTGAGTCAAACCGCTATCCACGC
core_56	CAGCATCTCGCGTTAAAAAACCCGCGCTTTCTCCCTTCGGGAAGCGGGGCGAATAAATCAG
core_57	GTTTCTGGTGATACCGATGAGATTCTGCAGGCCATTGATGTACTGCCAAAACCCGATCACC
core_58	GTGAGCAAAATTGTACGTGAACCTTCCGACCCTGCCGCTTACCGGAACGTAGATATTTTGT
core_59	ATGCCGCAGAGGGCTTATCCTTTTACTTACCATGGTAATAGCGATGACTAATACG
core_60	AAAAAGGGCCGCGCAGGTCGAGGCCCCCTGGAAGCTCCCTCGTGCG
core_61	GAAATGTTACTCCCCGCAATCTAATGGTCTCCACTGCCAAGTAGGAAAGTCCCATATATGAT
core_62	GAATACTATGATTTCCCTAAAGCCCCACAGGACTATAAAGATACCCAGCAAAATAAACAAA
core_63	ATTGGAACACCCCACTGCCTGCTATTGTCTGTTGAGTG
core_64	GCCACATCTCAGACAGGGAATTCGCCCTTAACCACTATT
core_65	CTTGCCCCGATTAGGAATACGTAGCCATGCTCTAAGGGCG
core_66	TGAGAATACAGGGTCATTAATTAAGGCAATGCCCCACTA
core_67	TGTGACTGACAACAGAGCGTAACCACCACACTTTTTTGGCATTTC
core_68	TACTGTCTCGAGGCAGGGCGCTGGCAAGTAATCGGAA
core_69	GTAAGTAGTCTCATCAAAGGAGCGAGTTCGCCCGTGGCAATAGGGACCTGCC
core_70	ACGTTGTTAATACCGCACCGATGGCTGTCCAGCAGCGGGCAAGGCAGACACCTAAGCAGAAC
core_71	GTCACGCTGAGTTGCTCACACCACACGGAATTGTCAGTGCCCAACAGCTCATTITGCGTCAA
core_72	AGCTCCGAGTCATTCACTCCAGGAAAGGACGATGATTTCCCGGCACCTTCGTGTATG
core_73	CATGATCTGCTTTTCTTATACGAATCCAGGTGGCAACACAGGCGAAGGGGCAAGTGAGTAC
core_74	TTAGCTCCAATTCTCCGGGAAGAAGGGACGTAGCAGAAGGACGTCAGGCACAGATGCCATC
core_75	TTCCGTTCCCAACATAGTTGTGCCCAAGGATGTTGCCGTC
core_76	AGATTACGCTCCATCCGACAGCAGCTTGCCGGTGGTGCAGA
core_77	CGCAGAAATGTTACGAAGTCCCGTTGATTTTGGTGCCAAAACAAAC
core_78	TCTACGGGGTCTGACCTCATTCTACTCGTG
core_79	GCTCAGTGCAGCAATTCGCCCTCCGGGGTAGCGGCTGA
core_80	AACTCACGTTAAGGGATAAAAATTTTACTTT
core_81	TTTTGGTCCGTGACCCCGTTTACGGACTTGAAGAAGTCG
core_82	TATCAAAAAGGATCTTGCCACCTAAAACAGG
core_83	AATGAAGTTAGGGGTTAATAAGGCACGCTGCTGGCTGGCAACTAGACCGCGCA
core_84	TTTAAATTCGTGTAGTTGCTCACTGTAGTTGCCGTCGTC
core_85	AGTATAAACACTTGAACCGTAAAAAGGCCGTGGCGAAA
core_86	TGAGTAAATATTTGACATACTCTCGGGCGCTAGATCTCGAAGACGCG
core_87	ACTTGGTCTTTCTGTTCTGGAGAGTCCGCTCGAACTTCA
core_89	CAGTTTACCGTAAATACTCCACCCATTGACGTCAATGGAAAGTCCCTATTGGCGTTACTATGGGAACATAC GTCATTATTGACGTCAATGGCGGGGTCGTTGGCGGTACGCCAGGCGGGCCATTTACCGTAAGTTAT GTAACGCGGAACCTC

polyT_1	TTTTTACGAGCATCACAGCCGGCGAACTTTTT
polyT_2	TTTTTAGCCCGACCGCTGCGCACGAACCGCTCTGCGAGATAGG
polyT_3	TTTTTTACCGTCATTGATTAATCAGTGTTTTT
polyT_4	TTTTTCGTCTTGAGTCCAACCTTATCCAAGCAGCTGAAGCCAGTTACTTTTT
polyT_5	CCCTTGGAACATTGGTAAGACACGACTTATTTTT
polyT_6	TTTTTGGTGCCTATCAGAAACTCAGTAAGACCAATATTTTT
polyT_7	TTTTTCGCCACTGGCAAGTGCTGGTAGCGGTGGTTTTTTTT
polyT_8	CGGTTCACTTGTCAGTCAGCGATTTGAAGCATTATTTTT
polyT_9	TTTTTCCCTTCAGCTCGATGCTCCTTGAAGTCGATGTTTTT
polyT_10	TTTTTTAGCAGAGCGAGGTATGTAGTGGTTGTCGGGCAGCGAGGGTGAAGCTAGATGCAACTT
polyT_11	TTTTTGATCCACATAGCGTAAGGCATTCTCATGGTCGCAGAG
polyT_12	TTTTTAGTTCTTGAAGTGGTGGAAAGCGAAAGGGTGCT
polyT_13	TTTTTCCGCGGGAAGGAAGGTCCGCTGGATTGAAGAGGCTATGGCAGCAATATTA
polyT_14	TTTTTCTACACCCCTTGCTGTGGGGGCCTAACTACGGTTTTT
polyT_15	TTTTTGTGGCGAGAAACCGGCAGCAGTTTTT
polyT_16	TTTTTTGGTATCTGCCCCCGTTCTTTTT
polyT_17	TTTTTTCAGGGTTATTGTGCGGGGAAAAAATCGACGCTCAAGTCAGAGGCGTTGCTGGCGGATAC
polyT_18	TTTTTCTTCGGAAAAAGCTCCTAGAAGGACAGTATTTTTT
polyT_19	AACGTTCTTCGGGTGGTTAATTGTCAGGTAGGCGGTGCTACAGTTTTT
polyT_20	TTTTTGCAGTGTTATCAAAGCAGCTTTTT
polyT_21	TTTTTAAACAAACCACCTATAGCTCTTGATCCGGCTTTTT
polyT_22	TGCCGGGGCCAGGGCACGGGCCACTGGTAACAGGATTTTT
polyT_23	CTGTCTATGACAGTTTTCTCTCTGTACTGGGCATAATGCCAGGCGGGCCATTTTT
polyT_24	TTTTTAGGCACCTATCAAGTAAGTTGGCCTTTTT
polyT_25	TTTTTTTTTTGTTGCGGTAACATTTTT
polyT_26	ACCAATGCCGTCAATCCATAGGCTCCGCCCCCTGTTTTT
Loop_1	AAGGAACCCCTAGTGATGGAGTTGGCCACTCCCTCT

2.2.5.4 sc_EGFP5 16HB_2

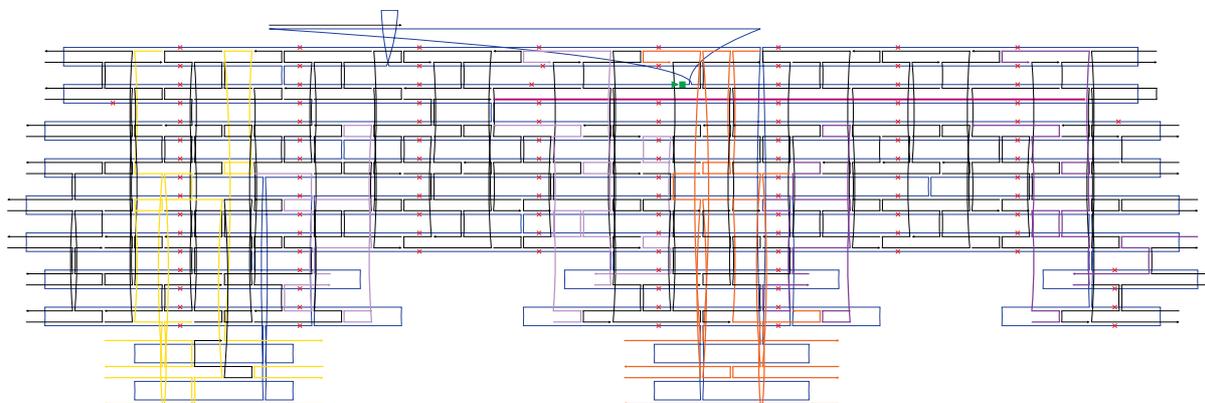


Fig. S36 | Scaffold routing and staple design for sc_EGFP5 16HB_2. Scaffold routing is shown in blue, staples are given in black, breakpoint for addition of sequence is marked by a green square (5') and arrow (3'). Long 154-mer 'primer' sequence at the promoter (5') region is given in pink. Protrusions 1 and 2 are shown in yellow and orange respectively, recess' 1 and 2 are given in light and dark purple, respectively. Design was prepared using caDNAo v0.1.

Table S23 | Individual staple sequences for sc_EGFP5 16HB_2.

Name	Sequence 5' – 3'
core_1	AGAAGATCCTTTGATGGTCATGCACGCTCAATTGTTGGGCAA
core_2	TTGCAAGCAGCAGATTACGCGCATCGACGCGTTGTTGCGCCAGT
core_3	TTTTTCCGTTTCAGCCCGACTCGACCAGGATGGGCACCTTTTT
core_4	TTTTTGAACGAAAACATATGAGTAAATTTTT
core_5	GGAAGCGTAGATAAGTAGCATGGCGGGTTAATTGGAAGGAAACGCACACCAGGG
core_6	TTTAAATTAATAAGGATCTTACCAGAAAAAAA
core_7	CCCCCTGAGAGGTGGCGAAACAGGTTTTTTGTGACTGGTG

core_8	CGCGTTGCGGCGTTTCCCCCTGGACGGCAAAC
core_9	AAGGCCAGGAACCGTAAAAAGGCGACCTCCCTTGTAACCACACCTAC
core_10	GGTACGTGGCGCTTTTCTCATAGCCTTATCGC
core_11	GAATTCCGATCTTCCCTAGAGCATCAAACCTCCGGATGTCAGCGCGGGTGATCTTAC
core_12	TTAAGGGCCTCAGTTCGGTGTAGGATCGTCTT
core_13	ATTACTATTAATAACTAGCTAGCCTTGGAAAGCCTGGACAGAAGATG
core_14	TTTTTCTTGGTCTGACAGTTACCAATGCTTAAATCAATCAGTCTATTGTGCTTTG
core_15	AGGCACCTATCTCAGCGATCTGTCTAGATCCTAAGTAGTTCATTGCTCTATTGTCGGTAACCA
core_16	TATTTGCCACGCTCACCGGCTCGATGAGTAGGGTCAAGGAATTGTTCAAAGGG
core_17	TTCATCCATAGTTGCCTGACTCCCGGCTCCGCGACGGTTCACGACCCCTGCCCAAAGAATA
core_18	CGTCGTGTGGCCCCAGTGCTGCGAAAGCGA
core_19	CATATATGGGCTATTTTTTTTTTTGAACATAATGACCCACGCCCA
core_20	GCCAGCCGAGCCGGCACAACAGATGAAAGGACTGGCCCACTACGTGAA
core_21	TTTTTCCAAAACCGCATCACCATGGTGCTCATCGCCGAT
core_22	TTTTTCTGCACTTTATCCGCCTCCATCCTAAAGTATCACGTTAAGGGATTTT
core_23	CAGCTCTGTTACCATCTAGATAACTACGATAC
core_24	CGCCATTGCCAGCAATCTCCTGTTCCGACCCCTTCGGAGCCCGGCG
core_25	ACGACATTTTCATTAACACCTGTCCGCCTTTCT
core_26	CATTGACCGATGTTATCCAGAGGTTGATTGGATCCAATGCCAAGTGGTGGAGA
core_27	GTCAATGGAGGAAAGTCCCATAAGGTCATGTATTTTGTA
core_28	CCGCTATCCGTAATTGTGGGCTGTGTGCACGAACCCCTTTTT
core_29	CAGTGCCACTAAACCATTTTAAAGTCCCGGAAAGGAGCTGACAGGGGAGGGCCTTATATA
core_30	ACCTGTCACCGTACGCTTGTGAATGCCCAACCAAGTGGGGGTTGATATGATA
core_31	CCCAGTTTTGAAGTCGTTTGTAAATTCCTTTTTCAATATTATTGAAGCAAAAACATCAGCTCG
core_32	CTCTGTCTTACCGGATTACCTAATTAAGGGTGAGCAAAAGTGCCTCAA
core_33	GGCGGCTCCCCGTTAGTGGTCTTGTACAGCTCGTCCATGCCGAGGTAATAG
core_34	CAGCTCCTCACGAGGGCAGATGAAGCGGCTGAAGCACTGCACCTTTTT
core_35	TTTTTACCCCGGTGAATTGATGTAAGTGTGTTTTT
core_36	TTTTTCCCAACGATTATCTACGTAGCTTTTT
core_37	GCAAAAACGTTGGTGTAGATTATCAAAAATGAAGTTTTAATCAGTGTAACCA
core_38	TAAGATGCTGCGCAACTCAAGTCACGAGCATCACAAAA
core_39	AGTACTCTAAATCAACCCACCCCCAGAAATTTCT
core_40	CATTCTGAAAACCCCGCTGGTAGCGGTGGTACTATAA
core_41	GAATAGTGTATGCGGACCAATACCCAGG
core_42	TCAATACTTAAATTATGCCCTGGAAGGCCCTGACAACGGGCCA
core_43	TCATTCATTCACTGGCAGCAGCCACTGGTAACCCCTTCGCCTATCATCCCGTTGATGCCG
core_44	GGAAAACGTTCTTCGGGGCGGCCAGAGAAAGGTAGGTAT
core_45	GGACACGCGAGTCCAACCCGTAAGACACGATCACGCTGCAAAGT
core_46	TGAAGTTGTGGCCGTCCGTAGGGTGCCT
core_47	GGCCGAAATCGGCAAAATCCCTTAAACCAAGTTACCTGCCAGATACCATGGCGTTTTTCCATA
core_48	ATCAGCTCATTTTTTACGACCGAGCTCCTTAATCGTGCGC
core_49	TGGCATCGCCCTCGCCCTCAAACCTCAAGCTTGTAGTTGCCGTGCGCGTCT
core_50	C TTCAGGGTCAGCTTGTTACGTCGCTCACCATCTCCAAGC
core_51	TTTTTTCAGCTTGCCGGTGGTGTGGGCCAGGGCACGGGTTTTT
core_52	TTTTTCATGCTCTAGGAACATAATCAATTTTT
core_53	TCGGGAATGTATGGCTAGTTACATCGGGGTCTGACGCTCAGTGTTTTT
core_54	AACTCGAAGGGCGAGATGATTTCCCGAC
core_55	CTCGAATTTTACCTTGATTTTTGGTAATGCCAGGCGGGCCATTTACC
core_56	ACCTTCTCCGCGGGGCAAGGCAGGCGGGCTGGATTAACACCACGGAAGGCACATGCCTG
core_57	TTAGGAAAGGACAGTGCTTGTGTAACCTCTGTTTTCTGT
core_58	TGTGGCTGCCAGTTGAAAATGCCGCAAAAATCATACTC
core_59	CCATGATAGAGTTGTTTCGTCAATAGGGGGCTACTTGGCCAGCGTAT
core_60	GTGGCGGATCTTGAAGTCACCTCGGTAAGACC
core_61	TTTTTAGATCTCGAAGACAATCCAGTTTTT
core_62	GGCCGACGGAAGGGCCGAGCGCAGAAGTGGTCTTTTT
core_63	GCAGGCCAAGGGACGTAGCAGAAGGACGTCCAGCCAAGGGCTGGCA
core_64	GTGGTGGCTCAGACAATGCGATGCATAGAATGTTGATACT
core_65	ATAGTTAATCTTCTGCATAAAGAAGCGTTTTCTGGGTGAGTCCCCGAA
core_66	GAATACCATGAAAGCCATACGGGAAGCAATCAACTCCTTGTGCGGATGCGGTT
core_67	GCTTATTACGGCGAGCTGCACGCTGCTTGAACCTGTGG
core_68	GAGGGCCGCGGGAAGGAAGGTCCCCAGATTTGCTAGAGT
core_69	AGCATGATGCGTAAAAGGAGCAACGTCATTGA
core_70	TTTTTCGTTGGGGTCTTTTTGTCCGGTTTTT
core_71	TTTTTTGGCAACACAGGCGAGCCGCGCAGGCGGAAGA
core_72	TCCAGCAGGGTGGTGGGCATGGTCCGGGTA
core_73	CGAAAAACGGGAGCCCCACACCTAAGGAGCGGCTCCTCCGATCGTTCATGGTTA
core_74	TTTTTCCGTAGGTCAGGACCATGTAGTCCGCTTCTTTTT
core_75	TTTTTGTTTTTTGGGGTTCGAGCCATCCCGATCCTTAACTAGAAGGCACAGTCCGAGGCTTTTT
core_76	GTGCCGTACAATGCGCCATGCTACTCAAGGCGTCATTCAGCTCCGGTTTTTTTT

core_77	ACCCTAAAGCTCTATCGTCCCCAGCGGGGGAGGCCGGAAATCAGCAA
core_78	CCGGTGTAGCGGTCAAGTGTGTACGTGGACTCCTCCCCGGAGTGGCTAATAGTTCCGCGTGAC
core_79	AAGGAACCCCTAGTGATGGAGTTGGCCACTCCCTCT
core_80	CAGTTTACCGTAAATACTCCACCCATTGACGTCAATGGAAAGTCCCTATTGGCGTTACTATGGGAACAT ACGTCAATTATTGACGTCAATGGCGGGGGTCTGTTGGGCGGTGAGCCAGGCGGGCCATTACCCTAAG TTATGTAACGCGGAACTC
rec1_1	CGACAAGTGGGGCGTAGACGTCCACATAACAAGGCATTAAGCGTCA
rec1_2	CTCTCATGCCATCCGCGCTGCGTTCCCAATCCAACGCAGTGCCCAACA
rec1_3	GCAGCTGGCAATAGGGAGGGGAATGATACCAGGATCT
rec1_4	CCACGTTGTAGTTGCTCCTCTATTGGTTTGTCTTAAAAGAGTTGGTAGCTCTTGATCAGCTCCC
rec1_5	ATTAAGATCCAGTTTGACCGAGATAGGGTTG
rec1_6	TTGAATAACGGGAATAAATGTTGCCGTACTCCAACGCCTACCACTTGTACTGCTGCAAGTGGG
rec2_1	TGTAAGCTGTGCGCTTTCACCGACAGCAACCAG
rec2_2	TTTTTCAGCAGCACGGGGCCGGGTAGTGGGCTCAGGGCGGACTGGAGTG
rec2_3	AGAAAAGTCAATCTTTCACAACTGGGCATGCCAAAA
rec2_4	CGAACGGGGGTGTTAGCCTTCCGCCCTTCCGTCCAGCCGCTGCGCCTTATCCGGTAACTTCGTTCCG
rec2_5	CTTCAGCAAACCCACTCGCTGTTGAGATCCAGAAAAGTGCAATAGGTG
rec2_6	TCATGTGGCGGACTTGCCTGGACGTCTGCTGGGAGTCAAACGATGACTAATACGTAGATGTAC
pro1_pass_1	CTTTCTAGATCCCCCTAAGTTGGTTTTT
pro1_pass_2	ACAGGCATGCGGTTAGGGATCTCATGGCAGCACTGCATAATTCTTTT
pro1_pass_3	TCGCCCTTAATTAAGGAAGCACTAGAACGTGGTTTTT
pro1_pass_4	TTTTCCGCAGTGTATCACTGTCAGAAGATGTTGTTAATGTT
pro1_pass_5	TTTTTCTTACTGCTTGGCGCTAGGGCTTTTT
pro1_pass_6	TTTTTCGAGAAAGGAAGGGAAAAGCCGGCAATCGGA
pro1_pass_7	TTTTTGCTGGCAAATTTAGAGCTTGACGGGGAGAAAAGCGAGCCGCGCT
pro2_pass_1	TTTGGTATACATAGCACGGTGCTAATATTTGAATGTATTTAGAAATTTTT
pro2_pass_2	AGAGTCTTACCGCGCCCTGCGCTCCACTAGAAGGACAGTAAGGATTAGCAGAGTTTTT
pro2_pass_3	TTTGTTAAGCGGATACCAGAGTTCCTGAAGTGGTGGCTTTTT
pro2_pass_4	GTTAATATAAGTGCCAGGGTCCGCGCACATTCATTATCAGGGTTTTTT
pro2_pass_5	TTTTTCTAACTACGGCTATGCTGAAGCCAGTTATGCCGCT
pro2_pass_6	TTTTTCGAGGTATGTAGGGAACTTT
pro2_pass_7	TTTTTTATTGTCTCATGAAATTCGCGGGGATAAT
pro2_pass_8	TTTTTAATAAACAAATAGCCTAAATTTTCGATGTTCTTTTAC
pro1_x8_1	CTTTCTAGATCCCCCTAAGTTGGAAGCATT
pro1_x8_2	ACAGGCATGCGGTTAGGGATCTCATGGCAGCACTGCATAATTCCTCGTTCA
pro1_x8_3	TCGCCCTTAATTAAGGAAGCACTAGAACGTGGCGAGTCCC
pro1_x8_4	TATTATTGCCGAGTGTATCACTGTCAGAAGATGTTGTTAATGTT
pro1_x8_5	CTGTCTATTCTTACTGCTTGGCGCTAGGGCACCTGCC
pro1_x8_6	CCTTAACTCGAGAAAGGAAGGGAAAAGCCGGCAATCGGA
pro1_x8_7	TGATACTTGCTGGCAAATTTAGAGCTTGACGGGGAGAAAAGCGAGCCGCGCT
pro2_x8_1	TTTGGTATACATAGCACGGTGCTAATATTTGAATGTATTTAGAAAACTGAGG
pro2_x8_2	AGAGTCTTACCGCGCCCTGCGCTCCACTAGAAGGACAGTAAGGATTAGCAGAGCTACGGCT
pro2_x8_3	TTTGTTAAGCGGATACCAGAGTTCCTGAAGTGGTGGCAGTTTTTAA
pro2_x8_4	GTTAATATAAGTGCCAGGGTCCGCGCACATTCATTATCAGGGTAATAGGTG
pro2_x8_5	AAAAATGACTAACTACGGCTATGCTGAAGCCAGTTATGCCGCT
pro2_x8_6	TGGCCTAACGAGGTATGTAGGGAACTTT
pro2_x8_7	GTAAGACCTATTGTCTCATGAAATTCGCGGGGATAAT
pro2_x8_8	CGTACATGAATAAACAAATAGCCTAAATTTTCGATGTTCTTTTAC
pro1_x5_1	CTTTCTAGATCCCCCTAAGTTGGAAGCA
pro1_x5_2	ACAGGCATGCGGTTAGGGATCTCATGGCAGCACTGCATAATTCCTCGT
pro1_x5_3	TCGCCCTTAATTAAGGAAGCACTAGAACGTGGCGAGT
pro1_x5_4	TATTGCCGAGTGTATCACTGTCAGAAGATGTTGTTAATGTT
pro1_x5_5	TCTATTCTTACTGCTTGGCGCTAGGGCACCTG
pro1_x5_6	TAACTCGAGAAAGGAAGGGAAAAGCCGGCAATCGGA
pro1_x5_7	TACTTGCTGGCAAATTTAGAGCTTGACGGGGAGAAAAGCGAGCCGCGCT
pro2_x5_1	TTTGGTATACATAGCACGGTGCTAATATTTGAATGTATTTAGAAAACTG
pro2_x5_2	AGAGTCTTACCGCGCCCTGCGCTCCACTAGAAGGACAGTAAGGATTAGCAGAGCTACG
pro2_x5_3	TTTGTTAAGCGGATACCAGAGTTCCTGAAGTGGTGGCAGTTTT
pro2_x5_4	GTTAATATAAGTGCCAGGGTCCGCGCACATTCATTATCAGGGTAATAG
pro2_x5_5	AATGACTAACTACGGCTATGCTGAAGCCAGTTATGCCGCT
pro2_x5_6	CCTAACGAGGTATGTAGGGAACTTT
pro2_x5_7	AGACCTATTGTCTCATGAAATTCGCGGGGATAAT
pro2_x5_8	ACATGAATAAACAAATAGCCTAAATTTTCGATGTTCTTTTAC

2.2.5.5 sc_EGFP5 16HB_3

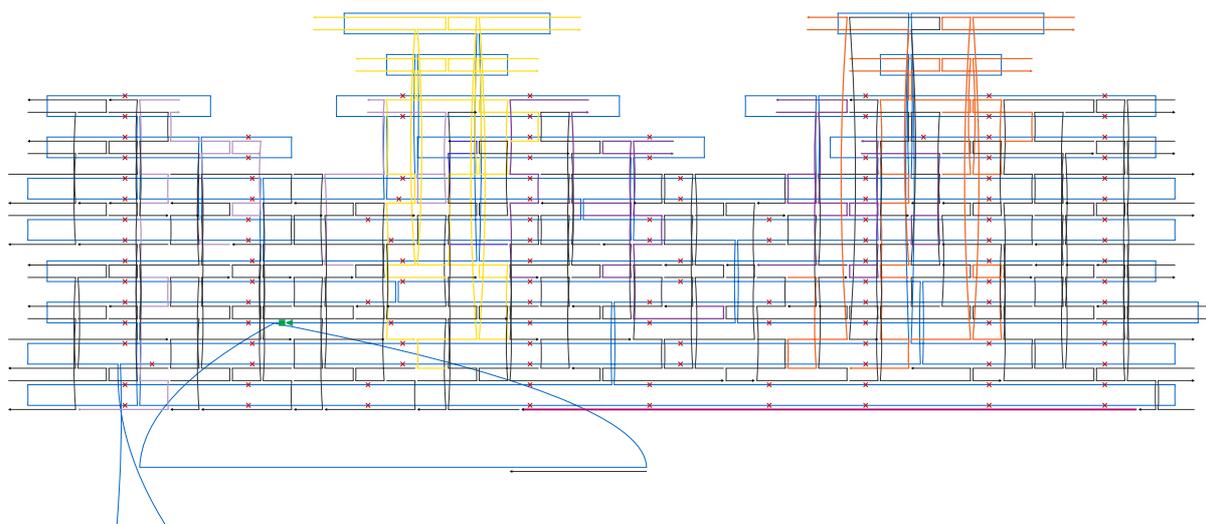


Fig. S37 | Scaffold routing and staple design for sc_EGFP5 16HB_3. Scaffold routing is shown in blue, staples are given in black, breakpoint for addition of sequence is marked by a green square (5') and arrow (3'). Long 154-mer 'primer' sequence at the promoter (5') region is given in pink. Protrusions 1 and 2 are shown in yellow and orange respectively, recess' 1 and 2 are given in light and dark purple, respectively. Design was prepared using caDNAo v0.1.

Table S24 | Individual staple sequences for sc_EGFP5 16HB_3.

Name	Sequence 5' – 3'
core_1	TTTTTAGGACCATAGTTGTA CTCCAGTTTTT
core_2	TTTTTCCGAGCGCATCCAGTCTATTATTTTT
core_3	AGAAGTGGCAGCCAGCCGGAAGGGTTTTT
core_4	TCCTGCACCGCGTGAGGCTACACTTCTTGAA
core_5	TTGATTGGATCCAAGCTTCTCTGAGGTCAGG
core_6	CGGCGGCGCGCTTCTCGTTGGGGTCTTTGCTCTCGGCCATGATGCCCTGAGTGGCA
core_7	GTGGATGTCAGTAAGAAGTGATCCTTGAAGTCGATATAGACTAGAAGG
core_8	TTTTTGGAGAGAATTCAGGGTCAGTTTTT
core_9	TTTTTGCGCTCTGACAGGATTAGCAGTTTTT
core_10	CTGAAGCCACAGTATTTGGTATCTTTTTT
core_11	CCCGGTGACAGAAACGCAAGAGTCTTATTACTACCAGG
core_12	TTTTTATTGTTGCCGGGAAGCTAGAGTAAGTAGAATAAAC
core_13	TTAATAGTATCGTGGTGTACGCTTCTATCAG
core_14	GCCCCAACAAAGGAGCTGACAGGTGATCACCT
core_15	CATACGGGAGAATCCAGGTGGCAGAGCCCC
core_16	CCTTGATGACGCGGAAAATGTTAATATCGGGACGGTCACGCTGCGCGTGCCTGGC
core_17	CGTTGTGGCTGTTGTGTGATCGGTACGAACTCCAGTTTTT
core_18	GGTCATGAGGGTCTGATTACCGGAGGCGCTTTCTCATAGCTCACGCT
core_19	TCAGCTCGCCAGGGTGCCTATACAGCTCCTCGCCCTTGCTCACCGCCGGACA
core_20	TTTTTCTTGTGCCCCAGGGGGGGAGTTTTT
core_21	TTTTTAGCGAGGTATGTAGGCGGTGCCCGCTCC
core_22	TACAGAGTAGAAGGAGTTACCTTCGGAAAAAGAGTTGGACTTATCGTGGTGGC
core_23	GCAGATTATTGAGTCTACCTAATTAAGGGTGTAGCTAGC
core_24	CGCTCAGGGGCTGTTGGCGTTTTTCCATAGGCTCCGCGTGCACCC
core_25	TGCTGCTTGTCTTGAAGAAGATTTAAAGCCGAAGGG
core_26	CACGGGCACGCTGAACAGACCTCCCACCGTACACGCCTACTTGATTTTGTGGTGCAGTCCTCC
core_27	GCTTGCCGGGTGCCAACCTTCCAGACACTACTGCTATTGTCTTCCCAATCCTCCACAGATG
core_28	GCCACTGGAGATAAGTAGCATGGCTAGAGCAT
core_29	GACCGCTAAGGCCAGGAACCGTAATGACCC
core_30	TTCAGCCCTTTGATCTGCGTTTTCTGGGTGCAG
core_31	TCCAAGCTTGAACGAAACTGATCGAATAAGGGTTAAAATCGATTTAGAGCTTGACGGGGAAG
core_32	TCGTTGCGGTAGGTATCTCTCTGTTCCGACCCTAAAGTAACGTTAAG
core_33	CTTGATACTACGTACAGAGGTGGCGAAACCCAGAAGTCGAAAACCGC
core_34	CAGCTCGATTACGGTTCACTAAACCAGCTCTGACGTCAATGAGGGTGGGATGCGGT
core_35	TTTTTGGCGTAGGTGGCTTGCCTCAATGGGGCGGAGTTTTT

core_36	CTACAGGCTTGCGCAACGTTGTTGTGGCAATCGGGAGGGAAAGCGGT
core_37	AAGCGAAACCGATCGTCCCCTCGTATCTCAAGAAGATCC
core_38	ACGTAGCACCGGCGAACTAAATTGTAAGCGTTCCAATAGCGATGACTAATACGTAGATGTACT
core_39	GCAGCAGGGCCGTCGCCGATGGCGGGCAG
core_40	CACAGTCGAGCATGCCTCAGACAATGCGATGTCATTGA
core_41	TTTTTGGCAAACACCCCTTGCTGTCCTTTTTT
core_42	TTTTTAGCTCCGGATTAAGAACGTGTTTTT
core_43	GCGACACGTCCCAGCGCAAGCAATAGCATGATACAAAGGCAGGTGCGCT
core_44	GAATACTCATCACCATGGTACTCCCCCTGAGGTGTAGG
core_45	ATACTCTGTGCCACCGTGGCGAGAAAGGAAGGAAGACCGCTGGA
core_46	CAATATTATTGCCGTCCATGTGGTCGGGGTA
core_47	TTGAAGCATCCACGCGGAAAGTCCATAAGGTCATGTAAAATAAACGGTTATTG
core_48	AAGGCACATGTTGCGATGAACAGGCAACGCCTGGACACCTGTTTTT
core_49	CATAATTCCATGATCCCGAGATAGGGCGATGGCCACTACGTGAACCGCCATTGGTGCAA
core_50	CACTCATGTTTTAAAAGTTAAGGGCGAATTCCGATCTCC
core_51	AGTGTTATTAGCTCCTGCAAAATCAATCAAGTTTTTGGGGTTCGAGGT
core_52	TTCAGCAGTTGAGATCCAGTTCGATGTAACAATATTT
core_53	CCATTGATCAACTGCCAGTGCCTCACGACCAACTTCTGGTGAGTCA
core_54	GACTTGGATAATGCCAGGCGGGCCATTTACCGCAATTTCTCATTATT
core_55	GGGGTGGACATTTATTAGAAAGGTCTCGAAG
core_56	CCCATTGCTTATATTTGTGGCCGTTTACG
core_57	AACAACTCGTCAATAGGGGGCGTACTTGGCAATAGAATGGGTCAAGGGCTGGCAA
core_58	TTTTTTGTTACGACATTTTGAACACTTGATGTGGG
core_59	TTTTTCTGTGACATAGTGTATGCGTTTTT
core_60	TGGTGAGTATCCGTAAGATGCTTTTTTTT
core_61	ACTCAACCAGCAGCCACTGGTA
core_62	GGGTTAATCAGCACTGTTTTGTTTTCTGGCCCCAGTGCTG
core_63	AGCAAAAGTTGGCCGCAAAAAGGGTAGATAACTACGATA
core_64	GCCAGCAAGCGCCTTATCCGGTAACTATCGTCCGCGCAGA
core_65	CGCCATATCGCCCTCGCCCTCATGGCGGC
core_66	TTTTTGACTCCAACGTCAAAGGGCGAAAAACCGCGTCTGTTGATCAAGACTTTAT
core_67	AAGCGAAAGTTCGGCGCACATTTCCGCAAGTA
core_68	GGTTGAGTGTTGAAGGCGTCAATCATGCC
core_69	CCTTATAAATCAAAAGCAGAACGTTATGGCATTAAACAACCCGG
core_70	TTTTAACCAATAGGCACGTTCTAAGTAAG
core_71	TCCGTTAAATTTTTTTTACCGCTTCTTTTACCGCGTTGC
core_72	CCCGAAAATCCTTTTTTTGAGGGCAGCGTATCCACATAGCGTAAAAGGTCTTGATG
core_73	TTTTTGCCCCACCCACCCCCAGATATGATAAGTCCCG
core_74	TTTTTCGACCGAGTTGCTCTTGCCGAGTCCACTTTCCCAACTGGTATGGCTTATTCTTTTT
core_75	AATACCGCGCCACATAGAATAGACCCCATGTT
core_76	TGCTCATCATTGGAACGAAATCGTCCGGTCT
core_77	TCGGGGCGCGTAATTGATTACTATTAATAAC
core_78	AAAACCTCAAGGATCGTTAAATCAAGGCAAA
core_79	CTGGGCAATCCCCAGGATCTCTCCTTAAACCTGTCTTGTAACGCGGCTGAAACCGCTA
core_80	CATATATGGGCTATGAACTAAAAGGCTTTCACCATTCTACGGATTATCATAAAAATG
core_81	TTTTTGCCAAGTACTTTTTT
core_82	AAGGAACCCCTAGTGATGGAGTTGGCCACTCCCTCT
core_96	CAGTTTACCCTAAACTCCACCCATTGACGTCAATGGAAAGTCCCTATTGGCGTTACTATGGGAACA TAGCTCATTATTGACGTCAATGGGCGGGGTCGTTGGGCGGTCAGCCAGGCGGGCCATTTACCGTA AGTTATGTAACGCGGAACCTC
rec1_1	GATTTATCAGCTTCGCCAGCAATGATAGCGAGTTATCTTACTGTACGGGATGGCTACGTCAAGTCATT CTGAGA
rec1_2	GACTCTAGATCCTTTTAAATAAAAGGATAGGATTTA
rec1_3	CTTACCAGCAAGCATGGTAGCTGATCCG
rec1_4	TAAGACACGTAGCTCTGGTGGTTTCTAACTACCCC
rec1_5	GTGCACGAACCCCCCGGAAGCGTTACCTGTC
rec1_6	GTCCCGGACAGTGGGGGTTGCGTCTTCGT
rec2_1	ACAGTTACCAATGCTTAATCTGAAAGCGGATTTTAAAAAGG
rec2_2	AAATTTTGTAAAGGCGGGGAGCAACATCCGCGGGAAGGAAGGT
rec2_3	TGGAAGCTCCGACTTGAGACAGGACGAGCATCACAAAAATCGACGCTGTACTGCC
rec2_4	TTGGCCAGGATGGGCACCACTCGCCGTCGTGGTAC
rec2_5	AAATTCGGGCATGGCGCTCGTGGCTCAGTTCTATAAAGAT
rec2_6	AGCACTGCTTACCTCTCCAGAGGCAGCACGGAGTTAAGAATA
rec2_7	GAAGGACGGAAATGTTCTGGACGTAGCCCTCTATAT
rec2_8	AGAGCCGTCTCATGCTCGAACACGCGGTTCTCG
pro1_pass_1	TTTTTGATGATTTCCCCGACAACACCTAAAGGACACAGGCATGCCGCA
pro1_pass_2	TTTTTGTCCAGCAGCGGGCAAGGCAAGGGGAGCCGTAA
pro1_pass_3	TTTTTACTCCTCAGGAGAAAAAGTGAGGCAAGTTTTTAAATCAATCTGCCCG
pro1_pass_4	TTTTTACCACGCTGTCTATAGCAAACCTTACCTGTAGCAAAAACAGG

pro1_pass_5	ACCTATCTCAGCGATCCACGTTGCCAGCAACCACAGTGCATTTTT
pro1_pass_6	CAATAGGGAGGCGGCGTTGTCAGTGCCCAACAGCCGAGCCCTTTTTT
pro1_pass_7	CGCCGTGGTACAAGGATAAAGAGACTGACAACGGGCCACATTTTT
pro1_pass_8	AGTCTATTAGCACTAAATCGGAACCCACGGAAATGAGTTTCGAGCAGCCAAGGAAAGGACTTTTT
pro2_pass_1	TTTTTTACGTAGCCATGCTCTAGGAGTCCCCAGGCAGAACAGTGG
pro2_pass_2	TTTTTACCACACCCGCCGCGCTTGAGGCCGCAAGTGTAGTGAATGTATTTAGAA
pro2_pass_3	TTTTTTGTTGTGGCTGCTTGAGGGCGGCTCGTCCATGCCGAGCCAATAGG
pro2_pass_4	TTTTTGTGGTTCGGGTGGTTGTGGGTGTTTCGTGTGCCAGCGGATA
pro2_pass_5	TTGTACAGACTGGGTGCTCAGGTACGAGCTGCGAAGTTCATGCTGGTATTTTT
pro2_pass_6	CCGTTCTTCGGATCTTACGCTGCCGTCTCGATTTTT
pro2_pass_7	TTTATCAGAAATAGGGGGAGCGGGCGCTAGGGAACTTTTT
pro2_pass_8	ATTCGCCCTTAACTCGAAGATCCTTTAAGGCAATGCGCCATGCTACTTATCTTTTT
pro1_x8_1	GCAAACAGGATGATTTCCCGACAACACCTAAAGGACACAGGCATGCCGCA
pro1_x8_2	GCCGAGCCGTCCAGCAGCGGGCAAGGCAGGGGGAGCCGTAA
pro1_x8_3	GGGCGACAACCTCCTCAGGAGAAAAAGTGAGGCAAGTTTTAAATCAATCTGCCGC
pro1_x8_4	GGAACAAGCACCACGCTGTCTATAGCAAACCTTACCTGTAGCAAAAAACAGG
pro1_x8_5	ACCTATCTCAGCGATCCACGTTGCCAGCAACCACAGTGAAGTCCACT
pro1_x8_6	CAATAGGGAGGCGGCGTTGTCAGTGCCCAACAGCCGAGCCCTCCTGTCCA
pro1_x8_7	CGCCGTGGTACAAGGATAAAGAGACTGACAACGGGCCACACGGAAATG
pro1_x8_8	AGTCTATTAGCACTAAATCGGAACCCACGGAAATGAGTTTCGAGCAGCCAAGGAAAGGACAGTGCACA
pro2_x8_1	ATCCCGGCTACGTAGCCATGCTCTAGGAGTCCCCAGGCAGAACAGTGG
pro2_x8_2	GATTTATACACCACACCCGCCGCGCTTGAGGCCGCAAGTGTAGTGAATGTATTTAGAA
pro2_x8_3	AAGAAGTCTGTTGTGGCTGCTTGAGGGCGGCTCGTCCATGCCGAGCCAATAGG
pro2_x8_4	CGTGACCGTGGTTCGGGTGGTTGTGGGTGTTTCGTGTGCCAGCGGATA
pro2_x8_5	TTGTACAGACTGGGTGCTCAGGTACGAGCTGCGAAGTTCATGCTGGTACAACCTGAT
pro2_x8_6	CCGTTCTTCGGATCTTACGCTGCCGTCTCGAGTGTCTGCT
pro2_x8_7	TTTATCAGAAATAGGGGGAGCGGGCGCTAGGGAACCAAGGAGG
pro2_x8_8	ATTCGCCCTTAACTCGAAGATCCTTTAAGGCAATGCGCCATGCTACTTATCGGCGGTCA
pro1_x5_1	AACACGATGATTTCCCGACAACACCTAAAGGACACAGGCATGCCGCA
pro1_x5_2	GAGCCGTCCAGCAGCGGGCAAGGCAGGGGGAGCCGTAA
pro1_x5_3	CGACAACCTCCTCAGGAGAAAAAGTGAGGCAAGTTTTAAATCAATCTGCCGC
pro1_x5_4	ACAAGCACCACGCTGTCTATAGCAAACCTTACCTGTAGCAAAAAACAGG
pro1_x5_5	ACCTATCTCAGCGATCCACGTTGCCAGCAACCACAGTGAAGTCC
pro1_x5_6	CAATAGGGAGGCGGCGTTGTCAGTGCCCAACAGCCGAGCCCTCCTGT
pro1_x5_7	CGCCGTGGTACAAGGATAAAGAGACTGACAACGGGCCACACGGAA
pro1_x5_8	AGTCTATTAGCACTAAATCGGAACCCACGGAAATGAGTTTCGAGCAGCCAAGGAAAGGACAGTGC
pro2_x5_1	CCGGCTACGTAGCCATGCTCTAGGAGTCCCCAGGCAGAACAGTGG
pro2_x5_2	TTATACACCACACCCGCCGCGCTTGAGGCCGCAAGTGTAGTGAATGTATTTAGAA
pro2_x5_3	AAGTCTGTTGTGGCTGCTTGAGGGCGGCTCGTCCATGCCGAGCCAATAGG
pro2_x5_4	GCACCGTGGTTCGGGTGGTTGTGGGTGTTTCGTGTGCCAGCGGATA
pro2_x5_5	TTGTACAGACTGGGTGCTCAGGTACGAGCTGCGAAGTTCATGCTGGTACAACCT
pro2_x5_6	CCGTTCTTCGGATCTTACGCTGCCGTCTCGAGTGTCT
pro2_x5_7	TTTATCAGAAATAGGGGGAGCGGGCGCTAGGGAACCAAGG
pro2_x5_8	ATTCGCCCTTAACTCGAAGATCCTTTAAGGCAATGCGCCATGCTACTTATCGGCGG

2.2.5.6 sc_EGFP5 16HB_4

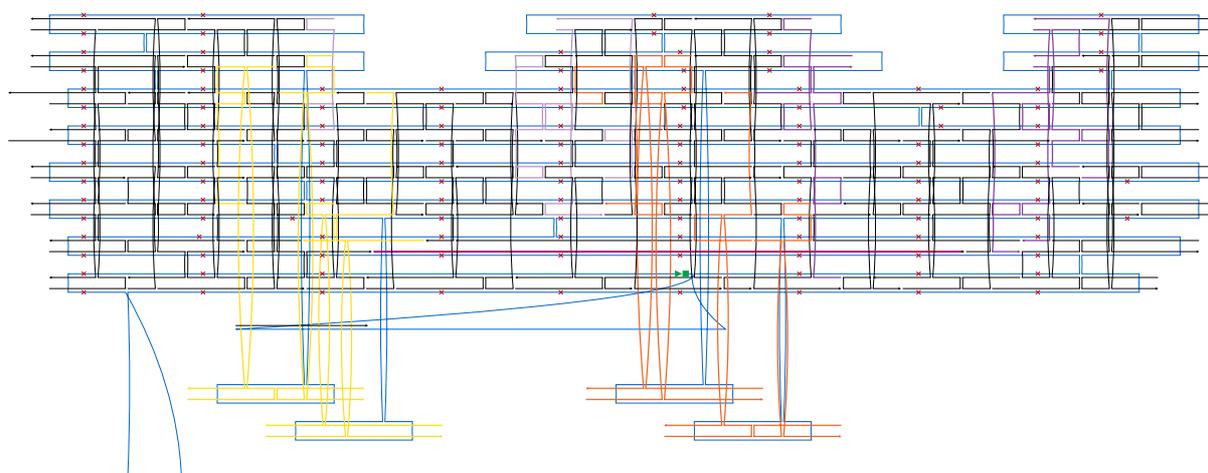


Fig. S38 | Scaffold routing and staple design for sc_EGFP5 16HB_4. Scaffold routing is shown in blue, staples are given in black, breakpoint for addition of sequence is marked by a green square (5') and arrow (3'). Long 154-mer 'primer' sequence at the promoter (5') region is given in pink. Protrusions 1 and 2 are shown in yellow and orange respectively, recess' 1 and 2 are given in light and dark purple, respectively. Design was prepared using caDNAo v0.1.

Table S25 | Individual staple sequences for sc_EGFP5 16HB_4.

Name	Sequence 5' – 3'
core_1	TTTTTATCACCGTAATCAAGTGCCCACTACGTGAACCTTTTT
core_2	ATACTTACGTCCTCGAGGTCGGCGCGATGGGGGTGTTCTGGTGACACAC
core_3	GTCAGCTTTTGTAAATCAAGGAGCAACATAGTCACGCCAGCGTCAG
core_4	TTTTTTTTTCTGTGACTGGTGGCCATCCGTAAGATGCTTTTT
core_5	CTCACGCTGTAGGTATCTCAGTTTTTGCTGGCCGTTTGGTGGGAAGCT
core_6	TTTTTAAGGCAAAGTGGGCGTTTCTTTTT
core_7	GTCGTTGCTCCAAGCTGGGCTGAGCAAAGCCAACGATTCGCCTC
core_8	TTTTTCGGGGCCGTCGCAGCTGCATTTTTTTTTTTTTTTTT
core_9	GCCACAACATCCACATCTGCTTGGCAGATGACTGTCTCGTACTTTCATCCCTCG
core_10	GGCAACAGGAAGGTTTTTCAATATTATT
core_11	CTACTCACTTCCAGGCCCTCGACGGGCATGCGGTTAGC
core_12	CACCACACAATCAAAGAATAGACTTAACCAAATGGTTATTATGCGGC
core_13	GATGATTAACAGCCGCGATGAGTTCGCCAATCCCCGTCTATTA
core_14	TAGCGTAACAGAGGTTGATTGGATCGAGAGTGCCAGCGTT
core_15	CCGCCGCGCTTAATGTTAATTAACACTTGGTCGCTCACCGGCTCCAG
core_16	TTTTTGTCTATCAGGGCGATGCAAAGGGCGAAAAACCTTTTT
core_17	TTTTTACTATTAAGAACGTGGACTTCGTTT
core_18	TTTTTCGCTGCCCTGCCAGTGCTTTTT
core_19	TGTTGTGGCCAAGGTAGTGGTTGTCGGGCAGCAGCATTTTT
core_20	ACGTTGTGGCTGTTGTTGGGCCAGTGGTGTCACTCGTGCA
core_21	CTCCAGCTAGGGAGGGGGAAAGCGAAAGTCCCCCATTGA
core_22	CGCGGGAAGGCAAACACGTGCTGTGCATGCGCAGAAGAGTGGAA
core_23	ACAAATTGCCGTAGGAAGTTCCTTAACAACCAGC
core_24	TAAGAATACGGATCTTGTGGCATCTAGGTGCCCTCACCAT
core_25	CAAACACAGTTCACTACTGTCTTGGACGTCAACCGACAGGACTATAAA
core_26	GGCAATGCGGGTCTTGTAGTTGCCGTGATCTGACCCACTGACAG
core_27	TTTAGAGCTTGACGGTTTTTGGGATTGTAAGCGTTAATTTTTTTTT
core_28	ACTTCAGGCACCACCCATCCCGCGGCGGTACGAACTC
core_29	GGGTTGAGGTAGCGGTGGGCGCTAGGGCGCTCCAACGTGGAAGCC
core_30	GGACTGGGTGCTCGTCAATCTTTCCAGCAGGACCGTCCAGCTCGACC
core_31	AATCGGCAAACCTTGAAGAAGATCAGAAGTATCAATACGGTAAGACA
core_32	TTTTTTGTTAAAATTCAACAAGAGTCTTTTT
core_33	TTTTTTTTTTTTTTTTTGAACCTCCTGTGGAGAGATTTTT
core_34	TCTGGGTCATCTTTTGTGGCCATCACAAATGTAAGTGGG
core_35	CCGCAAATAACCCACGCTCGTGTTTTTCCATAGGCTCCGCCCC
core_36	AAAGGGAAGGTTGGTCACGAGGGAGTTGTAAGAGTAACGTCAATGTACTGC

core_37	ATGTTGAGATCTTACCGGTTCCGCCAGGAACCGTAAAA
core_38	TCATGAGCATAGCATCCTTCGTAAGTAGCATGGCGGGTTAATCATGCGCCTT
core_39	GAAAAATCCCGGCGAGTTGGCTCCGATCTTCCTAG
core_40	GCCCTTGATCAGAGCGAAACTAGGGGGCGTACTTG
core_41	GGCGAAAACCTCTCAAGATACTCATTAGCGGCTCGCGCAGAGAAAGGAC
core_42	GAAAACGATCCCCCTAATTAAGGGTGAGCAAAGGCCTGTGCACG
core_43	AAAGTGCTCATCATTGGAAGCATTGAAGAAGTACAGATGGAAGGCACG
core_44	TTTTTTTTGCTCAGGGCGACACGCTTTTT
core_45	AATAGTGGGCAGCAACTATTAATACTAATGACCCCGTTCTGCTGA
core_46	TTCCGACCGGCGCGCCGCTGGACAGTGGCCGTCCTCGCCG
core_47	CCACTGGTAACAGATTAGCAGAAATTGATTCTGCATAAGGGCTTAC
core_48	CTGCCAAGTCTGCTTATATAGACCCCAACCAGGCCGTTCT
core_49	CAGTTTGGGCGTTAACTGTCATAGTACTCATGTAGGCGTTTTT
core_50	ATCGTTGTGGTGCGCTTATTTGAATGTATTTA
core_51	GCCCTCGCTTACGTCGCCATGTGATCGCGCTTCTCGTTGGGGTCTTTTT
core_52	GGAAGCGTGGCGCTTTGACGAGCATTGCTACTAGTTTGTGTGATATAGAGCTGACA
core_53	CGACTTATCGCCACTAGGGCGAACGAGTGTATACCGCGCCTTATA
core_54	AGGGTGTGGTCAAGGCTGGCAACTAGAAGGAGGCCGTTATTGTC
core_55	GTAGTTCCGCAGTTAAAGGCATCGGGCACGGGACAGGAAGGCAAAATG
core_56	TTGTTGCCATGGCTTCGTAGGTCATAAGGGCGACACGGAA
core_57	CTTCATGTGTTACATGTTCTTCGGAACCCCGTTCAGCCCGACCCG
core_58	TAACTACGGGCCTAACTACGGCTACACTTTTT
core_59	TTTTTCTCCCGTCGTGTAGAATCCATAGTTGCCTGATTTTT
core_60	TTTTTTCACGACCAACTATAATGCTTTTT
core_61	TCCCACCACAAGCCCTACACTTGAAATCGAC
core_62	GGGGTGGAGACTTGGAGTGGCAATTGTGCCCC
core_63	ACGGGGTCCGCGGCAAGTCGATGCCAGCCAAGATCCAGGT
core_64	GGGATTTTACCAGCCAGCCGGAAGCAAAAAG
core_65	TCAGCGATCTGTCTATTGGCAAGTTGTTGTTCCGGCAAC
core_66	TAGGAAAGTCCCATAAGGTCATGCCGTCATTTAACCTTGAAGACCAA
core_67	CTGCCGCTTACCAGGATACCTGTCCAGAGGTGAACGCAAG
core_68	TAATACGTAGATGTAGCATATGAAGTTTCTAAGTCTTCT
core_69	GCGGACTTTATCAGGGAGAGCCGGCAGCAGGC
core_70	TTACGCGCAGAAAAAAGGATCTCAAGAAGATCCTGTCAAACCGCTATCCACGCCCATTTGAT
core_71	GAACTTAATCCGGTAACTATCGTCTTGAGT
core_72	CTCTTGATCCGGCAAACAACCCAGCTGGTAAAGTATATATGAGTA
core_73	TACCTTCCGAAAAAGAGTTGGTAGTTACCAATGCTTAATC
core_74	TTTTTTAGAAGGACAGTATTTGGTATCTGCGCGAAGTGGTATACGGGA
core_75	TTTTTCAGGCGGGCCATTATACTGGGCTCTGCAGGATCTGACGCTGGTAGT
core_76	TCTCATAGCCCAACTGATCTTCAGGAGCAAAACAGCTTGC
core_77	CATATATCTGCAATGTATCACTCTAGGCCGAAGGGGTTCCGCGCACATTTCCCC
core_78	GCTCAAGTCGCCTTTCCGGTGAACAGTCCTCAGGATGGG
core_79	AGGCCGCGCGGTGTAGGAGATCCAGTTCGATG
core_80	TTAACTACATGTTGTGGGCGGAGCGGTTACCAGGAAGATCCCAATAGAATGACAC
core_81	ATAACTAGCTAGCTTAGGCAGCAGGACCGAGTTGCTCTTGAAACAAAT
core_82	AGCATGGCCCAACCCGGGATAATACCGCGCCACGGATACACCTGGACG
core_83	TTTTGTGCTACAGAGTCTTTCGCGAGGTAACCAAGTC
core_84	TTTTTCCCCTGGAAGCTCCGATACCAGATGTGAGT
core_85	AAGGAACCCCTAGTGATGGAGTTGGCCACTCCCTCT
core_96	CAGTTTACCGTAAATACTCCACCCATTGACGTCAATGGAAAGTCCCTATTGGCGTTACTATGGGAACA TACGTCATTATTGACGTCAATGGGCGGGGTCGTTGGGCGGTACGCCAGGCGGGCCATTTACCGTA AGTTATGTAACGCGGAACTC
rec1_1	CAAGGCGAGGTCGGGGACTCTTCCCGCTGGATTGAGGGCCGAAG
rec1_2	CGTCCGAAGCAGCCCGTCCCATCCAGTGATTGATCTTTTCT
rec1_3	CAGTGCCCTCCCCGACAGGATGTTTGCACGCCATTACGCTCCGCTGTT
rec1_4	GTCCATGCCAAGCTTCGGTGGTTCGGCCA
rec2_1	ATTCTGAGGAAAAGTGAATCAGCGCCCCCGAGTGGCGAGAAAAGG
rec2_2	AGGACAGTGGGAGTGGCAGCACAATGCACTTATCTACGTAGC
rec2_3	CGCGGAAGCACAGTCTGAGCCTTACTTAC
rec2_4	AGGGATCATTTTTCGAGATACATCTGGACCTATCAGCCAGT
rec2_5	TTCTCTTATTTTTGTTCCACCTAAGTTCGAGGTGCCGTAAGACAC
rec2_6	AGGAGCACGCTGCGCGTAACAGTGAGGCCCCAGTGGGGCTATG
rec2_7	TACGTAGAGTCTCCGATTTATCACTCGGCGGCCATGCTGATGCAATTC
pro1_pass_1	TTGGTCTCACCTTGATTGGGGGTTGTTGCCTGCAACCAGTTTTT
pro1_pass_2	CAAAACCGCATCACCATGGTATCCCGTTGATTTTGGTGCTTTTT
pro1_pass_3	TTTTTGACATTTTGGAAAGATACTACCGCCCATTACTCGGAAAGG
pro1_pass_4	TTTTTACGGGAAGTCCCTATAAGAAAATGGCAATGCC
pro1_pass_5	AAGCAGCGCAATAGCAGGTGGTGAAGCCATTTTTT
pro1_pass_6	TTTTTCAAACAAGCGTCAATGGGGCGGACGCGCGATGAC

pro1_pass_7	TTTTTGATTTATACAAGGAGGAAGAGACAGACAACGG
pro1_pass_8	GCAACGTGTACAGTTGTTACTTTTT
pro2_pass_1	GCAATAATCAATCTAGCGGTGAAAATGAATTTTT
pro2_pass_2	GGGGAGGCTCGAGTCCTTATCGCTGTCTGCCCCACCCCATTTTT
pro2_pass_3	TCCTTGAAGGCAGGCGGAGCCCCTGTCCAGCAGCCGTCTTCCCTTTTT
pro2_pass_4	TTTTTCTTACCTAGATCCTTGATTATCACACGTTAACATGCTCT
pro2_pass_5	TTTTTAATCTCCCCCTTGGGAATTCTGACGCTCTGGTCTGCAACTTTA
pro2_pass_6	TTTTTGTTTTAAAGGTCATGATTAATTAAGTTTTTTTT
pro2_pass_7	TTTTTCCCCCAGCAGCATGCCTGCTATTCTTAACAGGCGAGCCTTCAGC
pro2_pass_8	GTTTGCAAGCAGCAGACGAAAACAAAAGGATTTTTT
pro1_x8_1	TTGGTCTCACCTTGATTGGGGGTTTCGTTGCCTGCAACCAGCGGCTCCA
pro1_x8_2	CAAAACCGCATCACCATGGTATCCCGTTGATTTTGGTGCTCCCTTCG
pro1_x8_3	GCAAACAAGACATTTTGGAAAGATACTACCGCCATTTACTCGGAAAGG
pro1_x8_4	TCATCCATACGGGAAGTCCTCATAAGAAAATGGCAATGCC
pro1_x8_5	AAGCAGCGCAATAGCAGGTGGTAAAAGCCATAGTTGCCT
pro1_x8_6	CGCCTTTCCAAAACAAGCGTCAATGGGGCGGACGCGCGATGAC
pro1_x8_7	ACGCTCAGGATTTATACAAGGAGGAAGAGACAGACAACGG
pro1_x8_8	GCAACGTGTACAGTTGTTACACCACCGC
pro2_x8_1	GCAATAATCAATCTAGCGGTGAAAATGAAAGTTTCTA
pro2_x8_2	GGGGAGGCTCGAGTCCTTATCGCTGTCTGCCCCACCCCATCTTTCAC
pro2_x8_3	TCCTTGAAGGCAGGCGGAGCCCCTGTCCAGCAGCCGTCTTCCCTTGGTCTG
pro2_x8_4	ACCAGGCGCTTACCTAGATCCTTGATTATCACACGTTAACATGCTCT
pro2_x8_5	GAGTAAACAATCCTCCCCCTTGGGAATTCTGACGCTCTGGTCTGCAACTTTA
pro2_x8_6	ACAAGCCCGTTTTAAAGGTCATGATTAATTAAGTTTTTTTT
pro2_x8_7	CCAGTCAACCCCCCAGCAGCATGCCTGCTATTCTTAACAGGCGAGCCTTCAGC
pro2_x8_8	GTTTGCAAGCAGCAGACGAAAACAAAAGGATTTTCCCCC
pro1_x5_1	TTGGTCTCACCTTGATTGGGGGTTTCGTTGCCTGCAACCAGCGGCT
pro1_x5_2	CAAAACCGCATCACCATGGTATCCCGTTGATTTTGGTGCTCCCT
pro1_x5_3	AACAAGACATTTTGGAAAGATACTACCGCCATTTACTCGGAAAGG
pro1_x5_4	TCCATACGGGAAGTCCTCATAAGAAAATGGCAATGCC
pro1_x5_5	AAGCAGCGCAATAGCAGGTGGTAAAAGCCATAGTTG
pro1_x5_6	CTTTCCAAAACAAGCGTCAATGGGGCGGACGCGCGATGAC
pro1_x5_7	CTCACGATTTATACAAGGAGGAAGAGACAGACAACGG
pro1_x5_8	GCAACGTGTACAGTTGTTACACCAC
pro2_x5_1	GCAATAATCAATCTAGCGGTGAAAATGAAAGTTT
pro2_x5_2	GGGGAGGCTCGAGTCCTTATCGCTGTCTGCCCCACCCCATCTTT
pro2_x5_3	TCCTTGAAGGCAGGCGGAGCCCCTGTCCAGCAGCCGTCTTCCCTTGGT
pro2_x5_4	AGGCGCTTACCTAGATCCTTGATTATCACACGTTAACATGCTCT
pro2_x5_5	TAAACAATCCTCCCCCTTGGGAATTCTGACGCTCTGGTCTGCAACTTTA
pro2_x5_6	AGCCCGTTTTAAAGGTCATGATTAATTAAGTTTTTTTT
pro2_x5_7	GTCAACCCCCCAGCAGCATGCCTGCTATTCTTAACAGGCGAGCCTTCAGC
pro2_x5_8	GTTTGCAAGCAGCAGACGAAAACAAAAGGATTTTTCC

2.2.6 sc_EGFP6

Scaffold sequence for sc_EGFP6:

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ctccaggggaaacgctgtatctttagtctgctgggttccgacacctgactgagcgtgattttgtgatgctcgtcagggggggcgga
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Corresponding DNA origami designs and staple lists:

2.2.6.1 sc_EGFP6 20HB

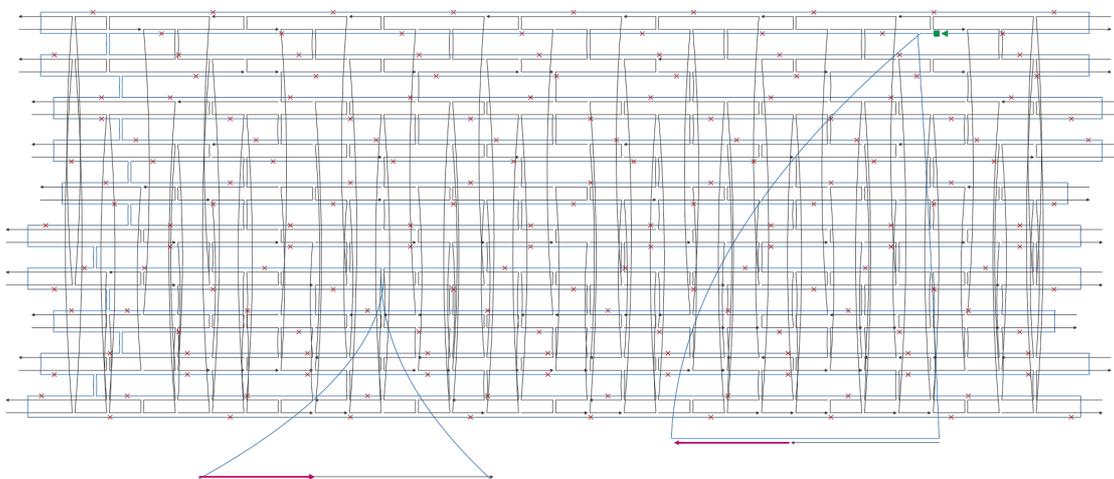


Fig. S39 | Scaffold routing and staple design for sc_EGFP6 20HB. Scaffold routing is shown in blue, staples are given in black, breakpoint for addition of sequence is marked by a green square (5') and arrow (3'). Staple sequences which position the ITR hairpins to flank the 5' promoter region and the 3' polyA region are given in pink (ITR hairpin is position on the left side of each of the loops). Design was prepared using caDNAo v0.1.

Table S26 | Individual staple sequences for sc_EGFP6 20HB.

Name	Sequence 5' - 3'
core_1	AATGACCCGACGTC AATGGAAAGCCCGTTGA
core_2	TAGCTAGCTGGGCAGTTTACCGTATTGACGT
core_3	CGGGTTAATACCGTCATTGACGTCTGATGTA
core_4	CATTATTGGGTTTTTATTTTTGTCTCATCATTAGCCATGCGCTGCTATTGTCTTCC
core_5	GGCGTTACACTCCATATATGGGCTCGACCCTG
core_6	CACCCATTTCGTAATTGATTACTATCCTGGAAG
core_7	AAATACTCTGATCTTTATTTCCCCAGTTCGAACACACAATGACACCTACTCA
core_8	CTGCCAAGTTAAGGGCGAATTCCGACAGGAC
core_9	GGCGTACTGGCTACGTAGATAAGATCGACGC
core_10	GGGCCATTTCACTAACACTAAGGCTCCG
core_11	CCCCGTGCCAGTTTCTATTGGTCTCGCCGCTCCCCGTTACAGGCAATGCCATCTGCGTCAG
core_12	CACTAAACCAGCTCTGGGTGGTCAGATTTATTTTATCCG
core_13	GACGGTTGTTGTTACGACATTTTGGAAAGTTCCTATTGCAGAAAGCGTTAA
core_14	CACGACCAACTTCTGAGCTTGCGATACCGTGCCGGGAGTGTATGCGGGAAAGC
core_15	TGTAACCTTGATACTGTCAGCTTAGGGCTTTAATAGT
core_16	ACCTGTCTCAATGGGGTGGAGACTTGGAAATTTGATGTAGAACGAAAAGAAAAAT
core_17	AAACGCAAGAGTCTTTACGTCGCCTGTCTATGGTATGGATGGCAG
core_18	GATGTCAGTAAGACCAACCACCCCGCTTAATCGATCAAGTAAGTTG
core_19	TGAAGAAGGGGCCAGGGCACGGGCCAGGATCTGATTACGC
core_20	TCCTGGACCAGATGAACTTCAGGTACCTGCCGATCCTT
core_21	GTTGCCGTTGGCATCGCCCTCGCCTCCTTAACTCAGTG
core_22	CCTCGAACGAACCTTGTGGCCGTTCTCTGTCTTTTGGTC
core_23	TCAGCTCTCGACCAGGATGGGCATAGGTGCACCTAGA
core_24	GGTCTTTGGGTCACGAACTCCAGGGCAGGCG
core_25	CGGCAGGTCAATCTTTTACAAGTGGGGGTCGTAAGT
core_26	GTTCTTCTACAAGGCATTAAGCCTCCTCATGCCGAGT
core_27	CCGGCCGGCCTCAGGGCGGACTGGGTGCTCAGGGCGGACTTTAATTGTCGTGACCC
core_28	GCTCGTCCATGCCGAAGGGAGGGGCGACCG
core_29	CTTGATACATGTCGGGCAGCAGCAGGGGCCGTTGGTGCCTTCGCCAGACCATCTG
core_30	GTTGATTGGATCCAAGCTGACTCAACCAACAGAATAG
core_31	ATCCAGAGTGGGGGTGTTCTGCTGGTAGTGGTTCTTTAGCCATTGCCGTGTAG
core_32	GAATACCACTGCACGCTGCCGTCCTCGATGTGTGTCGCCGTCGTTTTTCGTTCCACCAGCG
core_33	CGTAAAAGGAGCAACACCACGCCACACTGCAT
core_34	CCACATAGATCTTGAAGTTCACCTTGATGCCGATGCCCTTTCCCAACAGTGAGGCAGGCAAAA
core_35	CGTTGCCCTCCGCTGGATTGAGGGCCGAAGGGGAAAGGAAATCTCTTTACTTT

core_36	AAAGAGACCAGAGCCGGCAGCAGGCCGCGGGGTCAAGGAGTTATCACAACAGGA
core_37	AATTGTCAGTTCGCCCGTGGCAATGAGTGATCCAGTCTA
core_38	TGATTTCCCCGACAACACCACGGCACTCTAGTTGCTGTTCTTCACGCTCACAAGCAGCA
core_39	AGGACGAGAAAGTCCCGGAAAGGGCTTATTATAAGTAG
core_40	CAGGTGGGCAATGCCCAACCAATTTTGTAACTGTTGTT
core_41	AGAAGGACGTCCCGCGCAGAATCGACAATGCGCTTTTCTCACTCGTGATAACTACGTCTGACG
core_42	GACGTAGCCAAACACAGTGCACATAGTTAAGTACGCT
core_43	AAGGAAGGGACAACGGGCCACAAAGCGTATAGCTCCGG
core_44	AGAGGCCGAGCAACCAGGATTTAAAGCAATA
core_45	CCCCCTTGACCCGCCATGCTACTTATAGACCG
core_46	GATGCAATGTAGCGGTCACGCTGCTGGACTCTAGGGGTT
core_47	CAGTGGGAAAAGGAGCGGGCGCTCTATCAG
core_48	AGGCACGGAACGTGGCGAGAAAATCACCCCTTATCAGG
core_49	TGGCAACCCCGATTTAGAGCTTTGCCGTAA
core_50	CCGCCGCGCTTAATGTGTTTGAAGCCACCTA
core_51	TTATAAATCAAAGAATCTACGTGGAAAACCTTGCCCGCCTCCATC
core_52	GTTGAGTGTGTTCCATAAATTAAGAGGATCTTTGAGAATAAGCTAGAG
core_53	CCACTATTAAGAACGGCGTAACCTGTAACCGTGACTGTTGCCGA
core_54	AAGGGCGAAAACCGTAGGGCGCTAGCATCTTACTGTCTCGTGGT
core_55	CCCCTACGTGAACCGGAAGGGAGTGAGCAATCATGGTTCTTCATTC
core_56	TTTTTGGGGTTCGAGGGACGGGGAAAAAGGGGTCAGAAG
core_57	TTAACCAACAAACAGCCGACGCGACTGCACGCCGTAGGTCAGCTTATATA
core_58	CTCATTTGCCAGGCTGGGCGGGGTCGTTGGCATAACGCCCTACCGCCATTTGCGTCAAT
core_59	GCGTTAATTGTTTCCCGGCTCCACGAGGGTTCGTGCTGCTTCATG
core_60	TAAATTCACGCGGATATGGGAACATACGTGGGGCGGA
core_61	ATATTTGATAGAGCATTGGCATATGATACAC
core_62	CGGATACGGCGATGGTCAAGTCAGAGGTGGCGAAACCCGATCTTCCATGTATTT
core_63	CTCATGAGTTTCTGGAGAAAGCGGTGGCACCTTCCAGG
core_64	GAAGCATTAAATCAAGTCCCCCTGACGAGCATCACAATAAGCATGGGTTATTGTATCAAAAA
core_65	ATATTATTTGCCGCAAAGCCGGCGGGGAGGGGCAACAA
core_66	CTTTTTCAAATTAATTAACCAATGGTGAACCGTCTCCTTGAAGTC
core_67	ACTCTTCTGAGCAAAATGTACTGGGCATAATTAATAGCG
core_68	GGGGCGAATATTTTGTAGATAGGCCGCTTACCGGATACCTGTCCGCGTTATGTA
core_69	AACTCTCAAATGATTACTGTCTGCCCCACC
core_70	ACCGCTGGCCCCAGTCTCAAGAACAGTGCCTTTTGGTGCCAAAACAAACTCCC
core_71	TTGAGTCCGAAAAGTCAAGAGTCTCCCTCGTGCGCTCTCCTGTTGATGAACCTAATTGTAA
core_72	CACCCAACAAACAAACACCGTCAATATAAAGATACCAGGCGTTTCCCTAATAACCCGCGCAC
core_73	TGATCTTCGGCAAGTTTCTCATTTTATTA
core_74	ACGGGATCCAGCATGCTCTAGGAAGATCCTAAATCGGG
core_75	GCGTCAATGCGATGAGTGCCCAACAGCCGAGCCCCTGTGAGTCC
core_76	GTGAGTACAGGTGGTGAACACAGGCGAGCAGCAAGGAACCCCCCGTCATTC
core_77	GCGAGTTAGCATGATGCTTGTCCGCCATGATATAGACG
core_78	CATGATCCATGTTGACAGCTCCTCGCCCTTGCAGAAAAG
core_79	CAGCAATAAAAAGTGTAAATCAGAAAATCCCCCTTCGGGAAGCGTGCGCTTTCTGGCGGTCA
core_80	ATCCATAGTAAGGGACGACAAGCAGTCAAACCGCTATCCACGCCATAATAGGGATGAGATT
core_81	TCAGCGATCGTCCAGCGATGCGGTTTACCAGGTGTGGCGG
core_82	ACCTATCGGATCTTCTATCAGCTGCCAAAACCGCATCACCATGGGCCAGGCTCCTTTTA
core_83	GTAGCGGTACGTCAAGGGCCATTTACCGTAACTTCTC
core_84	AAAAGGATGTGCAATCGGTGGTGGTAGCCTTCGGGCATGTAGTGGT
core_85	TCTACGGGGATACGGGGCCGTAGGCGTCTTGAAGAAGATCGCCGA
core_86	ACTCACGTTTGCCTGAGACACGCTTTACCTCGGCGCGGG
polyT_1	TTTTTACCGTAAAAAGGATCGGAACCCTTTTT
polyT_2	TTTTTAGGTATCTCAGTTCGGCATAGCTTGGCCTAAATAGGCCGTATCGGGA
polyT_3	TTTTTAGGAAAGTCCCAAATCTAAAGTTTTTT
polyT_4	TTTTTCAAAGCTGGGCTGTGTTGTAGTTCATCCGGCACTACGGCTACACTTTTT
polyT_5	TTTTTCTGGACACCTGTGGATCACCATGGCGGCCGTTTTT
polyT_6	TTTTTGCCCGACCGCTGAGGTTTCGAAAAAGAGTTTTTTT
polyT_7	GCCCCAGGCCCATGTAACCTTGGTTCGACACGGAAATTTTTT
polyT_8	TTTTTTGTACTCCAGCTTGTGTTGTTGTTGTTGTTGTTT
polyT_9	TTTTTCGCTTGTAGTCCAACCCGATCGCGCTTCTCGTTGGTGGTGGGCTGCAAC
polyT_10	TTTTTATGAAAGCCATACGGGTACAAGGAGGTCTCT
polyT_11	TTTTTATCGCCACTGGCAGCGCAGCGGGCAACAGGACCA
polyT_12	TTTTTGTGAGGACAGATCTCGAAGACGCGGACAGATGGCCGATCGTTAATAAGGG
polyT_13	TTTTTGAGCGAGGTATGTAGGCGCCCTTAACTCCAAGCCACTGGTAAACAGGATTAGCATTTTT
polyT_14	TTTTTTAAAGGGAGCCTAGAAGGCACATTTTT
polyT_15	TTTTTCTGAAGTGGCACGCTGTTTTT
polyT_16	TTTTTGTGAATCATAAGCATAAGCCTGTTGCTGGCGTTTTTTCATATTAAGGG
polyT_17	TTTTTTAGAAGGACAGTAATTCGGTGCTACAGAGTTTTTTTT
polyT_18	TTTATAGCAGAACTTTAACCCAGCCACCGCTGGACCTCCACCGCGAACCCCGGTTCATTTTT

polyT_19	AATACCGCGCCACGGGAGCGCAGTGTGGTAAGACACGACTTTTTTT
polyT_20	TTTTTGGTTAGCTCCTTCGGAGAAATTTTT
polyT_21	TTTTTAAGCCAGTTACCGCCTATCTGCGCTCTGCTGTTTTT
polyT_22	AAGTGGTCGTAGCGGCTGAACCTATCCGGTAACTATTTTTT
polyT_23	CTGACAGAATGAAGTGCAAAGTGATGACTAATACGTAGATGTAAGTCCAAAGTTTTT
polyT_24	TTTTTATATATGAGTATGTGCAAAAAAGCTTTTT
polyT_25	TTTTTGGTAGCTCTTGGTTTCGCTTTTTT
polyT_26	TTTAAATCTAAGGTCAGGCCAGCAAAAGGCCAGGATTTTT
Loop_1	AGGAACCCCTAGTGATGGAGTTGGCCACTCCCTCT
Loop_2	AGAGGGAGTGGCCAACTCCATCACTAGGGGTTCTTGTAGTT
ITR_1	CTGCGCGCTCGCTCGCTCACTGAGGCCGCCCGGGCAAAGCCCGGGCGTCCGGGCGACCTTTGGTCGCC C
ITR_2	GGGCGACCAAAGGTCGCCCGACGCCCGGGCTTTGCCGGGCGGCCCTCAGTGAGCGAGCGAGCGCGC AG

2.2.6.2 sc_EGFP6 20HB-exLP

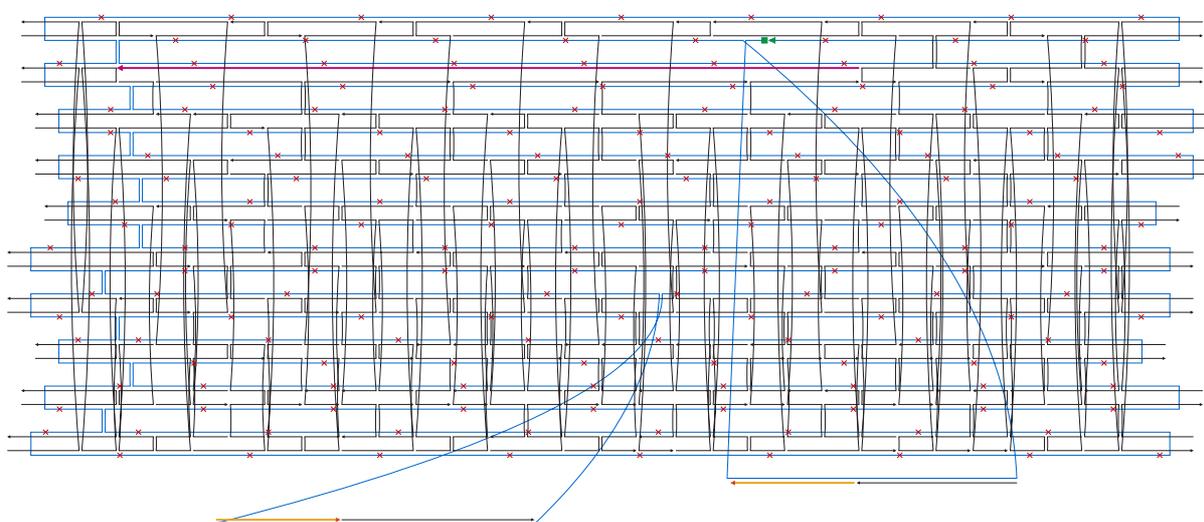


Fig. S40 | Scaffold routing and staple design for sc_EGFP6 20HB-exLP. Scaffold routing is shown in blue, staples are given in black, breakpoint for addition of sequence is marked by a green square (5') and arrow (3'). Staple sequences which position the ITR hairpins to flank the 5' promoter region and the 3' polyA region are given in orange (ITR hairpin is position on the left side of each of the loops). Long 154-mer 'primer' sequence at the promoter (5') region is given in pink. Design was prepared using caDNAno v0.1.

Table S27 | Individual staple sequences for sc_EGFP6 20HB-exLP.

Name	Sequence 5' - 3'
core_1	CCGATCTTCCTAGAGCATGGCTACACCTGTCC
core_2	AGTAGCATGGCGGGTTAATCATTTCCTGT
core_3	AACTACACTAATTAAGGGTGAGCAAAAGGCCGGCGTTTC
core_4	GCCAGGAATACTGCCAAGTGGGCAGATGTAC
core_5	ACTTGATGCCGTAAAAGGCCGGCGGAAAC
core_6	GTCCCCTTGATTTTGGTGCCAAAACAACCTCATTTGCGTCAATGGGGAACTTCA
core_7	TAGACCTCCCACCGTCGCCCTCCAGCCGTTGCCATT
core_8	CTGACGGTTCACCTAATGTGGCCGACCCGGCTCTCGTCGT
core_9	TGCAGGATATTGATGTACTGCCAAAACCGCATCACCAT
core_10	CCCAGTGCCTCACGACAGGATGGGTGCTGCAGGTTCCCATCTGAGAAAACCTCCTC
core_11	AAACCTGTCTTGAATCGCCCTTACGATACGCCCCATCTGTGAC
core_12	CTCGACAAGCCAGTTCACCTGTGAGTTGCCTTTCGGTCTTACTG
core_13	GCCGTAGGTTGCCGTAGGTGGCATAACGCCT
core_14	TGTGGTCGCGGACAGCTGAACCTACCAGCTC
core_15	ATGGCGGAGCCGTCCAGCTCGACCCAACTTCAAGGATC
core_16	GATGGTGCCCGGTGAACAGCTCCCCTTGATA
core_17	GGTCTTGTGGCGGCCGCTGGATCTATTGTATGAGTA
core_18	CACGGGGCCTCAGGGCGGACTGGCCCGGAAA

core_19	CTTGATGTACTTGTACAGCTCGCGGGCCACTAGTGAT
core_20	TCCAGCTTAAGAATACCAGTCAATGCCATACGTCATGCCA
core_21	GGTCTTTGCGTCGCGCATGGGGGTGTTCTGCCACTGCACCAACGTTGGAAGGGCC
core_22	CATGTATCGCGCTTCCCACTAGCAGAA
core_23	AGCAGGACGTGCGGCGAGCTGCACGCTGCCGTCTGCTTCAGTGTACGCCAGATTT
core_24	TCCCGGCGGCGGTACAGTGCACGGCGTCATAGGAAAG
core_25	GAGAGTGAGTTGTGGCGGATCTTGAAGTTCACTTCGGGCTCAGCTCCATGATAC
core_26	AAGCTTATCCGTTCTTCTGCTTGTGCGCCATTGAAGAATACATGATGGAGGGCGGAATAAG
core_27	TAATCCAGAGGTTGATCCAGGATTTGGTGAGT
core_28	AAATTTTGCCTTGTGGCTGTTGTAGTTGTACTCGGCGCGGTTAGCTCGACTCCCCTCATACTC
core_29	ACAGGTGGTGGCAATGCTCGTTGGGTTGCG
core_30	GTTGCGTCAGCAAACACGAACTCCCATCGTG
core_31	CACGTTGCCTGACAATCCATGCCGGCTTCAT
core_32	TATACAAGTCCAGGTGGCAACACAGGCGAGCGGGCAAACACTCAACCCAAAAAAG
core_33	GAGGAGAAAATGAACTTTACAAAAAGCG
core_34	GGAAGCAAGGGACGTAGCAGAAGGACGTCCCGCACAGTCTCCGTAAGTTGAATAC
core_35	AAAGGACGATGATTTCCCGGACAACCTCCAGCGCGACGGGTGAGCCGCGTGACTTATCAA
core_36	AGCCAAGGATAAAGAGACAGCAATGGATCCAGGCGAGT
core_37	GGGCCGAATAGCATGATACAAAAGAGGAGCAA
core_38	CAGACAATGAATTGCGCCTTAACTTTAAAGAA
core_39	GGGTCAAGAAGAATGATTAACCCGACGTGAAAAATTTTT
core_40	AACAGATGCCACCACCCCGCCGGGTGCGACCCGAAAA
core_41	GAGGCAGCTGGCAAGTGTAGCGAACCCCTAAATAACA
core_42	CAGAGCCGAAGAAAGCGAAAGGGACGGGGA
core_43	CCAGCATGCCAGAAATAGAATGACGGCGATGA
core_44	CTTATCGGGCGATGCAATTTCTAGCCCCT
core_45	ACGTAGCCATGCTCTAGAAAAACCGTTAATC
core_46	CTTATCTGACAGTGGGAGTGGCACACCAC
core_47	GCGCGTAAGCTGGCAACTAGAAGGCGCAGAA
core_48	AACAAGAGTCCACTACGAGTCTCCAGTTCAAGTCAAGTTAATA
core_49	TCCAACGTCAAAGGGCGGAAGTCACTGATCTATAATACCGCTACAGG
core_50	AGGGCGATGGCCACTCCATGCTACGTTTCTCGAGTTGTTGGTAT
core_51	CTAATCAAGTTTTTTGCGCTTAATAATGCCGAAGTCATACGATCA
core_52	AAAGCACTAAATCGGGTCACGCTCGGAAATGATGCTTTTTGTTGTGCA
core_53	CCCGATTTAGAGCTTAGCGGGCGTTCAATATATAATTCT
core_54	CGAGATAGCGCAGACAGTCTATCGAGCTTGCCCGTGCAGATGCGGAGTTG
core_55	GAATAGACCTATGAACTAATGACCCCGTAATCTCAGTT
core_56	GCAAAATTTTGTACTAAGTGGTGGGTCAGCTCAGGGTGGTCACGA
core_57	CGAAATCGATTAATAACTAGCTAGCTTAAGGGGCGCTTT
core_58	GTGCCACCGATCCTTTTGGCCCCAGCACCACCGCTCCTGGACGTAGCC
core_59	ACATTTCCGTGCCGTCCCCTGGAAGCTCCCCTCGTGCCG
core_60	TTAGAAAAGGGAGCCCGACAGGACTATAAAGATACCAAGCAAAGAATAGGGGTCAATCTA
core_61	GAATGTATTTCTTTTCTAGGGCGATCTCGAAGACGCGGA
core_62	ACATATTTCTGACAGTCAATCCATGAGAGAAGCCCTCGAACTTCACC
core_63	AGCGGATCGTTTTTTCGGGGCGTACTTGGCATGGTCATGT
core_64	GCTACTCTCAAGGATCCCTCCATCAAAAAAGGTTACGAC
core_65	GATGTAAGAGCGCAGTTTCTACGACCCGCCCATTTGACGTCAATGGGGTGGAGACTTGAA
core_66	CCCCTCGCAATAGGCCGTGGACCTCATAGCTCAGCGTGTAGGTATTGATTACT
core_67	TCAGCATCAGCAAAAACCTCACTGCTTATAATCCCGTGAGTCAAACCGCTATCCACGCC
core_68	CTTTTACTAAATTTTTGTCTATCGCCTTTCTCCCTTCGGGAAGCGTGCGAATTAGCTCATT
core_69	AAAAACAGAAGCGTTCATCACCTCCGACCCTGCCGCTTACCGGATGTAGATAGTTAAAT
core_70	GAAGGCAAGTTAATTGAAGGCACGGGGGAG
core_71	AAACGTTCCCCACCCCTGCTATTGTCTTCTGTTGTTT
core_72	GCGCCACACAGTGGGGTCCAGCAGCGGGCAAGGCAGGCACCTACTCTTTAAA
core_73	CTCTTGCCCAACCACGCGGAATTGTCAGTGCCCAACAGCCGATTTTATATACGGG
core_74	CCTCCGATCATAGTTGTGCCCCAGGATGTTGCCGCTCT
core_75	CGTTGTCAGGGTGTGAGCAAAGTGGATGTCAAGAGTC
core_76	TTTATCCGTTACCGCAATCAAAACAGTTTGGCGGTGTAGGTGCTTCCGCTCCAAGCTATATGGG
core_77	CCTGCAACAGTTTCCCTCATTGGAGGAGCTGGTTCCGCCGTGGCAATAGGGAGGTGCCCA
core_78	TTACCATCTAAATTAACCTGGGTAATAGCGATGACTAATACGTA
core_79	AGATAACTGCTCACCATAGTTGCCGTCGCTCTGATATAGA
core_80	GTCGTGTAAGTATAGTCTCCTTTGCCAAGTAGGAAAGTCCCATAAATGATACAACCTGGT
core_81	ATCTCAAGAAGATCCCCCTTATATGTTGAGA
core_82	GGGTCTGACGCTCAGTTTTAACTGCACCCA
core_83	TGGAACGATAAACCAGGCCCTCGCGGGTAGCGGCTGAAGTGGTAGTG
core_84	GTTAAGGGATTTTGGTCCGCTTTTACCAG
core_85	TCATGAGACCACGCTCTTACGTCCTTGAAGAAGTCGTGCCTCGAT
core_86	TTCACCTATAAATTGT
core_87	AAAATGAAGTTTTAAATTCGCGCGGGCAGCA

core_88	GTTTACCGTAAATACTCCACCCATTGACGTCAATGGAAAGTCCCTATTGGCGTTACTATGGGAACATACG TCATTATTGACGTCAATGGGCGGGGGTCTGTTGGGCGGTACGCCAGGCGGGCCATTTACCGTAAGTTATG TAACGCGGAACTCCA
polyT_1	TTTTTCGAGCATCACAAGAACGTGGCGTTTTT
polyT_2	TTTTTAACCCCGTTTCAGCCTGGGCTGTGAAGCCAGGGTTGAGCCAATCCT
polyT_3	TTTTTACCGTCATTGACGAGTGAGGCACTTTTT
polyT_4	TTTTTATCCGTAACACTATCGTCGACCGCTCAGATTACGTTACCTTCGGATTTTT
polyT_5	CTTGGAAAATTTTGAGTCCAACCCGGTATTTTT
polyT_6	TTTTTGCCTATCAGAAACGCAGTAAGACCAATAGGTTTTT
polyT_7	TTTTTAGACACGACTTATTAGCGGTGGTTTTTTTTGTTTTT
polyT_8	GTTACCAGAAGTAAATCTGTCTCATTTATCAGGGTTTTT
polyT_9	TTTTTCTTCAGCTCGATGCGCCTTGAAGTCGATGCCTTTTT
polyT_10	TTTTTCTGGTAACAGGATTAGCGTGGTTGTCTGGGCAGCAGGGGTGGGCAGTAAGT
polyT_11	TTTTTATCCACATAGCGTAAAGCATTAAATTATGGC
polyT_12	TTTTTGGCGGTGCTACAGAGAAAGCGAAAGTGTGCTCAG
polyT_13	TTTTTGCGGGAAGGAAGGTCCGCTGGATTGAAGAGGCCGAGCACTGCTATTGAAG
polyT_14	TTTTTCGGCTACACTAGAAGGCCTTGCTGTCCGGTCTTGAAGTGGTGGCCTAACTATTTTT
polyT_15	TTTTTAGAAAGGAAGGGGCAGCAGGCCTTTTT
polyT_16	TTTTTTCGCTCTGCTGTGCACGTTTTT
polyT_17	TTTTTTTATTGTCTCATGAAGCCGGCAAATCGACGCTCAAGTCAGAGGTGTTGCTGG
polyT_18	TTTTTAAAAGAGTTGGTACCCACAGTATTTGGTATCTTTTT
polyT_19	CTTCGGGGCGAAACTGTTGCCGGTAAGAGCGAGGTATGTATTTTT
polyT_20	TTTTTTTATCACTCATGGGCAGCGTTTTTT
polyT_21	TTTTTCCACCGCTGGTAATTTGATCCGGCAAACAAATTTTT
polyT_22	GAAGCTAGCAGGGCACGGGCCACTGGCAGCAGCCATTTTT
polyT_23	ATTTTCGTTTACCAATTTCTCTGTACTGGGCATAATGCCAGGCGGGCCATTTTTTTTT
polyT_24	TTTTTCTATCTCAGCGGTTGGCCGCAGTTTTTT
polyT_25	TTTTTTTTGCAAGCAGGCGCCTTTTTTT
polyT_26	GCTTAATCTCAATAGCATAGGCTCCGCCCCCTGATTTTT
Loop_1	AGGAACCCCTAGTGATGGAGTTGGCCACTCCCTCT
Loop_2	AGAGGGAGTGGCCAACTCCATCACTAGGGGTTCTTGTAGTT

2.2.7 sc_mCherry5

Scaffold sequence for sc_mCherry5:

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Corresponding DNA origami designs and staple lists:

2.2.7.1 sc_mCherry5 16HB_1

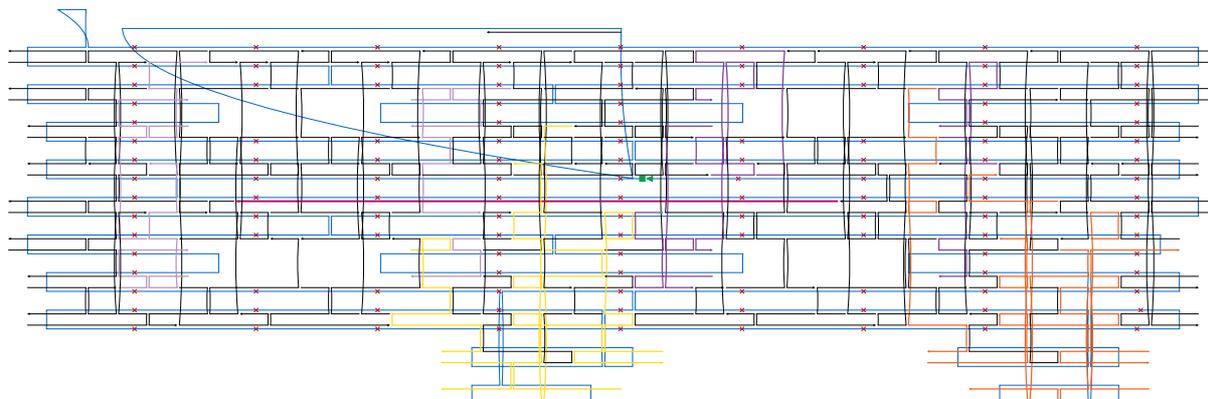


Fig. S41 | Scaffold routing and staple design for sc_EGFP5 16HB_3. Scaffold routing is shown in blue, staples are given in black, breakpoint for addition of sequence is marked by a green square (5') and arrow (3'). Long 154-mer 'primer' sequence at the promoter (5') region is given in pink. Protrusions 1 and 2 are shown in yellow and orange respectively, recess' 1 and 2 are given in light and dark purple, respectively. Design was prepared using caDNAo v0.1.

Table S28 | Individual staple sequences for sc_mCherry5 16HB_1

Name	Sequence 5' - 3'
core_1	TAATACCGAACTAATGACCCCGTAATTGATTGTCAAAGCCACTCG
core_2	ATGCTTTTATGGCGGGTTAATCATTAACTACATCCCTT
core_3	TTTTTCCCAACGATCAAGACCCGACATTTTT
core_4	TTTTTTCGGGGCGAAAACCTCTCATTGGAACGTTCTTTTTT
core_5	AACTTTACGTTAAGGGATTTTGGTCATCGA
core_6	TCCGCCTCCATCCAGTATAATTCTGGGTGAGCAAAA
core_7	TAGTTCGCCCCGCTTTAAACCCCGCACCTTGAAGCGCATCCAGCCCA
core_8	TTTGGTATAAACCACCGCTGGTAGTCTGCTGA
core_9	AAGGCAAAATGCCGGAATACTCTGAGCGGATACATATTTGATTTTT
core_10	TCACCAGCGTTTCTGGGTGAGCACGCACATTACCGTCTAGGAAGAAA
core_11	TTTTACTTTGCACCCAACCTGATCTCGAGTTGC
core_12	TACCGCGTGACCACTCAGCATCTGTAAGCGGAACGTGGGCTGGCAACGTCAATG
core_13	TGCAATGAATTTATCAGCAATAAAAAGTCATT
core_14	TTTTCTACGGGAAGCTAGAGTAAGGTGTTATC
core_15	GATCTCAAGAAGATCCTTTGATCTGTGCACGCTCCCTTCGCTATCCAGCCCAT
core_16	AAAAAAGTAGTTTTCGCAACGTTTCCGATCG
core_17	TTTTTGTTGGCATCGTGGTGTCACTGTGCAA
core_18	CGGTGGTTCCAACCCGGAAGCTCCACGTAGAT
core_19	TTTTTTAGCTCTTGATCCGGCAGCAGTTTTT
core_20	TTTTTGCGACACGGAATGTTAAAAAGGGAATAAGGTTTTT
core_21	GCGAGGTATGTAGGCGGCAGGGGGCCTCGCCC
core_22	TTTTTCCACTGGTAACAGGACGCCACTGGCAAACGGCTTCATTACGCTCCGGTTTTTTTT
core_23	TTTTTATGATTTAGAAACCATCACCTTTTTT
core_24	AAAATAAAAAGGCAATTAGGAAGACTTCCCAATCCTCCCCCTTTTTTT
core_25	ACCAATAGAAAGAATACCAAACAACTCCCA
core_26	GCCGAAATATGGTGGCGAATACACAAGAGGCC
core_27	CGGCAAAGTAACTAGGATCTCACCTAGATCCTTTTGTGGAACCGGGTGTAG
core_28	CGACTATTTAGCAGAAGCCAGTTACCTTCGGAAGAGTTGGTTTTT
core_29	GGCGAAAATCCCCGAAAACACCACCCCGCCG
core_30	CTATTAATTAATATTCCCATTGGTGTAGCG
core_31	TGTAGGTAATAAATACTTACTGTGCATGCCATCCGTAAGGGTCTGCATAAATCA
core_32	GCTCAGCTGGAGACTTGGAAATCCCTCTGCTTACATCCG
core_33	TTTTTGGACTATAATAAGGTGATGATTTTT
core_34	TTTTTAAATCAAGTTTTTTGGGGTCTAAAGGGGGCGAACGGAAAGCGACCACCCCA
core_35	CCGTAAGCGCCACATAGCAGAACCTTTAAAACAAGGATCACTACGTG
core_36	TAACTAGCTAGCTTAAGGGCGAAGAGTCCAGCTCCAG
core_37	TTCTAGAGCATGGCTACGTAGATAGGGTTCCGGAAG

core_38	TAAGTAGCCTGTGACTGGTGAGTACTCAACCCCAGCCAGGAGTGTGTTGTTAAA
core_39	ACCGTAAAAAGGCCGCGTTGCTGTACCTGTCAGTTAA
core_40	CCATAGGCTCCGCCCCGCATATGACCATGGTATTCTTGGCGGGGAAGT
core_41	AAAAATCGACGCTCAACCGTCATTGTACTGCC
core_42	TGGCGAAGCGAGTTACATGATCCCCATGTGCTCGTCCCTGGTAAGACAGGGGCGGC
core_43	CACTAAATCGGAACCCGAGGTGATGGCCCTTACCGC
core_44	GGGCTATGTGGGGTTGCGTCAGCAAACACACAGGCGG
core_45	CATATATGCGAAAGGCGATGAGTACTCAGACAATGCGATGCAATTTGTCACGCT
core_46	TTTTTCTGGGCATAATGCACCCGCTCTTTTT
core_47	TTTTTCTTGACGGGAAAGCCAGCCCCGATTTAGAGTTTTT
core_48	AGCGGGCGCGCTAAAGTGCCACCTAAAT
core_49	GCTAGGGCACTCCAACACTATTAATCTTGCCCGCGTCAATACGGGAGATGTAAC
core_50	GTTGTTACTGGAACAATTCCGATCCTGAGAATAGTGTATGCGGCGACGCTCACCG
core_51	CCCGTGATGATGATCTCCAAGCTGGGCTG
core_52	ATAGCGAGATCTCGACTATCGGAGTTCTAGGACAGTATTTGGTATCTGCGC
core_53	TGACTAATCTCGTGCAGCATCACAAGCGGTTAGCTCCTTCGGTCCGTTGCCATGTTCCGAC
core_54	CTCTGCTTCAGCTCATCAGTGCCGTCGTGTAGATAACTAGAGATTATCAAATGGTCTGA
core_55	GCCGTTACCGTCGGACTTGTAGGTGGTCTTGAGCAACATATGATTTTTTTGTCAGAAGTAAGTTGGC CGCA
core_56	TTTTTGGTCTGGGTGAAGTCCAAGATAC
core_57	TTTTTCTACGTAGCCATGCTCGCGCCATGCTACTTATTTTTT
core_58	CTGCAGGATCTGACGAATGCTTAATTTTTTATACGGGAGGGCCGAGCGCAGAAGT
core_59	TGTGGAGAGAAAGGCATATATGATCTCAGTTAAAACCTCA
core_60	TTTTTCTGGGTCACGGTCACCTGCAGGGAGGAGTTTTTT
core_61	TTTTTGCTGTCTGCCAAGTCCCGAATTTTT
core_62	TGACACCTTCCGCGGTGGCAATAGCCAACCAG
core_63	CCTCATTTCAGCAGCGGGCAAGGGTGCACACCACGCCACGTTGCCTGACAACGG
core_64	CTCGGAGGAGGCCTCGAACTCCTGTCAAACCCGGGAAGCG
core_65	TGGTCCCCTGAGGCTGTCCAGGCGAAGGGTGTACATCTTGAGT
core_66	TCACCTTGAGGCGCCGTGTCCAACCGTTCGTAAGTGTCCACGATGGTGCAATCTTACGGGC
core_67	GCCCCTGTTATTAGGAAAGGACAGTGGGAGTGTACACG
core_68	CAGCCGAGGGCGGACCTACCGTTGTTAAAATTCGCG
core_69	CACCACGGCCAGGGTCAAGGCGAGTCTCCT
core_70	AAGGACGAGCAACACAGGCCGAAGGGACGTAGCAGAAGGAGCAACCACCGACAACCGTTGA
core_71	GCCAAGGAGCAGAGCCGGCAGCATTCTCTG
core_72	TGATCTCGCCCTTCAGATACGGGAAGCAATAGCATGATACAAAGGCAGCTTCAG
core_73	TGGCCGCCGTCCTTCATTAAGCAGCGTATCCACATAGCGTAAAAGG
core_74	CGTCGTAGTGGTCTTCTCTGCATTACGGGGCGGAGCCCTGCCAAAATTACCGGAGCGCTGCCAAGT GGG
core_75	ACCTCAGGATGTACTCCATGTGTTTACGCCGACCGC
core_76	TTTTTACTTGTACAGCTCGTTCCAATCCAGGCGGGCCATTTAGTCAGAGGCAGGCGTT
core_77	TTTTTAGGAGCTGACAGGTGGTGGCAATGCCGAGGGGTGGCGAG
core_78	AAGGAACCCCTAGTGATGGAGTTGGCCACTCCCTCT
core_96	CAGTTTACCGTAAATACTCCACCCATTGACGTCAATGGAAAGTCCCTATTGGCGTTACTATGGGAACAT ACGTCATTATTGACGTCAATGGGCGGGGTGCGTTGGGCGGTGAGCCAGGCGGGCCATTTACCGTAAG TTATGTAACGCGGAACCTC
rec1_1	GGCTTACCATCTGGCCTGACTCCAGGCACCTATCTCAGCGAT
rec1_2	TCCATTAGTTGCCCCAGTGCTTAAATTTTTCCAGTTGACATTTTGAAAAGTC
rec1_3	ATCAGGGTTATGGGTTCCGAAAACAGGTGTTGAGATCCAGTTC
rec1_4	GTTAATTCAAATAGTGTCTCAATACTCTTCTTTTTCAA
rec1_5	TATATAGAACCAACTTTAAACCTGTCTTGTAAACCT
rec1_6	CAGCATGCCTGGAATAGAAGCCTTAATAAAGGAAGTCAGGGCG
rec1_7	CCCCCACTATTGTTCTTATCGGGAATTGCG
rec2_1	ATCAATCTAAAGTAAAGTGGATATCCTCCCTCGGGG
rec2_2	ATCCGGTAAACTCGTGGGACAGGACTTGGAGC
rec2_3	CTCTCCTTGCTACATGCAAGCAGCAGATTACACTAGATGAAGTGG
rec2_4	GGCCAGCAAAATGGCGCTTGTGCGGGGTCTGACGCTCAAAATT
rec2_5	GGCCAGGAACTCATGGTTATGGCAGCACTGCCTATTAATTTCTCATAGTTCGGGCCATGTTGTCA
rec2_6	CCTATCAGAAACGCAAGAGTCGGCGCCGTTCCGCTTCAATGGGG
pro1_pass_1	CAGTACCCTTATGCCTGGACACCTCTCGACAAGCCAGTATGGCTGGCAACTTTTTT
pro1_pass_2	AGTGCCTCAGCCTCCCACGGCACCTTAATTGTGAGTGCCAAGCCACAACCTCCTATAAAGAGACAC GTTTTT
pro1_pass_3	ACAAGGAGGAGAAAATGGATTGAGGGCGAGCAGGAAGGAATTTTT
pro1_pass_4	TTTTTGGTCCGCTGAAAGCCGGCCGCTTGACGTGCTCAC
pro1_pass_5	TTTTTCCGCGCAGAATCTAAGGCACGTTTTT
pro1_pass_6	TTTTTAGAAGGCACAGTCGAGGCAGATCCAGGTGTGATTCCGGATTTAT
pro1_pass_7	TTTTTGGGGAGGGGCAAACAACAGTTCTATTGAGACGCGGTAACCAGTTTTTGGTGGACCGAGA
pro2_pass_1	ACGCGCAGTGCCTTCTGCGCCCGCATCATACACTTGAGTTAAGAATACCAGTTAGTTTTT
pro2_pass_2	GACGTCAATTTGTAATCCAGAGGTCTCGGCGTTGATGTTCCGGTGGATTTTT

pro2_pass_3	CCACCCTTCGGGGATGTCGGCGGGGACAGCTTTCCCACTTGAAGCCCTTTTT
pro2_pass_4	GACGTTGTTAGATGAATCACGCGCCAAGTAGTCCGTCCTTTTT
pro2_pass_5	TTTTGTGGCGGCTGATTGGACCATGCCGAAGTAGGACCCTCGTA
pro2_pass_6	TTTTTCTCGTTGTGGAGGTGAGGCAGCTGCACAAATTAGGGGGCGTACTTGCCTGACG
pro2_pass_7	TTTTTGAAGTTCACTCGCCGTCCACGCCGGGTACCTTCAGCTTGGCTTTTT
pro2_pass_8	TTTTTCGGGGAAGGTGCTTCAGCGCAGCTTCGCCCTC
pro1_x8_1	TGATACAAGGGGAGGGGCAAACAACAGTTCTATTGAGACGCGGTAAACCAGTTTTGGTGGACCGAGA
pro1_x8_2	GGACGTAGGGTCCGCTGAAAGCCGGCCGCGTTGACGTGCTCACC
pro1_x8_3	ATTACTTGCCCGCGCAGAATCTAAGGCACGAGGCATTA
pro1_x8_4	CAGTTACCGTTATGCCTGGACACCTCTCGACAAGCCCAGTATGGCTGGCAACTGGAATTGT
pro1_x8_5	AACACCACAGAAGGCACAGTCGAGGCAGATCCAGGTGTGATTTCCGGATTTAT
pro1_x8_6	AGTGCCTCACGCCTCCACGGCACCTTAATTGTCAGTGCCCAAGCCACAACCTCCTCATAAAGAGACAC GTTACAGCTC
pro1_x8_7	ACAAGGAGGAGAAAATGGATTGAGGGCGAGCAGGAAGGAACAGAAGGA
pro2_x8_1	GACGTCAATTTGTAATCCAGAGGTCCTCGGCGTTGATGTTCCGGTGGAAACCCTAA
pro2_x8_2	CTCATTTCGGGGAAGGTGCTTCAGCGCAGCTTCGCCCTC
pro2_x8_3	GACGTTGTTAGATGAATCACGCGCCAAGTAGTCCGTCCTCAAAGCGAA
pro2_x8_4	AAGGGAAGGAAGTTCACCTCGCCGTCCACGCCGGGTACCTTCAGCTTGGCTTTTT
pro2_x8_5	ACGCGCAGTGCGCCTTCTGCCGCCCGCATCATACACTTGAGTTAAGAATACCAGTTAGCTCGAAGA
pro2_x8_6	AGGCAGATTCCTCGTTGTGGGAGGTGAGGCAGCTGCACAAATTAGGGGGCGTACTTGCCTGACG
pro2_x8_7	TAAATCGGGTGGCGGCTGATTGGACCATGCCGAAGTAGGACCCTCGTA
pro2_x8_8	CCACCCTTCGGGGATGTCGGCGGGGACAGCTTTCCCACTTGAAGCCCTATTAGGAA
pro1_x5_1	TACAAGGGGAGGGGCAAACAACAGTTCTATTGAGACGCGGTAAACCAGTTTTGGTGGACCGAGA
pro1_x5_2	CGTAGGGTCCGCTGAAAGCCGGCCGCGTTGACGTGCTCACC
pro1_x5_3	ACTTGCCCGCGCAGAATCTAAGGCACGAGGCA
pro1_x5_4	CAGTTACCGTTATGCCTGGACACCTCTCGACAAGCCCAGTATGGCTGGCAACTGGAAT
pro1_x5_5	ACCACAGAAGGCACAGTCGAGGCAGATCCAGGTGTGATTTCCGGATTTAT
pro1_x5_6	AGTGCCTCACGCCTCCACGGCACCTTAATTGTCAGTGCCCAAGCCACAACCTCCTCATAAAGAGACAC GTTACAG
pro1_x5_7	ACAAGGAGGAGAAAATGGATTGAGGGCGAGCAGGAAGGAACAGAA
pro2_x5_1	GACGTCAATTTGTAATCCAGAGGTCCTCGGCGTTGATGTTCCGGTGGAAACCC
pro2_x5_2	ATTTTCGGGGAAGGTGCTTCAGCGCAGCTTCGCCCTC
pro2_x5_3	GACGTTGTTAGATGAATCACGCGCCAAGTAGTCCGTCCTCAAAGC
pro2_x5_4	GGAAGGAAGTTCACCTCGCCGTCCACGCCGGGTACCTTCAGCTTGGCTTTTT
pro2_x5_5	ACGCGCAGTGCGCCTTCTGCCGCCCGCATCATACACTTGAGTTAAGAATACCAGTTAGCTCGA
pro2_x5_6	CAGATTCCTCGTTGTGGGAGGTGAGGCAGCTGCACAAATTAGGGGGCGTACTTGCCTGACG
pro2_x5_7	ATCGGGTGGCGGCTGATTGGACCATGCCGAAGTAGGACCCTCGTA
pro2_x5_8	CCACCCTTCGGGGATGTCGGCGGGGACAGCTTTCCCACTTGAAGCCCTATTAG