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## Supplemental Information

# DNA origami signposts for identifying proteins on cell membranes by electron cryotomography

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**Table S1** –List of oligonucleotide staples required to construct SPOTs and their precursors or derivatives, related to STAR Methods. This table is also available as a CSV file for ordering from commercial oligonucleotide suppliers (Table S4). All oligonucleotides require a standard desalting purification unless indicated.

| Name | Sequence | Mix | Notes |
|---|---|---|---|
| **Staple Oligonucleotides** | | | |
| A-0[111]16[102] | TCGCCCAGCAGGGAGACTTTTCAACATTGAAAGATTCA | 1 | 25 nmol |
| A-0[195]4[182] | TTGTATCGGATTTTCATAGTTAGCGTAAACCCTCATAGGTGT | 1 | 25 nmol |
| A-1[116]67[121] | AAGCATCTTTAATCACCGCCTTAACAATTTTTCAATATATATCTTTAG | 1 | 25 nmol |
| A-1[140]5[149] | CCCTCAGGATACCGAGTAACAGTG | 1 | 25 nmol |
| A-1[163]3[174] | ACGTTGGTGAATAGTTAATGCCCCCTTACTCAGCCCTCAG | 1 | 25 nmol |
| A-1[203]3[216] | AACTTTCAAAGGAGCATGAAAGTATTAAAGAGGGTGGGATAG | 1 | 25 nmol |
| A-1[85]13[90] | TTTTTCGGTCGCTTAGTAAATTGGGCTGAATTAC | 1 | 25 nmol |
| A-10[106]66[92] | CCAGCGGATATTCATTGTCATTTCTTAGAGTGAGTAACCGAAC | 1 | 25 nmol |
| A-10[136]66[130] | CCACGCCTTTAGCCATATGGAGCGGAGACTTTACA | 1 | 25 nmol |
| A-10[166]31[166] | TTTTCCTCAGAACTAAAAACAGTTTT | 1 | 25 nmol |
| A-11[105]6[98] | ACATCAATCATACAGAACGAGGAGGCTTCGCATAAGTGTCGACTGCTCC | 1 | 25 nmol |
| A-11[126]67[139] | GAGCCACCACCGGACGACTTGCAACGCTCCTCAAAACTAATA | 1 | 25 nmol |
| A-11[64]67[90] | TTTTGCTCATTCAGATTCCCGCAACTATCGACTCAGTGCCAAGCTTGCATAAAGG | 1 | 25 nmol |
| A-12[134]5[125] | TTTTATTAGCGTTTGCGCCGCTTAATGACATTTAACG | 1 | 25 nmol |
| A-13[64]69[95] | TTTTCAACTTTAAATGGTCATTTGACCCCATTAACGAACCACCAGCAGAAGATAAAACAG | 1 | 25 nmol |
| A-13[91]13[111] | CTTATGCGATTTTAAGGGCAT | 1 | 25 nmol |
| A-14[111]51[101] | TCATCAACTGGCTCAACGGAATCATGAGGCCACTAAACGGAGTAGCACCCACCGACCAGT | 1 | 25 nmol |
| A-14[127]13[134] | TTTTTGTAGCGCGTTTTTTCGGTCATAGCCCCCTTTTTT | 1 | 25 nmol |
| A-14[90]40[81] | CAGTCAGGAATTAGCAATAAAGCC | 1 | 25 nmol |
| A-15[68]15[90] | TTTTAAAAATCTACGTTAATAAA | 1 | 25 nmol |
| A-16[101]17[127] | TCAGGAATTACGAGGCAGAATCAAGTTTGCTTTT | 1 | 25 nmol |
| A-16[127]15[127] | TTTTCTTTAGCAGGTAATTACGTCAGACTTTT | 1 | 25 nmol |
| A-16[90]42[81] | TTAGGAAGAAGCCTGCAAGGATAA | 1 | 25 nmol |
| A-18[127]20[98] | TTTTTAATCAGTAGCGACATAGTAAGAGCAAGGCTTTGTTTTGC | 1 | 25 nmol |
| A-19[68]20[77] | TTTTACGACGATAAAAGTAAAAT | 1 | 25 nmol |
| A-2[132]21[131] | ACAGAGGCTTTGAGTAAAACGACCAAGCGGCCGGAAATTTT | 1 | 25 nmol |
| A-2[143]10[137] | TAGCATACACTTCTTTGACCCCCAGTGCCTTGATAGTTGGCAGGTCCCA | 1 | 25 nmol |
| A-2[167]1[162] | GTCGTCTTTGCATCAGACACCAG | 1 | 25 nmol |
| A-2[209]0[196] | CCACAGAATTTTCATGATATAAGTATAGTCTGAAACCTTTAA | 1 | 25 nmol |
| A-20[131]19[127] | TTTTCGTCACCAATGAAACCATAAGAATGCAACGATAGCAGCACCGTTTT | 1 | 25 nmol |
| A-21[84]45[107] | AGCAAAAGGTAATAACCAAAATAAAGTACAGCTAATGCAGAACGCGCTTTTT | 1 | 25 nmol |
| A-22[131]51[131] | TTTTCATTAAAGGCAGTAGGGCTTTT | 1 | 25 nmol |
| A-23[93]7[111] | TGAGGATGTTACCTTTGAAAGAGGAGTAAGCGT | 1 | 25 nmol |
| A-23[98]95[107] | GAGGGAAAAGGGCGTTAAATATACCGACTGCCGTAAAG | 1 | 25 nmol |
| A-24[131]53[131] | TTTTTGGTTTACCAATCATAATTTTT | 1 | 25 nmol |
| A-24[92]53[83] | CAACTGCTTTAAACAGAAGATTG | 1 | 25 nmol |
| A-25[112]59[127] | CAATAGAAAAGAAAACATACAGAATACCGATGATGAAACAAATCAATATCGATAGCTT | 1 | 25 nmol |
| A-25[54]51[69] | AATCAAAAATTCGCGTTAAATCAGCTCATTACGTTAAGCCCCAATGTACCCAGAGAAT | 1 | 25 nmol |
| A-25[86]26[90] | TAGTCAGAAGTTGGAATAAG | 1 | 25 nmol |
| A-26[104]55[107] | ACACCACTATTTTGCAAATCCTCAAATATAT | 1 | 25 nmol |
| A-26[131]25[131] | TTTTCAACATATAAAATTCATATTTT | 1 | 25 nmol |
| A-26[89]6[81] | CAAAGCTGACCAATTAGCCGGAA | 1 | 25 nmol |
| A-27[50]57[65] | TTTTCGAAAGACTGGGAACAATCAACATTA | 1 | 25 nmol |
| A-27[77]62[91] | TTAATTTACGCAGTATGTTAGTGATTAATCAATTAATCGCGC | 1 | 25 nmol |
| A-28[142]62[136] | AACCGAGGAAAAAACAAAATA | 1 | 25 nmol |
| A-28[166]61[166] | TTTTGAACAAAGTAACAATTTCTTTT | 1 | 25 nmol |
| A-28[85]61[76] | CTTATCGAGCTTCAAATCGCACT | 1 | 25 nmol |
| A-29[147]2[144] | AAGTAAGAATCCTCTGGCCTTACAGCTTCAGCGAAGGAACGAGGG | 1 | 25 nmol |
| A-29[54]29[83] | TTTTGGAAGCAAACTCCATTGAGTTAAGCC | 1 | 25 nmol |
| A-29[84]9[93] | CAATAATTGTACAGGAGTAATCTT | 1 | 25 nmol |
| A-3[162]4[156] | CCGCCAGAGGTTTAGTAC | 1 | 25 nmol |
| A-3[175]5[188] | AACCGCCCGATCTAGAATTTTCTGTATGGGTTTATTCGGAAC | 1 | 25 nmol |
| A-3[78]43[107] | TTTTGGGTAAAATCATAACCCATAACGAATAGATACCAATCAATAATCGGCTGTTTTT | 1 | 25 nmol |
| A-30[104]57[108] | GAGCGCTAGAAACAACTGGCACAAACGTAATAGTGCTTTTTAACCT | 1 | 25 nmol |
| A-30[153]11[166] | TTAGACGAATAACACGCCACCCCCTCAGAGCCGCCACTTTT | 1 | 25 nmol |
| A-30[166]63[166] | TTTTGGAAGCGCAACAGTAACATTTT | 1 | 25 nmol |
| A-31[54]65[76] | TTTTGAGAGTACCTTTAATTGCTATAATGCATGTGCT | 1 | 25 nmol |
| A-31[84]11[104] | ATAAGAGACCCAAAAGGCTTGCCCTGACGAGAAAC | 1 | 25 nmol |
| A-31[98]10[107] | TTGCGTTTTGTTTACACCA | 1 | 25 nmol |
| A-32[125]27[131] | CCCAATCAAATGAAGAACAAAGCTATCTAATAACGTAAAGGTGGTTTT | 1 | 25 nmol |
| A-32[166]65[166] | TTTTATTTGCCAGTCCTGATTATTTT | 1 | 25 nmol |
| A-33[105]35[115] | CCAGCTAATCAAGATACCGCGCCCA | 1 | 25 nmol |
| A-33[54]34[54] | TTTTTGTTTTAAATATAATTCTGCGAACTTTT | 1 | 25 nmol |
| A-34[153]35[166] | TTTAGCGCCGGTATTCTAAGAACTTTT | 1 | 25 nmol |
| A-34[166]33[166] | TTTTGCGAGGCGTCAGAGCCTATTTT | 1 | 25 nmol |
| A-34[99]8[77] | TGCTTTCATTCCATATAACAGTTGTGAATATCAACGTTCATCAAACCAGGC | 1 | 25 nmol |
| A-35[116]7[131] | ATAGCAAGCATTTTGAAGGAACCAGCCGCCAGACAGGAGCAGTCTGGCTTTTGATTTT | 1 | 25 nmol |
| A-35[133]8[143] | TATAGAAGGCTTATAACCTCCACCGCCTCTCAGAGAGACGATATTA | 1 | 25 nmol |
| A-35[54]36[54] | TTTTGAGTAGATTTAGATAACCTGTTTATTTT | 1 | 25 nmol |
| A-35[83]34[100] | TACTTTTCATCGTAGGAATCATTTAGT | 1 | 25 nmol |
| A-36[104]36[81] | TTTTTGCAAGCCGTTTTTAATTTC | 1 | 25 nmol |
| A-37[54]76[63] | TTTTGCTATATTTTCATCTACTATTGCGCTTTTCCAG | 1 | 25 nmol |
| A-37[83]38[81] | CTGCTCATCCGCAAAAAG | 1 | 25 nmol |
| A-38[107]37[104] | TTTTTTTAAACCAAGTACGAGAACAATTTTT | 1 | 25 nmol |
| A-39[47]78[63] | TTTTTTAACATCCAATAAATCATAGCTAAACGGGGAGCGTATTG | 1 | 25 nmol |
| A-4[111]1[115] | AAAGTACCGAAGGCACCAACCGACTAAAGTTA | 1 | 25 nmol |
| A-4[155]3[161] | CGCCACTCAAAAACACCCTCAGAA | 1 | 25 nmol |
| A-4[181]2[168] | ATCACCGGCCTATTCAGCTTGCTTTCGAAGTAAATAAGTTTT | 1 | 25 nmol |
| A-4[222]2[210] | CCGTCGGAGGCTGAAAAAAAGGCTCCAAACAGTTTAGCATT | 1 | 25 nmol |
| A-40[107]39[107] | TTTTTTCCAAGATTAAGCAAAAACGGGTATTTTT | 1 | 25 nmol |
| A-40[76]71[92] | AGCATAAACAGGCACATCAATTTTGGGGGAATGGCTAATGCGCGAACTGATAGCTTTT | 1 | 25 nmol |
| A-41[47]41[76] | TTTTAAAACATTATGACCCTGTAATACTTT | 1 | 25 nmol |
| A-42[107]41[107] | TTTTTTCTTTCCTCAACTTATTTTATCATTTTTT | 1 | 25 nmol |

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|---|---|---|---|
| A-42[72]80[66] | AGAACCCTCAGATTGCCTGGC | 1 | 25 nmol |
| A-42[80]47[102] | AAATTGTAGAAAAGTCCTACATGTTCCGACAAACAGTCAAATCAATTTTCGAGCCAGT | 1 | 25 nmol |
| A-43[47]44[47] | TTTTGCAATGCCCATCATATCCTGAGTATTTT | 1 | 25 nmol |
| A-44[107]14[91] | TTTTTCTGTTTATCAACCCAAAAGTTGAGATACGAACTATTATAC | 1 | 25 nmol |
| A-44[69]81[85] | AAAAATACTAATTTCCTACATACATTGGCAGATTCACCAGTTTT | 1 | 25 nmol |
| A-45[47]46[47] | TTTTATGTGTACAGACCTGTCGGTAAAGTTTT | 1 | 25 nmol |
| A-46[107]18[95] | TTTTTAATAAGAGAATATAGCGAGACA | 1 | 25 nmol |
| A-46[72]84[63] | TAAAGTAATTGACGACAGGCCTTG | 1 | 25 nmol |
| A-47[47]48[50] | TTTTATTCAAAAGGGTAGCTGATAATTTT | 1 | 25 nmol |
| A-48[100]89[89] | TTTTTCAGAGGCCCATCAATATGAGTTTTTATATTTT | 1 | 25 nmol |
| A-49[50]50[50] | TTTTATTAATGCCGAGAGTCTGTTTT | 1 | 25 nmol |
| A-49[77]49[100] | TTTTTGAGAGATATTTAGGTTTTT | 1 | 25 nmol |
| A-5[114]66[109] | TAAGTACAACCACAGCATTCCAGAACACGTCAACAAATAAGGAACAAAACTCGTATT | 1 | 25 nmol |
| A-5[126]1[139] | GGGTCAGCGATTATAAAGAGGCAAAAGAAACGGCTATCGTCA | 1 | 25 nmol |
| A-5[150]9[166] | CCCGTATAAACTTCTTAAGATATTCACTTTT | 1 | 25 nmol |
| A-5[189]1[202] | CTATTATCCCGGAAGAGCCACCACCCTCCAGCCCTGCTAAAC | 1 | 25 nmol |
| A-50[131]6[115] | TTTTTTAATTGAGAATCGCCCGCTCAATGAATTAATTGACGAGTG | 1 | 25 nmol |
| A-50[69]85[85] | ATTGCCTGGAGAGGCCGTTCTGAGAAAGTAACCGTCACTTGCCTGAGTAGAAGATTTT | 1 | 25 nmol |
| A-50[73]51[92] | GGTCCGATGAACGGTAATCGTAAAATT | 1 | 25 nmol |
| A-50[97]91[89] | CAACATGTACTACAAAGGCTAAGGAGGCCGTTTT | 1 | 25 nmol |
| A-51[102]94[91] | ATATCATATGGCTGCGCACGGGGAAAGCCGG | 1 | 25 nmol |
| A-51[50]52[50] | TTTTGAGCAAACACGGTTGATATTTT | 1 | 25 nmol |
| A-51[93]22[84] | CTTACTTGAGCCATTTGGG | 1 | 25 nmol |
| A-52[131]93[131] | TTTTTACTAGAAACGCCGCGCTTTTT | 1 | 25 nmol |
| A-52[92]94[77] | TACAAACTAGCATGTCGCTGGCAGGCGAGA | 1 | 25 nmol |
| A-53[50]95[65] | TTTTATCAGAAAATATTTTGCTACGTGAAC | 1 | 25 nmol |
| A-53[84]24[93] | TATAAAATAAGGCGACATT | 1 | 25 nmol |
| A-54[114]60[105] | TGGGTTAATTTGTAAATGCTTAGGCGCTGAGAACATAGATGTGAGTGAATAA | 1 | 25 nmol |
| A-54[131]95[131] | TTTTGACCTAAATAACCCTAAATTTT | 1 | 25 nmol |
| A-54[92]95[83] | GATAGCAAATATTTAATCAAGTT | 1 | 25 nmol |
| A-55[108]50[98] | TTTATTTGAAAAGAATAATTTAGTAAAGCCAAATATTTAACAACGC | 1 | 25 nmol |
| A-55[40]56[40] | TTTTTTAAATTTTTGTCTGGCCTTCCTGTTTT | 1 | 25 nmol |
| A-55[87]59[97] | AGAAAACTTTTAATCGCAAGACTACAATTTATTTTTCCC | 1 | 25 nmol |
| A-56[131]55[131] | TTTTATAACTATATCATCTTCTTTTT | 1 | 25 nmol |
| A-56[85]57[76] | AAGAGGAACGCCATCATGAGCGA | 1 | 25 nmol |
| A-57[109]4[112] | CCGGCTGATGTCACAATAAGACAAGGTAAATTCACCGTATTACCATTAGCAAGCGAAAC | 1 | 25 nmol |
| A-57[40]58[40] | TTTTTAGCCAGCTTTCAACGGCGGATTGTTTT | 1 | 25 nmol |
| A-57[66]24[50] | AATGAAAATAATCAGGTCAACGAGAATGACCATATTTT | 1 | 25 nmol |
| A-57[77]56[86] | GTAACAGGTCTGAGAGACA | 1 | 25 nmol |
| A-58[131]57[131] | TTTTAGATTAAGATTGGGTTATTTTT | 1 | 25 nmol |
| A-58[85]59[76] | TCATAACCCGTCGGATGTAGATG | 1 | 25 nmol |
| A-59[40]60[43] | TTTTACCGTAATGGGAAGGGGACGATTTT | 1 | 25 nmol |
| A-59[66]26[50] | TGGTTCTCCGTTCAAATAAGATTAAGAGGAAGCCTTTT | 1 | 25 nmol |
| A-59[77]58[86] | GGCGCATTAATTAACAAAA | 1 | 25 nmol |
| A-6[114]5[113] | TACTGGCGACAATCCGTAA | 1 | 25 nmol |
| A-6[131]23[131] | TTTTTGATACAGGGAAATTATTTTTT | 1 | 25 nmol |
| A-6[80]23[92] | CGAGATCCCCCTCAAACGAT | 1 | 25 nmol |
| A-60[104]26[105] | CCTTGCTTTAGAATCCTTGAAAAGAGTCAGAAAATCGCAAAG | 1 | 25 nmol |
| A-60[135]1[132] | AGTACATAAACATCAAGACGCAATTACCGAAGGAAAGCGGTTGAGCGCCGACTTGCGGG | 1 | 25 nmol |
| A-60[166]28[147] | TTTTATTTGAATTACCTTTTTACATTTTACCAGA | 1 | 25 nmol |
| A-60[69]28[54] | CATCTGCAGGAAGAGCGAACCAGACCTTTT | 1 | 25 nmol |
| A-60[97]60[70] | TCTGTAAATCGTCGCTATCGTAACCGTG | 1 | 25 nmol |
| A-61[105]8[98] | AAAAGAACAAAAGAATGAAATTACCGTTCCACAGA | 1 | 25 nmol |
| A-61[43]62[43] | TTTTCGACAGTATCGGAAACCATTTT | 1 | 25 nmol |
| A-61[77]28[86] | CCAGCTTATTCATTGACTC | 1 | 25 nmol |
| A-62[114]30[105] | GAATACCAAGAGAAATAGGTAATT | 1 | 25 nmol |
| A-62[135]10[126] | ACGGATTCGCATTTTCAACACCCTAATAGCACCTCAGA | 1 | 25 nmol |
| A-62[166]63[153] | TTTTGTACCTTTTACATCGGTGAATAT | 1 | 25 nmol |
| A-62[90]63[69] | AGAGGCGAACAGCTTTCCGGCACCGCTTCCATTCA | 1 | 25 nmol |
| A-63[43]64[43] | TTTTGGCAAAGCGTCTTCGCTATTTT | 1 | 25 nmol |
| A-63[70]30[84] | GGCTGCGCAACTTCAAAATTAGAGAGAT | 1 | 25 nmol |
| A-64[108]32[98] | ATAAATTTTGCGAAACGATTGATGG | 1 | 25 nmol |
| A-64[139]35[132] | ATAATCCTGATTGTACCAGAATATTTATATCTTACCGGGAGGAATCAGA | 1 | 25 nmol |
| A-64[166]33[153] | TTTTTCAGATGATGGCAATTATCATATTTACAAAGTCTTTC | 1 | 25 nmol |
| A-64[62]59[65] | GCGGGCCCCATTCGCTGGTGCCGGCCTCCAGTTTGTAGGTCACGT | 1 | 25 nmol |
| A-64[69]32[54] | GATCGGTAAGGGGGTGTAGCTCAACATTTT | 1 | 25 nmol |
| A-65[43]66[43] | TTTTTTACGCCAGTTTCCCAGTTTTT | 1 | 25 nmol |
| A-65[86]33[104] | AAGTTCTTAATTGTGTCTGGAAGTATTTTGCAC | 1 | 25 nmol |
| A-66[108]69[115] | AAATATCTAAACTGGTCAGGTCAGTATTA | 1 | 25 nmol |
| A-66[129]69[136] | AACAACTAACATATCAAACAACAGTGCCA | 1 | 25 nmol |
| A-66[166]67[150] | TTTTACATTTGAGGATTTAGGCCG | 1 | 25 nmol |
| A-66[76]67[69] | TAAGTTGGGTAACGGACGGCC | 1 | 25 nmol |
| A-66[91]65[85] | GTTATTAATTGCGATGCAAGTTAA | 1 | 25 nmol |
| A-67[122]62[115] | GAGCATTCGACAGAAACCTTGGATTTGCGTAGCTGATTGCTTT | 1 | 25 nmol |
| A-67[140]60[136] | GATTAGAAAGTATTAATTATCCATCAATCGTCAGAGAGAAACAATTAATTTAATGGAAAC | 1 | 25 nmol |
| A-67[151]69[166] | TCAATCACCTCAGCAGCAAATGAAAATTTT | 1 | 25 nmol |
| A-67[43]65[62] | TTTTCACGACGTTGTAAAACCCAGGGTCTGGCGA | 1 | 25 nmol |
| A-67[91]61[104] | AATTGAGGAAGGTTCCTTTGCCATTATCTGGAAGGGTAAAACTTACAAACCTGAGC | 1 | 25 nmol |
| A-68[166]67[166] | TTTTATCTAAAGCATAGATAATTTTT | 1 | 25 nmol |
| A-68[69]68[43] | TAGAGGATCCCCGGGTACCGAGCTTTT | 1 | 25 nmol |
| A-68[81]30[54] | GCAGGAAGTACGGCTGAATCCTTTTGAACCCACAAGAAACAGGTCAGGATTATTTT | 1 | 25 nmol |
| A-69[116]33[125] | ACACCGCCTGCCCTCAAATCCTGA | 1 | 25 nmol |
| A-69[137]28[143] | CGCTGAGAGCTGCTGAAAACGAGCATAAACACAGAGAGGGAGAATTTAAGAAAGGA | 1 | 25 nmol |
| A-69[43]69[69] | TTTTTCGAATTCGTAATCATGGCCGAA | 1 | 25 nmol |
| A-69[96]68[82] | AGGTGAGGCGTTGGCAAATCAACAGTTGGCCT | 1 | 25 nmol |
| A-7[112]64[109] | CATACATCTGAATTAGCAATAGTCAGAGAAGAAATATACTTCTGA | 1 | 25 nmol |
| A-7[77]27[76] | ACCGAACGGATTGCATCAAAATCGCGTT | 1 | 25 nmol |
| A-70[69]70[43] | AAATATCATAGCTGTTTCCTGTGTTTT | 1 | 25 nmol |
| A-70[92]35[82] | TTTTCCTAAAACATCGATTAGA | 1 | 25 nmol |
| A-71[43]71[69] | TTTTTGAAATTGTTATCCGCTCGTCTT | 1 | 25 nmol |
| A-72[69]72[43] | TATTAACAATTCCACACAACATATTTT | 1 | 25 nmol |
| A-72[92]37[82] | TTTTGACAATATTTTTCGCGAG | 1 | 25 nmol |
| A-73[43]74[56] | TTTTCGAGCCGGAAGCATAAAGGAAAGGACCTTGTAAAGCC | 1 | 25 nmol |
| A-74[85]73[92] | TTTTTAGAACCCTTCTCGTAAGAATACGTGGCACATTTT | 1 | 25 nmol |

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| A-75[36]76[36] | TTTTTGAGCTAACGTGCCAGCTTTTT | 1 | 25 nmol |
| A-75[49]74[36] | TCACATTTGGGGTGCCTAATGAGTTTT | 1 | 25 nmol |
| A-75[56]38[47] | AATTGCGATAGTAGTAGCATTTT | 1 | 25 nmol |
| A-76[62]40[47] | TCGGGAACAACGCGTCGGTTGTACCATTTT | 1 | 25 nmol |
| A-76[85]75[85] | TTTTCTGGCCACCCGCCACTGACAGAGATTTT | 1 | 25 nmol |
| A-77[36]76[49] | TTTTGCATTAATGAATCGGCACCTGTC | 1 | 25 nmol |
| A-78[62]42[47] | GGCGCCAAACAGCTTATATTTTAAATTTTT | 1 | 25 nmol |
| A-78[85]77[85] | TTTTATAAAAGGTTTGAGGCGGGACATTTTTT | 1 | 25 nmol |
| A-79[36]80[36] | TTTTCACCAGTGAAAGCGGTCCTTTT | 1 | 25 nmol |
| A-79[49]78[36] | GACGGGCGGGTGGTTTTTCTTTTTTTT | 1 | 25 nmol |
| A-8[142]63[139] | AAGCCAGAATGCCCTTTTAACTGAGGTTTAA | 1 | 25 nmol |
| A-8[166]29[166] | TTTTAAACAAATACAGATAGCCTTTT | 1 | 25 nmol |
| A-8[97]64[70] | TGAACGGAAGAGCAAATATCATTTGCACGTTAGAACCTACCATAGTTGGGAAGGGC | 1 | 25 nmol |
| A-80[65]84[56] | CCTGAGAGAGATTATTTTTTGACGCGCCAGCCTGGTAA | 1 | 25 nmol |
| A-80[85]79[85] | TTTTTCACACGCCGCCCTTCAACCAGTATTTT | 1 | 25 nmol |
| A-81[36]80[49] | TTTTACGCTGGCTGAAATGGTTGCAGC | 1 | 25 nmol |
| A-82[85]42[73] | TTTTCTCATGGAAATAACGAGCATTTT | 1 | 25 nmol |
| A-83[36]82[36] | TTTTCAGCAGGCAATATTACCTCAATCGTTTTGCCCTTTT | 1 | 25 nmol |
| A-83[63]83[85] | CATTGCAACAGGAAAAACGTTTT | 1 | 25 nmol |
| A-84[85]44[70] | TTTTACTCAAACTATCATAAACAGAACAAG | 1 | 25 nmol |
| A-85[36]84[36] | TTTTCCTGTTTTGATTAGTATATCCAGAACGAAAATTTTT | 1 | 25 nmol |
| A-85[56]88[40] | ATAACATTGTAGCAAATCCCTAAGAATAGCCCGAGATAGGTTTT | 1 | 25 nmol |
| A-86[85]46[73] | TTTTCATCACGCAAATGCCGGAGAAGG | 1 | 25 nmol |
| A-87[36]86[36] | TTTTGTTCCGAAATCGGCAAATACTTCTTGATGGTGTTTT | 1 | 25 nmol |
| A-87[63]87[85] | TATAATAAAAGAGTCTGTCTTTT | 1 | 25 nmol |
| A-88[89]88[63] | TTTTATCAGTGAGGCCACCGAGATCAA | 1 | 25 nmol |
| A-89[40]89[62] | TTTTGTTGAGTGTTGTTCCGCCA | 1 | 25 nmol |
| A-9[94]9[111] | GACAAGAACAGCCGCCGC | 1 | 25 nmol |
| A-90[62]90[40] | CGGTACAGTTTGGAACAAGTTTT | 1 | 25 nmol |
| A-90[89]89[69] | TTTTATTAAAGGGATTTTAGACAGGAAGAATCCT | 1 | 25 nmol |
| A-91[40]91[62] | TTTTAGTCCACTATTAAAATCAG | 1 | 25 nmol |
| A-92[100]52[93] | TGGTTGCTTTCGGTCACCGTTA | 1 | 25 nmol |
| A-92[131]24[112] | TTTTTAATGCGCCGCTACAGCCACACCAAGCCTGACACCGGAGCGCCA | 1 | 25 nmol |
| A-92[62]92[40] | TTAGAGAACGTGGACTCCATTTT | 1 | 25 nmol |
| A-92[76]25[85] | ACGTGCTCTAGGGCAATCATAAAACAGGTTCAGAATTTACCCTGACTATTA | 1 | 25 nmol |
| A-93[40]93[62] | TTTTACGTCAAAGGGCGAAAGGA | 1 | 25 nmol |
| A-94[131]54[115] | TTTTGGGAGCCCCCGATTTAAAATCGGTTAA | 1 | 25 nmol |
| A-94[62]94[40] | AGCGAAAAACCGTCTATCATTTT | 1 | 25 nmol |
| A-94[76]55[86] | AAGGAAGACCCAAAATTGTAATTTTAACCAATAACGCG | 1 | 25 nmol |
| A-94[90]92[77] | CGAACGTAGTGTAGGACGAGCACGTATA | 1 | 25 nmol |
| A-95[108]92[101] | CACTGAGCTTGGTAACCAGGCGCGTACTA | 1 | 25 nmol |
| A-95[40]54[40] | TTTTGGCGATGGCCCATTAAAATTCGCATTTT | 1 | 25 nmol |
| A-95[66]50[74] | CATCGGAAGAAGCGGGCGTTCCTCGAGCGGGAGCTAAACTCA | 1 | 25 nmol |
| A-95[84]54[93] | TTTTGGGGTCGAGGCGTGT | 1 | 25 nmol |
| A-5[231]3[253] | TCAAGAGCCAGGCGACCCATGTACCGTAACACTTTTT | 1 | 25 nmol |
| B-0[260]5[257] | TTTTCGAATAATAATTAGGATTAGCTTTT | 2 | 25 nmol |
| C-anchor-0[260]5[257] | CAACTAAGCCTTTTCGAATAATAATTAGGATTAGCTTTT | 3 | 25 nmol |
| G-9[64]10[64] | TTTTGGCTGACCTAACAAAGCTTTTT | 4 | 25 nmol |
| G-17[68]18[68] | TTTTCTAATGCAGATACTCGTTTACCAGTTTT | 4 | 25 nmol |
| G-18[94]2[78] | CTATACGTAATGAAGTTTCCATTAAACTTTT | 4 | 25 nmol |
| G-20[97]4[82] | CAGAGGGTCACCAGATTTGTATCATCTTTT | 4 | 25 nmol |
| G-21[61]22[61] | TTTTTAGCGTCCAATACTGAGCCAATTAGCGGAATCGTCATATTTT | 4 | 25 nmol |
| G-23[61]6[61] | TTTTAATATTCATTGAGCGCAGACGGTCTTTT | 4 | 25 nmol |
| G-36[80]12[64] | GCAATCATTGTTGAGATGGTTTAATTTTTTT | 4 | 25 nmol |
| G-38[80]14[68] | GTGGAGGCAAAGACGTTGGGAAGTTTT | 4 | 25 nmol |
| G-40[80]16[68] | TCAGTGCGGGATACCACATTCAATTTT | 4 | 25 nmol |
| G-5[82]0[85] | TTTTGCCTGATAAATTCCGATATATTTTT | 4 | 25 nmol |
| G-7[61]8[64] | TTTTAATCATAAGGGAGCATAGGCTTTTT | 4 | 25 nmol |
| G-89[70]20[61] | GAGAAGTTATTCAAGTAGCTAGTTTAGACTGGATTTT | 4 | 25 nmol |
| H-9[64]10[48]-FL | TTTTGGCTGACCTAACAAAGCTACCACCTCCTTACTACTCCC | 5 | 25 nmol |
| H-17[72]18[52]-FL | TTTTCTAATGCAGATACTCGTTTACCAGACCACCTCCTTACTACTCCC | 5 | 25 nmol |
| H-18[94]2[62]-FL | CTATACGTAATGAAGTTTCCATTAAACACCACCTCCTTACTACTCCC | 5 | 25 nmol |
| H-20[97]4[66]-FL | CAGAGGGTCACCAGATTTGTATCATCACCACCTCCTTACTACTCCC | 5 | 25 nmol |
| H-21[61]22[45]-FL | TTTTTAGCGTCCAATACTGAGCCAATTAGCGGAATCGTCATAACCACCTCCTTACTACTCCC | 5 | 25 nmol |
| H-23[61]6[45]-FL | TTTTAATATTCATTGAGCGCAGACGGTCACCACCTCCTTACTACTCCC | 5 | 25 nmol |
| H-36[80]12[48]-FL | GCAATCATTGTTGAGATGGTTTAATTTACCACCTCCTTACTACTCCC | 5 | 25 nmol |
| H-38[80]14[52]-FL | GTGGAGGCAAAGACGTTGGGAAGACCACCTCCTTACTACTCCC | 5 | 25 nmol |
| H-40[80]16[52]-FL | TCAGTGCGGGATACCACATTCAAACCACCTCCTTACTACTCCC | 5 | 25 nmol |
| H-5[82]0[69]-FL | TTTTGCCTGATAAATTCCGATATATACCACCTCCTTACTACTCCC | 5 | 25 nmol |
| H-7[61]8[48]-FL | TTTTAATCATAAGGGAGCATAGGCTACCACCTCCTTACTACTCCC | 5 | 25 nmol |
| H-89[70]20[45]-FL | GAGAAGTTATTCAAGTAGCTAGTTTAGACTGGAACCACCTCCTTACTACTCCC | 5 | 25 nmol |
| I-B-4[257]0[237] | TTTTGGGGTTTTGCTCAGTAAAGGATTTTTTCACG | 6 | 25 nmol |
| I-B-0[236]4[223] | TTGAAAATAGAAACAGTACAAACTACAAATAGGAGATAAGTG | 6 | 25 nmol |
| I-B-2[253]1[260] | TTTTGAGTTTCGTCACGGAACAACTAAAGGAATTGTTTT | 6 | 25 nmol |
| J-3'-0[244]1[237] | TTTTCACGTTGAAAATAGAAA | 7 | 25 nmol |
| J-3'-2[253]4[223] | TTTTGAGTTTCGTCACCAGTACAAACTACAAATAGGAGATAAGTG | 7 | 25 nmol |
| J-3'-4[257]5[244] | TTTTGGGGTTTTGCTCAGTAAAGGATT | 7 | 25 nmol |
| K-3'add | GGAACAACTAAAGGAATTGCCGGCCGGCGGC | 8 | 25 nmol |
| **Functionalized Oligonucleotides** | | | |
| cy5 label | /5Cy5/GGGAGTAGTAAGGAGGTGGT | N/A | 100 nmol, HPLC |
| texas red label | /5TexRd-XN/GGGAGTAGTAAGGAGGTGGT | N/A | 100 nmol, HPLC |
| **Aptamer Template Oligonucleotides** | | | |
| apt for SPOT, anti-sense strand | CCGGCCGGCGGCGGGAGAAGCTTCTGTGGTGGCCCTCTTTTAAGGACTTTCCCCACCCAATTGCGCCACGACGTTTC GTGCTCCCATCGCAGTCCAGAAGCTTCTCCCTATAGTGAGTCGTATTAG | N/A | 4 nmol Ultramer |
| apt for SPOT, sense strand | CTAATACGACTCACTATAGGGAGAAGCTTCTGGACTGCGATGGGAGCACGAAACGTCGTGGCGCAATTGGGTGGGGA AAGTCCTTAAAAGAGGGCCACCACAGAAGCTTCTCCCGCCGCCGGCCGG | N/A | 4 nmol Ultramer |
| free apt, anti-sense strand | CTAATACGACTCACTATAGGGAGCTTCTGGACTGCGATGGGAGCACGAAACGTCGTGGCGCAATTGGGTGGGGAAAG TCCTTAAAAGAGGGCCACCACAGAAGCTCCC | N/A | 4 nmol Ultramer |
| free apt, sense strand | GGGAGCTTCTGTGGTGGCCCTCTTTTAAGGACTTTCCCCACCCAATTGCGCCACGACGTTTCGTGCTCCCATCGCAGT CCAGAAGCTCCCTATAGTGAGTCGTATTAG | N/A | 4 nmol Ultramer |
| rigid apt for SPOT, anti-sense strand | CAACTAAGCCAGCTTCTGTGGTGGCCCTCTTTTAAGGACTTTCCCCACCCAATTGCGCCACGACGTTTCGTGCTCCCA TCGCAGTCCAGAAGCTCAATTCCTTTAGTTGTTCCTATAGTGAGTCGTATTAG | N/A | 4 nmol Ultramer |
| rigid apt, sense strand | CTAATACGACTCACTATAGGAACAACTAAA | N/A | 25 nmol |

**Table S2** – Mixtures of staples required to produce SPOTs and their derivatives or precursors, related to STAR Methods.

| Construct | Required Staple Mixes from Table S1 | Required Additional Reagents |
|---|---|---|
| Signpost origami (SPO) | 1, 2, 4, 6 | - |
| SPO with Texas red label | 1, 2, 5, 6 | Texas red labeled oligo |
| SPO with cy5 label | 1, 2, 5, 6 | cy5 labeled oligo |
| SPOT (SPO with aptamer) | 1, 2, 4, 7, 8 | Transcribed apatmer |
| SPOT with Texas red label | 1, 2, 5, 7, 8 | aptamer, Texas red label |
| SPOT with cy5 label | 1, 2, 5, 7, 8 | aptamer, cy5 label |
| SPOT with rigid aptamer attachment | 1, 3, 4, 8 | rigid aptamer |

**Table S3** – Primers used for cloning of fluorescent protein genes, related to STAR Methods.

| Description | Primer type | Sequence |
|---|---|---|
| Primers for insertion of fluorescent protein genes in pEP98-gB | forward | GGCGGGAATTCATGGTGAGCAAGGGCGAG |
| | reverse | GGCGGCTCGAGCTTGTACAGCTCGTCCATGC |
| Primers for insertion of TEV restriction site as linker into mCherry-gB in pEP98 | forward | GGCGGGAATTCCGCCGACGCCACCAGG |
| | reverse | gBGGCGGCTCGAGAACCTGTACTTCCAGTCCGCTCCGACTTCCCCCGG |
| Primers for ligation independent cloning of fluorescent proteins into pET-28, plasmid | forward | CACCACCACCACTGA |
| | reverse | AGTGGTGGTGGTGCTTACTTGTACAGCTC |
| Primers for ligation independent cloning of fluorescent proteins into pET-28, insert | forward | AGATTGGTGAGAATTTATATTTTCAGGGCCATATGGTGAGCAAGG |
| | reverse | AGTGGTGGTGGTGCTTACTTGTACAGCTC |