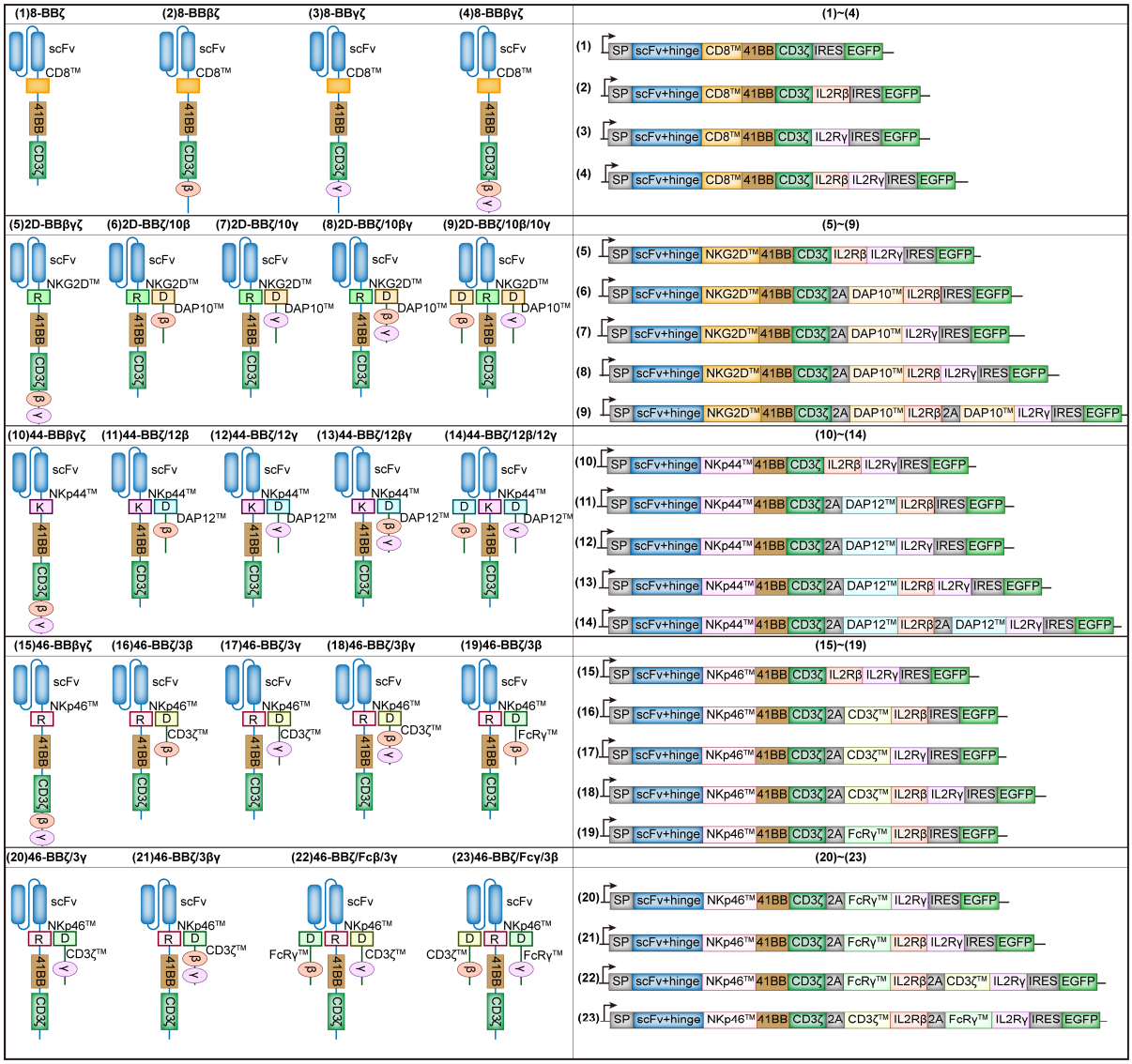
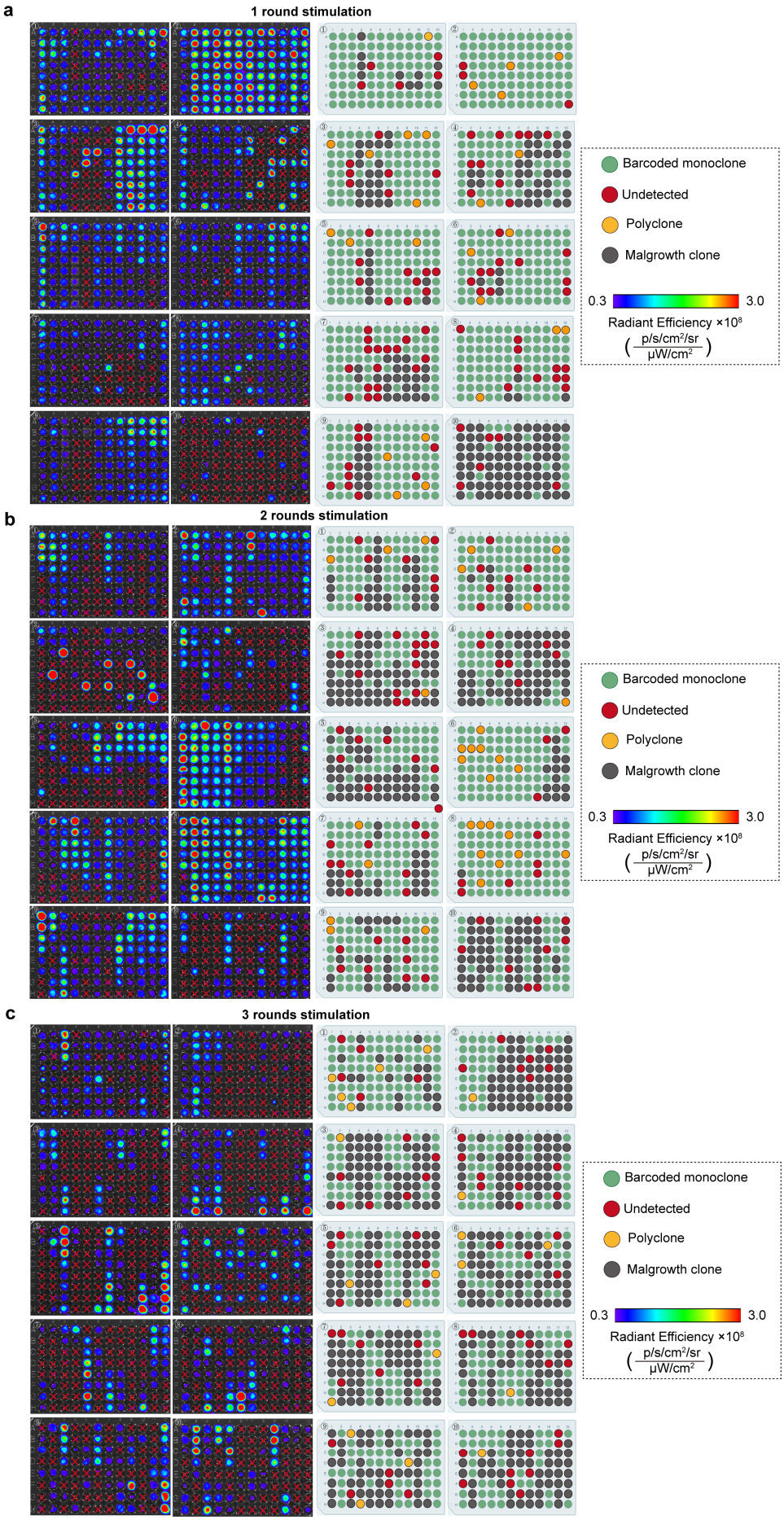
Extended figure and table

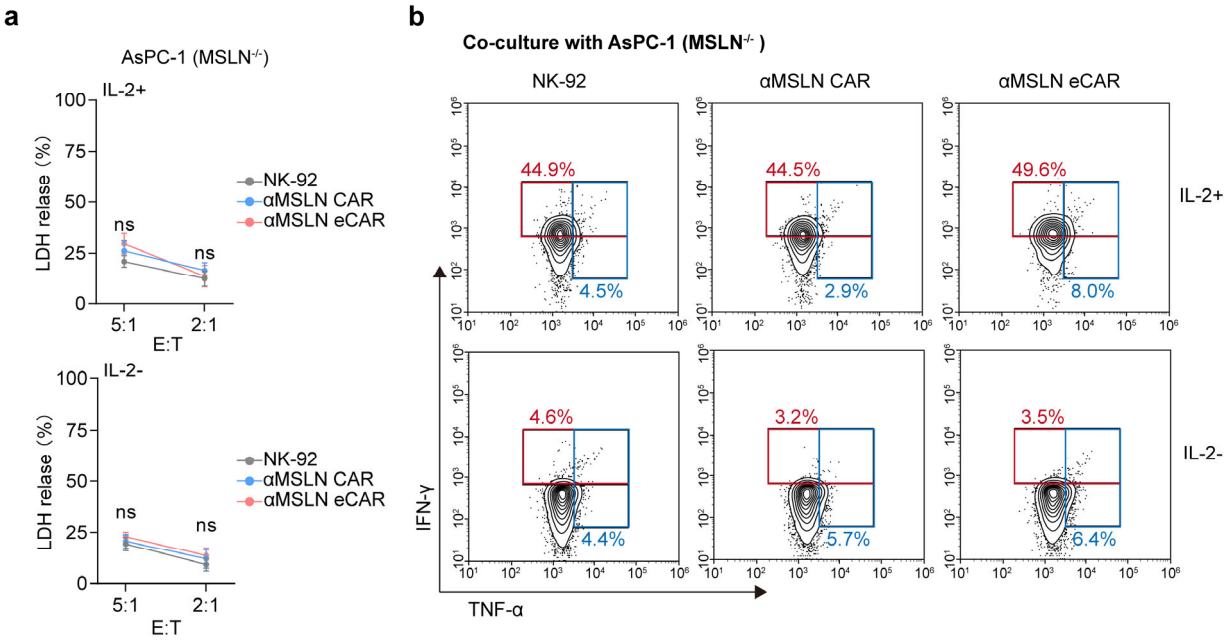


Extended Data Fig. 1 | Visualization of detailed Tandem IL2R-ICD CARs and Charge-attracting IL2R-ICD CARs.

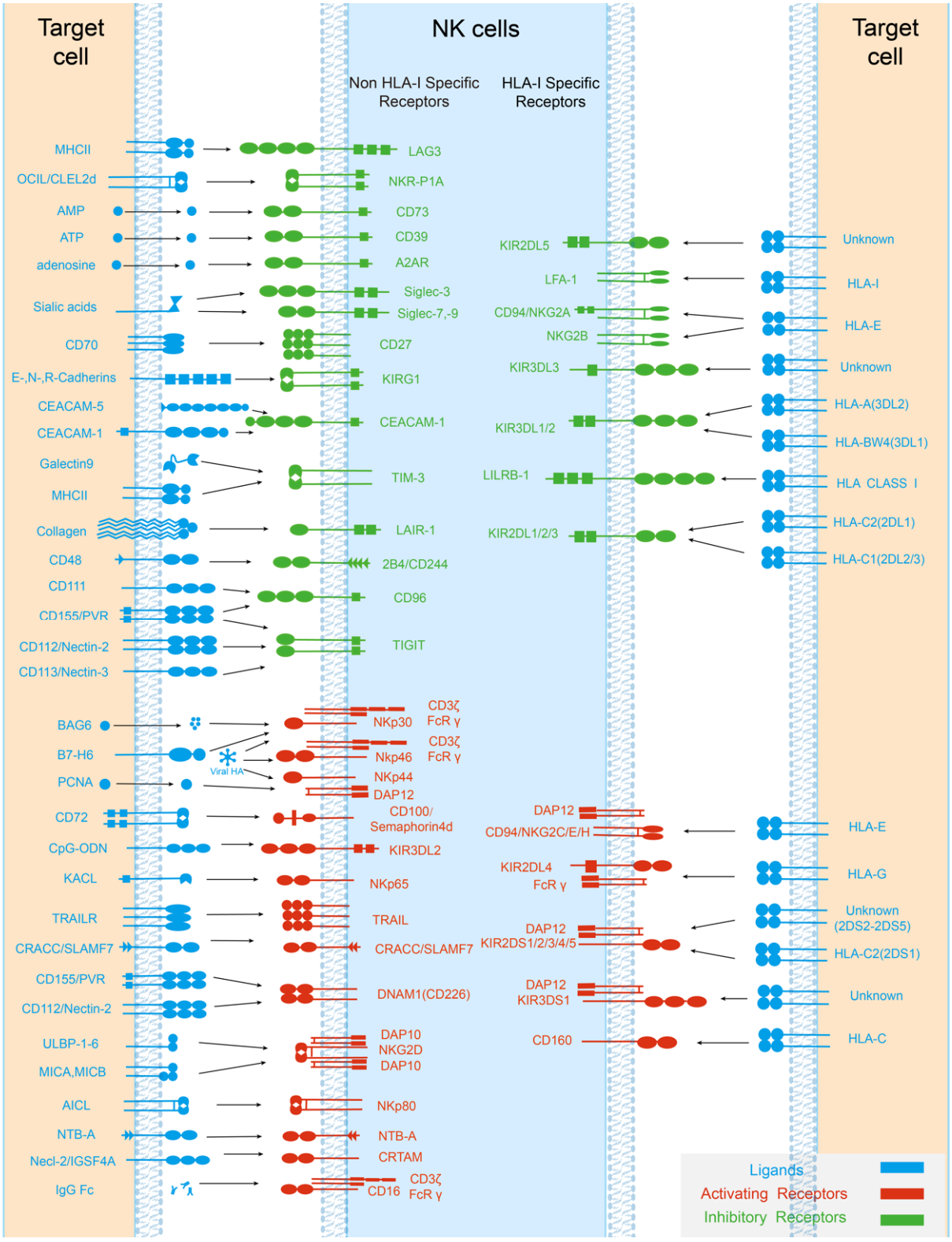
The left diagram illustrates how different chimeric antigen receptor structures oligomerize on the cell membrane. The right diagram details the genetic structures of CARs numbered 1-23. Each of these CARs corresponds to a specific barcode, which can be found in Extended Data Table 1. In brief, Anti-CD19 antibody scFv (FMC63), hinge of CD8a, transmembrane domain of CD8, NKG2D, NKp44 and NKp46; cytoplasmic signaling domain of 4-1BB, and CD3ζ were used to construct second-generation CARs; IL-2R ICDs are serially linked after CD3ζ or combined with negatively charged transmembrane regions from DAP10, DAP12, CD3ζ, or FcRγ, and then placed in parallel with second-generation CAR.


a
1 round stimulation
b
2 rounds stimulation
c
3 rounds stimulation
Barcoded monoclone
Undetected
Polyclone
Malgrowth clone
0.3
3.0
Radiant Efficiency ×10⁸
(p/s/cm²/sr / μW/cm²)

Extended Data Fig. 2 | The images of EGFP assay and Barcode analysis. a-c, The image of EGFP assay on the left of and the result of barcode analysis on the right of each picture after indicated stimulations. On the left of each picture, the radiant efficiency was detected by fluorography that indicated the expression levels of EGFP of different NK cells clones after sorting $EGFP^+$ NK cells, one cell for one well, and culturing for seven days with cytokines. On the right of each picture, the different colors indicated the result of barcodes analyzed by qPCR in correspond to each well on the right. The green label indicated that the barcodes were monoclonal in the well and the yellow label indicated polyclonal barcodes. While the red label indicated that we could not find the corresponding barcodes in our design according to the qPCR, and the black label indicated the malgrowth clone. Only the wells with high EGFP expression along with monoclonal barcode were picked up and counted in the following statistics.



Extended Data Fig. 3 | *In vitro* antitumor effect of the eCAR NK-92 cells. a, Killing activity of NK-92 (control), αMSLN CAR or αMSLN eCAR co-cultured with AsPC-1 ($MSLN^{-/-}$) (ns, not significant). Data are mean ± SD. **b,** Killing activity of NK-92 (control), αMSLN CAR or αMSLN eCAR co-cultured with AsPC-1 ($MSLN^{-/-}$). The IFN-γ and TNF-α expression levels of eCAR NK-92 cells and the control NK cells against AsPC-1 ($MSLN^{-/-}$) under the condition of $IL\text{-}2^{+}$ and $IL\text{-}2^{-}$ were assessed by flow cytometry.



Extended Data Fig. 4 | Simplified schematic of commonly expressed NK cell activating and inhibitory receptors and corresponding ligands. The green and red patterns indicate the NK cell inhibitory and activating receptors, respectively, and the blue patterns indicate the ligands on target cells. The left part indicates the non-HLA-1 specific receptors while the right part indicates the HLA-1 specific receptors.

Extended Data Table. 1 | Barcode for 1-23 CARs.

No.	Squence
1	CCGTAGGCTCGGTAAGTTCGACCAGCGTGGGCCATTGTAAGGCGCTAGGTCA GCCTACCGTGGAACCGCTGCGACCGGAAGTCCGATCGTAGGCCAAGGCCG
2	GTACGTCCATCGGCCGTGAGCTGACCGGTCGACCGGTAAGTCGACCTAGGCG TCAGCTCCGGTAGGCCGCCGTCGACCGATCGGACCTGACGCGTAGGCCG
3	CGAGCTGACCGTCGATGCCGGTAGCTGCGTCCGAGCTAGGCGTCCGTCGACC GTGCGGTCAGCGCTAGGCGGTCAGGCCGTCGACCGGATCGTCCGAGGCT
4	GTCGACCAGCGGTCGACGCTAGGCGTCGACCTAGGCGTCAGCTCCGGTAGCC TGACCGGTCGACTGAGCGTCAGCGGCTGCGAGTAGGCTGACCGGTCGGA
5	CCGGTCGATCGTCCGAGGCTAGGCGTCAGCCCGGTCAGCTCGGATCGTAGGC GTCAGCTCCGGTAGCCTGACCGGGCTAGGCGTCAGCTGACCGATCGGAC
6	GTCGACCGTAGCCTGACCGGTCAGCGCTAGGCGTCAGGCCGTCGACCGGATC GTCCGAGGCTAGGCGTCCGTCGACCGTGCGGTCAGCCCGGTCGACGCTA
7	GACCGGTAGCTGCGTCCGAGCTAGGCGTCCGTCGACCGTGCGGCTGCGAGTA GGCTGACCGGTCGACCCGGTAGCCTGACCGGTCGATCGTAGGCGTCGAC
8	GTAGGCGTCAGCCCGGTCAGCTCGGATCGTAGGCGTCAGCTCCGGTAGCCTG ACCGGTCGACTGAGCGTCAGCGGCTGCGAGTAGGCTGACCGGTCGACTG
9	GTCGGATCGTAGGCGTCAGCTCCGGTAGCCTGACCGGTCGACTGAGCGTCAG CGGCTGCGAGTAGGCTGACCGGTCGACCCGGTAGCCTGACCGGTCGATC
10	CGTAGGCGTCAGCTCCGGTAGCCTGACCGGTCGACTGAGCGTCAGCGGCTGC GAGTAGGCTGACCGGTCGACCCGGTAGCCTGACCGGTCGATCGTAGGCG
11	GTCAGCTCCGGTAGCCTGACCGGTCGACTGAGCGTCAGCGGCTGCGAGTAGG CTGACCGGTCGACCCGGTAGCCTGACCGGTCGATCGTAGGCGTCAGCCC
12	GGTAGCCTGACCGGTCGACTGAGCGTCAGCGGCTGCGAGTAGGCTGACCGGT CGACCCGGTAGCCTGACCGGTCGATCGTAGGCGTCAGCTCCGGTAGCCT
13	GACCGGTCGACTGAGCGTCAGCGGCTGCGAGTAGGCTGACCGGTCGACCCG GTAGCCTGACCGGTCGATCGTAGGCGTCAGCTCCGGTAGCCTGACCGGTC
14	CGACTGAGCGTCAGCGGCTGCGAGTAGGCTGACCGGTCGACCCGGTAGCCTG ACCGGTCGATCGTAGGCGTCAGCTCCGGTAGCCTGACCGGTCGACTGAG
15	CGTCAGCGGCTGCGAGTAGGCTGACCGGTCGACCCGGTAGCCTGACCGGTCG ATCGTAGGCGTCAGCTCCGGTAGCCTGACCGGTCGACTGAGCGTCAGCG
16	GCTGCGAGTAGGCTGACCGGTCGACCCGGTAGCCTGACCGGTCGATCGTAGG CGTCAGCTCCGGTAGCCTGACCGGTCGACTGAGCGTCAGCGGCTGCGAG
17	TAGGCTGACCGGTCGACCCGGTAGCCTGACCGGTCGATCGTAGGCGTCAGCT CCGGTAGCCTGACCGGTCGACTGAGCGTCAGCGGCTGCGAGTAGGCTGA
18	CCGGTCGACCCGGTAGCCTGACCGGTCGATCGTAGGCGTCAGCTCCGGTAGC CTGACCGGTCGACTGAGCGTCAGCGGCTGCGAGTAGGCTGACCGGTCGA
19	CCCGGTAGCCTGACCGGTCGATCGTAGGCGTCAGCTCCGGTAGCCTGACCGG TCGACTGAGCGTCAGCGGCTGCGAGTAGGCTGACCGGTCGACCCGGTAG
20	CCTGACCGGTCGATCGTAGGCGTCAGCTCCGGTAGCCTGACCGGTCGACTGA GCGTCAGCGGCTGCGAGTAGGCTGACCGGTCGACCCGGTAGCCTGACCG
21	GTCGATCGTAGGCGTCAGCTCCGGTAGCCTGACCGGTCGACTGAGCGTCAGC GGCTGCGAGTAGGCTGACCGGTCGACCCGGTAGCCTGACCGGTCGATCG
22	TAGGCGTCAGCTCCGGTAGCCTGACCGGTCGACTGAGCGTCAGCGGCTGCGA GTAGGCTGACCGGTCGACCCGGTAGCCTGACCGGTCGATCGTAGGCGTC
23	AGCTCCGGTAGCCTGACCGGTCGACTGAGCGTCAGCGGCTGCGAGTAGGCTG ACCGGTCGACCCGGTAGCCTGACCGGTCGATCGTAGGCGTCAGCTCCGG