

## Supplementary Figures and Data

**Table S1. Potency of BFP receptor binding and redirected T cell killing of tumor cells**

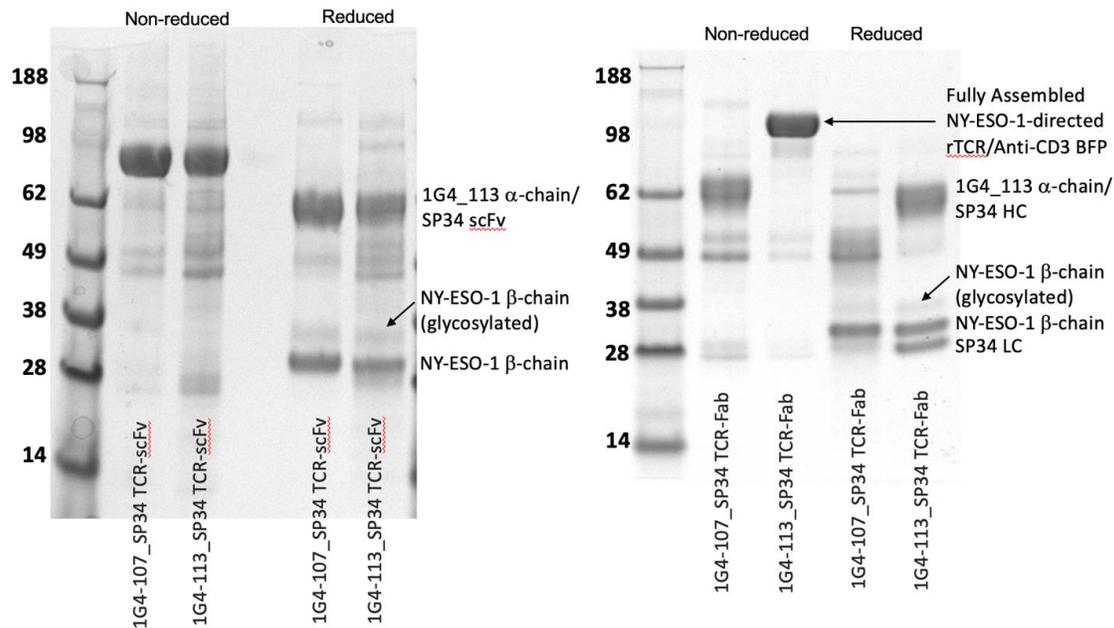
<b>BFP</b>	<b>CD3 binding IC<sub>50</sub> of SP34-PE (nM)</b>	<b>HLA binding EC<sub>50</sub> (nM)</b>	<b>Tumor cell killing with non-activated T cells and exogenous antigen peptide EC<sub>50</sub> (nM)<sup>a</sup></b>	<b>Tumor cell killing with pre-activated T cells and endogenous antigen peptide EC<sub>50</sub> (nM)<sup>b</sup></b>
NY-ESO-1 TCR-IgG	17	10	Inactive	n.d. <sup>c</sup> , Inactive
NY-ESO-1 TCR-Fab	130	1.4	0.1 (90%)	n.d., 20 (90%)
NY-ESO-1 TCR-scFv	230±30 <sup>d</sup>	2.5	7.0 (75%)	n.d., 200 (50%)
MAGE-A3 TCR-IgG	5.7	13	Inactive	Inactive, 0.6 (60%)
MAGE-A3 TCR-Fab	130	15	0.7 (100%)	0.7 (95%), 0.6 (100%)
MAGE-A3 TCR-scFv	110	14	2.1 (100%)	2.0 (95%), 1.0 (100%)

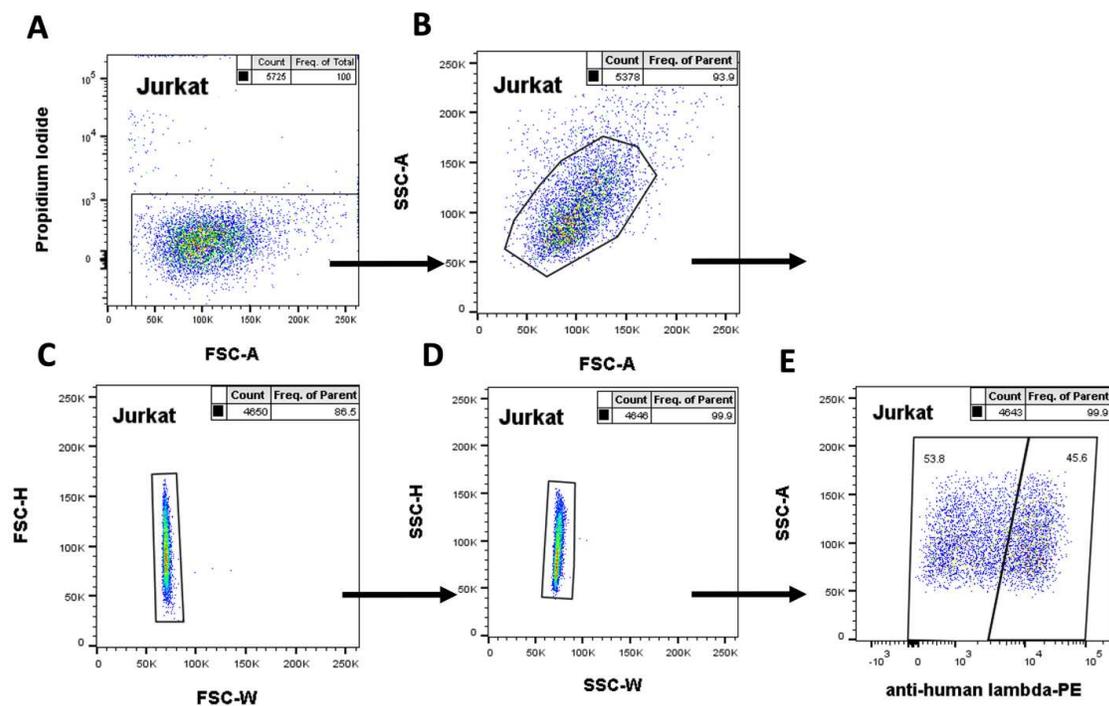
<sup>a</sup>NY-ESO-1<sub>157-165</sub> SLLMWITQC or MAGE-A3<sub>168-176</sub> EVDPIGHLY peptide was pre-incubated for 2 hours on Saos-2 or HCT116 cells, respectively, prior to the addition of T cells and BFP.

<sup>b</sup>T cells preactivated with anti-CD3/anti-CD28 and cultured with IL-2 were added with the BFPs onto HCT116 (left number) or A375 (right number) tumor cells. Numbers in parentheses indicates the percent tumor cell killing.

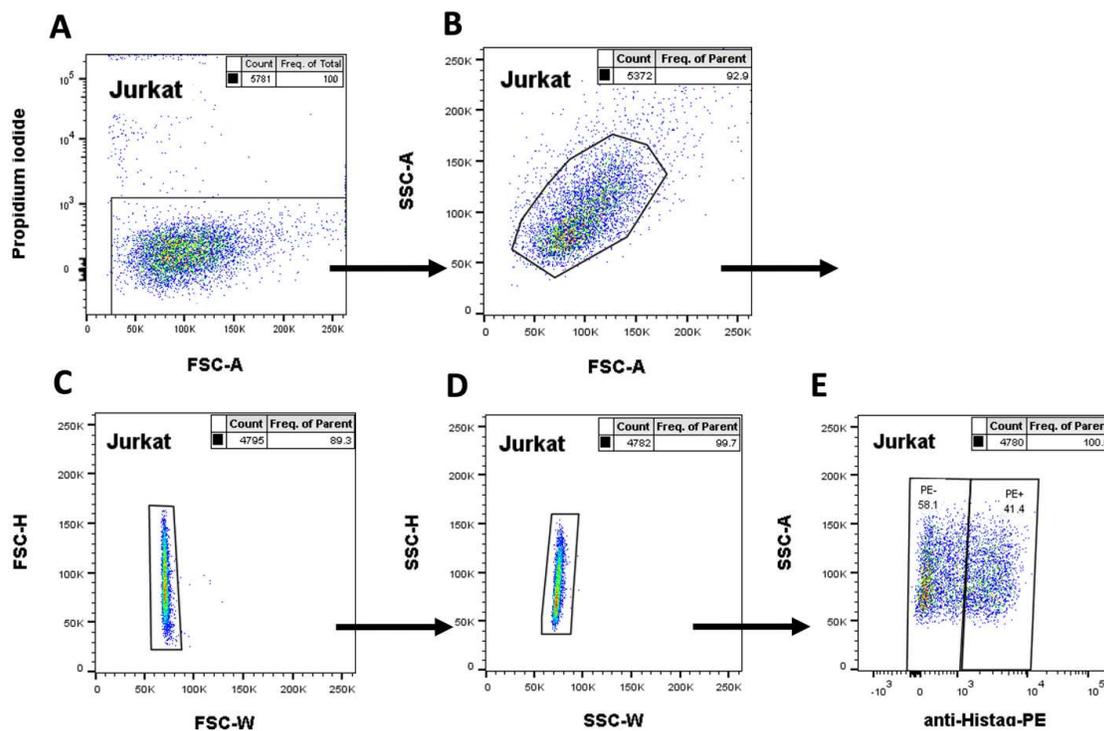
<sup>c</sup>n.d. = not done

<sup>d</sup>Average of 2 separate experiments

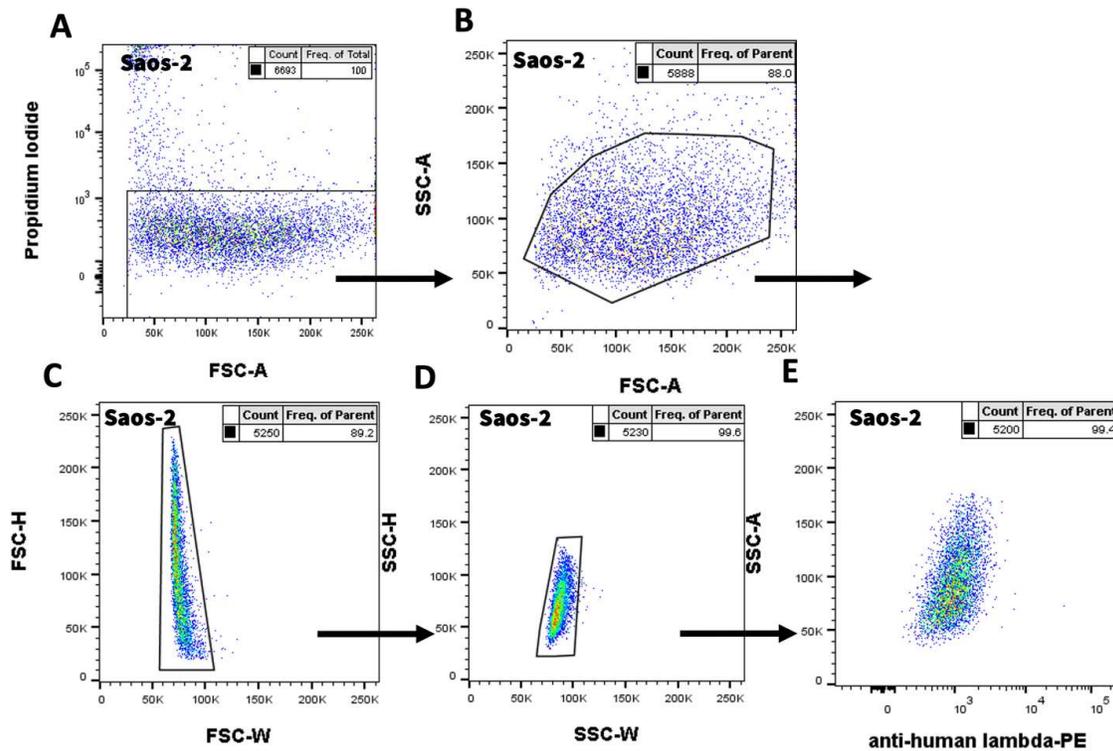




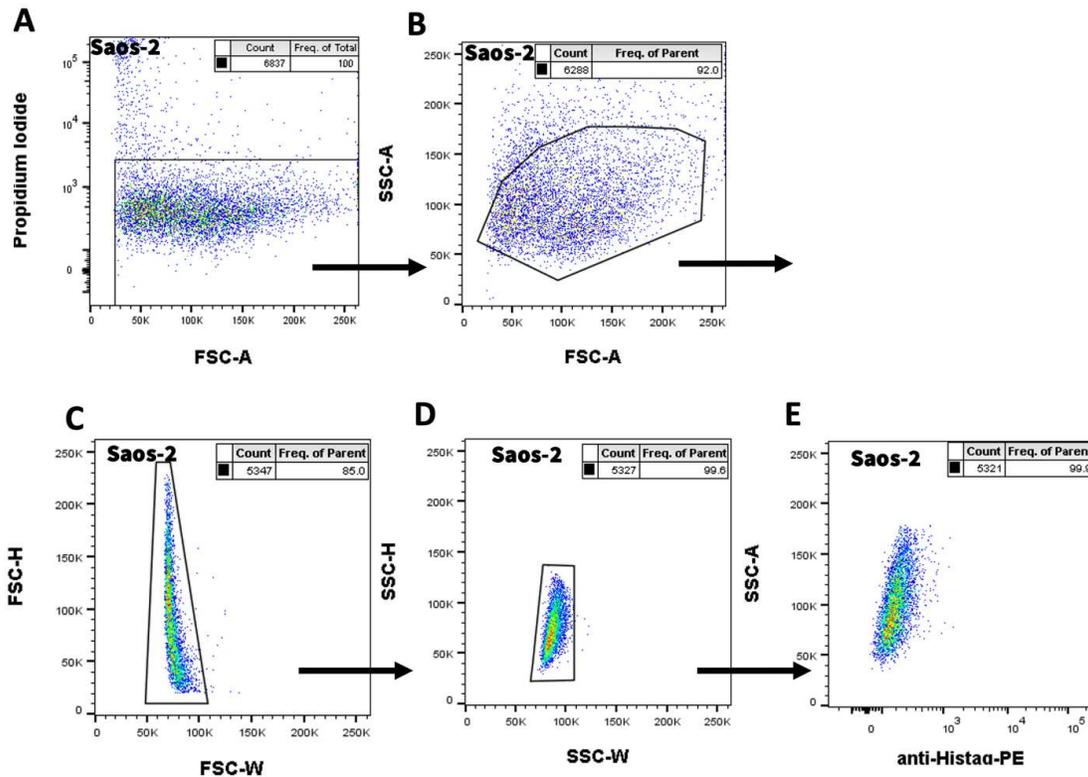
**Figure S2. Gating Strategy for SP34-PE, TCR-IgG and TCR-Fab binding to CD3+ Jurkat cells.** Jurkat cells were centrifuged and incubated for 45 minutes with rTCR/anti-CD3 BFPs. After washing, cells were stained with anti-human lambda-PE and incubated for 45 minutes. Propidium iodide was used as the live/dead cell discriminator. Acquisition was performed on a Becton Dickinson Fortessa using BD FACSDiva software v8.0.1. Analysis was performed using FlowJo v10.7.1. Cells were initially gated based on A: FSC-A vs Propidium Iodide to exclude dead cells. B: FSC-A vs SSC-A to identify cells based on light scattering. C and D: FSC-W vs FSC-H and SSC-W vs SSC-H for doublet discrimination. E: anti-human lambda-PE vs SSC-A was used to detect binding of the BFPs positivity while SP34-PE was directly labeled. Example shown uses the NY-ESO-1-directed TCR-IgG.



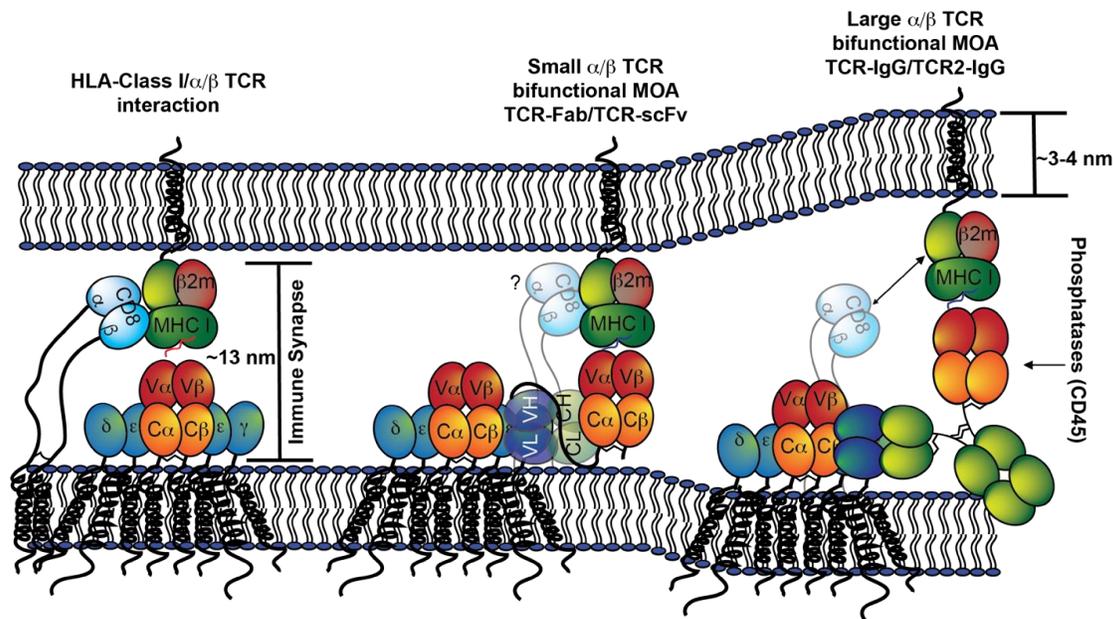
**Figure S3. Gating Strategy for TCR-scFv binding to CD3+ Jurkat cells.** Jurkat cells were centrifuged and incubated for 45 minutes with TCR BsAbs. After washing, cells were stained with anti-Histag-PE and incubated for 45 minutes. Propidium iodide was used as the live/dead cell discriminator. Acquisition was performed on a Becton Dickinson Fortessa using BD FACSDiva software v8.0.1. Analysis was performed using FlowJo v10.7.1. Cells were initially gated based on A: FSC-A vs Propidium Iodide to exclude dead cells. B: FSC-A vs SSC-A to identify the distribution of cells based on light scatter. C and D: FSC-W vs FSC-H and SSC-W vs SSC-H for doublet discrimination. E: anti-Histag-PE vs SSC-A for determination of positivity. Example shown uses the NY-ESO-1-directed TCR-scFv.



**Figure S4. Gating Strategy for TCR-IgG and TCR-Fab binding to HLA-A2+ Saos-2 cells preloaded with NY-ESO-1157-165 SLLMWITQC peptide.** Saos-2 cells were pulsed with peptide for 3 hrs., centrifuged and incubated for 45 minutes with TCR BsAbs. After washing, cells were stained with anti-human lambda-PE and incubated for 45 minutes. Propidium iodide was used as the live/dead discriminator. Acquisition was performed on a Becton Dickinson Fortessa using BD diva software v8.0.1. Analysis was performed using FlowJo v10.7.1. Cells were initially gated based on A: FSC-A vs Propidium Iodide to exclude dead cells. B: FSC-A vs SSC-A to identify the distribution of cells based on light scatter. C and D: FSC-H vs FSC-W and SSC-H vs SSC-W for doublet discrimination. E: anti-human lambda-PE vs SSC-A for determination of positivity for TCR-IgG and TCR-Fab BFPs.



**Figure S5. Gating Strategy for TCR-scFv binding to HLA-A2+ Saos-2 cells preloaded with NY-ESO-1157-165 SLLMWITQC peptide.** Saos2 cells were pulsed with peptide for 3 hrs., centrifuged and incubated for 45 minutes with TCR BsAbs. After washing, cells were stained with anti-Histag-PE and incubated for 45 minutes. Propidium iodide was used as the live/dead discriminator. Acquisition was performed on a Becton Dickinson Fortessa using BD diva software v8.0.1. Analysis was performed using FlowJo v10.7.1. Cells were initially gated based on A: FSC-A vs Propidium Iodide to exclude dead cells. B: FSC-A vs SSC-A to identify the distribution of cells based on light scatter. C and D: FSC-H vs FSC-W and SSC-H vs SSC-W for doublet discrimination. E: anti-Histag-PE vs SSC-A for determination of positivity.



**Figure S6. Schematic diagram depicting hypothetical challenges for the use of large and/or complex rTCR/Anti-CD3 BFPs.** The natural TCR/MHC/CD8 complex formation is depicted on the left side of the diagram including the synaptic distance bridged by the TCR/MHC-peptide interaction as well as the CD8-coreceptor interaction. The rTCR/Anti-CD3 TCR-Fab and TCR-scFv molecules are depicted in the center of the diagram enabling similarly tight synapse formation, while the larger rTCR/Anti-CD3 BFPs exemplified by the TCR-IgG on the right-side of the diagram do not. The potential challenge of co-localizing the TCR-coreceptor CD8 is clear based on the diagram if the synaptic distance is too large.

**Recombinant T cell Receptor/Anti-CD3 Bifunctional Protein Sequences**

**>MAGE-A3 TCR-scFv**

KQEVTVQIPAAALSVPEGENLVLNCSFTDSAIYNLQWFRQDPGKGLTSLLYVRPYQREQTSGRNLASLDKSSGRSTLYIAASQPGDSA  
TYLCAVRPGGAGPFFVVFVFGKGTKLSVIPIYIQNPDPVAVYQLRDSKSSDKFVCLFTDFDSQINVSQSKSDSVYITDKC  
VLDMRSMDFK  
SNSAVAWSNKSDFTCANAFNNSIIPEDTFFPSPESCGGGGSGGGGSGGGGSEVQLVESGGGLVQPKGSLKLSAASGFTFNTYAM  
NWVRQAPGKGLEWVARIRSKYNNYATYYADSVKDRFTISRDDSQSLLYLQMNKLTEDTAMYCVRHGNFGNSYVSWFAYWGQGLT  
VTVSAGGGGSGGGGSGGGGSGGGGSAVVTQESALTTSPGETVTLTCSRSTGAVTTSNYANWVQEKPDHLFTGLIGGTTNKRAPGVP  
ARFSGSLIGDKAALITGAQTEDEAIYFCALWYSNLWVFGGGTKLTVLGS DYKDDDDKLEHHHHHHHH

GVTQTPRYLIKTRGQVTLTSCSPISGHRVSVWYQQTPGQGLQFLFEYFSETQRNKGNFGRFSGRQFSNSRSEMNVSTLELGD  
SALYLCASSFNMATGQYFGPGTRLTVTEDLKNVFPPEVAVFEPSEKAEISRTQKATLVCLATGFYPHVELS  
WVWNGKEVHDGVCCTDPQP  
LKEQPALNDSRYALSSRLRVSATFWQDPRNHFRQCQVQFYGLSENDEWTQDRAKPVTVIVSAEAWGRADC

**>MAGE-A3 TCR-Fab**

HHHHHHHSGKQEVTVQIPAAALSVPEGENLVLNCSFTDSAIYNLQWFRQDPGKGLTSLLYVRPYQREQTSGRNLASLDKSSGRSTLY  
IAASQPGDSATYLCAVRPGGAGPFFVVFVFGKGTKLSVIPIYIQNPDPVAVYQLRDSKSSDKFVCLFTDFDSQINVSQSKSDSVYITDKC  
VLDMRSMDFK  
SNSAVAWSNKSDFTCANAFNNSIIPEDTFFPSPESCGGGGSGGGGSGGGGSEVQLVESGGGLVQPKGSLK  
LSCAASGFTFNTYAMNWVRQAPGKGLEWVARIRSKYNNYATYYADSVKDRFTISRDDSQSLLYLQMNKLTEDTAMYCVRHGNFG  
NSYVSWFAYWGQGLTVTVSAASTKGPSVFLAPCSRSTSESTAALGCLVKDYFPEPVTVSWNSGALTS  
SGVHTFFPAVLQSSGLYSLS  
SVVTVPSSSLGTQTYTCNVDHKPSNTKVDKRVESKYG

GVTQTPRYLIKTRGQVTLTSCSPISGHRVSVWYQQTPGQGLQFLFEYFSETQRNKGNFGRFSGRQFSNSRSEMNVSTLELGD  
SALYLCASSFNMATGQYFGPGTRLTVTEDLKNVFPPEVAVFEPSEKAEISRTQKATLVCLATGFYPHVELS  
WVWNGKEVHDGVCCTDPQP  
LKEQPALNDSRYALSSRLRVSATFWQDPRNHFRQCQVQFYGLSENDEWTQDRAKPVTVIVSAEAWGRADC

QAVVTQESALTTSPGETVTLTCSRSTGAVTTSNYANWVQEKPDHLFTGLIGGTTNKRAPGVPARFSGSLIGDKAALITGAQTEDEA  
IYFCALWYSNLWVFGGGTKLTVLQPKAAPSVTLFPPSSEELQANKATLVCLISDFYPGAVTVAWKADSSPVKAGVETTT  
PSKQSN  
NKYAASSYLSLTPQWKS  
HRYSYSCQVTHEGSTVEKTVAPTEC

**>MAGE-A3 TCR-IgG**

KQEVTVQIPAAALSVPEGENLVLNCSFTDSAIYNLQWFRQDPGKGLTSLLYVRPYQREQTSGRNLASLDKSSGRSTLYIAASQPGDSA  
TYLCAVRPGGAGPFFVVFVFGKGTKLSVIPIYIQNPDPVAVYQLRDSKSSDKFVCLFTDFDSQINVSQSKSDSVYITDKC  
VLDMRSMDFK  
SNSAVAWSNKSDFTCANAFNNSIIPEDTFFPSPPEPKSCDKTHTCPPCPAPEAAGGPSVFLFPPKPKDTLMI  
SRTPEVTCVVVDVSHEDPEVKFNWYVDGVEVHNAKTKPREEQYQSTYRVVSVLTVLHQD  
WLNQKEYKCKVSNKALPAPIEKTISKAKGQPREPQVYTLPPS  
REEMTQINQVSLTCLVKGFYPSDIAVEWESNGQPENNYKTTTPVPLVSDGSFFLAKSLTVDKSRWQQGNV  
FSCSVMEALHNNHYTQKSLSPGK

GVTQTPRYLIKTRGQVTLTSCSPISGHRVSVWYQQTPGQGLQFLFEYFSETQRNKGNFGRFSGRQFSNSRSEMNVSTLELGD  
SALYLCASSFNMATGQYFGPGTRLTVTEDLKNVFPPEVAVFEPSEKAEISRTQKATLVCLATGFYPHVELS  
WVWNGKEVHDGVCCTDPQP  
LKEQPALNDSRYALSSRLRVSATFWQDPRNHFRQCQVQFYGLSENDEWTQDRAKPVTVIVSAEAWGRADC

EVQLVESGGGLVQPKGSLKLSAASGFTFNTYAMNWVRQAPGKGLEWVARIRSKYNNYATYYADSVKDRFTISRDDSQSLLYLQMN  
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SGVHTFFPAVLQSSGLYSLSAVVTVPS  
SSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPAPEAAGGPSVFLFPPKPKDT  
LMI  
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PSKQSN  
NKYAASSYLSLTPQWKS  
HRYSYSCQVTHEGSTVEKTVAPTEC

**>NY-ESO-1 TCR-scFv**

QEVTVQIPAAALSVPEGENLVLNCSFTDSAIYNLQWFRQDPGKGLTSLLLITPWQREQTSGRNLASLDKSSGRSTLYIAASQPGDSAT  
YLCAVRPLLDGTYIPTFGRGTS  
LIVHPYIQNPDPVAVYQLRDSKSSDKFVCLFTDFDSQINVSQSKSDSVYITDKC  
VLDMRSMDFK  
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WVRQAPGKGLEWVARIRSKYNNYATYYADSVKDRFTISRDDSQSLLYLQMNKLTEDTAMYCVRHGNFGNSYVSWFAYWGQGLTV  
TVSAGGGGSGGGGSGGGGSGGGGSAVVTQESALTTSPGETVTLTCSRSTGAVTTSNYANWVQEKPDHLFTGLIGGTTNKRAPGVP  
ARFSGSLIGDKAALITGAQTEDEAIYFCALWYSNLWVFGGGTKLTVLGS DYKDDDDKLEHHHHHHHH

GVTQTPKFQVLKGTGQSMTLQCAQDMNHEYMSWYRQDPGMGLRLIHYSVAIQTTDRGEVPNGYNVSRSTIEDFPLRLLSAAAPSQTSV  
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>NY-ESO-1 TCR-Fab

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LDMRSMDFKNSAVAWSNKSDFTCANAFNNSIIPEDTFFPSPPESSCGGGGGGGGGGGGGGGGGGGSEVQLVESGGGLVQPKGSLKL  
SCAASGFTFNTYAMNWVRQAPGKGLEWVARIRSKYNNYATYYADSVKDRFTISRDDSQSLLYLQMNLLKTEDTAMYYCVRHGNFGN  
SYVSWFAYWGQGTTLVTVSAASTKGPSVFLAPCSRSTSESTAALGCLVKDYFPEPVTVSWNSGALTSVHTFFPAVLQSSGLYSLSS  
VVTVPSSSLGTKTYTCNVDHKPSNTKVDKRVESKYG

GVTQTPKFQVLKGTGQSMTLQCAQDMNHEYMSWYRQDPGMGLRLIHYSVAIQTTDRGEVPNGYNVSRSTIEDFPLRLLSAAAPSQTSV  
YFCASSYLNGTGEFFGEGSRLTVLEDLKNVFPPEVAVFEPSPKAEISRTQKATLVCLATGFYPPHVELSWSWVNGKEVHDGVCITDPO  
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>NY-ESO-1 TCR-IgG

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SLSFGK

EVQLVESGGGLVQPKGSLKLSCAASGFTFNTYAMNWVRQAPGKGLEWVARIRSKYNNYATYYADSVKDRFTISRDDSQSLLYLQMN  
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PQVYTLPPSR EEMTKNQVSLTCLVKGFYPSDIAVEWESNGQPENNYKTPPVLDSDGSFFFLYSLTVDKSRWQQGNVFCSCVMHEALHNHYTQKSL  
SLSFGK

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PLKEQPALNDSRYALSRRLRVSATFWQDPRNHFRQCQVQFYGLSENDEWTQDRAKPVVTQIVSAEAWGRADC

QAVVTQESALTTSPGETVTLTCSRSTGAVTTSNYANWVQEKPDHLFTGLIGGTNKRAPGVPARFSGSLIGDKAALTTGAQTEDEA  
IYFCALWYNSLWVFGGKTLTVLQPKAAPSVTLFPPSSEELQANKATLVCLISDFYPGAVTVAWKADSSPVKAGVETTTPSKQSN  
NKYAASSYLSLTPEQWKSHRSYSCQVTHEGSTVEKTVAPTEC

>NY-ESO-1/SP34 TCR-Fab-Fc

QEVQIIPAALSVPEGENLVLNCSTDSAIYNLQWFRQDPGKGLTSLLLISPWQREQTSGRNLNASLDKSSGRSTLYIAASQPGDSAT  
YLC AVRPLLDGTIYPTFGRGTS LIVHPYIQNPDPVAVYQLRDSKSSDKFVCLFTDFDSQINVSQSKDSVYITDKCVLDMRSMDFKNS  
NSAVAWSNKSDFTCANAFNNSIIPEDTFFPSPPESSCGGGGGGGGGGGGGGGGGGGSEVQLVESGGGLVQPKGSLKLSCAASGFTFN  
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QGTTLVTVSAASTKGPSVFLAPSSKSTSGGTAALGCLVADYFPEPVTVSWNSGALTSVHTFFPAVLQSSGLYSLASVVTVPSSSLG  
QTQYICNVNHKPSNTKVDKRVKPKSCDKTHTCPPCPAPEAAGGSPVFLFPPPKKDTLMI SRTPEVTCVVVDVSHEDPEVKFNWYVD  
GVEVHNAKTKPREEQYSTYRVVSVLTVLHQDWLNGKEYKCKVSNKALPAPIEKTISKAKGQPREPQVYTLPPSR EEMTKNQVSL  
CLVKGFYPSDIAVEWESNGQPENNYKTPPVLDSDGSFFFLYSLTVDKSRWQQGNVFCSCVMHEALHNHYTQKSLSLSFGK

DKTHTCPPCPAPEAAGGSPVFLFPPPKKDTLMI SRTPEVTCVVVDVSHEDPEVKFNWYVDGVEVHNAKTKPREEQYSTYRVVSVL  
TVLHQDWLNGKEYKCKVSNKALPAPIEKTISKAKGQPREPQVYTLPPSR EEMTKNQVSLTCLVKGFYPSDIAVEWESNGQPENNYK  
TTPPVPLSDGSFFLASKLTVDKSRWQQGNVFCSCVMHEALHNHYTQKSLSLSFGK

GVTQTPKFQVLKGTGQSMTLQCAQDMNHEYMSWYRQDPGMGLRLIHYSVAIQTTDRGEVPNGYNVSRSTIEDFPLRLLSAAAPSQTSV  
YFCASSYLNGTGEFFGEGSRLTVLEDLKNVFPPEVAVFEPSPKAEISRTQKATLVCLATGFYPPHVELSWSWVNGKEVHDGVCITDPO  
PLKEQPALNDSRYALSRRLRVSATFWQDPRNHFRQCQVQFYGLSENDEWTQDRAKPVVTQIVSAEAWGRADC

QAVVTQESALTTSPGETVTLTCRSSTGAVTTSNYANWVQEKPDHLFTGLIGGTNKRAPGVPARFSGSLIGDKAALTTGAQTEDEA  
IYFCALWYNLWVFGGGTKLTVLQPKAAPSVTLFPPSSEELQANKATLVCLISDFYPGAVTVAWKADSSPVKAGVETTTPSKQSN  
NKYAASSYLSLTPQWKSHRYSYSCQVTHEGSTVEKTVAPTEC

>NY-ESO-1/SP34 TCR-Fab-IgG

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YLC AVRPLLDGTYIPTFGRGTS LIVHPYIQNPDPVAVYQLRDSKSSDKFVCLFTDFDSQINVSQSKSDSVYITDKC VLDMRSMDFKS  
NSAVAWSNKSDFTCANAFNNSIIPEDTFFPSPESSCGGGGSGGGSGGGSGGGSEVQLVESGGGLVQPKGSLKLSCAASGFTFN  
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QGTTLVTVSAASTKGPSVFPLAPSSKSTSGGTAALGCLVADYFPEPVTVSWNSGALTSVHTFPAVLQSSGLYSLASVTVPSSSLG  
TQTYICNVNHKPSNTKVDKRVKPKSCDKHTHTCPPCPAPEAAGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWYVD  
GVEVHNAKTKPREEQYSTYRVVSVLTVLHQDWLNGKEYKCKVSNKALPAPIEKTIISKAKGQPRSPVYITLPPSREEMTKNQVSLV  
CLVKGFIYPSDIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSLTVDKSRWQQGNVFCSCVMHEALHNHYTQKLSLSLSPGK

QEVVTQIPAAALSVPEGENLVLNCSFTDSAIYNLQWFRQDPGKGLTSLLLITPWQREQTSGRNLASLDKSSGRSTLYIAASQPGDSAT  
YLC AVRPLLDGTYIPTFGRGTS LIVHPYIQNPDPVAVYQLRDSKSSDKFVCLFTDFDSQINVSQSKSDSVYITDKC VLDMRSMDFKS  
NSAVAWSNKSDFTCANAFNNSIIPEDTFFPSPKSCDKHTHTCPPCPAPEAAGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHED  
DPEVKFNWYVDGVEVHNAKTKPREEQYSTYRVVSVLTVLHQDWLNGKEYKCKVSNKALPAPIEKTIISKAKGQPREPQVYITLPPSR  
EEMTKNQVSLTCLVKGFIYPSDIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSLTVDKSRWQQGNVFCSCVMHEALHNHYTQKLS  
LSLSPGK

GVTQTPKFQVLKGTGQSMTLQCAQDMNHEYMSWYRQDPGMGLRLIHYSVAIQTTDRGEVPNGYNVSRSTIEDFPLRLLSAAAPSQTSV  
YFCASSYLNGTGEFFGEGSRLTVLEDLKNVFPPEVAVFEPSPKAEISRITQKATLVCLATGFYPPHVELS WVVNGKEVHDGVC TDPQ  
PLKEQPALNDSRYALSRLRVSATFWQDPRNHFRQCQVQFYGLSENDEWTQDRAKPVTVQIVSAEAWGRADC

QAVVTQESALTTSPGETVTLTCRSSTGAVTTSNYANWVQEKPDHLFTGLIGGTNKRAPGVPARFSGSLIGDKAALTTGAQTEDEA  
IYFCALWYNLWVFGGGTKLTVLQPKAAPSVTLFPPSSEELQANKATLVCLISDFYPGAVTVAWKADSSPVKAGVETTTPSKQSN  
NKYAASSYLSLTPQWKSHRYSYSCQVTHEGSTVEKTVAPTEC

>2TCR.1-Fab

HHHHHHHHSQEVVTQIPAAALSVPEGENLVLNCSFTDSAIYNLQWFRQDPGKGLTSLLLISPWQREQTSGRNLASLDKSSGRSTLYI  
AASQPGDSATYLC AVRPLLDGTYIPTFGRGTS LIVHPYIQNPDPVAVYQLRDSKSSDKFVCLFTDFDSQINVSQSKSDSVYITDKC V  
LDMRSMDFKSN SAVAWSNKSDFTCANAFNNSIIPEDTFFPSPESSCGGGGSGGGSGGGSGGGSEVTVQIPAAALSVPEGENLV  
NCSFTDSAIYNLQWFRQDPGKGLTSLLLISPWQREQTSGRNLASLDKSSGRSTLYIAASQPGDSATYLC AVRPLLDGTYIPTFGRG  
TSLIVHPYIQNPDPVAVYQLRDSKSSDKFVCLFTDFDSQINVSQSKSDSVYITDKC VLDMRSMDFKSN SAVAWSNKSDFTCANAFN  
SIIIPEDTFFPSPESSCGGGGSGGGSGGGSGGGSEVQLVESGGGLVQPKGSLKLSCAASGFTFN TYAMNWRQAPGKGLEWVAR  
IRSKYNNYATYYADSVKDRFTISRDDSQSLLYLQMNLLKTEDTAMYYCVRHGNFNGNSYVSWFAYWGQGTTLVTVSAASTKGPSVFPL  
APCSRSTSESTAALGCLVKDYFPEPVTVSWNSGALTSVHTFPAVLQSSGLYSLSSVTVPSSSLGKTKYTCNVDPKPSNTKVDK  
VESKYG

QAVVTQESALTTSPGETVTLTCRSSTGAVTTSNYANWVQEKPDHLFTGLIGGTNKRAPGVPARFSGSLIGDKAALTTGAQTEDEA  
IYFCALWYNLWVFGGGTKLTVLQPKAAPSVTLFPPSSEELQANKATLVCLISDFYPGAVTVAWKADSSPVKAGVETTTPSKQSN  
NKYAASSYLSLTPQWKSHRYSYSCQVTHEGSTVEKTVAPTEC

GVTQTPKFQVLKGTGQSMTLQCAQDMNHEYMSWYRQDPGMGLRLIHYSVAIQTTDRGEVPNGYNVSRSTIEDFPLRLLSAAAPSQTSV  
YFCASSYLNGTGEFFGEGSRLTVLEDLKNVFPPEVAVFEPSPKAEISRITQKATLVCLATGFYPPHVELS WVVNGKEVHDGVC TDPQ  
PLKEQPALNDSRYALSRLRVSATFWQDPRNHFRQCQVQFYGLSENDEWTQDRAKPVTVQIVSAEAWGRADC

>2TCR.2-Fab

HHHHHHHHSQEVVTQIPAAALSVPEGENLVLNCSFTDSAIYNLQWFRQDPGKGLTSLLLISPWQREQTSGRNLASLDKSSGRSTLYI  
AASQPGDSATYLC AVRPLLDGTYIPTFGRGTS LIVHPYIQNPDPVAVYQLRDSKSSDKFVCLFTDFDSQINVSQSKSDSVYITDKC V  
LDMRSMDFKSN SAVAWSNKSDFTCANAFNNSIIPEDTFFPSPESSCGGGGSGGGSGGGSGGGSEVQLVESGGGLVQPKGSLK  
SCAASGFTFN TYAMNWRQAPGKGLEWVARIRSKYNNYATYYADSVKDRFTISRDDSQSLLYLQMNLLKTEDTAMYYCVRHGNFNG  
SYVSWFAYWGQGTTLVTVSAASTKGPSVFPLAPCSRSTSESTAALGCLVKDYFPEPVTVSWNSGALTSVHTFPAVLQSSGLYSLSS  
VTVPSSSLGKTKYTCNVDPKPSNTKVDKRVESKYGGGGSGGGSGGGSGGGSEVTVQIPAAALSVPEGENLVLNCSFTDSAIY  
NLQWFRQDPGKGLTSLLLISPWQREQTSGRNLASLDKSSGRSTLYIAASQPGDSATYLC AVRPLLDGTYIPTFGRGTS LIVHPYIQ  
NPDPVAVYQLRDSKSSDKFVCLFVCLFTDFDSQINVSQSKSDSVYITDKC VLDMRSMDFKSN SAVAWSNKSDFTCANAFNNSIIPED  
TFFPSPESSC

QAVVTQESALTTSPGETVTLTCRSSTGAVTTSNYANWVQEKPDHLFTGLIGGTNKRAPGVPARFSGSLIGDKAALTTGAQTEDEA  
IYFCALWYNLWVFGGGTKLTVLQPKAAPSVTLFPPSSEELQANKATLVCLISDFYPGAVTVAWKADSSPVKAGVETTTPSKQSN  
NKYAASSYLSLTPQWKSHRYSYSCQVTHEGSTVEKTVAPTEC

GVTQTPKFQVLKGTGQSMTLQCAQDMNHEYMSWYRQDPGMGLRLIHYSVAIQTTDRGEVPPNGYNVSRSTIEDFPLRLLSAAPSQTSV  
YFCASSYLNGTGEFFGEGSRLTVLEDLKNVFPPEVAVFEPSEKAEISRTQKATLVCLATGFYPPHVELSWSWVNGKEVHDGVCITDPO  
PLKEQPALNDSRYALSRRLRVSATFWQDPRNHFRQCQVQFYGLSENDEWTQDRAKPVVTQIVSAEAWGRADC

### >2TCR.3-Fab

HHHHHHHGGSEVQLVESGGGLVQPKGSLKLSAASGFTFNNTYAMNWRQAPGKGLEWVARIRSKYNNYATYYADSVKDRFTISRDD  
SQSLLYLQMNLLKTEDTAMYCVRHGNFGNSYVSWFAYWQGTLVTVSAASTKGPSVFLAPCSRSTSESTAALGCLVKDYFPEPV  
TVSWNSGALTSGVHTFPAVLQSSGLYSLSSVTVPSSSLGKTYTCNVNPKSNTKVDKRVESKYGGGGGGGGGGGGGGGGGGGGGGGG  
QEVTVIQAALSVPEGENLVNCSFTDSAIYNLQWFRQDPGKGLTSLLLISPWQREQTSGRNLASLDKSSGRSTLYIAASQPGDSAT  
YLC AVRPLLDGTYIPTFGRGTS LIVHPYIQNPDPVAVYQLRDSKSSDKFVCLFTDFDSQINVSQSKSDSVYITDKCVLDMRSMDFKS  
NSAVAWSNKSDFTCANAFNNSIIPEDTFFPSPESPSCGG  
NLQWFRQDPGKGLTSLLLISPWQREQTSGRNLASLDKSSGRSTLYIAASQPGDSATYLC AVRPLLDGTYIPTFGRGTS LIVHPYIQ  
NPDPVAVYQLRDSKSSDKFVCLFTDFDSQINVSQSKSDSVYITDKCVLDMRSMDFKSN SAVAWSNKSDFTCANAFNNSIIPEDTFFP  
SPESPSC

QAVVTQESALTTSPGETVTLTCRSTGAVTTSNYANWVQEKPDHLFTGLIGGNTKRAPGVPARFSGSLIGDKAALITGAQTEDEA  
IYFCALWYNSLWVFGGKTLTVLQPKAAPSVTLPFSSSEELQANKATLVCLISDFYPGAVTVAWKADSSPVKAGVETTTPSKQSN  
NKYAASSYLSLTPQWKS HRYSYSCQVTHEGSTVEKTVAPTEC

GVTQTPKFQVLKGTGQSMTLQCAQDMNHEYMSWYRQDPGMGLRLIHYSVAIQTTDRGEVPPNGYNVSRSTIEDFPLRLLSAAPSQTSV  
YFCASSYLNGTGEFFGEGSRLTVLEDLKNVFPPEVAVFEPSEKAEISRTQKATLVCLATGFYPPHVELSWSWVNGKEVHDGVCITDPO  
PLKEQPALNDSRYALSRRLRVSATFWQDPRNHFRQCQVQFYGLSENDEWTQDRAKPVVTQIVSAEAWGRADC

### >2TCR.1-IgG

QEVTVIQAALSVPEGENLVNCSFTDSAIYNLQWFRQDPGKGLTSLLLISPWQREQTSGRNLASLDKSSGRSTLYIAASQPGDSAT  
YLC AVRPLLDGTYIPTFGRGTS LIVHPYIQNPDPVAVYQLRDSKSSDKFVCLFTDFDSQINVSQSKSDSVYITDKCVLDMRSMDFKS  
NSAVAWSNKSDFTCANAFNNSIIPEDTFFPSPESPSCGG  
QGTLVTVSAASTKGPSVFLAPSSKSTSGGTAALGCLVADYFPEPVTVSWNSGALTSGVHTFPAVLQSSGLYSLASVTVPSSSLG  
TQTYICNVNPKSNTKVDKRVESKCDKTHCPCPAPEAAGGPSVFLFPPKPKDTLMI SRTP ETV CVVVDVSHEDPEVKFNWYVD  
GVEVHNAKTKPREEQYSTYRVVSVLTVLHQDWLNGKEYKCKVSNKALPAPIEKTISKAKGQPRFPVYVYTLPPSREEMTKNQVSLV  
CLVKGFPYPSDIAVEWESNGQPENNYKTPPVLDSDGSFFLYSLLTVLTKSRWQQGNVFSCSVMHEALHNHYTQKSLSLSPGK

DKTHCPCPAPEAAGGPSVFLFPPKPKDTLMI SRTP ETV CVVVDVSHEDPEVKFNWYVDGVEVHNAKTKPREEQYSTYRVVSVL  
TVLHQDWLNGKEYKCKVSNKALPAPIEKTISKAKGQPREPQVYTLPPSREEMTINQVSLTCLVKGFPYPSDIAVEWESNGQPENNYK  
TTPPVLDSDGSFFLYSLLTVLTKSRWQQGNVFSCSVMHEALHNHYTQKSLSLSPGKGG  
VPEGENLVNCSFTDSAIYNLQWFRQDPGKGLTSLLLISPWQREQTSGRNLASLDKSSGRSTLYIAASQPGDSATYLC AVRPLLDG  
TYIPTFGRGTS LIVHPYIQNPDPVAVYQLRDSKSSDKFVCLFTDFDSQINVSQSKSDSVYITDKCVLDMRSMDFKSN SAVAWSNKSD  
FTCANAFNNSIIPEDTFFPSPESPSC

QAVVTQESALTTSPGETVTLTCRSTGAVTTSNYANWVQEKPDHLFTGLIGGNTKRAPGVPARFSGSLIGDKAALITGAQTEDEA  
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NKYAASSYLSLTPQWKS HRYSYSCQVTHEGSTVEKTVAPTEC

GVTQTPKFQVLKGTGQSMTLQCAQDMNHEYMSWYRQDPGMGLRLIHYSVAIQTTDRGEVPPNGYNVSRSTIEDFPLRLLSAAPSQTSV  
YFCASSYLNGTGEFFGEGSRLTVLEDLKNVFPPEVAVFEPSEKAEISRTQKATLVCLATGFYPPHVELSWSWVNGKEVHDGVCITDPO  
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### >2TCR.2-IgG

EVQLVESGGGLVQPKGSLKLSAASGFTFNNTYAMNWRQAPGKGLEWVARIRSKYNNYATYYADSVKDRFTISRDDSQSLLYLQMN  
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SGVHTFPAVLQSSGLYSLSSVTVPSSSLGKTYTCNVNPKSNTKVDKRVESKYGG  
VPEGENLVNCSFTDSAIYNLQWFRQDPGKGLTSLLLISPWQREQTSGRNLASLDKSSGRSTLYIAASQPGDSATYLC AVRPLLDG  
TYIPTFGRGTS LIVHPYIQNPDPVAVYQLRDSKSSDKFVCLFTDFDSQINVSQSKSDSVYITDKCVLDMRSMDFKSN SAVAWSNKSD  
FTCANAFNNSIIPEDTFFPSPESPSCDKTHCPCPAPEAAGGPSVFLFPPKPKDTLMI SRTP ETV CVVVDVSHEDPEVKFNWYVD  
GVEVHNAKTKPREEQYSTYRVVSVLTVLHQDWLNGKEYKCKVSNKALPAPIEKTISKAKGQPRFPVYVYTLPPSREEMTKNQVSLV  
CLVKGFPYPSDIAVEWESNGQPENNYKTPPVLDSDGSFFLYSLLTVLTKSRWQQGNVFSCSVMHEALHNHYTQKSLSLSPGK

DKTHCPCPAPEAAGGPSVFLFPPKPKDTLMI SRTP ETV CVVVDVSHEDPEVKFNWYVDGVEVHNAKTKPREEQYSTYRVVSVL  
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TTPPVLDSDGSFFLYSLLTVLTKSRWQQGNVFSCSVMHEALHNHYTQKSLSLSPGKGG  
VPEGENLVNCSFTDSAIYNLQWFRQDPGKGLTSLLLISPWQREQTSGRNLASLDKSSGRSTLYIAASQPGDSATYLC AVRPLLDG

TYIPTFGRGTS LIVHPYIQNPDPVAVYQLRDSKSSDKFVCLFTDFDSQINVSQSKSDSVYITDKCVLDMRSMDFKSNSAVAWSNKSD  
 FT CANAFNNSI IPEDTFFPSPEPKSC

QAVVTQESALTTSPGETVTLTCRSSTGAVTTSNYANWVQEKPDHLFTGLIGGTNKRAPGVPARFSGSLIGDKAALITGAQTEDEA  
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GVTQTPKFQVLKGTGQSMTLQCAQDMNHEYMSWYRQDPGMGLRLIHYSVAIQTTDRGEVPNGYNVSRSTIEDFLRLLSAAAPSQTSV  
 YFCASSYLNGTGFEGSRLTVLEDLKNVFPPEVAVFEPKAEISRTQKATLVCLATGFYPHVELS WVVNGKEVHDGVC TDPQ  
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>2TCR.3-IgG

EVQLVESGGGLVQPKGSLKLSCAASGFTFNTYAMNWRQAPGKGLEWVARIRSKYNNYATYYADSVKDRFTISRDDSQSLLYLQMN  
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 SGVHTFPAVLQSSGLYSLSSVTVTPSSSLGKTYTCNVNDRKPSNTKVDKRVESKYGGGGSGGGGSGGGGSGGGGSGQEVTPAALS  
 VPEGENLVLNCSTDSAIYNLQWFRQDPGKGLTSLLLISPWQREQTSGRNLASLDKSSGRSTLYIAASQPGDSATYLCVAVRPLLDG  
 TYIPTFGRGTS LIVHPYIQNPDPVAVYQLRDSKSSDKFVCLFTDFDSQINVSQSKSDSVYITDKCVLDMRSMDFKSNSAVAWSNKSD  
 FT CANAFNNSI IPEDTFFPSPEPKSCDKHTHTCPPCPAPEAAGGPSVFLFPPKPKDTLMI SRTEVTCVVDVSHEDPEVKFNWYVD  
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 CLVKGFPYPSDIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSLTVDKSRWQQGNVFCSCVMHEALHNHYTQKLSLSLSPGK

QEVTPAALSVP EGENLVLNCSTDSAIYNLQWFRQDPGKGLTSLLLITPWQREQTSGRNLASLDKSSGRSTLYIAASQPGDSAT  
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 PEVKFNWYVDGVEVHNAKTKPREEQYSTYRVVSVLTVLHQDWLNGKEYKCKVSNKALPAPIEKTISKAKGQPRRPVYVYTLPPSRE  
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 SLSLSPGK

QAVVTQESALTTSPGETVTLTCRSSTGAVTTSNYANWVQEKPDHLFTGLIGGTNKRAPGVPARFSGSLIGDKAALITGAQTEDEA  
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 NKYAASSYLSLTPQWKS HRYSYSCQVTHEGSTVEKTVAPTEC

GVTQTPKFQVLKGTGQSMTLQCAQDMNHEYMSWYRQDPGMGLRLIHYSVAIQTTDRGEVPNGYNVSRSTIEDFLRLLSAAAPSQTSV  
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>2TCR.4-IgG

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 NSAVAWSNKSDFT CANAFNNSI IPEDTFFPSPEKSCDKHTHTCPPCPAPEAAGGPSVFLFPPKPKDTLMI SRTEVTCVVDVSHED  
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 EMTKNQVSLVCLVKGFPYPSDIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSLTVDKSRWQQGNVFCSCVMHEALHNHYTQKLS  
 LSPGKGGGGSGGGGSGGGGSGGGGSEVQLVESGGGLVQPKGSLKLSCAASGFTFNTYAMNWRQAPGKGLEWVARIRSKYNNYATY  
 YADSVKDRFTISRDDSQSLLYLQMN NLKTEDTAMYCVRHGNFGNSYVSWFAYWGGTGLVTVSAASTKGPSVFLPAPCSRSTSEST  
 AALGCLVKDYFPEPVTVSWNSGALTS GVHTFPAVLQSSGLYSLSSVTVTPSSSLGKTYTCNVNDRKPSNTKVDKRVESKYG

QEVTPAALSVP EGENLVLNCSTDSAIYNLQWFRQDPGKGLTSLLLITPWQREQTSGRNLASLDKSSGRSTLYIAASQPGDSAT  
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 PEVKFNWYVDGVEVHNAKTKPREEQYSTYRVVSVLTVLHQDWLNGKEYKCKVSNKALPAPIEKTISKAKGQPREPQVYVYTLPPSR  
 EEMTKNQVSLTCLVKGFPYPSDIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSLTVDKSRWQQGNVFCSCVMHEALHNHYTQKLS  
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QAVVTQESALTTSPGETVTLTCRSSTGAVTTSNYANWVQEKPDHLFTGLIGGTNKRAPGVPARFSGSLIGDKAALITGAQTEDEA  
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 NKYAASSYLSLTPQWKS HRYSYSCQVTHEGSTVEKTVAPTEC

GVTQTPKFQVLKGTGQSMTLQCAQDMNHEYMSWYRQDPGMGLRLIHYSVAIQTTDRGEVPNGYNVSRSTIEDFLRLLSAAAPSQTSV  
 YFCASSYLNGTGFEGSRLTVLEDLKNVFPPEVAVFEPKAEISRTQKATLVCLATGFYPHVELS WVVNGKEVHDGVC TDPQ  
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