```
         10          20          30          40          50
MQIPQAPWPV VWAVLQLGWR PGWFLDSPDR PWNPPTFSPA LLVVTEGDNA
         60          70          80          90         100
TFTCSFSNTS ESFVLNWYRM SPSNQTDKLA AFPEDRSQPG QDCRFRVTQL
        110         120         130         140         150
PNGRDFHMSV VRARRNDSGT YLCGAISLAP KAQIKESLRA ELRVTERRAE
        160         170         180         190         200
VPTAHPSPSP RPAGQFQTLV VGVVGGLLGS LVLLVWVLAV ICSRAARGTI
        210         220         230         240         250
GARRTGQPLK EDPSAVPVFS VDYGELDFQW REKTPEPPVP CVPEQTEYAT
        260         270         280
IVFPSGMGTS SPARRGSADG PRSAQPLRPE DGHCSWPL
```

Human PD-1 (SEQ ID NO:1)

```
         10          20          30          40          50
MWVRQVPWSF TWAVLQLSWQ SGWLLEVPNG PWRSLTFYPA WLTVSEGANA
         60          70          80          90         100
TFTCSLSNWS EDLMLNWNRL SPSNQTEKQA AFCNGLSQPV QDARFQIIQL
        110         120         130         140         150
PNRHDFHMNI LDTRRNDSGI YLCGAISLHP KAKIEESPGA ELVVTERILE
        160         170         180         190         200
TSTRYPSPSP KPEGRFQGMV IGIMSALVGI PVLLLLAWAL AVFCSTSMSE
        210         220         230         240         250
ARGAGSKDDT LKEEPSAAPV PSVAYEELDF QGREKTPELP TACVHTEYAT
        260         270         280
IVFTEGLGAS AMGRRGSADG LQGPRPPRHE DGHCSWPL
```

Murine PD-1 (SEQ ID NO:2)

FIG. 1A

```
        10          20          30          40          50
MRIFAVFIFM TYWHLLNAFT VTVPKDLYVV EYGSNMTIEC KFPVEKQLDL
        60          70          80          90         100
AALIVYWEME DKNIIQFVHG EEDLKVQHSS YRQRARLLKD QLSLGNAALQ
       110         120         130         140         150
ITDVKLQDAG VYRCMISYGG ADYKRITVKV NAPYNKINQR ILVVDPVTSE
       160         170         180         190         200
HELTCQAEGY PKAEVIWTSS DHQVLSGKTT TTNSKREEKL FNVTSTLRIN
       210         220         230         240         250
TTTNEIFYCT FRRLDPEENH TAELVIPELP LAHPPNERTH LVILGAILLC
       260         270         280         290
LGVALTFIFR LRKGRMMDVK KCGIQDTNSK KQSDTHLEET
```

Human PD-L1 (SEQ ID NO:3)

```
        10          20          30          40          50
LQLHQIAALF TVTVPKELYI IEHGSNVTLE CNFDTGSHVN LGAITASLQK
        60          70          80          90         100
VENDTSPHRE RATLLEEQLP LGKASFHIPQ VQVRDEGQYQ CIIIYGVAWD
       110         120         130         140         150
YKYLTLKVKA SYRKINTHIL KVPETDEVEL TCQATGYPLA EVSWPNVSVP
       160         170         180         190         200
ANTSHSRTPE GLYQVTSVLR LKPPPGRNFS CVFWNTHVRE LTLASIDLQS
       210         220         230         240         250
QMEPRTHPTW LLHIFIPSCI IAFIFIATVI ALRKQLCQKL YSSKDTTKRP
       260
VTTTKREVNS AI
```

Human PD-L2 (SEQ ID NO:4)

FIG. 1B

Exemplary V_H and V_L sequences. CDRs underlined.

Mouse 12A11 V_H (SEQ ID NO:31)

QIQLVQSGPELKKPGETVMISCKASGYTFTHYGMNWVKQAPGKGLKWVGWVNT
YTGEPTYADDFKGRLAFSLETSASTAYLQINNLKNEDTATYFCTREGEGMGFGDW
GQGTTLTVSS

Mouse 12A11 V_L (SEQ ID NO:41)

DVLMTQIPLSLPVSLGDQASISCRSSQSIVHSHGDTYLEWYLQKPGQSPKLLIYKVS
NRFSGVPDRFSGSGSGTDFTLKISRVEAEDLGVYYCFQGSHIPVTFGAGTKLEIK

Humanized 12A11.1b V_H (SEQ ID NO:32)

EIQLVQSGSELKKPGASVKVSCKASGYTFTHYGMNWVRQAPGQGLEWVGWVNT
YTGEPTYADDFKGRLVFSLDTSVSTAYLQISSLKAEDTAVYYCTREGEGMGFGDW
GQGTTVTVSS

Humanized 12A11.1a V_L (SEQ ID NO:42)

DVVMTQSPLSLPVTPGEPASISCRSSQSIVHSHGDTYLEWYLQKPGQSPQLLIYKVS
NRFSGVPDRFSGSGSGTDFTLKISRVEAEDVGVYYCFQGSHIPVTFGQGTKLEIK

Humanized 12A11.2b V_H (SEQ ID NO:33)

EIQLVQSGAEVKKPGSSVKVSCKASGYTFTHYGMNWVRQAPGQGLEWVGWVNT
YTGEPTYADDFKGRLTFTLDTSTSTAYMELSSLRSEDTAVYYCTREGEGMGFGDW
GQGTTVTVSS

Humanized 12A11.2b V_H with M99I mutation (SEQ ID NO:34)

EIQLVQSGAEVKKPGSSVKVSCKASGYTFTHYGMNWVRQAPGQGLEWVGWVNT
YTGEPTYADDFKGRLTFTLDTSTSTAYMELSSLRSEDTAVYYCTREGEGIGFGDW
GQGTTVTVSS

Humanized 12A11.2b V_H with M99V mutation (SEQ ID NO:35)

EIQLVQSGAEVKKPGSSVKVSCKASGYTFTHYGMNWVRQAPGQGLEWVGWVNT
YTGEPTYADDFKGRLTFTLDTSTSTAYMELSSLRSEDTAVYYCTREGEGVGFGDW
GQGTTVTVSS

Humanized 12A11.2b V_H with M99L mutation (SEQ ID NO:36)

EIQLVQSGAEVKKPGSSVKVSCKASGYTFTHYGMNWVRQAPGQGLEWVGWVNT
YTGEPTYADDFKGRLTFTLDTSTSTAYMELSSLRSEDTAVYYCTREGEGLGFGDW
GQGTTVTVSS

FIG. 2

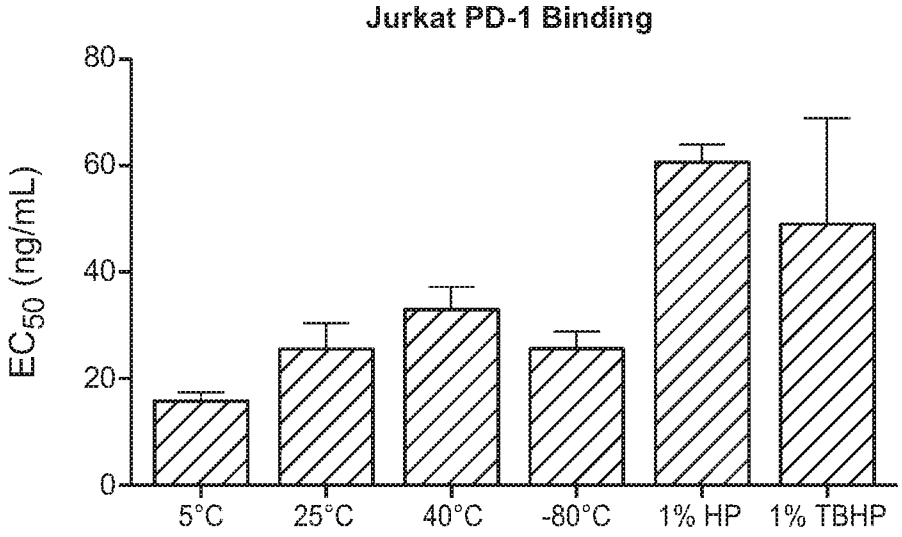

Jurkat PD-1 Binding
EC50 (ng/mL)
80
60
40
20
0
5°C
25°C
40°C
-80°C
1% HP
1% TBHP

FIG. 3A

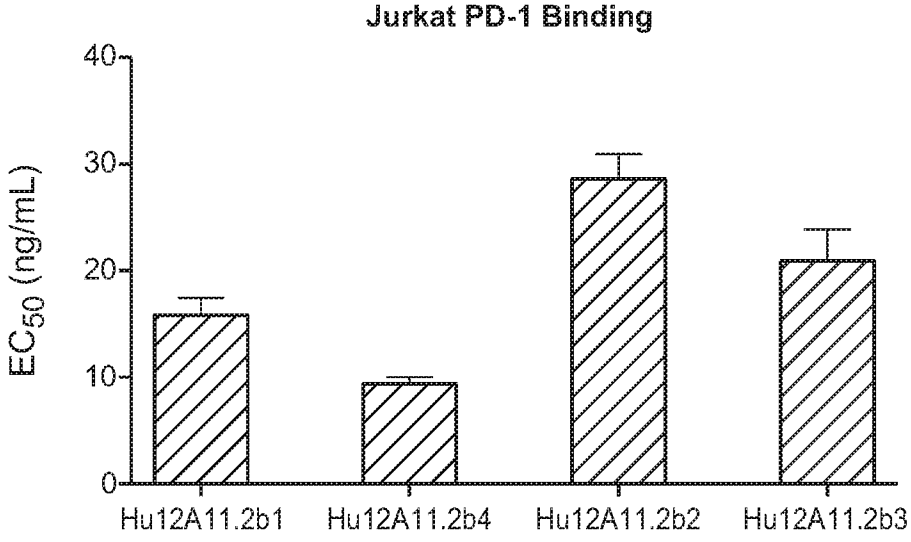

Jurkat PD-1 Binding
EC50 (ng/mL)
40
30
20
10
0
Hu12A11.2b1
Hu12A11.2b4
Hu12A11.2b2
Hu12A11.2b3

FIG. 3B

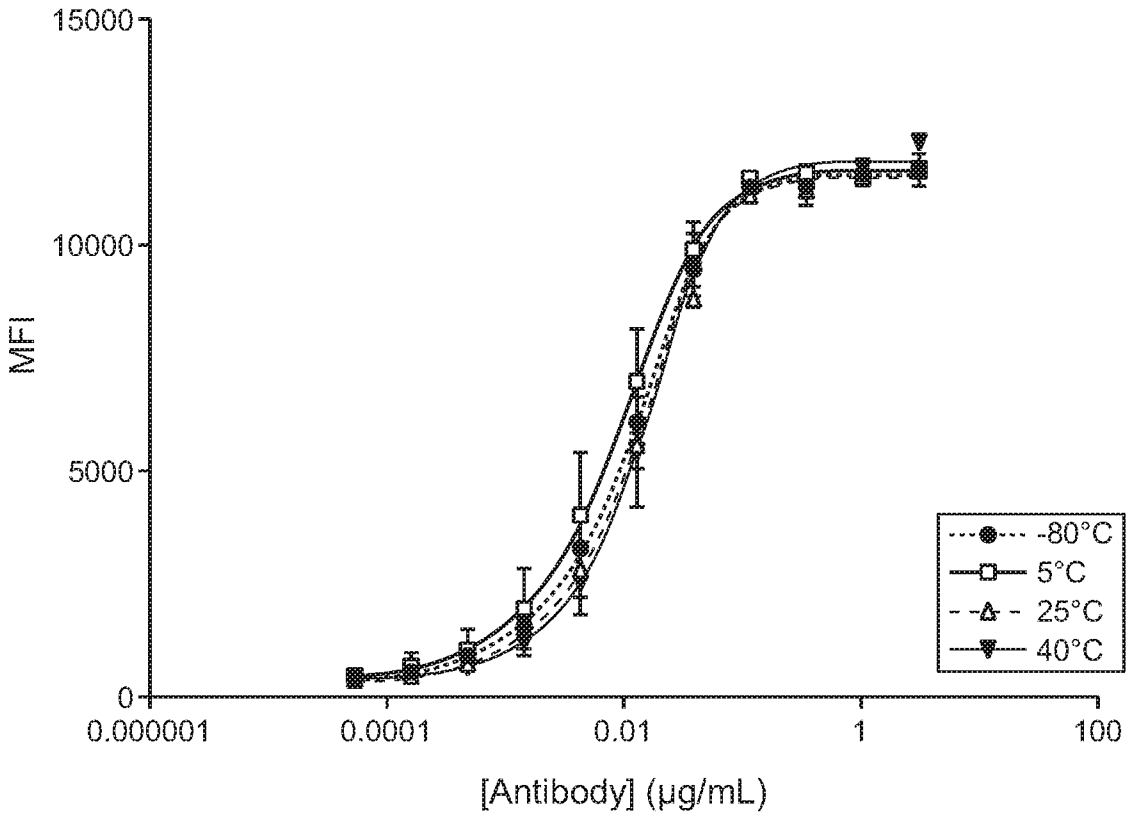

15000
10000
5000
0
MFI
0.000001
0.0001
0.01
1
100
[Antibody] (µg/mL)
-80°C
5°C
25°C
40°C

FIG. 3C

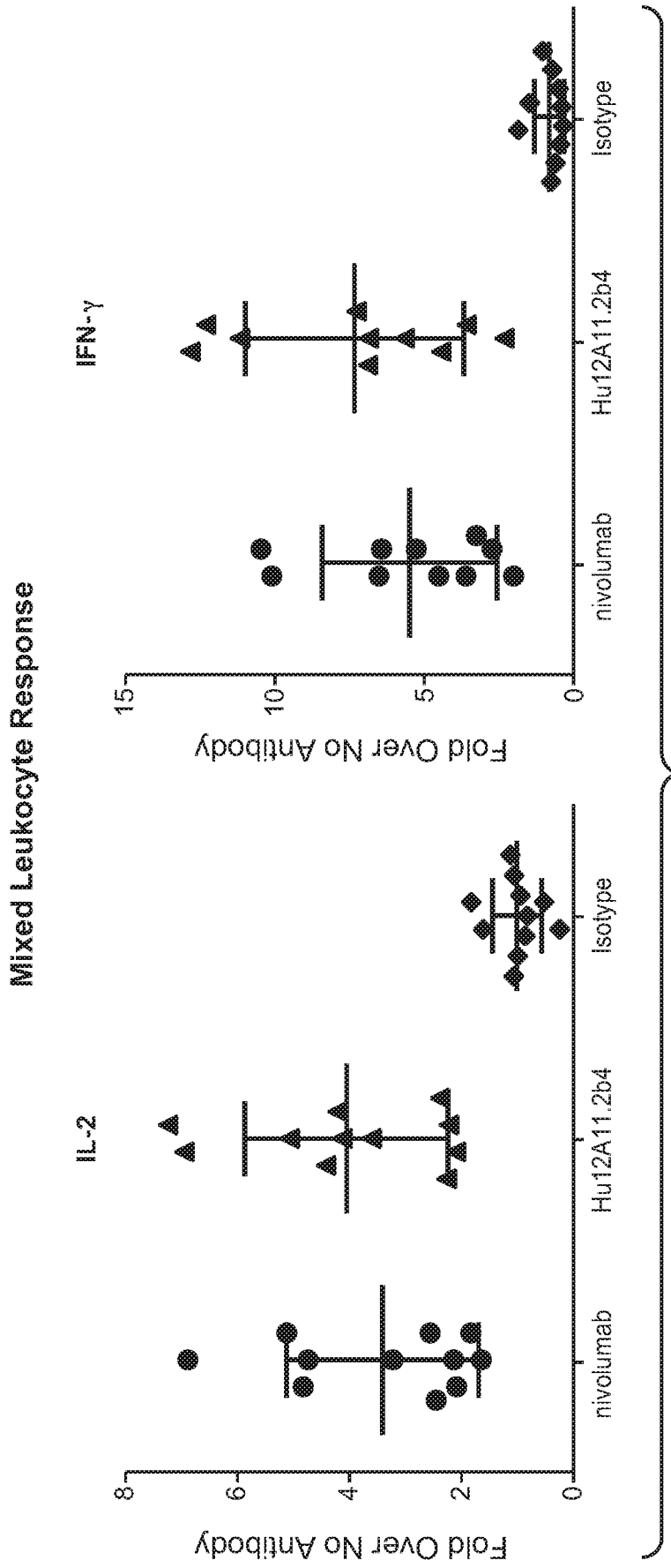

Mixed Leukocyte Response
IL-2
IFN-γ
Fold Over No Antibody
0
2
4
6
8
0
5
10
15
nivolumab
Hu12A11.2b4
Isotype

FIG. 4A

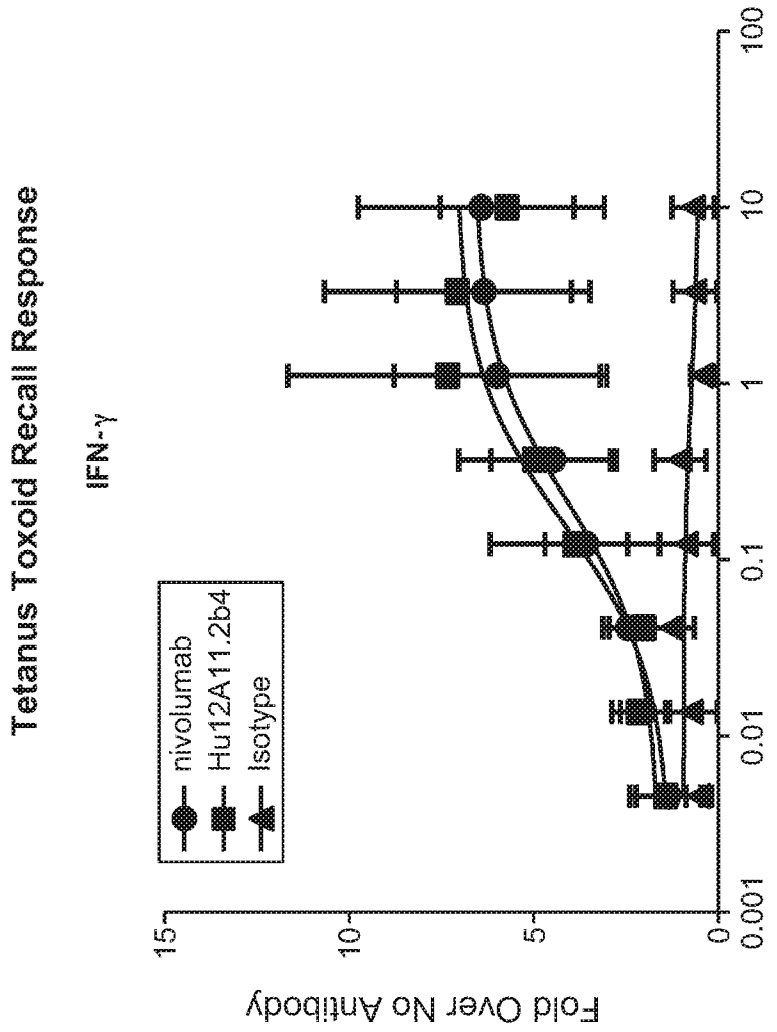

Tetanus Toxoid Recall Response
IFN-γ
nivolumab
Hu12A11.2b4
Isotype
Fold Over No Antibody
15
10
5
0
0.001
0.01
0.1
1
10
100
[Antibody] (µg/mL)

FIG. 4B