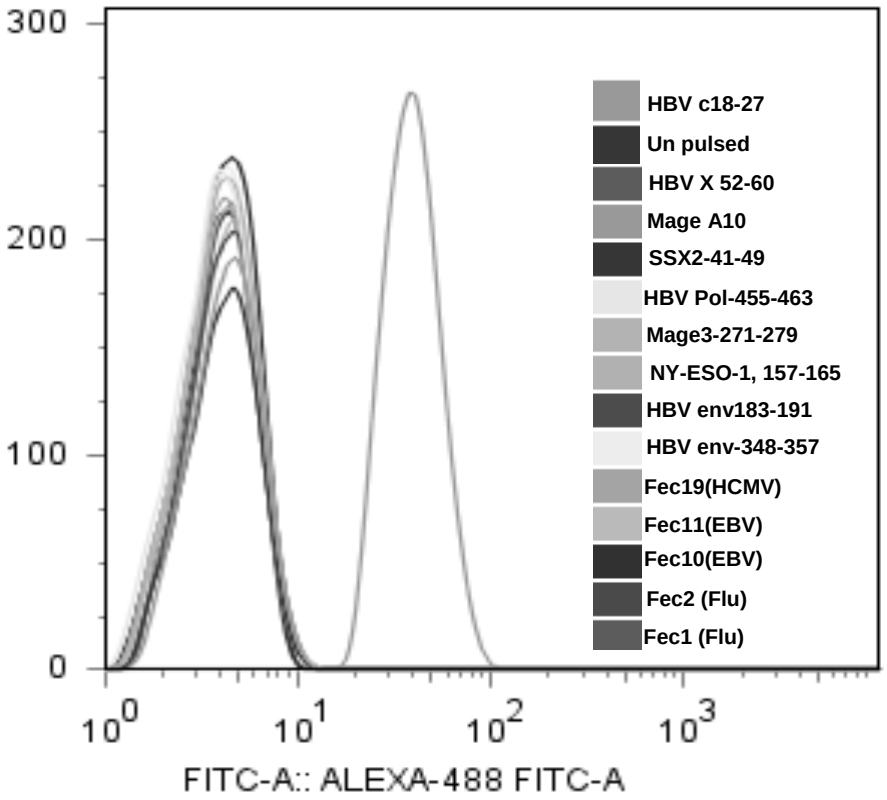
A


300
200
100
0
10^0
10^1
10^2
10^3
FITC-A:: ALEXA-488 FITC-A
HBV c18-27
Un pulsed
HBV X 52-60
Mage A10
SSX2-41-49
HBV Pol-455-463
Mage3-271-279
NY-ESO-1, 157-165
HBV env183-191
HBV env-348-357
Fec19(HCMV)
Fec11(EBV)
Fec10(EBV)
Fec2 (Flu)
Fec1 (Flu)

B

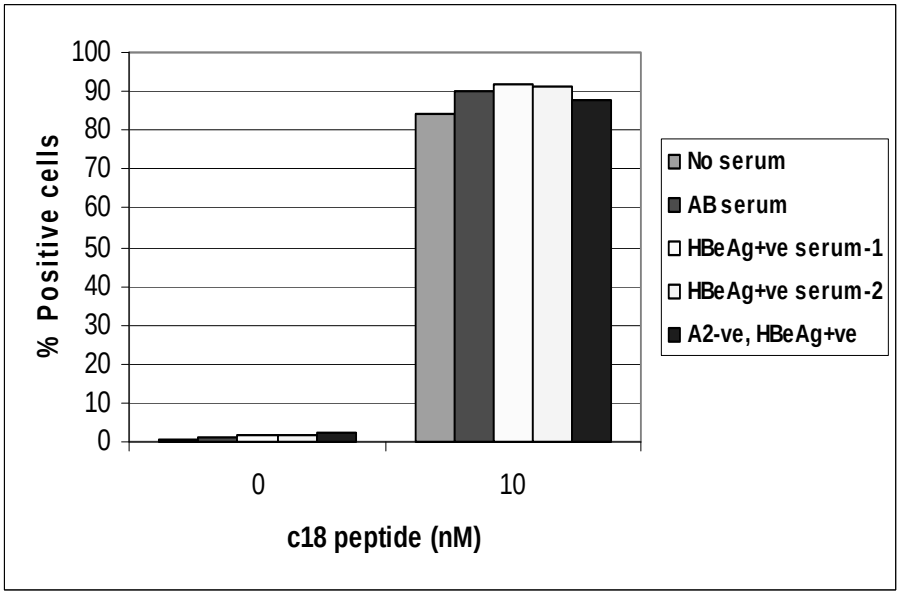
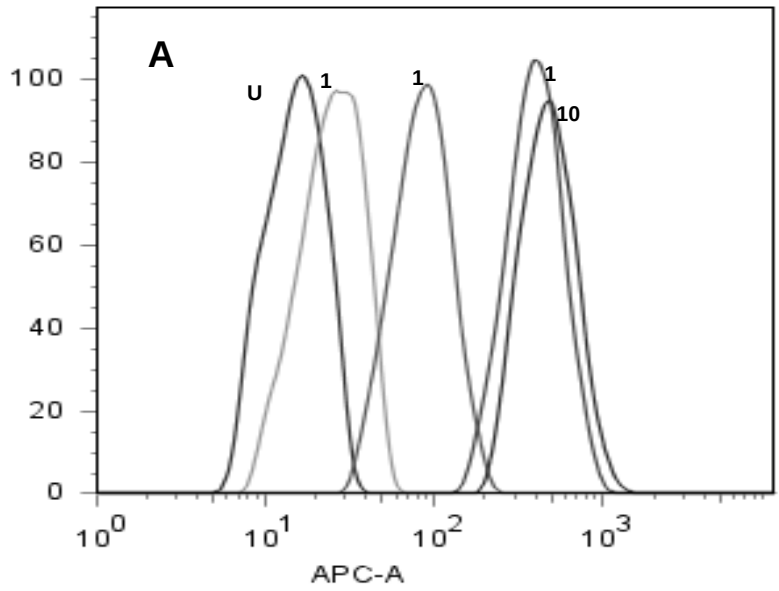
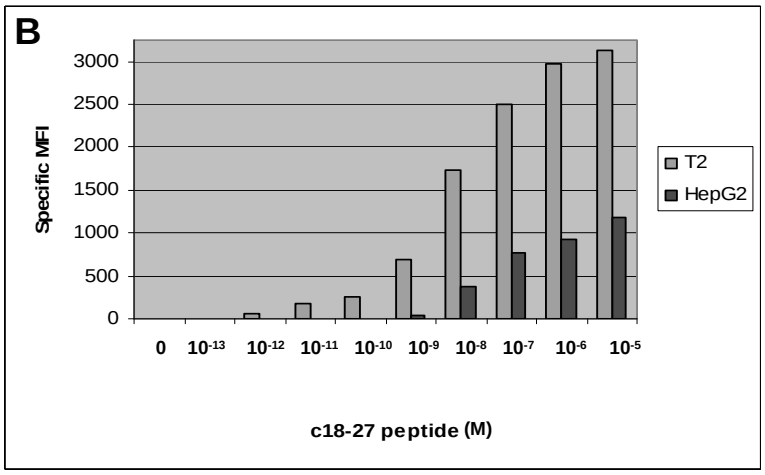

% Positive cells
100
90
80
70
60
50
40
30
20
10
0
0
10
c18 peptide (nM)
No serum
AB serum
HBeAg+ve serum-1
HBeAg+ve serum-2
A2-ve, HBeAg+ve

FIGURE 1


A
U
1
1
1
10
100
80
60
40
20
0
10^0
10^1
10^2
10^3
APC-A


B
Specific MFI
3000
2500
2000
1500
1000
500
0
T2
HepG2
0
10^{-13}
10^{-12}
10^{-11}
10^{-10}
10^{-9}
10^{-8}
10^{-7}
10^{-6}
10^{-5}
c18-27 peptide (M)

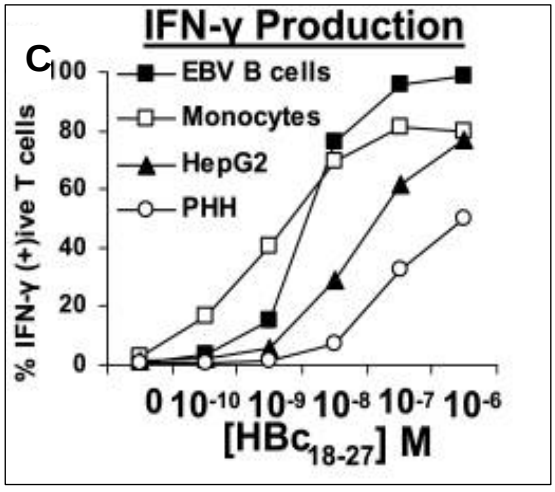

IFN-γ Production
C
EBV B cells
Monocytes
HepG2
PHH
% IFN-γ (+)ive T cells
100
80
60
40
20
0
0 10^{-10} 10^{-9} 10^{-8} 10^{-7} 10^{-6}
[HBc_{18-27}] M

FIGURE 2

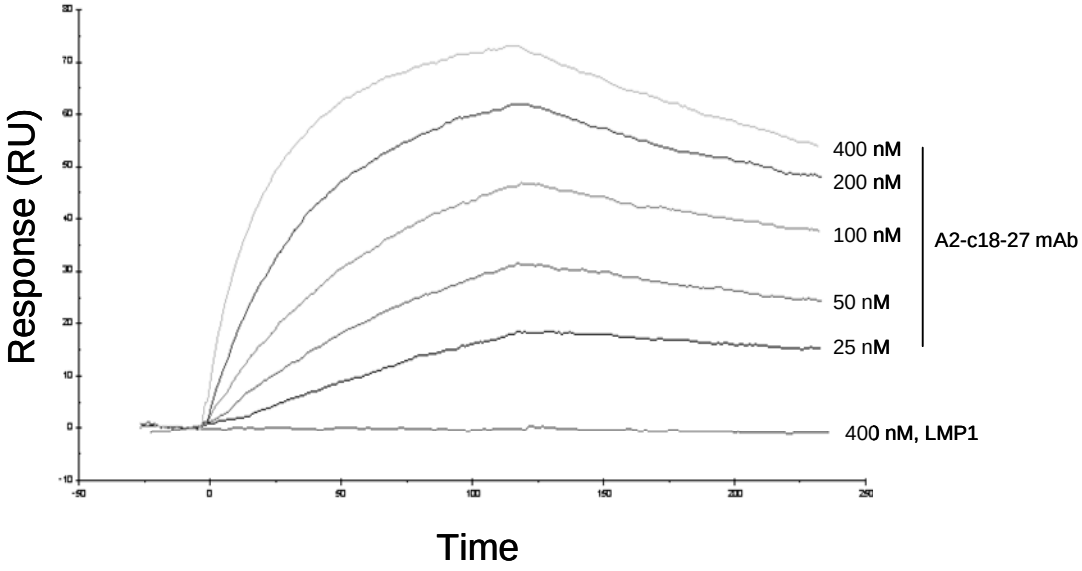


FIGURE 3

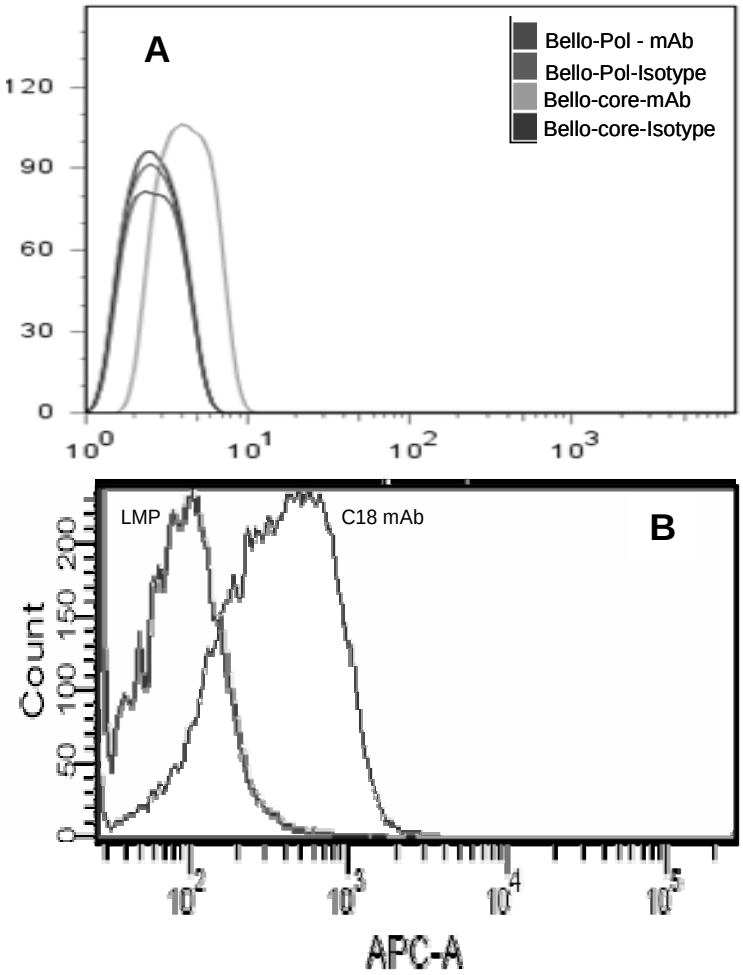


FIGURE 4

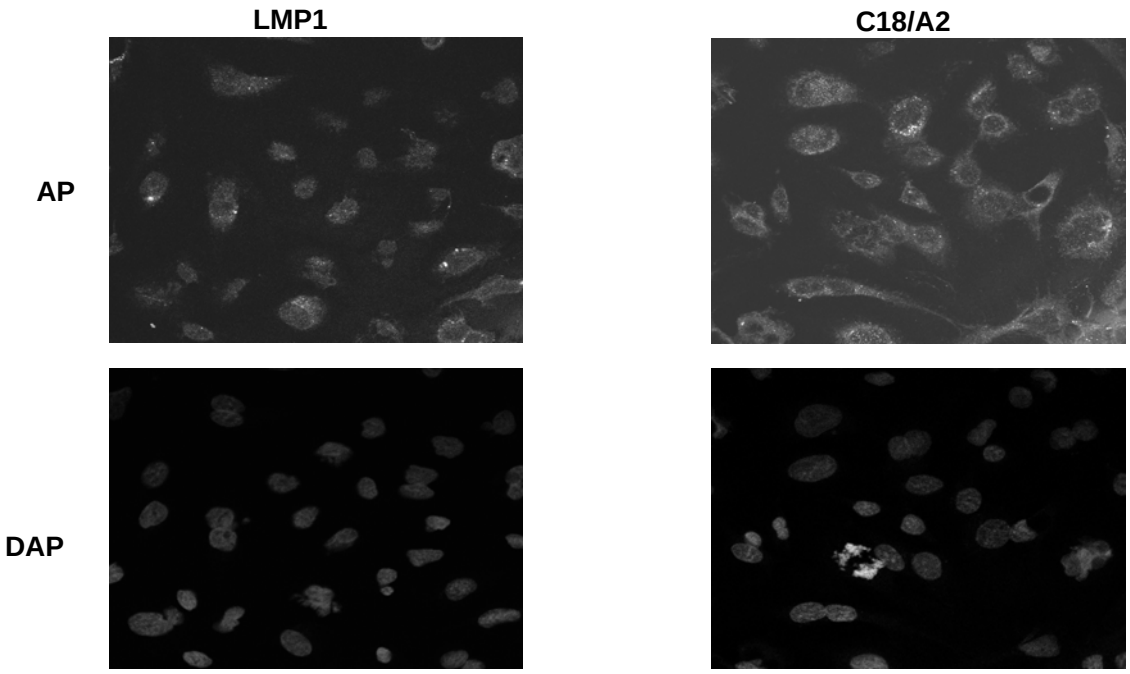


FIGURE 5

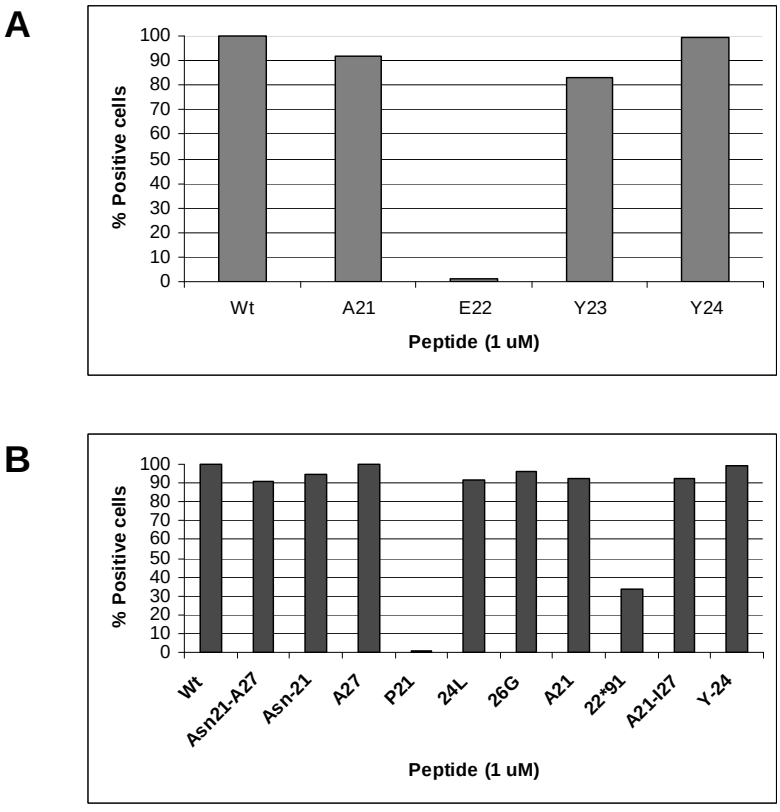
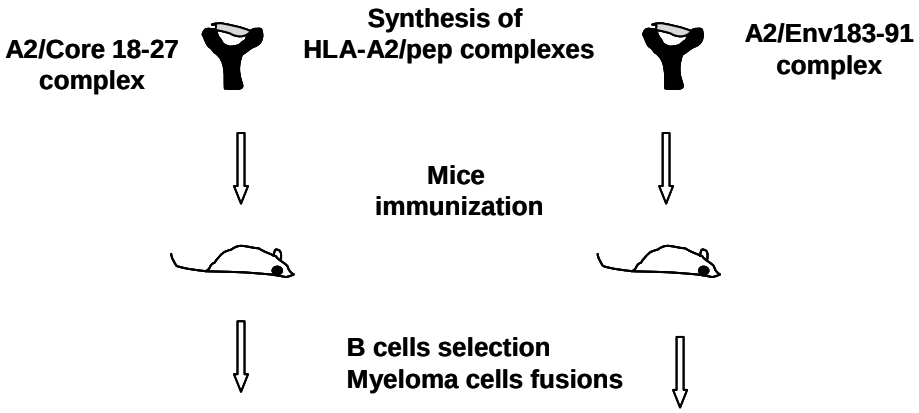


FIGURE 6

A



Ab c18/A2

Total	Negative	HLA-A2 specific	c18/A2 specific
1452	1200	249	3

Ab e183-91/A2

Total	Negative	HLA-A2 specific	e183/A2 specific
2000	1650	348	2

B

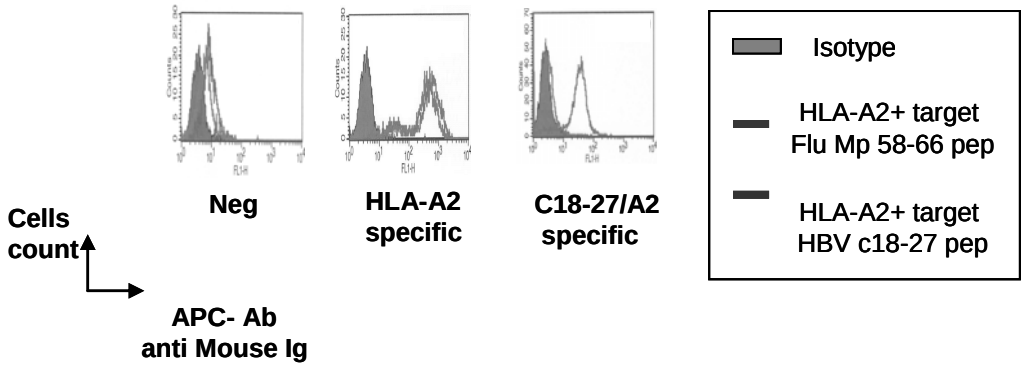
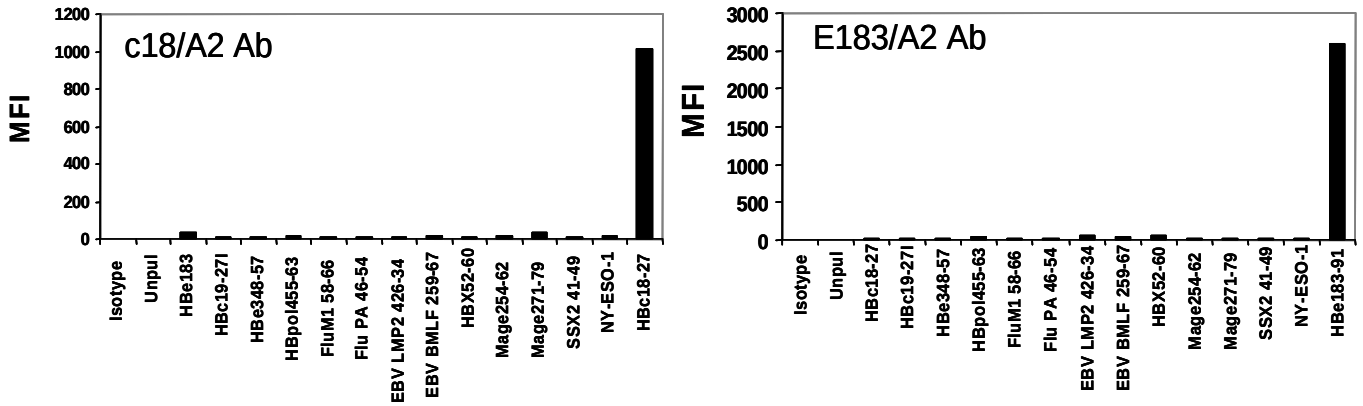
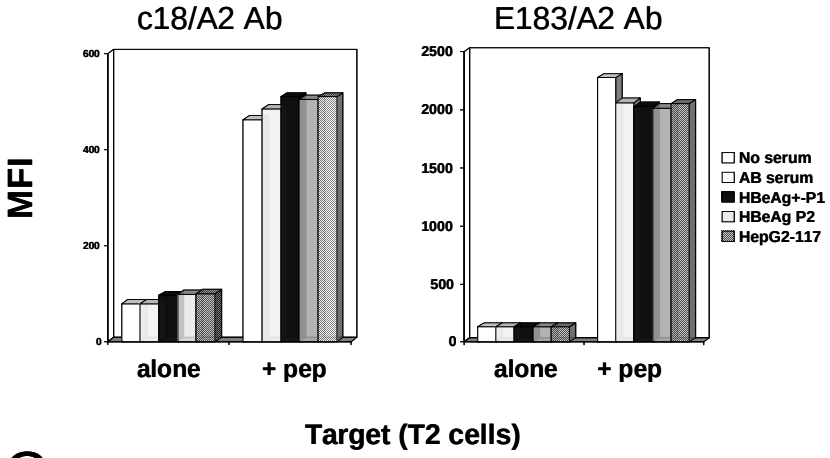


FIGURE 7

A


c18/A2 Ab
MFI
1200
1000
800
600
400
200
0
Isotype
Unpul
HBe183
HBc19-27l
HBe348-57
HBpol455-63
FluM1 58-66
Flu PA 46-54
EBV LMP2 426-34
EBV BMLF 259-67
HBX52-60
Mage254-62
Mage271-79
SSX2 41-49
NY-ESO-1
HBc18-27
E183/A2 Ab
MFI
3000
2500
2000
1500
1000
500
0
Isotype
Unpul
HBc18-27
HBc19-27l
HBe348-57
HBpol455-63
FluM1 58-66
Flu PA 46-54
EBV LMP2 426-34
EBV BMLF 259-67
HBX52-60
Mage254-62
Mage271-79
SSX2 41-49
NY-ESO-1
HBe183-91

B


c18/A2 Ab
E183/A2 Ab
MFI
600
400
200
0
2500
2000
1500
1000
500
0
alone
+ pep
alone
+ pep
No serum
AB serum
HBeAg+-P1
HBeAg P2
HepG2-117
Target (T2 cells)

C

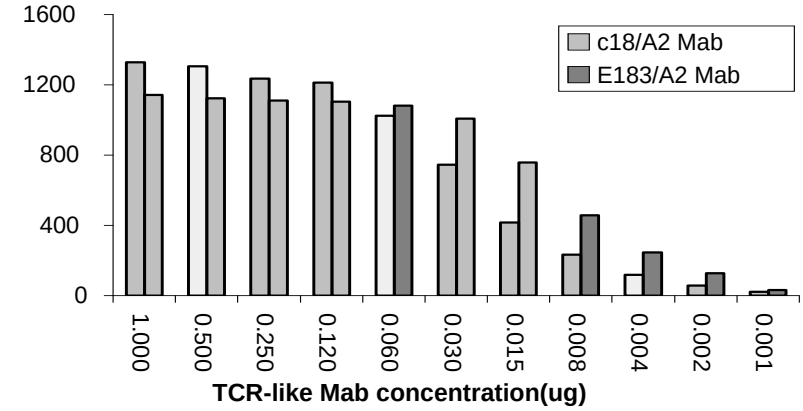

1600
1200
800
400
0
c18/A2 Mab
E183/A2 Mab
1.000
0.500
0.250
0.120
0.060
0.030
0.015
0.008
0.004
0.002
0.001
TCR-like Mab concentration(ug)

FIGURE 8

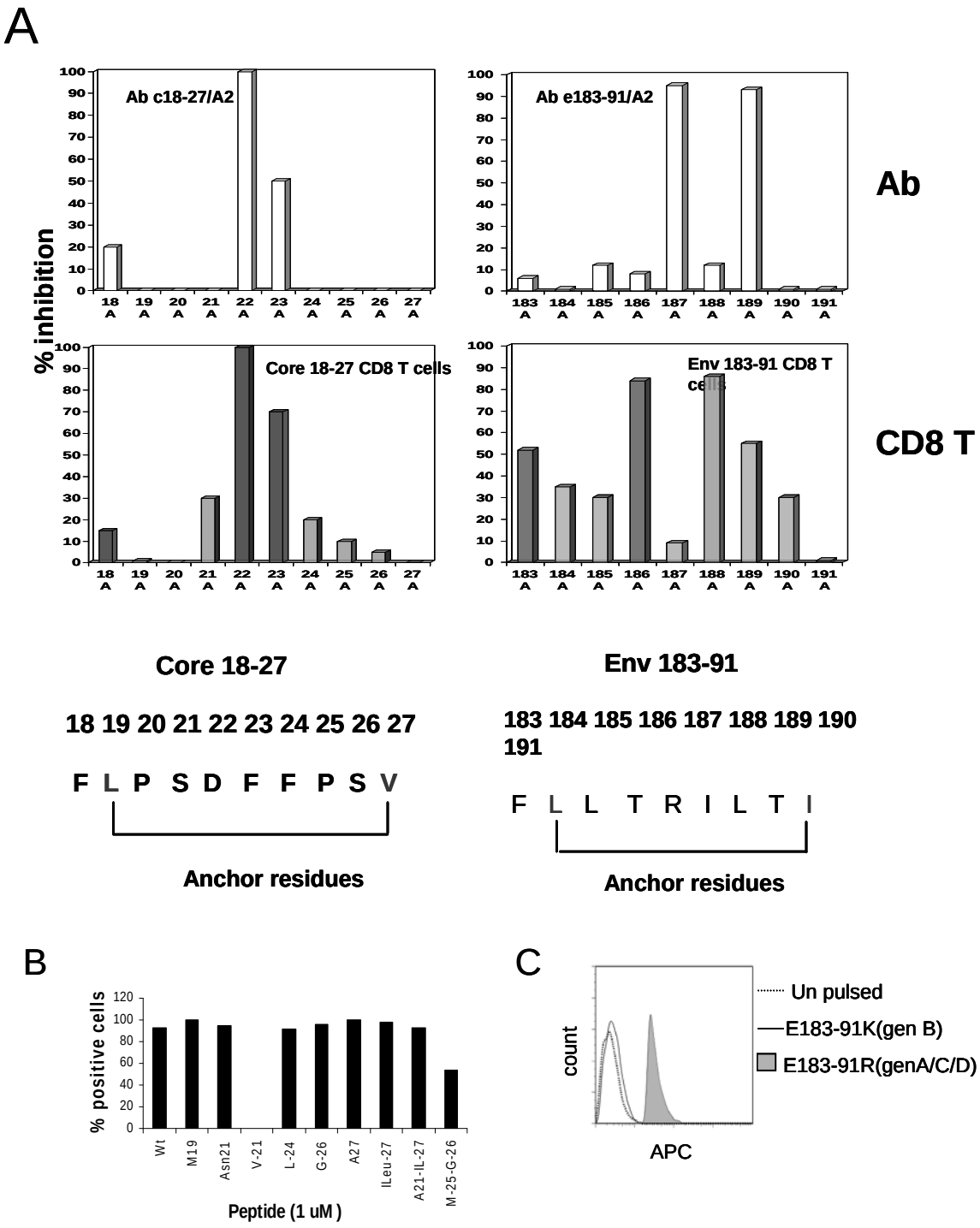

A
Ab c18-27/A2
Ab e183-91/A2
Ab
% inhibition
Core 18-27 CD8 T cells
Env 183-91 CD8 T cells
CD8 T
Core 18-27
18 19 20 21 22 23 24 25 26 27
F L P S D F F P S V
Anchor residues
Env 183-91
183 184 185 186 187 188 189 190 191
F L L T R I L T I
Anchor residues
B
% positive cells
Wt
M19
Asn21
V-21
L-24
G-26
A27
ILeu-27
A21-IL-27
M-25-G-26
Peptide (1 uM)
C
count
Un pulsed
E183-91K(gen B)
E183-91R(genA/C/D)
APC

FIGURE 9

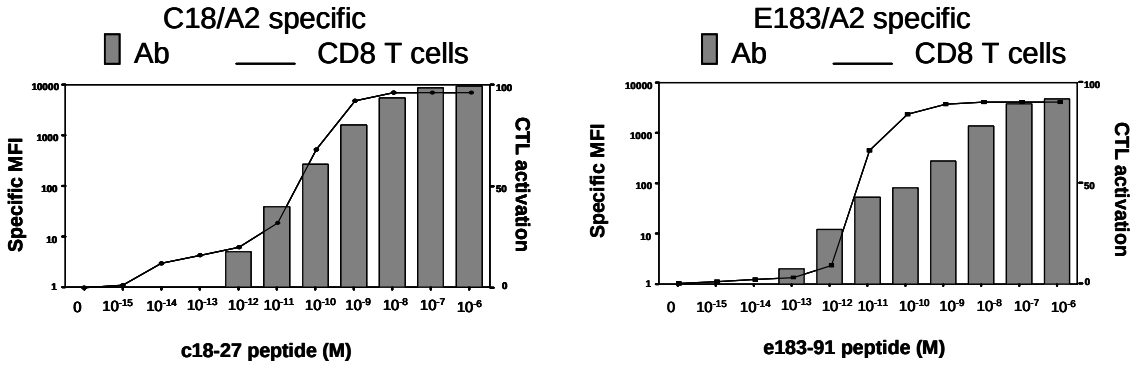


FIGURE 10

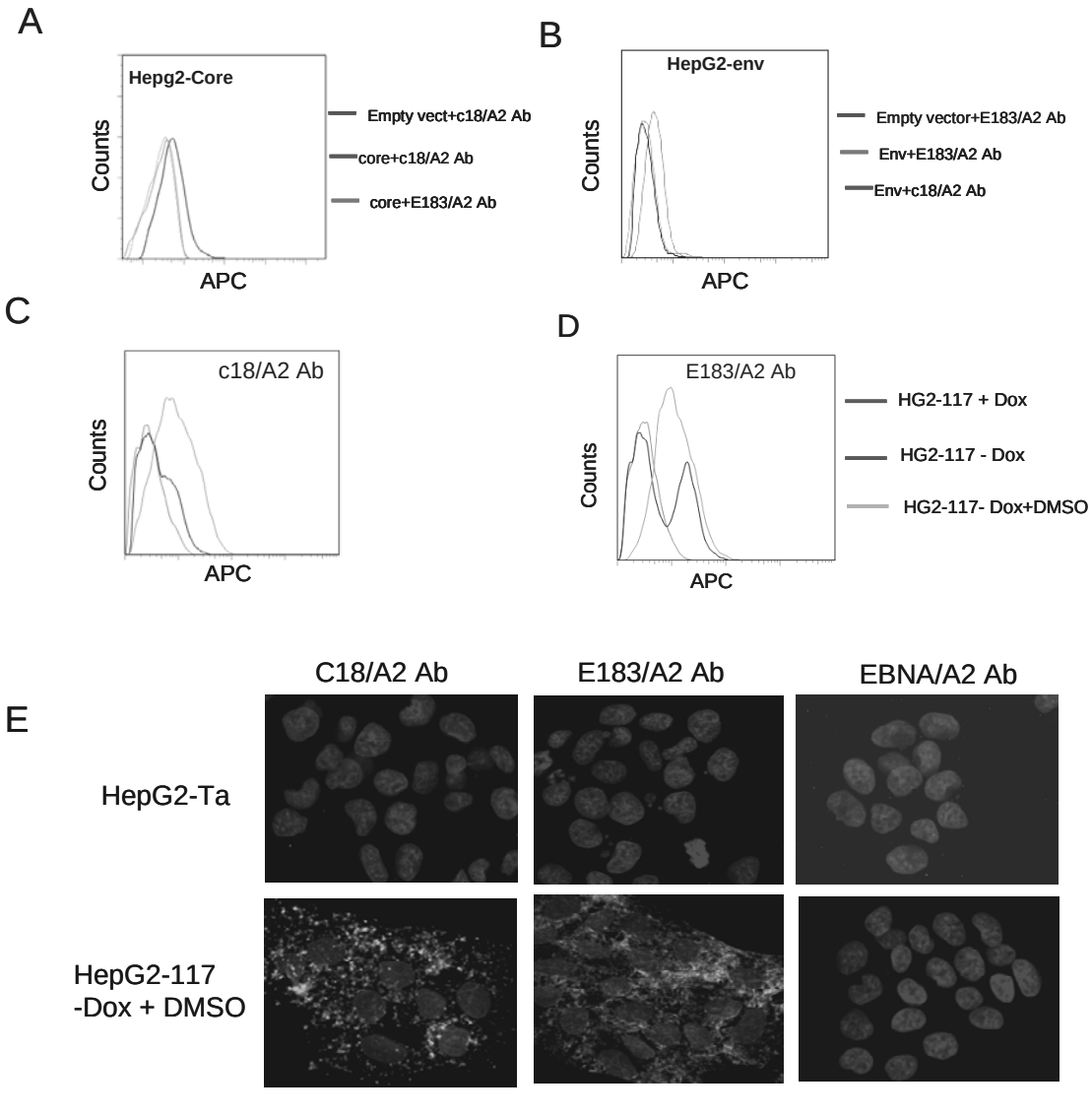


FIGURE 11

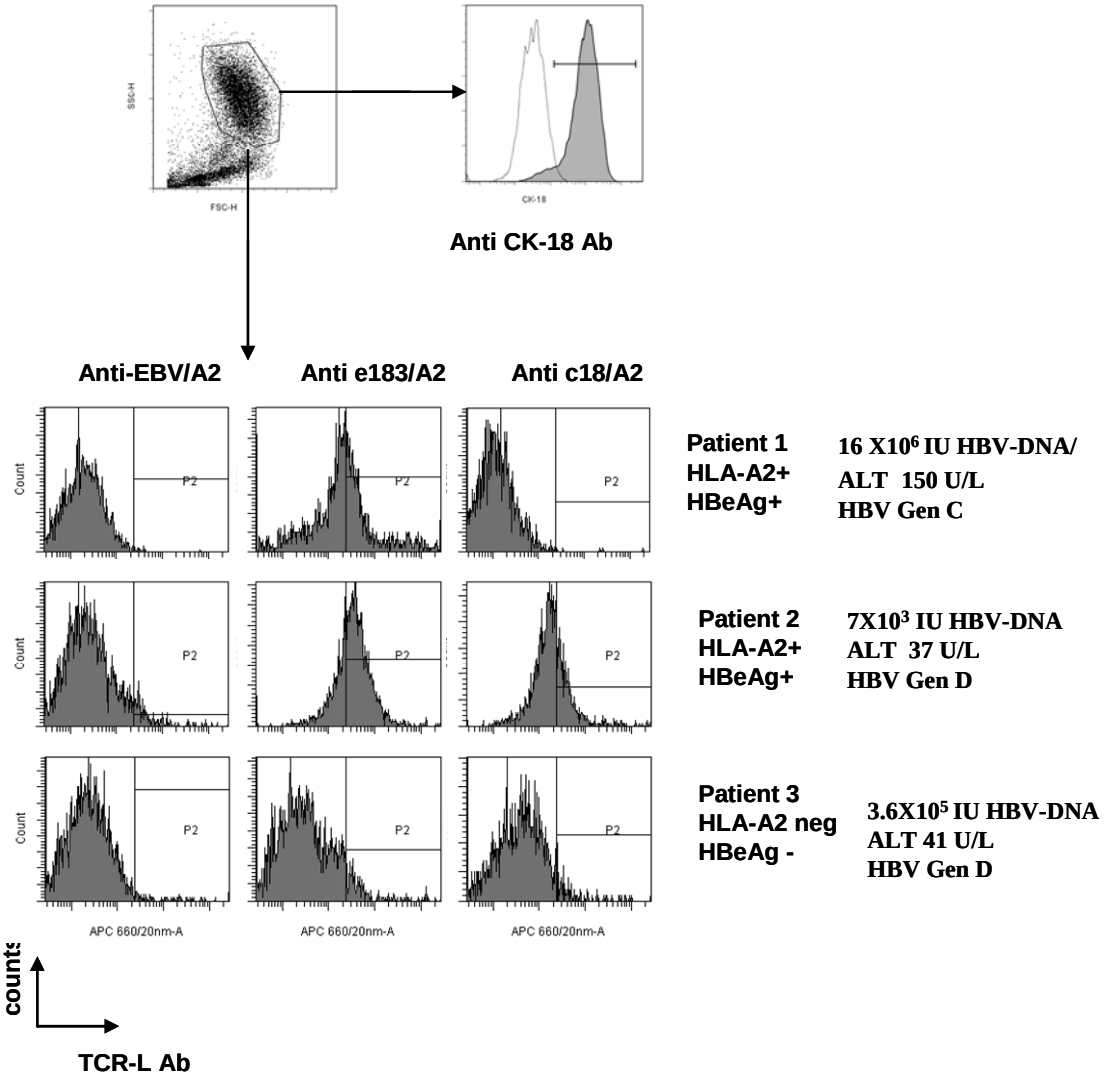

SSC-H
FSC-H
CK-18
Anti CK-18 Ab
Anti-EBV/A2
Anti e183/A2
Anti c18/A2
Count
P2
APC 660/20nm-A
counts
TCR-L Ab
Patient 1
HLA-A2+
HBeAg+
16 X10⁶ IU HBV-DNA/
ALT 150 U/L
HBV Gen C
Patient 2
HLA-A2+
HBeAg+
7X10³ IU HBV-DNA
ALT 37 U/L
HBV Gen D
Patient 3
HLA-A2 neg
HBeAg -
3.6X10⁵ IU HBV-DNA
ALT 41 U/L
HBV Gen D

FIGURE 12

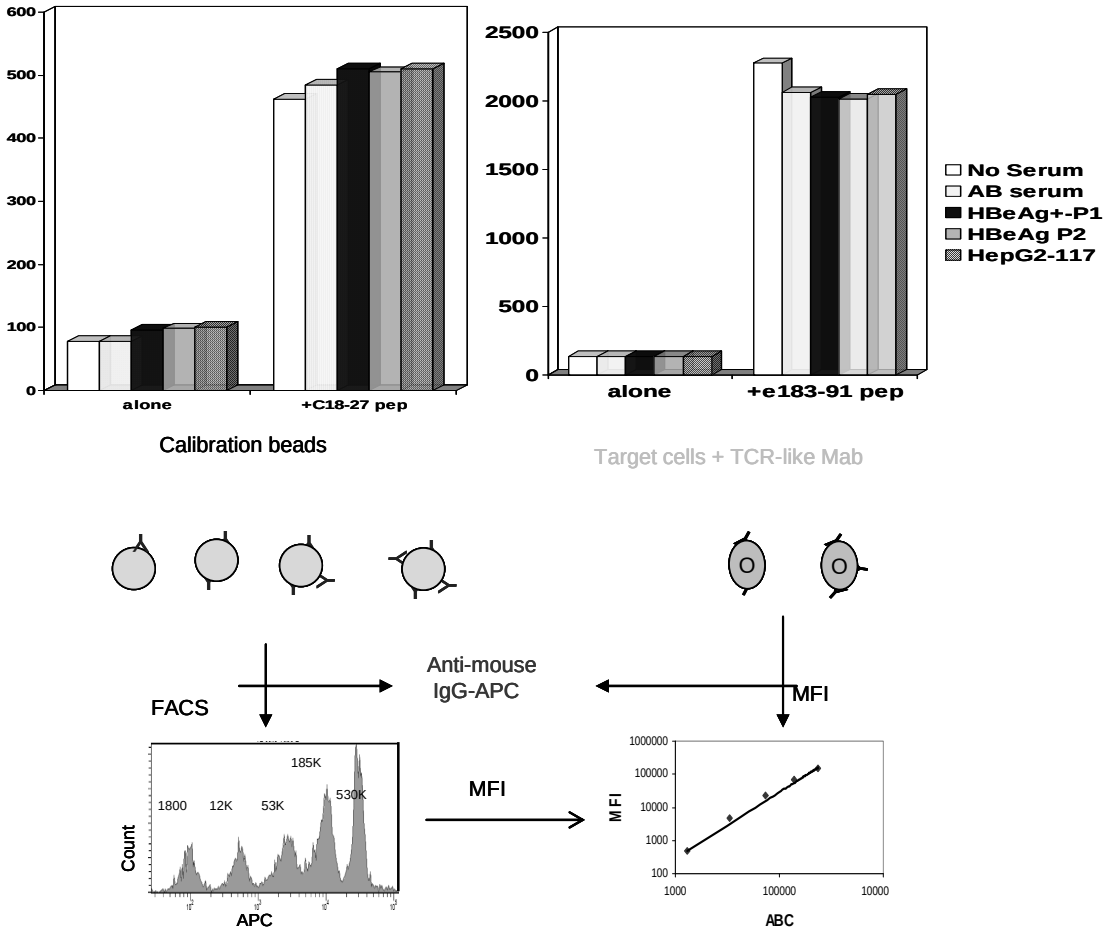


FIGURE 13

E183/A2 antibody

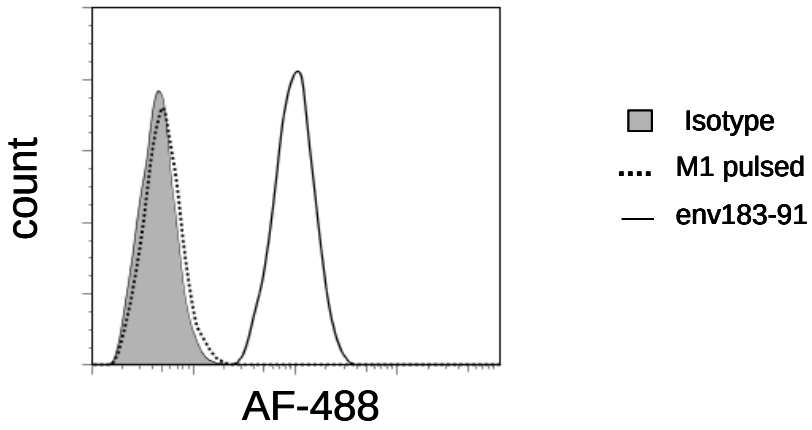


FIGURE 14

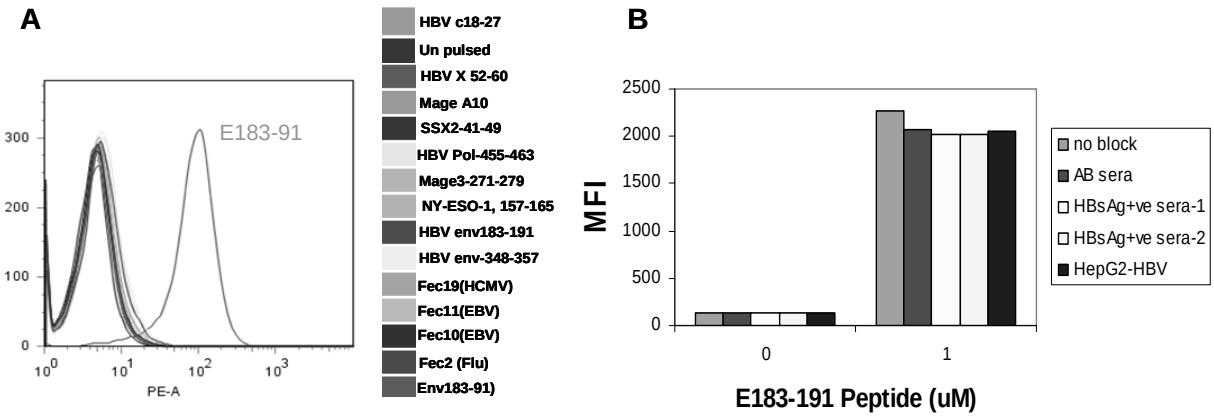


FIGURE 15

E183/A2 mAb

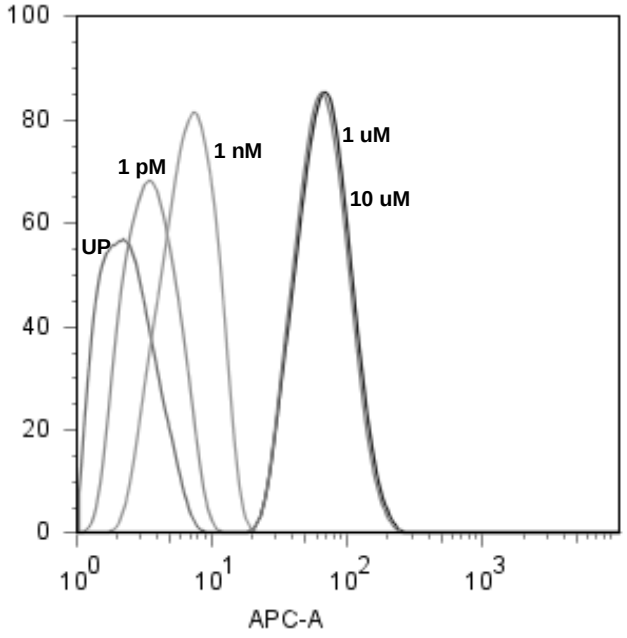


FIGURE 16

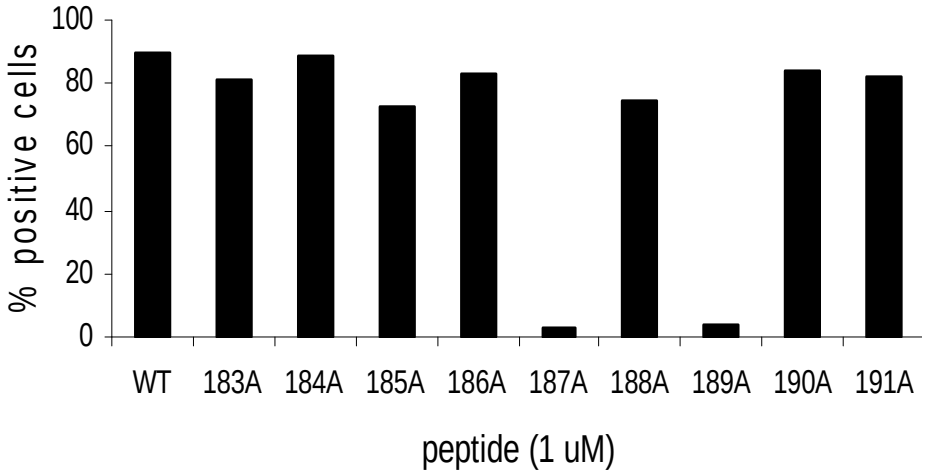


FIGURE 17

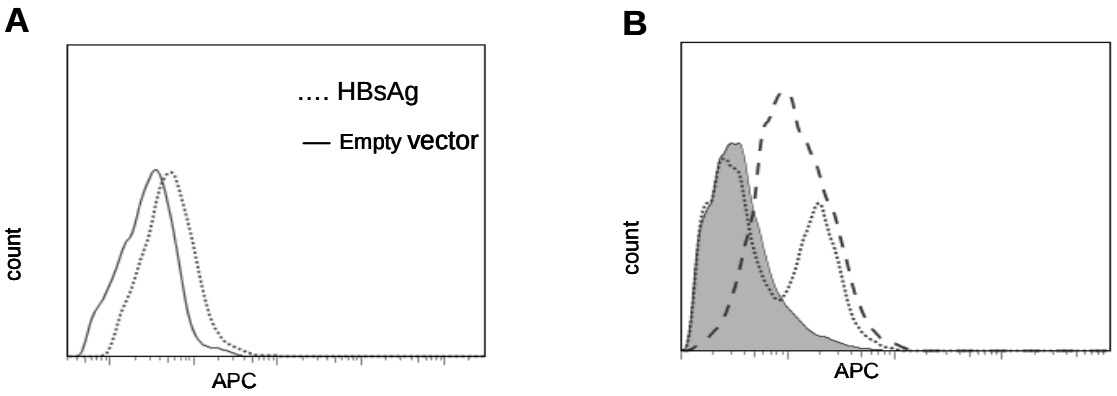


FIGURE 18

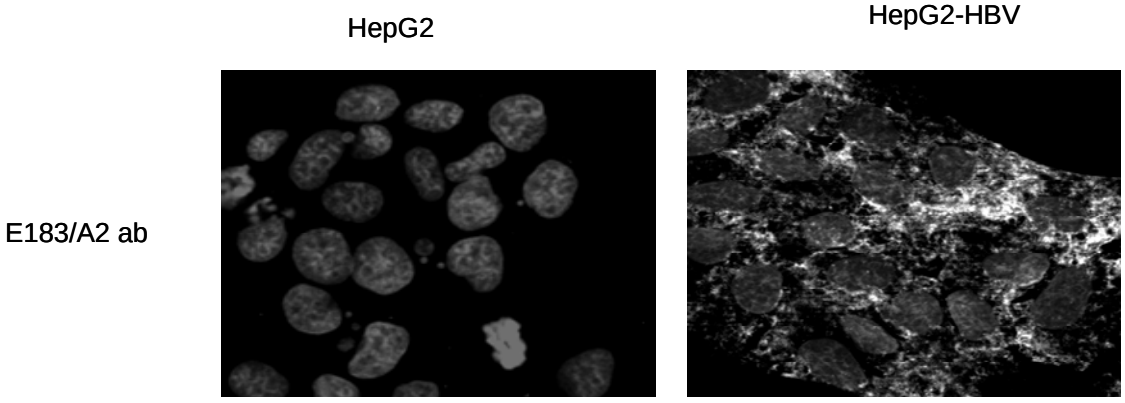


FIGURE 19



FIGURE 20

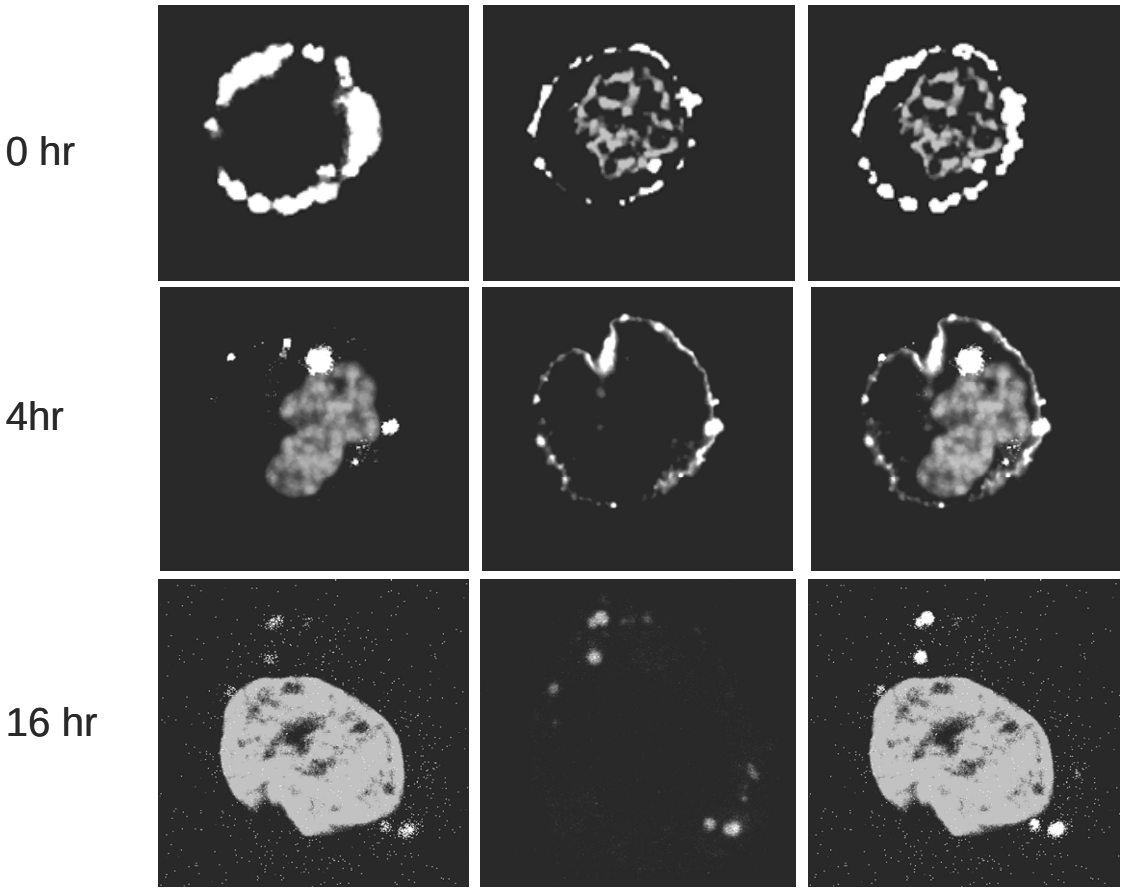


FIGURE 21

SEQUENCE LISTING

<110> Agency for Science, Technology and Research
National University of Singapore

<120> Hepatitis B virus specific antibody and uses thereof

<130> SG3134A

<160> 34

<170> PatentIn version 3.4

<210> 1
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<213> Murinae gen. sp.

<220>
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<223> Amino acid sequence of variable domain of c18/A2 mAb Light Chain

<400> 1

Asp Ile Gln Met Thr Gln Ser Pro Ala Ser Leu Ser Thr Ser Val Gly
1 5 10 15

Glu Thr Val Thr Ile Thr Cys Arg Ala Ser Gly Asn Ile His Asn Tyr
20 25 30

Leu Ala Trp Tyr Gln Gln Lys Gln Gly Lys Ser Pro Gln Leu Leu Val
35 40 45

Tyr Asn Ala Lys Thr Leu Ala Asp Gly Val Pro Ser Arg Phe Ser Gly
50 55 60

Ser Gly Ser Gly Thr Gln Phe Ser Leu Lys Ile Asn Ser Leu Gln Pro
65 70 75 80

Glu Asp Phe Gly Ser Tyr Tyr Cys Gln His Phe Trp Ser Thr Pro Phe
85 90 95

Thr Phe Gly Ser Gly Thr Lys Leu Glu Ile Lys
100 105

<210> 2
<211> 120
<212> PRT
<213> Murinae gen. sp.

<220>
<221> misc_feature
<223> Amino acid sequence of variable domain of c18/A2 mAb Heavy Chain

<400> 2

Gln Val Gln Leu Gln Gln Ser Gly Pro Glu Leu Val Lys Pro Gly Ala
1 5 10 15

Ser Val Lys Ile Ser Cys Lys Ala Ser Gly His Thr Phe Thr Ser Tyr
20 25 30

Asp Ile Asn Trp Val Lys Gln Arg Pro Gly Gln Gly Leu Glu Trp Ile
35 40 45

Gly Trp Ile Tyr Pro Gly Asp Gly Ser Thr Lys Tyr Asn Glu Lys Phe
50 55 60

Lys Gly Lys Ala Thr Leu Thr Ala Asp Lys Ser Ser Ser Thr Ala Tyr
65 70 75 80

Met Gln Leu Ser Ser Leu Thr Ser Glu Asn Ser Ala Val Tyr Phe Cys
85 90 95

Ala Arg Arg Asp Tyr Gly Tyr Ser Tyr Ala Met Asp Tyr Trp Gly Gln
100 105 110

Gly Thr Ser Val Thr Val Ser Ser
115 120

<210> 3
<211> 10
<212> PRT
<213> Hepatitis B virus

<220>
<221> misc_feature
<223> HBc18-27 peptide

<220>
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<223> HBc18-27 peptide (HBV genotype A/D/E/F)

<400> 3

Phe Leu Pro Ser Asp Phe Phe Pro Ser Val
1 5 10

<210> 4
<211> 10
<212> PRT
<213> Hepatitis B virus

<220>
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<223> HBc18-27 peptide (HBV genotype B/C)

<400> 4

Phe Leu Pro Ser Asp Phe Phe Pro Ser Ile
1 5 10

<210> 5
<211> 10
<212> PRT
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<220>
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<400> 5

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<220>
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<400> 9

Phe Leu Pro Asn Asp Phe Phe Pro Ser Val
1 5 10

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1 5 10

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1 5 10

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1 5 10

<210> 13
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<213> Hepatitis B virus

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Phe Leu Pro Ser Asp Phe Phe Pro Gly Val
1 5 10

<210> 14
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<212> PRT
<213> Hepatitis B virus

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<400> 14

Phe Leu Pro Ala Asp Phe Phe Pro Ser Ile
1 5 10

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<211> 10
<212> PRT
<213> Hepatitis B virus

<220>
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1 5 10

<210> 16
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<213> Hepatitis B virus

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<223> 22*91

<400> 16

Phe Leu Pro Ser Asp Thr Asn Met Gly Leu
1 5 10

<210> 17
<211> 9
<212> PRT
<213> Influenza virus

<220>
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<223> M1 peptide/ FEC1 (Flu)

<400> 17

Gly Ile Leu Gly Phe Val Phe Thr Leu
1 5

<210> 18
<211> 9
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<213> Hepatitis B virus

<220>
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<223> HBV X, 52-60

<400> 18

His Leu Ser Leu Arg Gly Leu Pro Val
1 5

<210> 19
<211> 9
<212> PRT
<213> Homo sapiens

<220>
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<223> Mage A10, 254-262

<400> 19

Gly Leu Tyr Asp Gly Met Glu His Leu
1 5

<210> 20
<211> 9
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<213> Homo sapiens

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<223> SSX2, 41-49

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Lys Ala Ser Glu Lys Ile Phe Tyr Val
1 5

<210> 21
<211> 9
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<213> Hepatitis B virus

<220>
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<223> HBV pol, 455-463

<400> 21

Gly Leu Ser Arg Tyr Val Ala Arg Leu
1 5

<210> 22
<211> 9
<212> PRT
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<220>
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<223> Mage3, 271-279

<400> 22

Phe Leu Trp Gly Pro Arg Ala Leu Val
1 5

<210> 23
<211> 9
<212> PRT
<213> Homo sapiens

<220>
<221> Misc_feature
<223> NY-ESO-1, 157-165

<400> 23

Ser Leu Leu Met Trp Ile Thr Gln Ala
1 5

<210> 24
<211> 9
<212> PRT
<213> Hepatitis B virus

<220>
<221> Misc_feature
<223> HBV env, 183-191

<400> 24

Phe Leu Leu Thr Arg Ile Leu Thr Ile
1 5

<210> 25
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<213> Hepatitis B virus

<220>
<221> Misc_feature
<223> HBV env, 348-357

<400> 25

Gly Leu Ser Pro Thr Val Trp Leu Ser Val
1 5 10

<210> 26
<211> 9
<212> PRT
<213> Human cytomegalovirus

<220>
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<223> FEC19 (HCMV)

<400> 26

Asn Leu Val Pro Met Val Ala Thr Val
1 5

<210> 27
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<212> PRT
<213> Epstein-Barr Virus

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<221> Misc_Feature
<223> FEC11 (EBV)

<400> 27

Gly Leu Cys Thr Leu Val Ala Met Leu
1 5

<210> 28
<211> 9
<212> PRT
<213> Epstein-Barr Virus

<220>
<221> Misc_feature
<223> FEC10 (EBV)

<400> 28

Cys Leu Gly Gly Leu Leu Thr Met Val
1 5

<210> 29
<211> 9
<212> PRT
<213> Influenza virus

<220>
<221> Misc_feature
<223> FEC2 (Flu)

<400> 29

Phe Met Tyr Ser Asp Phe His Phe Ile
1 5

<210> 30
<211> 321
<212> DNA
<213> Murinae gen. sp.

<220>
<221> misc_feature
<223> Nucleotide sequence of variable domain of c18/A2 mAb Light Chain

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atcacatgtc gagcaagtgg gaatattcac aattatttag catggtatca gcagaaacag 120

ggaaaatctc ctcagctcct ggtctataat gcaaaaacct tagcagatgg tgtgccatca 180

aggttcagtg gcagtggatc aggaacacaa ttttctctca agatcaacag cctgcagcct 240

gaagattttg ggagttatta ctgtcaacat ttttggagta ctccattcac gttcggctcg 300

gggacaaagt tggaaataaa a 321

<210> 31
<211> 360
<212> DNA
<213> Murinae gen. sp.

<220>
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<223> Nucleotide sequence of variable domain of c18/A2 mAb Heavy Chain

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cctggacagg gacttgagtg gattggatgg atttatcctg gagatggaag tactaagtac 180

aatgagaaat tcaagggcaa ggccacactg actgcagaca aatcctccag cacagcctac 240

atgcagctca gcagcctgac ttctgagaac tctgcagtct atttctgtgc aagaagggac 300

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<210> 32
<211> 108
<212> PRT
<213> Murinae gen. sp.

<220>
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<223> Amino acid sequence of variable domain of e183/A2 mAb Light Chain

<400> 32

Asp Ile Gln Met Thr Gln Ser Pro Ala Ser Leu Ser Ala Ser Val Gly
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Glu Thr Val Thr Ile Thr Cys Arg Ala Ser Glu Asn Ile Tyr Ser Tyr
20 25 30

Leu Ala Trp Tyr His Gln Lys Gln Gly Lys Ser Pro Gln Leu Leu Val
35 40 45

Tyr Ile Ala Lys Thr Leu Ala Glu Gly Val Pro Ser Arg Phe Ser Gly
50 55 60

Ser Gly Ser Gly Thr Gln Phe Ser Leu Lys Ile Asp Ser Leu Gln Pro
65 70 75 80

Glu Asp Val Gly Ser Tyr Tyr Cys Gln His His Tyr Gly Thr Pro Tyr
85 90 95

Thr Phe Gly Gly Gly Thr Lys Leu Glu Ile Lys Arg
100 105

<210> 33
<211> 107
<212> PRT
<213> Murinae gen. sp.

<220>
<221> Misc_feature
<223> Amino acid sequence of variable domain of e183/A2 mAb Heavy Chain

<400> 33

Gln Val Gln Leu Gln Gln Pro Gly Ala Glu Phe Val Lys Pro Gly Ala
1 5 10 15

Ser Val Arg Leu Ser Cys Lys Ala Ser Gly Tyr Thr Phe Ser Ser Tyr
20 25 30

Trp Met His Trp Val Lys Gln Arg Pro Gly Gln Gly Leu Glu Trp Ile
35 40 45

Gly Glu Ile Asp Pro Ser Asp Ser Tyr Thr Asn Tyr Asn Gln Lys Phe
50 55 60

Lys Gly Lys Ala Thr Leu Thr Val Asp Lys Ser Ser Ser Thr Ala Tyr
65 70 75 80

Met Gln Phe Ser Ser Leu Thr Ser Glu Asp Ser Ala Val Tyr Tyr Cys
85 90 95

Ala Thr Ser Gly Tyr Asp Val Gly Val Asp Tyr
100 105

<210> 34
<211> 9
<212> PRT
<213> Hepatitis B virus

<220>
<221> Misc_feature
<223> HBC 19-27I peptide

<400> 34

Leu Pro Ser Asp Phe Phe Pro Ser Ile
1 5