Fab-heteroFc-[scFv*]
(C1mAb)

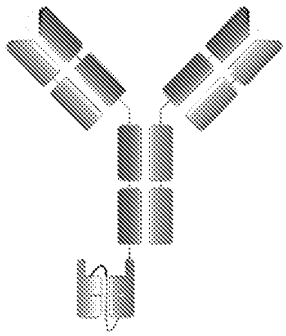


FIG. 1A

heteroIgG

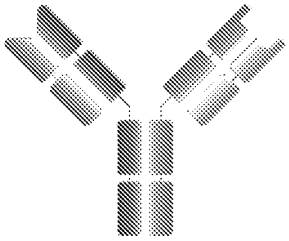


FIG. 1B

Antibody-Cytokine
Fusion, Asymmetric

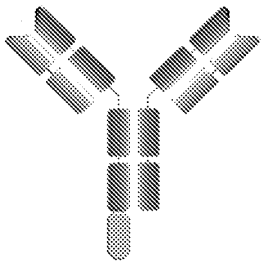


FIG. 1C

[VH-VH*Fab]-heteroFc

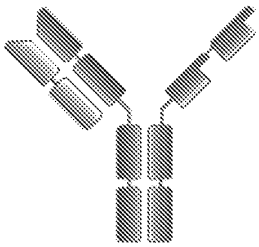


FIG. 1D

[VH*Fab]-heteroFc

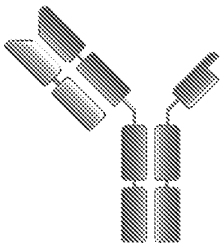


FIG. 1E

[Fab*]-heteroFc-[VH*VH]

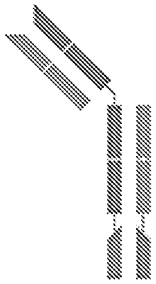


FIG. 1F

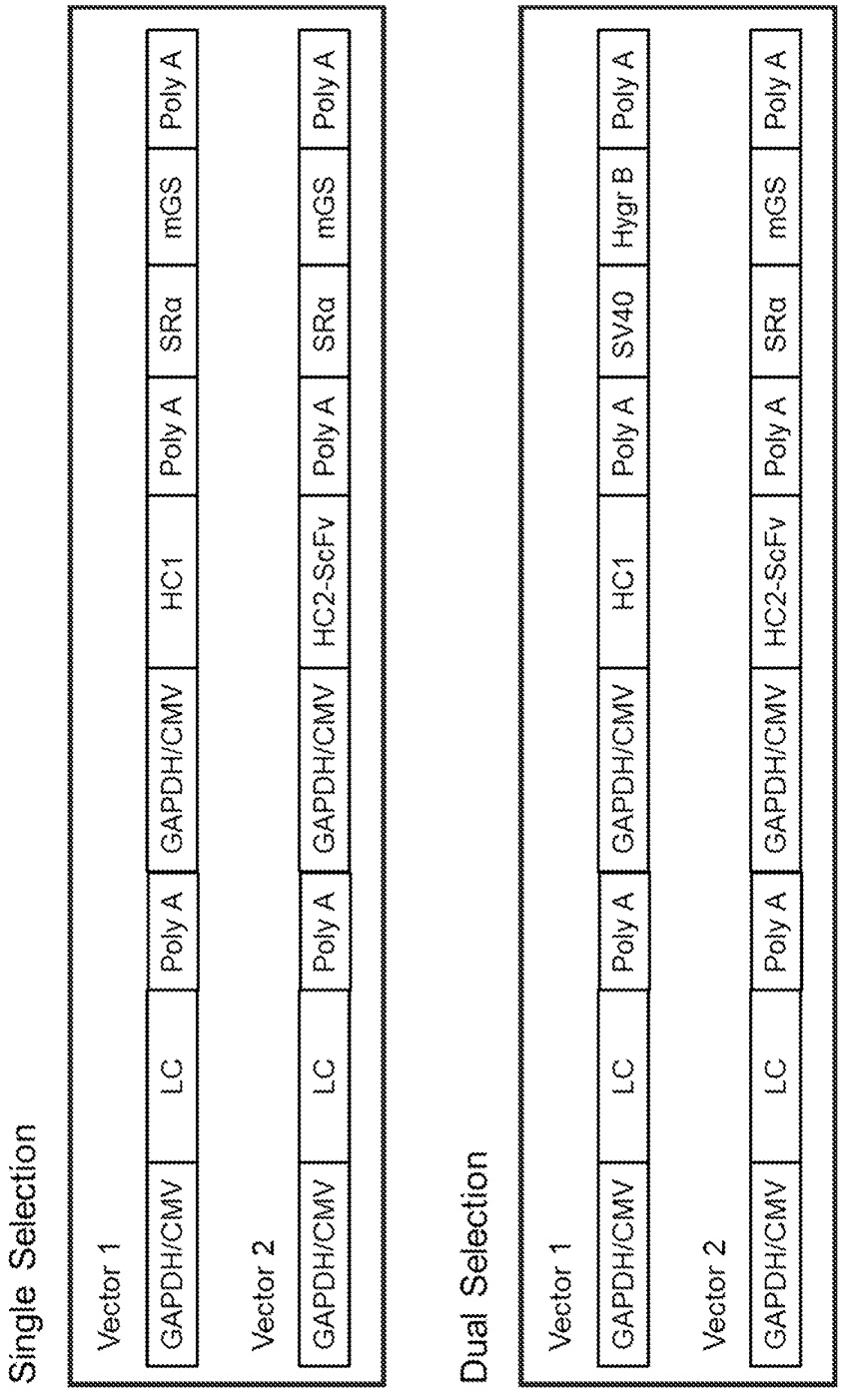

Single Selection
Vector 1
GAPDH/CMV
LC
Poly A
GAPDH/CMV
HC1
Poly A
SRα
mGS
Poly A
Vector 2
GAPDH/CMV
LC
Poly A
GAPDH/CMV
HC2-ScFv
Poly A
SRα
mGS
Poly A
Dual Selection
Vector 1
GAPDH/CMV
LC
Poly A
GAPDH/CMV
HC1
Poly A
SV40
Hygr B
Poly A
Vector 2
GAPDH/CMV
LC
Poly A
GAPDH/CMV
HC2-ScFv
Poly A
SRα
mGS
Poly A

FIG. 2A

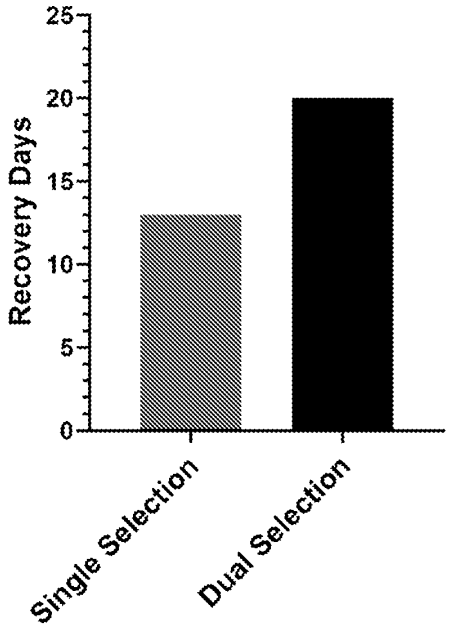

Recovery Days
25
20
15
10
5
0
Single Selection
Dual Selection

FIG. 2B

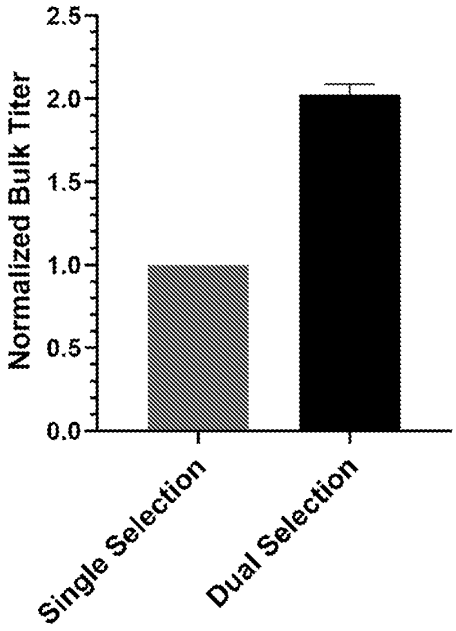

Normalized Bulk Titer
2.5
2.0
1.5
1.0
0.5
0.0
Single Selection
Dual Selection

FIG. 2C

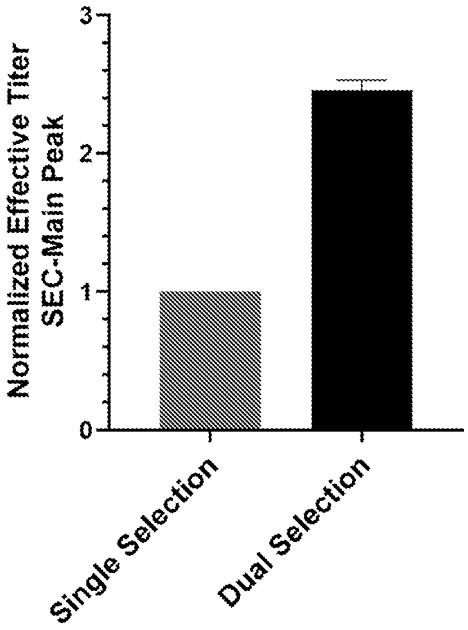

Normalized Effective Titer
SEC-Main Peak
3
2
1
0
Single Selection
Dual Selection

FIG. 2D

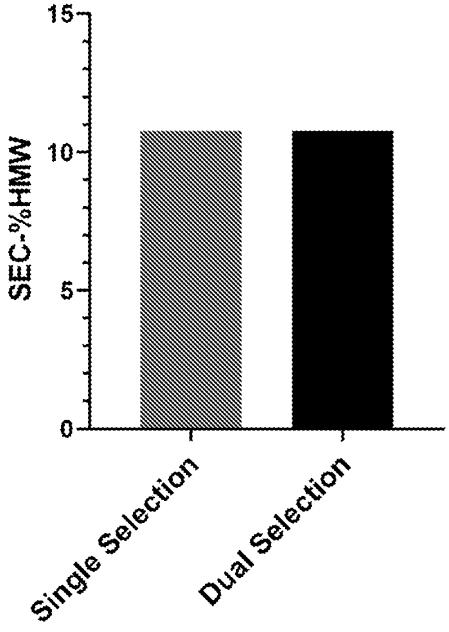

SEC-%HMW
15
10
5
0
Single Selection
Dual Selection

FIG. 2E

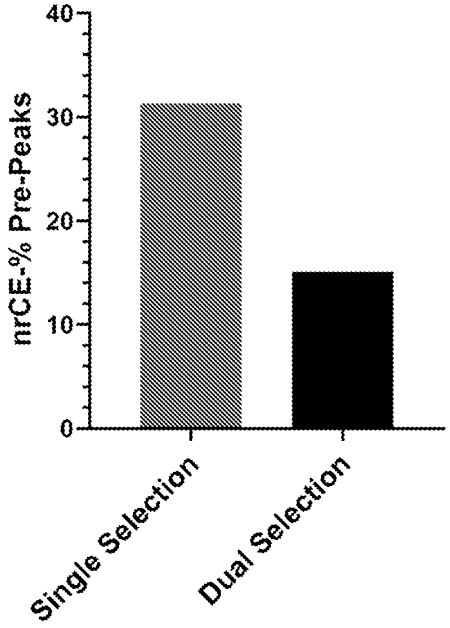

nrCE-% Pre-Peaks
40
30
20
10
0
Single Selection
Dual Selection

FIG. 2F

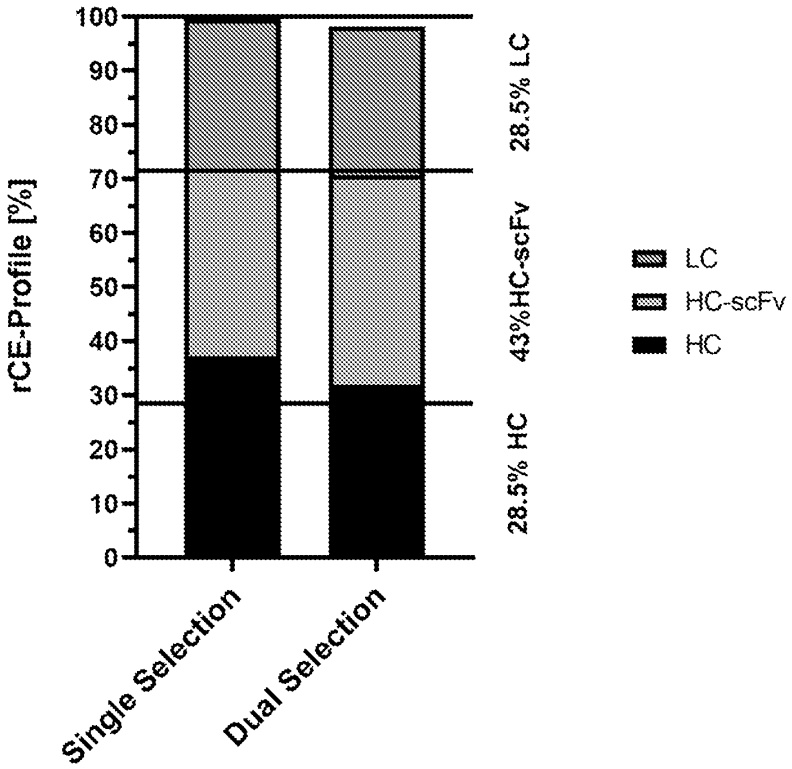

rCE-Profile [%]
100
90
80
70
60
50
40
30
20
10
0
28.5% LC
43%HC-scFv
28.5% HC
LC
HC-scFv
HC
Single Selection
Dual Selection

FIG. 2G

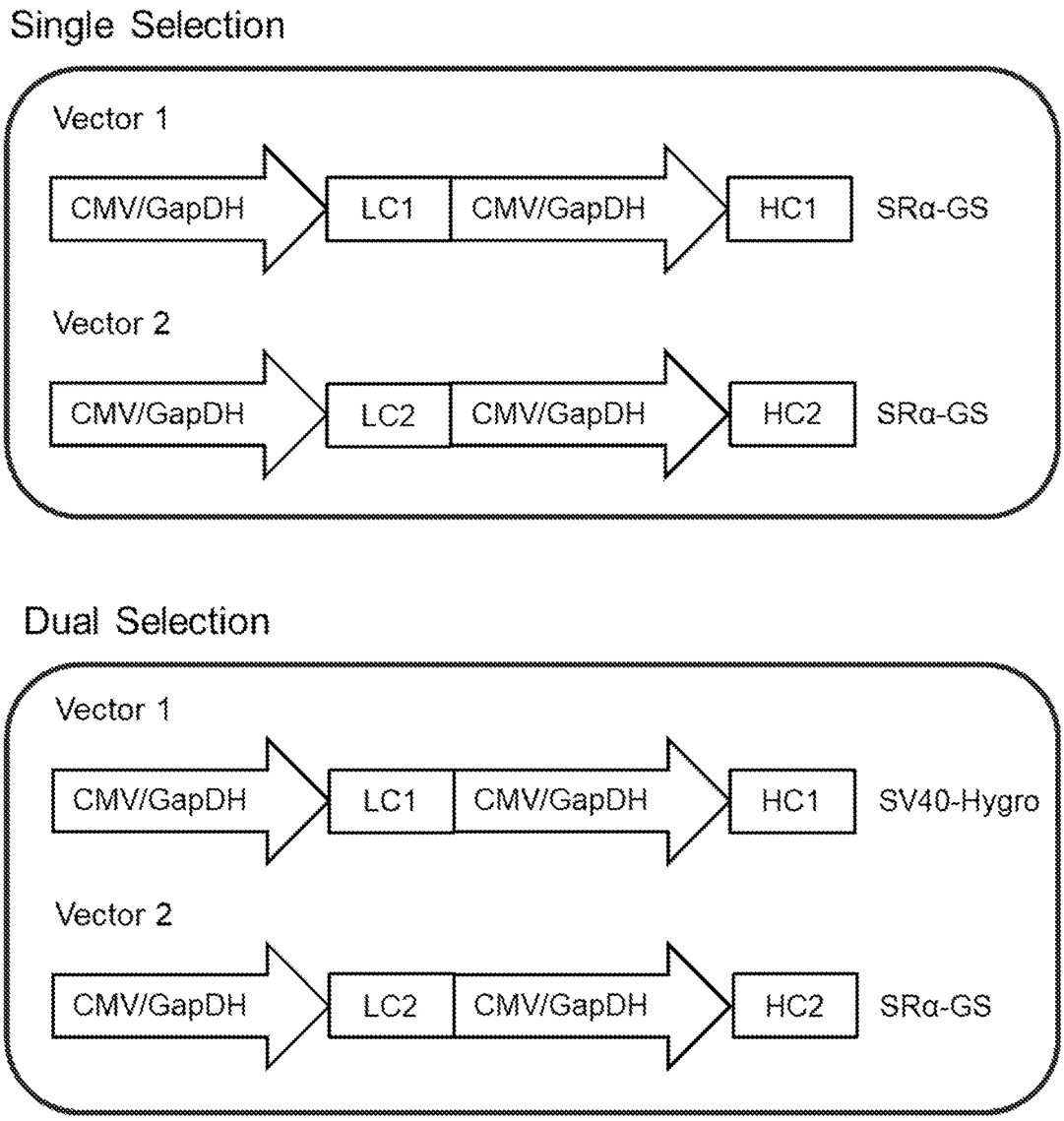

Single Selection
Vector 1
CMV/GapDH
LC1
CMV/GapDH
HC1
SRα-GS
Vector 2
CMV/GapDH
LC2
CMV/GapDH
HC2
SRα-GS
Dual Selection
Vector 1
CMV/GapDH
LC1
CMV/GapDH
HC1
SV40-Hygro
Vector 2
CMV/GapDH
LC2
CMV/GapDH
HC2
SRα-GS

FIG. 3A

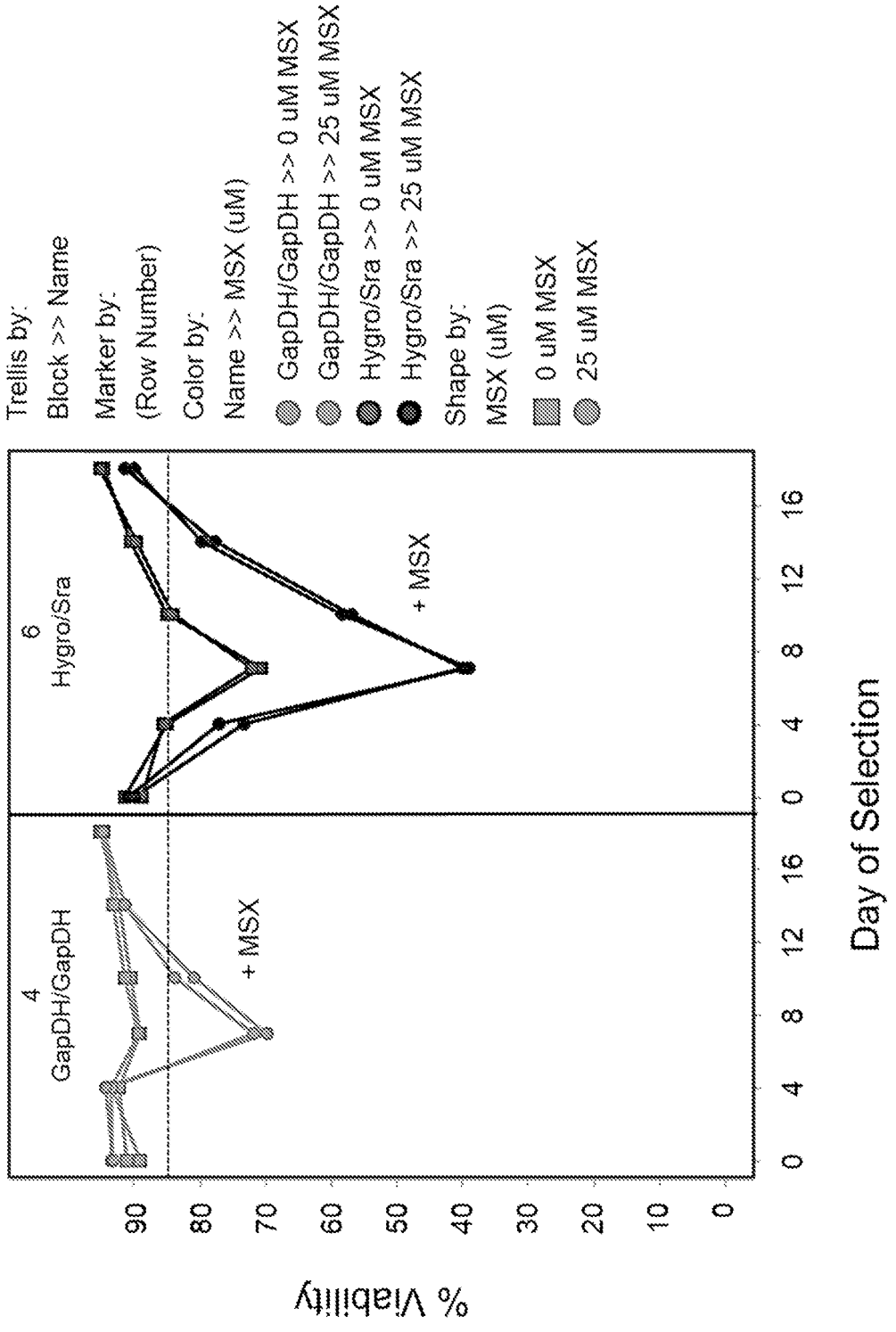

Trellis by:
Block >> Name
Marker by:
(Row Number)
Color by:
Name >> MSX (uM)
GapDH/GapDH >> 0 uM MSX
GapDH/GapDH >> 25 uM MSX
Hygro/Sra >> 0 uM MSX
Hygro/Sra >> 25 uM MSX
Shape by:
MSX (uM)
0 uM MSX
25 uM MSX
4
GapDH/GapDH
6
Hygro/Sra
+ MSX
+ MSX
% Viability
Day of Selection
0
10
20
30
40
50
60
70
80
90
0
4
8
12
16
0
4
8
12
16

FIG. 3B

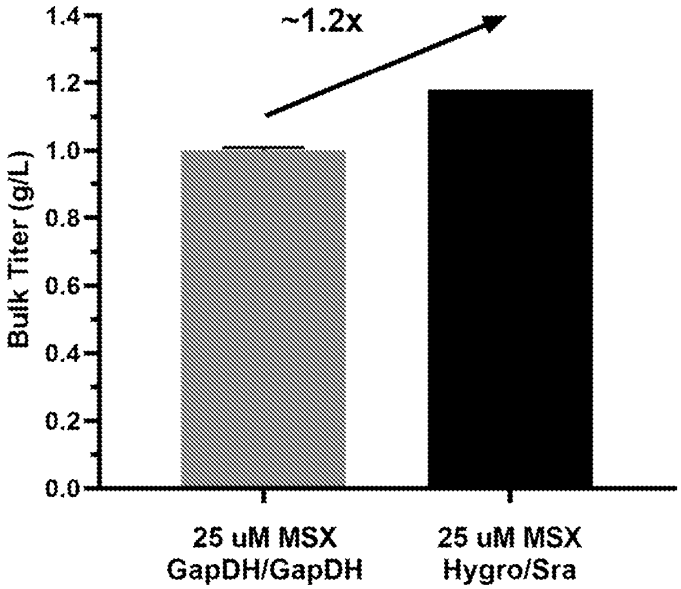

~1.2x
Bulk Titer (g/L)
1.4
1.2
1.0
0.8
0.6
0.4
0.2
0.0
25 uM MSX
GapDH/GapDH
25 uM MSX
Hygro/Sra

FIG. 3C

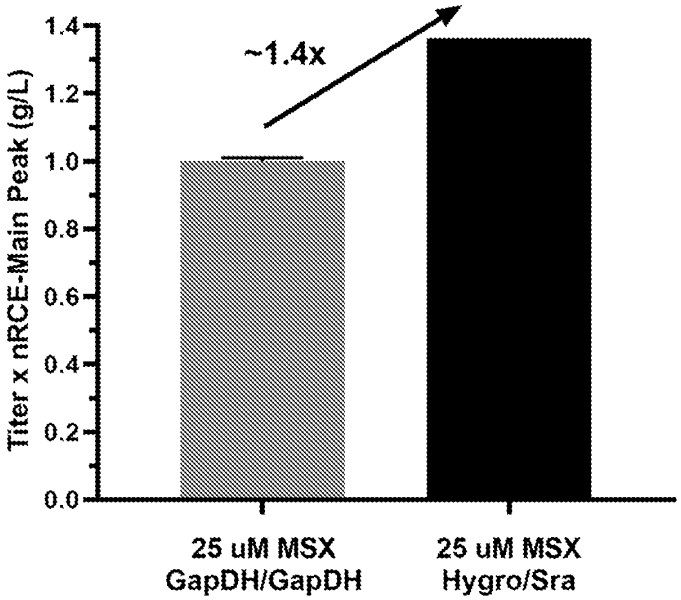

~1.4x
Titer x nRCE-Main Peak (g/L)
1.4
1.2
1.0
0.8
0.6
0.4
0.2
0.0
25 uM MSX
GapDH/GapDH
25 uM MSX
Hygro/Sra

FIG. 3D

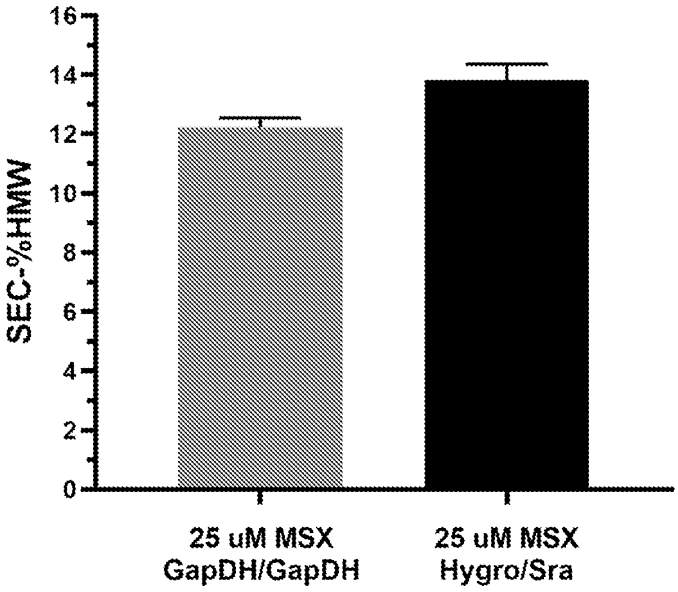

SEC-%HMW
16
14
12
10
8
6
4
2
0
25 uM MSX
GapDH/GapDH
25 uM MSX
Hygro/Sra

FIG. 3E

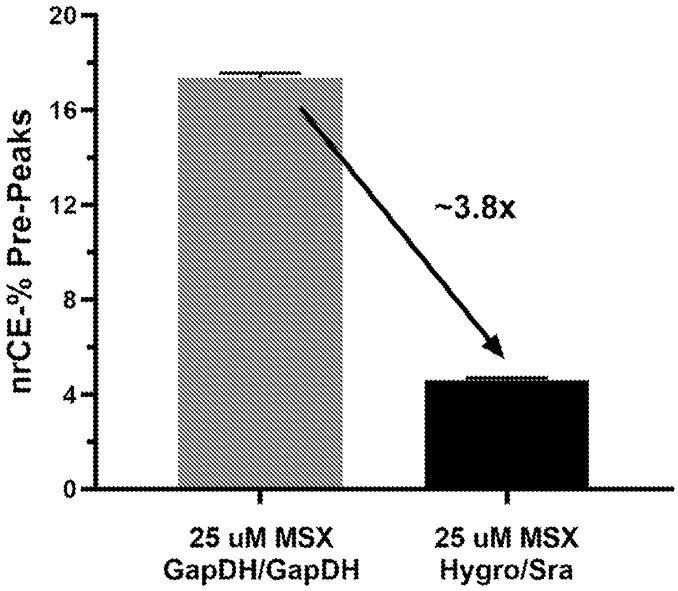
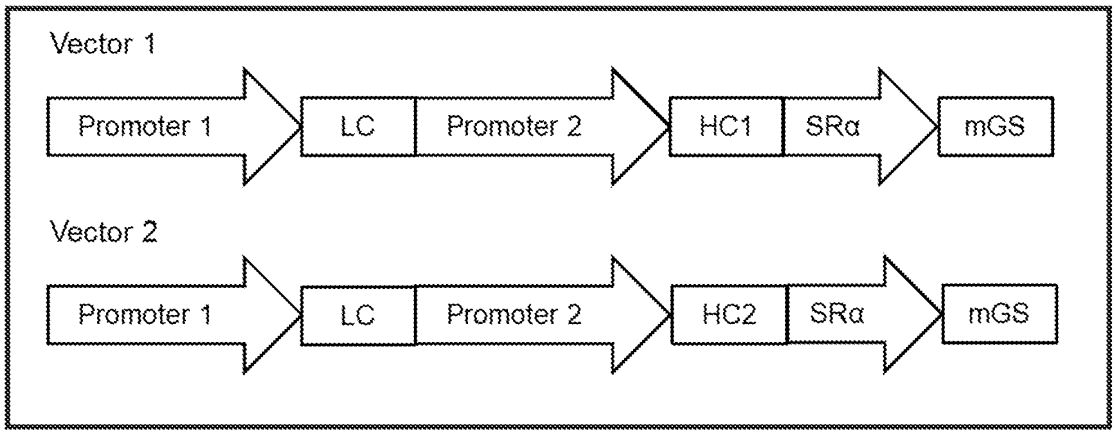

nrCE-% Pre-Peaks
20
16
12
8
4
0
~3.8x
25 uM MSX
GapDH/GapDH
25 uM MSX
Hygro/Sra

FIG. 3F

Single Selection



Dual Selection

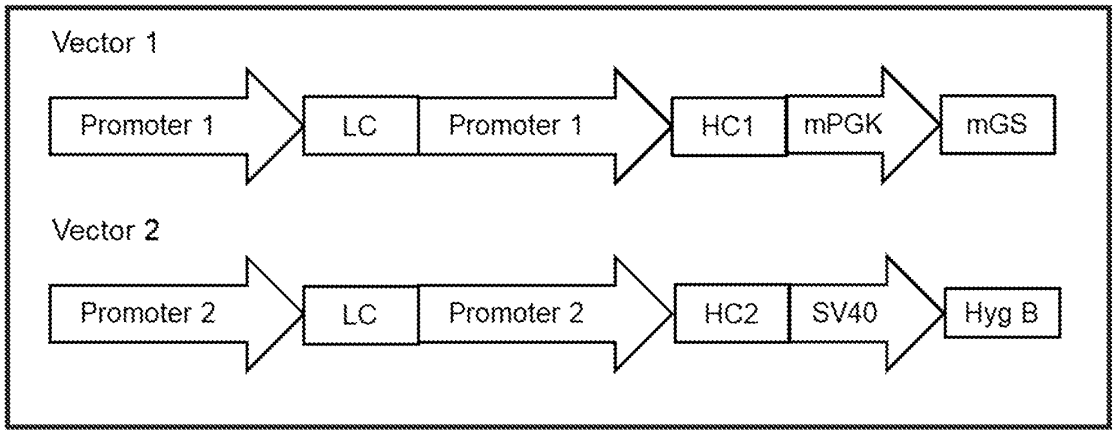


FIG. 4A

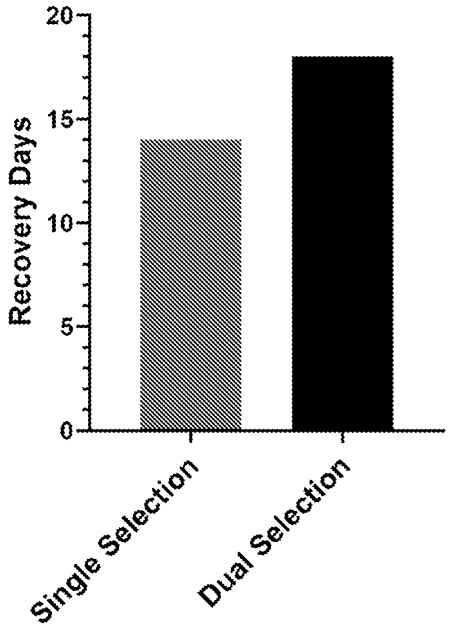

Recovery Days
20
15
10
5
0
Single Selection
Dual Selection

FIG. 4B

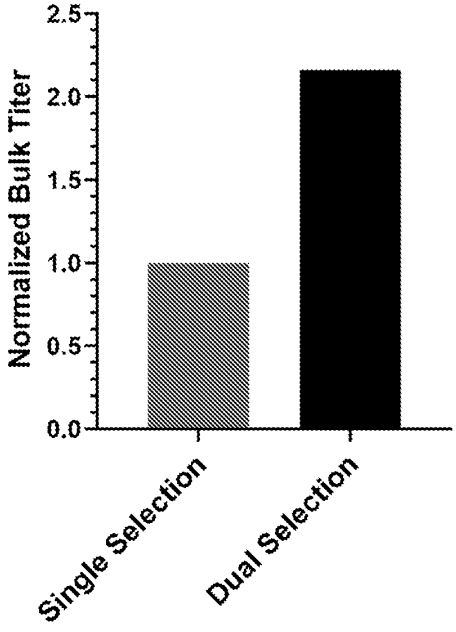

Normalized Bulk Titer
2.5
2.0
1.5
1.0
0.5
0.0
Single Selection
Dual Selection

FIG. 4C

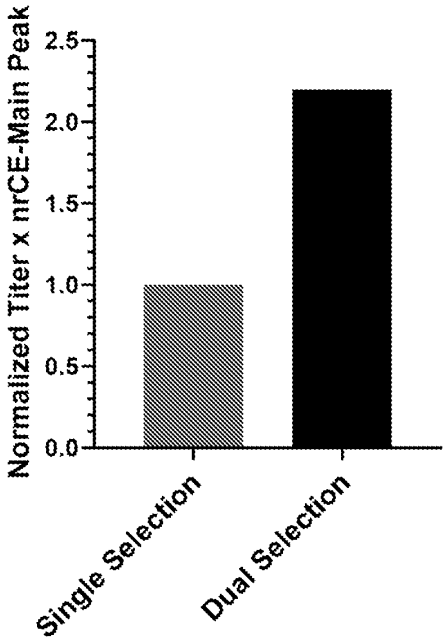

Normalized Titer x nrCE-Main Peak
2.5
2.0
1.5
1.0
0.5
0.0
Single Selection
Dual Selection

FIG. 4D

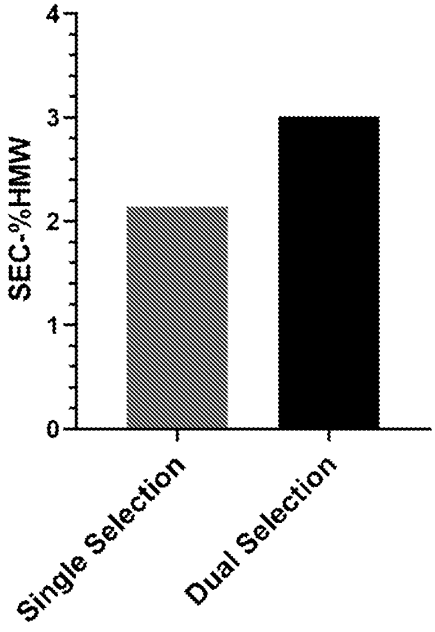

SEC-%HMW
4
3
2
1
0
Single Selection
Dual Selection

FIG. 4E

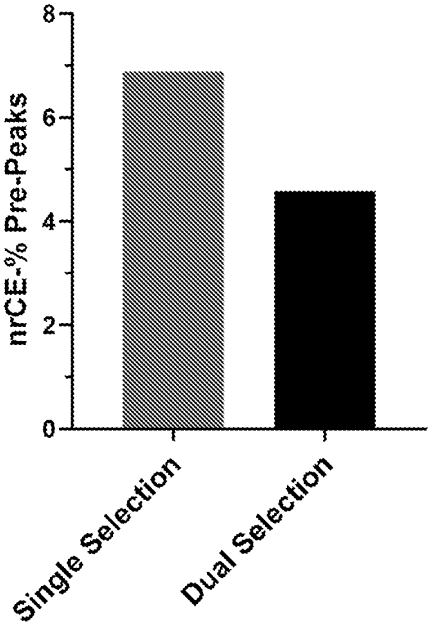

nrCE-% Pre-Peaks
0
2
4
6
8
Single Selection
Dual Selection

FIG. 4F

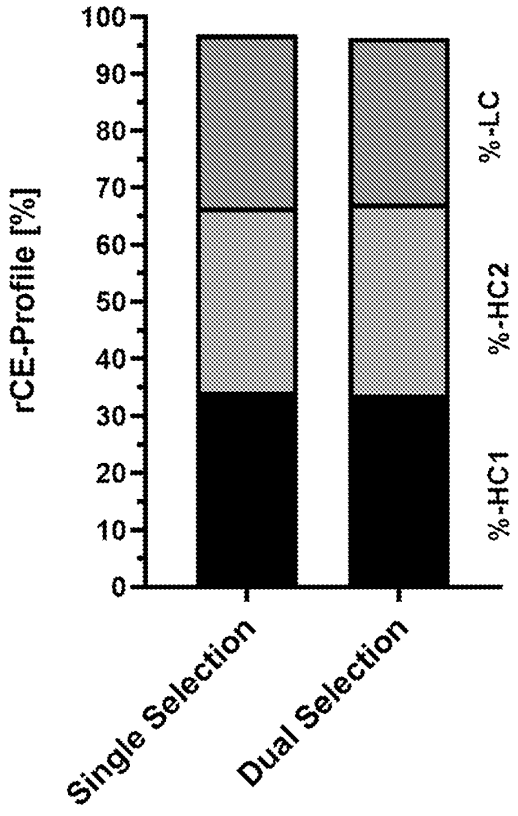

rCE-Profile [%]
0
10
20
30
40
50
60
70
80
90
100
%-HC1
%-HC2
%-LC
Single Selection
Dual Selection

FIG. 4G

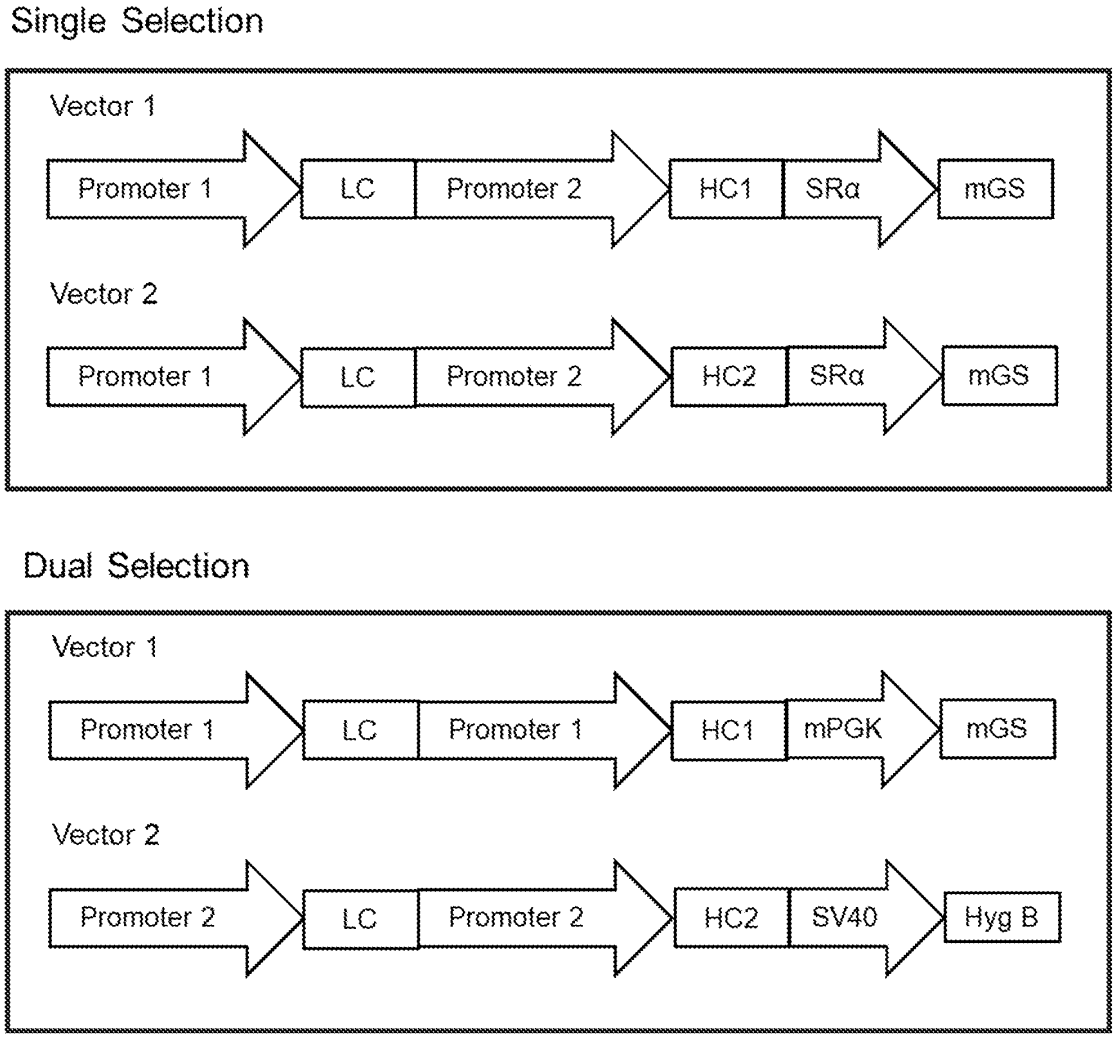

Single Selection
Vector 1
Promoter 1
LC
Promoter 2
HC1
SRα
mGS
Vector 2
Promoter 1
LC
Promoter 2
HC2
SRα
mGS
Dual Selection
Vector 1
Promoter 1
LC
Promoter 1
HC1
mPGK
mGS
Vector 2
Promoter 2
LC
Promoter 2
HC2
SV40
Hyg B

FIG. 5A

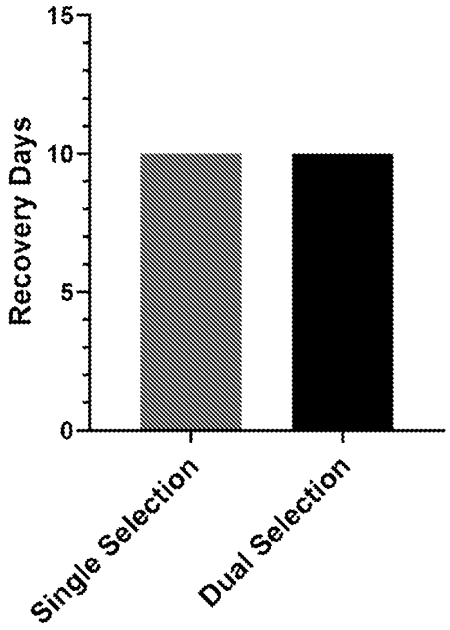

15
10
5
0
Recovery Days
Single Selection
Dual Selection

FIG. 5B

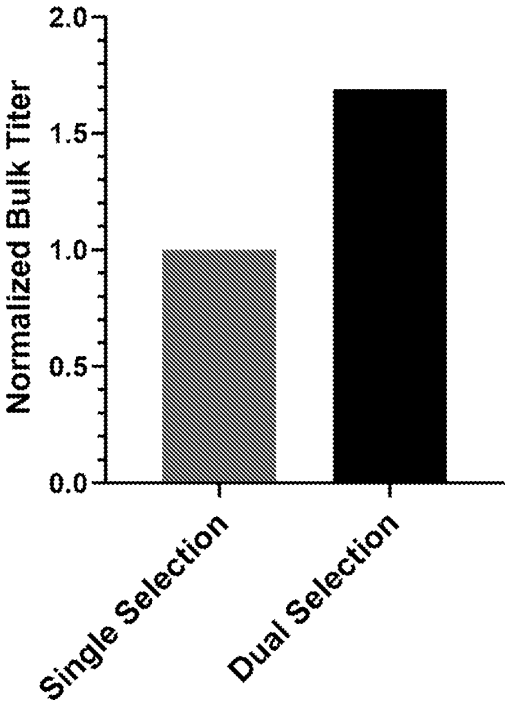

2.0
1.5
1.0
0.5
0.0
Normalized Bulk Titer
Single Selection
Dual Selection

FIG. 5C

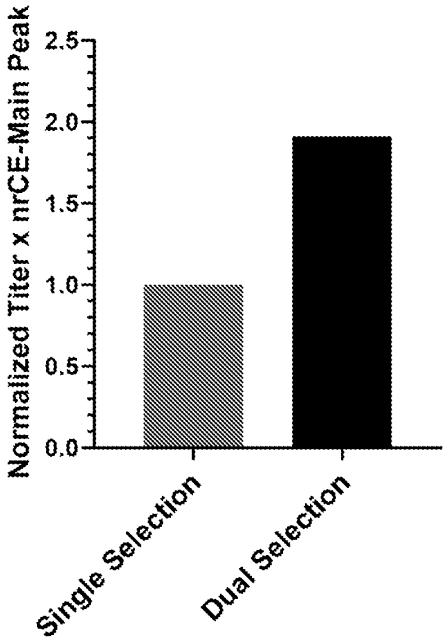

Normalized Titer x nrCE-Main Peak
2.5
2.0
1.5
1.0
0.5
0.0
Single Selection
Dual Selection

FIG. 5D

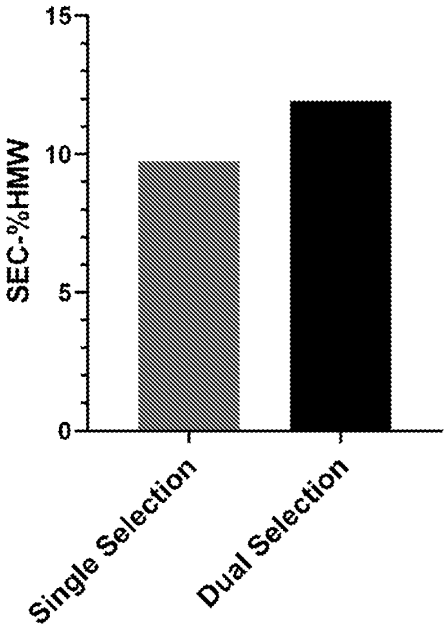

SEC-%HMW
15
10
5
0
Single Selection
Dual Selection

FIG. 5E

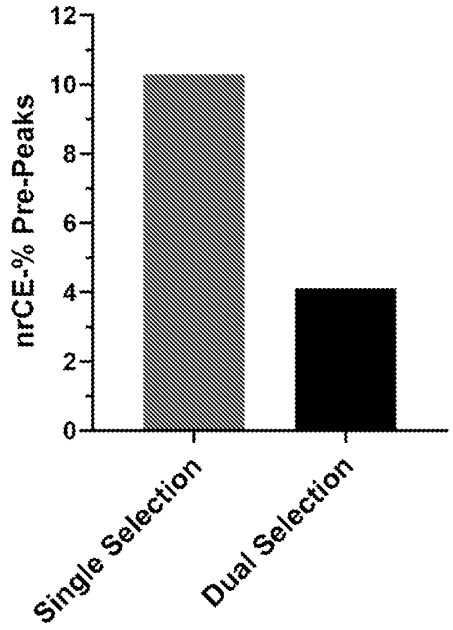

nrCE-% Pre-Peaks
12
10
8
6
4
2
0
Single Selection
Dual Selection

FIG. 5F

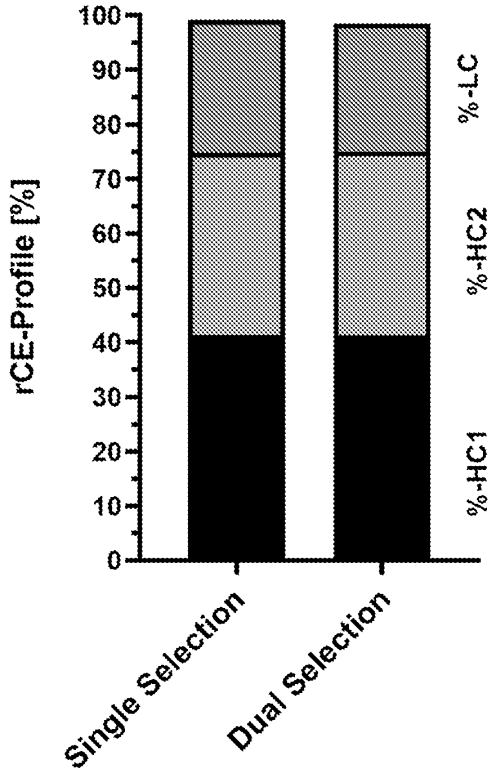

rCE-Profile [%]
100
90
80
70
60
50
40
30
20
10
0
%-LC
%-HC2
%-HC1
Single Selection
Dual Selection

FIG. 5G

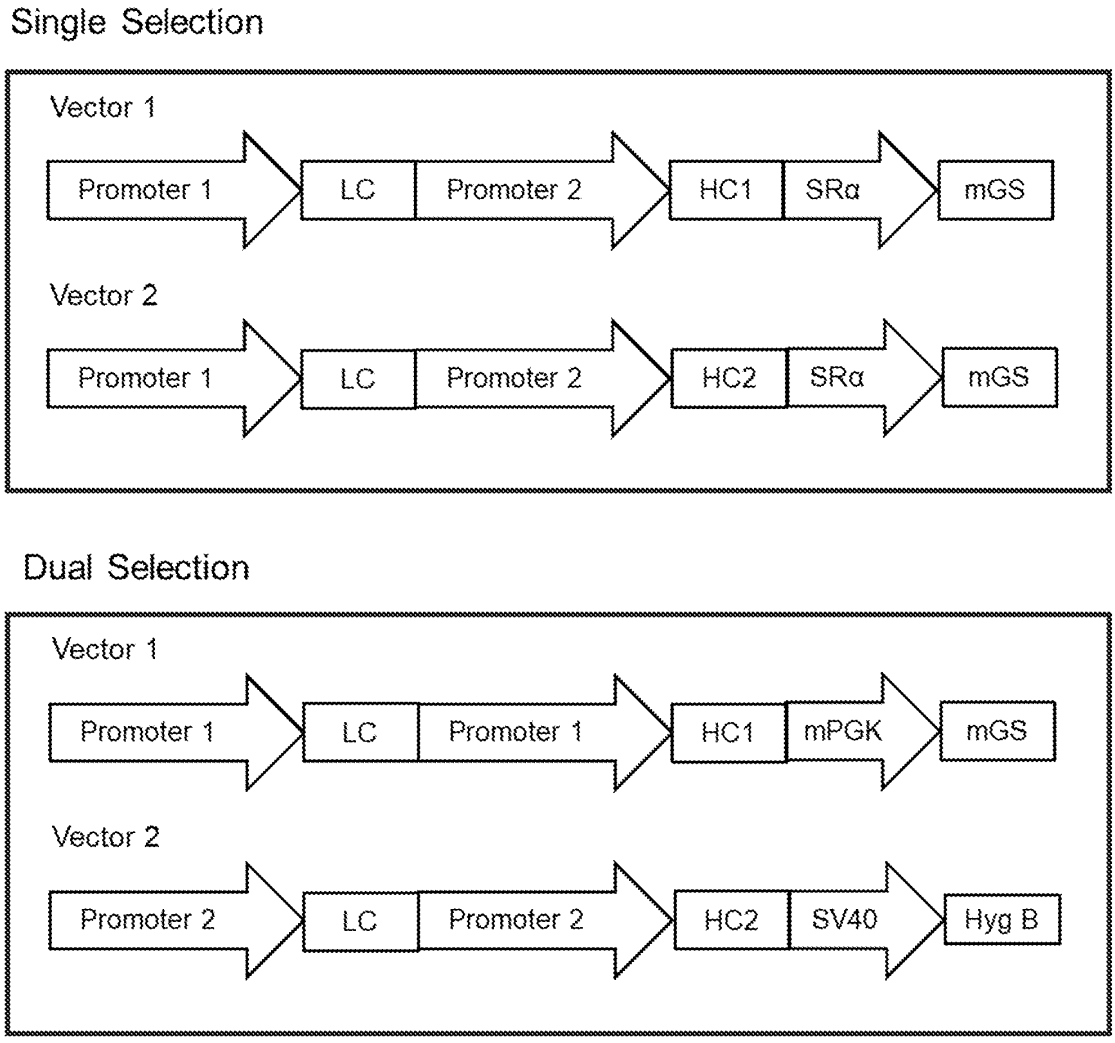

Single Selection
Vector 1
Promoter 1
LC
Promoter 2
HC1
SRα
mGS
Vector 2
Promoter 1
LC
Promoter 2
HC2
SRα
mGS
Dual Selection
Vector 1
Promoter 1
LC
Promoter 1
HC1
mPGK
mGS
Vector 2
Promoter 2
LC
Promoter 2
HC2
SV40
Hyg B

FIG. 6A

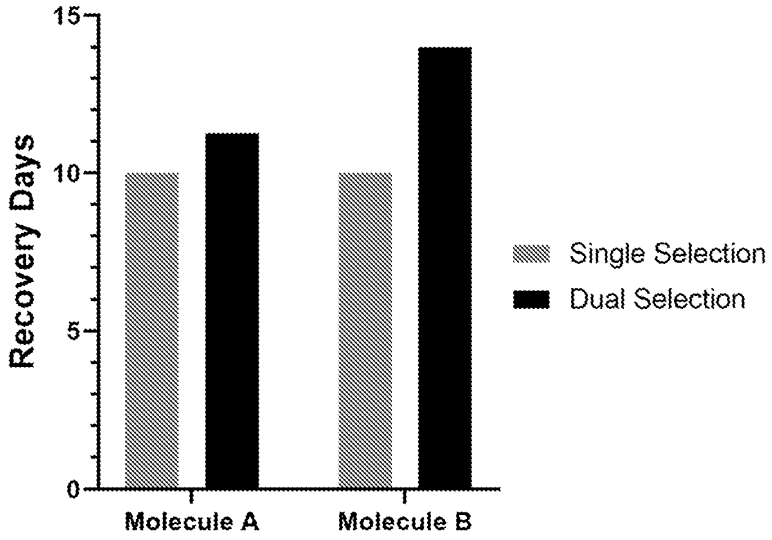

15
10
5
0
Recovery Days
Molecule A
Molecule B
Single Selection
Dual Selection

FIG. 6B

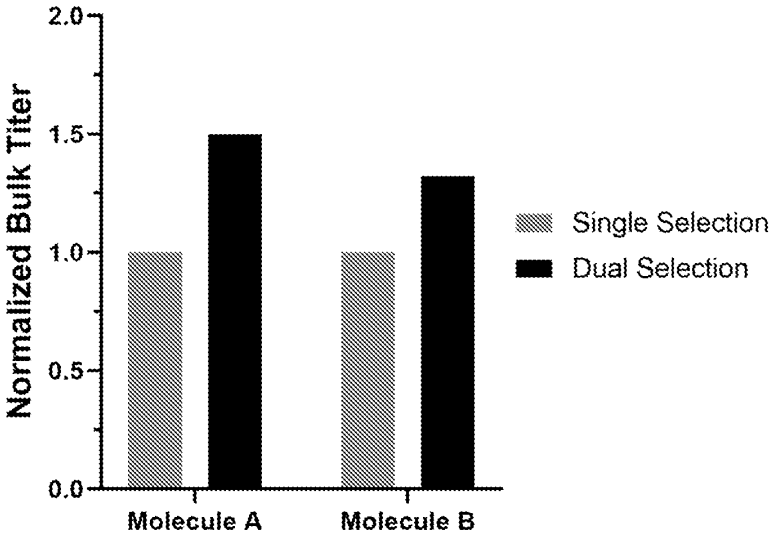

2.0
1.5
1.0
0.5
0.0
Normalized Bulk Titer
Molecule A
Molecule B
Single Selection
Dual Selection

FIG. 6C

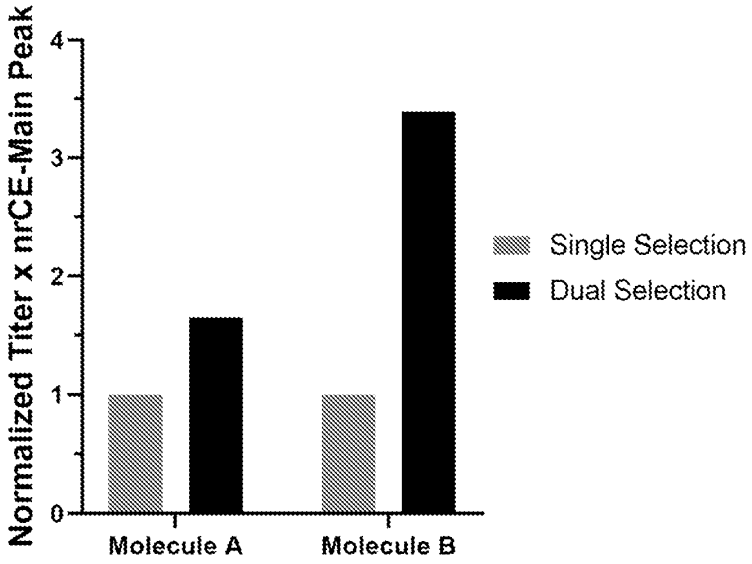

Normalized Titer x nrCE-Main Peak
0
1
2
3
4
Molecule A
Molecule B
Single Selection
Dual Selection

FIG. 6D

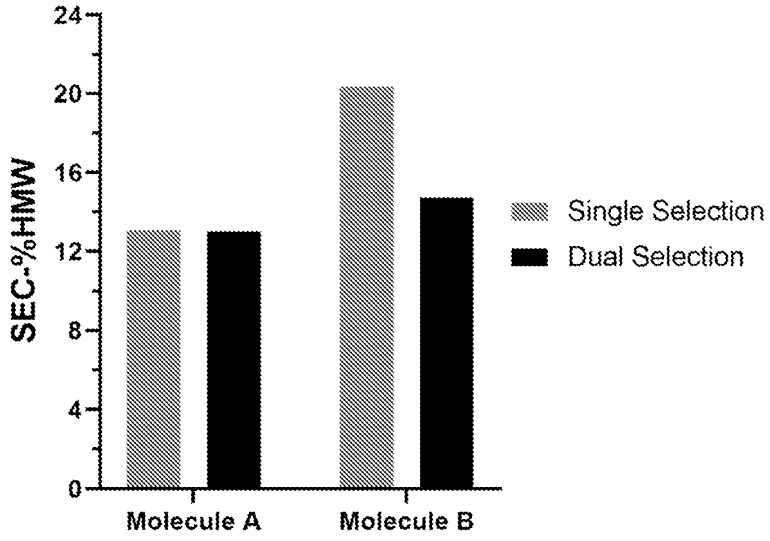

SEC-%HMW
0
4
8
12
16
20
24
Molecule A
Molecule B
Single Selection
Dual Selection

FIG. 6E

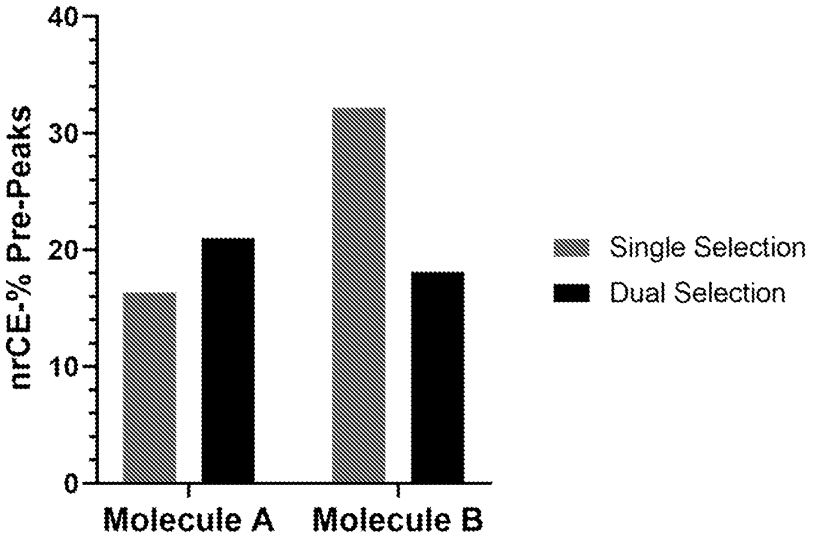

40
30
20
10
0
nrCE-% Pre-Peaks
Molecule A
Molecule B
Single Selection
Dual Selection

FIG. 6F

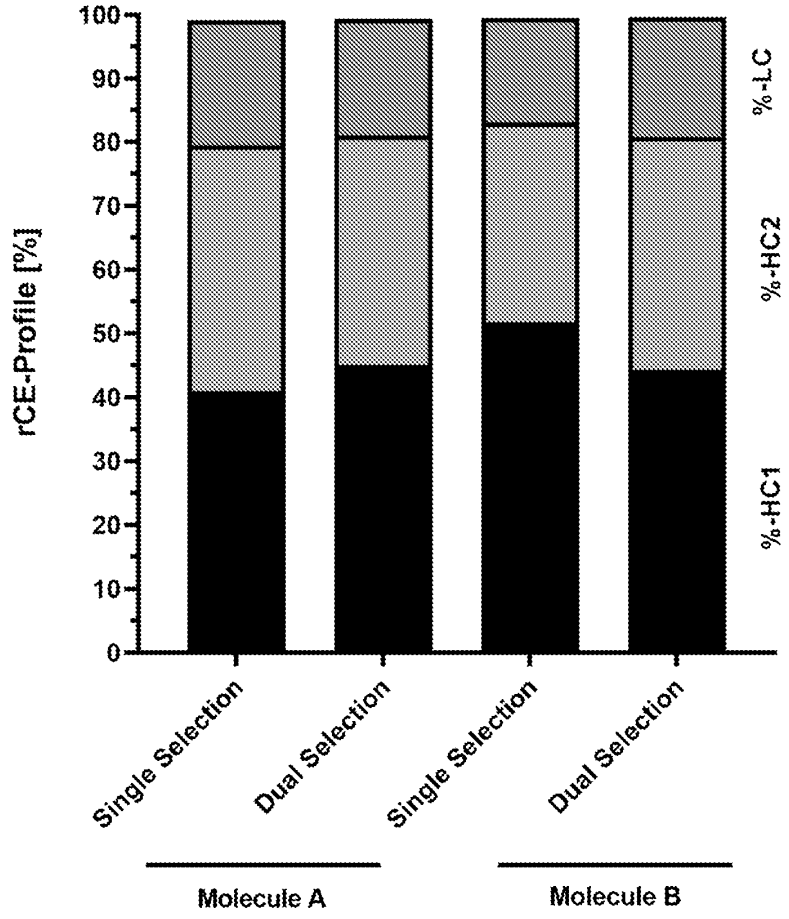

100
90
80
70
60
50
40
30
20
10
0
rCE-Profile [%]
%-LC
%-HC2
%-HC1
Single Selection
Dual Selection
Single Selection
Dual Selection
Molecule A
Molecule B

FIG. 6G

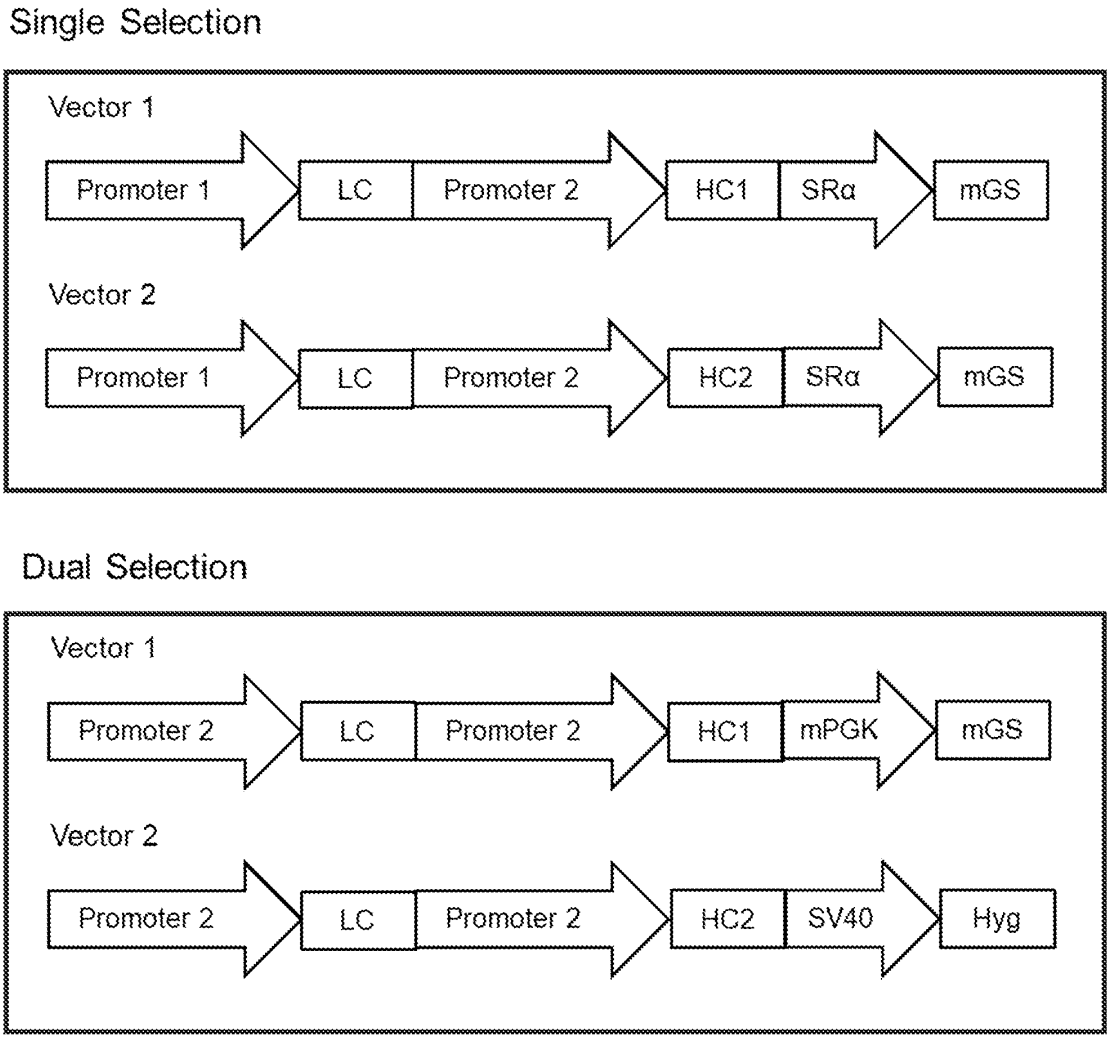

Single Selection
Vector 1
Promoter 1
LC
Promoter 2
HC1
SRα
mGS
Vector 2
Promoter 1
LC
Promoter 2
HC2
SRα
mGS
Dual Selection
Vector 1
Promoter 2
LC
Promoter 2
HC1
mPGK
mGS
Vector 2
Promoter 2
LC
Promoter 2
HC2
SV40
Hyg

FIG. 7A

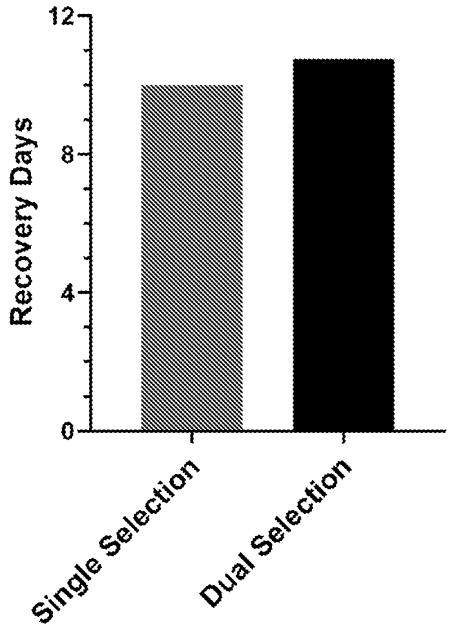

12
8
4
0
Recovery Days
Single Selection
Dual Selection

FIG. 7B

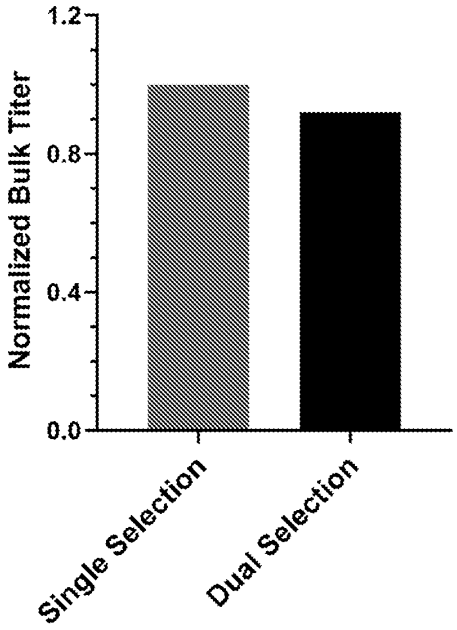

1.2
0.8
0.4
0.0
Normalized Bulk Titer
Single Selection
Dual Selection

FIG. 7C

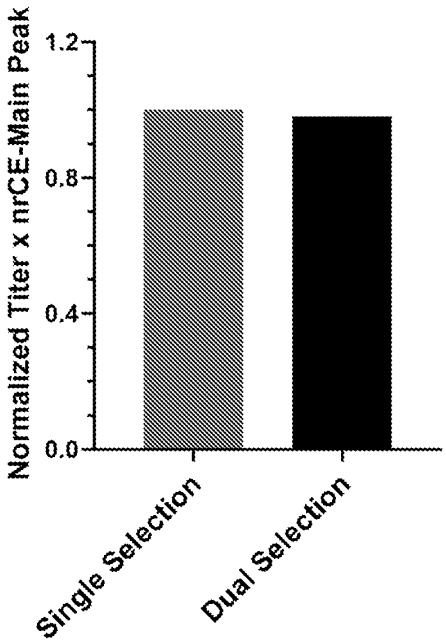

Normalized Titer x nrCE-Main Peak
1.2
0.8
0.4
0.0
Single Selection
Dual Selection

FIG. 7D

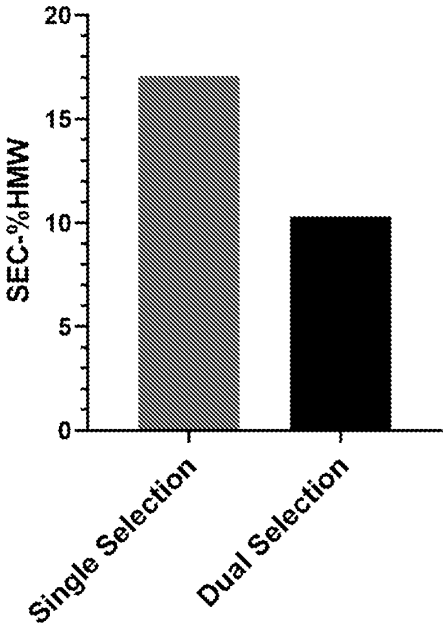

SEC-%HMW
20
15
10
5
0
Single Selection
Dual Selection

FIG. 7E

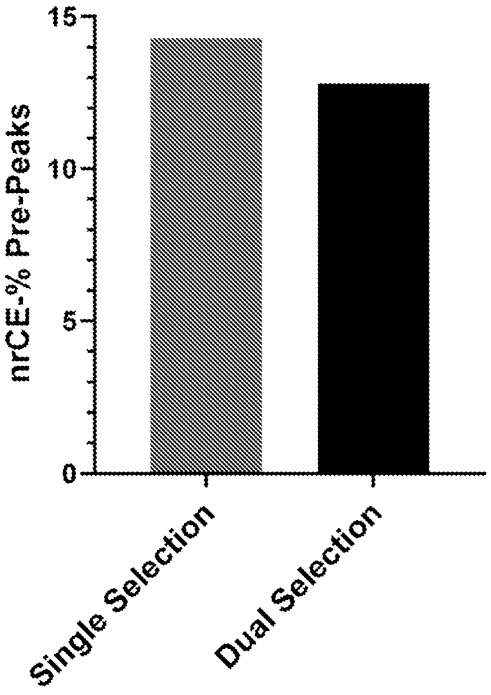

15
10
5
0
nrCE-% Pre-Peaks
Single Selection
Dual Selection

FIG. 7F

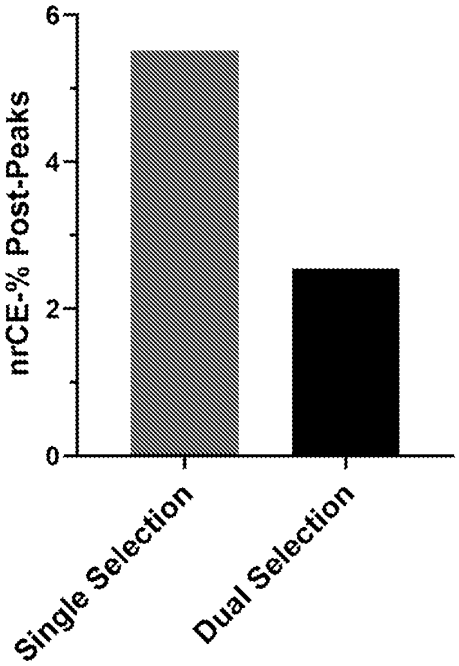

6
4
2
0
nrCE-% Post-Peaks
Single Selection
Dual Selection

FIG. 7G

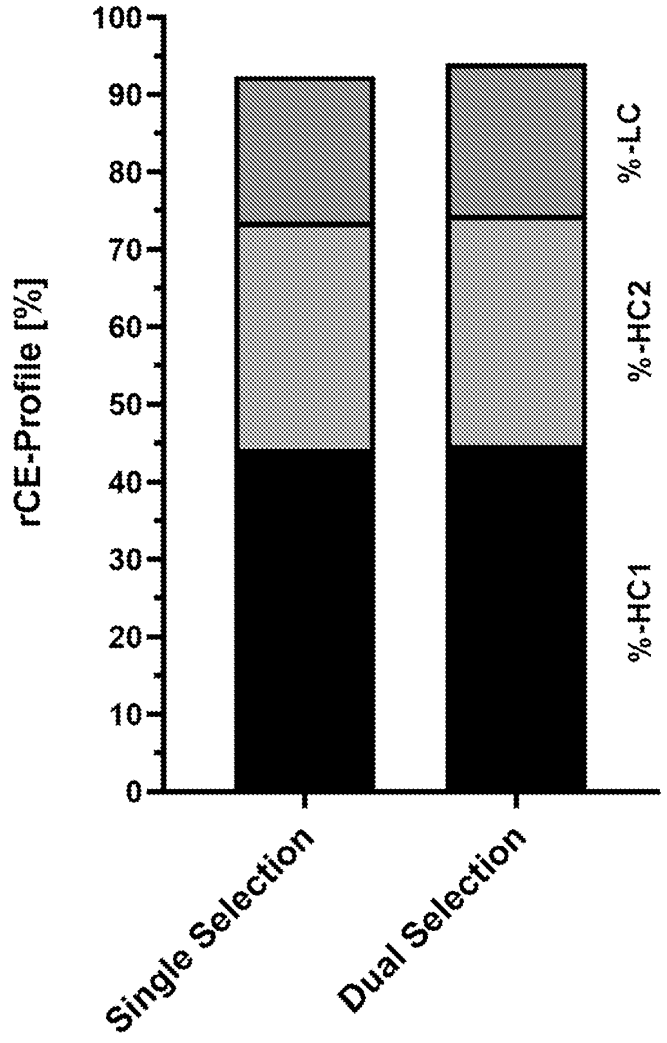

100
90
80
70
60
50
40
30
20
10
0
rCE-Profile [%]
%-LC
%-HC2
%-HC1
Single Selection
Dual Selection

FIG. 7H