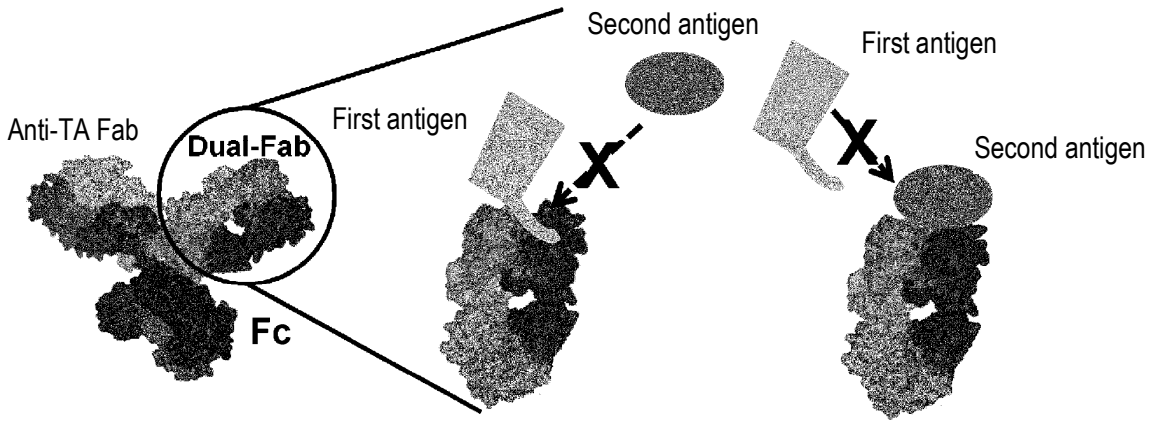

Second antigen
First antigen
Anti-TA Fab
Dual-Fab
First antigen
Second antigen
Fc

FIG. 1

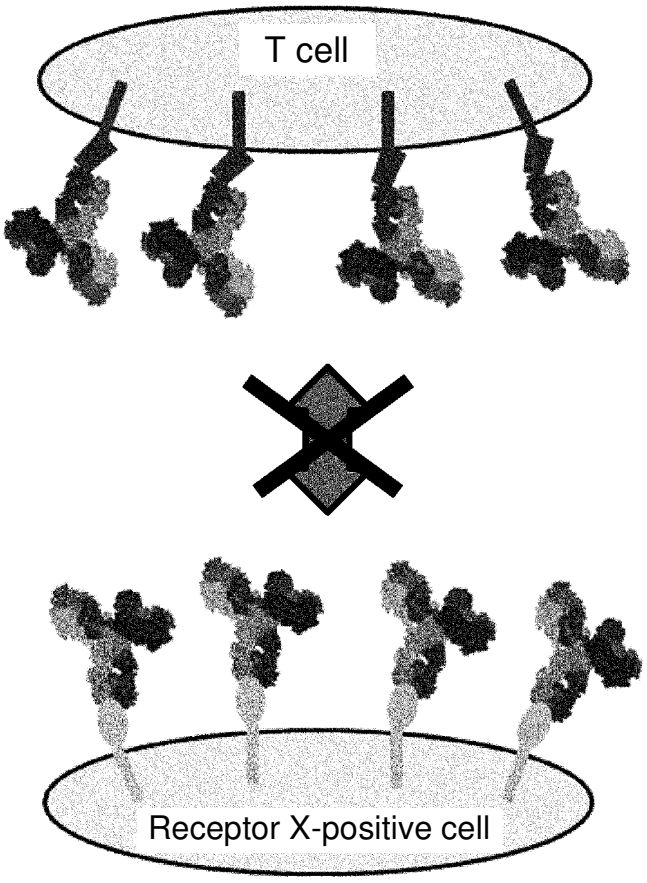


FIG. 2

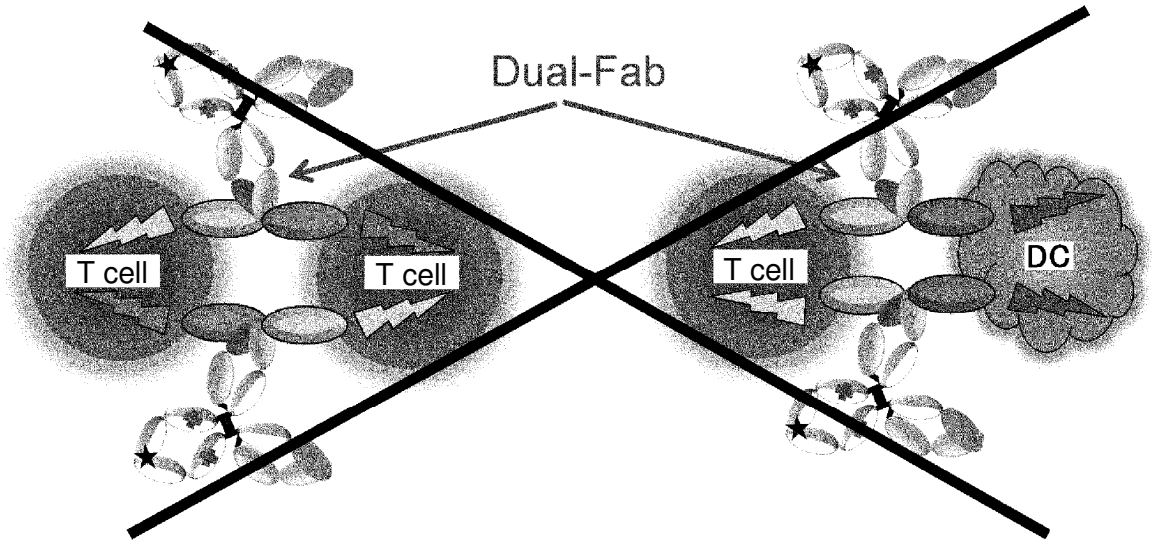


FIG. 3

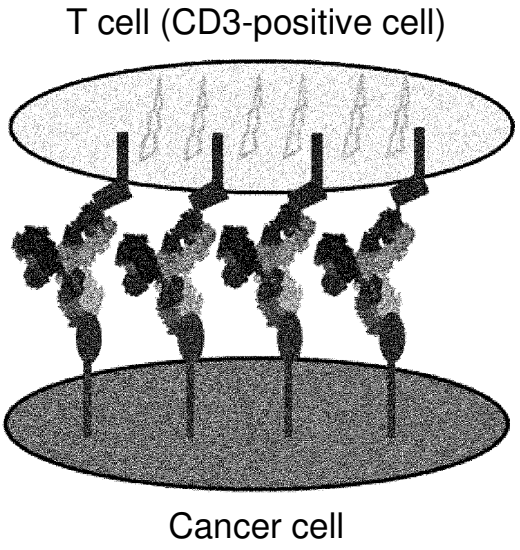


FIG. 4

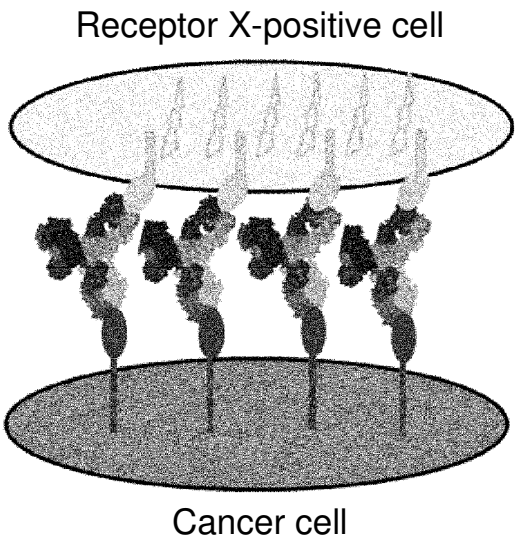


FIG. 5

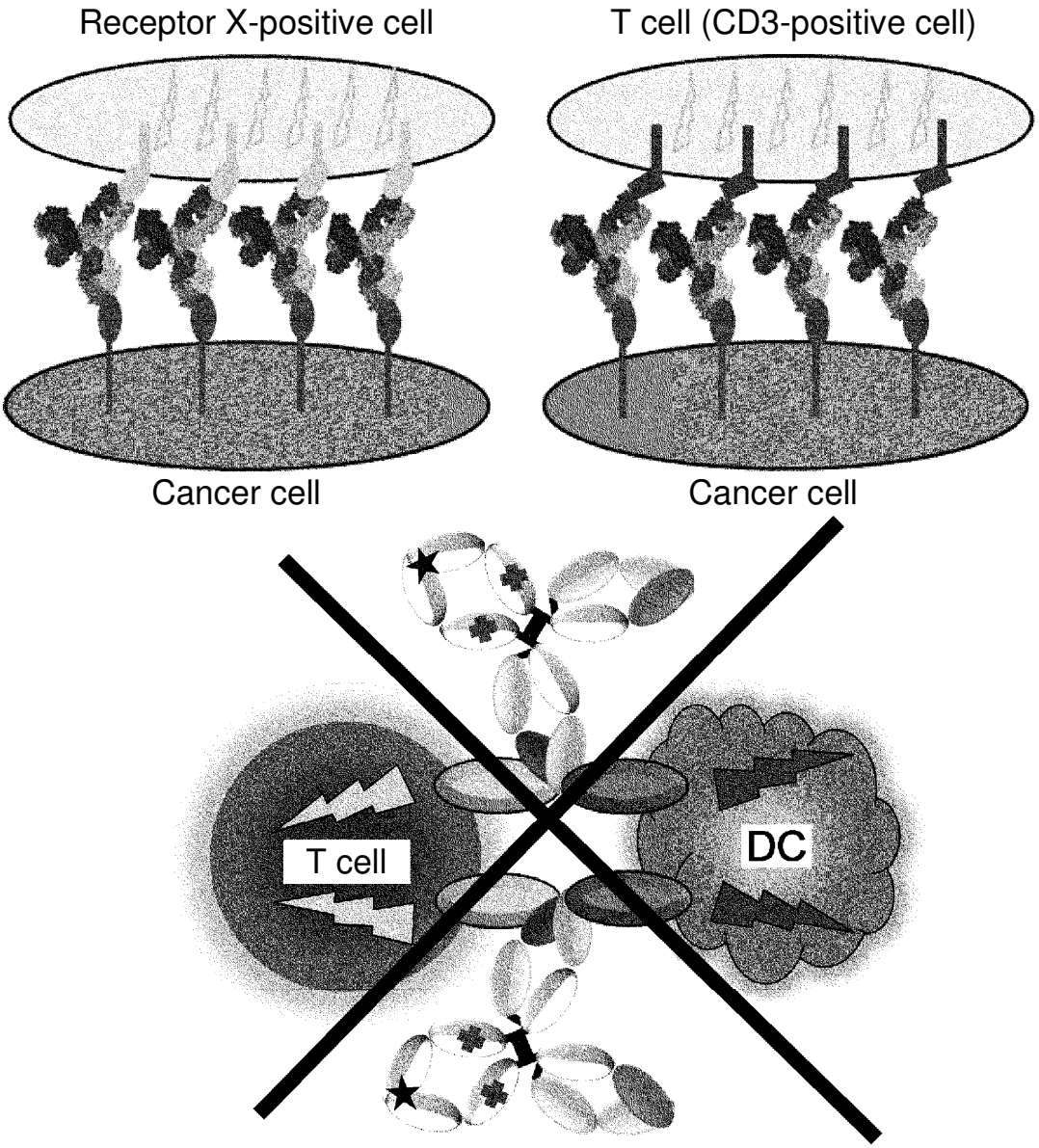

Receptor X-positive cell
T cell (CD3-positive cell)
Cancer cell
Cancer cell
T cell
DC

FIG. 6

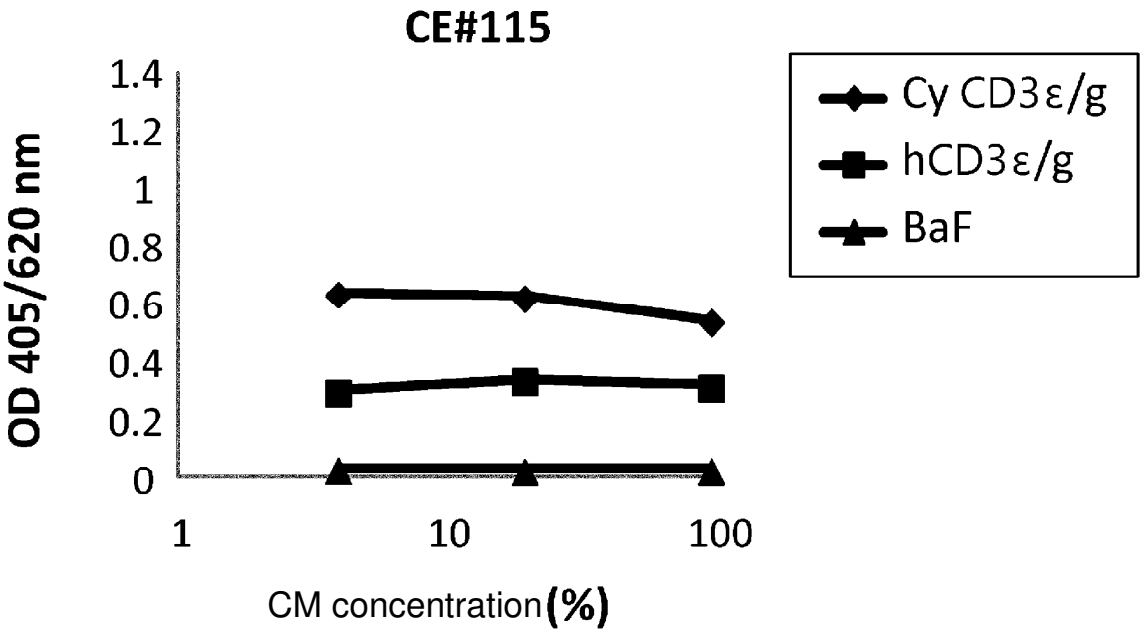

CE#115
OD 405/620 nm
1.4
1.2
1
0.8
0.6
0.4
0.2
0
1
10
100
CM concentration (%)
Cy CD3ε/g
hCD3ε/g
BaF

FIG. 7

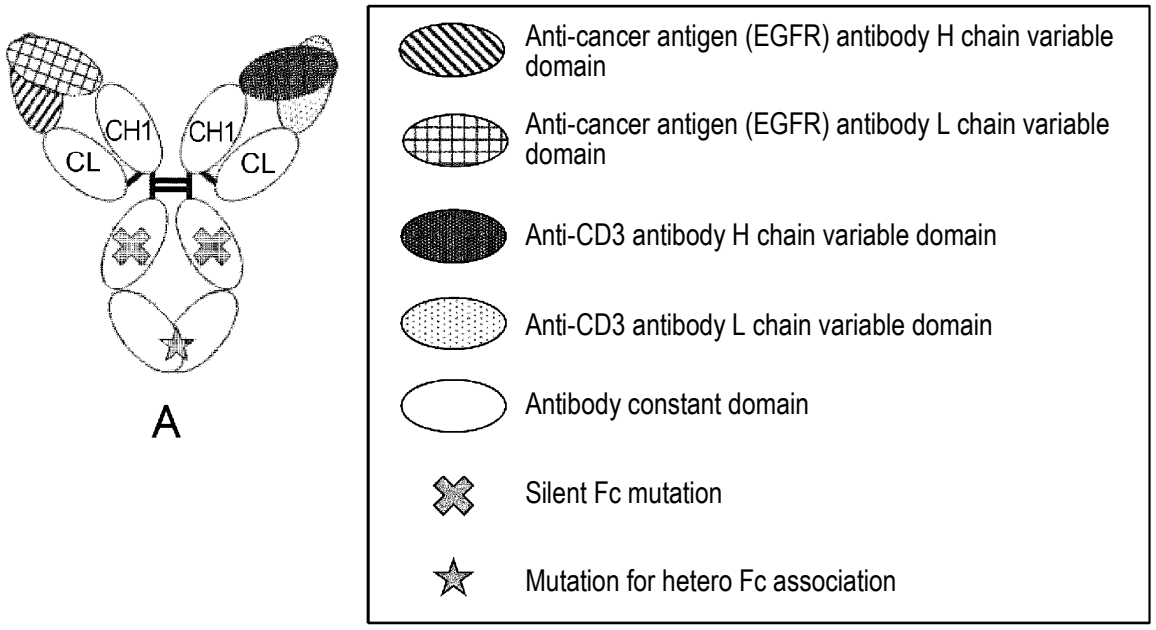

CH1
CL
CH1
CL
A
Anti-cancer antigen (EGFR) antibody H chain variable domain
Anti-cancer antigen (EGFR) antibody L chain variable domain
Anti-CD3 antibody H chain variable domain
Anti-CD3 antibody L chain variable domain
Antibody constant domain
Silent Fc mutation
Mutation for hetero Fc association

FIG. 8

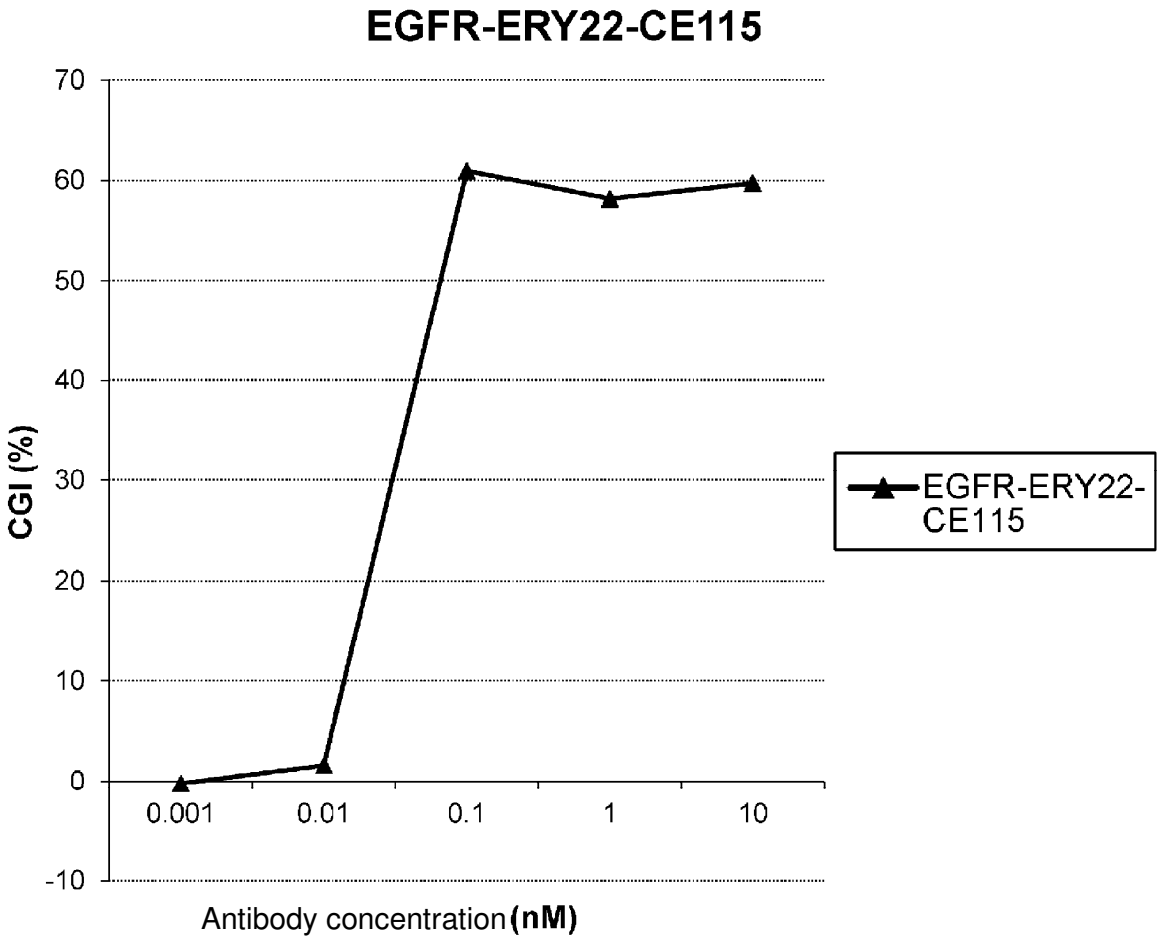

EGFR-ERY22-CE115
70
60
50
40
30
20
10
0
-10
CGI (%)
0.001
0.01
0.1
1
10
Antibody concentration (nM)
EGFR-ERY22-CE115

FIG. 9

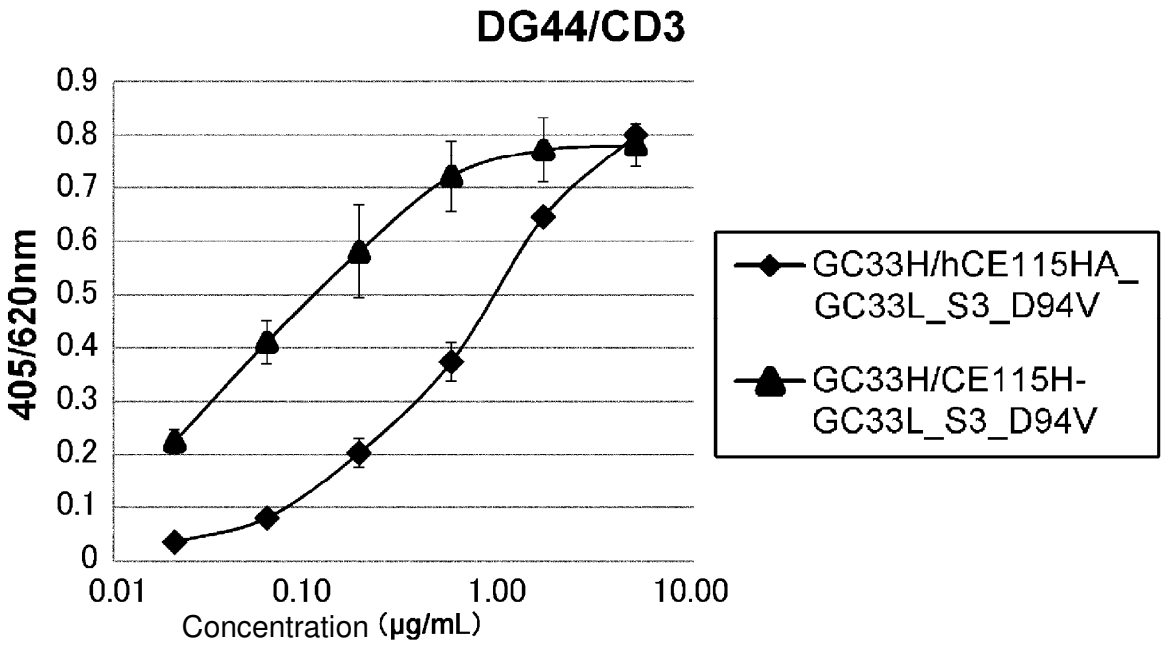

DG44/CD3
405/620nm
0.9
0.8
0.7
0.6
0.5
0.4
0.3
0.2
0.1
0
0.01
0.10
1.00
10.00
Concentration (μg/mL)
GC33H/hCE115HA_GC33L_S3_D94V
GC33H/CE115H-GC33L_S3_D94V

FIG. 10

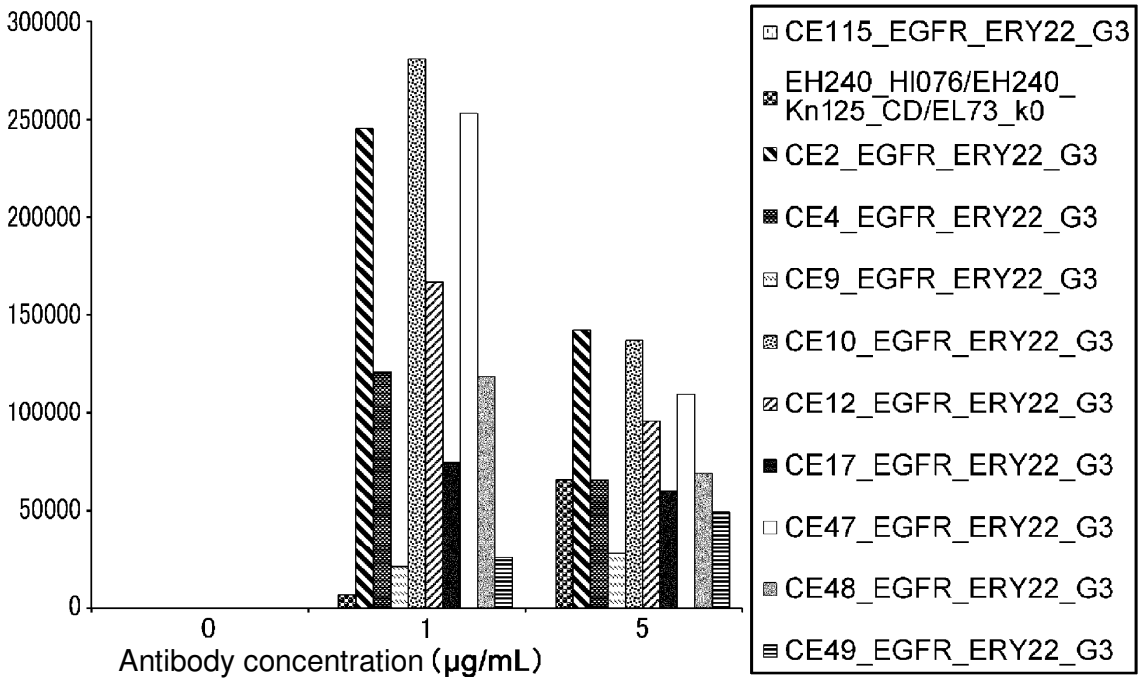

300000
250000
200000
150000
100000
50000
0
0
1
5
Antibody concentration (μg/mL)
CE115_EGFR_ERY22_G3
EH240_HI076/EH240_Kn125_CD/EL73_k0
CE2_EGFR_ERY22_G3
CE4_EGFR_ERY22_G3
CE9_EGFR_ERY22_G3
CE10_EGFR_ERY22_G3
CE12_EGFR_ERY22_G3
CE17_EGFR_ERY22_G3
CE47_EGFR_ERY22_G3
CE48_EGFR_ERY22_G3
CE49_EGFR_ERY22_G3

FIG. 11

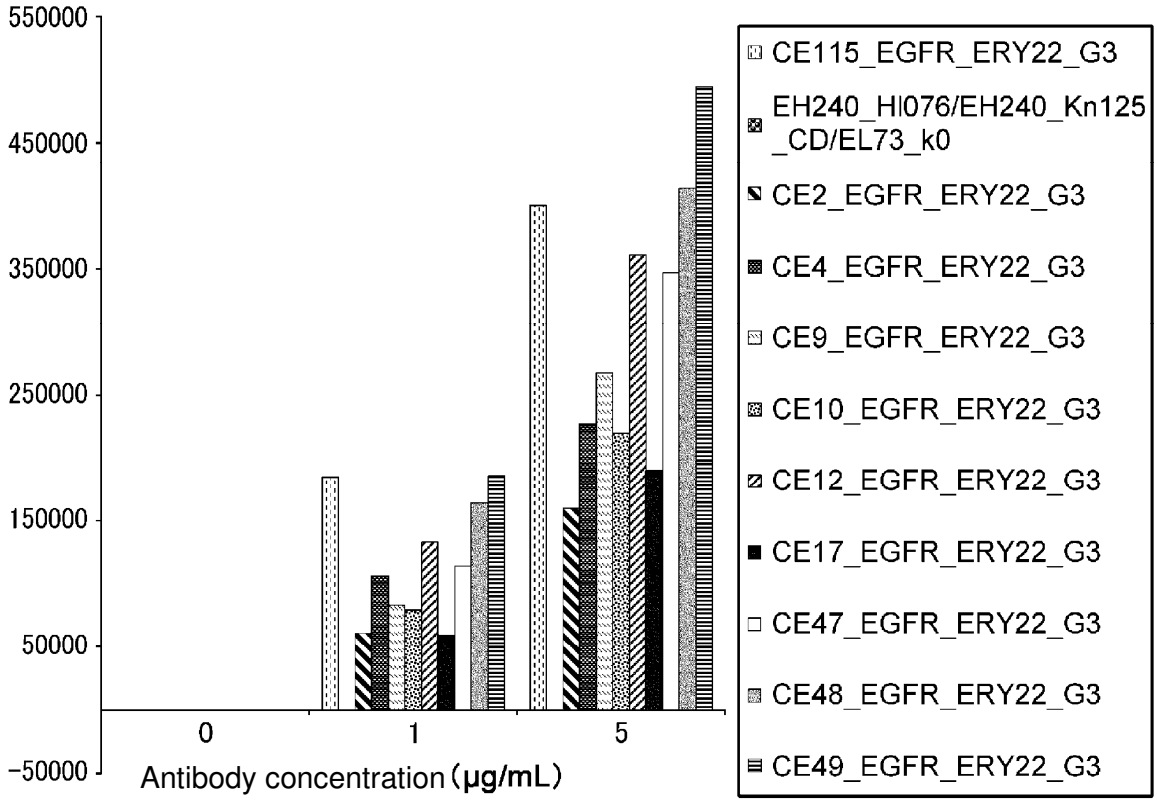

550000
450000
350000
250000
150000
50000
−50000
0
1
5
Antibody concentration (μg/mL)
CE115_EGFR_ERY22_G3
EH240_Hl076/EH240_Kn125_CD/EL73_k0
CE2_EGFR_ERY22_G3
CE4_EGFR_ERY22_G3
CE9_EGFR_ERY22_G3
CE10_EGFR_ERY22_G3
CE12_EGFR_ERY22_G3
CE17_EGFR_ERY22_G3
CE47_EGFR_ERY22_G3
CE48_EGFR_ERY22_G3
CE49_EGFR_ERY22_G3

FIG. 12

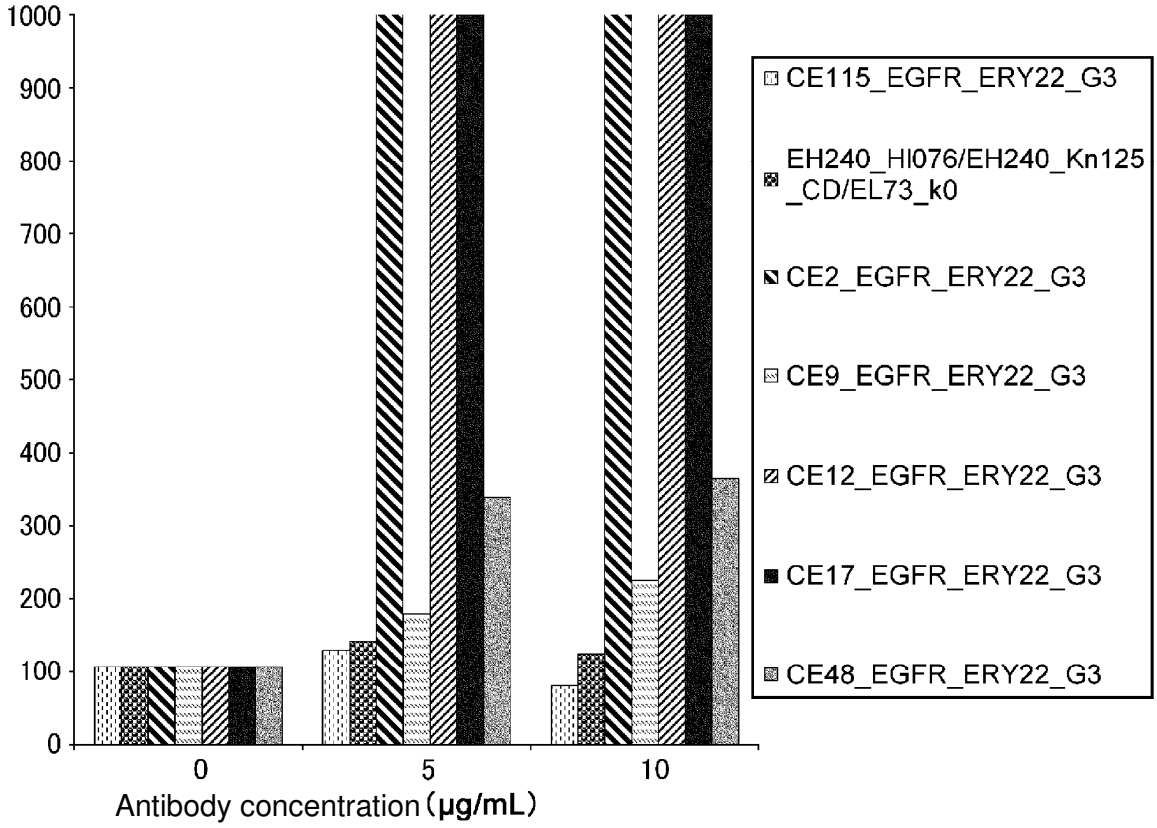

1000
900
800
700
600
500
400
300
200
100
0
0
5
10
Antibody concentration (μg/mL)
CE115_EGFR_ERY22_G3
EH240_HI076/EH240_Kn125_CD/EL73_k0
CE2_EGFR_ERY22_G3
CE9_EGFR_ERY22_G3
CE12_EGFR_ERY22_G3
CE17_EGFR_ERY22_G3
CE48_EGFR_ERY22_G3

FIG. 13

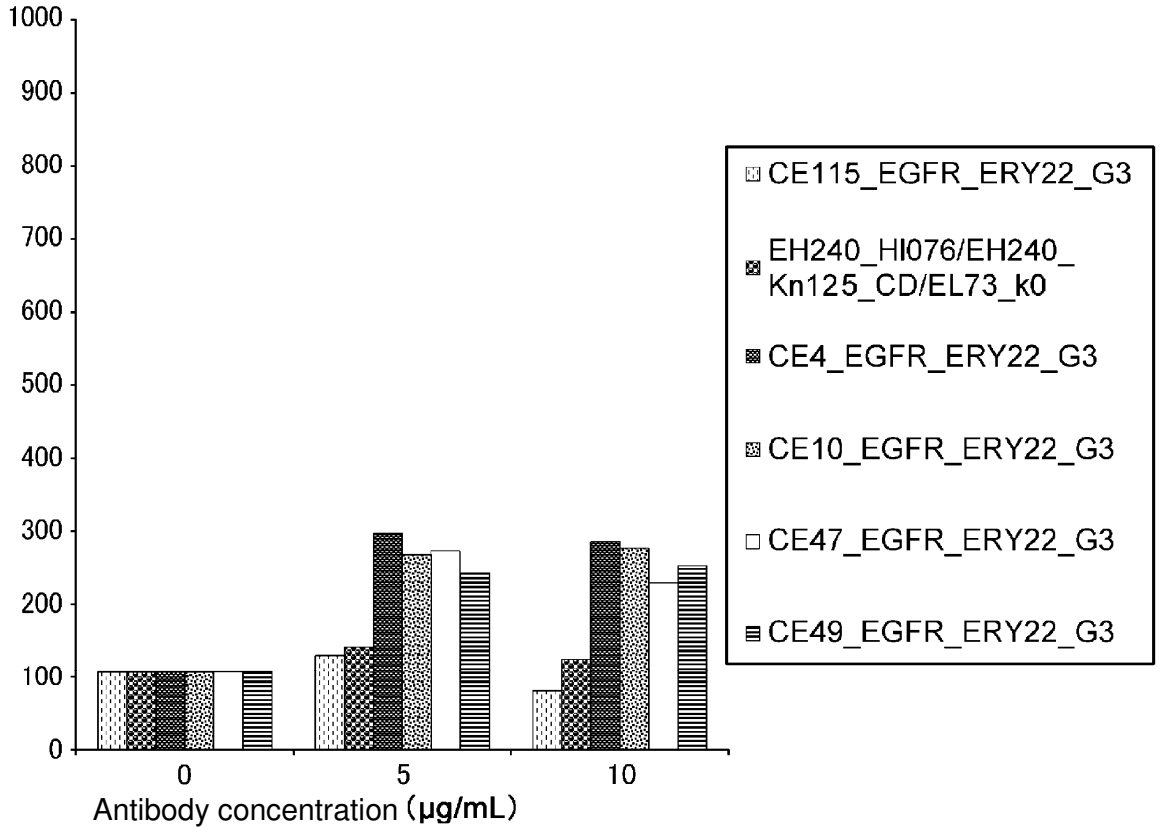

1000
900
800
700
600
500
400
300
200
100
0
0
5
10
Antibody concentration (μg/mL)
CE115_EGFR_ERY22_G3
EH240_HI076/EH240_Kn125_CD/EL73_k0
CE4_EGFR_ERY22_G3
CE10_EGFR_ERY22_G3
CE47_EGFR_ERY22_G3
CE49_EGFR_ERY22_G3

FIG. 14

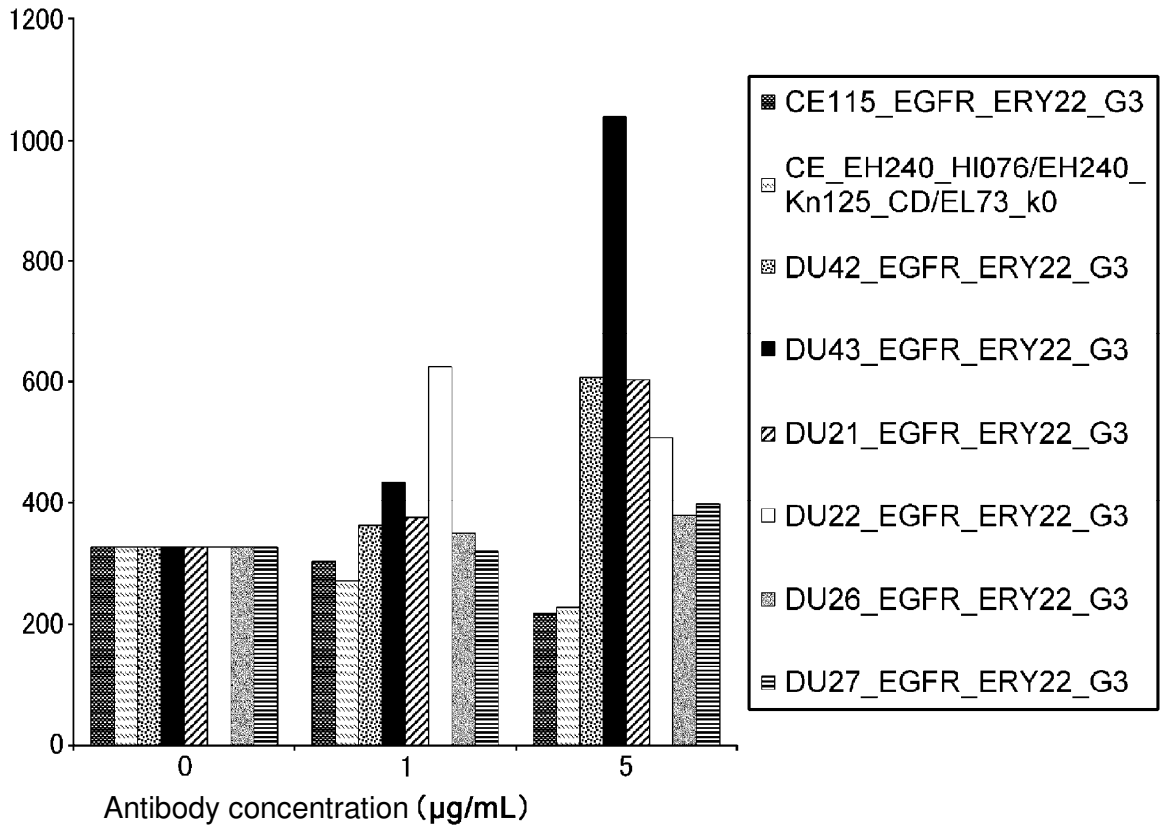

1200
1000
800
600
400
200
0
0
1
5
Antibody concentration (μg/mL)
CE115_EGFR_ERY22_G3
CE_EH240_HI076/EH240_Kn125_CD/EL73_k0
DU42_EGFR_ERY22_G3
DU43_EGFR_ERY22_G3
DU21_EGFR_ERY22_G3
DU22_EGFR_ERY22_G3
DU26_EGFR_ERY22_G3
DU27_EGFR_ERY22_G3

FIG. 15

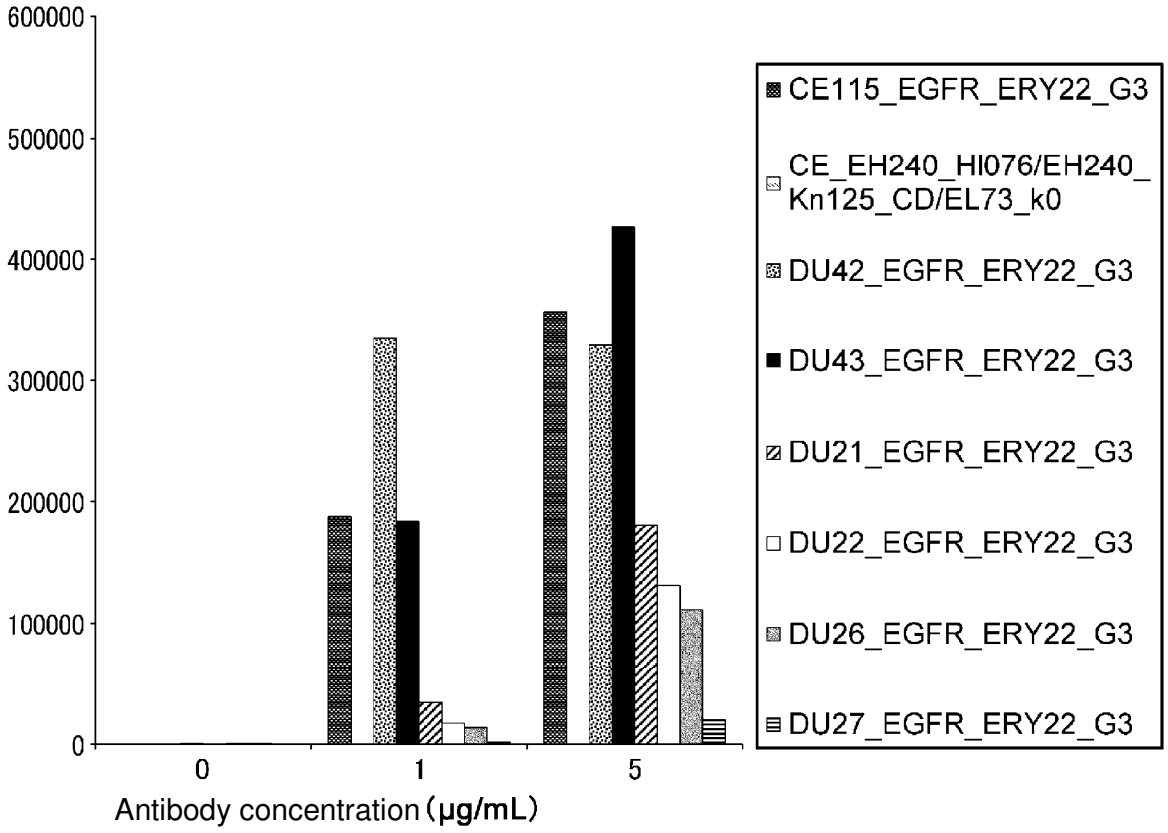

600000
500000
400000
300000
200000
100000
0
0
1
5
Antibody concentration (μg/mL)
CE115_EGFR_ERY22_G3
CE_EH240_HI076/EH240_Kn125_CD/EL73_k0
DU42_EGFR_ERY22_G3
DU43_EGFR_ERY22_G3
DU21_EGFR_ERY22_G3
DU22_EGFR_ERY22_G3
DU26_EGFR_ERY22_G3
DU27_EGFR_ERY22_G3

FIG. 16

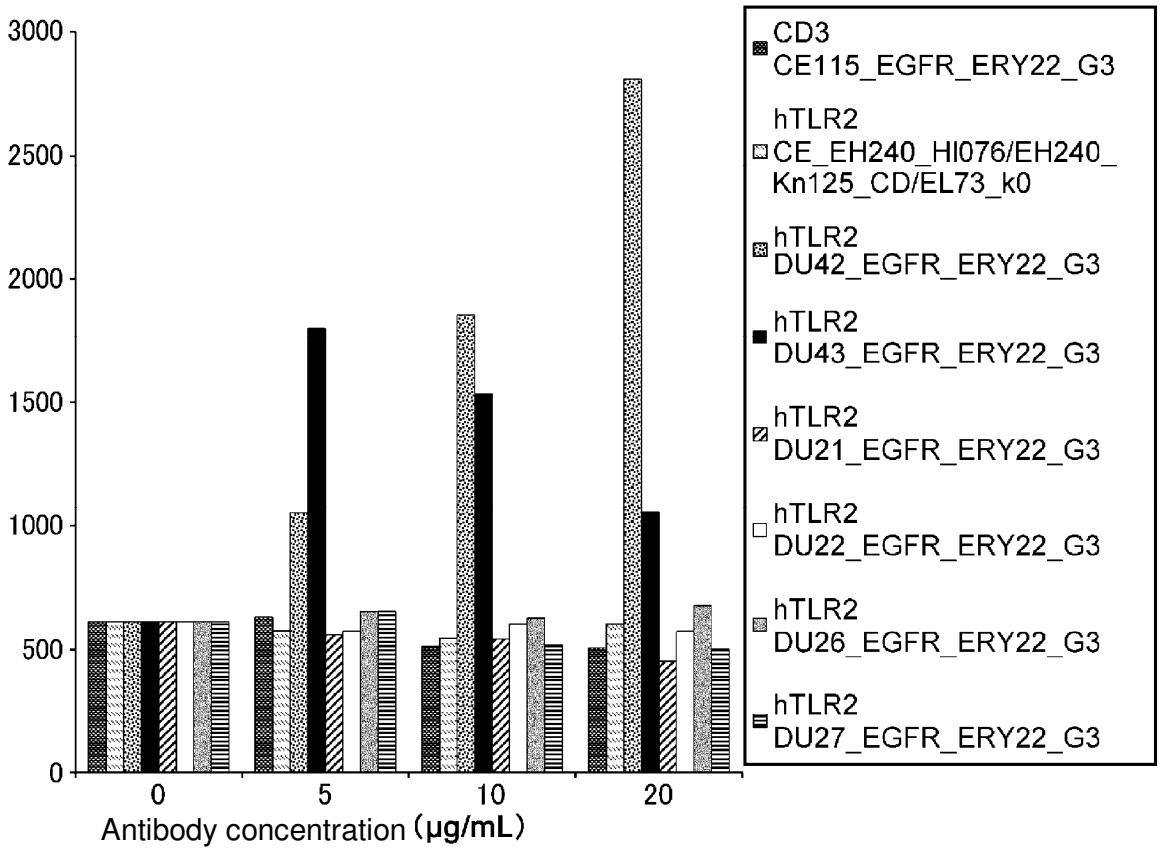

3000
2500
2000
1500
1000
500
0
0
5
10
20
Antibody concentration (μg/mL)
CD3
CE115_EGFR_ERY22_G3
hTLR2
CE_EH240_HI076/EH240_
Kn125_CD/EL73_k0
hTLR2
DU42_EGFR_ERY22_G3
hTLR2
DU43_EGFR_ERY22_G3
hTLR2
DU21_EGFR_ERY22_G3
hTLR2
DU22_EGFR_ERY22_G3
hTLR2
DU26_EGFR_ERY22_G3
hTLR2
DU27_EGFR_ERY22_G3

FIG. 17

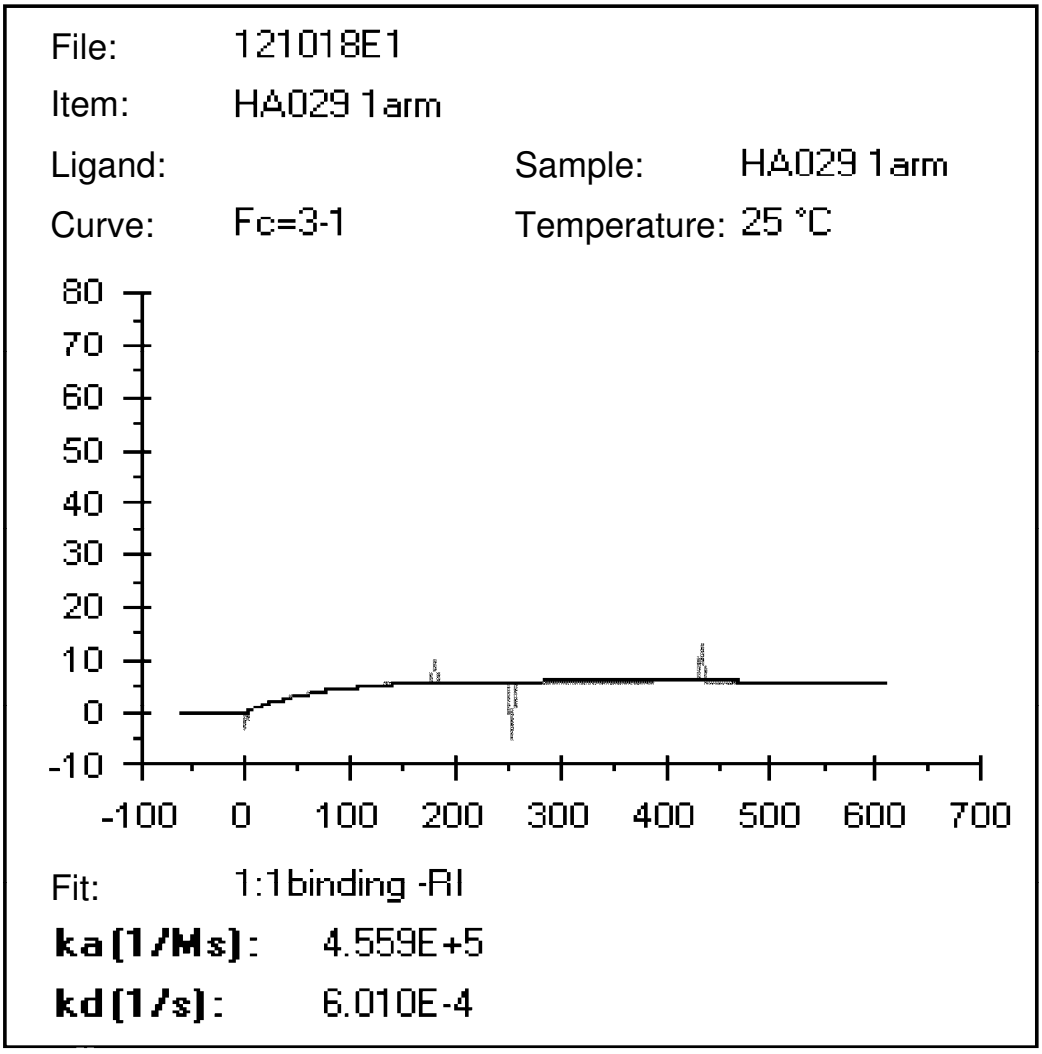

File: 121018E1
Item: HA029 1arm
Ligand:
Sample: HA029 1arm
Curve: Fc=3-1
Temperature: 25 °C
80
70
60
50
40
30
20
10
0
-10
-100
0
100
200
300
400
500
600
700
Fit: 1:1binding -RI
ka[1/Ms]: 4.559E+5
kd[1/s]: 6.010E-4

FIG. 18

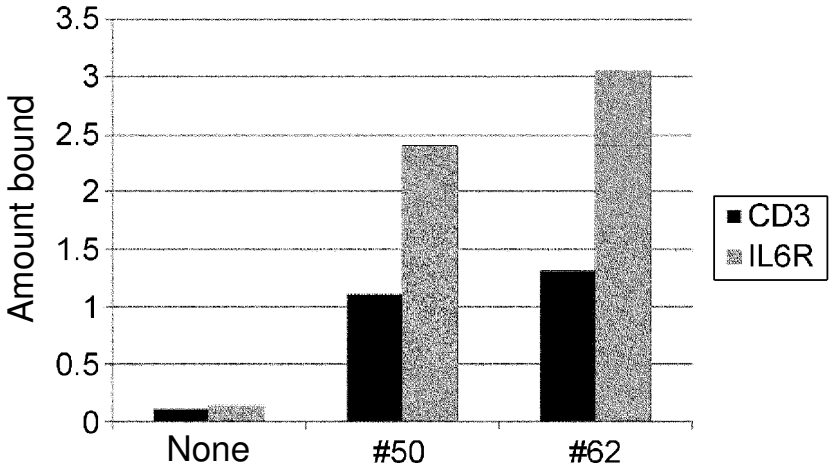


FIG. 19

hIADHB2b-4

#	Value of ELISA (absorbance at 450 nm)			Ratio to NC	
	hIgA	CD3	NC	S/N of hIgA	S/N of CD3
1	0.069	0.049	0.065	1.1	0.8
2	0.064	0.053	0.061	1.0	0.9
3	0.058	0.057	0.056	1.0	1.0
4	0.066	0.053	0.064	1.0	0.8
5	0.067	0.061	0.065	1.0	0.9
6	0.102	0.545	0.056	1.8	**9.7**
7	0.110	0.624	0.054	**2.0**	**11.6**
8	0.077	0.075	0.077	1.0	1.0
9	0.123	0.806	0.052	**2.4**	**15.5**
10	0.051	0.372	0.051	1.0	**7.3**
11	0.056	0.055	0.053	1.1	1.0
12	0.054	0.053	0.052	1.0	1.0
13	0.056	0.052	0.056	1.0	0.9
14	0.063	0.059	0.062	1.0	1.0
15	0.069	0.060	0.062	1.1	1.0
16	0.066	0.062	0.063	1.0	1.0
17	0.067	0.057	0.061	1.1	0.9
18	0.061	0.055	0.057	1.1	1.0
19	0.067	0.060	0.067	1.0	0.9
20	0.058	0.526	0.057	1.0	**9.2**
21	0.059	0.051	0.054	1.1	0.9
22	0.064	0.059	0.061	1.0	1.0
23	0.067	0.063	0.069	1.0	0.9
24	0.065	0.062	0.062	1.0	1.0
25	0.068	0.056	0.062	1.1	0.9
26	0.055	0.051	0.053	1.0	1.0
27	0.061	0.061	0.060	1.0	1.0
28	0.065	0.063	0.063	1.0	1.0
29	0.065	0.061	0.063	1.0	1.0
30	0.067	0.060	0.062	1.1	1.0
31	0.066	0.060	0.064	1.0	0.9
32	0.071	0.070	0.067	1.1	1.0
33	0.071	0.061	0.063	1.1	1.0
34	0.053	0.063	0.052	1.0	1.2
35	0.051	0.049	0.048	1.1	1.0
36	0.121	0.794	0.049	**2.5**	**16.2**
37	0.050	0.346	0.049	1.0	**7.1**
38	0.071	0.061	0.060	1.2	1.0
39	0.063	0.059	0.061	1.0	1.0
40	0.069	0.067	0.067	1.0	1.0
41	0.075	0.067	0.071	1.1	0.9
42	0.051	0.391	0.048	1.1	**8.1**
43	0.121	0.820	0.048	**2.5**	**17.1**
44	0.126	0.816	0.053	**2.4**	**15.4**
45	0.054	0.050	0.052	1.0	1.0
46	0.067	0.063	0.063	1.1	1.0
47	0.054	0.051	0.053	1.0	1.0
48	0.083	0.081	0.092	0.9	0.9

#	Value of ELISA (absorbance at 450 nm)			Ratio to NC	
	hIgA	CD3	NC	S/N of hIgA	S/N of CD3
49	0.062	0.055	0.061	1.0	0.9
50	0.106	0.807	0.048	**2.2**	**16.8**
51	0.061	0.059	0.059	1.0	1.0
52	0.063	0.060	0.060	1.1	1.0
53	0.063	0.061	0.061	1.0	1.0
54	0.070	0.063	0.064	1.1	1.0
55	0.122	0.821	0.049	**2.5**	**16.8**
56	0.126	0.889	0.052	**2.4**	**17.1**
57	0.060	0.050	0.053	1.1	0.9
58	0.052	0.048	0.050	1.0	1.0
59	0.068	0.064	0.067	1.0	1.0
60	0.062	0.070	0.055	1.1	1.3
61	0.063	0.061	0.061	1.0	1.0
62	0.065	0.062	0.061	1.1	1.0
63	0.134	0.832	0.050	**2.7**	**16.6**
64	0.061	0.058	0.055	1.1	1.1
65	0.075	0.067	0.074	1.0	0.9
66	0.063	0.057	0.060	1.1	1.0
67	0.083	0.672	0.050	1.7	**13.4**
68	0.064	0.060	0.063	1.0	1.0
69	0.055	0.050	0.053	1.0	0.9
70	0.069	0.071	0.070	1.0	1.0
71	0.053	0.050	0.053	1.0	0.9
72	0.059	0.056	0.055	1.1	1.0
73	0.102	0.785	0.051	2.0	**15.4**
74	0.060	0.055	0.058	1.0	0.9
75	0.061	0.057	0.059	1.0	1.0
76	0.061	0.058	0.058	1.1	1.0
77	0.063	0.062	0.069	0.9	0.9
78	0.054	0.073	0.049	1.1	1.5
79	0.143	0.844	0.054	**2.6**	**15.6**
80	0.068	0.068	0.063	1.1	1.1
81	0.059	0.056	0.058	1.0	1.0
82	0.061	0.056	0.059	1.0	0.9
83	0.064	0.059	0.059	1.1	1.0
84	0.068	0.060	0.064	1.1	0.9
85	0.065	0.061	0.064	1.0	1.0
86	0.058	0.051	0.052	1.1	1.0
87	0.055	0.052	0.052	1.1	1.0
88	0.062	0.060	0.056	1.1	1.1
89	0.067	0.131	0.061	1.1	**2.1**
90	0.065	0.060	0.058	1.1	1.0
91	0.133	0.826	0.049	**2.7**	**16.9**
92	0.080	0.087	0.083	1.0	1.0
93	0.060	0.054	0.053	1.1	1.0
94	0.076	0.069	0.068	1.1	1.0
95	0.088	0.089	0.082	1.1	1.1
96	0.076	0.069	0.071	1.1	1.0

FIG. 20

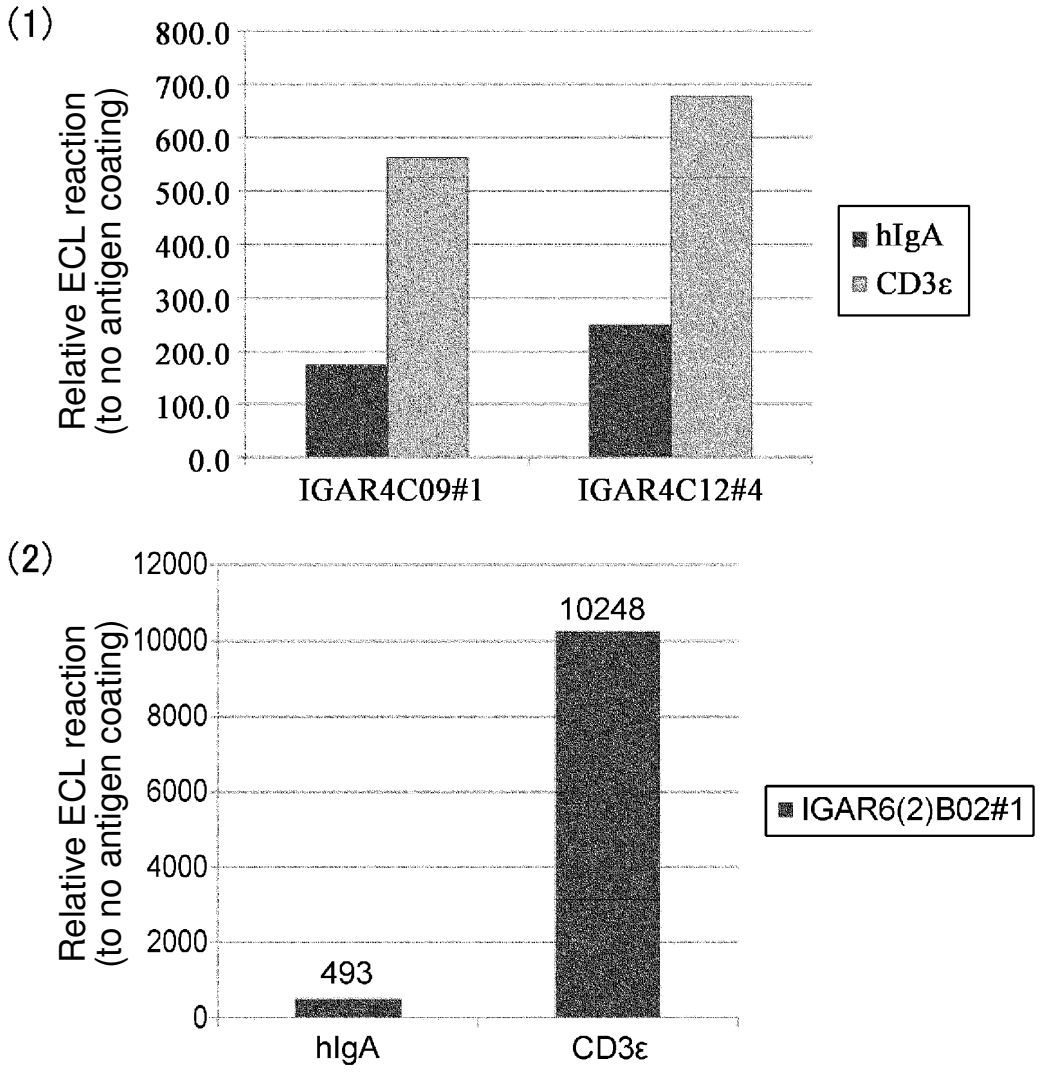

(1)
Relative ECL reaction
(to no antigen coating)
800.0
700.0
600.0
500.0
400.0
300.0
200.0
100.0
0.0
IGAR4C09#1
IGAR4C12#4
hIgA
CD3ε
(2)
Relative ECL reaction
(to no antigen coating)
12000
10000
8000
6000
4000
2000
0
10248
493
hIgA
CD3ε
IGAR6(2)B02#1

FIG. 21

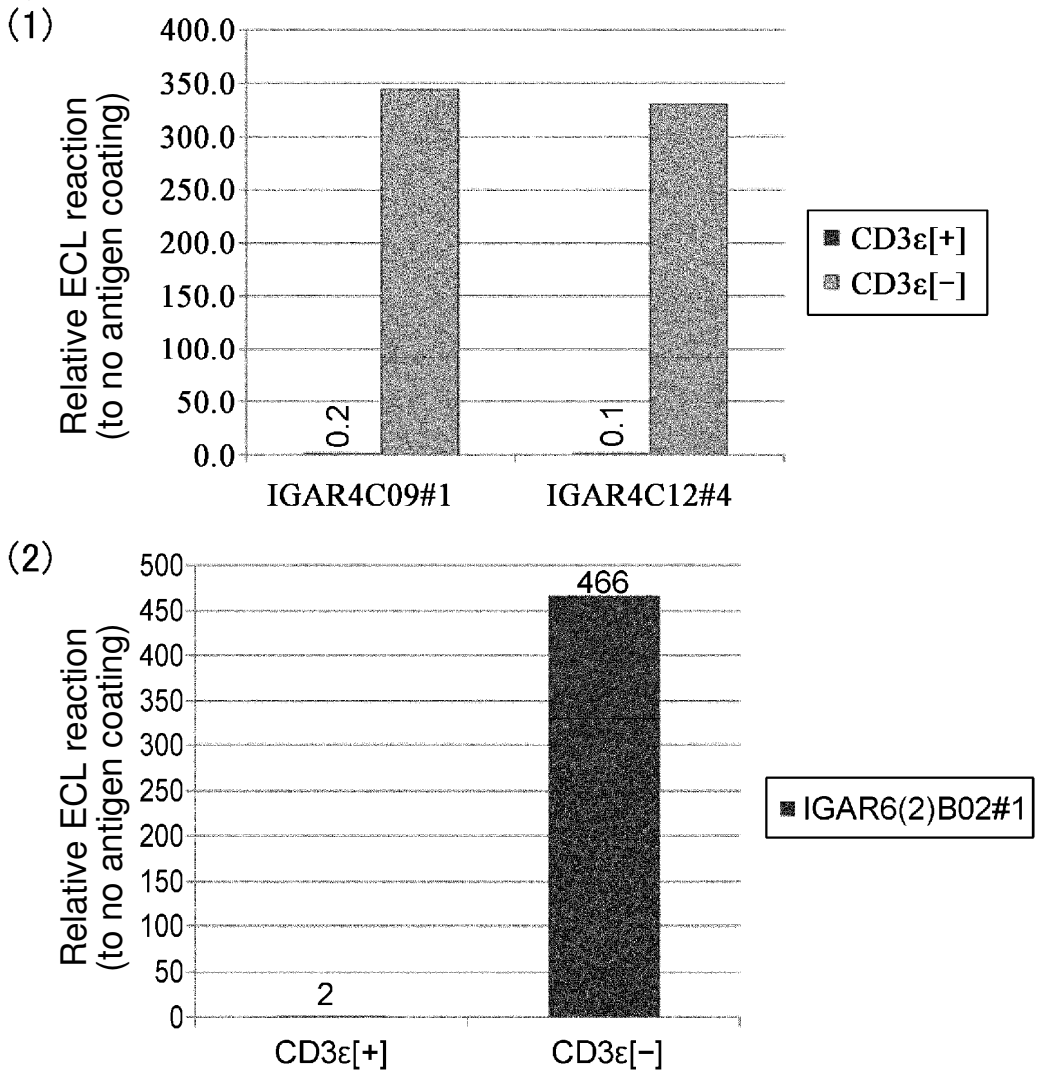

(1)
Relative ECL reaction
(to no antigen coating)
400.0
350.0
300.0
250.0
200.0
150.0
100.0
50.0
0.0
0.2
0.1
IGAR4C09#1
IGAR4C12#4
CD3ε[+]
CD3ε[−]
(2)
Relative ECL reaction
(to no antigen coating)
500
450
400
350
300
250
200
150
100
50
0
466
2
CD3ε[+]
CD3ε[−]
IGAR6(2)B02#1

FIG. 22

154DHB2b-4

#	Value of ELISA (absorbance at 450 nm)			Ratio to NC	
	CD154	CD3	NC	S/N of CD154	S/N of CD3
1	0.077	0.061	0.062	1.2	1.0
2	0.060	0.114	0.050	1.2	**2.3**
3	0.053	0.048	0.050	1.1	1.0
4	0.048	0.045	0.049	1.0	0.9
5	0.053	0.050	0.057	0.9	0.9
6	0.057	0.052	0.054	1.1	1.0
7	0.086	0.075	0.079	1.1	0.9
8	0.060	0.059	0.059	1.0	1.0
9	0.053	0.062	0.060	0.9	1.0
10	0.048	0.047	0.048	1.0	1.0
11	0.049	0.062	0.052	0.9	1.2
12	0.059	0.047	0.047	1.3	1.0
13	0.058	0.292	0.052	1.1	**5.6**
14	0.059	0.110	0.059	1.0	1.9
15	0.053	0.053	0.058	0.9	0.9
16	0.061	0.117	0.064	1.0	1.8
17	0.064	0.058	0.058	1.1	1.0
18	0.163	0.255	0.083	2.0	**3.1**
19	0.054	0.067	0.059	0.9	1.1
20	0.057	0.179	0.049	1.2	**3.7**
21	0.051	0.051	0.053	1.0	1.0
22	0.050	0.680	0.054	0.9	**12.6**
23	0.058	0.094	0.063	0.9	1.5
24	0.059	0.234	0.061	1.0	**3.8**
25	0.056	0.050	0.051	1.1	1.0
26	0.050	0.057	0.049	1.0	1.2
27	0.046	0.046	0.048	1.0	1.0
28	0.054	0.169	0.051	1.1	**3.3**
29	0.049	0.183	0.047	1.0	**3.9**
30	0.051	0.052	0.050	1.0	1.0
31	0.045	0.045	0.046	1.0	1.0
32	0.051	0.050	0.053	1.0	0.9
33	0.073	0.125	0.074	1.0	1.7
34	0.052	0.050	0.053	1.0	0.9
35	0.067	0.121	0.072	0.9	1.7
36	0.055	0.648	0.053	1.0	**12.2**
37	0.051	0.381	0.053	1.0	**7.2**
38	0.050	0.047	0.051	1.0	0.9
39	0.051	0.047	0.051	1.0	0.9
40	0.072	0.104	0.070	1.0	1.5
41	0.052	0.051	0.051	1.0	1.0
42	0.054	0.055	0.058	0.9	0.9
43	0.052	0.086	0.052	1.0	1.7
44	0.061	0.050	0.055	1.1	0.9
45	0.049	0.047	0.048	1.0	1.0
46	0.043	0.043	0.046	0.9	0.9
47	0.056	0.217	0.058	1.0	**3.7**
48	0.073	0.115	0.073	1.0	1.6

#	Value of ELISA (absorbance at 450 nm)			Ratio to NC	
	CD154	CD3	NC	S/N of CD154	S/N of CD3
49	0.069	0.210	0.066	1.0	**3.2**
50	0.110	0.751	0.049	**2.2**	**15.3**
51	0.046	0.044	0.049	0.9	0.9
52	0.050	0.178	0.052	1.0	**3.4**
53	0.050	0.048	0.059	0.8	0.8
54	0.063	0.057	0.064	1.0	0.9
55	0.053	0.975	0.054	1.0	**18.1**
56	0.056	0.057	0.061	0.9	0.9
57	0.050	0.052	0.049	1.0	1.1
58	0.050	0.045	0.049	1.0	0.9
59	0.062	0.067	0.059	1.1	1.1
60	0.060	0.067	0.052	1.2	1.3
61	0.057	0.065	0.058	1.0	1.1
62	0.109	0.749	0.052	**2.1**	**14.4**
63	0.062	0.068	0.069	0.9	1.0
64	0.049	0.052	0.051	1.0	1.0
65	0.065	0.059	0.059	1.1	1.0
66	0.054	0.055	0.058	0.9	0.9
67	0.049	0.045	0.048	1.0	0.9
68	0.065	0.175	0.064	1.0	**2.7**
69	0.055	0.052	0.057	1.0	0.9
70	0.047	0.046	0.048	1.0	1.0
71	0.059	0.058	0.061	1.0	1.0
72	0.057	0.090	0.059	1.0	1.5
73	0.059	0.077	0.057	1.0	1.4
74	0.052	0.192	0.057	0.9	**3.4**
75	0.059	0.130	0.071	0.8	1.8
76	0.055	0.046	0.048	1.1	1.0
77	0.049	0.054	0.050	1.0	1.1
78	0.054	0.052	0.054	1.0	1.0
79	0.054	0.056	0.054	1.0	1.0
80	0.125	0.816	0.057	**2.2**	**14.3**
81	0.048	0.046	0.049	1.0	0.9
82	0.052	0.056	0.051	1.0	1.1
83	0.053	0.410	0.054	1.0	**7.6**
84	0.065	0.057	0.064	1.0	0.9
85	0.054	0.048	0.053	1.0	0.9
86	0.053	0.102	0.056	0.9	1.8
87	0.051	0.050	0.054	0.9	0.9
88	0.054	0.054	0.053	1.0	1.0
89	0.065	0.087	0.070	0.9	1.2
90	0.047	0.072	0.051	0.9	1.4
91	0.062	0.047	0.052	1.2	0.9
92	0.052	0.052	0.052	1.0	1.0
93	0.048	0.047	0.048	1.0	1.0
94	0.056	0.803	0.058	1.0	**13.8**
95	0.059	0.183	0.058	1.0	**3.2**
96	0.074	0.069	0.067	1.1	1.0

FIG. 23

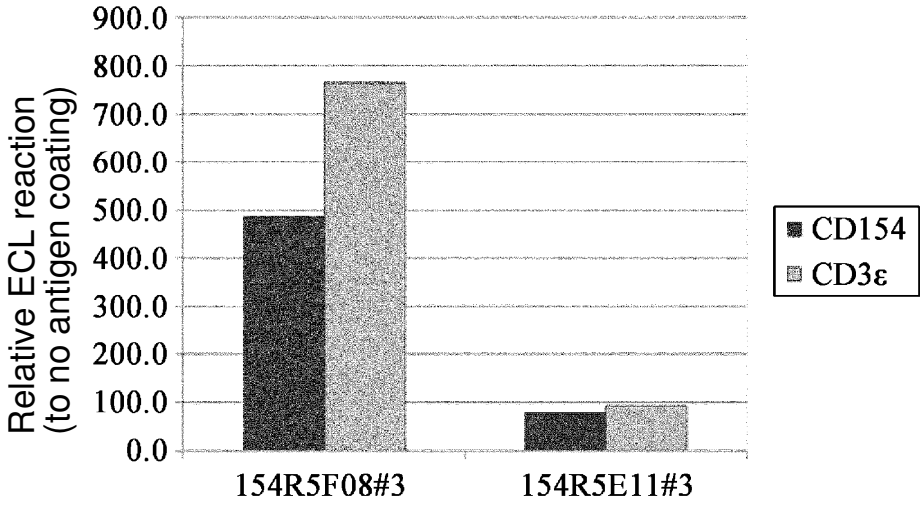

900.0
800.0
700.0
600.0
500.0
400.0
300.0
200.0
100.0
0.0
Relative ECL reaction
(to no antigen coating)
154R5F08#3
154R5E11#3
CD154
CD3ε

FIG. 24

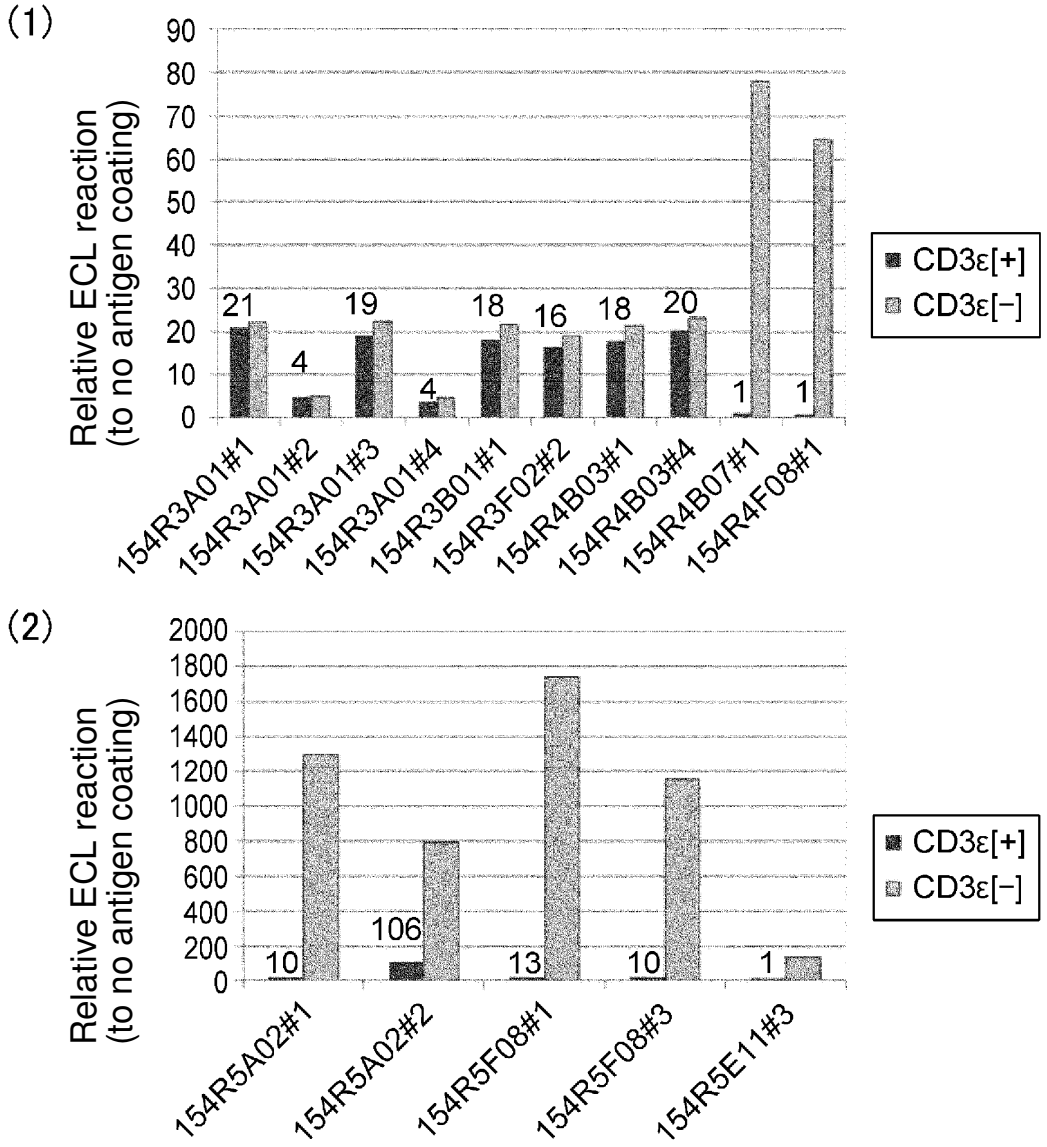

(1)
Relative ECL reaction
(to no antigen coating)
90
80
70
60
50
40
30
20
10
0
21
4
19
4
18
16
18
20
1
1
CD3ε[+]
CD3ε[−]
154R3A01#1
154R3A01#2
154R3A01#3
154R3A01#4
154R3B01#1
154R3F02#2
154R4B03#1
154R4B03#4
154R4B07#1
154R4F08#1
(2)
Relative ECL reaction
(to no antigen coating)
2000
1800
1600
1400
1200
1000
800
600
400
200
0
10
106
13
10
1
CD3ε[+]
CD3ε[−]
154R5A02#1
154R5A02#2
154R5F08#1
154R5F08#3
154R5E11#3

FIG. 25