RSV-F long (GC) R1691

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ACCUGCUCCGCCGUCAGCAAGGGCUACCUGUCCGCCCUCCGGACCGGGUGGUACACGAGC
GUGAUCACCAUCGAGCUGUCCAACAUCAAGGAGAACAAGUGCAACGGCACCGACGCGAAG
GUGAAGCUGAUCAACCAGGAGCUCGACAAGUACAAGAACGCCGUCACCGAGCUGCAGCUG
CUCAUGCAGAGCACGACCGCCGCCAACAACCGCGCGCGGCGCGAGCUGCCGCGGUUCAUG
AACUACACCCUGAACAACACCAAGAAGACGAACGUGACCCUCUCCAAGAAGCGCAAGCGG
CGCUUCCUGGGGUUCCUGCUCGGCGUGGGGAGCGCCAUCGCCUCCGGCAUCGCCGUCAGC
AAGGUGCUGCACCUGGAGGGCGAGGUGAACAAGAUCAAGUCCGCCCUCCUGAGCACCAAC
AAGGCGGUCGUGUCCCUGAGCAACGGGGUGUCCGUCCUCACCAGCAAGGUGCUGGACCUG
AAGAACUACAUCGACAAGCAGCUCCUGCCCAUCGUGAACAAGCAGUCCUGCCGGAUCAGC
AACAUCGAGACGGUCAUCGAGUUCCAGCAGAAGAACAACCGCCUGCUCGAGAUCACCCGG
GAGUUCAGCGUGAACGCCGGCGUGACCACCCCCGUCUCCACGUACAUGCUGACCAACAGC
GAGCUGCUCUCCCUGAUCAACGACAUGCCCAUCACCAACGACCAGAAGAAGCUGAUGAGC
AACAACGUGCAGAUCGUGCGCCAGCAGUCCUACAGCAUCAUGUCCAUCAUCAAGGAGGAG
GUCCUCGCCUACGUGGUGCAGCUGCCGCUGUACGGGGUCAUCGACACCCCCUGCUGGAAG
CUCCACACGAGCCCCCUGUGCACCACCAACACCAAGGAGGGCUCCAACAUCUGCCUGACG
CGGACCGACCGCGGGUGGUACUGCGACAACGCCGGCAGCGUGUCCUUCUUCCCCCAGGCC
GAGACCUGCAAGGUCCAGAGCAACCGGGUGUUCUGCGACACCAUGAACUCCCUCACGCUG
CCGAGCGAGGUGAACCUGUGCAACGUCGACAUCUUCAACCCCAAGUACGACUGCAAGAUC
AUGACCUCCAAGACCGACGUGAGCUCCAGCGUGAUCACCUCCCUCGGCGCGAUCGUCAGC
UGCUACGGGAAGACGAAGUGCACCGCCAGCAACAAGAACCGCGGCAUCAUCAAGACCUUC
UCCAACGGGUGCGACUACGUGAGCAACAAGGGCGUGGACACCGUCUCCGUGGGCAACACC
CUGUACUACGUGAACAAGCAGGAGGGGAAGAGCCUGUACGUCAAGGGCGAGCCCAUCAUC
AACUUCUACGACCCCCUCGUGUUCCCGUCCGACGAGUUCGACGCCAGCAUCUCCCAGGUG
AACGAGAAGAUCAACCAGAGCCUGGCCUUCAUCCGGAAGUCCGACGAGCUGCUGCACCAC
GUCAACGCCGGGAAGAGCACGACCAACAUCAUGAUCACCACCAUCAUCAUCGUGAUCAUC
GUGAUCCUCCUGUCCCUGAUCGCGGUCGGCCUCCUGCUGUACUGCAAGGCCCGCAGCACG
CCCGUGACCCUCUCCAAGGACCAGCUGAGCGGGAUCAACAACAUCGCCUUCUCCAACUGA
GGACUAGUUAUAAGACUGACUAGCCCGAUGGGCCUCCCAACGGGCCCUCCUCCCCCUCCUU
GCACCGAGAUUAAUAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAA
AAAAAAAAAAAAAAAAAAUGCAUCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCAAAGGC
UCUUUUCAGAGCCACCAGAAUU

Fig. 1

RSV-F long (GC) R2510

GGGGCGCUGCCUACGGAGGUGGCAGCCAUCUCCUUCUCGGCAUCAAGCUUACCAUGGAGC
UGCCCAUCCUCAAGGCCAACGCCAUCACCACCAUCCUGGCGGCCGUGACGUUCUGCUUCG
CCAGCUCCCAGAACAUCACCGAGGAGUUCUACCAGAGCACCUGCUCCGCCGUCAGCAAGG
GCUACCUGUCCGCCCUCCGGACCGGGUGGUACACGAGCGUGAUCACCAUCGAGCUGUCCA
ACAUCAAGGAGAACAAGUGCAACGGCACCGACGCGAAGGUGAAGCUGAUCAACCAGGAGC
UCGACAAGUACAAGAACGCCGUCACCGAGCUGCAGCUGCUCAUGCAGAGCACGACCGCCG
CCAACAACCGCGCGCGGCGCGAGCUGCCGCGGUUCAUGAACUACACCCUGAACAACACCA
AGAAGACGAACGUGACCCUCUCCAAGAAGCGCAAGCGGCGCUUCCUGGGGUUCCUGCUCG
GCGUGGGGAGCGCCAUCGCCUCCGGCAUCGCCGUCAGCAAGGUGCUGCACCUGGAGGGCG
AGGUGAACAAGAUCAAGUCCGCCCUCCUGAGCACCAACAAGGCGGUCGUGUCCCUGAGCA
ACGGGGUGUCCGUCCUCACCAGCAAGGUGCUGGACCUGAAGAACUACAUCGACAAGCAGC
UCCUGCCCAUCGUGAACAAGCAGUCCUGCCGGAUCAGCAACAUCGAGACGGUCAUCGAGU
UCCAGCAGAAGAACAACCGCCUGCUCGAGAUCACCCGGGAGUUCAGCGUGAACGCCGGCG
UGACCACCCCCGUCUCCACGUACAUGCUGACCAACAGCGAGCUGCUCUCCCUGAUCAACG
ACAUGCCCAUCACCAACGACCAGAAGAAGCUGAUGAGCAACAACGUGCAGAUCGUGCGCC
AGCAGUCCUACAGCAUCAUGUCCAUCAUCAAGGAGGAGGUCCUCGCCUACGUGGUGCAGC
UGCCGCUGUACGGGGUCAUCGACACCCCCUGCUGGAAGCUCCACACGAGCCCCCUGUGCA
CCACCAACACCAAGGAGGGCUCCAACAUCUGCCUGACGCGGACCGACCGCGGGUGGUACU
GCGACAACGCCGGCAGCGUGUCCUUCUUCCCCCAGGCCGAGACCUGCAAGGUCCAGAGCA
ACCGGGUGUUCUGCGACACCAUGAACUCCCUCACGCUGCCGAGCGAGGUGAACCUGUGCA
ACGUCGACAUCUUCAACCCCAAGUACGACUGCAAGAUCAUGACCUCCAAGACCGACGUGA
GCUCCAGCGUGAUCACCUCCCUCGGCGCGAUCGUCAGCUGCUACGGGAAGACGAAGUGCA
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UGGCCUUCAUCCGGAAGUCCGACGAGCUGCUGCACCACGUCAACGCCGGGAAGAGCACGA
CCAACAUCAUGAUCACCACCAUCAUCAUCGUGAUCAUCGUGAUCCUCCUGUCCCUGAUCG
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AAAUGCAUCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCAAAGGCUCUUUUCAGAGCCAC
CAGAAUU

Fig. 2

RSV-Fdel554-574 long (GC) R2821

GGGGCGCUGCCUACGGAGGUGGCAGCCAUCUCCUUCUCGGCAUCAAGCUUACCAUGGAGC
UGCCCAUCCUCAAGGCCAACGCCAUCACCACCAUCCUGGCGGCCGUGACGUUCUGCUUCG
CCAGCUCCCAGAACAUCACCGAGGAGUUCUACCAGAGCACCUGCUCCGCCGUCAGCAAGG
GCUACCUGUCCGCCCUCCGGACCGGGUGGUACACGAGCGUGAUCACCAUCGAGCUGUCCA
ACAUCAAGGAGAACAAGUGCAACGGCACCGACGCGAAGGUGAAGCUGAUCAACCAGGAGC
UCGACAAGUACAAGAACGCCGUCACCGAGCUGCAGCUGCUCAUGCAGAGCACGACCGCCG
CCAACAACCGCGCGCGGCGCGAGCUGCCGCGGUUCAUGAACUACACCCUGAACAACACCA
AGAAGACGAACGUGACCCUCUCCAAGAAGCGCAAGCGGCGCUUCCUGGGGUUCCUGCUCG
GCGUGGGGAGCGCCAUCGCCUCCGGCAUCGCCGUCAGCAAGGUGCUGCACCUGGAGGGCG
AGGUGAACAAGAUCAAGUCCGCCCUCCUGAGCACCAACAAGGCGGUCGUGUCCCUGAGCA
ACGGGGUGUCCGUCCUCACCAGCAAGGUGCUGGACCUGAAGAACUACAUCGACAAGCAGC
UCCUGCCCAUCGUGAACAAGCAGUCCUGCCGGAUCAGCAACAUCGAGACGGUCAUCGAGU
UCCAGCAGAAGAACAACCGCCUGCUCGAGAUCACCCGGGAGUUCAGCGUGAACGCCGGCG
UGACCACCCCCGUCUCCACGUACAUGCUGACCAACAGCGAGCUGCUCUCCCUGAUCAACG
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CCACCAACACCAAGGAGGGCUCCAACAUCUGCCUGACGCGGACCGACCGCGGGUGGUACU
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GCAACAAGGGCGUGGACACCGUCUCCGUGGGCAACACCCUGUACUACGUGAACAAGCAGG
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UCCCGUCCGACGAGUUCGACGCCAGCAUCUCCCAGGUGAACGAGAAGAUCAACCAGAGCC
UGGCCUUCAUCCGGAAGUCCGACGAGCUGCUGCACCACGUCAACGCCGGGAAGAGCACGA
CCAACAUCAUGAUCACCACCAUCAUCAUCGUGAUCAUCGUGAUCCUCCUGUCCCUGAUCG
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AUCUCAGCCUACCAUGAGAAUAAGAGAAAGAAAAUGAAGAUCAAUAGCUUAUUCAUCUCU
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AUUUUGCCUCUUUUCUCUGUGCUUCAAUUAAUAAAAAAUGGAAAGAACCUAGAUCUAAAA
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UGCAUCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCAAAGGCUCUUUUCAGAGCCACCAG
AAUU

Fig. 3

RSV-N (GC) R2831

GGGGCGCUGCCUACGGAGGUGGCAGCCAUCUCCUUCUCGGCAUCAAGCUUACCAUGGCCC
UGAGCAAGGUGAAGCUCAACGACACCCUGAACAAGGACCAGCUGCUCUCCAGCUCCAAGU
ACACCAUCCAGCGCAGCACGGGCGACUCCAUCGACACCCCCAACUACGACGUCCAGAAGC
ACAUCAACAAGCUGUGCGGGAUGCUGCUCAUCACCGAGGACGCCAACCACAAGUUCACCG
GCCUGAUCGGGAUGCUGUACGCGAUGAGCCGGCUCGGCCGCGAGGACACGAUCAAGAUCC
UGCGGGACGCCGGGUACCACGUGAAGGCCAACGGCGUGGACGUCACCACCCACCGCCAGG
ACAUCAACGGCAAGGAGAUGAAGUUCGAGGUGCUGACCCUCGCCUCCCUGACGACCGAGA
UCCAGAUCAACAUCGAGAUCGAGAGCCGGAAGUCCUACAAGAAGAUGCUGAAGGAGAUGG
GGGAGGUGGCCCCGGAGUACCGCCACGACAGCCCCGACUGCGGCAUGAUCAUCCUCUGCA
UCGCGGCCCUGGUCAUCACCAAGCUGGCCGCCGGGGACCGGUCCGGCCUCACCGCGGUGA
UCCGCCGGGCCAACAACGUGCUGAAGAACGAGAUGAAGCGCUACAAGGGGCUGCUCCCCA
AGGACAUCGCCAACAGCUUCUACGAGGUCUUCGAGAAGCACCCCCACUUCAUCGACGUGU
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UCGCCGGGCUGUUCAUGAACGCGUACGGCGCCGGCCAGGUGAUGCUGCGGUGGGGCGUGC
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AGGUGGUCGAGGUGUACGAGUACGCGCAGAAGCUGGGCGGCGAGGCCGGGUUCUACCACA
UCCUCAACAACCCGAAGGCCAGCCUGCUGUCCCUCACCCAGUUCCCGCACUUCAGCAGCG
UGGUCCUGGGGAACGCCGCCGGCCUGGGGAUCAUGGGCGAGUACCGCGGGACCCCGCGGA
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UCAACUACUCCGUGCUGGACCUCACCGCCGAGGAGCUGGAGGCGAUCAAGCACCAGCUGA
ACCCCAAGGACAACGACGUCGAGCUCUGAGGACUAGUGCAUCACAUUUAAAAGCAUCUCA
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UUUUCGUUGGUGUAAAGCCAACACCCUGUCUAAAAAACAUAAAUUUCUUUAAUCAUUUUG
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AAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAUGCAUC
CCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCAAAGGCUCUUUUCAGAGCCACCAGAAUU

Fig. 4

RSV-M_{2-1} (GC) R2833

GGGGCGCUGCCUACGGAGGUGGCAGCCAUCUCCUUCUCGGCAUCAAGCUUACCAUGAGCC

GCCGGAACCCCUGCAAGUUCGAGAUCCGCGGCCACUGCCUGAACGGGAAGCGGUGCCACU

UCUCCCACAACUACUUCGAGUGGCCGCCCCACGCCCUCCUGGUGCGCCAGAACUUCAUGC

UGAACCGGAUCCUCAAGAGCAUGGACAAGUCCAUCGACACCCUGAGCGAGAUCUCCGGCG

CCGCGGAGCUGGACCGCACCGAGGAGUACGCCCUCGGGGUCGUGGGCGUGCUGGAGAGCU

ACAUCGGGUCCAUCAACAACAUCACGAAGCAGAGCGCCUGCGUCGCCAUGUCCAAGCUGC

UCACCGAGCUGAACAGCGACGACAUCAAGAAGCUGCGGGACAACGAGGAGCUCAACUCCC

CCAAGAUCCGCGUGUACAACACCGUGAUCAGCUACAUCGAGUCCAACCGGAAGAACAACA

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ACACCCUGGACAUCCACAAGAGCAUCACCAUCAACAACCCGAAGGAGCUCACCGUGUCCG

ACACGAACGACCACGCGAAGAACAACGACACCACCUGAGGACUAGUGCAUCACAUUUAAA

AGCAUCUCAGCCUACCAUGAGAAUAAGAGAAAGAAAAUGAAGAUCAAUAGCUUAUUCAUC

UCUUUUUCUUUUUCGUUGGUGUAAAGCCAACACCCUGUCUAAAAAACAUAAAUUUCUUUA

AUCAUUUUGCCUCUUUUCUCUGUGCUUCAAUUAAUAAAAAAUGGAAAGAACCUAGAUCUA

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CAGAAUU

Fig. 5

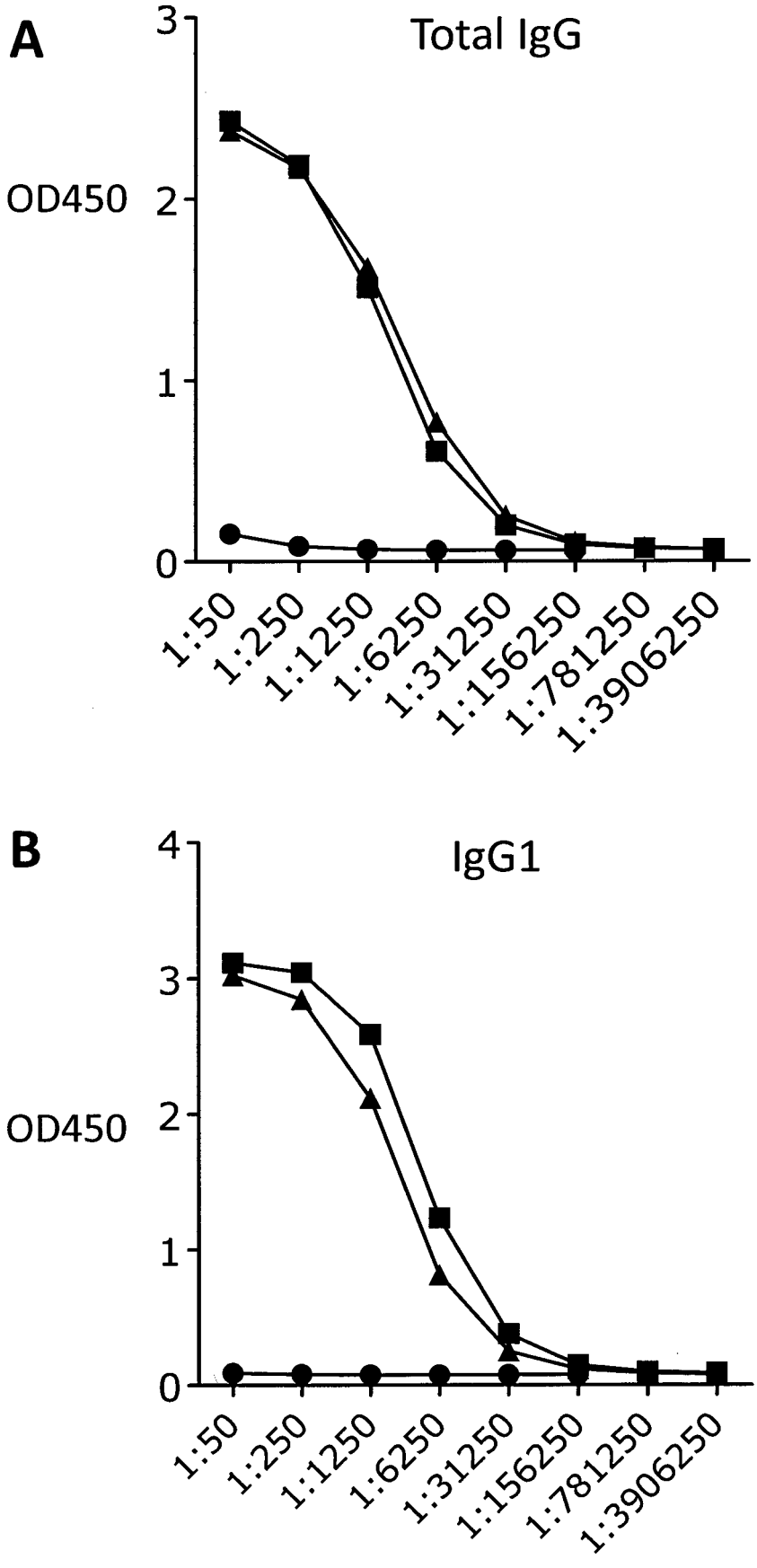

A
Total IgG
OD450
3
2
1
0
1:50
1:250
1:1250
1:6250
1:31250
1:156250
1:781250
1:3906250
B
IgG1
OD450
4
3
2
1
0
1:50
1:250
1:1250
1:6250
1:31250
1:156250
1:781250
1:3906250

Fig. 6

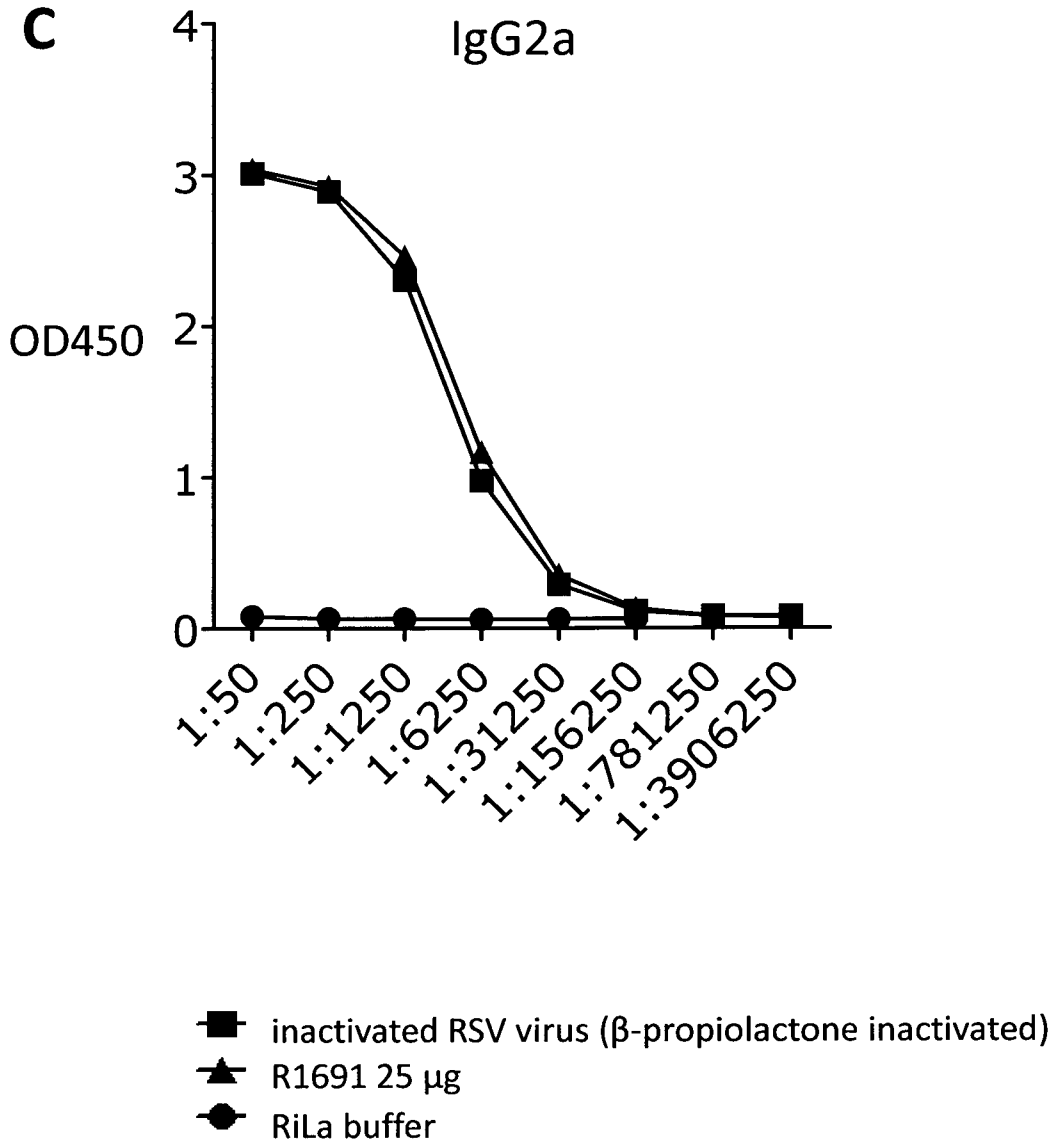


Fig. 6 - continued

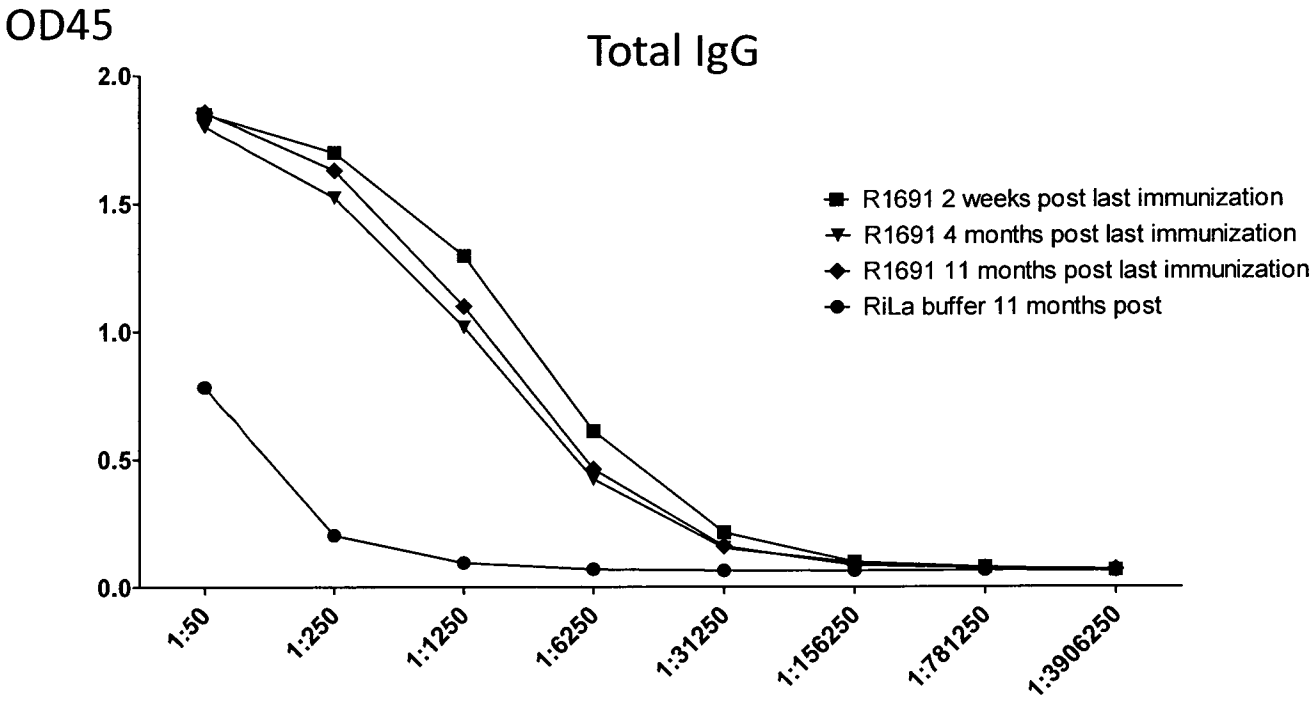

OD45
Total IgG
2.0
1.5
1.0
0.5
0.0
1:50
1:250
1:1250
1:6250
1:31250
1:156250
1:781250
1:3906250
R1691 2 weeks post last immunization
R1691 4 months post last immunization
R1691 11 months post last immunization
RiLa buffer 11 months post

Fig. 7

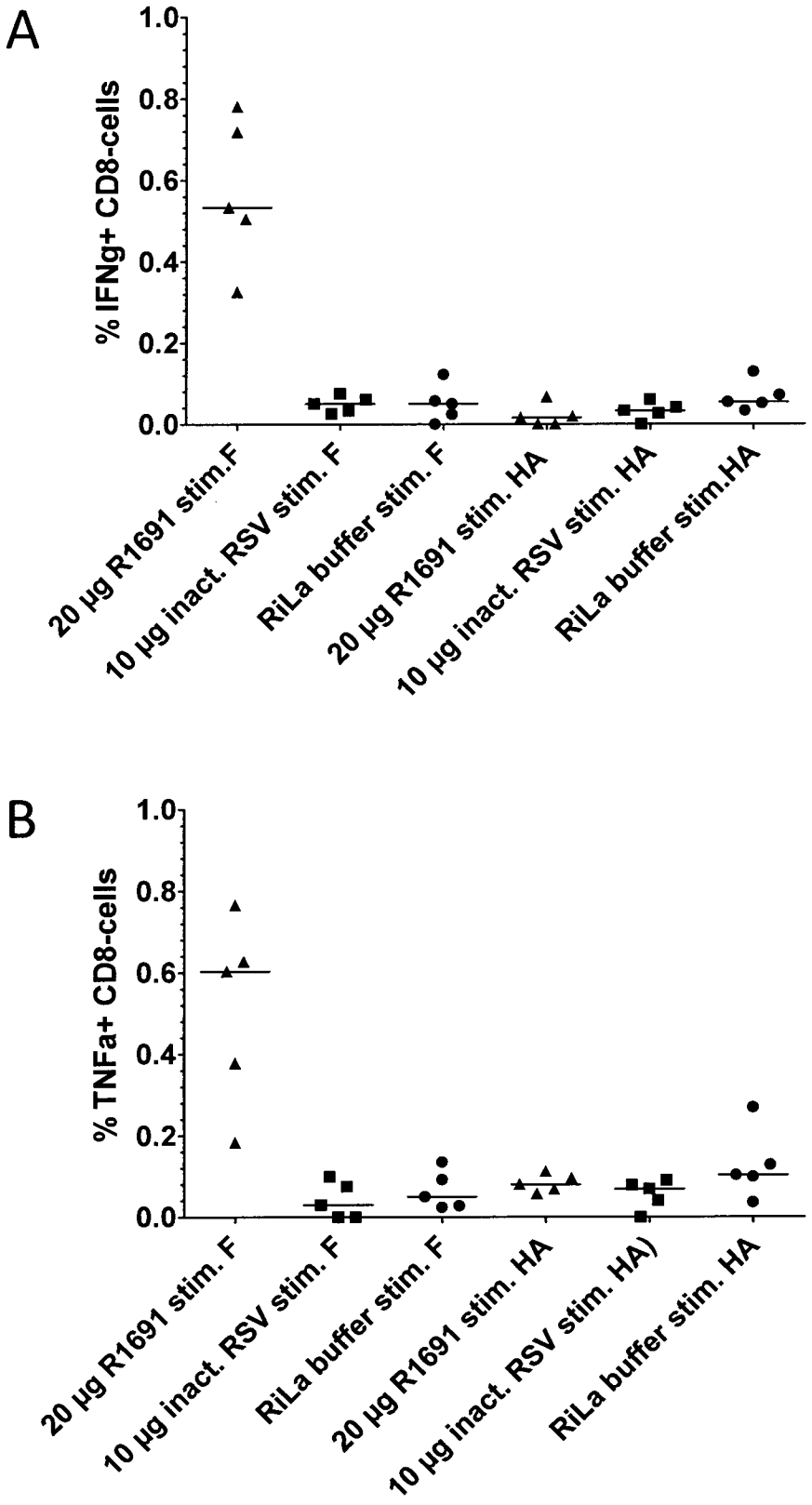
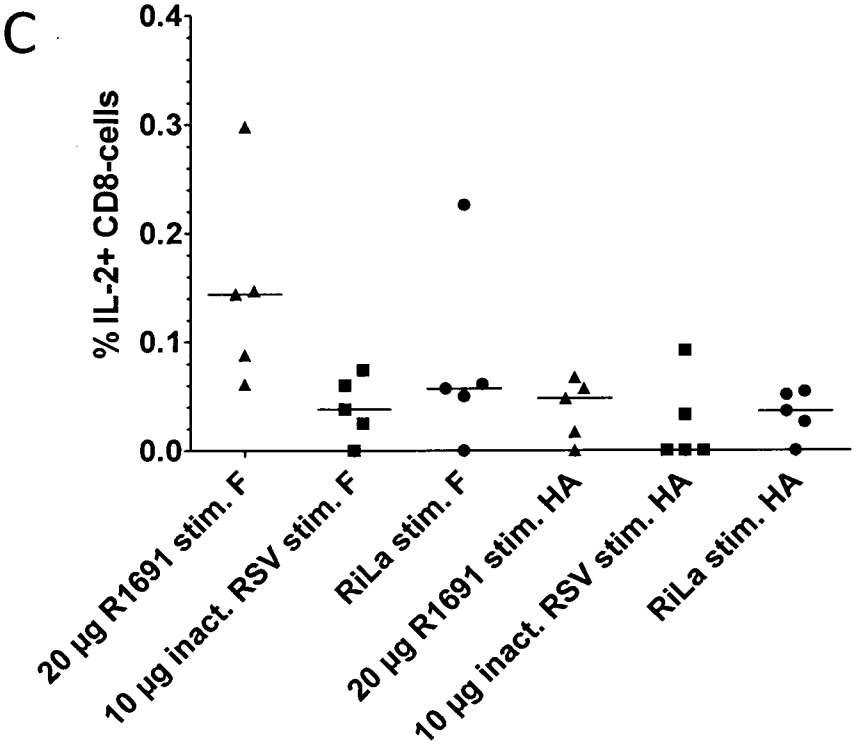

A
% IFNg+ CD8-cells
1.0
0.8
0.6
0.4
0.2
0.0
20 µg R1691 stim.F
10 µg inact. RSV stim. F
RiLa buffer stim. F
20 µg R1691 stim. HA
10 µg inact. RSV stim. HA
RiLa buffer stim.HA
B
% TNFa+ CD8-cells
1.0
0.8
0.6
0.4
0.2
0.0
20 µg R1691 stim. F
10 µg inact. RSV stim. F
RiLa buffer stim. F
20 µg R1691 stim. HA
10 µg inact. RSV stim. HA)
RiLa buffer stim. HA

Fig. 8



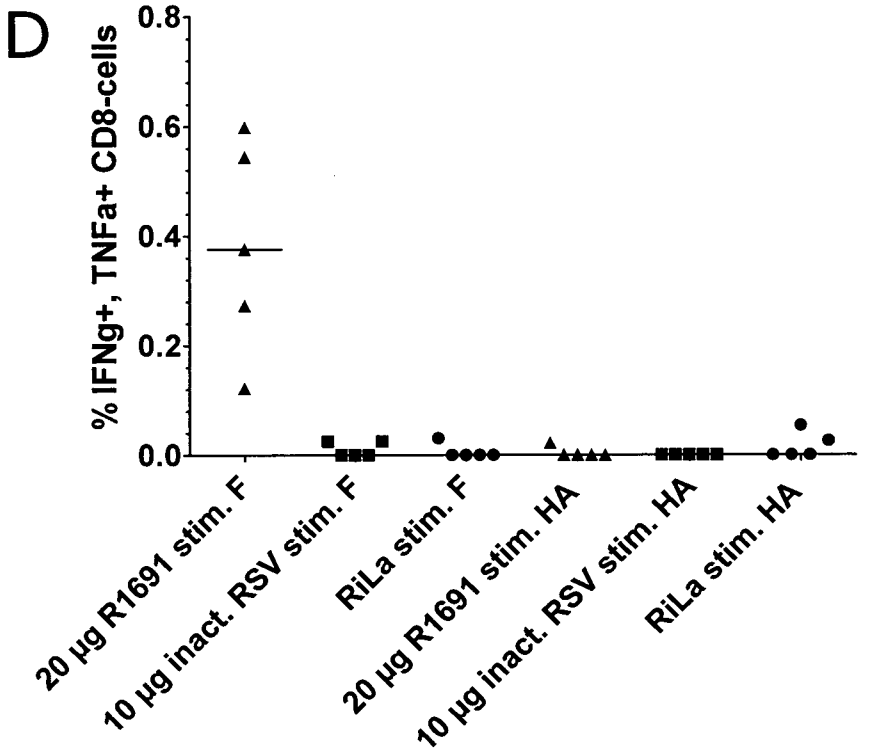
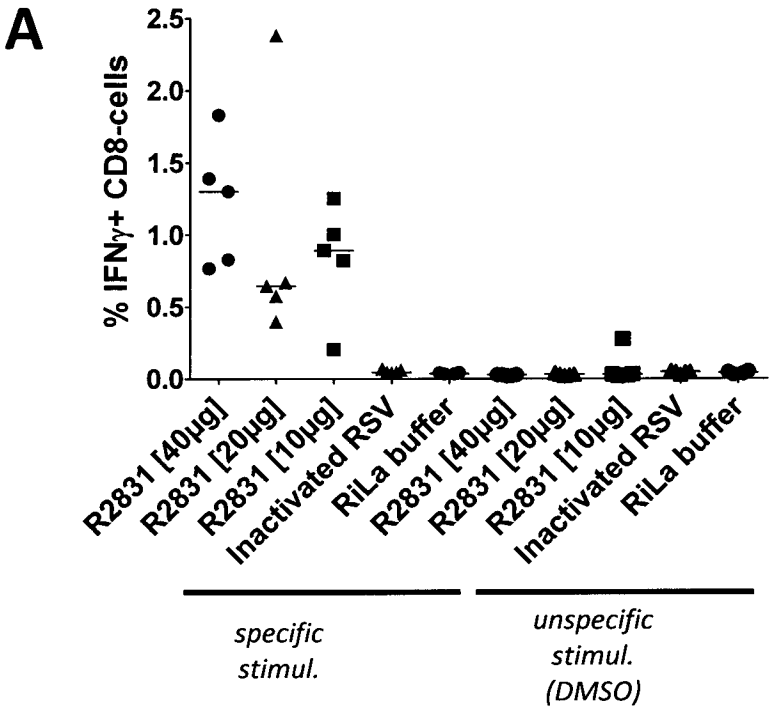


Fig. 8 – continued


A
% IFNγ+ CD8-cells
2.5
2.0
1.5
1.0
0.5
0.0
R2831 [40µg]
R2831 [20µg]
R2831 [10µg]
Inactivated RSV
RiLa buffer
R2831 [40µg]
R2831 [20µg]
R2831 [10µg]
Inactivated RSV
RiLa buffer
specific stimul.
unspecific stimul. (DMSO)

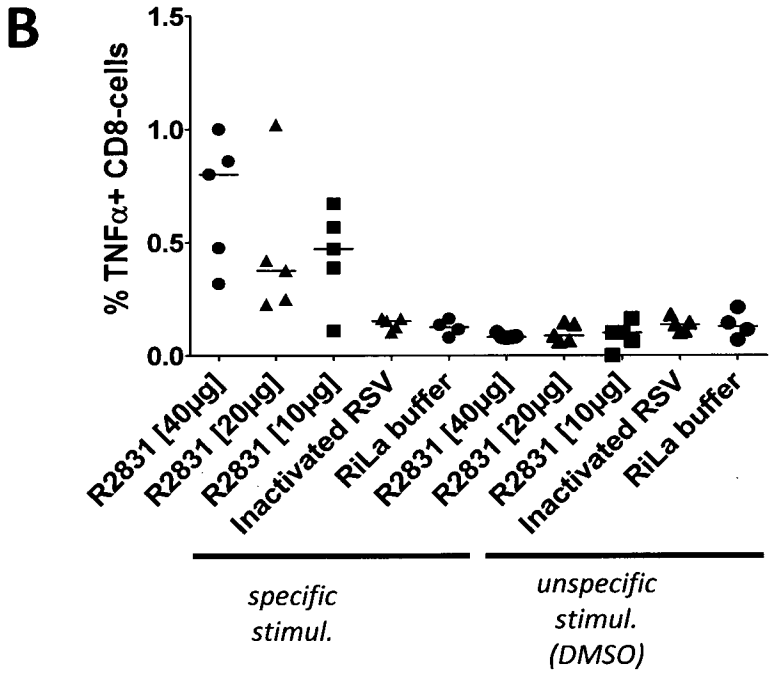

B
% TNFα+ CD8-cells
1.5
1.0
0.5
0.0
R2831 [40µg]
R2831 [20µg]
R2831 [10µg]
Inactivated RSV
RiLa buffer
R2831 [40µg]
R2831 [20µg]
R2831 [10µg]
Inactivated RSV
RiLa buffer
specific stimul.
unspecific stimul. (DMSO)

Fig. 9

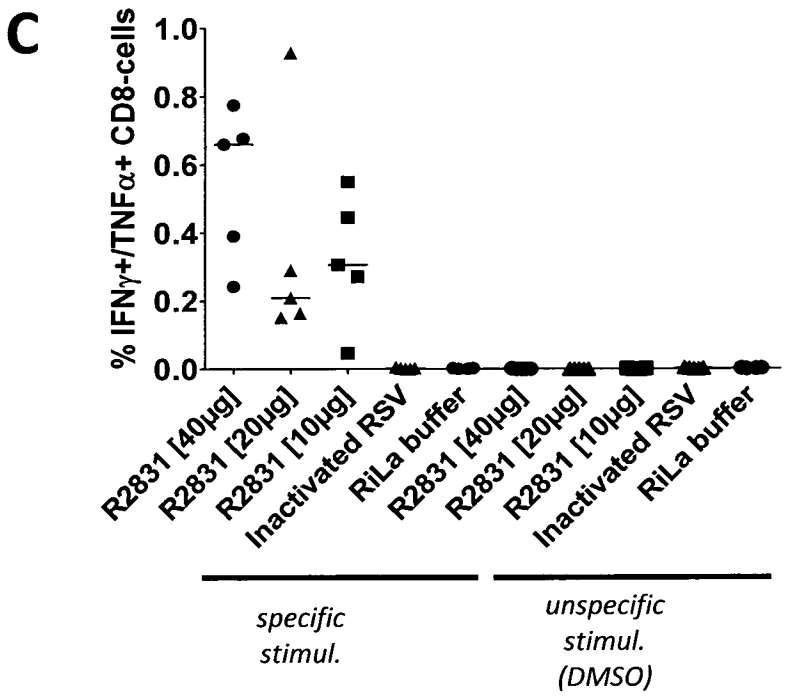
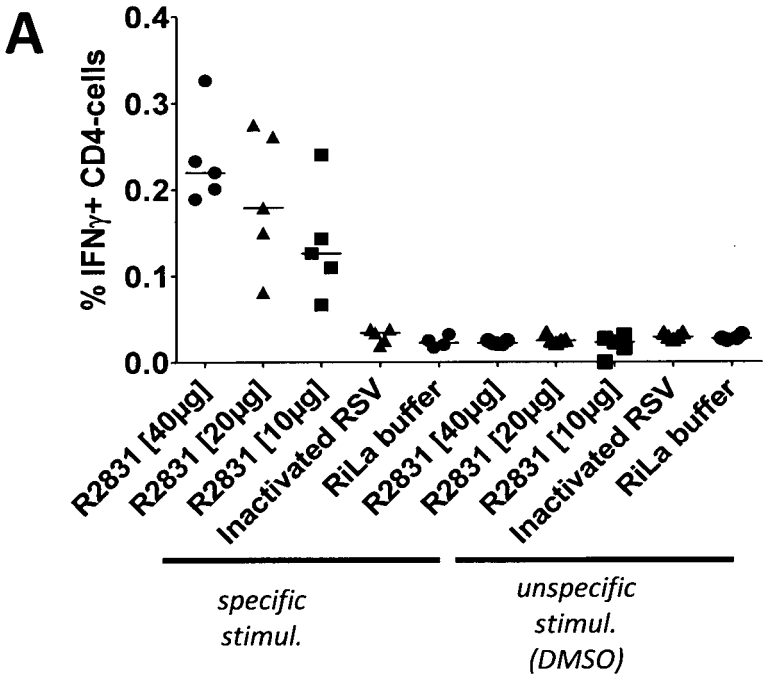


Fig. 9 – continued


A
% IFNγ+ CD4-cells
0.4
0.3
0.2
0.1
0.0
R2831 [40µg]
R2831 [20µg]
R2831 [10µg]
Inactivated RSV
RiLa buffer
R2831 [40µg]
R2831 [20µg]
R2831 [10µg]
Inactivated RSV
RiLa buffer
specific stimul.
unspecific stimul. (DMSO)

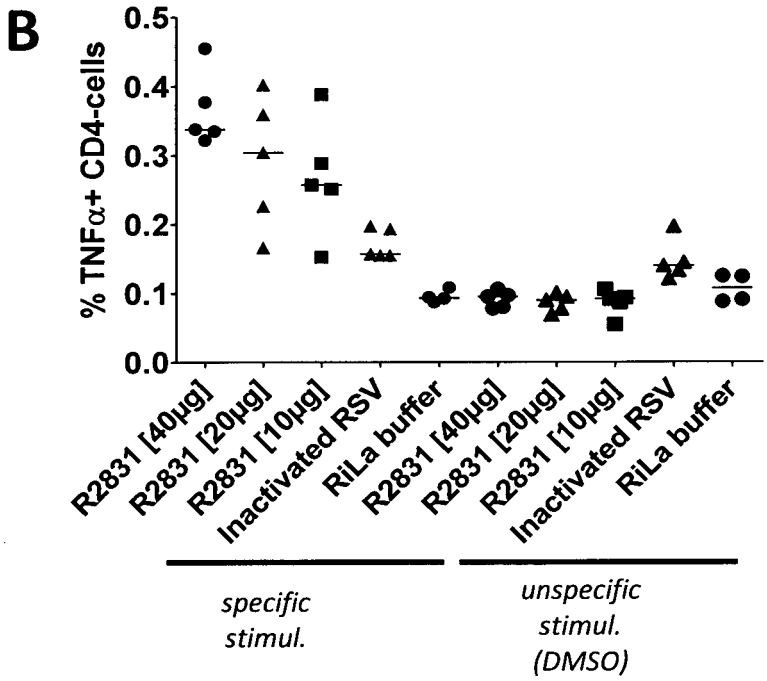

B
% TNFα+ CD4-cells
0.5
0.4
0.3
0.2
0.1
0.0
R2831 [40µg]
R2831 [20µg]
R2831 [10µg]
Inactivated RSV
RiLa buffer
R2831 [40µg]
R2831 [20µg]
R2831 [10µg]
Inactivated RSV
RiLa buffer
specific stimul.
unspecific stimul. (DMSO)

Fig. 10

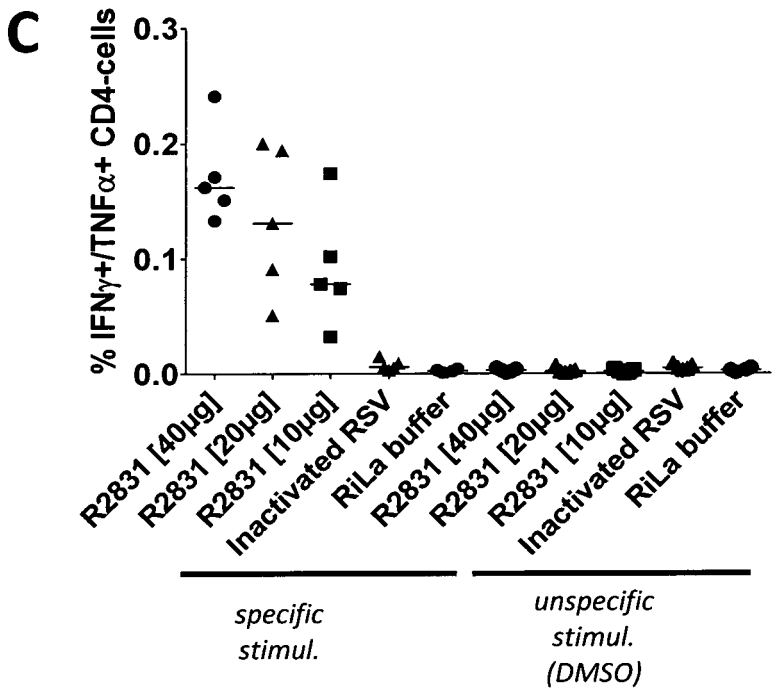
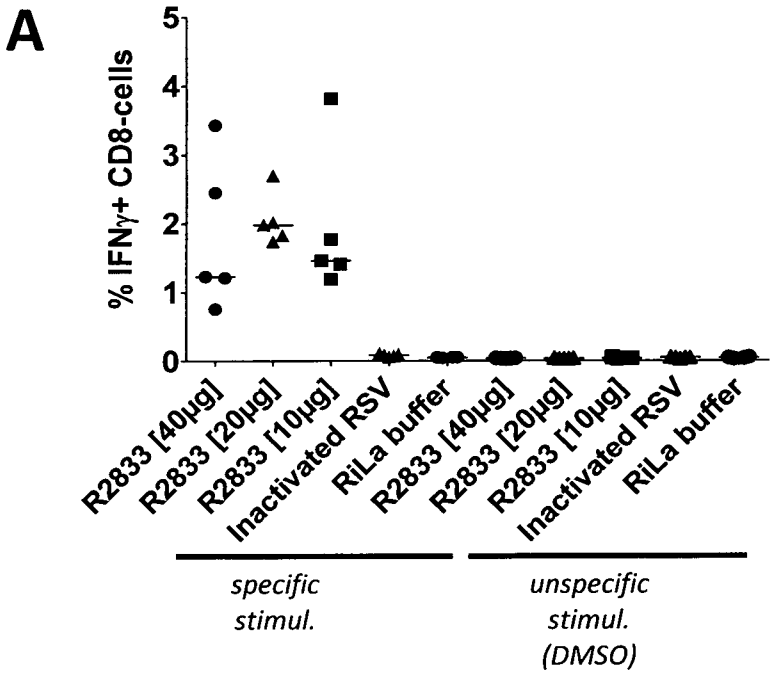


Fig. 10 – continued


A
% IFNγ+ CD8-cells
5
4
3
2
1
0
R2833 [40µg]
R2833 [20µg]
R2833 [10µg]
Inactivated RSV
RiLa buffer
R2833 [40µg]
R2833 [20µg]
R2833 [10µg]
Inactivated RSV
RiLa buffer
specific stimul.
unspecific stimul. (DMSO)

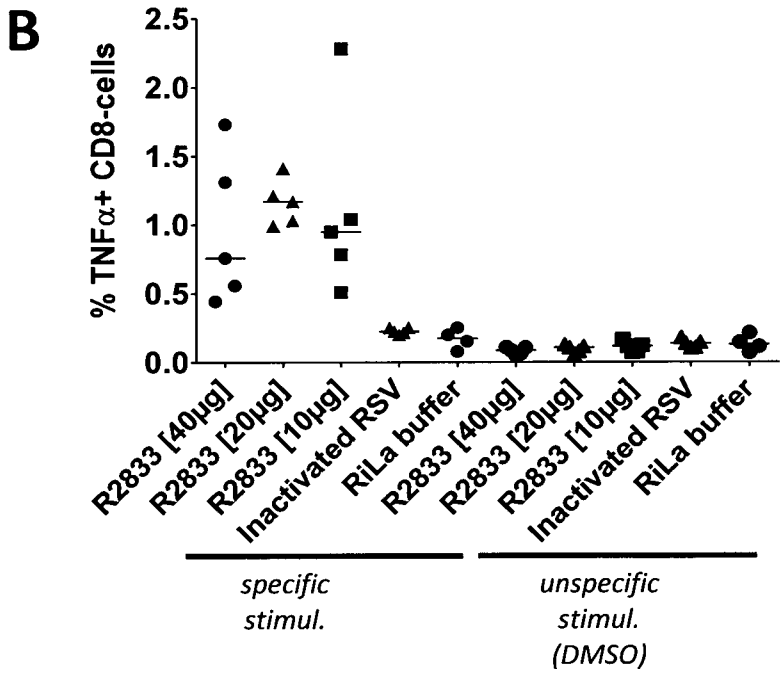

B
% TNFα+ CD8-cells
2.5
2.0
1.5
1.0
0.5
0.0
R2833 [40µg]
R2833 [20µg]
R2833 [10µg]
Inactivated RSV
RiLa buffer
R2833 [40µg]
R2833 [20µg]
R2833 [10µg]
Inactivated RSV
RiLa buffer
specific stimul.
unspecific stimul. (DMSO)

Fig. 11

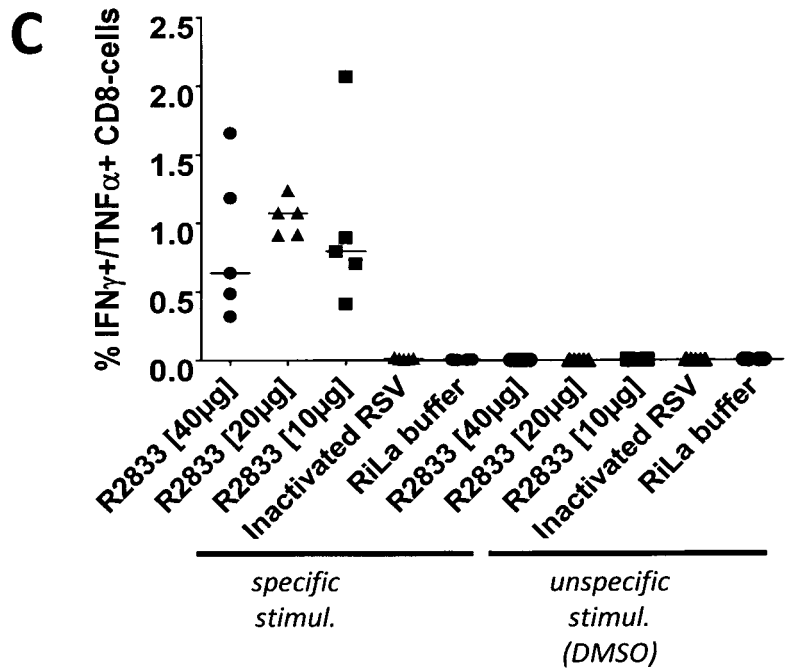


Fig. 11 – continued

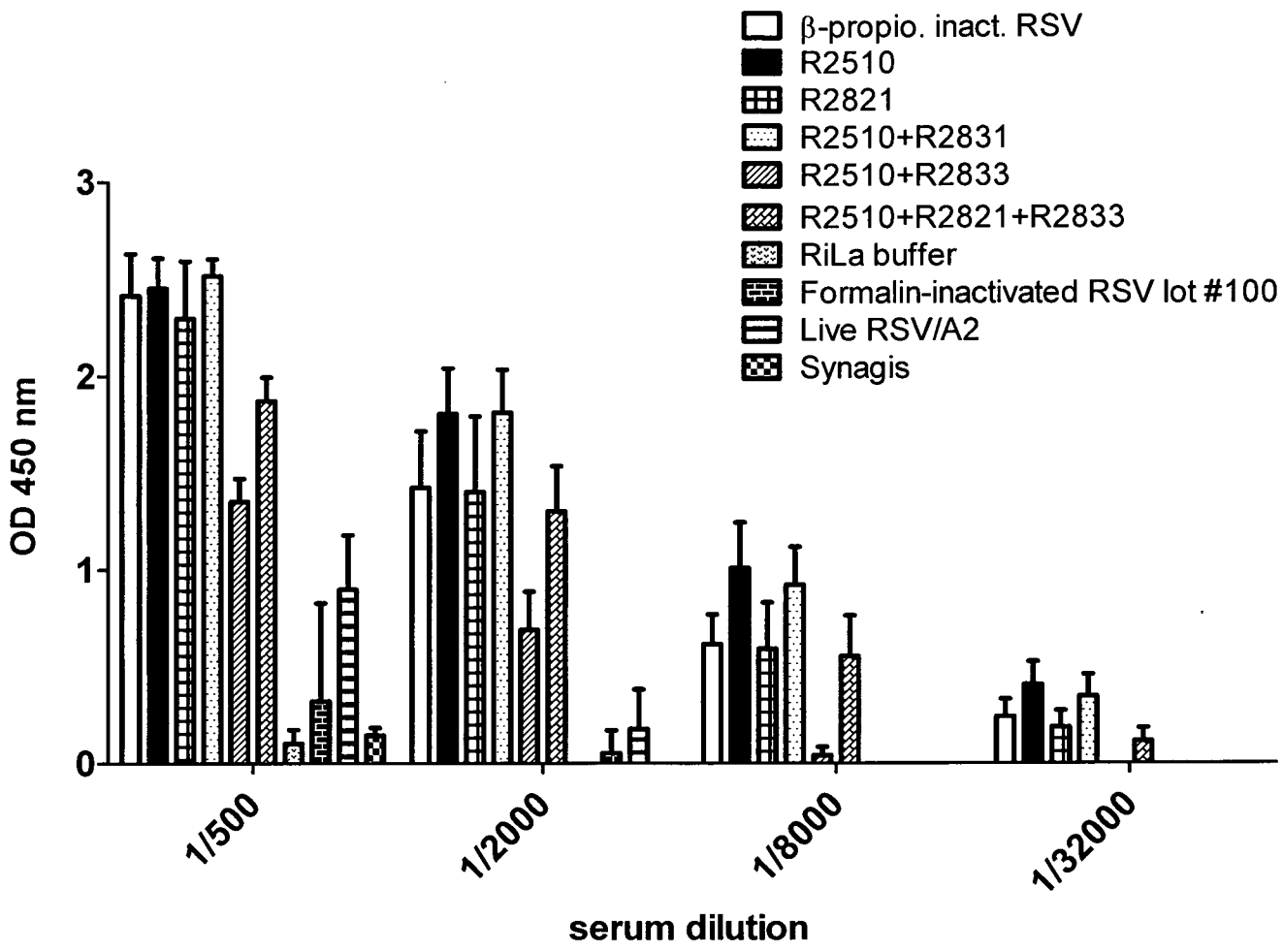

β-propio. inact. RSV
R2510
R2821
R2510+R2831
R2510+R2833
R2510+R2821+R2833
RiLa buffer
Formalin-inactivated RSV lot #100
Live RSV/A2
Synagis
OD 450 nm
3
2
1
0
1/500
1/2000
1/8000
1/32000
serum dilution

Fig. 12

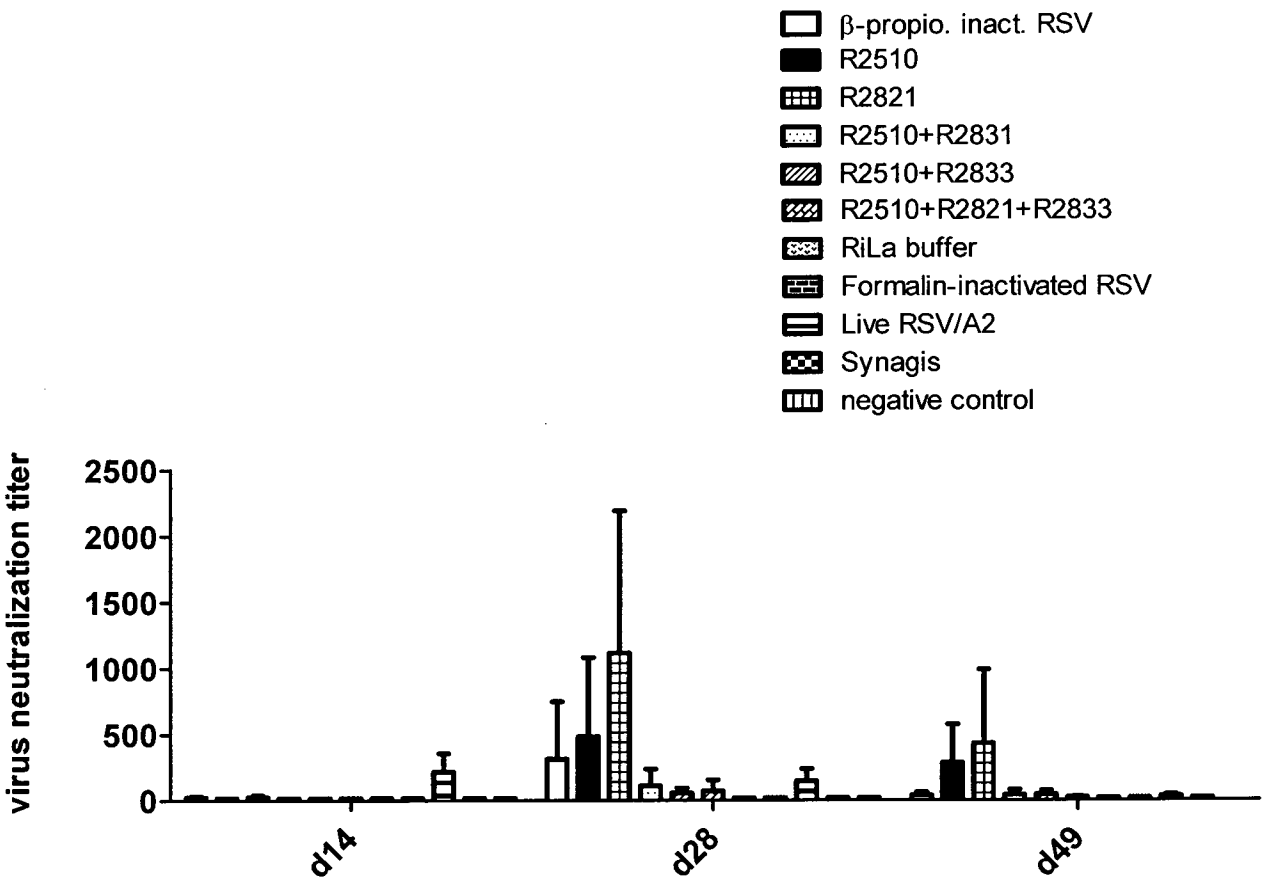

β-propio. inact. RSV
R2510
R2821
R2510+R2831
R2510+R2833
R2510+R2821+R2833
RiLa buffer
Formalin-inactivated RSV
Live RSV/A2
Synagis
negative control
virus neutralization titer
2500
2000
1500
1000
500
0
d14
d28
d49

Fig. 13

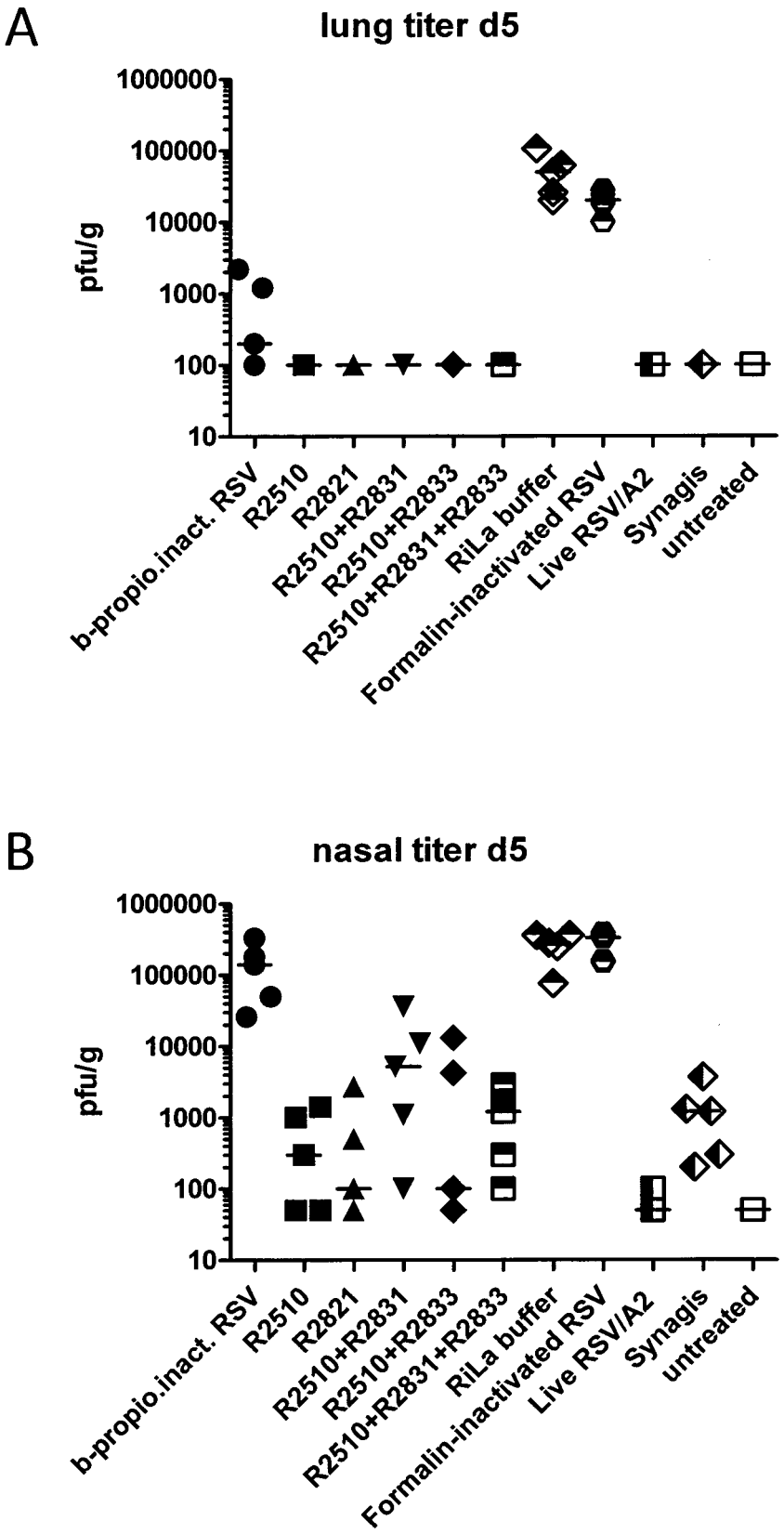

A
lung titer d5
pfu/g
1000000
100000
10000
1000
100
10
b-propio.inact. RSV
R2510
R2821
R2510+R2831
R2510+R2833
R2510+R2831+R2833
RiLa buffer
Formalin-inactivated RSV
Live RSV/A2
Synagis
untreated
B
nasal titer d5
pfu/g
1000000
100000
10000
1000
100
10
b-propio.inact. RSV
R2510
R2821
R2510+R2831
R2510+R2833
R2510+R2831+R2833
RiLa buffer
Formalin-inactivated RSV
Live RSV/A2
Synagis
untreated

Fig. 14

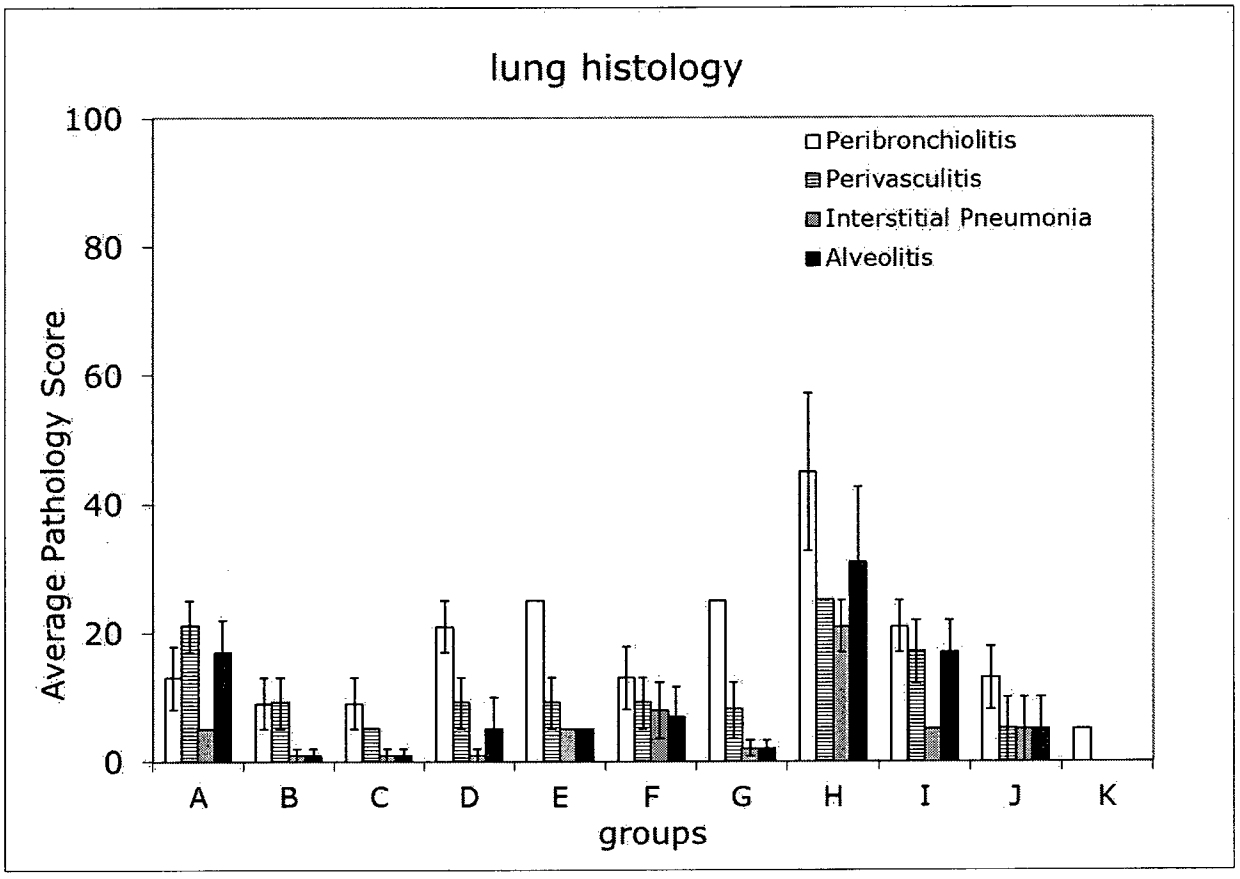

lung histology
Average Pathology Score
100
80
60
40
20
0
A
B
C
D
E
F
G
H
I
J
K
groups
Peribronchiolitis
Perivasculitis
Interstitial Pneumonia
Alveolitis

Fig. 15

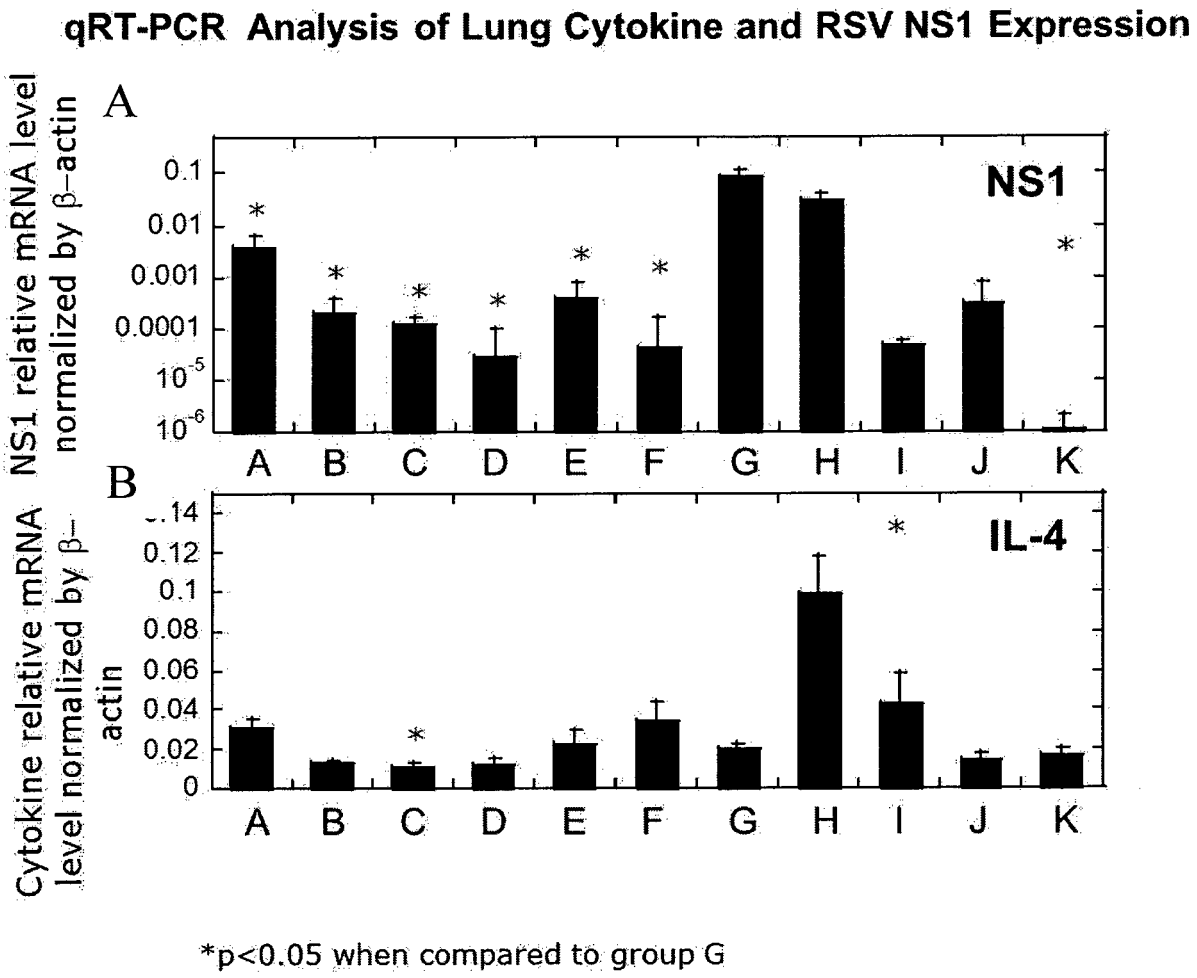

qRT-PCR Analysis of Lung Cytokine and RSV NS1 Expression
A
NS1 relative mRNA level normalized by β-actin
0.1
0.01
0.001
0.0001
10-5
10-6
NS1
A B C D E F G H I J K
B
Cytokine relative mRNA level normalized by β-actin
0.14
0.12
0.1
0.08
0.06
0.04
0.02
0
IL-4
A B C D E F G H I J K
*p<0.05 when compared to group G

Fig. 16

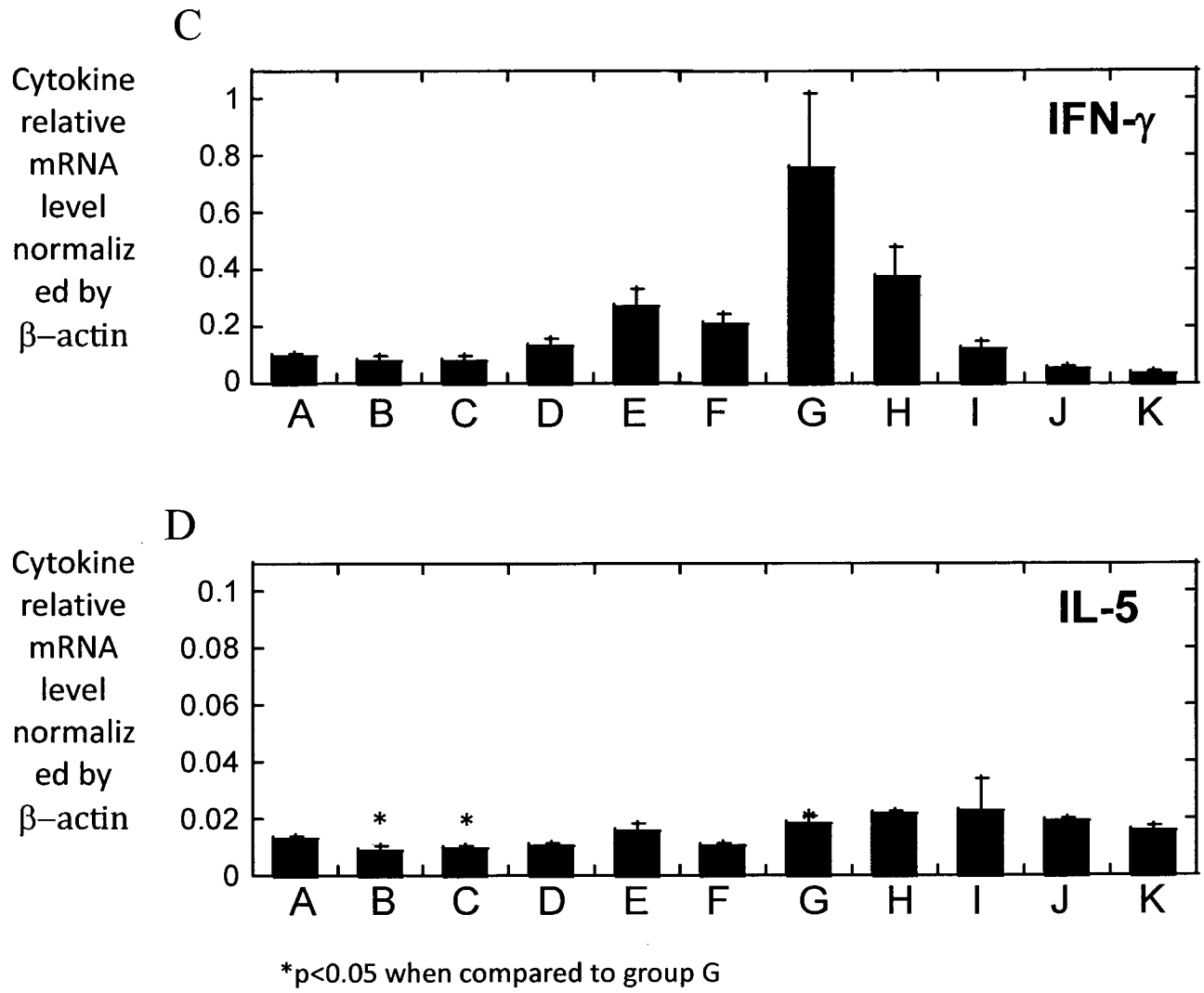
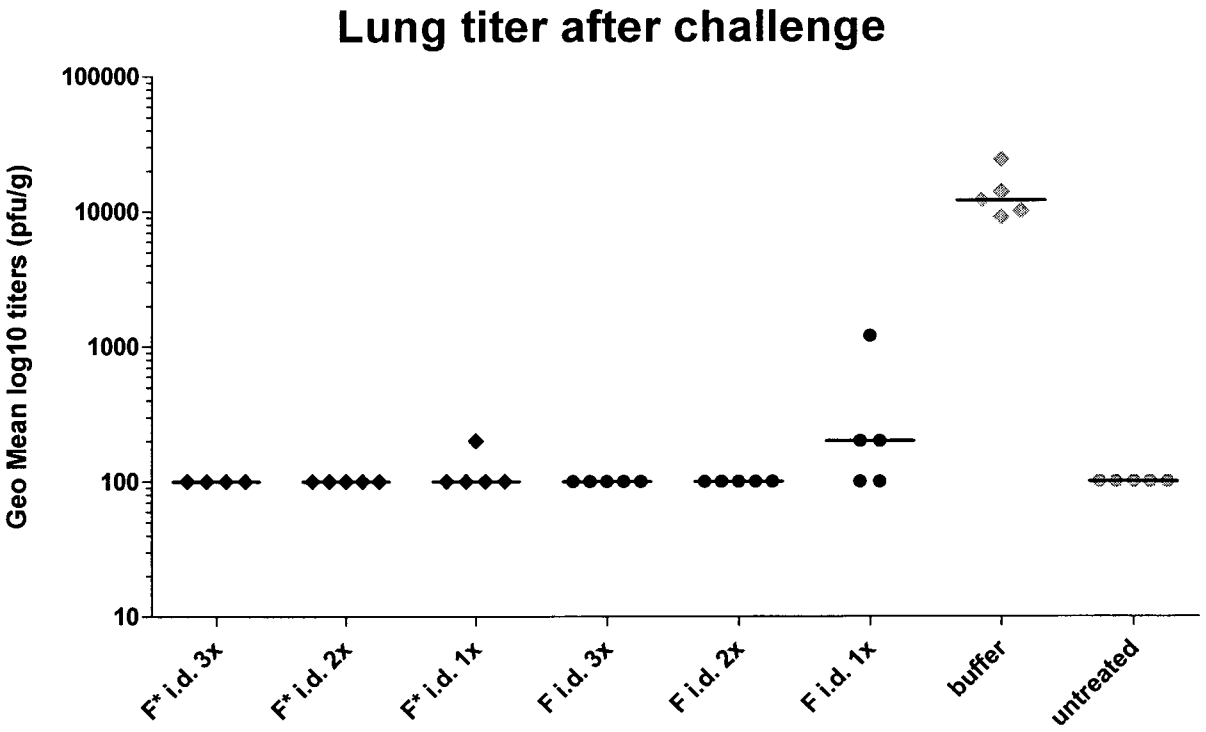


Fig. 16 –continued

A


Lung titer after challenge
Geo Mean log10 titers (pfu/g)
100000
10000
1000
100
10
F* i.d. 3x
F* i.d. 2x
F* i.d. 1x
F i.d. 3x
F i.d. 2x
F i.d. 1x
buffer
untreated

B

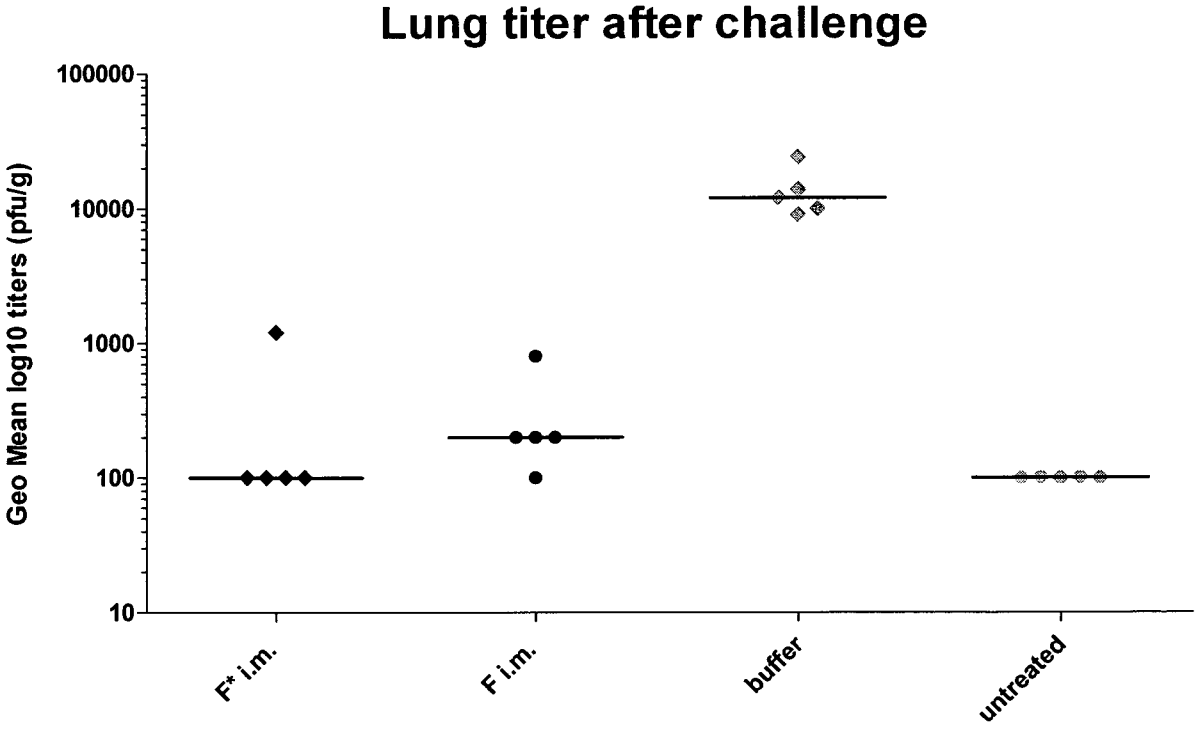

Lung titer after challenge
Geo Mean log10 titers (pfu/g)
100000
10000
1000
100
10
F* i.m.
F i.m.
buffer
untreated

Fig. 17