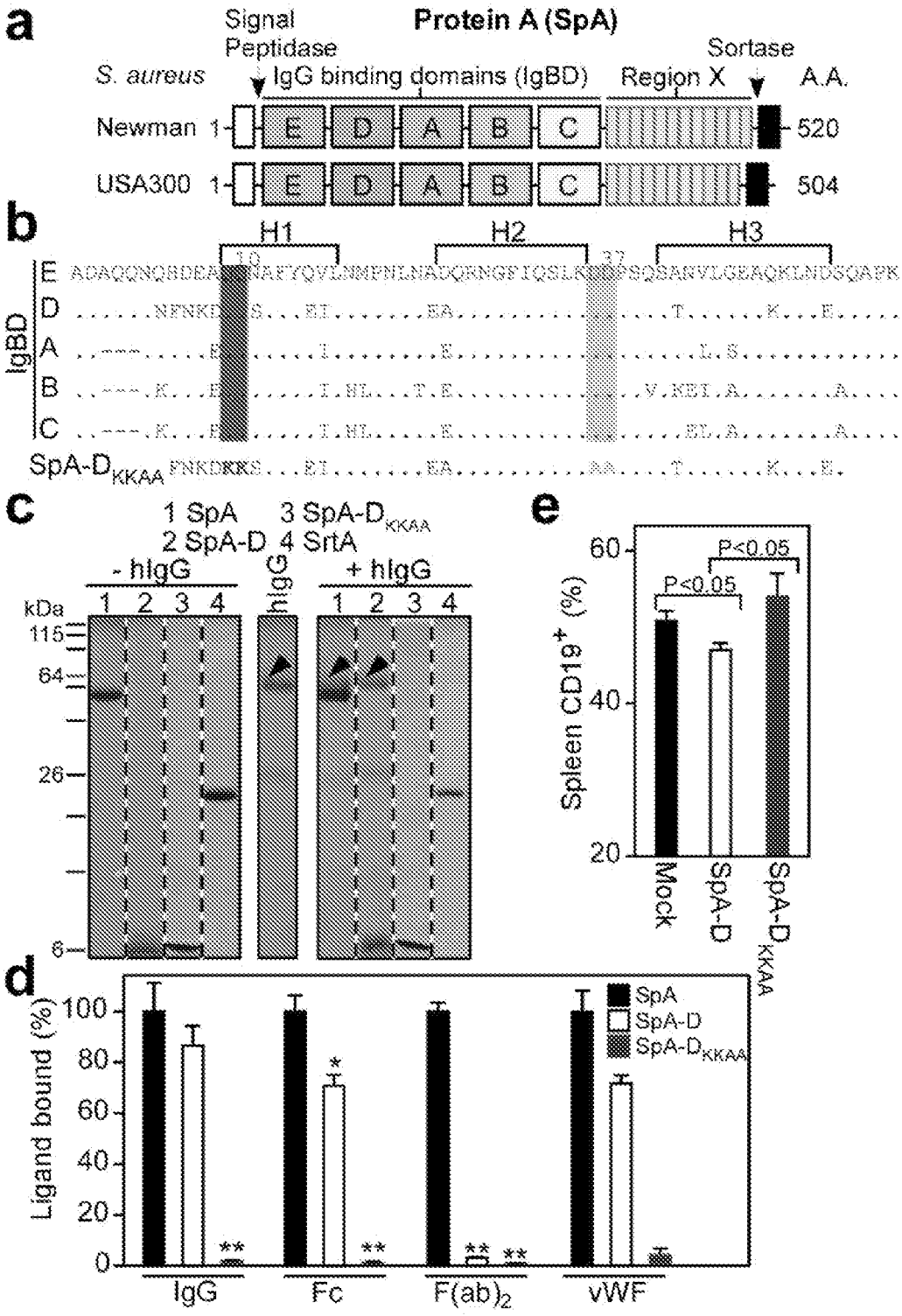

a
Signal Peptidase
Protein A (SpA)
Sortase
S. aureus
IgG binding domains (IgBD)
Region X
A.A.
Newman 1
E D A B C
520
USA300 1
E D A B C
504
b
H1
H2
H3
IgBD
E
D
A
B
C
SpA-D KKAA
c
1 SpA
2 SpA-D
3 SpA-D KKAA
4 SrtA
- hIgG
hIgG
+ hIgG
kDa
115
64
26
6
e
Spleen CD19+ (%)
60
40
20
P<0.05
P<0.05
Mock
SpA-D
SpA-D KKAA
d
Ligand bound (%)
100
80
60
40
20
0
SpA
SpA-D
SpA-D KKAA
IgG
Fc
F(ab)2
vWF

FIG. 1

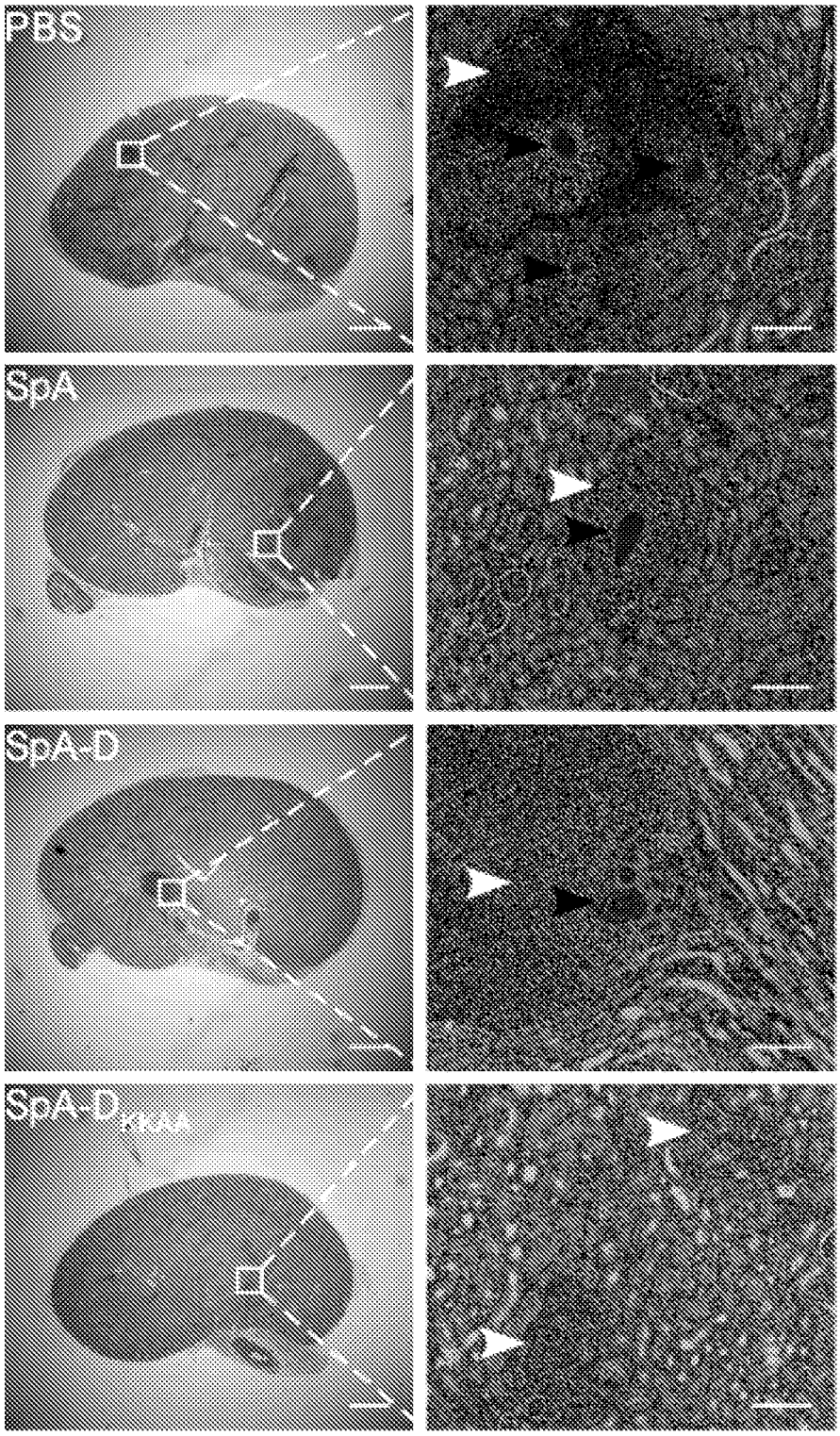


FIG. 2

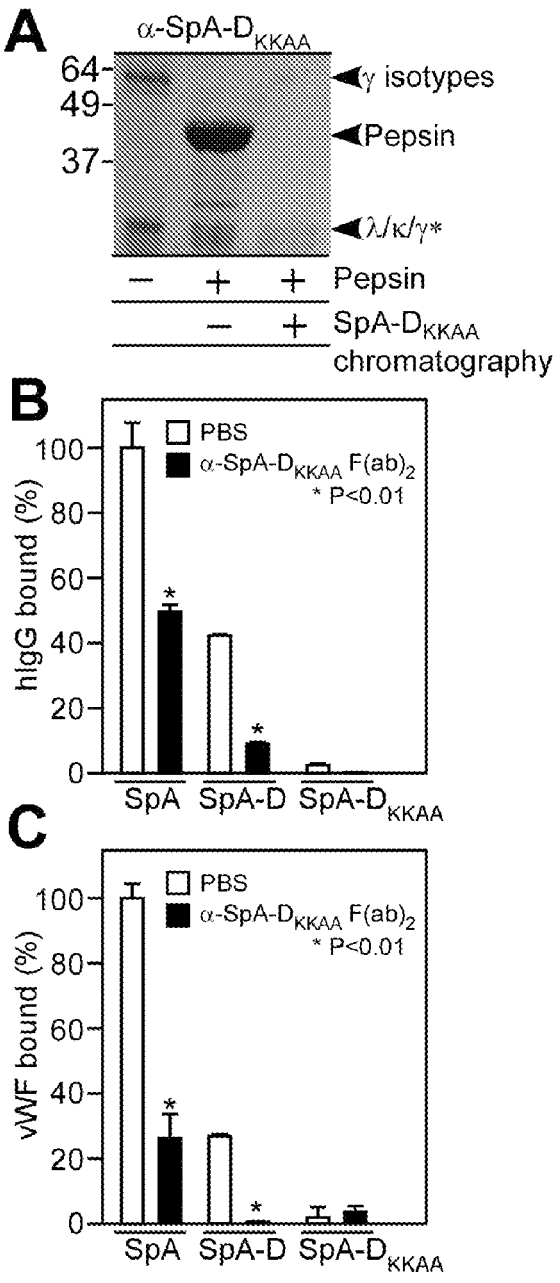

A
α-SpA-D$_{KKAA}$
64
49
37
γ isotypes
Pepsin
λ/κ/γ*
— + + Pepsin
— + SpA-D$_{KKAA}$
chromatography
B
PBS
α-SpA-D$_{KKAA}$ F(ab)$_2$
* P<0.01
hIgG bound (%)
100
80
60
40
20
0
SpA SpA-D SpA-D$_{KKAA}$
C
PBS
α-SpA-D$_{KKAA}$ F(ab)$_2$
* P<0.01
vWF bound (%)
100
80
60
40
20
0
SpA SpA-D SpA-D$_{KKAA}$

FIG. 3

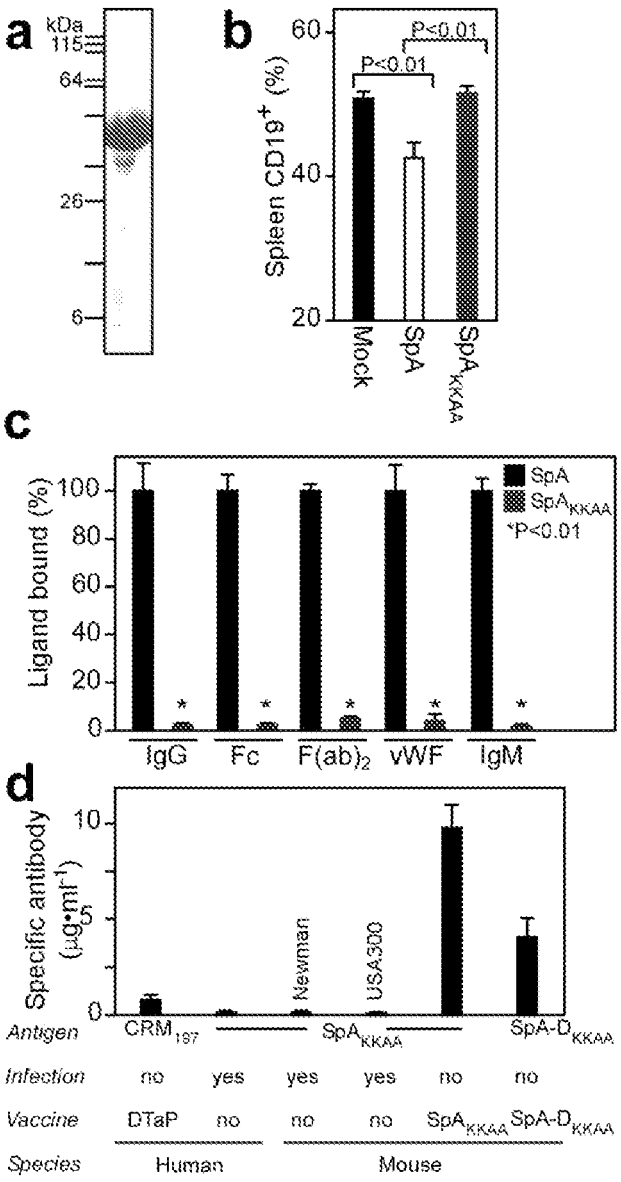

a
kDa
115
64
26
6
b
Spleen CD19+ (%)
60
40
20
P<0.01
P<0.01
Mock
SpA
SpAKKAA
c
Ligand bound (%)
100
80
60
40
20
0
SpA
SpAKKAA
*P<0.01
IgG
Fc
F(ab)2
vWF
IgM
d
Specific antibody (μg•ml-1)
10
5
0
Newman
USA300
Antigen
CRM197
SpAKKAA
SpA-DKKAA
Infection
no
yes
yes
yes
no
no
Vaccine
DTaP
no
no
no
SpAKKAA
SpA-DKKAA
Species
Human
Mouse

FIG. 4

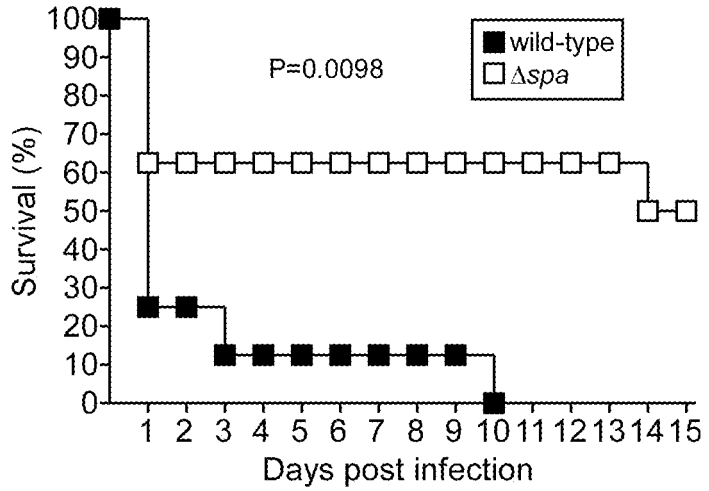

P=0.0098
wild-type
Δspa
Survival (%)
Days post infection

FIG. 5

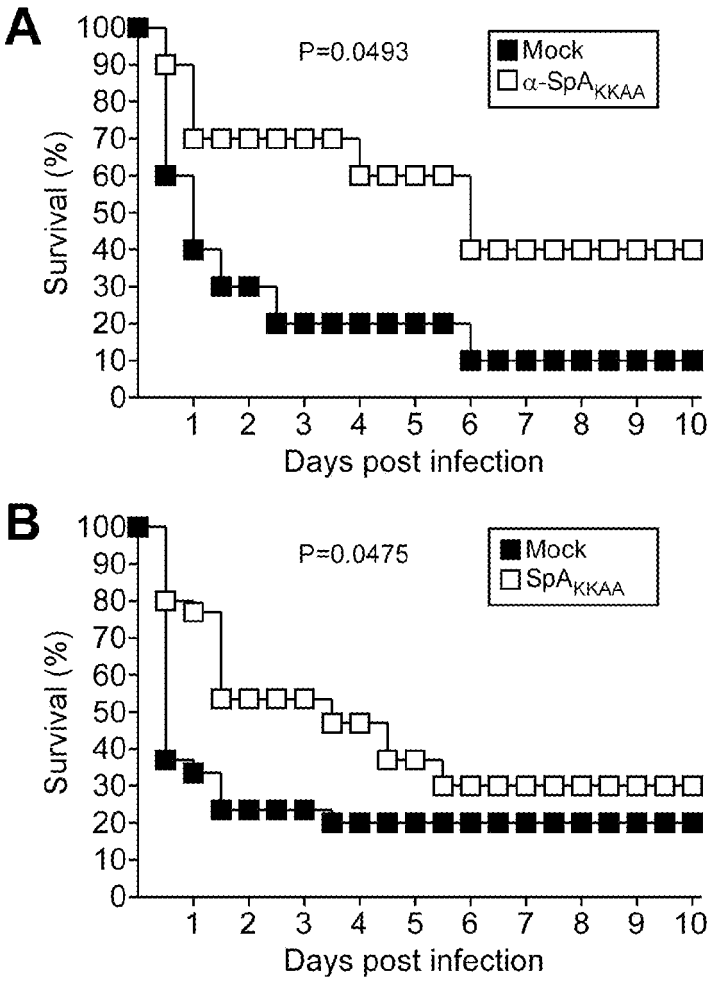

A
P=0.0493
Mock
α-SpA$_{KKAA}$
Survival (%)
Days post infection
B
P=0.0475
Mock
SpA$_{KKAA}$
Survival (%)
Days post infection

FIG. 6

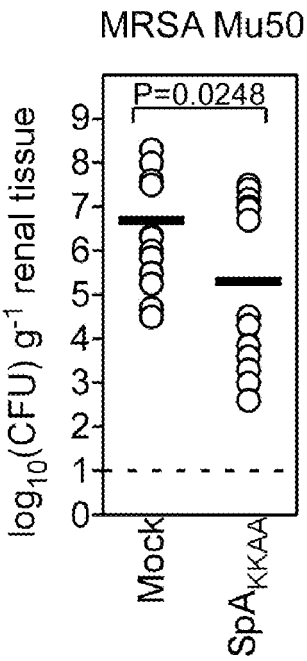

MRSA Mu50
P=0.0248
log10(CFU) g-1 renal tissue
0
1
2
3
4
5
6
7
8
9
Mock
SpAKKAA

FIG. 7

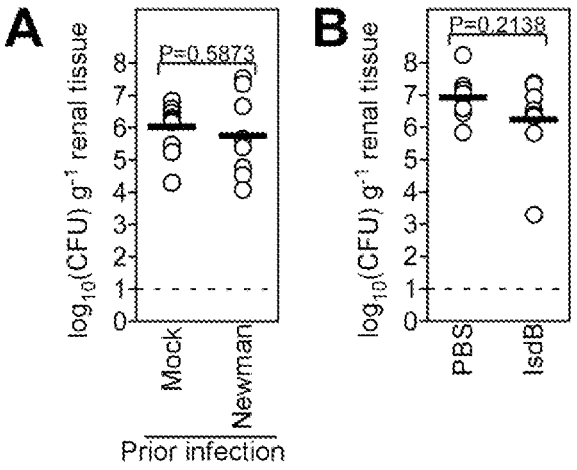

A
P=0.5873
log10(CFU) g-1 renal tissue
Mock
Newman
Prior infection
B
P=0.2138
log10(CFU) g-1 renal tissue
PBS
IsdB

FIG. 8

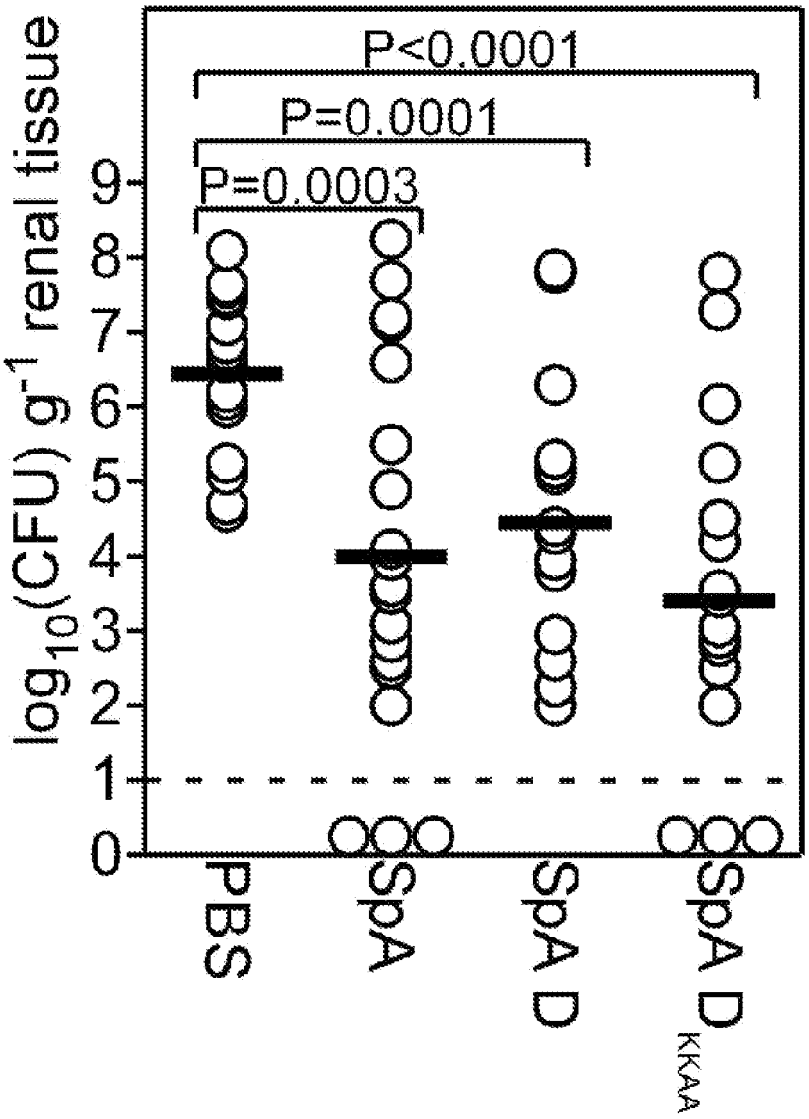

P<0.0001
P=0.0001
P=0.0003
log10(CFU) g-1 renal tissue
0
1
2
3
4
5
6
7
8
9
PBS
SpA
SpA D
SpA DKKAA

FIG. 9

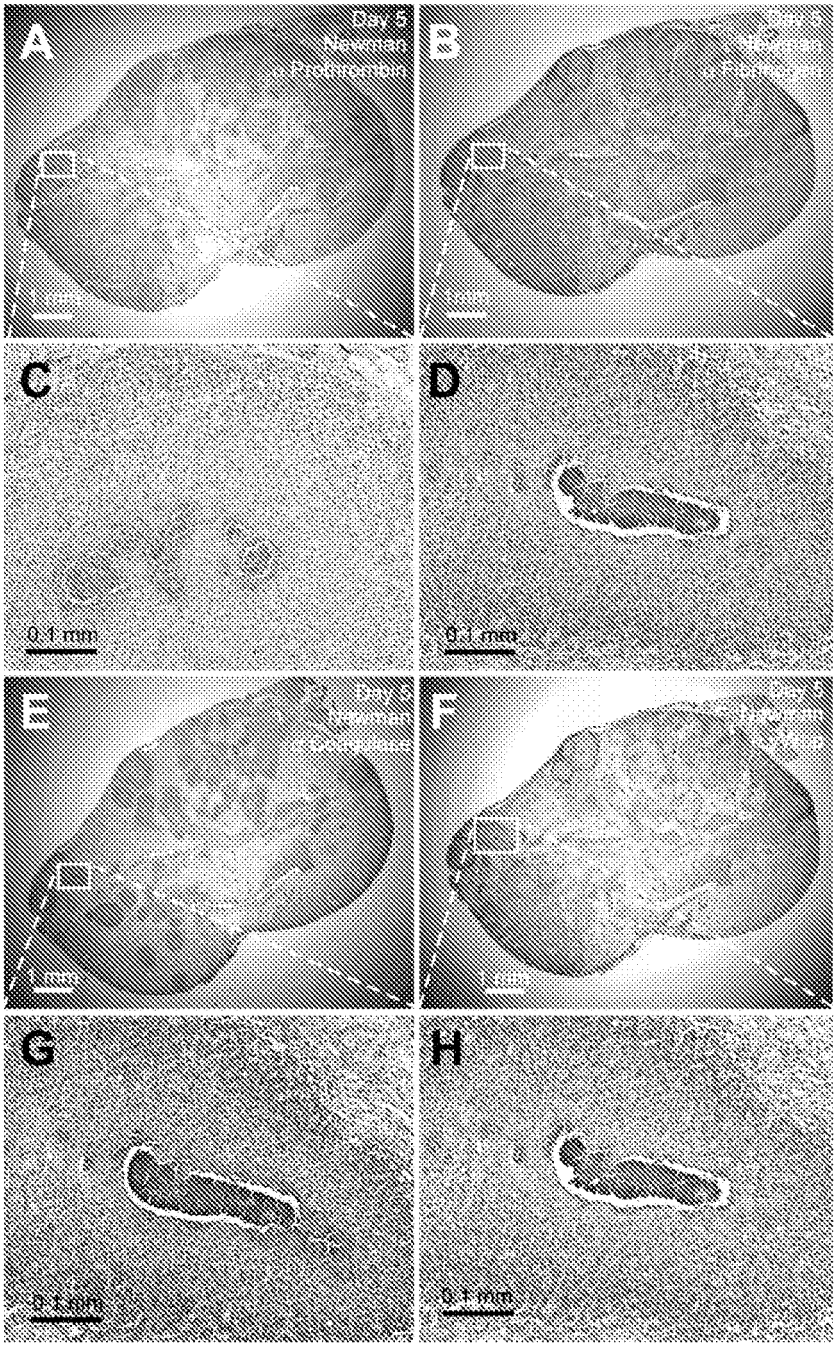
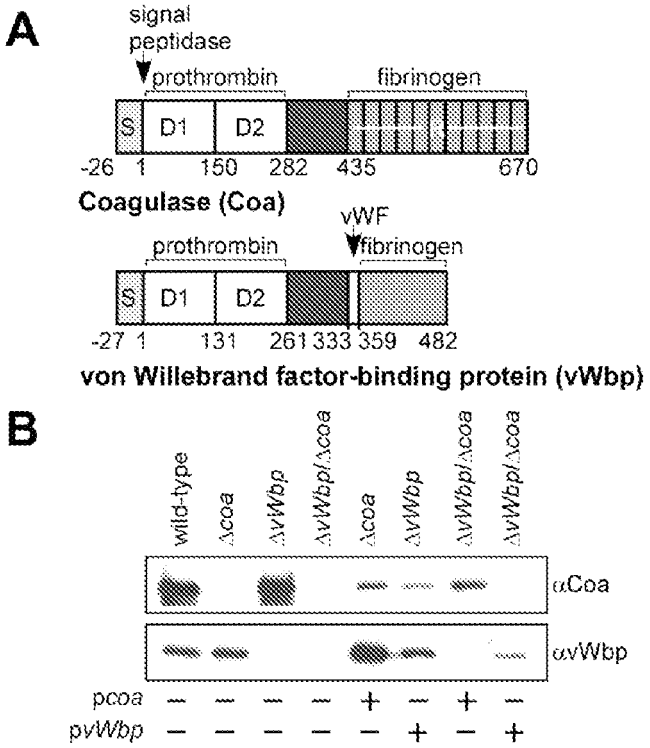


FIG. 10



C

***S. aureus* infection of lepirudin-treated mouse blood**

S. aureus strain	*24 hours*	*Coagulation (hours)*
Mock		>48
Newman		<12
Δ*vWbp*		24
Δ*coa*		36
Δ*vWbp*/Δ*coa*		>48
Δ*vWbp*/Δ*coa*+ p*vWbp*		<12
Δ*vWbp*/Δ*coa* + p*coa*		24
Δ*vWbp* + p*vWbp*		36
Δ*coa* + p*coa*		24

FIG. 11

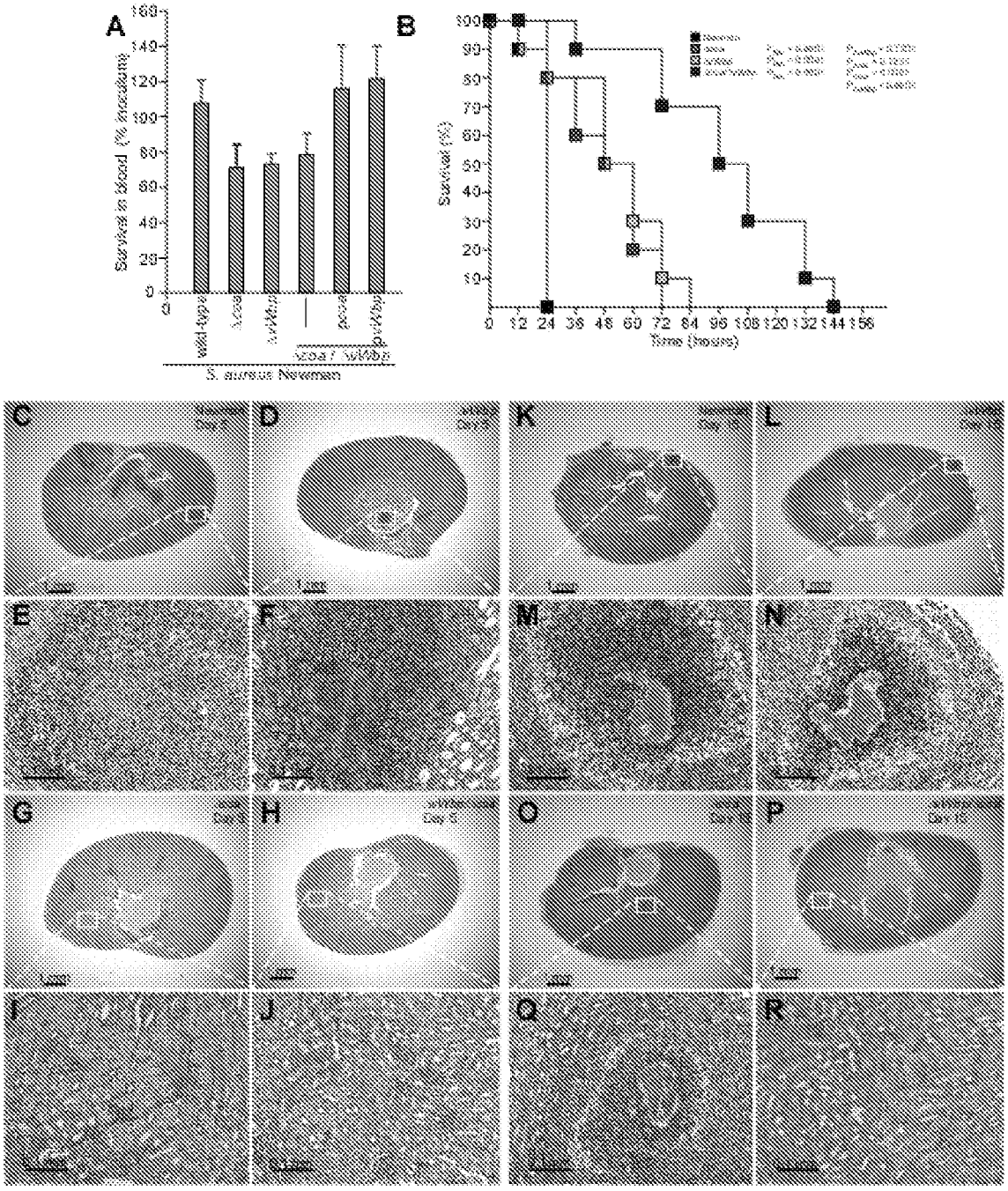
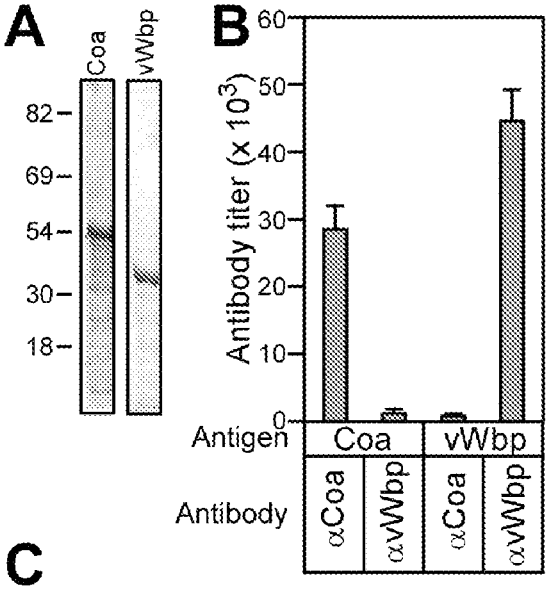


FIG. 12



C

***S. aureus* infection of mouse blood**

Antibody	Coagulation 24 hours	Coagulation Completed
Mock	—	24
αV10		24
αCoa		>48
αvWbp		>48
αCoa/αvWbp		>48

D

Mouse blood with	Coagulation time
PBS	indefinite
Coagulase	< 30 min
Coagulase + αV10	< 30 min
Coagulase + αCoa	2 hrs
Coagulase + αvWbp	1 hr
vWbp	< 30 min
vWbp + αV10	< 30 min
vWbp + αCoa	2 hrs
vWbp + αvWbp	2 hrs

FIG. 13

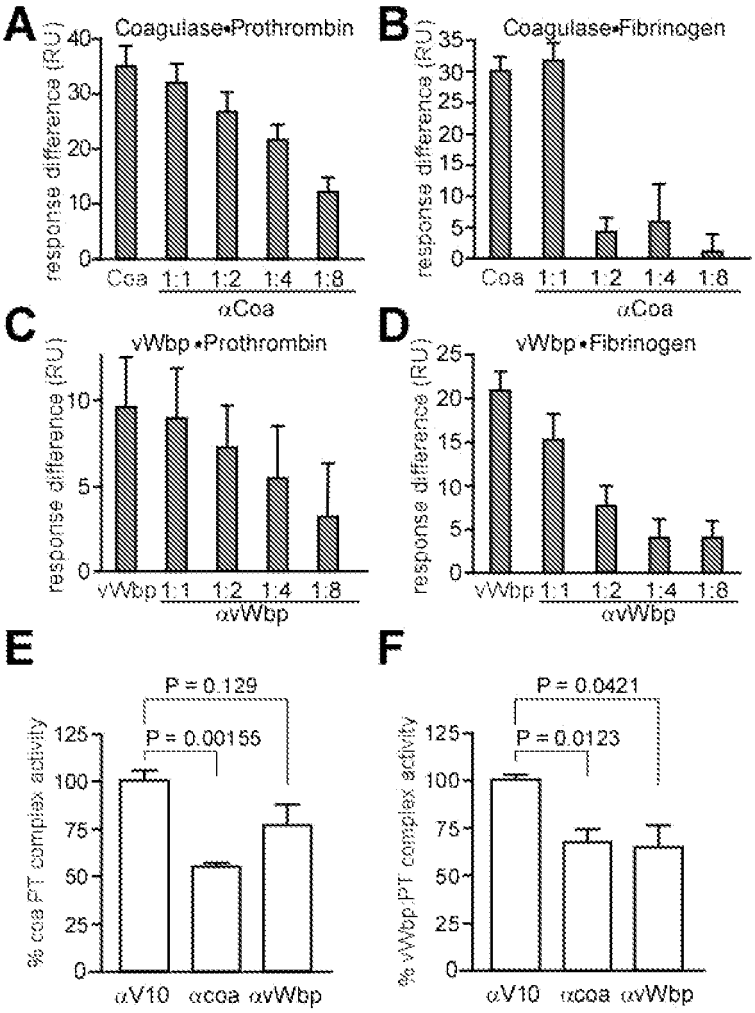

A
Coagulase•Prothrombin
response difference (RU)
Coa 1:1 1:2 1:4 1:8
αCoa
B
Coagulase•Fibrinogen
response difference (RU)
Coa 1:1 1:2 1:4 1:8
αCoa
C
vWbp•Prothrombin
response difference (RU)
vWbp 1:1 1:2 1:4 1:8
αvWbp
D
vWbp•Fibrinogen
response difference (RU)
vWbp 1:1 1:2 1:4 1:8
αvWbp
E
P = 0.129
P = 0.00155
% coa PT complex activity
αV10 αcoa αvWbp
F
P = 0.0421
P = 0.0123
% vWbp PT complex activity
αV10 αcoa αvWbp

FIG. 14

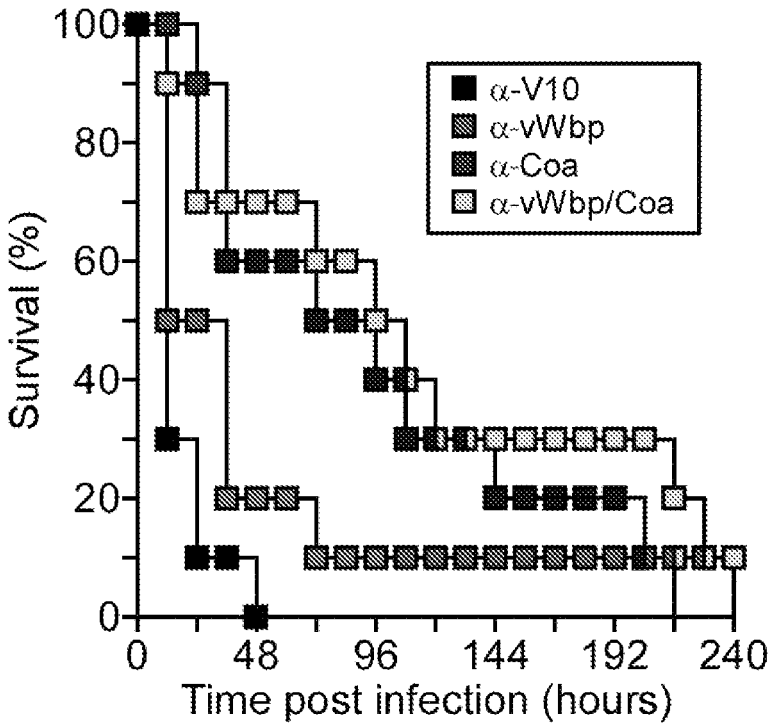

100
80
60
40
20
0
Survival (%)
0
48
96
144
192
240
Time post infection (hours)
α-V10
α-vWbp
α-Coa
α-vWbp/Coa

FIG. 15

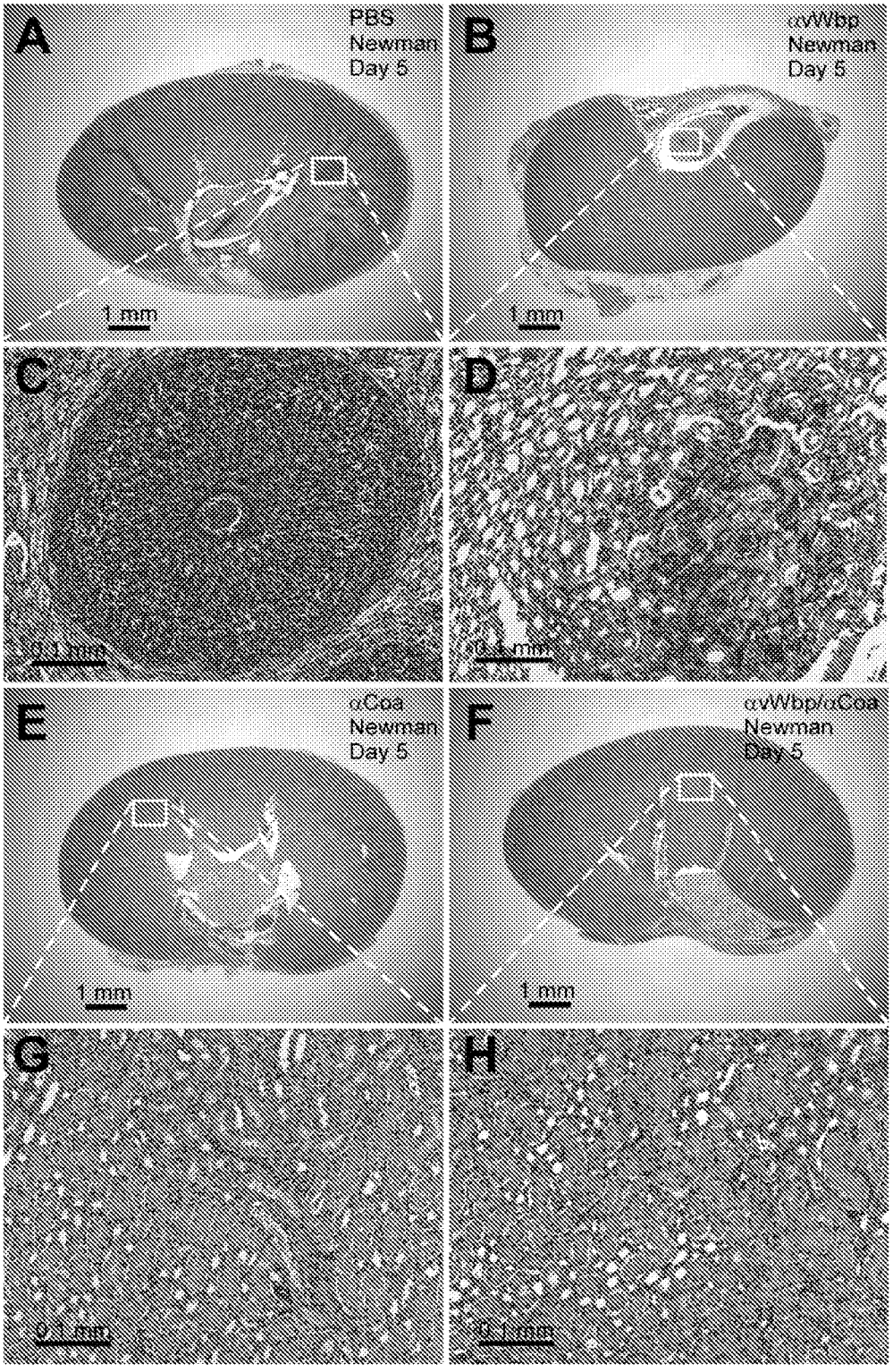

A
PBS
Newman
Day 5
1 mm
B
αvWbp
Newman
Day 5
1 mm
C
50 μm
D
50 μm
E
αCoa
Newman
Day 5
1 mm
F
αvWbp/αCoa
Newman
Day 5
1 mm
G
50 μm
H
50 μm

FIG. 16

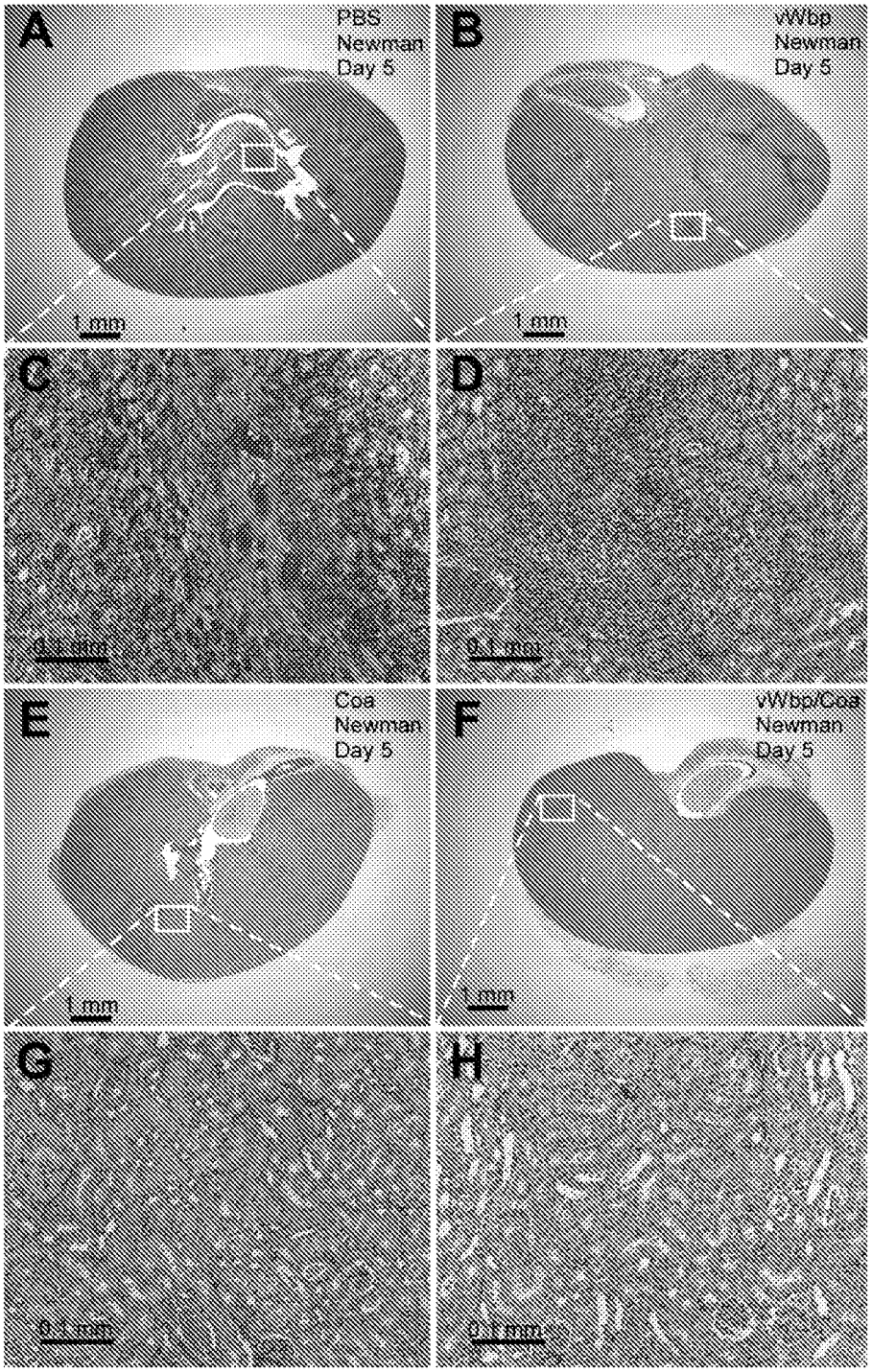

A
PBS
Newman
Day 5
1 mm
B
vWbp
Newman
Day 5
1 mm
C
0.1 mm
D
0.1 mm
E
Coa
Newman
Day 5
1 mm
F
vWbp/Coa
Newman
Day 5
1 mm
G
0.1 mm
H
0.1 mm

FIG. 17