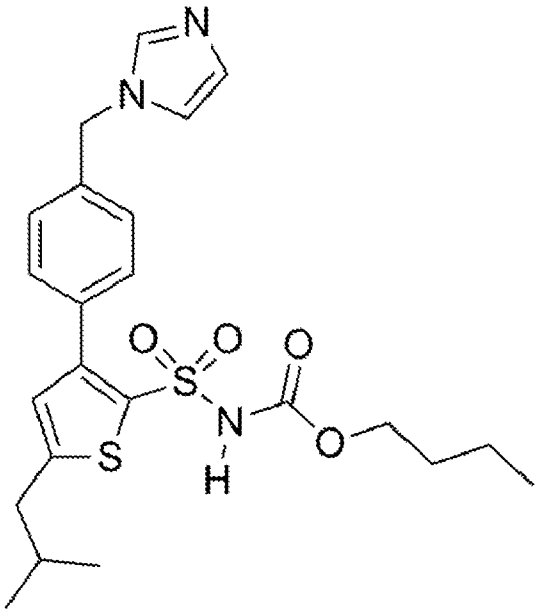
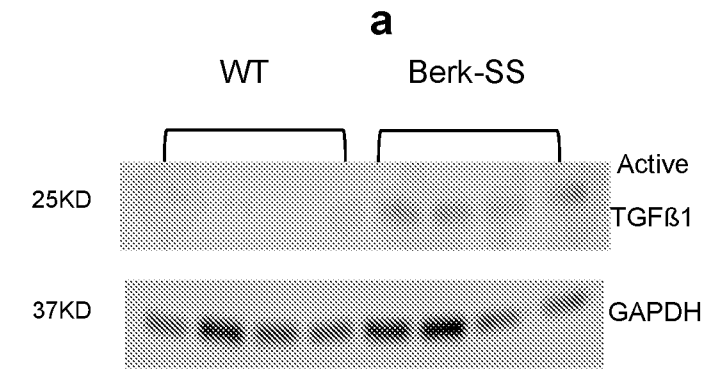
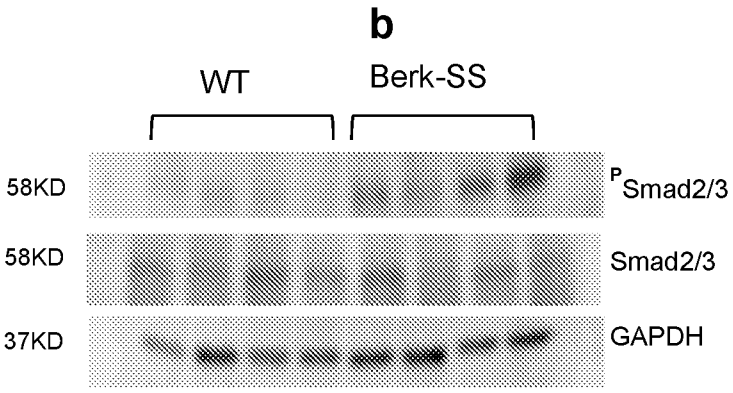


FIGURE 1

5


a
WT
Berk-SS
25KD
Active
TGFß1
37KD
GAPDH


b
WT
Berk-SS
58KD
PSmad2/3
58KD
Smad2/3
37KD
GAPDH

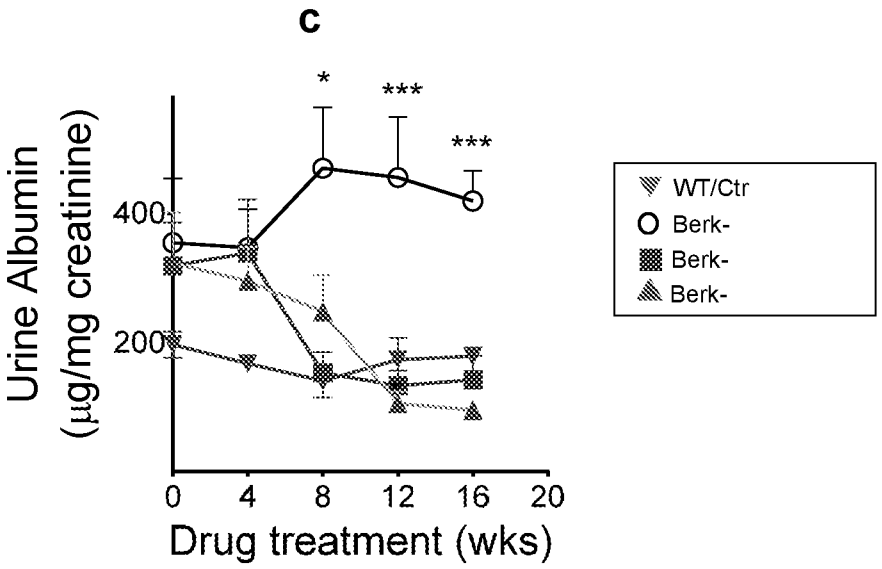
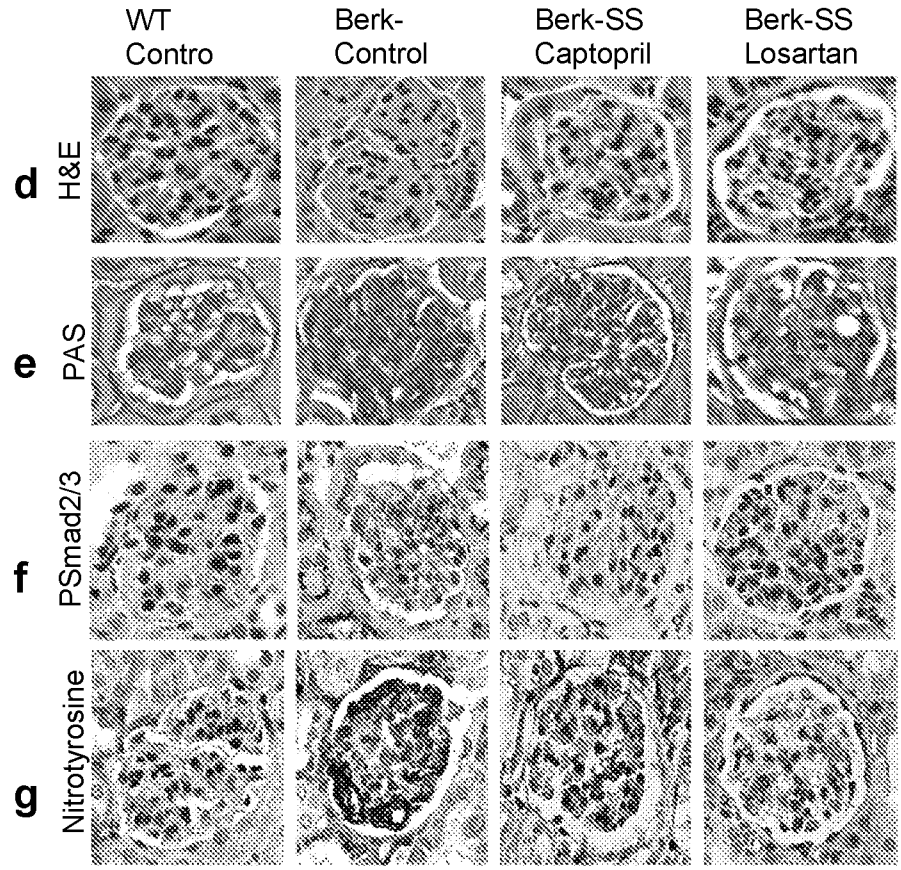

c
Urine Albumin (µg/mg creatinine)
400
200
0
4
8
12
16
20
Drug treatment (wks)
*
***
***
WT/Ctr
Berk-
Berk-
Berk-

FIGURE 2 (a – c)


WT Contro
Berk-Control
Berk-SS Captopril
Berk-SS Losartan
d H&E
e PAS
f PSmad2/3
g Nitrotyrosine

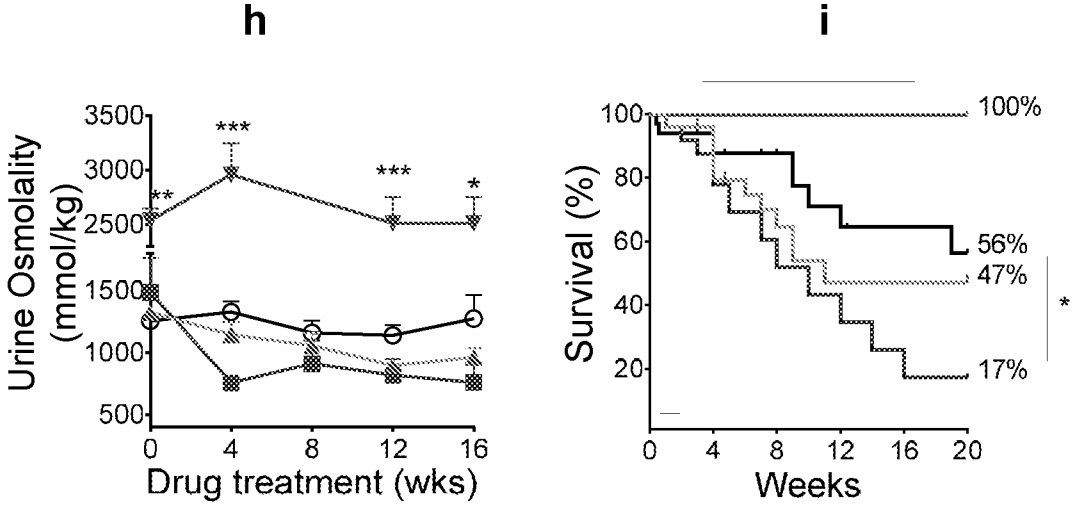

h
i
Urine Osmolality (mmol/kg)
3500
3000
2500
1500
1000
500
**
***
***
*
0
4
8
12
16
Drug treatment (wks)
Survival (%)
100
80
60
40
20
100%
56%
47%
17%
*
0
4
8
12
16
20
Weeks

FIGURE 2 (d – i)

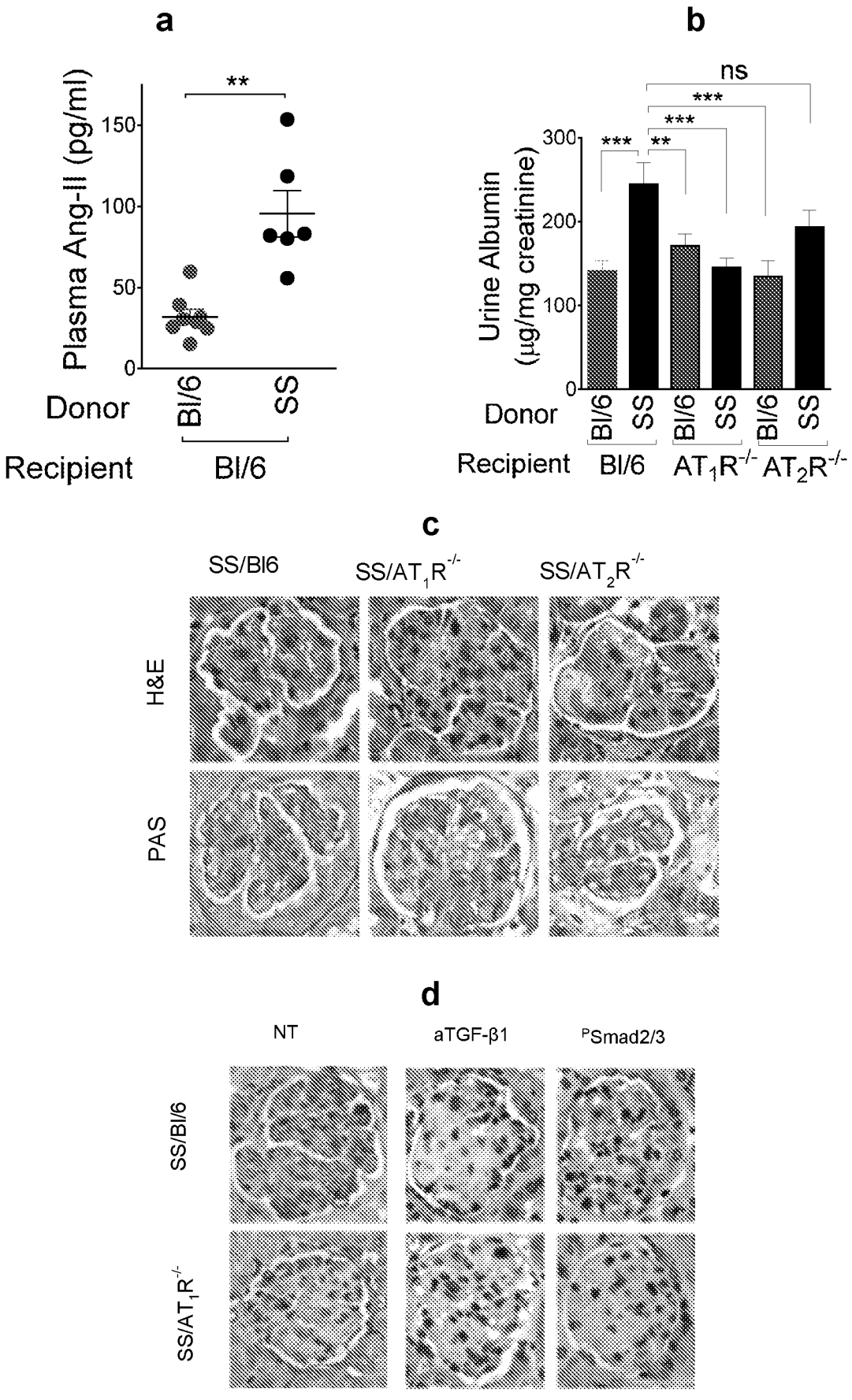
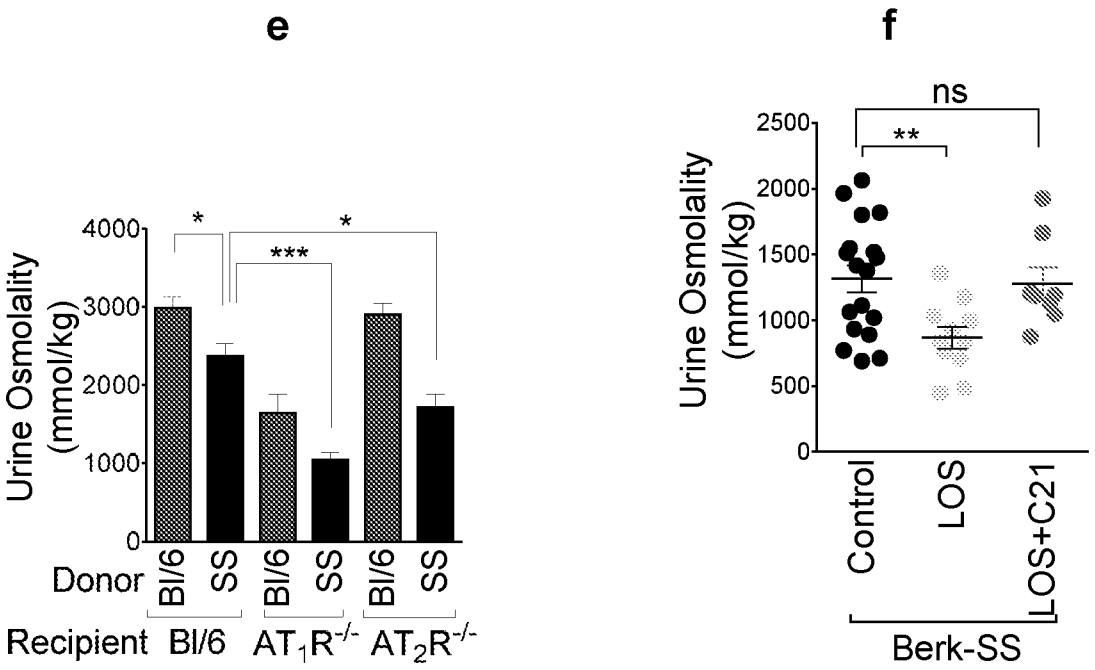

a
Plasma Ang-II (pg/ml)
150
100
50
0
**
Donor
Bl/6
SS
Recipient
Bl/6
b
Urine Albumin (µg/mg creatinine)
300
200
100
0
ns
***
***
***
**
Donor
Bl/6
SS
Bl/6
SS
Bl/6
SS
Recipient
Bl/6
AT1R-/-
AT2R-/-
c
SS/Bl6
SS/AT1R-/-
SS/AT2R-/-
H&E
PAS
d
NT
aTGF-β1
PSmad2/3
SS/Bl/6
SS/AT1R-/-

FIGURE 3 (a – d)


e
Urine Osmolality (mmol/kg)
4000
3000
2000
1000
0
*
***
*
Donor
Bl/6
SS
Bl/6
SS
Bl/6
SS
Recipient Bl/6
AT1R-/-
AT2R-/-
f
ns
**
2500
2000
1500
1000
500
0
Urine Osmolality (mmol/kg)
Control
LOS
LOS+C21
Berk-SS

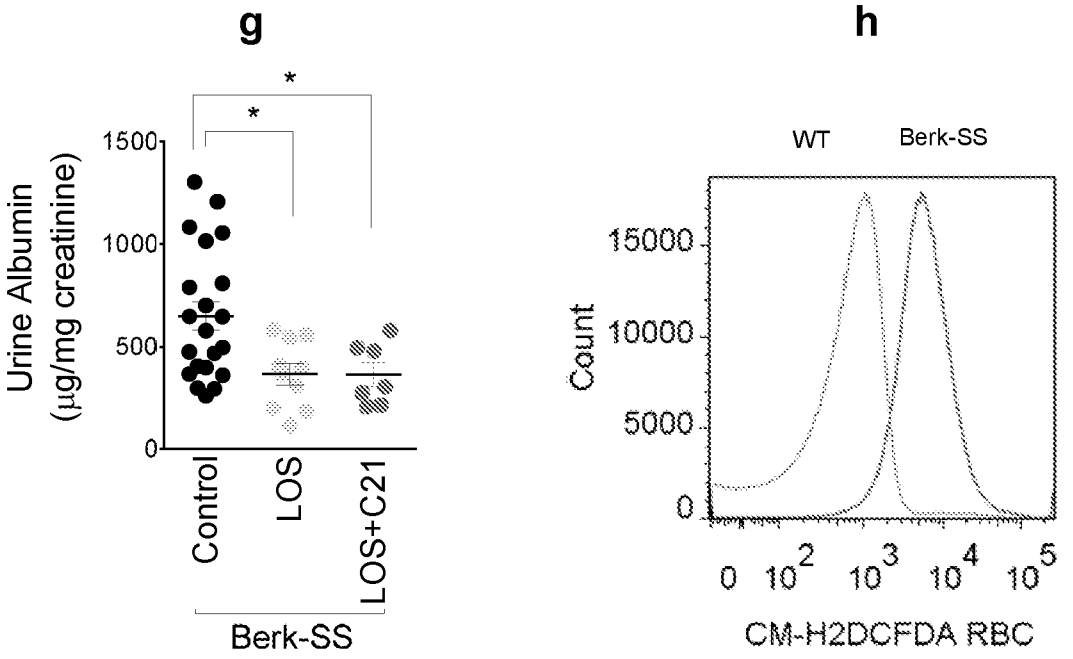

g
*
*
1500
1000
500
0
Urine Albumin (µg/mg creatinine)
Control
LOS
LOS+C21
Berk-SS
h
WT
Berk-SS
15000
10000
5000
0
Count
0
10^2
10^3
10^4
10^5
CM-H2DCFDA RBC

FIGURE 3 (e – h)

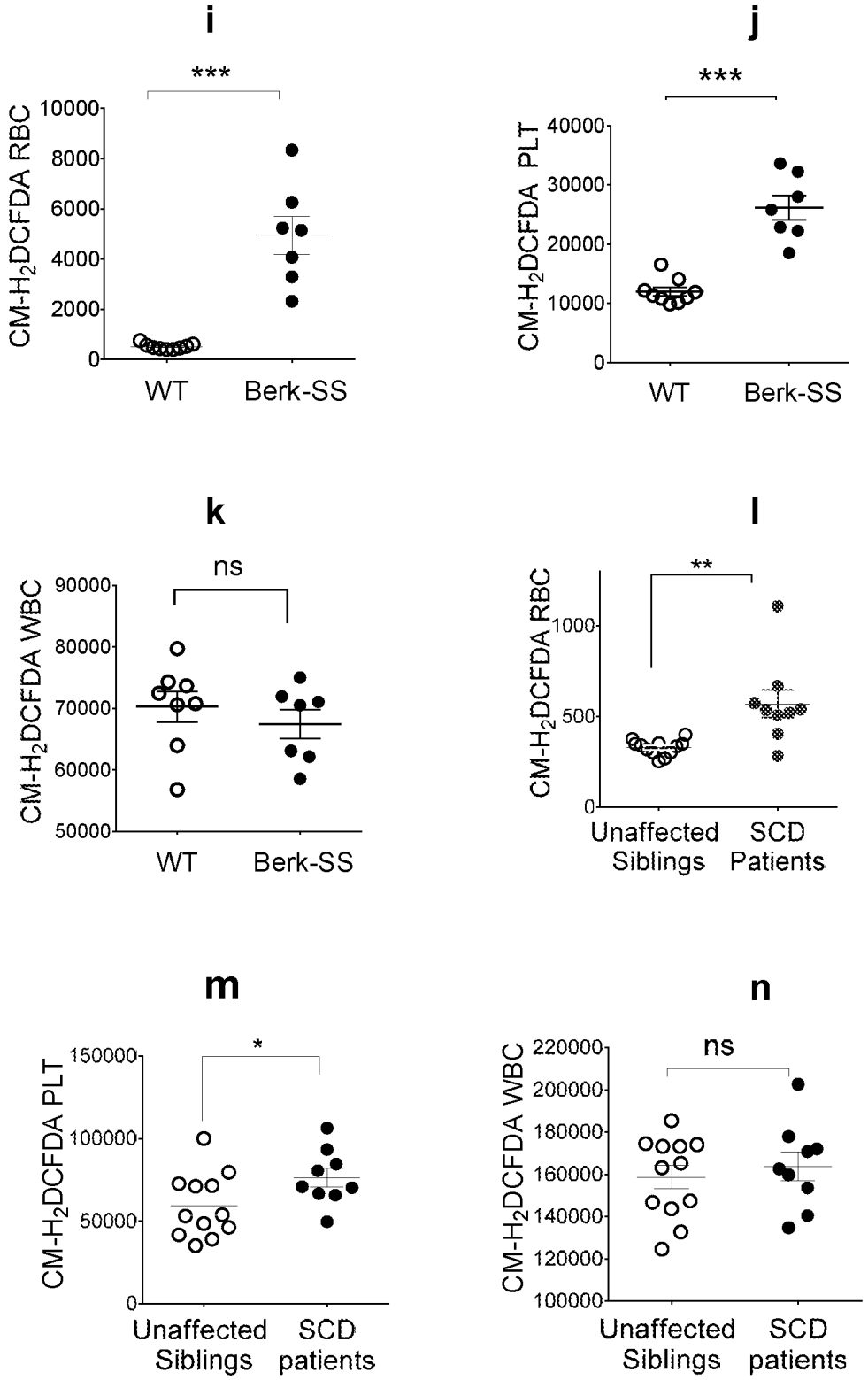

i
***
CM-H2DCFDA RBC
10000
8000
6000
4000
2000
0
WT
Berk-SS
j
***
CM-H2DCFDA PLT
40000
30000
20000
10000
0
WT
Berk-SS
k
ns
CM-H2DCFDA WBC
90000
80000
70000
60000
50000
WT
Berk-SS
l
**
CM-H2DCFDA RBC
1000
500
0
Unaffected Siblings
SCD Patients
m
*
CM-H2DCFDA PLT
150000
100000
50000
0
Unaffected Siblings
SCD patients
n
ns
CM-H2DCFDA WBC
220000
200000
180000
160000
140000
120000
100000
Unaffected Siblings
SCD patients

FIGURE 3 (i – n)

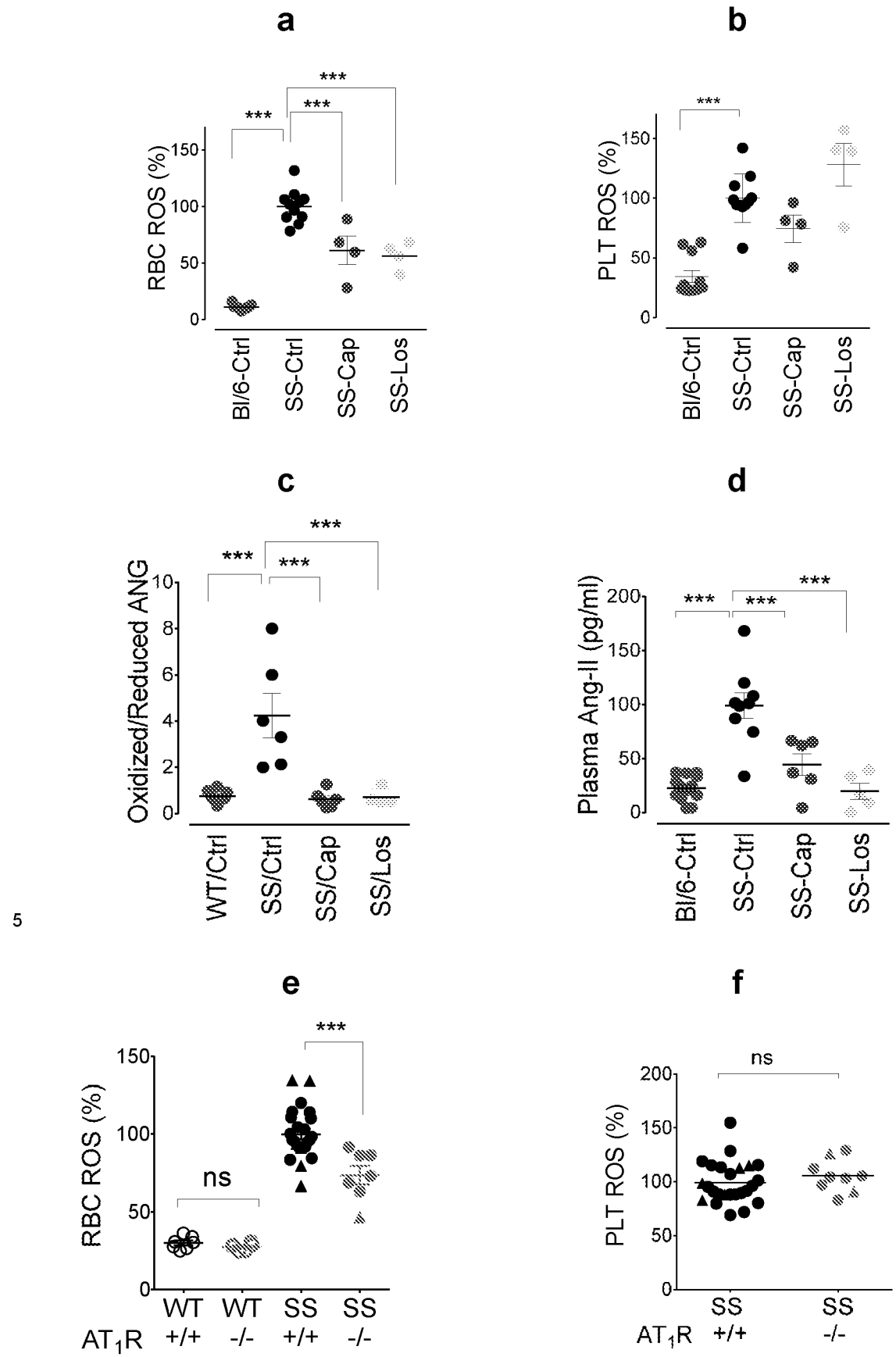

a
RBC ROS (%)
150
100
50
0
***
***
***
Bl/6-Ctrl
SS-Ctrl
SS-Cap
SS-Los
b
PLT ROS (%)
150
100
50
0
***
Bl/6-Ctrl
SS-Ctrl
SS-Cap
SS-Los
c
Oxidized/Reduced ANG
10
8
6
4
2
0
***
***
***
WT/Ctrl
SS/Ctrl
SS/Cap
SS/Los
d
Plasma Ang-II (pg/ml)
200
150
100
50
0
***
***
***
Bl/6-Ctrl
SS-Ctrl
SS-Cap
SS-Los
e
RBC ROS (%)
150
100
50
0
ns
***
WT +/+
WT -/-
SS +/+
SS -/-
AT1R
f
PLT ROS (%)
200
150
100
50
0
ns
SS +/+
SS -/-
AT1R

FIGURE 4 (a – f)

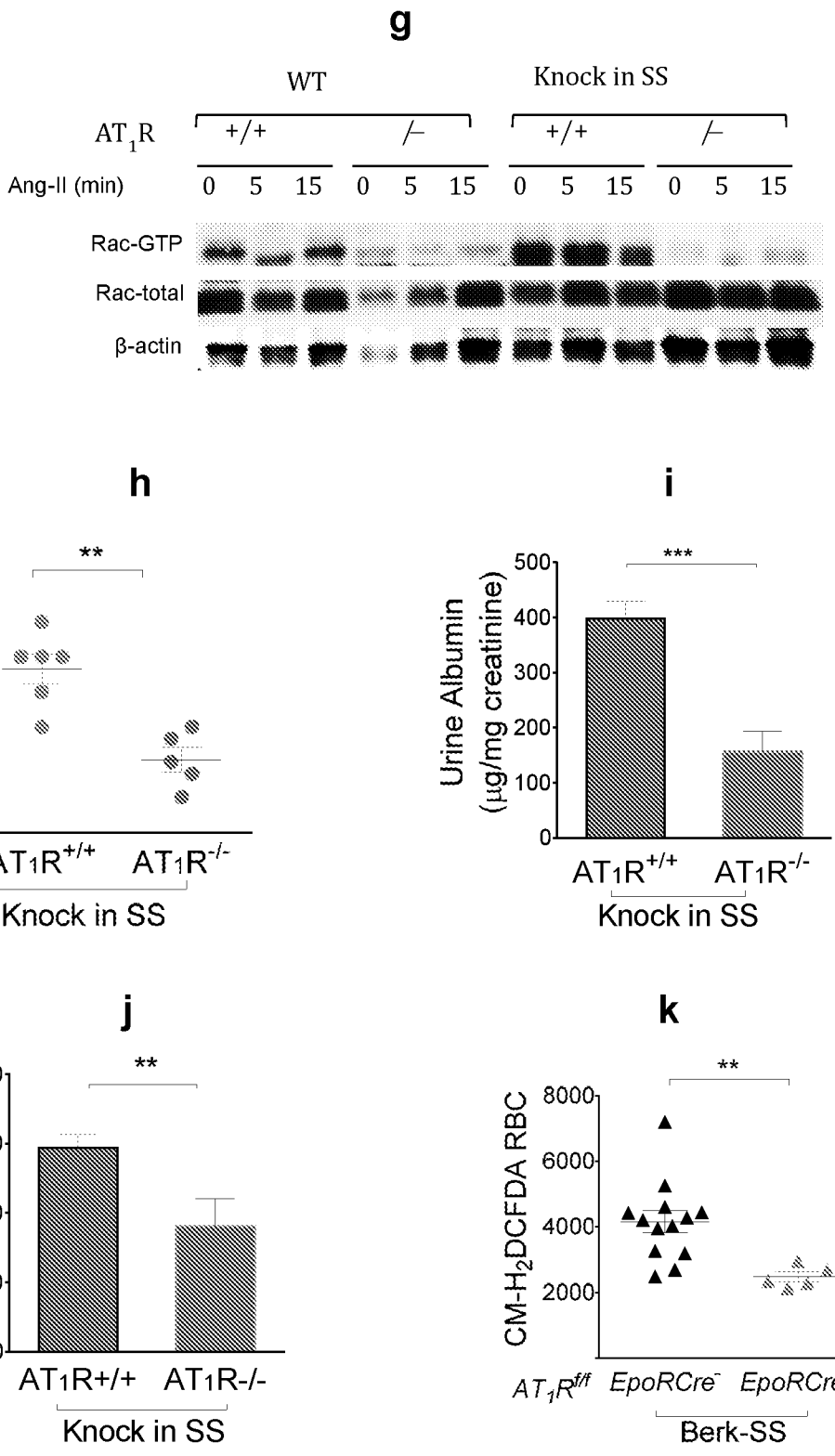
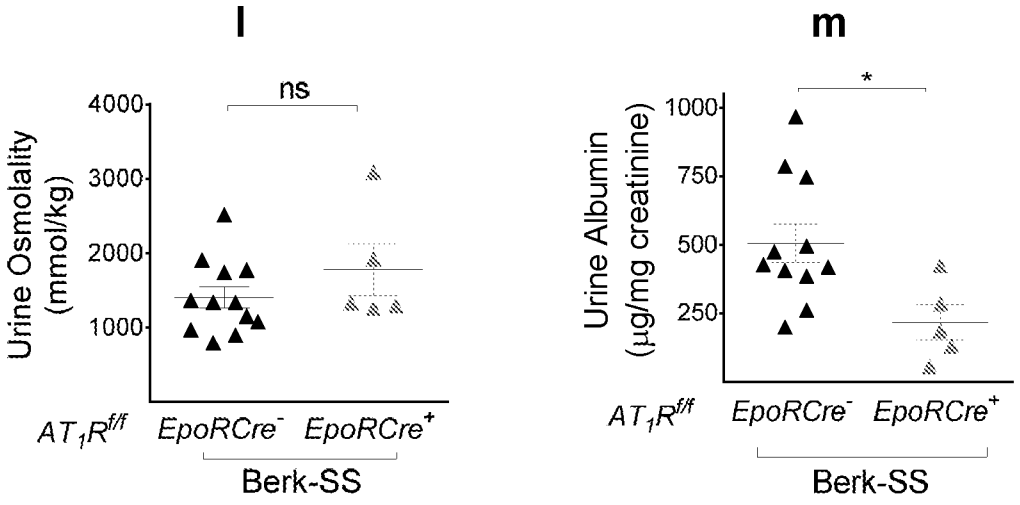

g
WT
Knock in SS
AT1R
+/+
/–
+/+
/–
Ang-II (min)
0 5 15 0 5 15 0 5 15 0 5 15
Rac-GTP
Rac-total
β-actin
h
Oxidized/Reduced ANG
**
AT1R+/+
AT1R-/-
Knock in SS
i
Urine Albumin (μg/mg creatinine)
***
AT1R+/+
AT1R-/-
Knock in SS
j
Urine Osmolality (mmol/kg)
**
AT1R+/+
AT1R-/-
Knock in SS
k
CM-H2DCFDA RBC
**
AT1Rf/f
EpoRCre-
EpoRCre+
Berk-SS

FIGURE 4 (g – k)


l
ns
Urine Osmolality (mmol/kg)
4000
3000
2000
1000
AT1Rf/f
EpoRCre-
EpoRCre+
Berk-SS
m
*
Urine Albumin (µg/mg creatinine)
1000
750
500
250
AT1Rf/f
EpoRCre-
EpoRCre+
Berk-SS

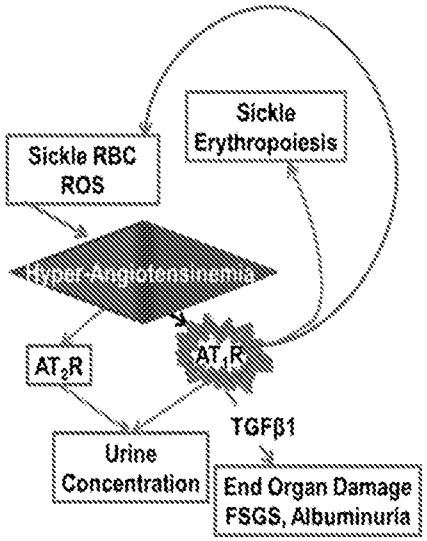

n
Sickle Erythropoiesis
Sickle RBC ROS
Hypoxia
AT2R
AT1R
TGFβ1
Urine Concentration
End Organ Damage FSGS, Albuminuria

FIGURE 4 (l – n)

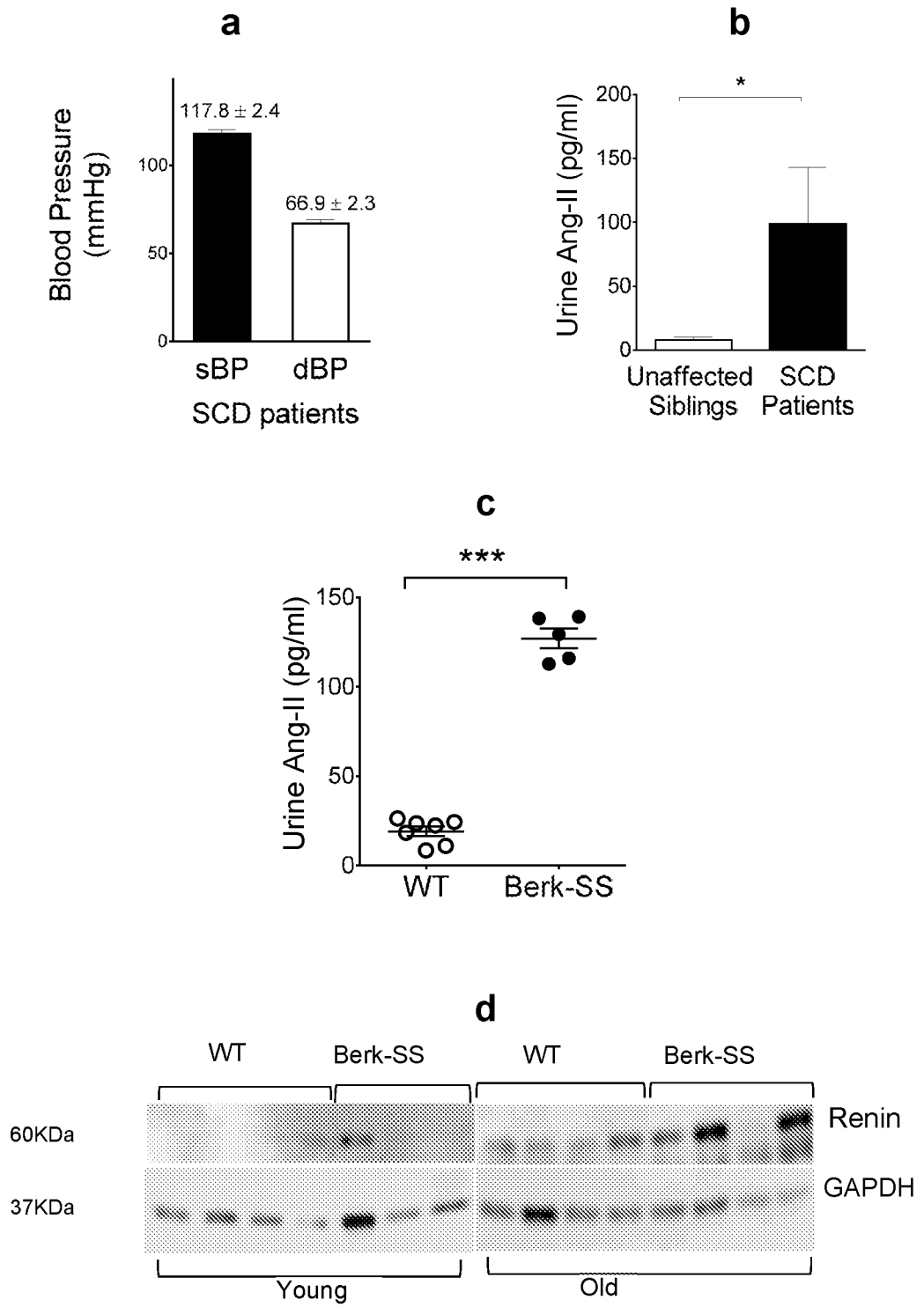
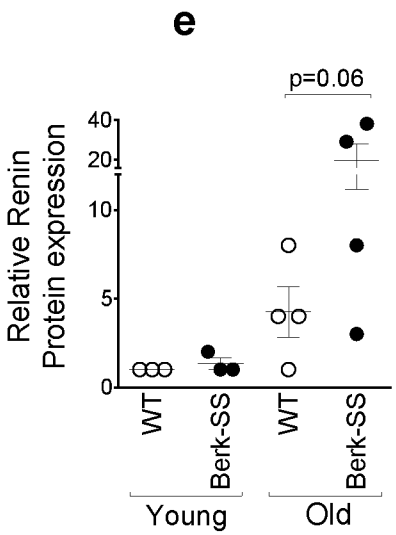
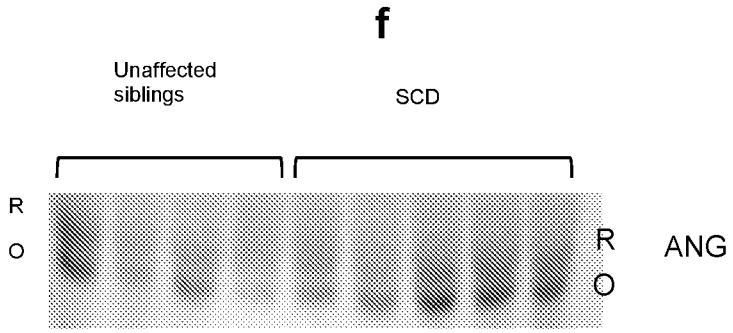

a
Blood Pressure (mmHg)
117.8 ± 2.4
66.9 ± 2.3
100
50
0
sBP
dBP
SCD patients
b
*
Urine Ang-II (pg/ml)
200
150
100
50
0
Unaffected Siblings
SCD Patients
c
***
Urine Ang-II (pg/ml)
150
100
50
0
WT
Berk-SS
d
WT
Berk-SS
WT
Berk-SS
60KDa
37KDa
Renin
GAPDH
Young
Old

FIGURE 5 (a - d)


e
p=0.06
Relative Renin
Protein expression
40
20
10
5
0
WT
Berk-SS
WT
Berk-SS
Young
Old


f
Unaffected
siblings
SCD
R
O
R
O
ANG

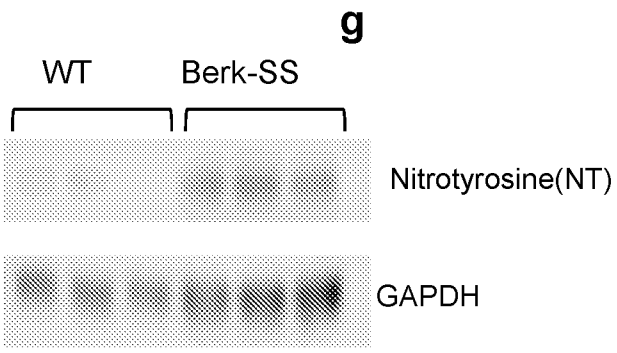

g
WT
Berk-SS
Nitrotyrosine(NT)
GAPDH

FIGURE 5 (e – g)

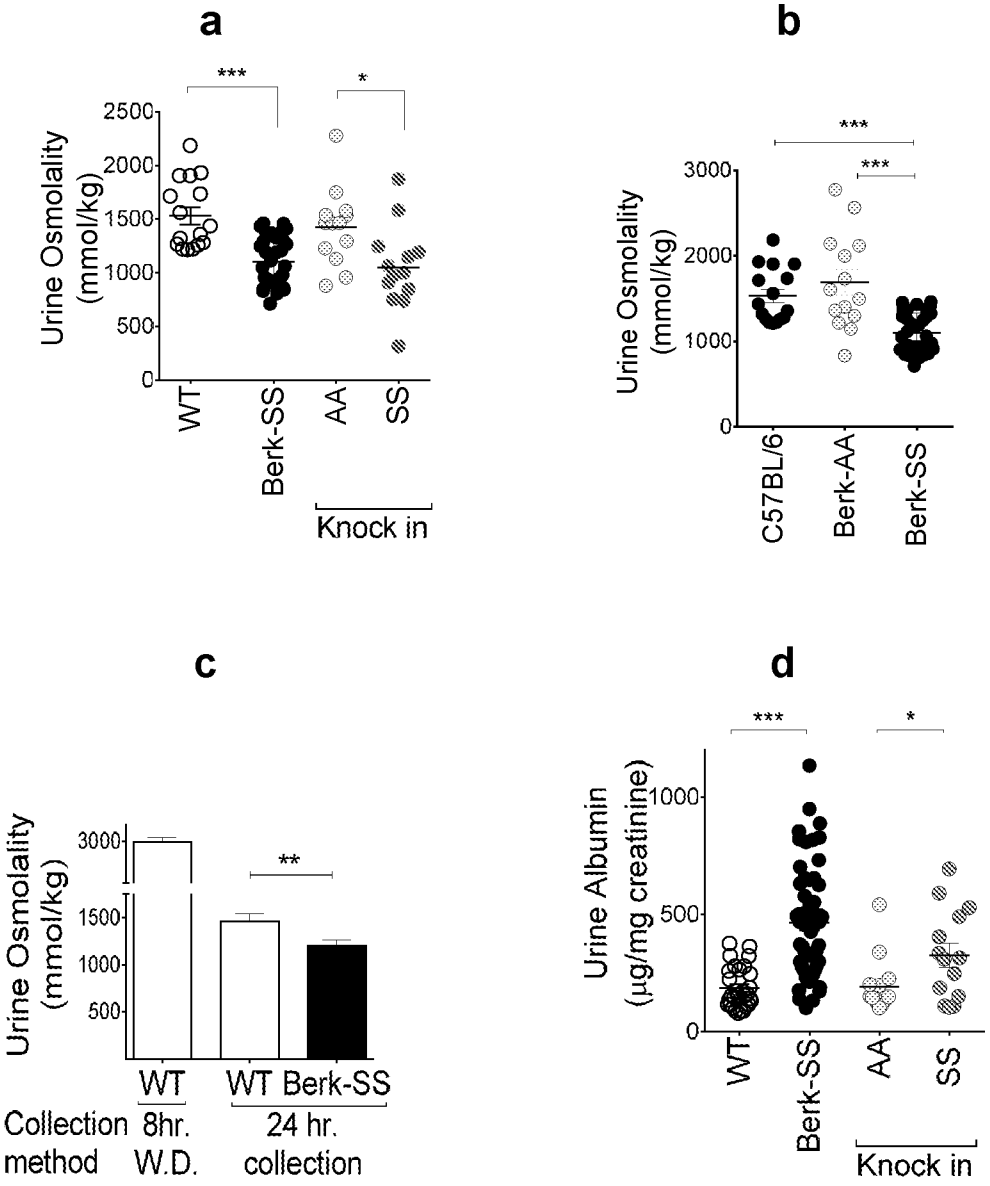

a
***
*
Urine Osmolality (mmol/kg)
2500
2000
1500
1000
500
0
WT
Berk-SS
AA
SS
Knock in
b
***
***
Urine Osmolality (mmol/kg)
3000
2000
1000
0
C57BL/6
Berk-AA
Berk-SS
c
Urine Osmolality (mmol/kg)
3000
1500
1000
500
**
WT
WT Berk-SS
Collection method
8hr. W.D.
24 hr. collection
d
***
*
Urine Albumin (µg/mg creatinine)
1000
500
0
WT
Berk-SS
AA
SS
Knock in

FIGURE 6 (a - d)

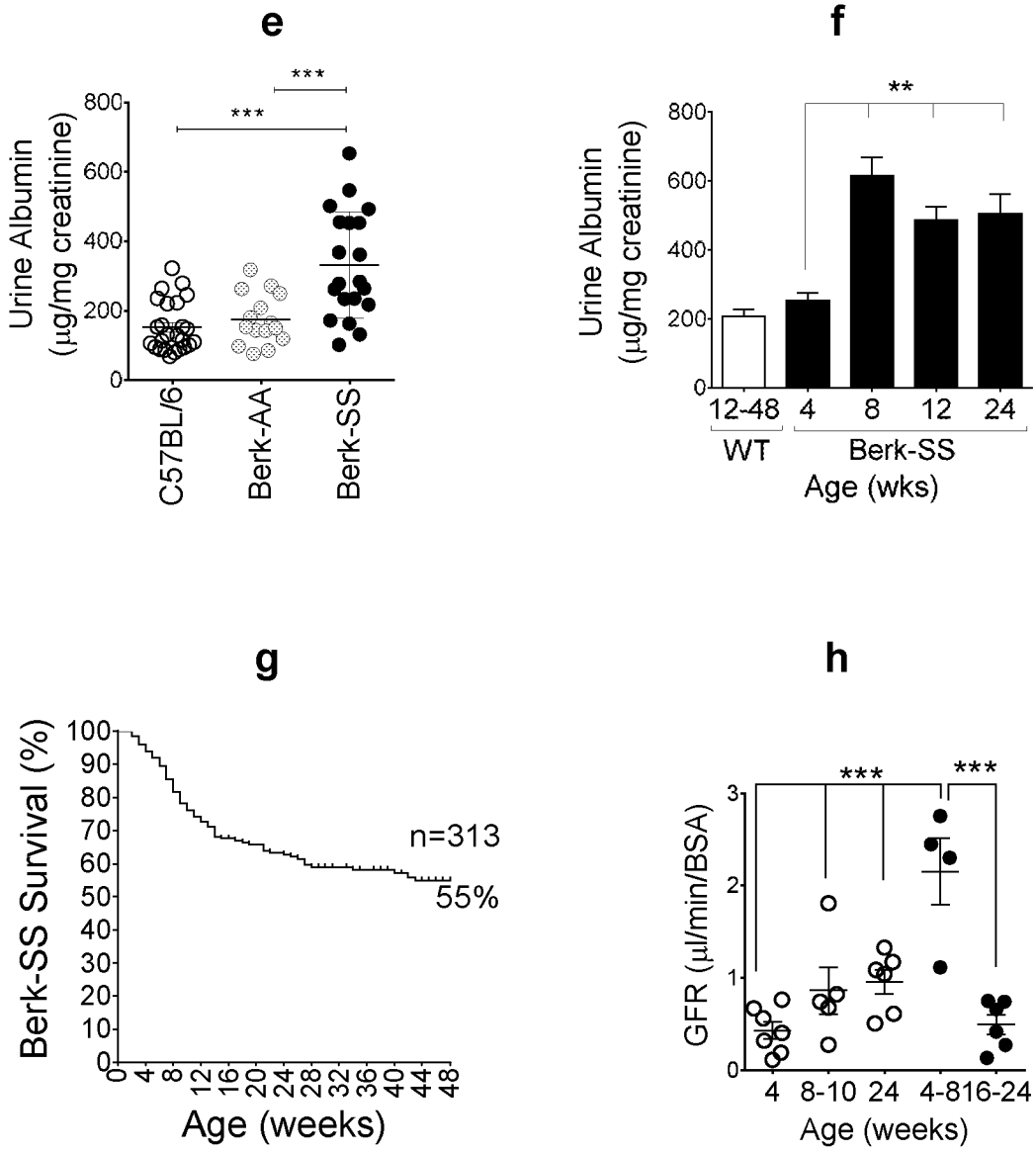

e
Urine Albumin (µg/mg creatinine)
800
600
400
200
0
***
***
C57BL/6
Berk-AA
Berk-SS
f
Urine Albumin (µg/mg creatinine)
800
600
400
200
0
**
12-48
4
8
12
24
WT
Berk-SS
Age (wks)
g
Berk-SS Survival (%)
100
90
80
70
60
50
40
30
20
10
0
n=313
55%
0 4 8 12 16 20 24 28 32 36 40 44 48
Age (weeks)
h
GFR (µl/min/BSA)
3
2
1
0
***
***
4
8-10
24
4-816-24
Age (weeks)

FIGURE 6 (e - h)

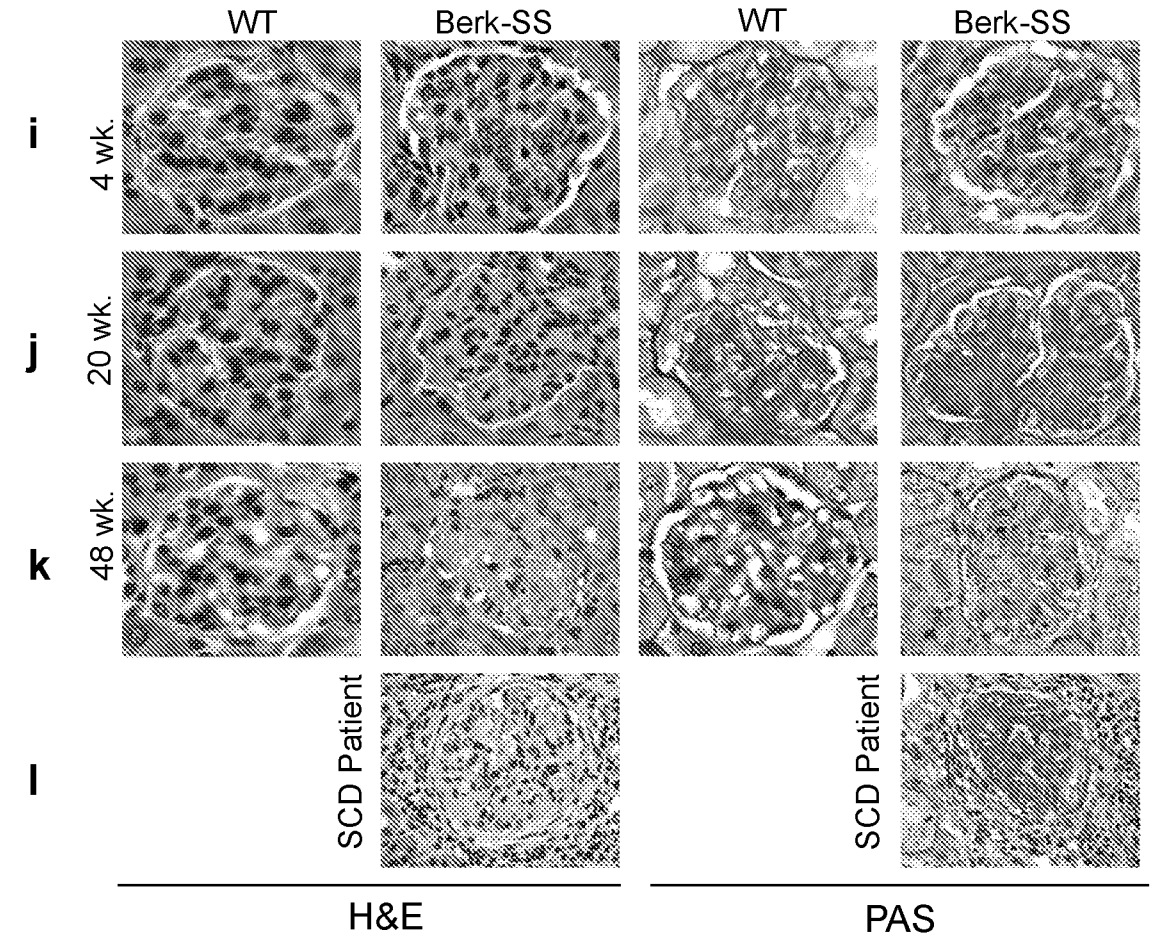

WT
Berk-SS
WT
Berk-SS
i
4 wk.
j
20 wk.
k
48 wk.
l
SCD Patient
SCD Patient
H&E
PAS

FIGURE 6 (i - l)

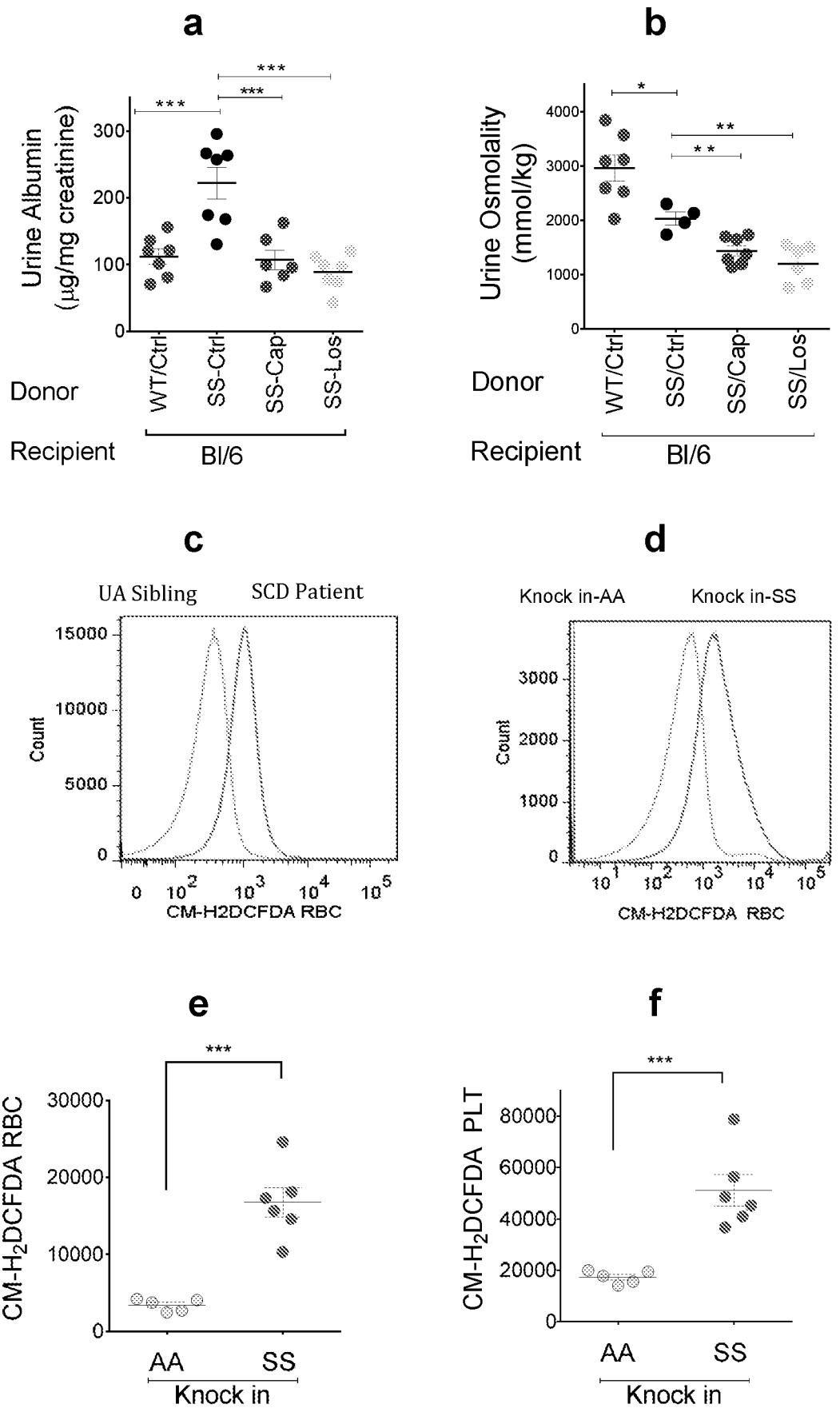

a
Urine Albumin (μg/mg creatinine)
300
200
100
0
***
***
***
WT/Ctrl
SS-Ctrl
SS-Cap
SS-Los
Donor
Recipient
Bl/6
b
Urine Osmolality (mmol/kg)
4000
3000
2000
1000
0
*
**
**
WT/Ctrl
SS/Ctrl
SS/Cap
SS/Los
Donor
Recipient
Bl/6
c
UA Sibling
SCD Patient
Count
15000
10000
5000
0
0 10^2 10^3 10^4 10^5
CM-H2DCFDA RBC
d
Knock in-AA
Knock in-SS
Count
3000
2000
1000
0
10^1 10^2 10^3 10^4 10^5
CM-H2DCFDA RBC
e
***
CM-H2DCFDA RBC
30000
20000
10000
0
AA
SS
Knock in
f
***
CM-H2DCFDA PLT
80000
60000
40000
20000
0
AA
SS
Knock in

FIGURE 7 (a – f)

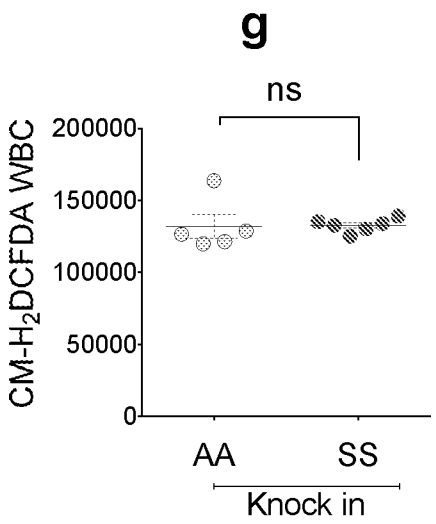

g
ns
200000
150000
100000
50000
0
CM-H2DCFDA WBC
AA
SS
Knock in

FIGURE 7 (g)

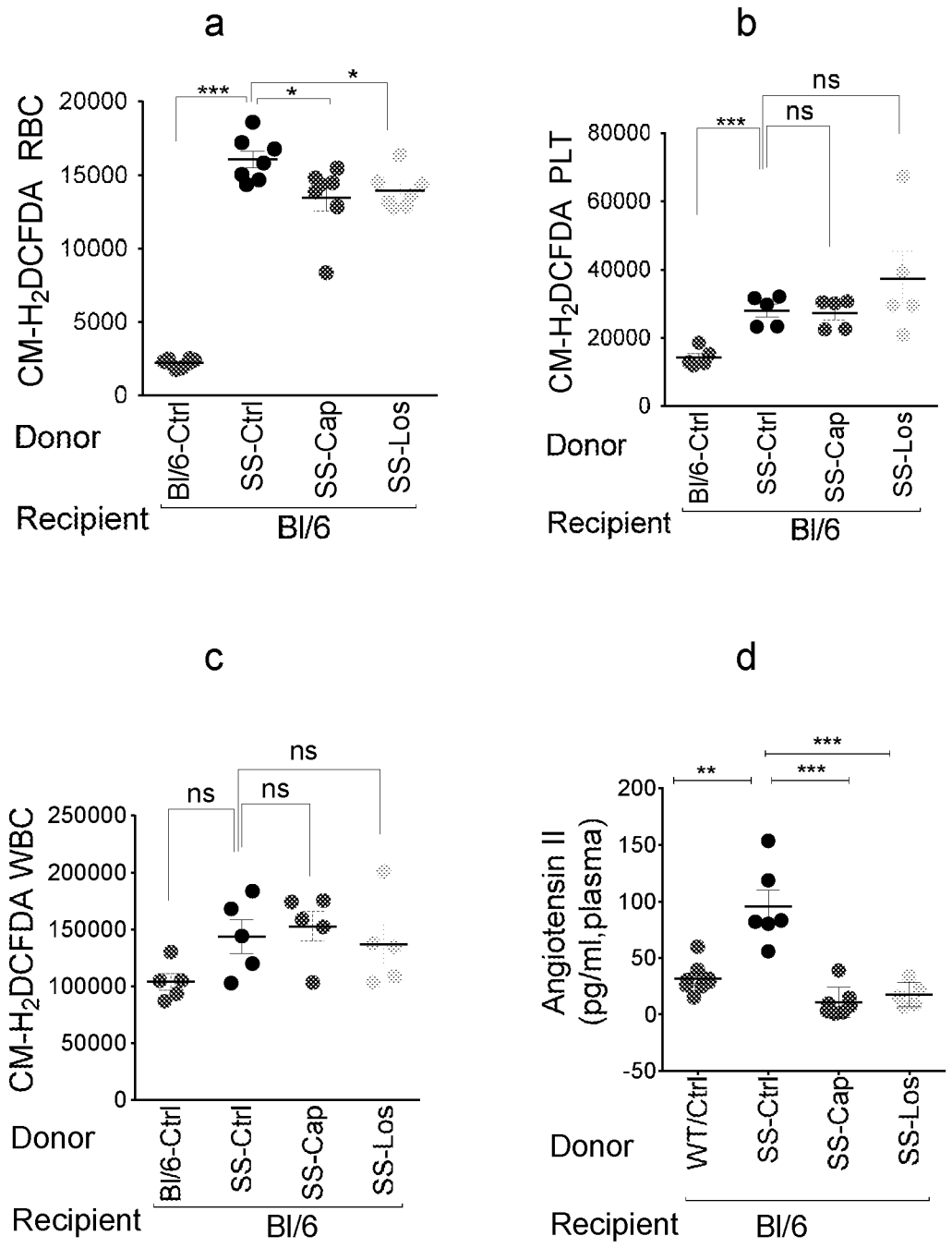

a
b
c
d
CM-H2DCFDA RBC
CM-H2DCFDA PLT
CM-H2DCFDA WBC
Angiotensin II (pg/ml,plasma)
Donor
Recipient
Bl/6-Ctrl
SS-Ctrl
SS-Cap
SS-Los
WT/Ctrl
Bl/6
ns
***
**
*

FIGURE 8 (a - d)

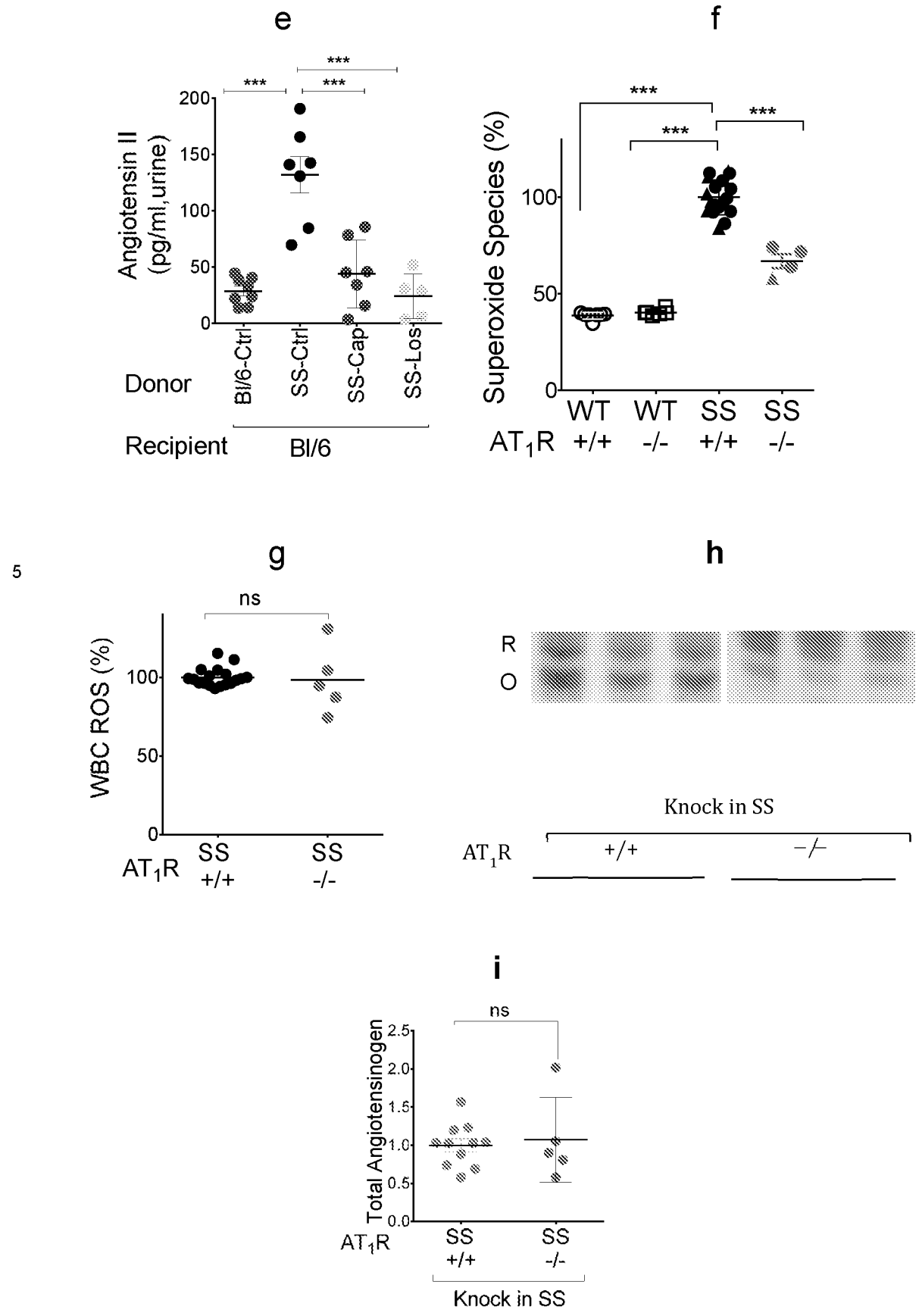

e
***
***
***
Angiotensin II (pg/ml,urine)
200
150
100
50
0
Bl/6-Ctrl
SS-Ctrl
SS-Cap
SS-Los
Donor
Recipient
Bl/6
f
***
***
***
Superoxide Species (%)
100
50
0
WT
WT
SS
SS
AT1R +/+ -/- +/+ -/-
g
ns
WBC ROS (%)
100
50
0
SS +/+
SS -/-
AT1R
h
R
O
Knock in SS
AT1R
+/+
−/−
i
ns
Total Angiotensinogen
2.5
2.0
1.5
1.0
0.5
0.0
AT1R
SS +/+
SS -/-
Knock in SS

FIGURE 8 (e - i)