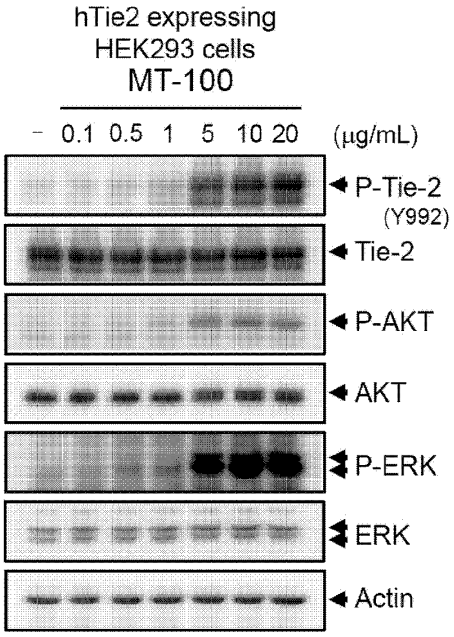
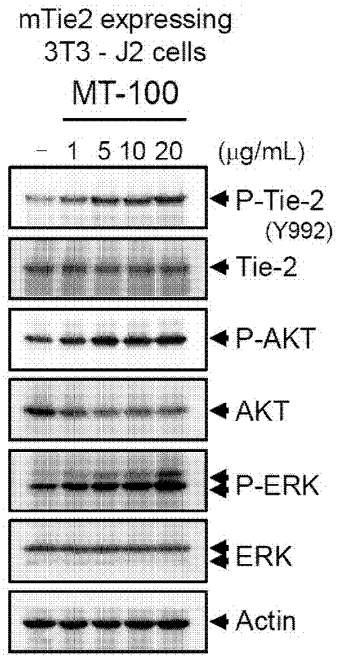
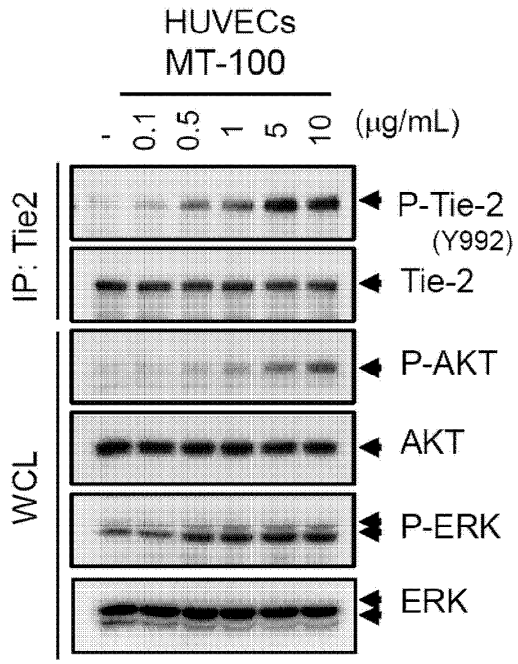
[도1a]


hTie2 expressing
HEK293 cells
MT-100
- 0.1 0.5 1 5 10 20 (μg/mL)
P-Tie-2
(Y992)
Tie-2
P-AKT
AKT
P-ERK
ERK
Actin

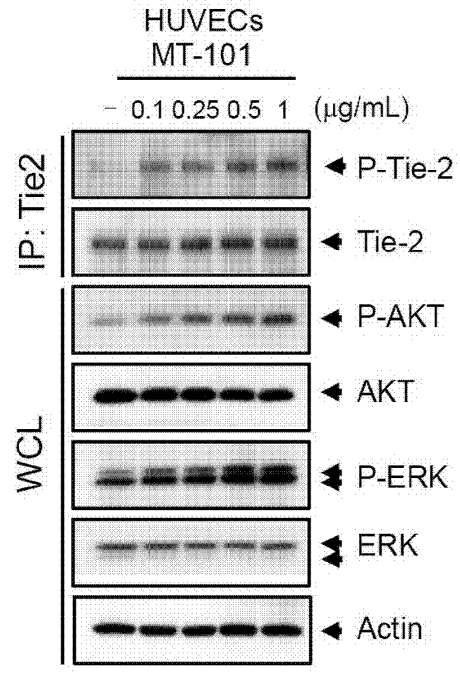
[도1b]


mTie2 expressing
3T3 - J2 cells
MT-100
- 1 5 10 20 (μg/mL)
P-Tie-2
(Y992)
Tie-2
P-AKT
AKT
P-ERK
ERK
Actin

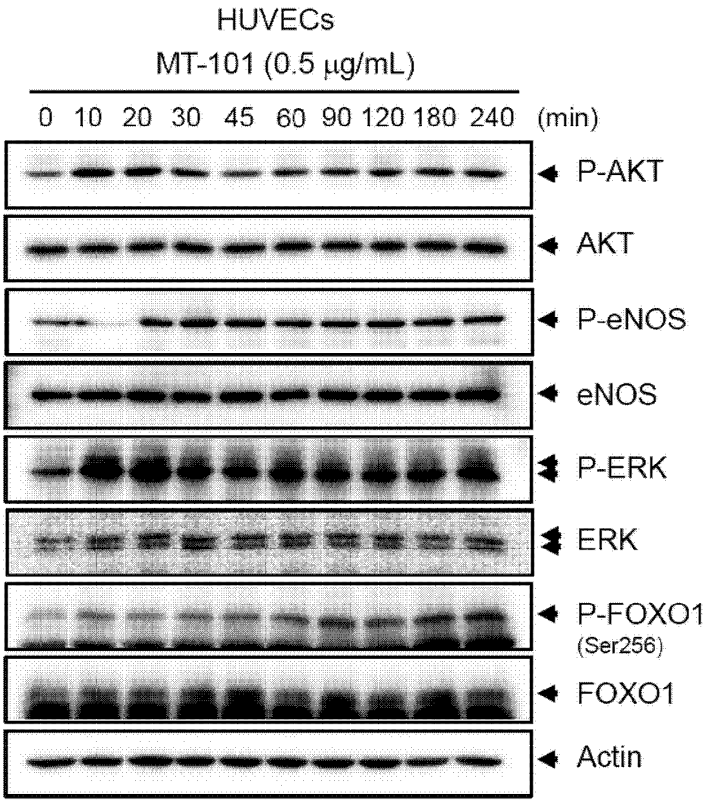
[도1c]


HUVECs
MT-100
- 0.1 0.5 1 5 10 (μg/mL)
IP: Tie2
WCL
P-Tie-2
(Y992)
Tie-2
P-AKT
AKT
P-ERK
ERK

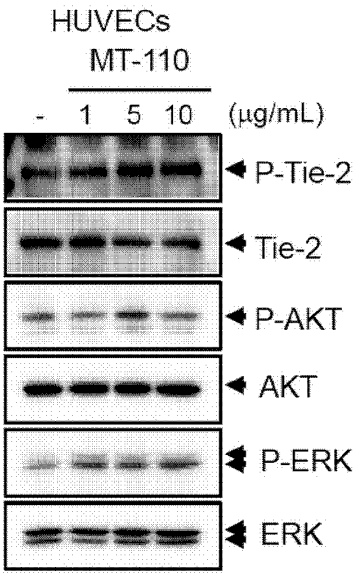
[도2a]


HUVECs
MT-101
- 0.1 0.25 0.5 1 (μg/mL)
IP: Tie2
WCL
P-Tie-2
Tie-2
P-AKT
AKT
P-ERK
ERK
Actin

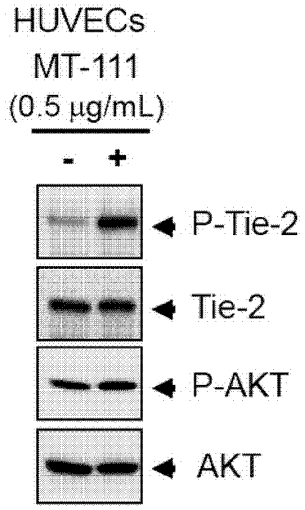
[도2b]


HUVECs
MT-101 (0.5 μg/mL)
0 10 20 30 45 60 90 120 180 240 (min)
P-AKT
AKT
P-eNOS
eNOS
P-ERK
ERK
P-FOXO1
(Ser256)
FOXO1
Actin

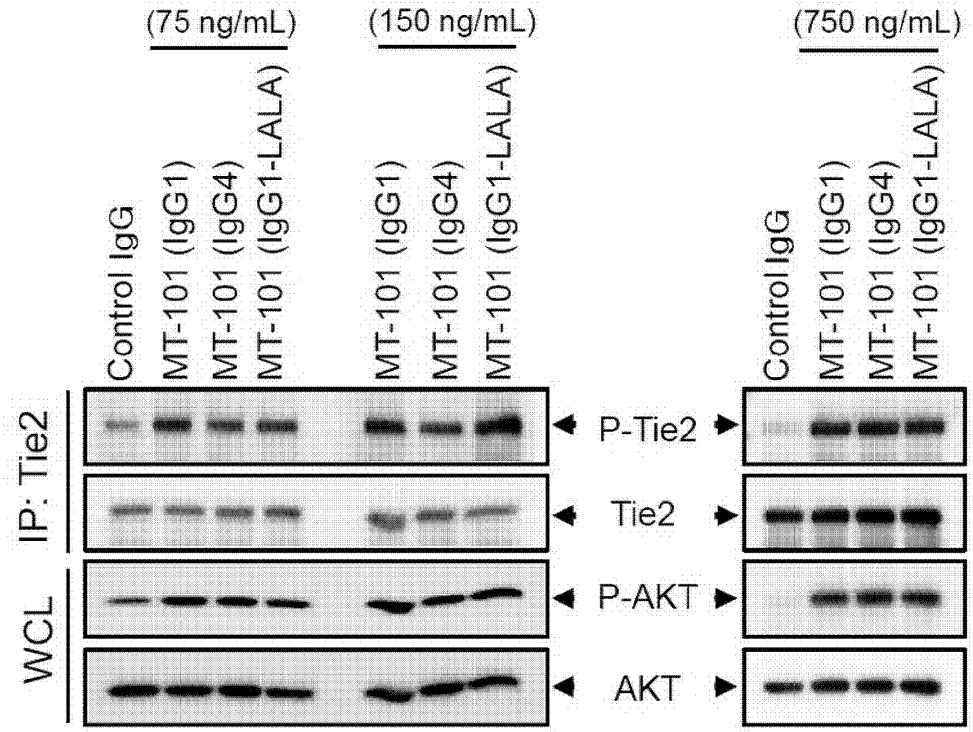
[도2c]


HUVECs
MT-110
- 1 5 10 (μg/mL)
P-Tie-2
Tie-2
P-AKT
AKT
P-ERK
ERK

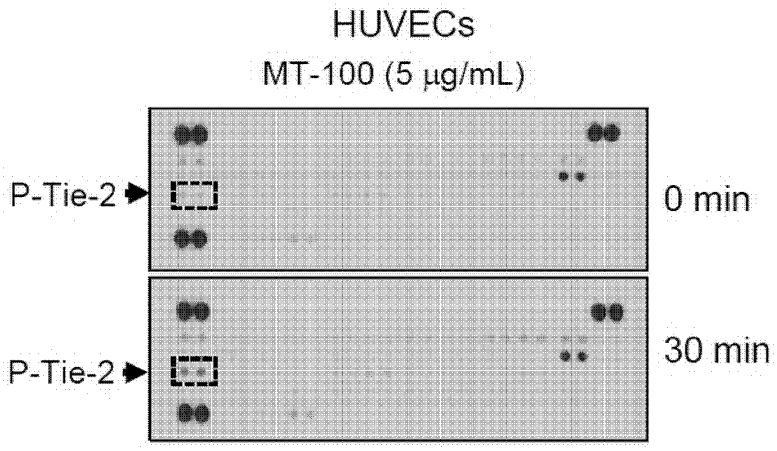
[도2d]


HUVECs
MT-111
(0.5 μg/mL)
-
+
P-Tie-2
Tie-2
P-AKT
AKT

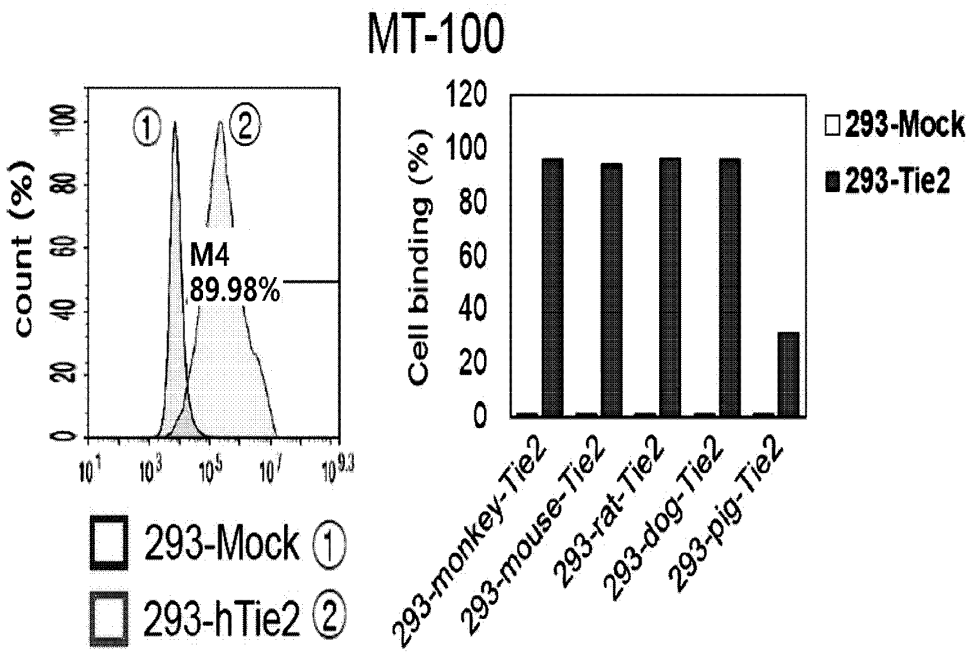
[도3]


(75 ng/mL)
(150 ng/mL)
(750 ng/mL)
Control IgG
MT-101 (IgG1)
MT-101 (IgG4)
MT-101 (IgG1-LALA)
IP: Tie2
WCL
P-Tie2
Tie2
P-AKT
AKT

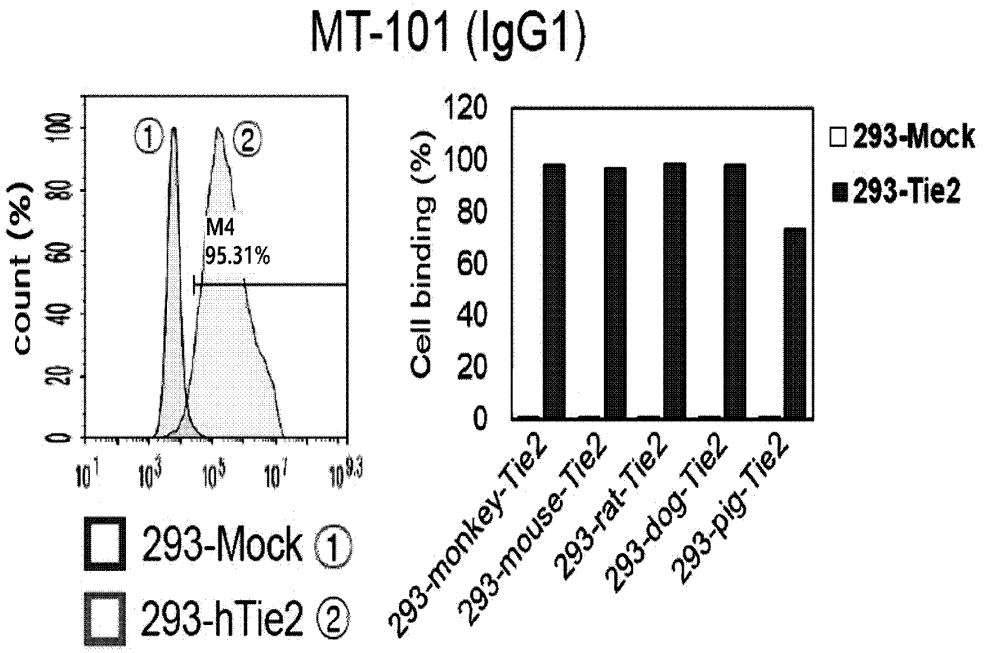
[도4]


HUVECs
MT-100 (5 μg/mL)
P-Tie-2
0 min
P-Tie-2
30 min

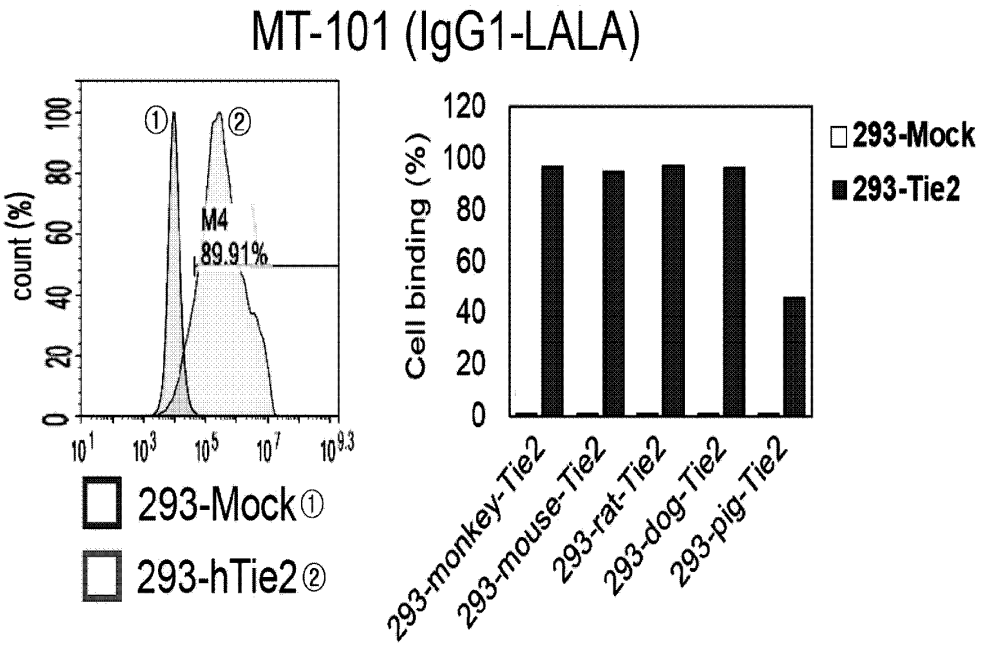
[도5a]


MT-100
count (%)
M4
89.98%
293-Mock ①
293-hTie2 ②
Cell binding (%)
293-Mock
293-Tie2
293-monkey-Tie2
293-mouse-Tie2
293-rat-Tie2
293-dog-Tie2
293-pig-Tie2

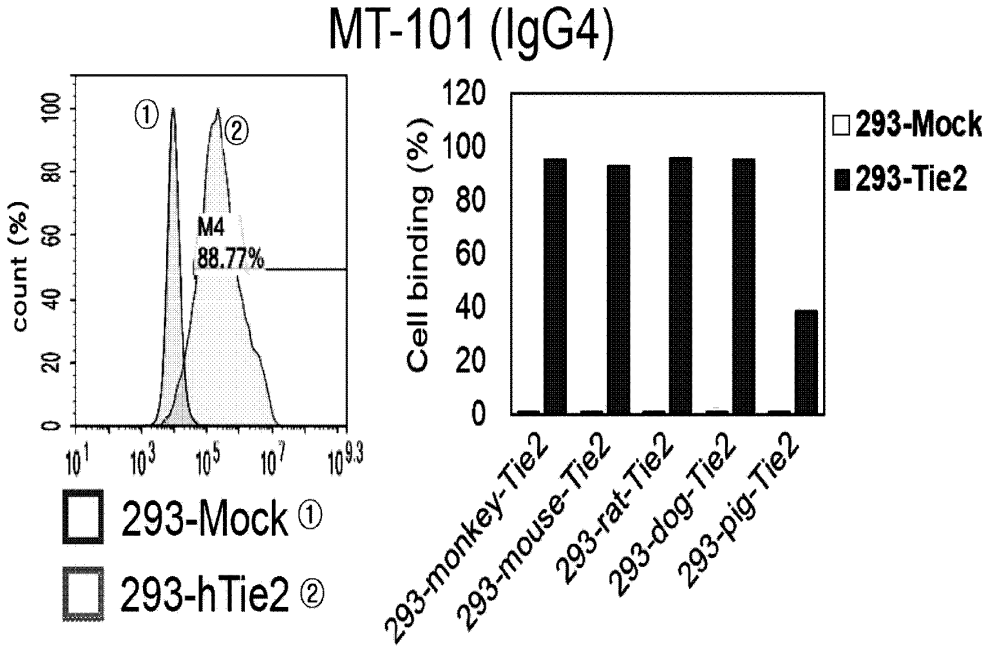
[도5b]


MT-101 (IgG1)
count (%)
M4
95.31%
293-Mock ①
293-hTie2 ②
Cell binding (%)
293-Mock
293-Tie2
293-monkey-Tie2
293-mouse-Tie2
293-rat-Tie2
293-dog-Tie2
293-pig-Tie2

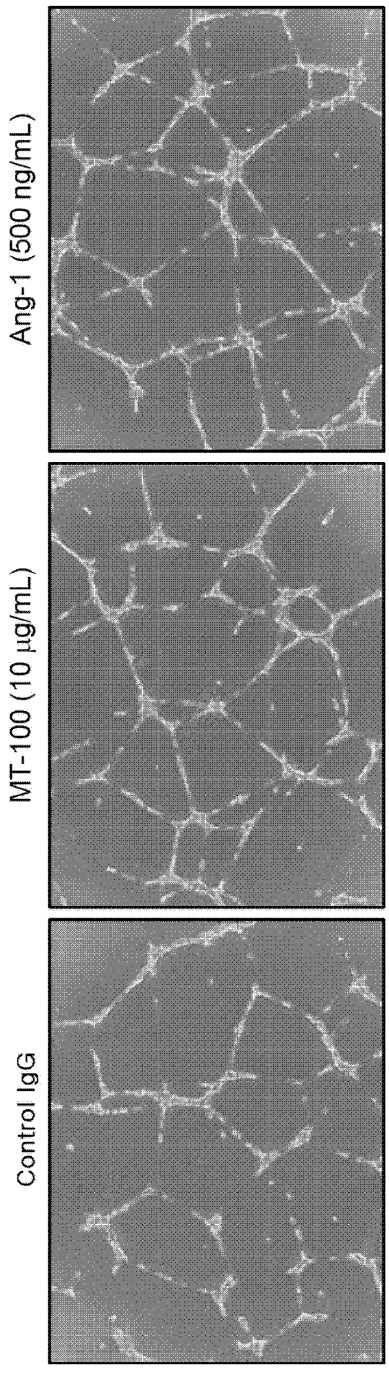
[도5c]


MT-101 (IgG1-LALA)
count (%)
M4
89.91%
293-Mock①
293-hTie2②
Cell binding (%)
293-Mock
293-Tie2
293-monkey-Tie2
293-mouse-Tie2
293-rat-Tie2
293-dog-Tie2
293-pig-Tie2

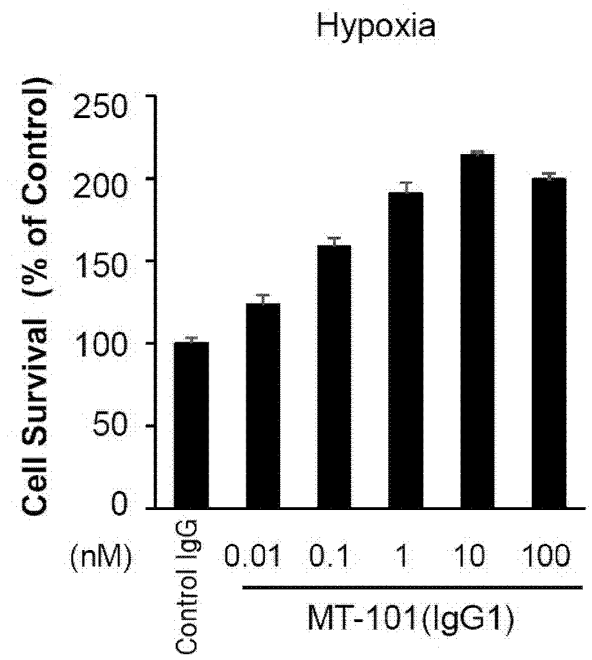
[도5d]


MT-101 (IgG4)
count (%)
M4
88.77%
293-Mock ①
293-hTie2 ②
Cell binding (%)
293-Mock
293-Tie2
293-monkey-Tie2
293-mouse-Tie2
293-rat-Tie2
293-dog-Tie2
293-pig-Tie2

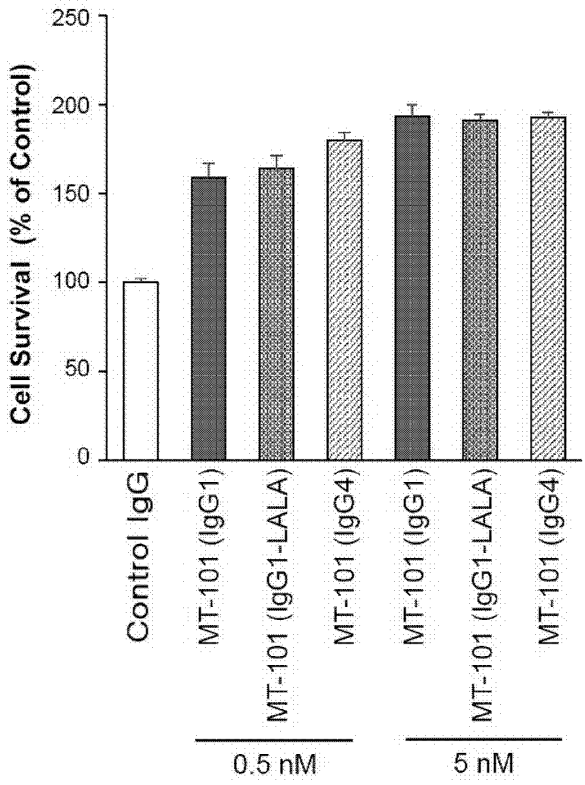
[도6]



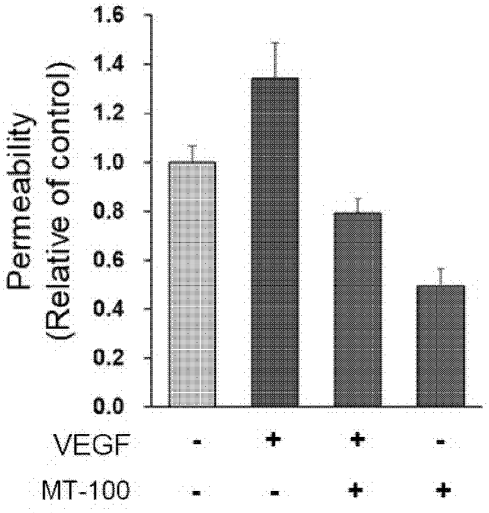
[도7a]


Hypoxia
Cell Survival (% of Control)
250
200
150
100
50
0
(nM)
Control IgG
0.01
0.1
1
10
100
MT-101(IgG1)

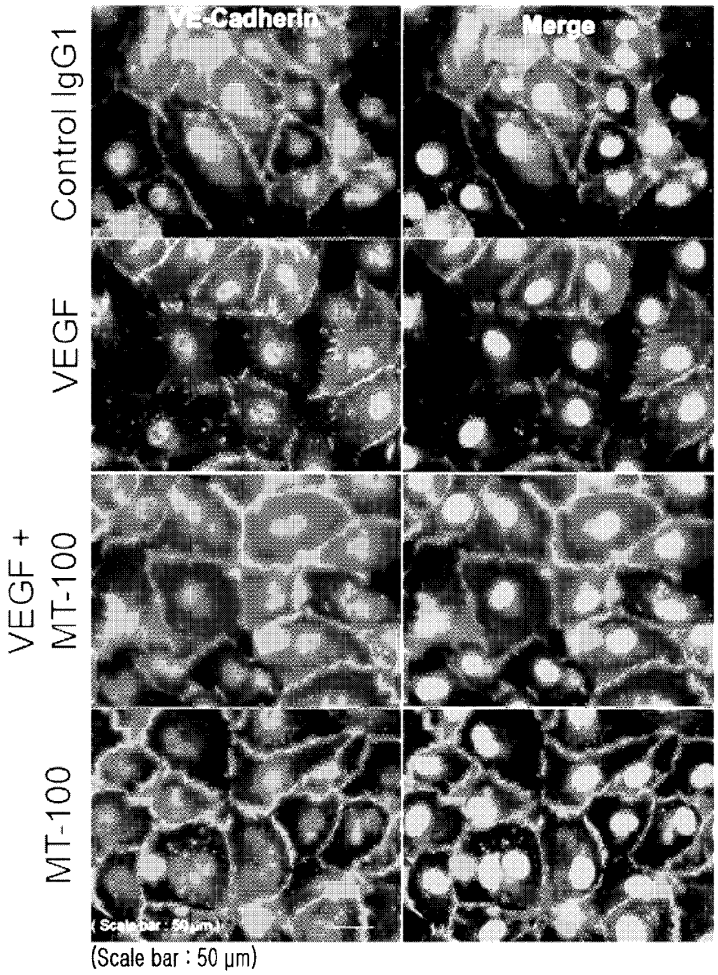
[도7b]


Cell Survival (% of Control)
250
200
150
100
50
0
Control IgG
MT-101 (IgG1)
MT-101 (IgG1-LALA)
MT-101 (IgG4)
MT-101 (IgG1)
MT-101 (IgG1-LALA)
MT-101 (IgG4)
0.5 nM
5 nM

[도8a]

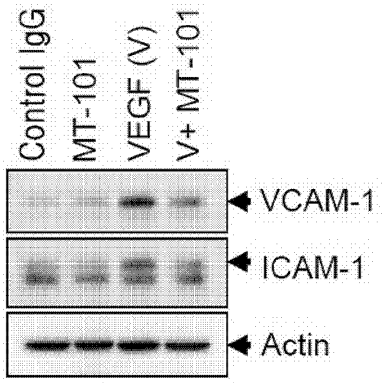


[도8b]

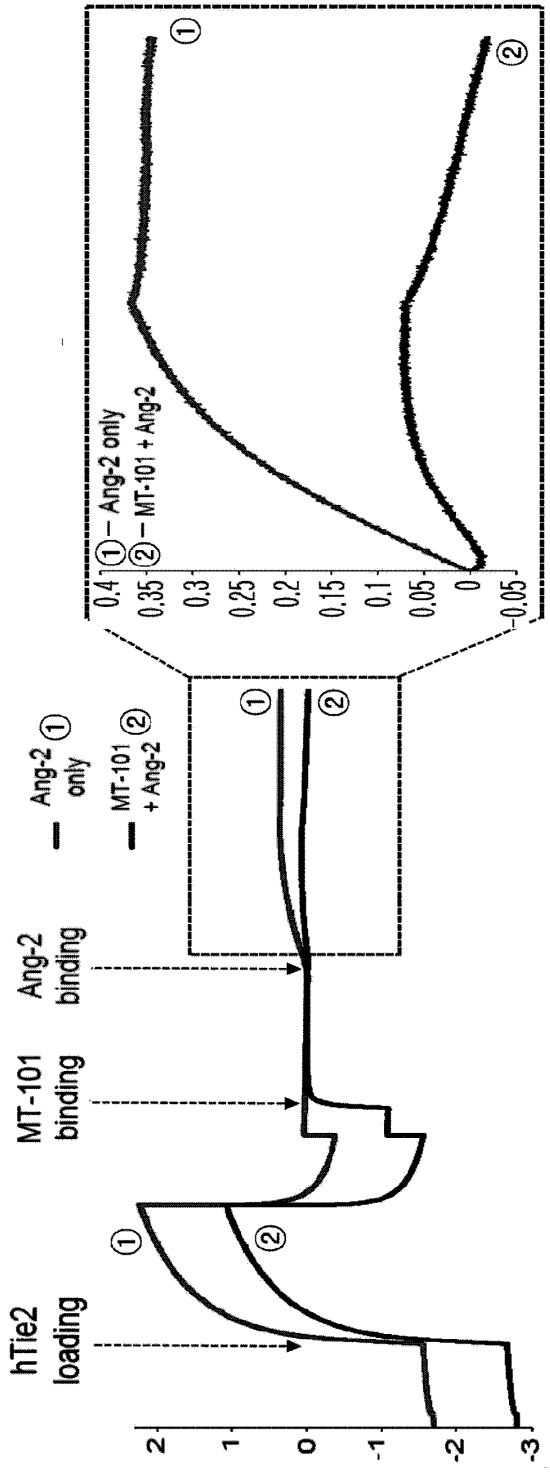


(Scale bar : 50 μm)

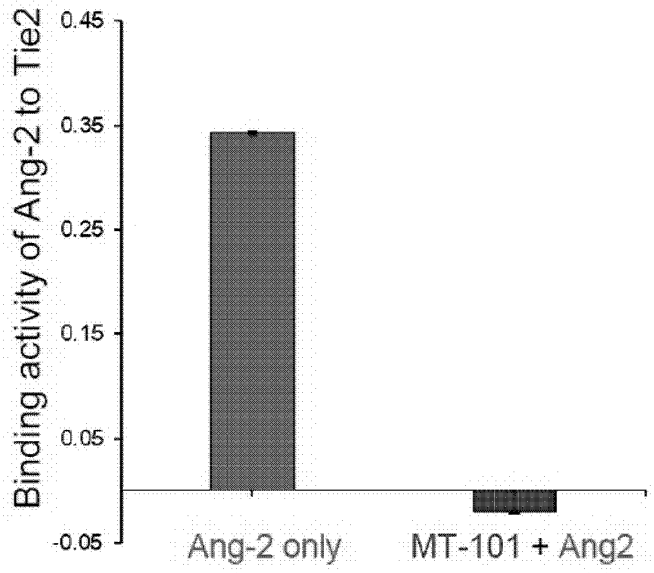
[도8c]



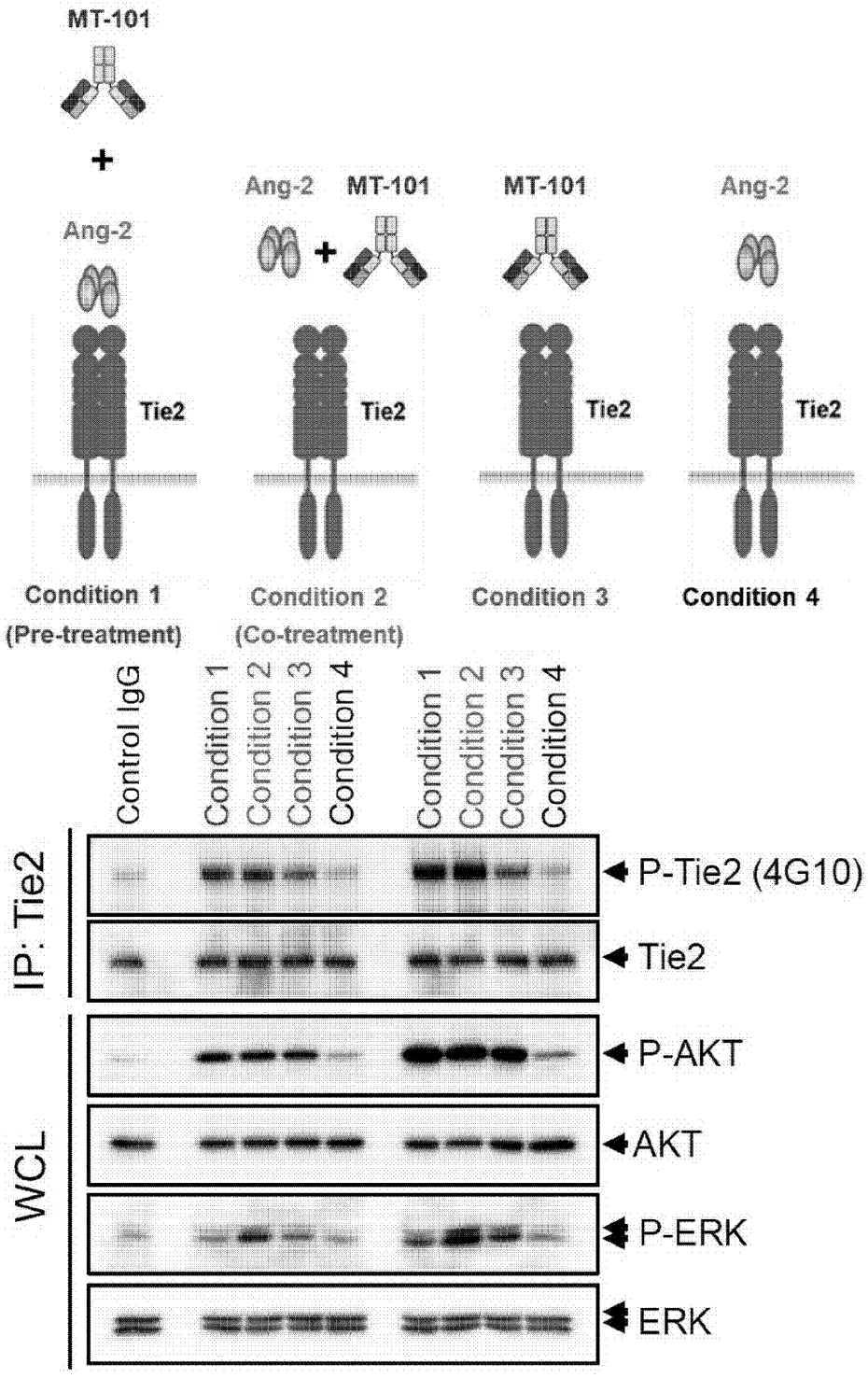
[도9a]


hTie2 loading
MT-101 binding
Ang-2 binding
Ang-2 only
MT-101 + Ang-2
① – Ang-2 only
② – MT-101 + Ang-2

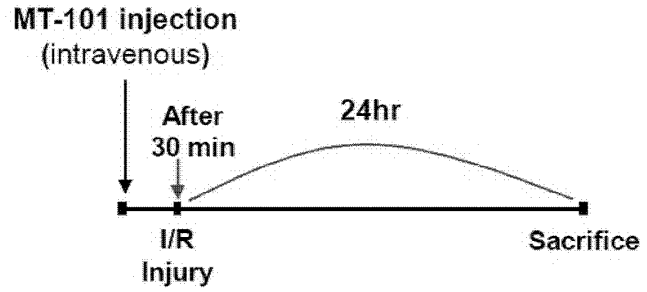
[도9b]


Binding activity of Ang-2 to Tie2
0.45
0.35
0.25
0.15
0.05
-0.05
Ang-2 only
MT-101 + Ang2

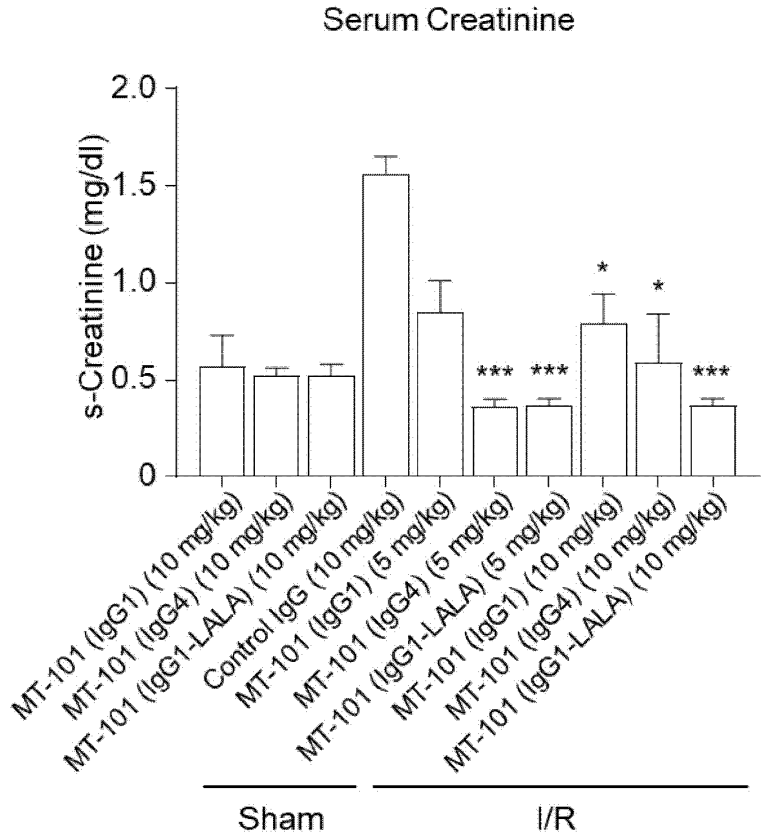
[도10]


MT-101
+
Ang-2
Tie2
Condition 1
(Pre-treatment)
Ang-2
MT-101
+
Tie2
Condition 2
(Co-treatment)
MT-101
Tie2
Condition 3
Ang-2
Tie2
Condition 4
Control IgG
Condition 1
Condition 2
Condition 3
Condition 4
Condition 1
Condition 2
Condition 3
Condition 4
IP: Tie2
P-Tie2 (4G10)
Tie2
WCL
P-AKT
AKT
P-ERK
ERK

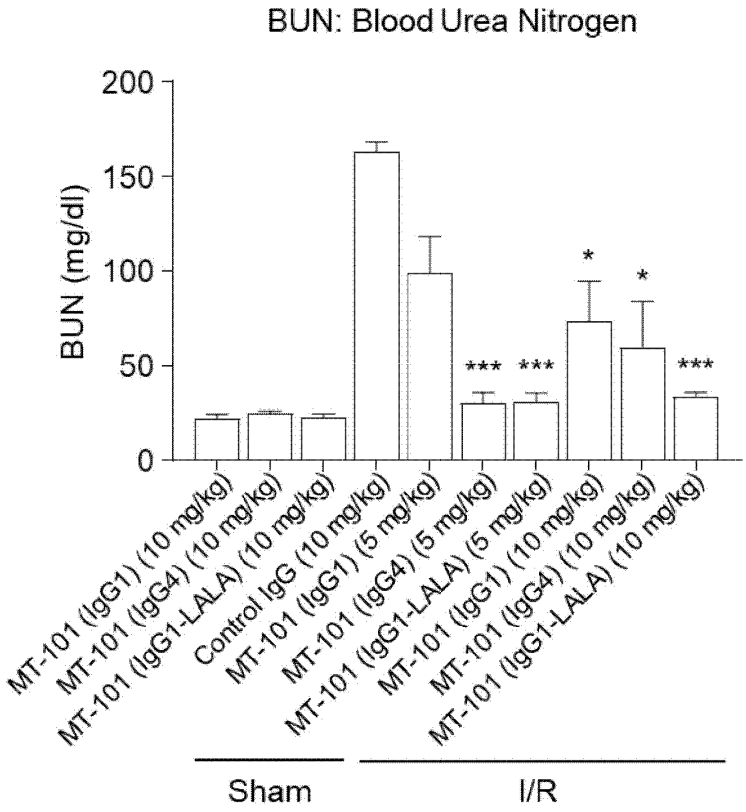
[도11a]


MT-101 injection
(intravenous)
After
30 min
24hr
I/R
Injury
Sacrifice

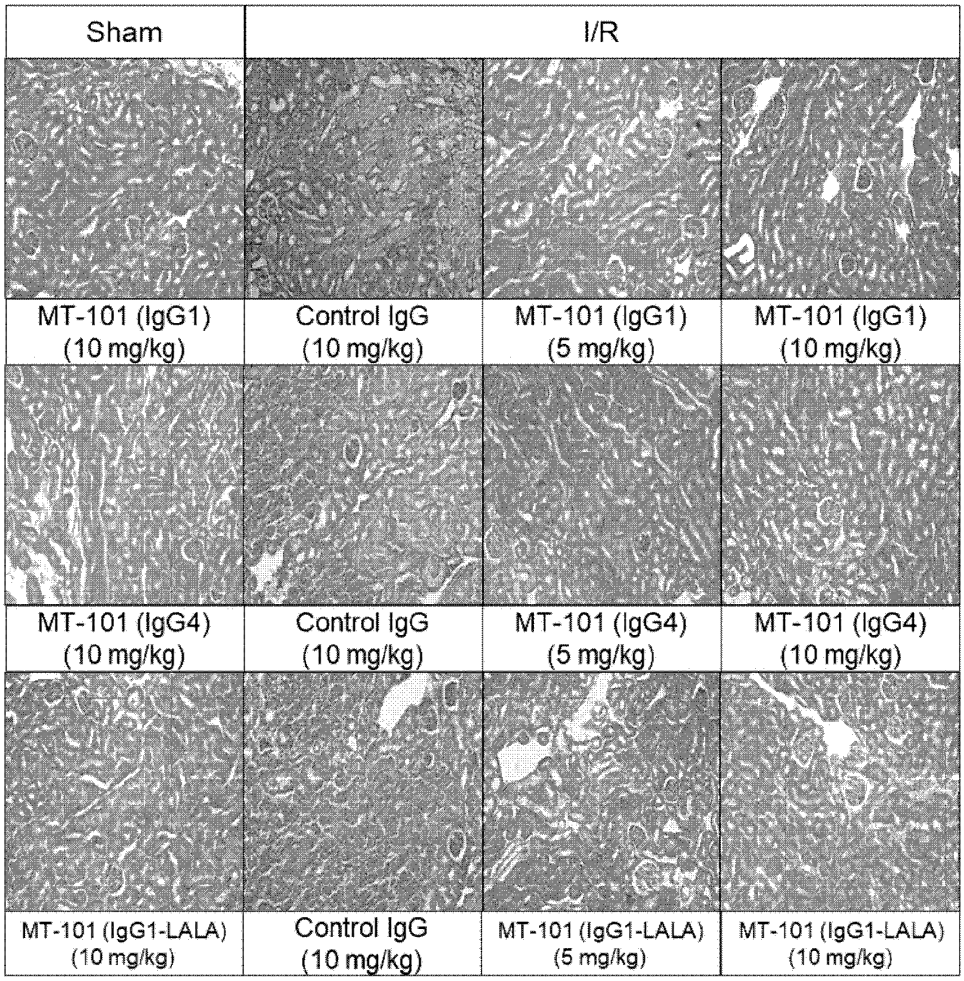
[도11b]


Serum Creatinine
s-Creatinine (mg/dl)
2.0
1.5
1.0
0.5
0
***
***
*
*
***
MT-101 (IgG1) (10 mg/kg)
MT-101 (IgG4) (10 mg/kg)
MT-101 (IgG1-LALA) (10 mg/kg)
Control IgG (10 mg/kg)
MT-101 (IgG1) (5 mg/kg)
MT-101 (IgG4) (5 mg/kg)
MT-101 (IgG1-LALA) (5 mg/kg)
MT-101 (IgG1) (10 mg/kg)
MT-101 (IgG4) (10 mg/kg)
MT-101 (IgG1-LALA) (10 mg/kg)
Sham
I/R

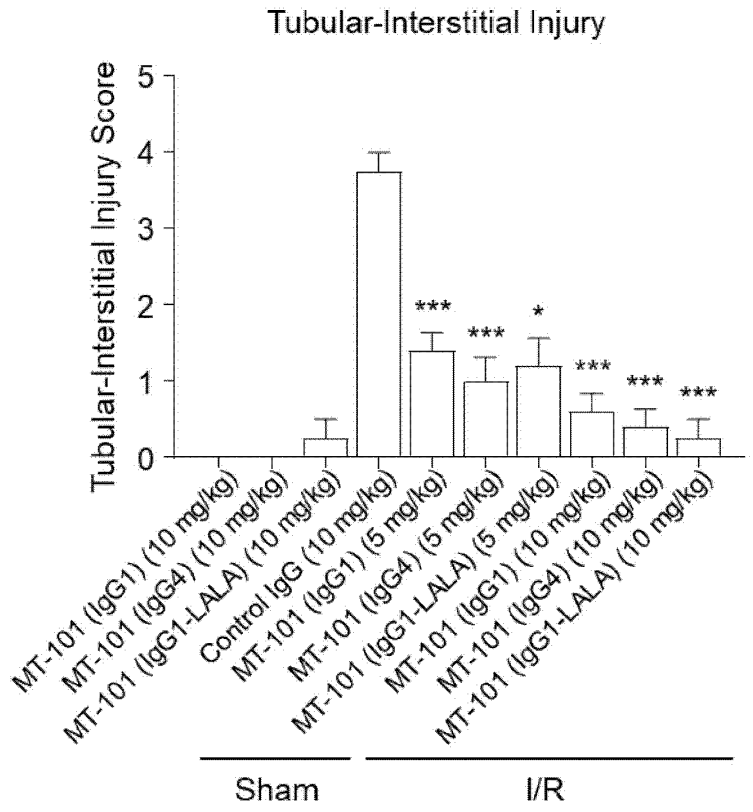
[도11c]


BUN: Blood Urea Nitrogen
BUN (mg/dl)
200
150
100
50
0
***
***
*
*
***
MT-101 (IgG1) (10 mg/kg)
MT-101 (IgG4) (10 mg/kg)
MT-101 (IgG1-LALA) (10 mg/kg)
Control IgG (10 mg/kg)
MT-101 (IgG1) (5 mg/kg)
MT-101 (IgG4) (5 mg/kg)
MT-101 (IgG1-LALA) (5 mg/kg)
MT-101 (IgG1) (10 mg/kg)
MT-101 (IgG4) (10 mg/kg)
MT-101 (IgG1-LALA) (10 mg/kg)
Sham
I/R

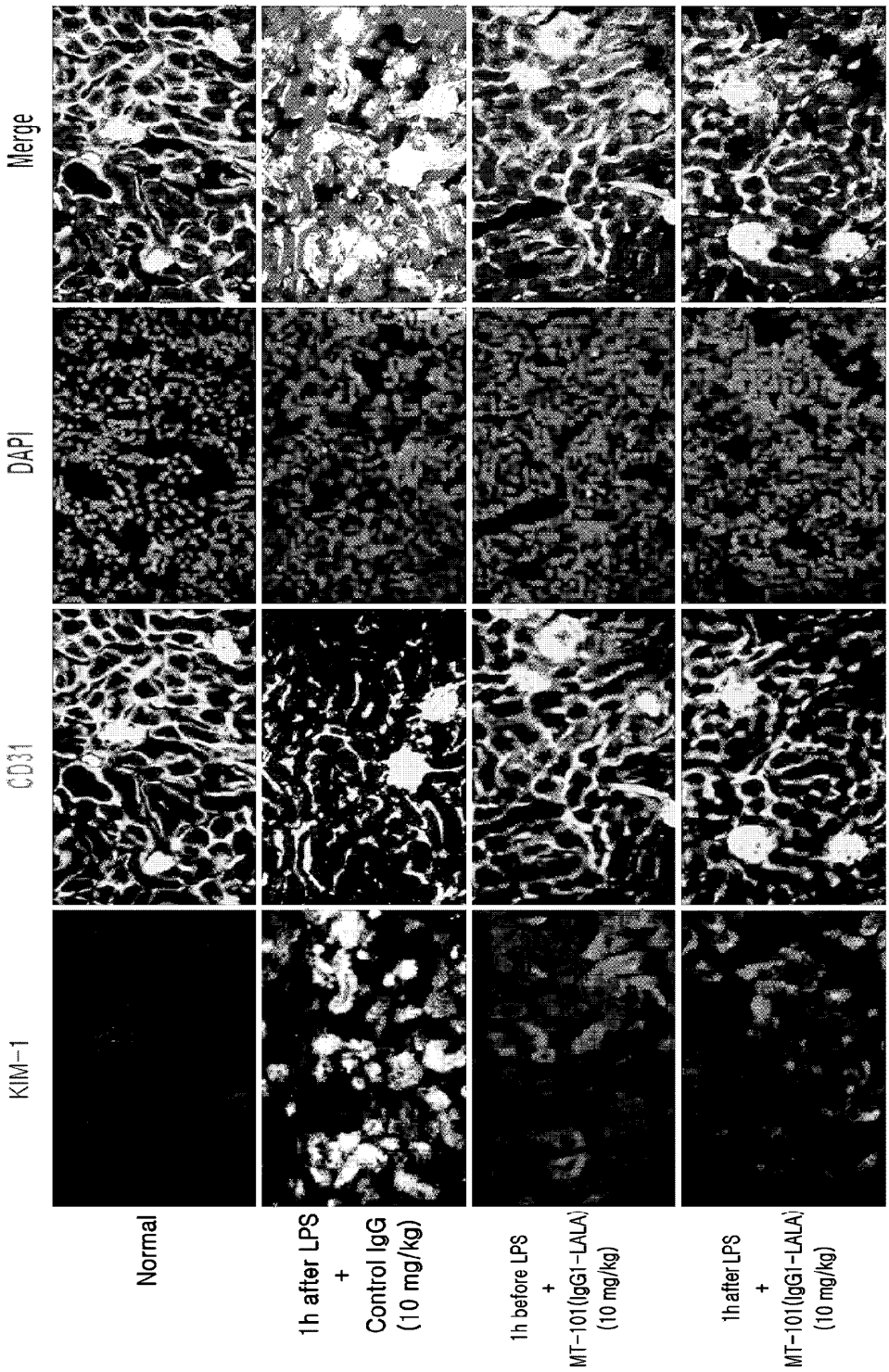
[도12a]


Sham
I/R
MT-101 (IgG1) (10 mg/kg)
Control IgG (10 mg/kg)
MT-101 (IgG1) (5 mg/kg)
MT-101 (IgG1) (10 mg/kg)
MT-101 (IgG4) (10 mg/kg)
Control IgG (10 mg/kg)
MT-101 (IgG4) (5 mg/kg)
MT-101 (IgG4) (10 mg/kg)
MT-101 (IgG1-LALA) (10 mg/kg)
Control IgG (10 mg/kg)
MT-101 (IgG1-LALA) (5 mg/kg)
MT-101 (IgG1-LALA) (10 mg/kg)

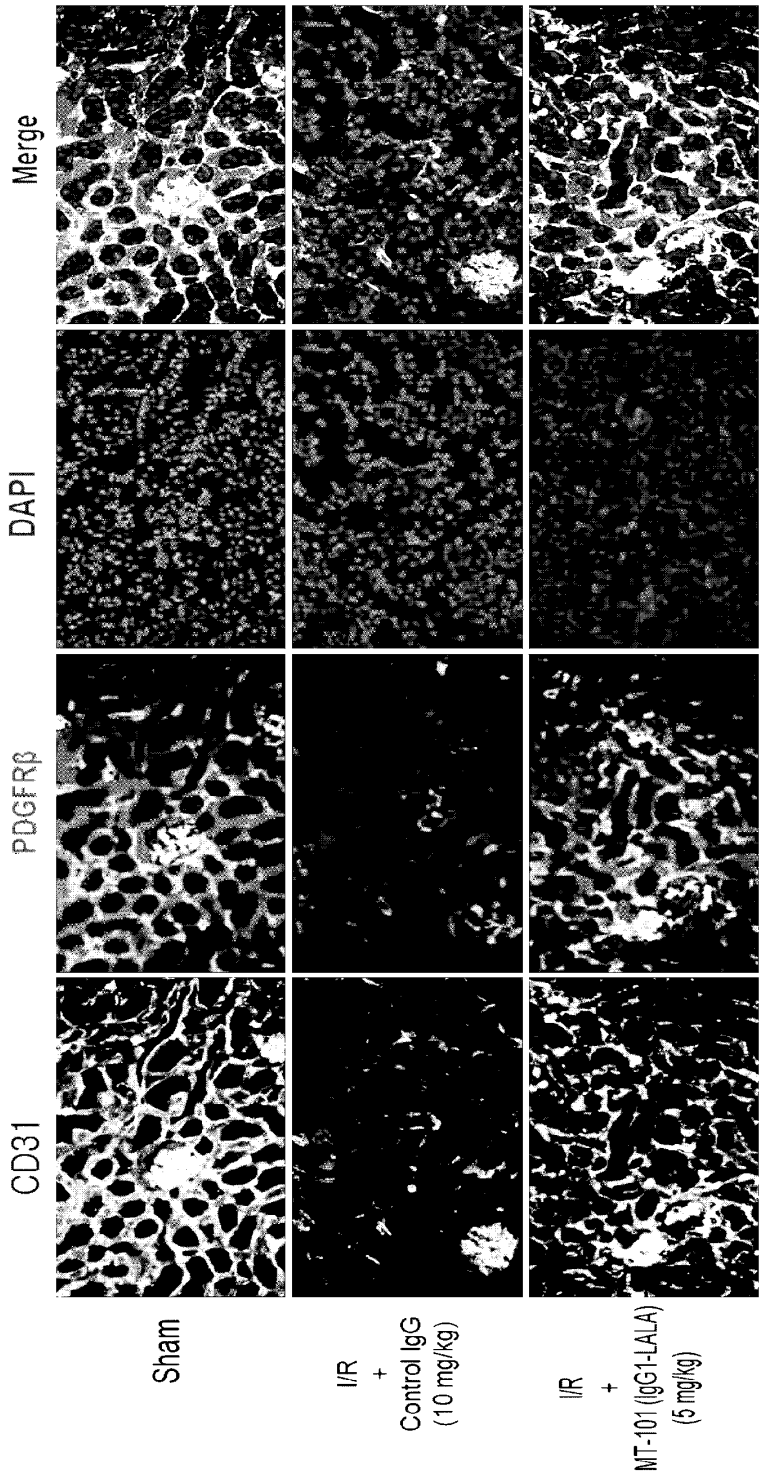
[도12b]


Tubular-Interstitial Injury
Tubular-Interstitial Injury Score
0
1
2
3
4
5
***
***
*
***
***
***
MT-101 (IgG1) (10 mg/kg)
MT-101 (IgG4) (10 mg/kg)
MT-101 (IgG1-LALA) (10 mg/kg)
Control IgG (10 mg/kg)
MT-101 (IgG1) (5 mg/kg)
MT-101 (IgG4) (5 mg/kg)
MT-101 (IgG1-LALA) (5 mg/kg)
MT-101 (IgG1) (10 mg/kg)
MT-101 (IgG4) (10 mg/kg)
MT-101 (IgG1-LALA) (10 mg/kg)
Sham
I/R

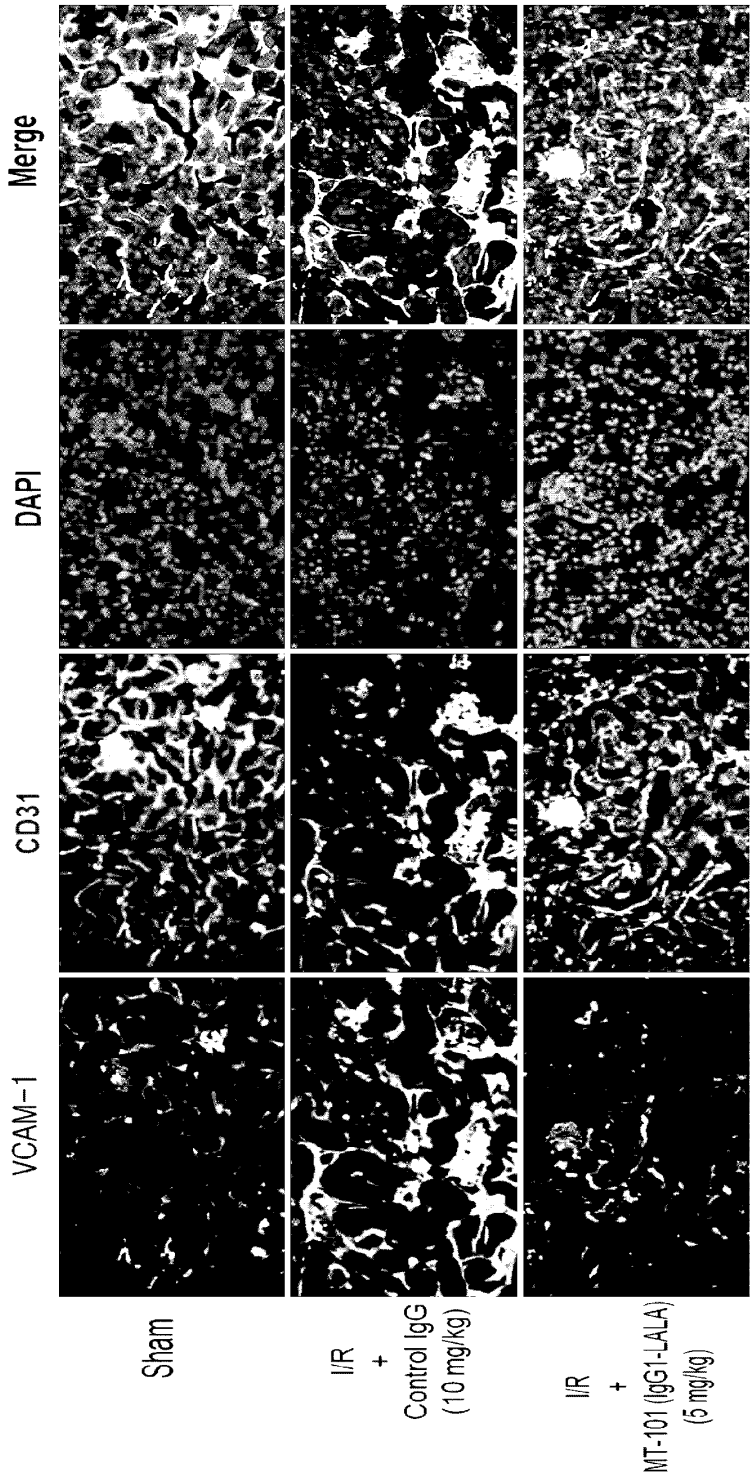
[도13a]


KIM-1
CD31
DAPI
Merge
Normal
1h after LPS + Control IgG (10 mg/kg)
1h before LPS + MT-101(IgG1-LALA) (10 mg/kg)
1h after LPS + MT-101(IgG1-LALA) (10 mg/kg)

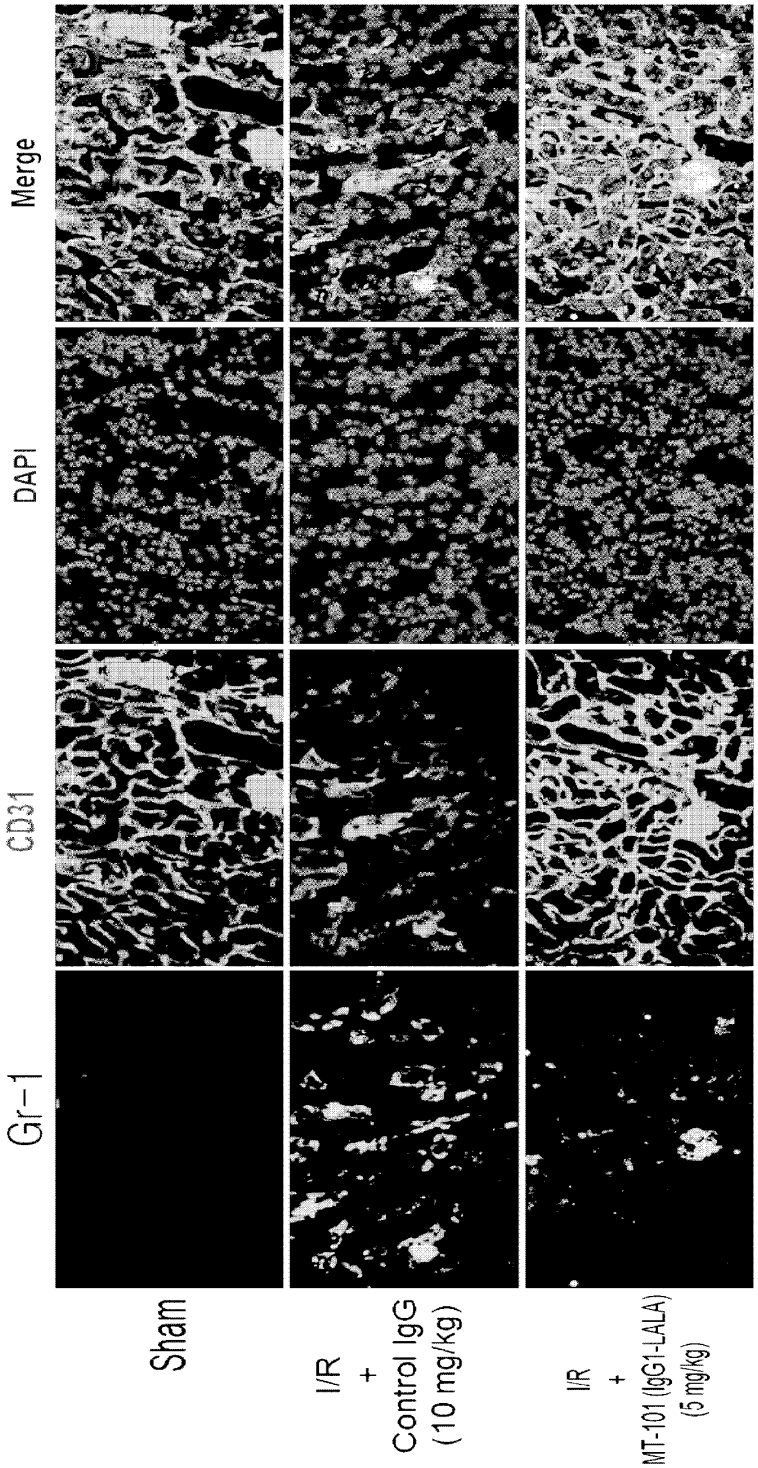
[도13b]


CD31
PDGFRβ
DAPI
Merge
Sham
I/R + Control IgG (10 mg/kg)
I/R + MT-101 (IgG1-LALA) (5 mg/kg)

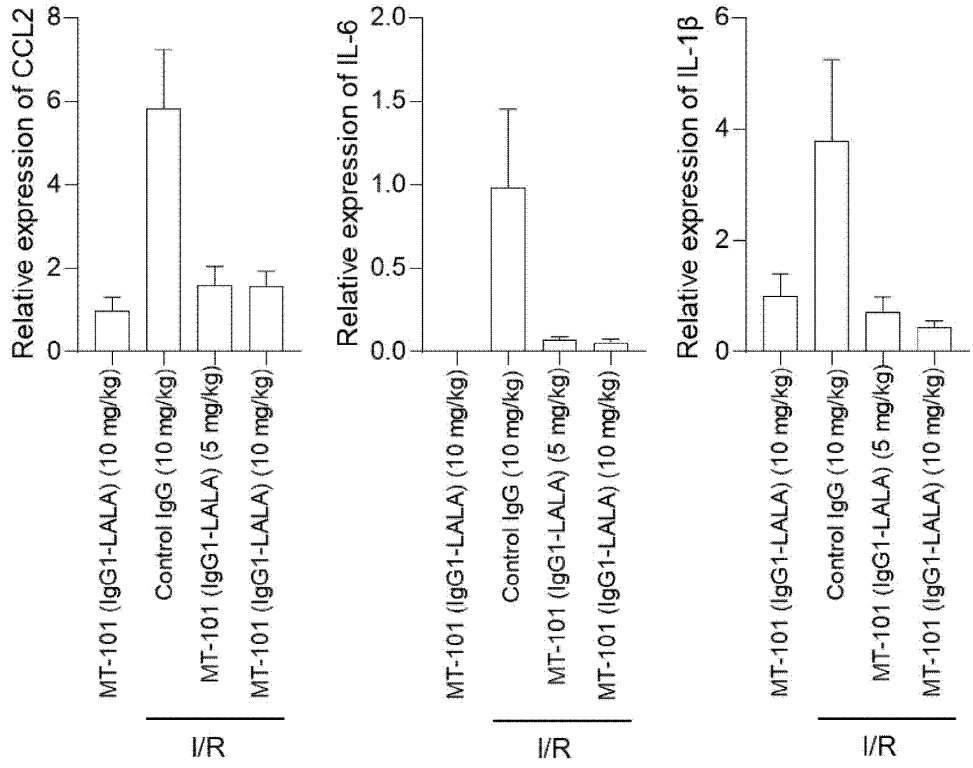
[도14a]


Merge
DAPI
CD31
VCAM-1
Sham
I/R
+
Control IgG
(10 mg/kg)
I/R
+
MT-101 (IgG1-LALA)
(5 mg/kg)

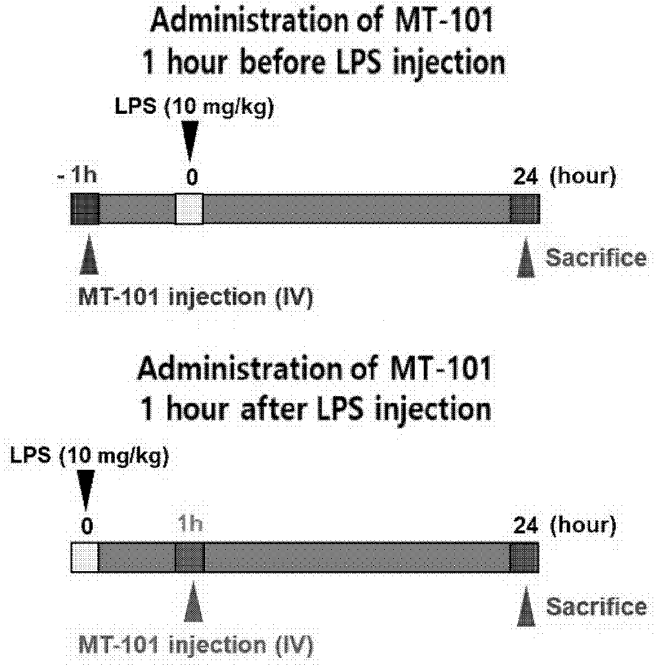
[도14b]


Merge
DAPI
CD31
Gr-1
Sham
I/R + Control IgG (10 mg/kg)
I/R + MT-101 (IgG1-LALA) (5 mg/kg)

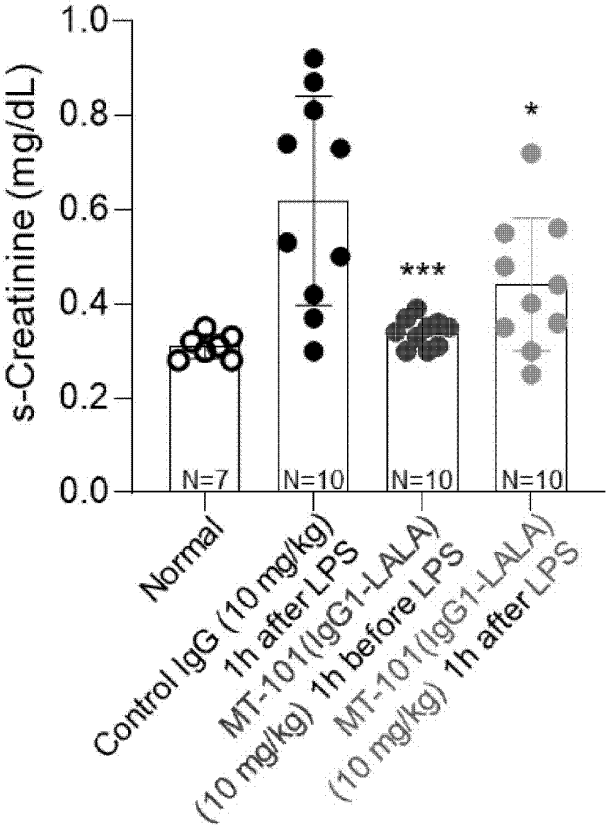
[도14c]


Relative expression of CCL2
Relative expression of IL-6
Relative expression of IL-1β
MT-101 (IgG1-LALA) (10 mg/kg)
Control IgG (10 mg/kg)
MT-101 (IgG1-LALA) (5 mg/kg)
MT-101 (IgG1-LALA) (10 mg/kg)
I/R

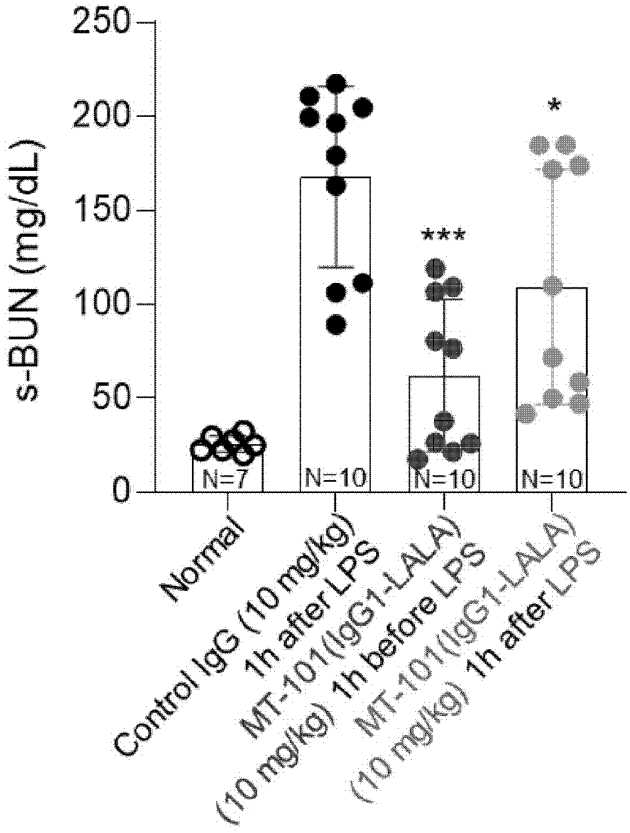
[도15a]


Administration of MT-101
1 hour before LPS injection
LPS (10 mg/kg)
- 1h
0
24 (hour)
Sacrifice
MT-101 injection (IV)
Administration of MT-101
1 hour after LPS injection
LPS (10 mg/kg)
0
1h
24 (hour)
Sacrifice
MT-101 injection (IV)

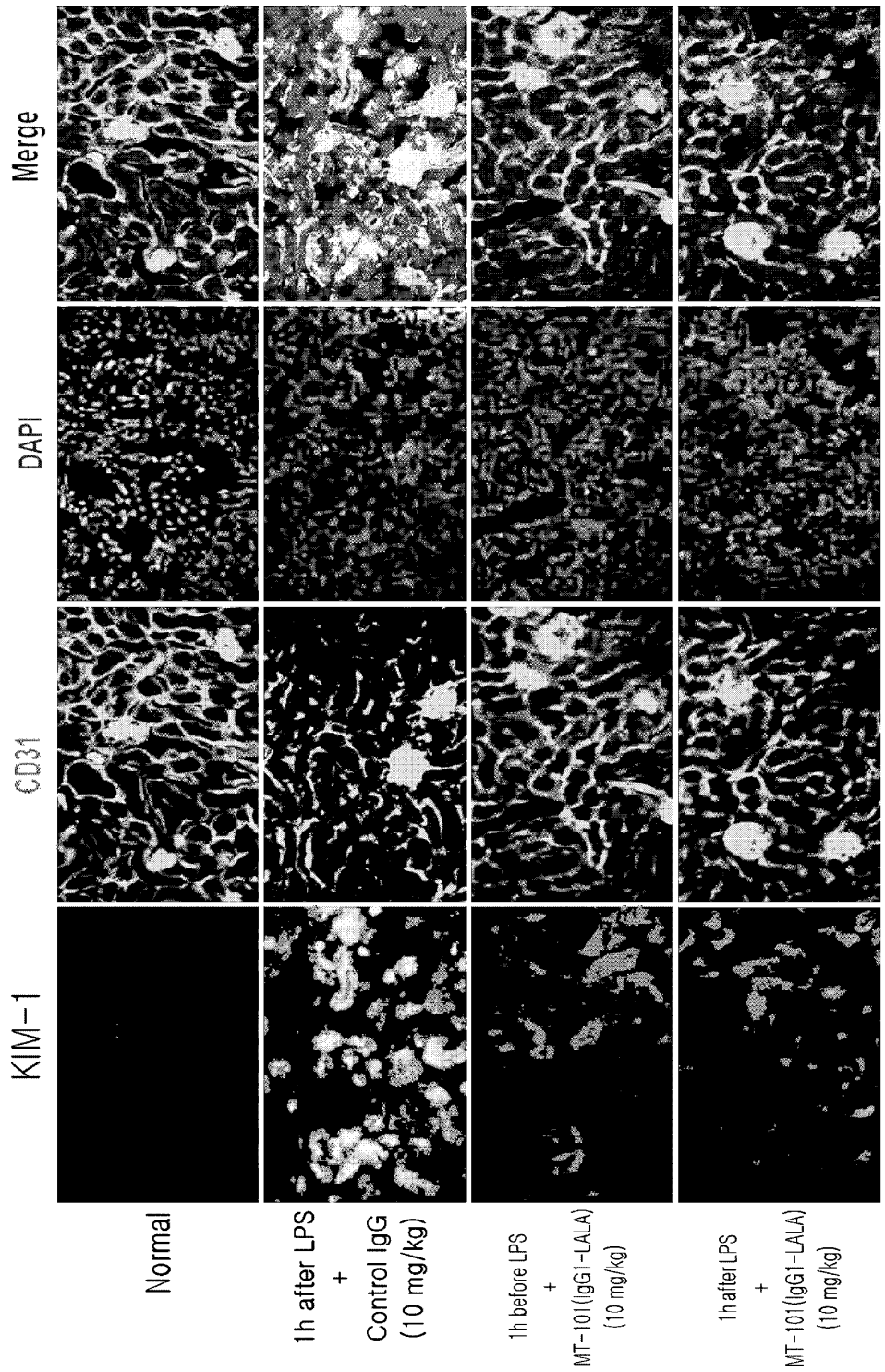
[도15b]


s-Creatinine (mg/dL)
1.0
0.8
0.6
0.4
0.2
0.0
N=7
N=10
N=10
N=10
***
*
Normal
Control IgG (10 mg/kg) 1h after LPS
MT-101(IgG1-LALA) (10 mg/kg) 1h before LPS
MT-101(IgG1-LALA) (10 mg/kg) 1h after LPS

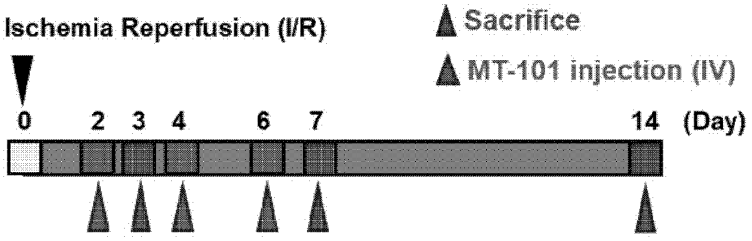
[도15c]


s-BUN (mg/dL)
250
200
150
100
50
0
N=7
N=10
N=10
N=10
***
*
Normal
Control IgG (10 mg/kg) 1h after LPS
MT-101(IgG1-LALA) (10 mg/kg) 1h before LPS
MT-101(IgG1-LALA) (10 mg/kg) 1h after LPS

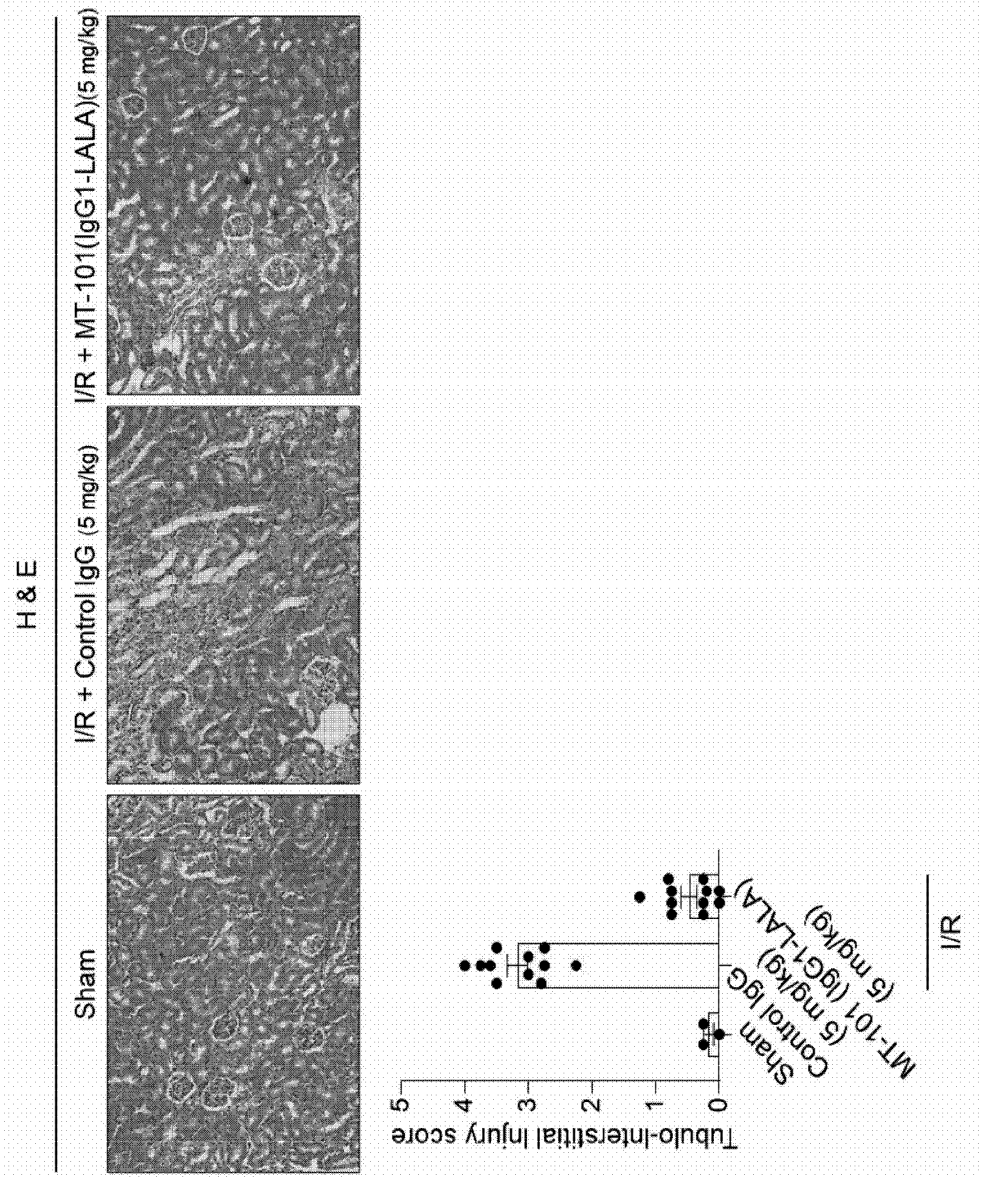
[도15d]


KIM-1
CD31
DAPI
Merge
Normal
1h after LPS + Control IgG (10 mg/kg)
1h before LPS + MT-101(IgG1-LALA) (10 mg/kg)
1h after LPS + MT-101(IgG1-LALA) (10 mg/kg)

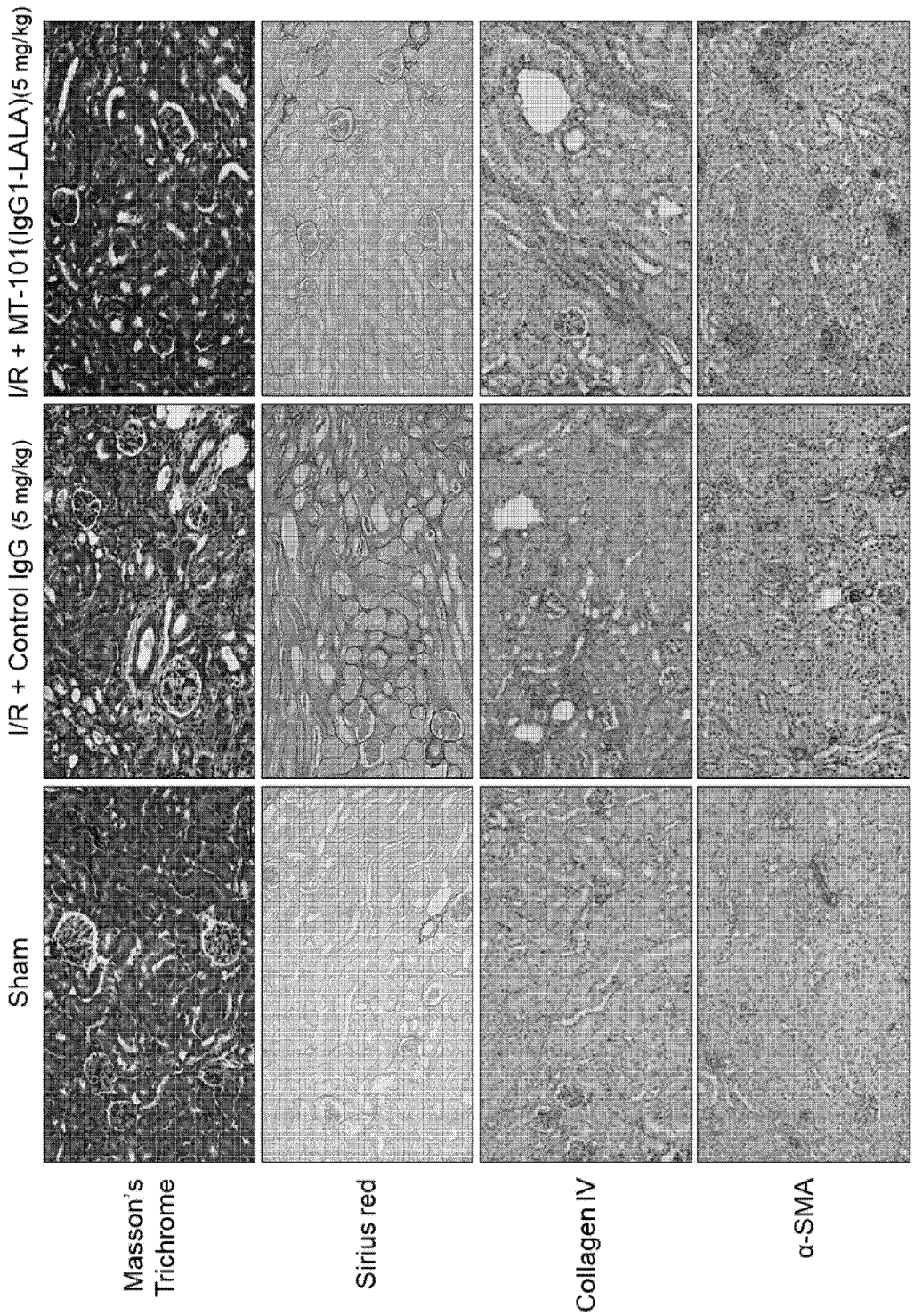
[도16a]


Ischemia Reperfusion (I/R)
Sacrifice
MT-101 injection (IV)
0
2
3
4
6
7
14
(Day)

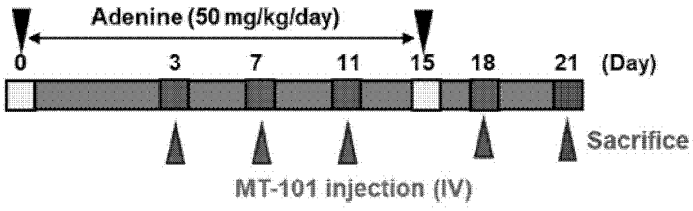
[도16b]


H & E
Sham
I/R + Control IgG (5 mg/kg)
I/R + MT-101(IgG1-LALA)(5 mg/kg)
Tubulo-Interstitial injury score
0
1
2
3
4
5
Sham
Control IgG (5 mg/kg)
MT-101 (IgG1-LALA) (5 mg/kg)
I/R

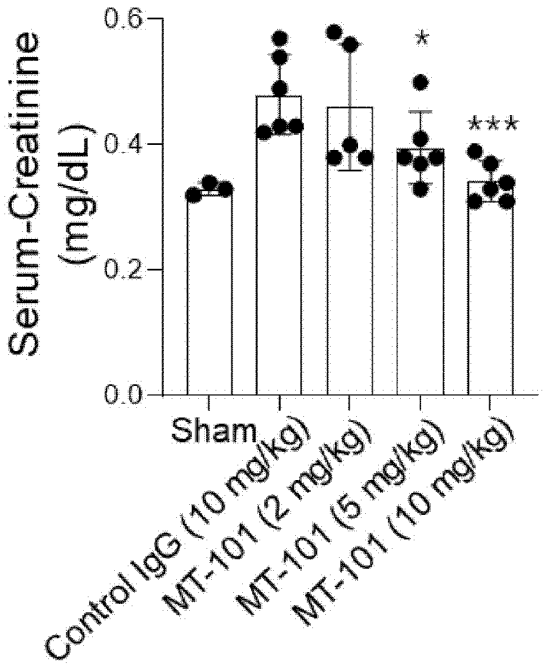
[도16c]


Sham
I/R + Control IgG (5 mg/kg)
I/R + MT-101(IgG1-LALA)(5 mg/kg)
Masson's Trichrome
Sirius red
Collagen IV
α-SMA

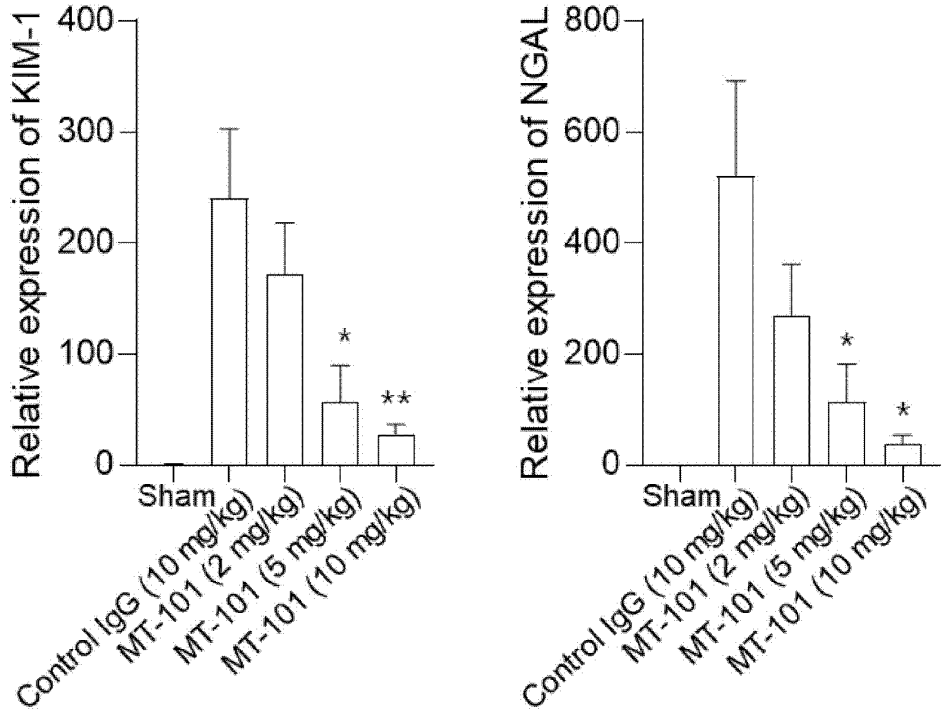
[도17a]


Adenine (50 mg/kg/day)
0
3
7
11
15
18
21
(Day)
Sacrifice
MT-101 injection (IV)

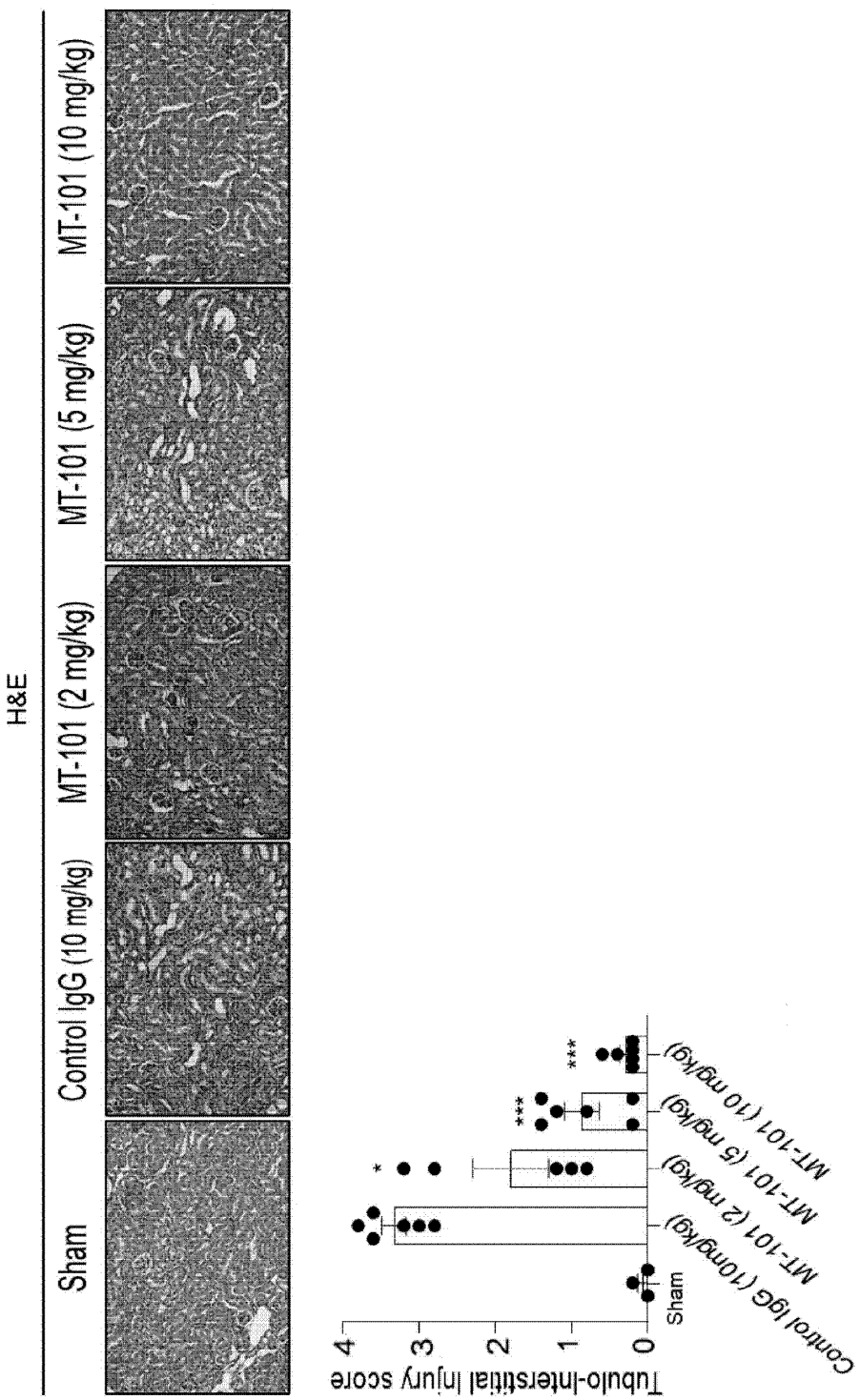
[도17b]


Serum-Creatinine (mg/dL)
0.6
0.4
0.2
0.0
*
***
Sham
Control IgG (10 mg/kg)
MT-101 (2 mg/kg)
MT-101 (5 mg/kg)
MT-101 (10 mg/kg)

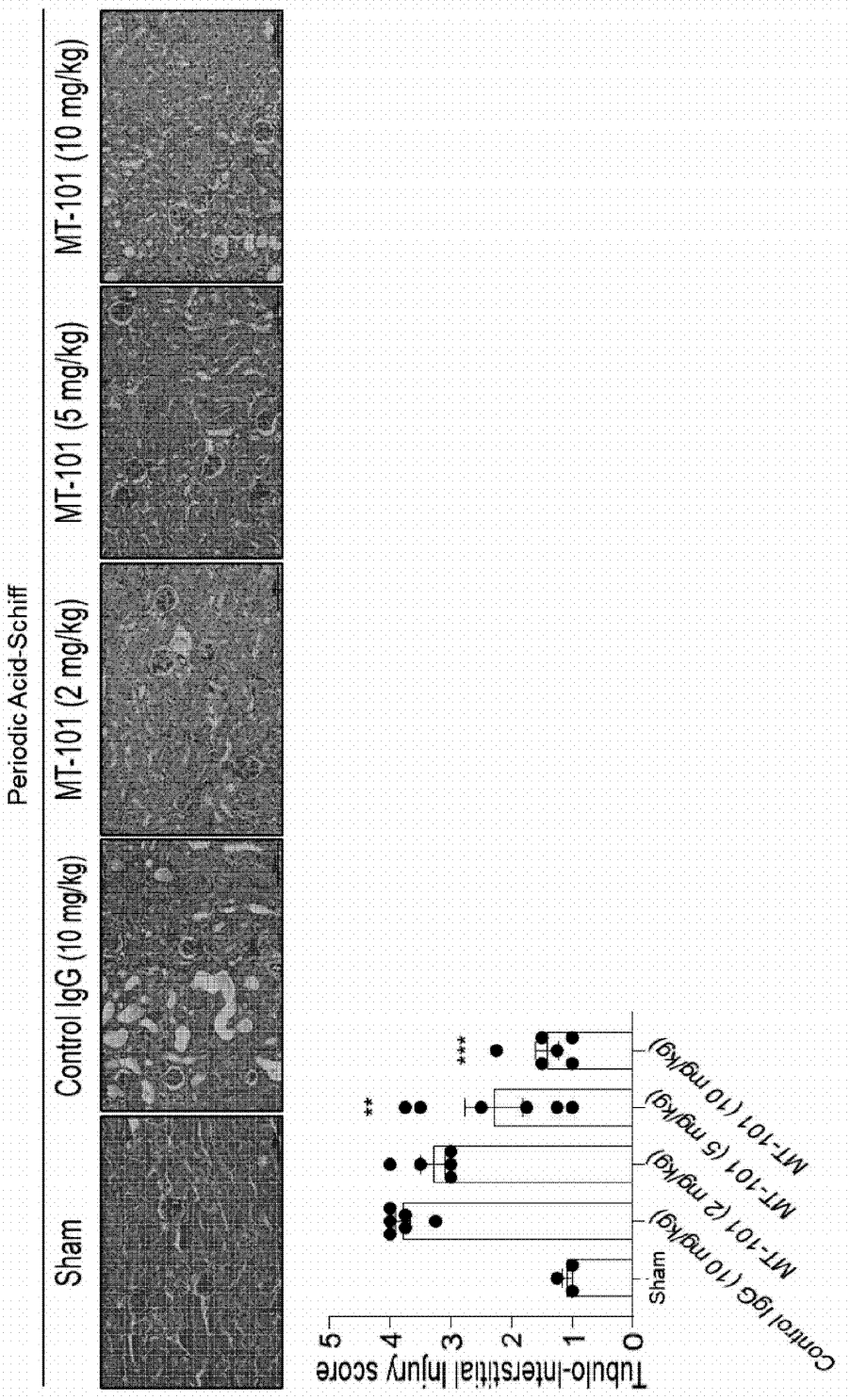
[도17c]


Relative expression of KIM-1
400
300
200
100
0
*
**
Sham
Control IgG (10 mg/kg)
MT-101 (2 mg/kg)
MT-101 (5 mg/kg)
MT-101 (10 mg/kg)
Relative expression of NGAL
800
600
400
200
0
*
*
Sham
Control IgG (10 mg/kg)
MT-101 (2 mg/kg)
MT-101 (5 mg/kg)
MT-101 (10 mg/kg)

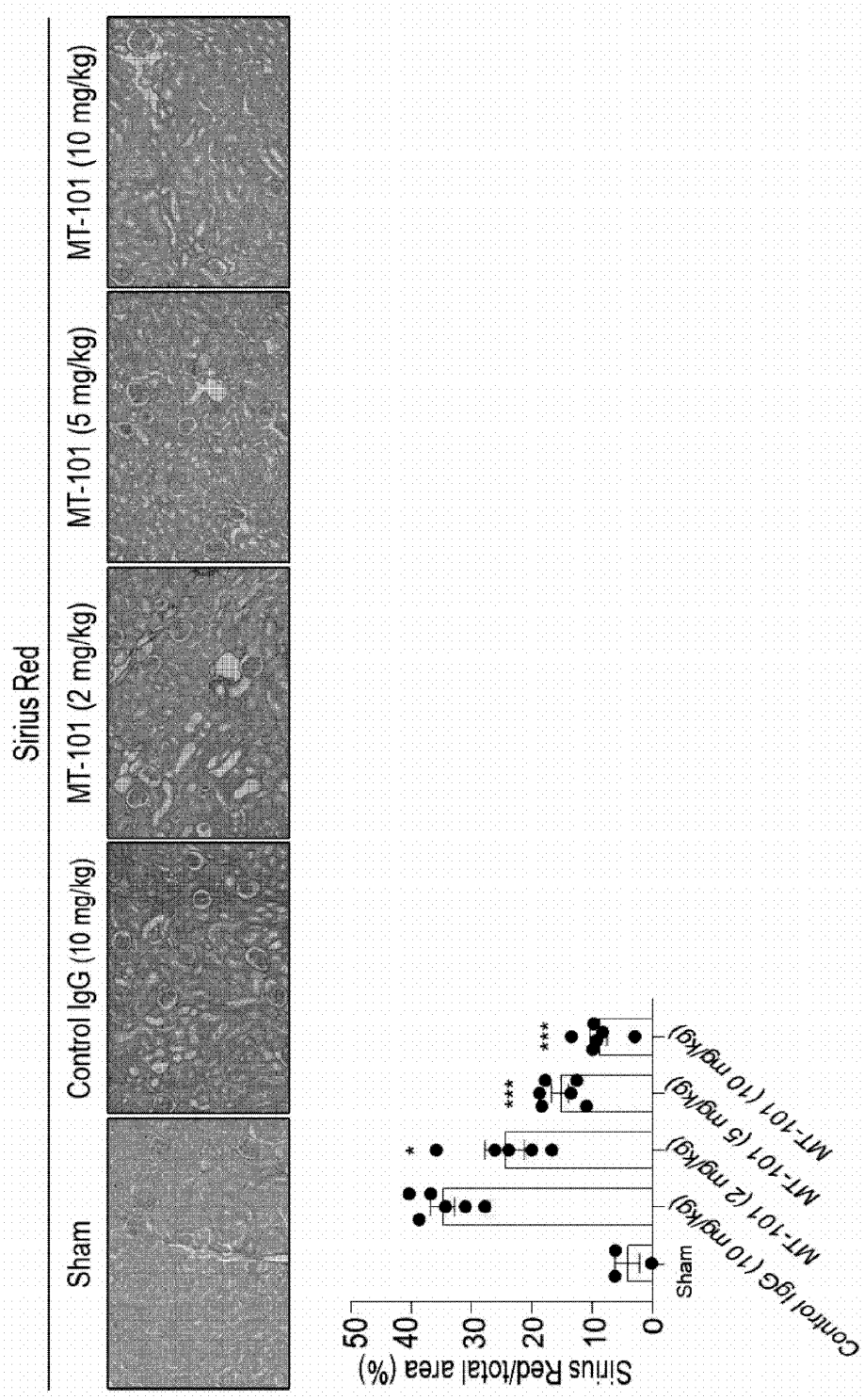
[도17d]


H&E
Sham
Control IgG (10 mg/kg)
MT-101 (2 mg/kg)
MT-101 (5 mg/kg)
MT-101 (10 mg/kg)
Tubulo-interstitial injury score
0
1
2
3
4
Sham
Control IgG (10mg/kg)
MT-101 (2 mg/kg)
MT-101 (5 mg/kg)
MT-101 (10 mg/kg)
*
***
***

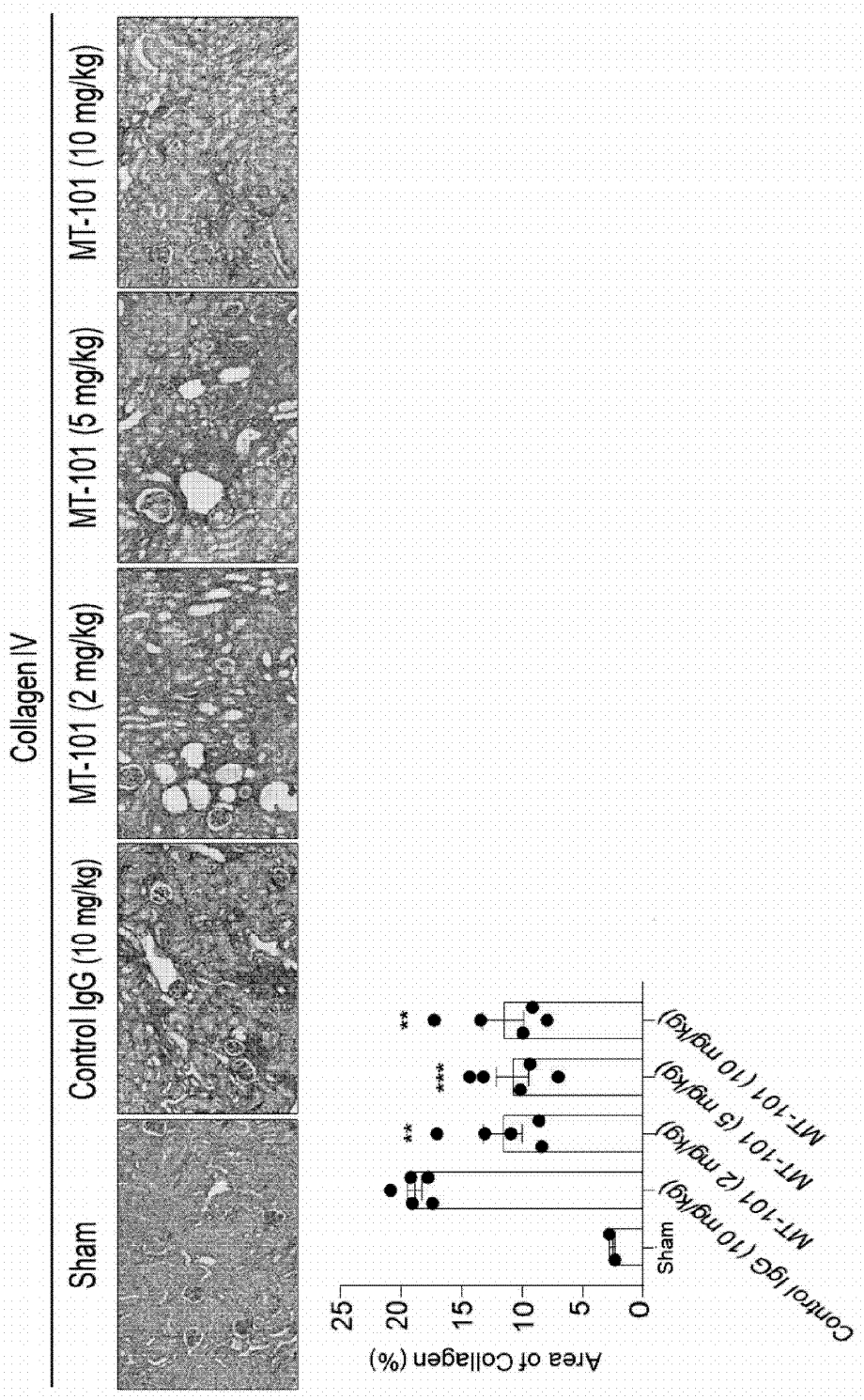
[도17e]


Periodic Acid-Schiff
Sham
Control IgG (10 mg/kg)
MT-101 (2 mg/kg)
MT-101 (5 mg/kg)
MT-101 (10 mg/kg)
Tubulo-interstitial injury score
0
1
2
3
4
5
**
***
Sham
Control IgG (10 mg/kg)
MT-101 (2 mg/kg)
MT-101 (5 mg/kg)
MT-101 (10 mg/kg)

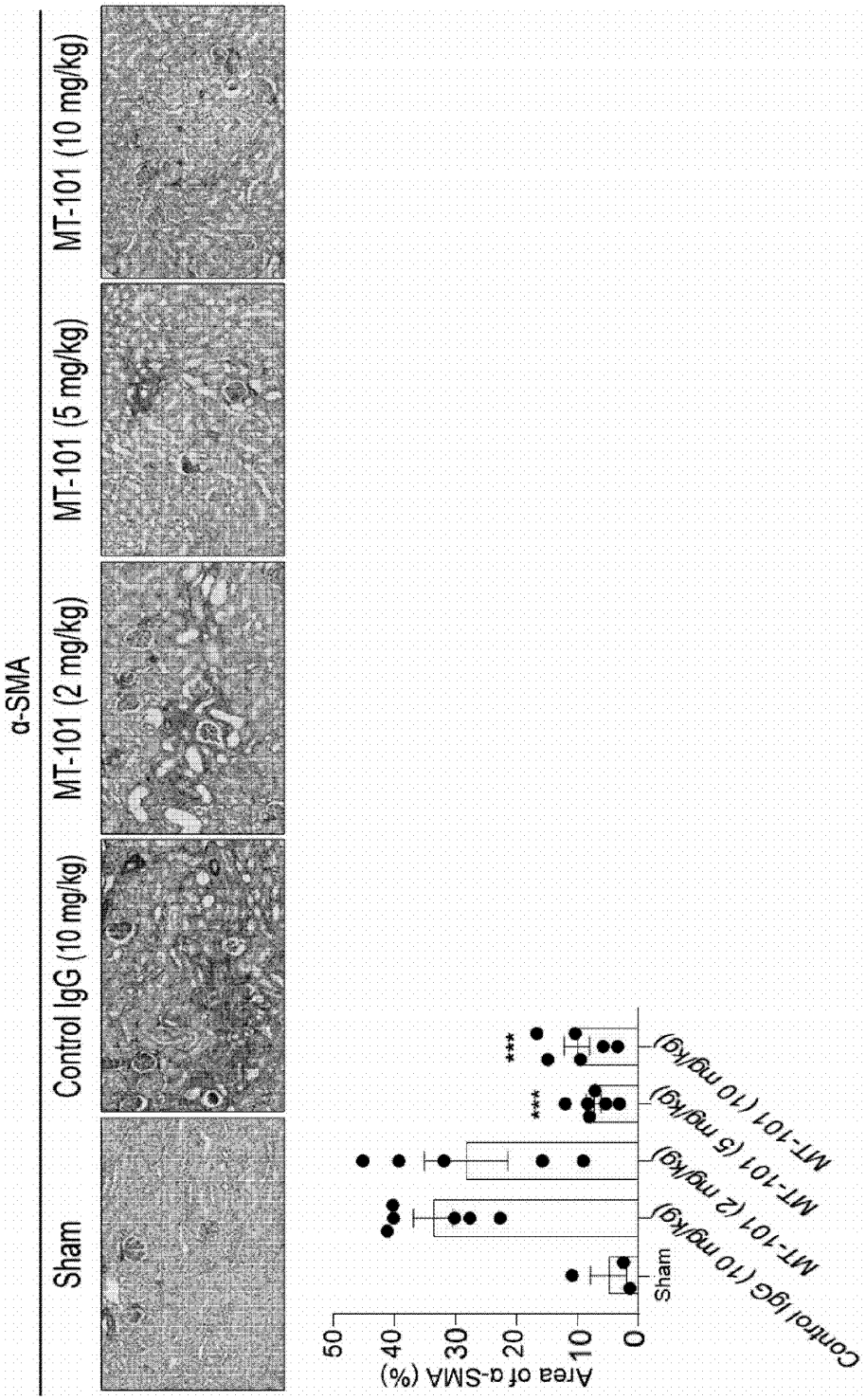
[도18a]


Sirius Red
Sham
Control IgG (10 mg/kg)
MT-101 (2 mg/kg)
MT-101 (5 mg/kg)
MT-101 (10 mg/kg)
Sirius Red/total area (%)
0
10
20
30
40
50
Sham
Control IgG (10 mg/kg)
MT-101 (2 mg/kg)
MT-101 (5 mg/kg)
MT-101 (10 mg/kg)
*
***
***

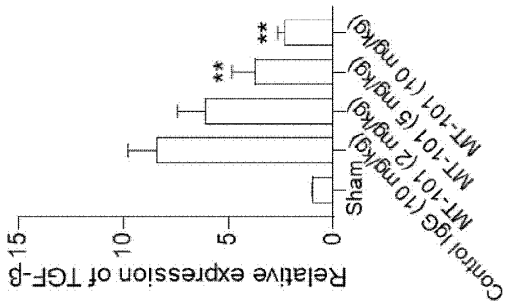
[도18b]

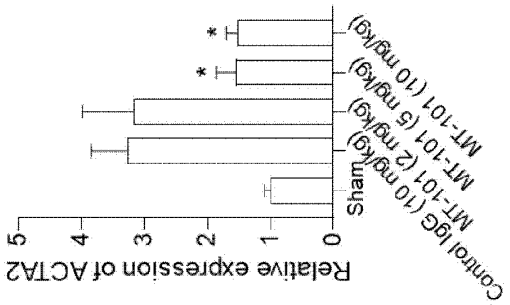

Collagen IV
Sham
Control IgG (10 mg/kg)
MT-101 (2 mg/kg)
MT-101 (5 mg/kg)
MT-101 (10 mg/kg)
Area of Collagen (%)
0
5
10
15
20
25
**
**
***
**
Sham
Control IgG (10 mg/kg)
MT-101 (2 mg/kg)
MT-101 (5 mg/kg)
MT-101 (10 mg/kg)

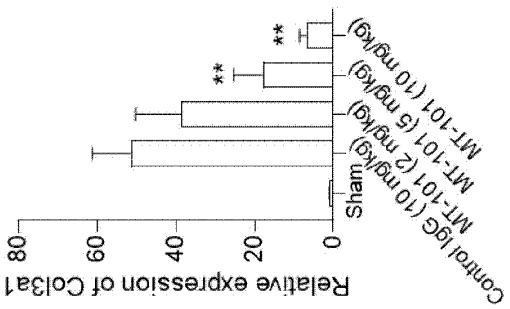
[도18c]

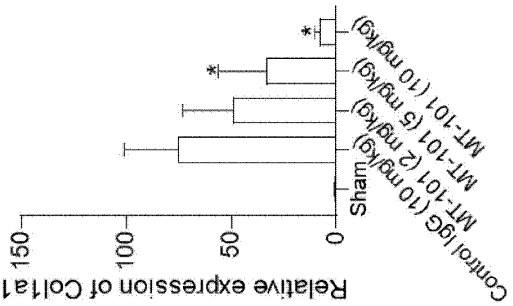

α-SMA
Sham
Control IgG (10 mg/kg)
MT-101 (2 mg/kg)
MT-101 (5 mg/kg)
MT-101 (10 mg/kg)
Area of α-SMA (%)
0
10
20
30
40
50
Sham
Control IgG (10 mg/kg)
MT-101 (2 mg/kg)
MT-101 (5 mg/kg)
MT-101 (10 mg/kg)
***
***

[도18d]

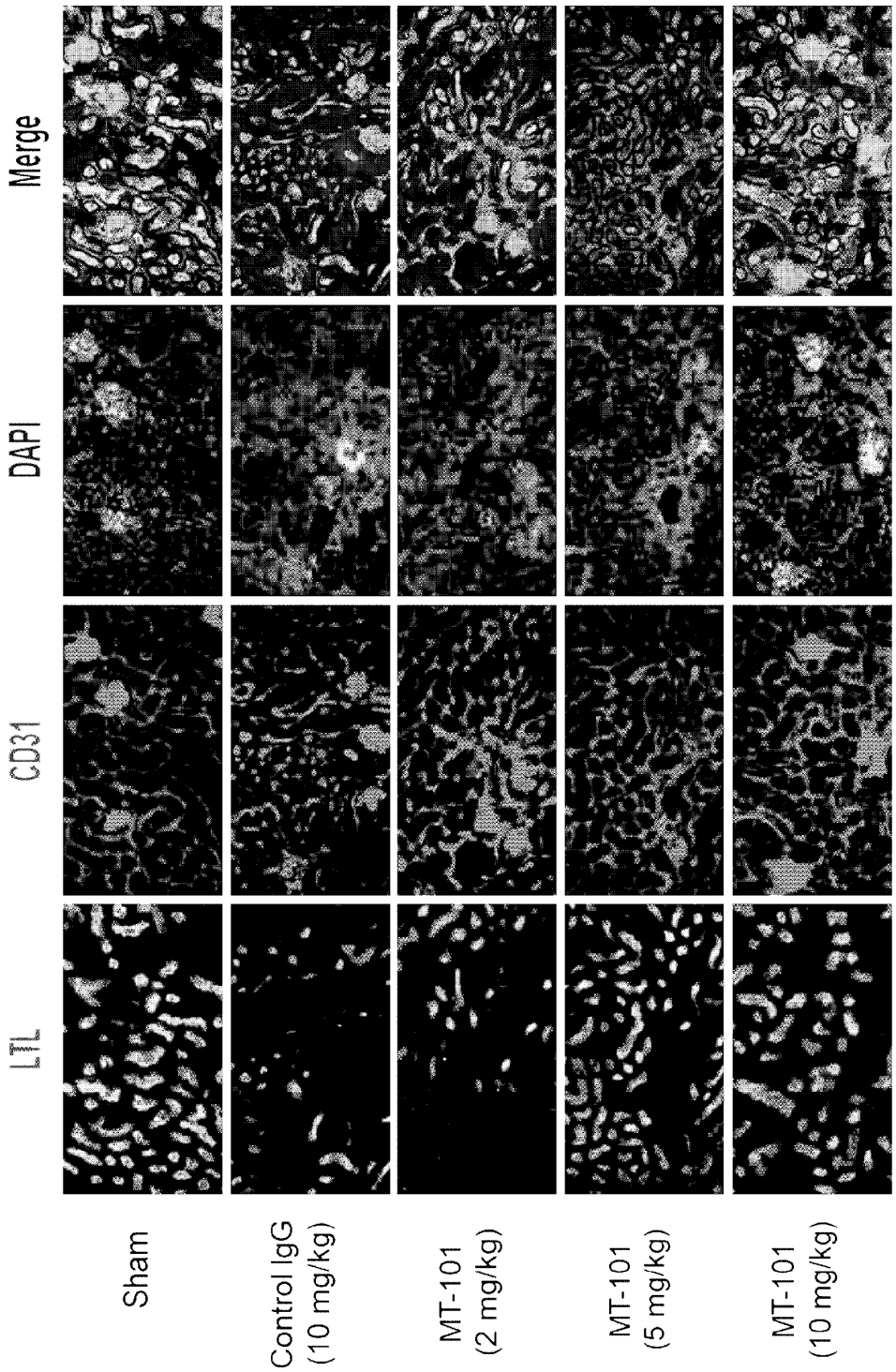

Relative expression of TGF-β
0
5
10
15
Sham
MT-101 (2 mg/kg)
MT-101 (5 mg/kg)
MT-101 (10 mg/kg)
Control IgG (10 mg/kg)
**
**


Relative expression of ACTA2
0
1
2
3
4
5
Sham
MT-101 (2 mg/kg)
MT-101 (5 mg/kg)
MT-101 (10 mg/kg)
Control IgG (10 mg/kg)
*
*

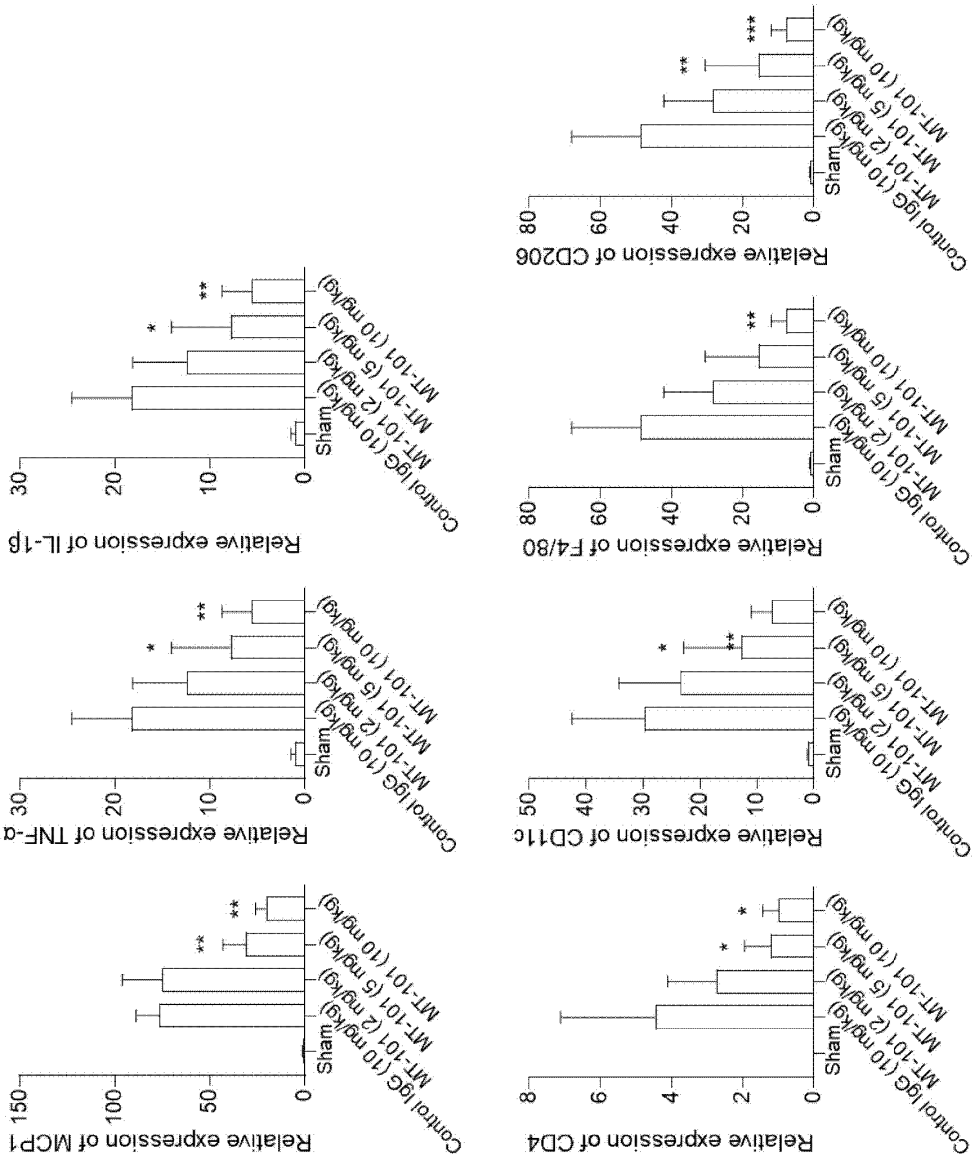

Relative expression of Col3a1
0
20
40
60
80
Sham
MT-101 (2 mg/kg)
MT-101 (5 mg/kg)
MT-101 (10 mg/kg)
Control IgG (10 mg/kg)
**
**


Relative expression of Col1a1
0
50
100
150
Sham
MT-101 (2 mg/kg)
MT-101 (5 mg/kg)
MT-101 (10 mg/kg)
Control IgG (10 mg/kg)
*
*

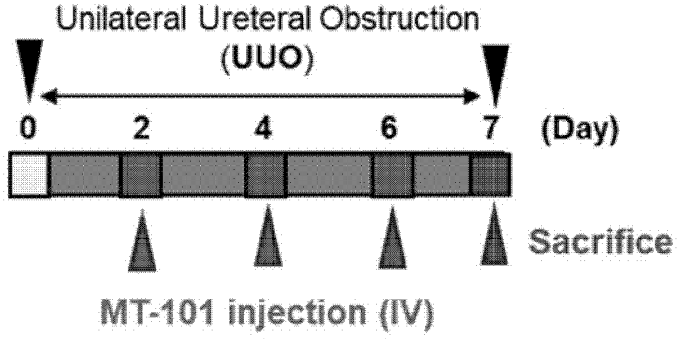
[도19a]


Merge
DAPI
CD31
LTL
Sham
Control IgG (10 mg/kg)
MT-101 (2 mg/kg)
MT-101 (5 mg/kg)
MT-101 (10 mg/kg)

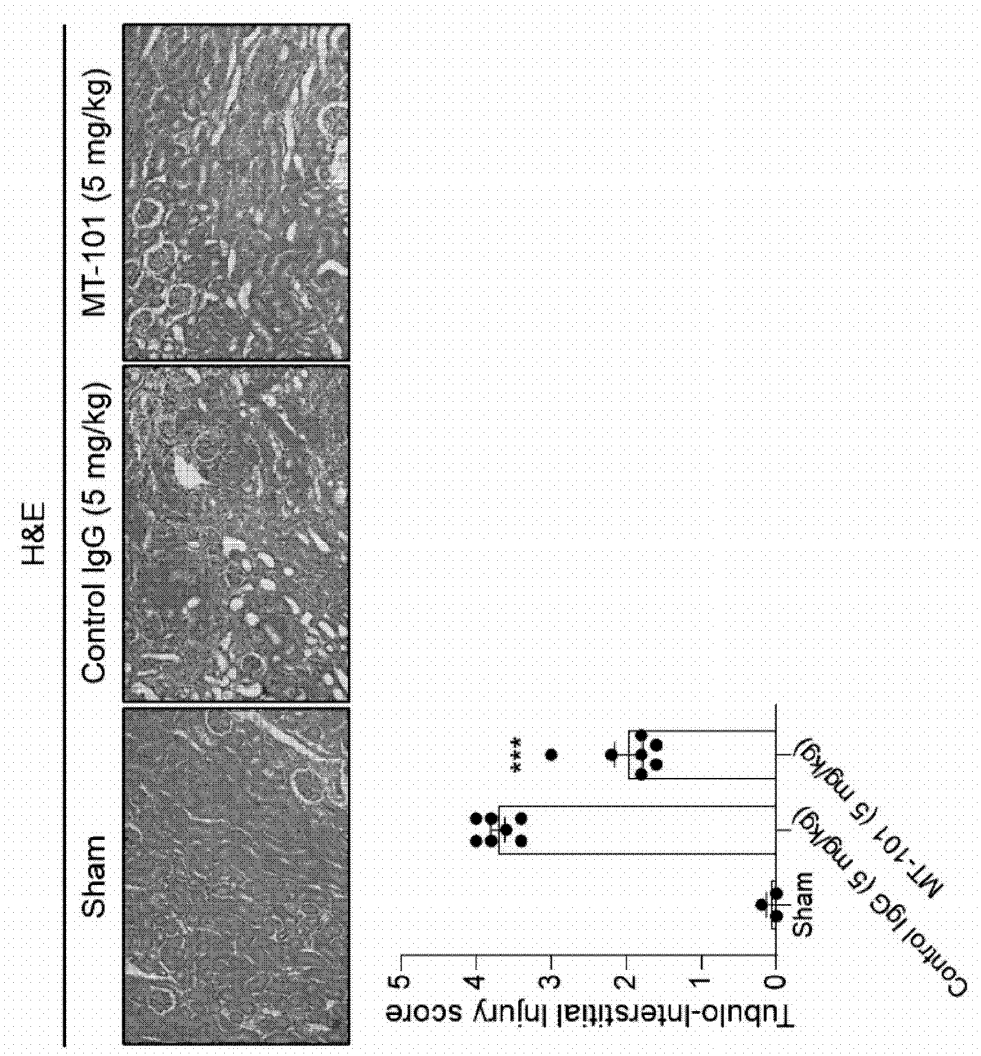
[도19b]


Relative expression of CD206
Relative expression of F4/80
Relative expression of CD11c
Relative expression of CD4
Relative expression of IL-1β
Relative expression of TNF-α
Relative expression of MCP1
Control IgG (10 mg/kg)
MT-101 (2 mg/kg)
MT-101 (5 mg/kg)
MT-101 (10 mg/kg)
Sham

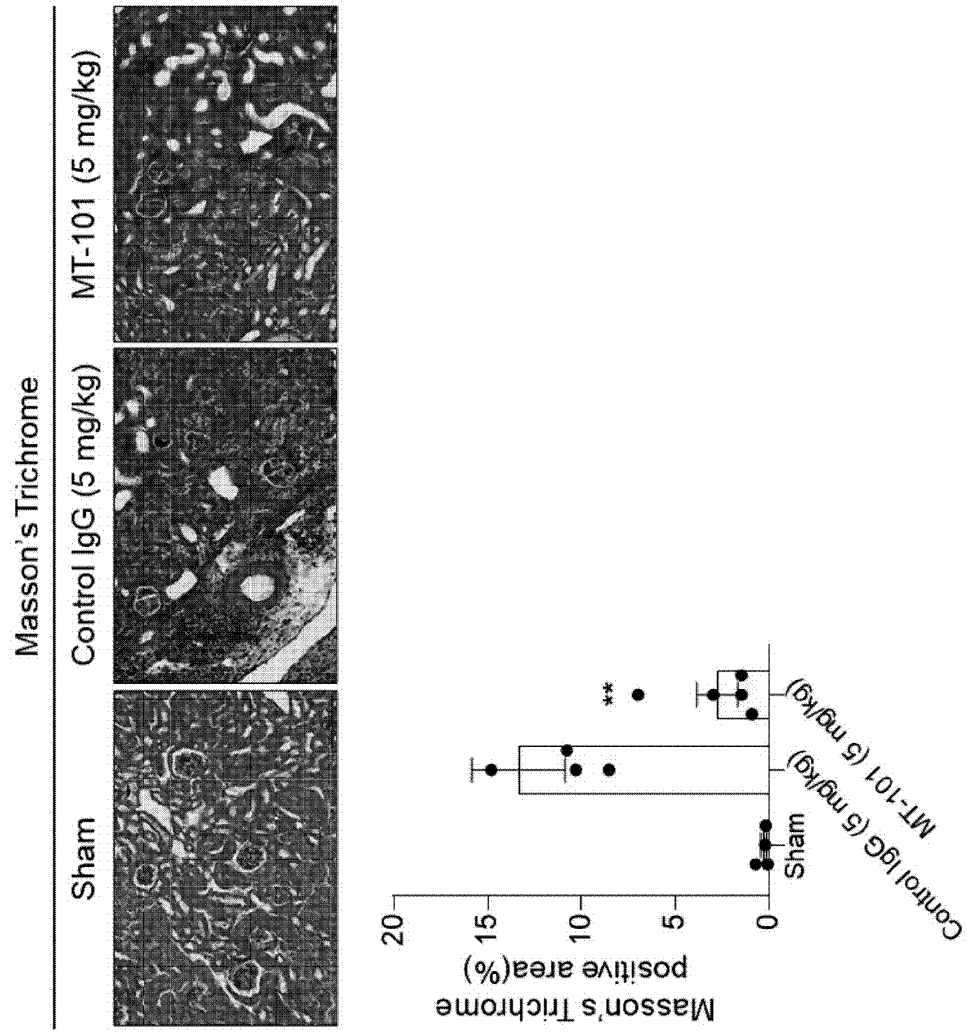
[도20a]


Unilateral Ureteral Obstruction
(UUO)
0
2
4
6
7
(Day)
Sacrifice
MT-101 injection (IV)

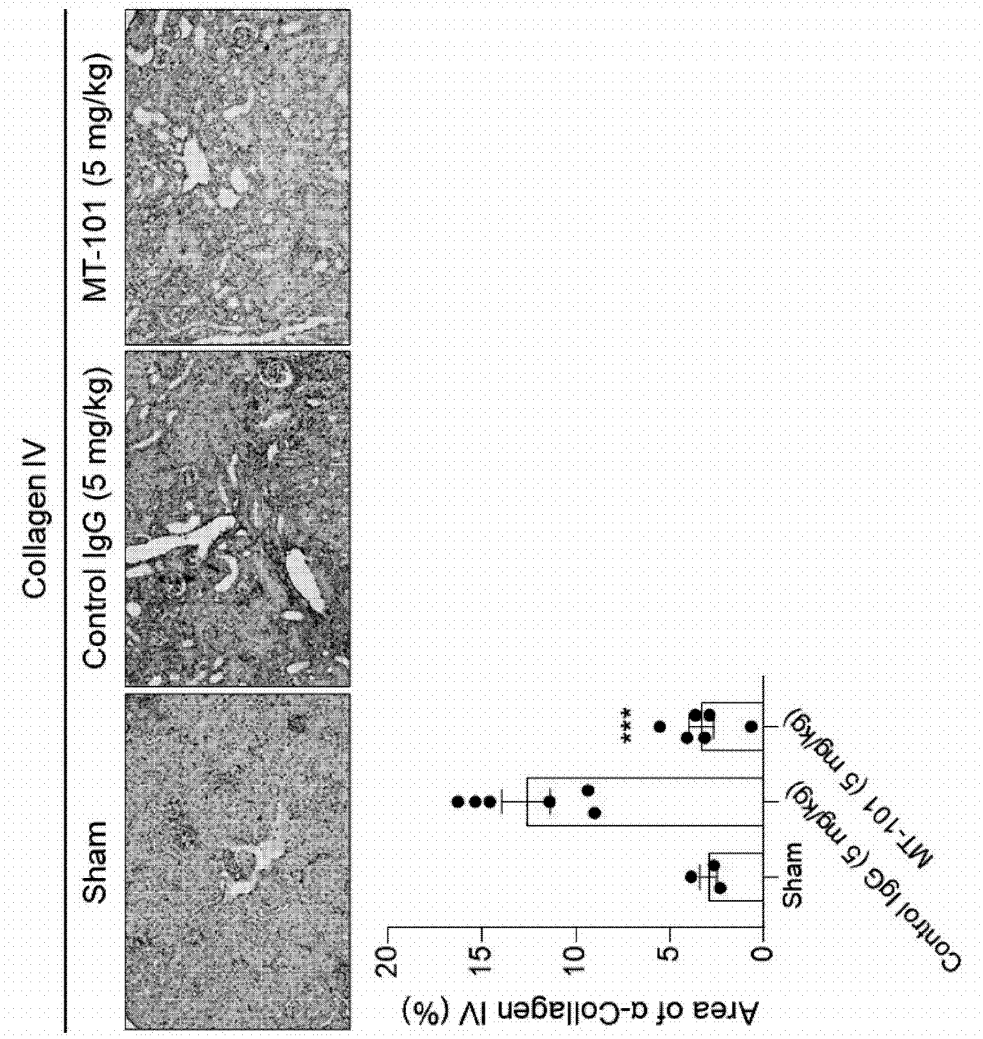
[도20b]


H&E
Sham
Control IgG (5 mg/kg)
MT-101 (5 mg/kg)
Tubulo-Interstitial Injury score
0
1
2
3
4
5
***
Sham
Control IgG (5 mg/kg)
MT-101 (5 mg/kg)

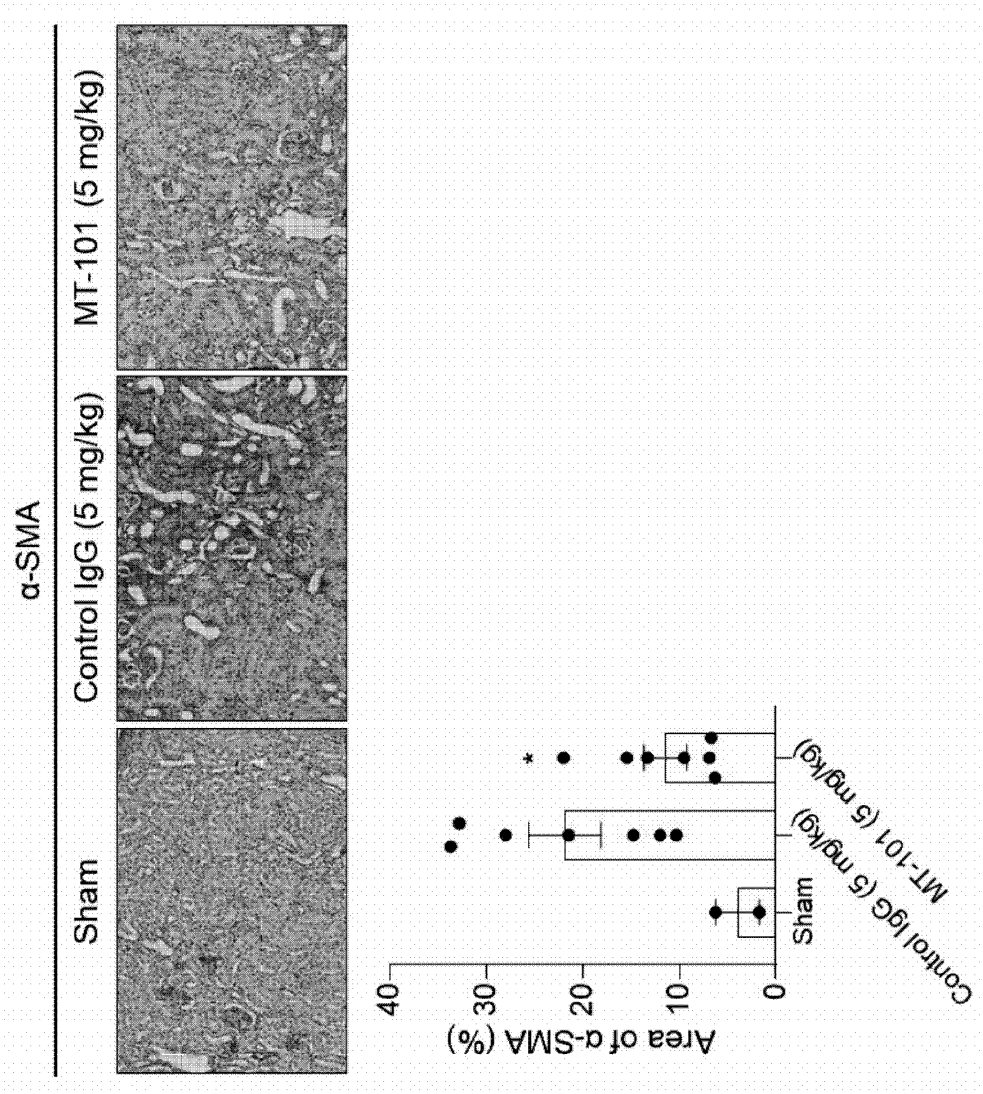
[도20c]


Masson's Trichrome
Sham
Control IgG (5 mg/kg)
MT-101 (5 mg/kg)
Masson's Trichrome positive area(%)
20
15
10
5
0
**
Sham
Control IgG (5 mg/kg)
MT-101 (5 mg/kg)

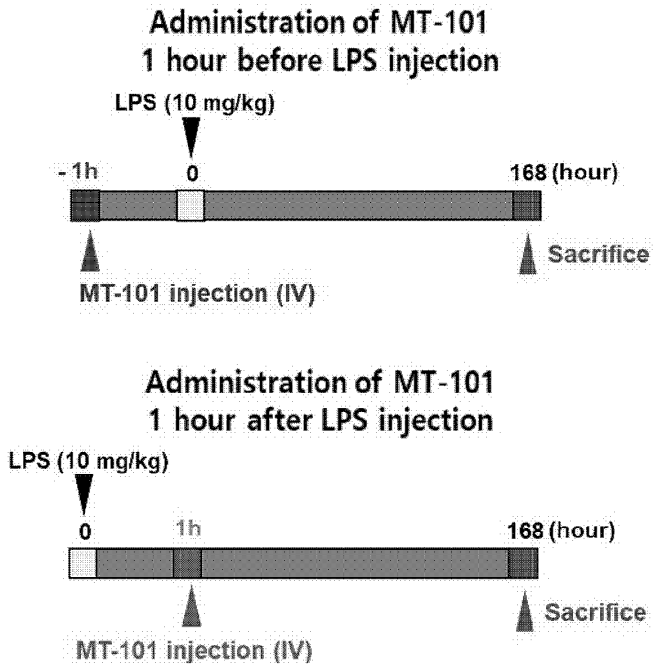
[도20d]


Collagen IV
Sham
Control IgG (5 mg/kg)
MT-101 (5 mg/kg)
Area of α-Collagen IV (%)
20
15
10
5
0
***
Sham
Control IgG (5 mg/kg)
MT-101 (5 mg/kg)

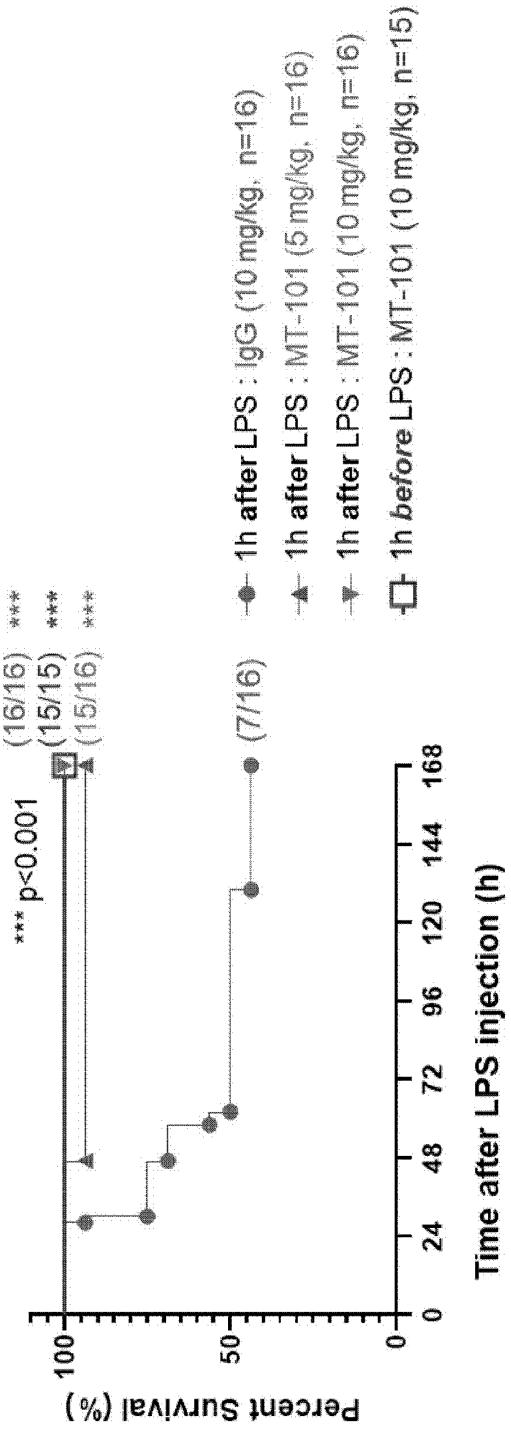
[도20e]


α-SMA
Sham
Control IgG (5 mg/kg)
MT-101 (5 mg/kg)
Area of α-SMA (%)
0
10
20
30
40
Sham
Control IgG (5 mg/kg)
MT-101 (5 mg/kg)

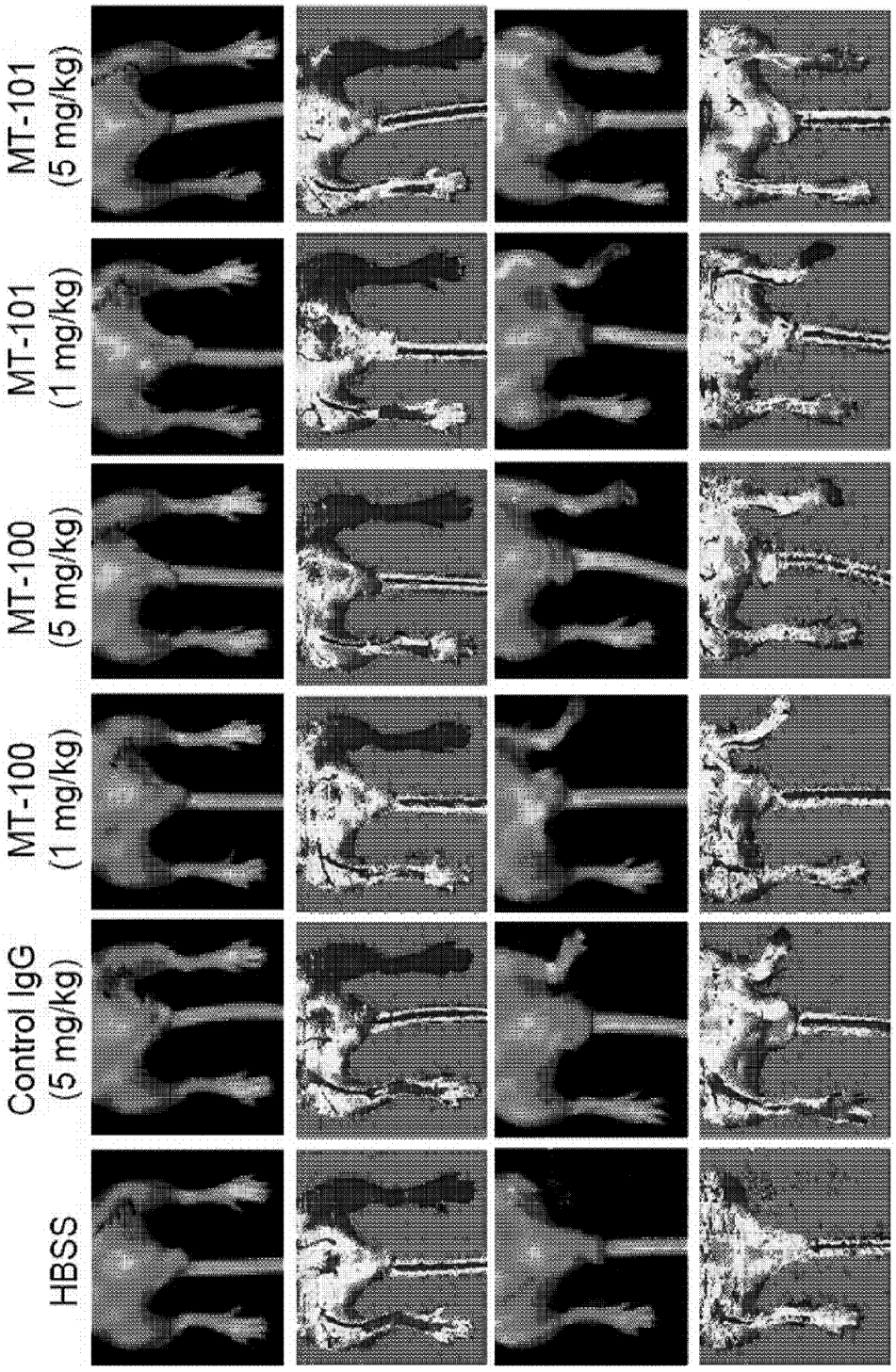
[도21a]


Administration of MT-101
1 hour before LPS injection
LPS (10 mg/kg)
-1h
0
168 (hour)
Sacrifice
MT-101 injection (IV)
Administration of MT-101
1 hour after LPS injection
LPS (10 mg/kg)
0
1h
168 (hour)
Sacrifice
MT-101 injection (IV)

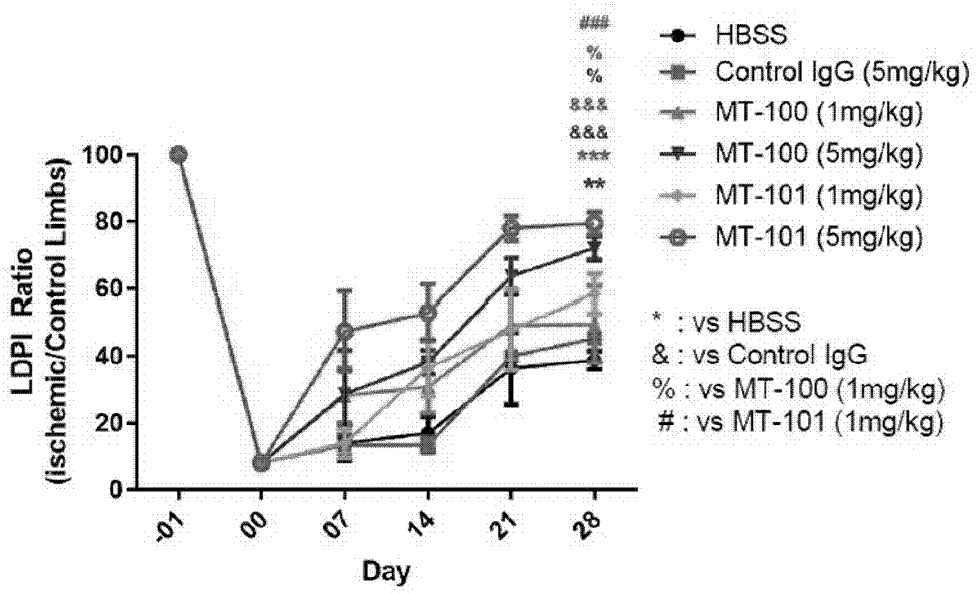
[도21b]


Percent Survival (%)
100
50
0
0
24
48
72
96
120
144
168
Time after LPS injection (h)
*** p<0.001
(16/16)
(15/15)
(15/16)
***
***
***
(7/16)
1h after LPS : IgG (10 mg/kg, n=16)
1h after LPS : MT-101 (5 mg/kg, n=16)
1h after LPS : MT-101 (10 mg/kg, n=16)
1h before LPS : MT-101 (10 mg/kg, n=15)

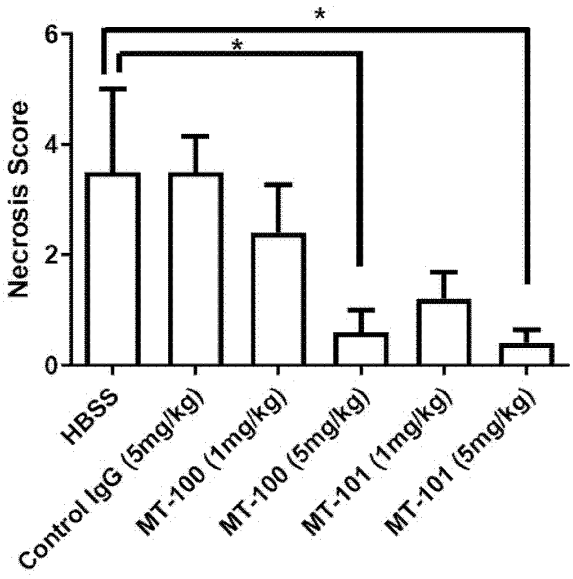
[도22a]


MT-101 (5 mg/kg)
MT-101 (1 mg/kg)
MT-100 (5 mg/kg)
MT-100 (1 mg/kg)
Control IgG (5 mg/kg)
HBSS

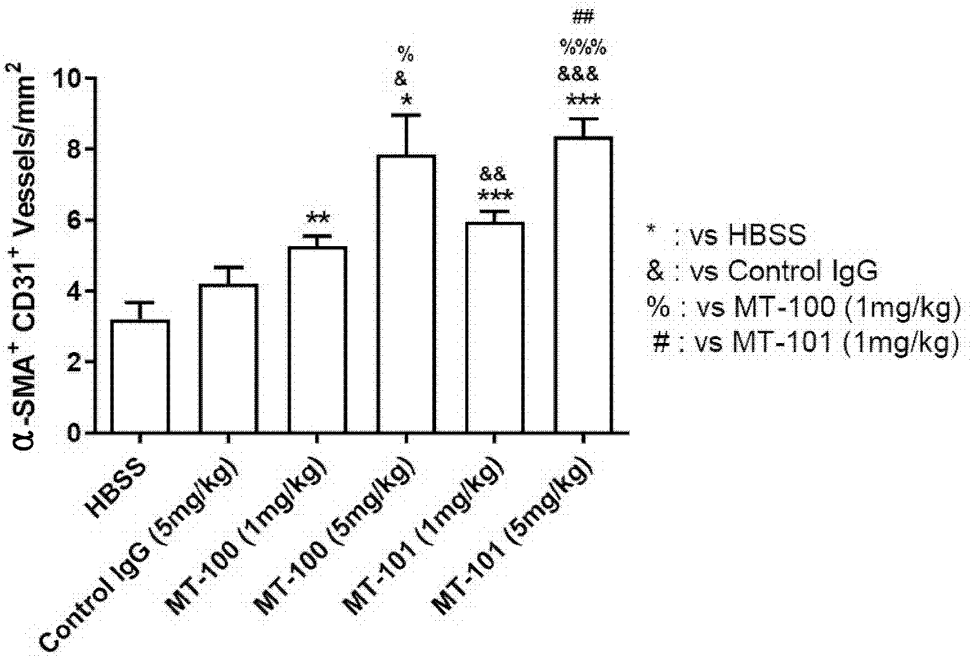
[도22b]


LDPI Ratio
(Ischemic/Control Limbs)
100
80
60
40
20
0
-01
00
07
14
21
28
Day
###
%
%
&&&
&&&
***
**
HBSS
Control IgG (5mg/kg)
MT-100 (1mg/kg)
MT-100 (5mg/kg)
MT-101 (1mg/kg)
MT-101 (5mg/kg)
* : vs HBSS
& : vs Control IgG
% : vs MT-100 (1mg/kg)
# : vs MT-101 (1mg/kg)

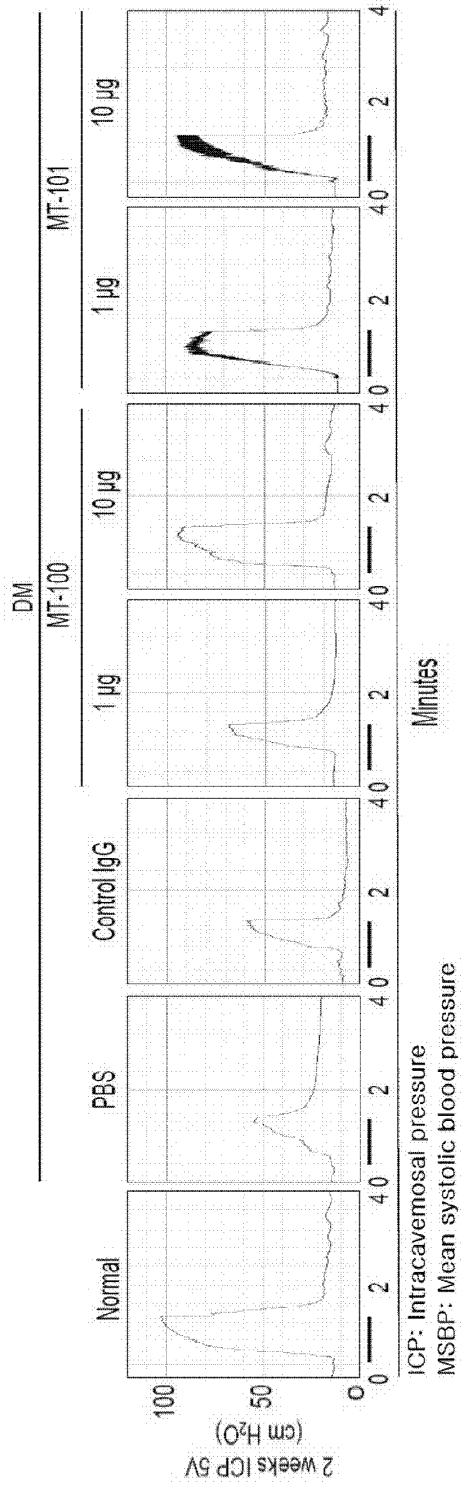
[도22c]


Necrosis Score
6
4
2
0
*
*
HBSS
Control IgG (5mg/kg)
MT-100 (1mg/kg)
MT-100 (5mg/kg)
MT-101 (1mg/kg)
MT-101 (5mg/kg)

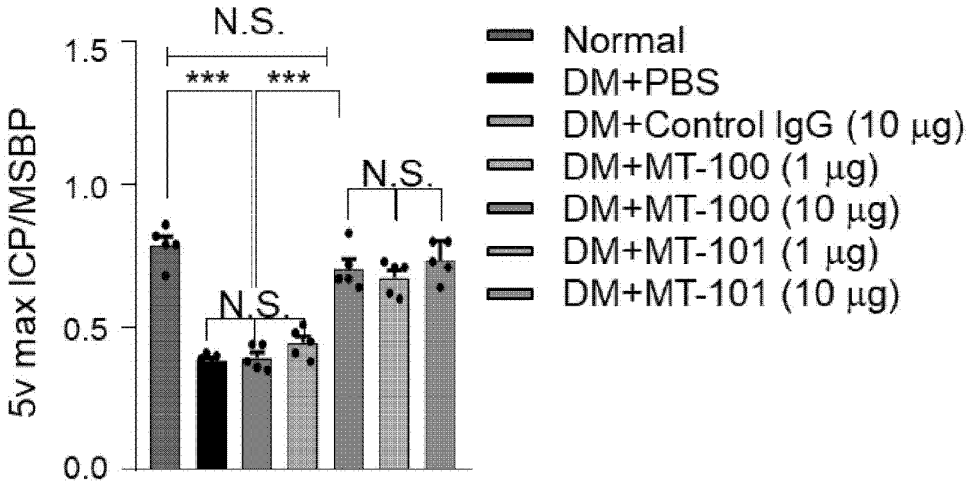
[도22d]


α-SMA+ CD31+ Vessels/mm2
10
8
6
4
2
0
**
%
&
*
&&
***
##
%%%
&&&
***
HBSS
Control IgG (5mg/kg)
MT-100 (1mg/kg)
MT-100 (5mg/kg)
MT-101 (1mg/kg)
MT-101 (5mg/kg)
* : vs HBSS
& : vs Control IgG
% : vs MT-100 (1mg/kg)
# : vs MT-101 (1mg/kg)

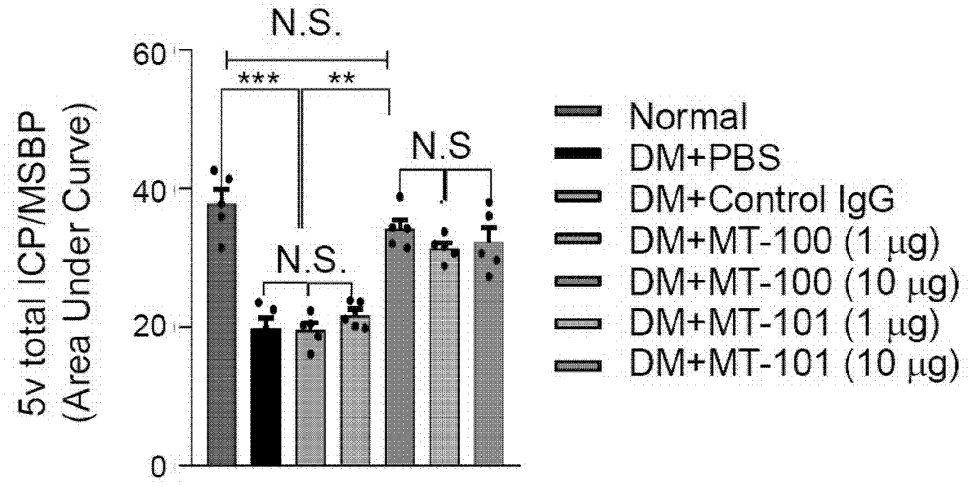
[도23a]


DIM
MT-100
MT-101
Normal
PBS
Control IgG
1 μg
10 μg
1 μg
10 μg
2 weeks ICP 5V
(cm H2O)
100
50
0
0
2
4
Minutes
ICP: Intracavemosal pressure
MSBP: Mean systolic blood pressure

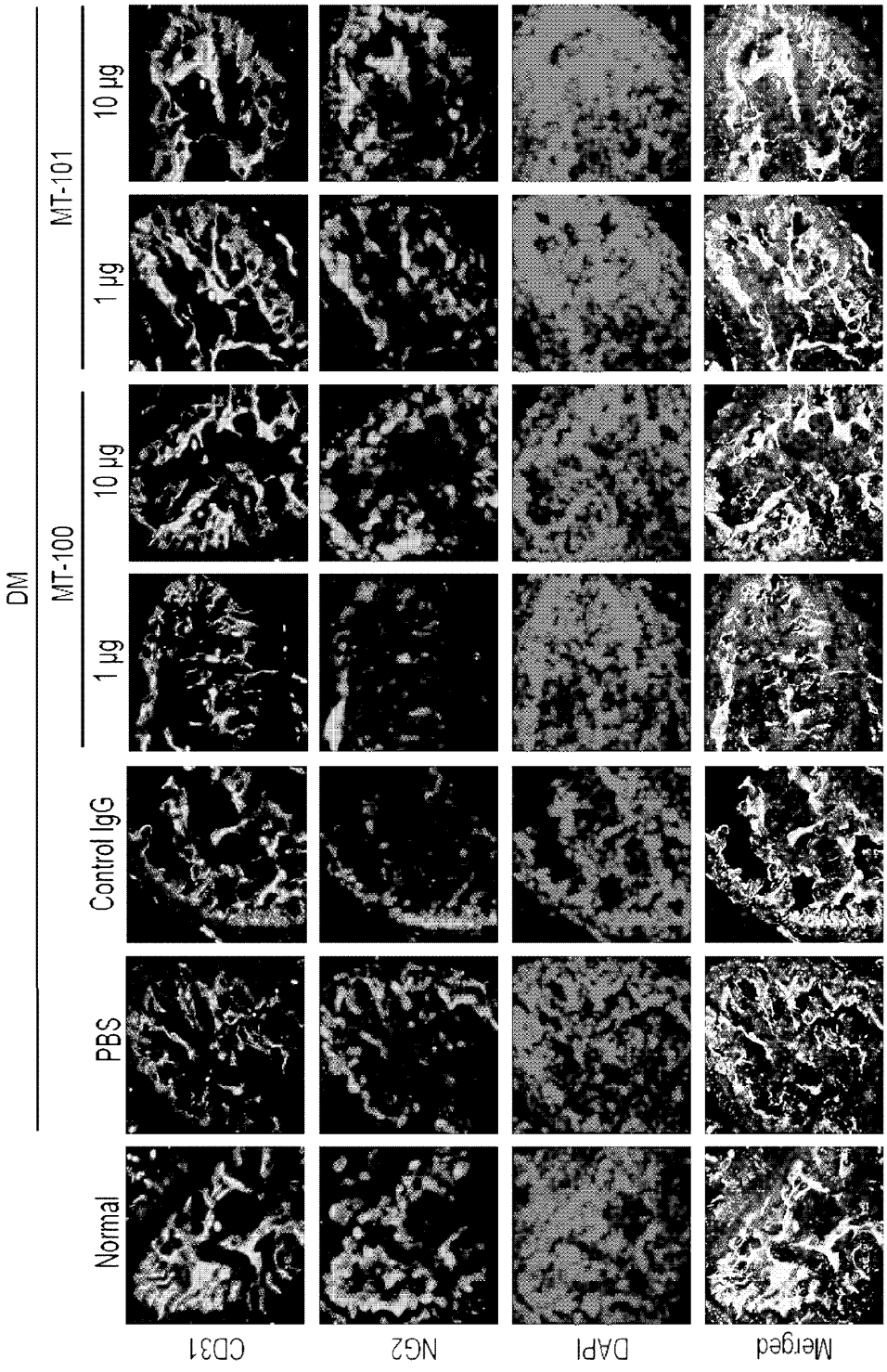
[도23b]


N.S.
***
***
N.S.
N.S.
1.5
1.0
0.5
0.0
5v max ICP/MSBP
Normal
DM+PBS
DM+Control IgG (10 μg)
DM+MT-100 (1 μg)
DM+MT-100 (10 μg)
DM+MT-101 (1 μg)
DM+MT-101 (10 μg)

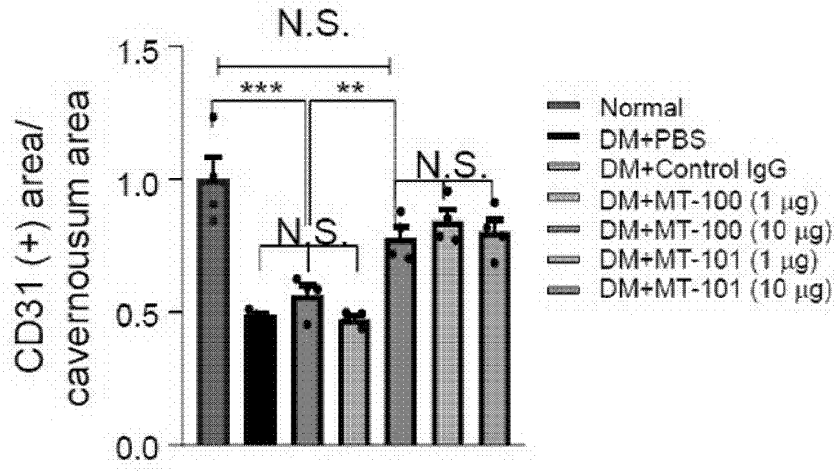
[도23c]


N.S.
***
**
N.S
N.S.
60
40
20
0
5v total ICP/MSBP
(Area Under Curve)
Normal
DM+PBS
DM+Control IgG
DM+MT-100 (1 μg)
DM+MT-100 (10 μg)
DM+MT-101 (1 μg)
DM+MT-101 (10 μg)

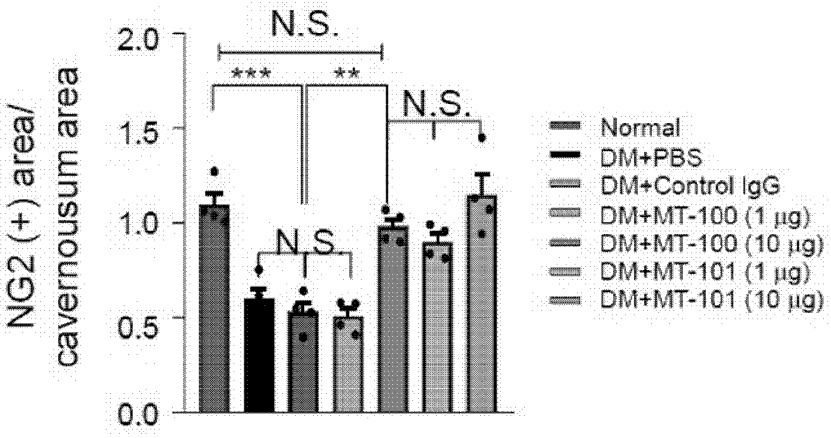
[도24a]


DM
MT-101
10 μg
1 μg
MT-100
10 μg
1 μg
Control IgG
PBS
Normal
CD31
NG2
DAPI
Merged

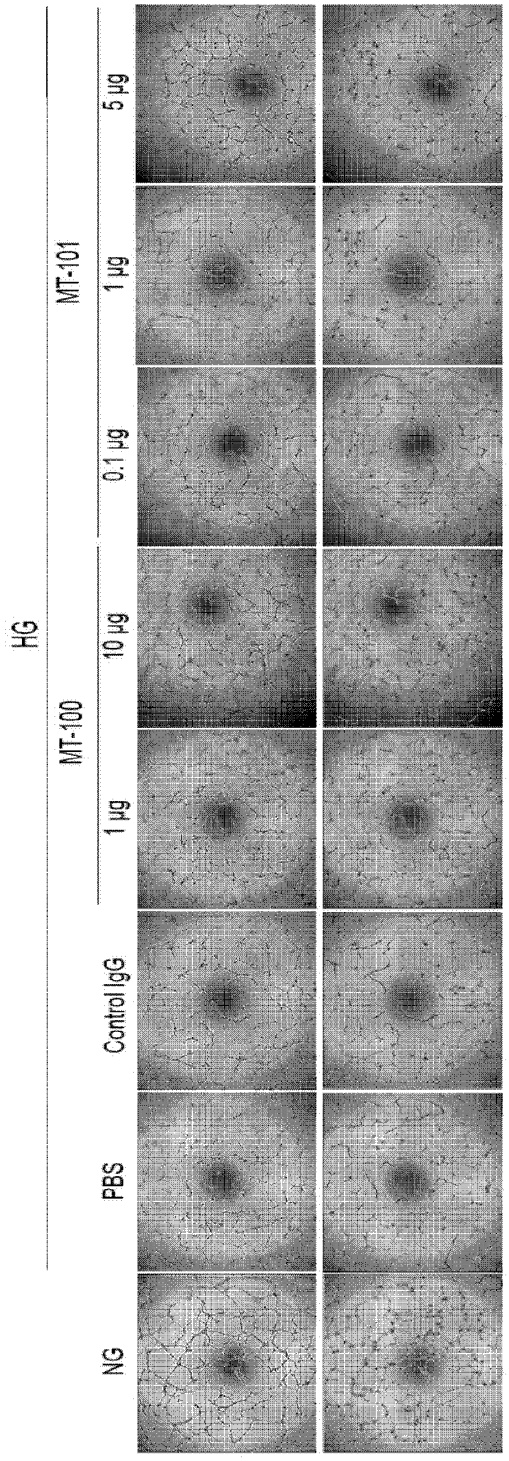
[도24b]


N.S.
***
**
N.S.
N.S.
1.5
1.0
0.5
0.0
CD31 (+) area/
cavernousum area
Normal
DM+PBS
DM+Control IgG
DM+MT-100 (1 μg)
DM+MT-100 (10 μg)
DM+MT-101 (1 μg)
DM+MT-101 (10 μg)

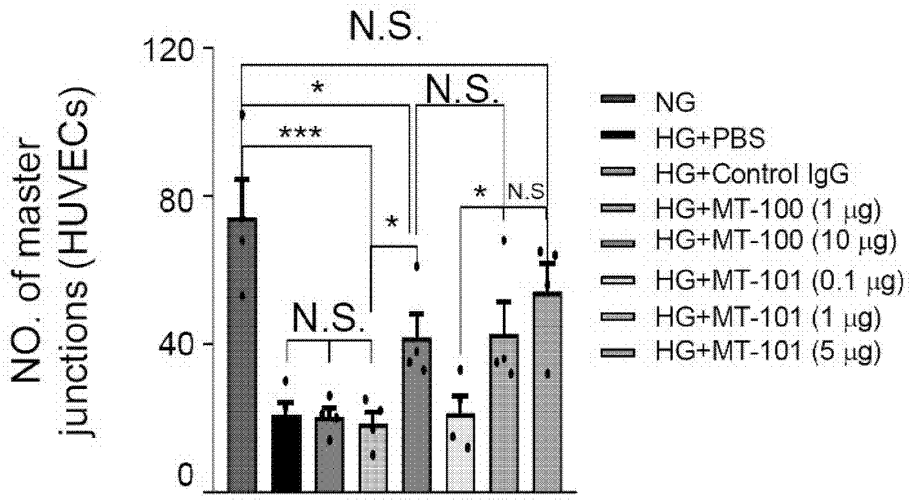
[도24c]


N.S.
***
**
N.S.
N.S.
2.0
1.5
1.0
0.5
0.0
NG2 (+) area/
cavernousum area
Normal
DM+PBS
DM+Control IgG
DM+MT-100 (1 μg)
DM+MT-100 (10 μg)
DM+MT-101 (1 μg)
DM+MT-101 (10 μg)

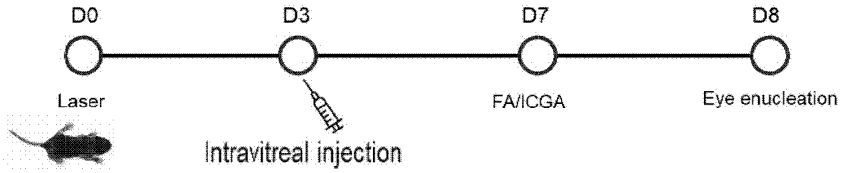
[도25a]



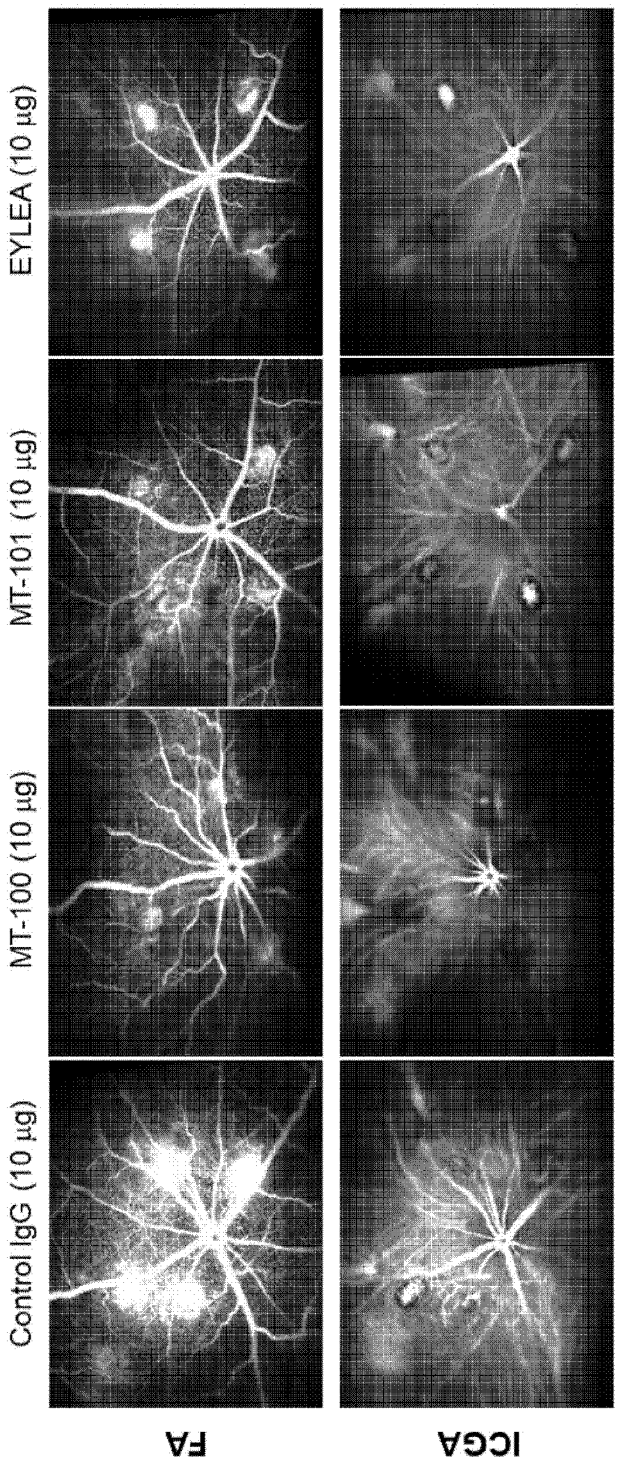
[도25b]


N.S.
120
80
40
0
NO. of master junctions (HUVECs)
*
N.S.
***
*
N.S.
*
N.S
NG
HG+PBS
HG+Control IgG
HG+MT-100 (1 μg)
HG+MT-100 (10 μg)
HG+MT-101 (0.1 μg)
HG+MT-101 (1 μg)
HG+MT-101 (5 μg)

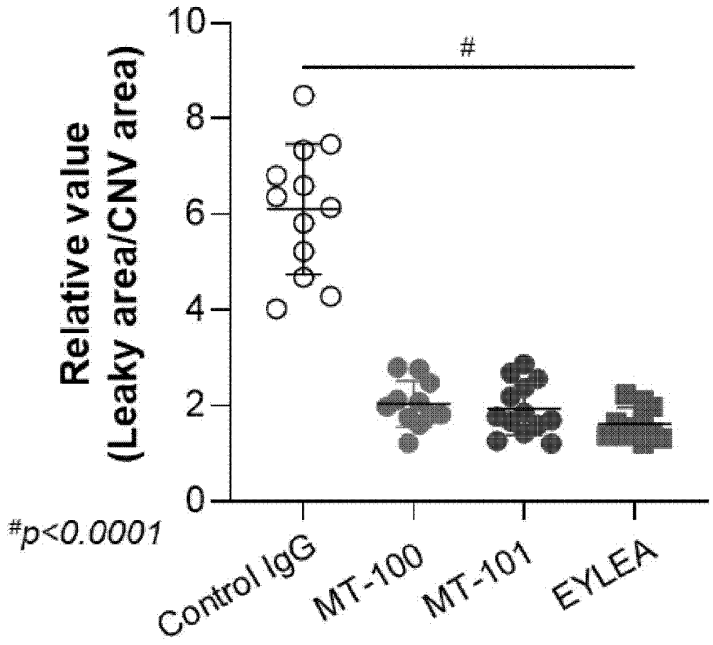
[도26a]


D0
D3
D7
D8
Laser
Intravitreal injection
FA/ICGA
Eye enucleation

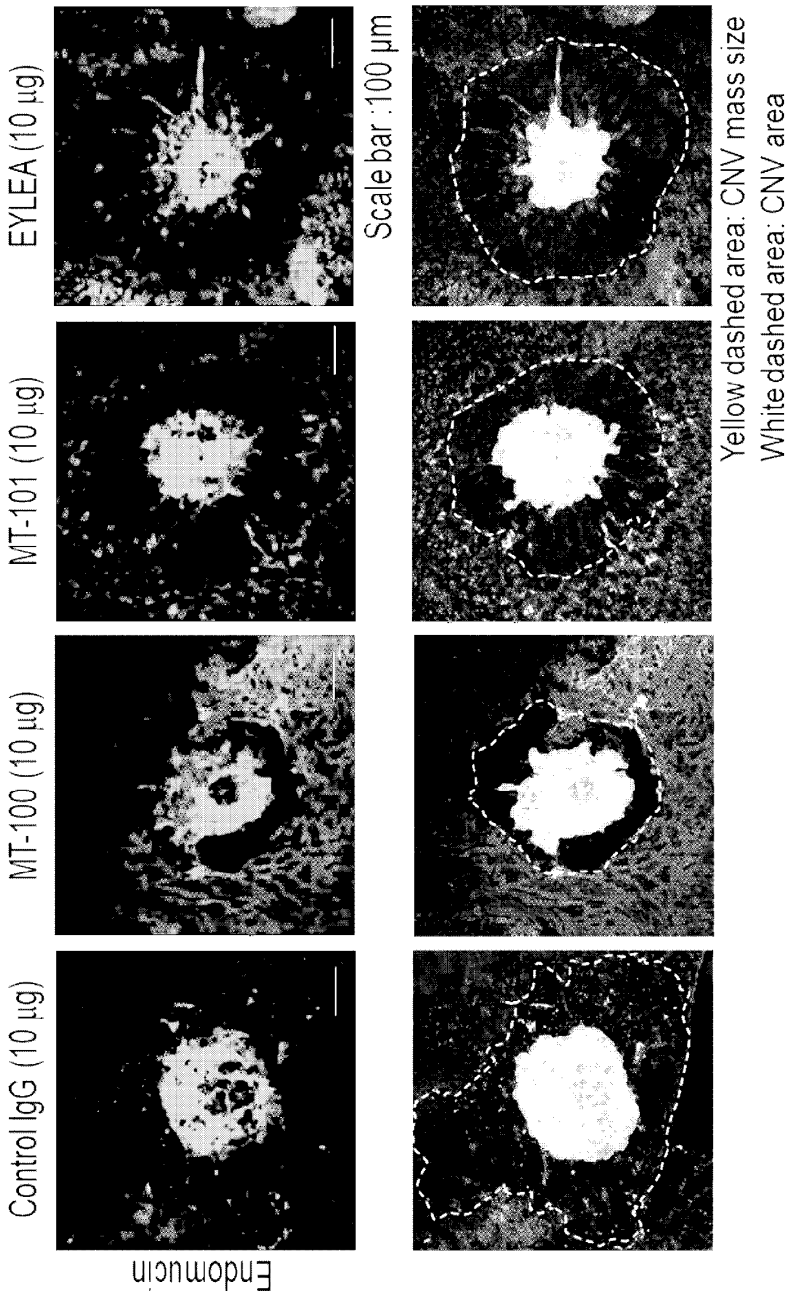
[도26b]



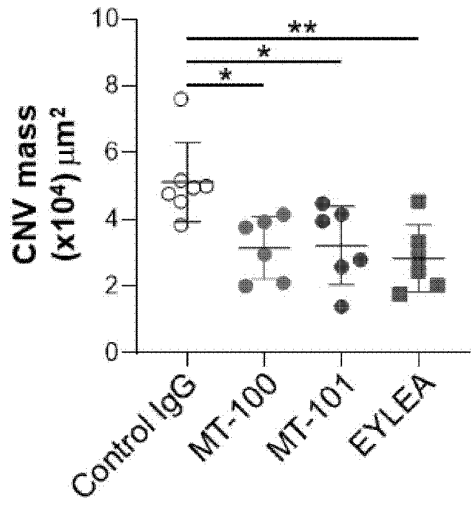
[도26c]


Relative value
(Leaky area/CNV area)
10
8
6
4
2
0
#
#p<0.0001
Control IgG
MT-100
MT-101
EYLEA

[도27a]

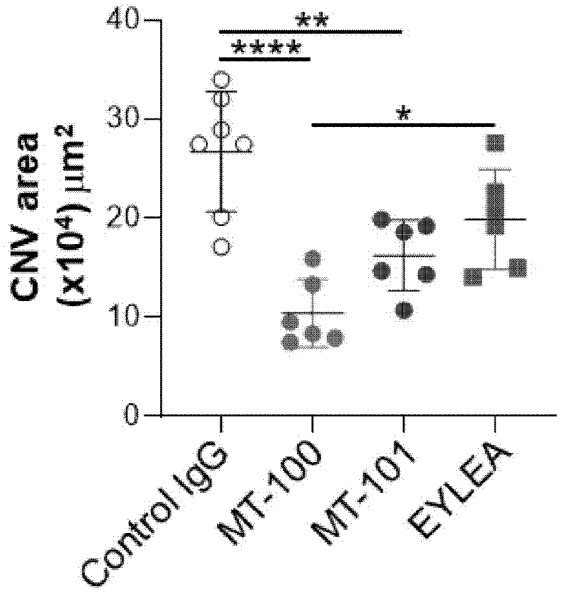

EYLEA (10 μg)
MT-101 (10 μg)
MT-100 (10 μg)
Control IgG (10 μg)
Endomucin
Scale bar :100 μm
Yellow dashed area: CNV mass size
White dashed area: CNV area

[도27b]



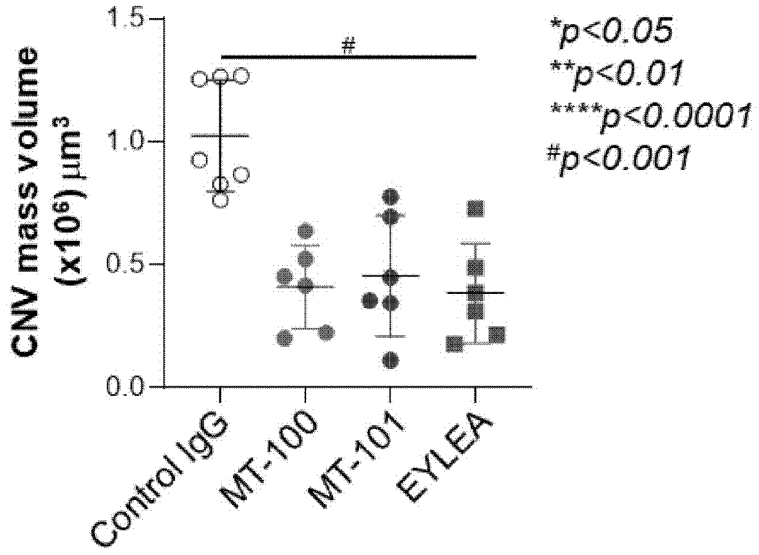
*p<0.05
**p<0.01
****p<0.0001
#p<0.001

[도27c]

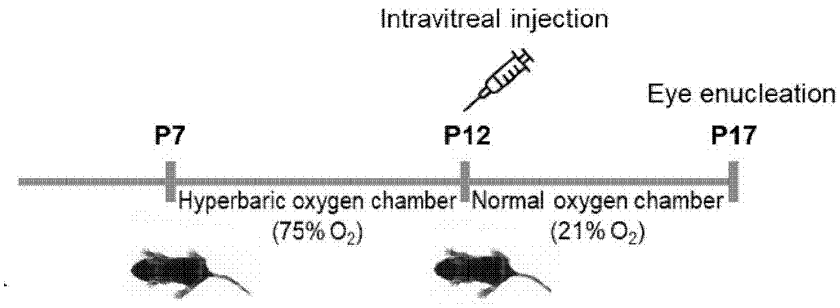


*p<0.05
**p<0.01
****p<0.0001
#p<0.001

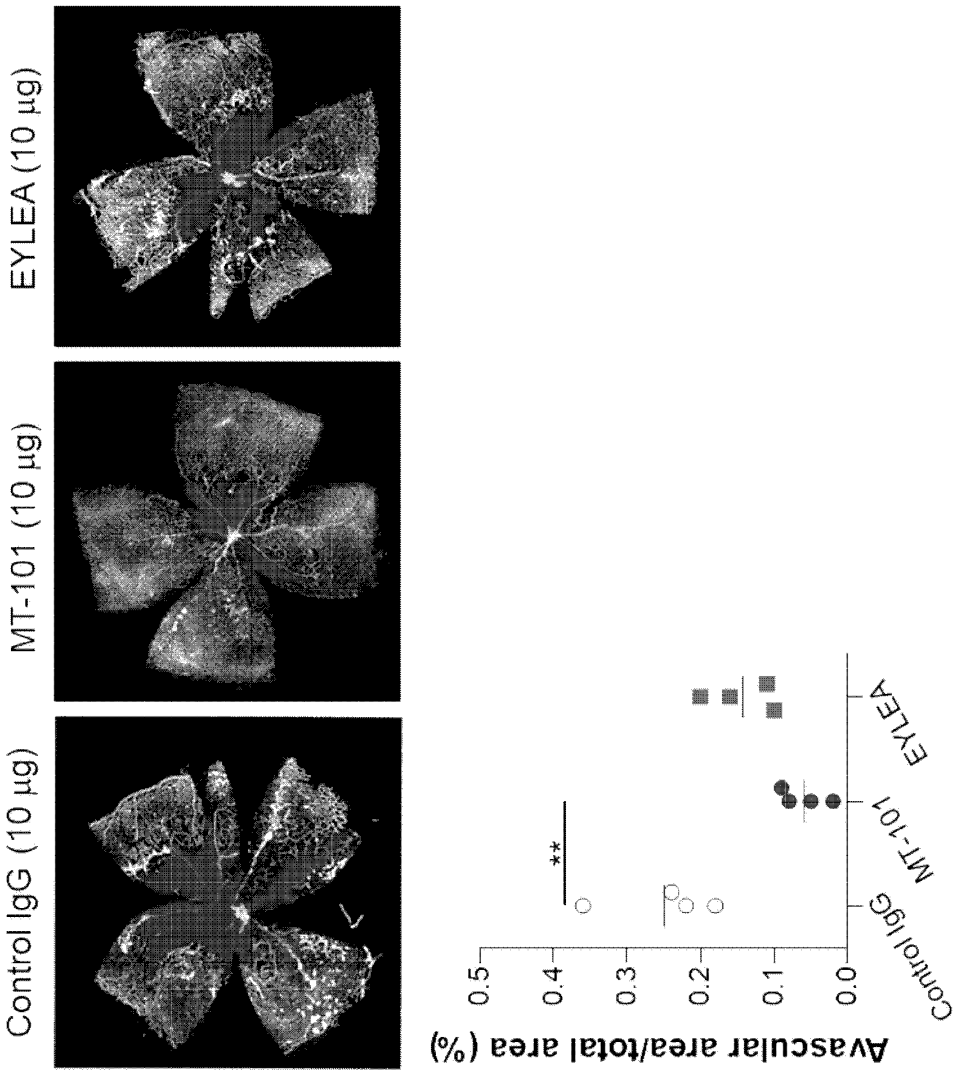
[도27d]


CNV mass volume
(x10⁶) μm³
1.5
1.0
0.5
0.0
#
*p<0.05
**p<0.01
****p<0.0001
#p<0.001
Control IgG
MT-100
MT-101
EYLEA

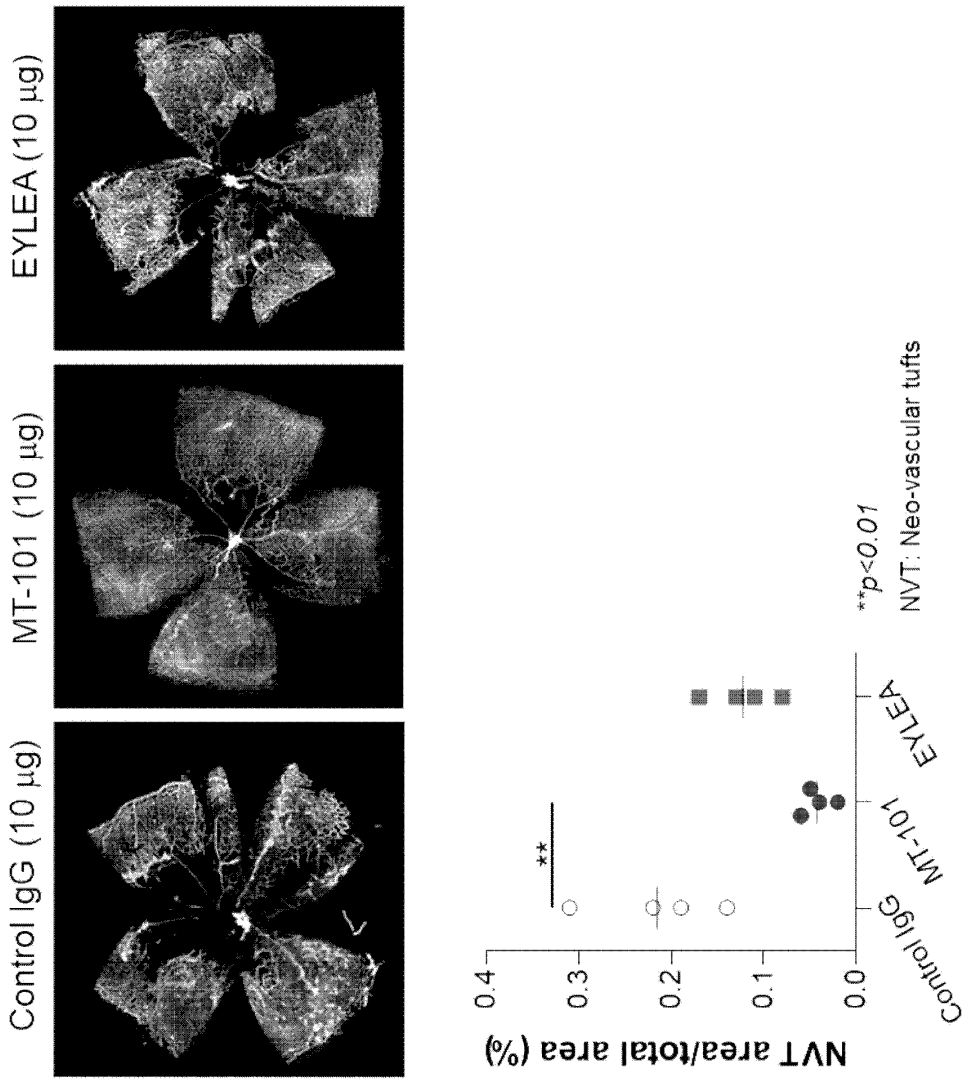
[도28a]


Intravitreal injection
Eye enucleation
P7
P12
P17
Hyperbaric oxygen chamber
(75% O2)
Normal oxygen chamber
(21% O2)

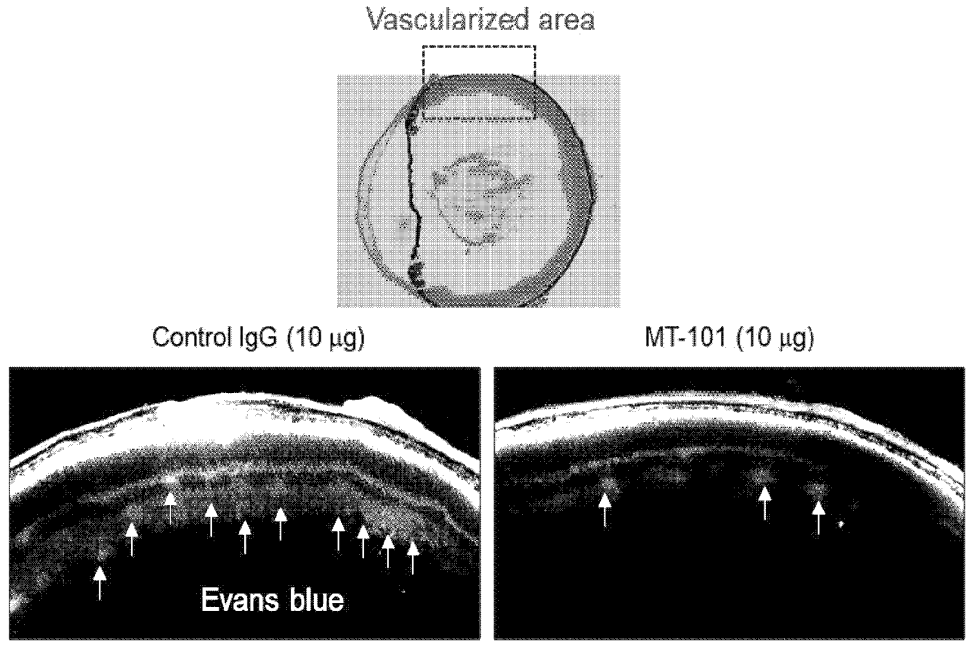
[도28b]


Control IgG (10 μg)
MT-101 (10 μg)
EYLEA (10 μg)
Avascular area/total area (%)
0.0
0.1
0.2
0.3
0.4
0.5
**
Control IgG
MT-101
EYLEA

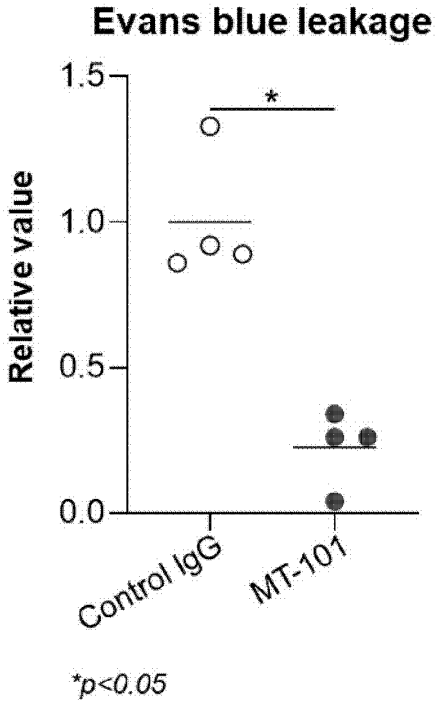
[도28c]


Control IgG (10 μg)
MT-101 (10 μg)
EYLEA (10 μg)
NVT area/total area (%)
0.0
0.1
0.2
0.3
0.4
**
Control IgG
MT-101
EYLEA
**p<0.01
NVT: Neo-vascular tufts

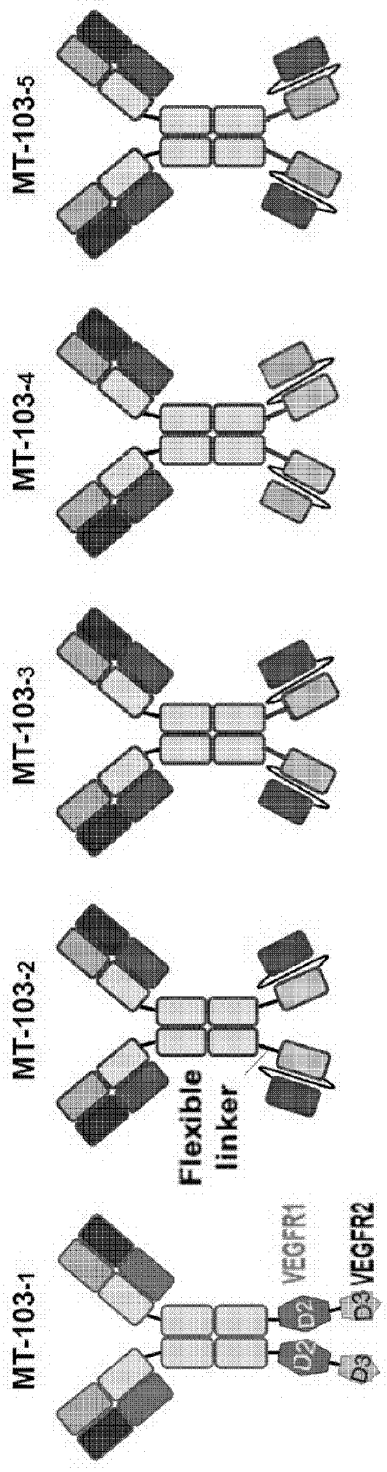
[도29a]


Vascularized area
Control IgG (10 μg)
MT-101 (10 μg)
Evans blue

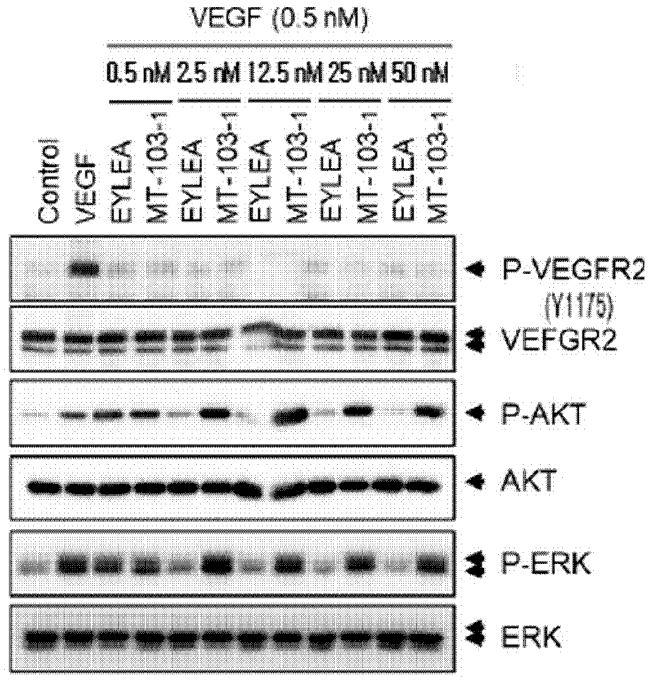
[도29b]


Evans blue leakage
Relative value
1.5
1.0
0.5
0.0
*
Control IgG
MT-101
*p<0.05

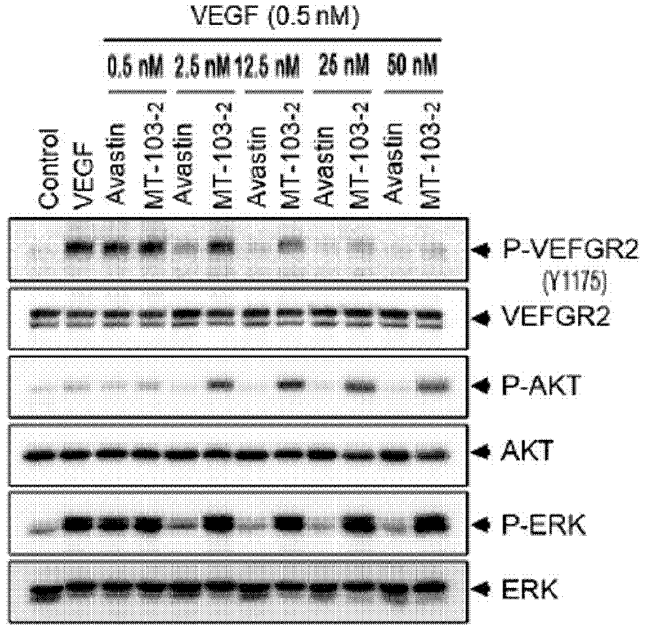
[도30]


MT-103-5
MT-103-4
MT-103-3
MT-103-2
Flexible linker
MT-103-1
VEGFR1
VEGFR2
D2
D3
D2
D3

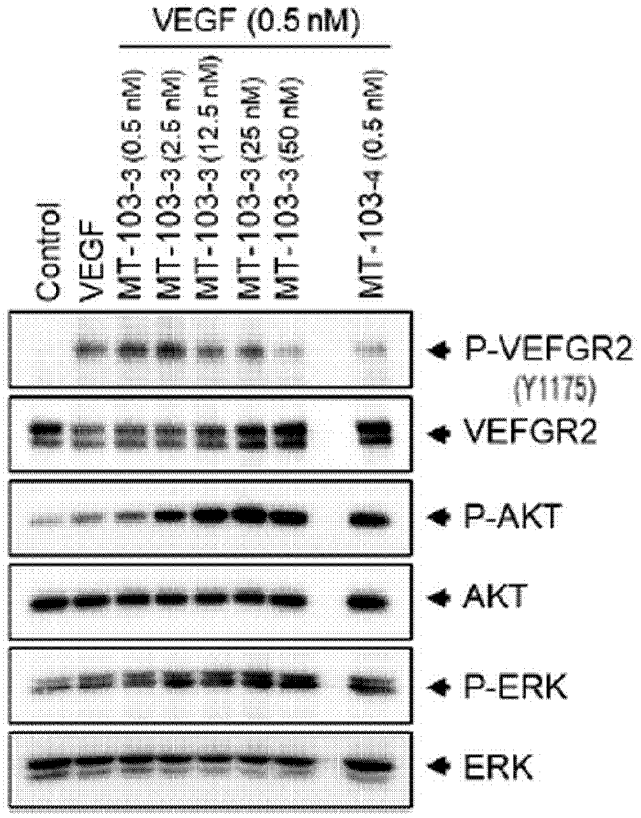
[도31a]


VEGF (0.5 nM)
0.5 nM
2.5 nM
12.5 nM
25 nM
50 nM
Control
VEGF
EYLEA
MT-103-1
P-VEGFR2 (Y1175)
VEFGR2
P-AKT
AKT
P-ERK
ERK

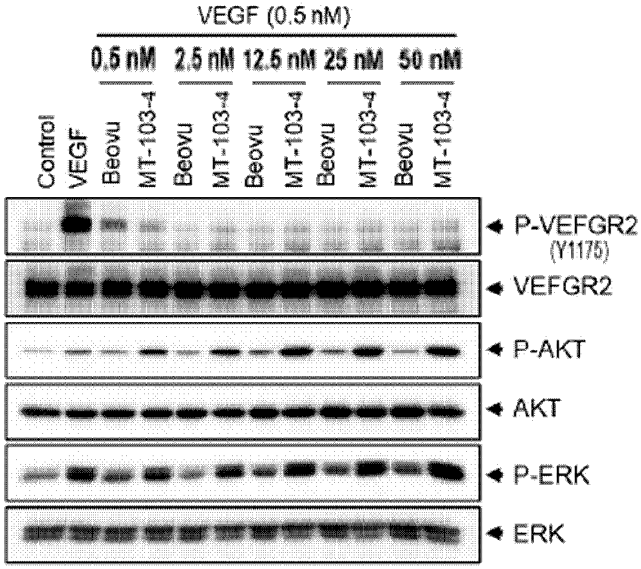
[도31b]


VEGF (0.5 nM)
0.5 nM
2.5 nM
12.5 nM
26 nM
50 nM
Control
VEGF
Avastin
MT-103-2
P-VEFGR2 (Y1175)
VEFGR2
P-AKT
AKT
P-ERK
ERK

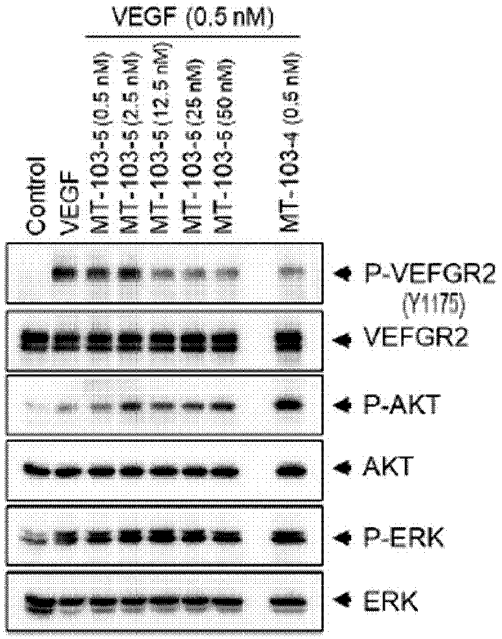
[도31c]


VEGF (0.5 nM)
Control
VEGF
MT-103-3 (0.5 nM)
MT-103-3 (2.5 nM)
MT-103-3 (12.5 nM)
MT-103-3 (25 nM)
MT-103-3 (50 nM)
MT-103-4 (0.5 nM)
P-VEFGR2 (Y1175)
VEFGR2
P-AKT
AKT
P-ERK
ERK

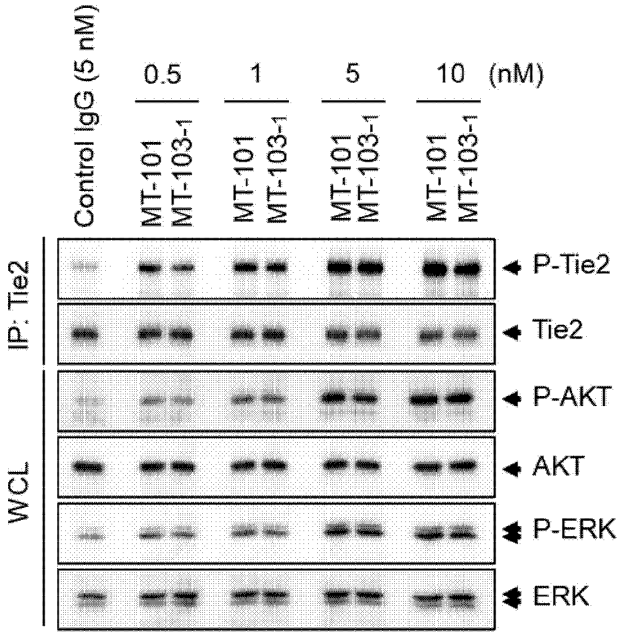
[도31d]


VEGF (0.5 nM)
0.5 nM
2.5 nM
12.5 nM
25 nM
50 nM
Control
VEGF
Beovu
MT-103-4
P-VEFGR2 (Y1175)
VEFGR2
P-AKT
AKT
P-ERK
ERK

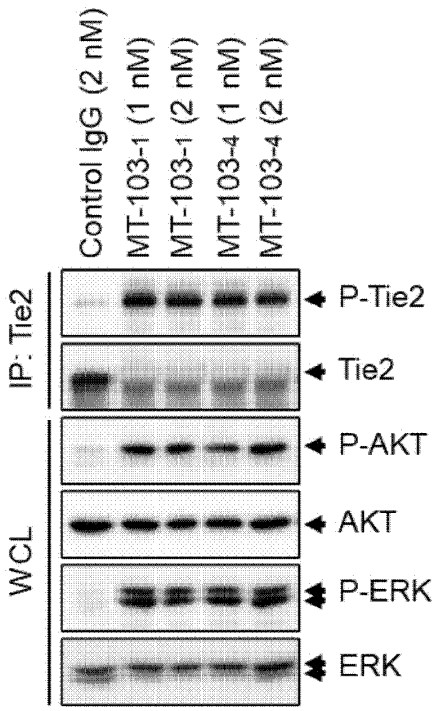
[도31e]


VEGF (0.5 nM)
Control
VEGF
MT-103-5 (0.5 nM)
MT-103-5 (2.5 nM)
MT-103-5 (12.5 nM)
MT-103-5 (25 nM)
MT-103-5 (50 nM)
MT-103-4 (0.5 nM)
P-VEFGR2 (Y1175)
VEFGR2
P-AKT
AKT
P-ERK
ERK

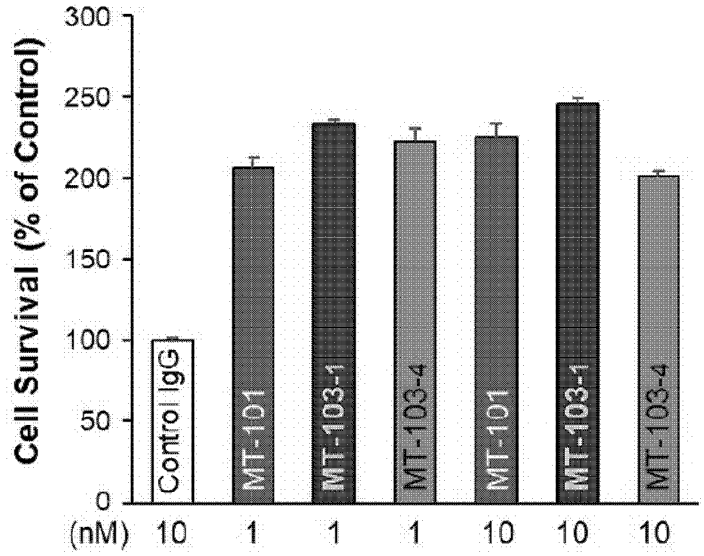
[도32a]


Control IgG (5 nM)
0.5
1
5
10
(nM)
MT-101
MT-103-1
IP: Tie2
WCL
P-Tie2
Tie2
P-AKT
AKT
P-ERK
ERK

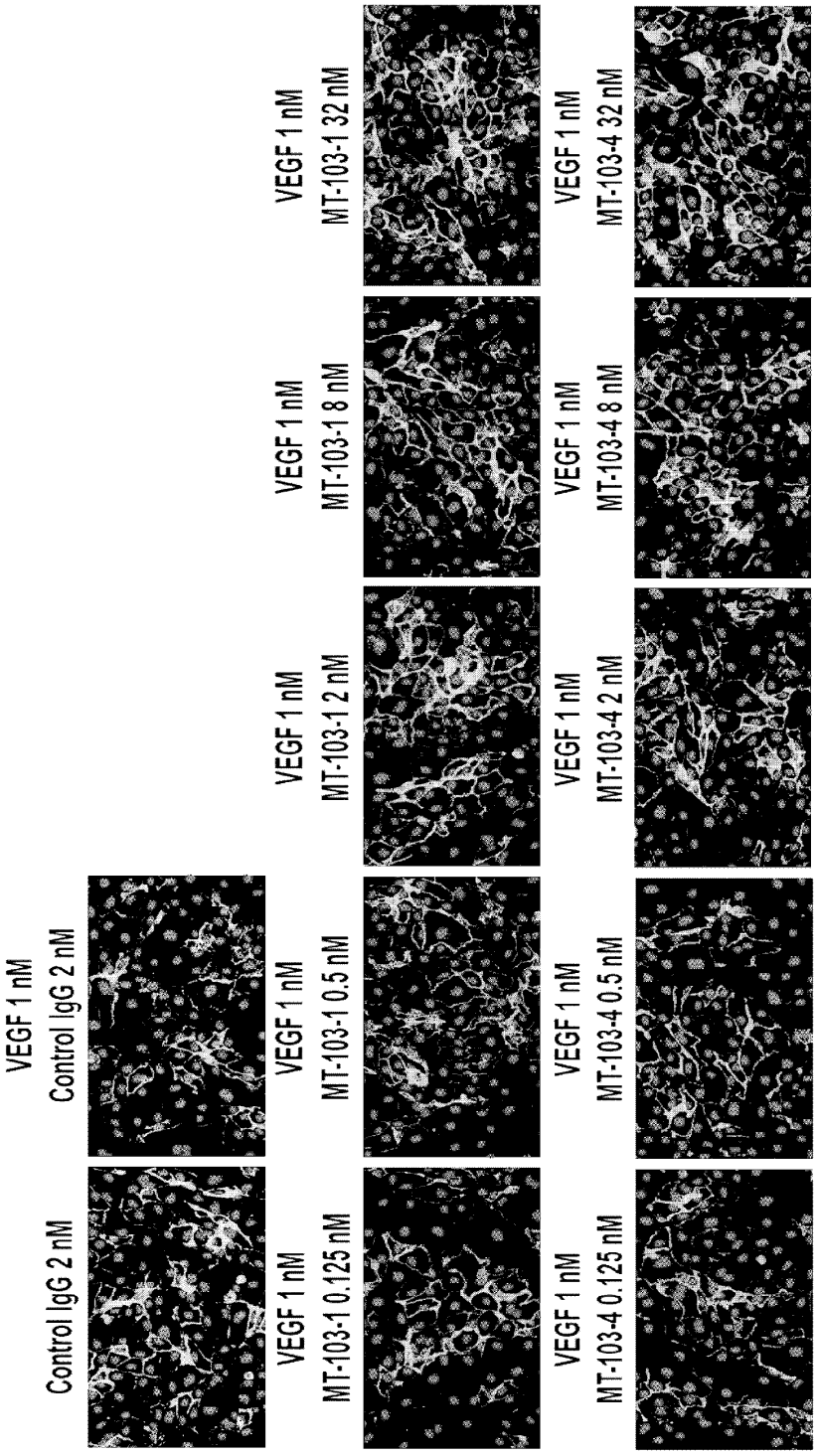
[도32b]


Control IgG (2 nM)
MT-103-1 (1 nM)
MT-103-1 (2 nM)
MT-103-4 (1 nM)
MT-103-4 (2 nM)
IP: Tie2
WCL
P-Tie2
Tie2
P-AKT
AKT
P-ERK
ERK

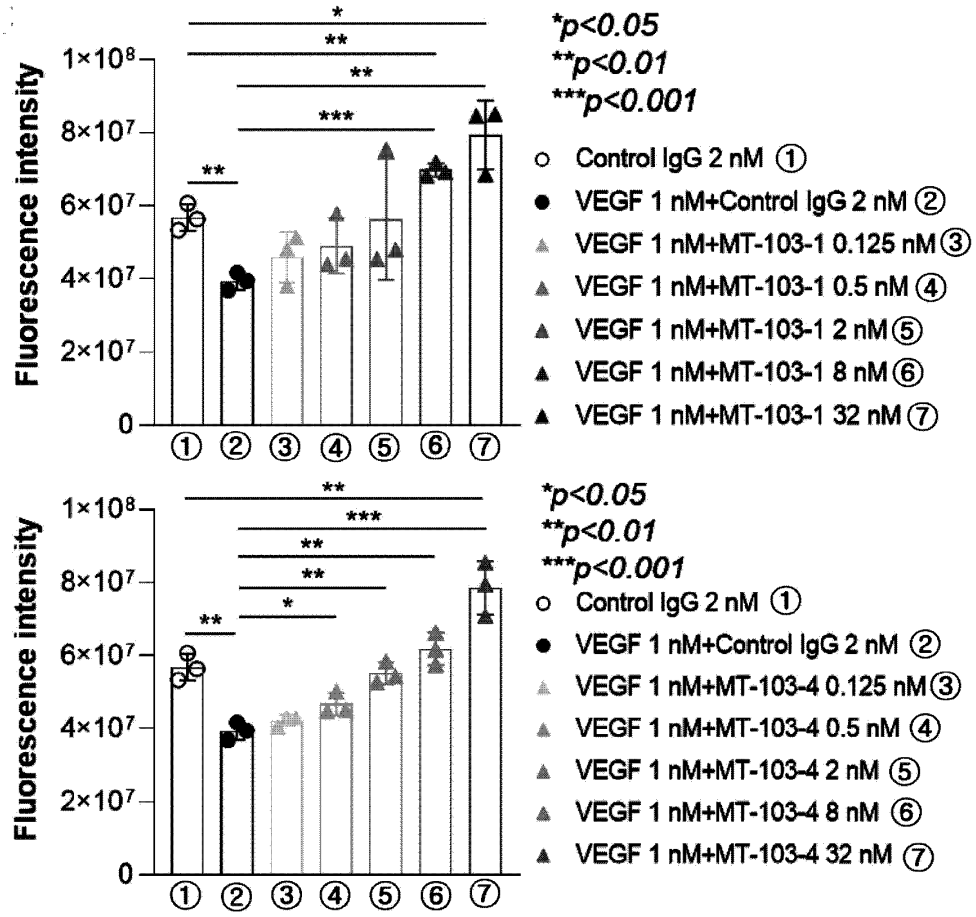
[도33a]


Cell Survival (% of Control)
300
250
200
150
100
50
0
Control IgG
MT-101
MT-103-1
MT-103-4
MT-101
MT-103-1
MT-103-4
(nM) 10 1 1 1 10 10 10

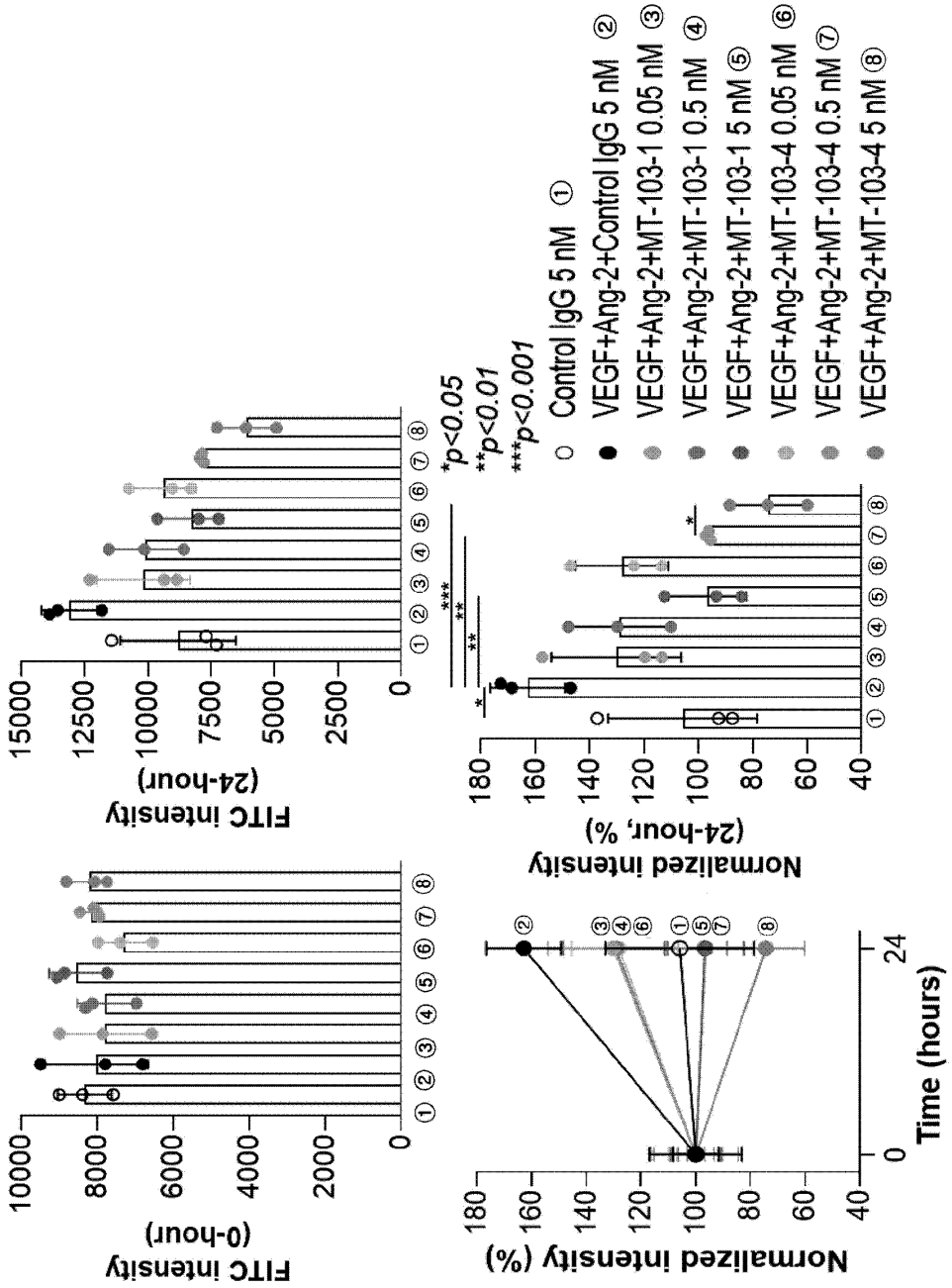
[도33b]


VEGF 1 nM
Control IgG 2 nM
VEGF 1 nM
MT-103-1 0.5 nM
VEGF 1 nM
MT-103-1 2 nM
VEGF 1 nM
MT-103-1 8 nM
VEGF 1 nM
MT-103-1 32 nM
Control IgG 2 nM
VEGF 1 nM
MT-103-1 0.125 nM
VEGF 1 nM
MT-103-4 0.5 nM
VEGF 1 nM
MT-103-4 2 nM
VEGF 1 nM
MT-103-4 8 nM
VEGF 1 nM
MT-103-4 32 nM

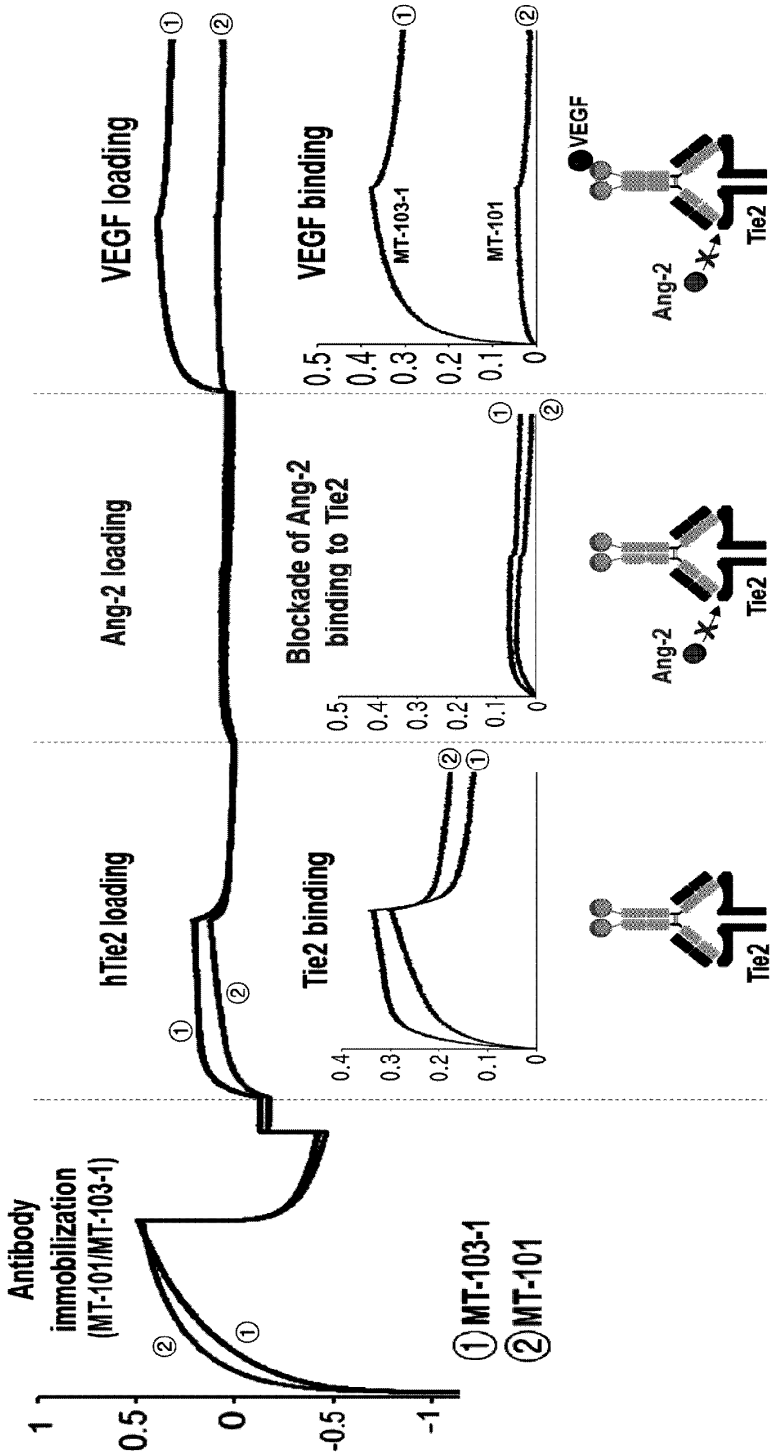
[도33c]


Fluorescence intensity
1×10⁸
8×10⁷
6×10⁷
4×10⁷
2×10⁷
0
①②③④⑤⑥⑦
*p<0.05
**p<0.01
***p<0.001
Control IgG 2 nM ①
VEGF 1 nM+Control IgG 2 nM ②
VEGF 1 nM+MT-103-1 0.125 nM ③
VEGF 1 nM+MT-103-1 0.5 nM ④
VEGF 1 nM+MT-103-1 2 nM ⑤
VEGF 1 nM+MT-103-1 8 nM ⑥
VEGF 1 nM+MT-103-1 32 nM ⑦
Fluorescence intensity
1×10⁸
8×10⁷
6×10⁷
4×10⁷
2×10⁷
0
①②③④⑤⑥⑦
*p<0.05
**p<0.01
***p<0.001
Control IgG 2 nM ①
VEGF 1 nM+Control IgG 2 nM ②
VEGF 1 nM+MT-103-4 0.125 nM ③
VEGF 1 nM+MT-103-4 0.5 nM ④
VEGF 1 nM+MT-103-4 2 nM ⑤
VEGF 1 nM+MT-103-4 8 nM ⑥
VEGF 1 nM+MT-103-4 32 nM ⑦

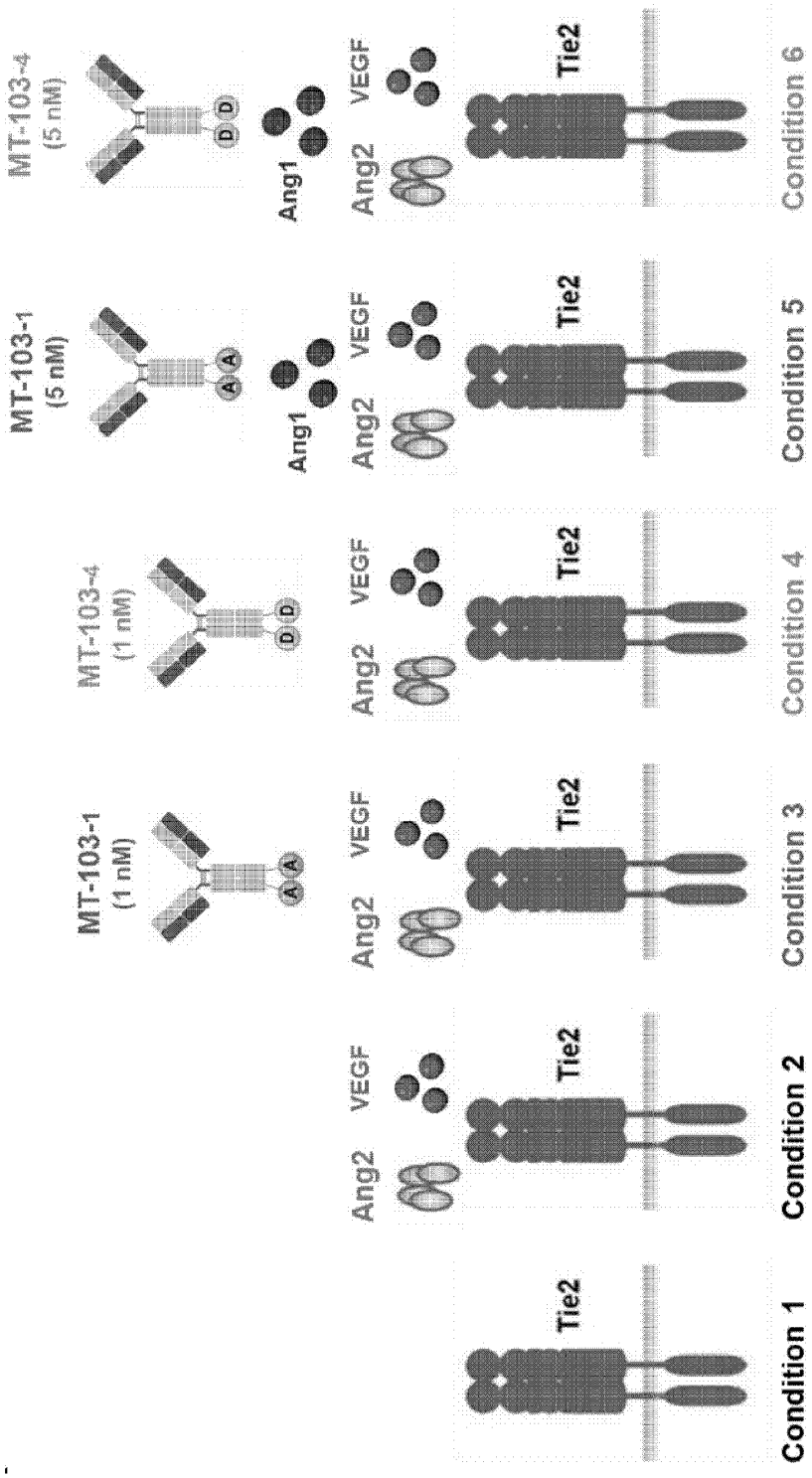
[도33d]


Normalized intensity (%)
Time (hours)
0
24
40
60
80
100
120
140
160
180
① Control IgG 5 nM
② VEGF+Ang-2+Control IgG 5 nM
③ VEGF+Ang-2+MT-103-1 0.05 nM
④ VEGF+Ang-2+MT-103-1 0.5 nM
⑤ VEGF+Ang-2+MT-103-1 5 nM
⑥ VEGF+Ang-2+MT-103-4 0.05 nM
⑦ VEGF+Ang-2+MT-103-4 0.5 nM
⑧ VEGF+Ang-2+MT-103-4 5 nM
*p<0.05
**p<0.01
***p<0.001
Normalized intensity (24-hour, %)
FITC intensity (0-hour)
FITC intensity (24-hour)

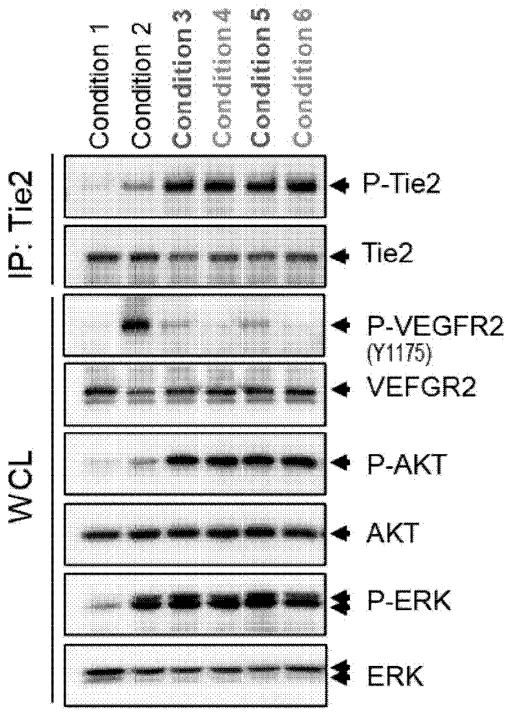
[도34]


Antibody immobilization (MT-101/MT-103-1)
1
0.5
0
-0.5
-1
① MT-103-1
② MT-101
hTie2 loading
Tie2 binding
Tie2
Ang-2 loading
Blockade of Ang-2 binding to Tie2
Ang-2
Tie2
VEGF loading
VEGF binding
MT-103-1
MT-101
VEGF
Ang-2
Tie2

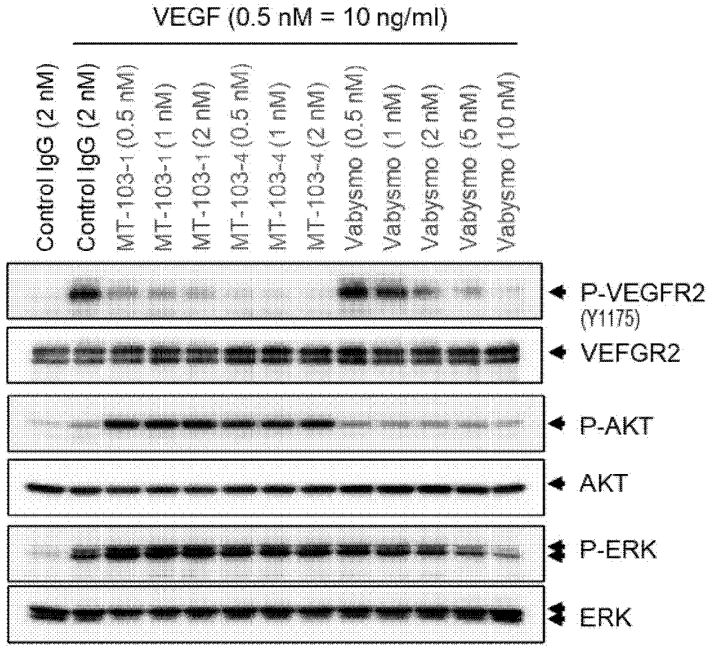
[도35a]


Condition 1
Tie2
Condition 2
Ang2
VEGF
Tie2
Condition 3
MT-103-1
(1 nM)
A A
Ang2
VEGF
Tie2
Condition 4
MT-103-4
(1 nM)
D D
Ang2
VEGF
Tie2
Condition 5
MT-103-1
(5 nM)
A A
Ang1
Ang2
VEGF
Tie2
Condition 6
MT-103-4
(5 nM)
D D
Ang1
Ang2
VEGF
Tie2

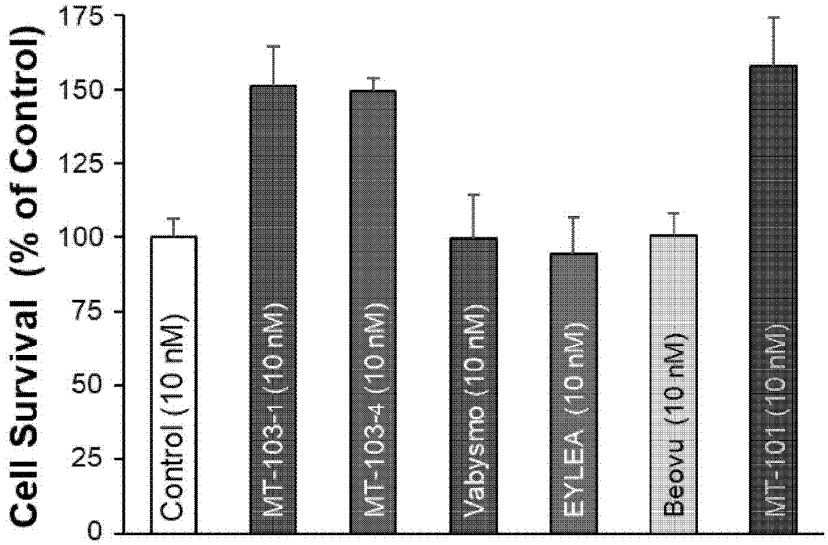
[도35b]


Condition 1
Condition 2
Condition 3
Condition 4
Condition 5
Condition 6
IP: Tie2
WCL
P-Tie2
Tie2
P-VEGFR2
(Y1175)
VEFGR2
P-AKT
AKT
P-ERK
ERK

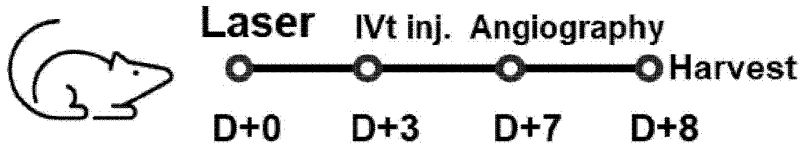
[도36a]


VEGF (0.5 nM = 10 ng/ml)
Control IgG (2 nM)
Control IgG (2 nM)
MT-103-1 (0.5 nM)
MT-103-1 (1 nM)
MT-103-1 (2 nM)
MT-103-4 (0.5 nM)
MT-103-4 (1 nM)
MT-103-4 (2 nM)
Vabysmo (0.5 nM)
Vabysmo (1 nM)
Vabysmo (2 nM)
Vabysmo (5 nM)
Vabysmo (10 nM)
P-VEGFR2
(Y1175)
VEFGR2
P-AKT
AKT
P-ERK
ERK

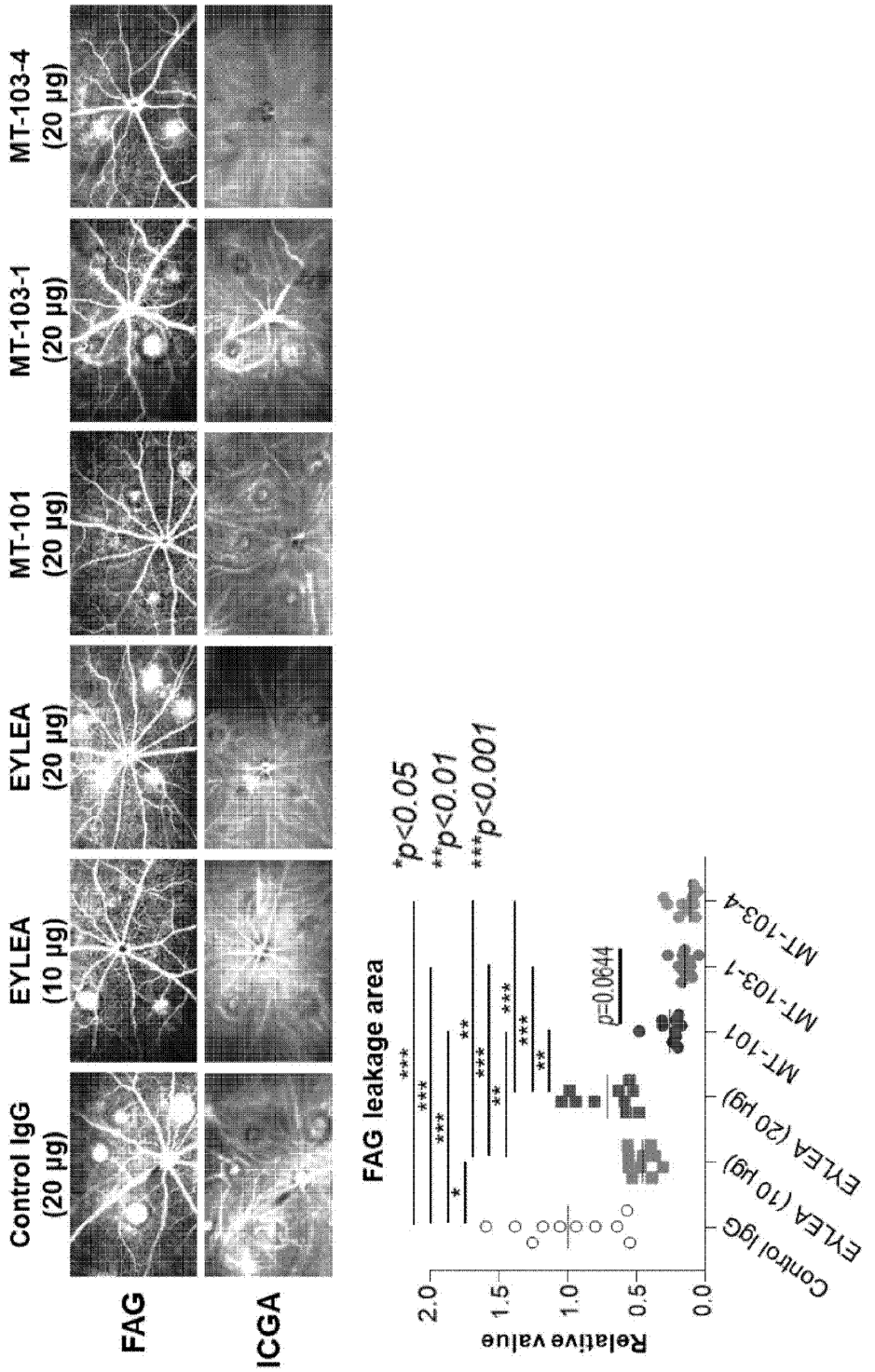
[도36b]


Cell Survival (% of Control)
175
150
125
100
75
50
25
0
Control (10 nM)
MT-103-1 (10 nM)
MT-103-4 (10 nM)
Vabysmo (10 nM)
EYLEA (10 nM)
Beovu (10 nM)
MT-101 (10 nM)

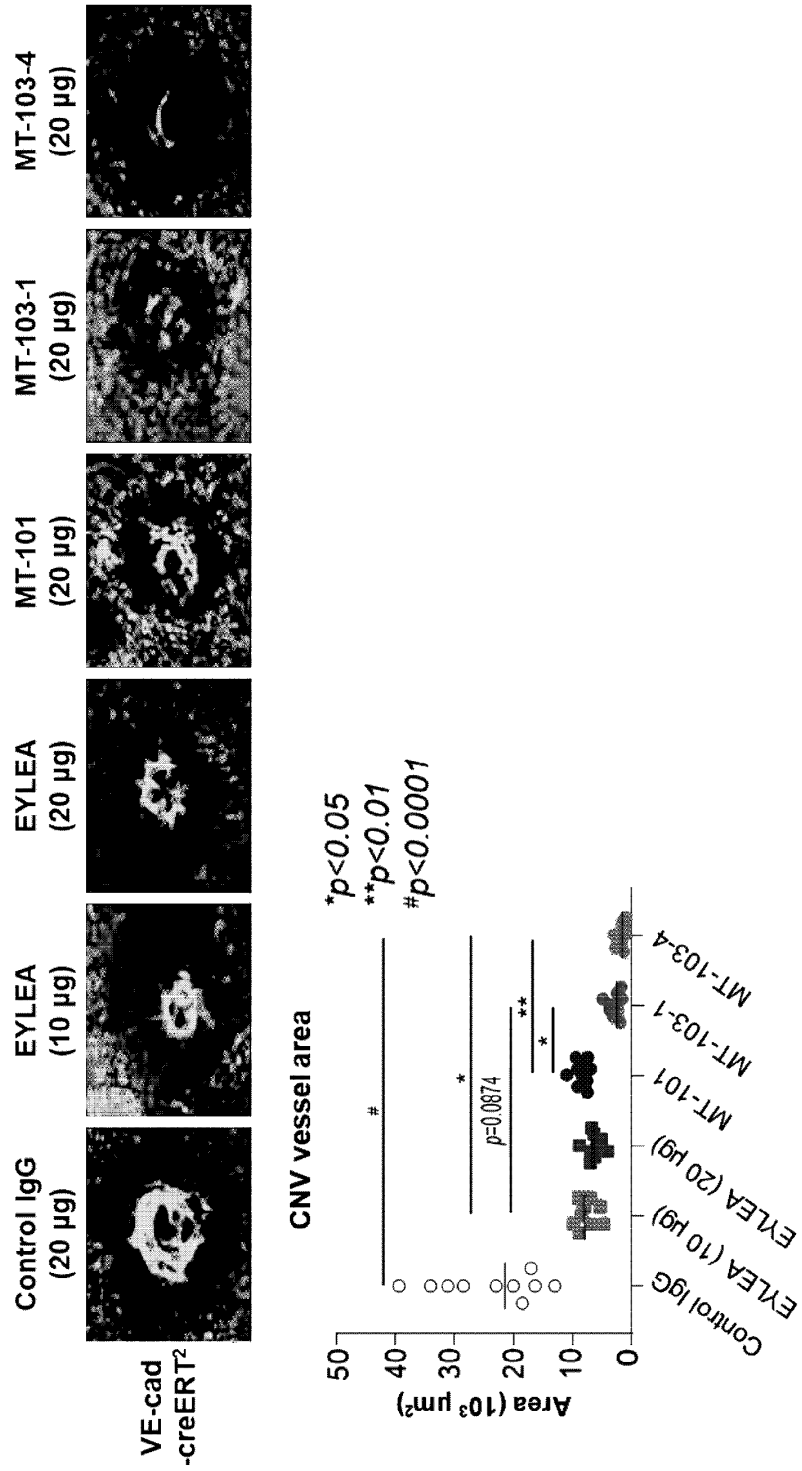
[도37a]


Laser
IVt inj.
Angiography
Harvest
D+0
D+3
D+7
D+8

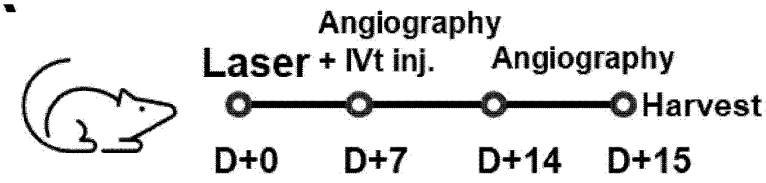
[도37b]


Control IgG (20 μg)
EYLEA (10 μg)
EYLEA (20 μg)
MT-101 (20 μg)
MT-103-1 (20 μg)
MT-103-4 (20 μg)
FAG
ICGA
FAG leakage area
*p<0.05
**p<0.01
***p<0.001
p=0.0644
Relative value
2.0
1.5
1.0
0.5
0.0
Control IgG
EYLEA (10 μg)
EYLEA (20 μg)
MT-101
MT-103-1
MT-103-4

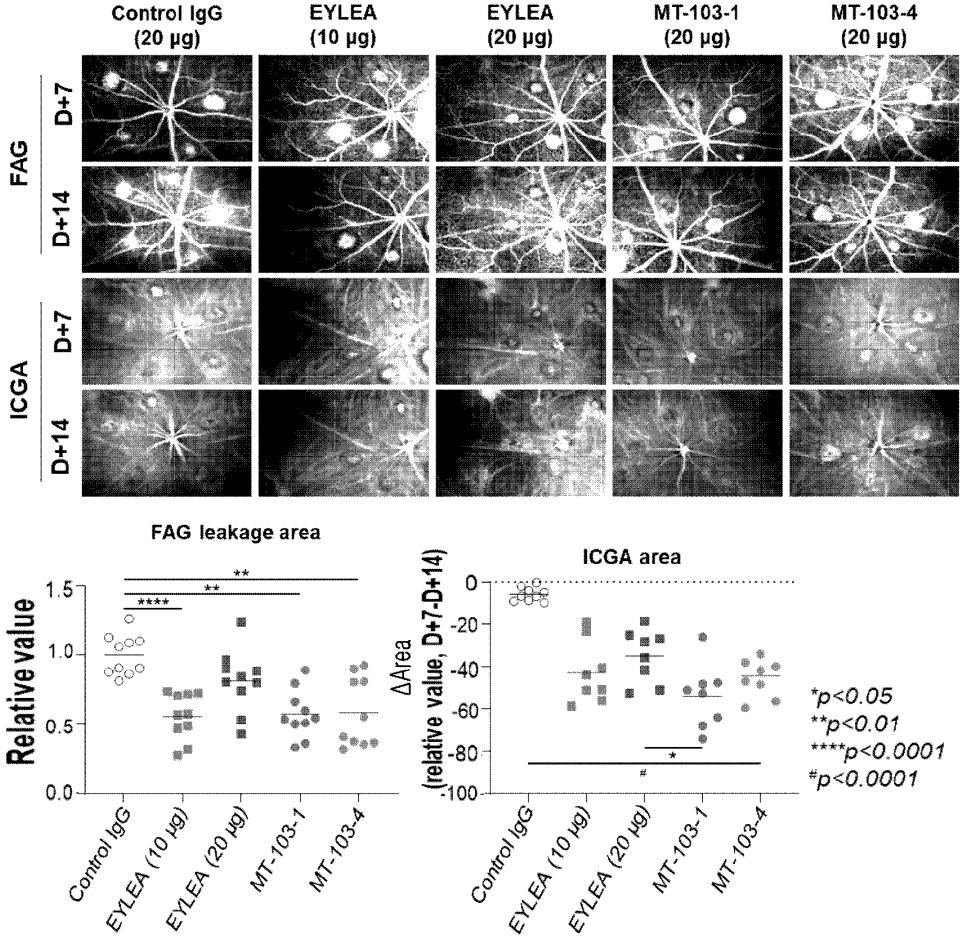
[도37c]


Control IgG (20 μg)
EYLEA (10 μg)
EYLEA (20 μg)
MT-101 (20 μg)
MT-103-1 (20 μg)
MT-103-4 (20 μg)
VE-cad -creERT2
CNV vessel area
*p<0.05
**p<0.01
#p<0.0001
p=0.0874
Area (10³ μm²)
0
10
20
30
40
50
Control IgG
EYLEA (10 μg)
EYLEA (20 μg)
MT-101
MT-103-1
MT-103-4

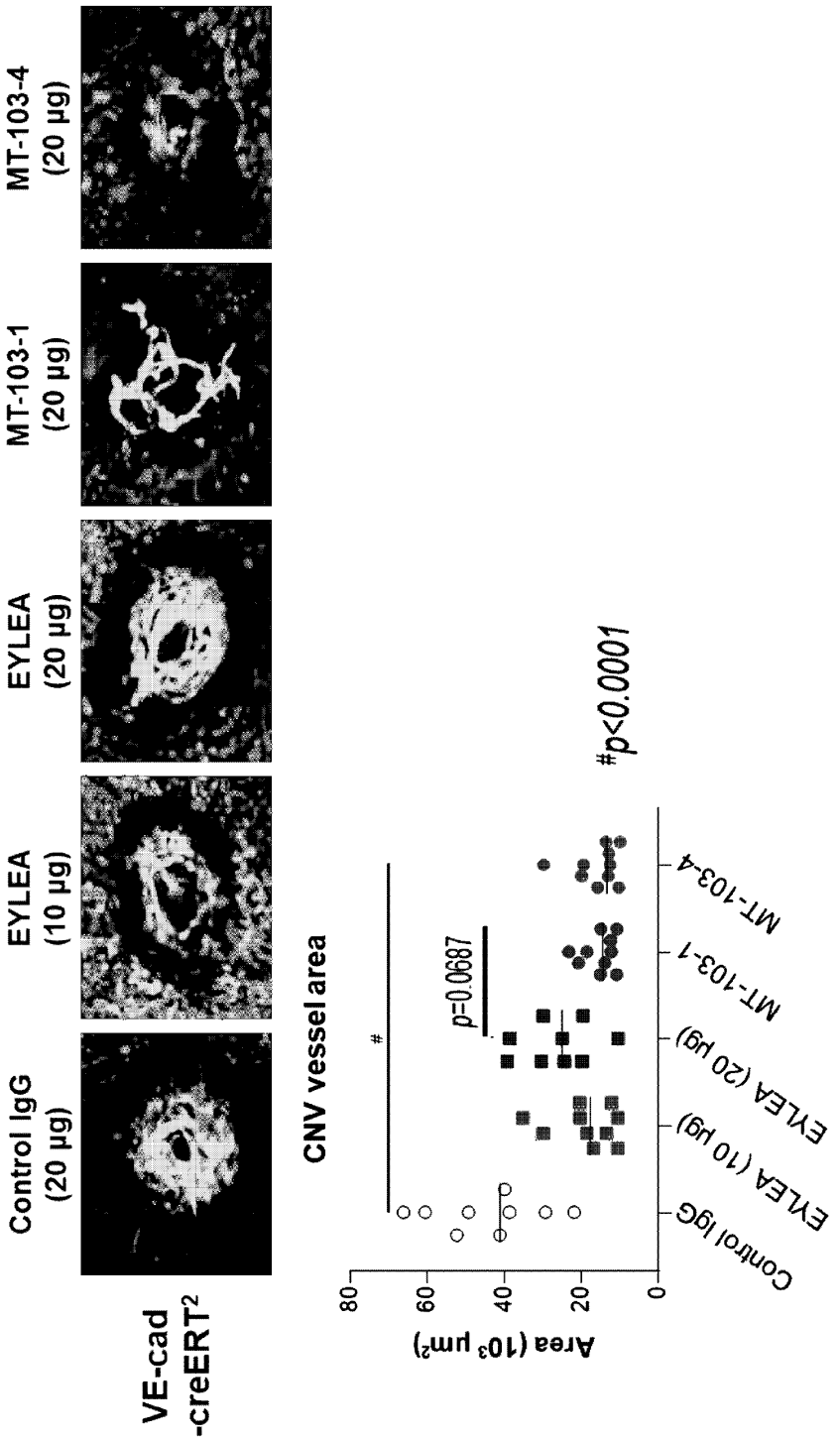
[도38a]


Angiography
Laser + IVt inj.
Angiography
Harvest
D+0
D+7
D+14
D+15

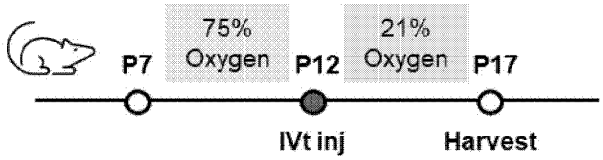
[도38b]


Control IgG (20 μg)
EYLEA (10 μg)
EYLEA (20 μg)
MT-103-1 (20 μg)
MT-103-4 (20 μg)
FAG
ICGA
D+7
D+14
FAG leakage area
Relative value
ICGA area
ΔArea (relative value, D+7-D+14)
Control IgG
EYLEA (10 μg)
EYLEA (20 μg)
MT-103-1
MT-103-4
*p<0.05
**p<0.01
****p<0.0001
#p<0.0001

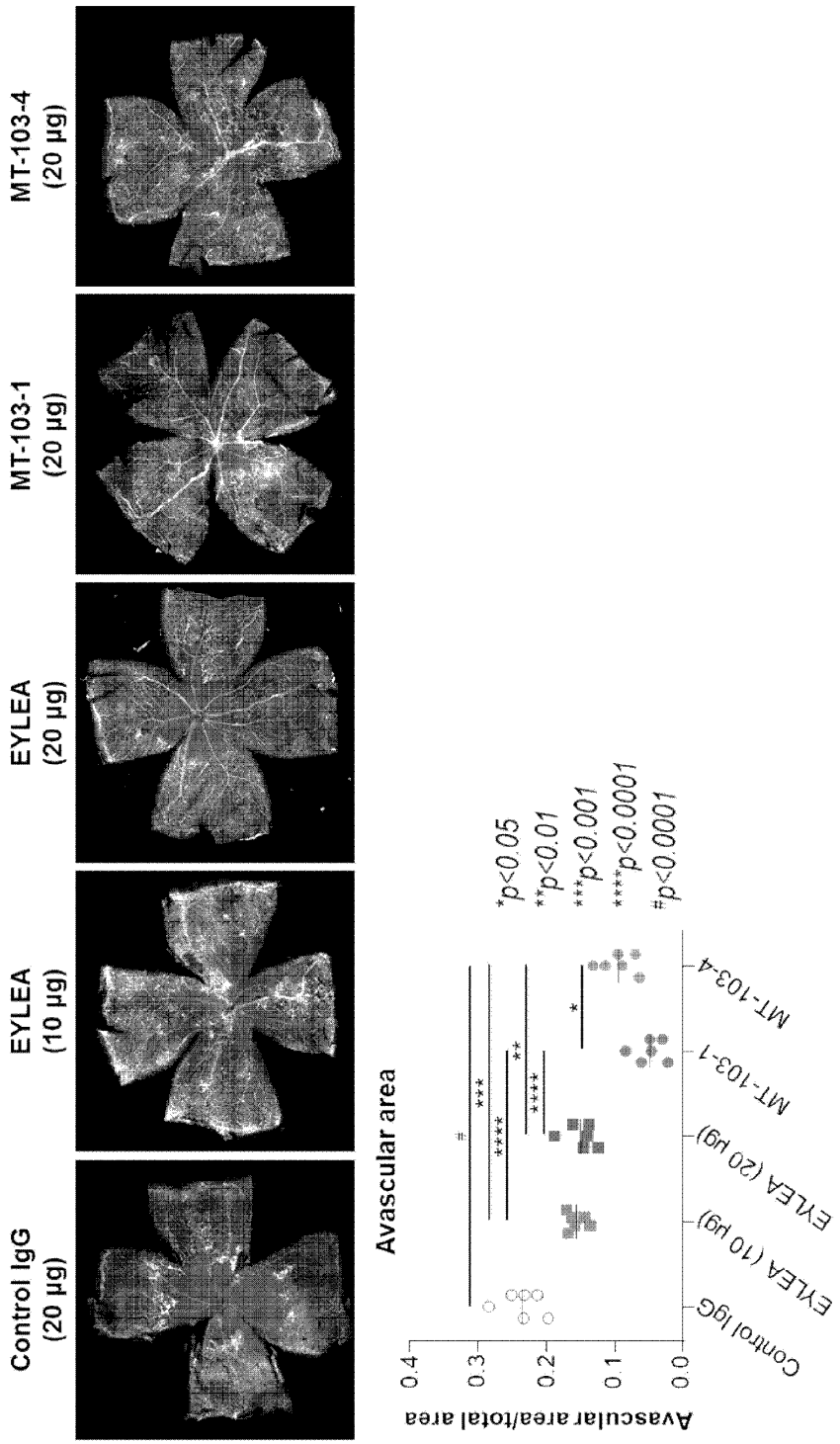
[도38c]


VE-cad -creERT2
Control IgG (20 μg)
EYLEA (10 μg)
EYLEA (20 μg)
MT-103-1 (20 μg)
MT-103-4 (20 μg)
CNV vessel area
Area (10³ μm²)
80
60
40
20
0
#
p=0.0687
#p<0.0001
Control IgG
EYLEA (10 μg)
EYLEA (20 μg)
MT-103-1
MT-103-4

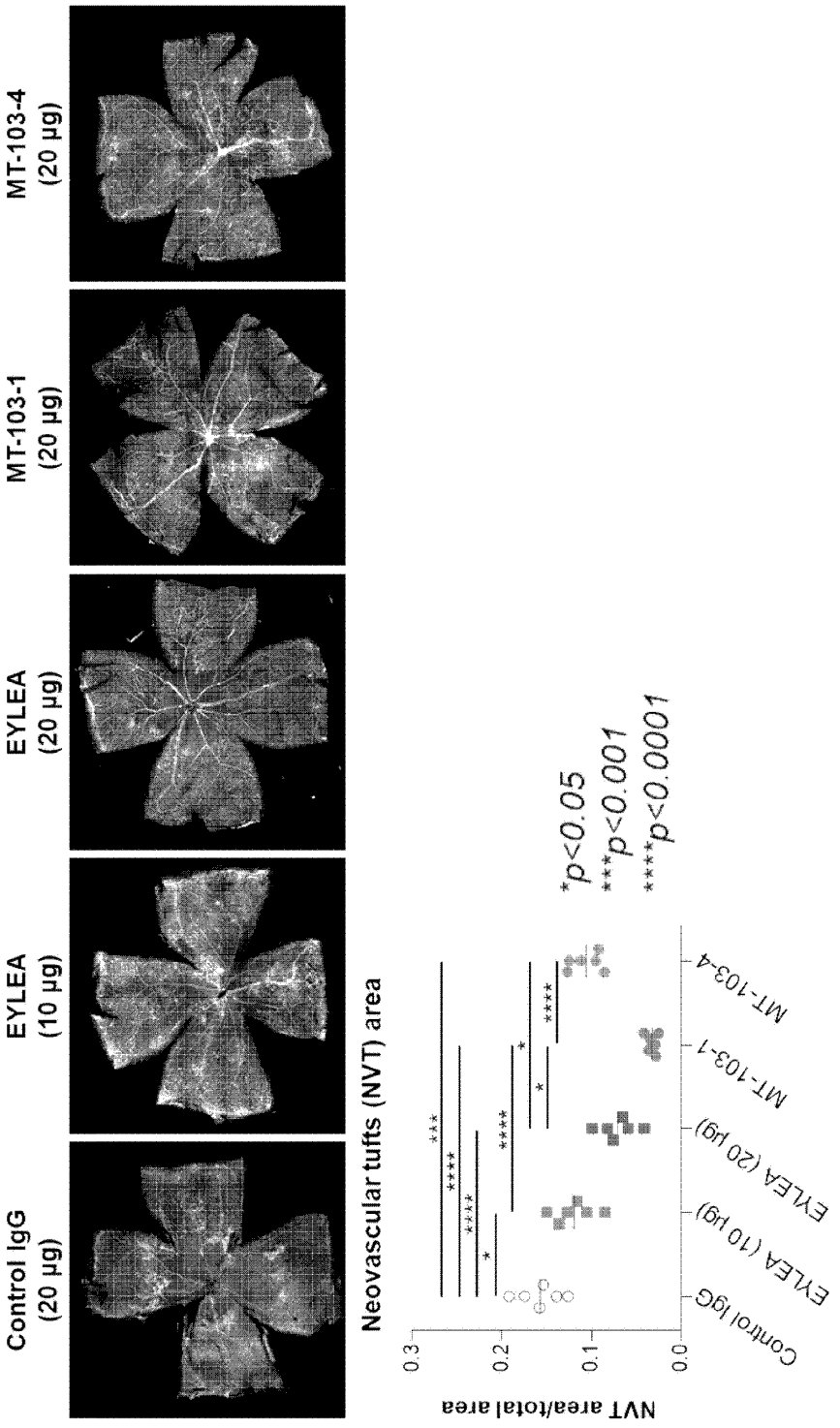
[도39a]



[도39b]


Control IgG (20 μg)
EYLEA (10 μg)
EYLEA (20 μg)
MT-103-1 (20 μg)
MT-103-4 (20 μg)
Avascular area
Avascular area/total area
0.0
0.1
0.2
0.3
0.4
Control IgG
EYLEA (10 μg)
EYLEA (20 μg)
MT-103-1
MT-103-4
*p<0.05
**p<0.01
***p<0.001
****p<0.0001
#p<0.0001

[도39c]


Control IgG (20 μg)
EYLEA (10 μg)
EYLEA (20 μg)
MT-103-1 (20 μg)
MT-103-4 (20 μg)
Neovascular tufts (NVT) area
NVT area/total area
0.0
0.1
0.2
0.3
Control IgG
EYLEA (10 μg)
EYLEA (20 μg)
MT-103-1
MT-103-4
*p<0.05
***p<0.001
****p<0.0001