SUPPLEMENTAL MATERIAL

Supplemental Figures

Figure S1. Conditional deletion of AT1a receptors in vascular smooth muscle cells of arteries. A: Schematic representation of the mouse allele containing loxP sequences, and the floxed allele after the action of Cre recombinase. **B:** Immunofluorescence staining results show that AT1R (red) is highly expressed in the mesenteric artery of SM-*Agtr1a*$^{+/+}$ mice. In SM-*Agtr1a*$^{-/-}$ mouse mesenteric artery, the expression of AT1R is specifically reduced in smooth muscle cells. **C:** Expression of AT1R is completely reduced in the mesenteric artery of global *Agtr1a*$^{-/-}$ mice. DAPI (4',6-diamino-2-phenylindole) nuclear staining, alpha-SMA (alpha smooth muscle actin), Scale bar: 40 μm.

Figure S2. Myogenic tone in cerebral arteries. A, B: Representative recordings of middle/posterior cerebral arteries diameter at the pressure of 80 mmHg in control conditions (WT), Ang II 100 nmol/L, and in Ca^{2+} free solution. **C:** Myogenic tone (at 80 mmHg) expressed as dilation of vessels induced by external Ca^{2+} free solution. **D, E:** Response to Ang II **(D)** and 60 mM KCl **(E)** in middle/posterior cerebral arteries of SM-*Agtr1a*$^{+/+}$ and SM-*Agtr1a*$^{-/-}$ mice. SM-*Agtr1a*$^{+/+}$, n=6 vessels from 6 mice, and SM-*Agtr1a*$^{-/-}$, n=6 vessels from 6 mice. **F:** Average myogenic constriction of cerebral arteries in physiological salt solution (PSS) in the absence and presence of 1 μmol/L FR900359 (n=6 each, from 6 mice each). **G:** Response to Ang II in the absence and presence of 1 μmol/L FR900359 at 80 mmHg (n=6 each, from 6 mice each). *$p<0.05$; n.s., not significant.

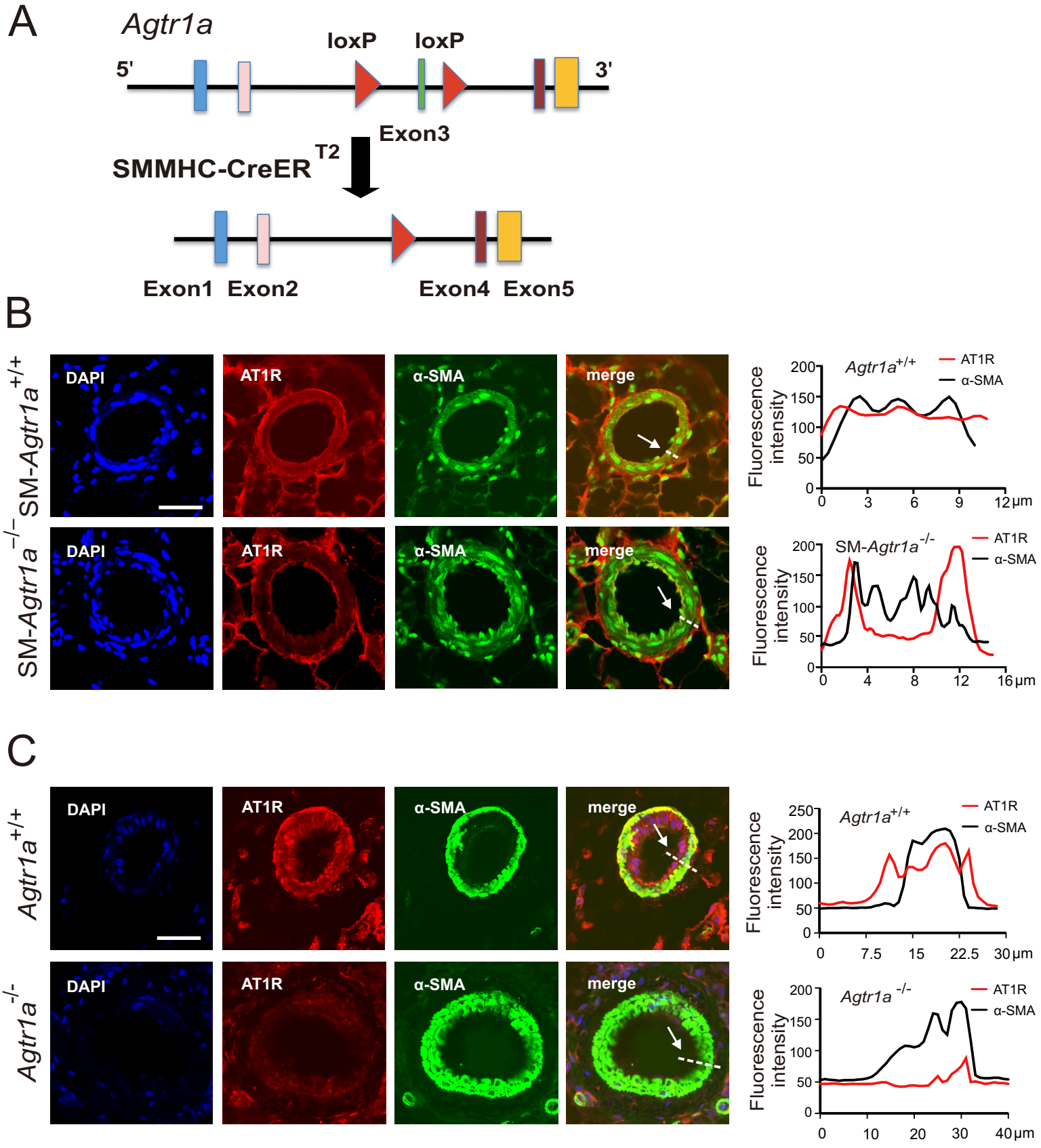
A
Agtr1a
loxP
loxP
5'
3'
Exon3
SMMHC-CreER T2
Exon1 Exon2
Exon4 Exon5
B
SM-Agtr1a +/+
SM-Agtr1a −/−
DAPI
AT1R
α-SMA
merge
Agtr1a+/+
SM-Agtr1a -/-
AT1R
α-SMA
Fluorescence intensity
200
150
100
50
0
0 3 6 9 12 μm
0 4 8 12 16 μm
C
Agtr1a +/+
Agtr1a -/-
DAPI
AT1R
α-SMA
merge
Agtr1a+/+
Agtr1a -/-
AT1R
α-SMA
Fluorescence intensity
250
200
150
100
50
0
0 7.5 15 22.5 30 μm
0 10 20 30 40 μm

Figure S1

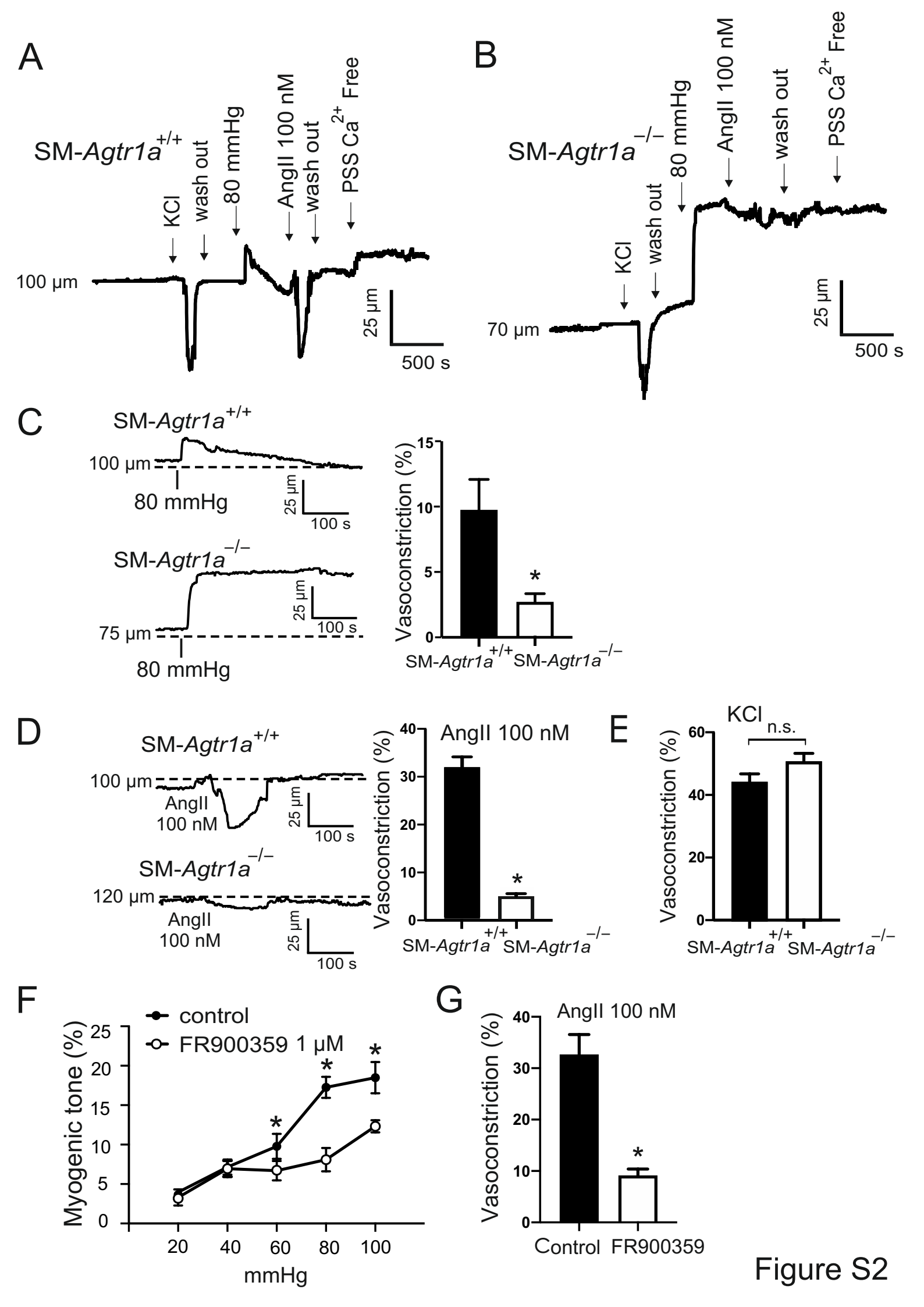
A
SM-Agtr1a+/+
KCl
wash out
80 mmHg
AngII 100 nM
wash out
PSS Ca2+ Free
100 µm
25 µm
500 s
B
SM-Agtr1a−/−
KCl
wash out
80 mmHg
AngII 100 nM
wash out
PSS Ca2+ Free
70 µm
25 µm
500 s
C
SM-Agtr1a+/+
100 µm
80 mmHg
25 µm
100 s
SM-Agtr1a−/−
75 µm
80 mmHg
25 µm
100 s
Vasoconstriction (%)
15
10
5
0
*
SM-Agtr1a+/+
SM-Agtr1a−/−
D
SM-Agtr1a+/+
100 µm
AngII
100 nM
25 µm
100 s
SM-Agtr1a−/−
120 µm
AngII
100 nM
25 µm
100 s
AngII 100 nM
Vasoconstriction (%)
40
30
20
10
0
*
SM-Agtr1a+/+
SM-Agtr1a−/−
E
KCl
n.s.
Vasoconstriction (%)
60
40
20
0
SM-Agtr1a+/+
SM-Agtr1a−/−
F
control
FR900359 1 µM
Myogenic tone (%)
25
20
15
10
5
0
*
*
*
20
40
60
80
100
mmHg
G
AngII 100 nM
Vasoconstriction (%)
40
30
20
10
0
*
Control
FR900359

Figure S2