SUPPLEMENTAL FIGURES

Suppl Fig. 1 Gestational time course of EPC development in HSD, DSS, and BN rats. Trophoblast cells were identified by cytokeratin immunostaining of HSD (*Panels A, D, G*), DSS (*Panels B, E, H*) and BN (*Panels C, E, I*) rat tissue sections from gestation d7.5 (*Panels A-C*), d8.5 (*Panels D-F*), and d9.5 (*Panels G-I*) placentation sites. Bars = 500 μm. EPC: ectoplacental cone; Emb: embryo; MD: mesometrial decidua; AD: antimesometrial decidua; Epi: epiblast; Cho: chorion.

Suppl Fig. 2 NK cells and vasculature within the placentation sites of gestation d8.5 DSS (*Panels A, C, E*) and BN (*Panels B, D, F*) rats. Uterine NK cell distributions were detected by perforin immunoreactivity (*Panels A and B*) and vasculature was assessed by immunostaining for smooth muscle actin (ACTA2; *Panels C and D*) and a rat endothelial cell antigen (RECA1; *Panels E and F*). Bars = 500 μm. MD: mesometrial decidua; EPC: ectoplacental cone.

Suppl Fig. 3 Proliferation and cell death in the decidual compartments of gestation d8.5 DSS (*Panels A and C*) and BN (*Panels B and D*) rats. Proliferation was monitored by MKI67 immunoreactivity (Panels A and B) and cell death as assessed by TUNEL positive cells (*Panels C and D*). EPC: ectoplacental cone; Emb: embryo; MD: mesometrial decidua; AD: antimesometrial decidua.

Suppl Fig. 4 In vitro differentiation of DSS and BN rat uterine stromal cells. Morphologies of DSS and BN uterine stromal cells following 72 h of in vitro

differentiation are shown in the upper panels. Transcript levels for decidualization-associated genes were measured by qRT-PCR and are presented in the bar graphs (A: *Prl6a1*; B: *Prl8a2*, C: *Prl3c1*; D: *Gja1*).

Suppl Fig. 5 Expression of transcripts for the decidual PRL family in deciduomal tissue from d8.5 pseudopregnant DSS and BN rats. Deciduomal transcript levels were monitored by qRT-PCR. *A*: *Prl*; *B*: *Prl6a1*; *C*: *Prl8a2*; D: *Prl3c1*.

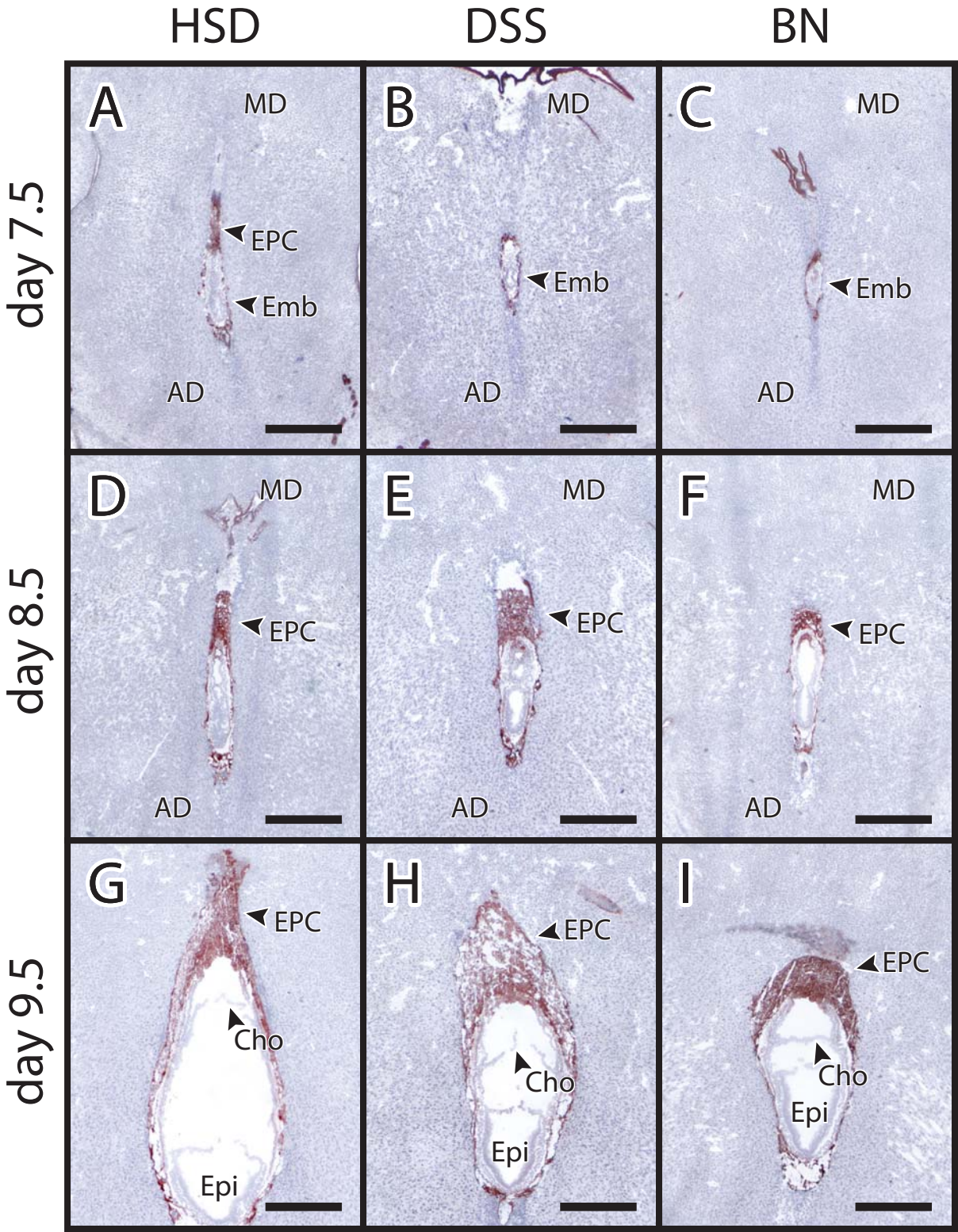
Suppl Fig. 6 Expression of transcripts for steroidogenic steroid metabolizing enzymes in deciduomal tissue from d8.5 pseudopregnant DSS and BN rats. Deciduomal transcript levels were determined by qRT-PCR. *A: Cyp11a1; B: Cyp17a1; C: Hsd17b2; D: Hsd3b1; E: Cyp19a1; F: Akr1c18*.

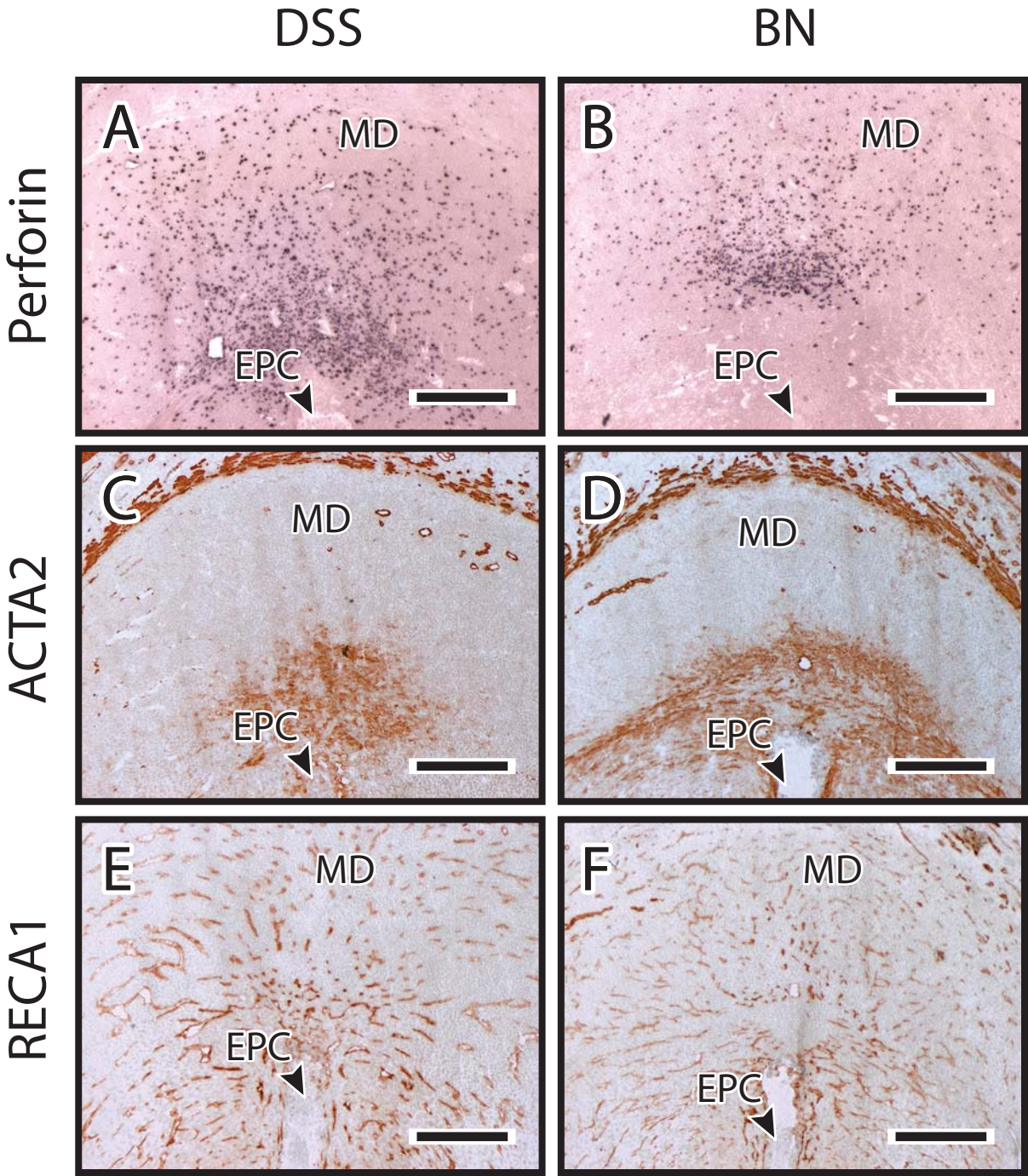
Suppl Fig. 7 Evaluation of mammary gland *Areg* responsiveness to P4. Rats from DSS and BN strains were ovariectomized, rested for two weeks, and acutely treated with a subcutaneous injection of P4 (40 mg/kg body weight) or vehicle. Twenty-four h post injection rats were sacrificed, mammary gland collected, and *Areg* gene expression assessed by qRT-PCR. Data was analyzed with the Wilcoxon rank sum test, *P<0.01, DSS control: n=10, DSS P4-treated: n=12, BN control: n=7, BN P4-treated: n=7.

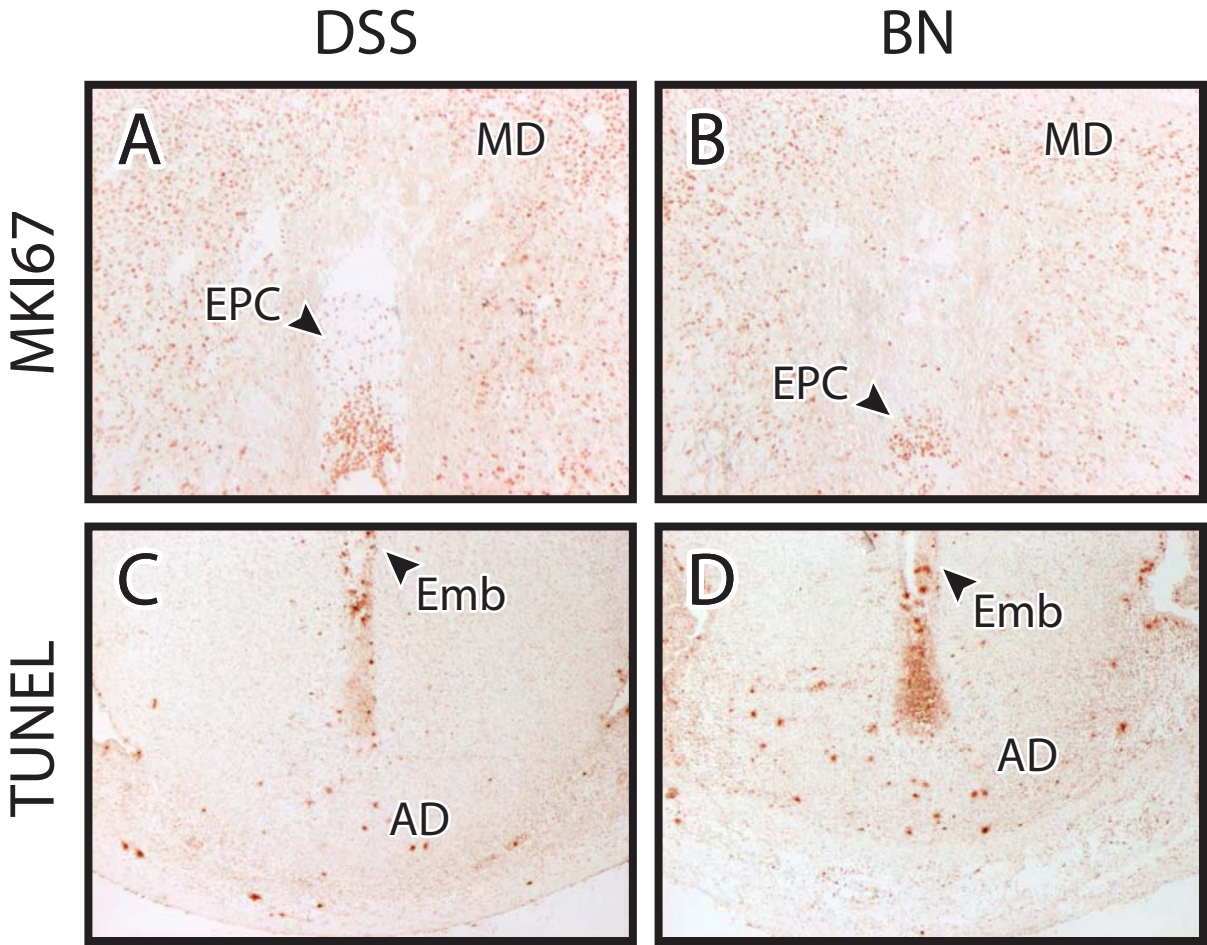
Suppl Fig. 8 *Pgr* and *Ncoa1* gene expression in gestation d4.5 rat uterus. Transcript levels for total *Pgr* (*Panel A*), *Pgr B* (*Panel B*), and *Ncoa1* (Panel C) in uteri of gestation d4.5 DSS and BN rats. Wilcoxon rank sum test. *P<0.05, n=8 for each group.

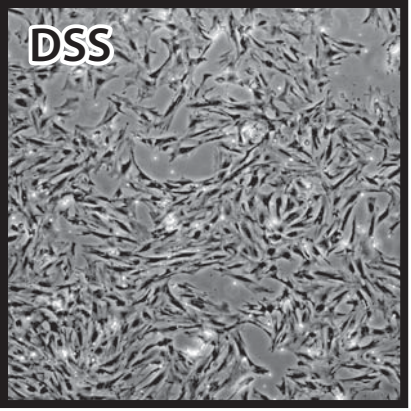
Suppl Fig. 9 PGR Immunolocalization in gestation d4.5 and d8.5 uterine tissues. PGR was detected by immunohistochemistry in gestation d4.5 (Panels A and B) and d8.5 (*Panels C-H*) of DSS (*Panels A, C, E, G*) and BN (*Panels B, D, F, H*) rat uterine tissues. The schematic in the center of the figure shows the location of the gestation d8.5 tissue sections presented. *Panels C and D*, mesometrial; *Panels E and F*, central; *Panels G and H*, antimesometrial. EPC: ectoplacental cone; Dec: decidua; Emb: embryo.

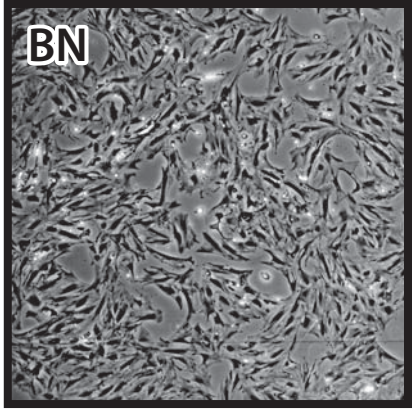
Suppl Fig. 10 *Pgr* promoter analyses. *Panel A*, Sequence analysis of *Pgr* regulatory DNA from DSS, F344, and BN rat strains (GenBank Accession: HM037358, HM037359, HM037360). *Pgr* promoter-reporter constructs (*Panel B*) were evaluated in rat U1 uterine stromal cells (*Panel C*) and human MCF7 breast cancer cells (*Panel D*). Note that each rat *Pgr* promoter reporter construct exhibited similar activities in the progesterone responsive rat U1 uterine stromal and human MCF7 breast cancer cells.


HSD
DSS
BN
day 7.5
day 8.5
day 9.5
A
B
C
D
E
F
G
H
I
MD
AD
EPC
Emb
Cho
Epi

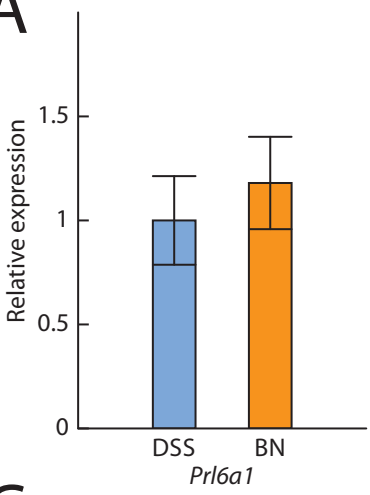

DSS
BN
Perforin
ACTA2
RECA1
A
B
C
D
E
F
MD
EPC


DSS
BN
MKI67
TUNEL
A
MD
EPC
B
MD
EPC
C
Emb
AD
D
Emb
AD

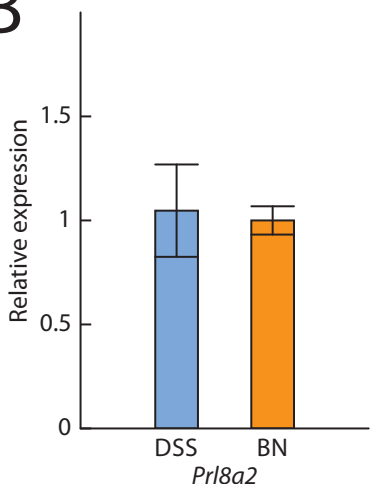

DSS


BN

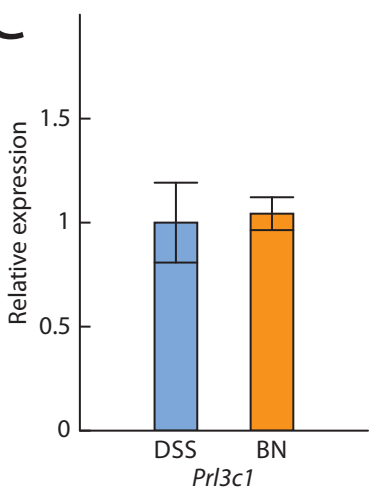
A


Relative expression
1.5
1
0.5
0
DSS
BN
Prl6a1

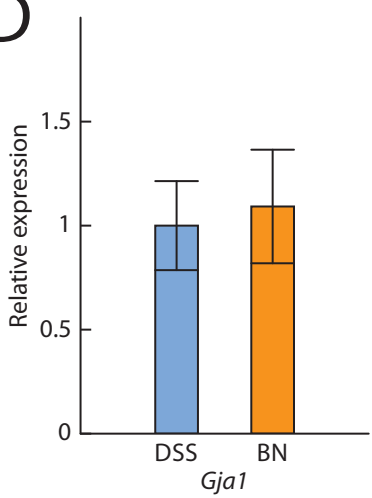
B

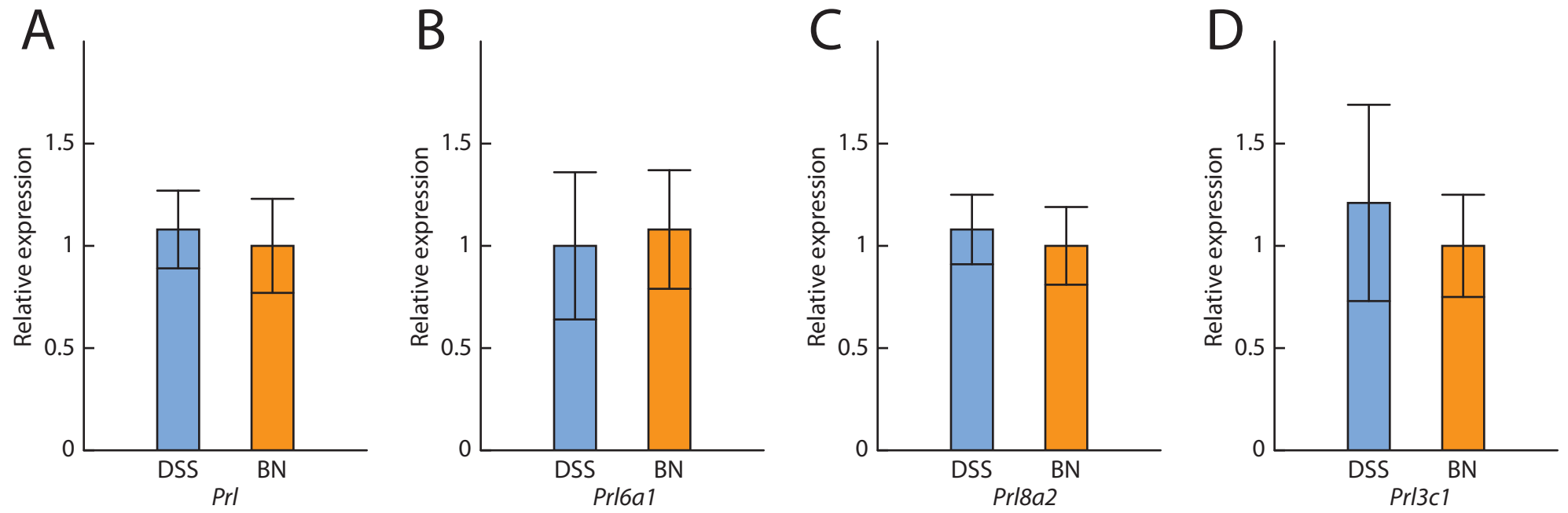

Relative expression
1.5
1
0.5
0
DSS
BN
Prl8a2

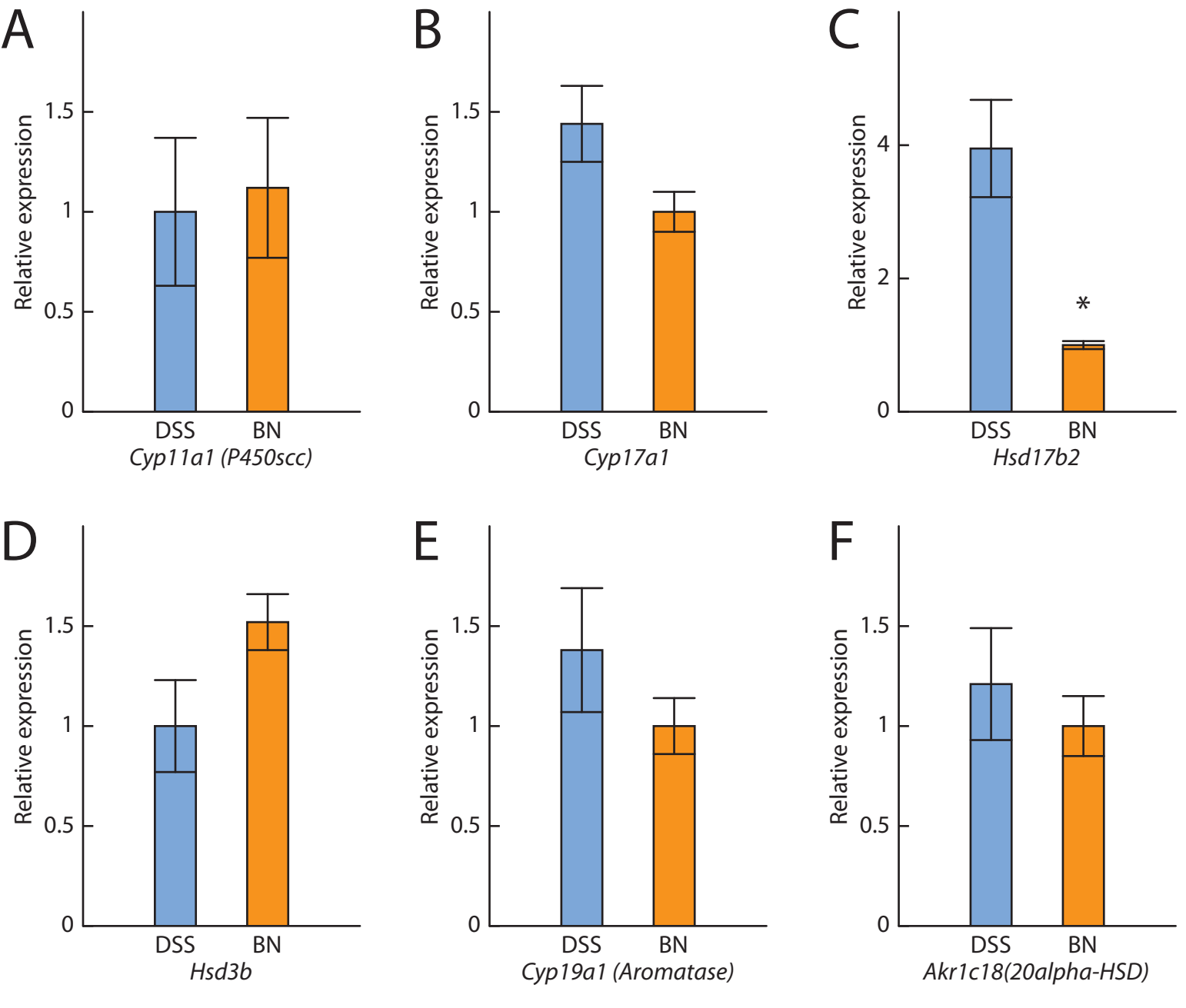
C

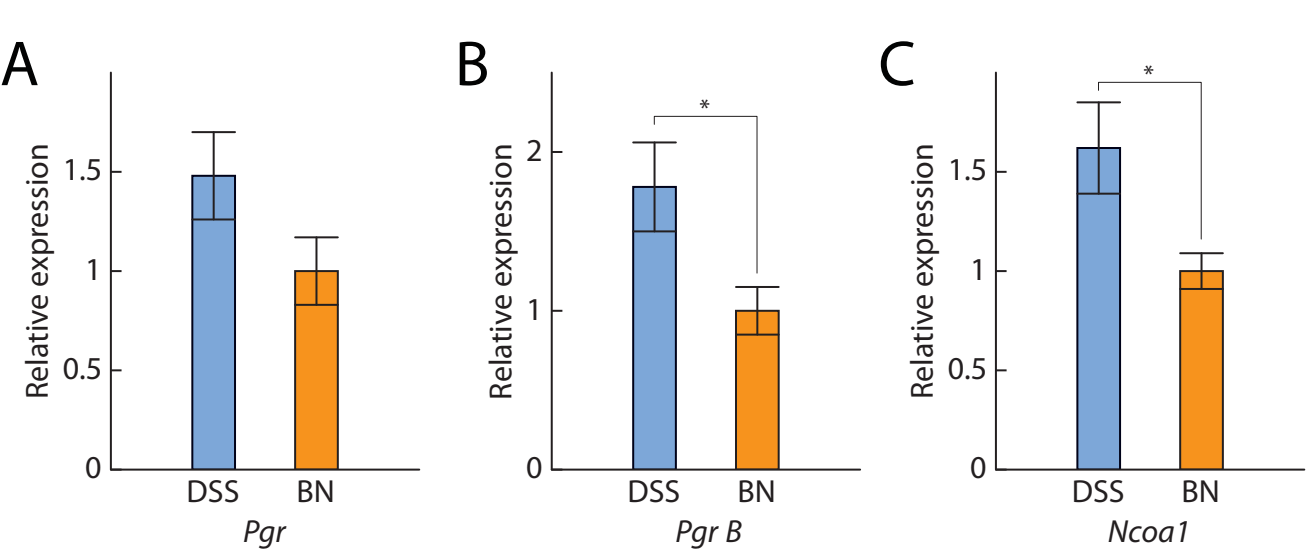

Relative expression
1.5
1
0.5
0
DSS
BN
Prl3c1

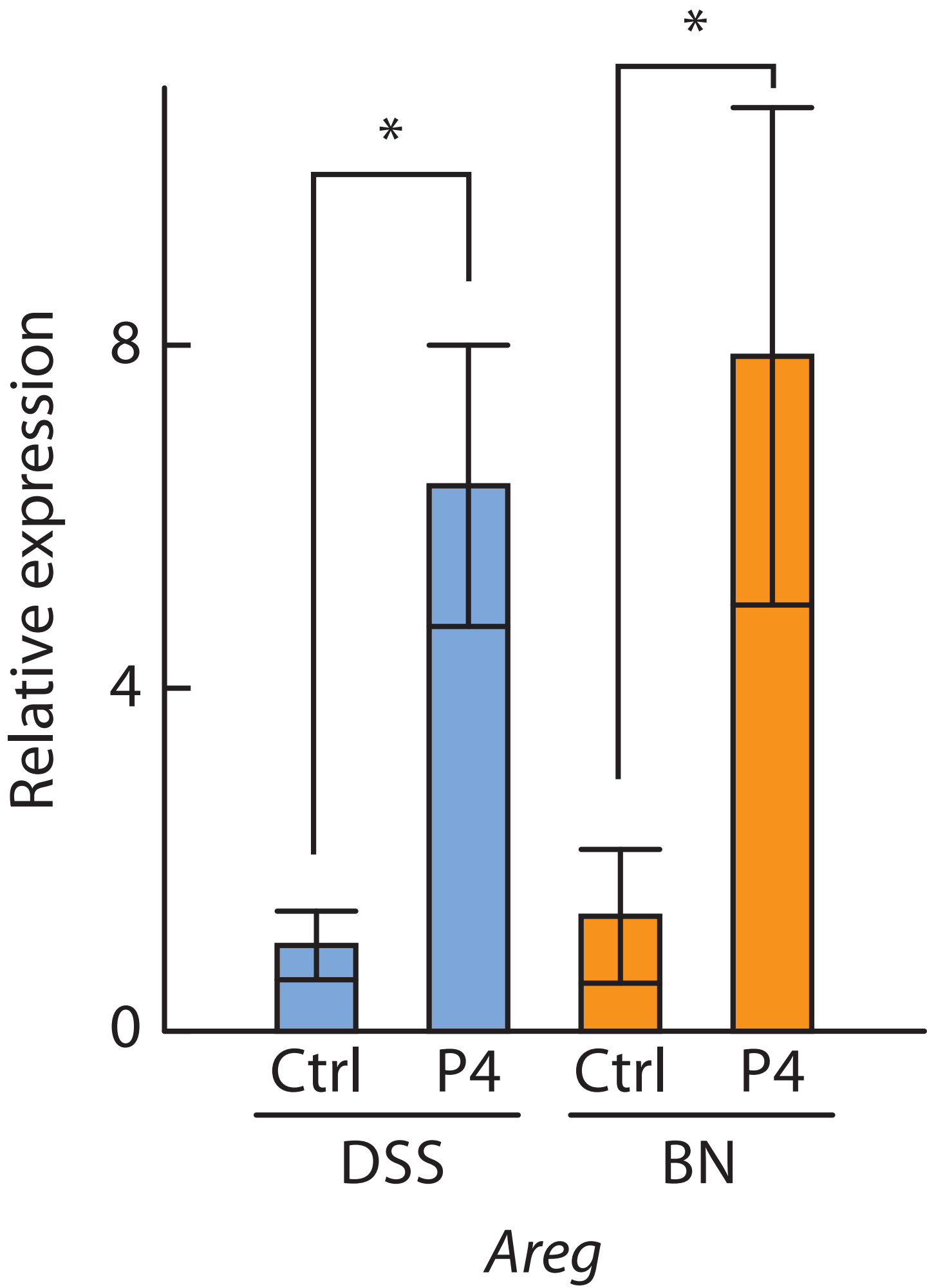
D

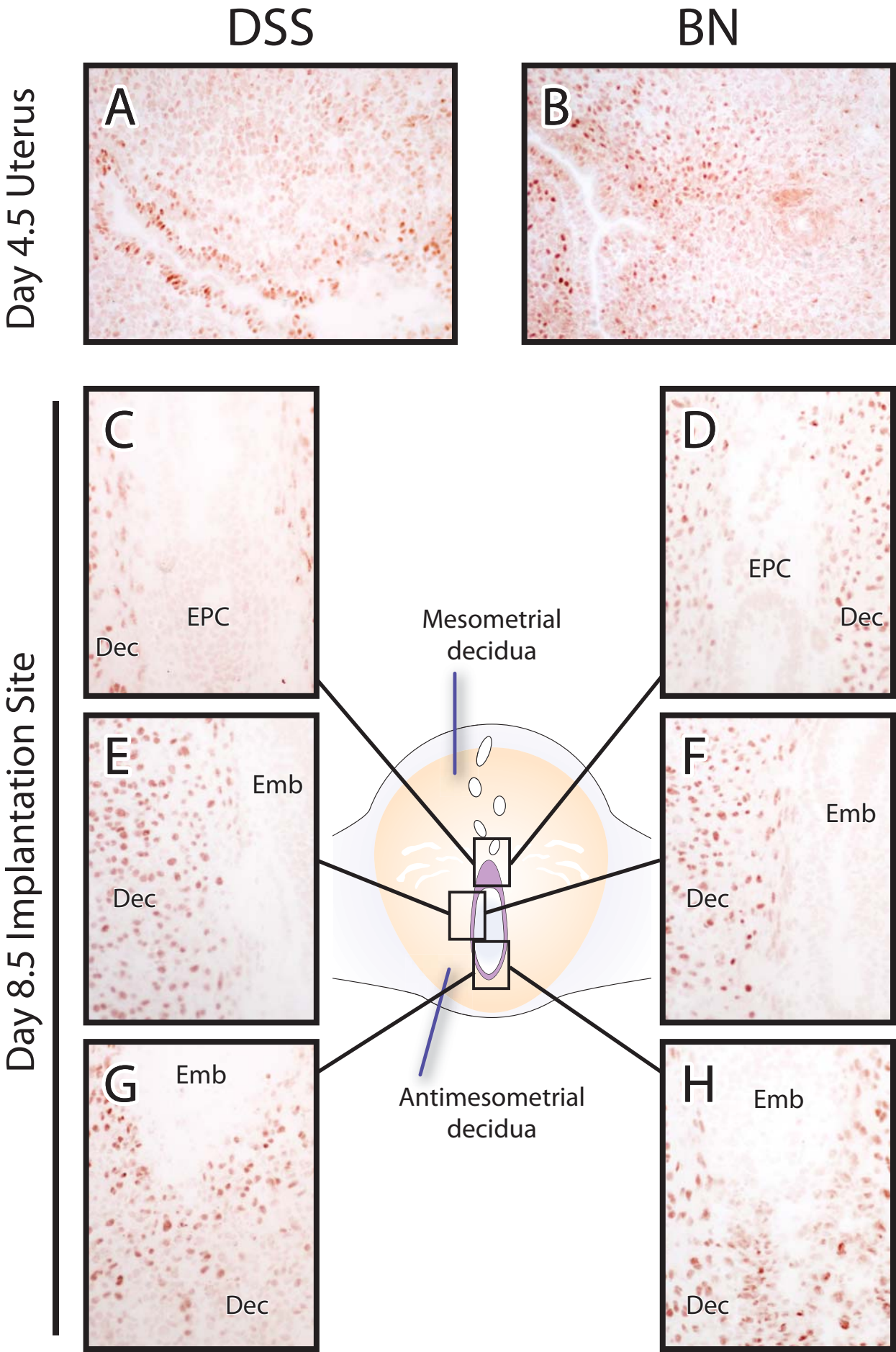

Relative expression
1.5
1
0.5
0
DSS
BN
Gja1


A
B
C
D
Relative expression
1.5
1
0.5
0
DSS
BN
Prl
Prl6a1
Prl8a2
Prl3c1


A
Relative expression
1.5
1
0.5
0
DSS
BN
Cyp11a1 (P450scc)
B
Relative expression
1.5
1
0.5
0
DSS
BN
Cyp17a1
C
Relative expression
4
2
0
*
DSS
BN
Hsd17b2
D
Relative expression
1.5
1
0.5
0
DSS
BN
Hsd3b
E
Relative expression
1.5
1
0.5
0
DSS
BN
Cyp19a1 (Aromatase)
F
Relative expression
1.5
1
0.5
0
DSS
BN
Akr1c18(20alpha-HSD)


A
Relative expression
1.5
1
0.5
0
DSS
BN
Pgr
B
Relative expression
2
1
0
*
DSS
BN
Pgr B
C
Relative expression
1.5
1
0.5
0
*
DSS
BN
Ncoa1


*
*
Relative expression
8
4
0
Ctrl
P4
Ctrl
P4
DSS
BN
Areg


DSS
BN
Day 4.5 Uterus
A
B
Day 8.5 Implantation Site
C
EPC
Dec
D
EPC
Dec
Mesometrial
decidua
E
Emb
Dec
F
Emb
Dec
G
Emb
Dec
Antimesometrial
decidua
H
Emb
Dec

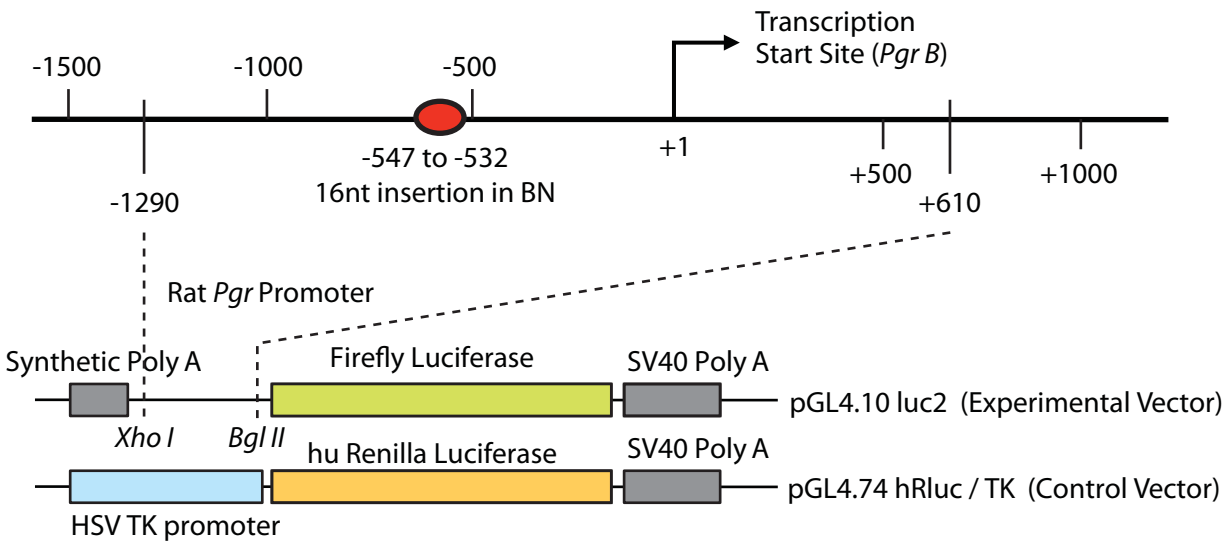
A

-547 to -532 nt insertion

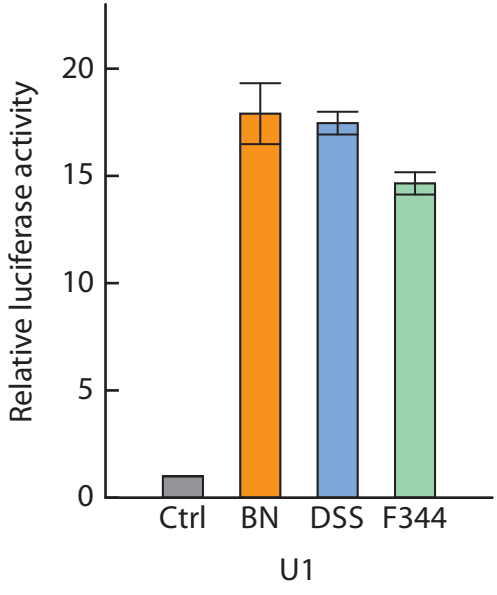
```
BN       -1308…//…gtagttggcc…//…gggcatgct…//…tatagaata…//…gctcattag…//…ttggtactatatgcacagtgacac…/
DSS      -1308…//…gtagctggcc…//…gggcgtgct…//…tataaaata…//…gctcgttag…//…ttgg……c……acac…/
Fisher   -1308…//…gtagctggcc…//…gggcgtgct…//…tataaaata…//…gctcgttag…//…ttgg……c……acac…/

BN       /…aaaattttg…//…tgcttcagc…//…agggaagtc…//…cgcctatac…//…gagtcgaag…+1060
DSS      /…aaaaatttg…//…tgctccagc…//…aggggagtc…//…cgccaatac…//…gagtggaag…+1060
Fisher   /…aaaaatttg…//…tgctccagc…//…aggggagtc…//…cgccaatac…//…gagtggaag…+1060
```

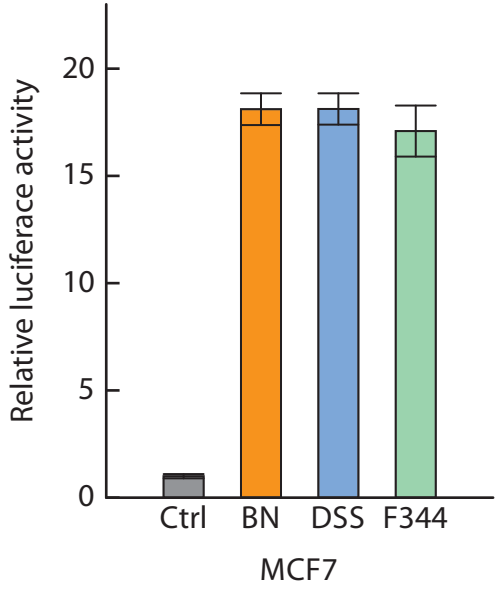
B



C



D



Suppl Table 1. Primers used for qRT-PCR analyses of deciduomal gene expression

Symbol	Accession No.	Forward primer	Reverse primer
Fmo3	NM_053433	TGACGAGAAAATGGGGAAGA	ACCTGGGGTCCTTGAGAAAC
Car4	NM_019174	CTCTACTGGCGCTGGCTTAC	TCTCCAGTCCATTGTTCAGG
Fubp3	BF282112	GTTTCAAACGAGCGTTCCTC	AAAGGGATGATTTCCAATGTTT
Cdkn1a	U24174	TCCACAGCGATATCGAGACA	GCTCAACTGCTCACTGTCCA
Slpi	NM_053372	GAATCCTGTTCCCATTCGTG	TTCCCACACATACCCTCACA
Rhox5	NM_022175	TGGGTGATGTGAAAGCAGAG	GAACACCAGGACCAAAGTGG
Naprt1	BF416417	ATACCAGGTCGCATTGTCAGAG	ATGACACGTGGCTGACAAAGTT
Col6a2	AI030021	TGCCACACTGTCCCTAATGA	CCACTAGGCCATCTGAGAGC
Ptgfrn	NM_019243	AGCCCGTCAACATATTCTGG	TGGAAGACACTTTGCAAGTCA
Sulf1	NM_134378	GTTGCAGGCAAAATCCAAGT	TTGCCAAATTCACCTTCTCC
Clu	AF314657	CAGCTGGCTAACCTCACACA	TGTGATGGGGTCAGAGTCAA
Wnt2	BF556985	CTCCCTCTGCTCTTGACCTG	GACCTGGCACATTGTCACAC
Tgfbi	BG379319	CTTTCCTGGACGATGGACTG	ACAGATCTGGCGGTCGTATC
Slit3	BF386446	ACATGGAGGATTGACGTGTG	ATTTCTCAGGGGTGATGGTG
Foxp1	AI072641	CCTGAAGGTTTGCTGTGCTT	CTGGCTACTGCCTGCACAT
Rsad2	AF134409	CCCAGAGCAATCACTGAGGT	GTCACTGCAGGACAGAAGCA

Suppl Table 2. Primers used for qRT-PCR analyses of steroidogenesis enzymes

Symbol	Accession No.	Forward primer	Reverse primer
Akr1c18	NM_138510	GGAGGCCATGGAGAAGTGTA	ATGGCATTCTACCTGGTTGC
Cyp11a1	NM_017286	ACCTATTCCGCTTTGCCTTT	CATGTTGAGCATGGGAACAC
Cyp17a1	NM_012753	GACCTGTCCACGCCTATCTT	TCGAACTTCTCCCTGCACTT
Cyp19a1	NM_017085	TTAACGAGAGCCTGCGGTAT	ACTCGAGCCTGTGCATTCTT
Hsd3b1	NM_001007719	GGTGCAGGAGAAAGAACTGC	TGGGCATCCAGAATATCTCC
Hsd17b2	NM_024391	GTCACCAAGCCAGAGCAGAT	AAGACCCCAGCATTGTTGAC

Suppl Table 3. Primers used for qRT-PCR analyses of decidual differentiation

Symbol	Accession No.	Forward primer	Reverse primer
Prl	NM_012629	ATCAATGACTGCCCCACTTC	ATTCCAGGAGTGCACCAAAC
Prl6a1	M31155	CCAACAGAGGCTGGGTGTAT	GGGGGTTCCTCCATATGACT
Prl8a2	NM_022846	ATCCAGCGAGCTGAAGTCAT	ATGCCTATACATGCGTGCAA
Prl3c1	NM_031316	GGATTCAGCCTGGAATTGAA	TTGCGCAAGCAGTAGAAGAA
Gja1	NM_012567	CCTTTGACTTCAGCCTCCAA	CTTGGACCTTGTCCAGAAGC

Suppl Table 4. Primers used for qRT-PCR analyses of P4 and E2 responsive genes

Symbol	Accession No.	Forward primer	Reverse primer
Ihh	NM_053384	GAGCTCACCCCCAACTACAA	TGACAGAGATGGCCAGTGAG
S100g	NM_012521	ATCCAAACCAGCTGTCCAAG	TCCATCACCGTTCTTATCCAG
Calca	NM_017338	CCTTTCCTGGTTGTCAGCAT	GGCGAGCTTCTTCTTCACTG
Areg	NM_017123	CCGGCTATATTGTGGACGAC	CCTGTTTCTTCTGCCTTTTCC

Suppl Table 5. Primers used for qRT-PCR analyses of the P4 signaling pathway

Symbol	Accession No.	Forward primer	Reverse primer
Pgr A/B	NM_022847	CTACCTGAGGCCAGATTCAG	CCTCTTAAAGAAGACCTTGCA
Pgr B	NM_022847	GAAGAAGCAGAAATCCCAGAG	CCAAAGAGACACCAAGAAGTG
Fkbp4	XM_342763	AGAAGCTGGAGCAGAGCAAC	GGACCTTTTGCATTTCCTCA
Hsp90	NM_175761	CTGCGTATTTGGTTGCTGAG	CATTGGTTCACCTGTGTCTG
Ncoa1	NM_001108012	AATCGACTAGCACCATCTCTG	ATGAACTTCACACCTGGGAG
Ncoa2	NM_031822	CAAAGTCCATGGTGAATGGG	ATCGTCTCGTATTTCTGATGTG

Suppl Table 6. *Pgr* coding sequence (PCR using cDNA)

Primer	Amplicon	Primer Sequence
+770 Fwd	+770 to +1739	GTCTCGCCAATACCGATCTC
+1739 Rev	+770 to +1739	CCTCTTTAGGGTCGCCTTCT
+1632Fwd	+1632 to +2606	AAGTCCCTTTTGCTCCACCT
+2606 Rev	+1632 to +2606	CCAGGGTCTGGCTCTCATTA
+2493 Fwd	+2493 to +3475	TGGTCCTTGGAGGTCGTAAG
+3475Rev	+2493 to +3475	GAGCTGACTGTCCTGACTGAGA

Suppl Table 7. Sequence upstream of transcription start site and 5' flanking region (PCR using genomic DNA)

Primer	Amplicon	Primer Sequence
-1308 Fwd	-1308 to -474	CCCAATGGTCCAATGTGACAG
-474 Rev	-1308 to -474	GTGGCTGGACAGGTAGCCAGTAA
-662 Fwd	-662 to +196	CCTCTGAAGGAGCAGCAAGT
+196 Rev	-662 to +196	CACCAAAACCCTGGGACTAAGA
+82 Fwd	+82 to +1060	AGACCAACCTGCAACCAGAACT
+1060 Rev	+82 to +1060	GCCCAAAGAGACACCAAGAAGT