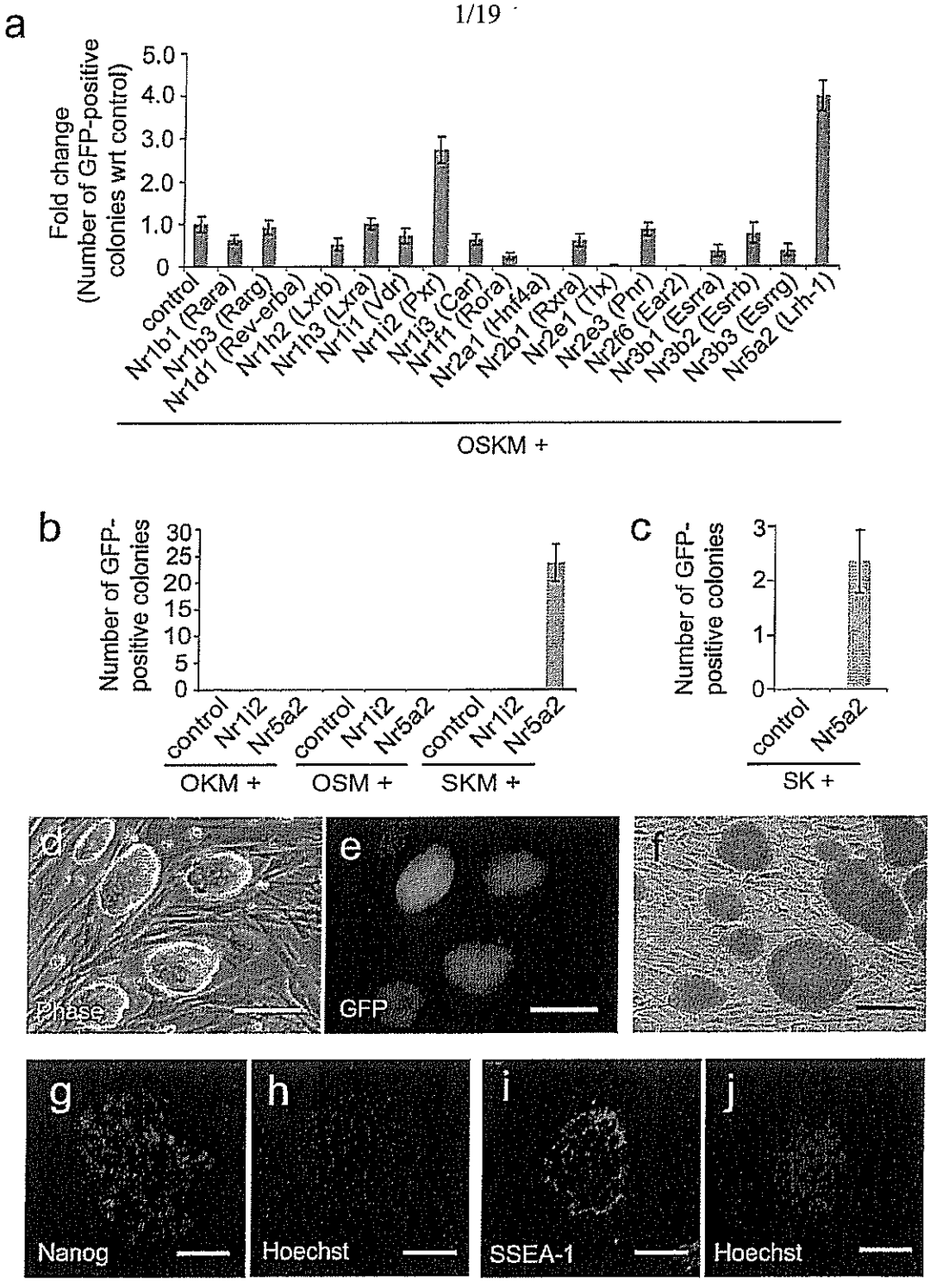
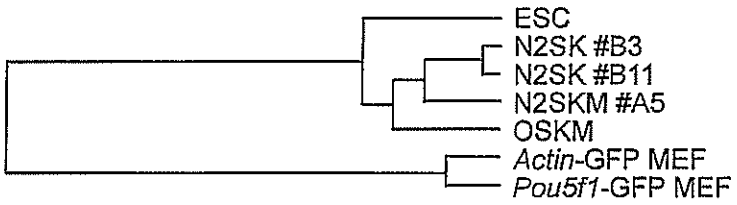

a
Fold change (Number of GFP-positive colonies wrt control)
5.0
4.0
3.0
2.0
1.0
0
control
Nr1b1 (Rara)
Nr1b3 (Rarg)
Nr1d1 (Rev-erba)
Nr1h2 (Lxrb)
Nr1h3 (Lxra)
Nr1i1 (Vdr)
Nr1i2 (Pxr)
Nr1i3 (Car)
Nr1f1 (Rora)
Nr2a1 (Hnf4a)
Nr2b1 (Rxra)
Nr2e1 (Tlx)
Nr2e3 (Pnr)
Nr2f6 (Ear2)
Nr3b1 (Esrra)
Nr3b2 (Esrrb)
Nr3b3 (Esrrg)
Nr5a2 (Lrh-1)
OSKM +
b
Number of GFP-positive colonies
30
25
20
15
10
5
0
control
Nr1i2
Nr5a2
OKM +
OSM +
SKM +
c
Number of GFP-positive colonies
3
2
1
0
control
Nr5a2
SK +
d
Phase
e
GFP
f
g
Nanog
h
Hoechst
i
SSEA-1
j
Hoechst

Figure 1

a


ESC
N2SK #B3
N2SK #B11
N2SKM #A5
OSKM
Actin-GFP MEF
Pou5f1-GFP MEF

b

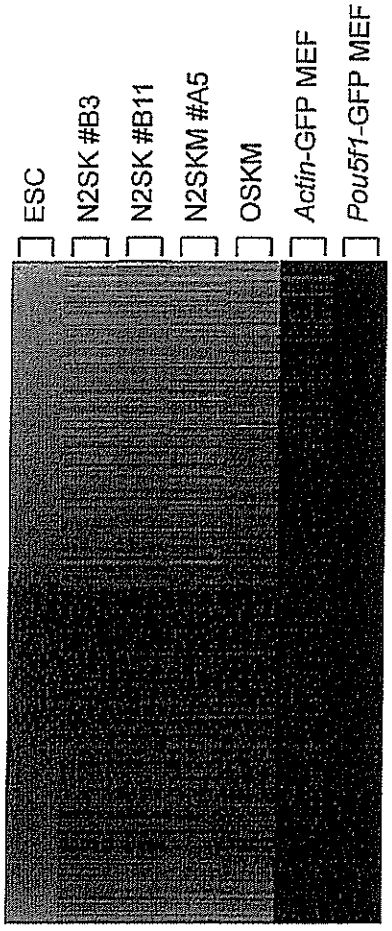

ESC
N2SK #B3
N2SK #B11
N2SKM #A5
OSKM
Actin-GFP MEF
Pou5f1-GFP MEF

Figure 2

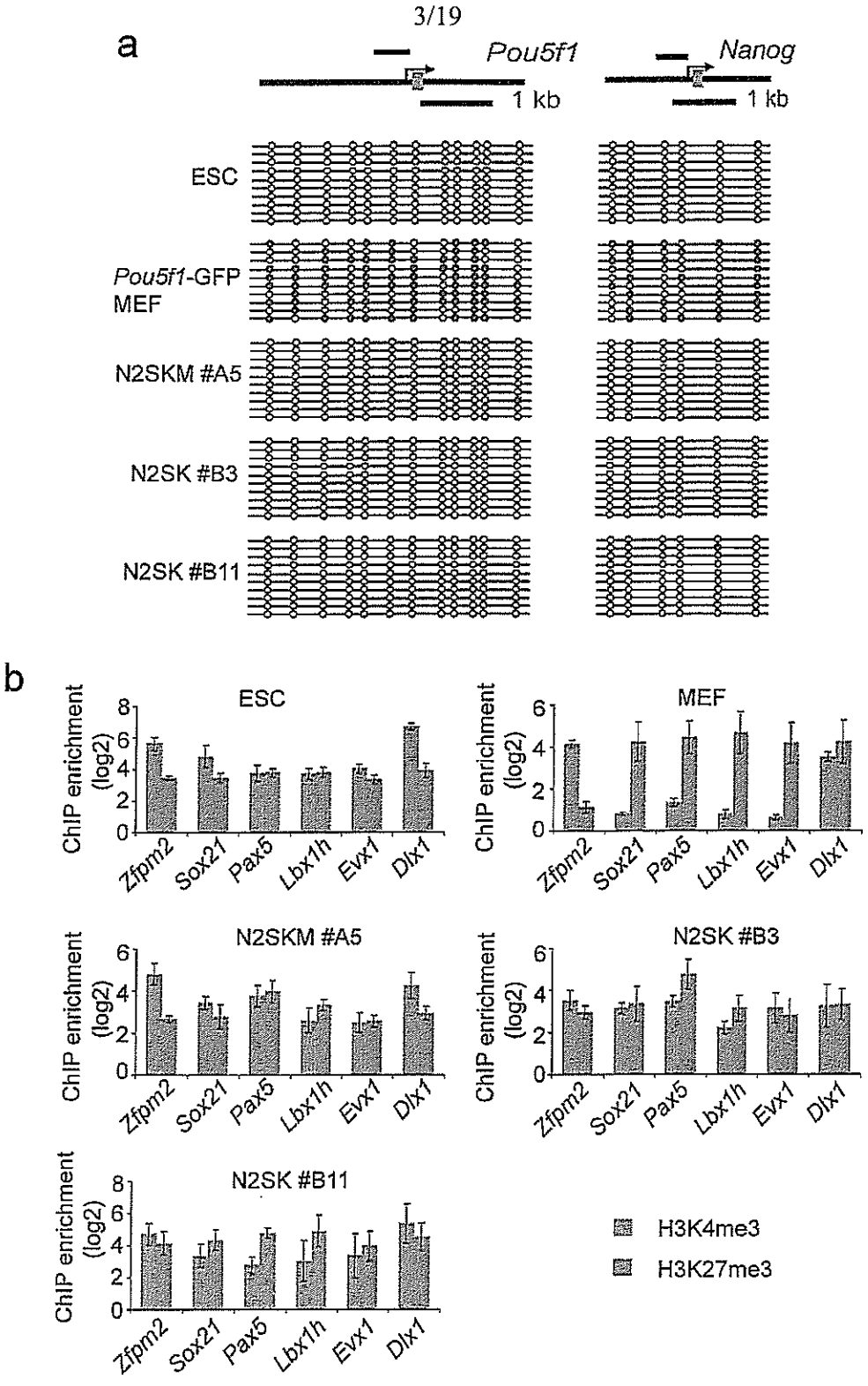

a
Pou5f1
Nanog
1 kb
1 kb
ESC
Pou5f1-GFP MEF
N2SKM #A5
N2SK #B3
N2SK #B11
b
ESC
MEF
N2SKM #A5
N2SK #B3
N2SK #B11
ChIP enrichment (log2)
Zfpm2
Sox21
Pax5
Lbx1h
Evx1
Dlx1
H3K4me3
H3K27me3

Figure 3

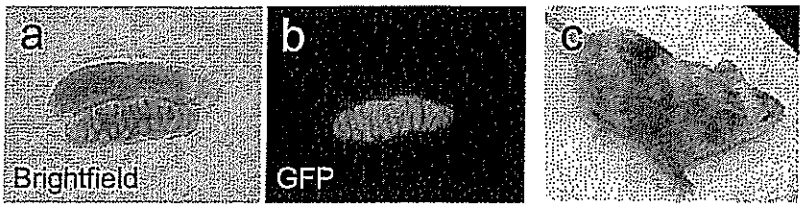


Figure 4

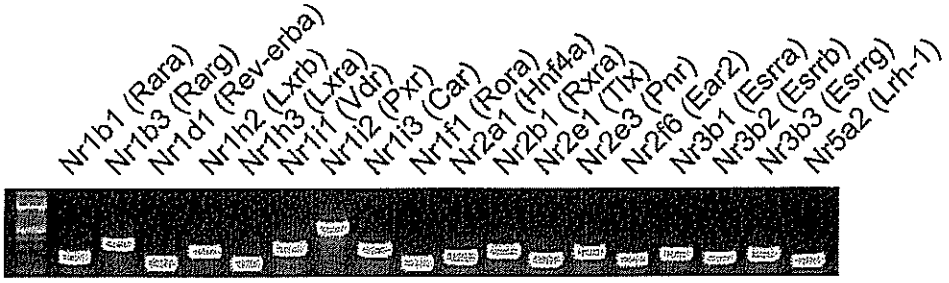
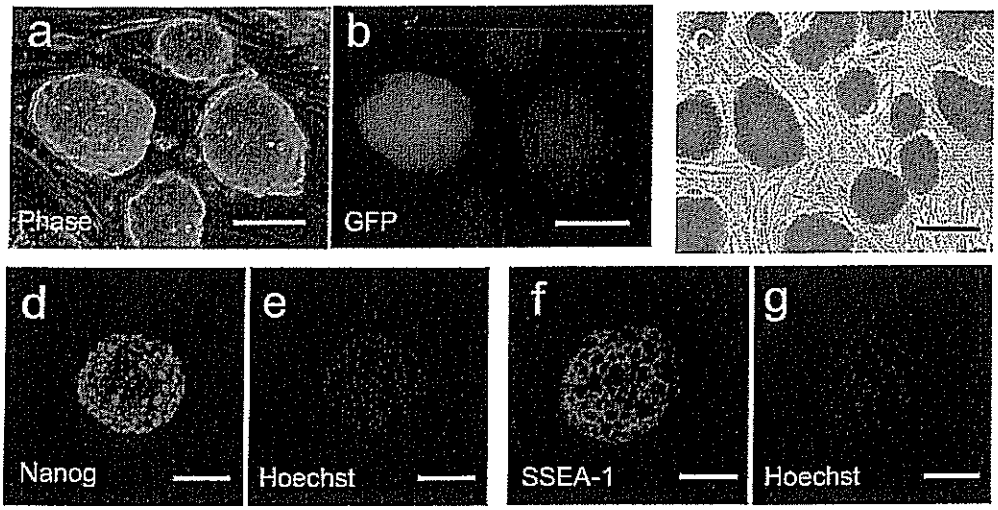
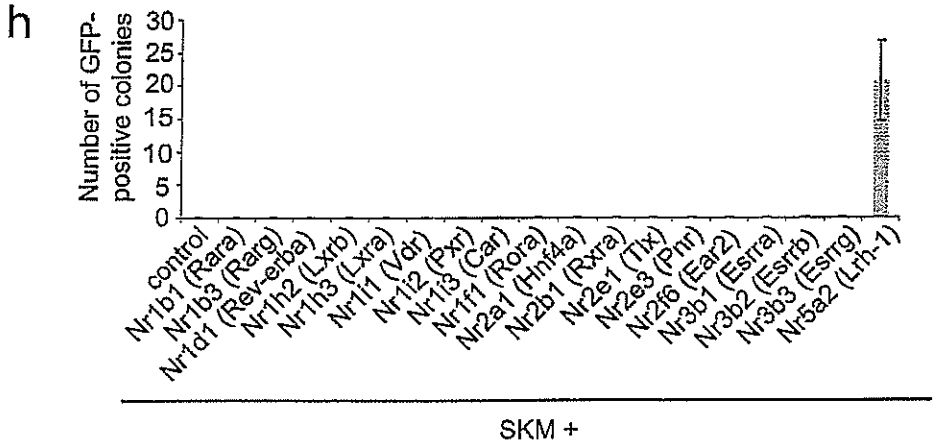

Nr1b1 (Rara)
Nr1b3 (Rarg)
Nr1d1 (Rev-erba)
Nr1h2 (Lxrb)
Nr1h3 (Lxra)
Nr1i1 (Vdr)
Nr1i2 (Pxr)
Nr1i3 (Car)
Nr1f1 (Rora)
Nr2a1 (Hnf4a)
Nr2b1 (Rxra)
Nr2e1 (Tlx)
Nr2e3 (Pnr)
Nr2f6 (Ear2)
Nr3b1 (Esrra)
Nr3b2 (Esrrb)
Nr3b3 (Esrrg)
Nr5a2 (Lrh-1)

Figure 5


a
Phase
b
GFP
c
d
Nanog
e
Hoechst
f
SSEA-1
g
Hoechst


h
Number of GFP-positive colonies
30
25
20
15
10
5
0
control
Nr1b1 (Rara)
Nr1b3 (Rarg)
Nr1d1 (Rev-erba)
Nr1h2 (Lxrb)
Nr1h3 (Lxra)
Nr1i1 (Vdr)
Nr1i2 (Pxr)
Nr1i3 (Car)
Nr1f1 (Rora)
Nr2a1 (Hnf4a)
Nr2b1 (Rxra)
Nr2e1 (Tlx)
Nr2e3 (Pnr)
Nr2f6 (Ear2)
Nr3b1 (Esrra)
Nr3b2 (Esrrb)
Nr3b3 (Esrrg)
Nr5a2 (Lrh-1)
SKM +

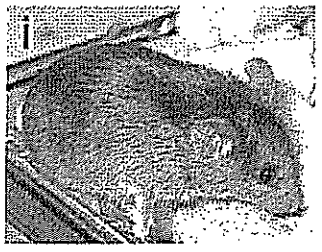

i

Figure 6

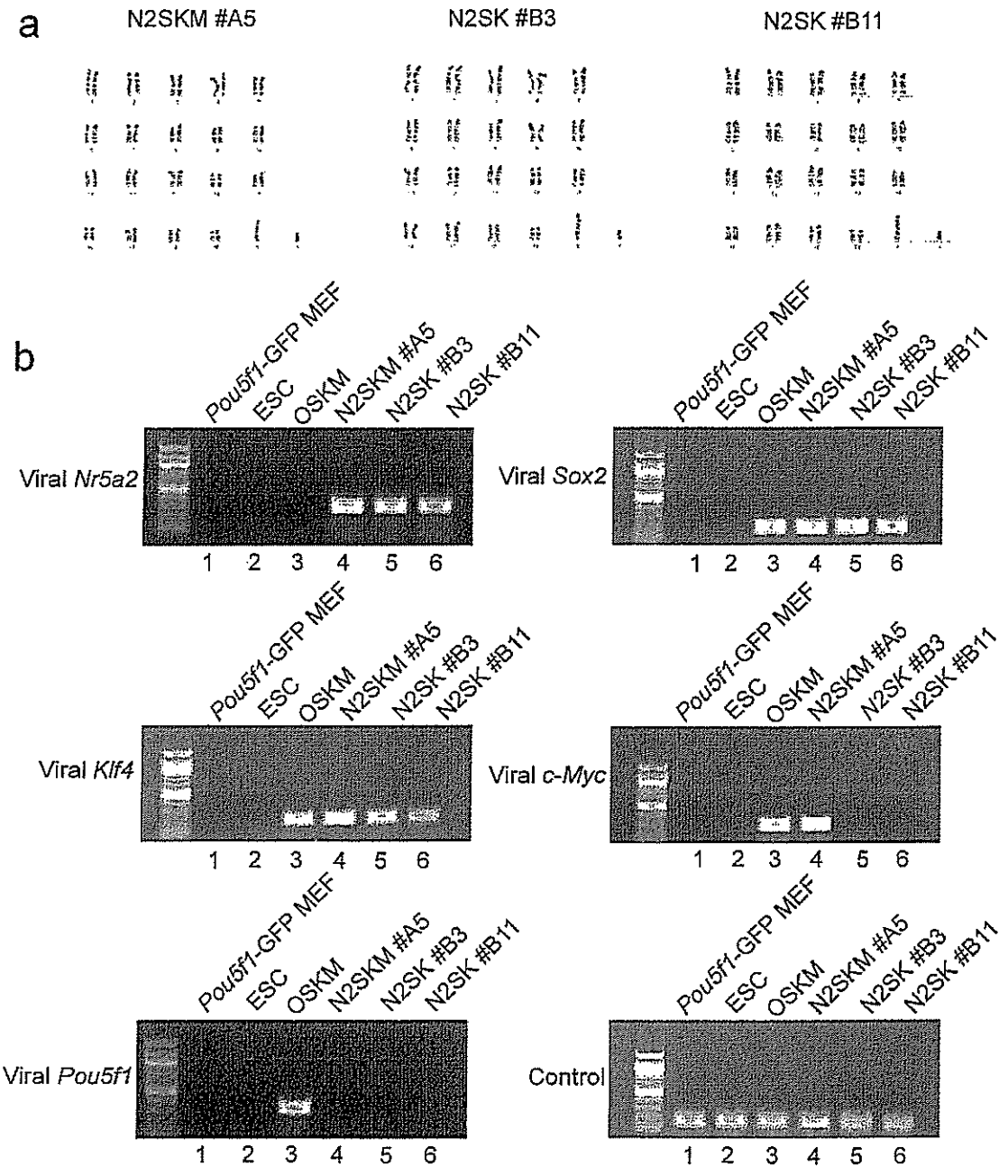

a
N2SKM #A5
N2SK #B3
N2SK #B11
b
Pou5f1-GFP MEF
ESC
OSKM
N2SKM #A5
N2SK #B3
N2SK #B11
Viral Nr5a2
Viral Sox2
Viral Klf4
Viral c-Myc
Viral Pou5f1
Control
1 2 3 4 5 6

Figure 7

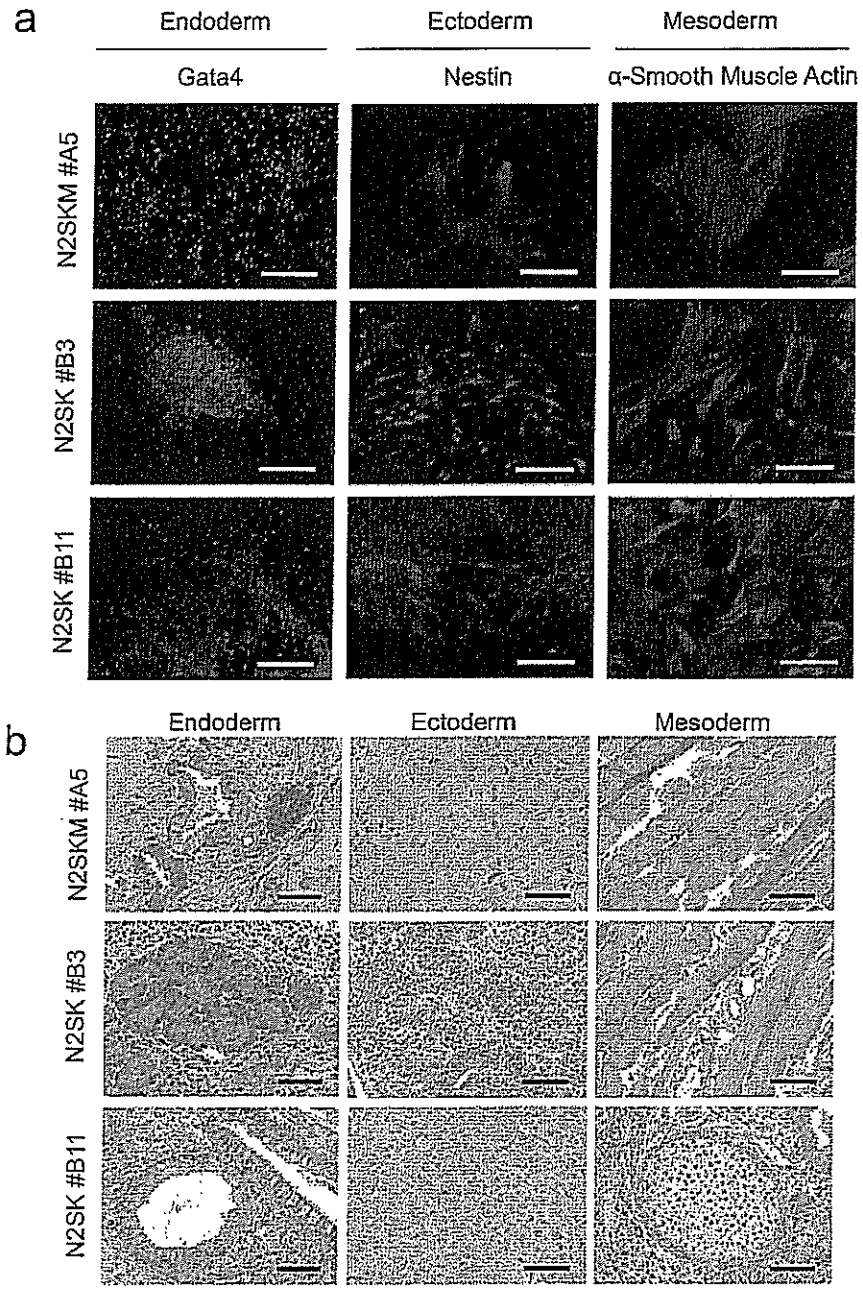

a
Endoderm
Ectoderm
Mesoderm
Gata4
Nestin
α-Smooth Muscle Actin
N2SKM #A5
N2SK #B3
N2SK #B11
b
Endoderm
Ectoderm
Mesoderm
N2SKM #A5
N2SK #B3
N2SK #B11

Figure 8

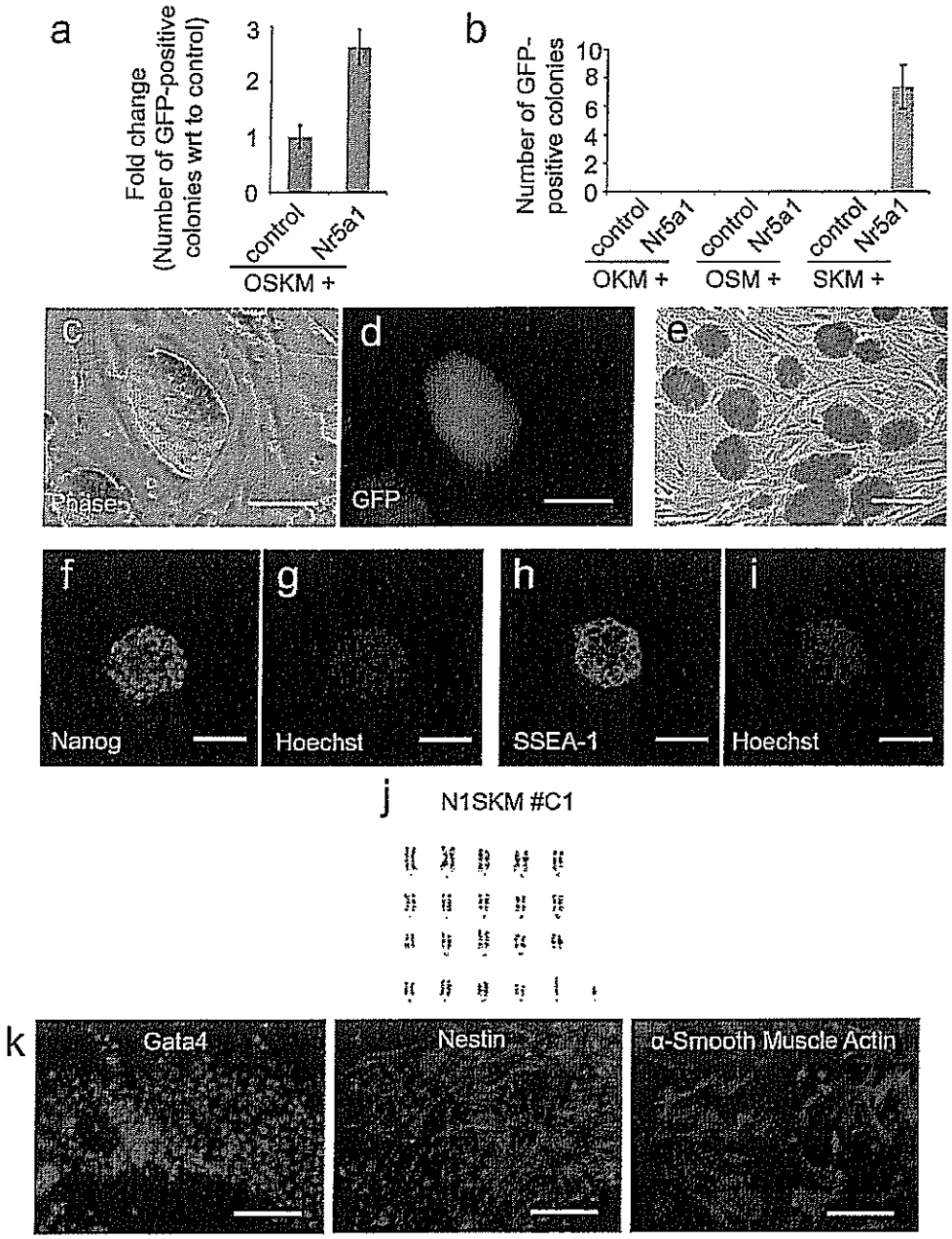

a
Fold change (Number of GFP-positive colonies wrt to control)
3
2
1
0
control
Nr5a1
OSKM +
b
Number of GFP-positive colonies
10
8
6
4
2
0
control
Nr5a1
control
Nr5a1
control
Nr5a1
OKM +
OSM +
SKM +
c
Phase
d
GFP
e
f
Nanog
g
Hoechst
h
SSEA-1
i
Hoechst
j
N1SKM #C1
k
Gata4
Nestin
α-Smooth Muscle Actin

Figure 9

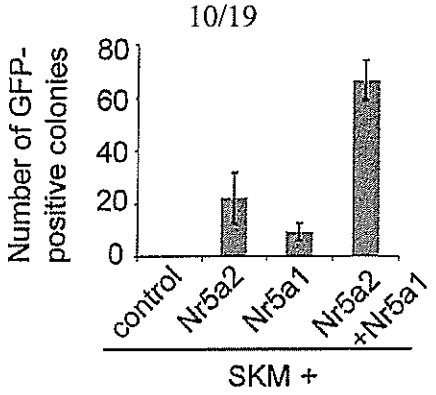

Number of GFP-positive colonies
80
60
40
20
0
control
Nr5a2
Nr5a1
Nr5a2 +Nr5a1
SKM +

Figure 10

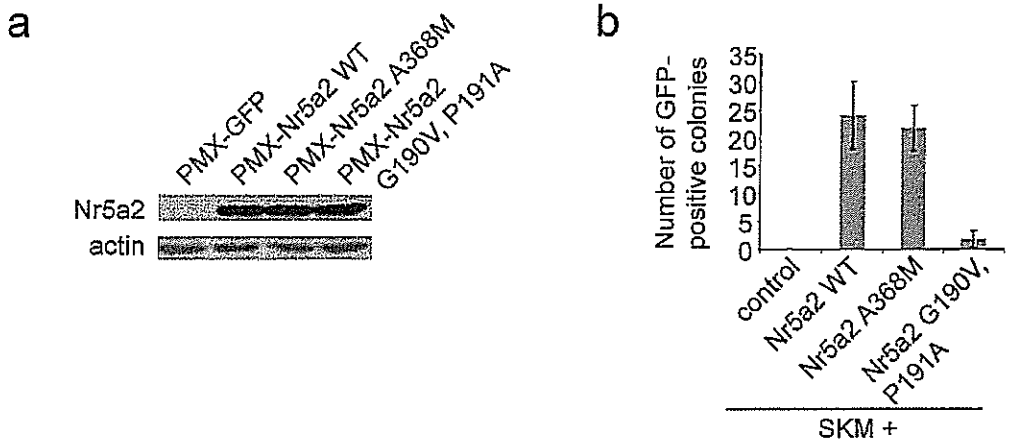

a
PMX-GFP
PMX-Nr5a2 WT
PMX-Nr5a2 A368M
PMX-Nr5a2 G190V, P191A
Nr5a2
actin
b
Number of GFP-positive colonies
35
30
25
20
15
10
5
0
control
Nr5a2 WT
Nr5a2 A368M
Nr5a2 G190V, P191A
SKM +

Figure 11

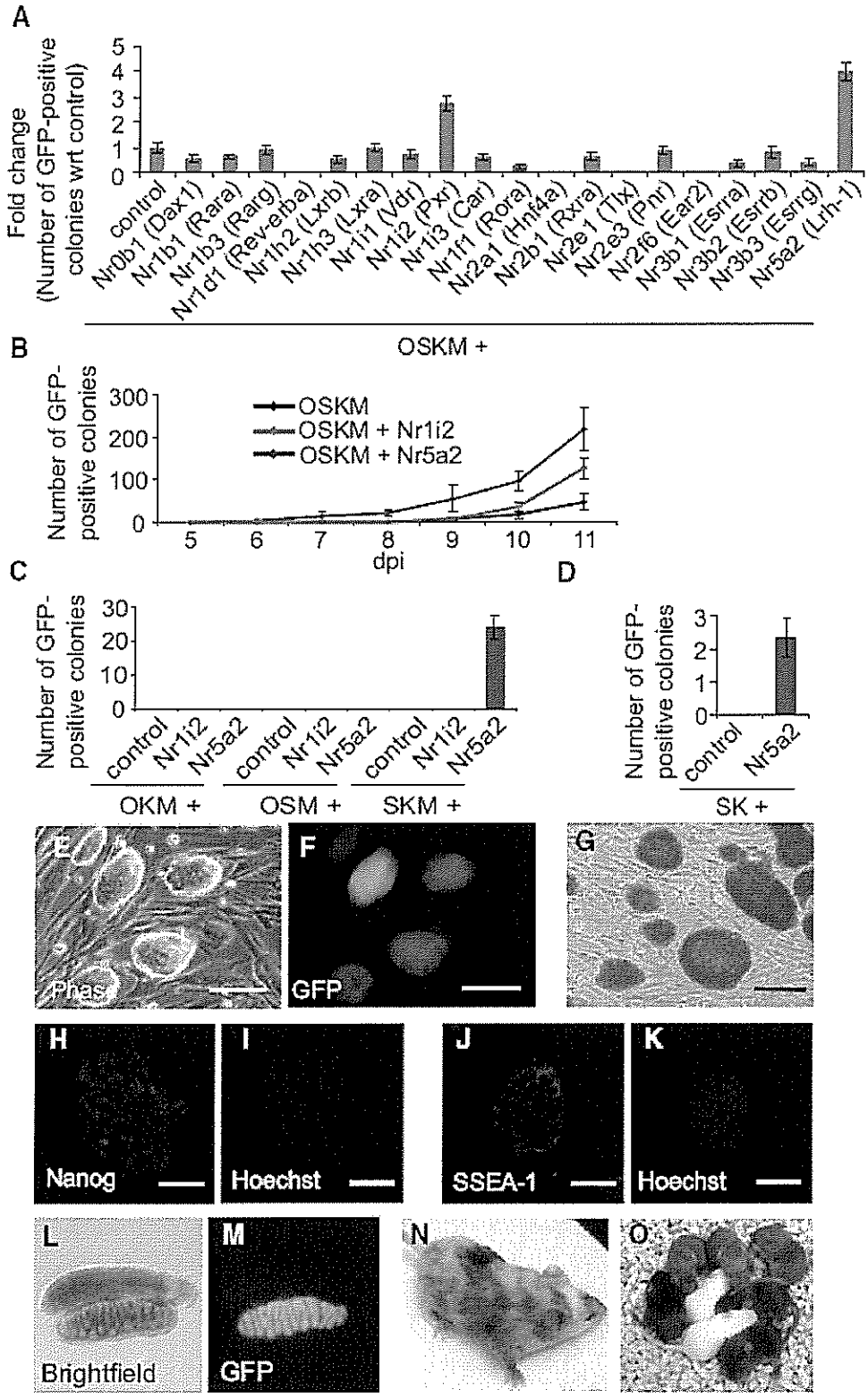

A
Fold change (Number of GFP-positive colonies wrt control)
control
Nr0b1 (Dax1)
Nr1b1 (Rara)
Nr1b3 (Rarg)
Nr1d1 (Rev-erba)
Nr1h2 (Lxrb)
Nr1h3 (Lxra)
Nr1i1 (Vdr)
Nr1i2 (Pxr)
Nr1i3 (Car)
Nr1f1 (Rora)
Nr2a1 (Hnf4a)
Nr2b1 (Rxra)
Nr2e1 (Tlx)
Nr2e3 (Pnr)
Nr2f6 (Ear2)
Nr3b1 (Esrra)
Nr3b2 (Esrrb)
Nr3b3 (Esrrg)
Nr5a2 (Lrh-1)
OSKM +
B
Number of GFP-positive colonies
OSKM
OSKM + Nr1i2
OSKM + Nr5a2
dpi
C
Number of GFP-positive colonies
control
Nr1i2
Nr5a2
OKM +
OSM +
SKM +
D
Number of GFP-positive colonies
control
Nr5a2
SK +
E
Phase
F
GFP
G
H
Nanog
I
Hoechst
J
SSEA-1
K
Hoechst
L
Brightfield
M
GFP
N
O

Figure 12

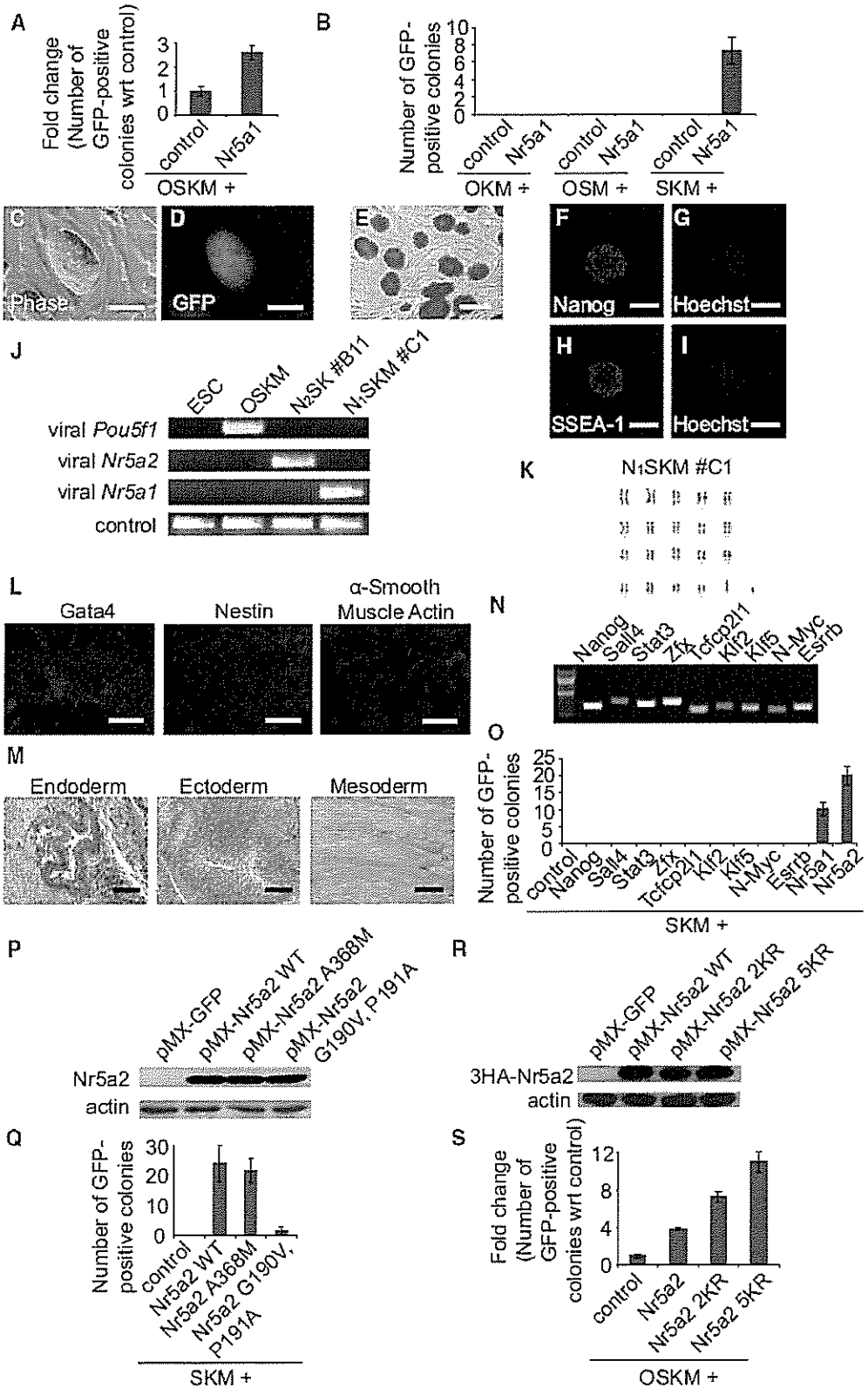

A
Fold change (Number of GFP-positive colonies wrt control)
control
Nr5a1
OSKM +
B
Number of GFP-positive colonies
control
Nr5a1
OKM +
OSM +
SKM +
C
Phase
D
GFP
E
F
Nanog
G
Hoechst
H
SSEA-1
I
Hoechst
J
ESC
OSKM
N2SK #B11
N1SKM #C1
viral Pou5f1
viral Nr5a2
viral Nr5a1
control
K
N1SKM #C1
L
Gata4
Nestin
α-Smooth Muscle Actin
M
Endoderm
Ectoderm
Mesoderm
N
Nanog
Sall4
Stat3
Zfx
Tcfcp2l1
Klf2
Klf5
N-Myc
Esrrb
O
Number of GFP-positive colonies
control
Nanog
Sall4
Stat3
Zfx
Tcfcp2l1
Klf2
Klf5
N-Myc
Esrrb
Nr5a1
Nr5a2
SKM +
P
pMX-GFP
pMX-Nr5a2 WT
pMX-Nr5a2 A368M
pMX-Nr5a2 G190V, P191A
Nr5a2
actin
R
pMX-GFP
pMX-Nr5a2 WT
pMX-Nr5a2 2KR
pMX-Nr5a2 5KR
3HA-Nr5a2
actin
Q
Number of GFP-positive colonies
control
Nr5a2 WT
Nr5a2 A368M
Nr5a2 G190V, P191A
SKM +
S
Fold change (Number of GFP-positive colonies wrt control)
control
Nr5a2
Nr5a2 2KR
Nr5a2 5KR
OSKM +

Figure 13

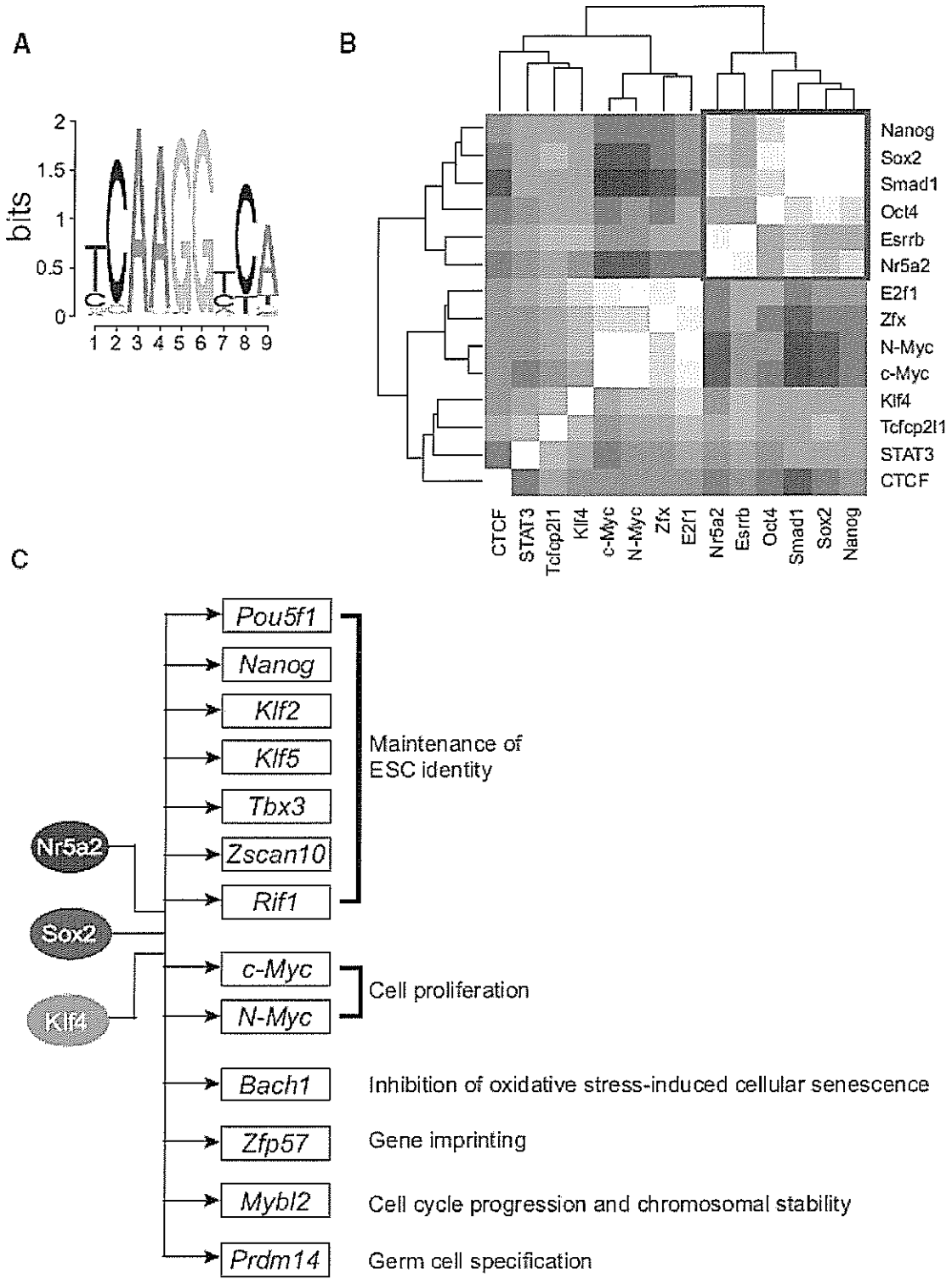

A
bits
2
1.5
1
0.5
0
1 2 3 4 5 6 7 8 9
B
Nanog
Sox2
Smad1
Oct4
Esrrb
Nr5a2
E2f1
Zfx
N-Myc
c-Myc
Klf4
Tcfcp2l1
STAT3
CTCF
CTCF
STAT3
Tcfcp2l1
Klf4
c-Myc
N-Myc
Zfx
E2f1
Nr5a2
Esrrb
Oct4
Smad1
Sox2
Nanog
C
Nr5a2
Sox2
Klf4
Pou5f1
Nanog
Klf2
Klf5
Tbx3
Zscan10
Rif1
c-Myc
N-Myc
Bach1
Zfp57
Mybl2
Prdm14
Maintenance of ESC identity
Cell proliferation
Inhibition of oxidative stress-induced cellular senescence
Gene imprinting
Cell cycle progression and chromosomal stability
Germ cell specification

Figure 14

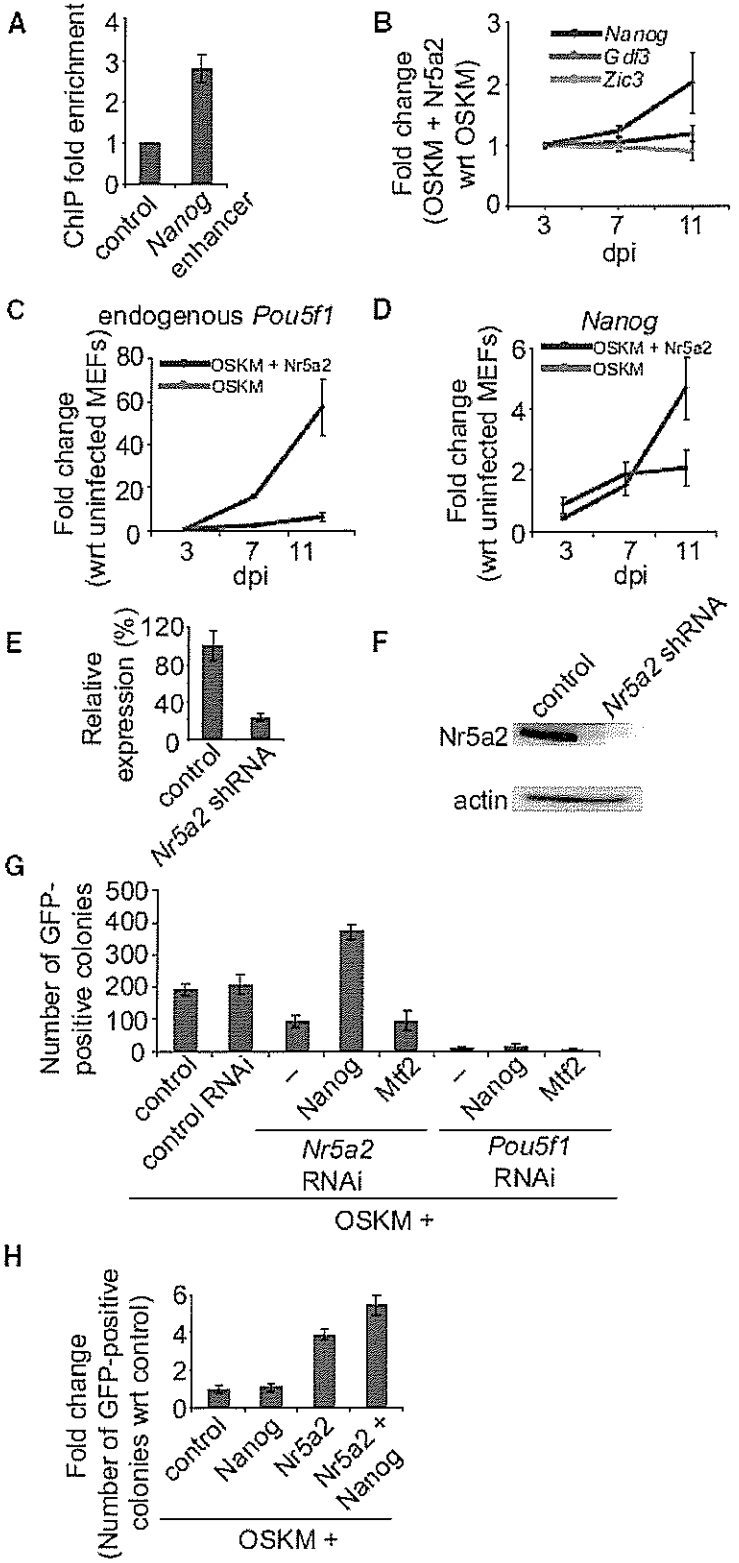

A
ChIP fold enrichment
control
Nanog enhancer
B
Fold change (OSKM + Nr5a2 wrt OSKM)
Nanog
Gdf3
Zic3
dpi
C
endogenous Pou5f1
Fold change (wrt uninfected MEFs)
OSKM + Nr5a2
OSKM
dpi
D
Nanog
Fold change (wrt uninfected MEFs)
OSKM + Nr5a2
OSKM
dpi
E
Relative expression (%)
control
Nr5a2 shRNA
F
control
Nr5a2 shRNA
Nr5a2
actin
G
Number of GFP-positive colonies
control
control RNAi
–
Nanog
Mtf2
Nr5a2 RNAi
Pou5f1 RNAi
OSKM +
H
Fold change (Number of GFP-positive colonies wrt control)
control
Nanog
Nr5a2
Nr5a2 + Nanog
OSKM +

Figure 15

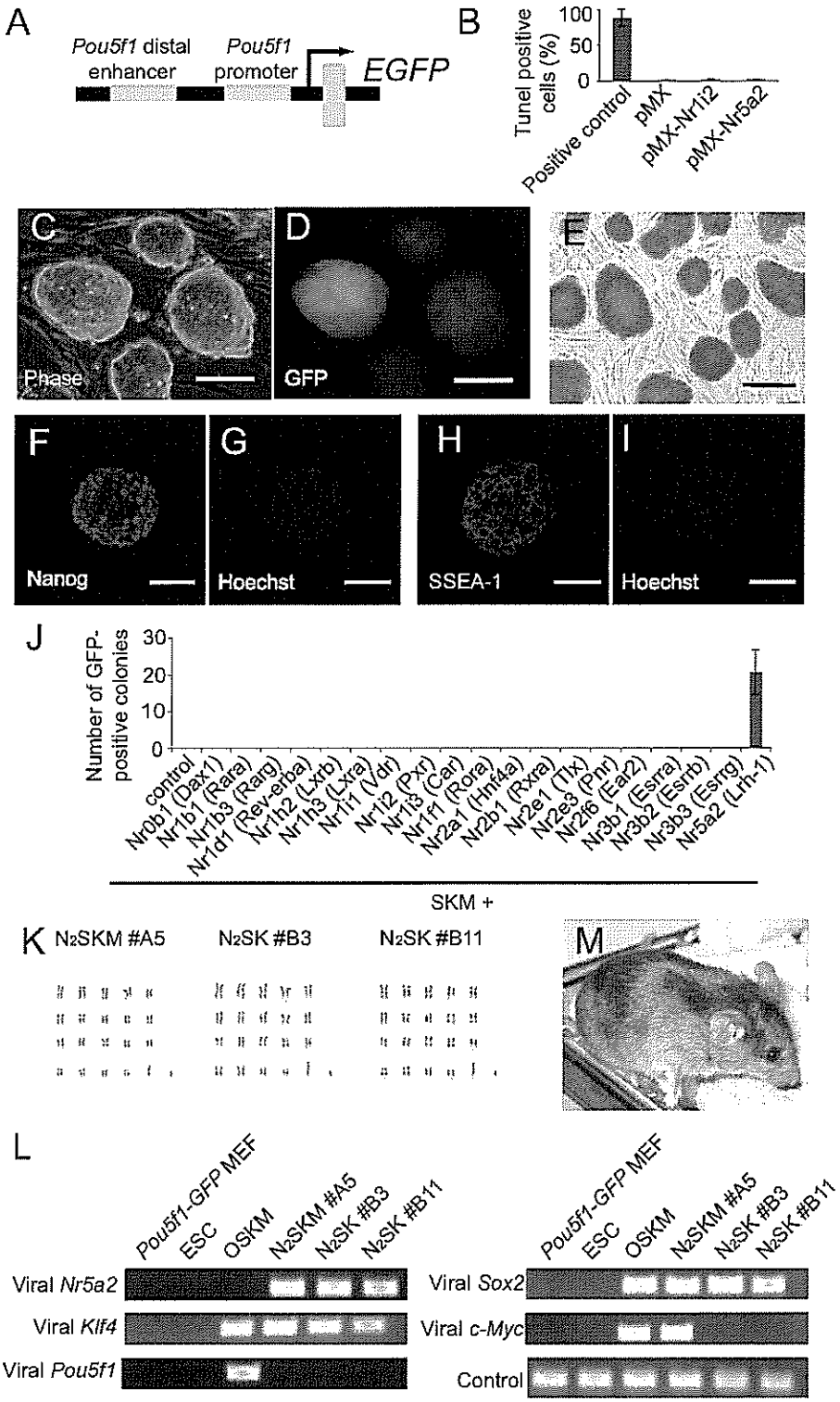

A
Pou5f1 distal enhancer
Pou5f1 promoter
EGFP
B
Tunel positive cells (%)
100
50
0
Positive control
pMX
pMX-Nr1i2
pMX-Nr5a2
C
Phase
D
GFP
E
F
Nanog
G
Hoechst
H
SSEA-1
I
Hoechst
J
Number of GFP-positive colonies
30
20
10
0
control
Nr0b1 (Dax1)
Nr1b1 (Rara)
Nr1b3 (Rarg)
Nr1d1 (Rev-erba)
Nr1h2 (Lxrb)
Nr1h3 (Lxra)
Nr1i1 (Vdr)
Nr1i2 (Pxr)
Nr1i3 (Car)
Nr1f1 (Rora)
Nr2a1 (Hnf4a)
Nr2b1 (Rxra)
Nr2e1 (Tlx)
Nr2e3 (Pnr)
Nr2f6 (Ear2)
Nr3b1 (Esrra)
Nr3b2 (Esrrb)
Nr3b3 (Esrrg)
Nr5a2 (Lrh-1)
SKM +
K
N2SKM #A5
N2SK #B3
N2SK #B11
M
L
Pou5f1-GFP MEF
ESC
OSKM
N2SKM #A5
N2SK #B3
N2SK #B11
Viral Nr5a2
Viral Klf4
Viral Pou5f1
Viral Sox2
Viral c-Myc
Control

Figure 16

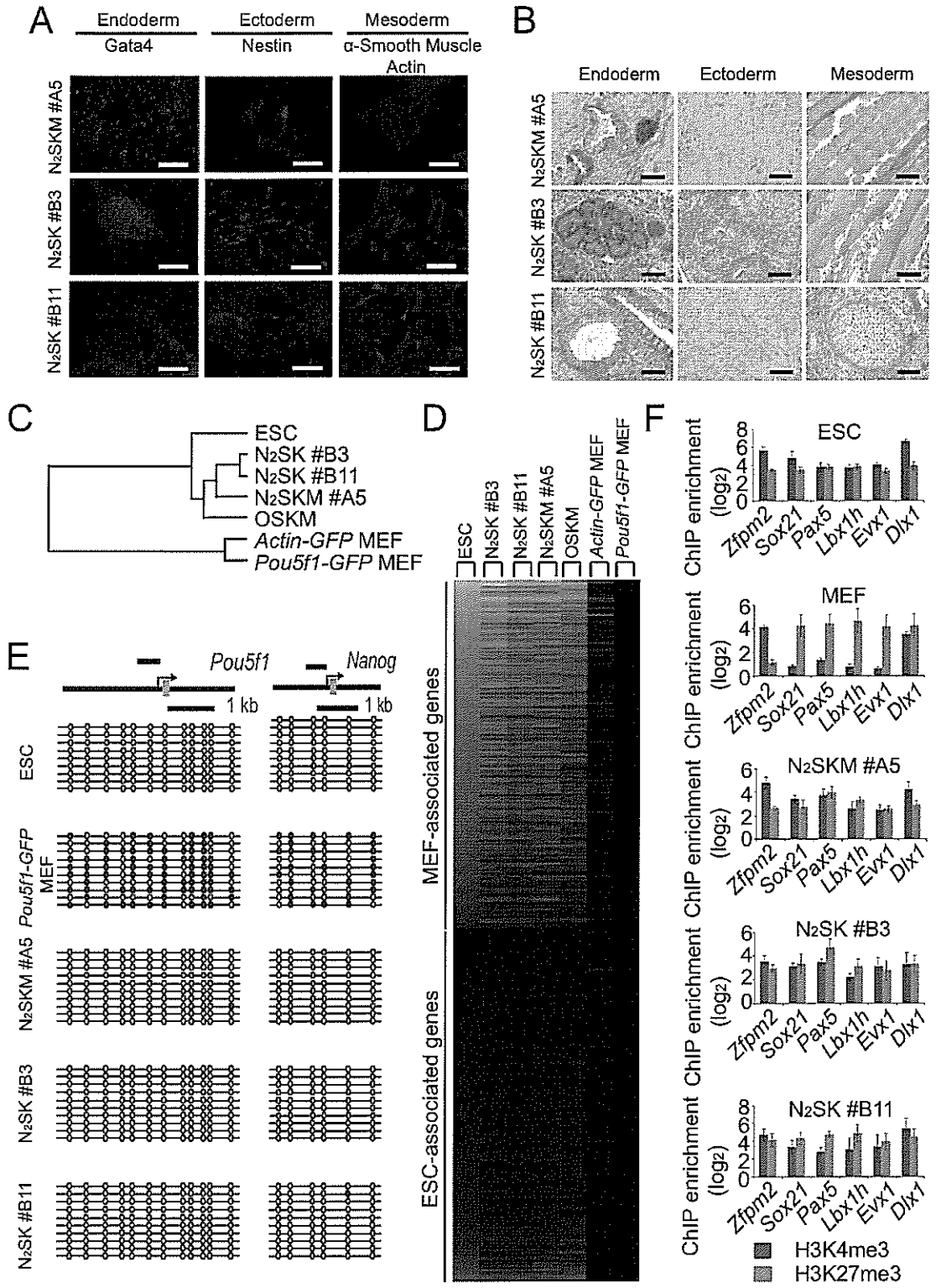

A
Endoderm
Ectoderm
Mesoderm
Gata4
Nestin
α-Smooth Muscle Actin
N2SKM #A5
N2SK #B3
N2SK #B11
B
Endoderm
Ectoderm
Mesoderm
N2SKM #A5
N2SK #B3
N2SK #B11
C
ESC
N2SK #B3
N2SK #B11
N2SKM #A5
OSKM
Actin-GFP MEF
Pou5f1-GFP MEF
D
ESC
N2SK #B3
N2SK #B11
N2SKM #A5
OSKM
Actin-GFP MEF
Pou5f1-GFP MEF
MEF-associated genes
ESC-associated genes
E
Pou5f1
Nanog
1 kb
1 kb
ESC
Pou5f1-GFP MEF
N2SKM #A5
N2SK #B3
N2SK #B11
F
ESC
MEF
N2SKM #A5
N2SK #B3
N2SK #B11
ChIP enrichment (log2)
Zfpm2
Sox21
Pax5
Lbx1h
Evx1
Dlx1
H3K4me3
H3K27me3

Figure 17

Figure 18

A

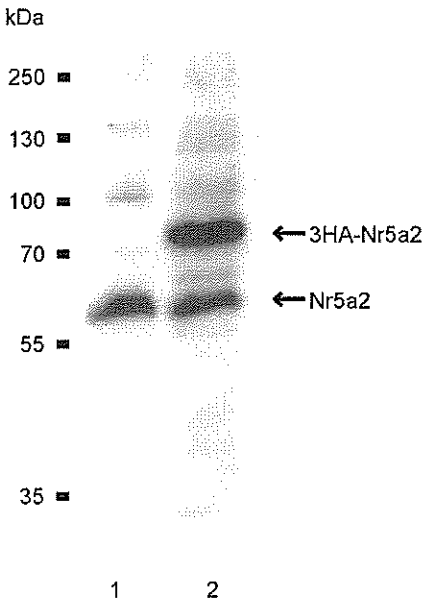
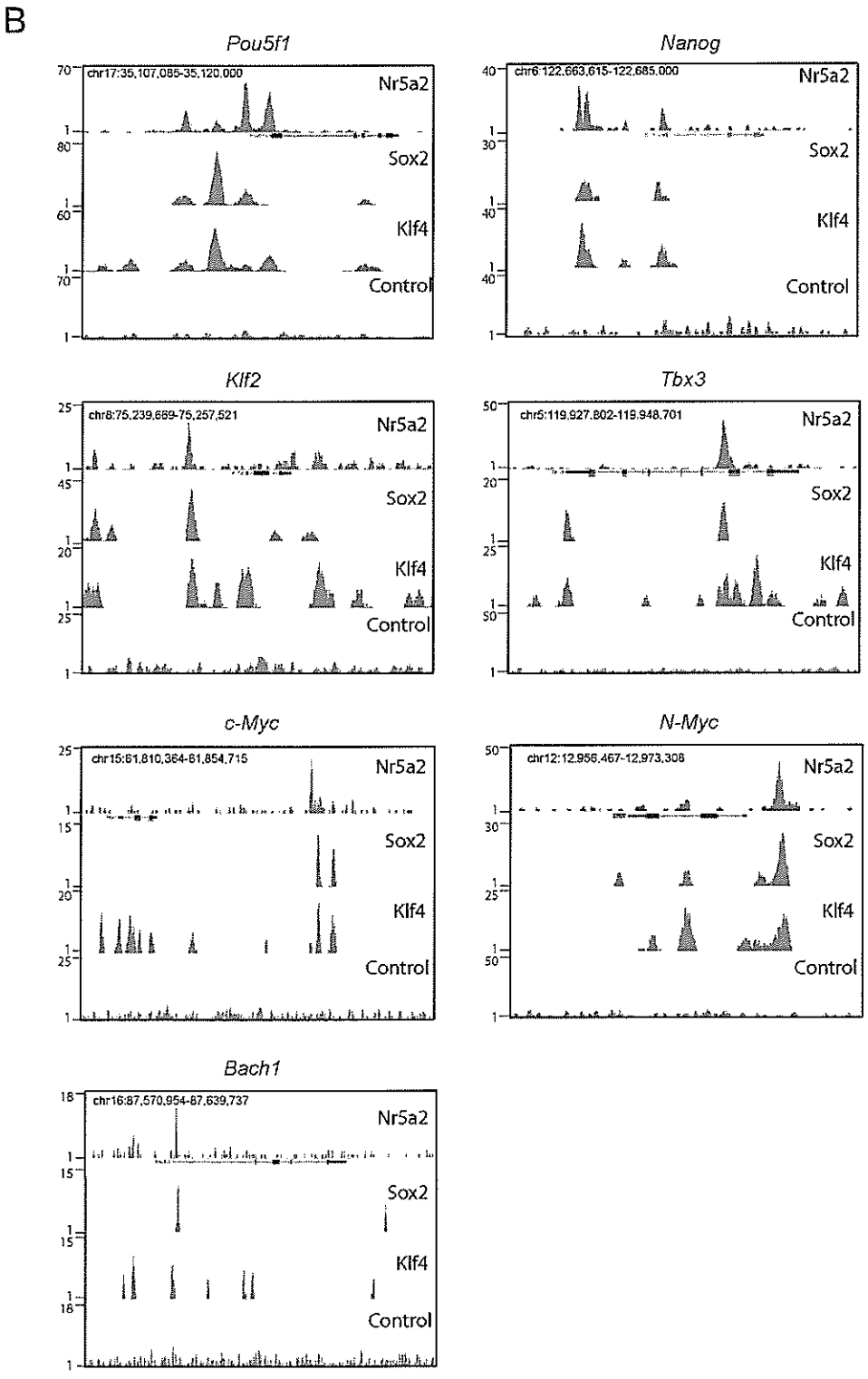


Figure 18


B
Pou5f1
chr17:35,107,085-35,120,000
Nr5a2
Sox2
Klf4
Control
Nanog
chr6:122,663,615-122,685,000
Klf2
chr8:75,239,689-75,257,521
Tbx3
chr5:119,927,802-119,948,701
c-Myc
chr15:61,810,364-61,854,715
N-Myc
chr12:12,956,467-12,973,308
Bach1
chr16:87,570,954-87,639,737