FIGURE 1A

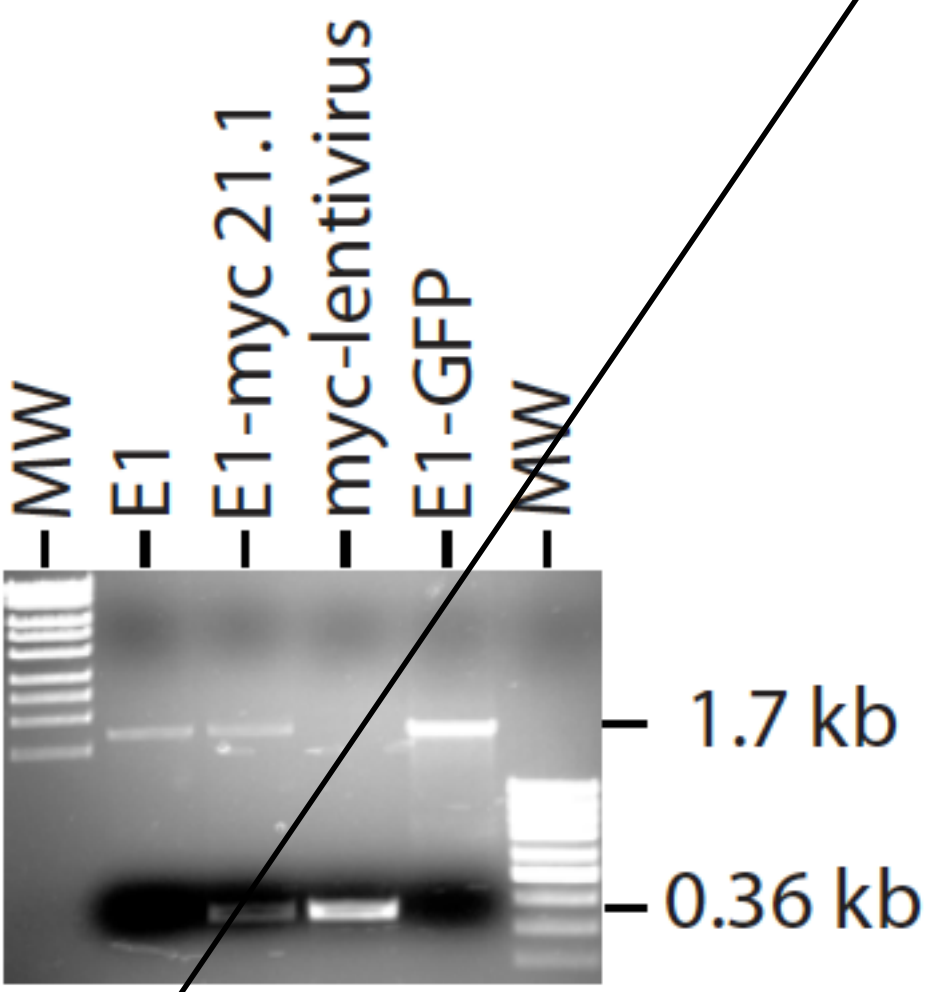


FIGURE 1B

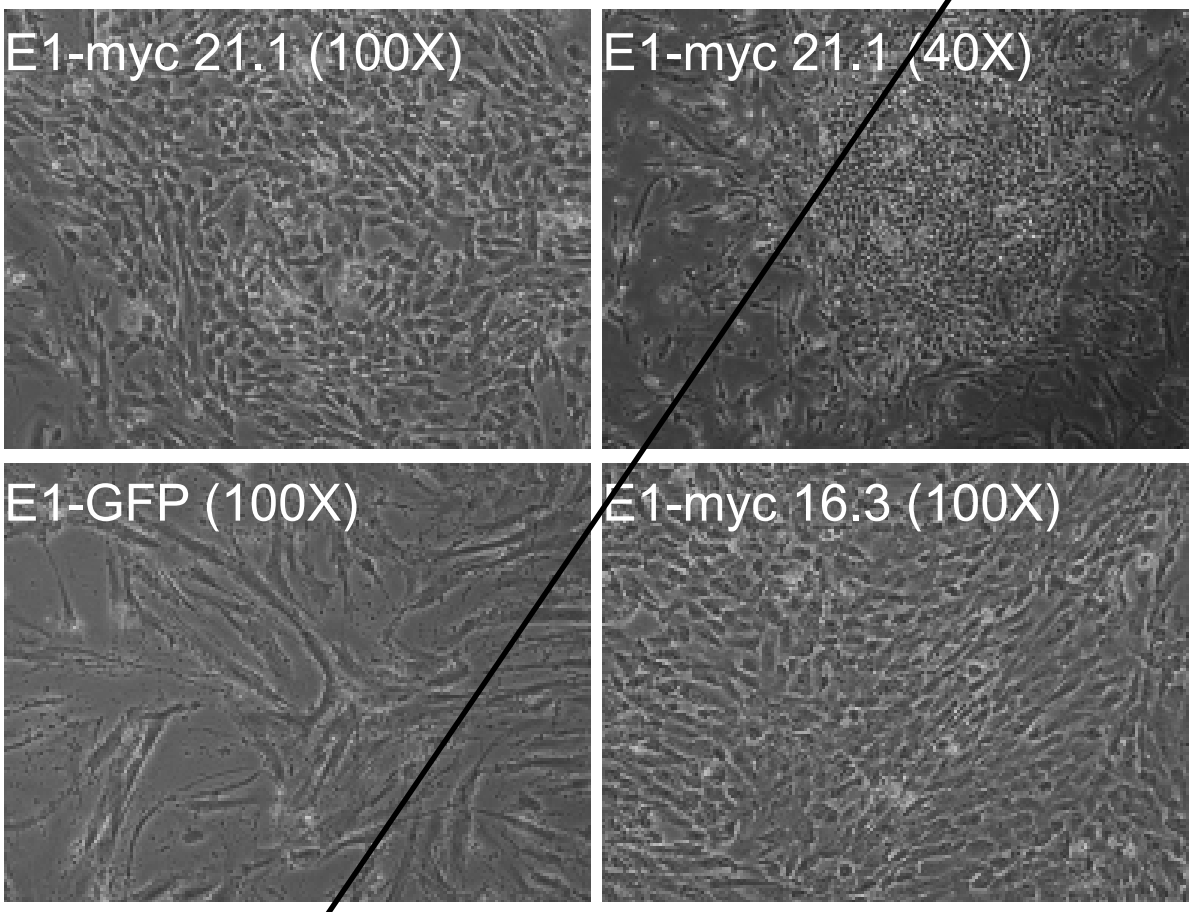


FIGURE 1C

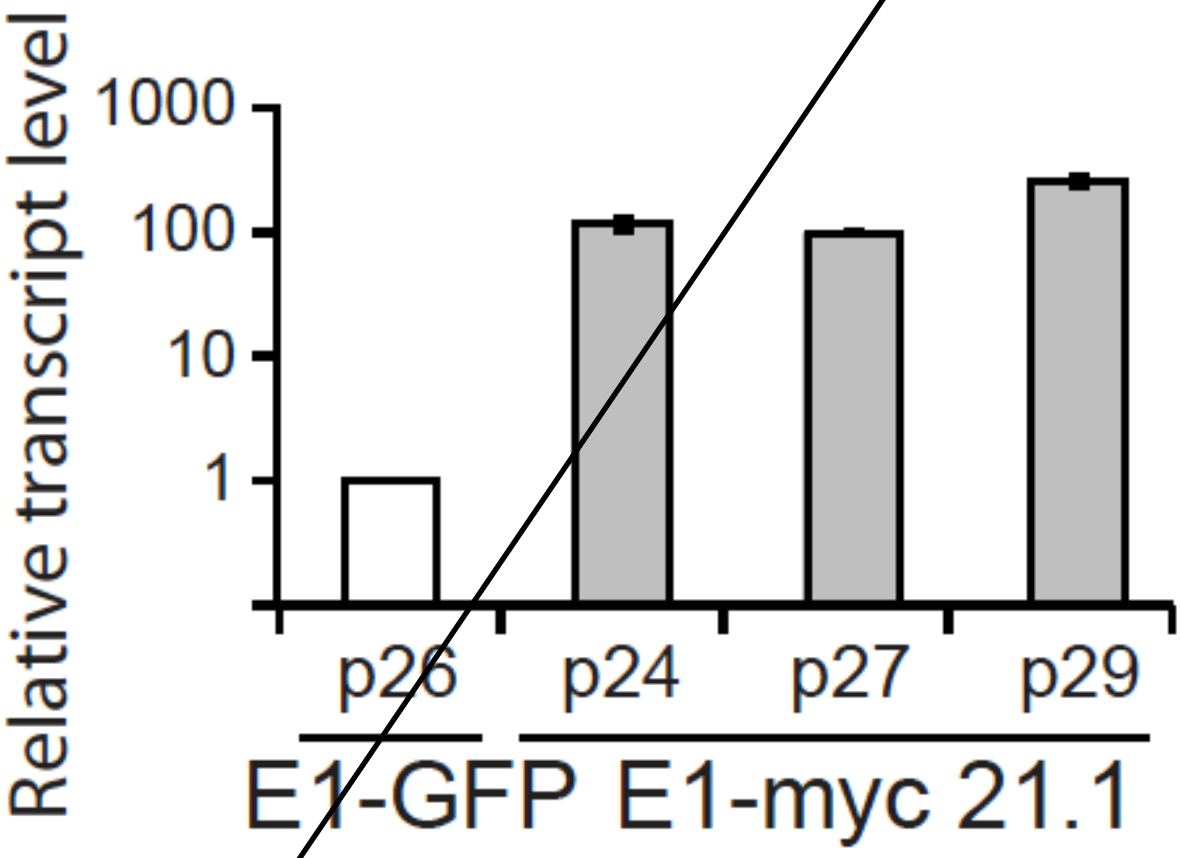

Relative transcript level
1000
100
10
1
p26
p24
p27
p29
E1-GFP
E1-myc 21.1

FIGURE 1D

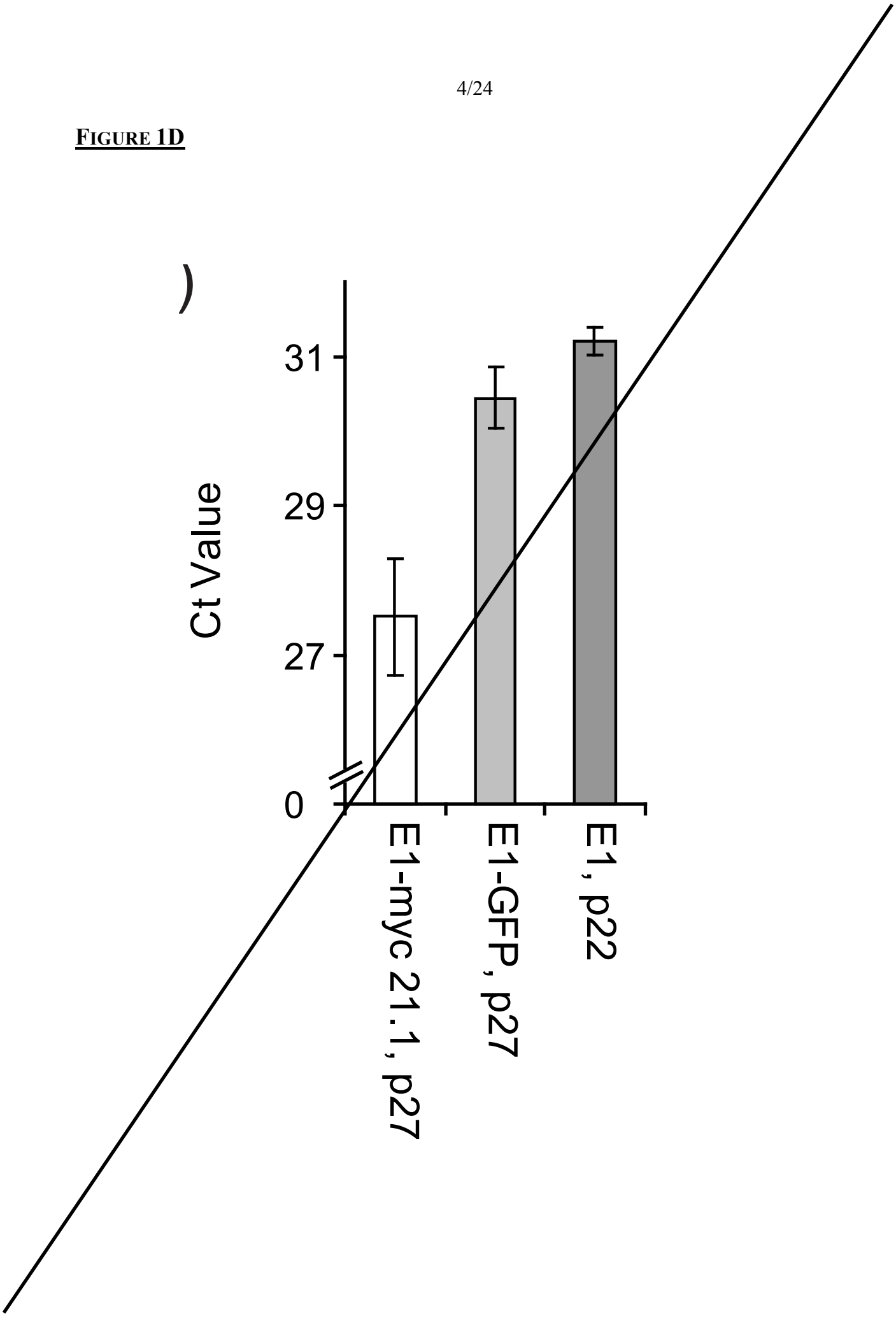


FIGURE 1E

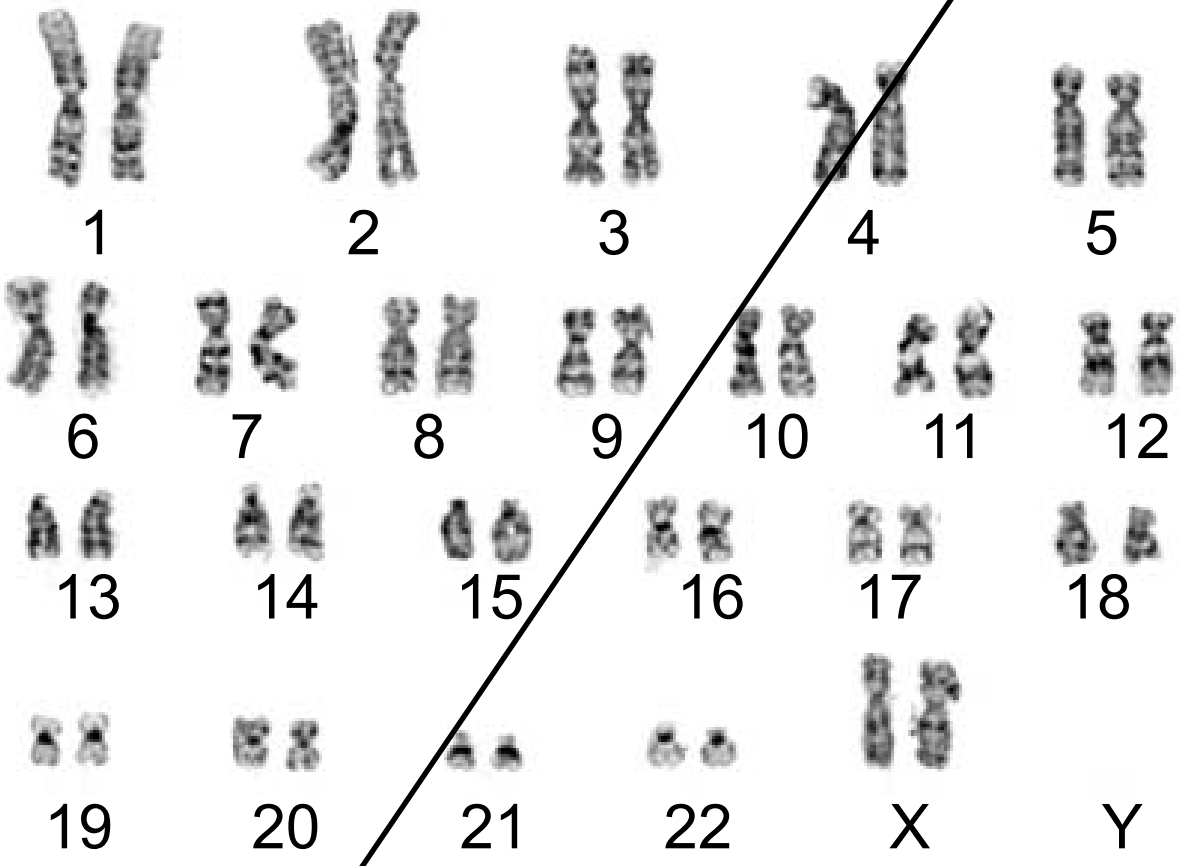


FIGURE 1F

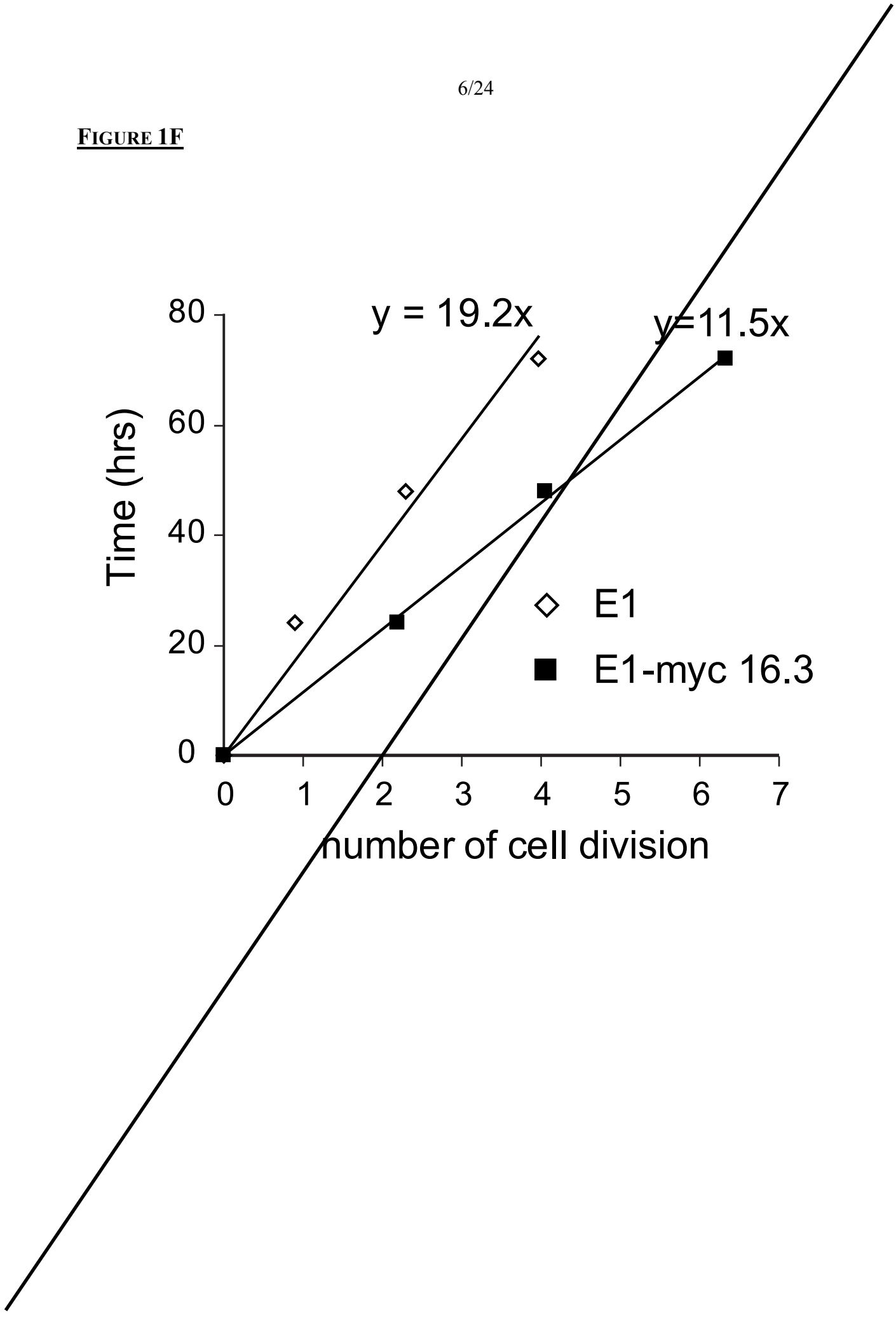

y = 19.2x
y=11.5x
Time (hrs)
80
60
40
20
0
0
1
2
3
4
5
6
7
number of cell division
E1
E1-myc 16.3

FIGURE 2

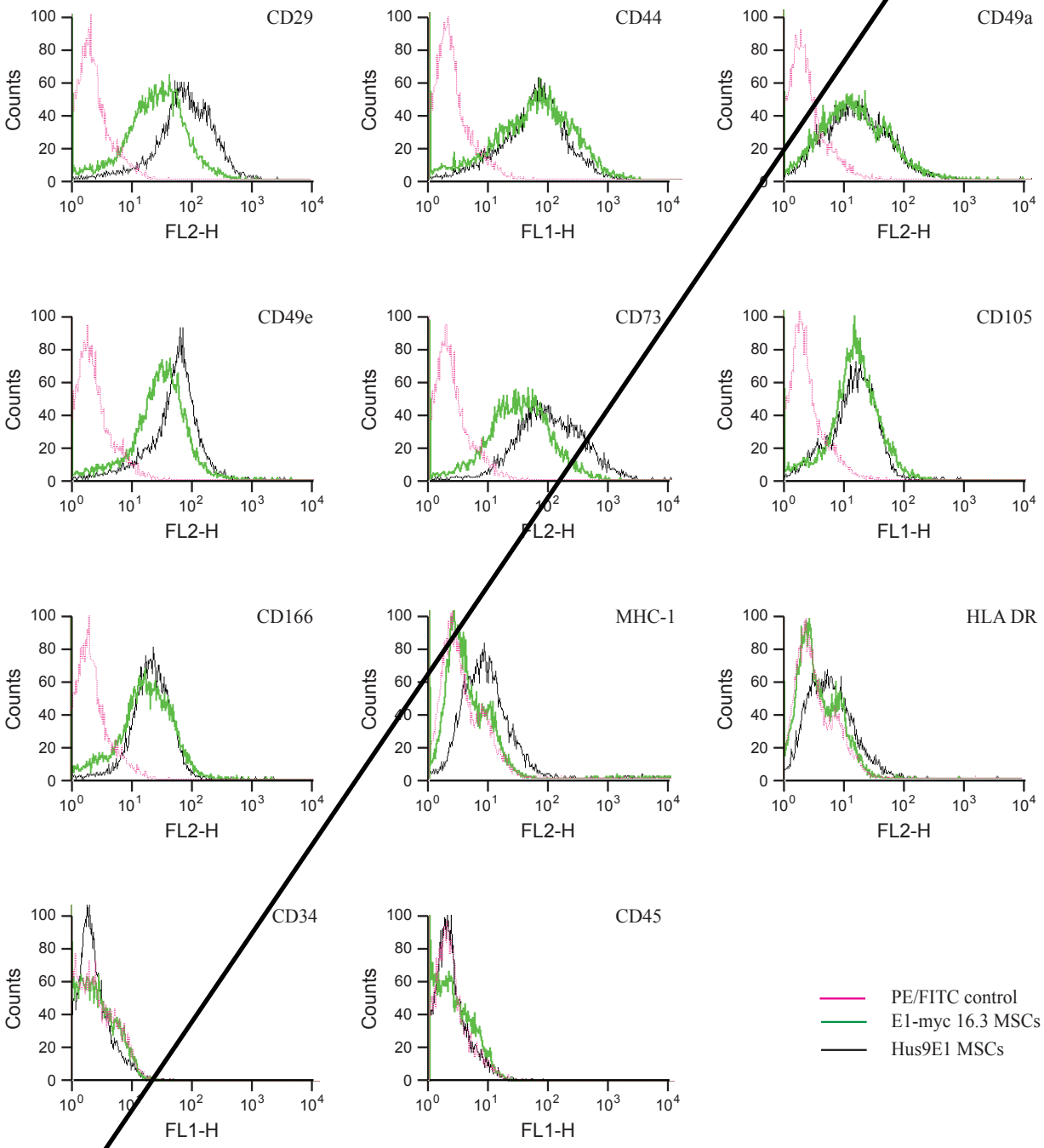

CD29
CD44
CD49a
CD49e
CD73
CD105
CD166
MHC-1
HLA DR
CD34
CD45
Counts
FL2-H
FL1-H
PE/FITC control
E1-myc 16.3 MSCs
Hus9E1 MSCs

FIGURE 3A

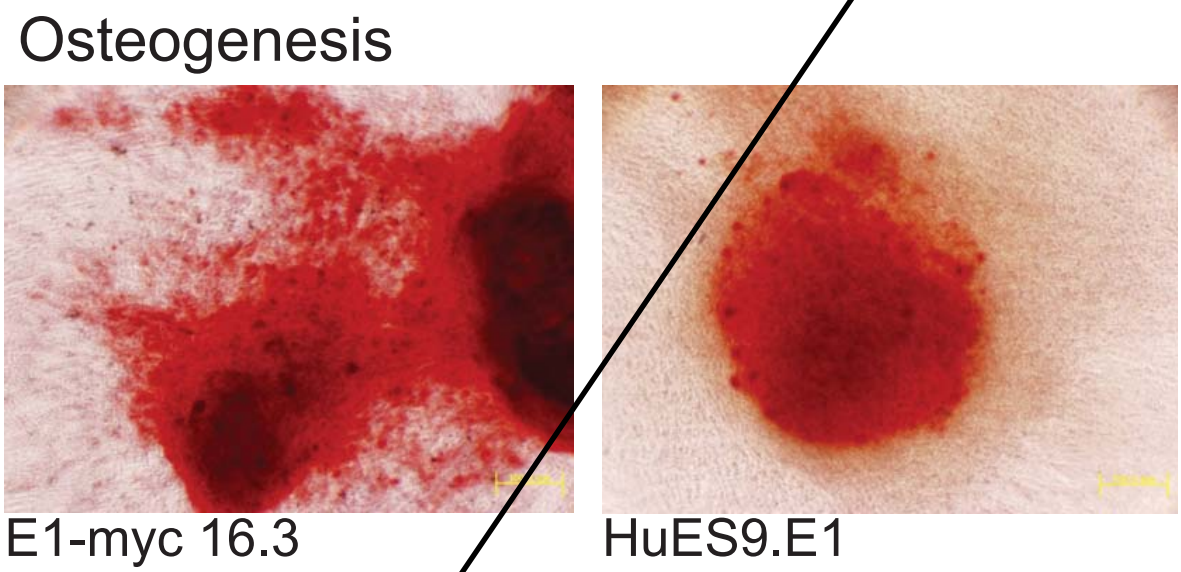
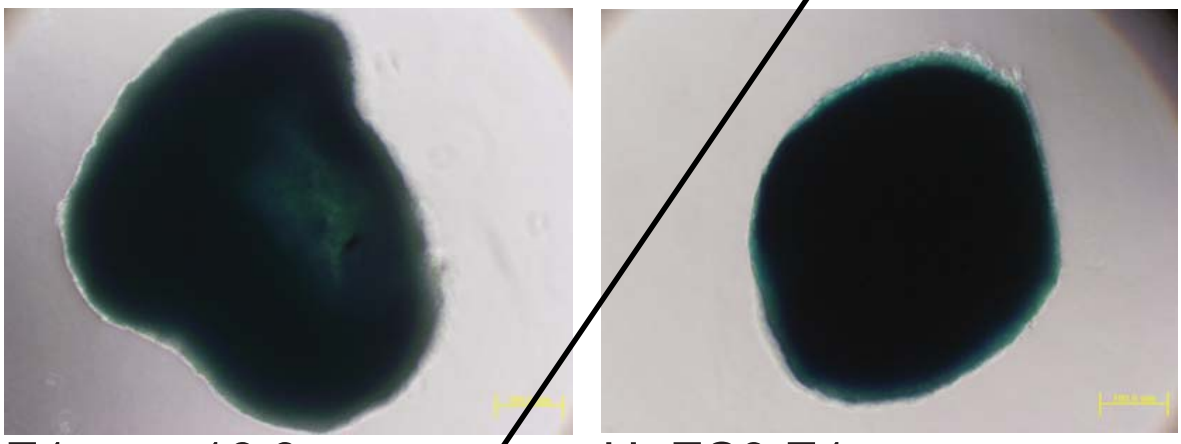


FIGURE 3B

Chondrogenesis



E1-myc 16.3 HuES9.E1

FIGURE 3C

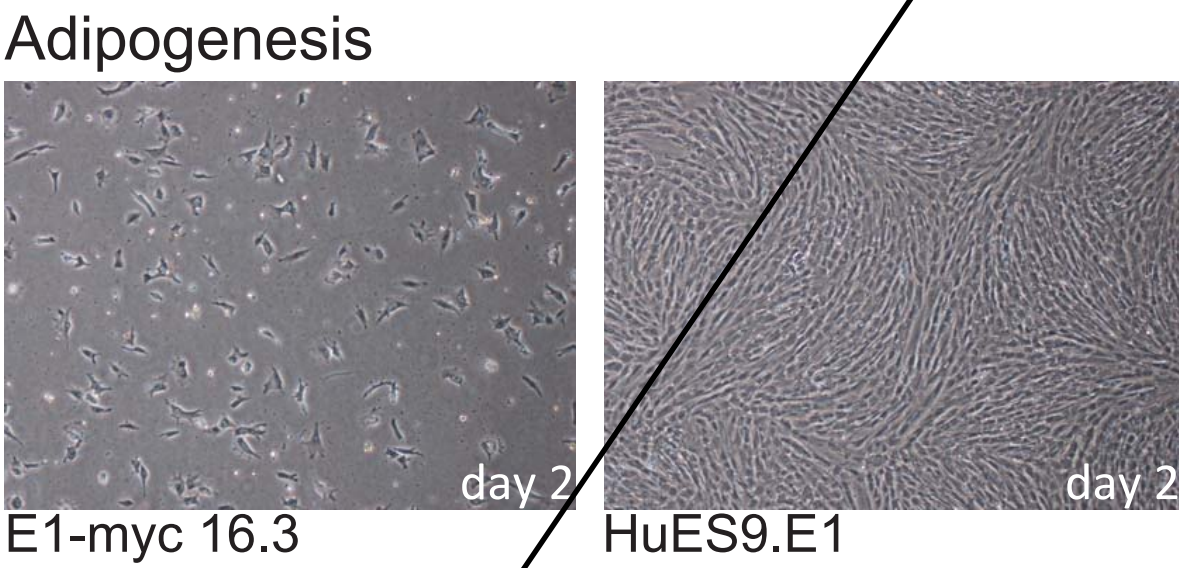


FIGURE 4A

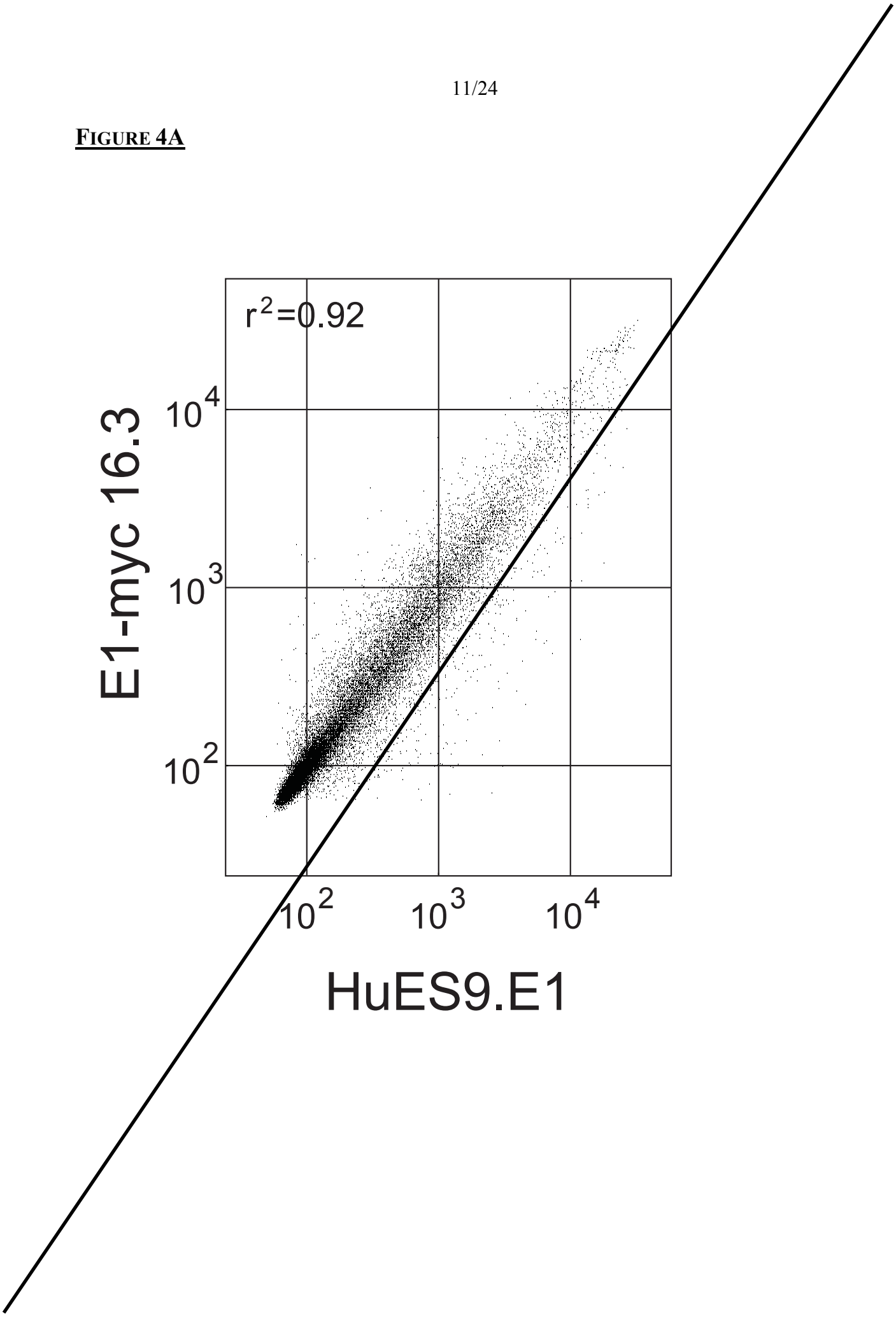


FIGURE 4B

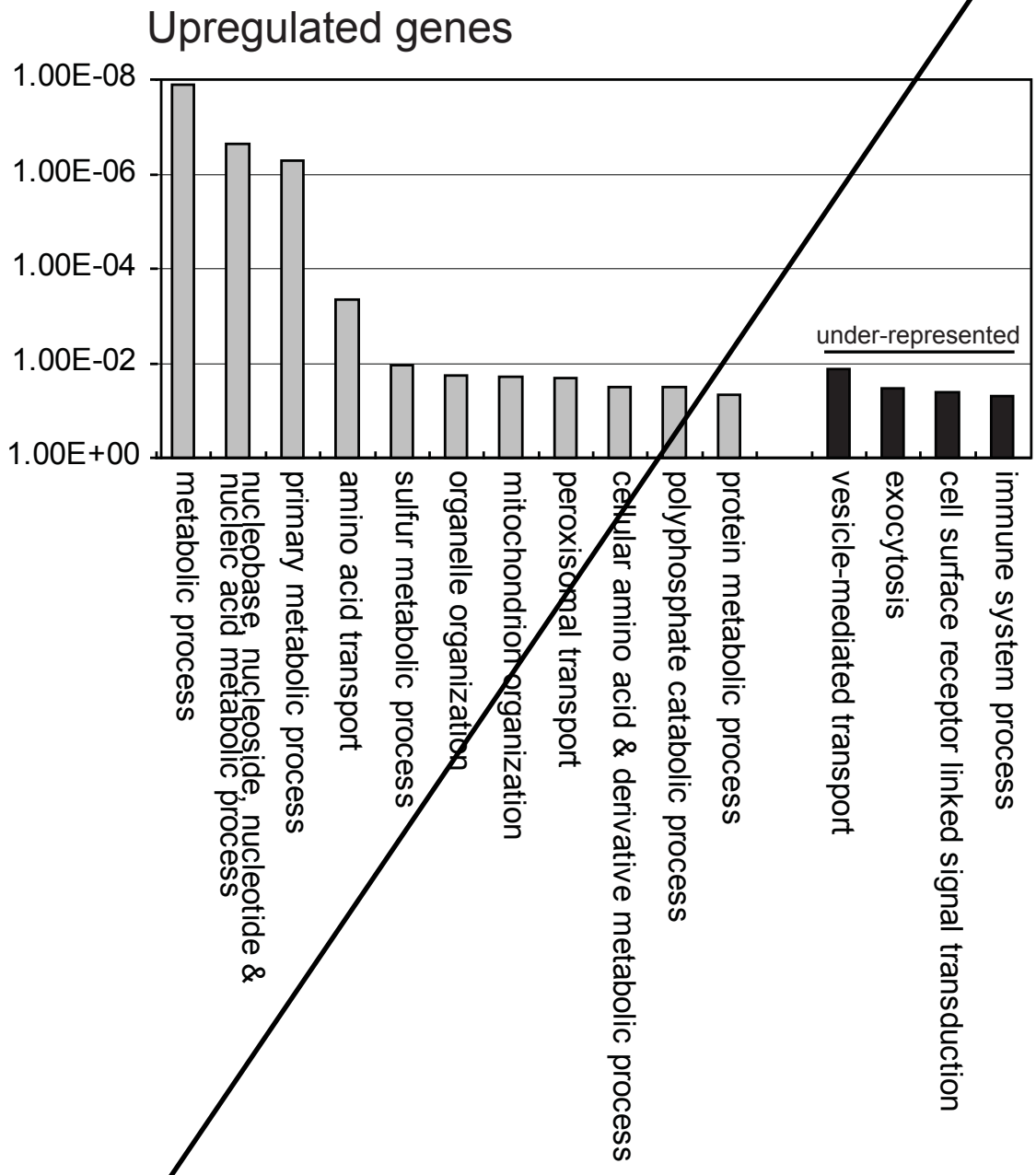

Upregulated genes
1.00E-08
1.00E-06
1.00E-04
1.00E-02
1.00E+00
under-represented
metabolic process
nucleobase, nucleoside, nucleotide & nucleic acid metabolic process
primary metabolic process
amino acid transport
sulfur metabolic process
organelle organization
mitochondrion organization
peroxisomal transport
cellular amino acid & derivative metabolic process
polyphosphate catabolic process
protein metabolic process
vesicle-mediated transport
exocytosis
cell surface receptor linked signal transduction
immune system process

FIGURE 4C

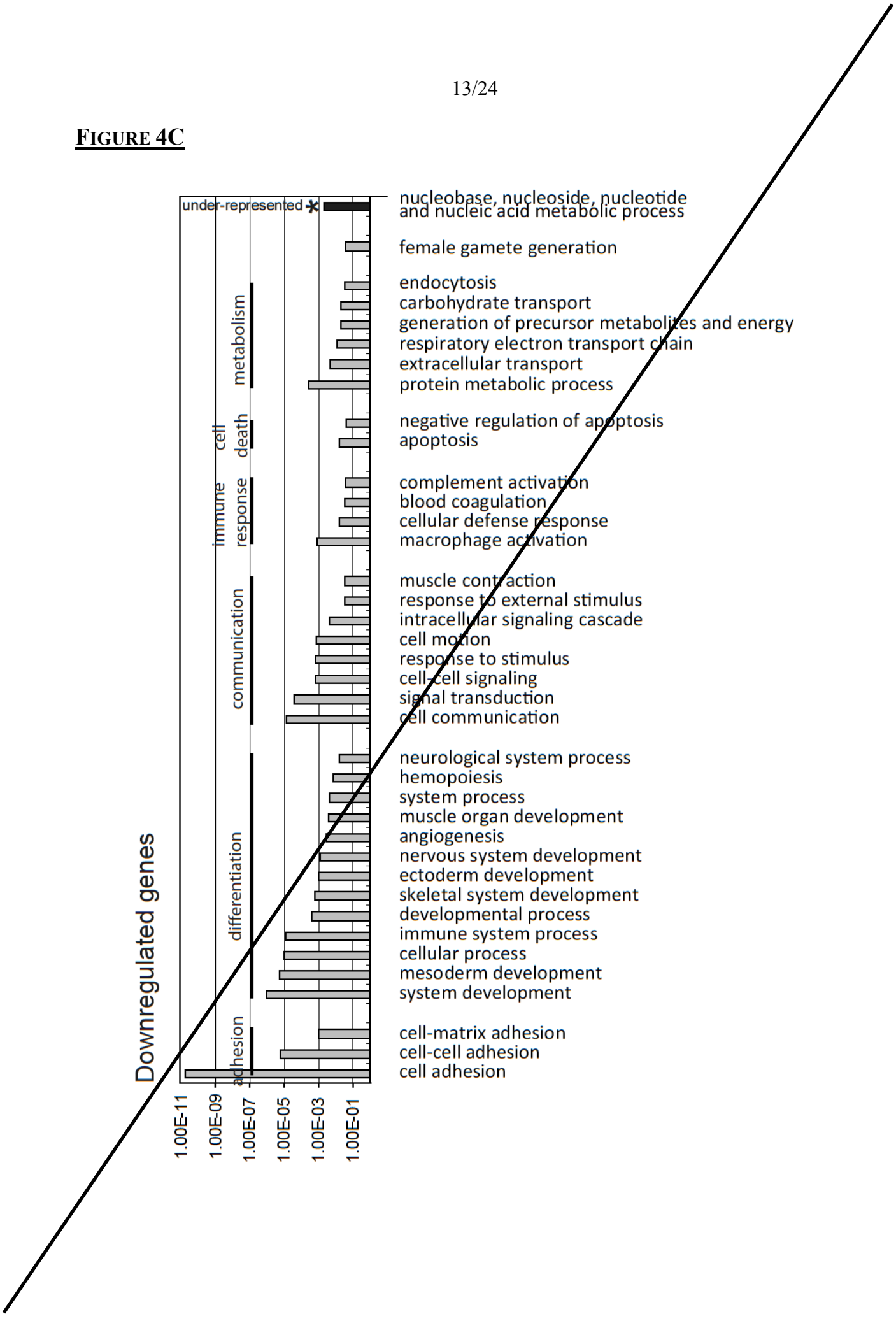

under-represented *
nucleobase, nucleoside, nucleotide and nucleic acid metabolic process
female gamete generation
endocytosis
carbohydrate transport
generation of precursor metabolites and energy
respiratory electron transport chain
extracellular transport
protein metabolic process
negative regulation of apoptosis
apoptosis
complement activation
blood coagulation
cellular defense response
macrophage activation
muscle contraction
response to external stimulus
intracellular signaling cascade
cell motion
response to stimulus
cell-cell signaling
signal transduction
cell communication
neurological system process
hemopoiesis
system process
muscle organ development
angiogenesis
nervous system development
ectoderm development
skeletal system development
developmental process
immune system process
cellular process
mesoderm development
system development
cell-matrix adhesion
cell-cell adhesion
cell adhesion
metabolism
cell death
immune response
communication
differentiation
adhesion
Downregulated genes
1.00E-11
1.00E-09
1.00E-07
1.00E-05
1.00E-03
1.00E-01

FIGURE 5A

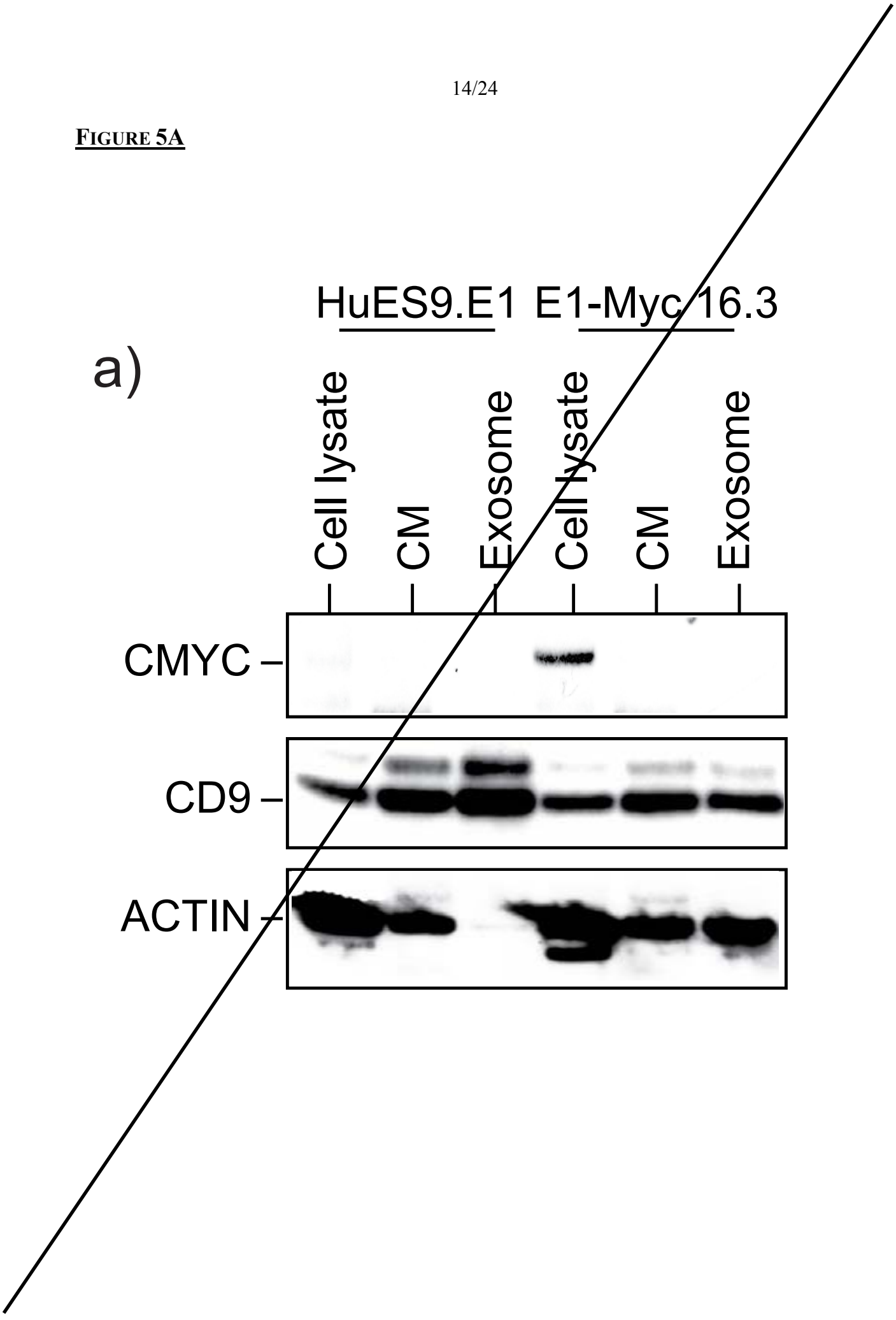

HuES9.E1
E1-Myc 16.3
a)
Cell lysate
CM
Exosome
Cell lysate
CM
Exosome
CMYC
CD9
ACTIN

FIGURE 5B

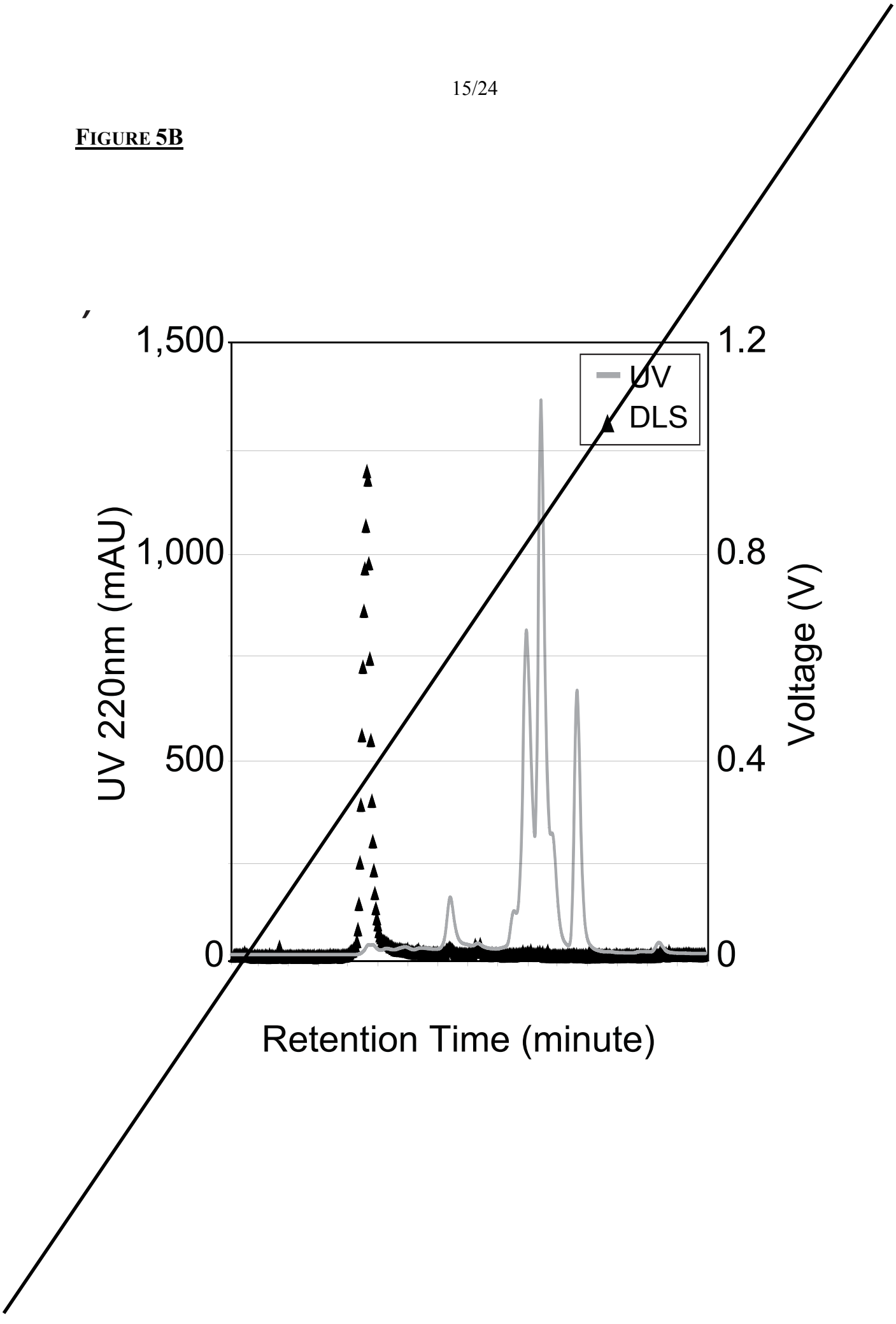

1,500
1,000
500
0
UV 220nm (mAU)
1.2
0.8
0.4
0
Voltage (V)
UV
DLS
Retention Time (minute)

FIGURE 5C

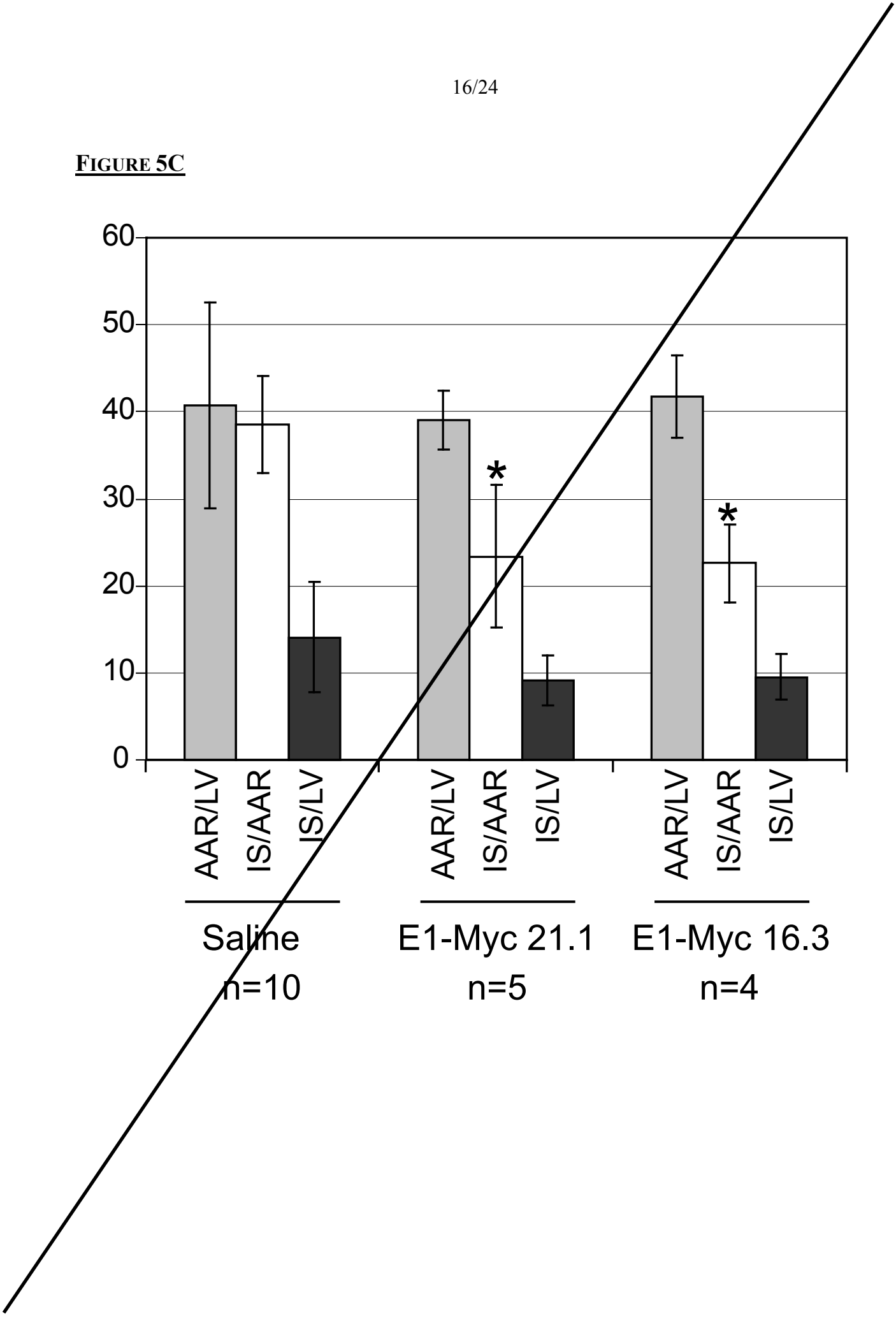

60
50
40
30
20
10
0
AAR/LV
IS/AAR
IS/LV
AAR/LV
IS/AAR
IS/LV
AAR/LV
IS/AAR
IS/LV
*
*
Saline
n=10
E1-Myc 21.1
n=5
E1-Myc 16.3
n=4

FIGURE 6

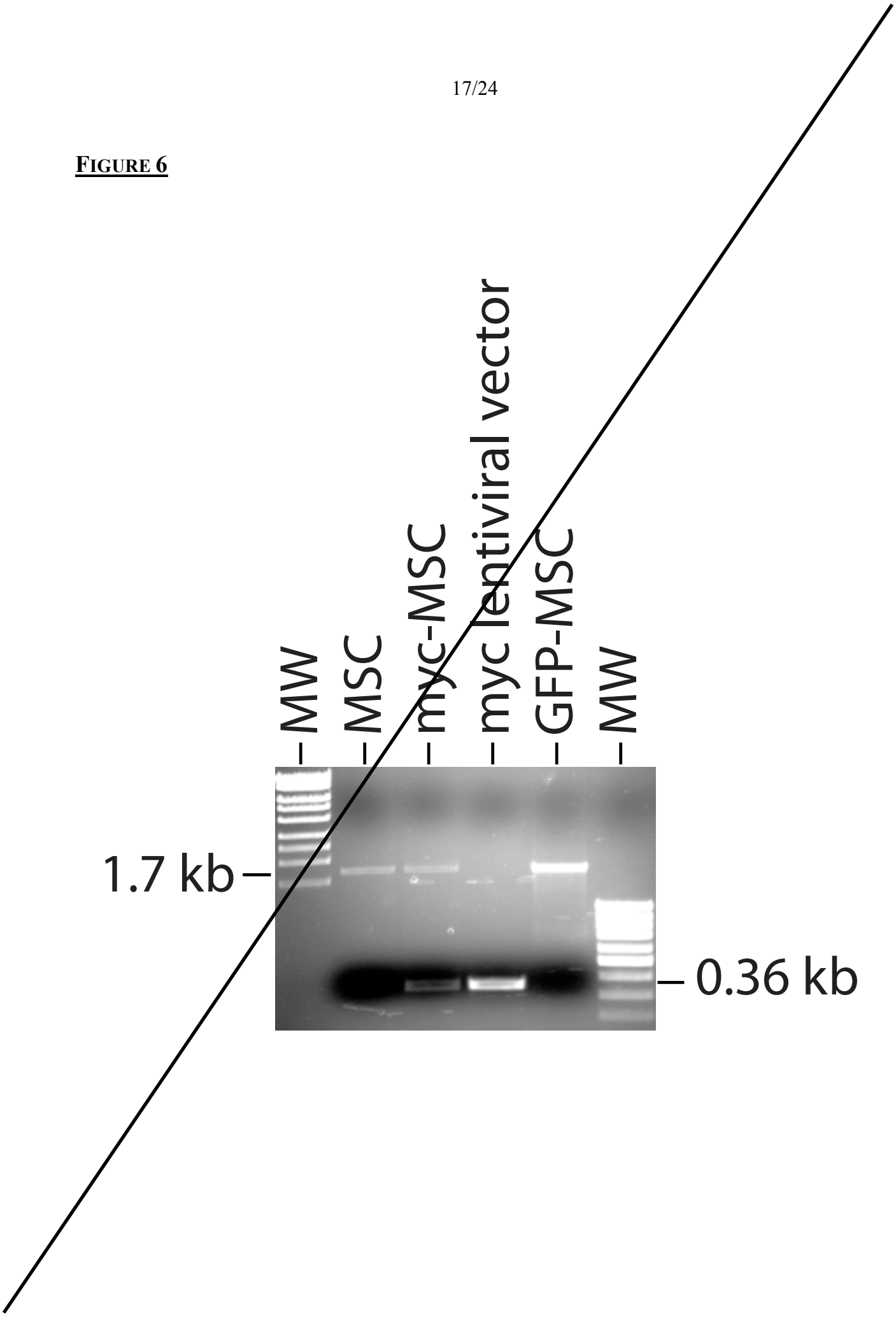


FIGURE 7

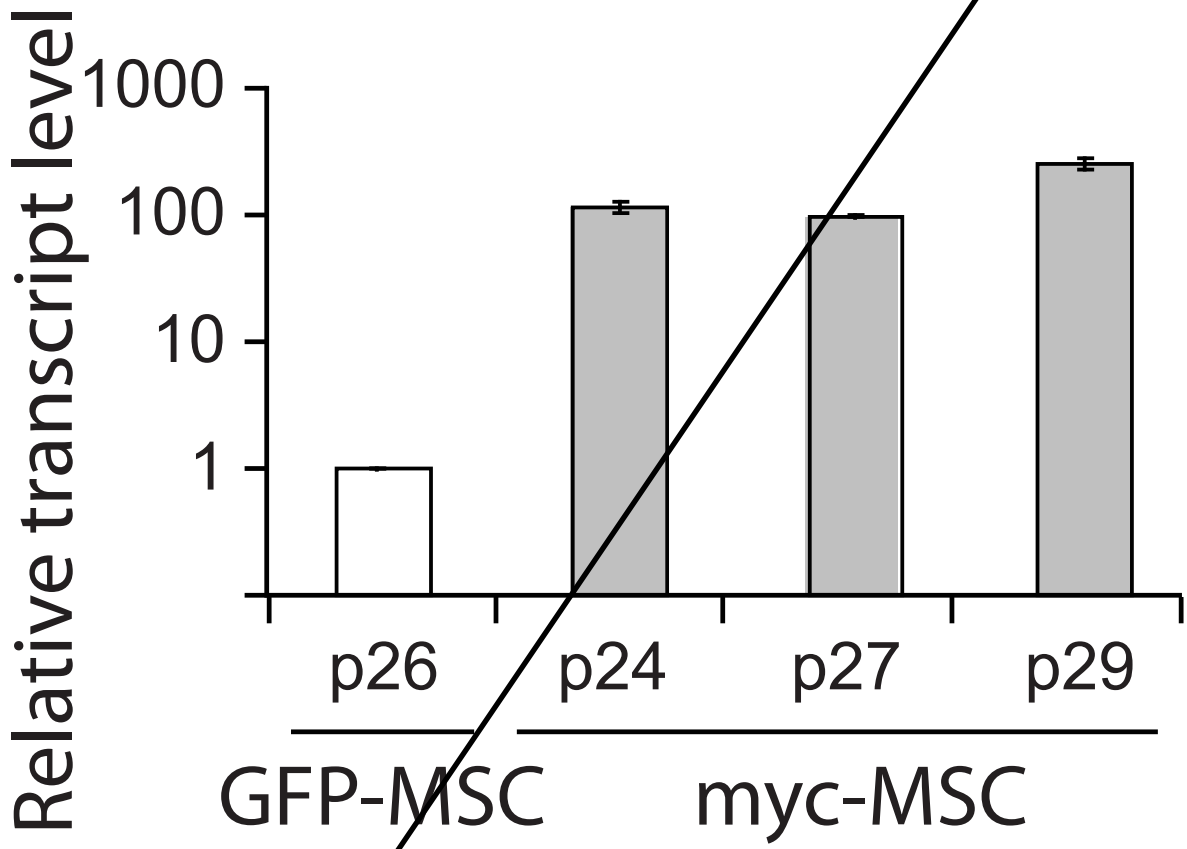

Relative transcript level
1000
100
10
1
p26
p24
p27
p29
GFP-MSC
myc-MSC

FIGURE 8

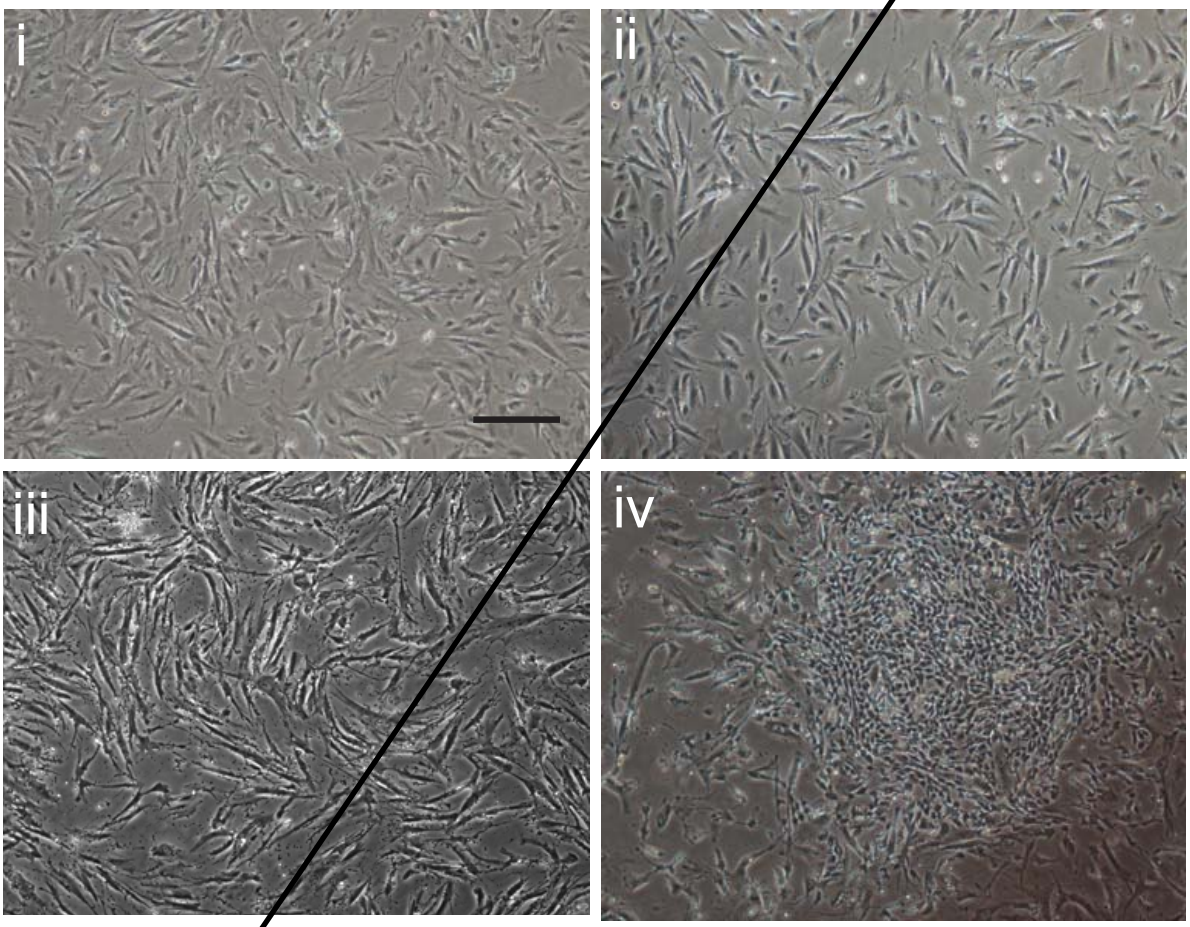


FIGURE 9

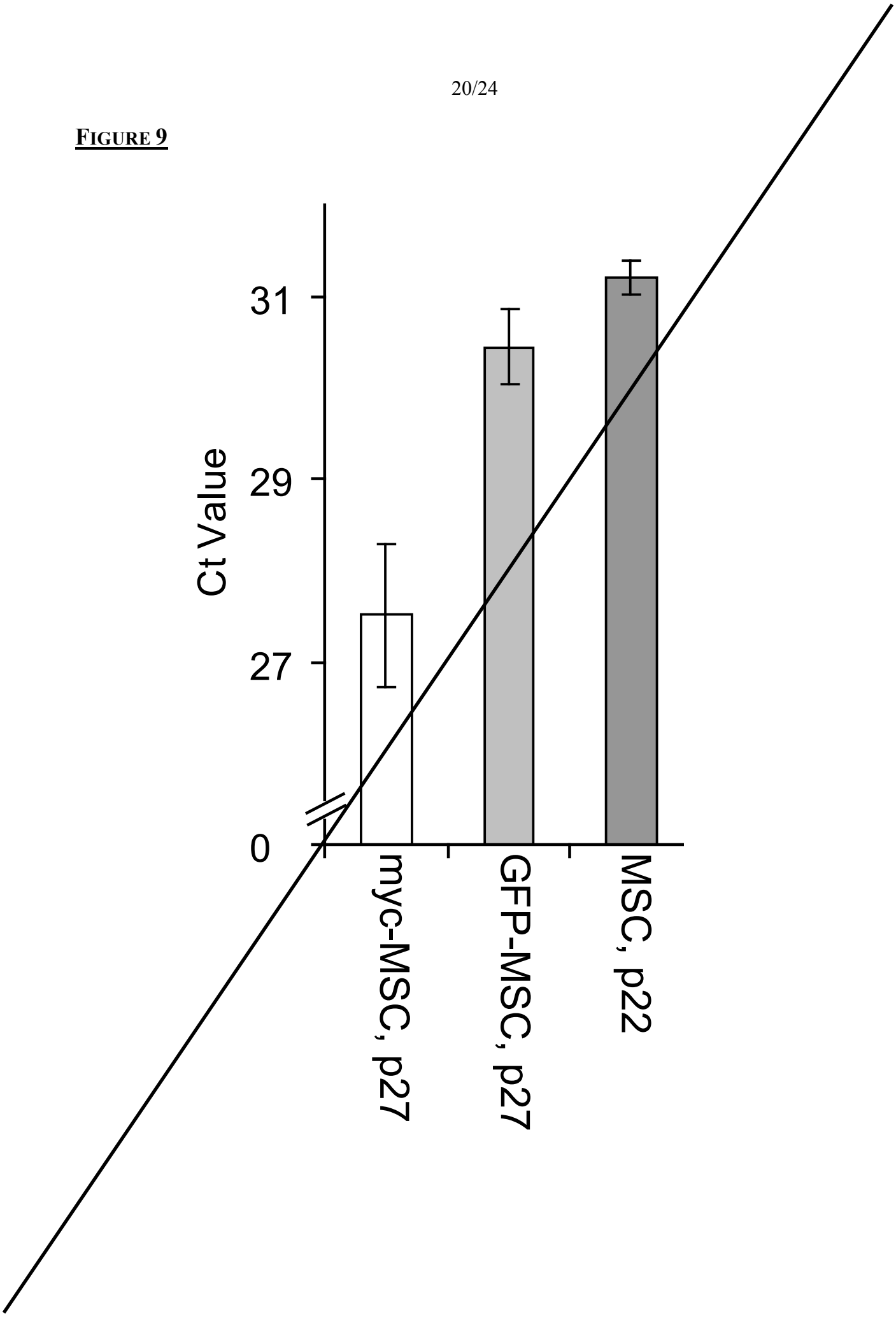


FIGURE 10

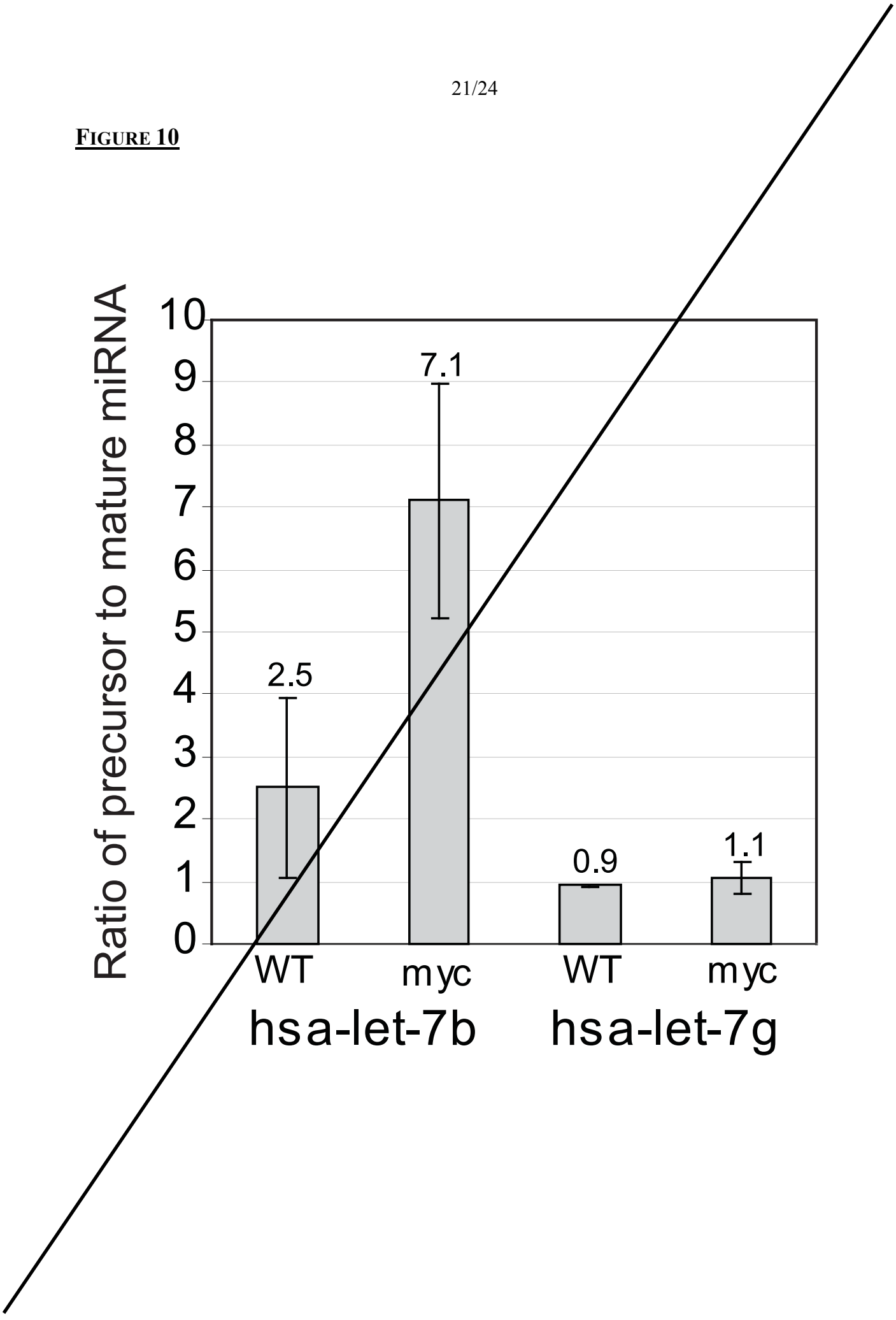

Ratio of precursor to mature miRNA
10
9
8
7
6
5
4
3
2
1
0
2.5
7.1
0.9
1.1
WT
myc
WT
myc
hsa-let-7b
hsa-let-7g

FIGURE 11

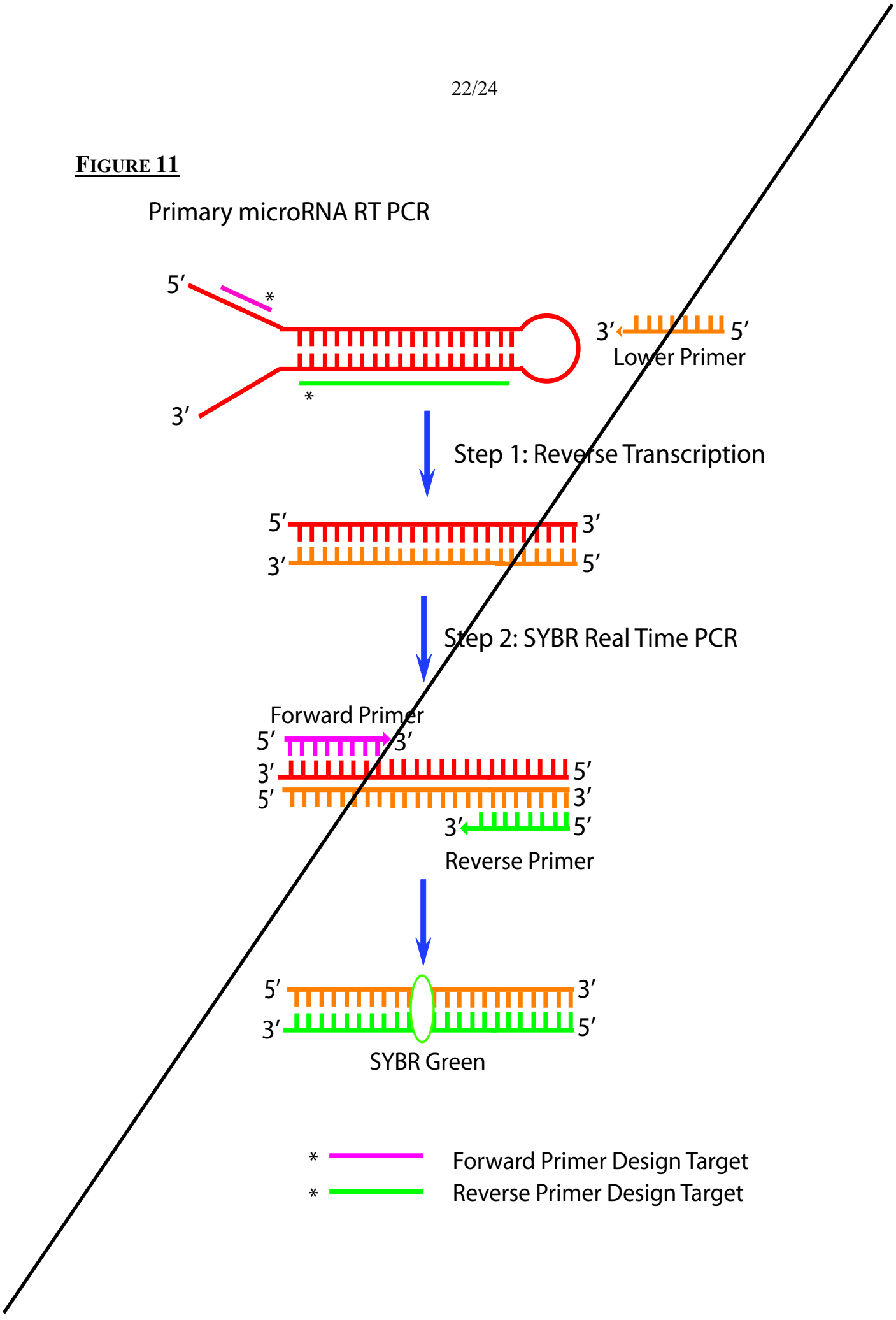

Primary microRNA RT PCR
5′
*
3′
*
3′
5′
Lower Primer
Step 1: Reverse Transcription
5′
3′
3′
5′
Step 2: SYBR Real Time PCR
Forward Primer
5′
3′
3′
5′
5′
3′
3′
5′
Reverse Primer
5′
3′
3′
5′
SYBR Green
* Forward Primer Design Target
* Reverse Primer Design Target

FIGURE 12

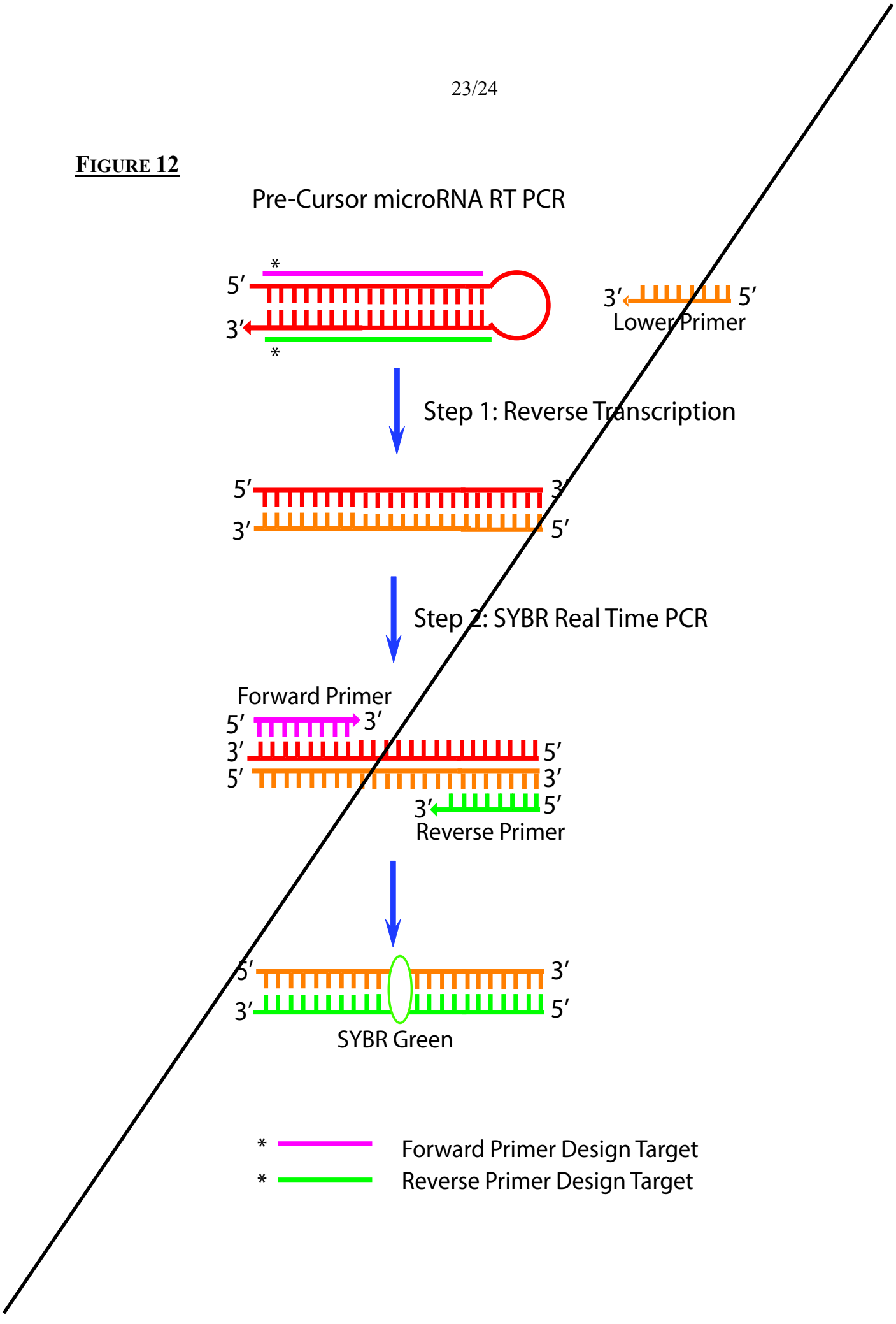

Pre-Cursor microRNA RT PCR
*
5′
3′
*
3′
5′
Lower Primer
Step 1: Reverse Transcription
5′
3′
3′
5′
Step 2: SYBR Real Time PCR
Forward Primer
5′
3′
3′
5′
5′
3′
3′
5′
Reverse Primer
5′
3′
3′
5′
SYBR Green
* Forward Primer Design Target
* Reverse Primer Design Target

FIGURE 13

Mature microRNA RT PCR

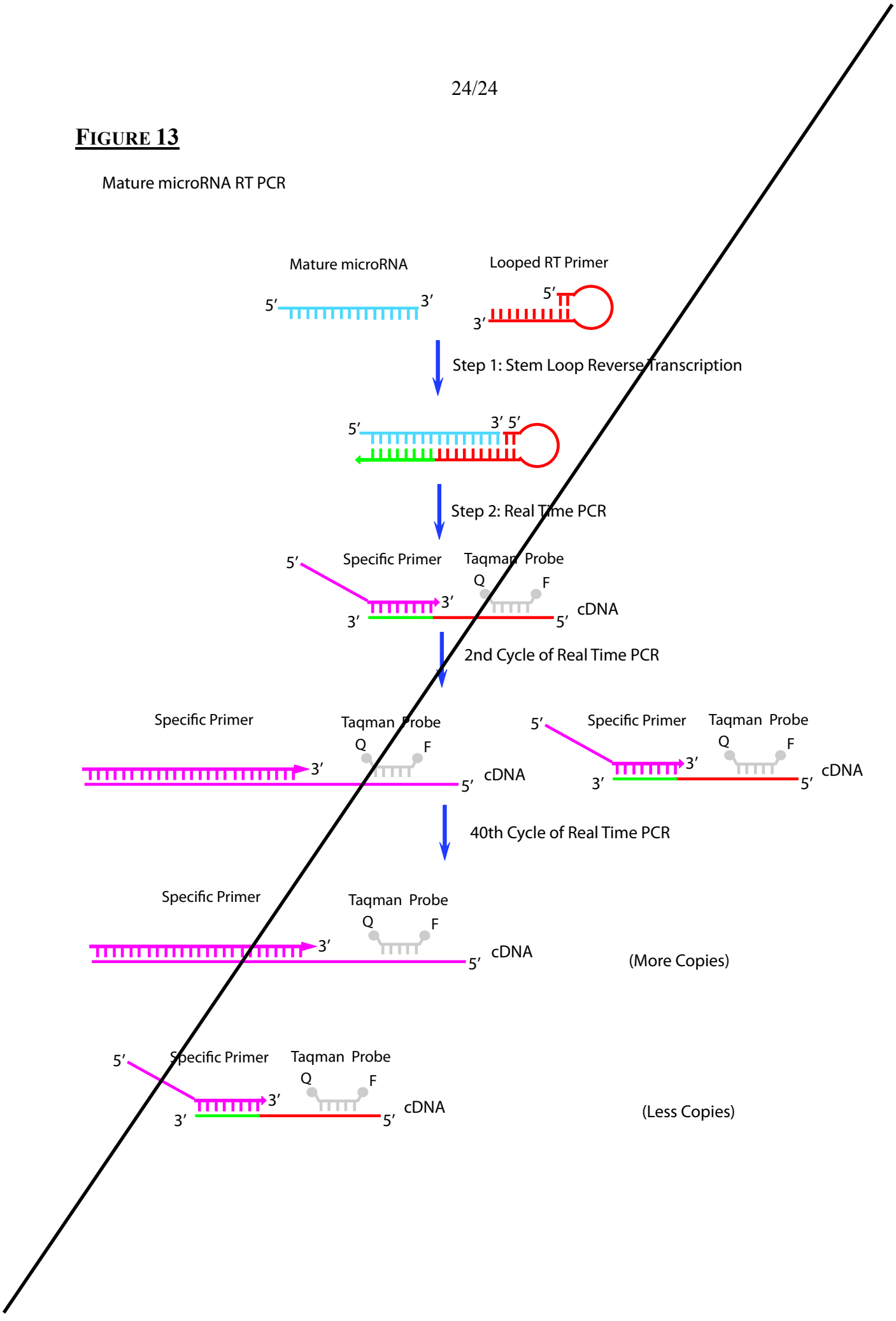


FIGURE 1A

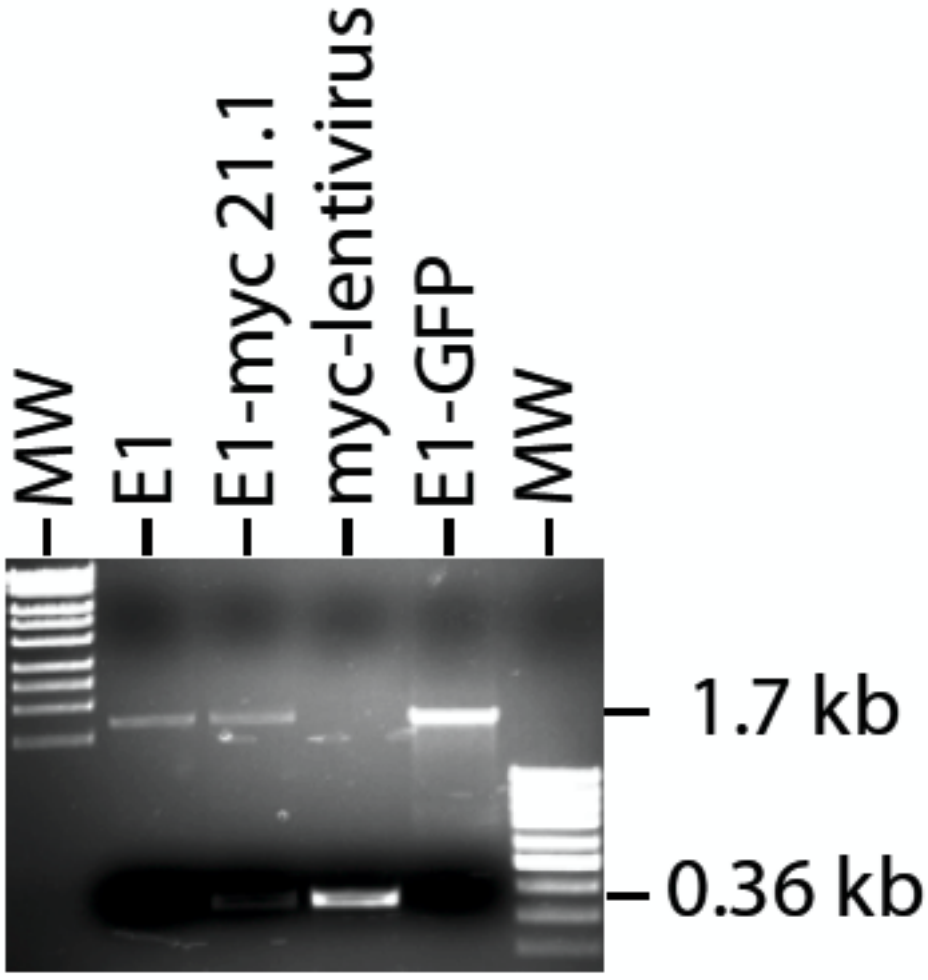


FIGURE 1B

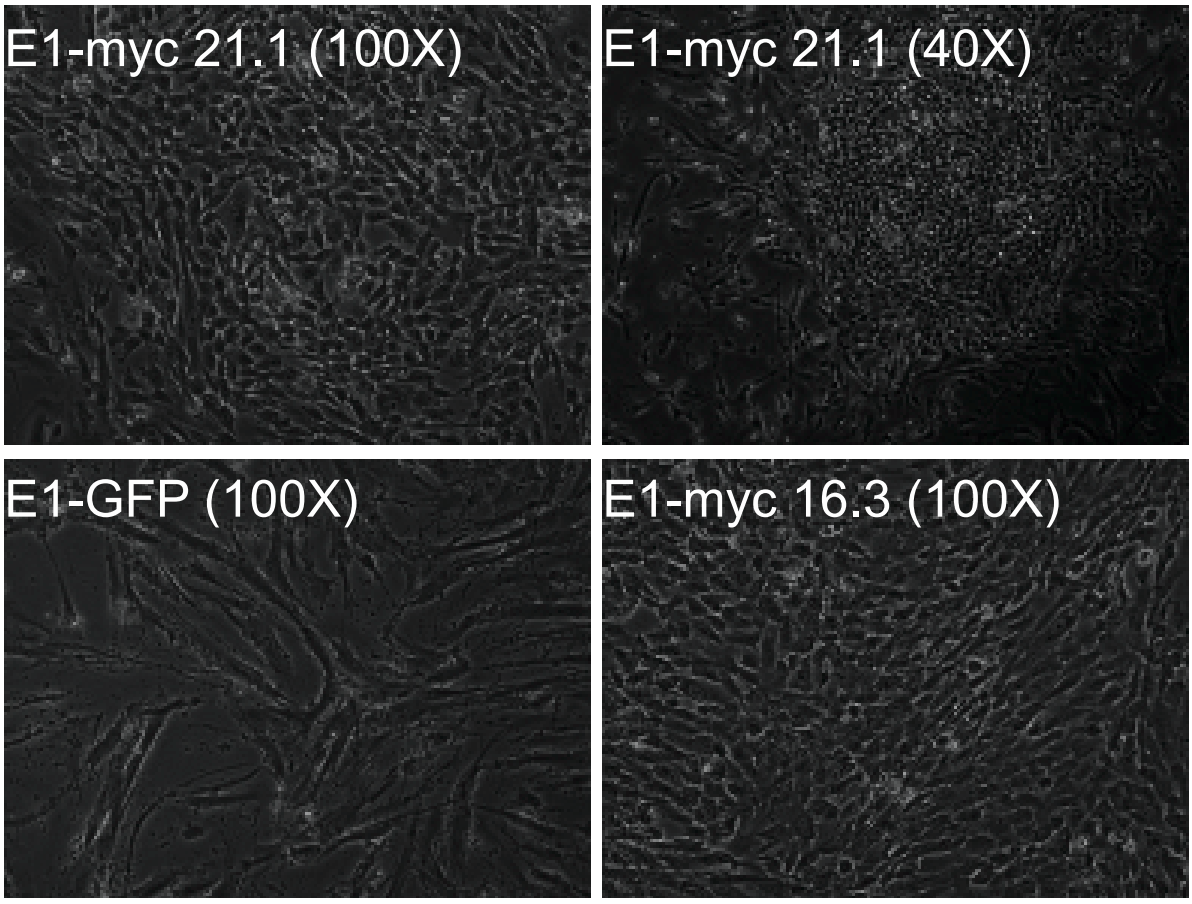


FIGURE 1C

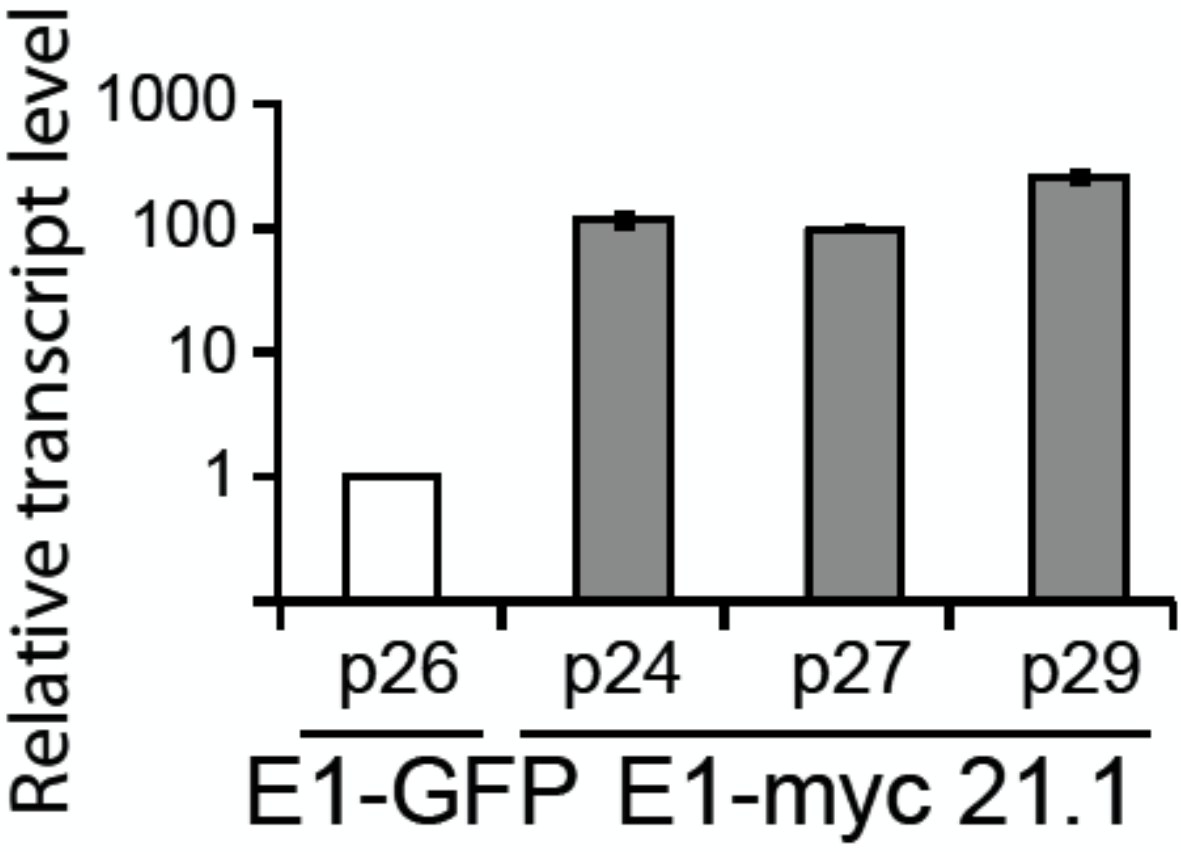

Relative transcript level
1000
100
10
1
p26
p24
p27
p29
E1-GFP
E1-myc 21.1

FIGURE 1D

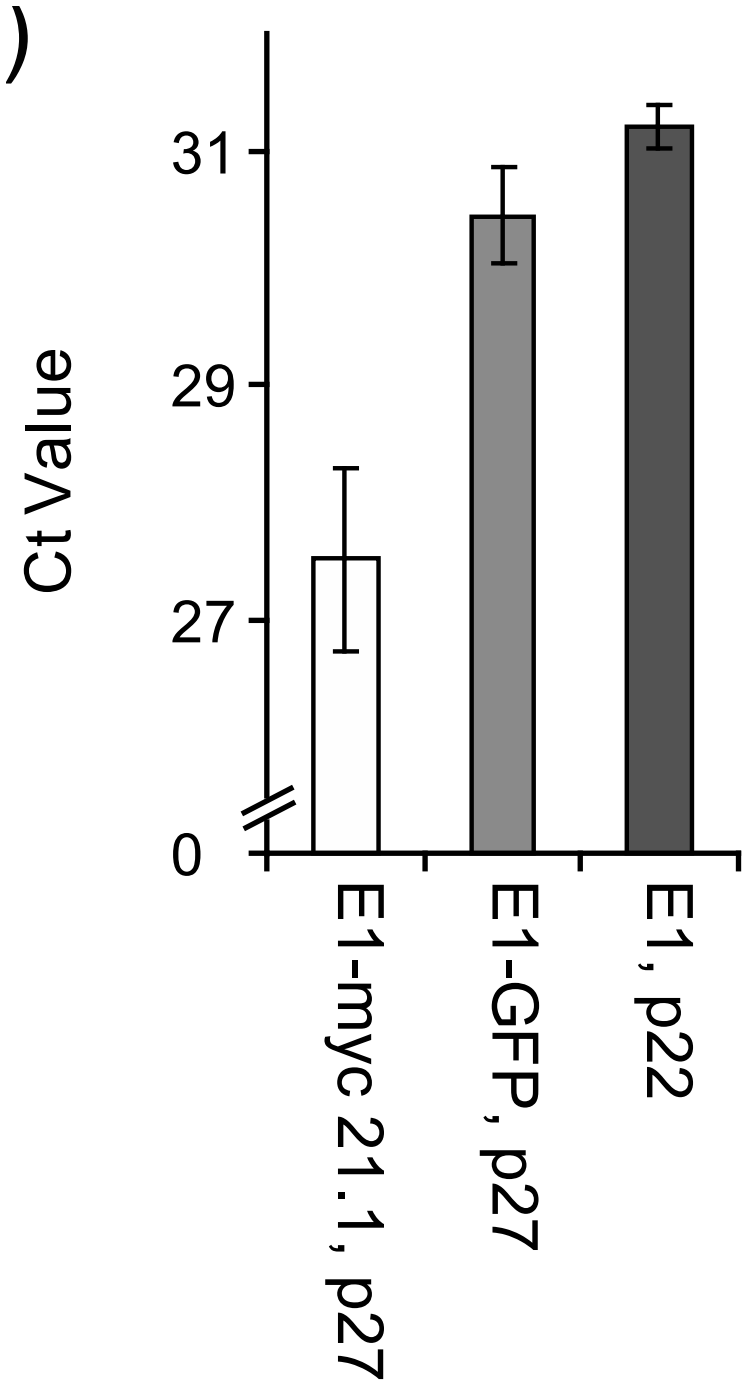


FIGURE 1E

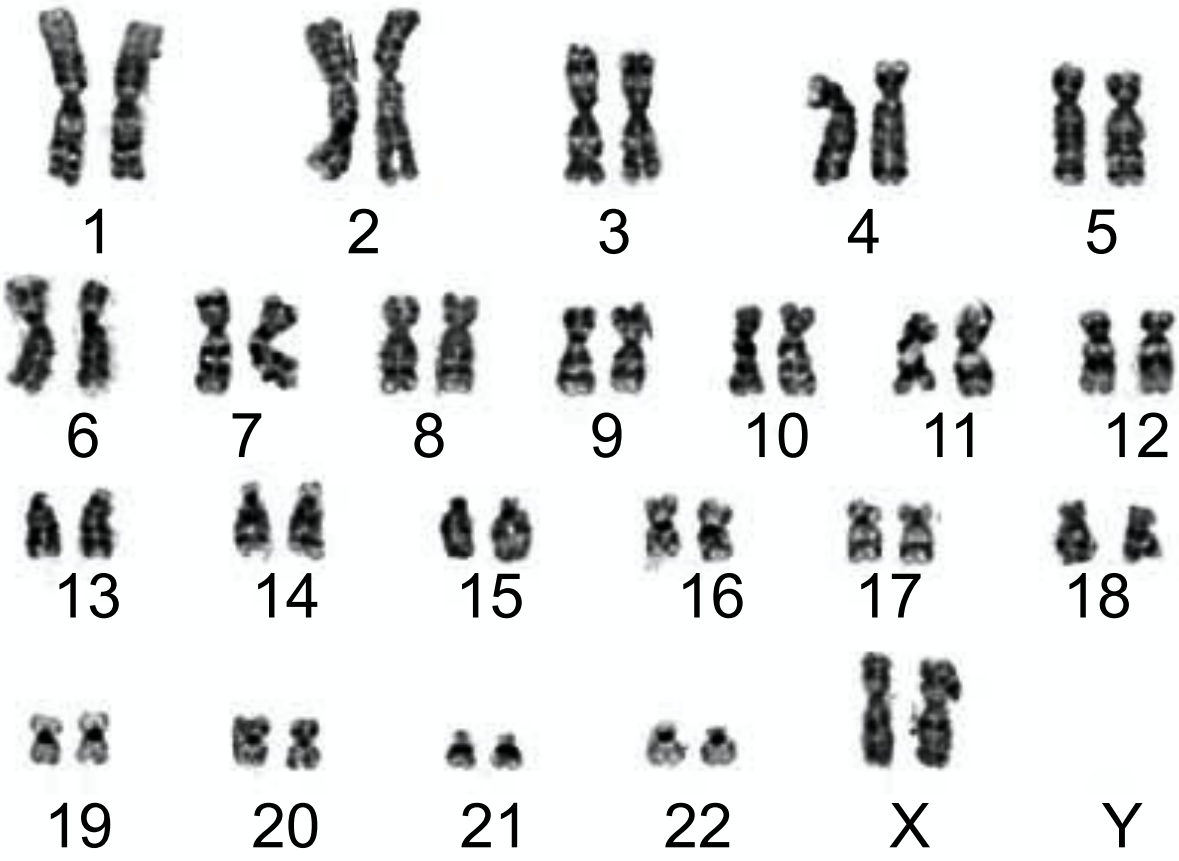


FIGURE 1F

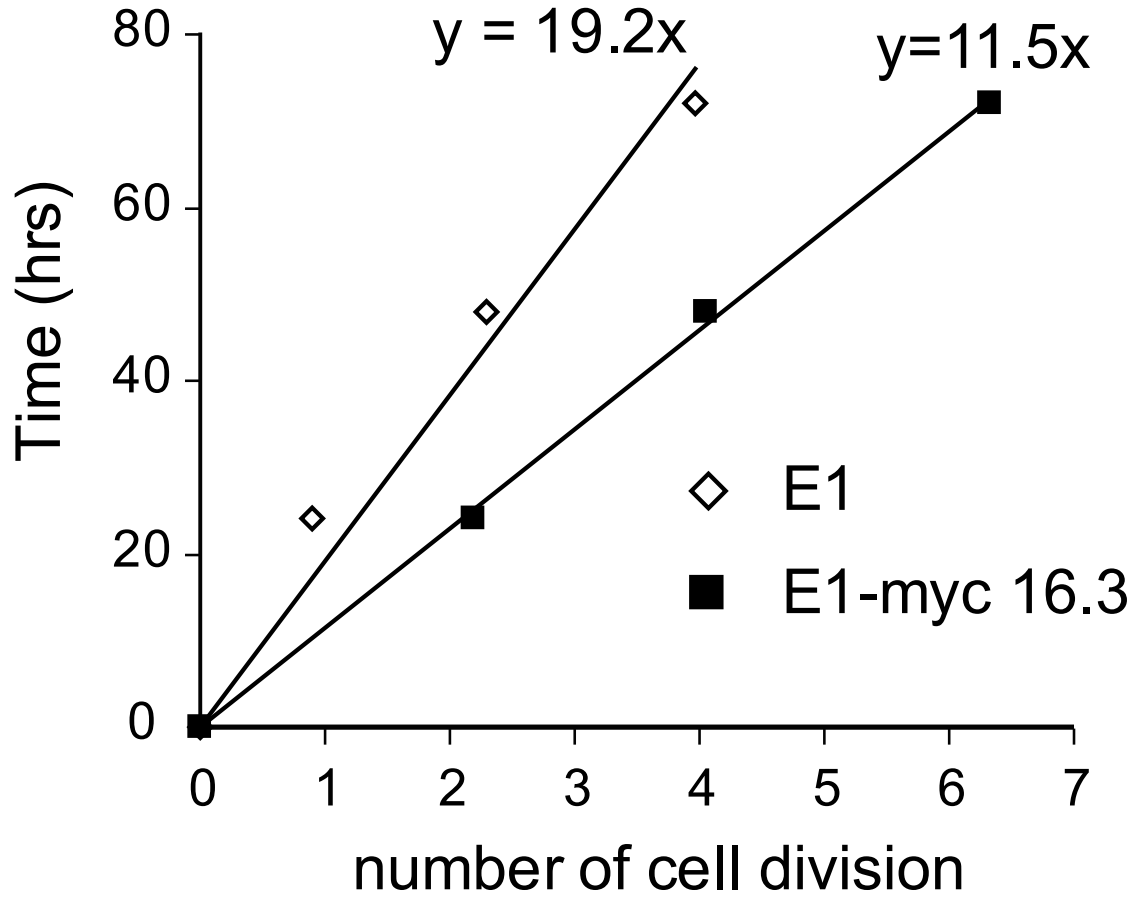

y = 19.2x
y=11.5x
Time (hrs)
80
60
40
20
0
0
1
2
3
4
5
6
7
number of cell division
E1
E1-myc 16.3

FIGURE 2

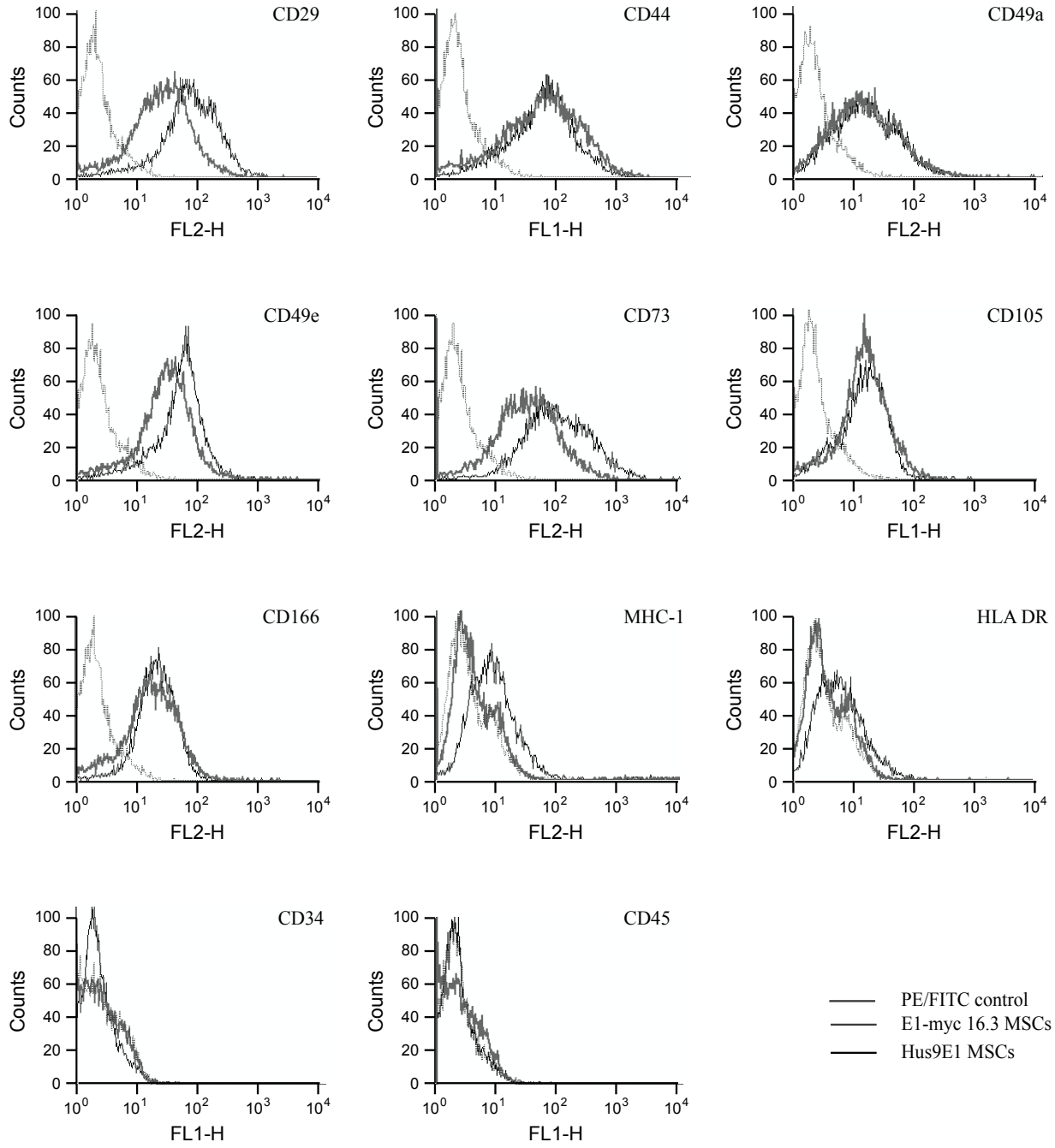
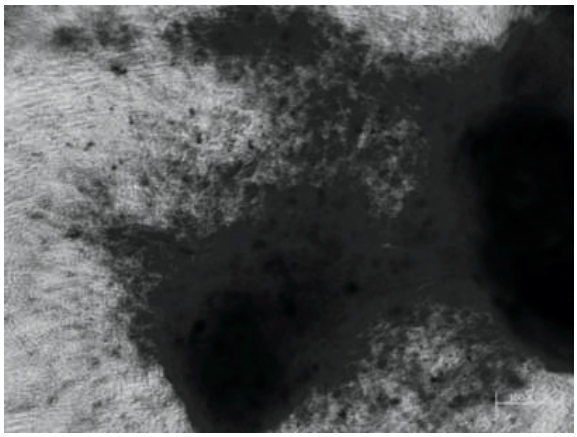
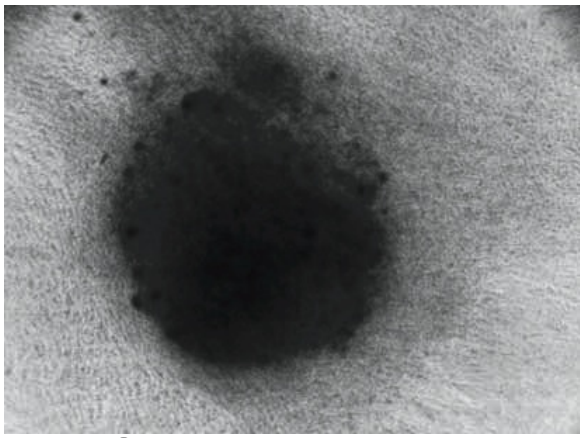

CD29
CD44
CD49a
CD49e
CD73
CD105
CD166
MHC-1
HLA DR
CD34
CD45
Counts
FL2-H
FL1-H
PE/FITC control
E1-myc 16.3 MSCs
Hus9E1 MSCs

FIGURE 3A

Osteogenesis



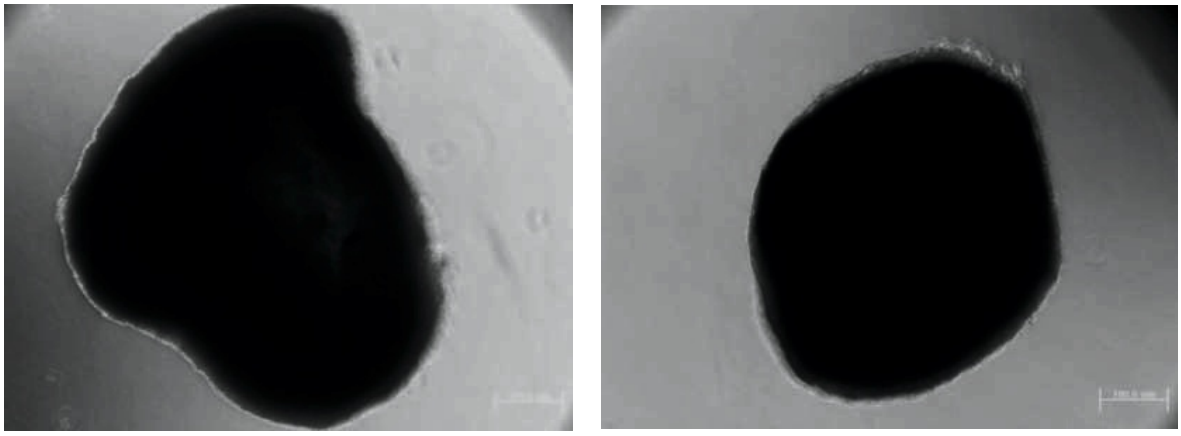
E1-myc 16.3



HuES9.E1

FIGURE 3B

Chondrogenesis



E1-myc 16.3 HuES9.E1

FIGURE 3C

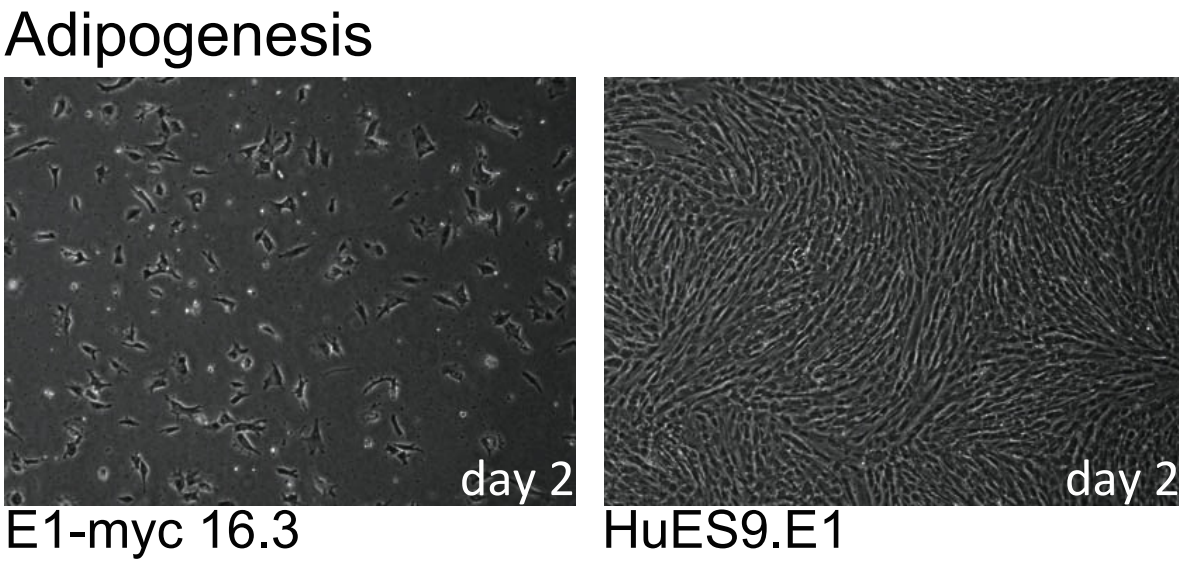


FIGURE 4A

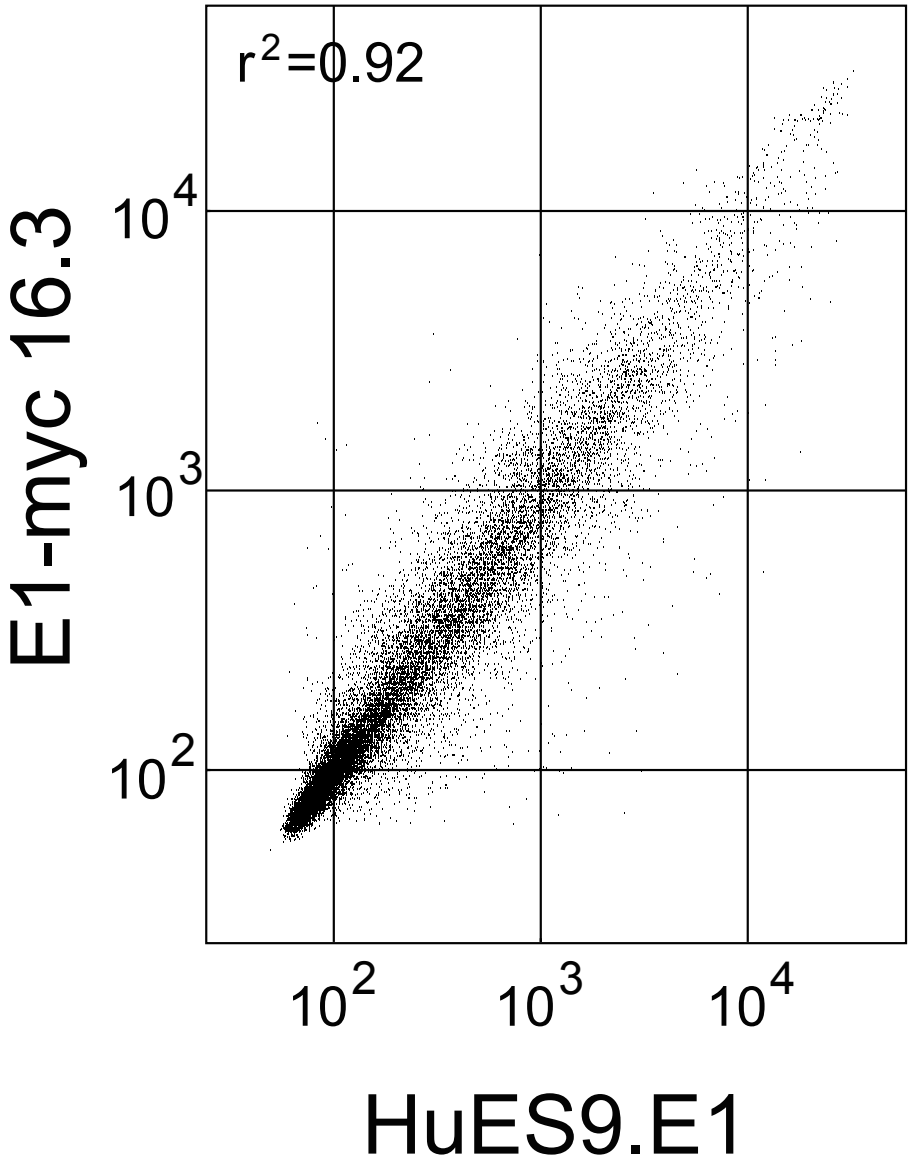


FIGURE 4B

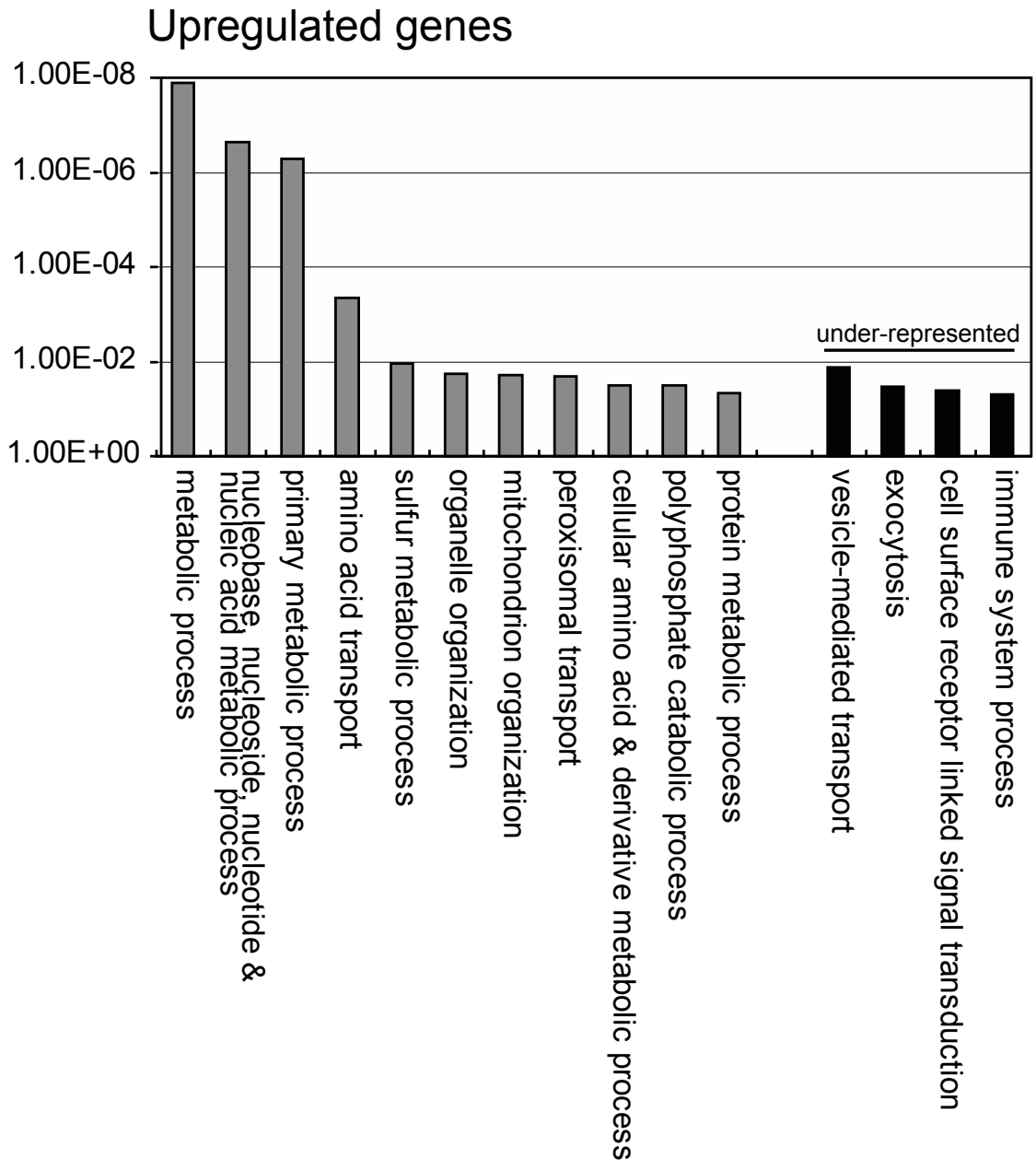

Upregulated genes
1.00E-08
1.00E-06
1.00E-04
1.00E-02
1.00E+00
under-represented
metabolic process
nucleobase, nucleoside, nucleotide & nucleic acid metabolic process
primary metabolic process
amino acid transport
sulfur metabolic process
organelle organization
mitochondrion organization
peroxisomal transport
cellular amino acid & derivative metabolic process
polyphosphate catabolic process
protein metabolic process
vesicle-mediated transport
exocytosis
cell surface receptor linked signal transduction
immune system process

FIGURE 4C

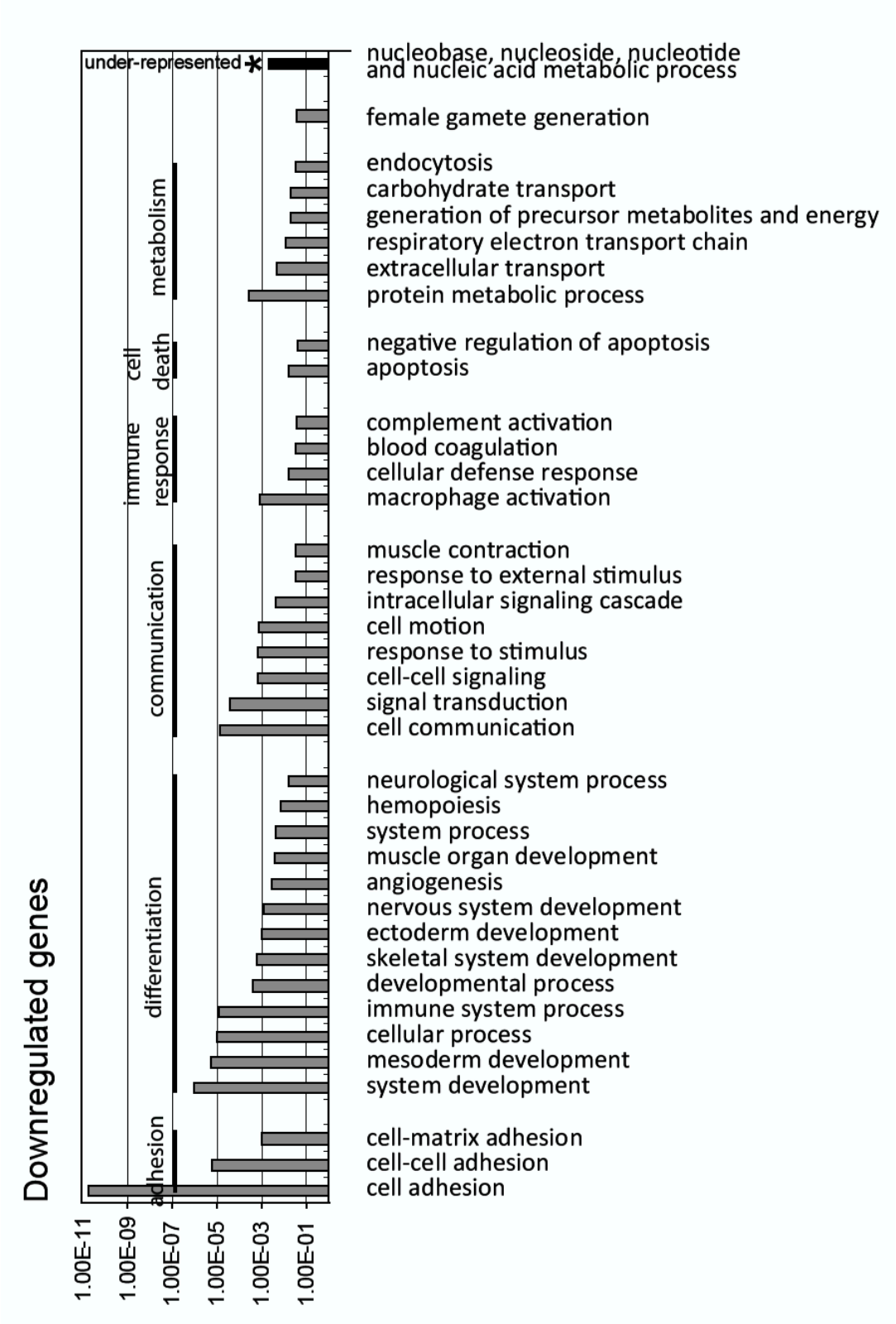

Downregulated genes
1.00E-11
1.00E-09
1.00E-07
1.00E-05
1.00E-03
1.00E-01
under-represented *
adhesion
differentiation
communication
immune response
cell death
metabolism
nucleobase, nucleoside, nucleotide and nucleic acid metabolic process
female gamete generation
endocytosis
carbohydrate transport
generation of precursor metabolites and energy
respiratory electron transport chain
extracellular transport
protein metabolic process
negative regulation of apoptosis
apoptosis
complement activation
blood coagulation
cellular defense response
macrophage activation
muscle contraction
response to external stimulus
intracellular signaling cascade
cell motion
response to stimulus
cell-cell signaling
signal transduction
cell communication
neurological system process
hemopoiesis
system process
muscle organ development
angiogenesis
nervous system development
ectoderm development
skeletal system development
developmental process
immune system process
cellular process
mesoderm development
system development
cell-matrix adhesion
cell-cell adhesion
cell adhesion

FIGURE 5A

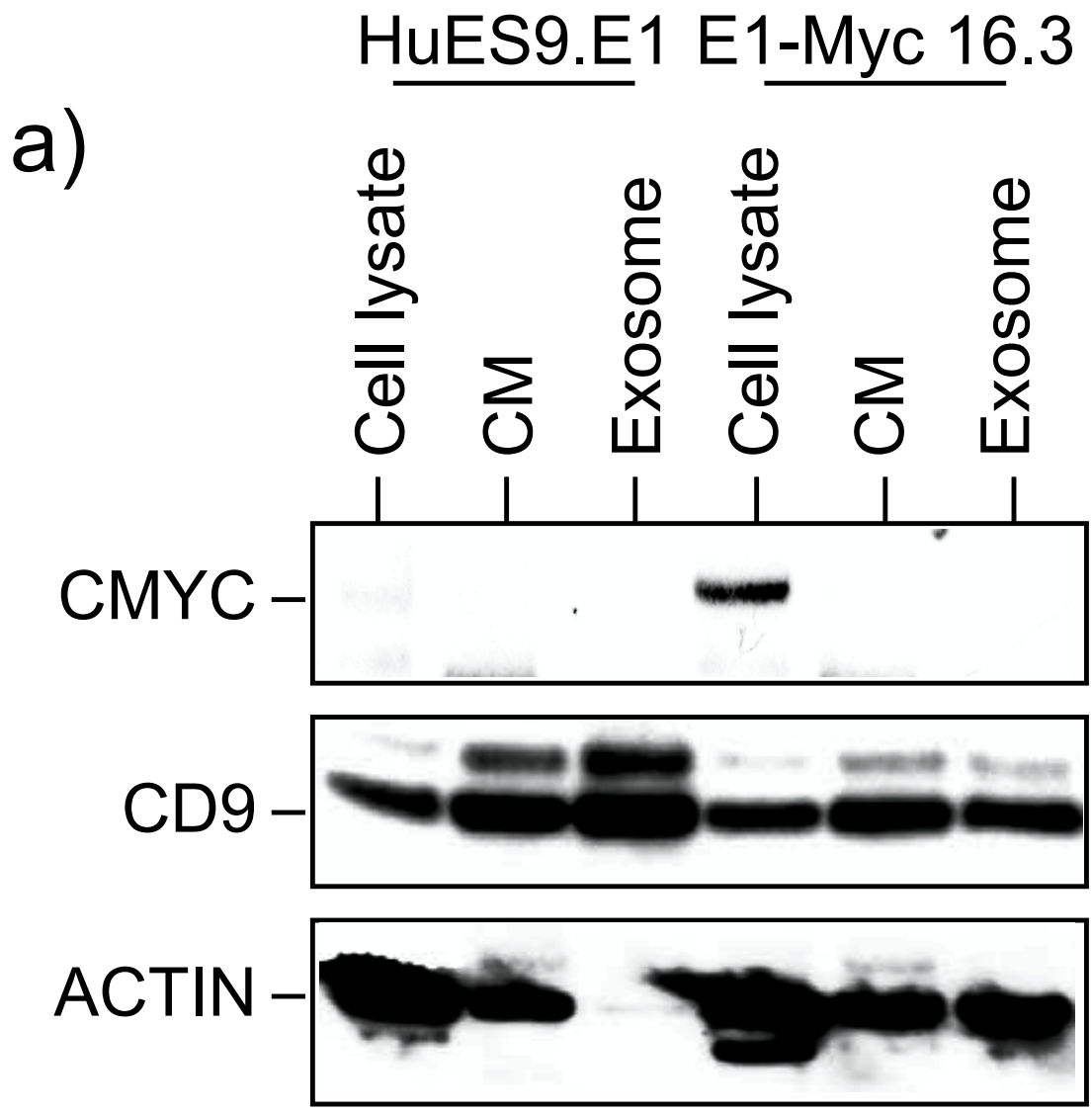

HuES9.E1
E1-Myc 16.3
a)
Cell lysate
CM
Exosome
Cell lysate
CM
Exosome
CMYC
CD9
ACTIN

FIGURE 5B

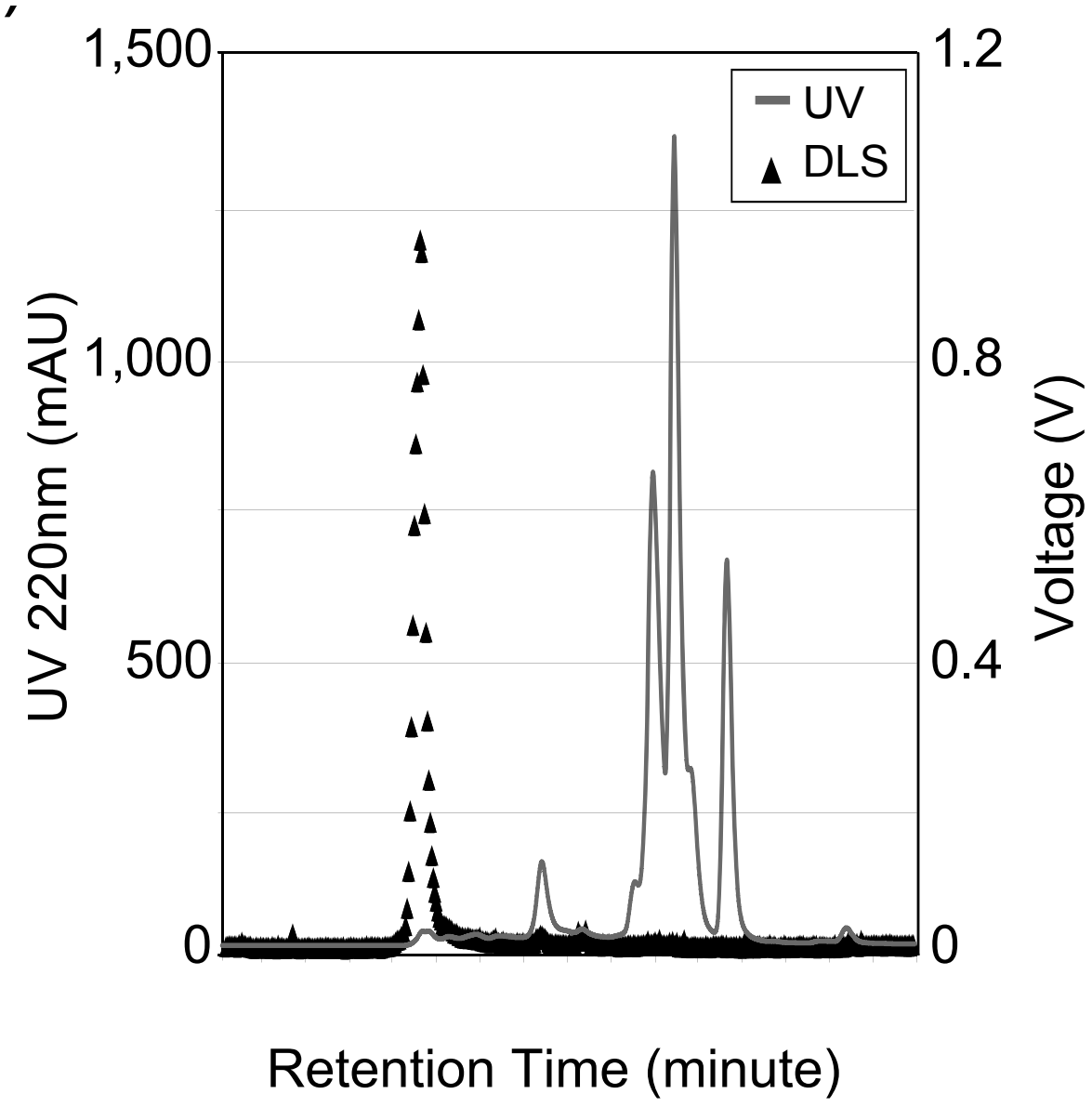

1,500
1,000
500
0
1.2
0.8
0.4
0
UV
DLS
UV 220nm (mAU)
Voltage (V)
Retention Time (minute)

FIGURE 5C

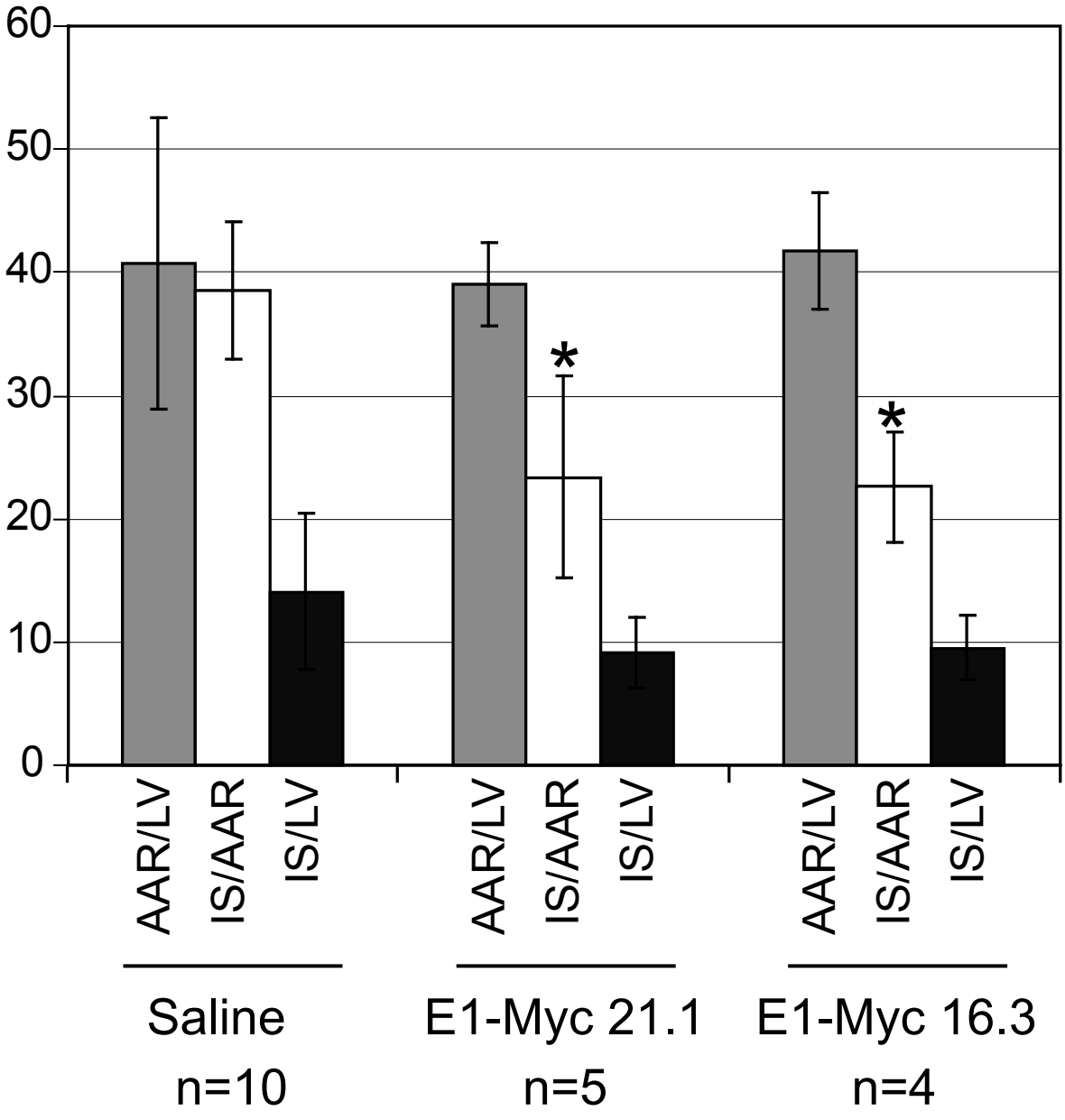

60
50
40
30
20
10
0
AAR/LV
IS/AAR
IS/LV
AAR/LV
IS/AAR
IS/LV
AAR/LV
IS/AAR
IS/LV
*
*
Saline
n=10
E1-Myc 21.1
n=5
E1-Myc 16.3
n=4

FIGURE 6

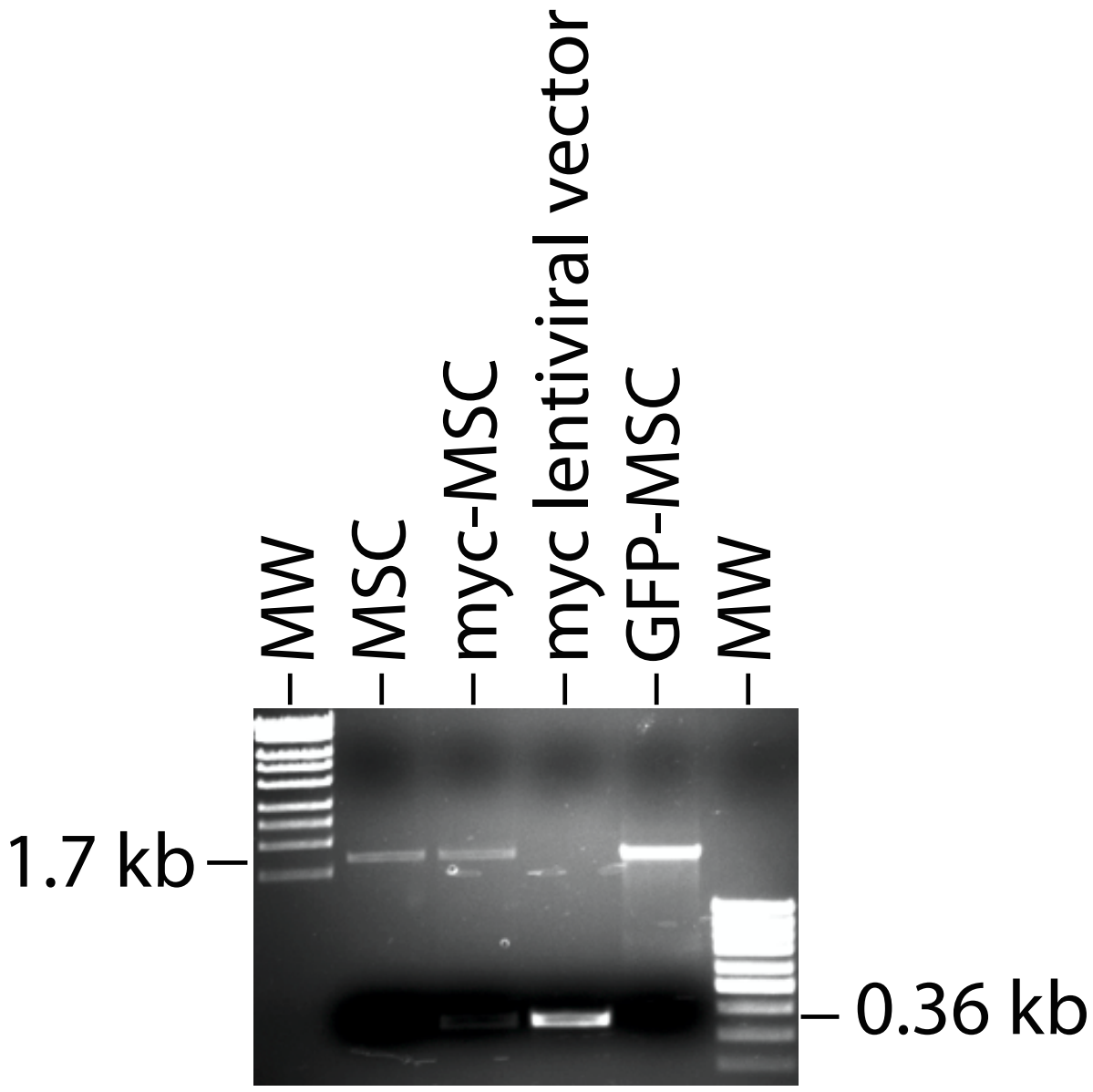


FIGURE 7

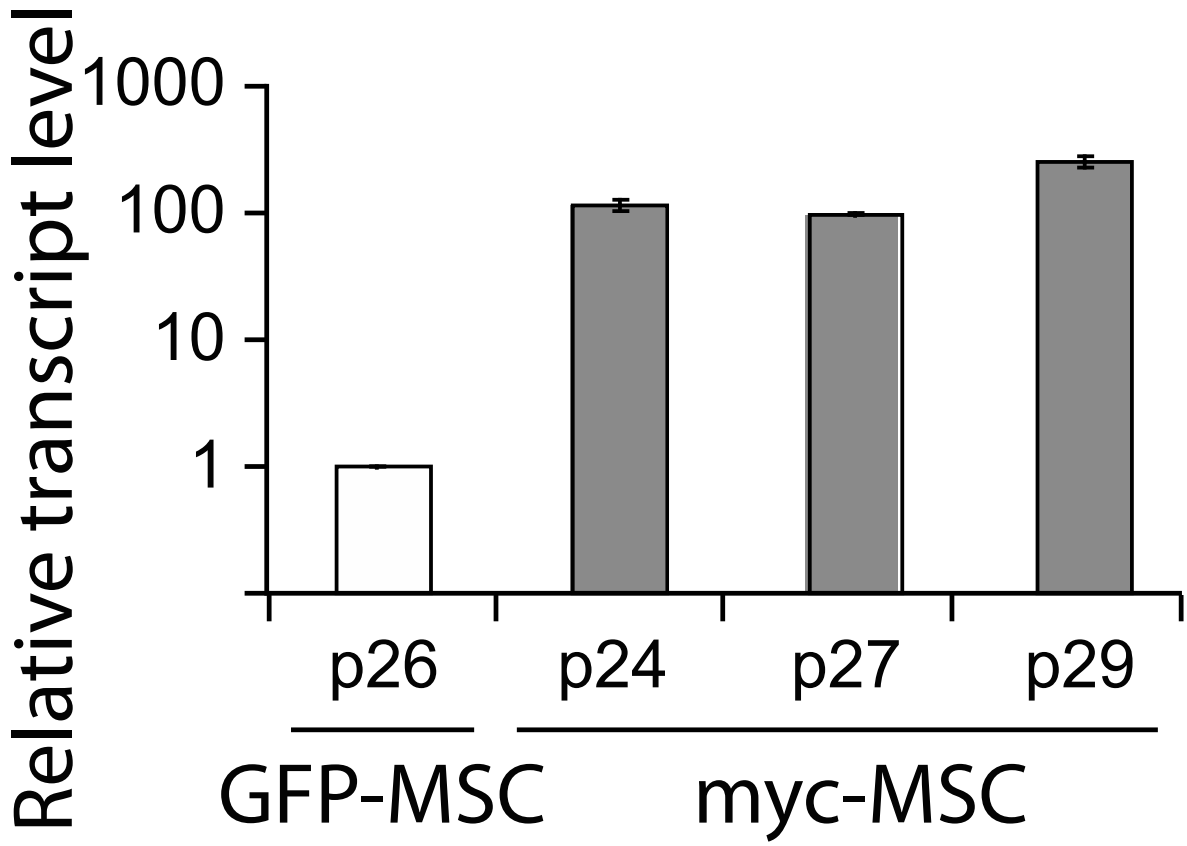

Relative transcript level
1000
100
10
1
p26
p24
p27
p29
GFP-MSC
myc-MSC

FIGURE 8

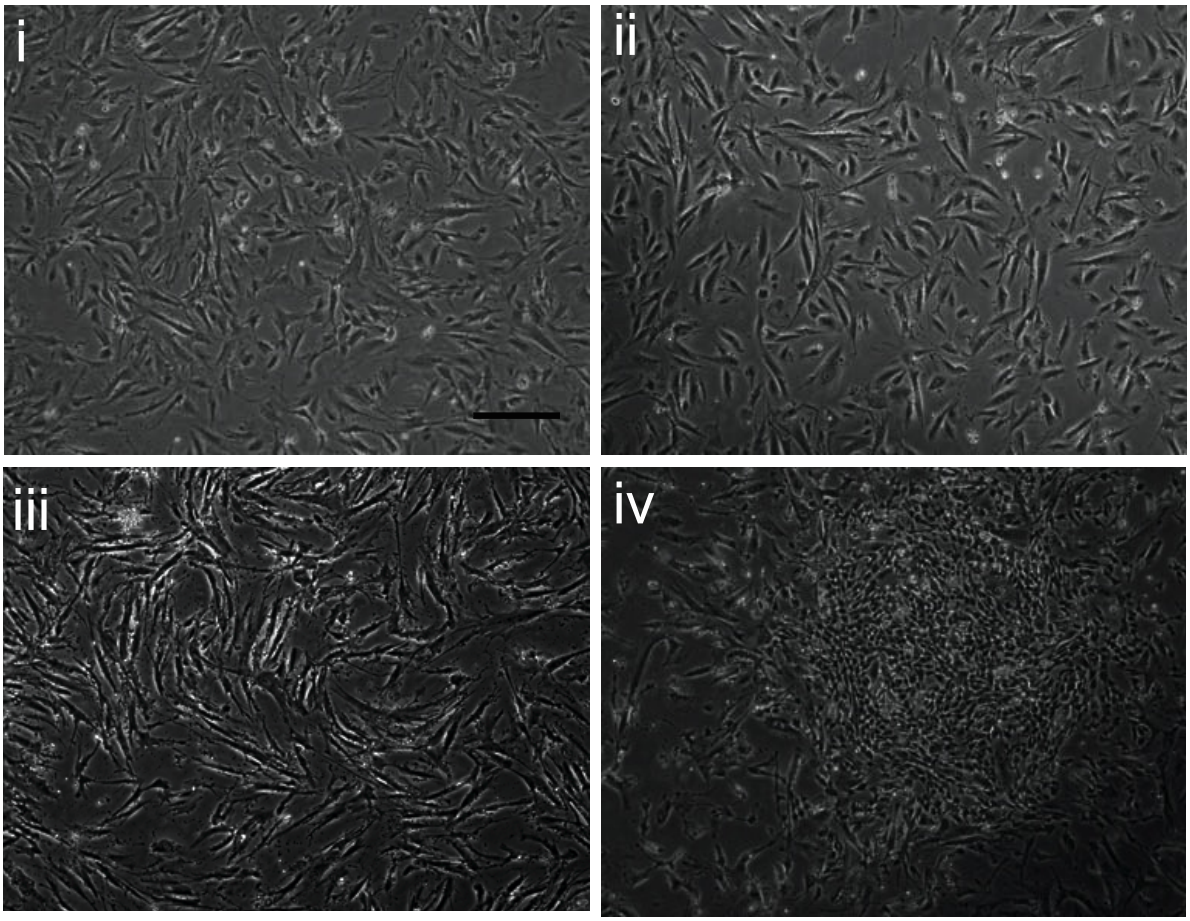


FIGURE 9

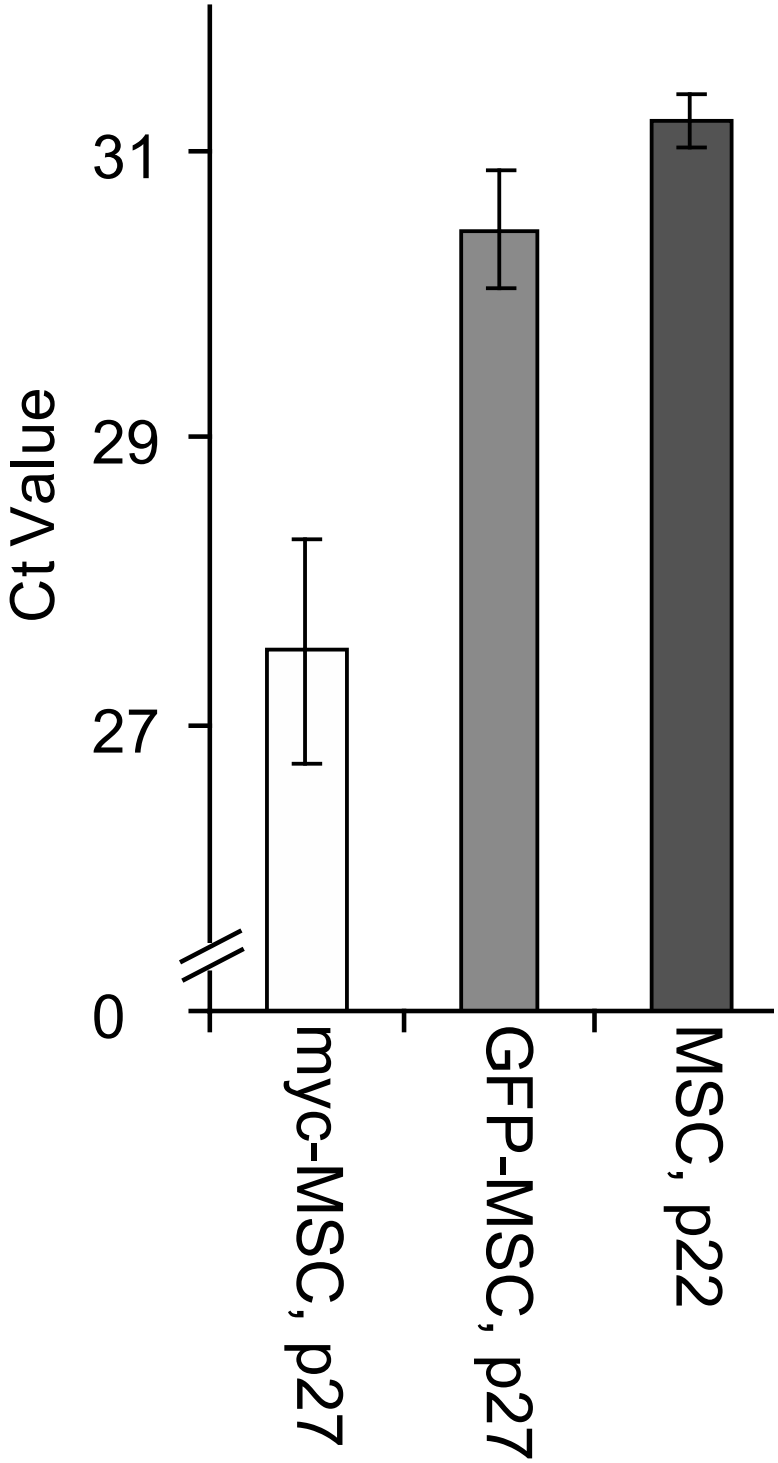


FIGURE 10

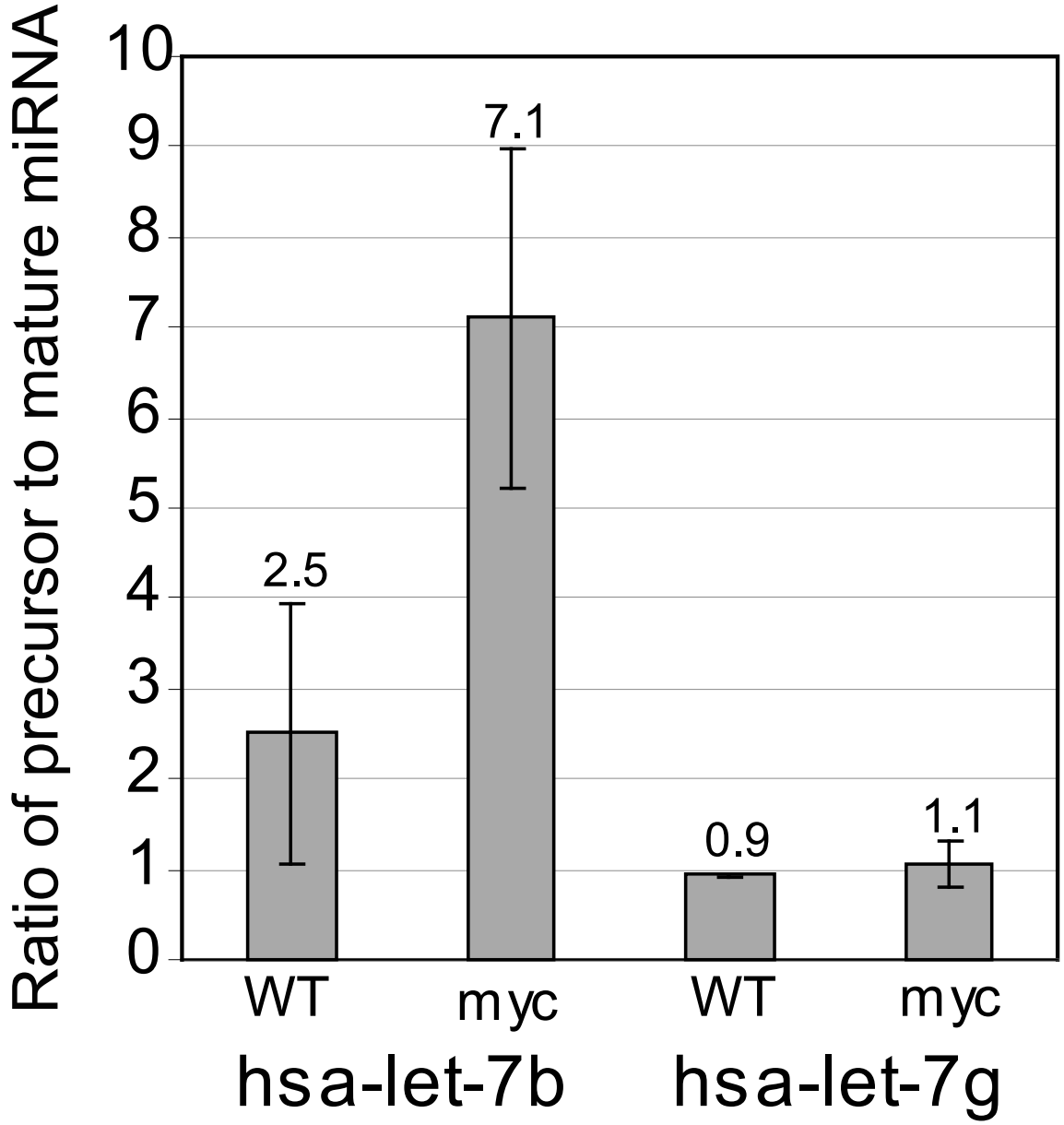

Ratio of precursor to mature miRNA
10
9
8
7
6
5
4
3
2
1
0
7.1
2.5
0.9
1.1
WT
myc
WT
myc
hsa-let-7b
hsa-let-7g

FIGURE 11

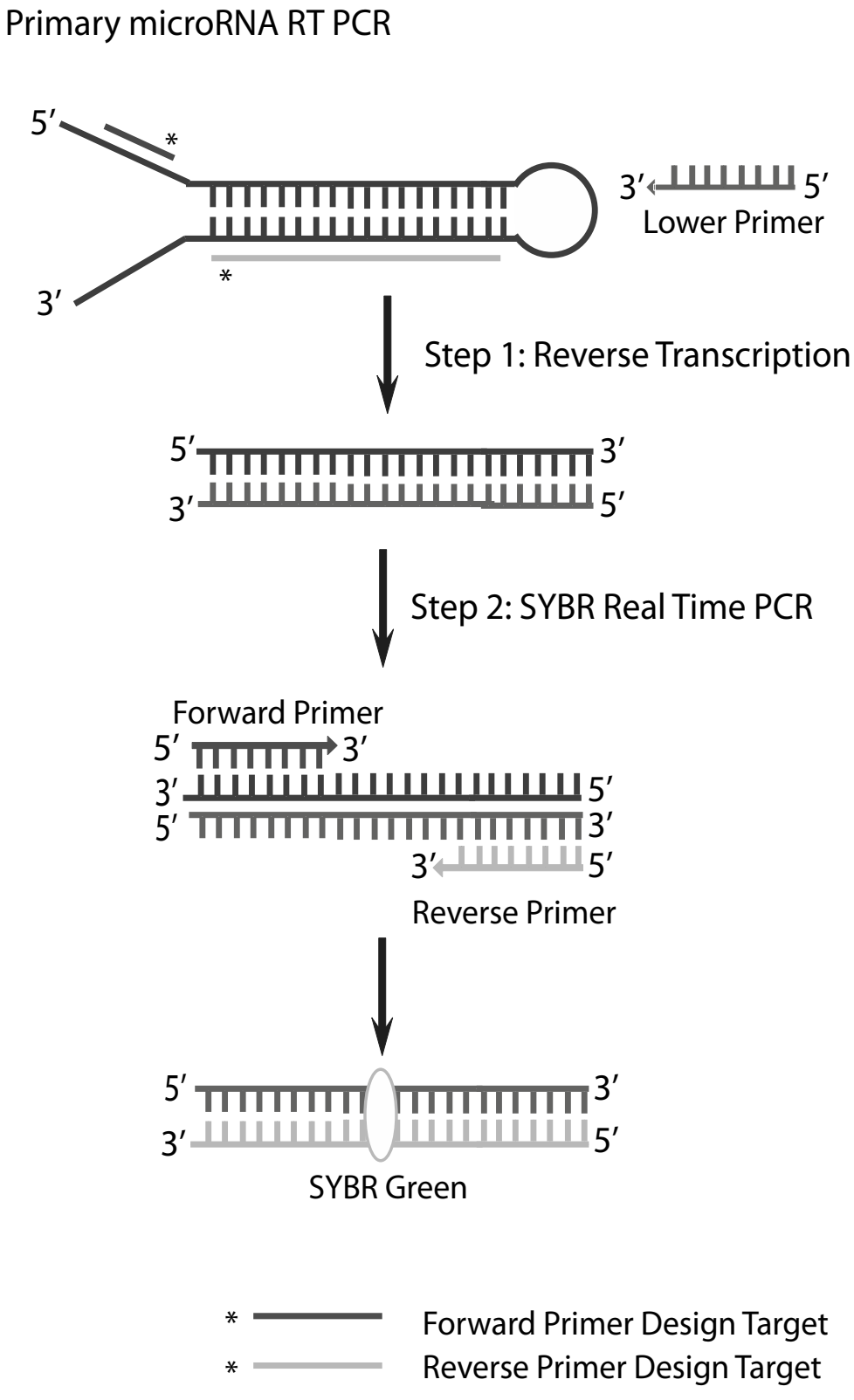

Primary microRNA RT PCR
5′
*
3′
3′
5′
Lower Primer
*
Step 1: Reverse Transcription
5′
3′
3′
5′
Step 2: SYBR Real Time PCR
Forward Primer
5′
3′
3′
5′
5′
3′
3′
5′
Reverse Primer
5′
3′
3′
5′
SYBR Green
*
Forward Primer Design Target
*
Reverse Primer Design Target

FIGURE 12

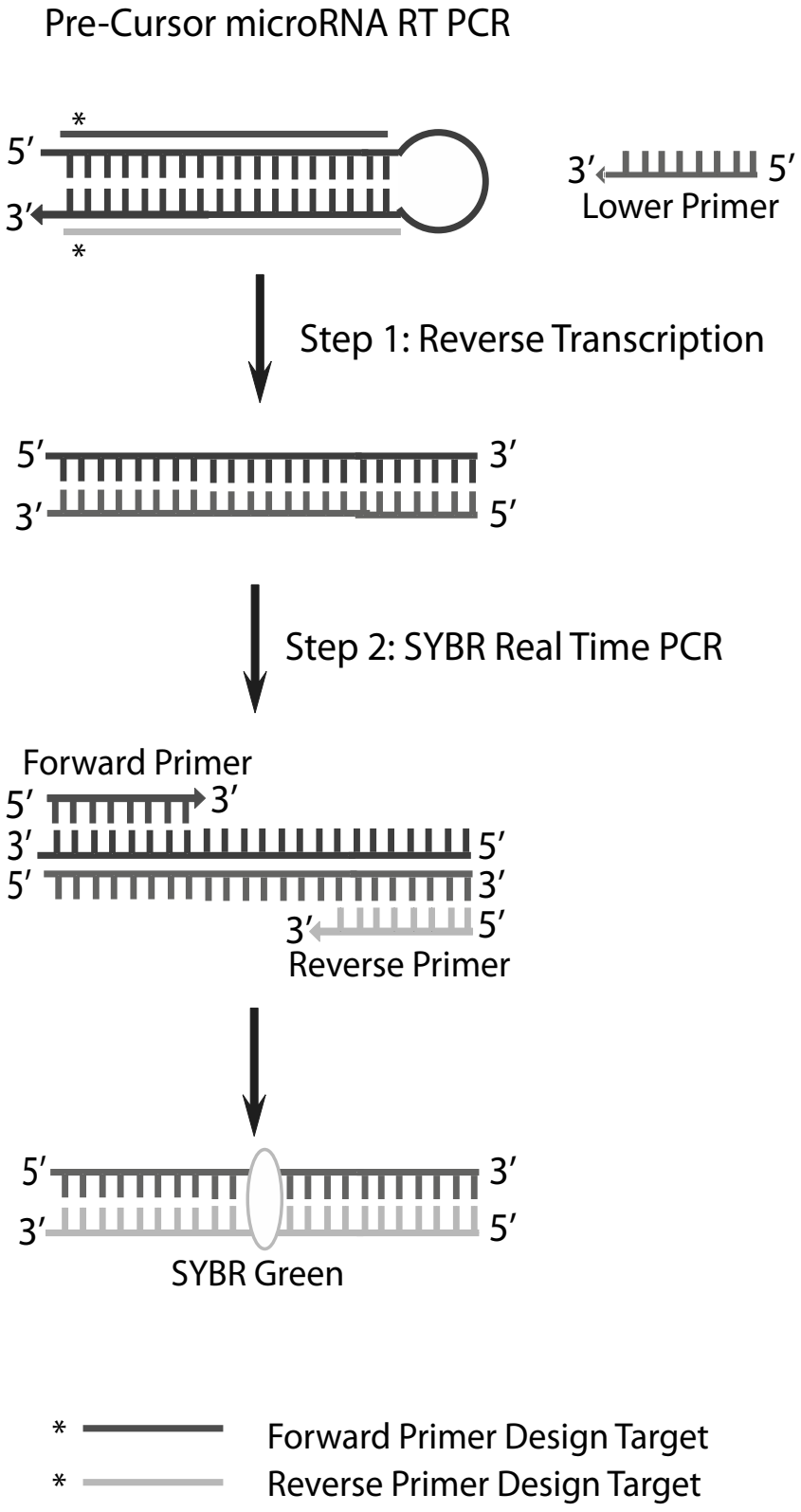
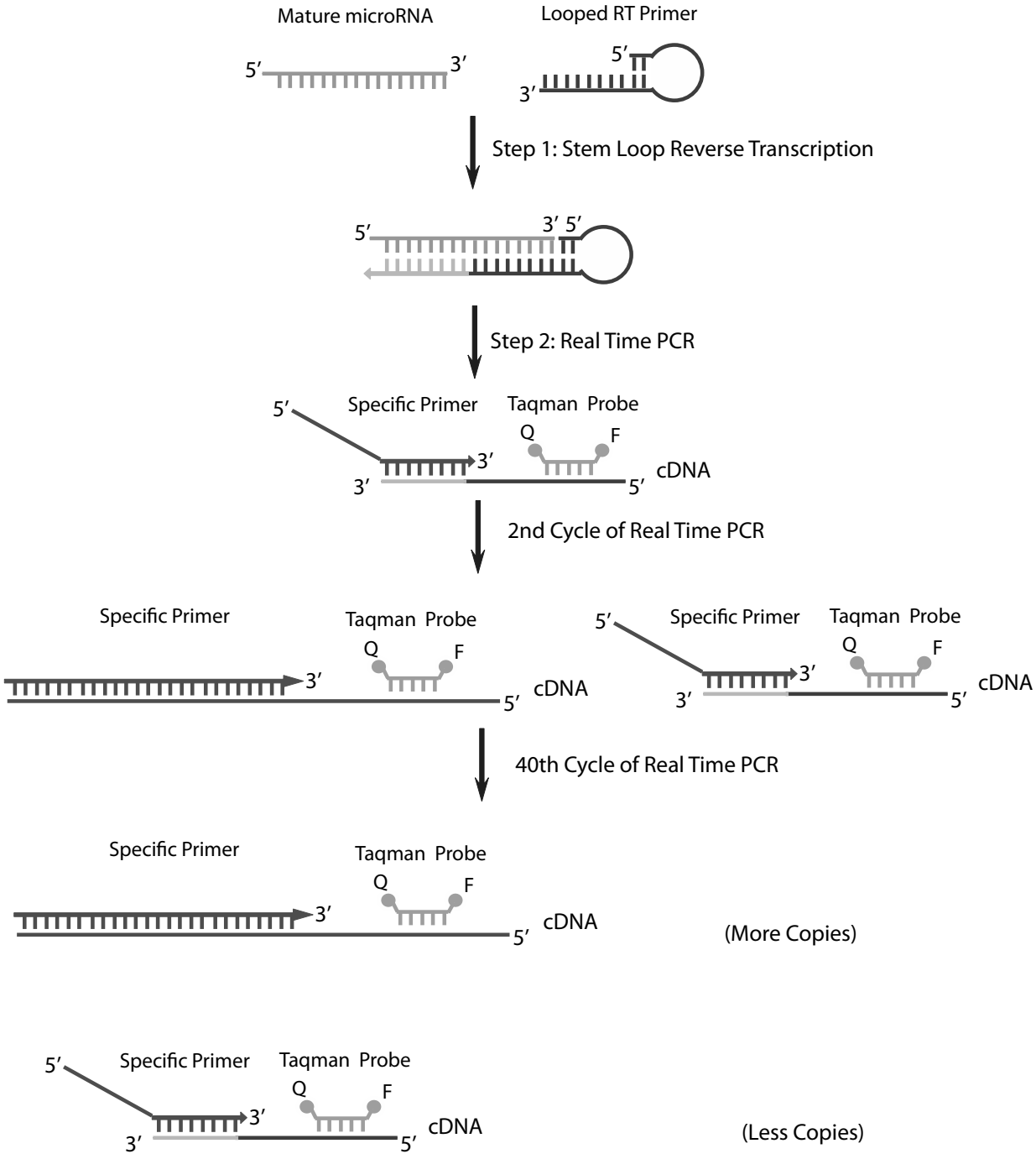

Pre-Cursor microRNA RT PCR
5′
3′
*
*
3′ 5′
Lower Primer
Step 1: Reverse Transcription
5′ 3′
3′ 5′
Step 2: SYBR Real Time PCR
Forward Primer
5′ 3′
3′ 5′
5′ 3′
3′ 5′
Reverse Primer
5′ 3′
3′ 5′
SYBR Green
* Forward Primer Design Target
* Reverse Primer Design Target

FIGURE 13

Mature microRNA RT PCR