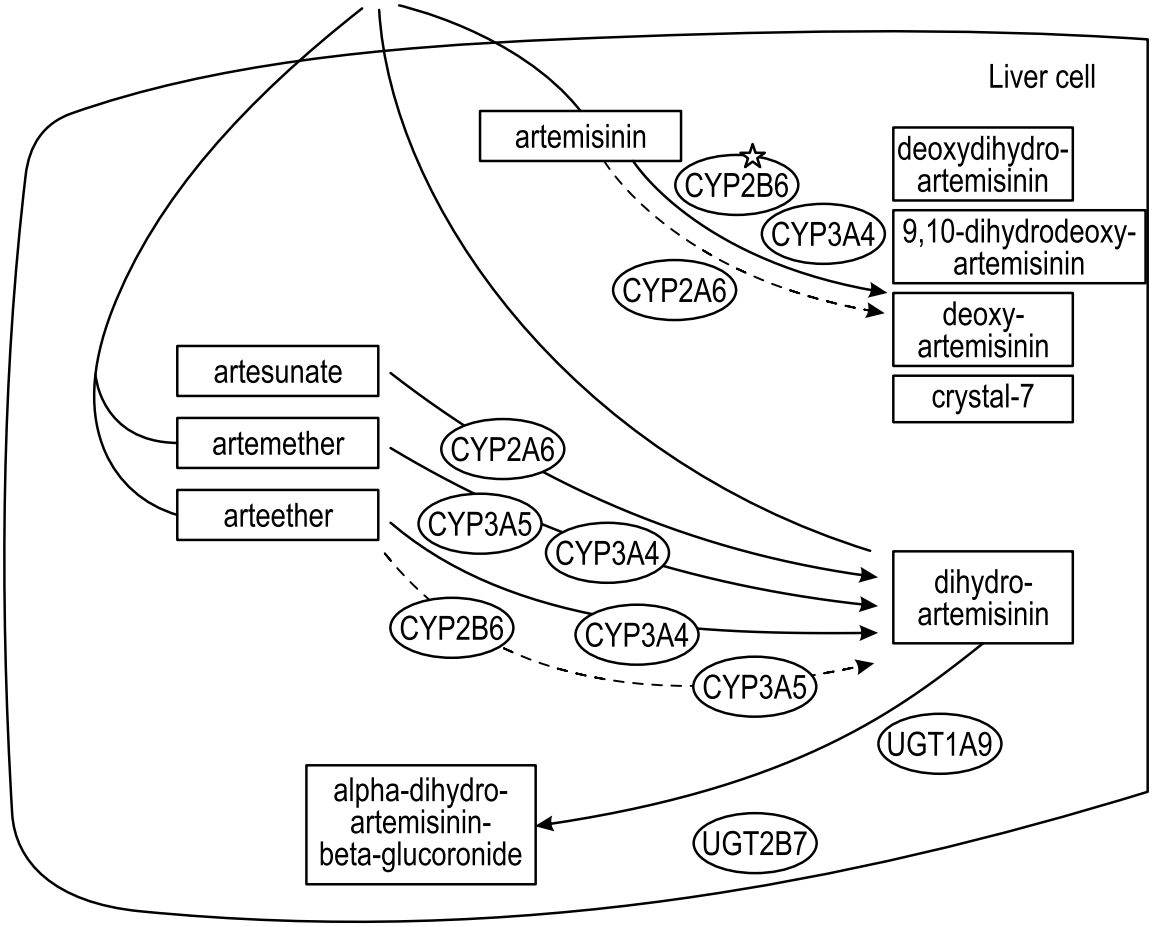

Liver cell
artemisinin
CYP2B6
CYP3A4
CYP2A6
deoxydihydro-artemisinin
9,10-dihydrodeoxy-artemisinin
deoxy-artemisinin
crystal-7
artesunate
artemether
arteether
CYP2A6
CYP3A5
CYP3A4
CYP2B6
CYP3A4
CYP3A5
dihydro-artemisinin
UGT1A9
alpha-dihydro-artemisinin-beta-glucoronide
UGT2B7

FIG. 1

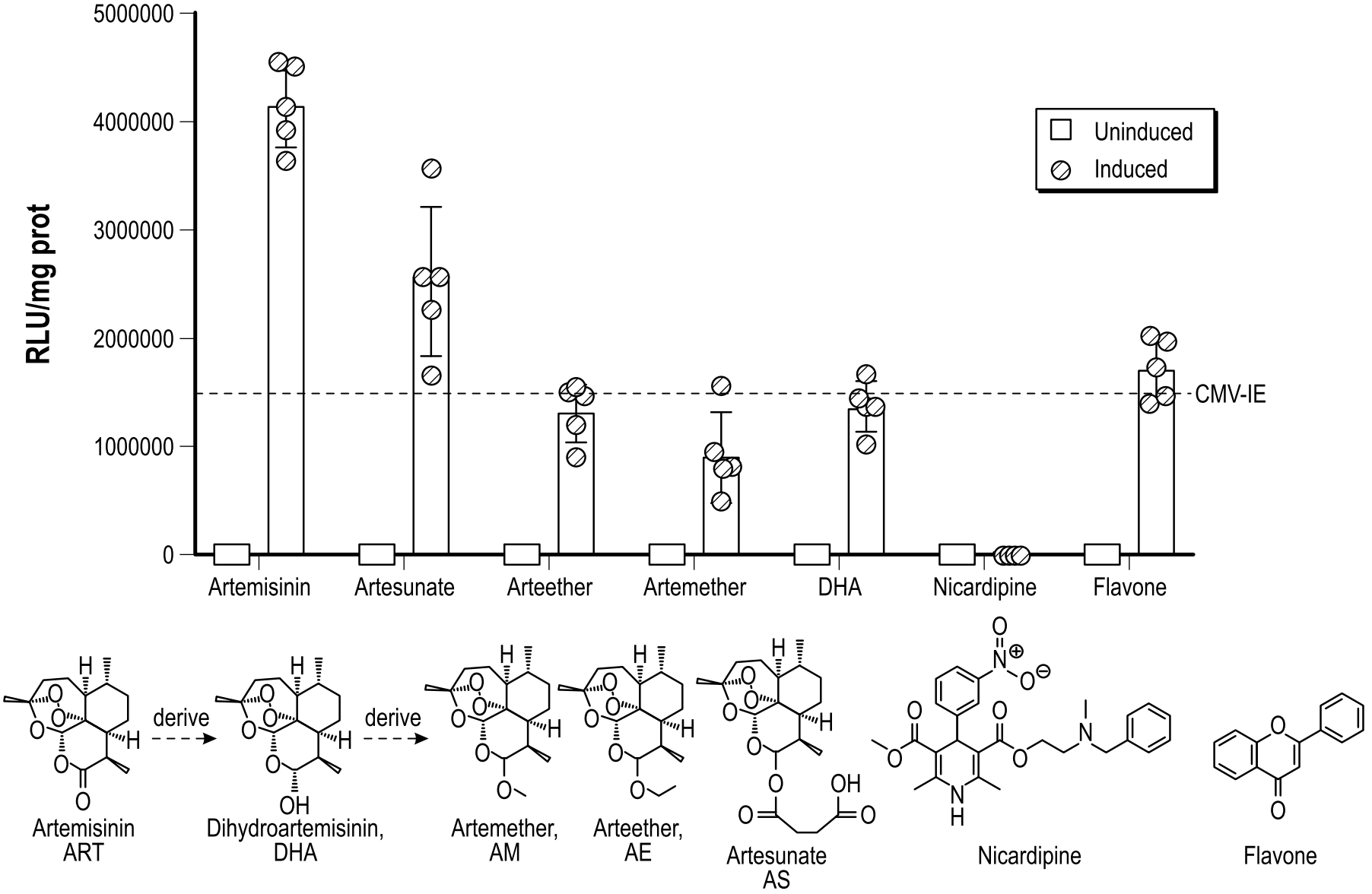

RLU/mg prot
5000000
4000000
3000000
2000000
1000000
0
Uninduced
Induced
CMV-IE
Artemisinin
Artesunate
Arteether
Artemether
DHA
Nicardipine
Flavone
derive
derive
Artemisinin
ART
Dihydroartemisinin,
DHA
Artemether,
AM
Arteether,
AE
Artesunate
AS
Nicardipine
Flavone

FIG. 2

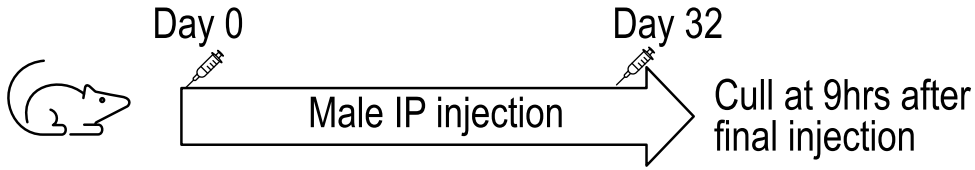


FIG. 3A

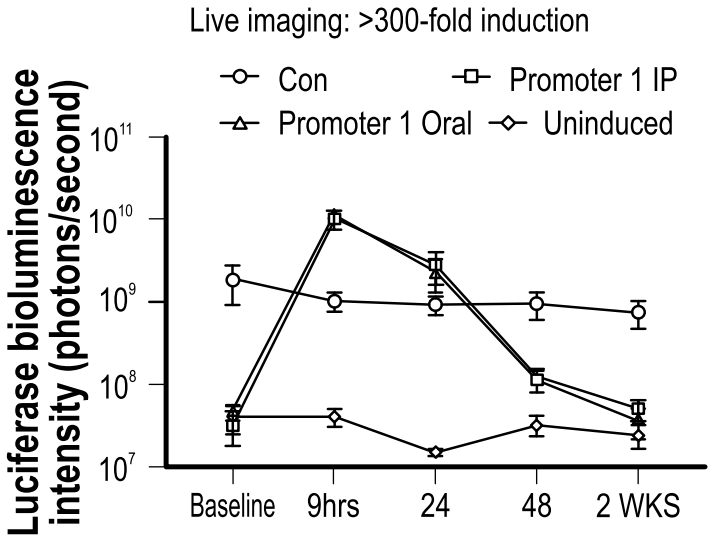


FIG. 3B

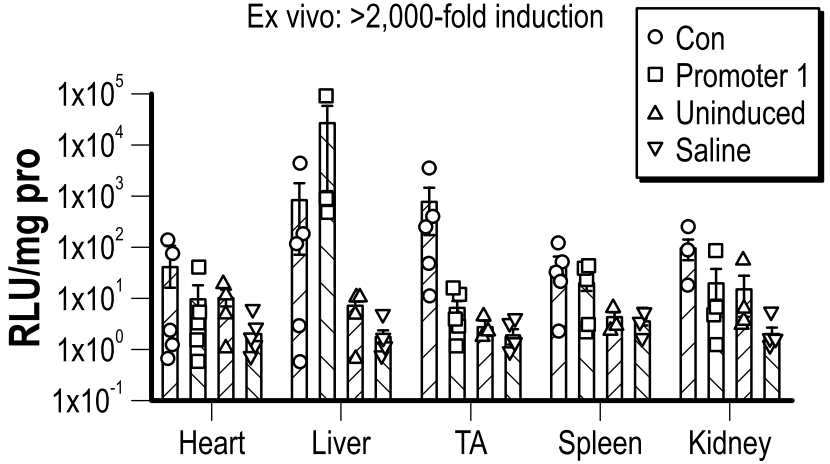


FIG. 3C

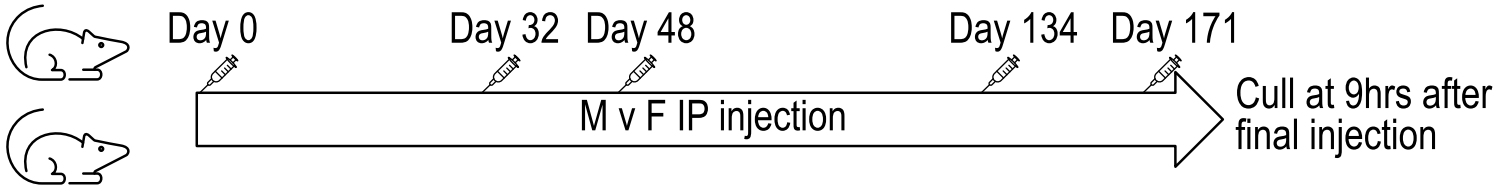


FIG. 4A

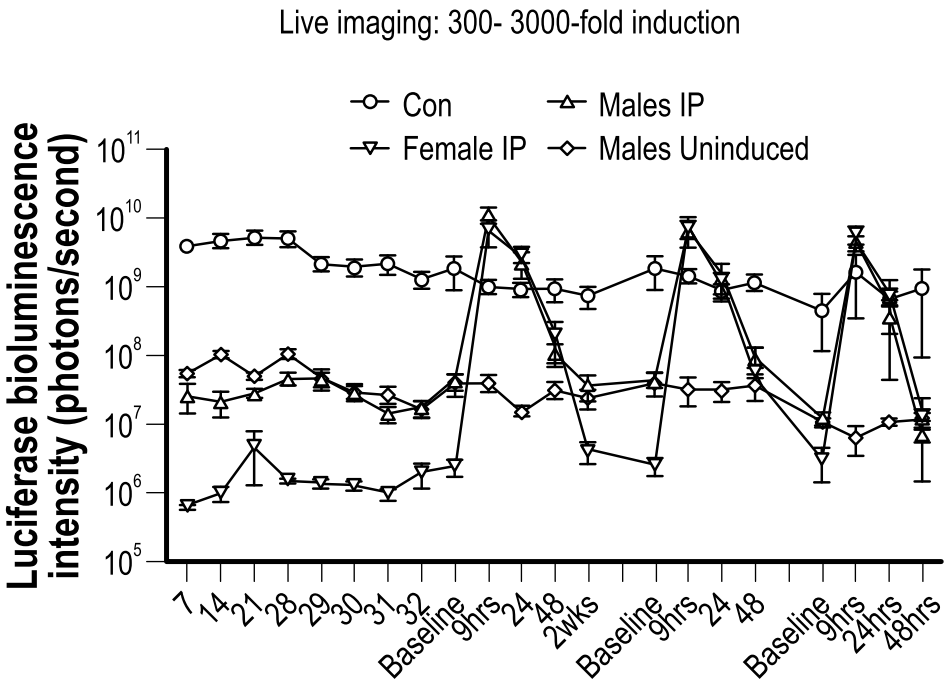


FIG. 4B

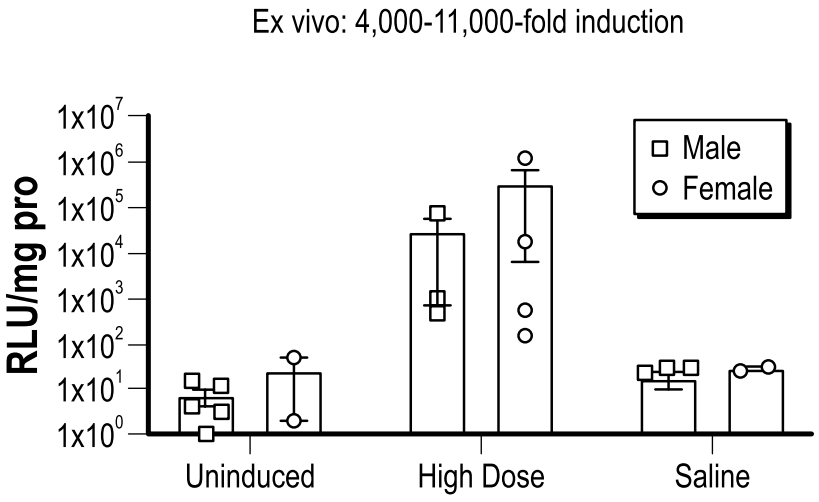


FIG. 4C

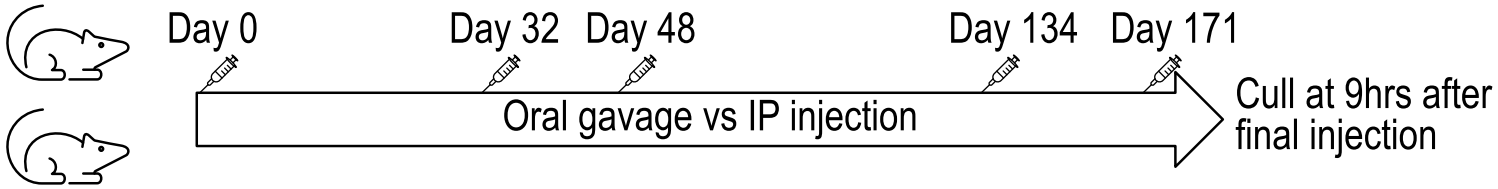


FIG. 5A

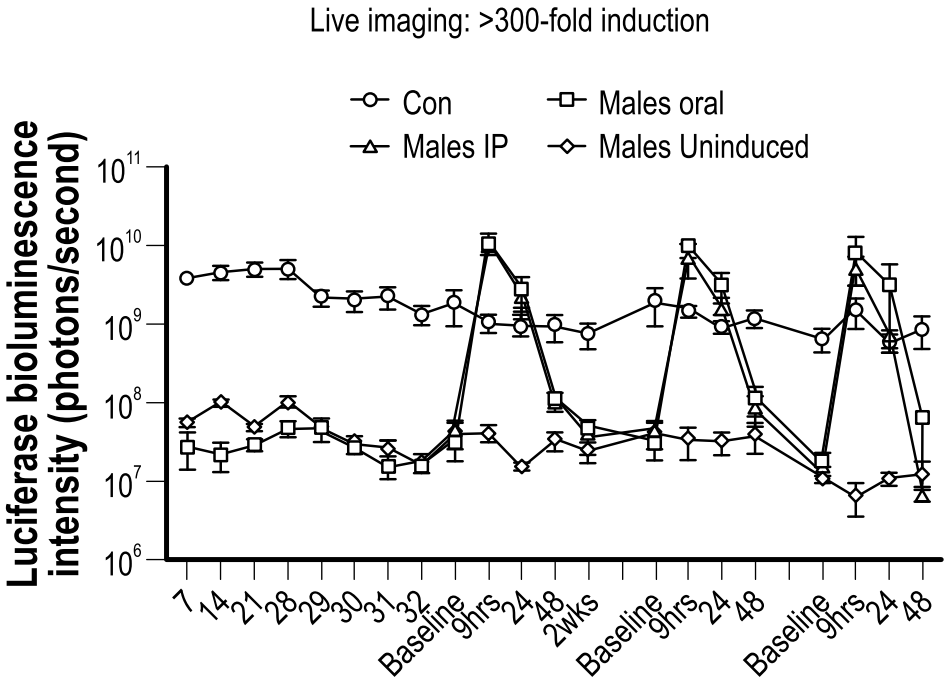


FIG. 5B

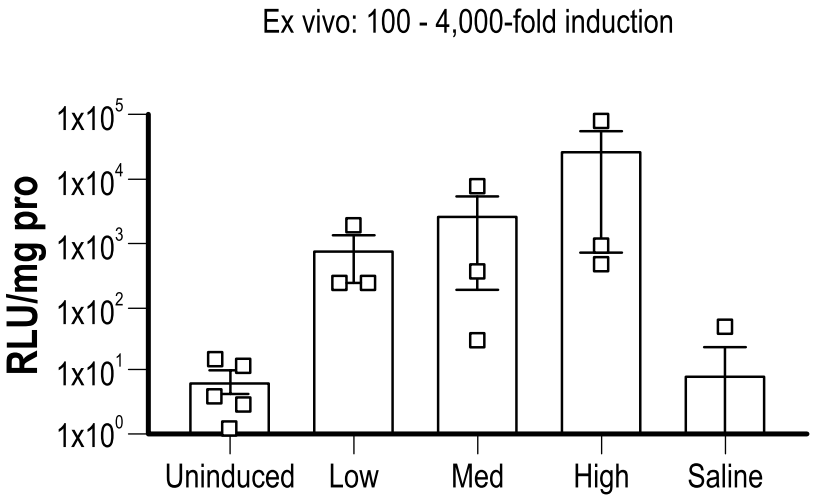


FIG. 5C

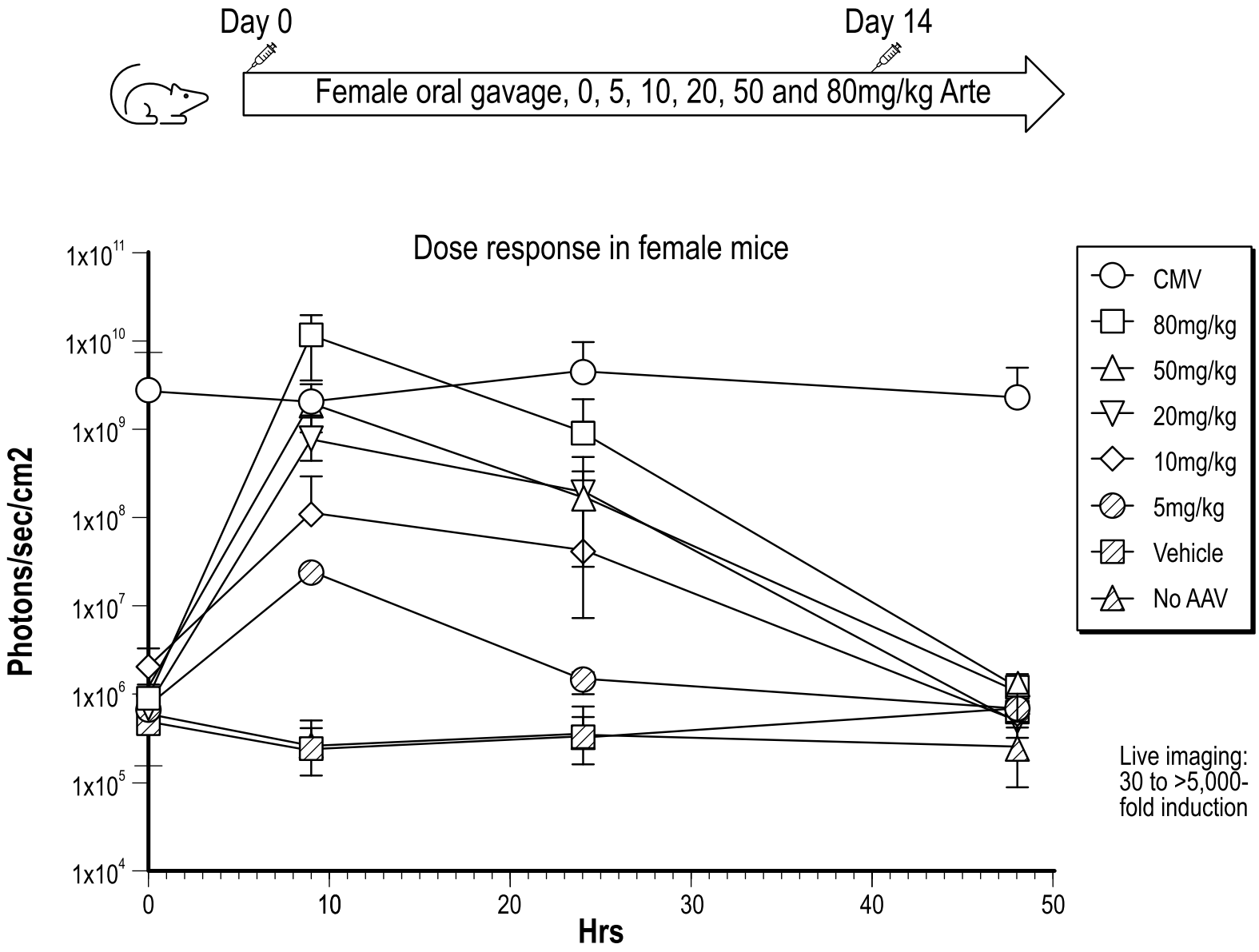

Day 0
Day 14
Female oral gavage, 0, 5, 10, 20, 50 and 80mg/kg Arte
Dose response in female mice
Photons/sec/cm2
1x10^11
1x10^10
1x10^9
1x10^8
1x10^7
1x10^6
1x10^5
1x10^4
0
10
20
30
40
50
Hrs
CMV
80mg/kg
50mg/kg
20mg/kg
10mg/kg
5mg/kg
Vehicle
No AAV
Live imaging:
30 to >5,000-
fold induction

FIG. 6

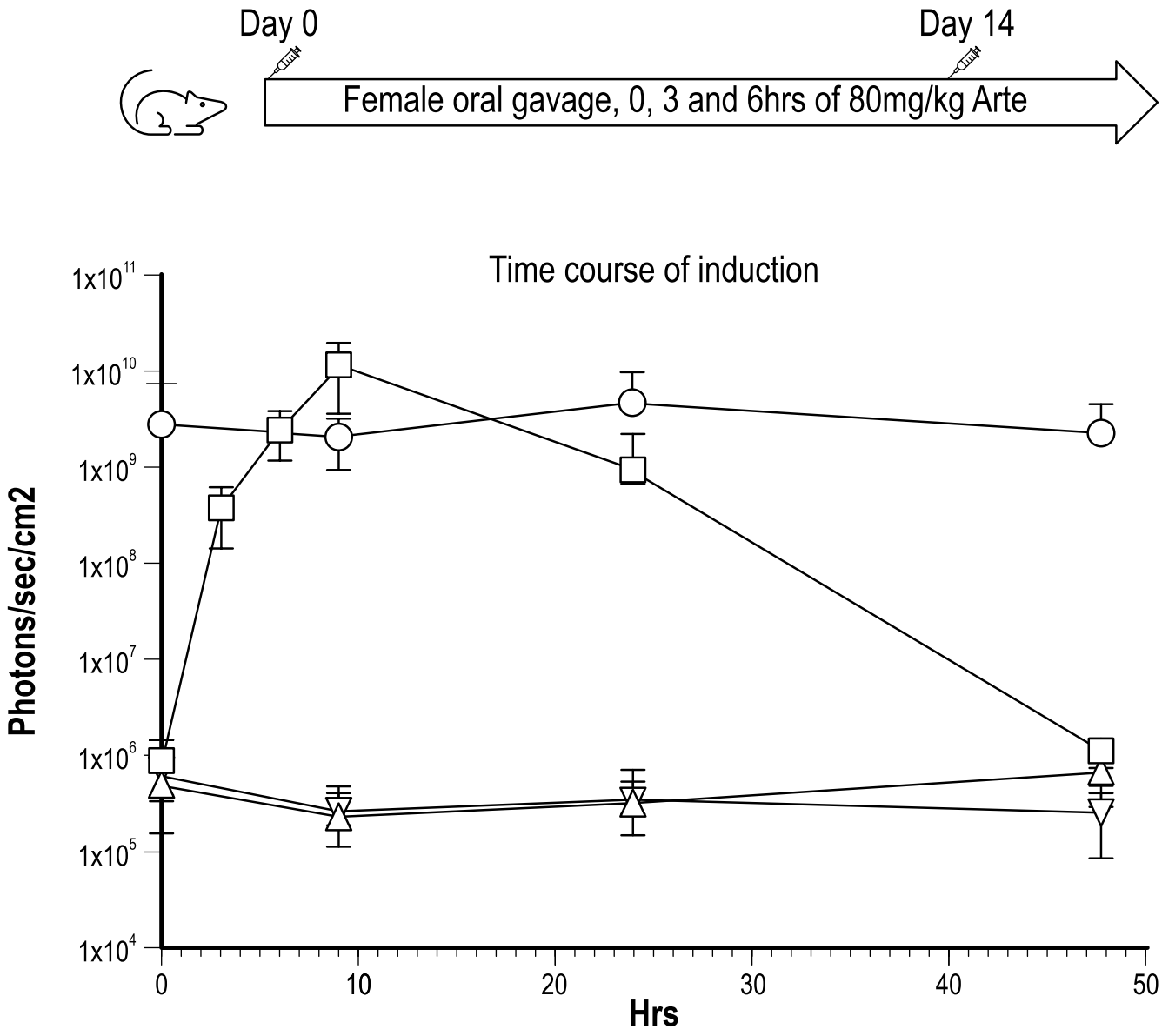

Day 0
Day 14
Female oral gavage, 0, 3 and 6hrs of 80mg/kg Arte
Time course of induction
Photons/sec/cm2
1x10^11
1x10^10
1x10^9
1x10^8
1x10^7
1x10^6
1x10^5
1x10^4
0
10
20
30
40
50
Hrs
CMV
Ind-80mg/kg
Vehicle
No AAV
Live imaging:
9hrs is peak induction time
but onset is rapid with
significant induction
observed at 3hrs.

FIG. 7

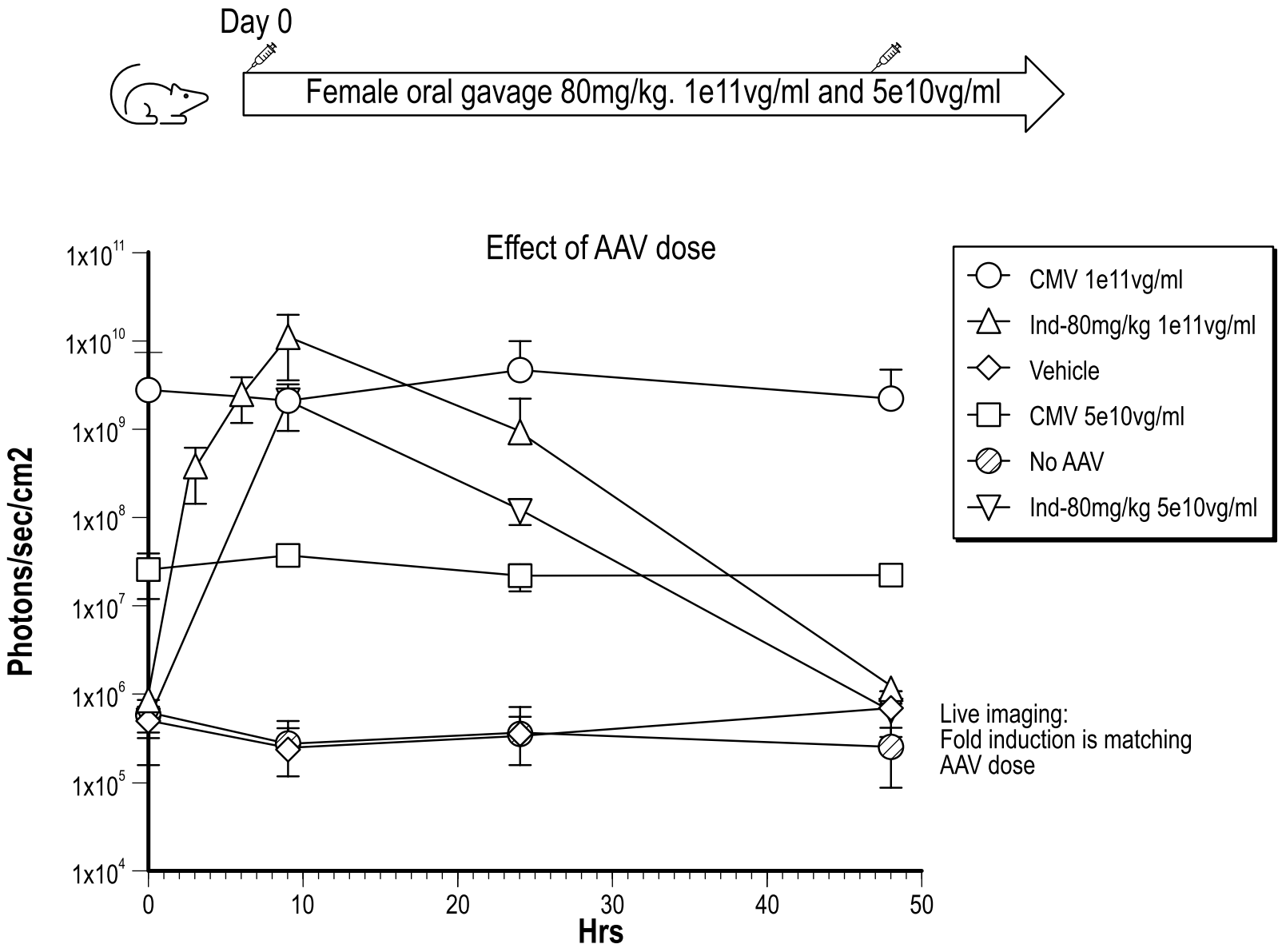

Day 0
Female oral gavage 80mg/kg. 1e11vg/ml and 5e10vg/ml
Effect of AAV dose
Photons/sec/cm2
1x10^11
1x10^10
1x10^9
1x10^8
1x10^7
1x10^6
1x10^5
1x10^4
0
10
20
30
40
50
Hrs
CMV 1e11vg/ml
Ind-80mg/kg 1e11vg/ml
Vehicle
CMV 5e10vg/ml
No AAV
Ind-80mg/kg 5e10vg/ml
Live imaging:
Fold induction is matching
AAV dose

FIG. 8

Groups	Dose vg/kg	No of NHPs	Transgene	AAV	Drug/Route	Analytics
Control Promoter	1x10^13	2 (1M, 1F)	Chorionic Gonadotropin	9	IP-80mg/kg	ELISA of Serum
Inducible Promoter	1x10^13	4 (2M, 2F)	Chorionic Gonadotropin	9	IP-80mg/kg	ELISA of Serum
Negative	Zero	2 (1M, 1F)	N/A	N/A	N/A	ELISA of Serum

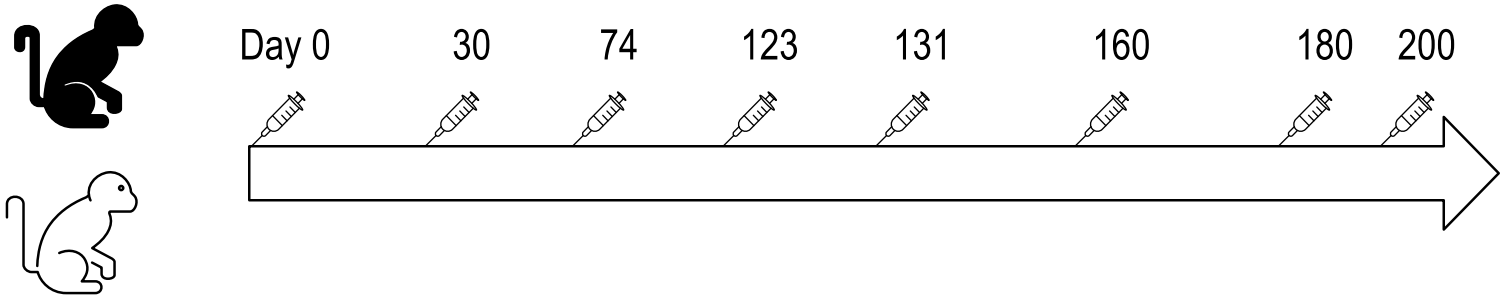


FIG. 9

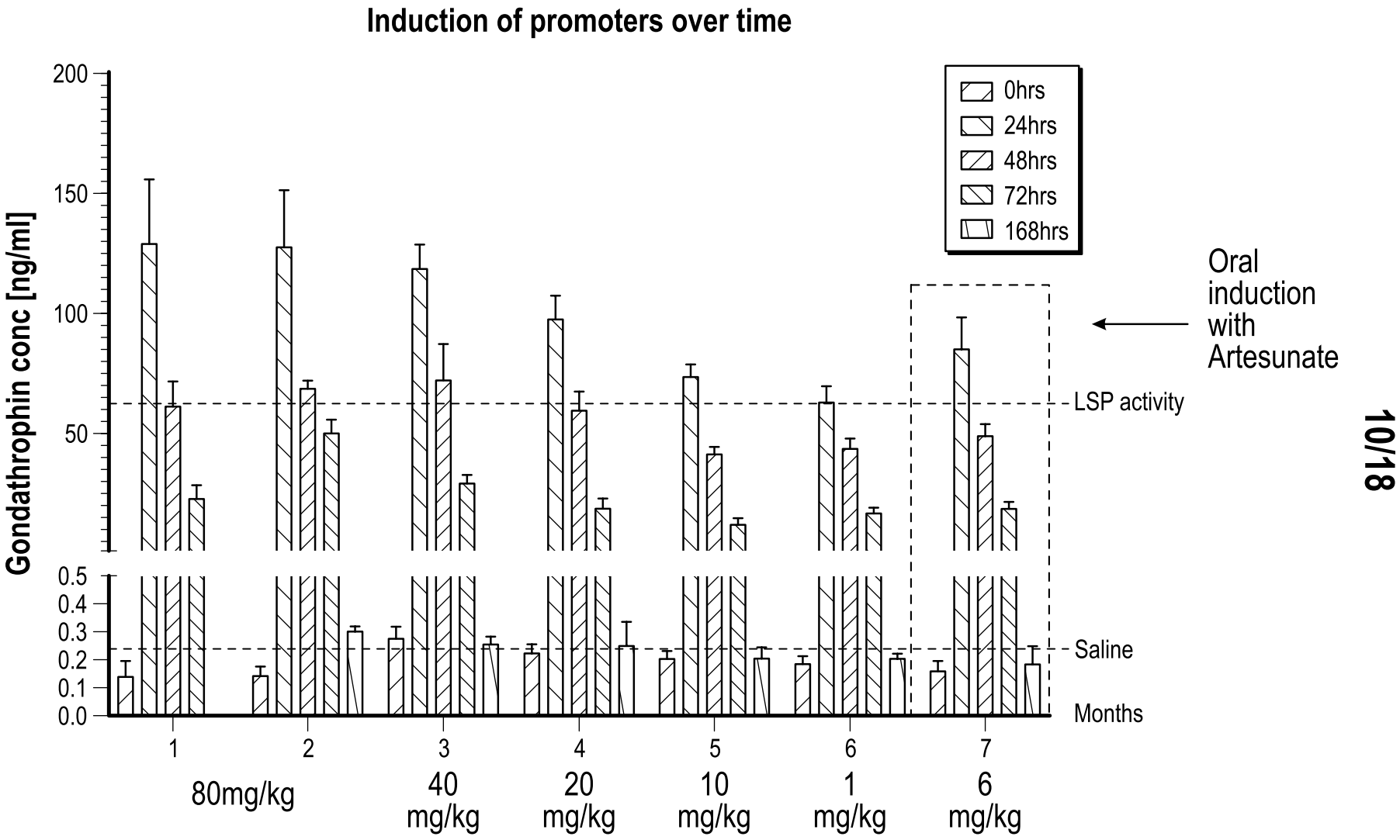

Induction of promoters over time
Gondathrophin conc [ng/ml]
200
150
100
50
0.5
0.4
0.3
0.2
0.1
0.0
0hrs
24hrs
48hrs
72hrs
168hrs
Oral induction with Artesunate
LSP activity
Saline
Months
1
2
3
4
5
6
7
80mg/kg
40 mg/kg
20 mg/kg
10 mg/kg
1 mg/kg
6 mg/kg

FIG. 10

Groups	Dose vg/kg	No of NHPs	Transgene	AAV	Drug/Route	Analytics
Control Promoter	1x10^13	2 (1M, 1F)	Chorionic Gonadotropin	9	IP-80mg/kg	ELISA of Serum
Inducible Promoter	1x10^13	4 (2M, 2F)	Chorionic Gonadotropin	9	IP-80mg/kg	ELISA of Serum
Negative	Zero	2 (1M, 1F)	N/A	N/A	N/A	ELISA of Serum

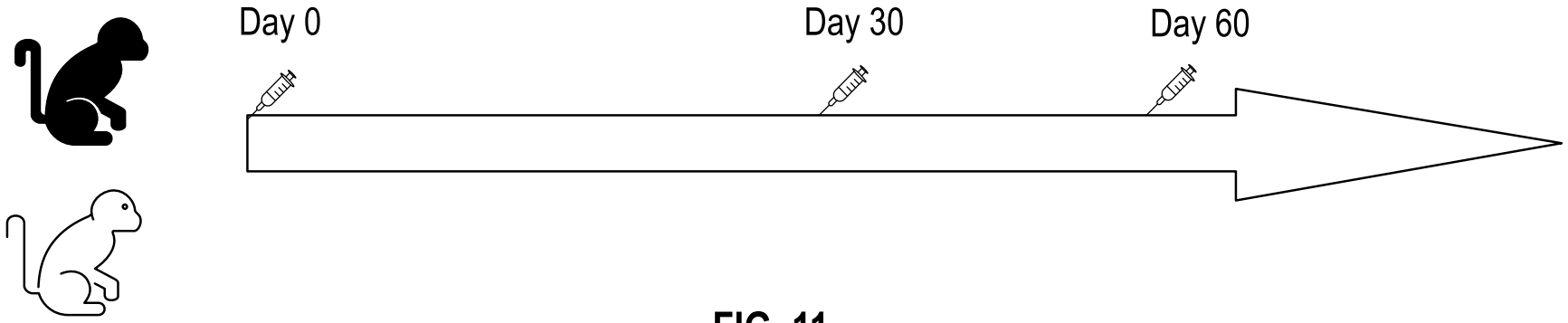


FIG. 11

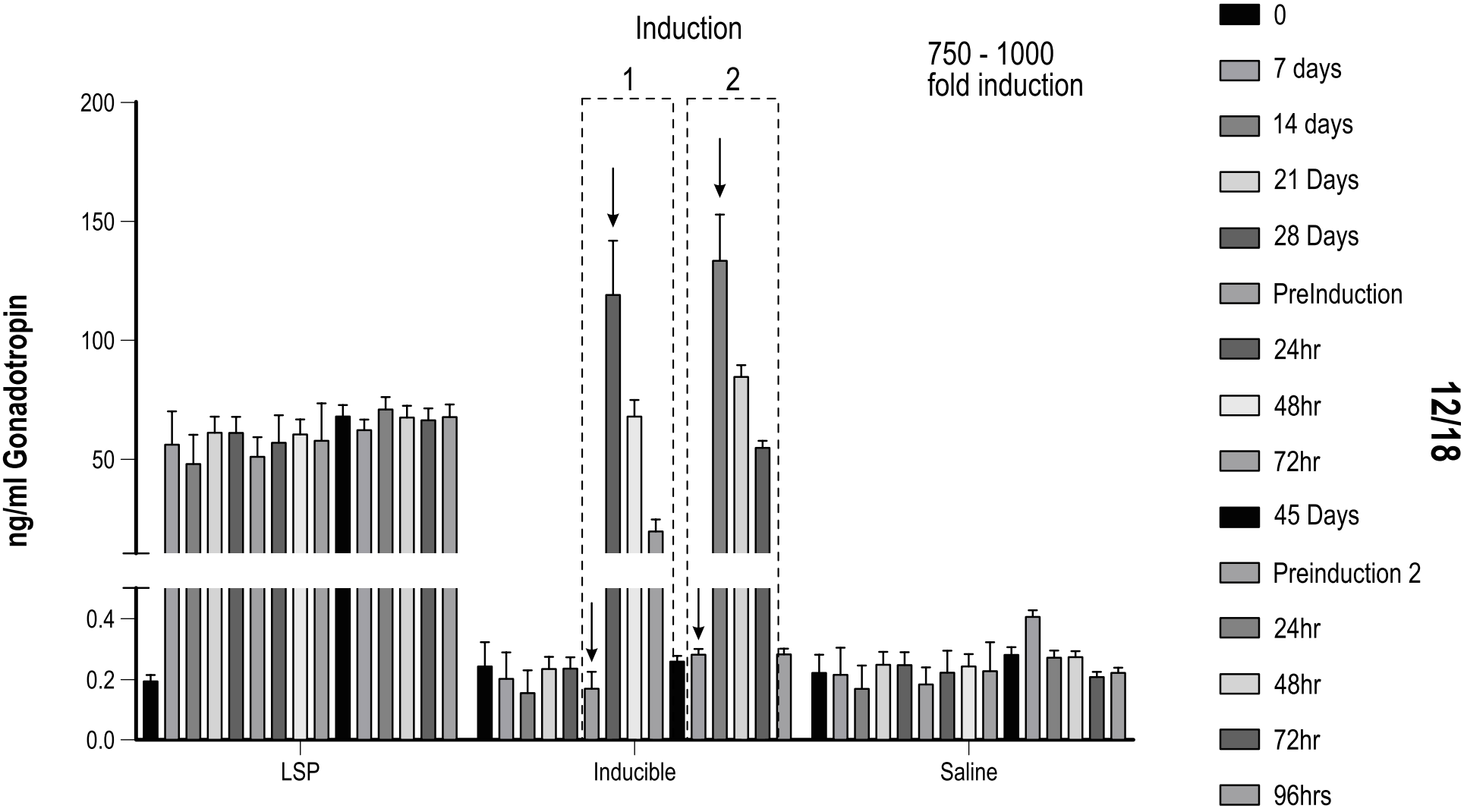

Induction
1
2
750 - 1000
fold induction
ng/ml Gonadotropin
200
150
100
50
0.4
0.2
0.0
LSP
Inducible
Saline
0
7 days
14 days
21 Days
28 Days
PreInduction
24hr
48hr
72hr
45 Days
Preinduction 2
24hr
48hr
72hr
96hrs

FIG. 12

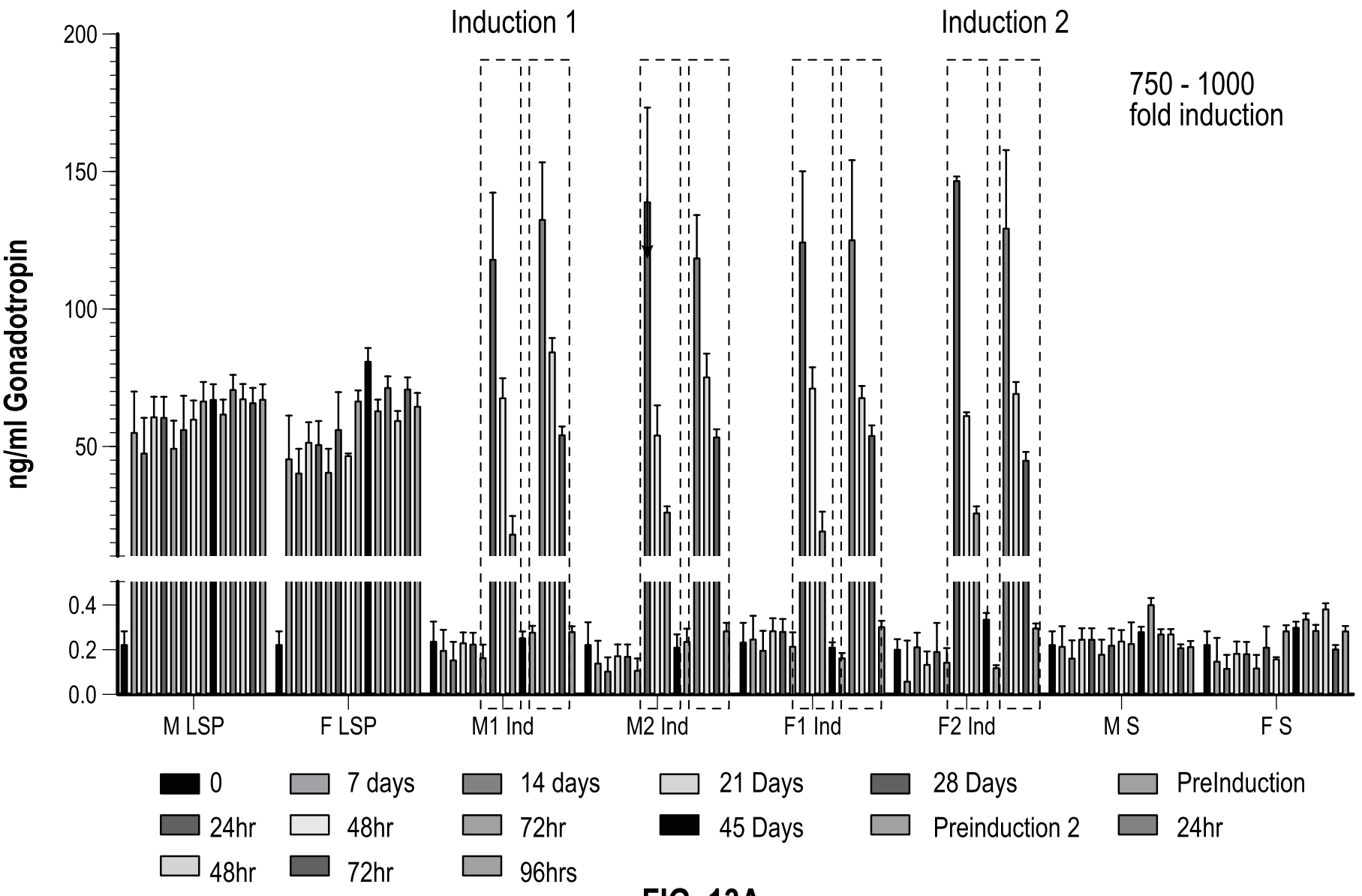

Induction 1
Induction 2
750 - 1000
fold induction
ng/ml Gonadotropin
200
150
100
50
0.4
0.2
0.0
M LSP
F LSP
M1 Ind
M2 Ind
F1 Ind
F2 Ind
M S
F S
0
7 days
14 days
21 Days
28 Days
PreInduction
24hr
48hr
72hr
45 Days
Preinduction 2
24hr
48hr
72hr
96hrs

FIG. 13A

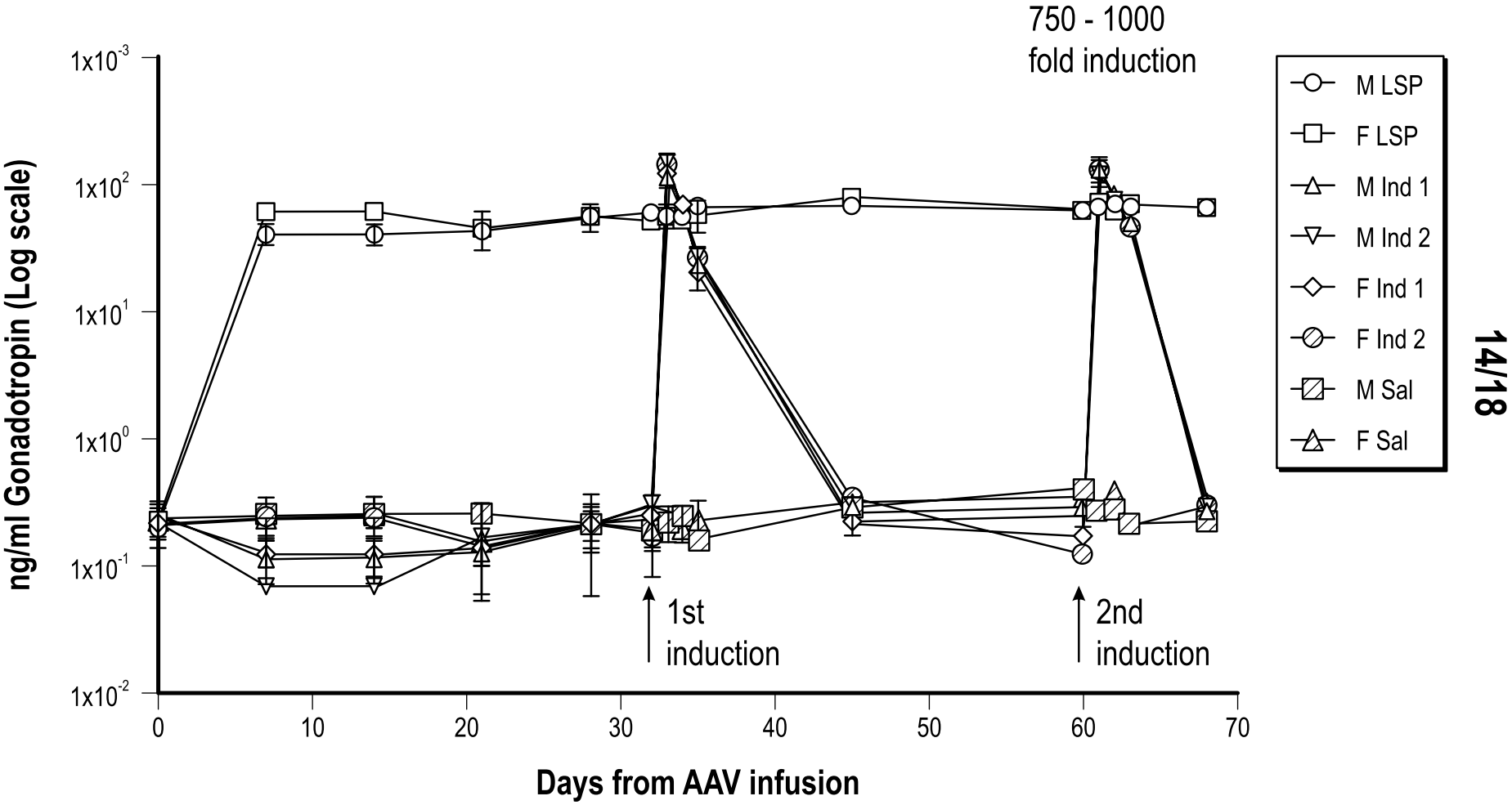
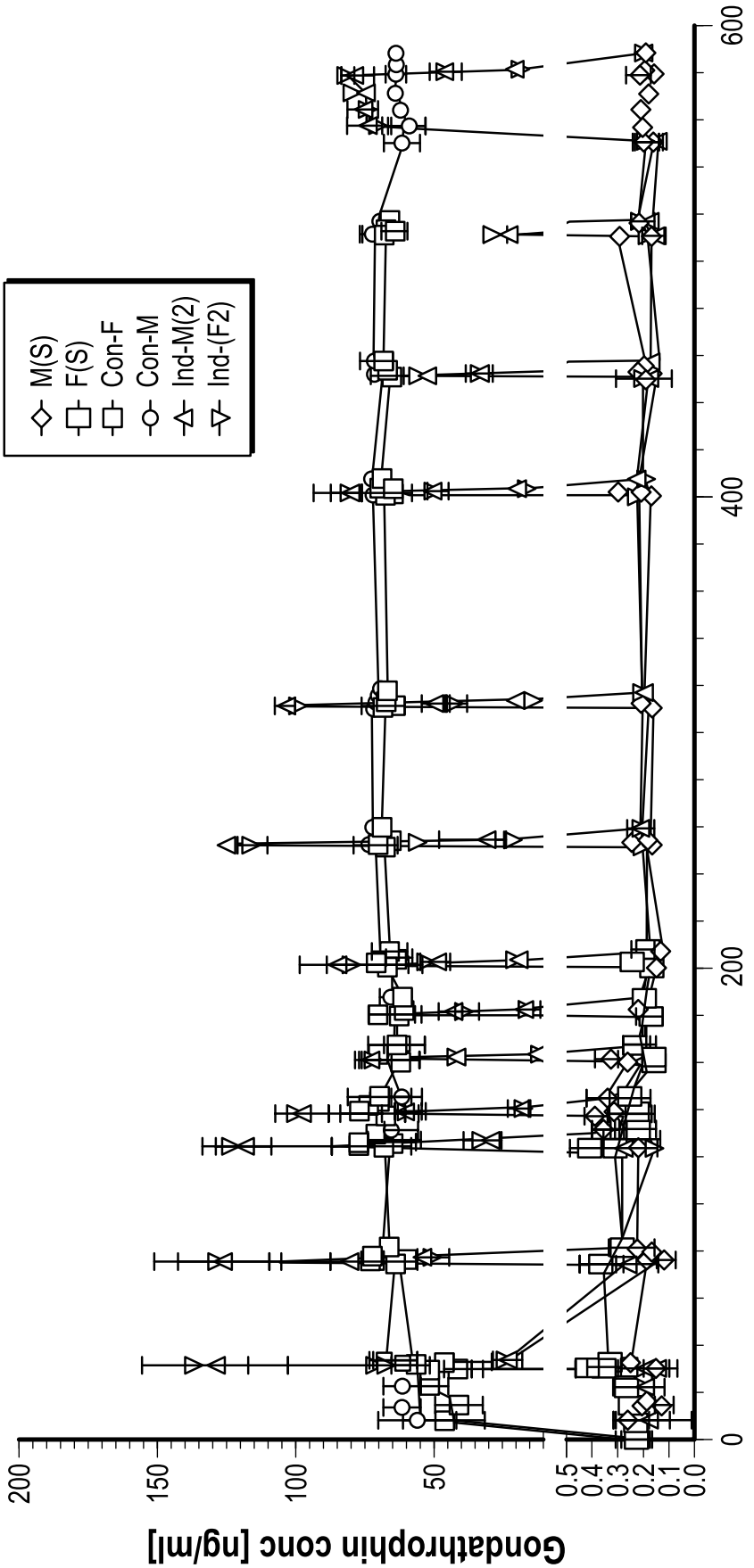

750 - 1000
fold induction
ng/ml Gonadotropin (Log scale)
1x10-3
1x10-2
1x10-1
1x100
1x101
1x102
0
10
20
30
40
50
60
70
Days from AAV infusion
1st
induction
2nd
induction
M LSP
F LSP
M Ind 1
M Ind 2
F Ind 1
F Ind 2
M Sal
F Sal

FIG. 13B

Groups	Dose vg/kg	No of NHPs	Transgene	AAV	Drug/Route	Analytics
Control Promoter	1x10^13	2 (1M, 1F)	Chorionic Gonadotropin	9	IP-80mg/kg	ELISA of Serum
Inducible Promoter	1x10^13	4 (2M, 2F)	Chorionic Gonadotropin	9	IP-80mg/kg	ELISA of Serum
Negative	Zero	2 (1M, 1F)	N/A	N/A	N/A	ELISA of Serum

Induction was performed with the following concentrations:
Artemisinin: IP injection at 80, 80, 40, 20, 10, 1mg/kg
Artesunate: oral ingestion at 20, 10, 6, 5, 1, 0.5, 0.1mg/kg
30 days continuous induction was performed via daily administration of 1mg/kg of artesunate orally.

FIG. 14



Inductions performed in following order: 80, 80, 40, 20, 10, 1 (IP), 6 (oral), 20, 10, 1, 0.5 and 0.1 (oral), mg/kg. Last induction set was 30-days continuous 1mg/kg oral.

FIG. 15

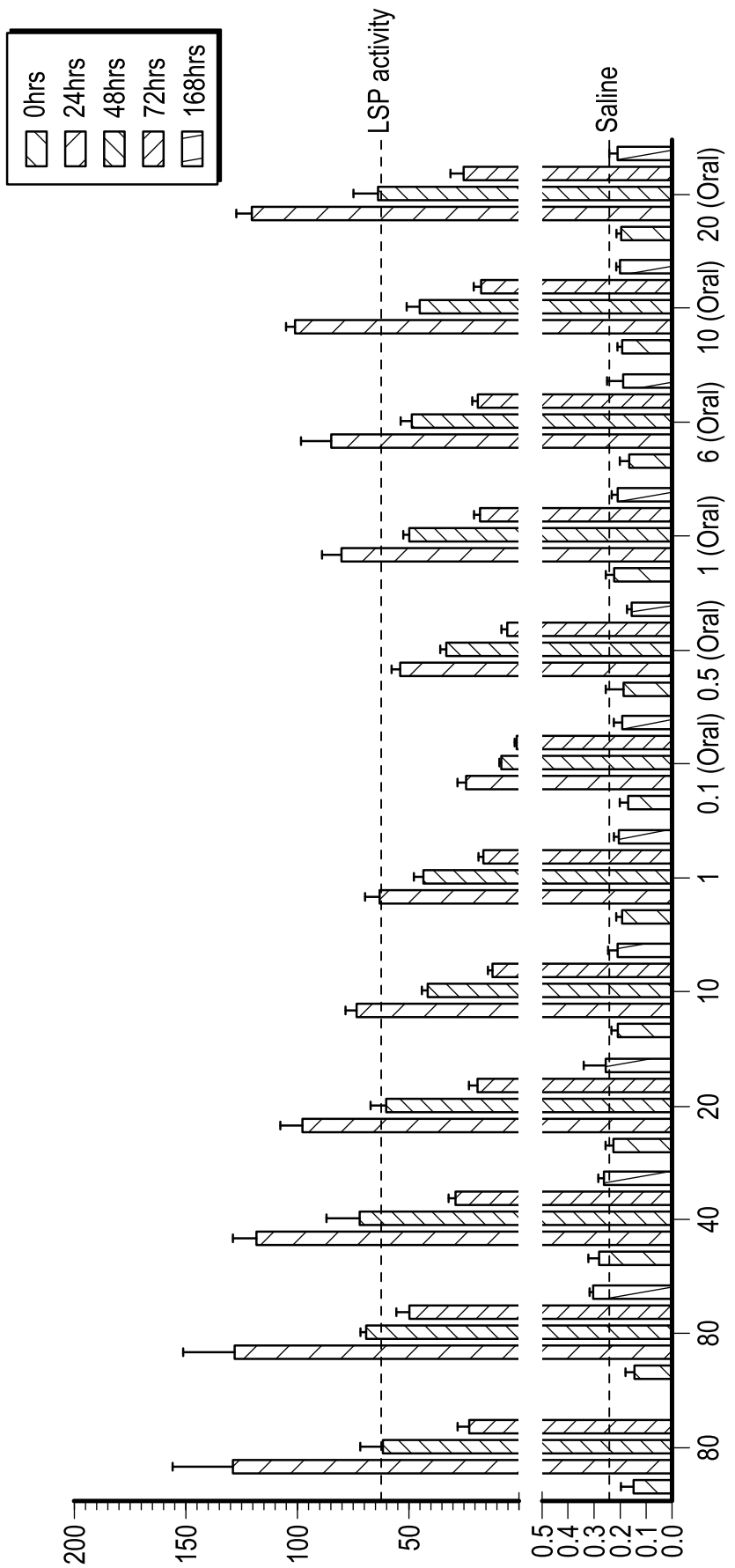

0hrs
24hrs
48hrs
72hrs
168hrs
LSP activity
Saline
Gondatrophin conc [ng/ml]
200
150
100
50
0.5
0.4
0.3
0.2
0.1
0.0
80
40
20
10
1
0.1 (Oral)
0.5 (Oral)
1 (Oral)
6 (Oral)
10 (Oral)
20 (Oral)
mg/kg

FIG. 16

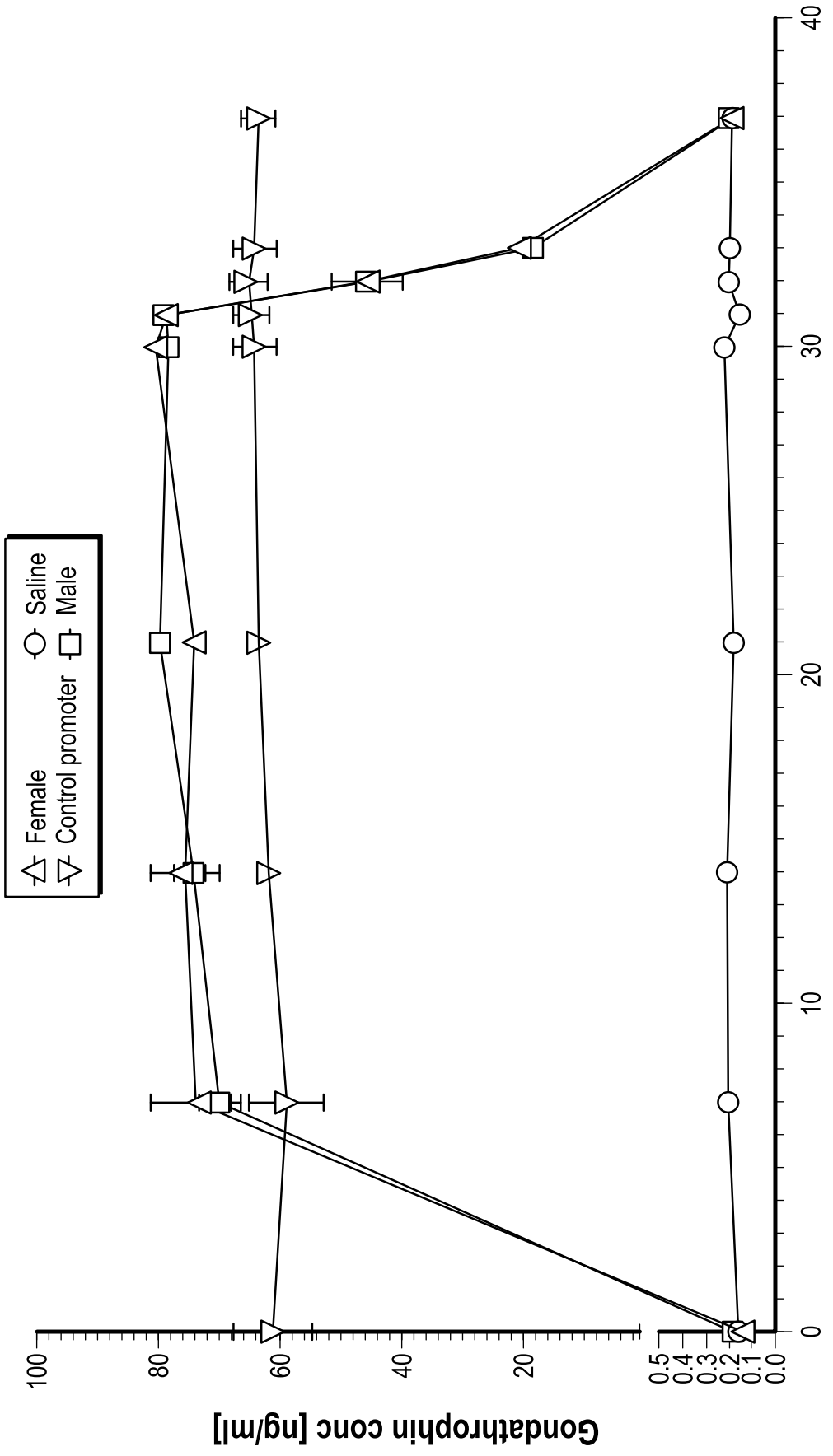

Female
Control promoter
Saline
Male
Gondathrophin conc [ng/ml]
100
80
60
40
20
0.5
0.4
0.3
0.2
0.1
0.0
0
10
20
30
40
Days

FIG. 17