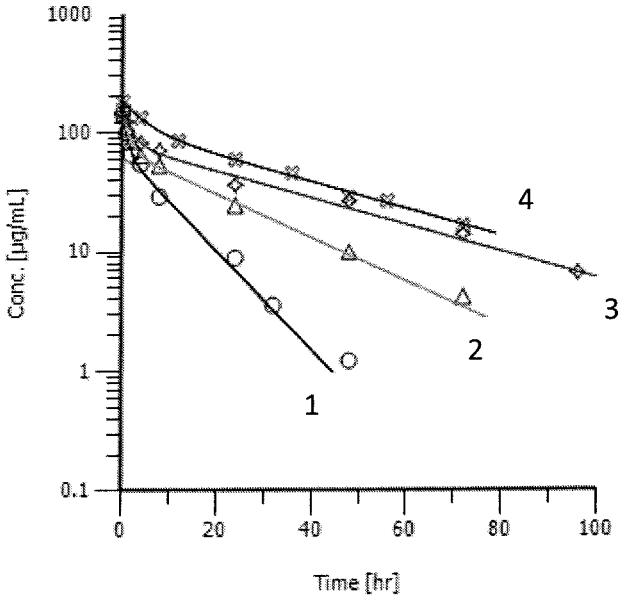
Figure 1

a. Mouse



b. Rat

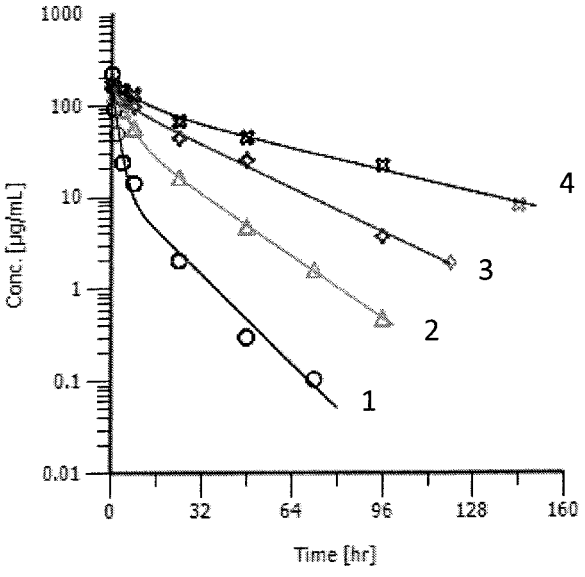
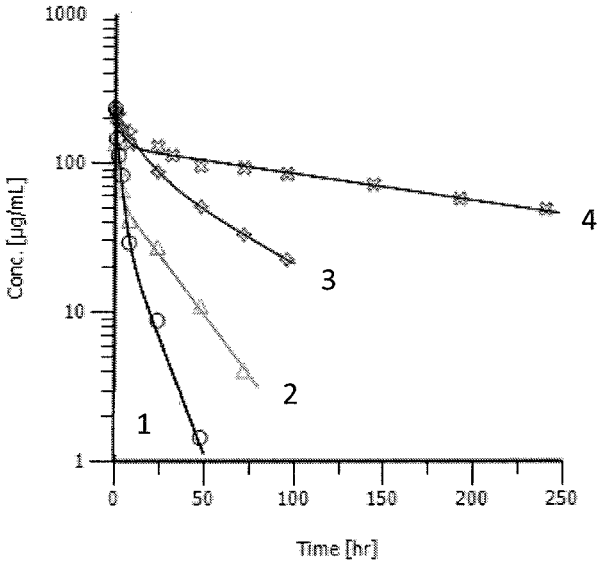


Figure 1 (cont'd)

c. Cyno

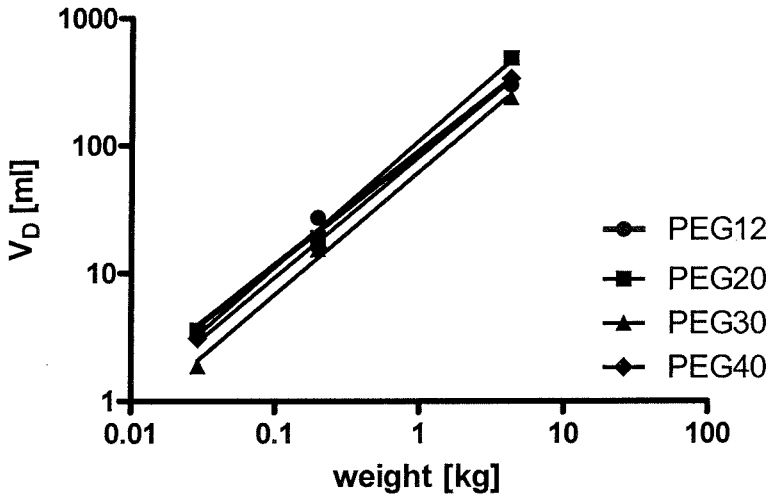


d.

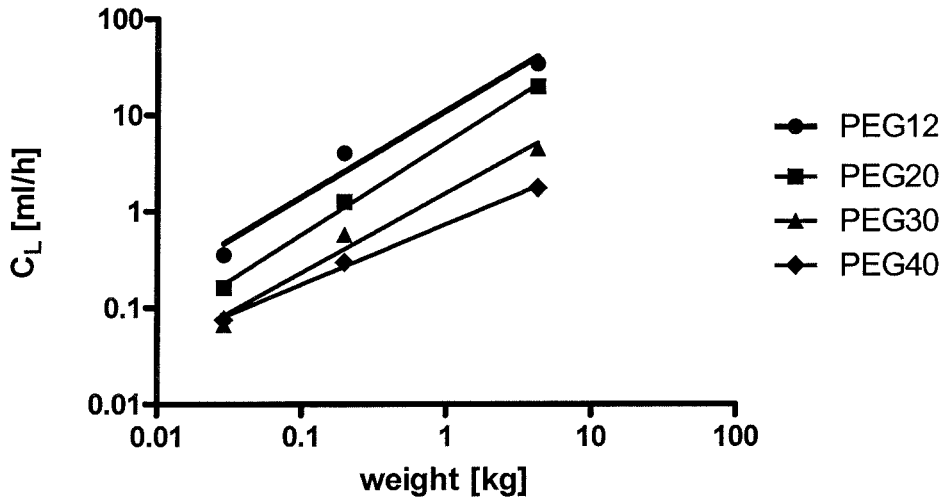
		mouse		rat		cyno	
		V_D [ml]	C_L [ml/h]	V_D [ml]	C_L [ml/h]	V_D [ml]	C_L [ml/h]
1	SEQ ID NO: 1 + PEG12	3.32	0.353	27.33	4.025	300.89	34.09
2	SEQ ID NO: 1 + PEG20	3.58	0.161	19.05	1.253	488.67	19.73
3	SEQ ID NO: 1 + PEG30	1.88	0.067	15.47	0.572	239.0	4.5
4	SEQ ID NO: 1 + PEG40	3.1	0.075	15.85	0.297	337.6	1.75

Figure 2

a. Allometric scaling of V_D



b. Allometric scaling of C_L



c. Goodness of fit (R^2)

	SEQ ID NO: 1 + PEG12	SEQ ID NO: 1 + PEG20	SEQ ID NO: 1 + PEG30	SEQ ID NO: 1 + PEG40
V_D	0.99	0.99	0.99	1.0
C_L	0.98	1.0	0.98	0.99

Figure 2 (cont'd)

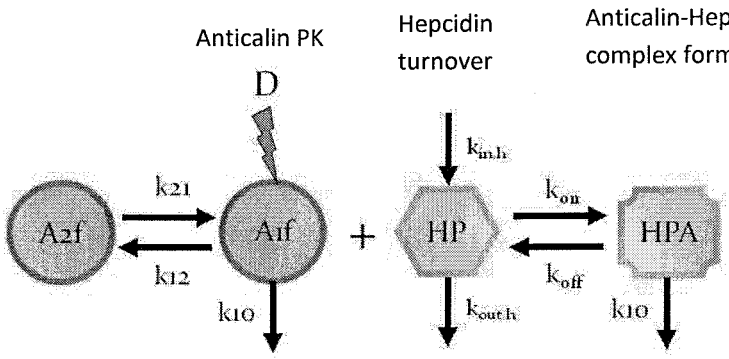
d. **Extrapolated values for human V_D and C_L and calculated values for K_{el} and $t_{1/2}\beta$**

	V_D (ml)	C_L (ml/h)	K_{el} (1/h)	$t_{1/2}\beta$ (h)
SEQ ID NO: 1 + PEG12	5093.3	633.8	0.124	5.6
SEQ ID NO: 1 + PEG20	9572	389.9	0.041	17
SEQ ID NO: 1 + PEG30	3723.9	51.4	0.0138	50
SEQ ID NO: 1 + PEG40	4497.8	10.5	0.0023	298

Figure 3

Antagonist	IC50 [pM]	STDV [pM]	mean of n=X
SEQ ID NO: 2	56	23	6
SEQ ID NO: 1 linked to PEG12	48	14	5
SEQ ID NO: 1 linked to PEG20	47	11	5
SEQ ID NO: 1 linked to PEG30	58	6	6
SEQ ID NO: 1 linked to PEG40	54	10	5

Figure 4



Free anticalin $$\frac{dA1f}{dt} = -k_{10} \cdot A1f - k_{12} \cdot A1f + k_{21} \cdot A2f - k_{on} \cdot HP \cdot \frac{A1f}{Vc} + k_{off} \cdot HPA$$

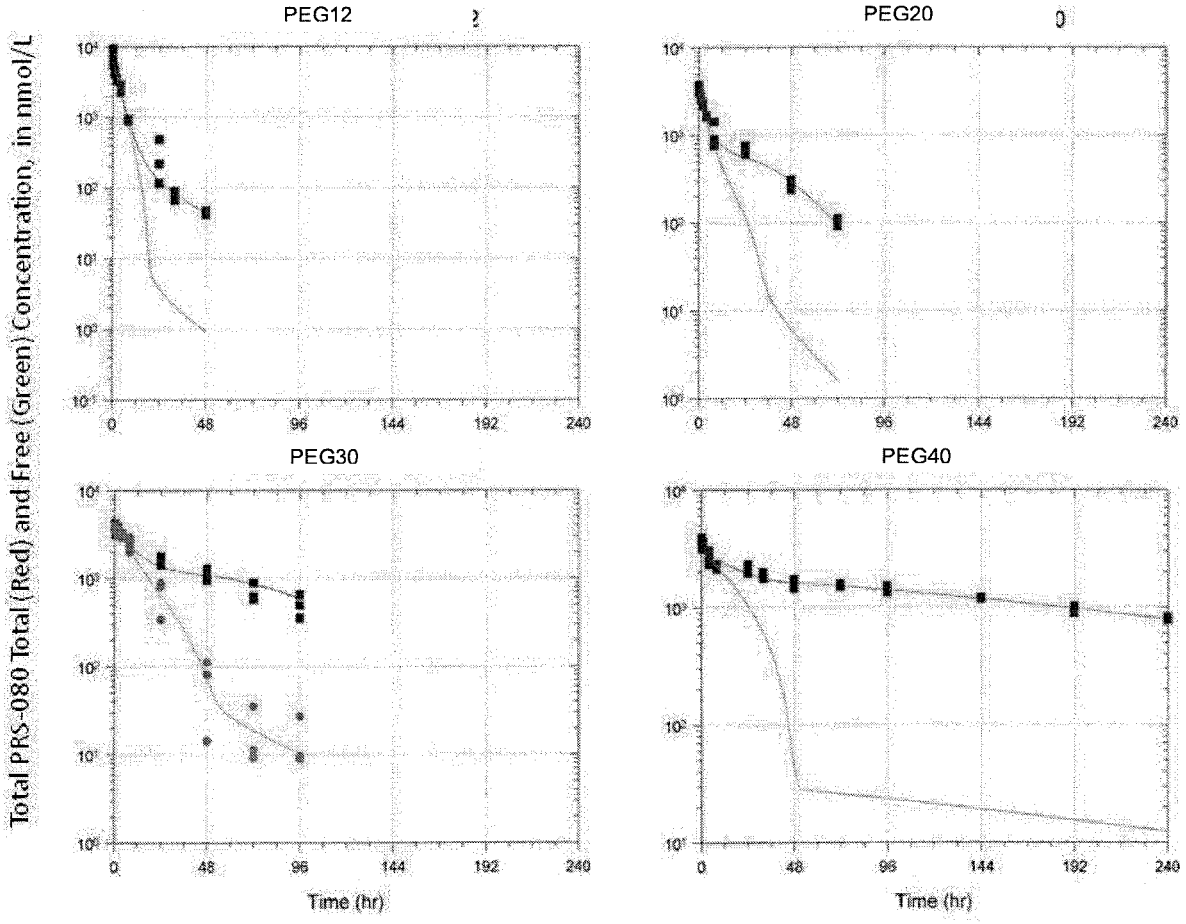
Free hepcidin $$\frac{dHP}{dt} = k_{in,h} - k_{out,h} \cdot HP - k_{on} \cdot HP \cdot \frac{A1f}{Vc} + k_{off} \cdot HPA$$

Complex $$\frac{dHPA}{dt} = k_{on} \cdot HP \cdot \frac{A1f}{Vc} - k_{off} \cdot HPA - k_{10} \cdot HPA$$

Total anticalin (measured) $$C_{tot} = \frac{A1f + HPA}{Vc}$$

Figure 5

a.



b.

Compound		PEG12	PEG20	PEG30	PEG40
Dose	mg/kg	10	10	10	10
MW	Da	32292	40292	50292	60292
Dose for 3.6 kg monkey	nmol	1115	894	716	597
Vc	L	0.184	0.285	0.172	0.191
K_{10}	h^{-1}	0.204	0.0759	0.0277	0.00639
K_{12}	h^{-1}	0.0434	0.115	0.0503	0.0194
K_{21}	h^{-1}	0.0382	0.0660	0.0328	0.00757
K_{ON}	$nM^{-1}\ h^{-1}$	*13.5*	*13.5*	*13.5*	*13.5*
K_{OFF}	h^{-1}	*0.882*	*0.882*	*0.882*	*0.882*
$K_{out,h}$	hr^{-1}	*9.35*	*9.35*	*9.35*	*9.35*

Figure 6

a. PEG40

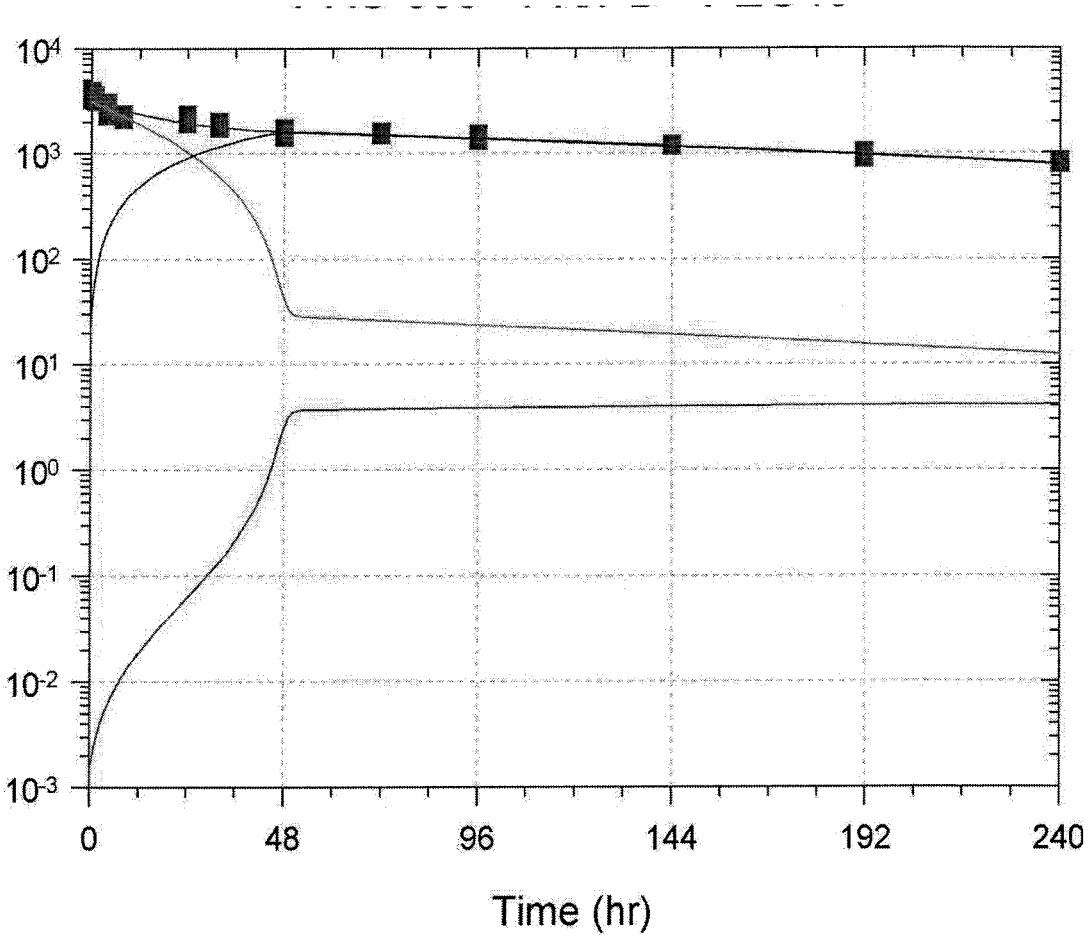


Figure 6 (cont'd)

b. PEG30

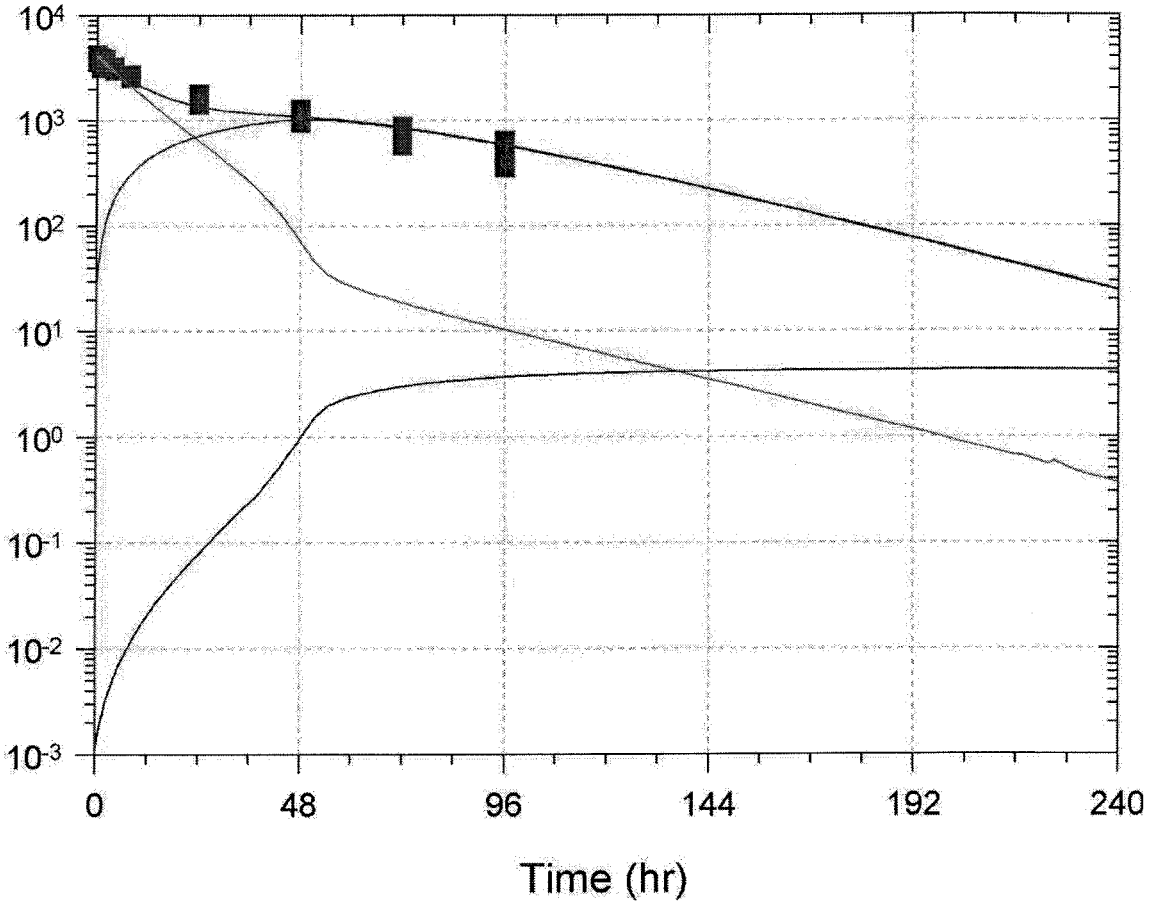


Figure 6 (cont'd)

c. PEG20

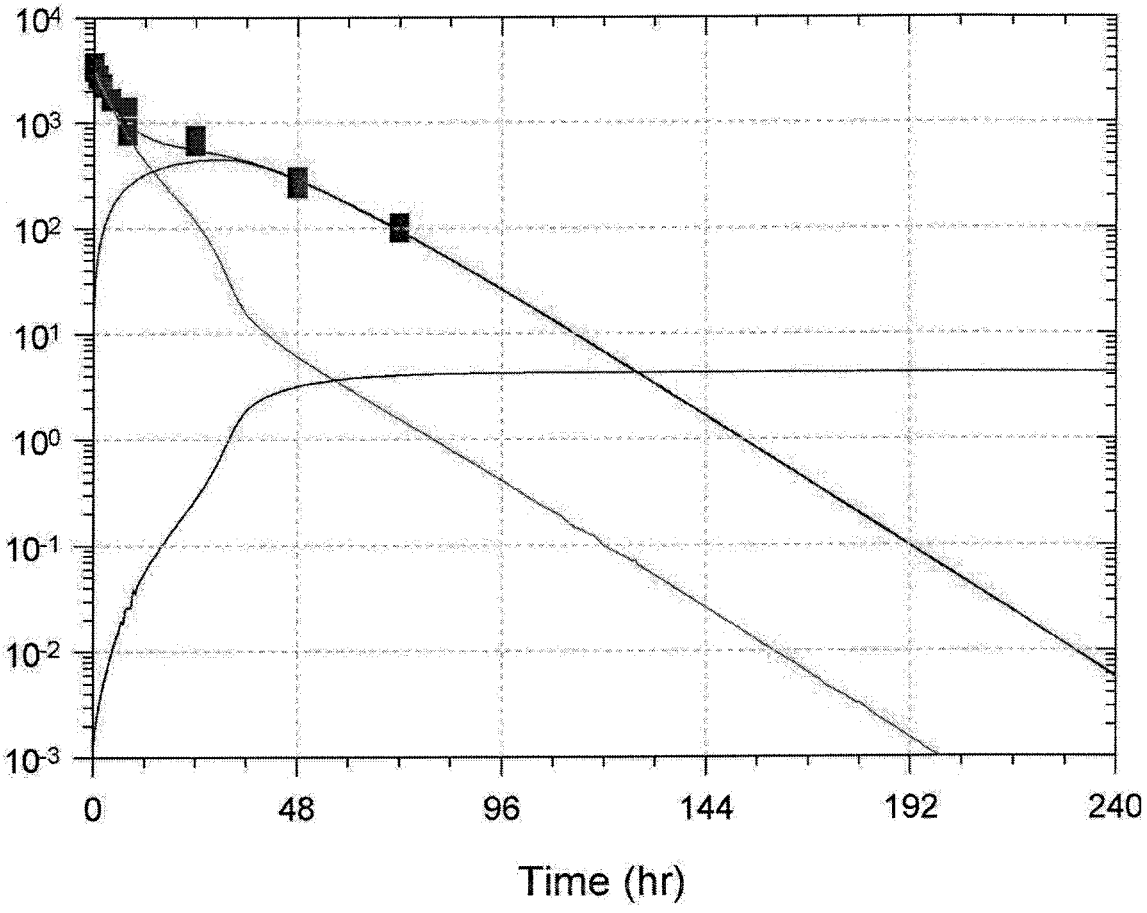


Figure 6 (cont'd)

d. PEG10

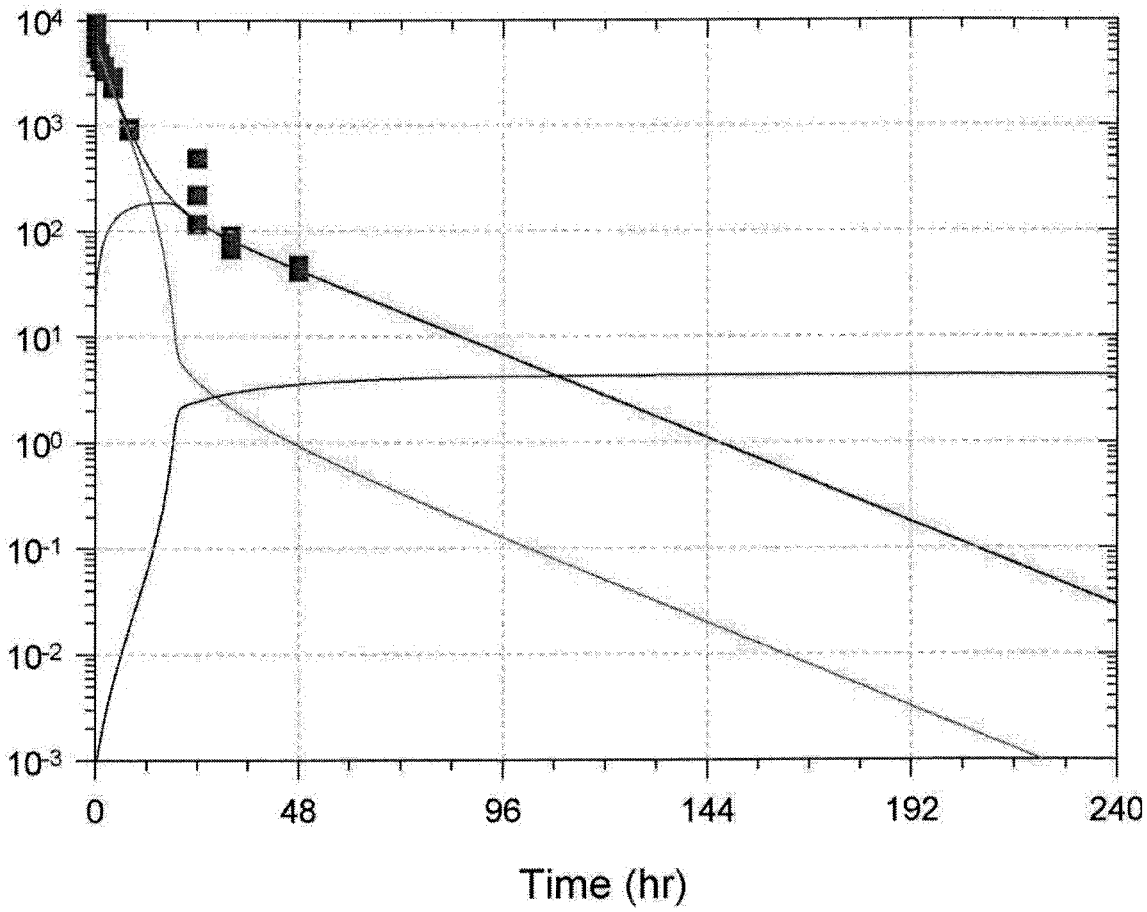
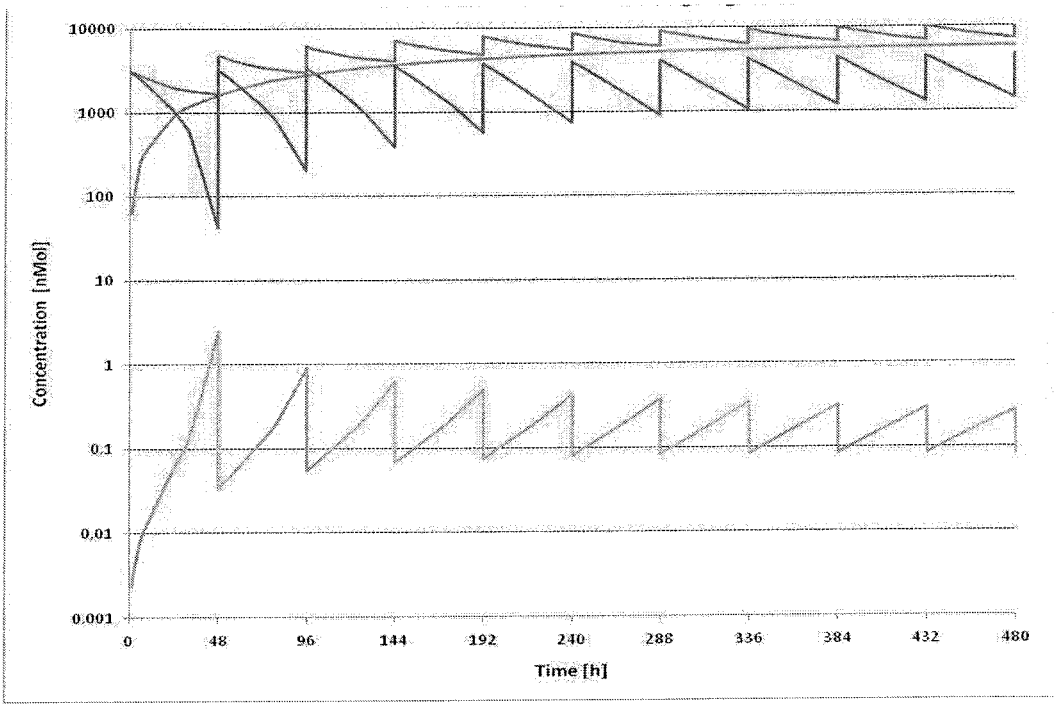


Figure 7

a) PEG40



b) PEG30

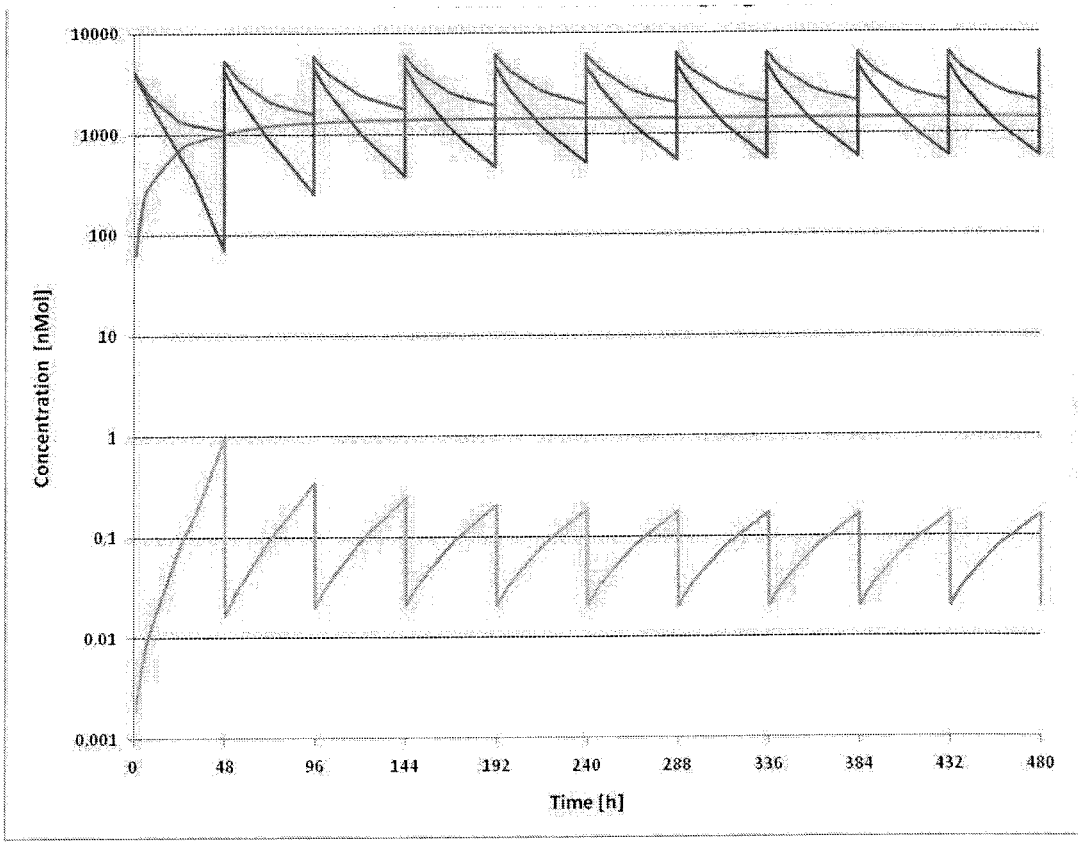


Figure 8

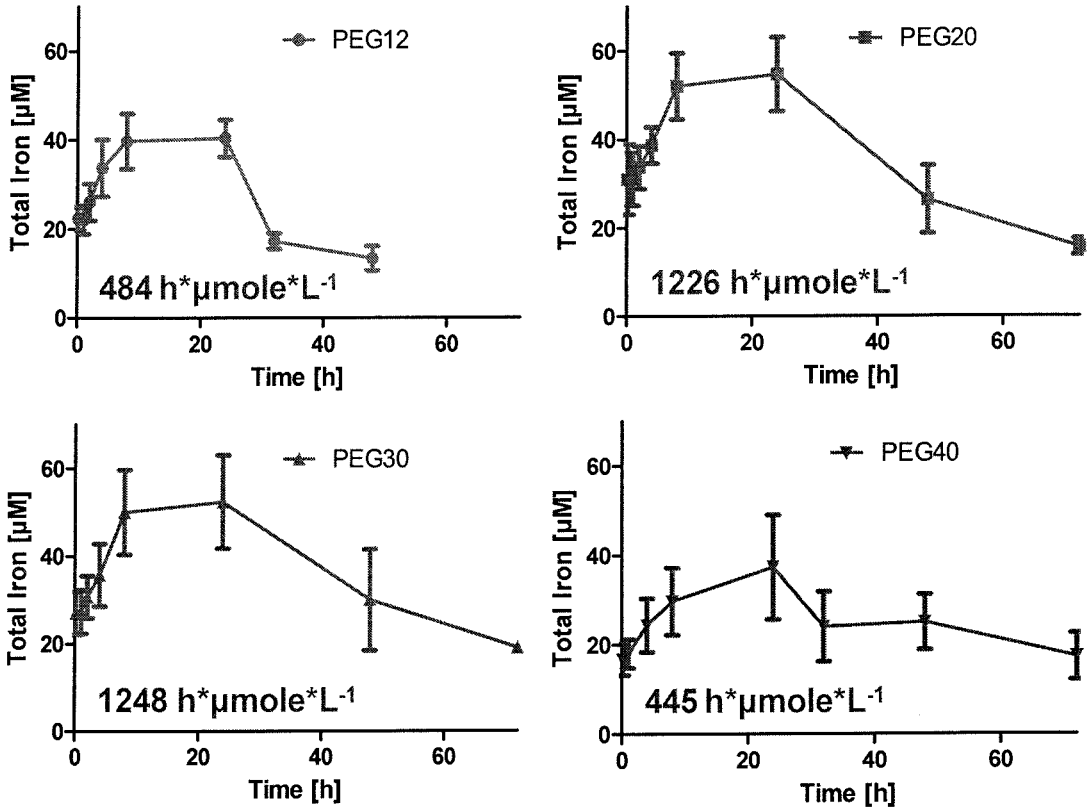

PEG12
Total Iron [µM]
Time [h]
484 h*µmole*L-1
PEG20
1226 h*µmole*L-1
PEG30
1248 h*µmole*L-1
PEG40
445 h*µmole*L-1

Figure 9

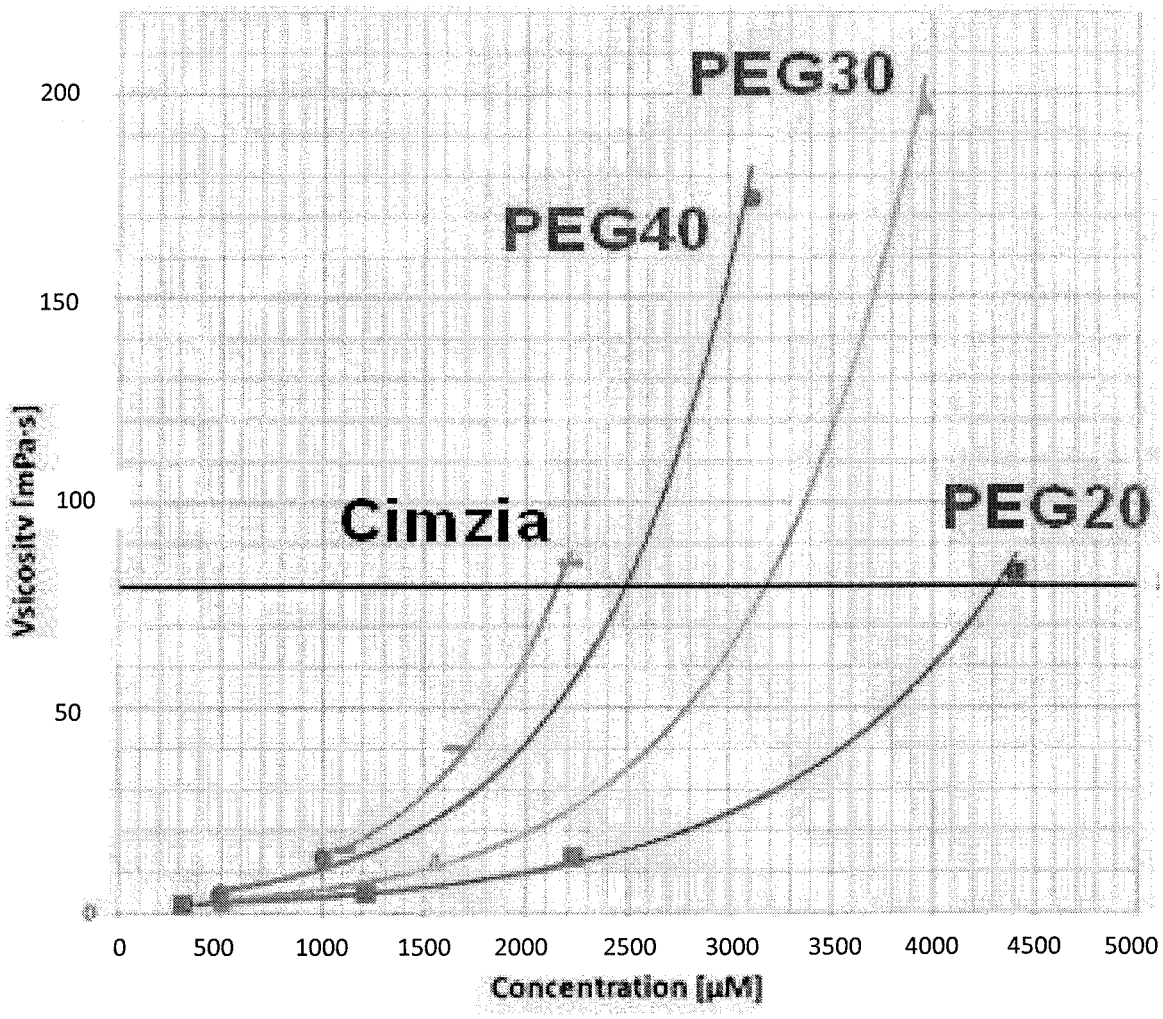
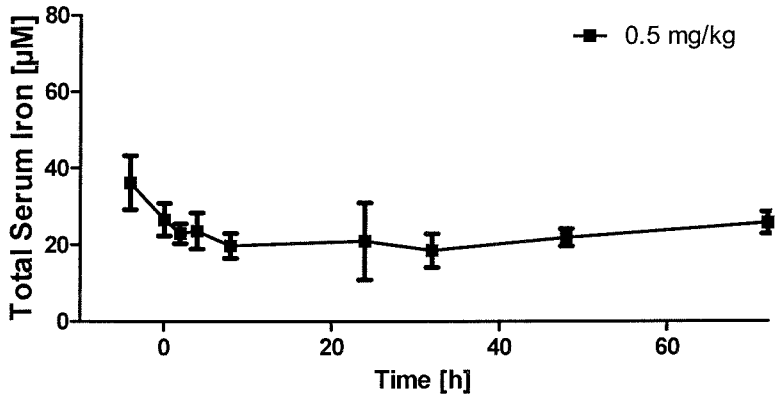

PEG30
PEG40
Cimzia
PEG20
200
150
100
50
0
Vsicositv [mPa·s]
0
500
1000
1500
2000
2500
3000
3500
4000
4500
5000
Concentration [µM]

Figure 10

a.


Total Serum Iron [µM]
80
60
40
20
0
0
20
40
60
Time [h]
0.5 mg/kg

b.

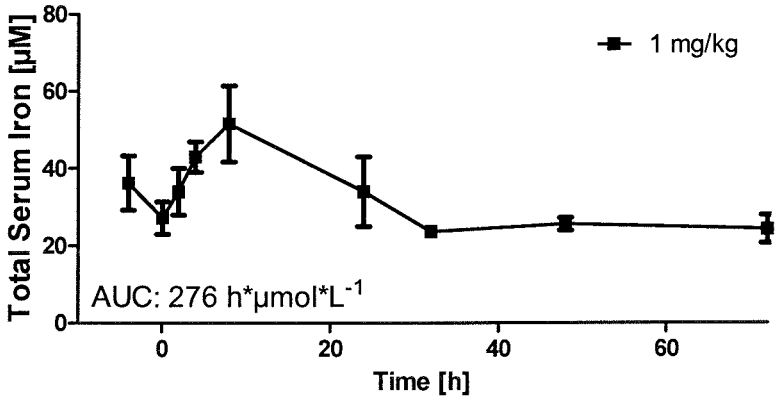
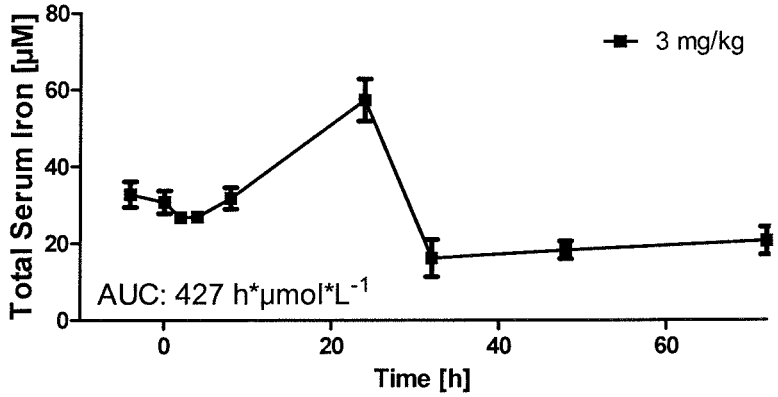

Total Serum Iron [µM]
80
60
40
20
0
0
20
40
60
Time [h]
1 mg/kg
AUC: 276 h*µmol*L-1

Figure 10 (cont'd)

c.



d.

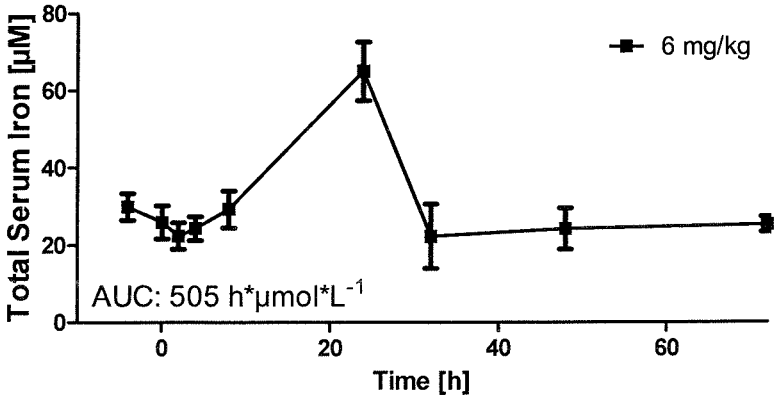
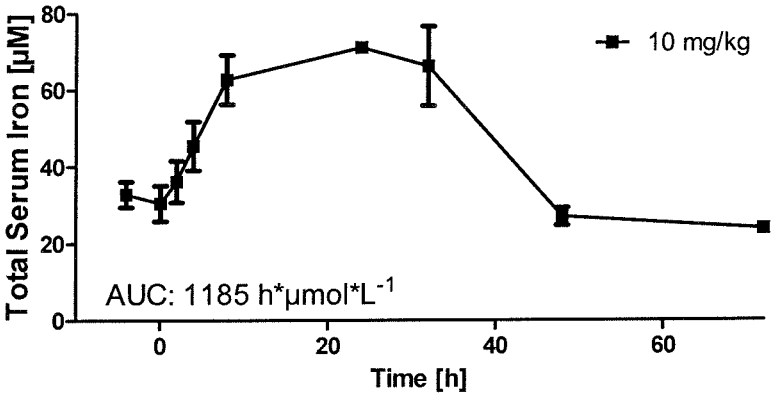


Figure 10 (cont'd)

e.



f.

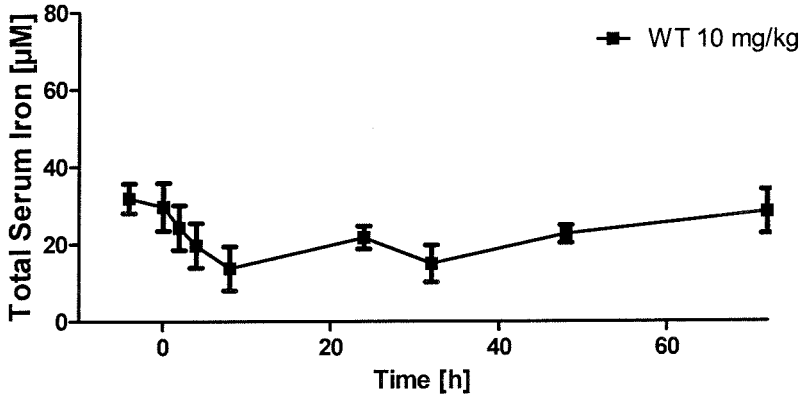
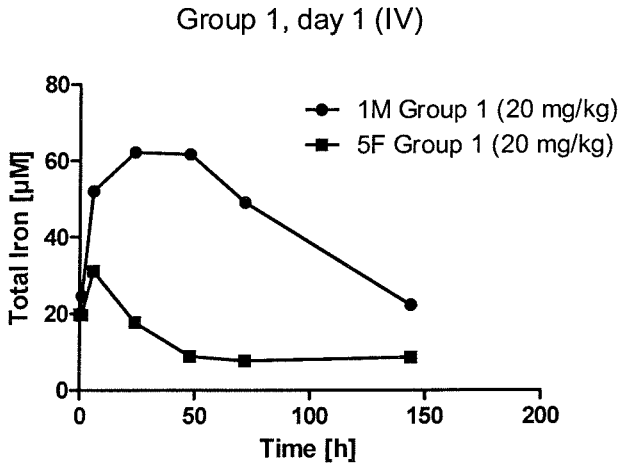


Figure 11

a.


Group 1, day 1 (IV)
1M Group 1 (20 mg/kg)
5F Group 1 (20 mg/kg)
Total Iron [µM]
Time [h]
0
20
40
60
80
0
50
100
150
200

b.

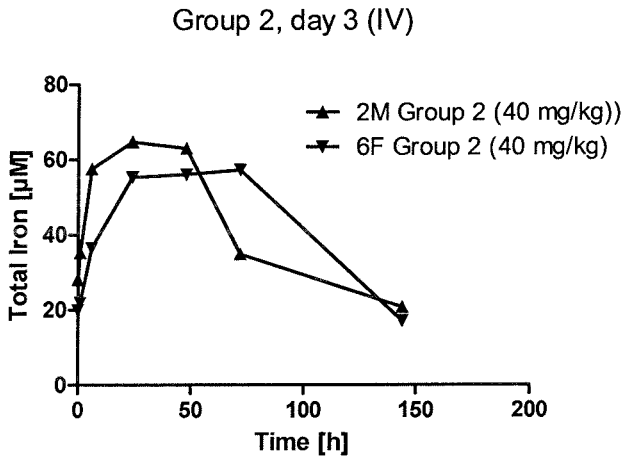
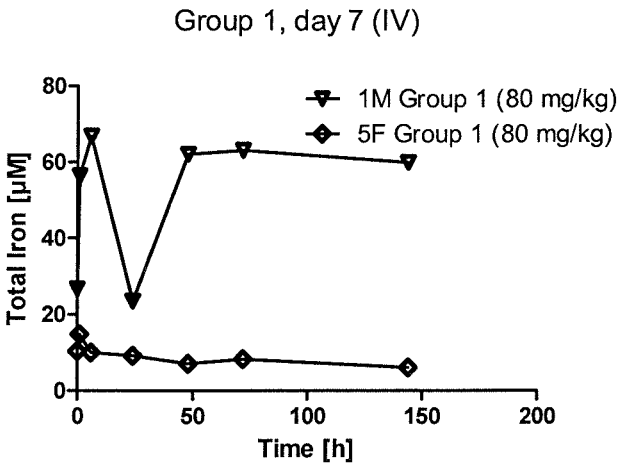

Group 2, day 3 (IV)
2M Group 2 (40 mg/kg))
6F Group 2 (40 mg/kg)
Total Iron [µM]
Time [h]
0
20
40
60
80
0
50
100
150
200

Figure 11 (cont'd)

c.



d.

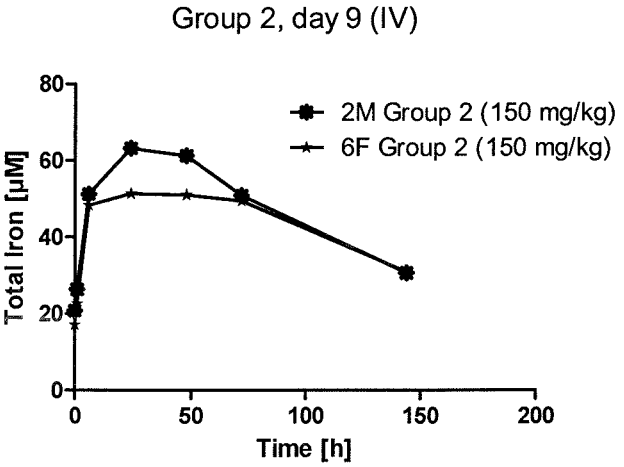


Figure 11 (cont'd)

e.

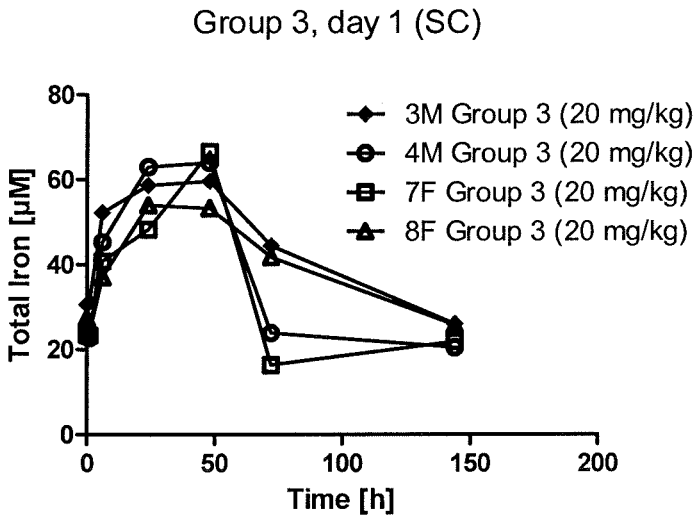


Figure 12

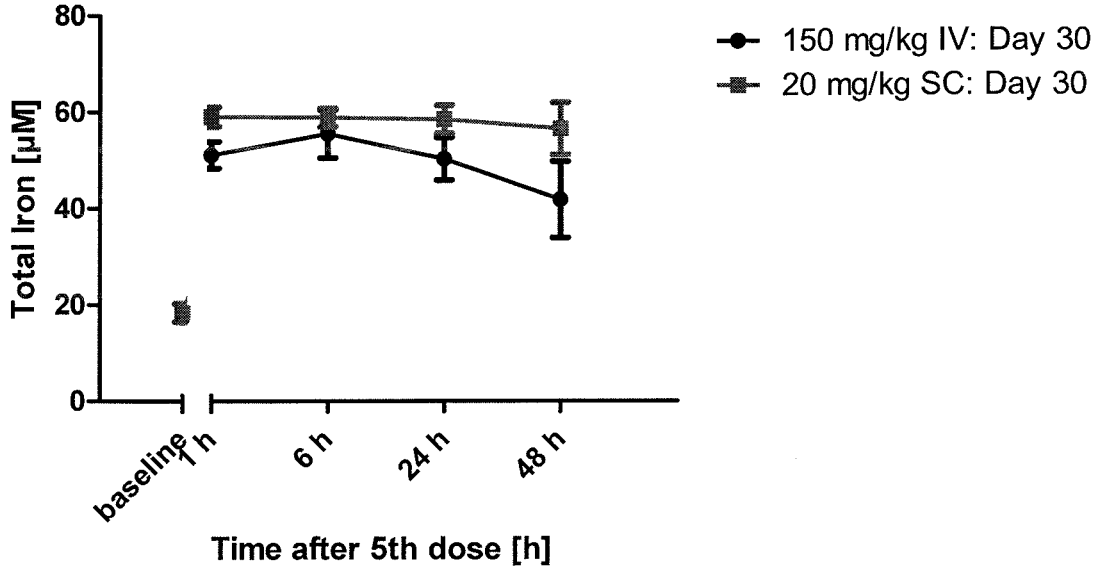


Figure 13

a.

Binder	*Analyte*	*ka [$M^{-1}*s^{-1}$]*	*kd [s-1]*	*Kd [nM]*	*N*
SEQ ID NO: 1	*Hepcidin-25*	*3,8 (± 1,5) • 10^6*	*2,0 (± 0,7) • 10^{-4}*	*0,07 (± 0,05)*	*6*
SEQ ID NO: 1	*Fe-enterobactin*	*No binding*			*8*
SEQ ID NO: 1	*ß-defensin*	*No binding*			*3*
SEQ ID NO: 1	*VEGF8-109*	*No binding*			*3*
SEQ ID NO: 1	*HSA*	*No binding*			*3*
SEQ ID NO: 4	*Hepcidin-25*	*No binding*			*6*
SEQ ID NO: 4	*Fe-enterobactin*	*2,1 (± 0,7) • 10^6*	*8,3 (± 1,1) • 10^{-4}*	*0,42 (± 0,14)*	*3*

b.

Binder	Analyte	Kd [nM]	N
SEQ ID NO: 1	*Hepcidin-25*	*0.10(±0.07)*	*3*
SEQ ID NO: 1	*cynomolgus hepcidin 25*	*0.07(±0.06)*	*3*