Behar et al Supplemental Materials

Supplemental Material

Supplemental Tables

Supplemental Figures

Feedback hub		TF2	
k1	0.125	k5a	0.014
k2a	0.0066	k5b	0.0006
k2b	0.75	TF1	
k3	0.07	k6	0.5
k4	0.025	k7a	0.01
km1	0.075	k7b	20
km2b	0.015	k8	0.1
km3	0.0025	k9	0.001
km4	0.0025	km8	0.05
Perturbation A		km9	0.05
k1	0.01		
Perturbation FBR			
k4	0.00004		

Supplemental Table S1: Rate constants used to simulate the simple network model (See Figure 1). The names of the rate constants correspond to equations 1-5 in Methods.

M1		
k1	0.085	R2
k2	0.085	R2
km1	0.1	N
km2	0.1	N

M2		
k1	0.1	R2
k2a	0.05	R2
k3	0.04	R2
km1	0.1	N
km2	0.1	N
km3	0.1	N

M3 - M3'			
k1	0.075	0.125	R5
k2a	0.025	0.01	R5
k2b	0.075	0.25	R7
k3	0.025	0.02	R5
k4	0.01	0.01	R4
km1	0.1	0.1	N
km2	0.1	0.1	N
km3	0.1	0.025	N
km4	0.1	0.025	N

M4 - M4'			
k1	0.075	0.25	R3
k2a	0.025	0.05	R5
k2b	0.075	1.0	R4
k3	0.025	0.04	R3
k4	0.01	0.04	R2
km1	0.1	0.05	N
km2	0.1	0.05	N
km3	0.1	0.05	N
km4	0.1	0.05	N

M5		
k1	0.075	R6
k2a	0.025	R6
k2i	5.0	R8
k3	0.025	R8
k4	0.01	R5
km1	0.1	N
km2	0.1	N
km3	0.1	N
km4	0.1	N

M6		
k1	0.075	R5
k2a	0.025	R5
k2i	2.5	R5
k3	0.025	R5
k4	0.01	R2
km1	0.1	N
km2	0.1	N
km3	0.1	N
km4	0.1	N

M7		
k1	0.075	R6
k2a	0.01	R6
k2i	0.25	R6
k3	0.25	R6
k4	0.25	R6
km1	0.1	N
km2	0.1	N
km3	0.1	N
km4	0.1	N
km2i	0.1	N

Supplemental Table S2: Rate constants for modules M1-7 used in the virtual screen (relate to Figure 2). The parameter names correspond to equations 10-25 in Methods. Multiplier range used to simulate perturbations are indicated next to each parameter ($R_n = 2^{-n}$ to 2^n in n/20 increments). Parameters indicated as N were not varied. Models M3' and M4' were used to generate Figure 4.

S	Function	sl	sb	sh	tr	td	tp1	tc	j	h
1	F1	0	0.005	1	0.5	0	10	0.5	NA	NA
2	F1	0	0.005	1	0.5	0	60	0.5	NA	NA
3	F1	0	0.005	1	0.5	0	180	5	NA	NA
4	F1	0	0.005	1	2.5	0	15	2.5	NA	NA
5	F1	0	0.005	1	10	0	15	300	NA	NA
6	F1	0	0.005	1	0.5	0	300	0.5	NA	NA
7	F2	0	0.005	NA	NA	0	NA	NA	0.01	9
8	F1	0	0.005	1	300	0	15	0.5	NA	NA
9	F1	0	0.005	1	180	0	15	0.5	NA	NA
10	F1	0	0.005	1	125	0	15	125	NA	NA

Supplemental Table S3: Parameters used to define the input functions S1-10 in the virtual screen. Parameter names correspond to equations 6-7 in Methods (F1 and F2, respectively). Parameters marked as NA do not apply to that particular function.

		Response		
		Out-of-Equilibrium	**Quasi-equilibrium**	**Steady-State**
Perturbations that affect	**Time-scale**	**Affected** if the change in the balance of internal and stimulus-related time scales is substantial.	**Typically affected** with simple perturbations because of their collateral effects on dose responses. Change in the kinetics may delay them but otherwise leave the amplitude unaffected.	**Not affected**
	Dose-response	**Not affected** unless the areas of the dose response curves that bound the response are changed significantly.	**Typically affected**.	**Typically affected** unless the change in the shape of the curve does not alter the equilibrium level of the signaling species.

Supplemental Table S4: Principles governing dynamic effecrs of simple perturbations (relate to Figure 3). How a simple perturbation affects out-of-equilibrium, quasi-equilibrium dynamics, or steady state depends on whether the primarily effect is on time-scales or dose-response surfaces. Complex perturbations (involving multimple reacions) allow for more selective control

See attached spreadsheet file.

Supplemental Table S5. Reactions and Rate Constants for the NFκB model (relate to Figure 5 and 6). The 73 reactions that make up the system are listed, with addition details. Two RNA synthesis reactions (69,70) also have a hill coefficient of 3, to represent transcriptional nonlinearity (cooperative binding). The parameter ID number correspond to the diagram in Figure S5 and Table S7.

Min	TNFc	TNFp	LPS	LPS-TNF
0	1	1	1	1
5	59.38	59.38	1.5	3
10	100	100	2	4
15	64.57	64.57	2.5	5
20	50.15	50.15	3	5
25	35.73	35.73	3.5	5
30	30	21.3	4	5
35	30	19.66	4.5	5
40	30	18.03	5	5
45	30	16.39	5.5	5
50	30	14.16	6	4.33
55	30	11.93	6.5	3.67
60	30	9.7	7	3.00
65	30	8.25	7.5	2.67
70	30	6.8	8	2.33
75	30	5.35	8.5	2.00
80	30	3.9	9	1.67
85	30	2.45	9.5	1.33
90	30	1	10	1
95	30	1	10.5	1
100	30	1	11	1
105	30	1	11.5	1
110	30	1	12	1
115	30	1	12.5	1
120	30	1	13	1
125	30	1	14	1
130	30	1	15	1
135	30	1	16	1
140	30	1	17	1
145	30	1	18	1
150	30	1	19	1
155	30	1	20	1
160	30	1	21	1
165	30	1	22	1
170	30	1	23	1
175	30	1	24	1
180	30	1	25	1

Supplemental Table S6: IKK Profiles for the NFκB simulations (relate to Figures 5 and 6). The figures shown are percentages, and represent the percent of total IKK which is phosphorylated and thus active. LPS-TNF was derived from measurements of IKK activity in TNF ko cells (Werner et al., 2005) and used for simulations in Figure 6.

Parameter #	Description	Grouping	
		biochem.	pharm.
8,10,12,13,14,15	Association IkB-NFkB	b1	
16,23,28	Dissociation IkBα-NFkB	b6	
18,25,29	Dissociation IkBβ-NFkB	b7	
20,27,30	Dissociation IkBε-NFkB	b8	
7,9,11	Association IKK-IkB	b26	
17,22	Dissociation IKK-IkBα	b29	
19,24	Dissociation IKK-IkBβ	b30	
21,26	Dissociation IKK-IkBε	b31	
1,3,5	Association IKKIkB+NFkB	b27	
2,4,6	Association IkBNFkB+IKK	b28	
37	Export NFkB	b14	
44	Import NFkB	b13	
31,32,33	Export IkB-NFkB	b11	
38,39,40	Import IkB-NFkB	b9	
34,35,36	Export IkB	b12	
41,42,43	Import IkB	b10	
54,55,56	Degradation IkB	b4	MG132
46,48,50	Degradation IkB-NFkB	b2	MG132
45,47,49	Degradation IKK-IkB	b5	MG132
51,52,53	Degrad. IKK-NFkB-IkB	b3	MG132
63	IkB Protein synthesis	b15	CHX
64		b16	CHX
65		b17	CHX
66	IkB RNA degradation	b23	
67		b24	
68		b25	
71	IkB RNA synthesis	b20	TSA
72		b21	TSA
73		b22	TSA
69	NF-kB responsive RNA synthesis	b18	PDTC
70		b19	PDTC

Supplemental Table S7: NFκB model: Biochemical & Pharmacological Grouping (relate to Figures 5 and 6). Related biochemical parameters identified as biochemical groups b1-b31. Selected pharmacologic agents affect the indicated parameters, defining the pharmacologic groupings. The parameter number corresponds to the labels in Figure S5.