Table I. Kinetic description of MAPK cascade reactions

The maximal rates (V_i^{max}), Michaelis (K_{mi}) and catalytic (k_i^{cat}) constants are expressed in nM·s^{-1}, nM and s^{-1}, respectively. The total protein concentrations (the sums of differently phosphorylated forms) are assumed to be constant on the time scale considered, Raf_{total} = 300 nM; MEK_{total} = 300 nM; ERK_{total} = 300 nM. Input signal starts at $t = 0$ when [*RasGTP*] changes from 0 to 10 nM, and resulting responses of *ppERK* are shown in Fig. 1 of the main text. The parameters F and K_f describe feedback regulation by *ppERK*; $F < 1$ for negative feedback, $F > 1$ for positive feedback and $F = 1$, if this feedback is absent.

N	Reaction	Rate	Kinetic Constants
1	Raf → pRaf	$v_1 = \frac{k_1^{cat}[RasGTP][Raf]/K_{m1}}{1+[Raf]/K_{m1}+[pRaf]/K_{m2}} \cdot \frac{1+F[ppERK]/K_f}{1+[ppERK]/K_f}$	$k_1^{cat} = 1$ K_{m1}=100, K_{m2} = 200
2	pRaf → ppRaf	$v_2 = \frac{k_2^{cat}[RasGTP][pRaf]/K_{m2}}{1+[Raf]/K_{m1}+[pRaf]/K_{m2}} \cdot \frac{1+F[ppERK]/K_f}{1+[ppERK]/K_f}$	k_2^{cat}=0.25
3	ppRaf → pRaf	$v_3 = \frac{V_3^{max}[ppRaf]/K_{m3}}{1+[ppRaf]/K_{m3}+[pRaf]/K_{m4}}$	V_3^{max}=2.5, K_{m3}=50, K_{m4}=100
4	pRaf → Raf	$v_4 = \frac{V_4^{max}[pRaf]/K_{m4}}{1+[ppRaf]/K_{m3}+[pRaf]/K_{m4}}$	V_4^{max}=3.75
5	MEK → pMEK	$v_5 = \frac{k_5^{cat}[ppRaf][MEK]/K_{m5}}{1+[MEK]/K_{m5}+[pMEK]/K_{m6}}$	k_5^{cat}=2.5, K_{m5}=250, K_{m6}=250
6	pMEK → ppMEK	$v_6 = \frac{k_6^{cat}[ppRaf][pMEK]/K_{m6}}{1+[MEK]/K_{m5}+[pMEK]/K_{m6}}$	k_6^{cat}=0.5
7	ppMEK → pMEK	$v_7 = \frac{V_7^{max}[ppMEK]/K_{m7}}{1+[ppMEK]/K_{m7}+[pMEK]/K_{m8}}$	V_7^{max}=3, K_{m7}=250, K_{m8}=80
8	pMEK → MEK	$v_8 = \frac{V_8^{max}[pMEK]/K_{m8}}{1+[ppMEK]/K_{m7}+[pMEK]/K_{m8}}$	V_8^{max}=3.75
9	ERK → pERK	$v_9 = \frac{k_9^{cat}[ppMEK][ERK]/K_{m9}}{1+[ERK]/K_{m9}+[pERK]/K_{m10}}$	k_9^{cat}=0.125, K_{m9}=250, K_{m10}=250
10	pERK → ppERK	$v_{10} = \frac{k_{10}^{cat}[ppMEK][pERK]/K_{m10}}{1+[ERK]/K_{m9}+[pERK]/K_{m10}}$	k_{10}^{cat}=0.125
11	ppERK → pERK	$v_{11} = \frac{V_{11}^{max}[ppERK]/K_{m11}}{1+[ppERK]/K_{m11}+[pERK]/K_{m12}+[ERK]/K_{m13}}$	V_{11}^{max}=3.75, K_{m11}=120, K_{m12}=20
12	pERK → ERK	$v_{12} = \frac{V_{12}^{max}[pERK]/K_{m12}}{1+[ppERK]/K_{m11}+[pERK]/K_{m12}+[ERK]/K_{m13}}$	V_{12}^{max}=5, K_{m13}=300

Differential equation system that describes the MAPK dynamics

$$\frac{d[Raf]}{dt} = v_4 - v_1$$
$$\frac{d[pRaf]}{dt} = v_1 - v_2 + v_3 - v_4$$
$$\frac{d[ppRaf]}{dt} = v_2 - v_3$$
$$\frac{d[MEK]}{dt} = v_8 - v_5$$
$$\frac{d[pMEK]}{dt} = v_5 - v_6 + v_7 - v_8$$
$$\frac{d[ppMEK]}{dt} = v_6 - v_7$$
$$\frac{d[ERK]}{dt} = v_{12} - v_9$$
$$\frac{d[pERK]}{dt} = v_9 - v_{10} + v_{11} - v_{12}$$
$$\frac{d[ppERK]}{dt} = v_{10} - v_{11}$$

The initial conditions at t=0 are the following. Figs. 1A-1E: $[Raf] = Raf_{total}$ = 300 nM; $[MEK] = MEK_{total}$ = 300 nM; $[ERK] = ERK_{total}$ = 300 nM; all phosphorylated forms are assumed to equal zero. Fig.1F. Blue curves (below threshold) $[ppERK](t=0)$ = 50, 100, 105 nM; Red curves (above threshold) $[ppERK](t=0)$ = 111, 120, 150 nM. The threshold value is about 108 nM. The values of $[ERK](t=0)$ are calculated as [*ERK*] = 300 - $[ppERK](t=0)$ (nM); $[pERK](t=0)$ = 0.