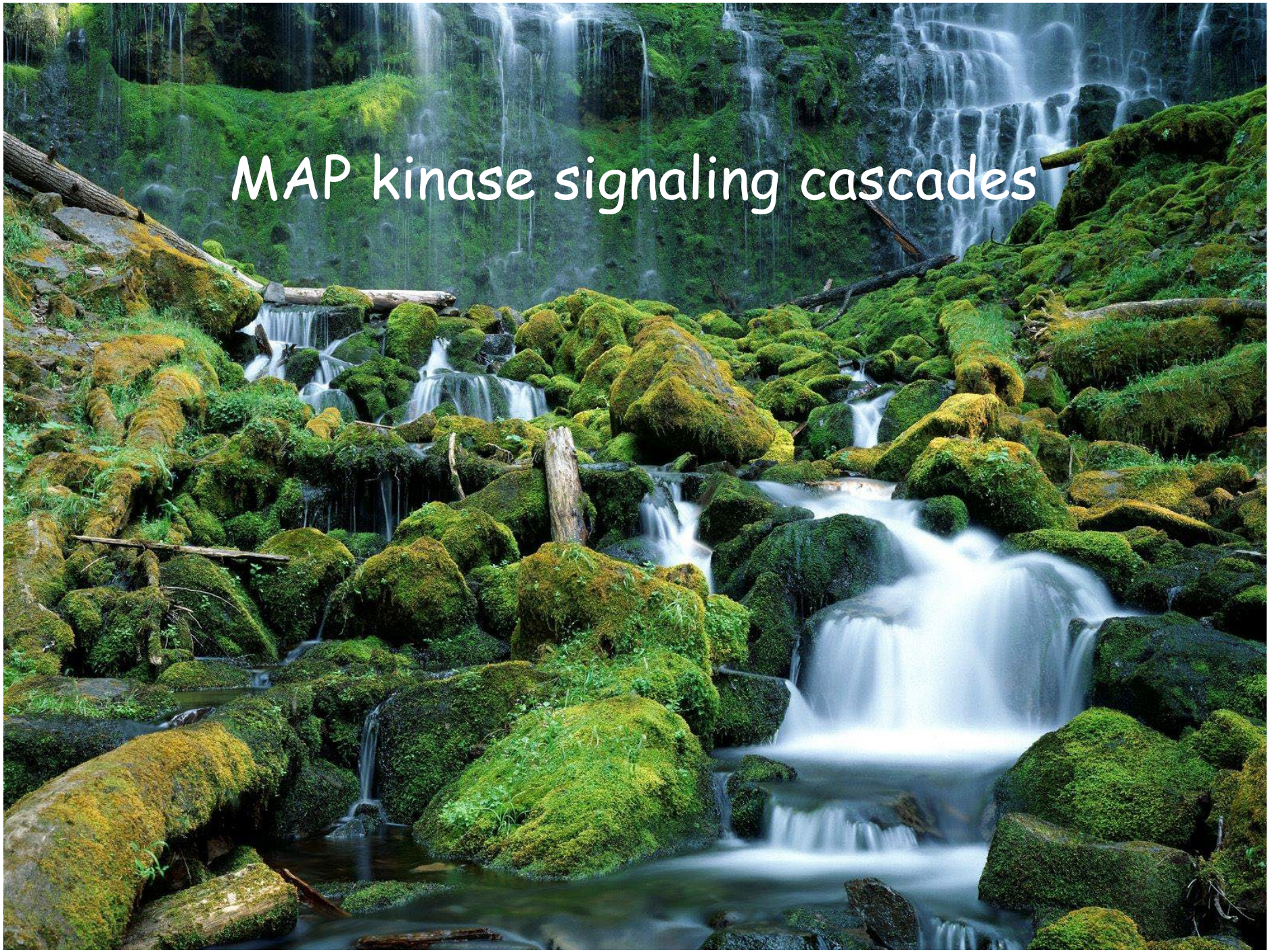
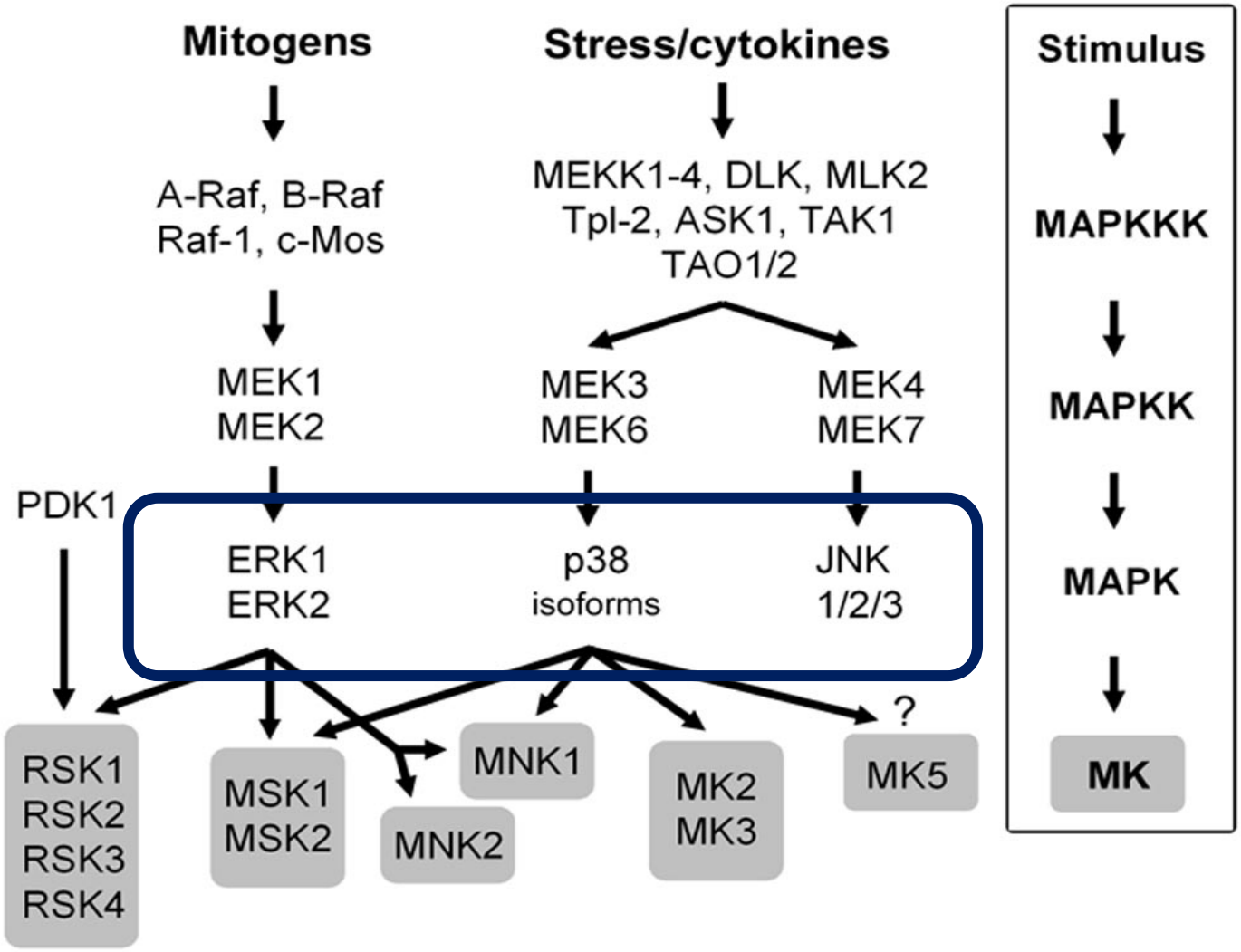

MAP kinase signaling cascades

MAP kinase (Mitogen activated kinase) signaling cascades



MAP kinase signaling cascades

Transduce a large variety of external signals, leading to a wide range of cellular responses, including growth, differentiation, inflammation and apoptosis. These pathways are characterized by the following general path:

Stimulus > MAPKKK > MAPKK > MAPK > Response

Where MAPKK is the kinase of MAPK and MAPKKK is the kinase of MAPKK. In most cases, the MAPKKK is activated by small G proteins such as Ras, Rac and Rap1.

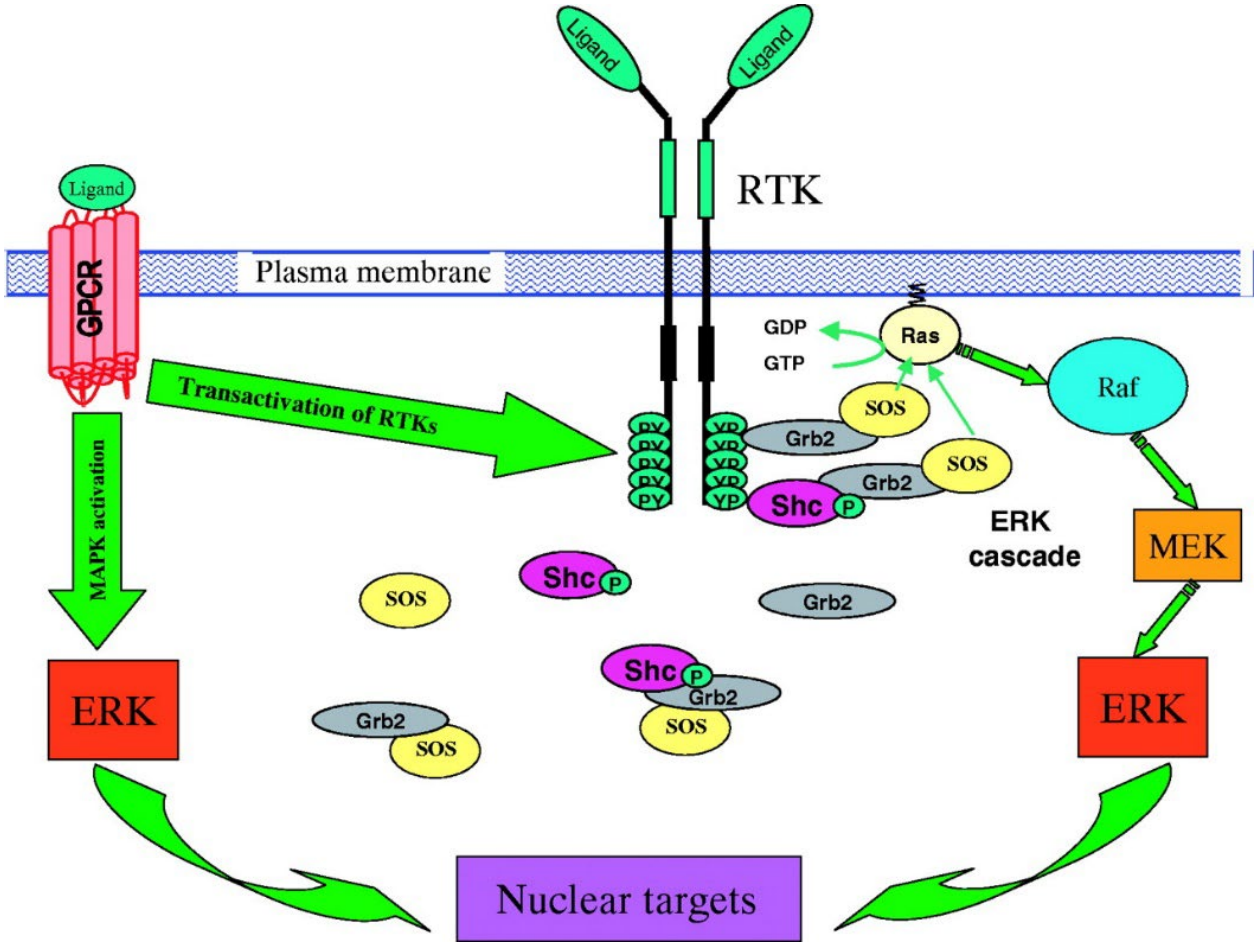
However, it may also be activated by other enzymes.

Table 1 Phenotypes of MAPKK and MAPK knockout mice

MAPKK/MAPK	Phenotypes	Similar to
MEK1	Defective placental vascularization[37]	ERK2?
MKK4	Defective liver development[40]	c-Jun knockout[41]
MKK7	Embryonic lethality of unknown cause[48]	
MKK3	Defective IL-12 production[52]	
ERK1	Defective T-cell development (positive selection)[35]	MEK1 dn negative transgenics
JNK1	Defective T-cell differentiation to Th2 cells[46]	
JNK2	Defective T-cell differentiation to Th1 cells[47]	
JNK1 or JNK2	Defective T-cell proliferation and IL-2 production[25]	JNK1 dn negative transgenics MKK4 knockouts
JNK1 or JNK2	Defective activation induced death of thymocytes[25]	JNK1 dn negative transgenics
JNK1 & JNK2	IL-2 overproduction[48]	MKK7 knockout
JNK1 & JNK2	Neural tube disclosure[38,39]	
JNK3	Resistance to excitotoxic neuronal cell death[28]	c-Jun$^{\Delta 63/73}$ knockin[29]
p38α	Placental defect[51] (trophoblast cells)	
p38α	Insufficient production of erythropoietein[50]	

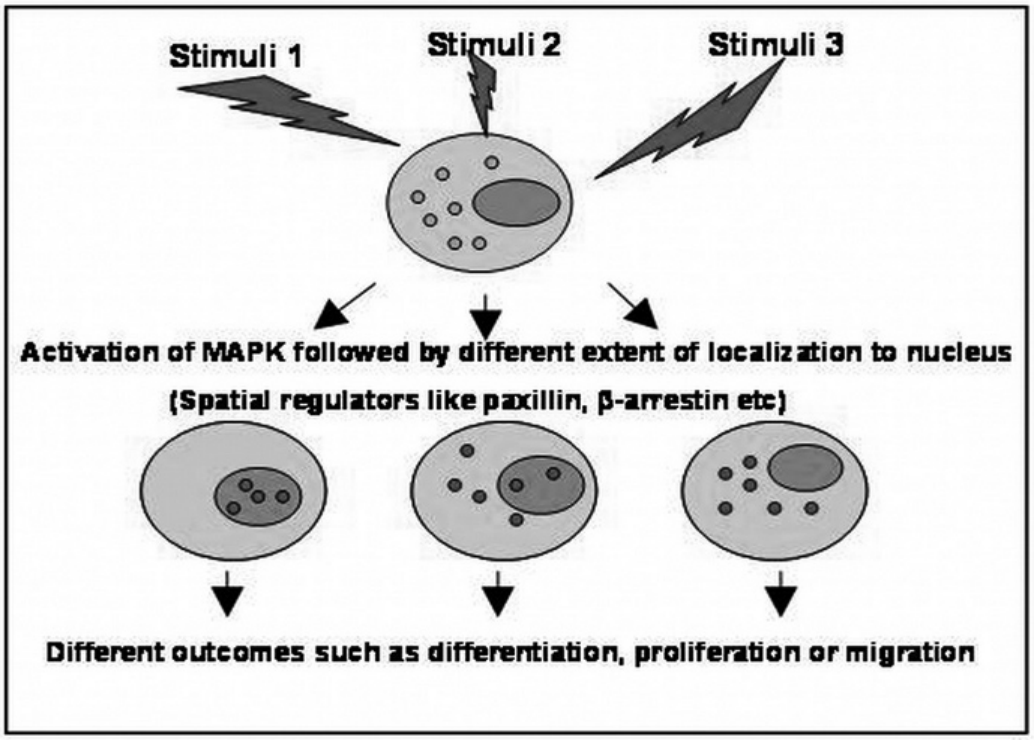
dn, dominant-negative

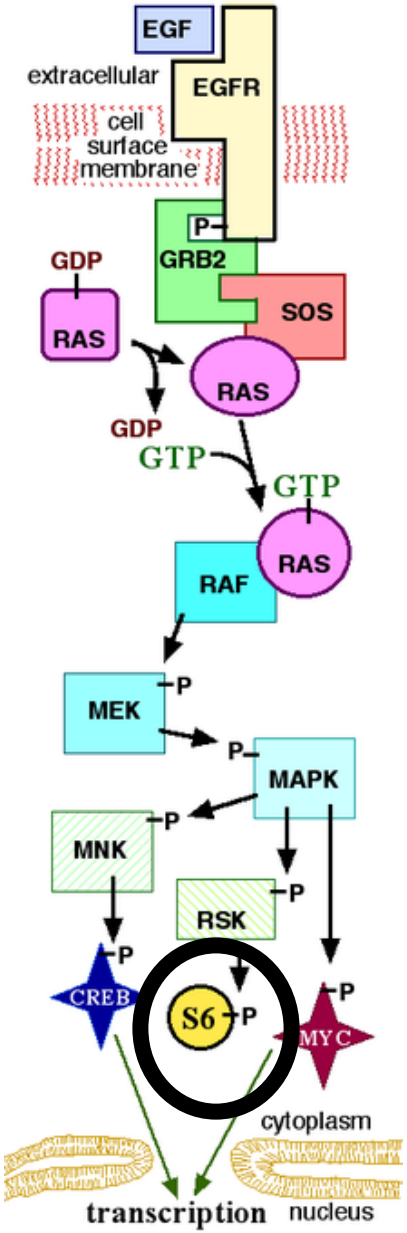
MAP kinase activation



MAP kinase signaling cascades

Respond to different stimuli by phosphorylating cytoplasmic components and nuclear transcription factors depending on the cellular context.





To identify relevant components in the signaling pathway

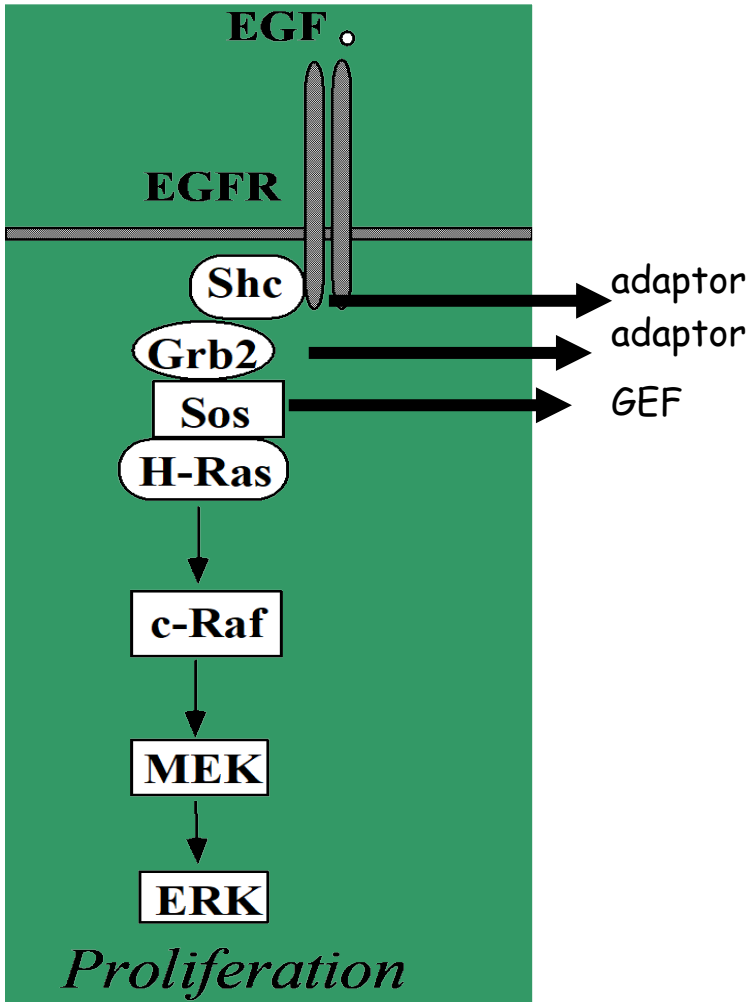
work "down"from the receptor in a stepwise manner.

start with a cellular event known to be affected bythe growth factors and to work "up" toward the receptor.

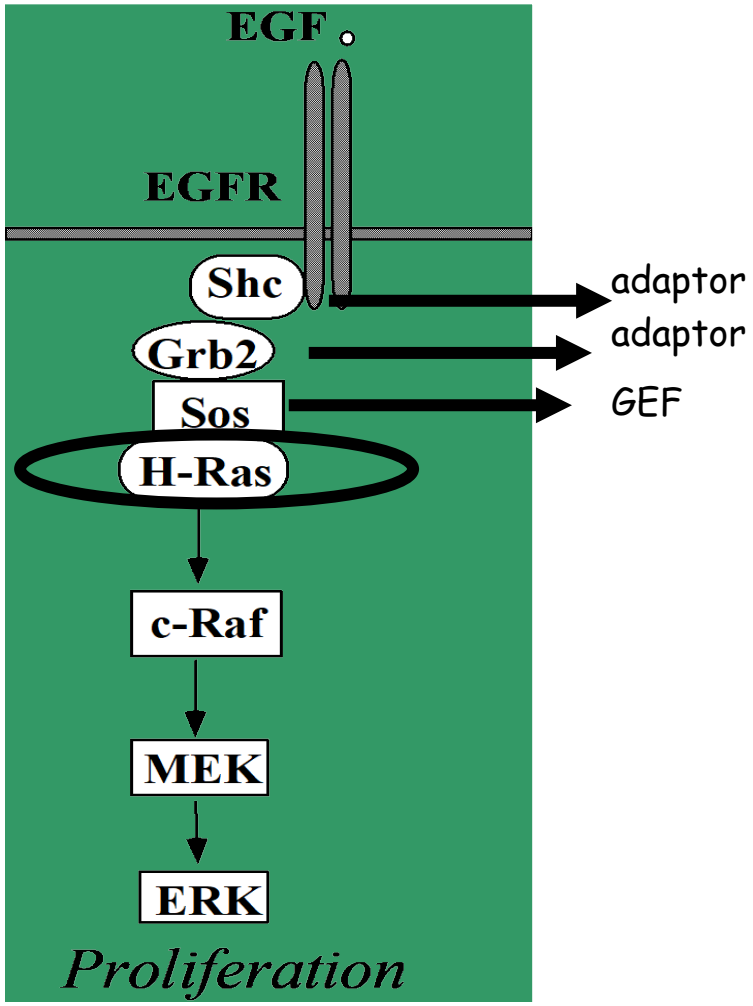
First substrate identified to be phosphorylated upon growth factor stimulation (ribosomal protein S6).

Cobb, M. H. and Rosen, 0. M. (1983) Novak-Hofer, L and Thomas, C. (1984) Kozma, S. C., Ferrari, S., Bassand, P., Siegmann, M., Totty, N., and Thomas, C. (1990)

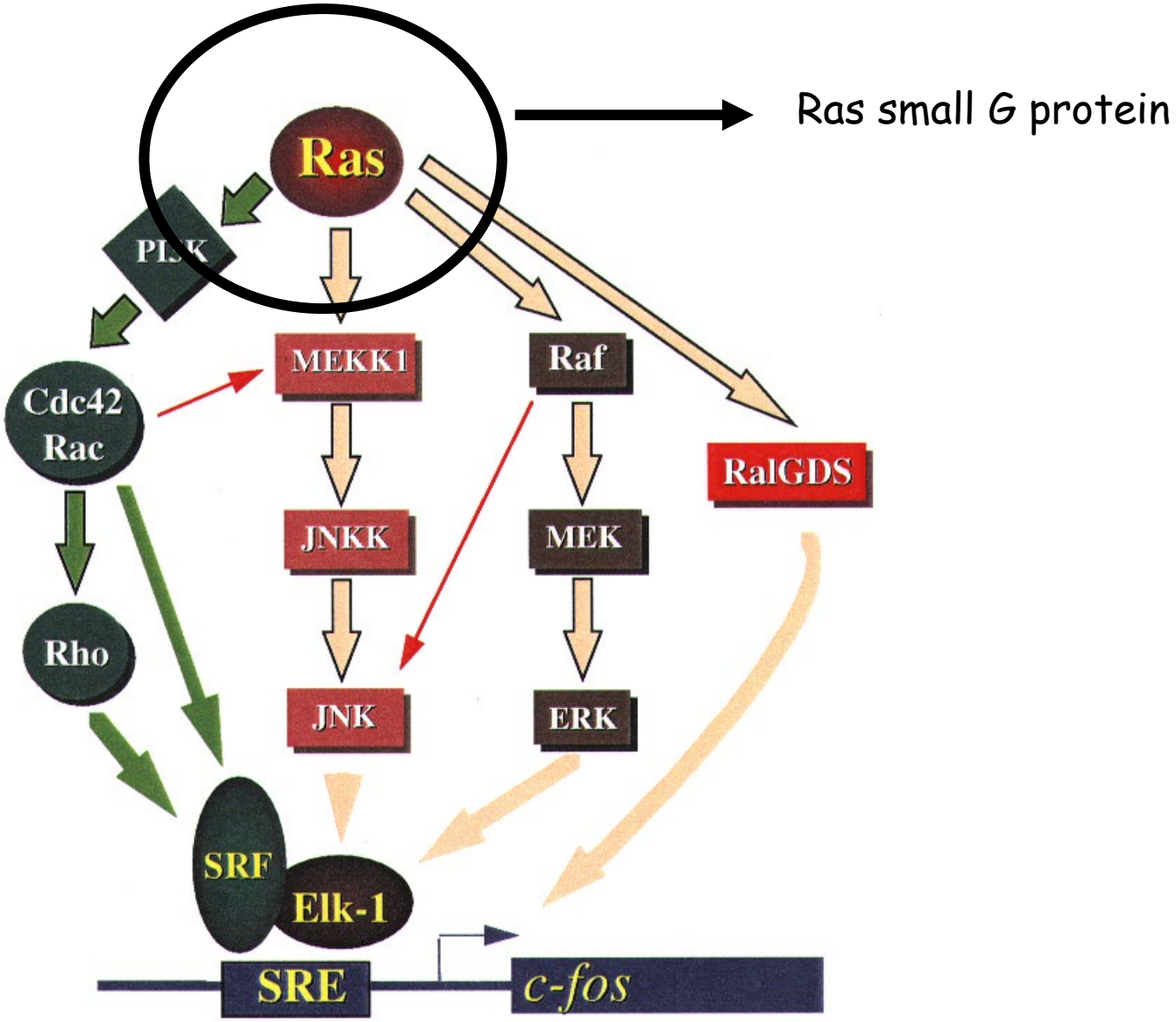
ERK kinase signaling cascades



ERK kinase signaling cascades



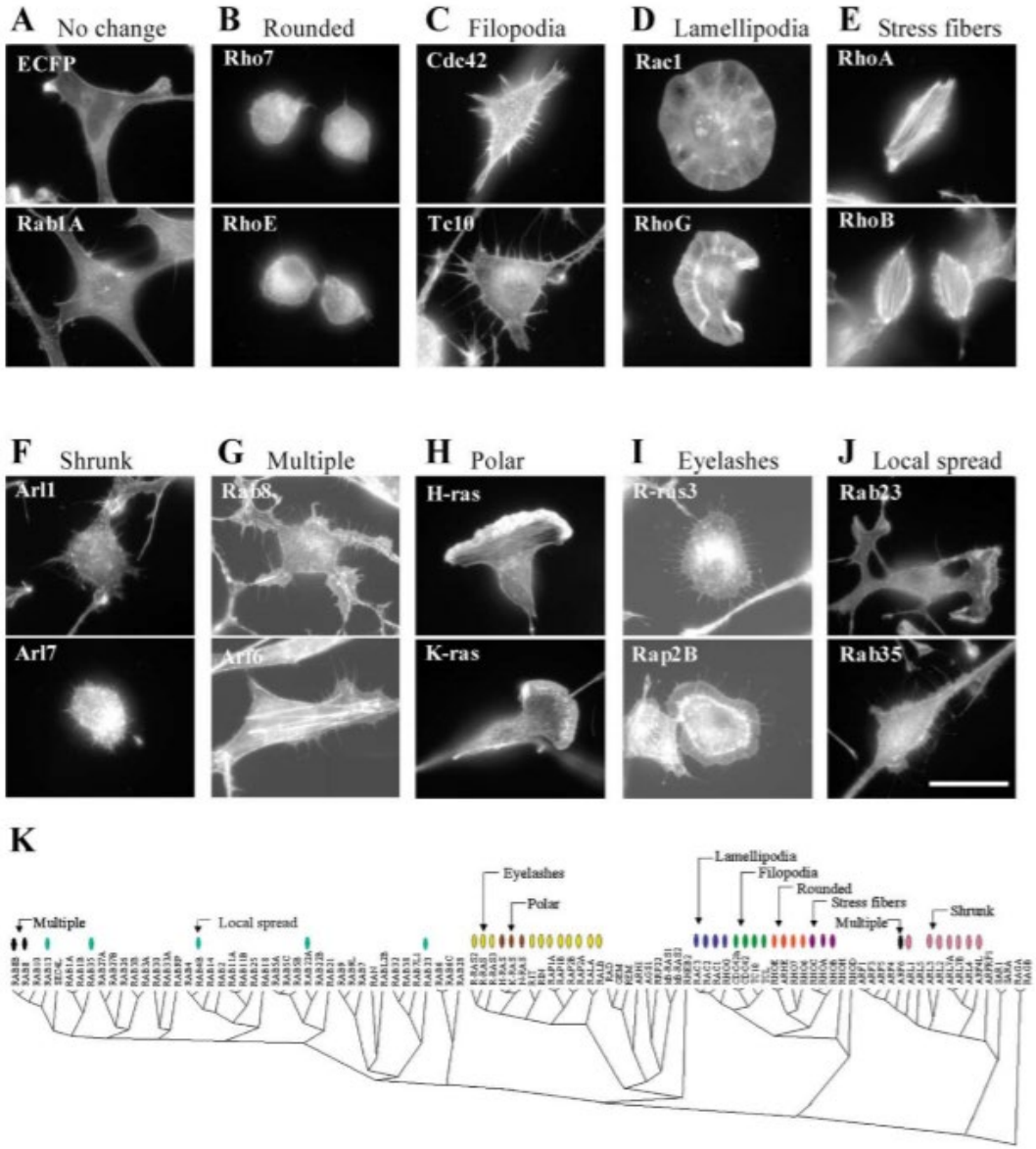
ERK kinase signaling cascades



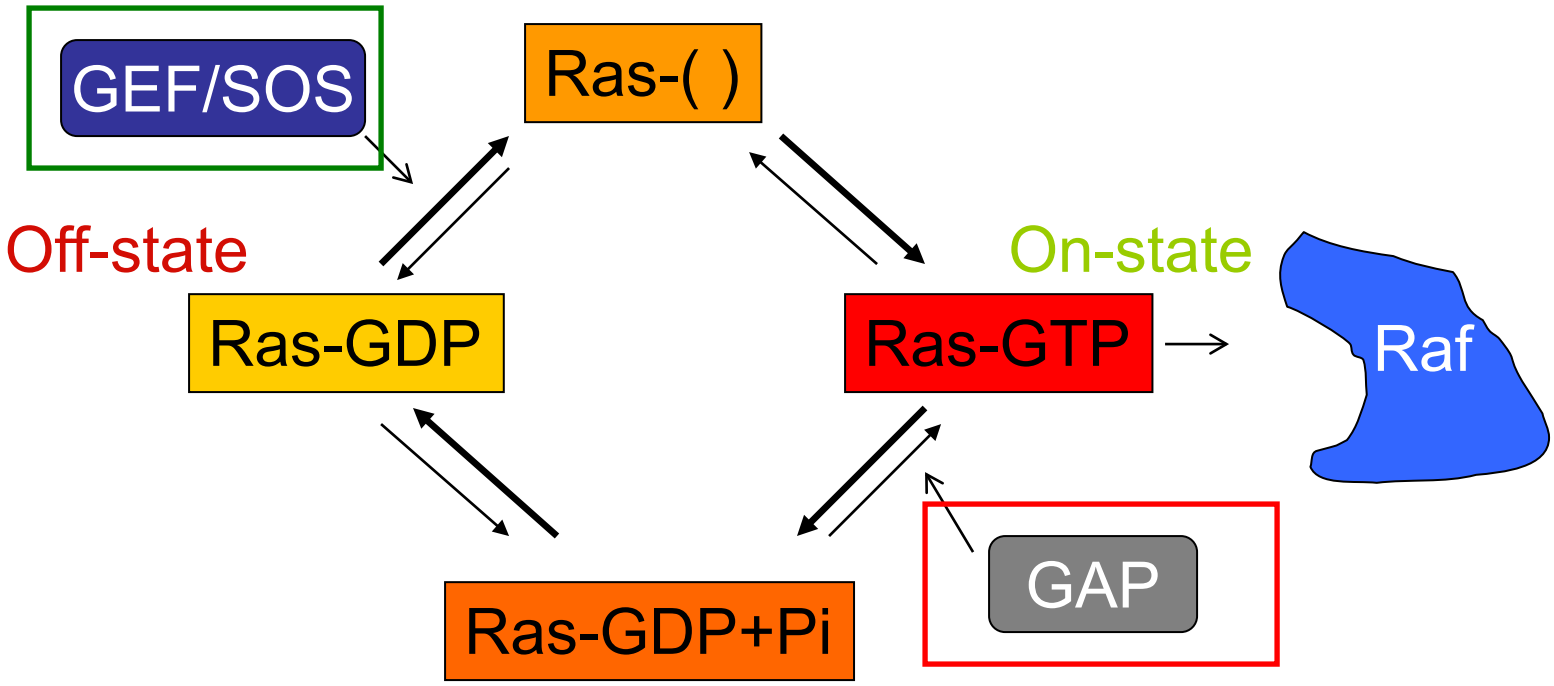
Cell, Vol. 113, 315–328, May 2, 2003, Copyright ©2003 by Cell Press

Switch-of-Function Mutants Based on Morphology Classification of Ras Superfamily Small GTPases

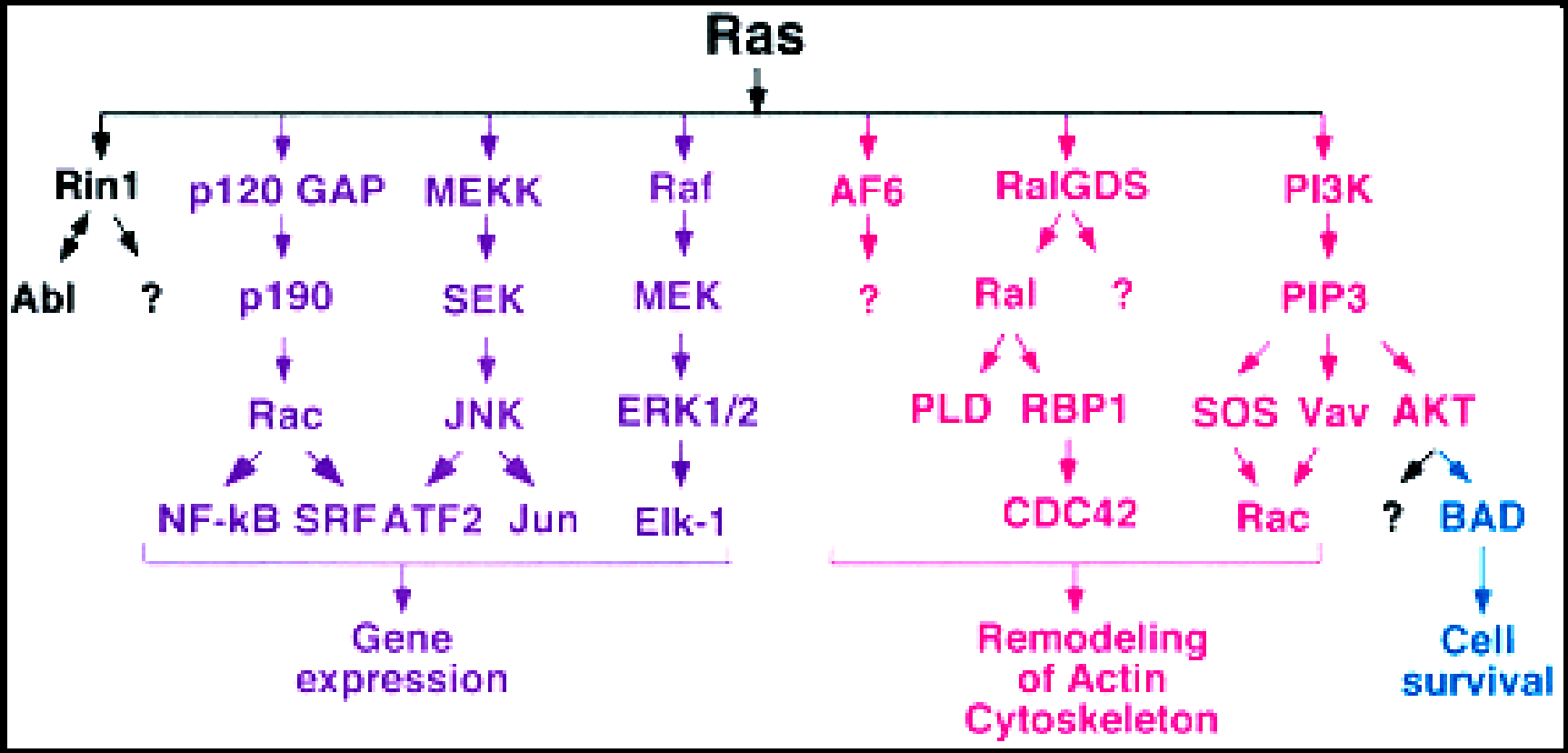
Won Do Heo and Tobias Meyer*
Department of Molecular Pharmacology
Stanford University School of Medicine
269 Campus Drive, Room 3215
Stanford, California, 94305



RAS GTP Cycle



Ras has multiple downstream substrates

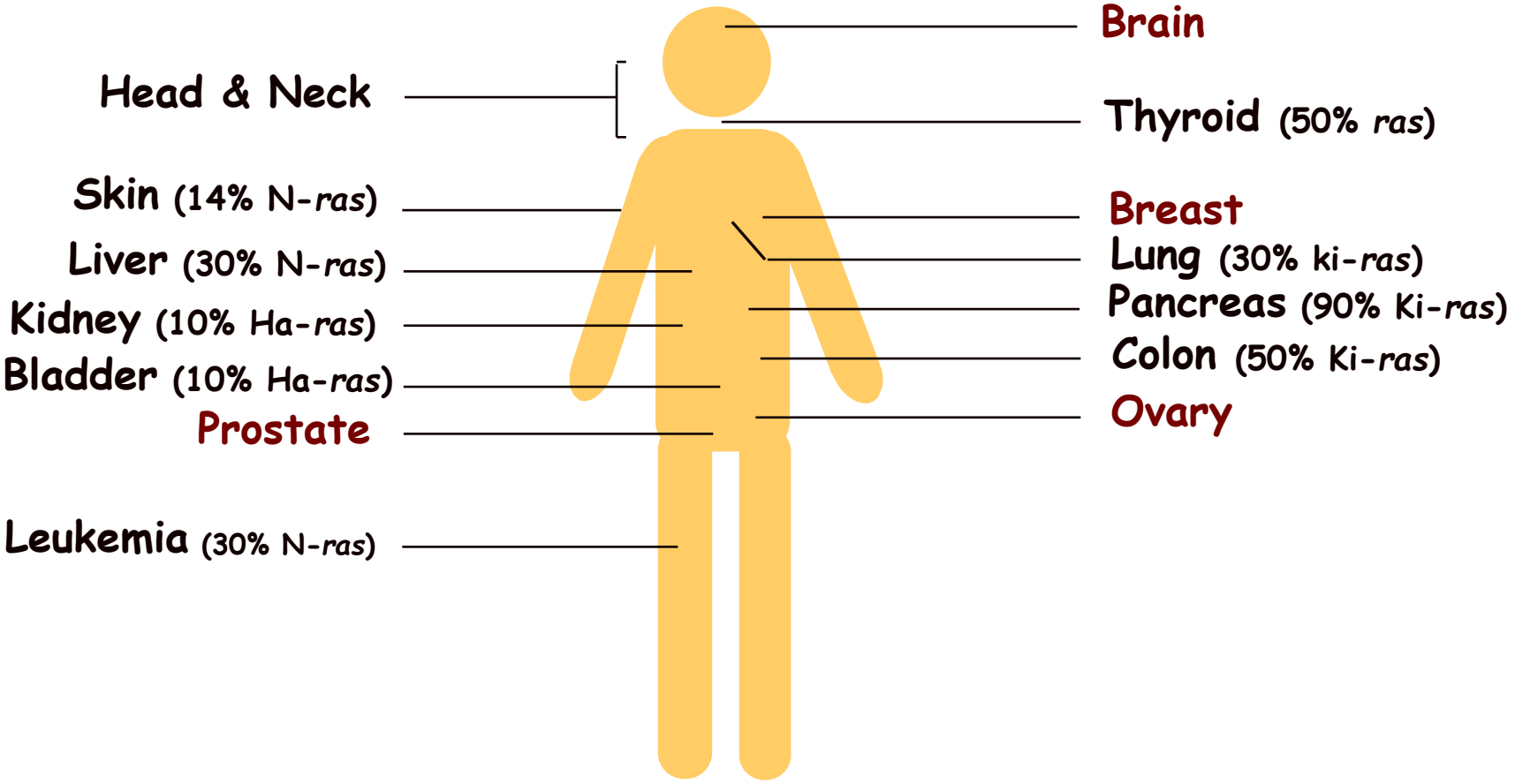


Vojtek & Der (1998) J. Biol. Chem 273: 19925

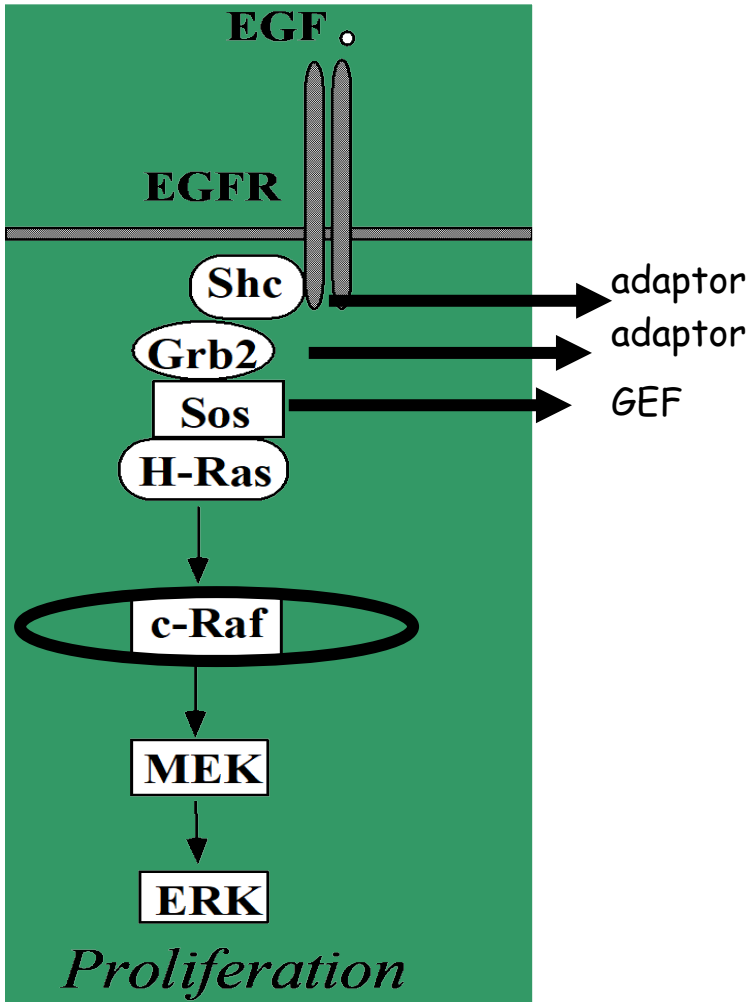
Amino acid substitutions in Ras family proteins

	amino acid position			
Ras gene	**12**	**59**	**61**	**Tumor**
c-ras (H, K, N)	Gly	Ala	Gln	normal cells
H-ras	Gly	Ala	**Leu**	lung carcinoma
	Val	Ala	Gln	bladder carcinoma
K-ras	**Cys**	Ala	Gln	lung carcinoma
	Arg	Ala	Gln	lung carcinoma
	Val	Ala	Gln	colon carcinoma
N-ras	Gly	Ala	**Lys**	neuroblastoma
	Gly	Ala	**Arg**	lung carcinoma
				Murine sarcoma virus
H-ras	**Arg**	**Thr**	Gln	Harvey strain
K-ras	**Ser**	**Thr**	Gln	Kirsten strain

Incidence of *RAS* mutations in human cancer



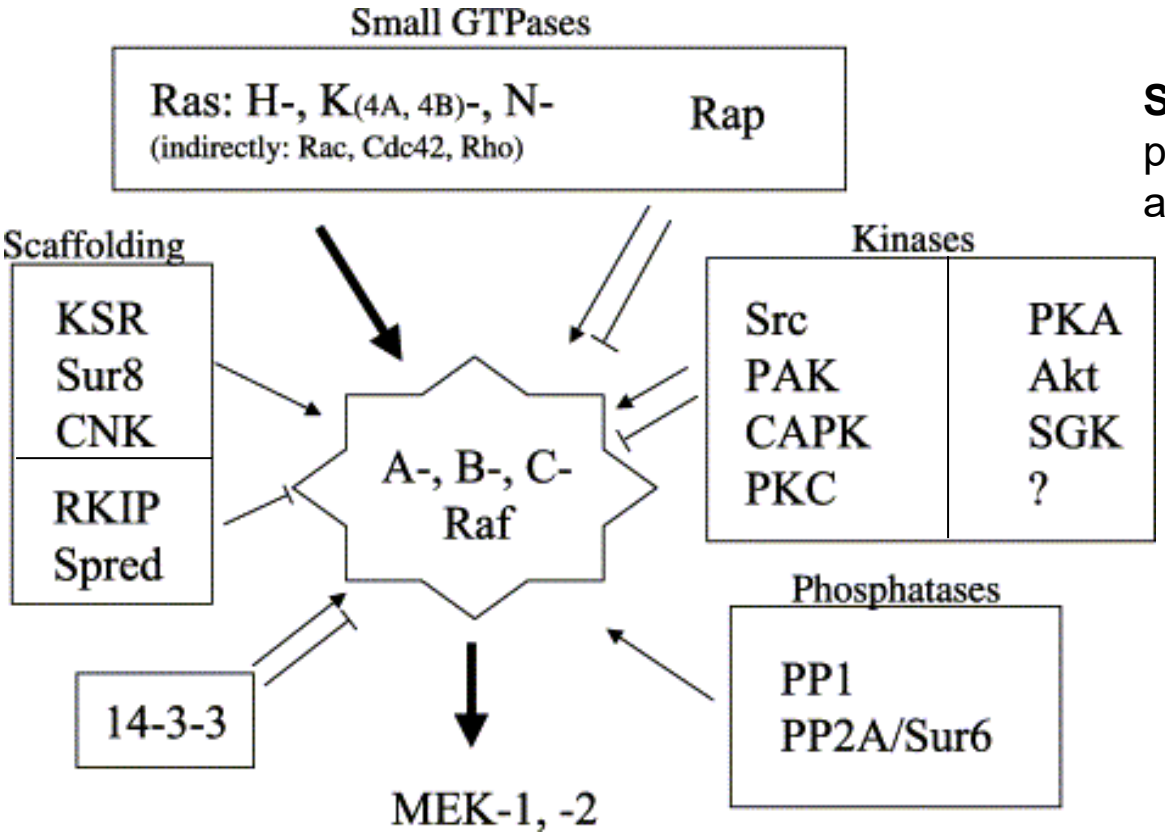
ERK kinase signaling cascades



Raf is a point of regulation in the MAP kinase pathway

Rap1 activates B-Raf but inhibits c-Raf-1.

Sur8: enhancing Ras and EGF-stimulated Raf activity in C. elegans.



Src, **PAK** and **PKC** phosphorylate c-Raf-1 and activate it.

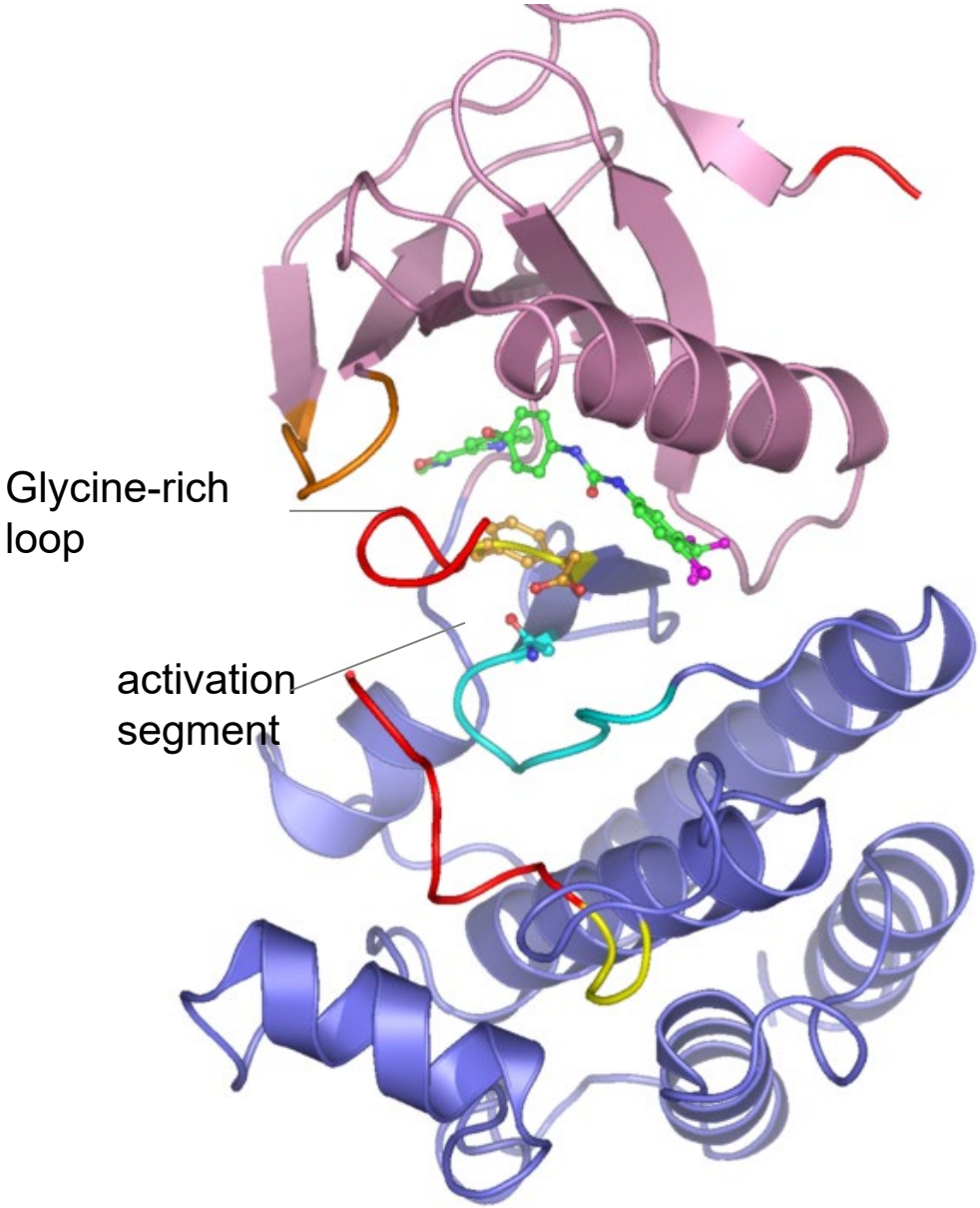
PKA inhibits c-Raf-1 activity

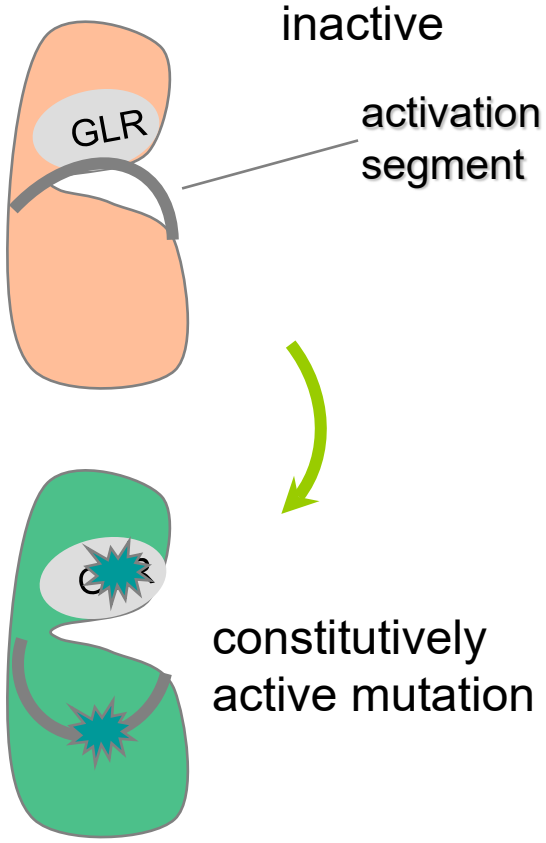
SGK: serum and glucocorticoid-inducible kinase. Homologous to Akt. Specifically target S364 on B-Raf.

RKIP: raf kinase inhibitor protein
Spred: Sprouty-related protein in *Drosophila*. Inhibiting Raf phosphorylation.

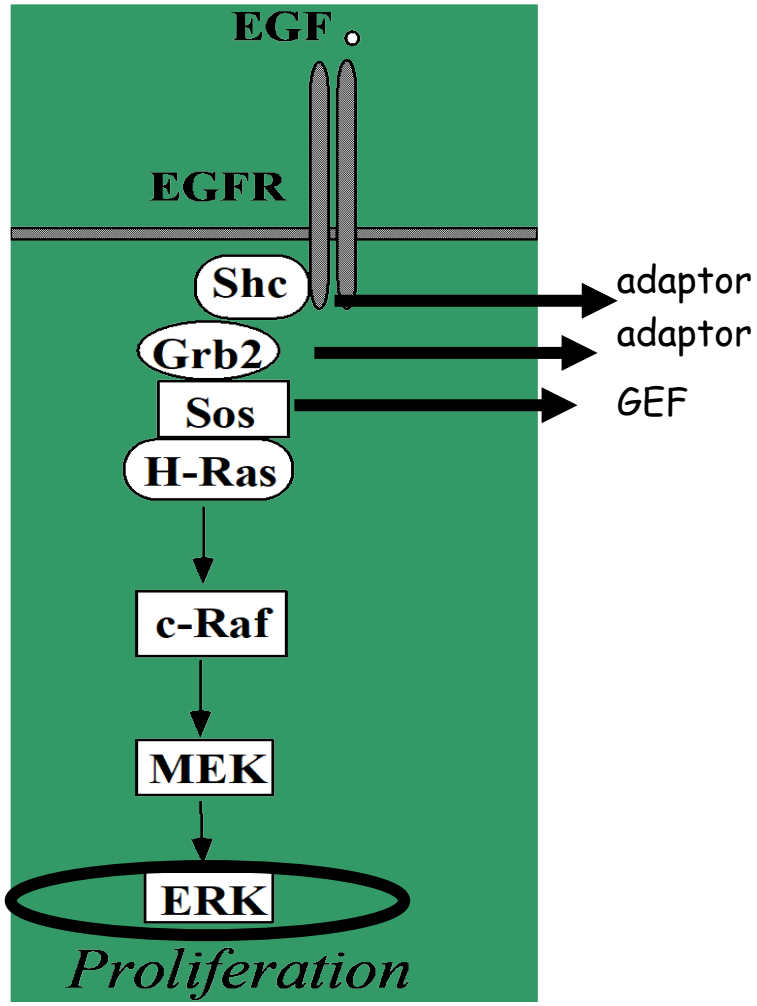
Sur6; a subunit of PP2A in C. elegans, an activator of Raf-1.

B-RAF activation by mutation



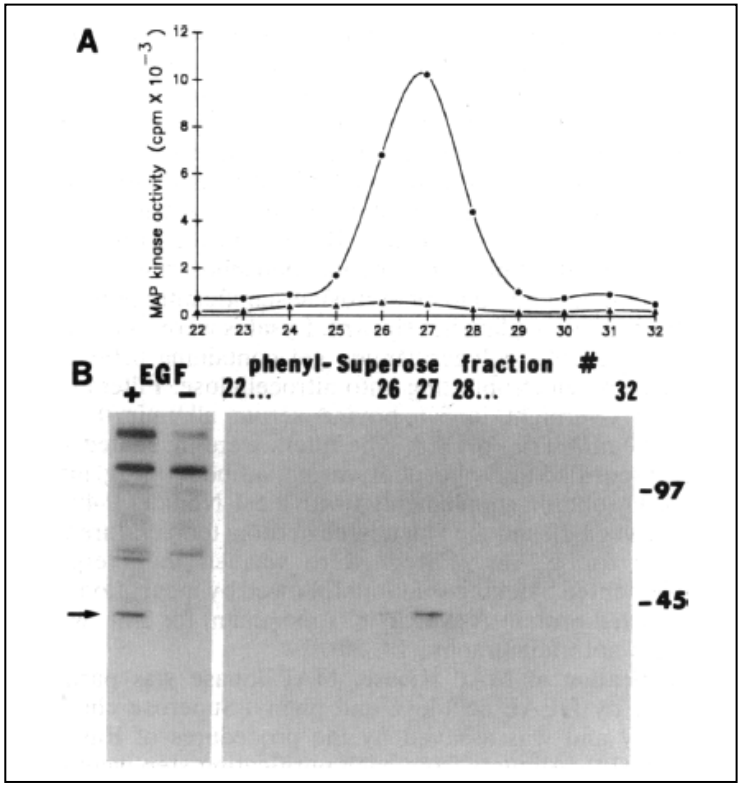


ERK kinase signaling cascades



ERK kinase signaling cascades

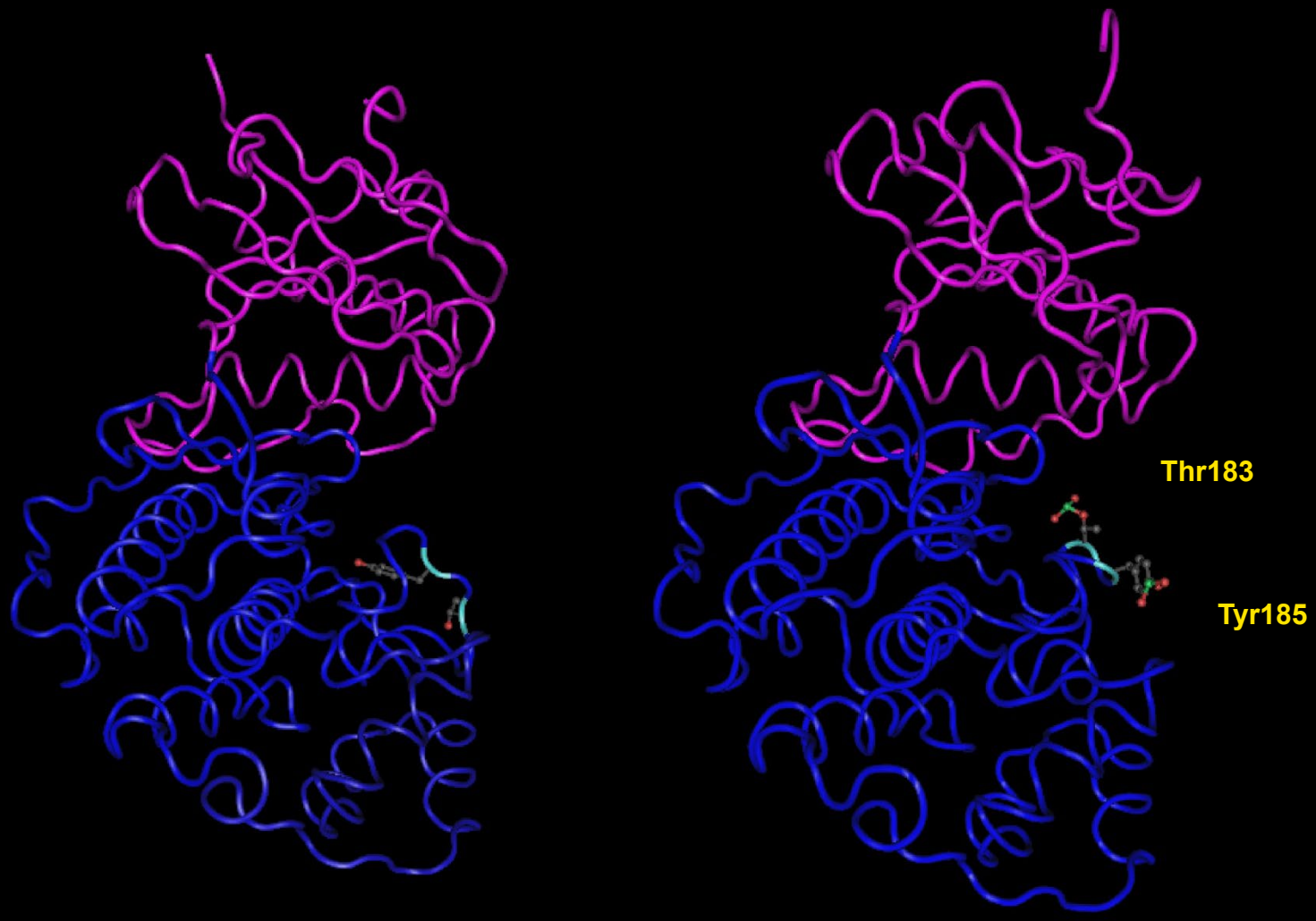
pp42 MAP Kinase/MAPK/ErK1/2
Myelin Basic Protein Kinase
Microtubule Associated Protein 2 Kinase
Mitogen-activated Protein Kinase



Rossomando et. al. (1989) *PNAS* 86: 6940

Canagarajah et al Cell 90, 859-869 (1997)

ERK2

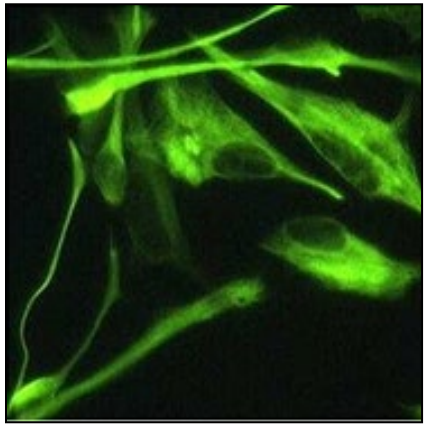


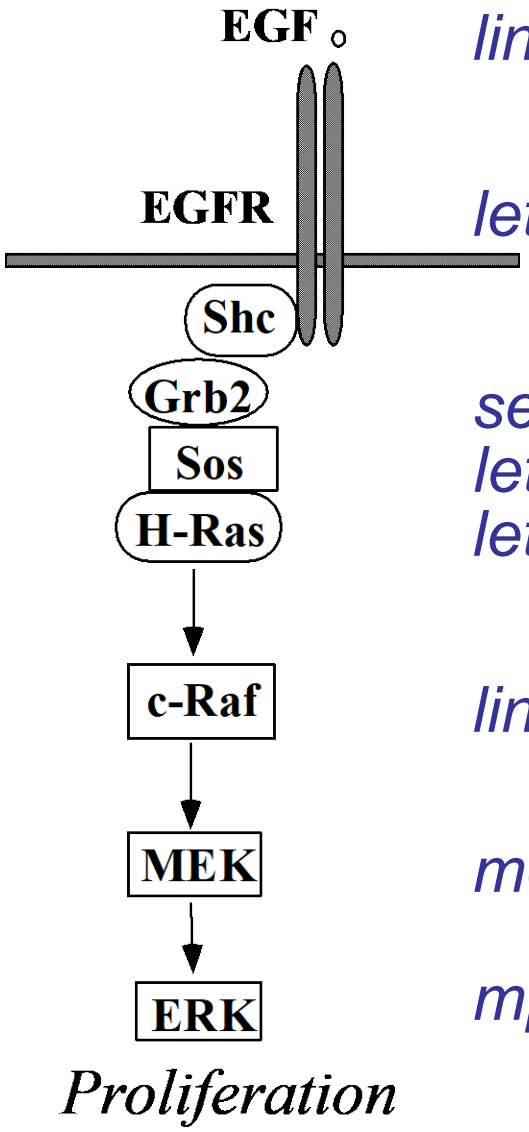
Unphosphorylated

phosphorylated

Evolutionary Conservation of the MAPK Signaling pathway

mammalian





lin-3

let-23

sem-5

let-341

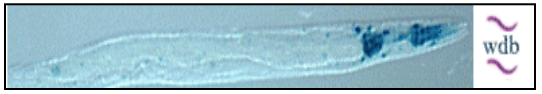
let-60

lin-45

mek-2

mpk-1

C. Elegans



Chardin et. al. (1993) *Science* 260: 1338
VanAelst et. al. (1993) *PNAS* 90: 6213
Han et. al. (1993) *Nature* 363: 133
Warne et. al. (1993) *Nature* 364: 352
Vojtek et. al. (1993) *Cell* 74: 205

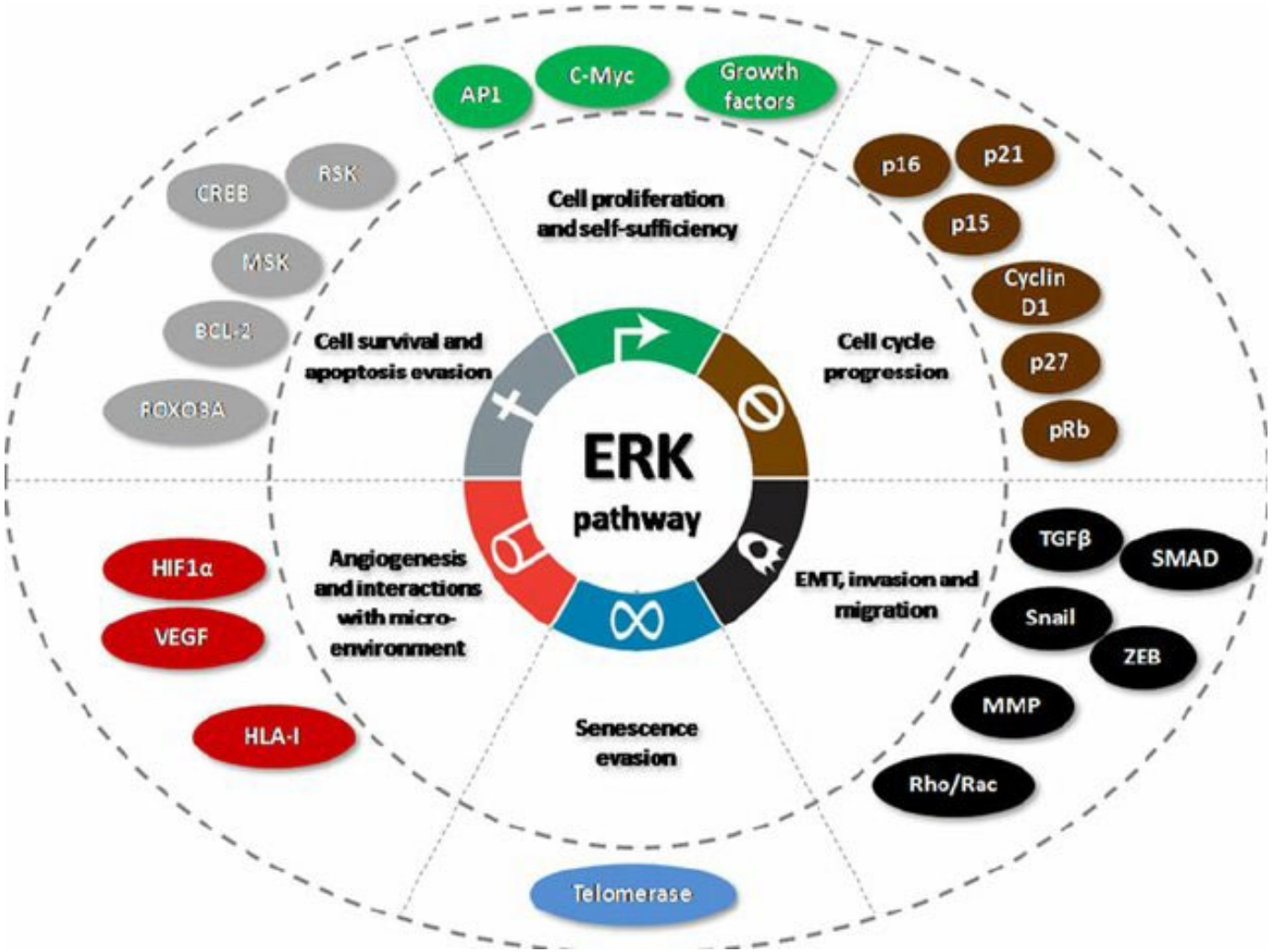
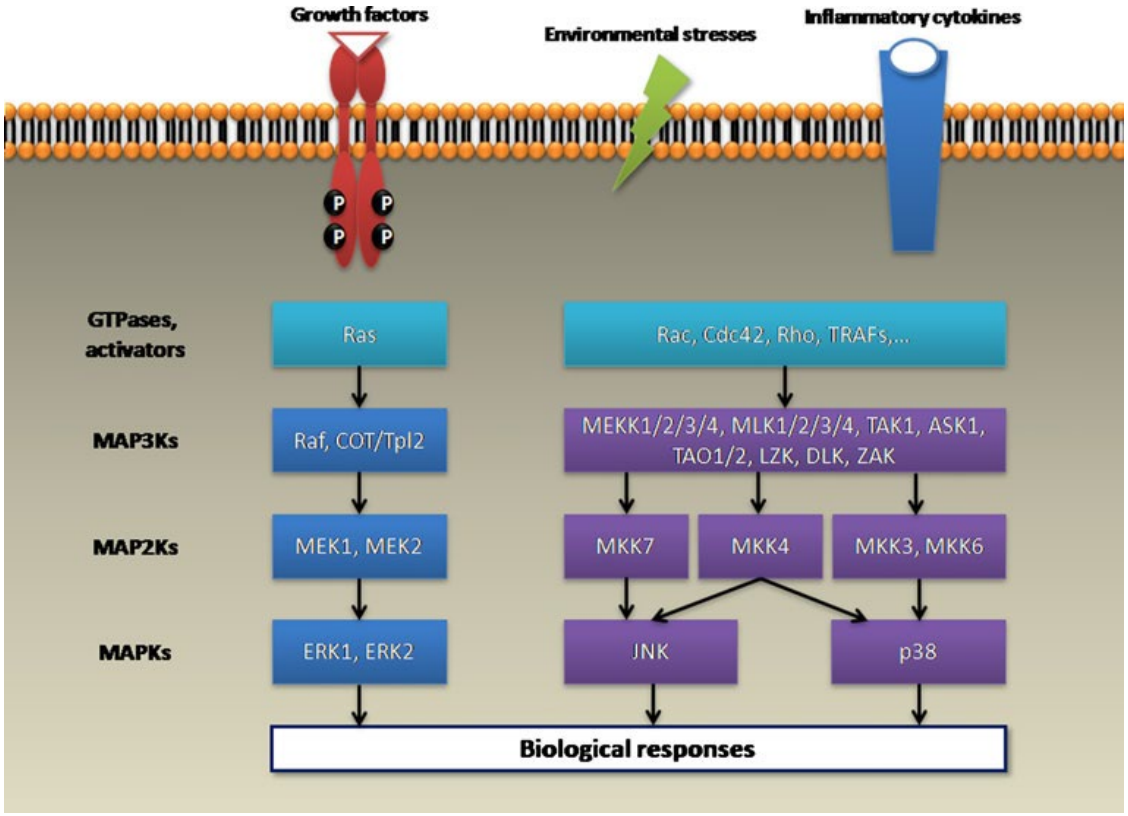


Fig. 3 Biological consequences of the Ras–ERK pathway activation and the main targets involved (adapted from D. Hanahan and Weinberg) [19]

ERK, JNK and P38



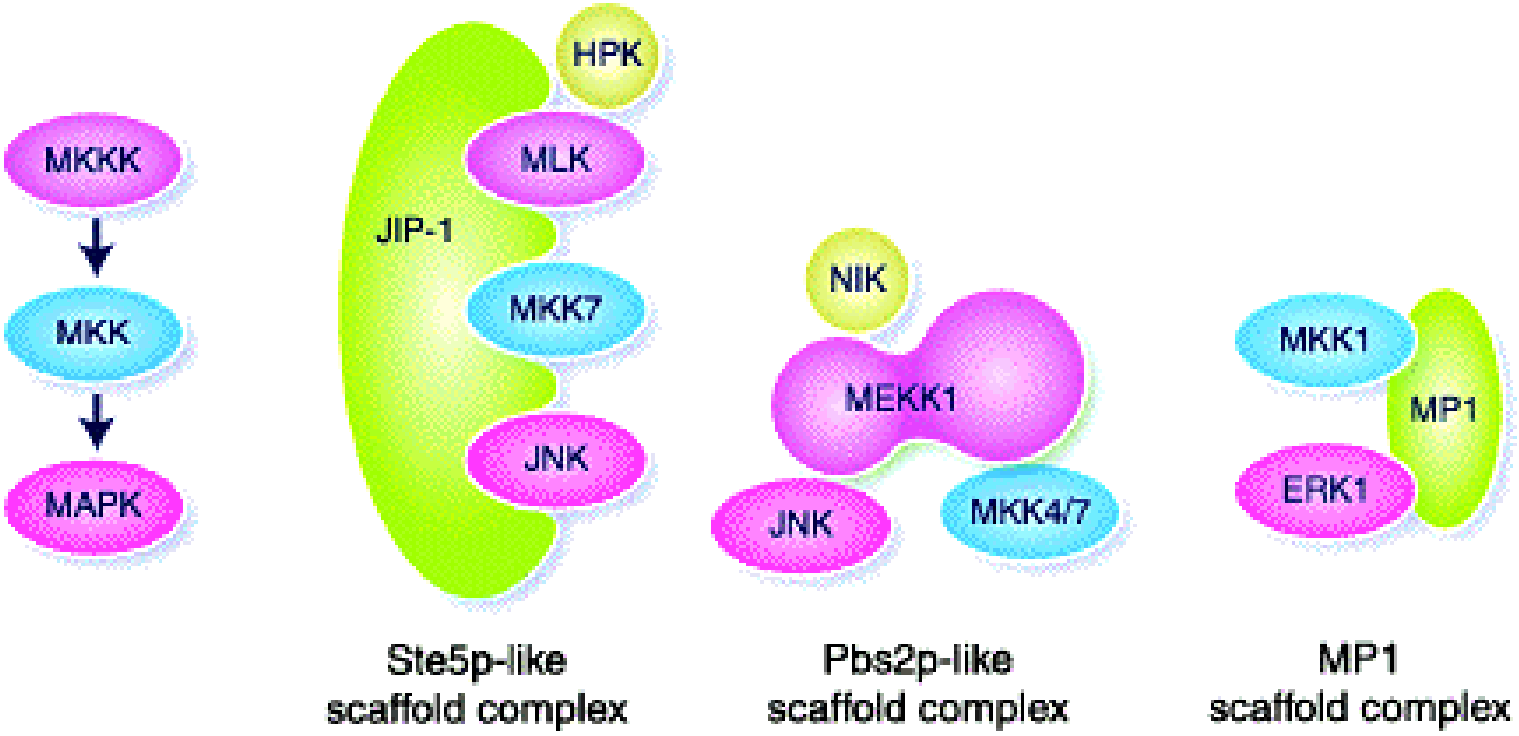
NON-THEMATIC REVIEW

Targeting the Ras–ERK pathway in pancreatic adenocarcinoma

Cindy Neuzillet · Pascal Hammel · Annemilaï Tijeras-Raballand · Anne Couvelard · Eric Raymond

Published online: 21 October 2012

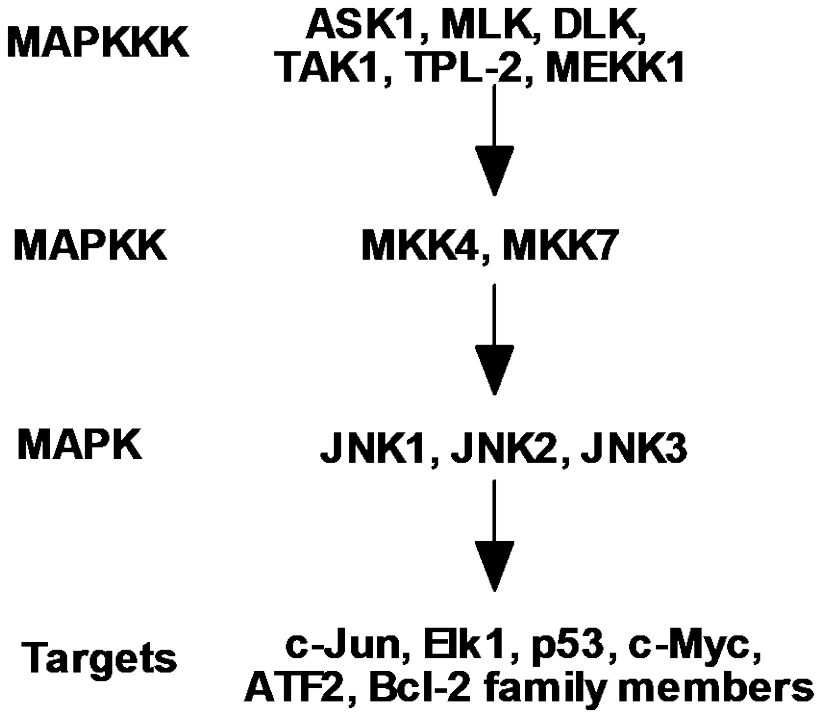
JNK kinase signaling cascades



JNK kinase signaling cascades

c-Jun N-terminal kinases (JNKs), also known as stress-activated protein kinases (SAPKs).

JNK kinase signaling cascades

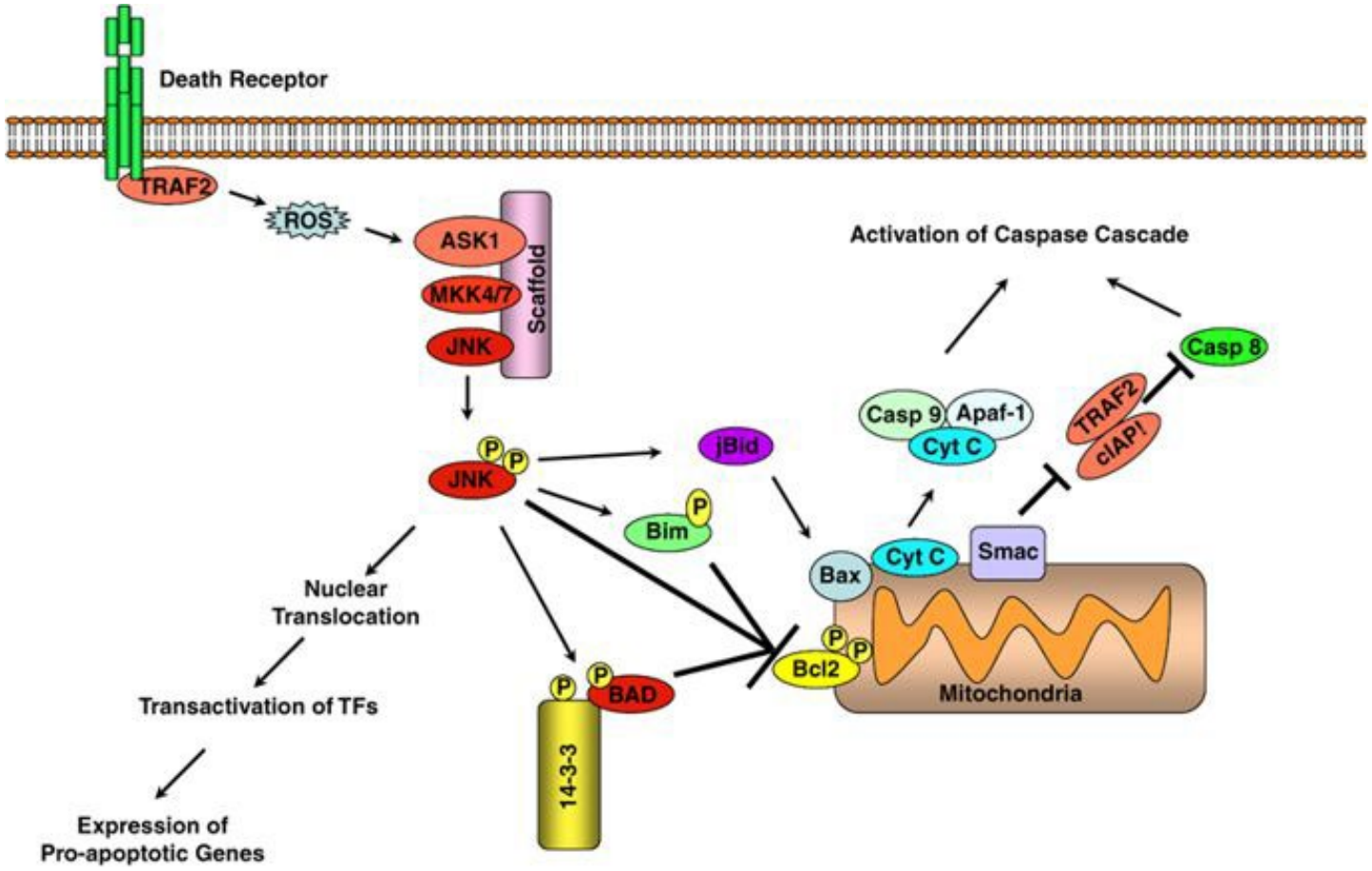


-JNK1 and JNK2 are widely expressed. JNK3 is mainly expressed in neuronal and heart tissues.

-JNK plays a central role in the regulation of proliferation, differentiation, migration, transformation, immune and inflammatory responses, and programmed cell death (apoptosis, autophagy and necrosis).

-Deregulation of the JNK activity has been implicated in many human diseases including certain types of cancer, cardiac hypertrophy and ischemia, immune disorders, liver injury, obesity, and neurodegenerative disorders such as Alzheimer's and Parkinson's diseases.

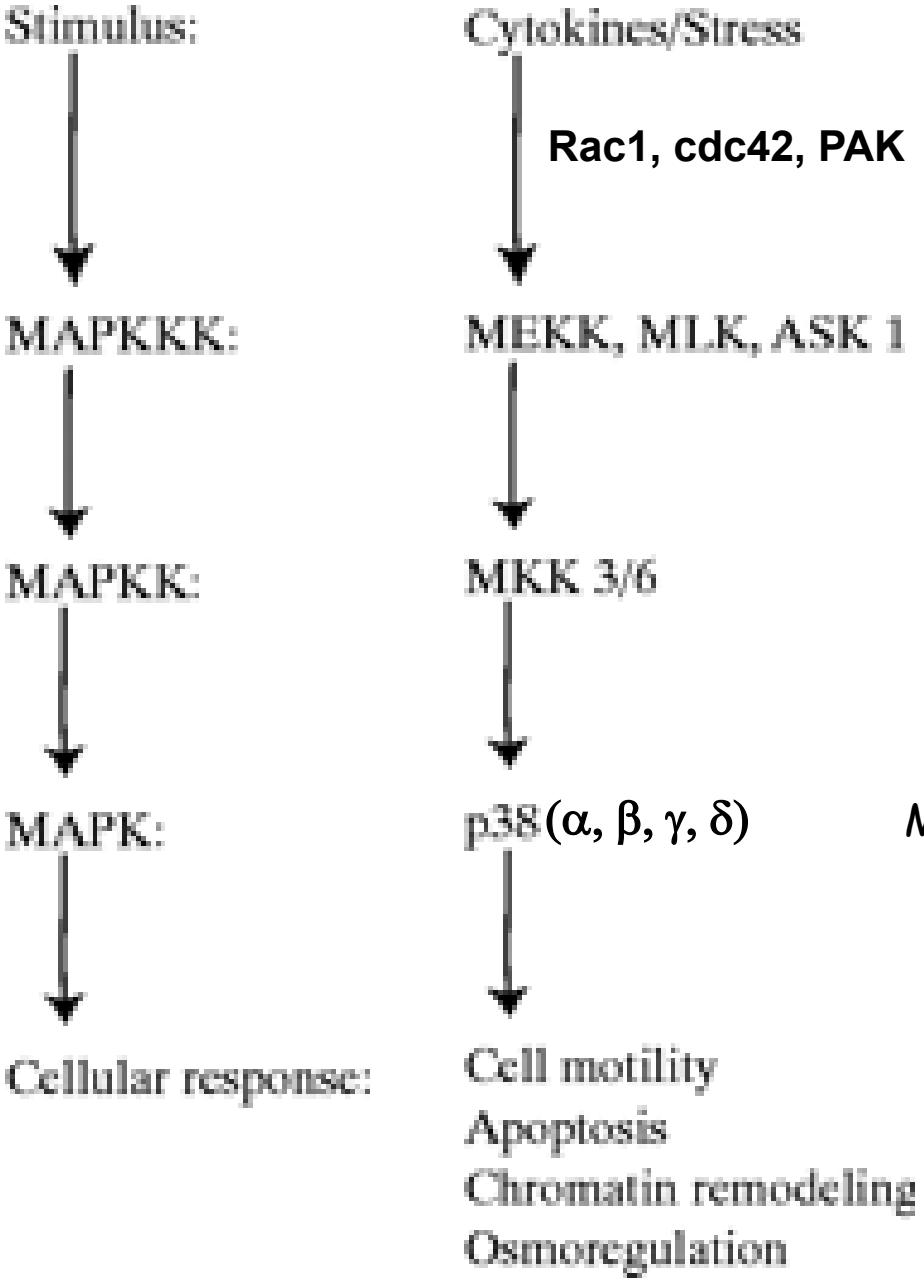
JNK kinase signaling cascades and apoptosis



p38 kinase signaling cascades

p38 isoforms.(p38-α (MAPK14), -β (MAPK11), -γ (MAPK12 or ERK6) and -δ (MAPK13 or SAPK4)) Both JNK and p38 signaling pathways are responsive to stress stimuli, such as cytokines, ultraviolet irradiation, heat shock, and osmotic shock, and are involved in cell differentiation and apoptosis.

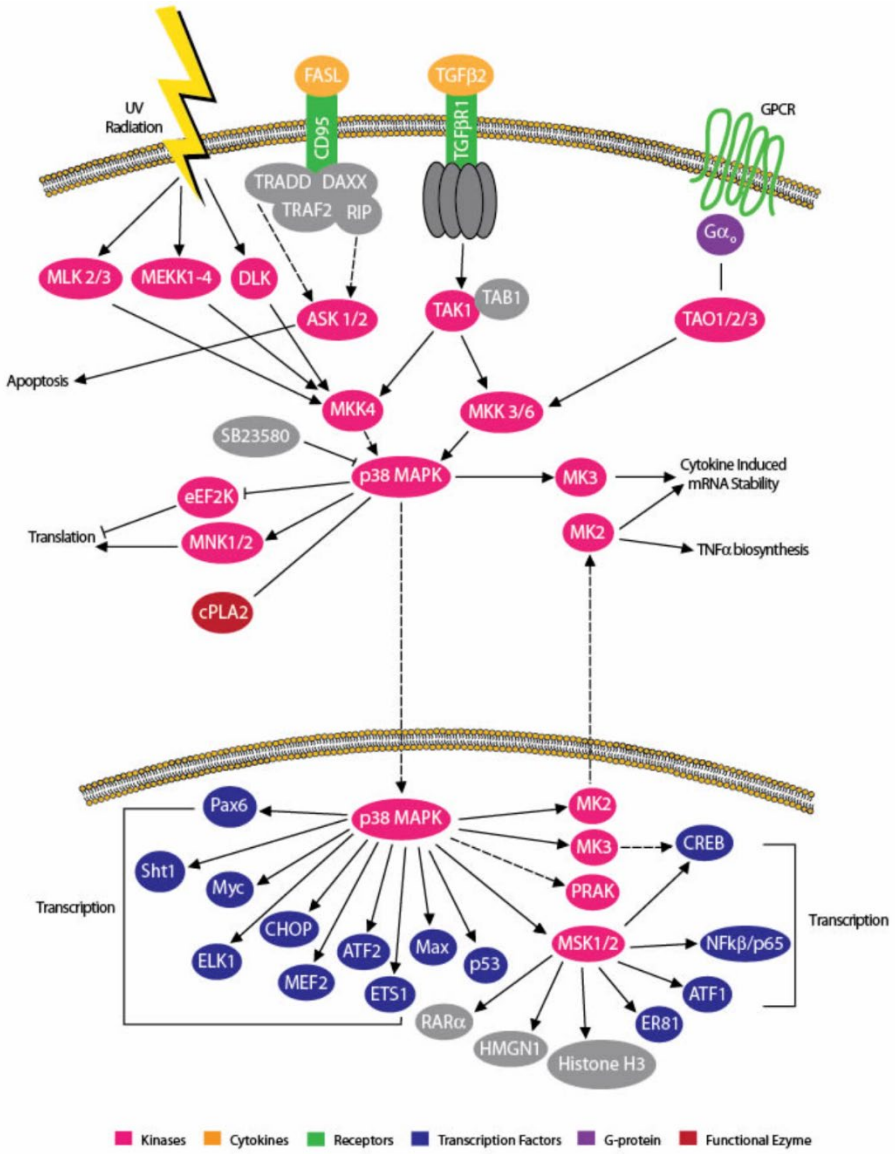
p38 kinase signaling cascades



Environmental stress: heat, osmotic and oxidative stresses, Ionizing radiation, ischemia, inflammation or cytokine and TNF receptor signaling

MKK4 can also phosphorylate p38a

Inflammation
cell cycle regulation (arrest)
cardiomyocyte hypertrophy
Development
cell differentiation
tumor suppression


UV Radiation
FASL
CD95
TGFβ2
TGFβR1
GPCR
TRADD
DAXX
TRAF2
RIP
Gα$_o$
MLK 2/3
MEKK1-4
DLK
ASK 1/2
TAK1
TAB1
TAO1/2/3
Apoptosis
MKK4
MKK 3/6
SB23580
p38 MAPK
MK3
Cytokine Induced mRNA Stability
eEF2K
Translation
MNK1/2
MK2
TNFα biosynthesis
cPLA2
Pax6
p38 MAPK
MK2
MK3
CREB
Sht1
Myc
PRAK
Transcription
CHOP
ATF2
Max
MSK1/2
NFkβ/p65
Transcription
ELK1
MEF2
p53
ETS1
ATF1
RARα
ER81
HMGN1
Histone H3
Kinases
Cytokines
Receptors
Transcription Factors
G-protein
Functional Ezyme

Targeting RTK-Ras-MAPK signaling pathways for cancer treatment

- Antagonists
- Monoclonal antibodies
- Small molecule inhibitors
- Cell-permeable peptide inhibitors (interfere with protein/ protein interactions).
- Dominant-negative regulators
- Antisense oligonuleotides
- Small interfering RNA

Development of inhibitors to target MAPK pathways

- **Dominant negative mutants of protein kinases** defective in ATP binding domains, membrane localization or catalytic activity.

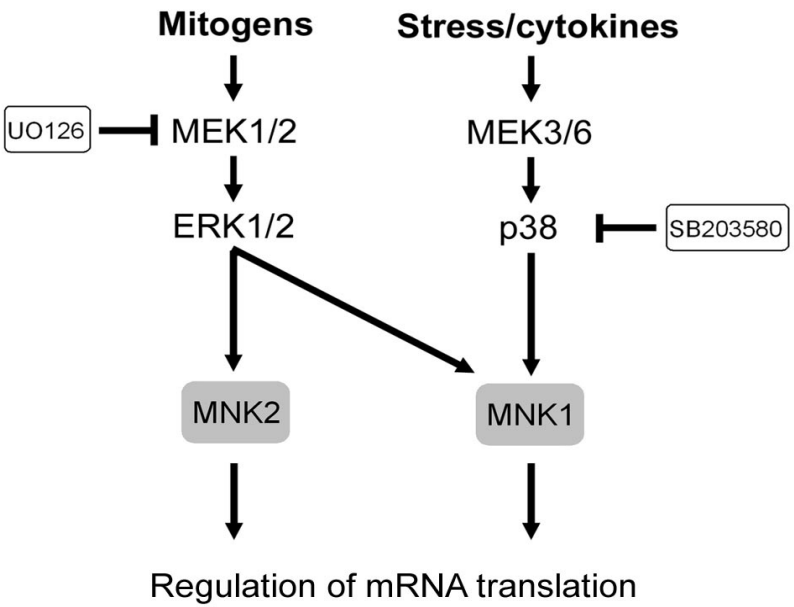
- **Pharmacological inhibitor of MEK1 and MEK2:** PD98059, PD184352 and U0126.

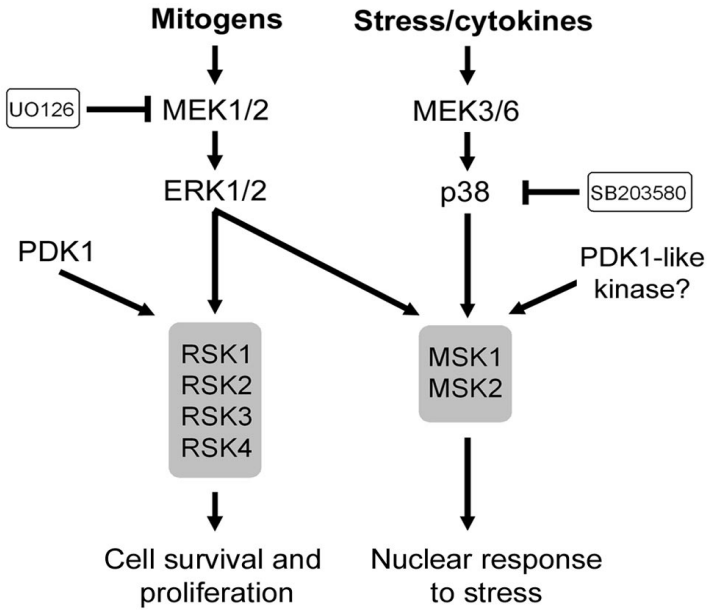
- **Small-molecule inhibitors of RAF:** sorafenib (BAY 43-9006) and antisense oligonucleotides of Raf; B-Raf^{V600E} inhibitors (PLX4032, GSK2118432)

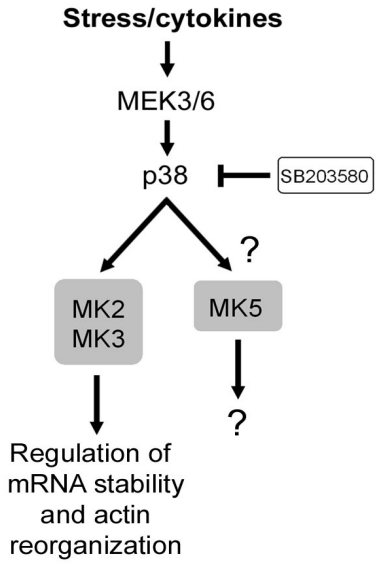
- **Inhibitors of JNK pathways**: SP600125 inhibits Jun-N-terminal kinase 2 (JNK2). CEP1347 (KT7515) is an inhibitor of mixed lineage kinases 1, 2 and 3 (MLK1, 2 and 3)

- **Inhibitors of p38 evaluated in clinical trials:** Vertex 745 (VX745) and RPR200765A are in clinical trials for rheumatoid arthritis, whereas SB235699 (HEP689) has been evaluated for the treatment of psoriasis. SCIO469 is a p38 inhibitor also in clinical trials for rheumatoid arthritis.

Development of inhibitors to target MAPK pathways





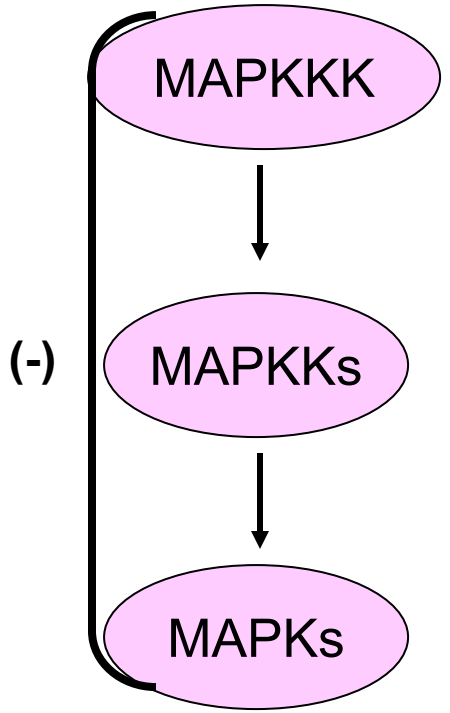


MAPK Inactivation

1) Regulation by phosphatases

A. MAPK phosphatases (MKPs):
Dual Specificity Phosphatases

B. Serine/Threonine phosphatases
PP1, PP2A (substrates: MKK, ERK)

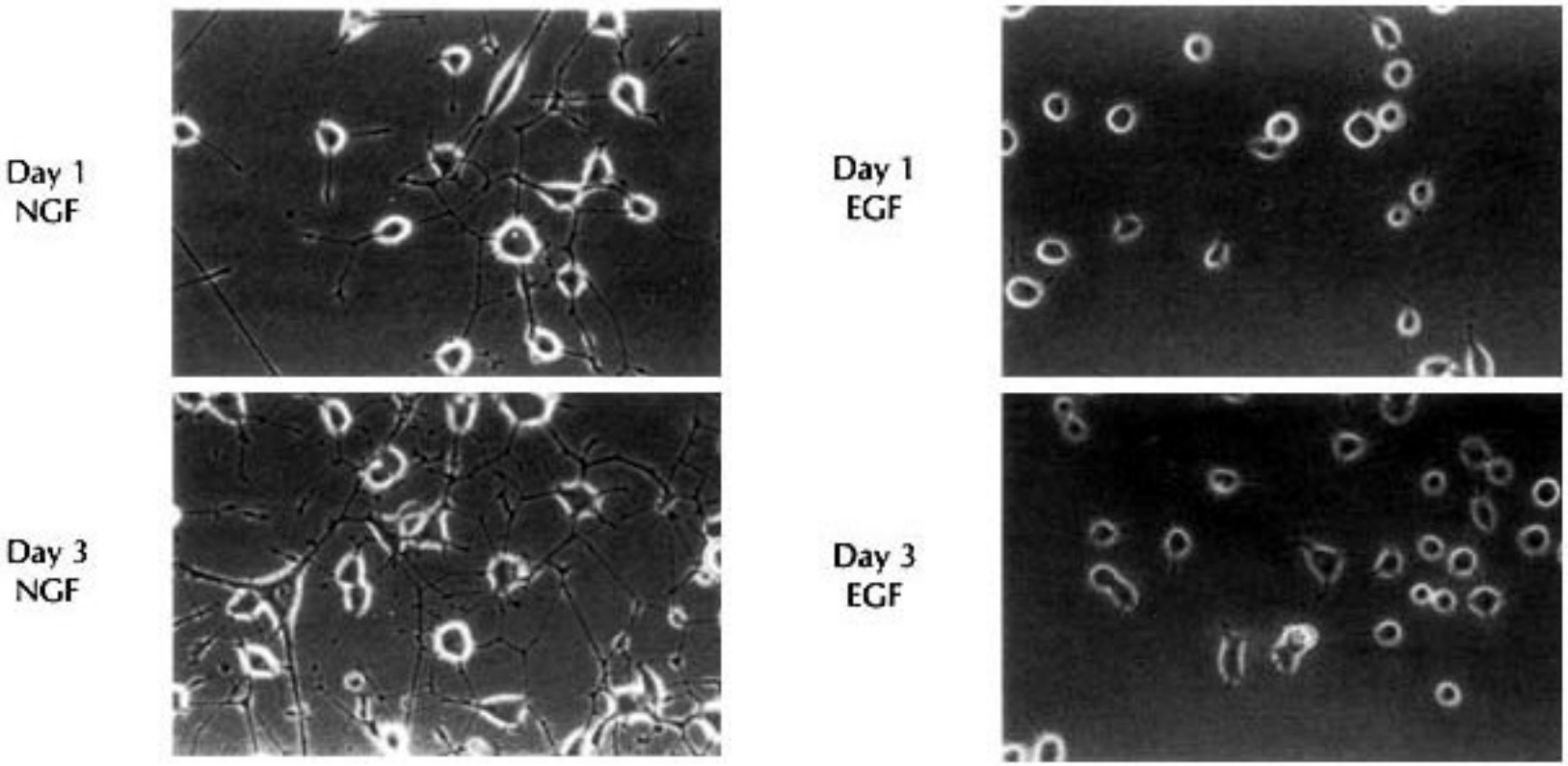


2) Negative feedback
phosphorylation and inactivation of MEK1, C-RAF and SOS by ERK

Cell fate determination by the MAPK Pathway

How is specificity of MAPK signaling determined ?

PC12 pheochromoytoma cell line



EGF triggers neuronal differentiation of PC12 cells that overexpress the EGF receptor
rg/10.1016/S0960-9822(00)00154-8

A avaliação de células individuais é importante para se compreender o tipo de resposta a um determinado estímulo

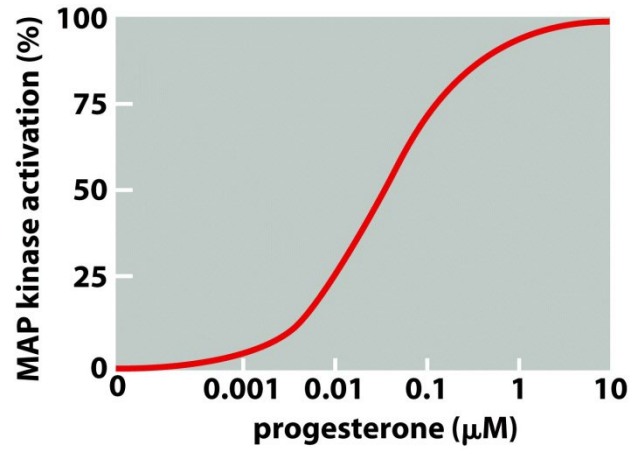


Figure 15-24a Molecular Biology of the Cell 5/e (© Garland Science 2008)

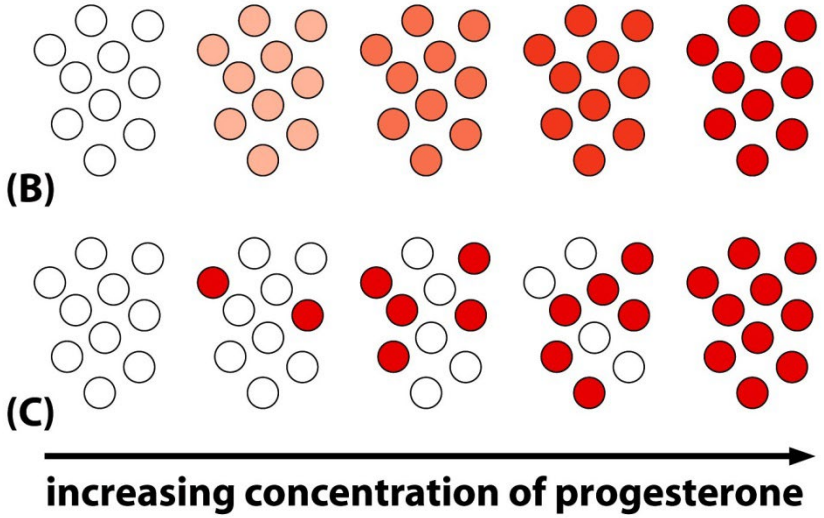
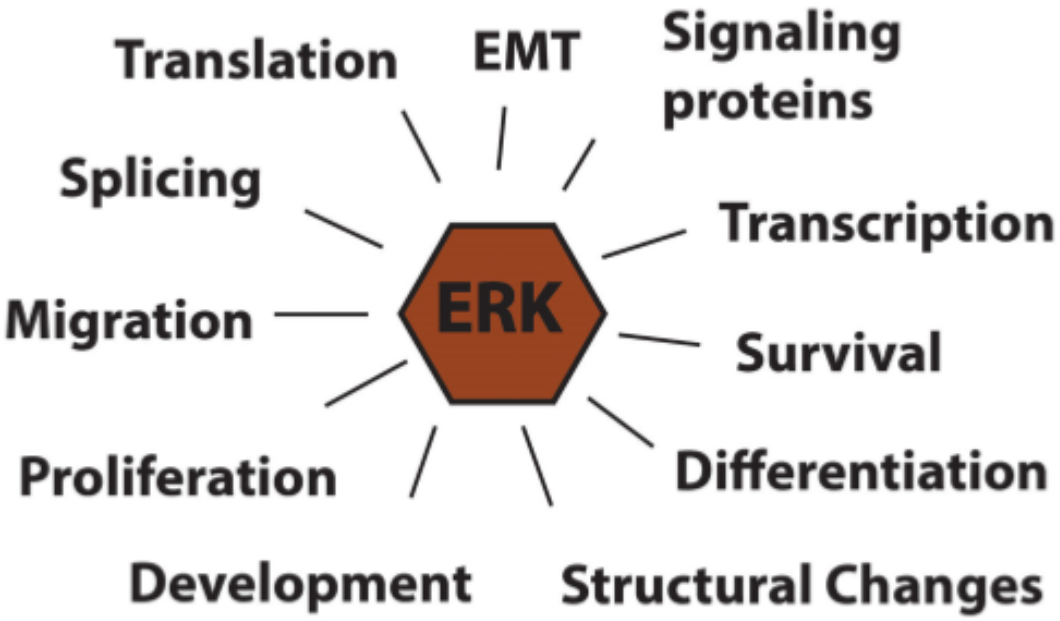
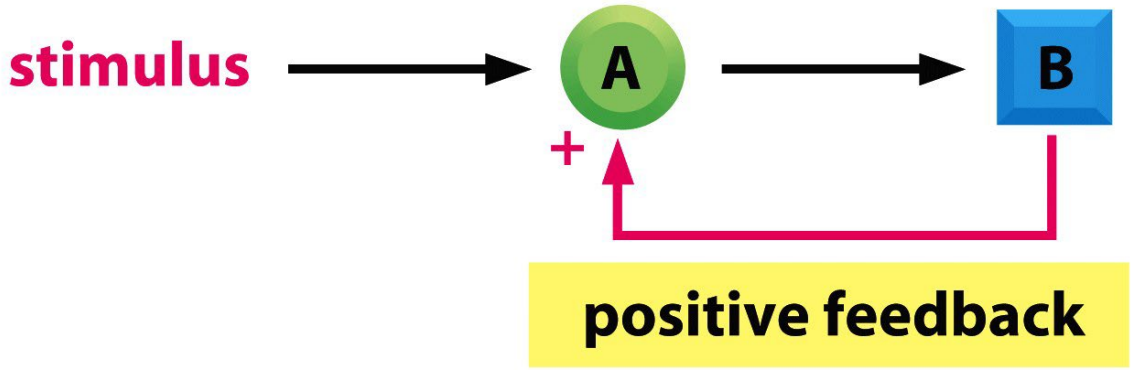


Figure 15-24bc Molecular Biology of the Cell 5/e (© Garland Science 2008)

ERK activation



Feedback



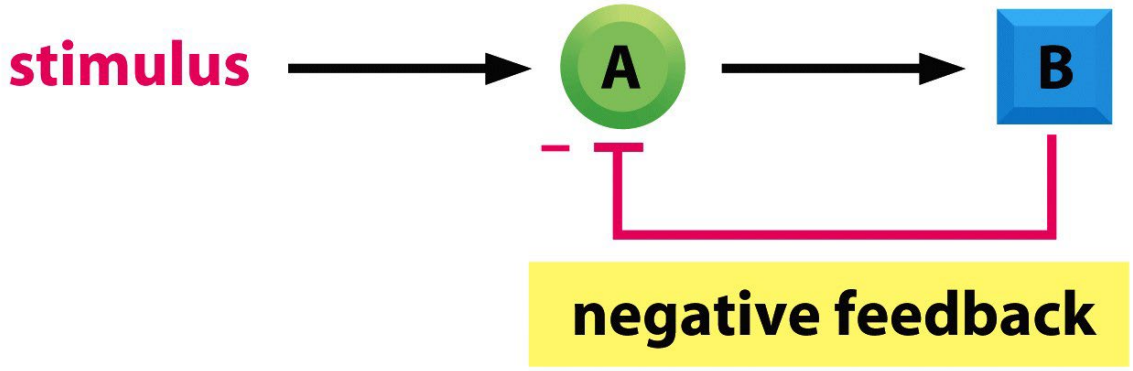
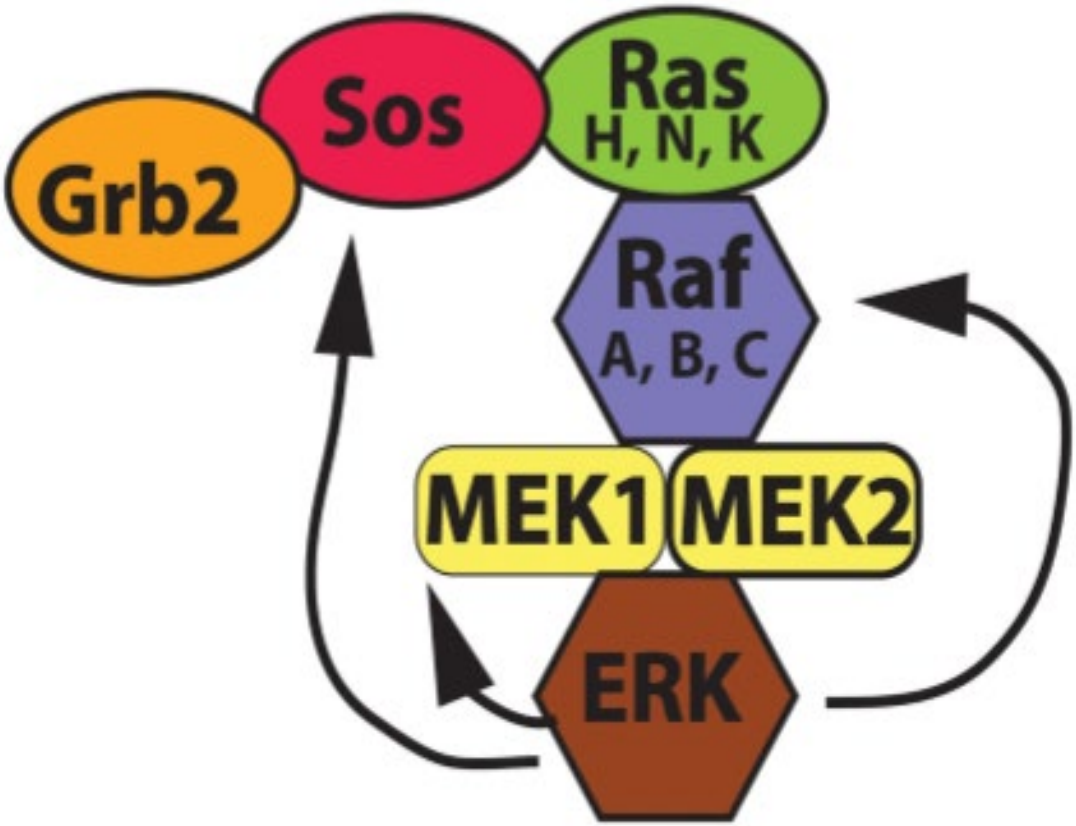
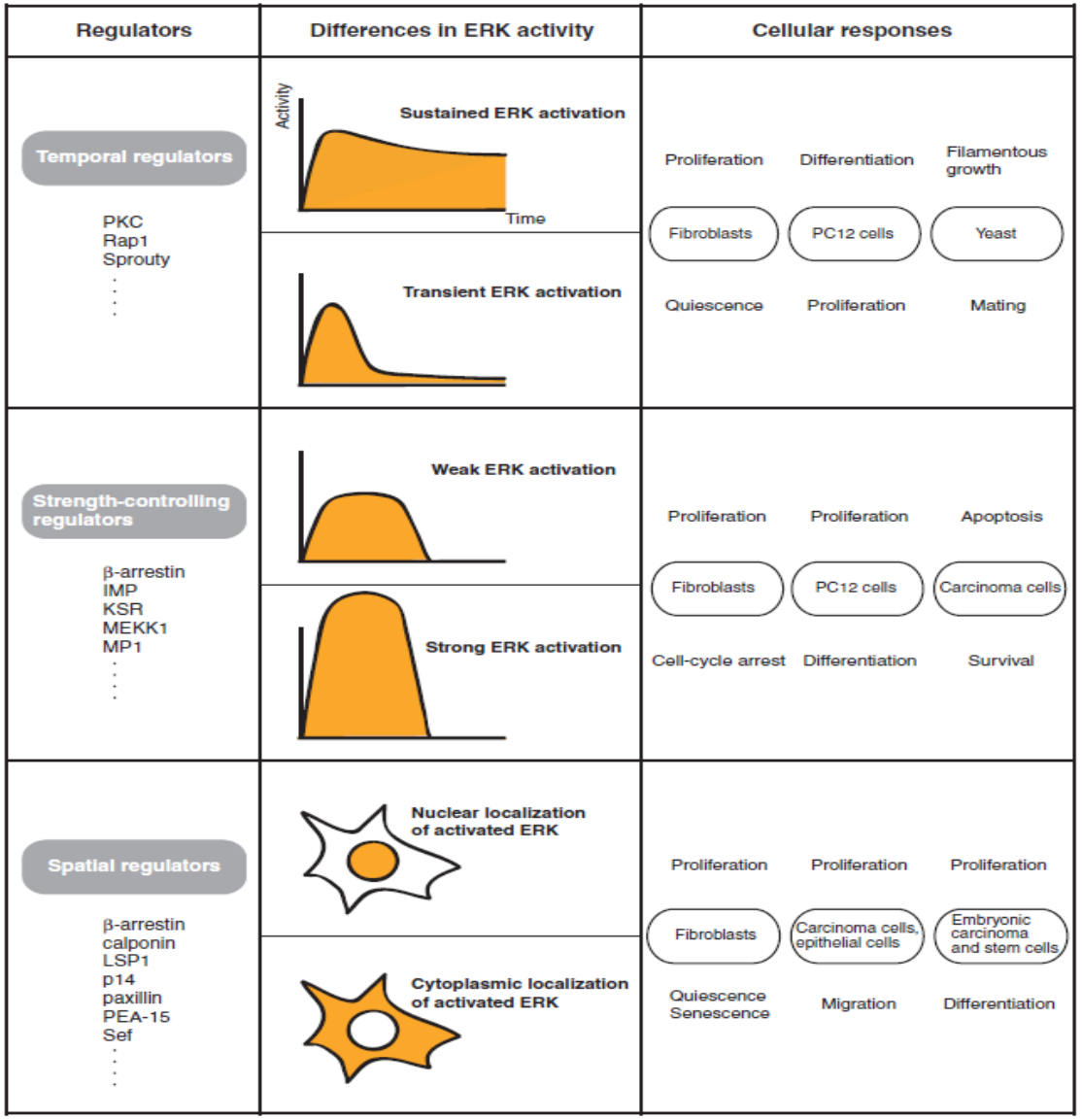


Figure 15-26 Molecular Biology of the Cell 5/e (© Garland Science 2008)

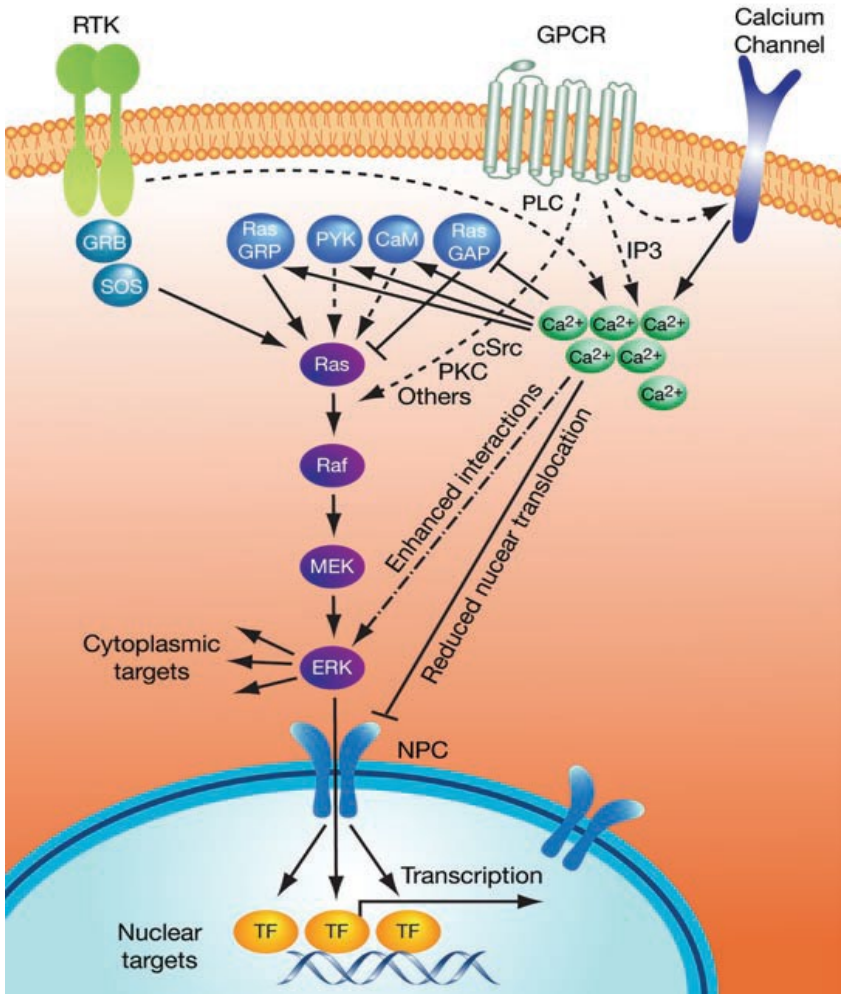
Feedback



Cell fate determination by the MAPK Pathway

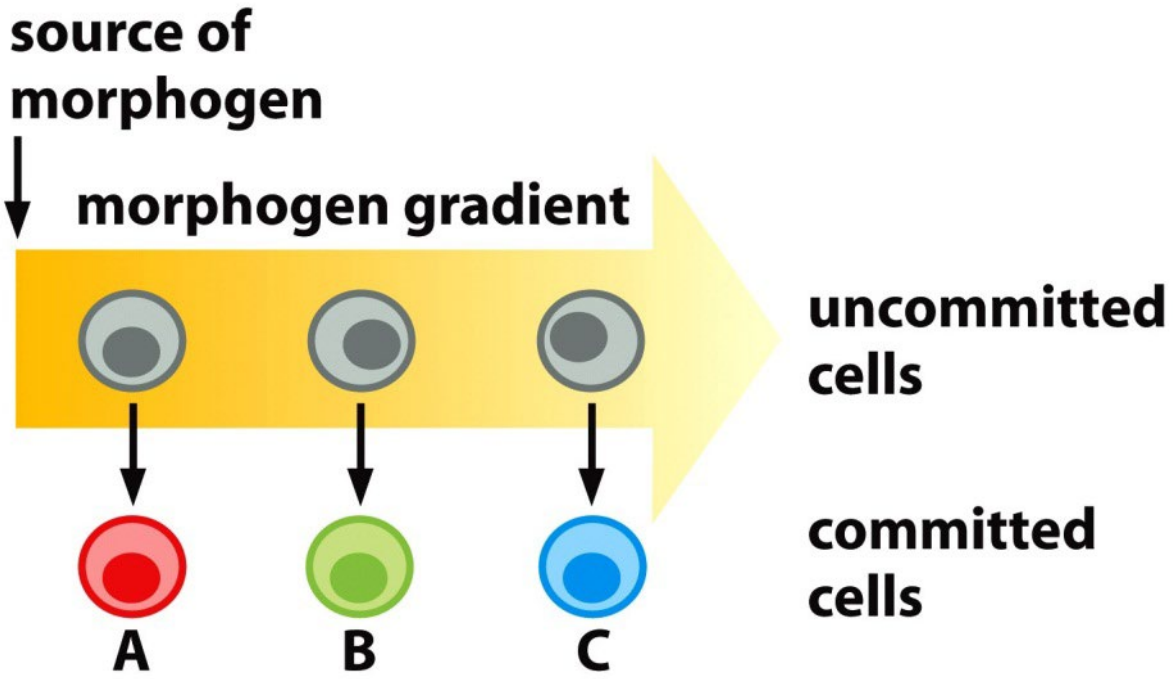


Cell fate determination by the MAPK Pathway



Elevated calcium concentrations induce cytoplasmic substrate phosphorylation (RSK), but not the nuclear substrate.

Diferentes concentrações do primeiro mensageiro podem induzir diferentes respostas

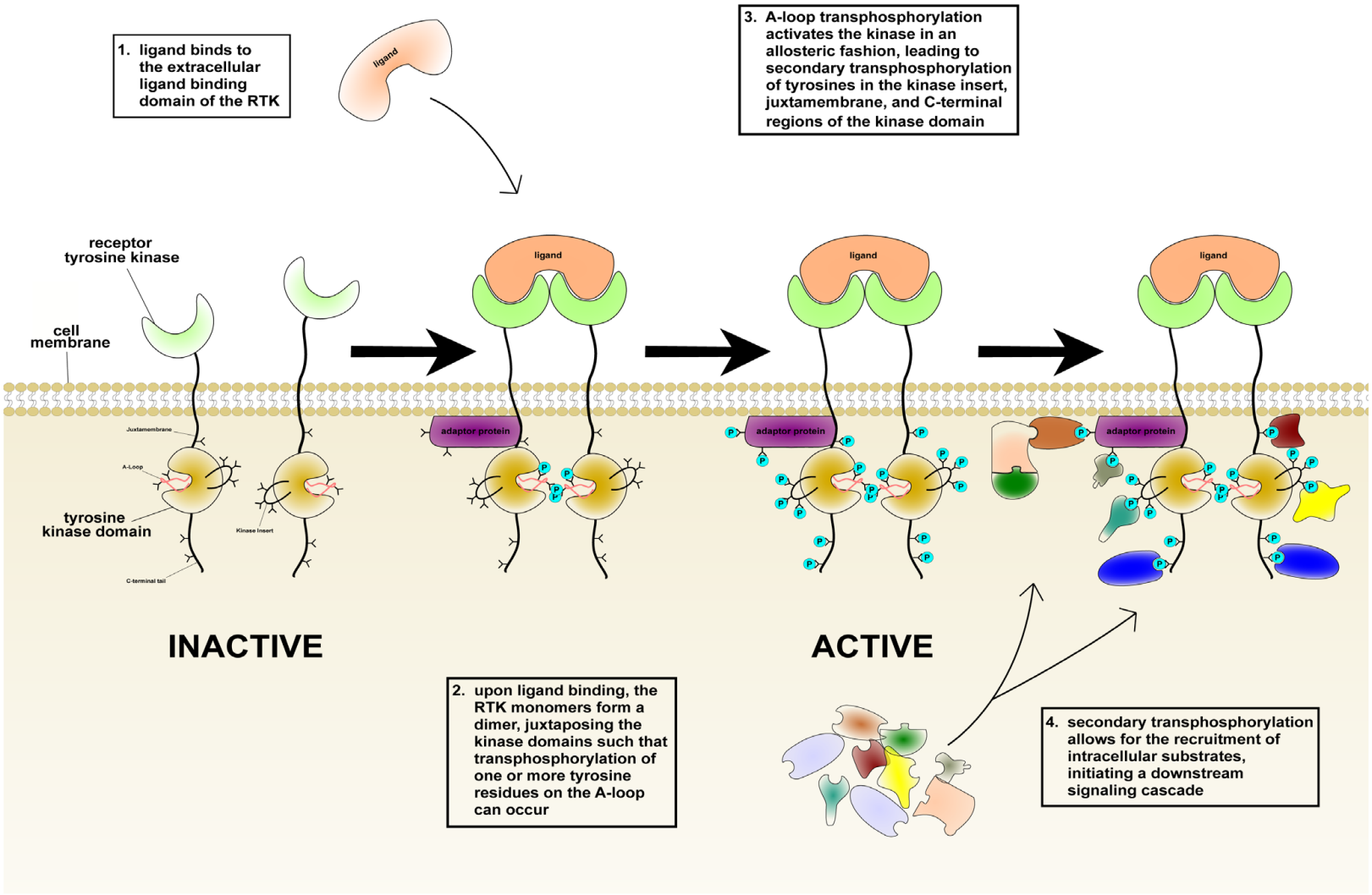


REVIEW

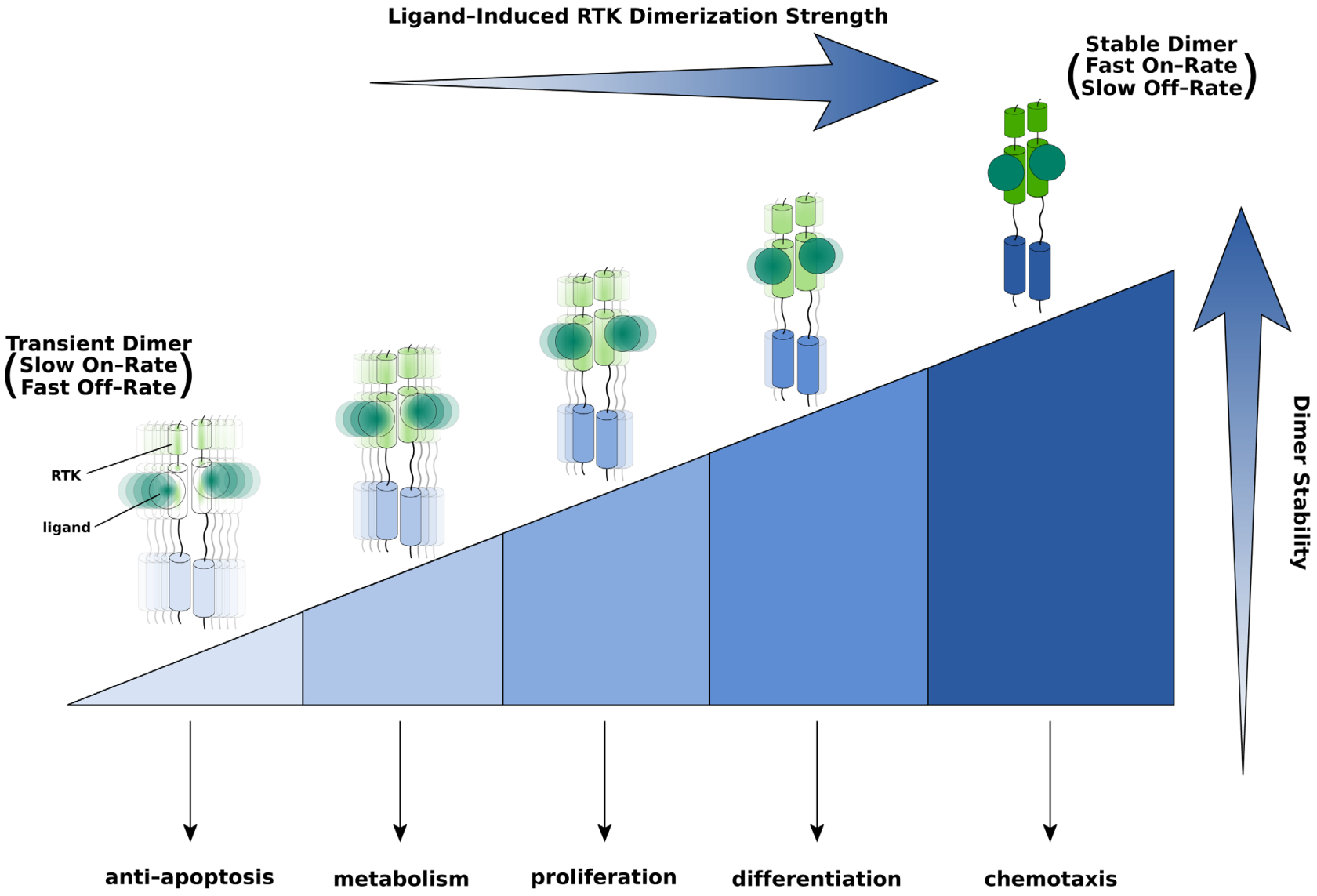
A threshold model for receptor tyrosine kinase signaling specificity and cell fate determination [version 1; referees: 4 approved]

Allen Zinkle, Moosa Mohammadi
Department of Biochemistry & Molecular Pharmacology, New York University School of Medicine, New York, NY, USA

Dimerização de RTK



Estabilidade do dímero altera a resposta



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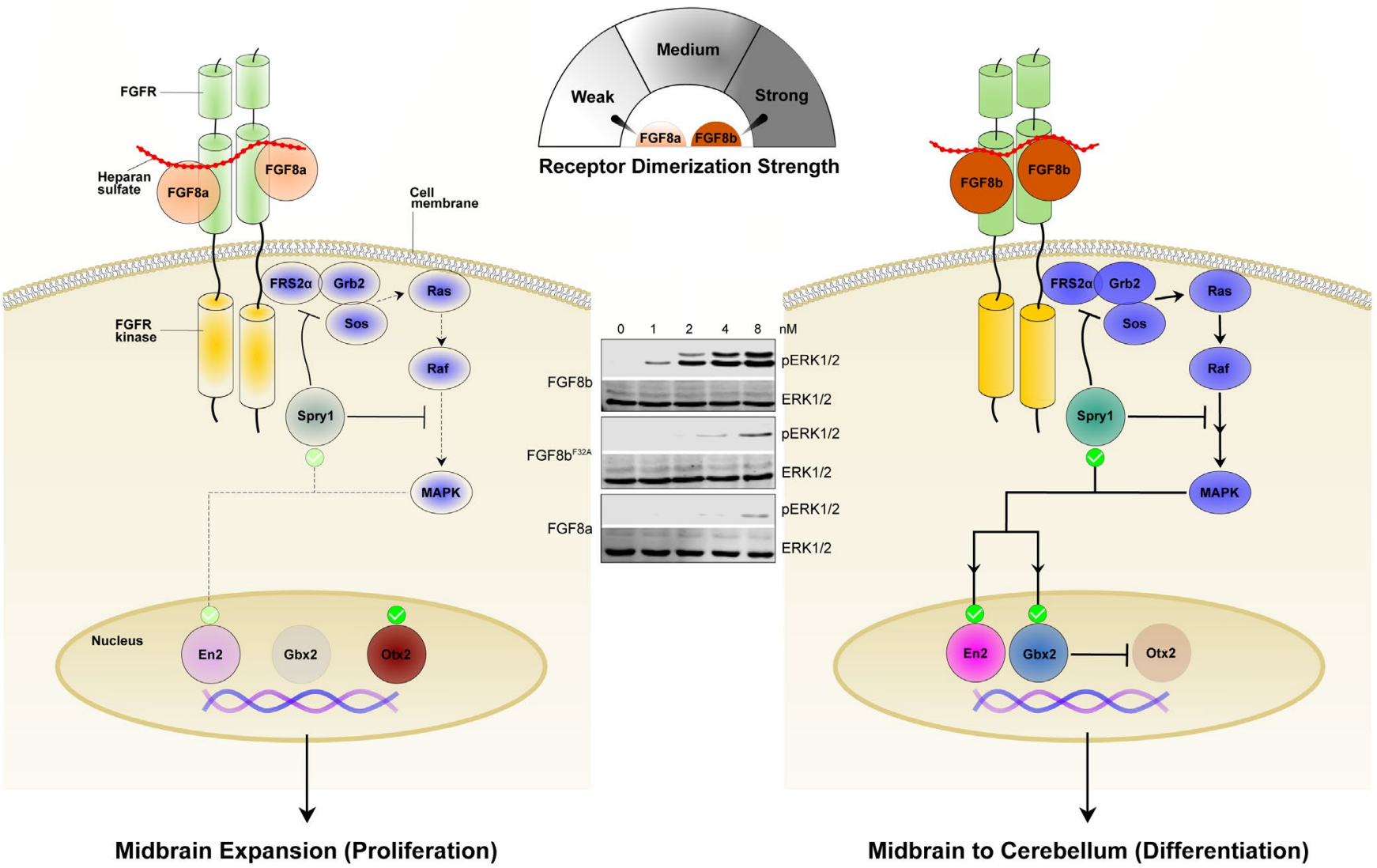
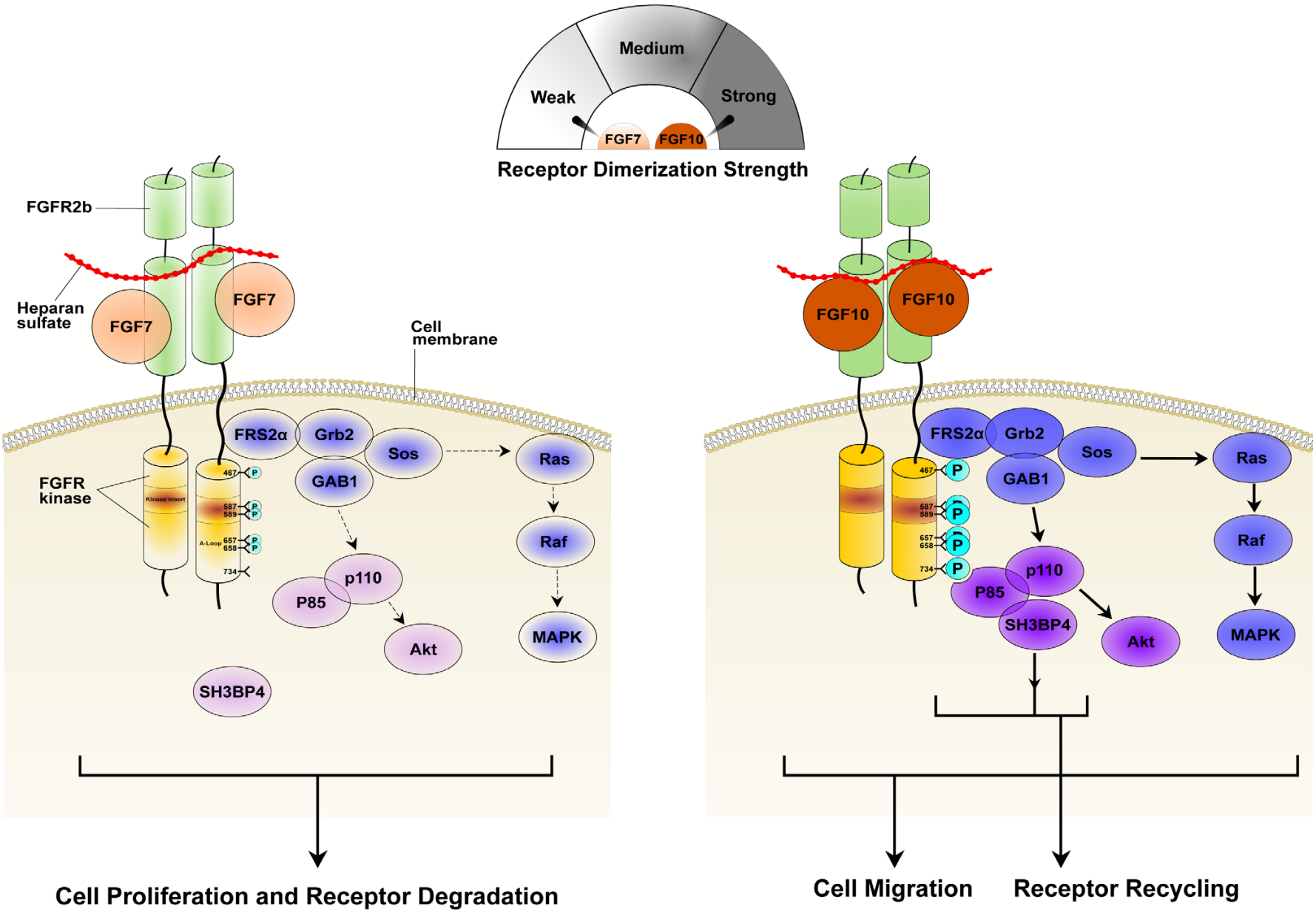
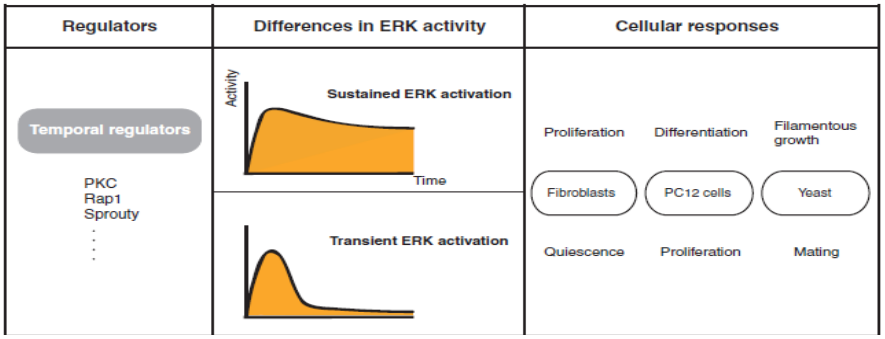


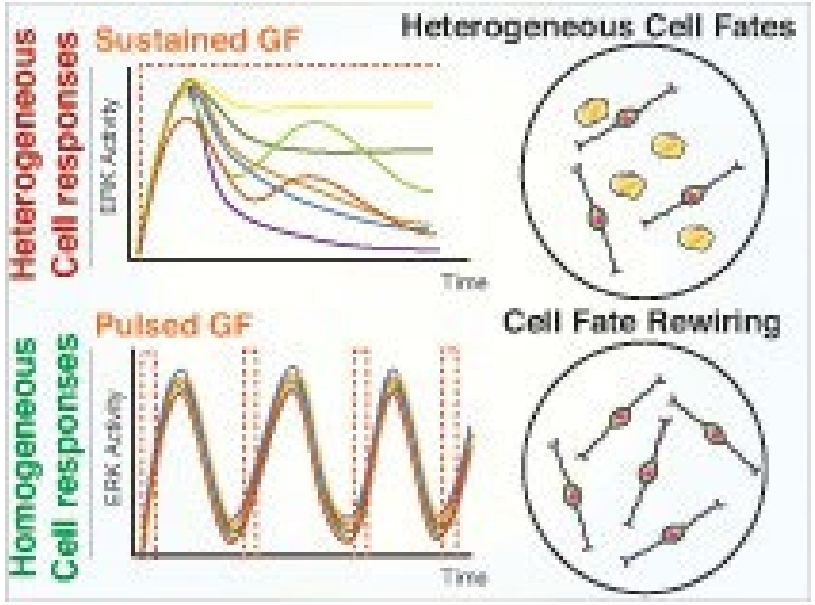
Estabilidade do dímero altera a resposta

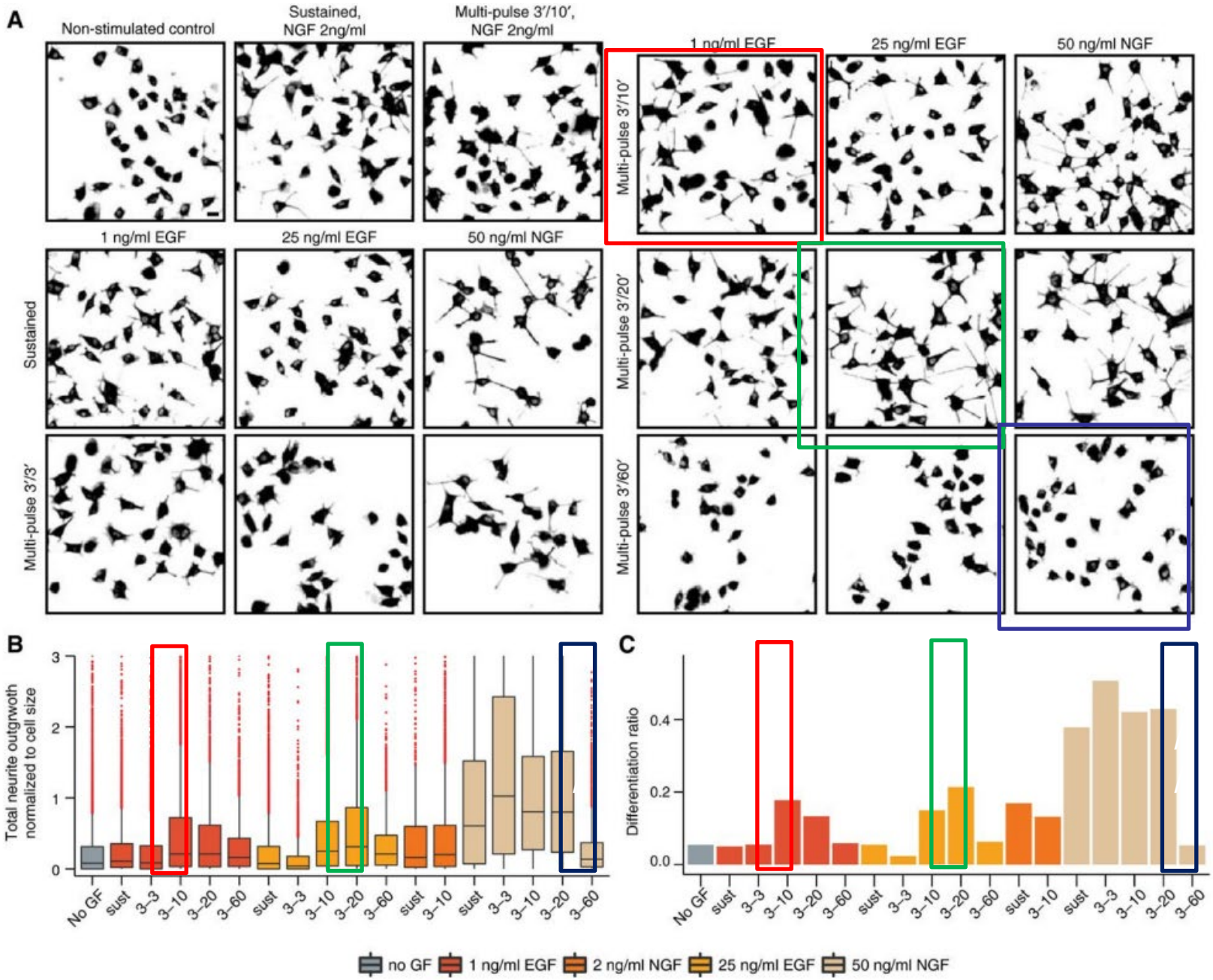


Frequency modulation of ERK activation dynamics rewires cell fate

Hyunryul Ryu[1,2], Minhwan Chung[1], Maciej Dobrzyński[3], Dirk Fey[3], Yannick Blum[4], Sung Sik Lee[5], Matthias Peter[5], Boris N Kholodenko[3,*], Noo Li Jeon[1,2,**] & Olivier Pertz[4,†,***]







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