Programmed Cell Death
- Apoptosis -

iGEM seminar

Mon, 2009-05-18

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Introduction – Programmed Cell Death

- Type I cell death = apoptosis
- Type II cell death = autophagy

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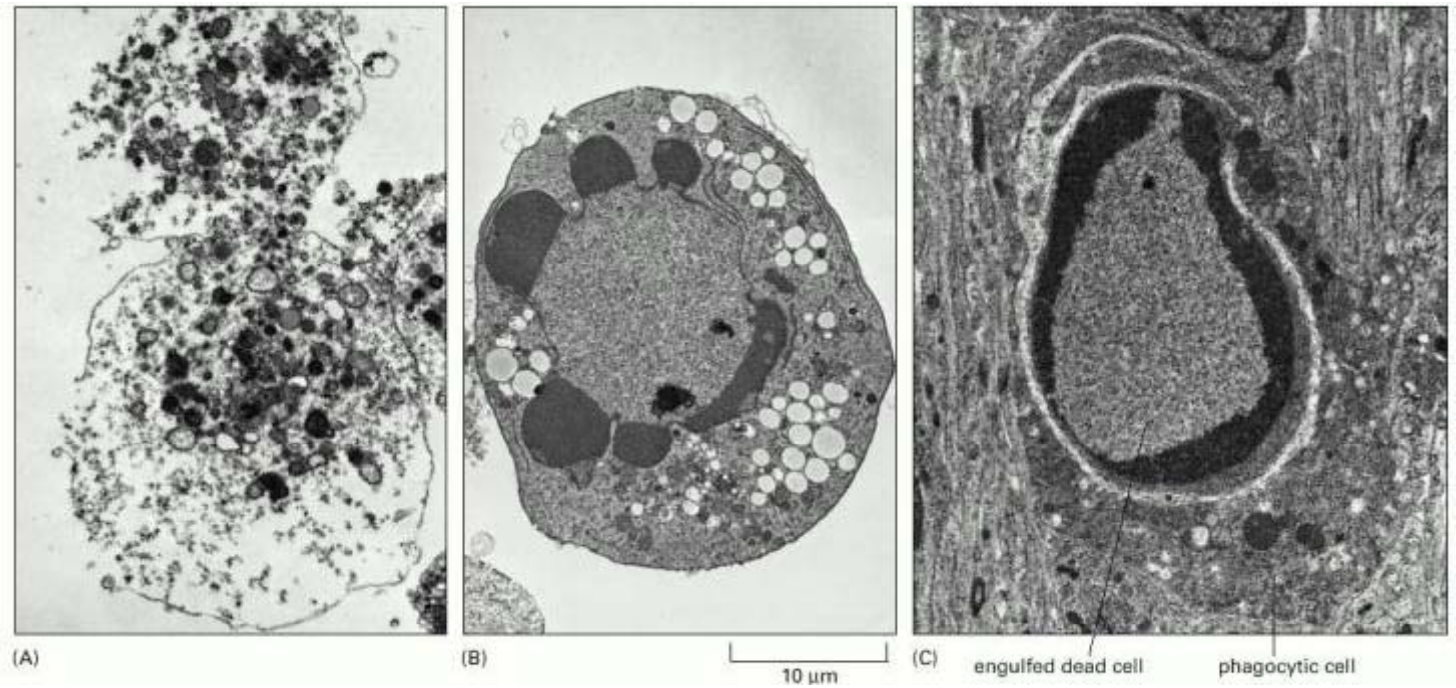
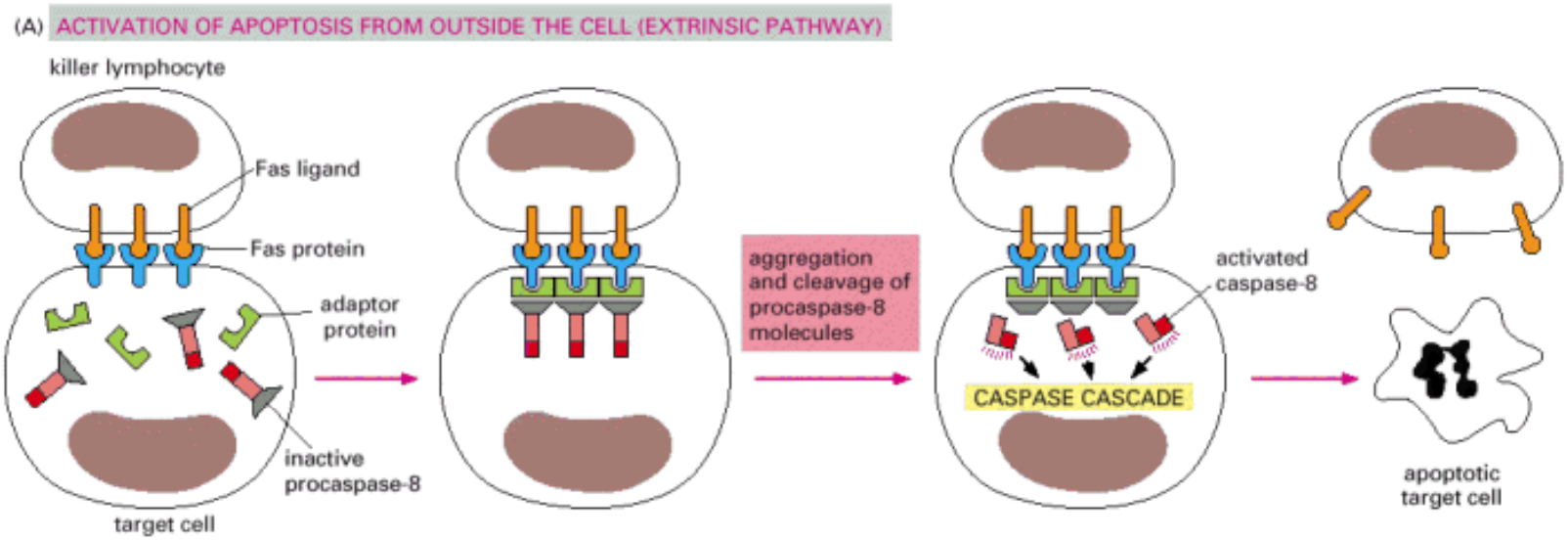


Introduction - Apoptosis

- Biochemical visualization
  - TUNEL assay
  - Annexin V staining – phosphatidylserine binding
- Important in
  - maintenance of tissue homeostasis
  - developmental processes
- Main mechanism: proteolytic caspase cascade
- 2 types: extrinsic vs intrinsic

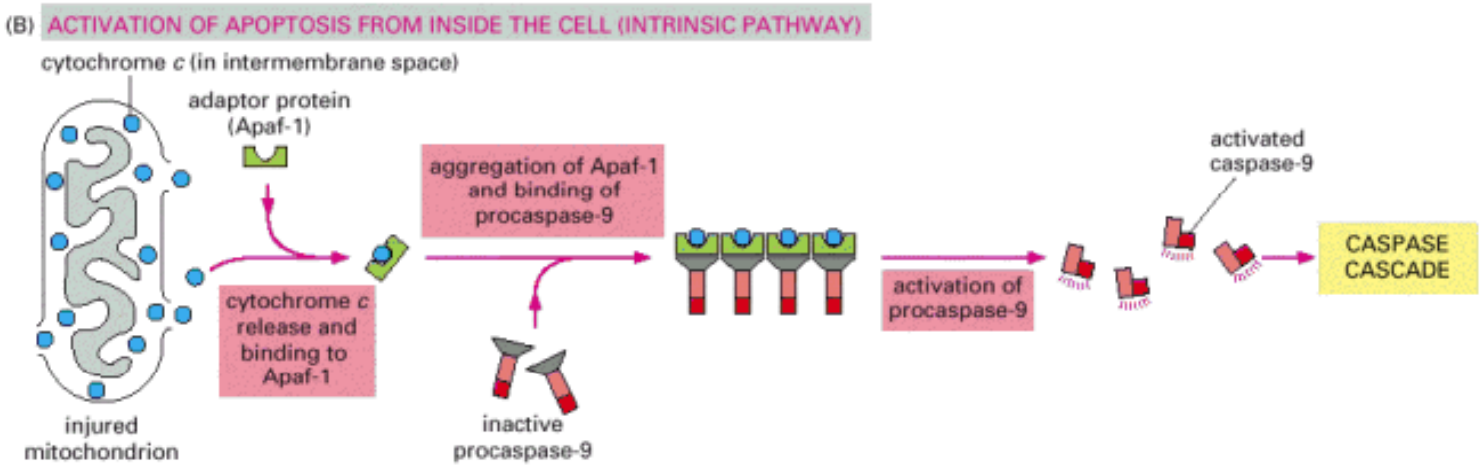
Extrinsic vs. intrinsic pathways



- Death receptors, Death Domains
  - TNF-alphaR, CD95
- DISC (death inducing signalling complexes)

Extrinsic vs. intrinsic pathways

- Cytochrome c release
- Apaf1
- Apoptosome



Intrinsic = transcription mediated

- Bcl2 family
- BH proteins (bcl-homology)

- IAP (inhibitors of apoptosis)
  - IIAP
  - Survivin
  - [illegible]CE, NAIP

blo

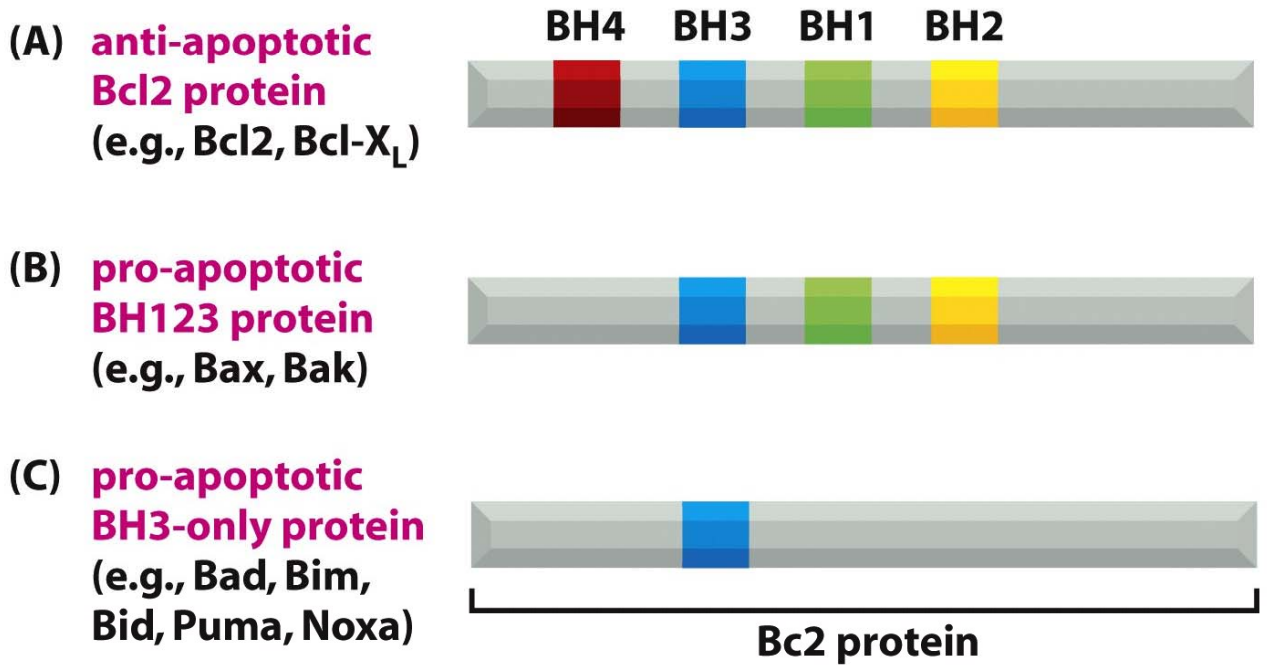


Figure 18-9 Molecular Biology of the Cell 5/e (© Garland Science 2008)

Intrinsic = transcription mediated

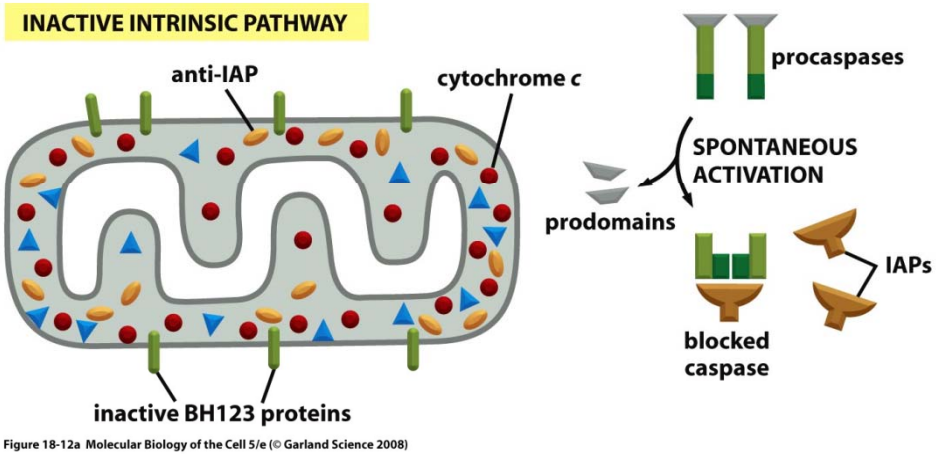


Figure 18-12a Molecular Biology of the Cell 5/e (© Garland Science 2008)

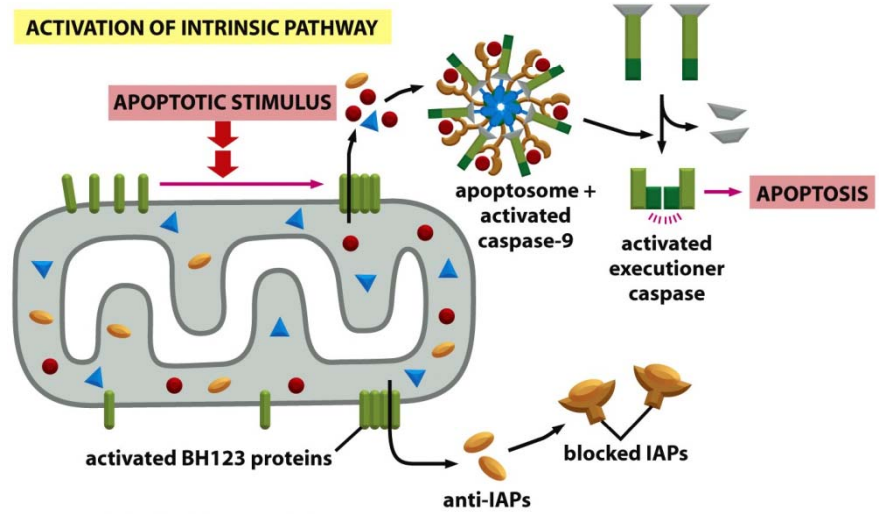
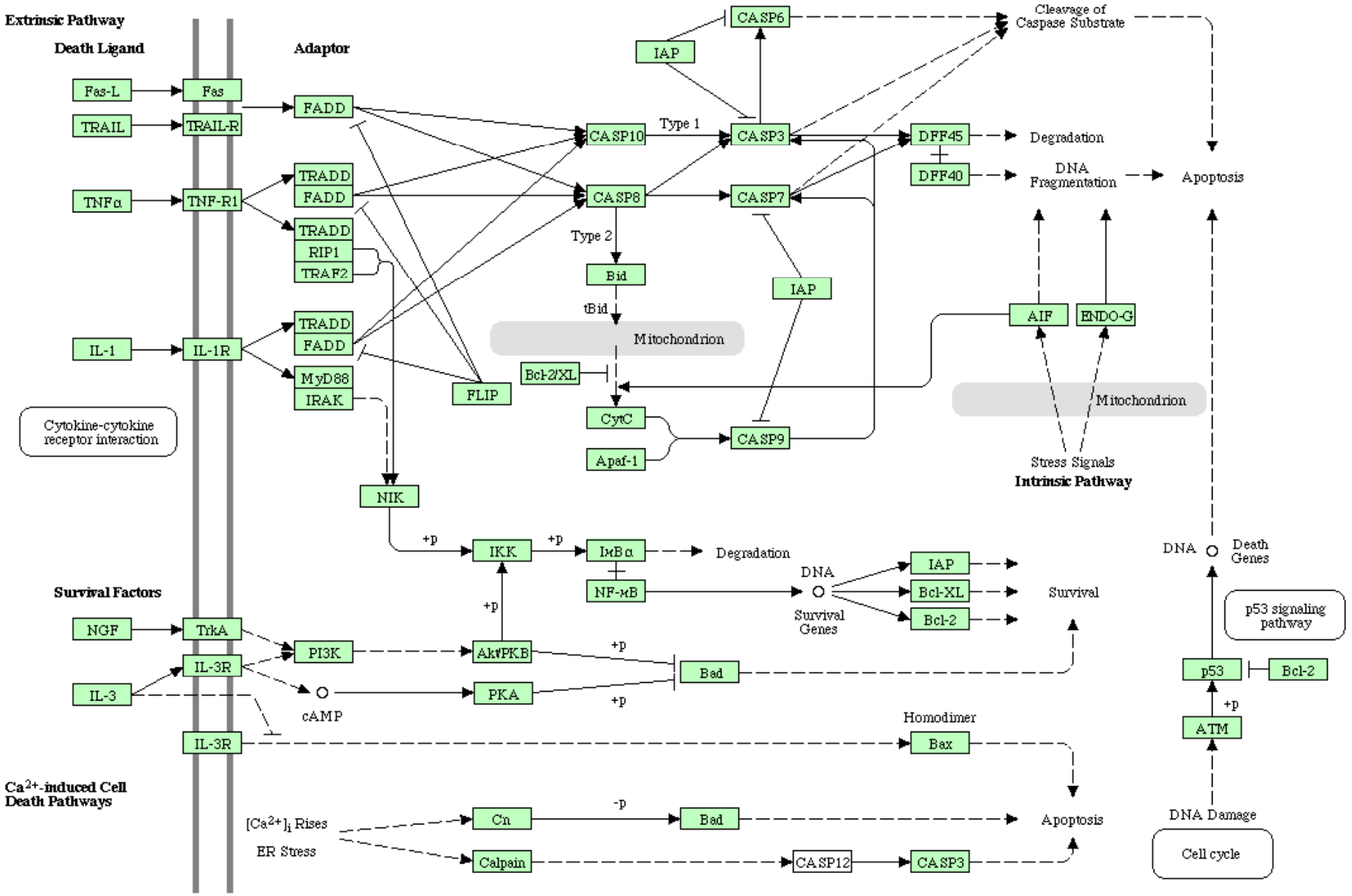


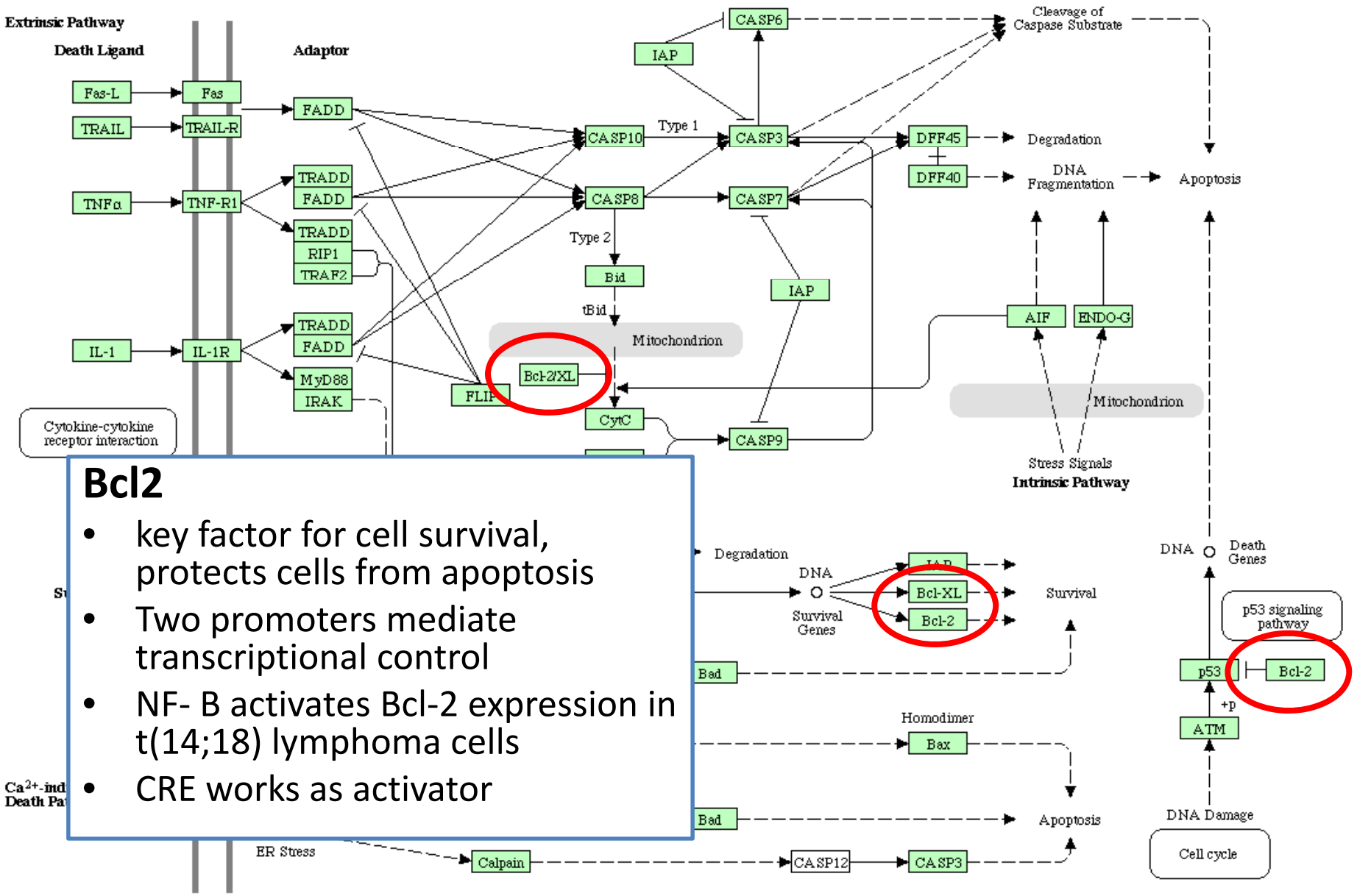
Figure 18-12b Molecular Biology of the Cell 5/e (© Garland Science 2008)

- IAP (inhibitors of apoptosis)
  - IIAP
  - Survivin
  - ILP2, BRUCE, NAIP
- Anti-IAP
  - Smac/Diablo
  - Omi

Apoptosis network



Promotors / Plasmids found on Addgene



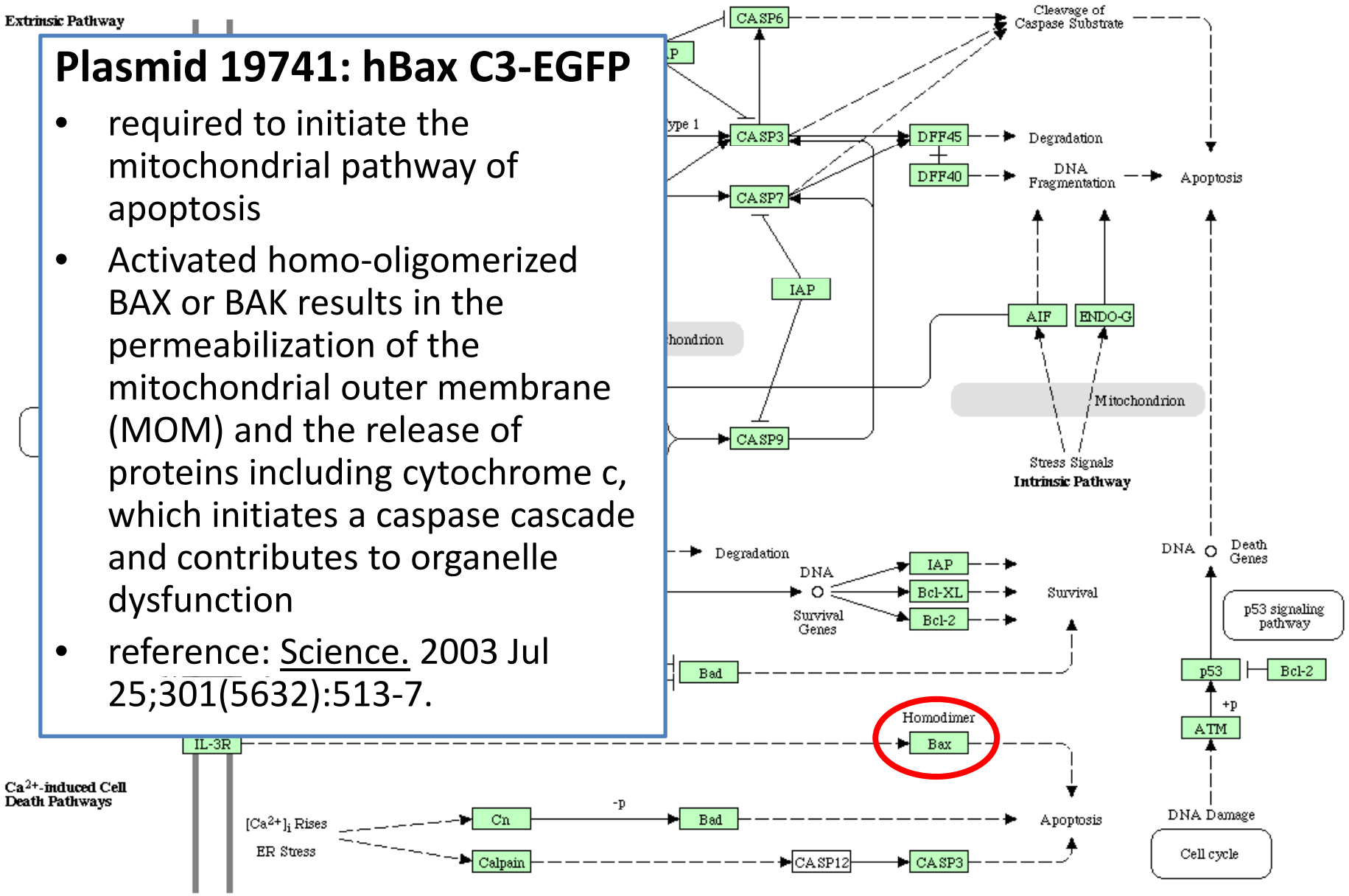
Bcl2

- key factor for cell survival, protects cells from apoptosis
- Two promoters mediate transcriptional control
- NF- B activates Bcl-2 expression in t(14;18) lymphoma cells
- CRE works as activator

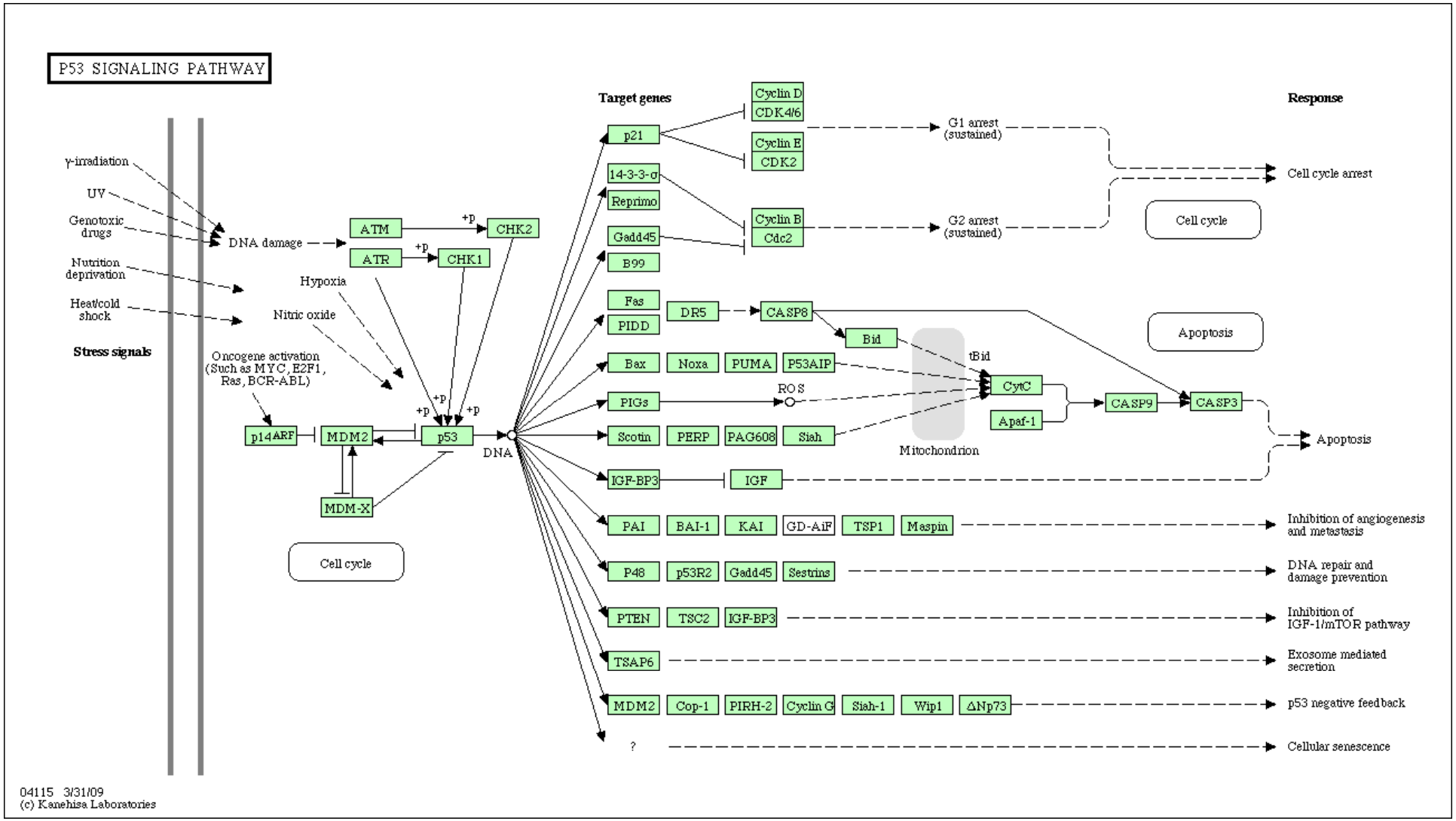
Promotors / Plasmids found on Addgene

Plasmid 19741: hBax C3-EGFP

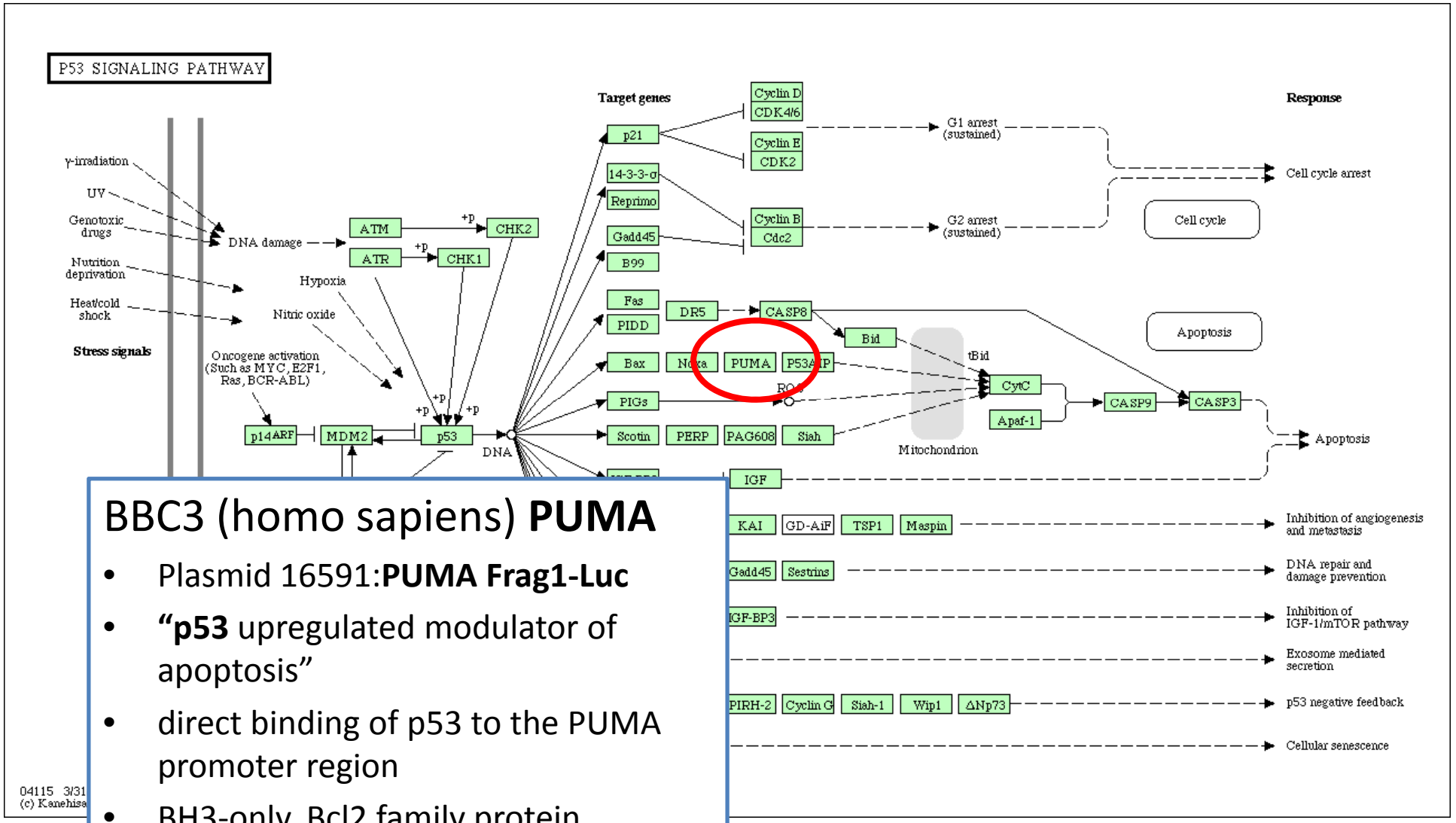
- required to initiate the mitochondrial pathway of apoptosis
- Activated homo-oligomerized BAX or BAK results in the permeabilization of the mitochondrial outer membrane (MOM) and the release of proteins including cytochrome c, which initiates a caspase cascade and contributes to organelle dysfunction
- reference: Science. 2003 Jul 25;301(5632):513-7.



p53 network


P53 SIGNALING PATHWAY
γ-irradiation
UV
Genotoxic drugs
Nutrition deprivation
Heat/cold shock
Stress signals
DNA damage
ATM
ATR
CHK2
CHK1
+p
Hypoxia
Nitric oxide
Oncogene activation (Such as MYC, E2F1, Ras, BCR-ABL)
p14ARF
MDM2
p53
MDM-X
DNA
Cell cycle
Target genes
Cyclin D
CDK4/6
Cyclin E
CDK2
Cyclin B
Cdc2
p21
14-3-3-σ
Reprimo
Gadd45
B99
Fas
PIDD
DR5
CASP8
Bid
tBid
Bax
Noxa
PUMA
P53AIP
PIGs
ROS
Scotin
PERP
PAG608
Siah
CytC
Apaf-1
Mitochondrion
CASP9
CASP3
IGF-BP3
IGF
PAI
BAI-1
KAI
GD-AiF
TSP1
Maspin
P48
p53R2
Gadd45
Sestrins
PTEN
TSC2
IGF-BP3
TSAP6
MDM2
Cop-1
PIRH-2
Cyclin G
Siah-1
Wip1
ΔNp73
?
Response
G1 arrest (sustained)
G2 arrest (sustained)
Cell cycle arrest
Cell cycle
Apoptosis
Apoptosis
Inhibition of angiogenesis and metastasis
DNA repair and damage prevention
Inhibition of IGF-1/mTOR pathway
Exosome mediated secretion
p53 negative feedback
Cellular senescence
04115 3/31/09
(c) Kanehisa Laboratories

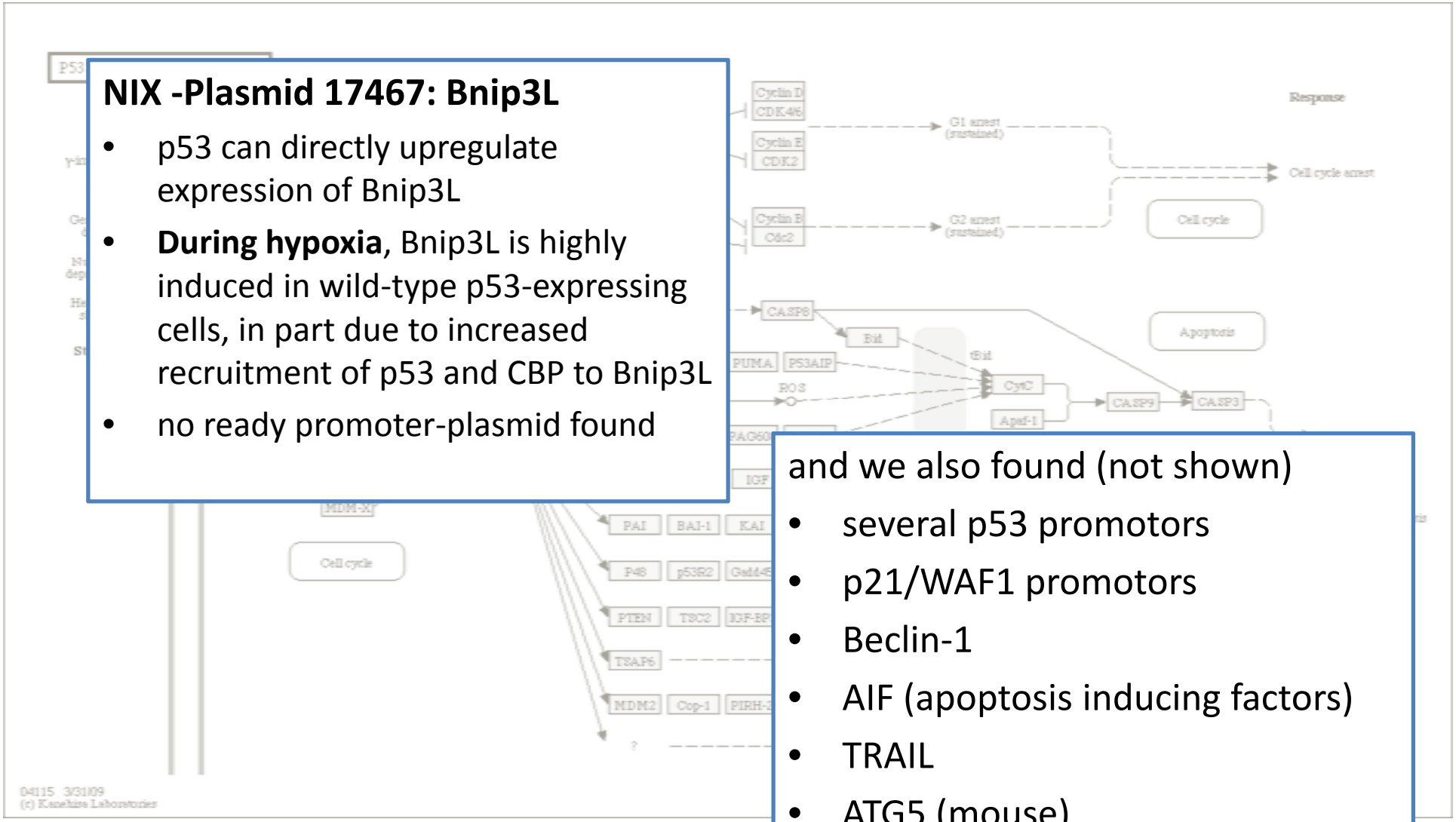
Promotors / Plasmids found on Addgene



BBC3 (homo sapiens) **PUMA**

- Plasmid 16591:**PUMA Frag1-Luc**
- **"p53** upregulated modulator of apoptosis"
- direct binding of p53 to the PUMA promoter region
- BH3-only, Bcl2 family protein

Promotors / Plasmids found on Addgene



NIX -Plasmid 17467: Bnip3L

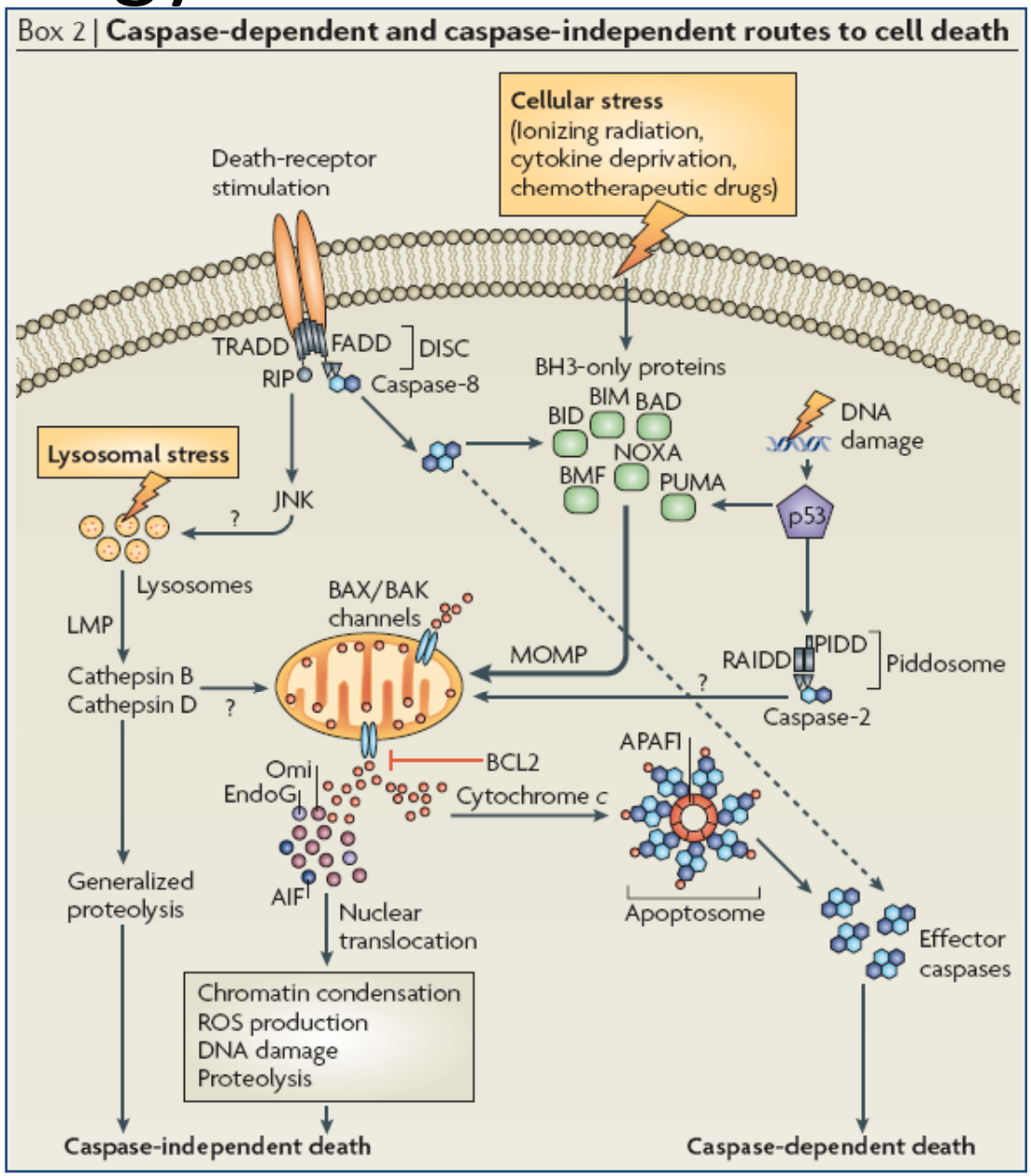
- p53 can directly upregulate expression of Bnip3L
- **During hypoxia**, Bnip3L is highly induced in wild-type p53-expressing cells, in part due to increased recruitment of p53 and CBP to Bnip3L
- no ready promoter-plasmid found

and we also found (not shown)

- several p53 promotors
- p21/WAF1 promotors
- Beclin-1
- AIF (apoptosis inducing factors)
- TRAIL
- ATG5 (mouse)

Crosstalk to autophagy

- Many crosstalks
- Molecular switches



Crosstalk to autophagy

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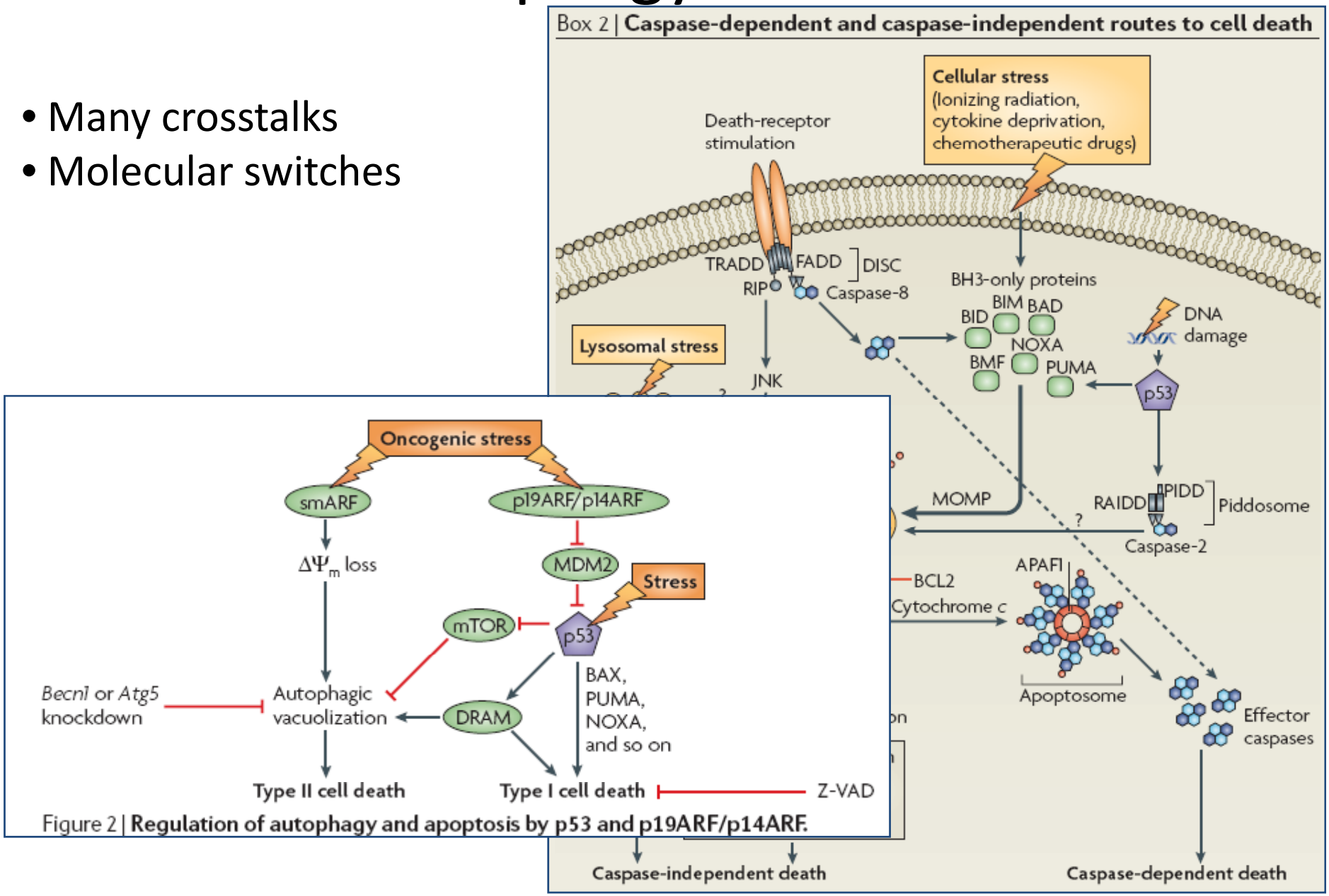
- Molecular switches



Molecular switches

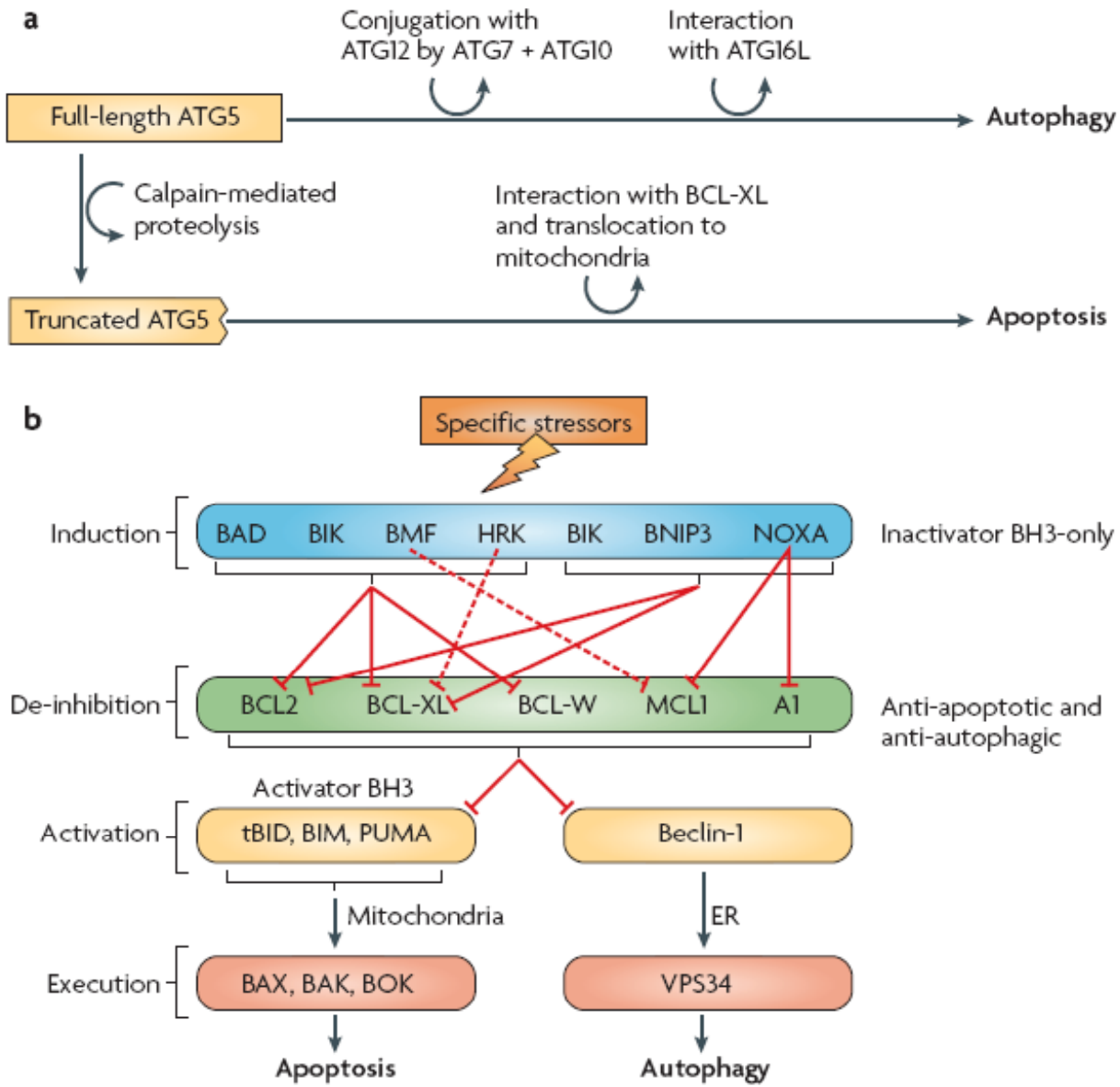


Figure 4 | **Molecular switches between apoptosis and autophagy. a** | Dual function of

Molecular switches

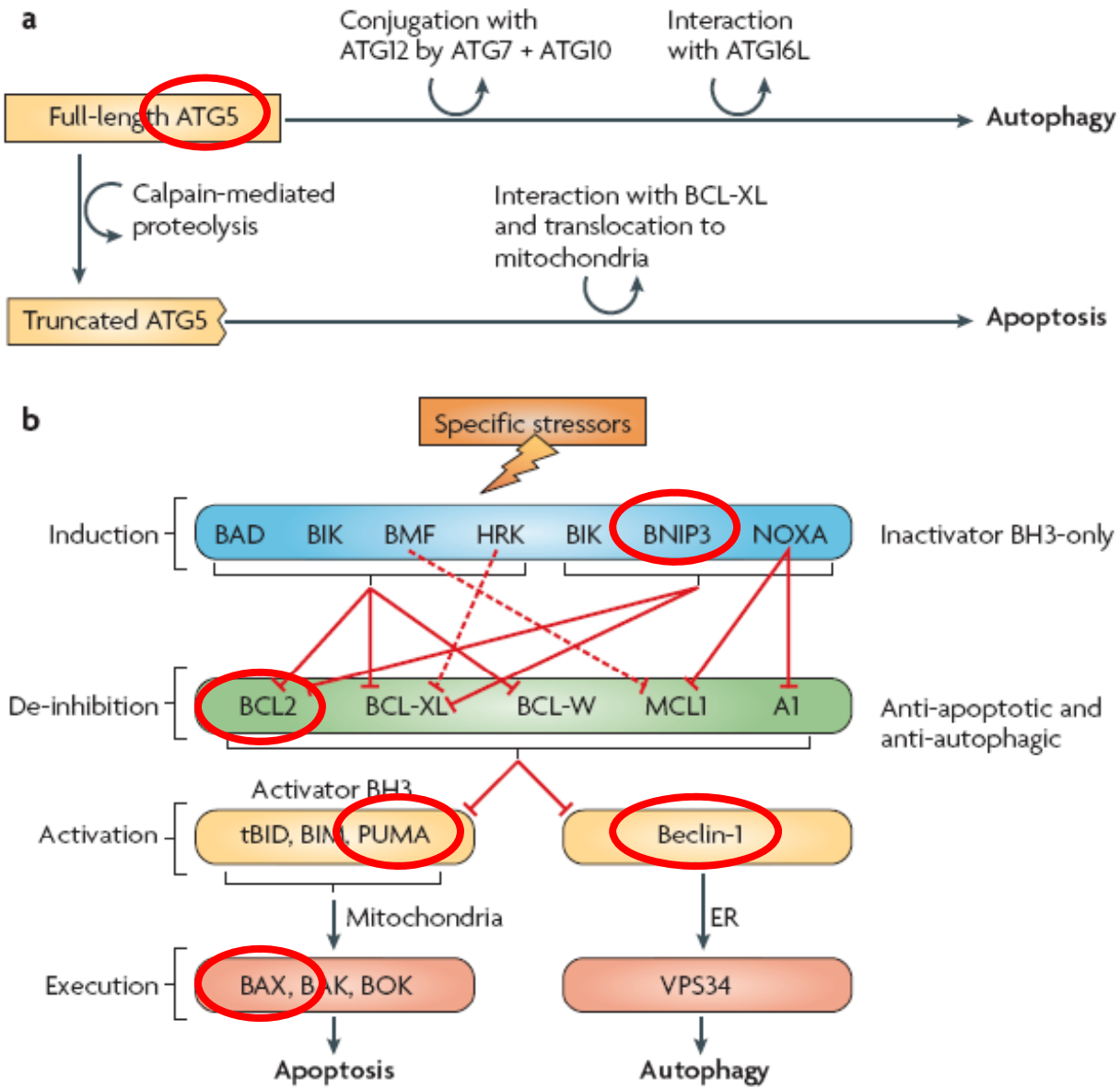


Figure 4 | **Molecular switches between apoptosis and autophagy. a** | Dual function of