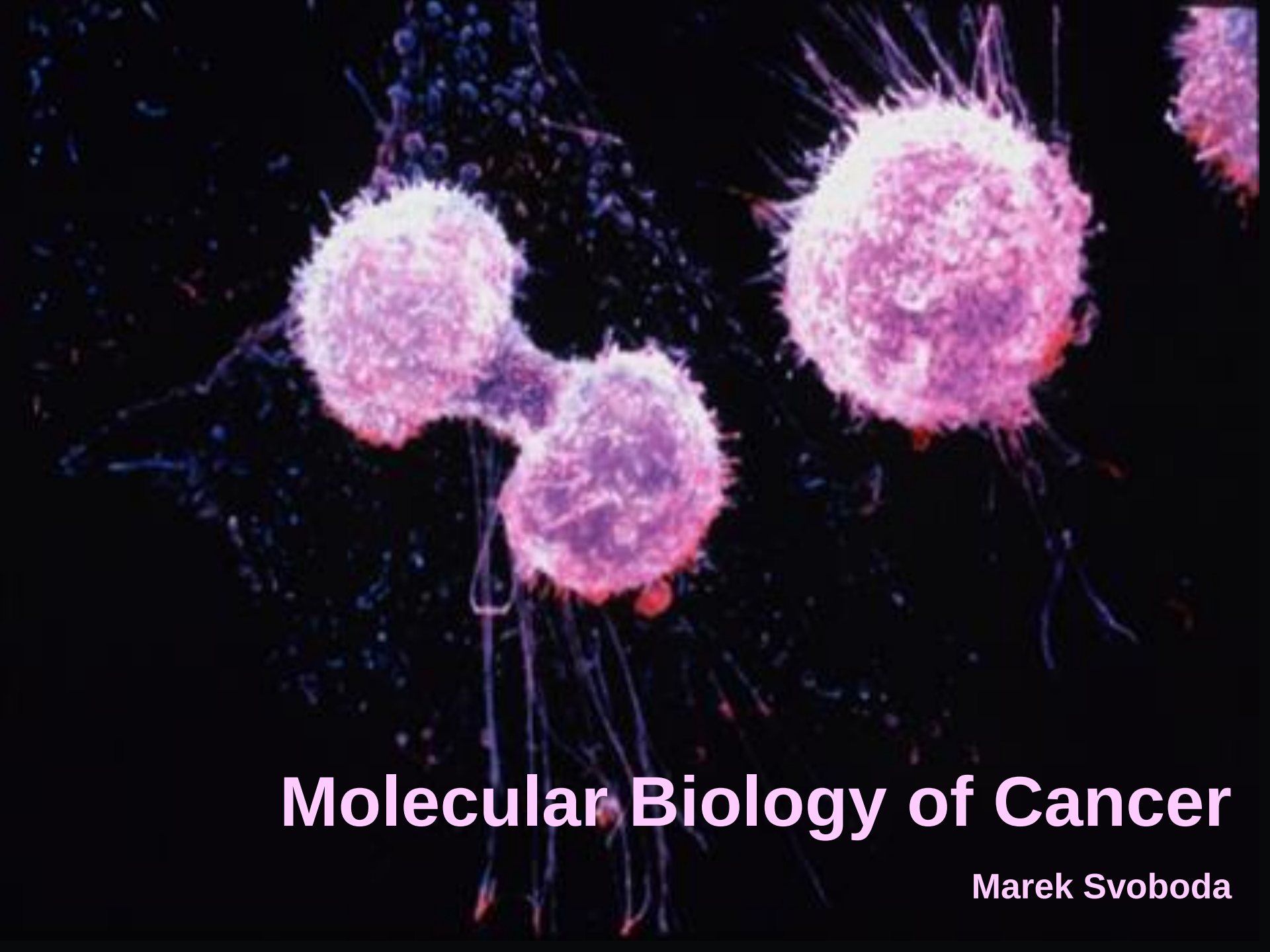
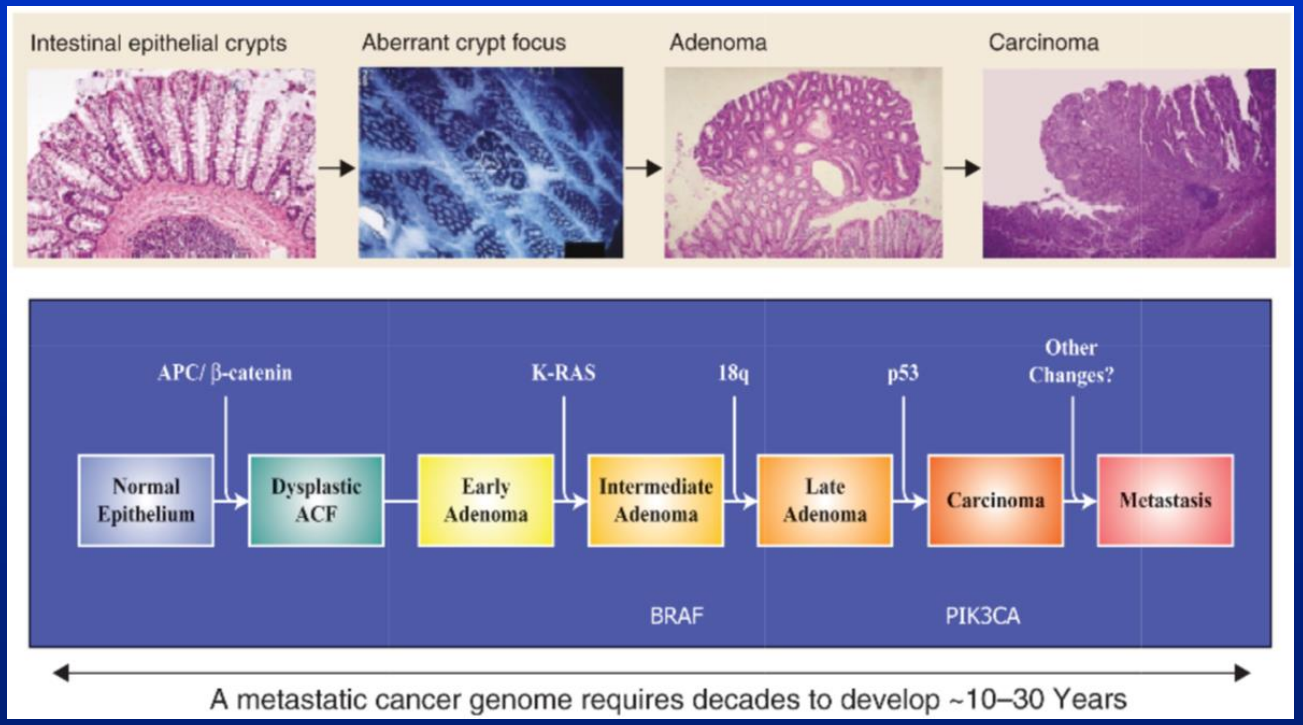


Molecular Biology of Cancer

Marek Svoboda

Molecular Biology of Cancer

- **There is a broad consensus that cancer is, in essence, a genetic disease**
- The development of fully malignant cancers requires accumulation of molecular alterations in the genome of somatic cells
  → **Multistep tumorigenesis**



Molecular Biology of Cancer

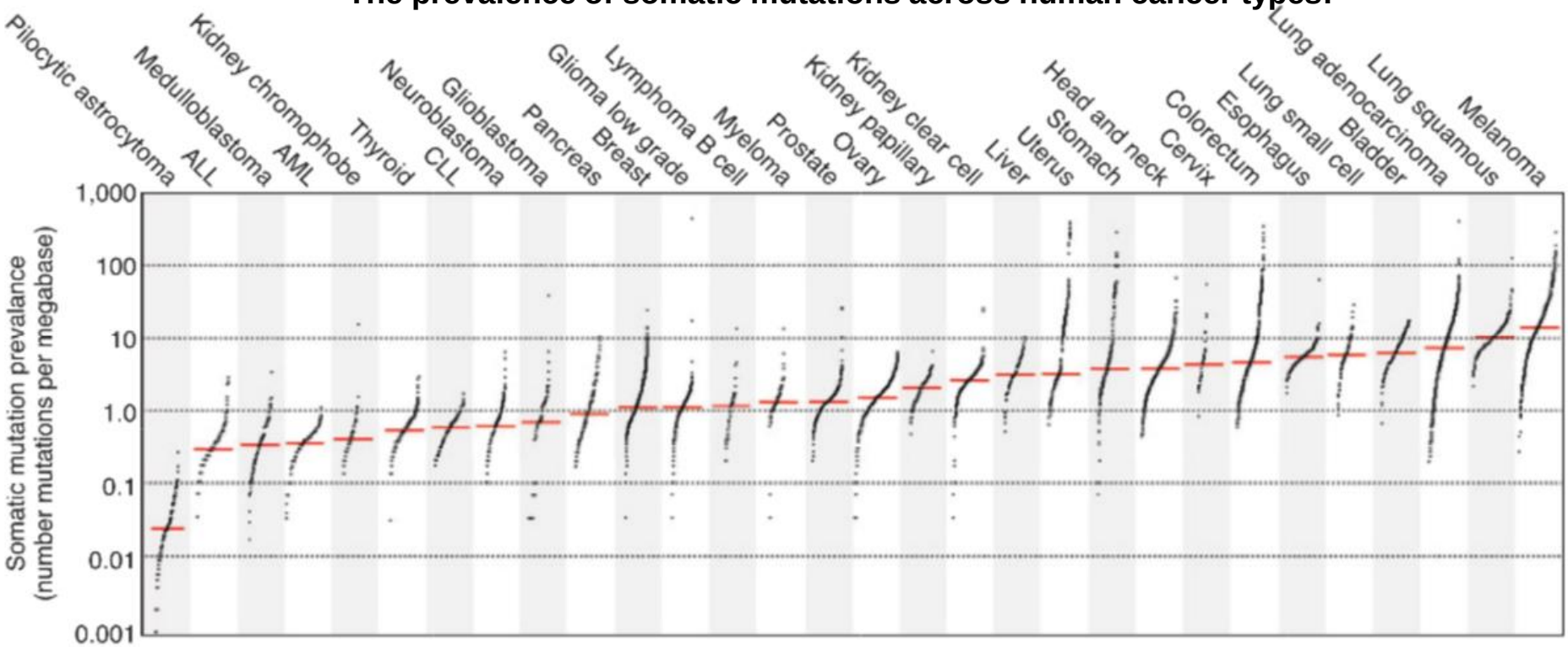
- **Multistep tumorigenesis → How many and what kind of genes are affected?**
  - Human genome contains approximately **$3x10^9$ nucleotides (3 000 Mb)** in 23 chromosomes → **~ 25 000 genes** → ~ 180 000 exomes (= ~ **1 % of genome = 30 Mb in coding regions**)

- **The variation in mutational frequency between different tumors is extraordinary:**
  - **Cancer genomes have an average of 30 to 100 somatic alterations per tumor (= 1-3 per Mb in coding regions)**

Molecular Biology of Cancer

- The variation in mutational frequency between different tumors is extraordinary:
  - **the lowest mutation rates: pediatric and hematologic cancers (0,001 per Mb of DNA in coding regions), the highest mutation rates: melanoma, lung cancers (400 per Mb of DNA in coding regions)**

The prevalence of somatic mutations across human cancer types:



Molecular Biology of Cancer

- Although the specific mutations that cause human cancers vary greatly between types of cancers and individuals, **the broad consequences of these mutations are abnormal phenotypes that are shared by most cancers → „Hallmarks of cancer“**
  - Sustaining proliferative signaling
    Soběstačnost v produkci růstových faktorů
  - Evading growth suppresors
    Necitlivost k supresorům růstu
  - Resisting cell death
    Poškození apoptózy
  - Enabling replicative immortality
    Neomezený replikační potenciál
  - Inducing angiogenesis
    Indukce angiogeneze
  - Activating invasion and metastasis
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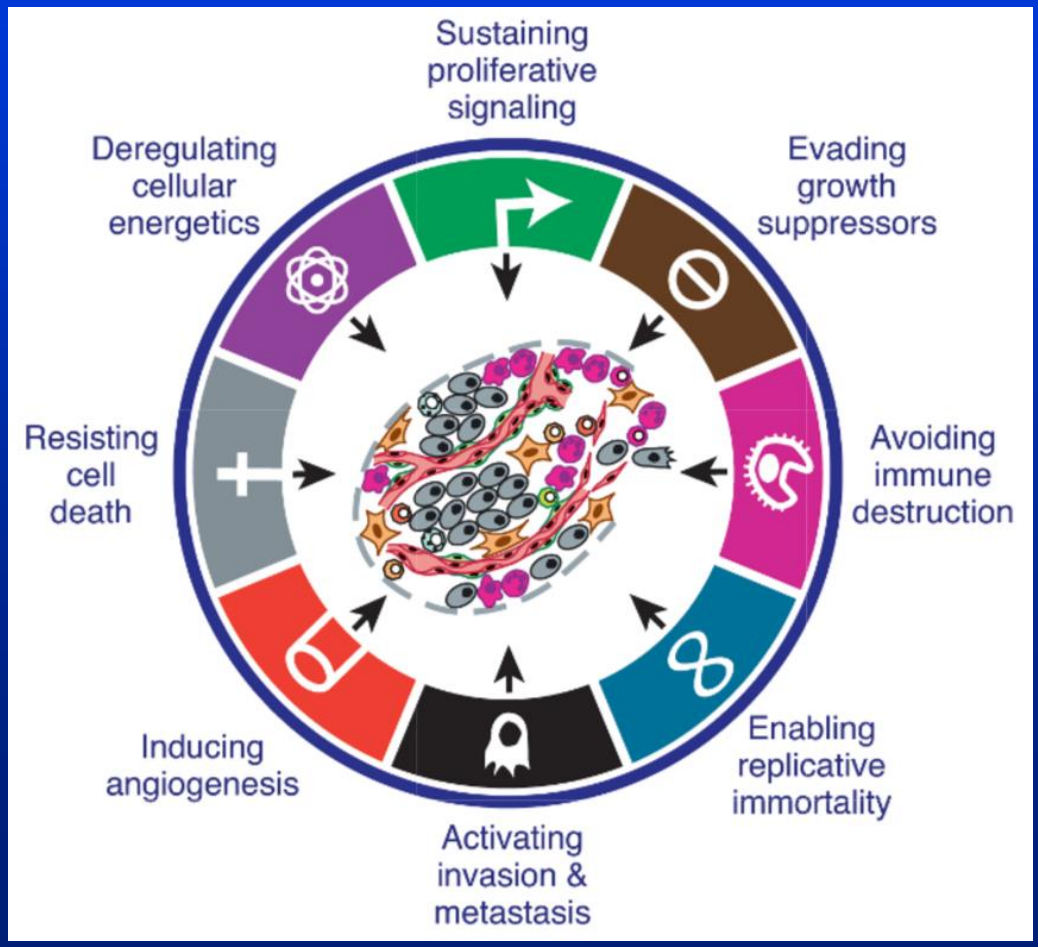
  - Reprogramming energy metabolism
    Změna energetického metabolismu
  - Evading immune destruction
    Únik před imunitním systémem

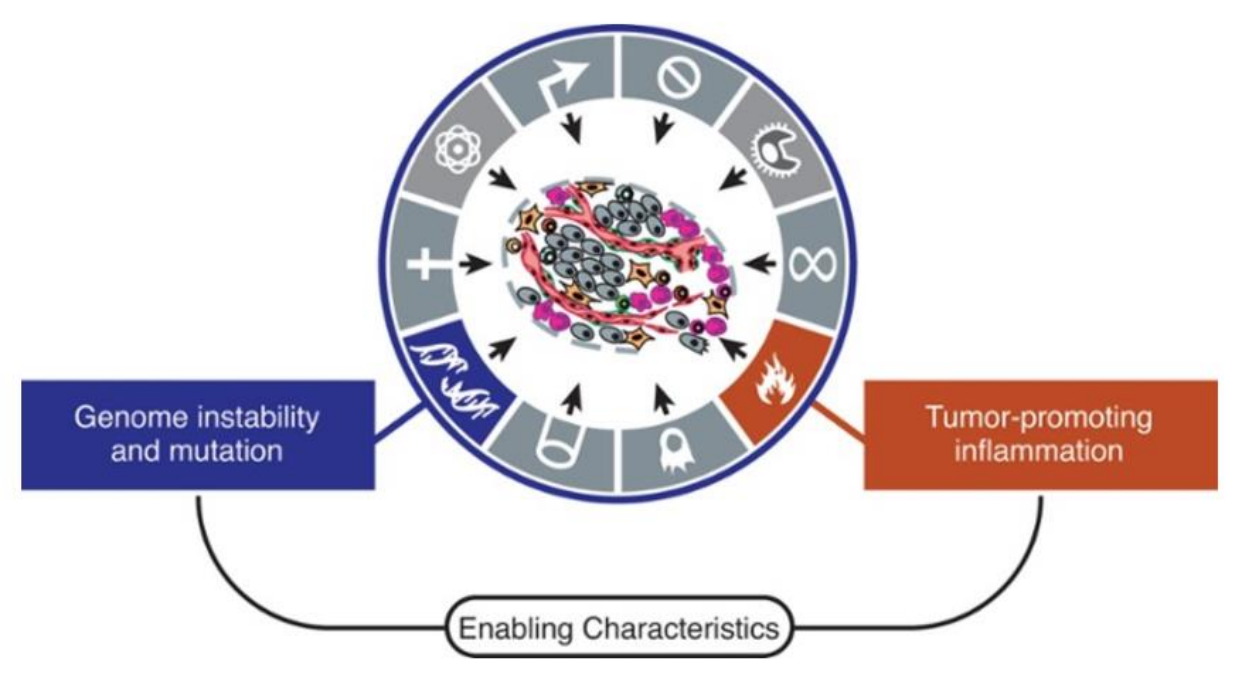


Molecular Biology of Cancer

- Two ubiquitous characteristics facilitate the acquisition of hallmark capabilities
  - Genome instability and mutation

    Nestabilita genomu
  - Tumor-promoting inflammation

    Nádorem vyvolaný zánět



Molecular Biology of Cancer

Genome instability and mutation

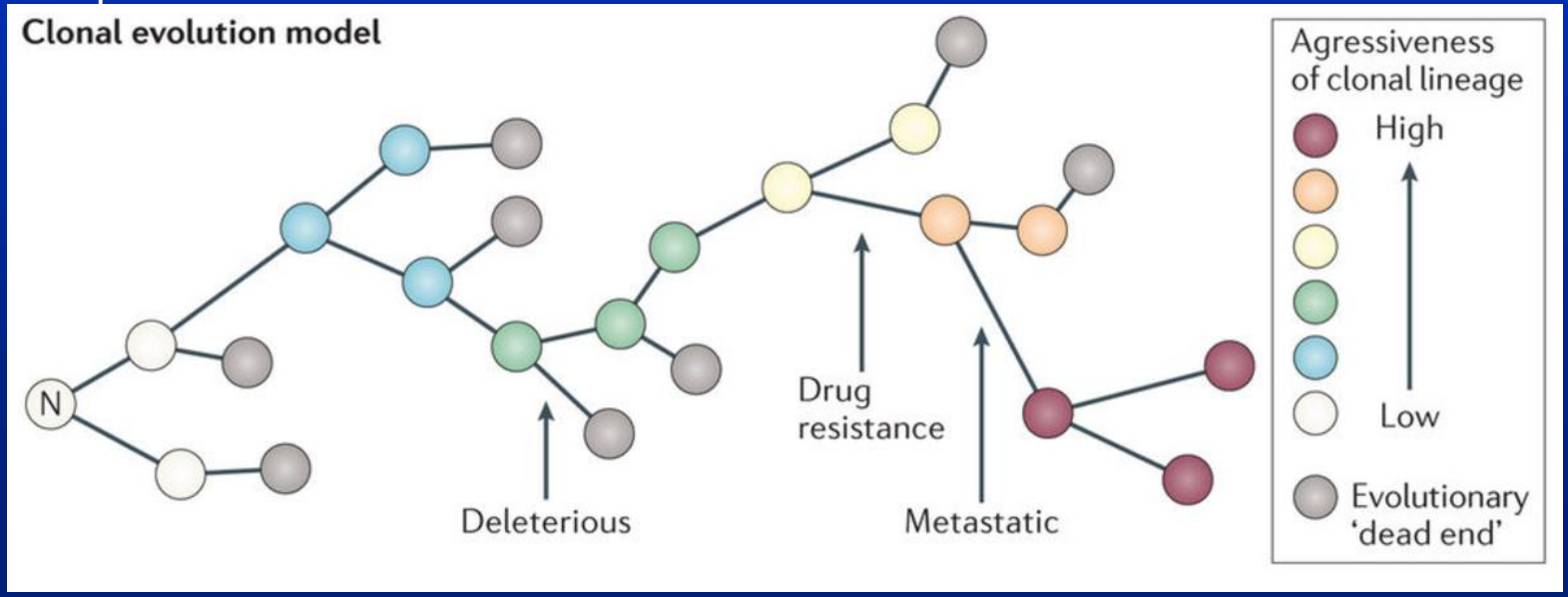
- **DNA damage can occur in several different forms, including SSBs or DSBs**
- **In higher eukaryotes, genomic stability is essential for healthy functioning and cell survival. DNA damage may induce mutations and can lead to cell death via apoptosis. Therefore, several repair mechanisms have evolved to maintain the integrity of the genome**
  - **BER** is a key pathway in the repair of SSBs and is reliant on the enzyme poly(ADP-ribose) polymerase (PARP)
  - **For DSB repair,** two pathways predominate:
    1. **Homologous recombination (HR)** that involves a protein kinase, ataxia-telangiectasia mutated (ATM)
    2. **Non-homologous end-joining (NHEJ)** that requires DNA-dependent protein kinase (DNA-PK)[2]

    HR is the most accurate mechanism for repairing DSBs, whereas NHEJ is rarely error-free[2]

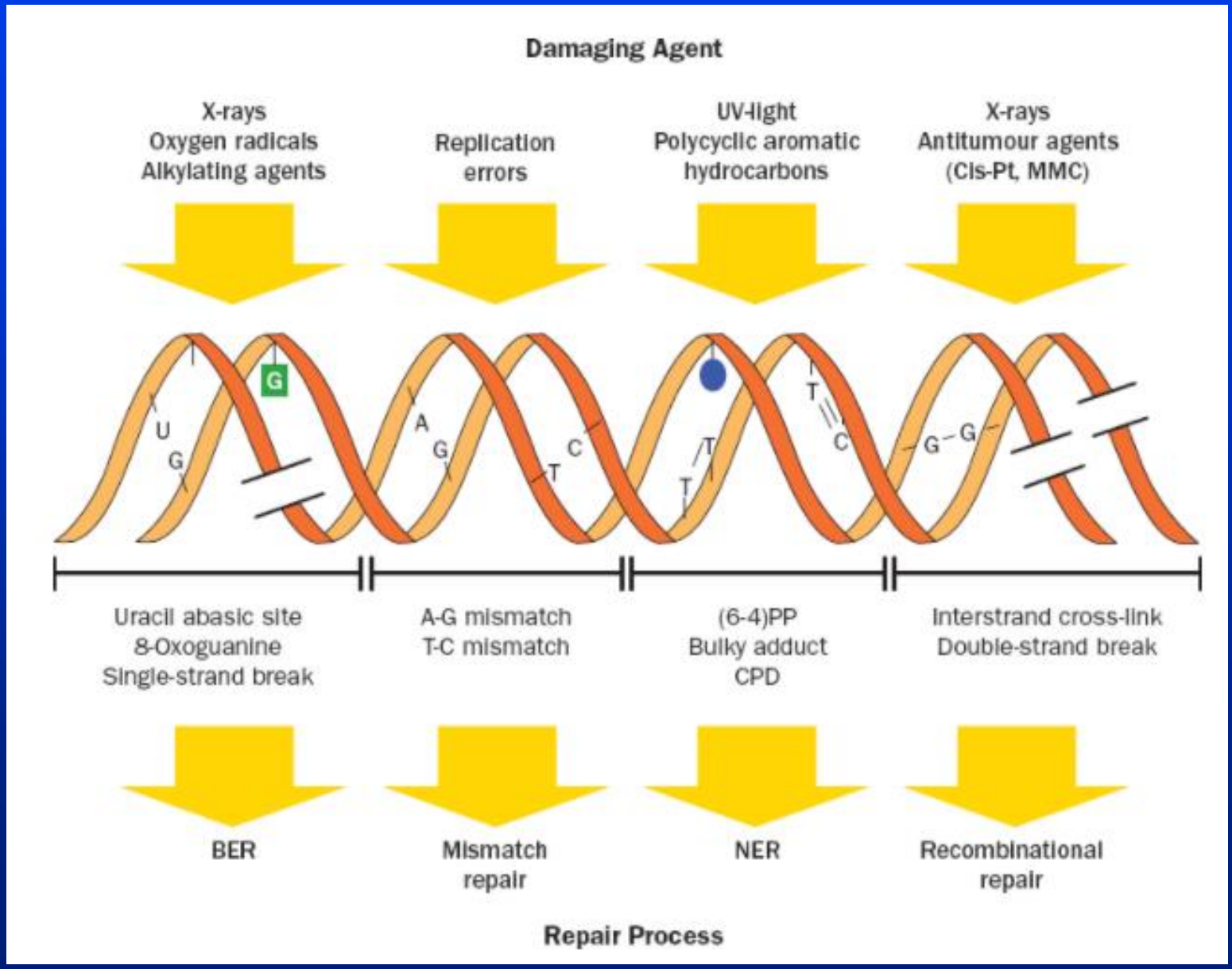
Molecular Biology of Cancer

Genome instability and mutation

- The ability of genome maintenance systems to detect and resolve defects in the DNA ensures that **rates of spontaneous mutation in normal cells of the body are typically very low.**
- The genomes of **most cancer cells, by contrast, are full of these alterations**, reflecting loss of genomic integrity with concomitantly increased rates of mutation.
- This heightened mutability appears to accelerate **the generation of variant cells, facilitating the selection** of those cells whose advantageous phenotypes enable their clonal expansion.

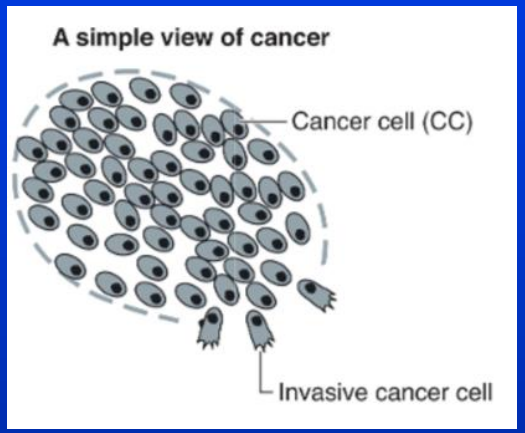


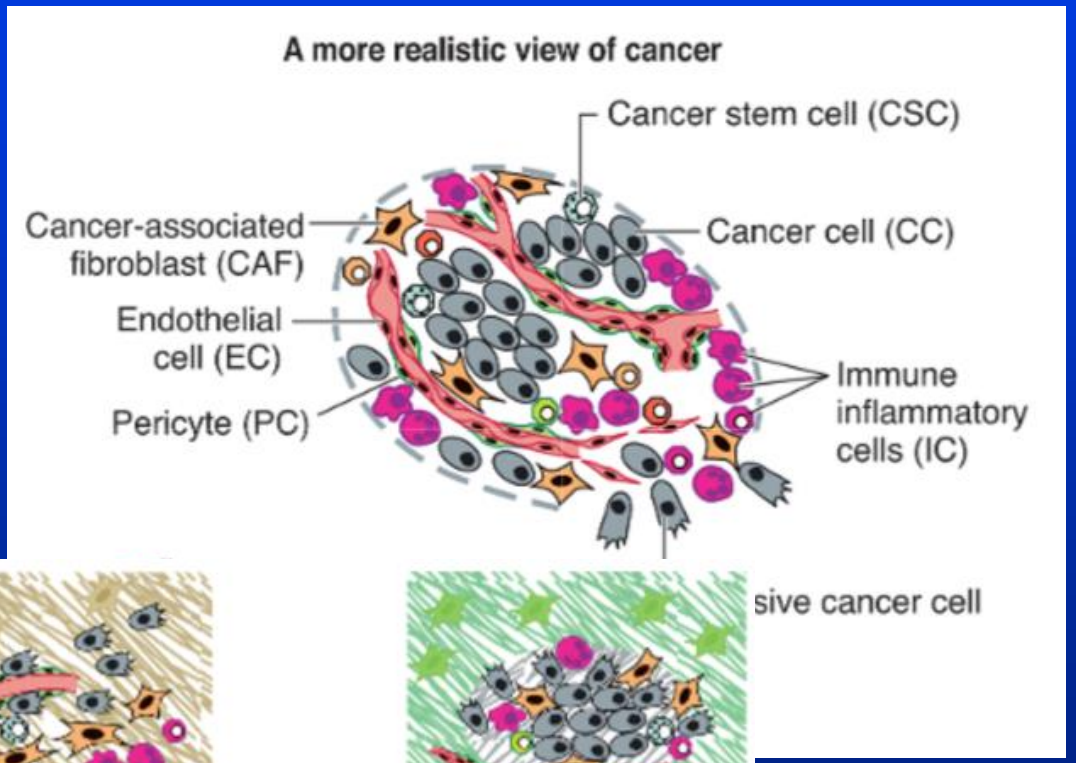
Molecular Biology of Cancer

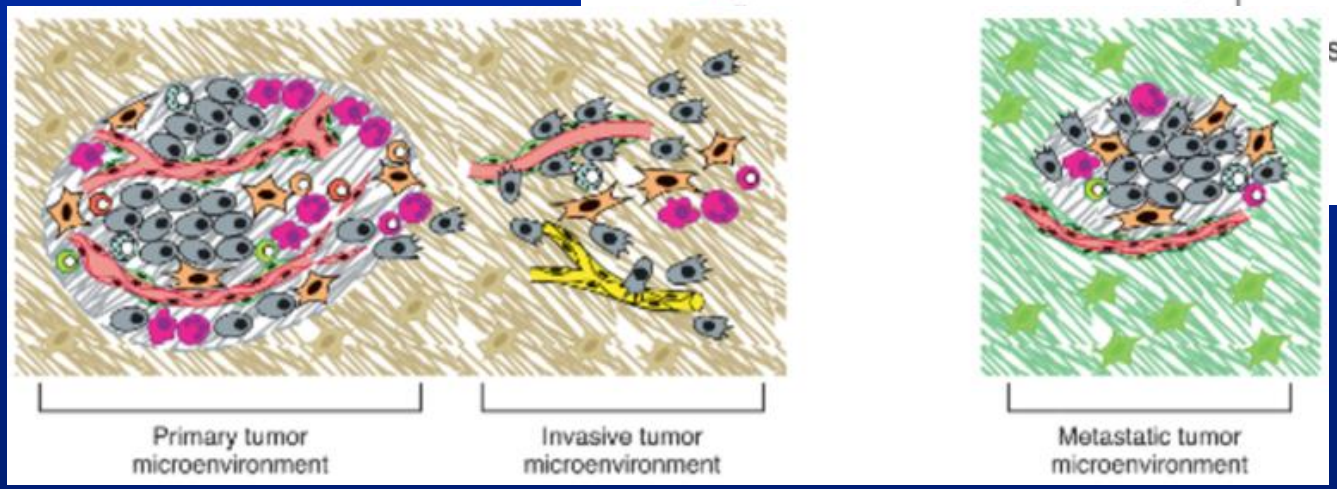


Molecular Biology of Cancer

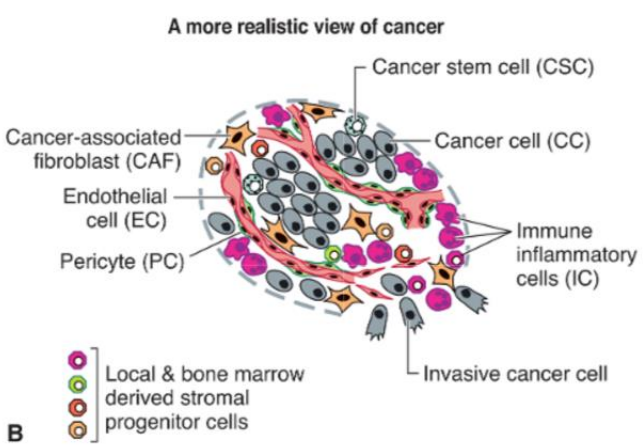
- Tumor promoting inflamation



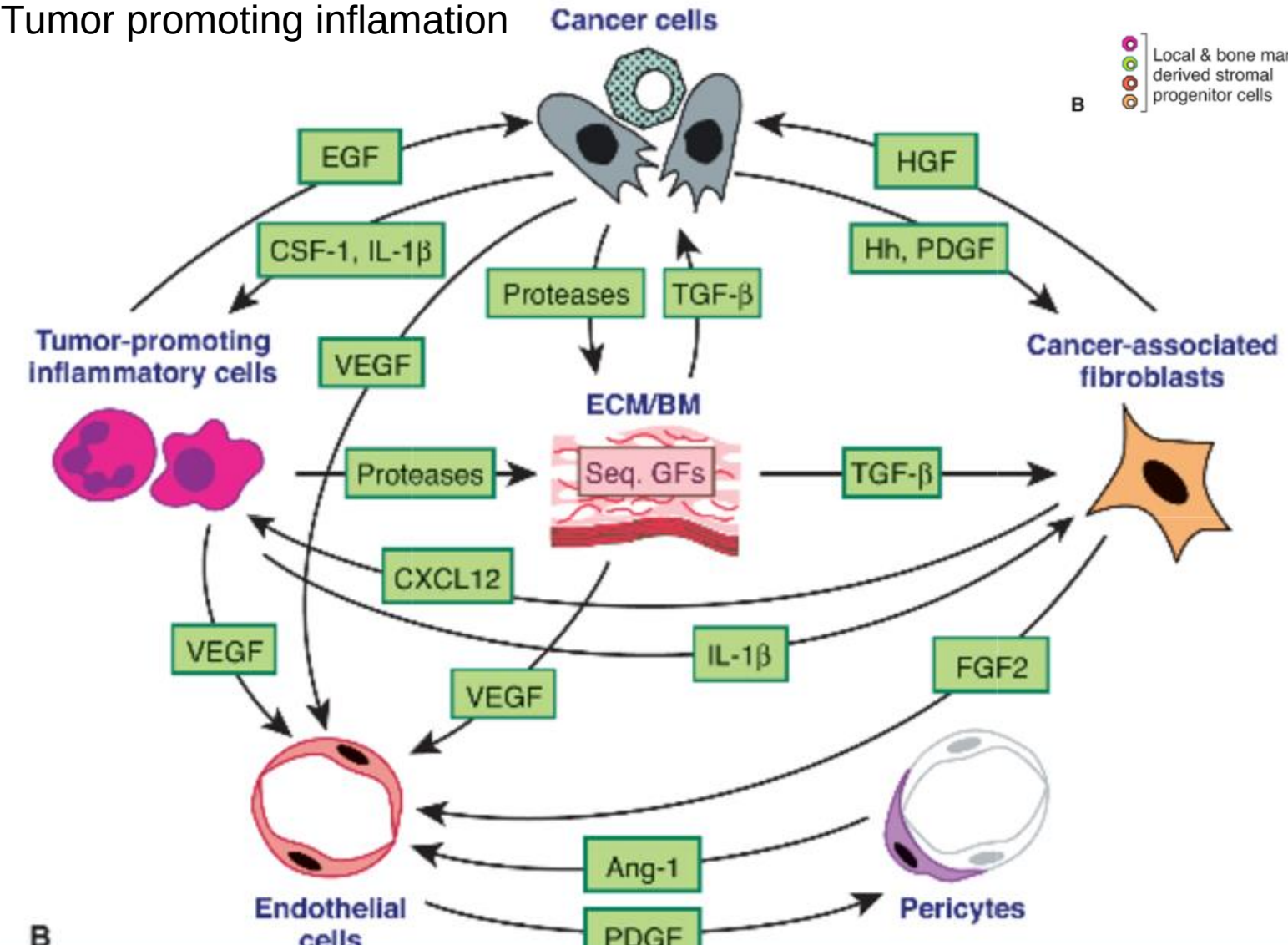




Molecular Biology of Cancer

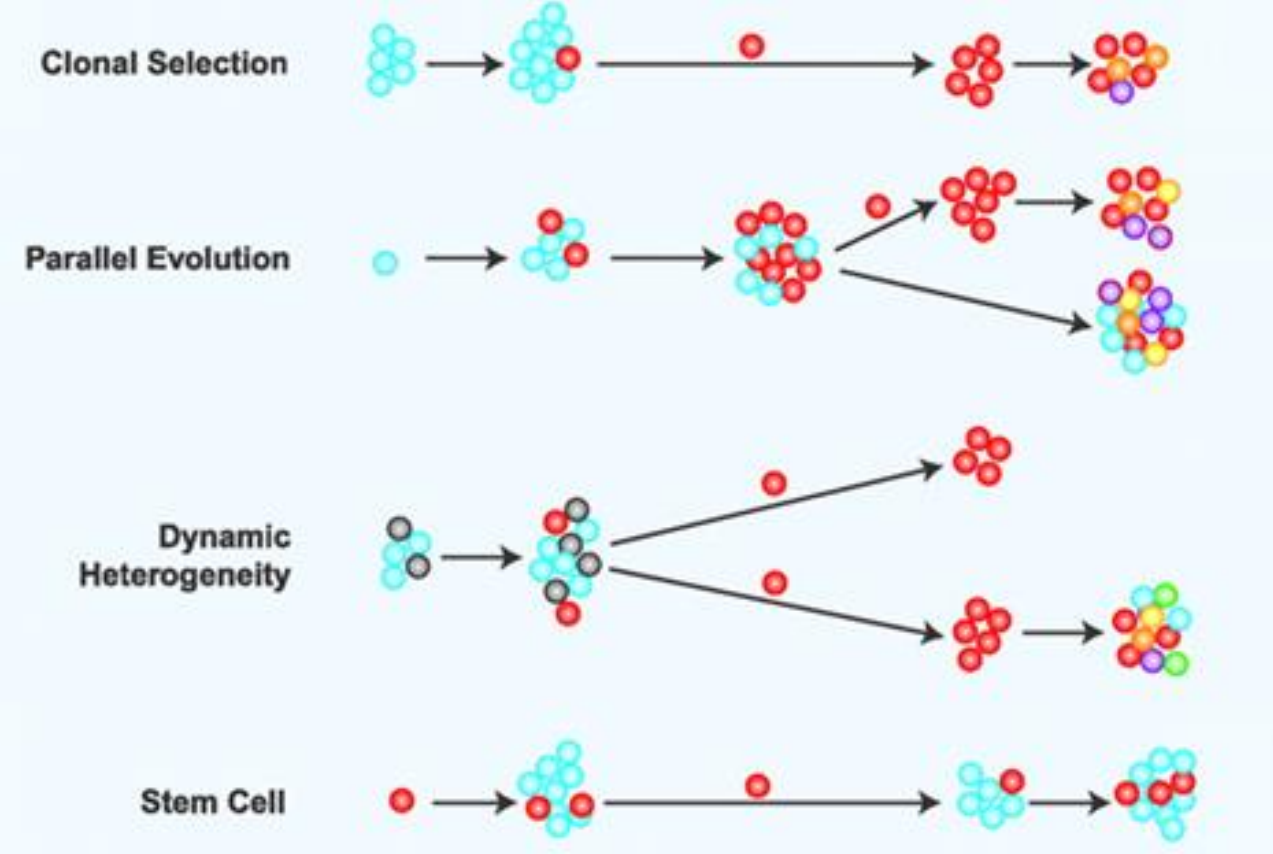


Tumor promoting inflamation



Molecular Biology of Cancer

- Genome instability and mutation
  → Clonal evolution models
- **The *clonal selection* model**
- **The *parallel evolution* model**
- **The *dynamic heterogeneity* model**
- **In the *stem cell* model**



Molecular Biology of Cancer

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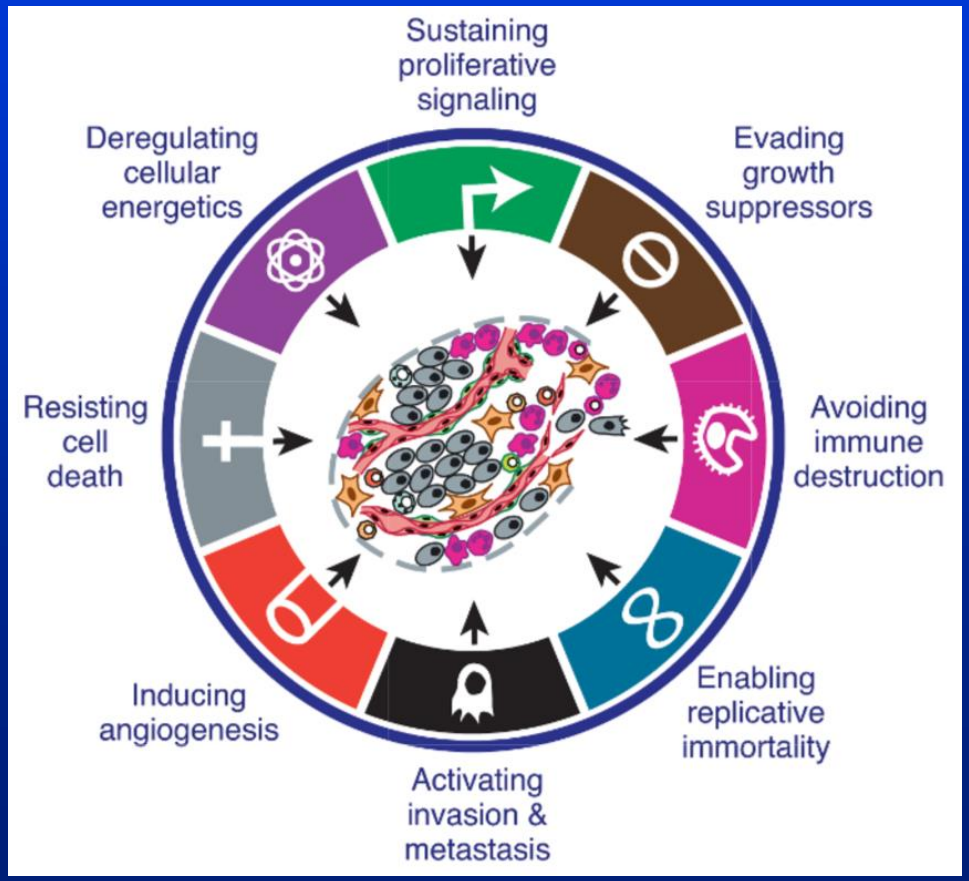
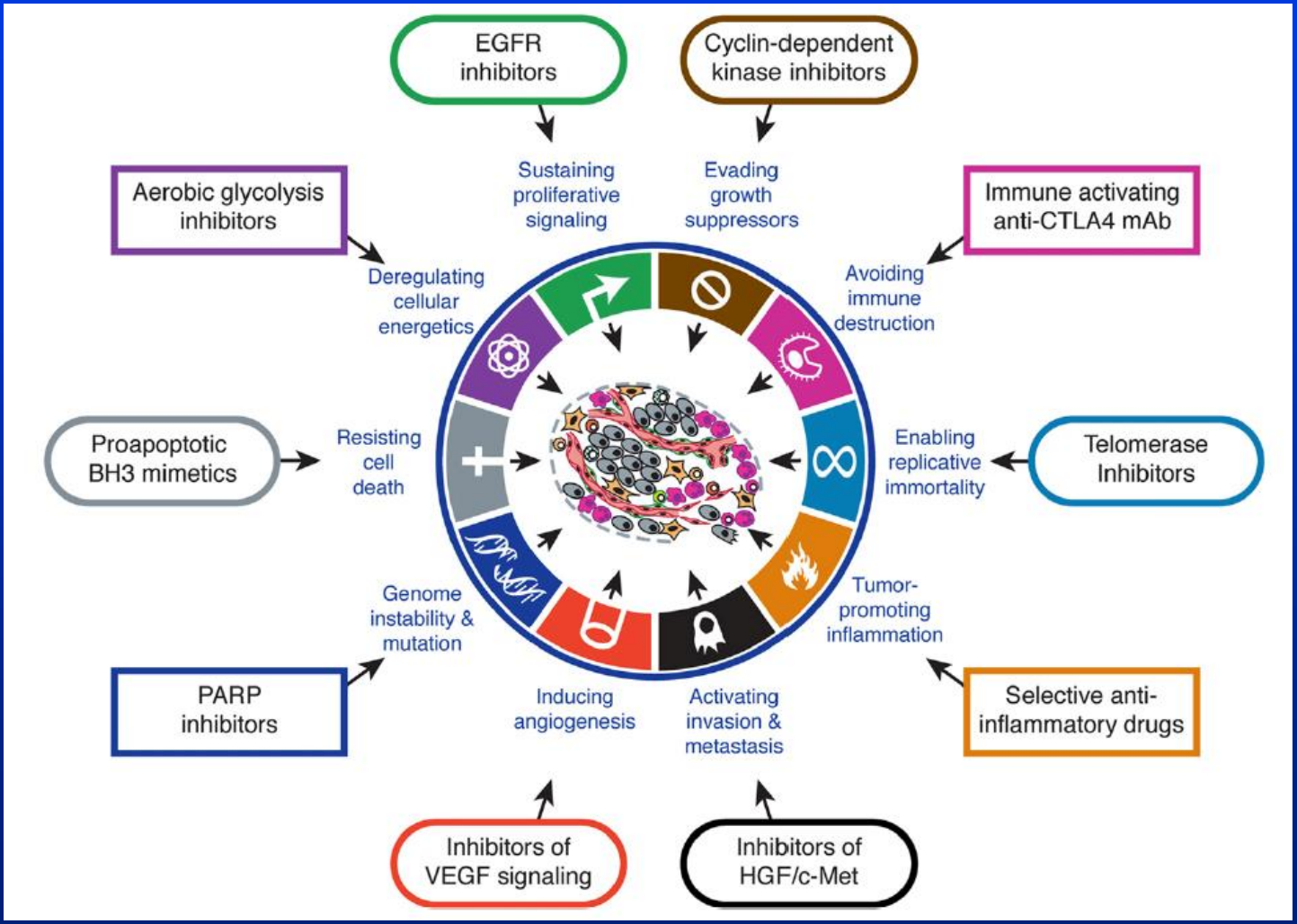


Molecular Biology of Cancer

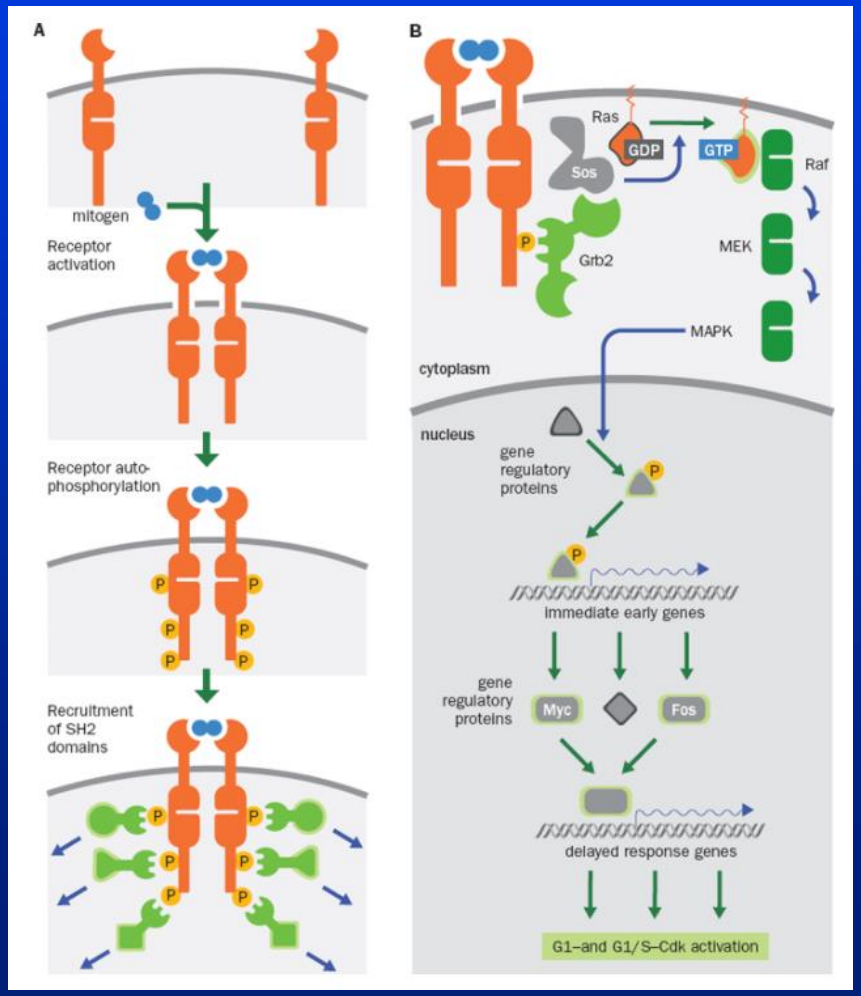
- „Hallmarks of cancer“

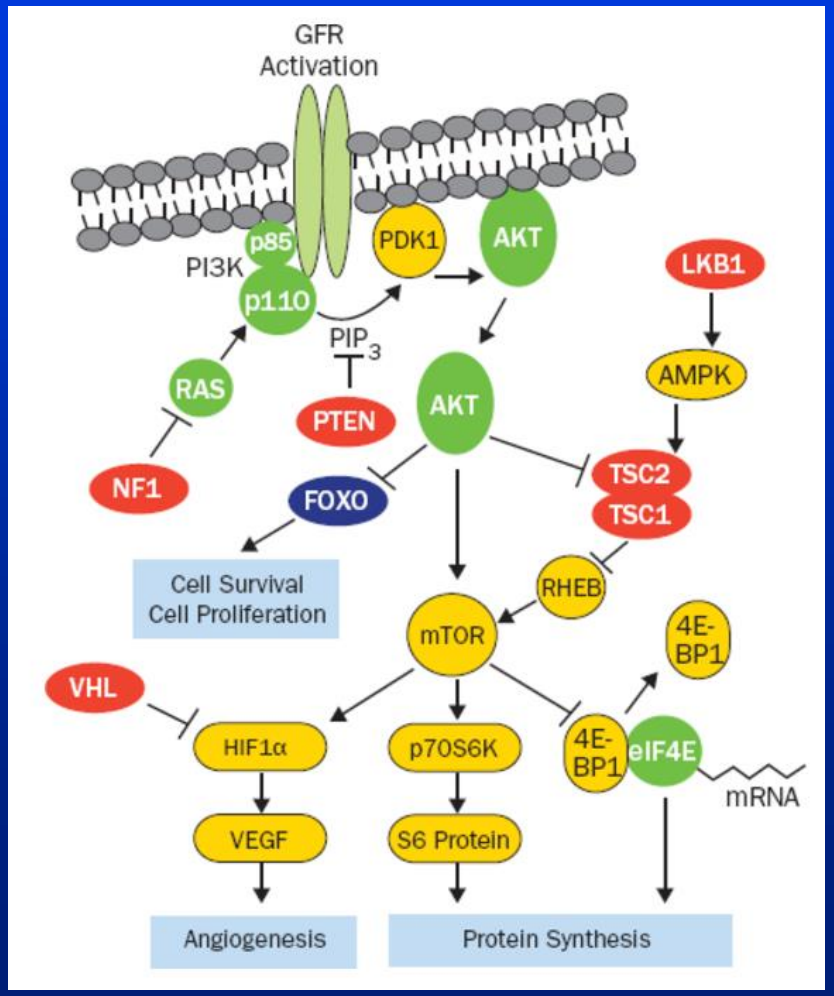


Molecular Biology of Cancer

Sustaining proliferative signaling:

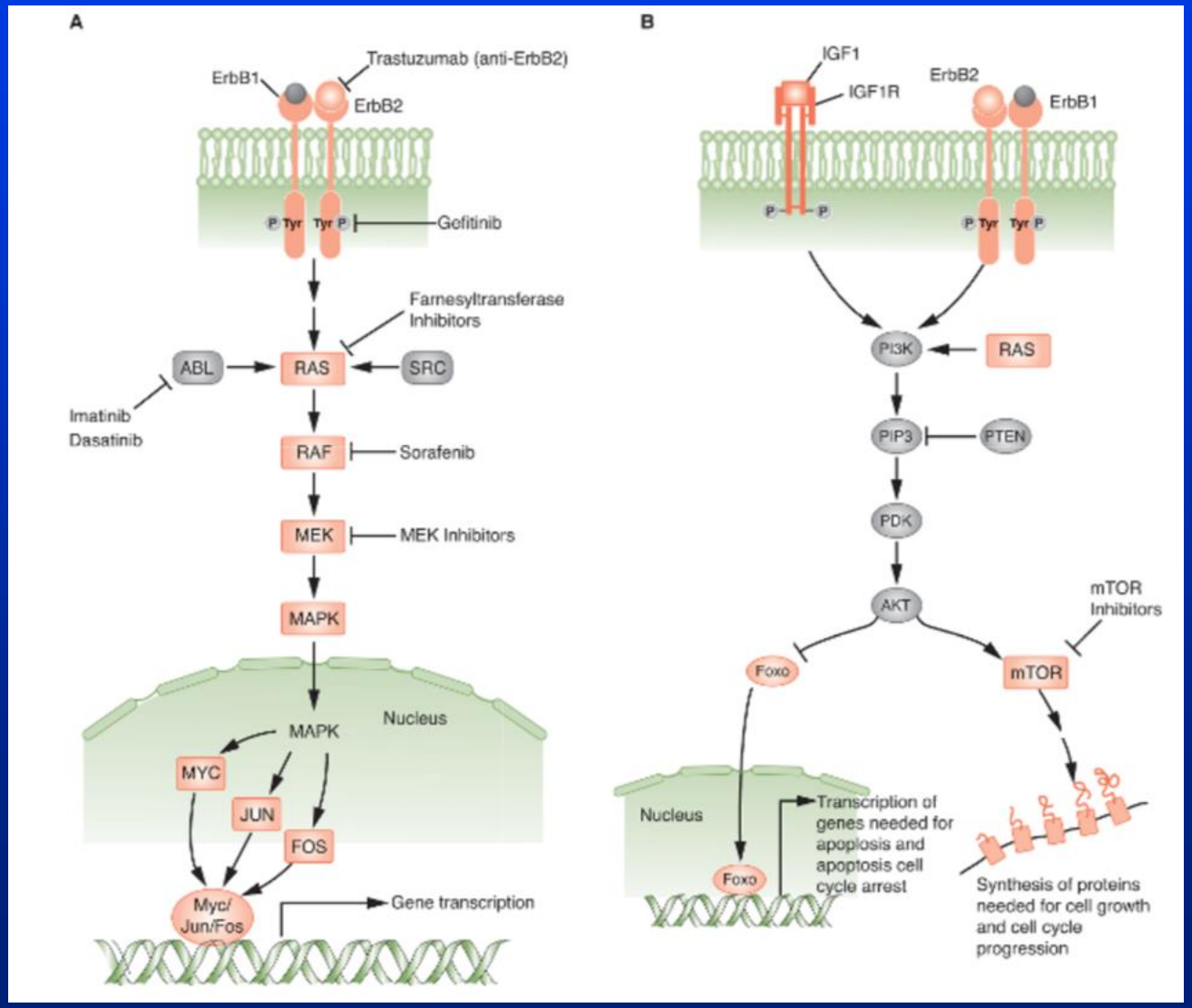
- *Somatic activation mutations in oncogenes (e.g. HER2,PI3K,RAS,RAF,MYC)*
- *Disruption of negative-feedback mechanisms (e,g, PTEN, mTOR)*





Molecular Biology of Cancer

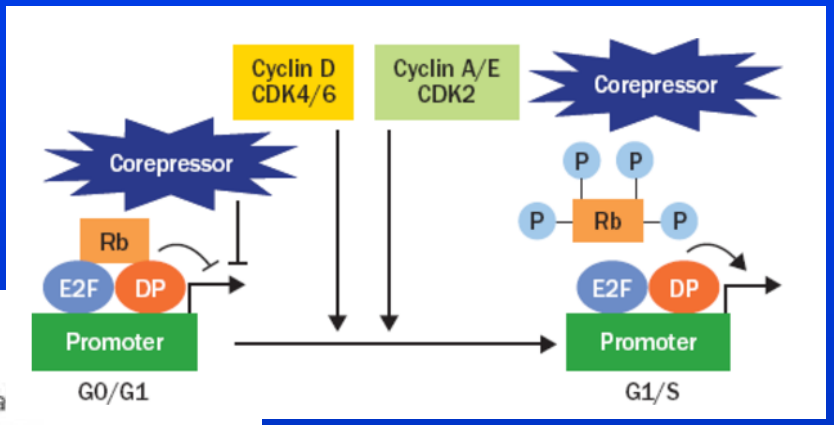
Sustaining proliferative signaling: RAS and PI3K signaling pathways

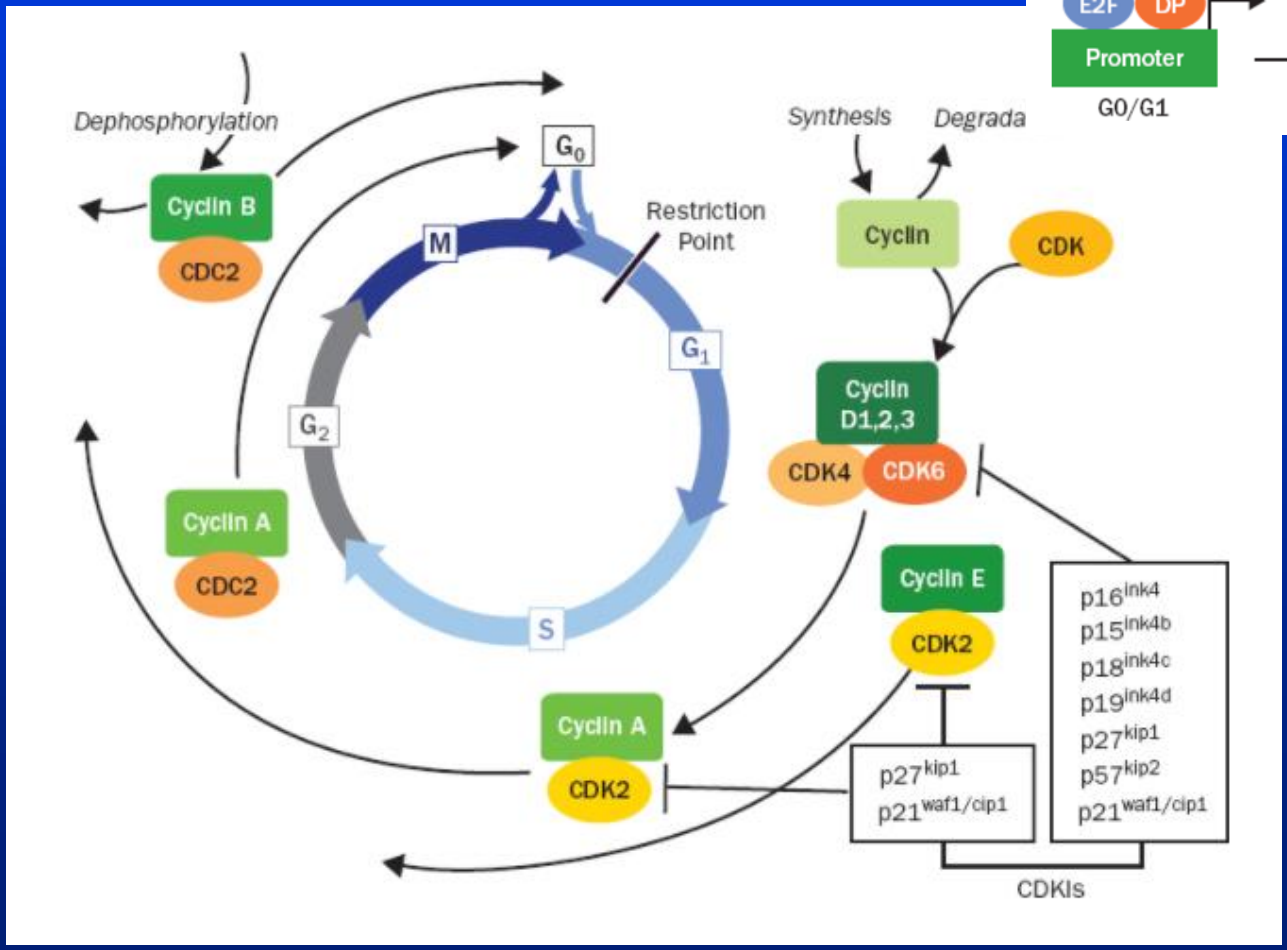


Molecular Biology of Cancer

Evading growth suppresors :

- Cell cycle control (e.g. Rb, TGF-b)
- Cell Death control (e.g. p53)

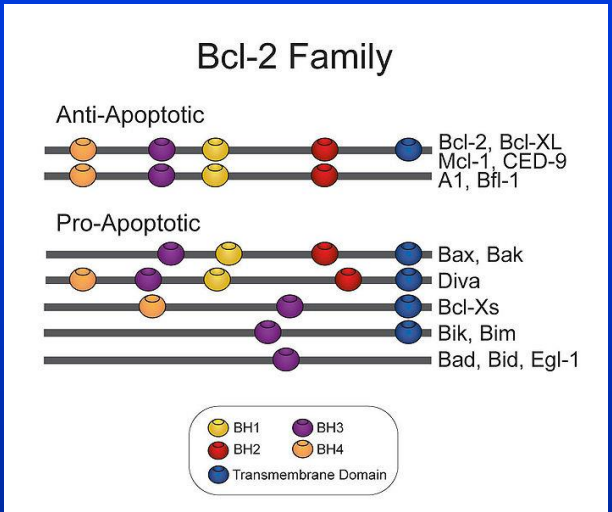


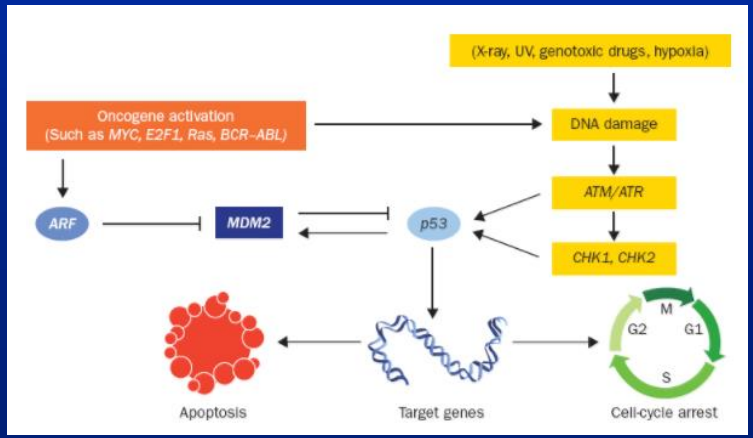


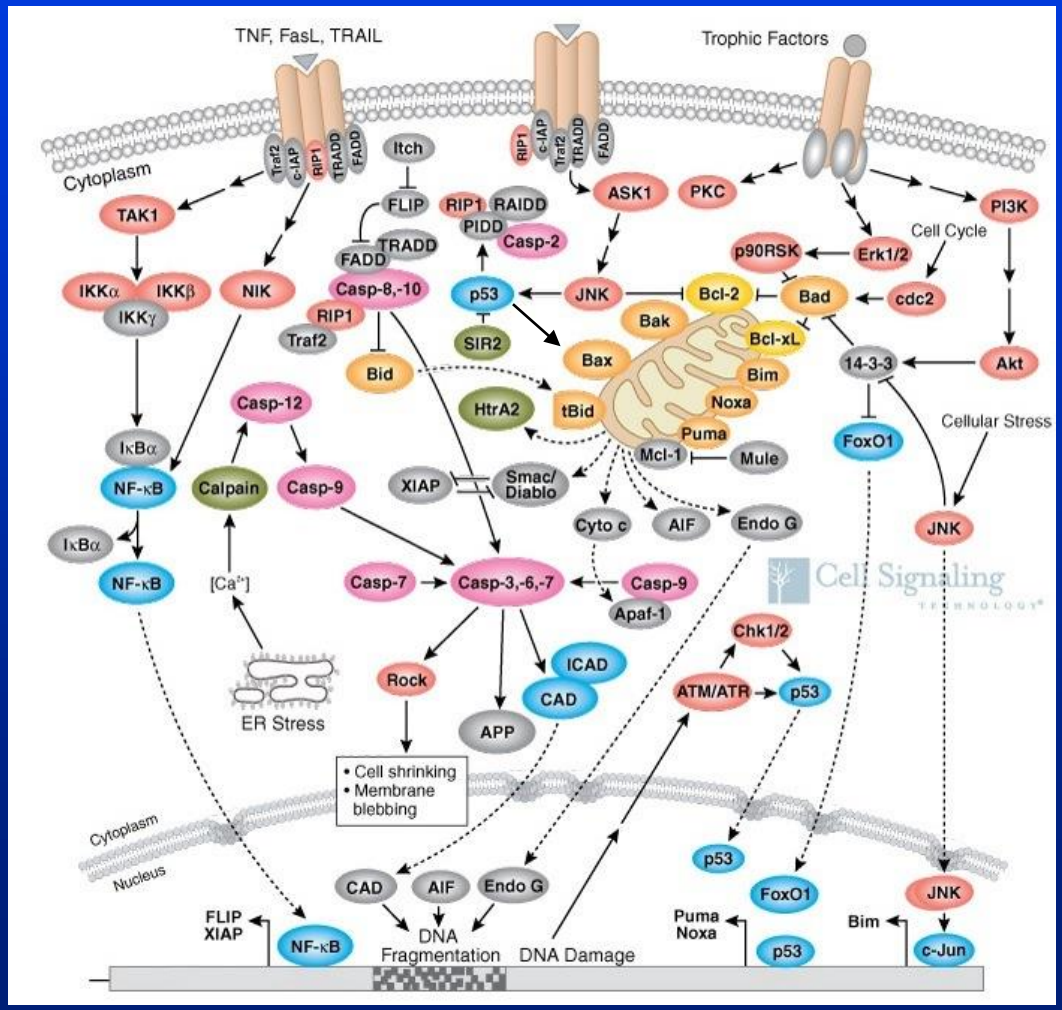
Molecular Biology of Cancer

Resisting cell death:

- Extrinsic apoptotic program (Death receptors: TNF, FASL, TRAIL → Casp8)
- Intrinsic apoptotic program (Bcl-2/Bax, Cytochrom C, p53 → Casp9)







Molecular Biology of Cancer

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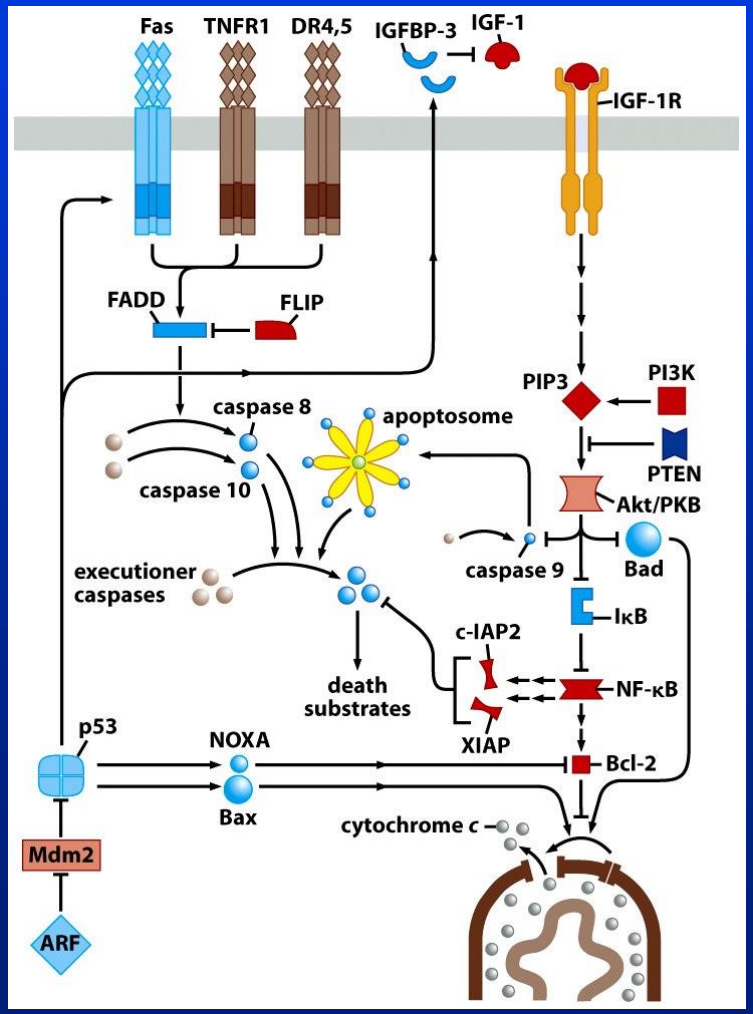
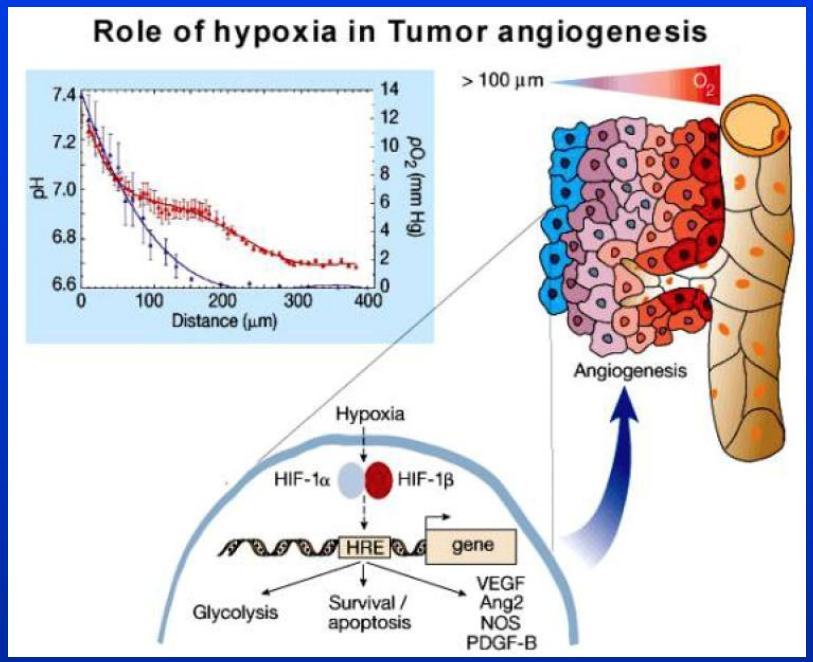
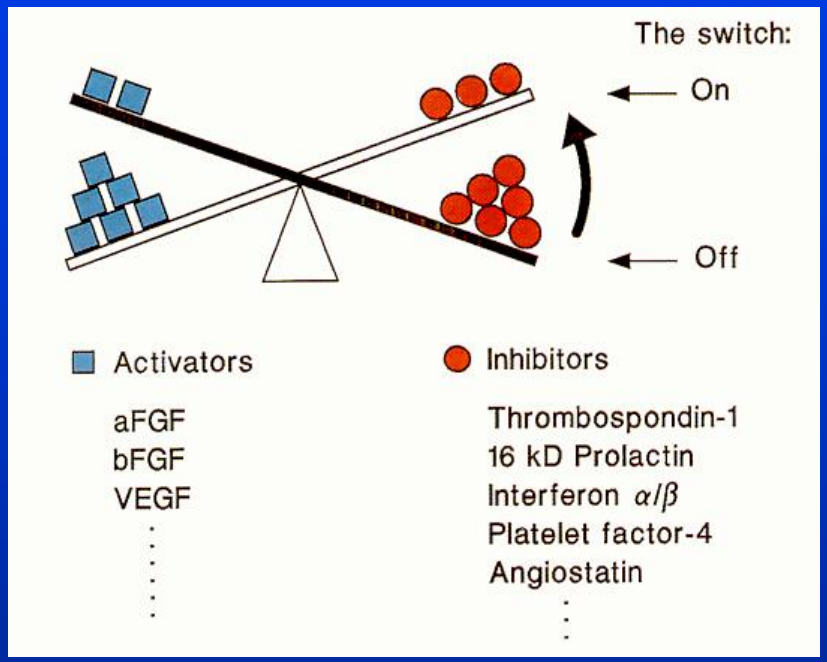
Apoptosis inhibition
Cancer cell strategy

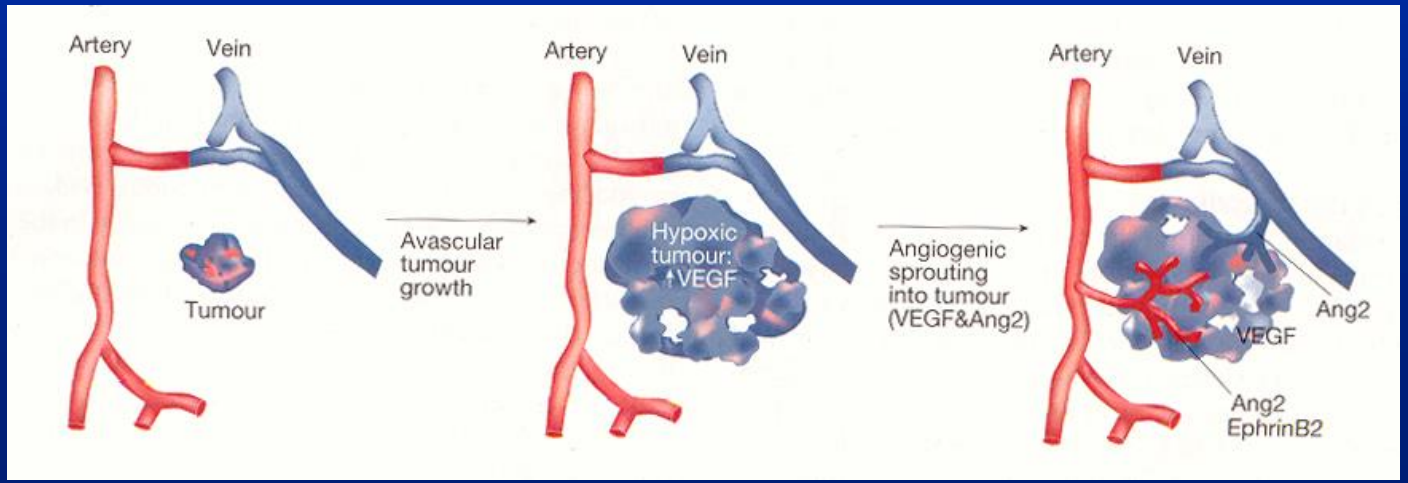


Molecular Biology of Cancer

Inducing angiogenesis:



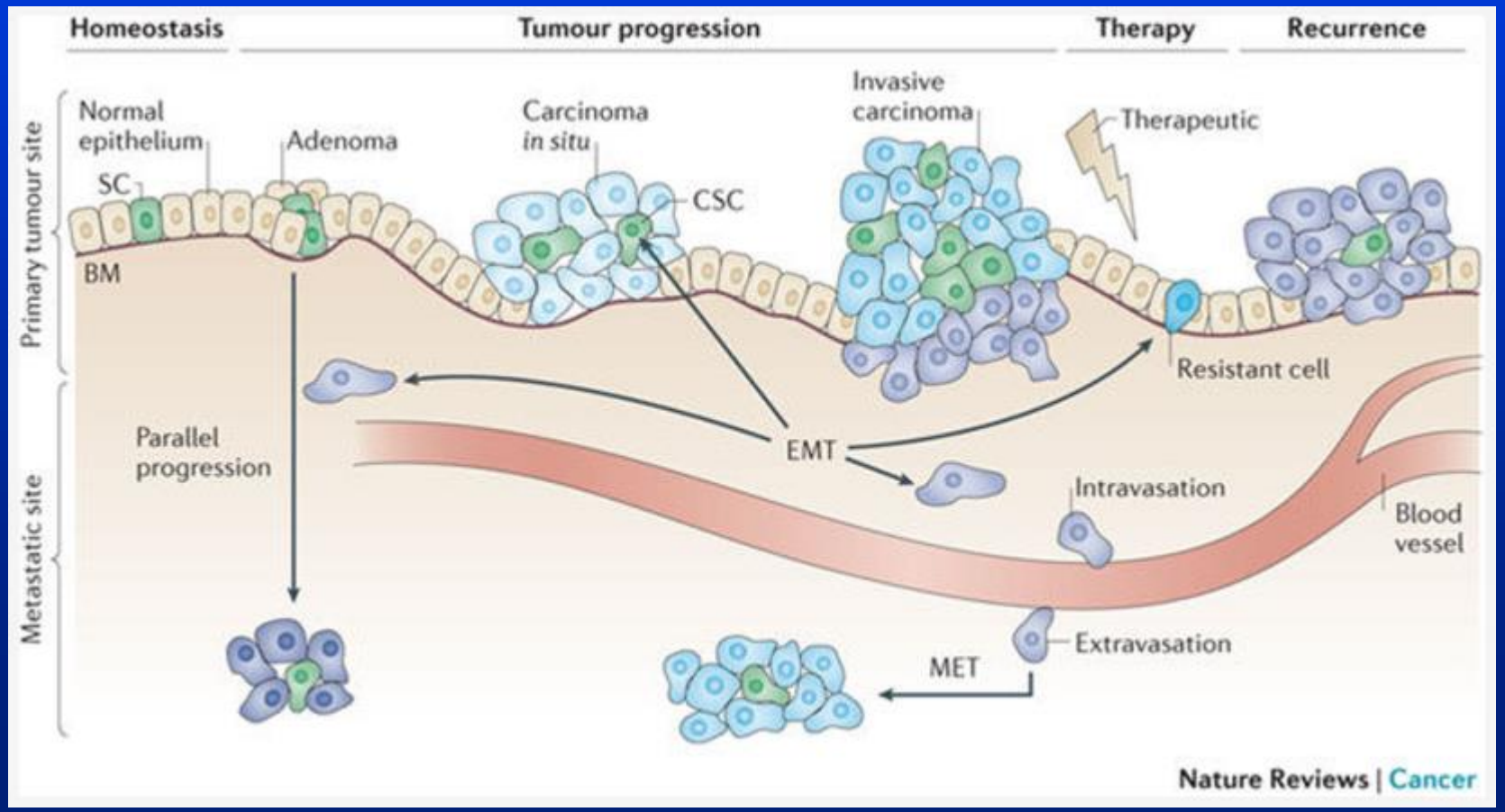




Molecular Biology of Cancer

Activating invasion and metastasis:

- Epithelial-to-Mesenchymal Transition
  - Loss of contact inhibition (E-cadherin/CDH1, integrins)
  - Activation of EMT program (Snail, Slug, Twist,…)
- Stromal cell contribution (mesenchymal stem cells, macrophages)



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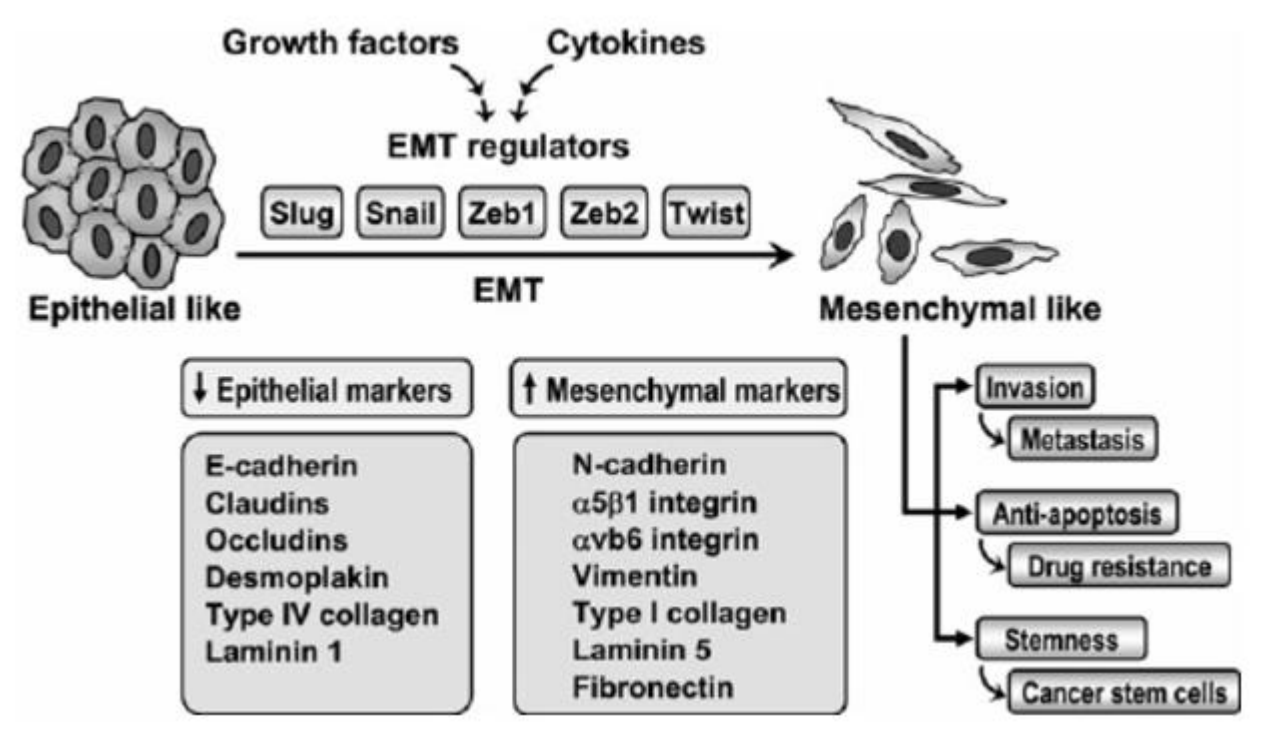
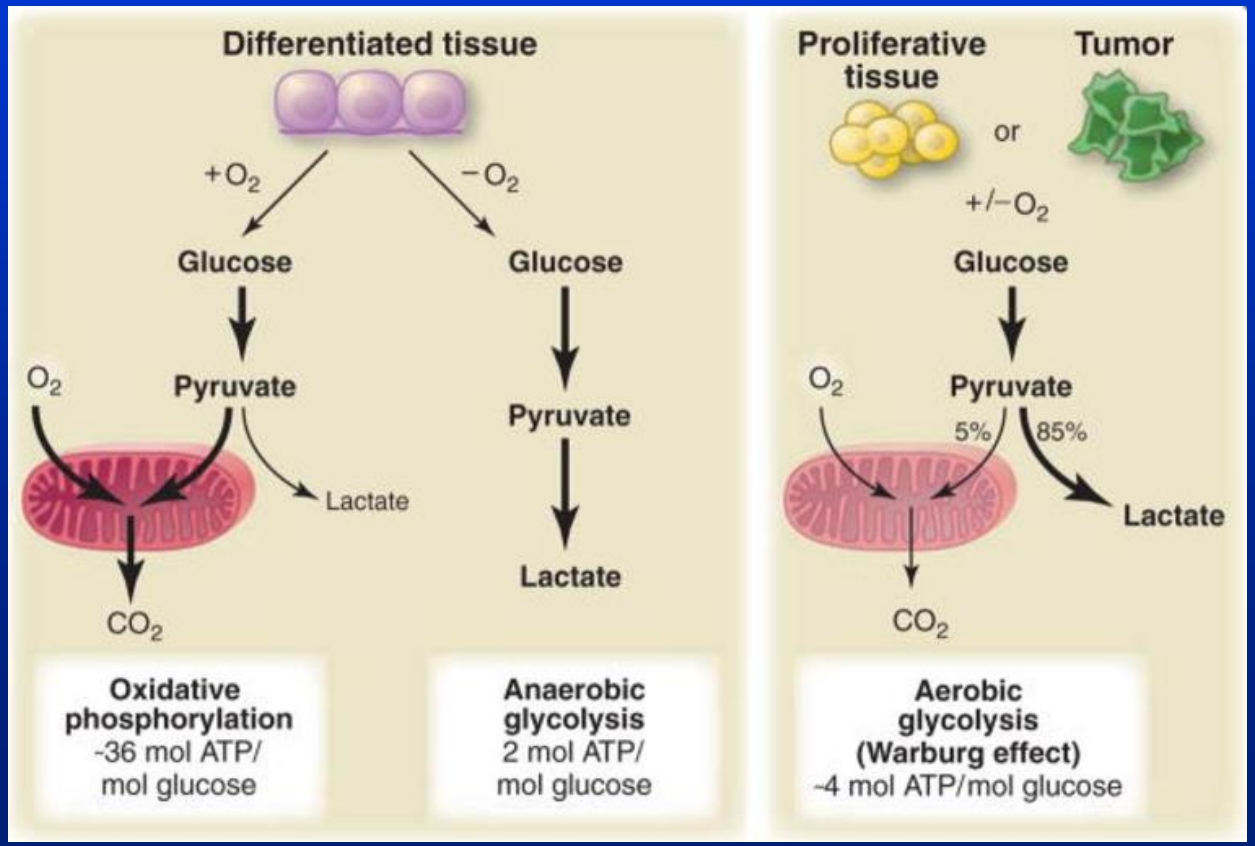


Molecular Biology of Cancer

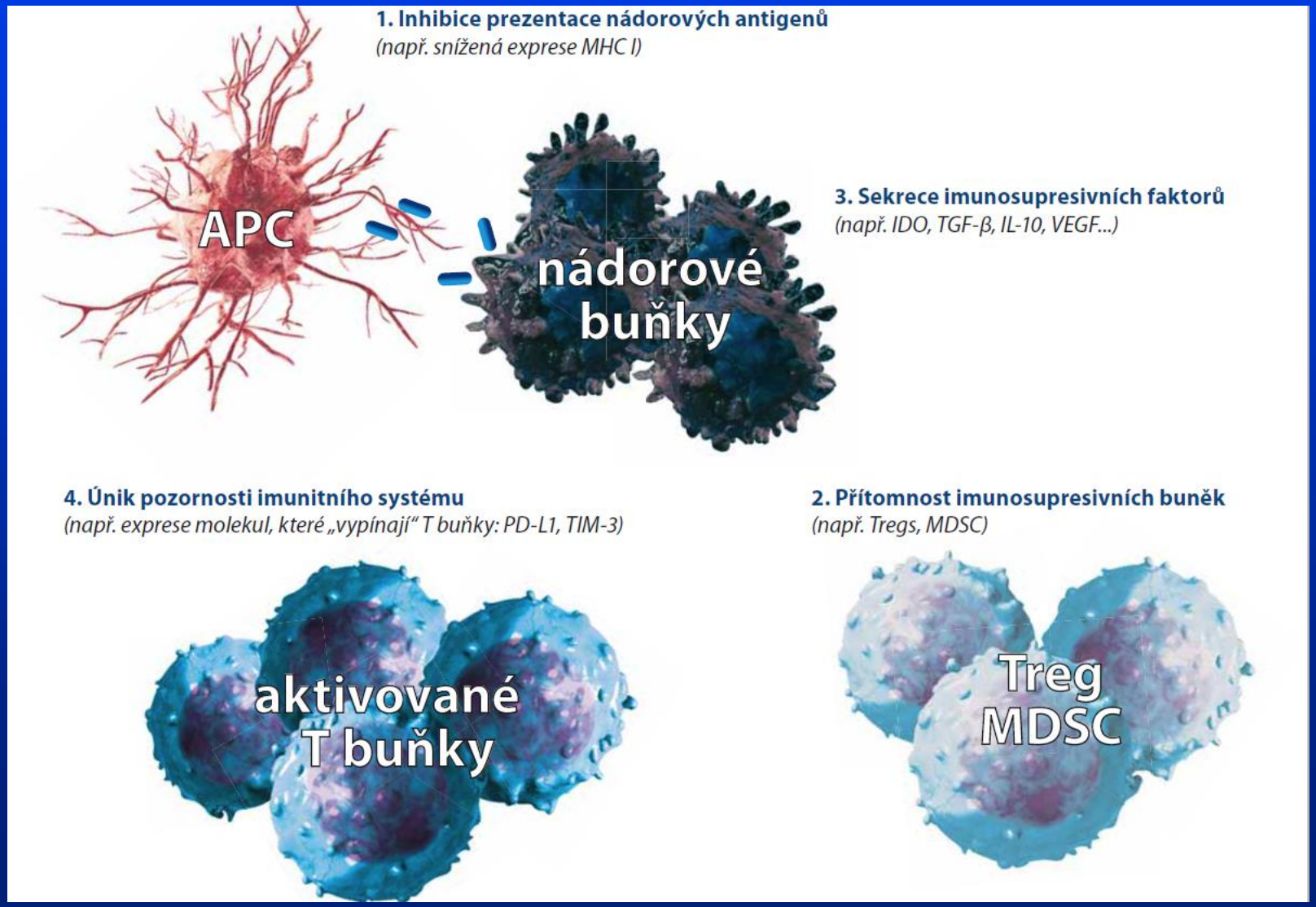
Reprogramming energy metabolism:

- Glycolysis via oxydative phosphorylation → arebic glycolysis
- The common feature of this altered metabolism is increased glucose uptake and fermentation of glucose to lactate. This phenomenon is observed even in the presence of completely functioning mitochondria and together is known as the Warburg Effect.
- Upregulation of glucose transporter GLUT1



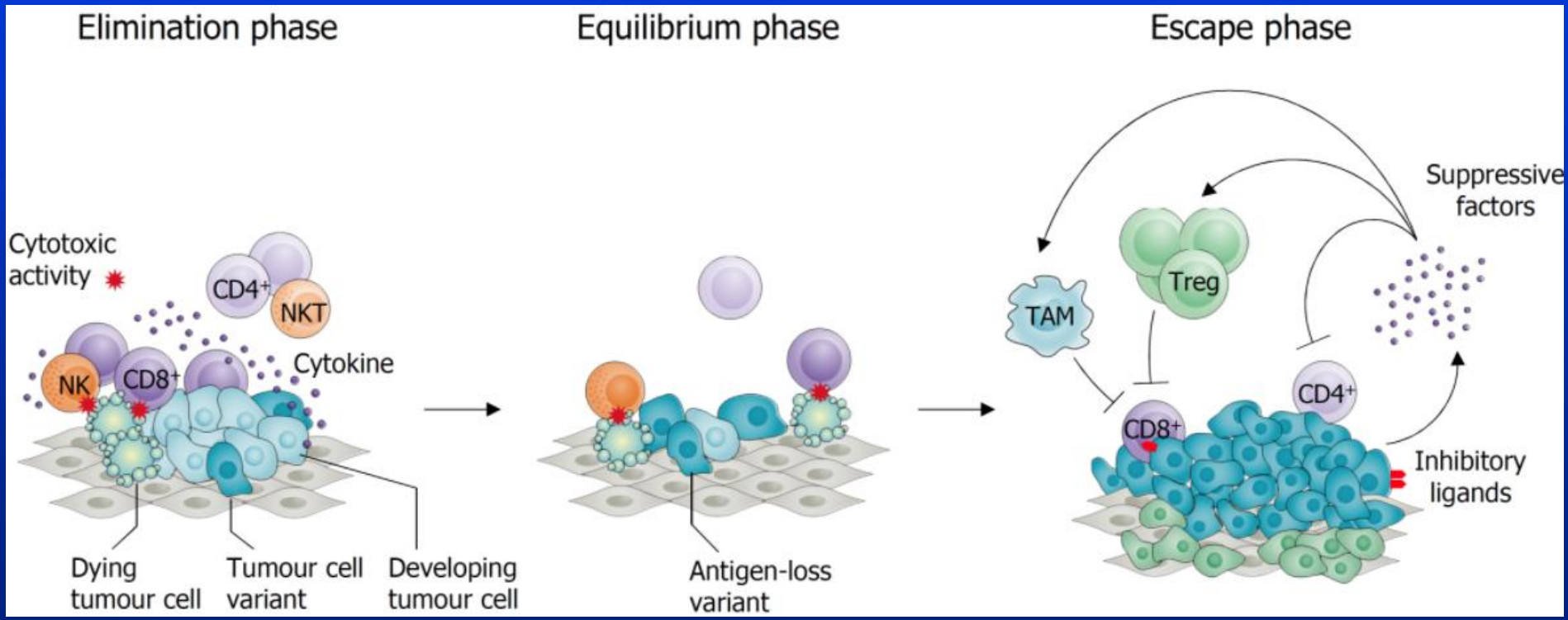
Molecular Biology of Cancer

– Evading immune destruction



Molecular Biology of Cancer

- Evading immune destruction:
  - Defective antigen presentation
  - Tolerance and Adaptation
  - Immunosuppressive Cells



Molecular Biology of Cancer

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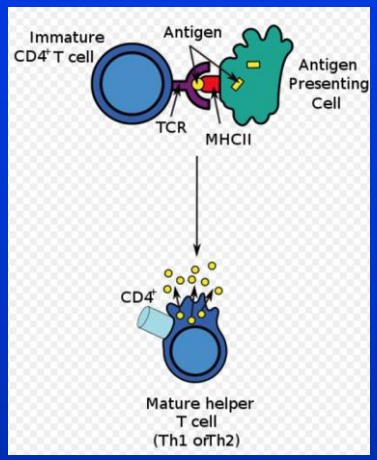
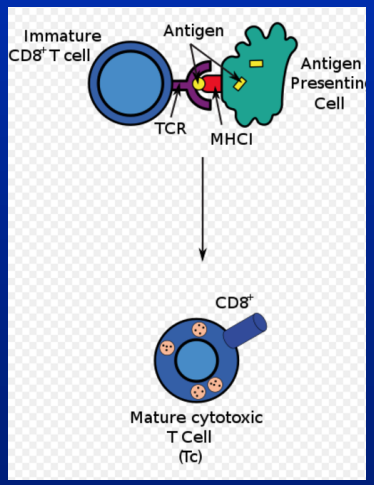
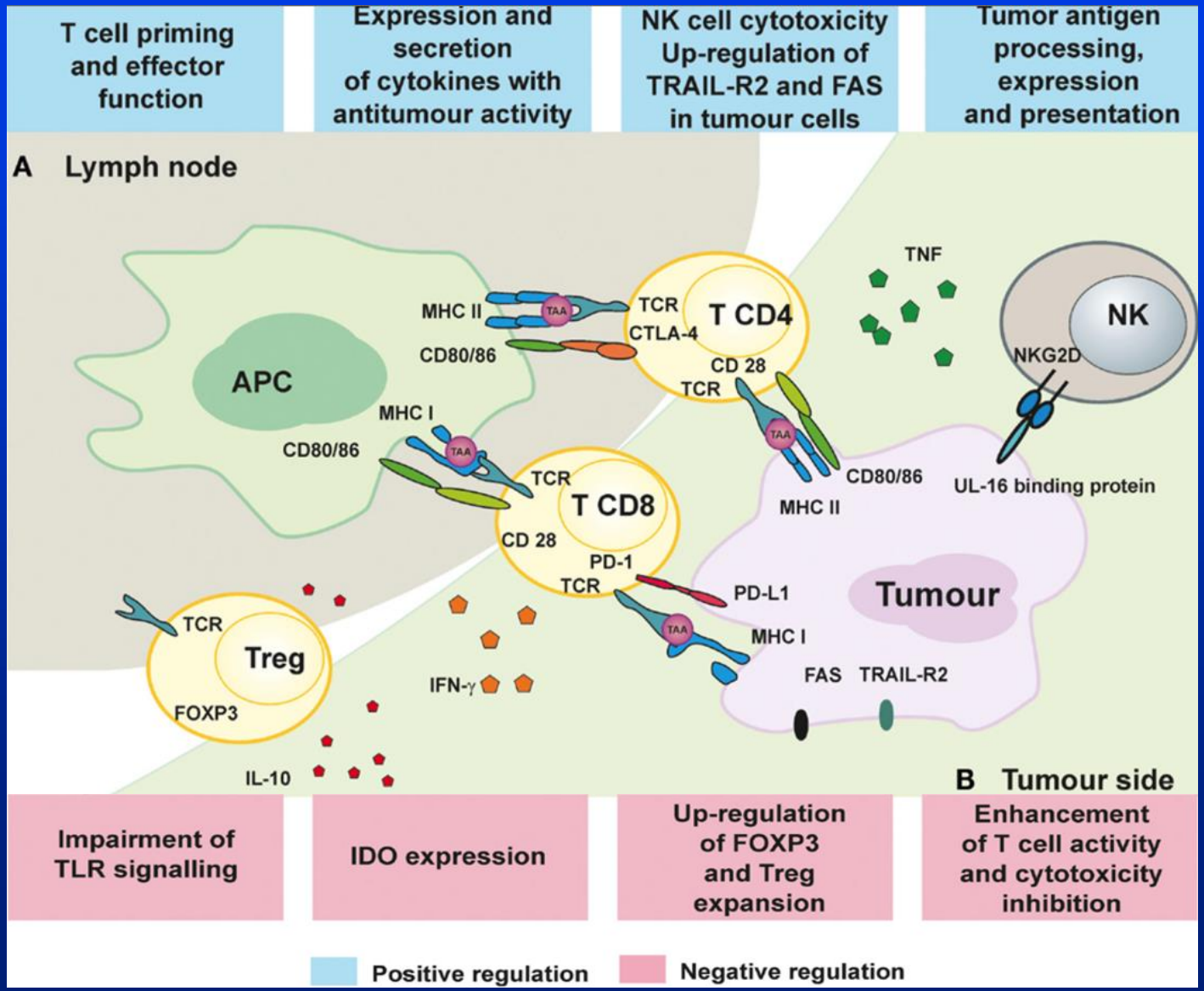
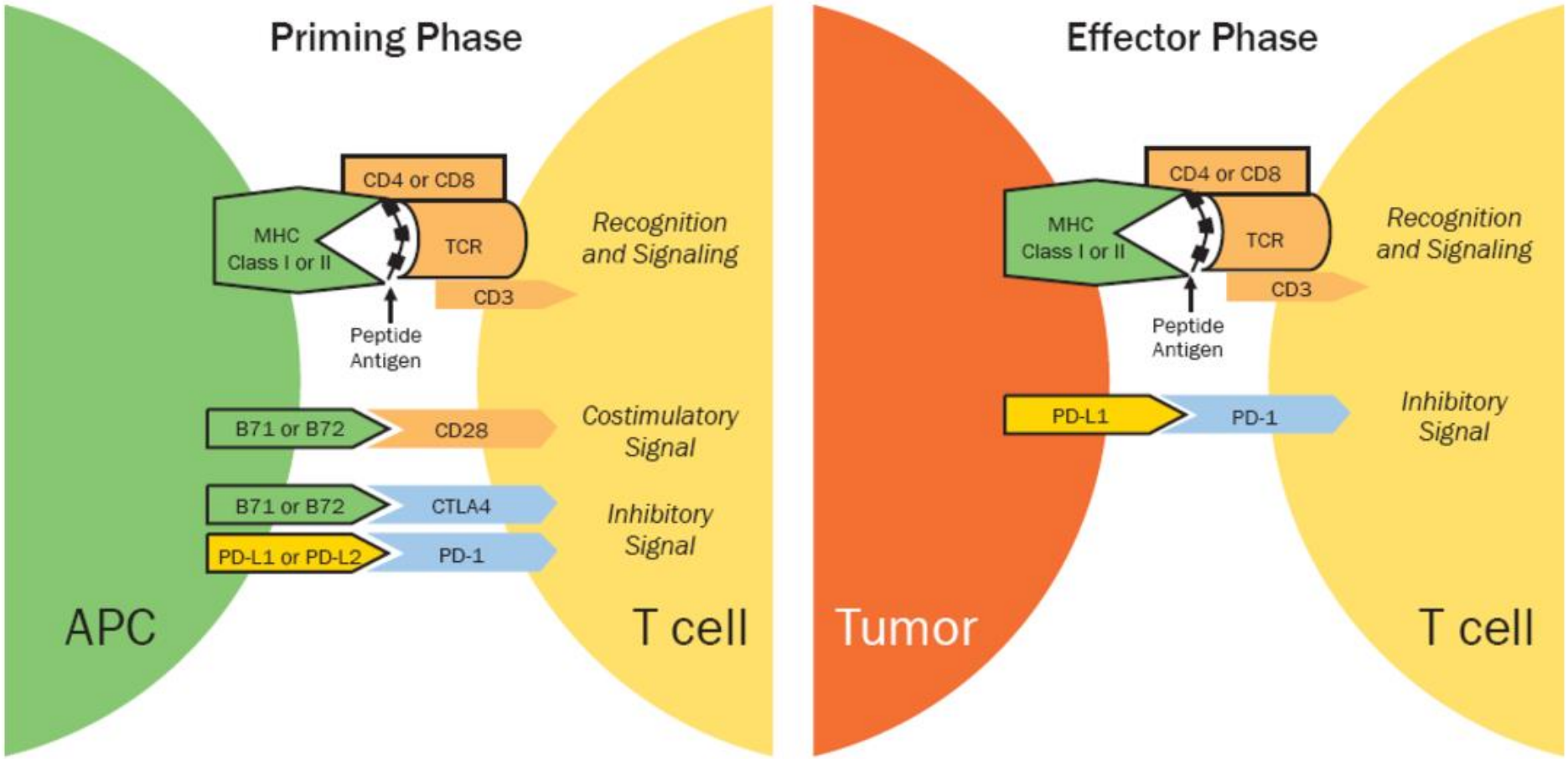






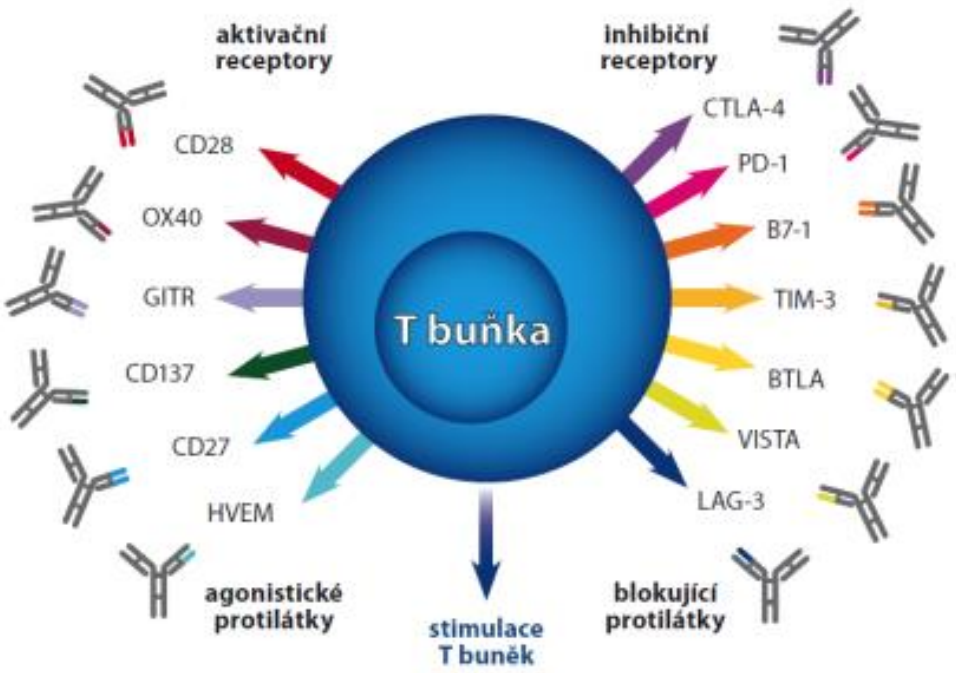
Molecular Biology of Cancer

Evading immune destruction



Molecular Biology of Cancer

Evading immune destruction



Anti-CTLA-4
Ipilimumab

Anti-PD-1
Pembrolizumab
Nivolumab

Molecular Biology of Cancer: TKI

Target	Drug	Approved Indications
ALK	Crizotinib Ceritinib	ALK mutant lung cancer
BCR-ABL	Imatinib Dasatinib Nilotinib Bosutinib Ponatinib	Chronic myeloid leukemia Philadelphia chromosome–positive acute lymphoid leukemia T315 mutation only (ponatinib)
BRAF	Vemurafenib Dabrafenib	BRAF mutant melanoma
BTK	Ibrutinib	Chronic lymphocytic leukemia Mantle cell lymphoma
EGFR	Gefitinib Erlotinib Afatinib	Lung adenocarcinoma with EGFR mutation
HER2	Lapatinib	$Her2^{+}$ breast cancer
JAK2	Ruxolitinib	JAK2 mutant myelofibrosis
KIT	Imatinib Sunitinib	Gastrointestinal stromal tumor
MEK	Trametinib	BRAF mutant melanoma

Molecular Biology of Cancer: TKI

PI3K delta[a]	Idelalisib	Chronic lymphocytic leukemia Indolent non-Hodgkin lymphoma
PDGFR- α/β	Imatinib	Chronic myelomonocytic leukemia (with TEL-PDGFR-β fusion) hypereosinophilic syndrome (with PDGFR-β fusion) Dermatofibrosarcoma protuberans
RET	Vandetanib Sorafenib Cabozantinib	Medullary thyroid cancer
TORC1	Sirolimus (rapamycin)	Kidney cancer
(mTOR)	Everolimus Temsirolimus	Breast cancer Tuberous sclerosis
VEGF Receptor	Sorafenib Sunitinib Axitinib Pazopanib	Kidney cancer Hepatocellular carcinoma (sorafenib only) Pancreatic neuroendocrine tumors (sunitinib)

Molecular Biology of Cancer: Angiogenesis-I

Drug	Class	(Cellular Targets)	Approval	Indications
Bevacizumab (Avastin)	Anti-VEGF mAB	VEGF	2004	First- and second-line metastatic CRC
			2006	First-line NSCLC
			2009	Second-line GBM
			2009	Metastatic RCC
			2013	Second-line metastatic CRC (after prior bevacizumab-containing regimen)
Ziv-aflibercept (Zaltrap, VEGF Trap)	Anti-VEGF mAB	VEGFA, VEGFB, PlGF1, PlGF2	2012	Metastatic CRC (after prior oxaliplatin-containing regimen)
Sorafenib (Nexavar, BAY439006)	Small-molecule TKI	VEGFR2, VEGFR3, PDGFR, FLT3, c-Kit	2005	Advanced RCC
			2007	Unresectable HCC
			2013	RAI-refractory DTC
Sunitinib (Sutent, SU11248)	Small-molecule TKI	VEGFR1, VEGFR2, VEGFR3, PDGFR, FLT3, c-Kit, RET	2006	Imatinib-resistant or -intolerant GIST
			2006	Advanced RCC
			2011	Advanced pNET
Pazopanib (Votrient)	Small-molecule TKI	VEGFR1, VEGFR2, VEGFR3, PDGFR, Itk, Lck, c-Fms	2009	Advanced RCC
			2012	Advanced soft tissue sarcoma
Vandetanib (Caprelsa)	Small molecule	RET, VEGFR, EGFR, BRK,	2011	Advanced MTC

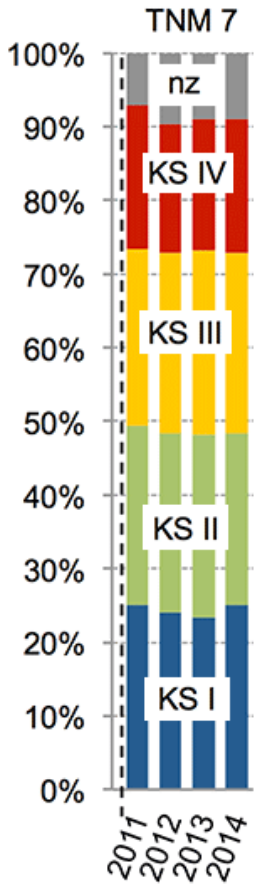
Molecular Biology of Cancer: Angiogenesis-I

Axitinib (Inlyta)	Small molecule TKI	VEGFR1, VEGFR2, VEGFR3	2012	Advanced RCC (after failure of prior therapy)
Cabozantinib (XL184, Cometriq)	Small molecule TKI	MET, VEGFR2, RET, KIT, AXL, FLT3	2012	Progressive, metastatic MTC
Regorafenib (Stivarga)	Small molecule TKI	RET, VEGFR1, VEGFR2, VEGFR3, TIE2, KIT, PDGFR	2012 2013	Previously treated metastatic CRC GIST
Temsirolimus (Torisel)	mTOR inhibitor	mTOR	2007	Advanced RCC
Everolimus (Afinitor, RAD-001)[a]	mTOR inhibitor	mTOR	2009 2010 2011 2012 2012	Second-line advanced RCC (after VEGFR TKI failure) SEGA associated w/TSC pNET Advanced HR+, HER2- breast cancer AML associated w/TSC

Molecular Biology of Cancer: others

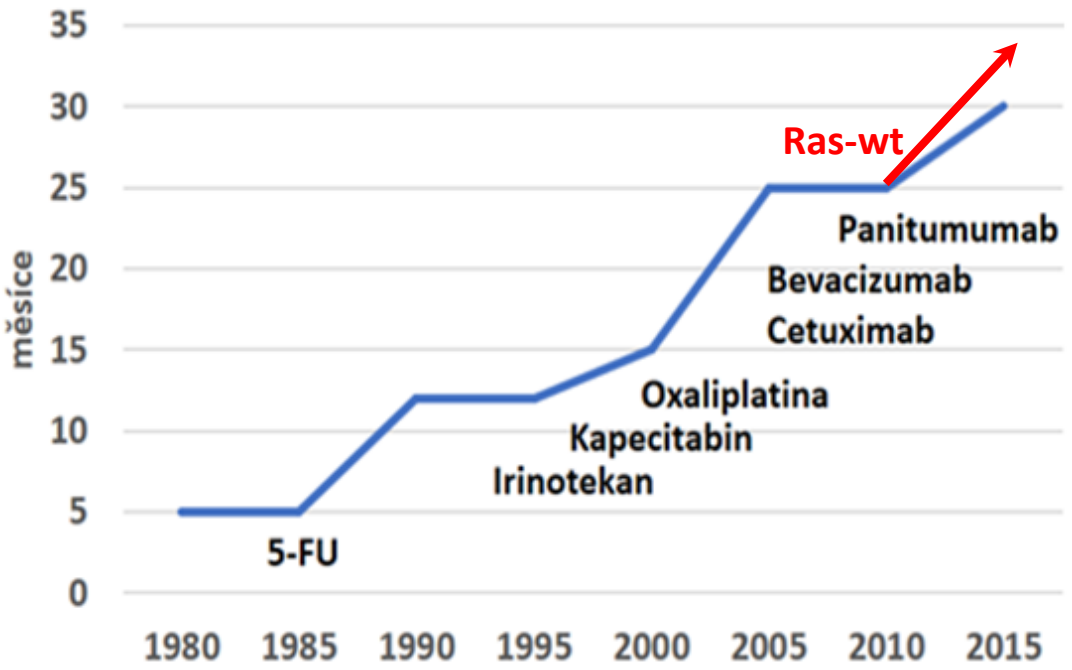
Drug	Class
Cetuximab Panitumumab Trastuzumab	EGFR/HER monoclonal antibodies
Gefitinib Erlotinib	EGFR small-molecule tyrosine–kinase receptor inhibitors
Everolimus Temsirolimus	mTOR inhibitors
Thalidomide Lenalidomide Pomalidomide	Immunomodulatory agents
Belinostat (PXD101) LBH589 Vorinostat (SAHA)	HDAC inhibitors
Celecoxib	COX-2 inhibitors
Bortezomib	Proteasome inhibitors
Zoledronic acid	Bisphosphonates
Rosiglitazone	PPAR-γ agonists
Doxycycline	Antibiotic

Colorectal cancer



5 % → 14 %

Overall survival of patients with metastatic colorectal carcinoma

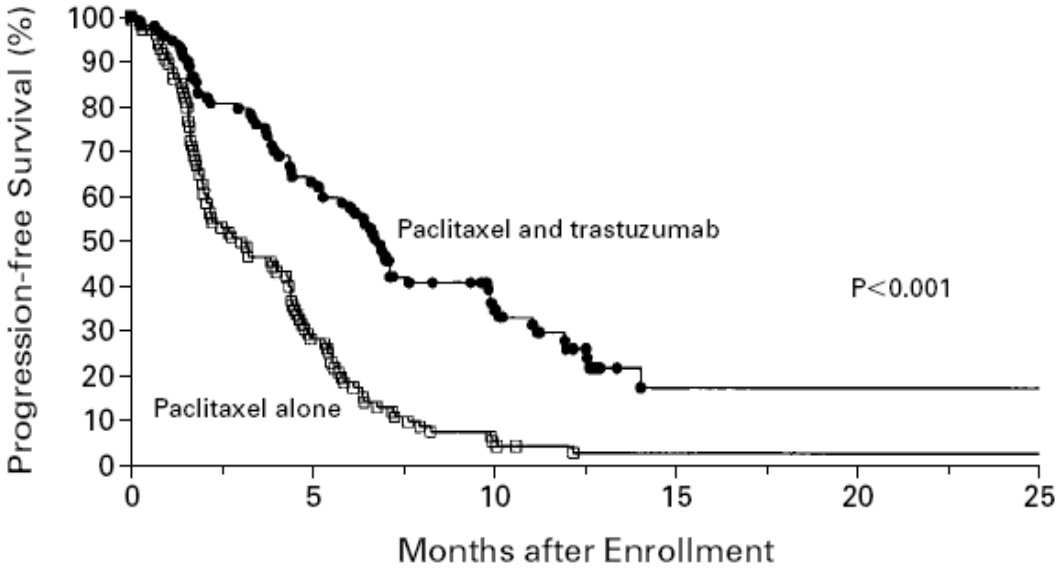


Palliative care, including targeted treatment, has led to prolongation median survival from 5 to 30 months

SEER Cancer Statistics 1975-2000, 2000 – 2013 Gustavsson et al., Clinical Colorectal Cancer 2015 (14),1:1-10

Targeted anti-HER2 therapy breast cancer

1. Line of palliative therapy (metastatic breast cancer)



Variable	Chemotherapy plus Trastuzumab (N=235)	Chemotherapy Alone (N=234)	Paclitaxel and Trastuzumab (N=92)	Paclitaxel Alone (N=96)
Complete and partial responses — no. (% [95% CI])	118 (50 [44–57])	74 (32 [26–38])	38 (41 [31–51])	16 (17 [9–24])
P value	<0.001		<0.001	
Median duration of response — mo	9.1	6.1	10.5	4.5
P value	<0.001		<0.01	

Smith et al. Anticancer Drugs 2001