Cancer

SBL101

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All Figures in this Lecture are taken from

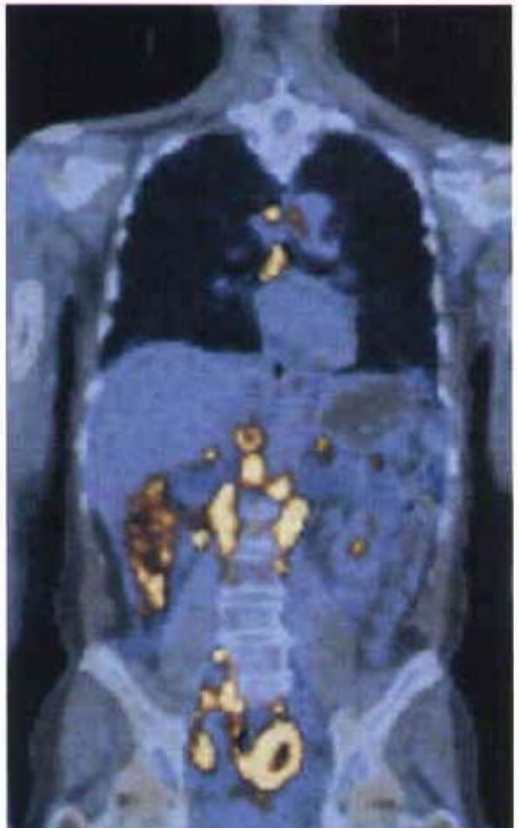
1. Molecular biology of the cell / Bruce Alberts et al., 5th ed.

2. Research papers as cited OR

3. Constructed

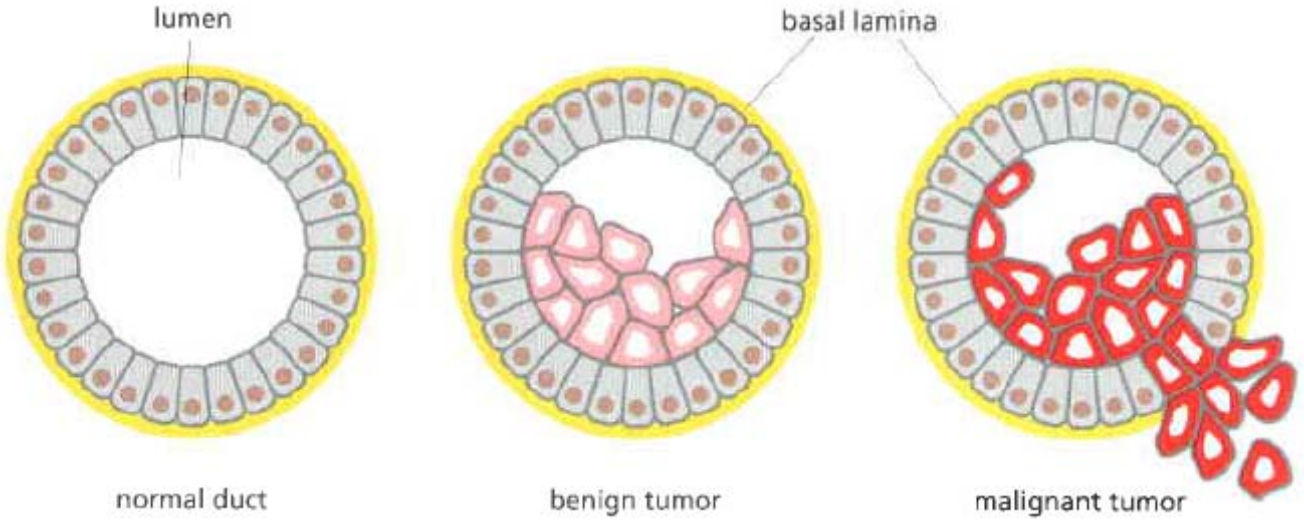
Basic property of cancer cells

- Cancer cells possess two heritable properties
  - Reproduce in defiance to normal constraints on growth and proliferation
  - Invade and colonize territories reserved for other cells
- The abnormal cells give rise to a tumor or *neoplasm*
  - *Benign*
  - *Malignant*
    - *Metastases*



Metastasis in Non-Hodgkin Lymphoma. Fluorodeoxyglucose shows up as yellow in regions of high glucose activity typical of tumor cells.

Cancer originates from a single abnormal cell

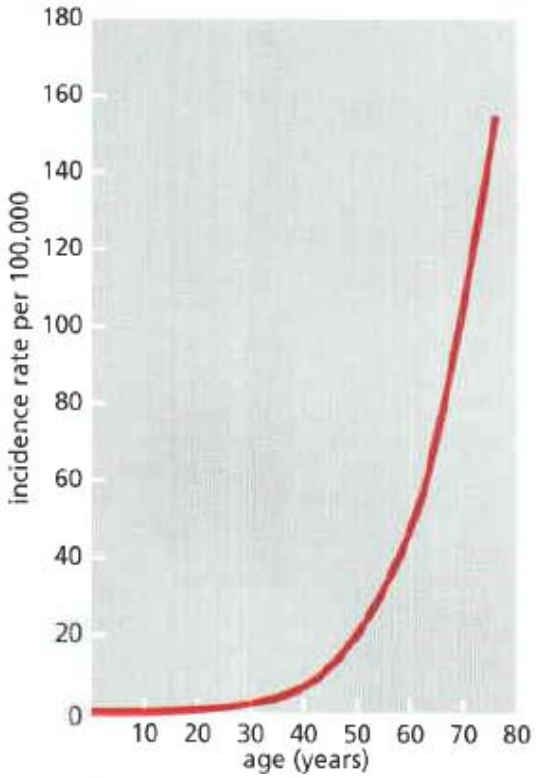


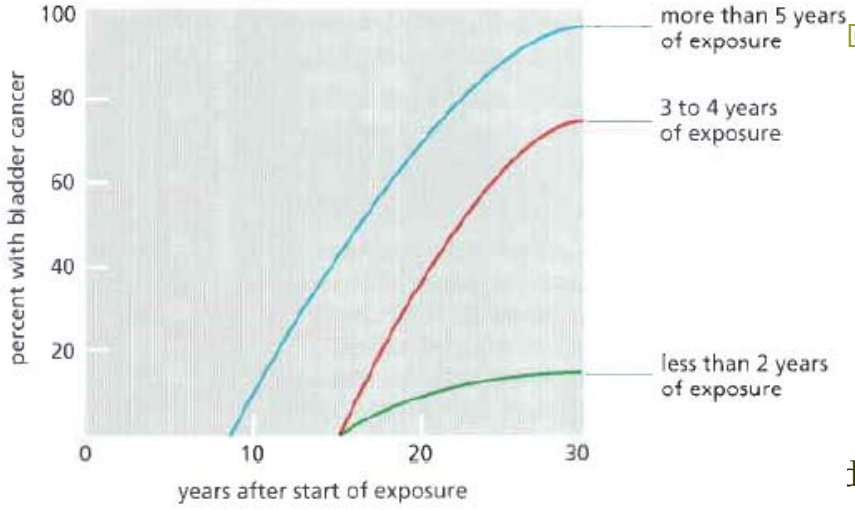
- Cancer develops from a single cell that has acquired a heritable change
  - This is passed on to its descendents allowing them to outgrow, out-divide and out-live their neighbors
  - By the time these cells are detected, there are about a billion of them

Causes of Cancer

- Genetic and Epigenetic changes
- Carcinogenesis
  - Chemical Carcinogenesis
  - Radiation Carcinogenesis
- Genetic defect in DNA repair mechanisms
  - People with *xeroderma pigmintosum* are more prone to cancer
  - Mice lacking certain DNA repair genes are abnormally prone to cancer

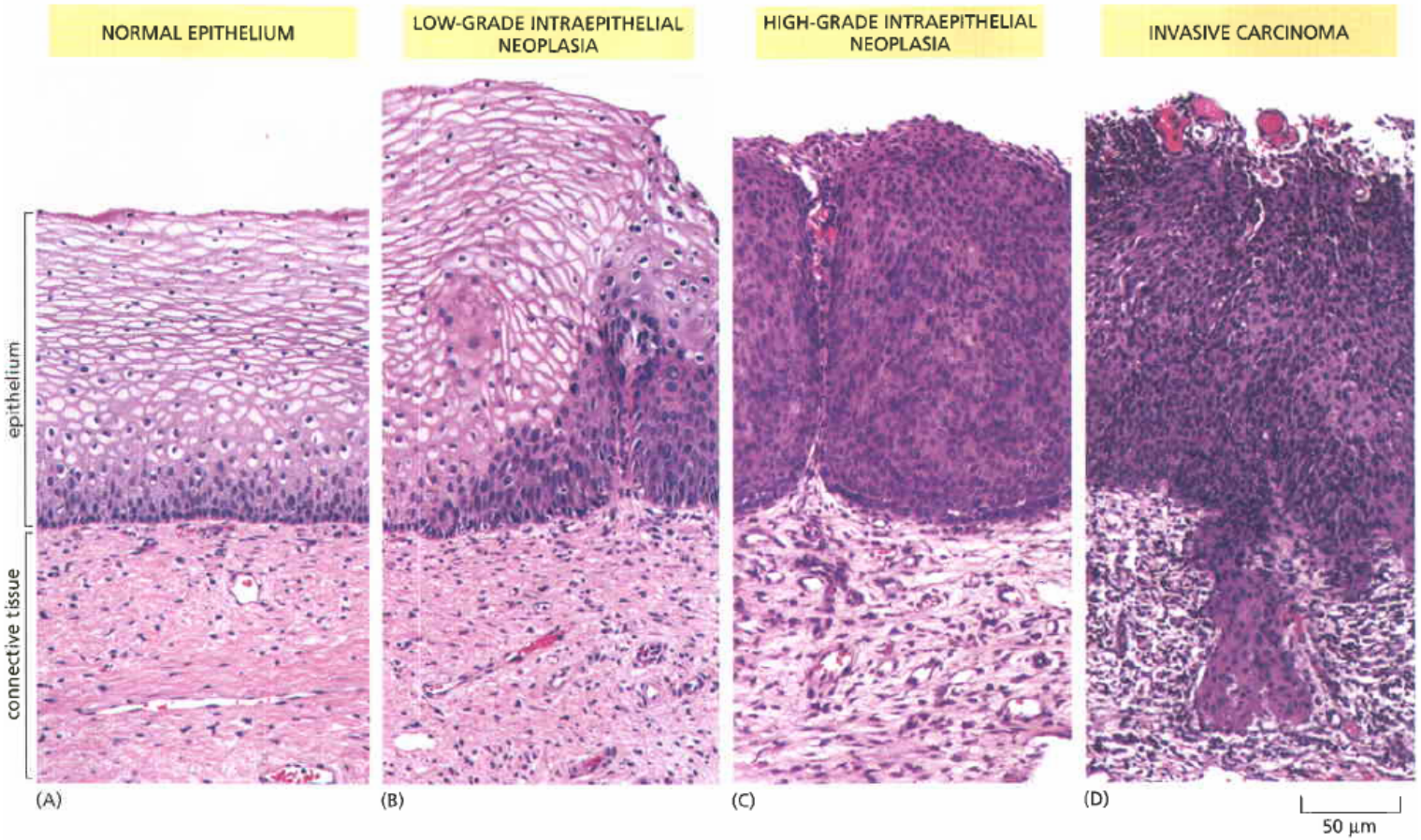
Mutations and Cancer





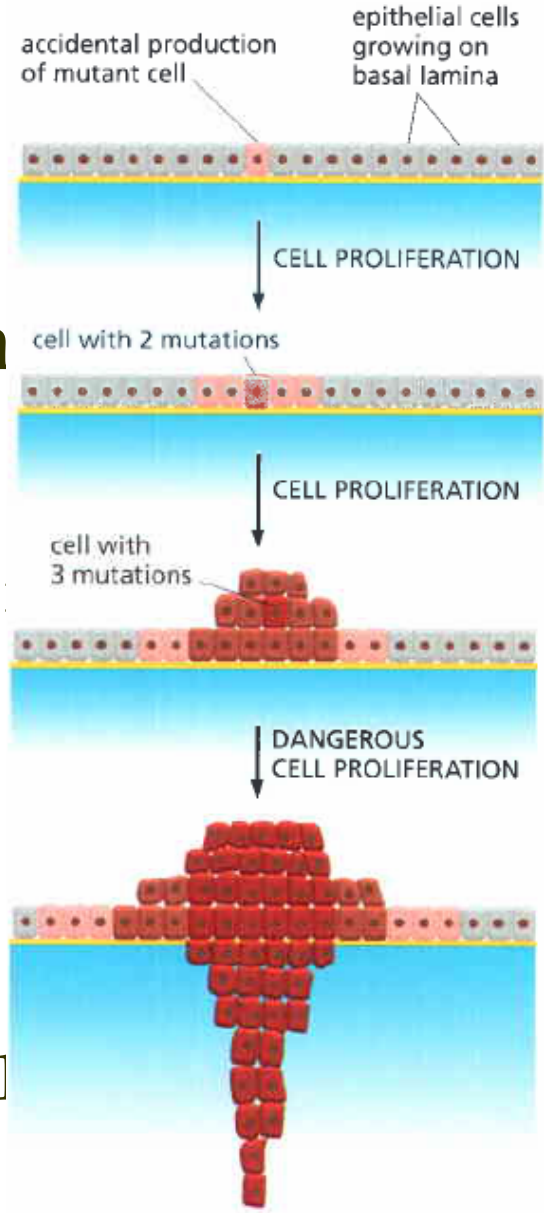
- It is estimated that 10^{16} cell divisions occur in a human body in one lifetime
- Error rate of DNA coding 10^{-6}
  - Every gene is likely to have undergone 10^{10} mutations
- So why does cancer occur so *In-frequently?*
  - It means that a number of *rare genetic* accidents must occur in the lineage of one cell i.e. progressive accumulation of random mutations in a single lineage of cells

Progression of cervical cancer



Tumor progression

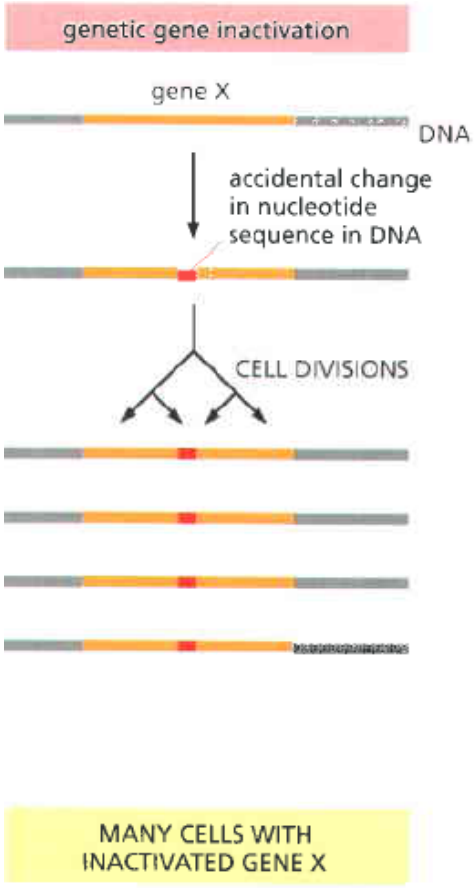
- Involves succession of random inherited changes followed by natural selection
  - At each stage the cells acquire a mutation or epigenetic change
  - The environment inside a tumor harsh and inhibits the growth of normal cells
- Is this expected?
  - Higher organisms have stringent gene regulation
  - The cells have to cross these level of regulation

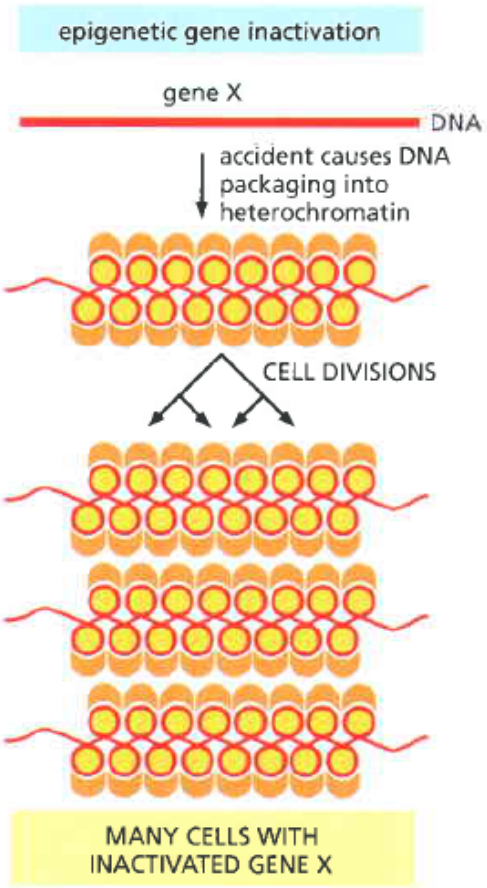


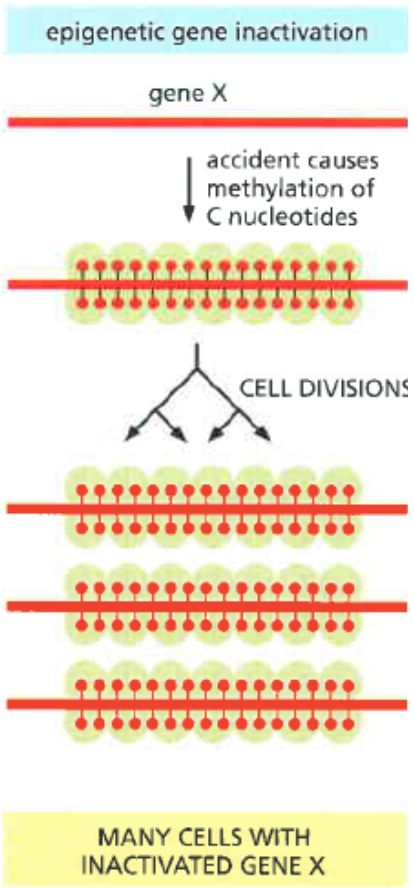
Epigenetic Changes: Inherited chromatin structure

- Cells are identified by abnormal appearance in tumor biopsies
  - Contain unusual amount of heterochromatin
  - Associated with gene silencing
  - Genes are switched off in a cell-to-cell inherited manner
- In reality this is the same principle by which fetus grows in higher animals

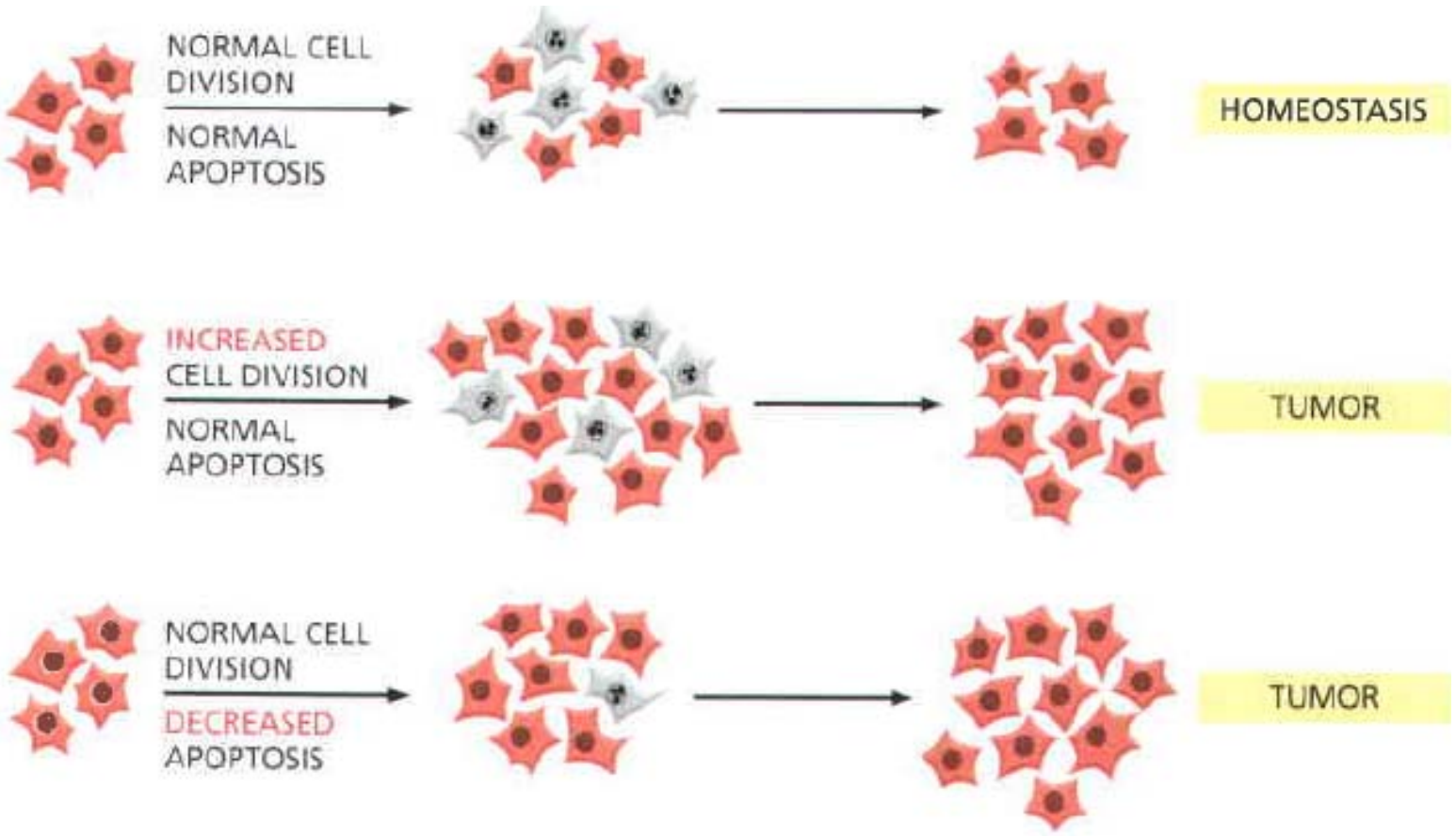
Genetic & Epigentic changes







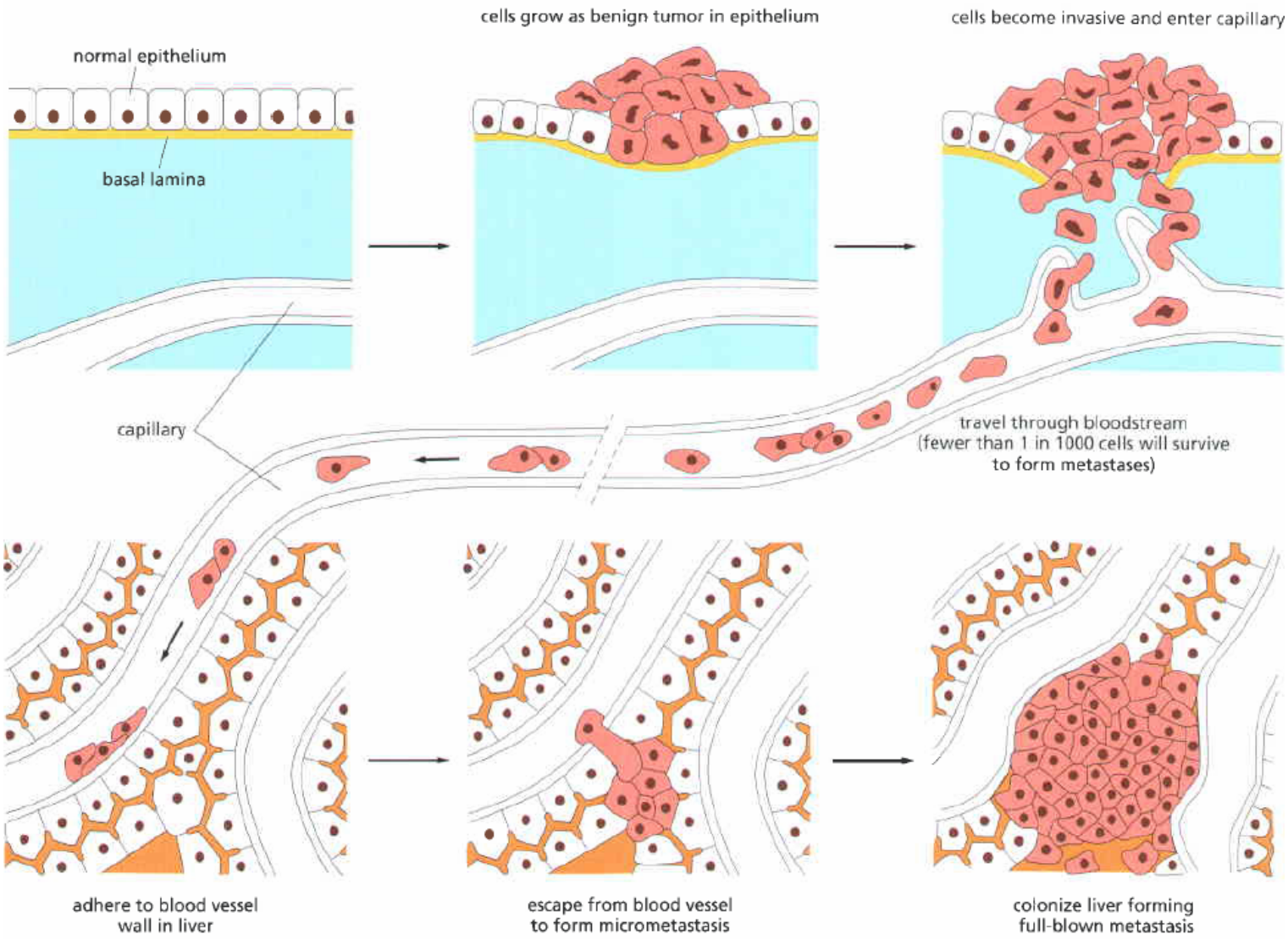
Defective Control of Programmed Cell Death



Escapes limits of replication

- Normal "primary" cells proliferate in culture, but soon stop dividing after a number of dividing cycles – replicative senescence
  - Cell division counting mechanism depends on the shortening of *telomeres*
  - Cells have the enzyme *telomerase,* the promoted the formation of protein caps to protect the ends of the chromosomes
  - Many proliferation cells (with the exception of *stem cells*) are deficient in telomerase and so it ultimately results in the cell cycle arrest of the cell
- Cancer cells either
  - Block the control point so that cell cycle continues in the absence of telomeres
  - Acquire/maintain telomerase activity to continue indefinite cell dividion

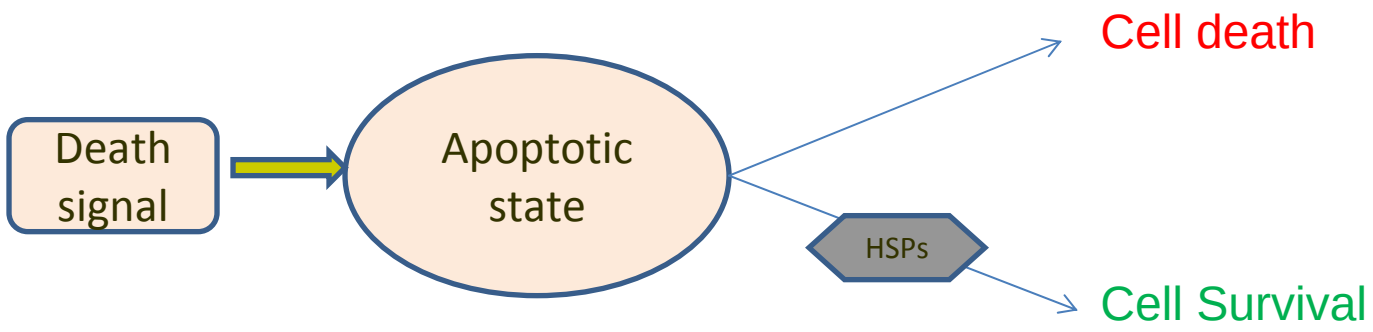
Tumors induce angiogenesis

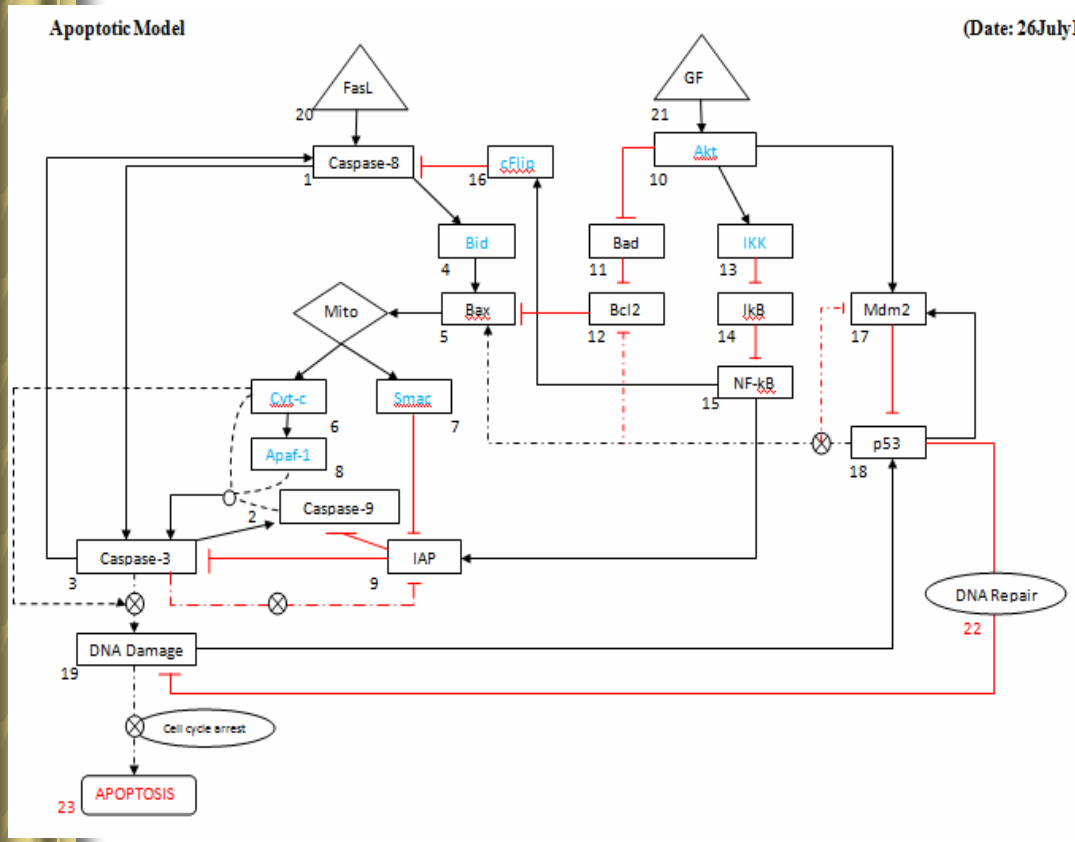


Viruses can cause cancer

VIRUS	ASSOCIATED CANCER	AREAS OF HIGH INCIDENCE
DNA viruses		
Papovavirus family		
Papillomavirus (many distinct strains)	warts (benign)	worldwide
	carcinoma of the uterine cervix	worldwide
Hepadnavirus family		
Hepatitis-B virus	liver cancer (hepatocellular carcinoma)	Southeast Asia, tropical Africa
Hepatitis-C virus	liver cancer (hepatocellular carcinoma)	worldwide
Herpesvirus family		
Epstein–Barr virus	Burkitt's lymphoma (cancer of B lymphocytes)	West Africa, Papua New Guinea
	nasopharyngeal carcinoma	Southern China, Greenland
RNA viruses		
Retrovirus family		
Human T-cell leukemia virus type I (HTLV-1)	adult T-cell leukemia/ lymphoma	Japan, West Indies
Human immuno-deficiency virus (HIV, the AIDS virus)	Kaposi's sarcoma	Central and Southern Africa

Analysis of stable states in Apoptotic pathways dependent upon the presence or absence of heat shock proteins for predicting drug targets





p53_excess = p53(t) & p53(t-1)

Casp3_excess = casp3(t) & casp3(t-1)

DNA damage(DD) = DD(t+1) &DD(t) & DD(t-1)

Boolean Rules:

1. Casp8 = FasL – cFlip + casp3 – HSP70
2. Casp9 = casp8 + casp3 –IAP
3. Casp3 = (Casp9 AND Apaf-1 AND Cyt-c) – IAP
4. Bid = Casp8 – HSP27
5. Bax = Bid –Bcl2 + p53_excess – HSP70 – HSP40
6. Cyto-c = Bax – HSP27
7. Smac = Bax – HSp27
8. Apaf-1 = cyt-c – HSP70 –HSP90
9. IAP = -Smac + Nf-kB –casp3_excess
10. AKT = GF + HSP27 + HSP90
11. Bad = Not(Akt) - Akt
12. Bcl2 = Not(Bad) – Bad - p53_excess
13. IKK = Akt
14. IkB = Not(IKK) + Nf-kB – IKK
15. Nf-kB = Not(IkB) - IkB
16. cFlip = Nf-kB
17. Mdm2 = (Akt or p53) – p53_excess
18. P53 = DNA Damage –Mdm2
19. DNA damage = Casp3 - p53 + Casp3_excess
20. FasL = [1,0]
21. Growth factor = [1,0]
22. DNA Reapir = p53
23. APOPTOSIS = DNA damage_excess

Hypothesis

(i) Apoptosis regulated at multiple steps, cFLIP is the first level, IAP is second and Bcl2 at third level.

(ii) Inhibition of Bcl2 signaling induces apoptosis.

(iii) Bcl2 should be constitutively activated concurrently with p53 is essential for the long-term survival of cells and to reproduce cancer phenotype.

(iv) Most stable state in pathway is where cFLIP, IAP, and Bcl2 are ON.

(v) HSPs can prevent apoptotic pathway.

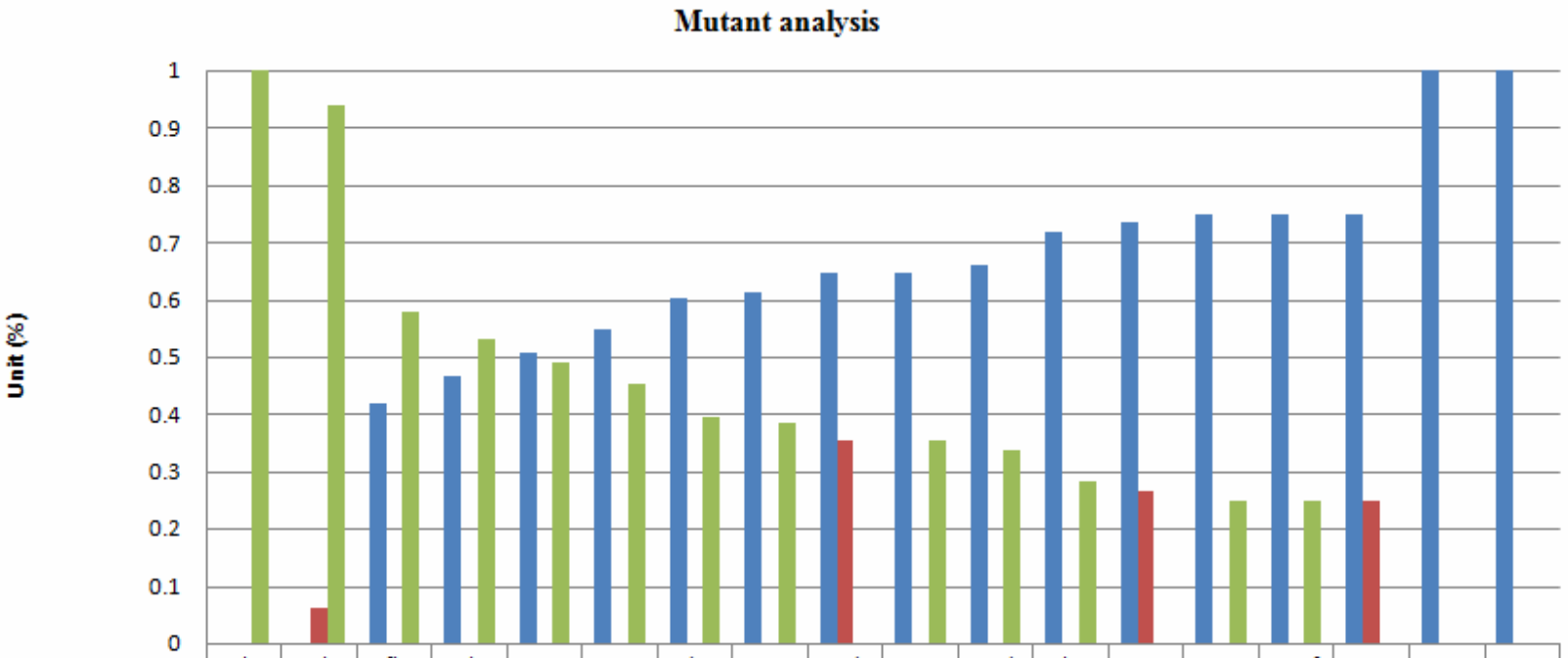
Stable states

						Transient												
	Survival											Death						
Casp8		x		x	x							x	x	x	x	x	x	
Casp9					x	x	x		x		x	x	x	x	x	x	x	
Casp3						x	x					x	x	x	x	x	x	
Bid		x		x	x							x	x	x	x	x	x	
Bax				x	x	x	x	x	x	x	x	x	x	x	x	x	x	
Cytc				x	x	x	x	x	x	x	x	x	x	x	x	x	x	
Smac				x	x	x	x	x	x	x	x	x	x	x	x	x	x	
Apaf				x	x	x	x	x	x	x	x	x	x	x	x	x	x	
IAP	x	x	x	x	x			x	x	x	x							
Akt	x	x	x	x	x	x	x	x	x	x	x	x	x	x			x	
Bad															x	x		
Bcl2	x	x	x	x	x		x	x	x	x	x			x			x	
IKK		x	x	x	x	x	x	x	x	x	x	x	x	x			x	
IkB	x														x	x		
NfkB	x	x	x	x	x	x	x	x	x	x	x	x	x	x			x	
cFlip	x	x	x	x	x	x	x	x	x	x	x	x	x	x			x	
Mdm2	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x	
P53				x	x	x	x	x	x	x	x	x	x	x	x	x	x	
DD*				x	x	x	x	x	x	x	x	x	x	x	x	x	x	
FasL		x	x	x	x					x	x	x				x	x	
GF	x	x	x	x	x	x	x	x	x	x	x	x	x	x			x	
	0	0	0	10	10	0	0	0	0	0	0	11	11	11	11	11	11	
	7025	1186675	7027	1309567	1833855	911741	912253	129917	654205	129919	654207	2091391	2091389	2091901	2090140	2090142	2091903	

Transient

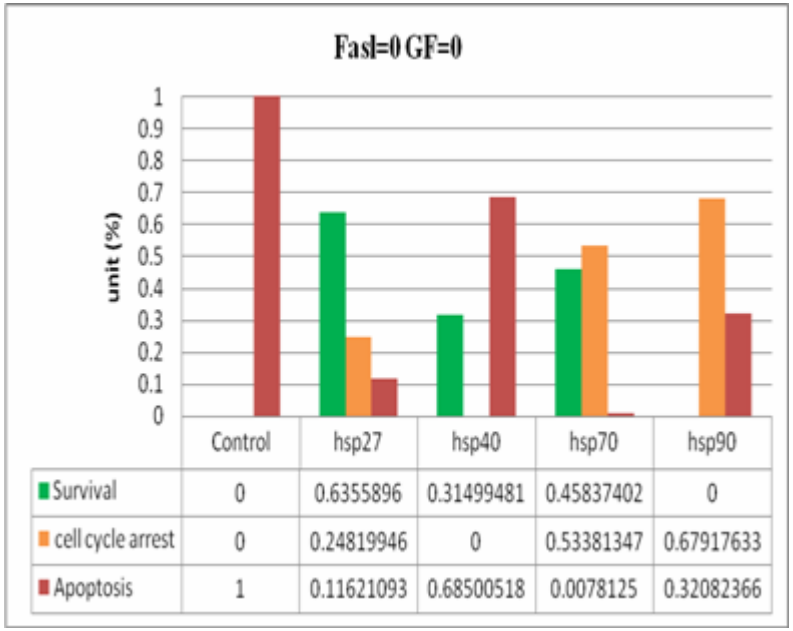
Survival		Death	
7025	911741	2091391	2091901
1186675	912253	2091389	2090140
7027	129917		2090142
1309567	654205		2091903
1833855	129919		
	654207		

Node Mutation

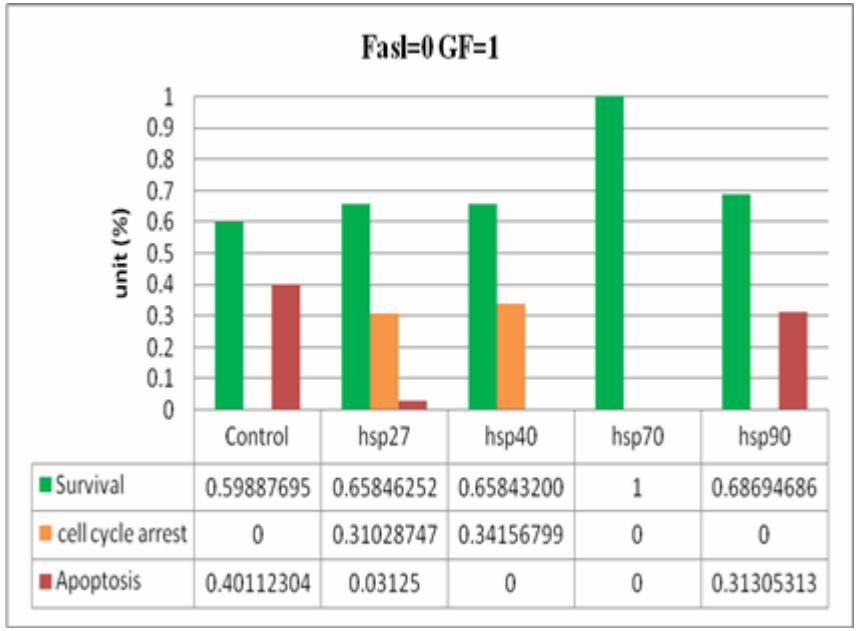


	Akt	Bcl2	NfkB	cFlip	IKK	IAP	Mdm2	P53	Bid	Smac	Bad	IkB	Bax	Casp9	Apaf	Cytc	Casp8	Casp3
Survival	0	0	0.42	0.468	0.508	0.548	0.603	0.613	0.647	0.647	0.661	0.718	0.734	0.75	0.75	0.75	1	1
Cell cycle arrest	0	0.061	0	0	0	0	0	0	0.352	0	0	0	0.265	0	0	0.25	0	0
Apoptosis	1	0.938	0.58	0.531	0.491	0.451	0.396	0.386	0	0.352	0.338	0.281	0	0.25	0.25	0	0	0

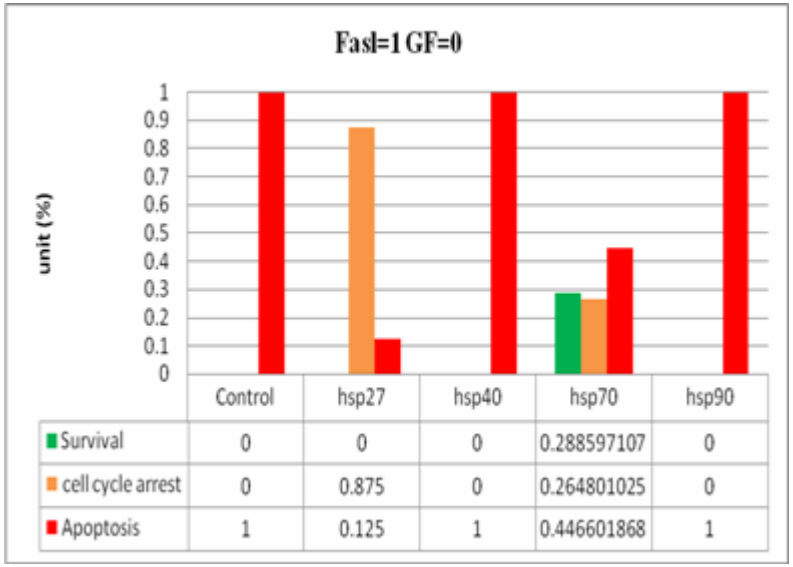
HSPs



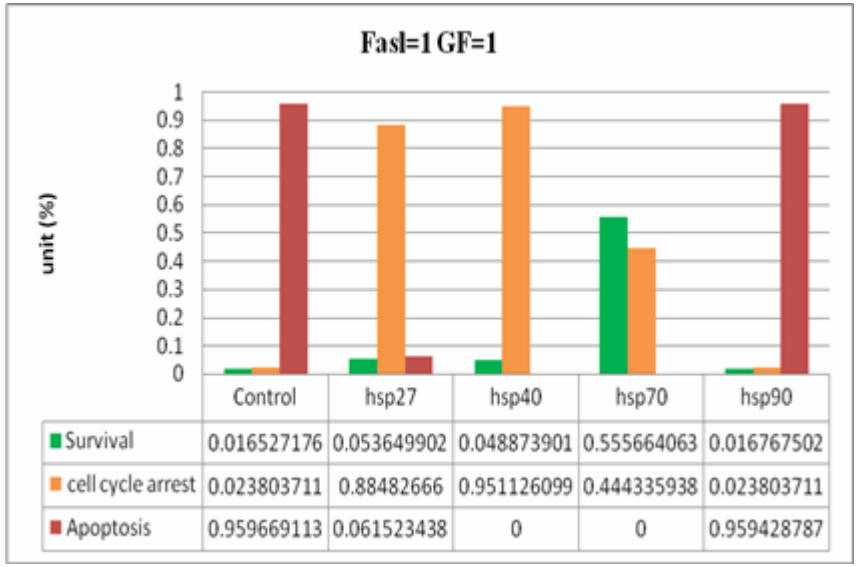
	Control	hsp27	hsp40	hsp70	hsp90
Survival	0	0.6355896	0.31499481	0.45837402	0
cell cycle arrest	0	0.24819946	0	0.53381347	0.67917633
Apoptosis	1	0.11621093	0.68500518	0.0078125	0.32082366



	Control	hsp27	hsp40	hsp70	hsp90
Survival	0.59887695	0.65846252	0.65843200	1	0.68694686
cell cycle arrest	0	0.31028747	0.34156799	0	0
Apoptosis	0.40112304	0.03125	0	0	0.31305313



	Control	hsp27	hsp40	hsp70	hsp90
Survival	0	0	0	0.288597107	0
cell cycle arrest	0	0.875	0	0.264801025	0
Apoptosis	1	0.125	1	0.446601868	1



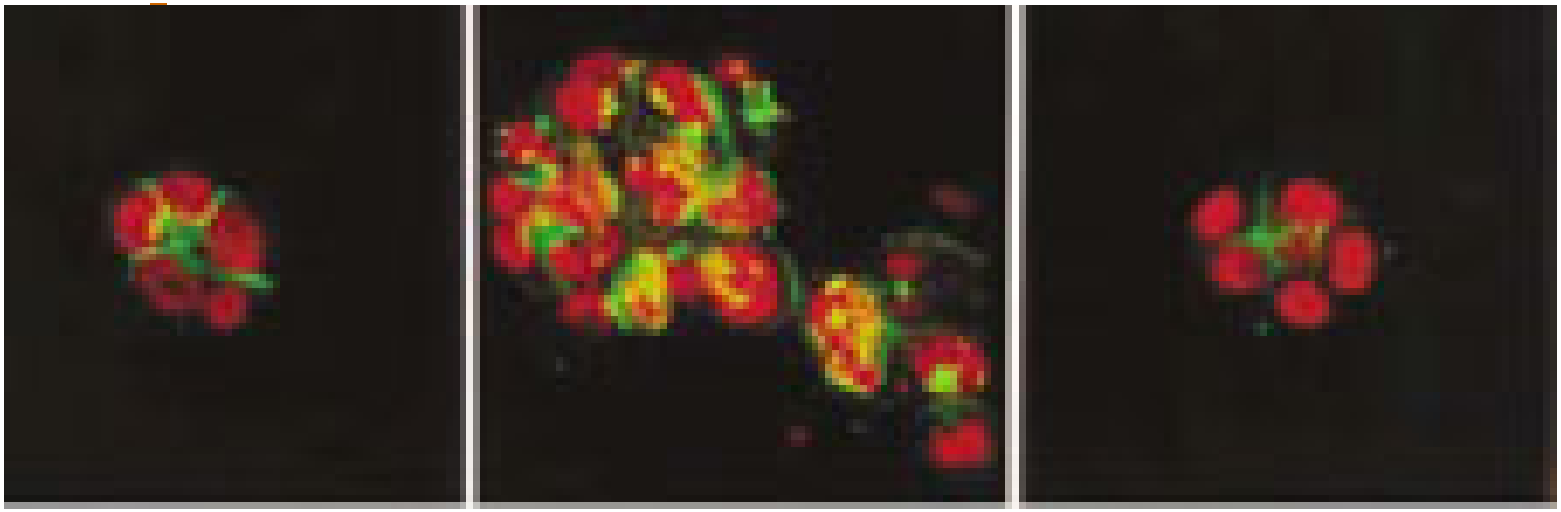
	Control	hsp27	hsp40	hsp70	hsp90
Survival	0.016527176	0.053649902	0.048873901	0.555664063	0.016767502
cell cycle arrest	0.023803711	0.88482666	0.951126099	0.444335938	0.023803711
Apoptosis	0.959669113	0.061523438	0	0	0.959428787

Experimental methods

Differences between 2-D and 3-D cultures

- Phenotype and morphology
- Metabolic and gene expression pattern
  - Tumor cells grown in 3-D show pronounced resistance to drugs as observed *in vivo*
  - Alters pluripotency of stem cells
  - Responsible for developmental changes (which means this area of research cannot be carried out in 2-D)
  - Spatio-temporal consistency of information and heirarchy

Variability in 2-D & 3-D

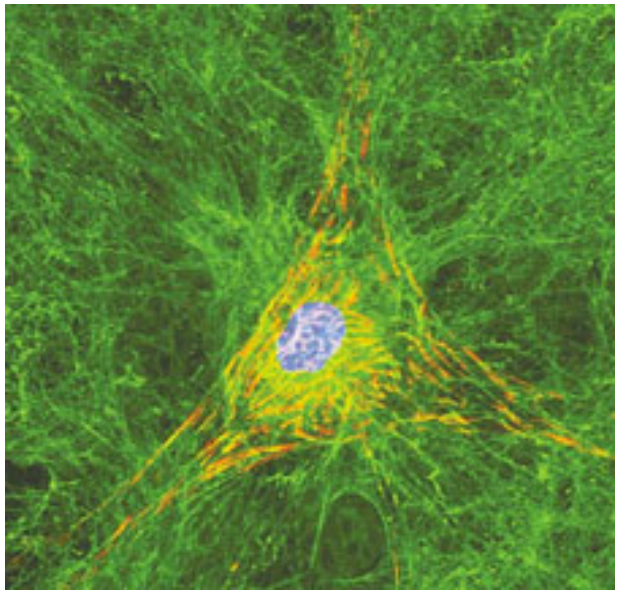


Role reversal: unlike in 2-D cultures, breast tumour cells in 3-D culture (left) that become malignant (centre) can be made to revert to their original state (right) when an antibody against β1-integrin is added to the system.

M. BISSELL, the Lawrence Berkeley National Laboratory in California

Testing Cancer drugs

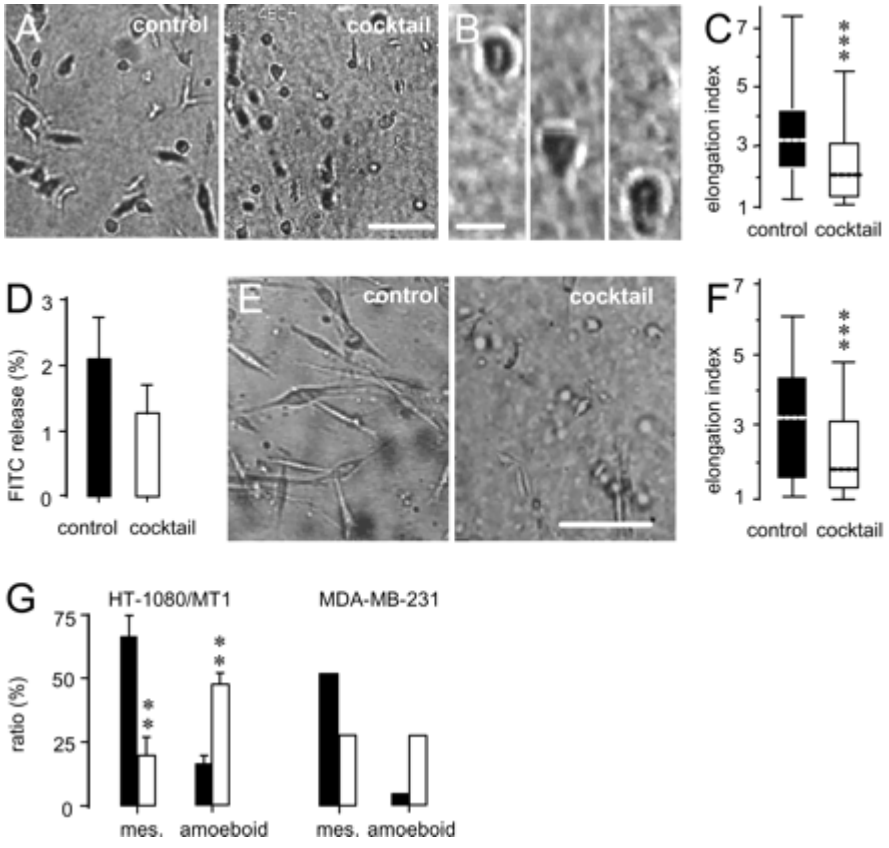
- Receptors for growth factors play a key role in the initial development of tumours
  - the migration of cells away from primary tumours to cause secondary cancers around the body
- Cancer cells undergoing metastasis normally cut themselves free from a tumour's ECM using protein-digesting enzymes
  - formation of amoeba-like cells depends on a particular signalling pathway in a range of different tumour cell lines



Joined up: a cell in a 3-D culture forming links by means of β1-integrin (orange) with the scaffolding.

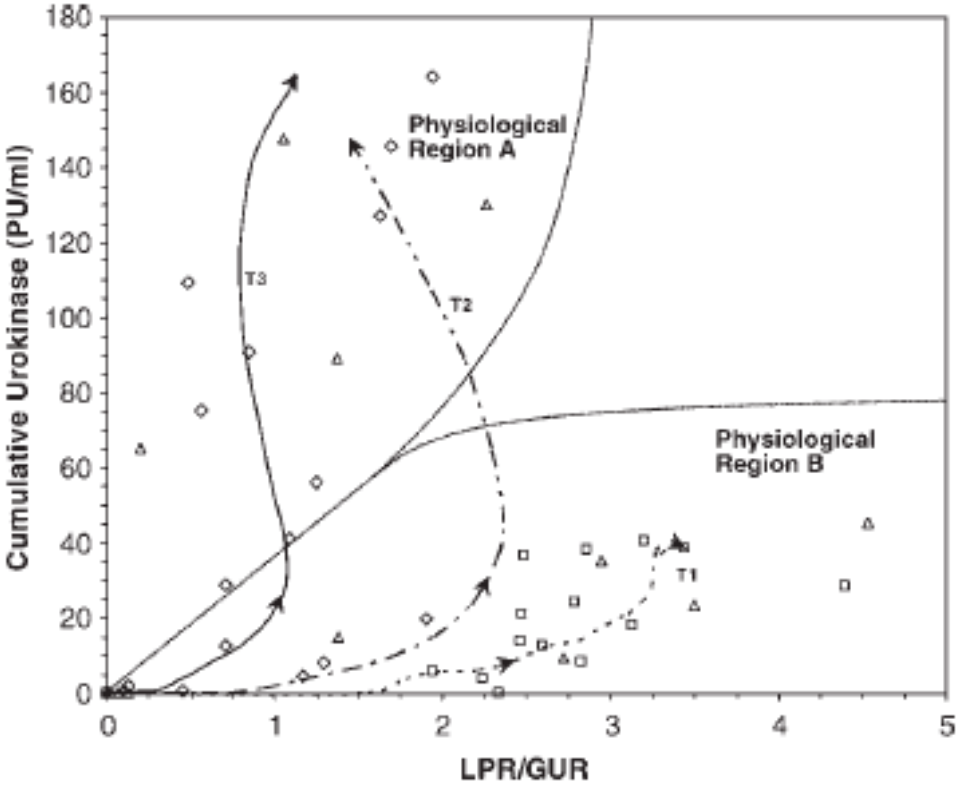
Changes in Morphology : Metastatis

- The invasion and migration of tumor cells involves coordinated adhesion as well as proteolytic interaction with the ECM substrate, resulting in the degradation and remodeling of interstitial tissue barriers

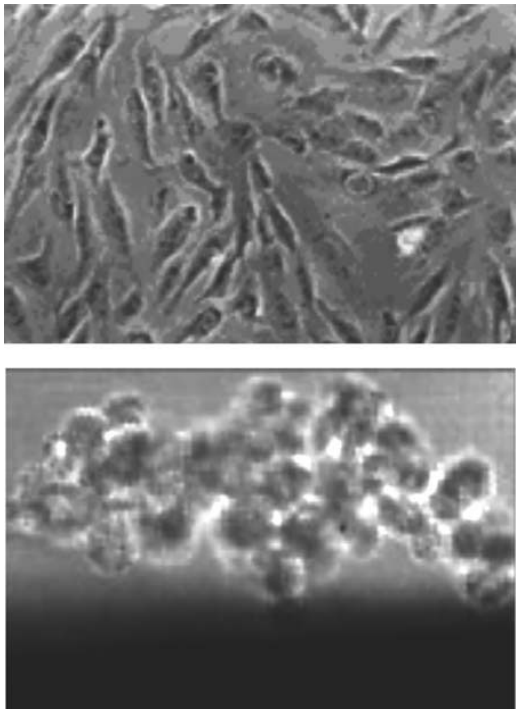


Transition of spindle-shaped (mesenchymal) to more spherical (amoeboid) migration in HT1080/MT1 and MDA-MB-231 cells for migration (metastatis) Wolf et al. JCB 2003 160 (2): 267

Change in Morphology of HT 1080 in Hollow Fiber Reactor



Trajectories showing the course of physiological states during the external modulation of HFR for urokinase production



S. S. Khaparde, P. K. Roychoudhury, J. Gomes*, A. Mukhopdhyay, Biotechnol. Prog. 2008, 24, 1325

- Adhered cells produce urokinase
- Amoeboidal cells migrate