Inflammation and carcinogenesis

Prof. Barry Campbell

Infection Biology & Microbiomes, IVES

bjcampbl@liverpool.ac.uk

https://pcwww.liv.ac.uk/~bjcampbl/Inflammation and cancer.htm

Learning objectives

- **LO1 - How does inflammation contribute to carcinogenesis?**

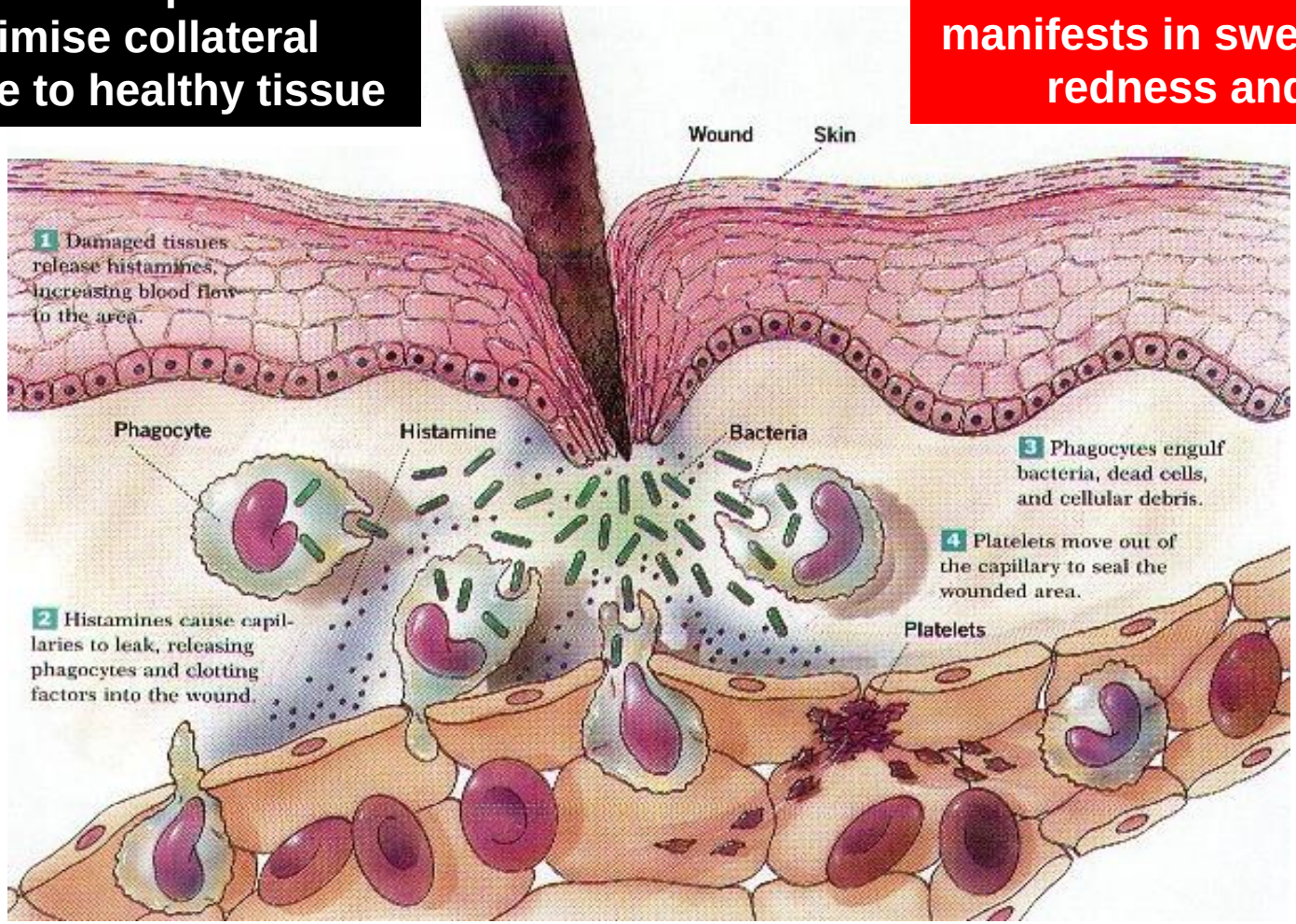
- **LO2 - How do infectious agents (such as the bacterium *H. pylori*) promote cancer?**

LO1

Inflammation is normal:
It is part of the our immune response

Brutal and quick to minimise collateral damage to healthy tissue

From the outside, this manifests in swelling, heat, redness and pain



Repair teams move in to direct healing.

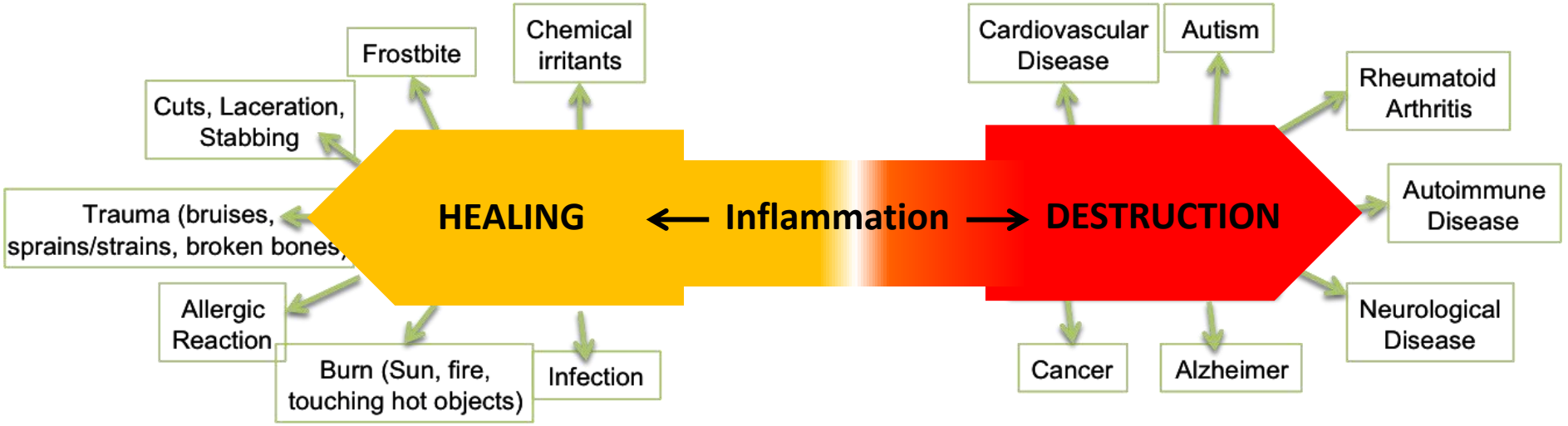
Blood vessels sprout.

A scab/plug forms and tissue cells grow

Before long, calm returns!
Signals urge victorious immune cells to return to base camp.

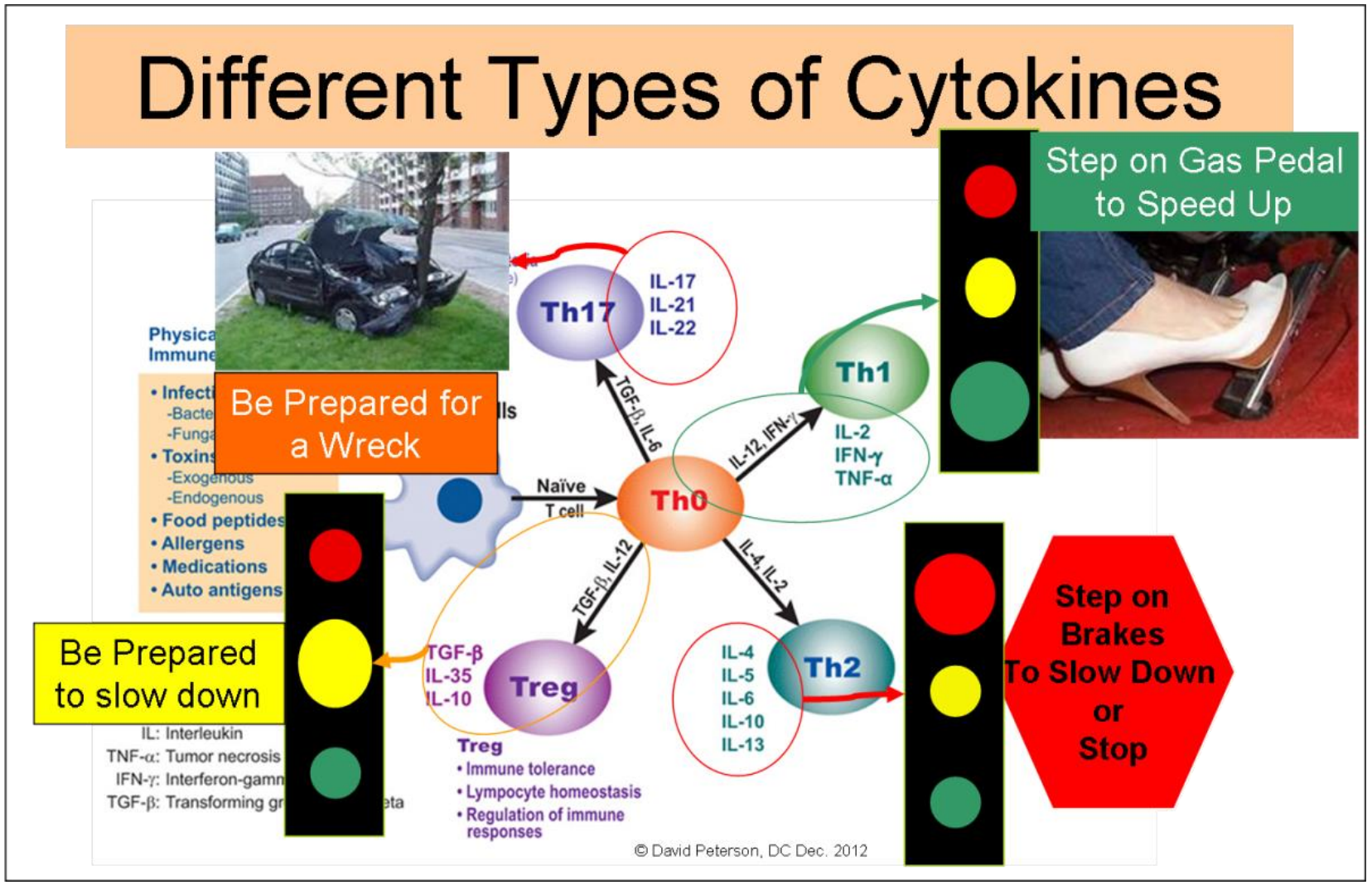
LO1

Inflammation - Where there is smoke, there is fire!



LO1

Inflammatory signals/messages – cytokine responses



LO1

Inflammation and cancer

RUDOLF LUDWIG KARL VIRCHOW (1821-1902)

1863 – **Rudolf Virchow was the first to ask whether inflammation might also contribute to cancer**

Virchow noted leukocytes in neoplastic tissue samples and made a connection between inflammation and cancer.

Balkwill & Mantovani 2001. *Lancet* 357:539-45

LO1

Hallmarks of cancer-related inflammation

- **Inflammatory cells and inflammatory mediators (e.g, chemokines, cytokines and prostaglandins) present in tumour tissues**

- **Tissue remodelling is similar to that seen in chronic inflammatory responses**

- **Angiogenesis is similar to that seen in tissue repair**
  (required for the survival of cells within tumours of a certain size & tumour metastasis)

- **Inflammatory cells and mediators are present in the tumour microenvironment**

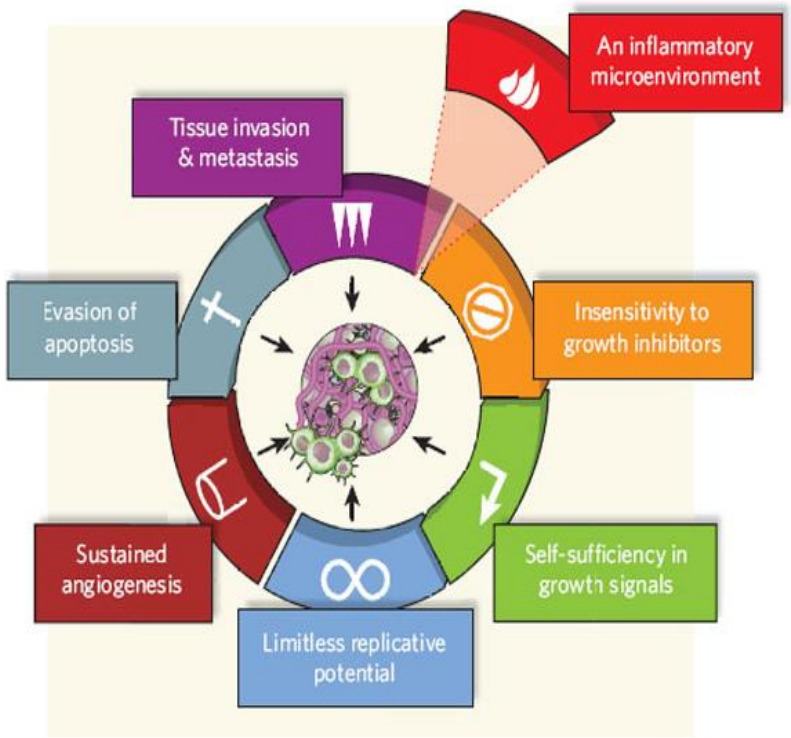
LO1

Molecular pathways associated with inflammation are also fundamental for cancer

1990 - Chronic IBD associated with an increased risk of colorectal cancer
Ekbom et al. *Lancet* 1990;336:357-9

2004 - NFκB functions as a tumour promoter in inflammation-associated liver cancer.
Pikarsky et al. *Nature* 2004;431:461-6

2004 - IKKβ/NFκB pathway links inflammation and tumorigenesis in colitis-associated cancer.
Greten et al. *Cell* 2004;118:285-96



2008 - Inflammation was suggested as a 7th hallmark of cancer to the 6 originally proposed

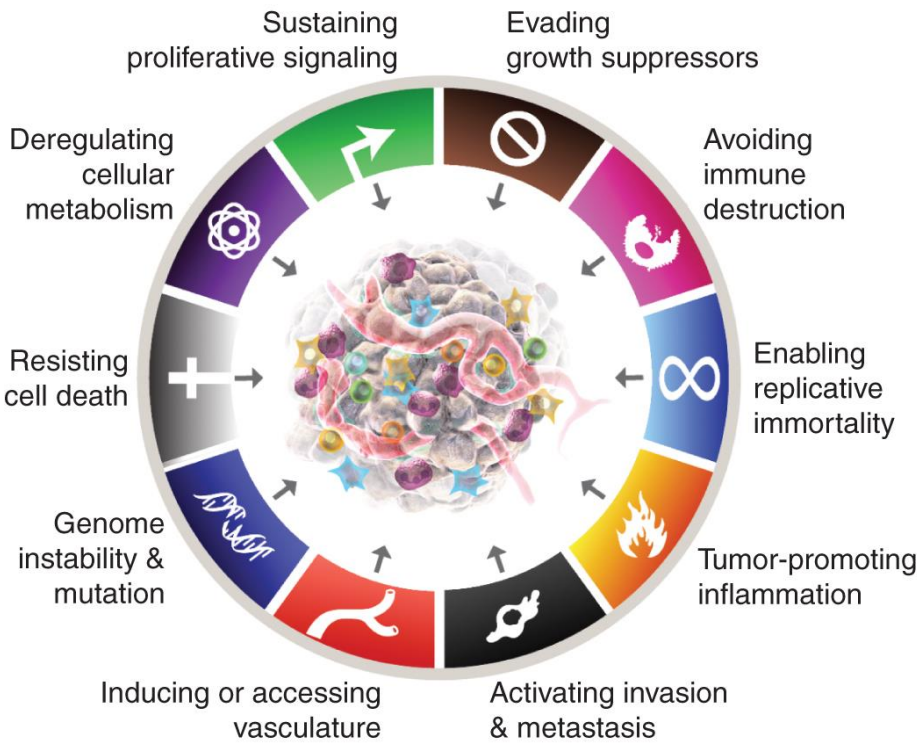
Hanahan & Weinberg. *Cell* 2000 ;100:57–70
Mantovani. *Nature* 2009; 457, 36-37

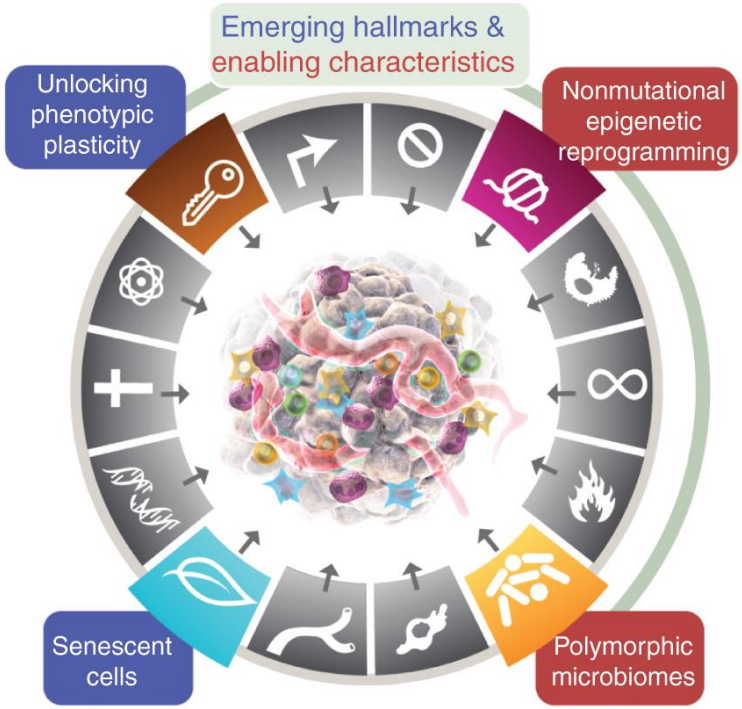
LO1

Molecular pathways associated with inflammation are also fundamental for cancer

2011 – **Tumour promoting inflammation firmly established as one of 10 Hallmarks of cancer** Hanahan & Weinberg. *Cell* 2011;144:646-74.

2021 - Hallmarks of Cancer: New Dimensions

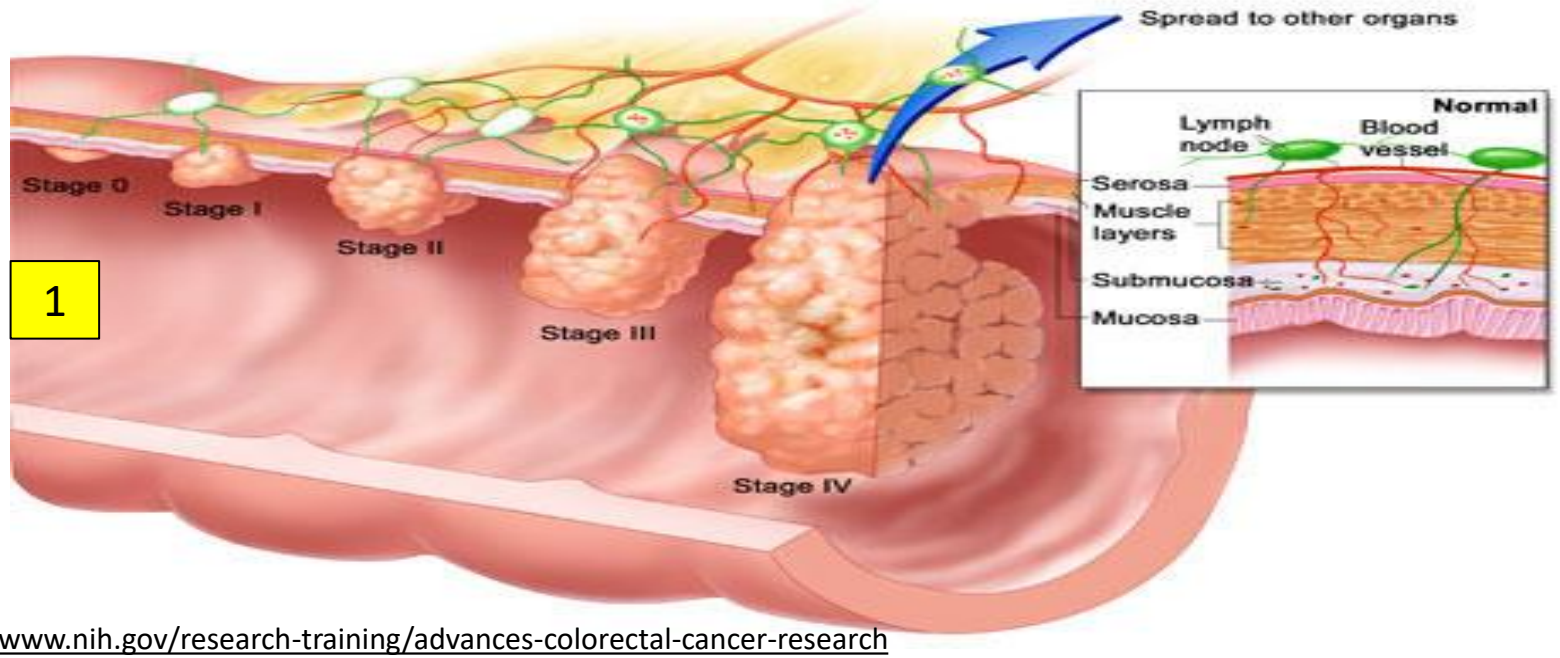




Hanahan, D. *Cancer Discov.* 2022;12(1):31-46.

LO1

So how does inflammation lead to cancer?

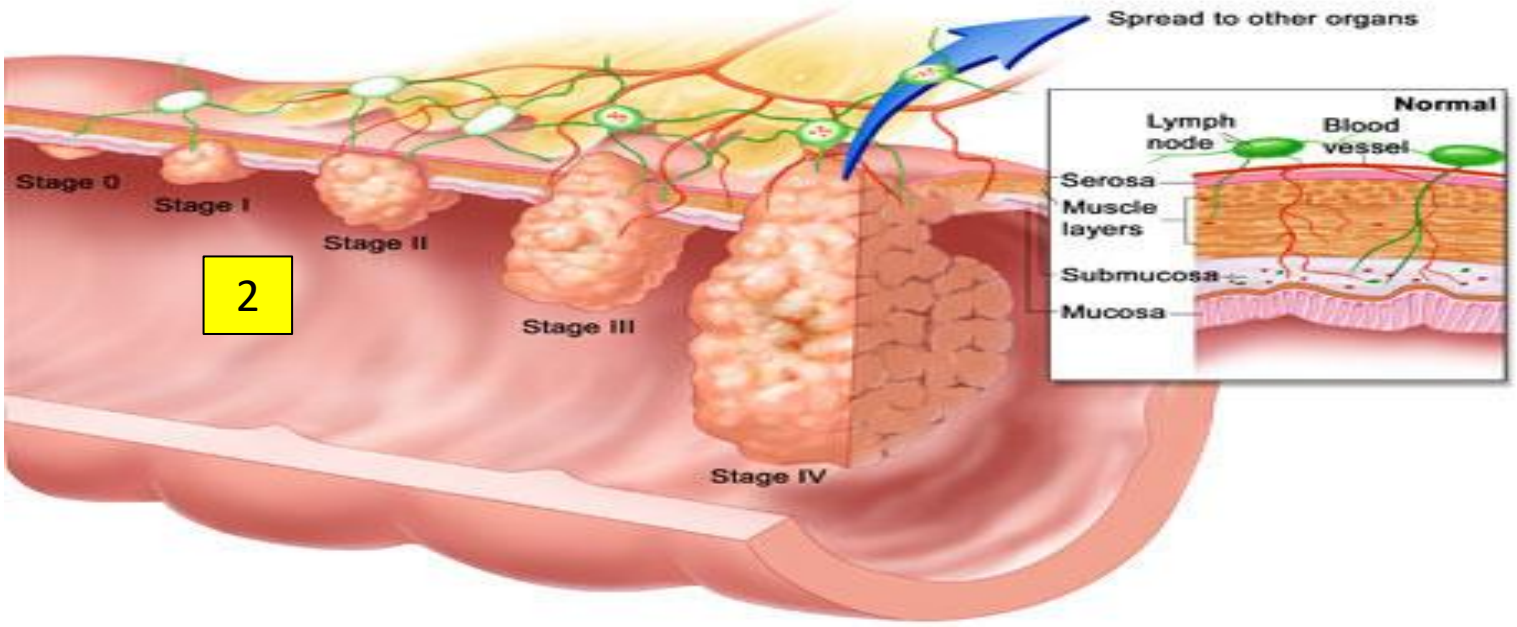


http://www.nih.gov/research-training/advances-colorectal-cancer-research

1. **Tiny tumour (few rogue cells scavenging O_2 and nutrients)**
   - **As the tumour grows bigger the cancer cells struggle to survive (demand outstrips supply)**
   - **more and more genetic faults accumulate**
   - **cancer cells release chemical signals that lure immune cells to infiltrate the tumour.**

LO1

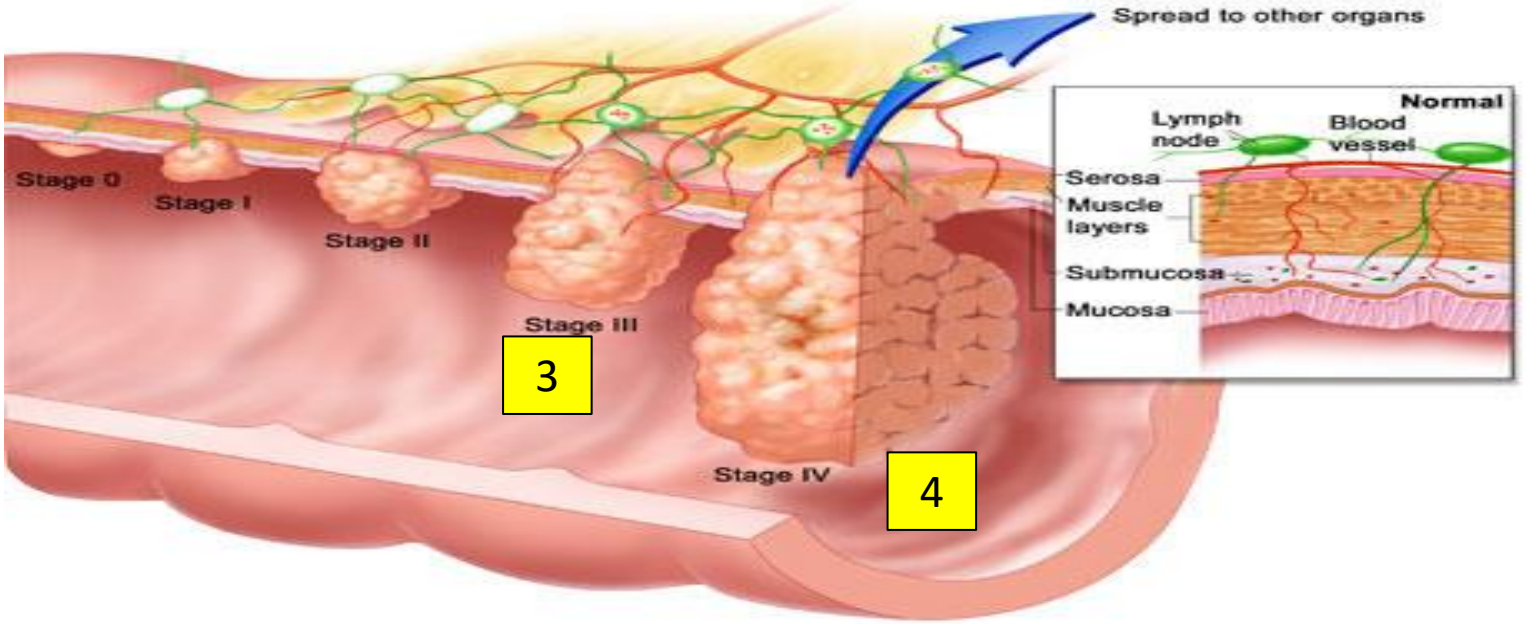
So how does inflammation lead to cancer?



2. **Release of cytokines from growing tumour kick starts the growth of new blood vessels (angiogenesis)**
   - **Brings in much-needed O_2 and nutrients**
   - **Cytokines released also encourage growth of the stroma - the cellular 'cushion' against which the tumour nestles.**

LO1

So how does inflammation lead to cancer?

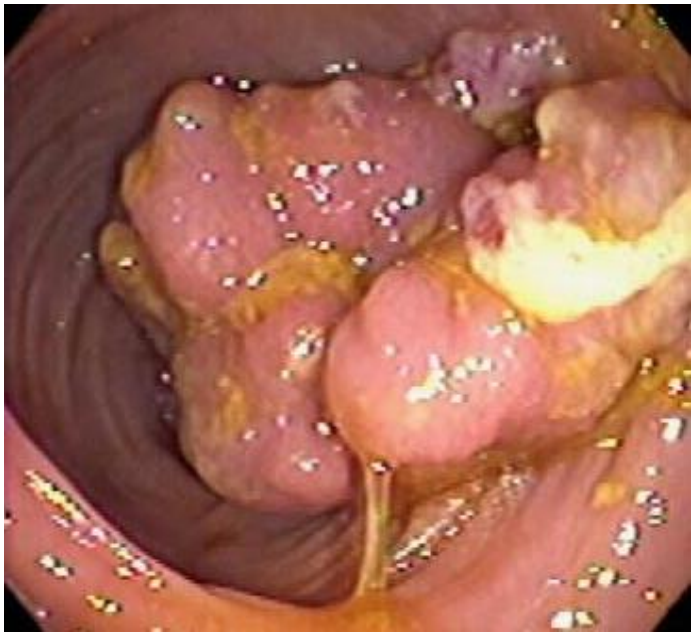


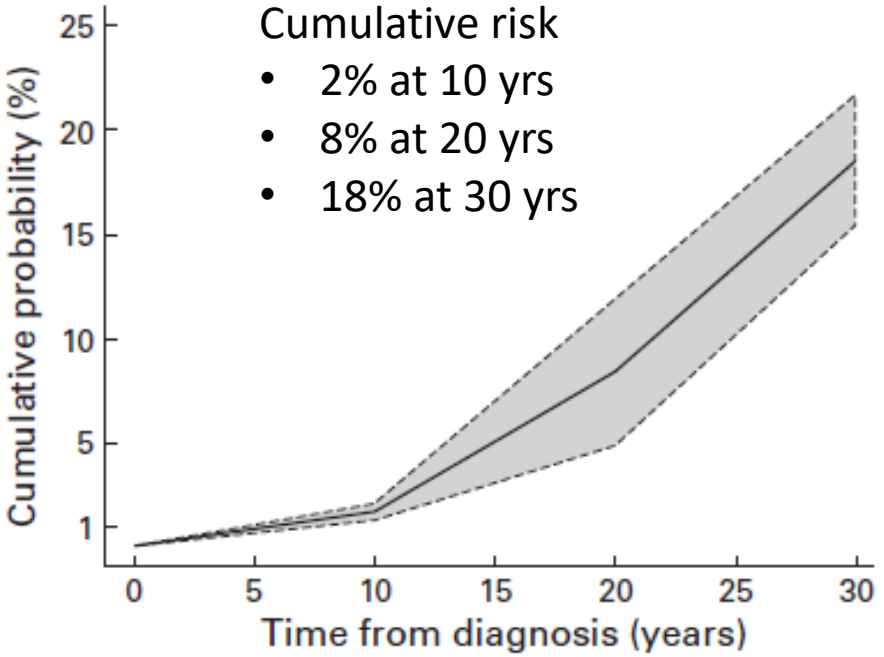
3. **Incoming inflammatory cells also hit the developing tumour with free radicals that further damage cellular DNA.**

4. **Inflammation can also initiate tumour spread (metastasis) by stimulating enzyme production that help tumour cells eat through the molecules tethering them to their surroundings.**

LO1

Colitis-associated bowel cancer





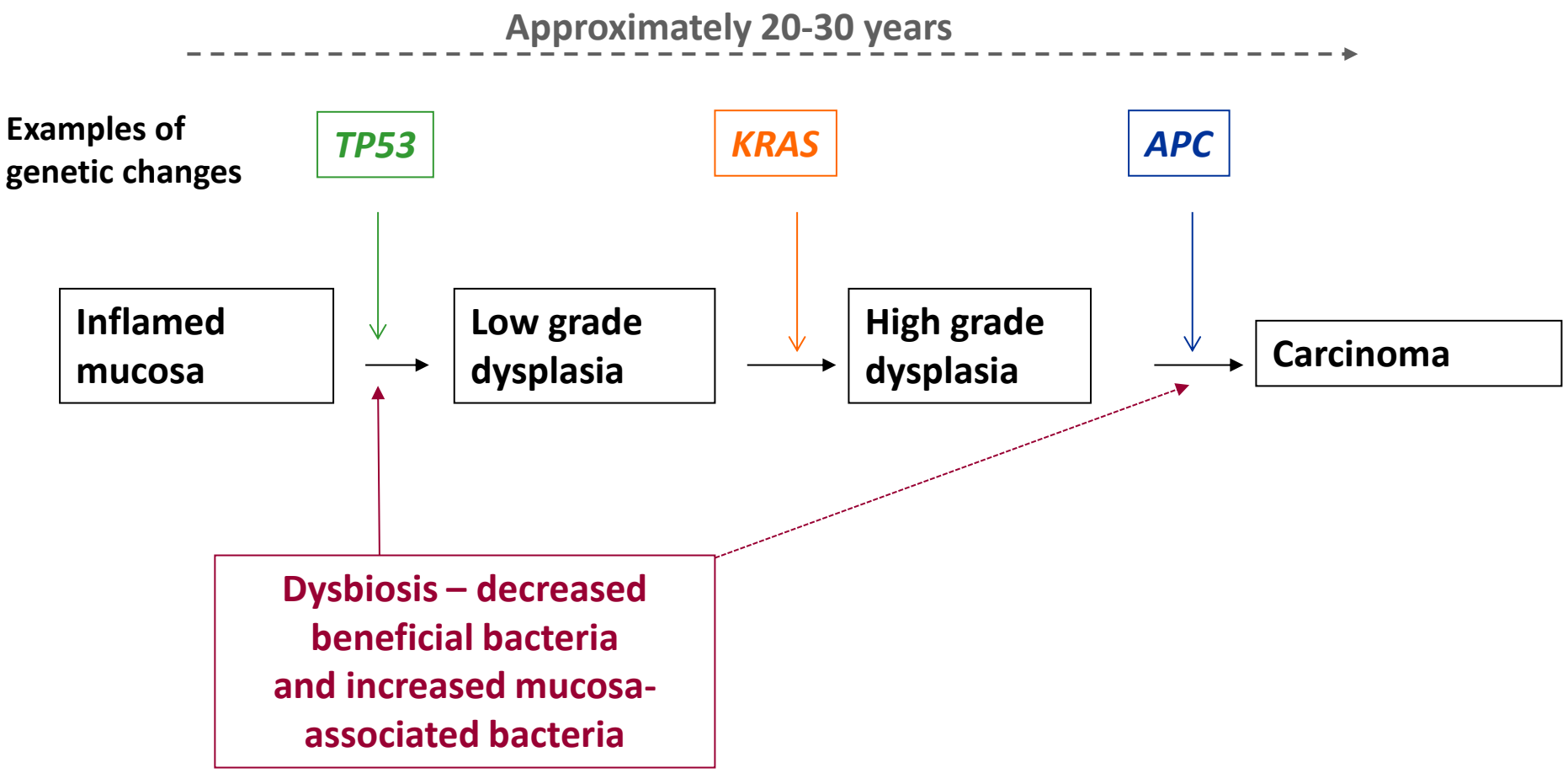
	O	E	Relative risk (95% CI)	
Ulcerative Colitis (n=486)	29	1.51	19.2***	(12.9-27.5)
Crohn's Disease (N=125)	8	0.44	18.2***	(7.8-35.8)

O=observed cancers, E= expected cancers. ***p<0.001

Gillen *et al*. 1994. *Gut* 35:1590-2; Eaden *et al*. 2001 *Gut* 48:526-35.

LO1

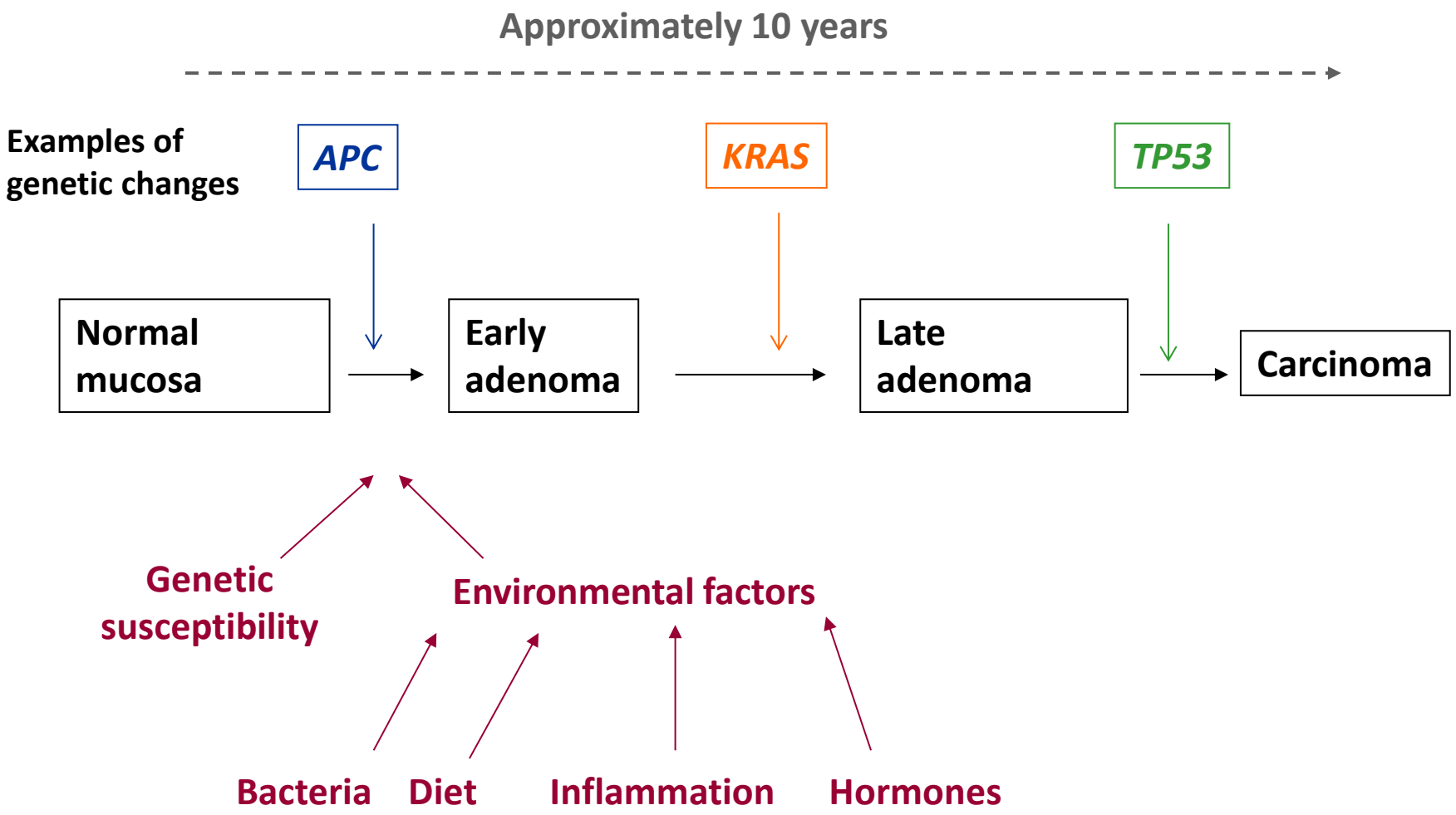
Inflammatory bowel disease (IBD) –associated colorectal cancer



Adapted from **Rhodes & Campbell 2002; *Trends Mol Med* 8(1):10-6.**

LO1

Sporadic colorectal cancer



Fearon & Vogelstein B. 1990 *Cell*; 61: 759-67.

Arends. 2000 *J Pathol*; 190: 412-16 (Review)

LO1

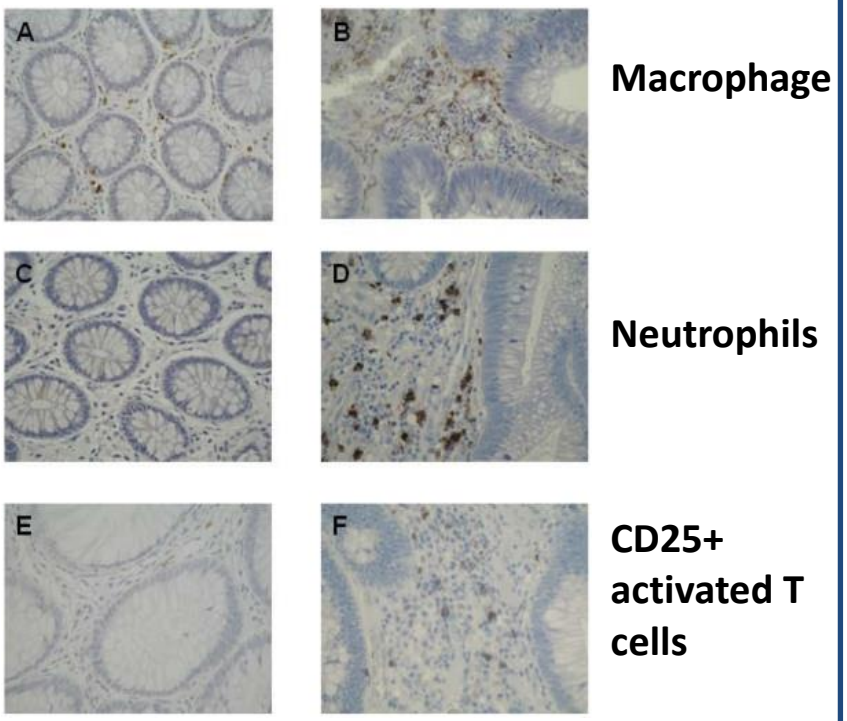
Inflammation and sporadic colorectal cancer development

- Increased serum concentrations of pro-inflammatory cytokines (e.g. TNFα etc.) in patients with CRC

- Inflammation in sporadic colonic adenomas →

- Increased intestinal expression of cyclooxygenase 2 (COX-2) with NSAIDs/aspirin being chemopreventive (↓ COX)

Drew *et al. Nat Rev Cancer*. 2016;16(3):173-86.



Associated with upregulation of proinflammatory cytokines

McLean MH…. El-Omar EM.. *PLoS One* 2011; 6:e15366

LO2

Infectious agents and cancer

- **~2 million new cancer cases of all cancer cases in 2008 (16.1%) were caused by infectious agents.**
- **~2·2 million (15·4%) in 2012 were attributable to carcinogenic infections**
- **~2·2 million (13%) new cancer cases in 2018 were attributable to infections (excluding non-melanoma skin cancers)**

Panel 1: Major cancer sites associated with group 1 infectious agents*

- Stomach: *Helicobacter pylori*
- Liver: Hepatitis B virus, hepatitis C virus (HCV), *Opisthorchis viverrini, Clonorchis sinensis*
- Cervix uteri: Human papillomavirus (HPV) with or without HIV
- Anogenital (penile, vulva, vagina, anus): HPV with or without HIV
- Nasopharynx: Epstein-Barr virus (EBV)
- Oropharynx: HPV with or without tobacco or alcohol consumption
- Kaposi's sarcoma: Human herpes virus type 8 with or without HIV
- Non-Hodgkin lymphoma: *H pylori*, EBV with or without HIV, HCV, human T-cell lymphotropic virus type 1
- Hodgkin's lymphoma: EBV with or without HIV
- Bladder: *Schistosoma haematobium*

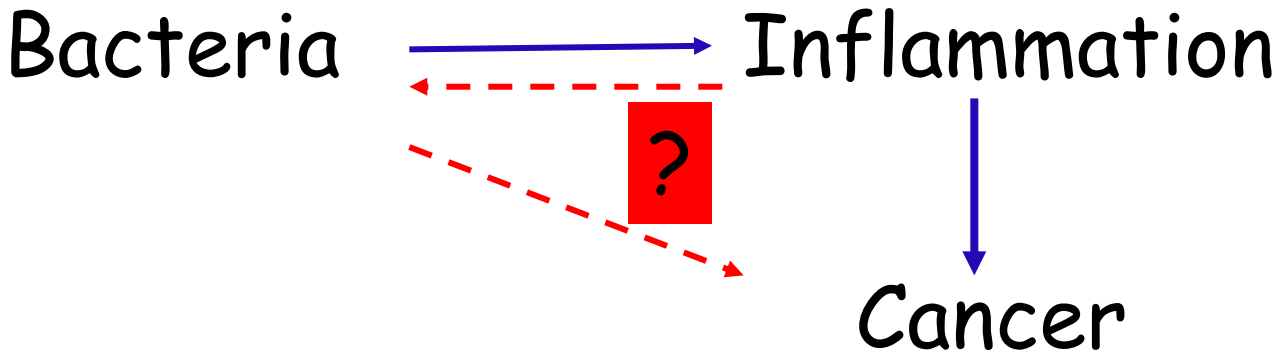
*Classified as carcinogenic to humans in International Agency for Research on Cancer Monograph 100B.[3]

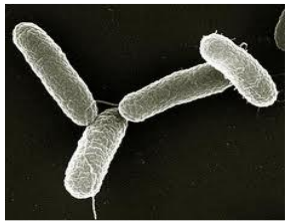
Other infectious agents likely but not evaluated by IARC.

De Martel *et al.* *Lancet Oncology* 2012;13(6):607-15
Plummer et al. *Lancet Glob Health* 2016;4(9):e609-16
De Martel et al. Lancet Glob Health 2020 Feb;8(2):e180-e190

LO2

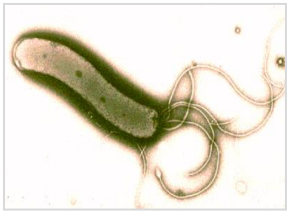
Bacteria, inflammation and cancer





***Salmonella* typhi** → Gallbladder cancer

Lancet 343, 83–84 (1994); *Am. J. Gastroenterol.* 95, 784–787 (2000); *J. Surg. Oncol.* 93, 633–639 (2006)



Helicobacter pylori → Gastric cancer/ MALT lymphoma

Nat. Rev. Cancer 2, 28–37 (2002)

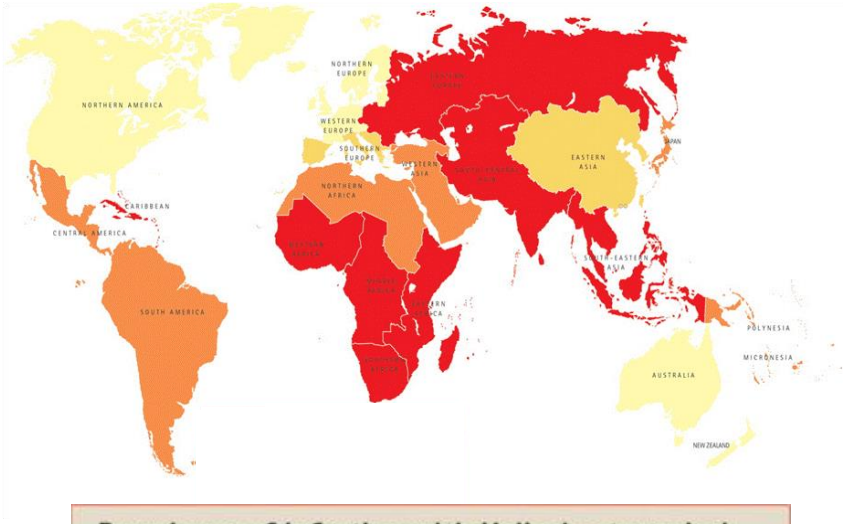
Streptococcus gallolyticus (bovis) → *colon cancer*

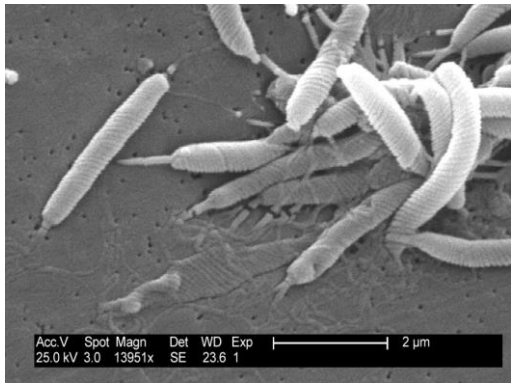
J Exp Clin Cancer Res. 2011; 20;30:11.

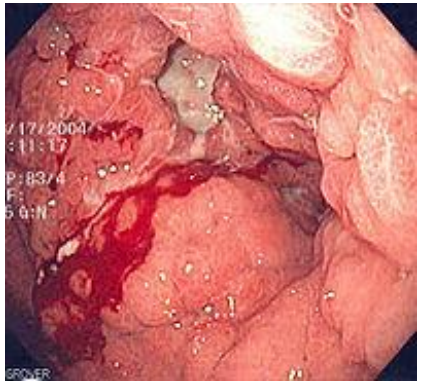
Clin Infect Dis. 2011;53(9):870-8.

LO2

Helicobacter pylori-induced gastric cancer







Prevalence of infection with Helicobacter pylori

In adults, by UN area
1994–2000

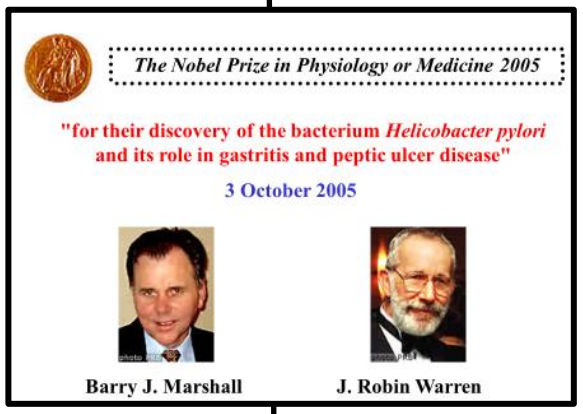
- 75% and above
- 65% – 74%
- 55% – 64%
- below 55%

Observed by Warren

Cultured by Marshall and Warren

Designated class I biological carcinogen by WHO/IARC

Marshall & Warren awarded Nobel Prize



The Nobel Prize in Physiology or Medicine 2005

"for their discovery of the bacterium *Helicobacter pylori* and its role in gastritis and peptic ulcer disease"

3 October 2005

Barry J. Marshall

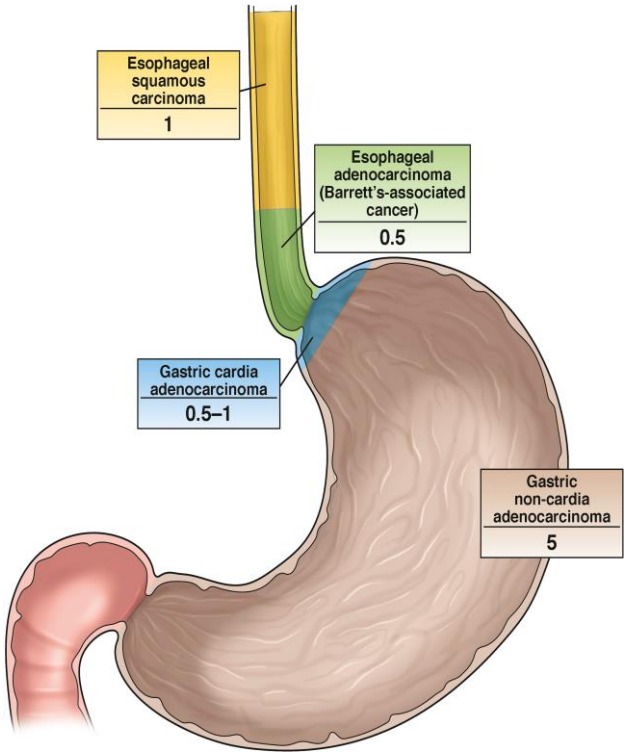
J. Robin Warren

1975 1980 1985 1990 1995 2000 2005 2010

LO2

Epidemiology

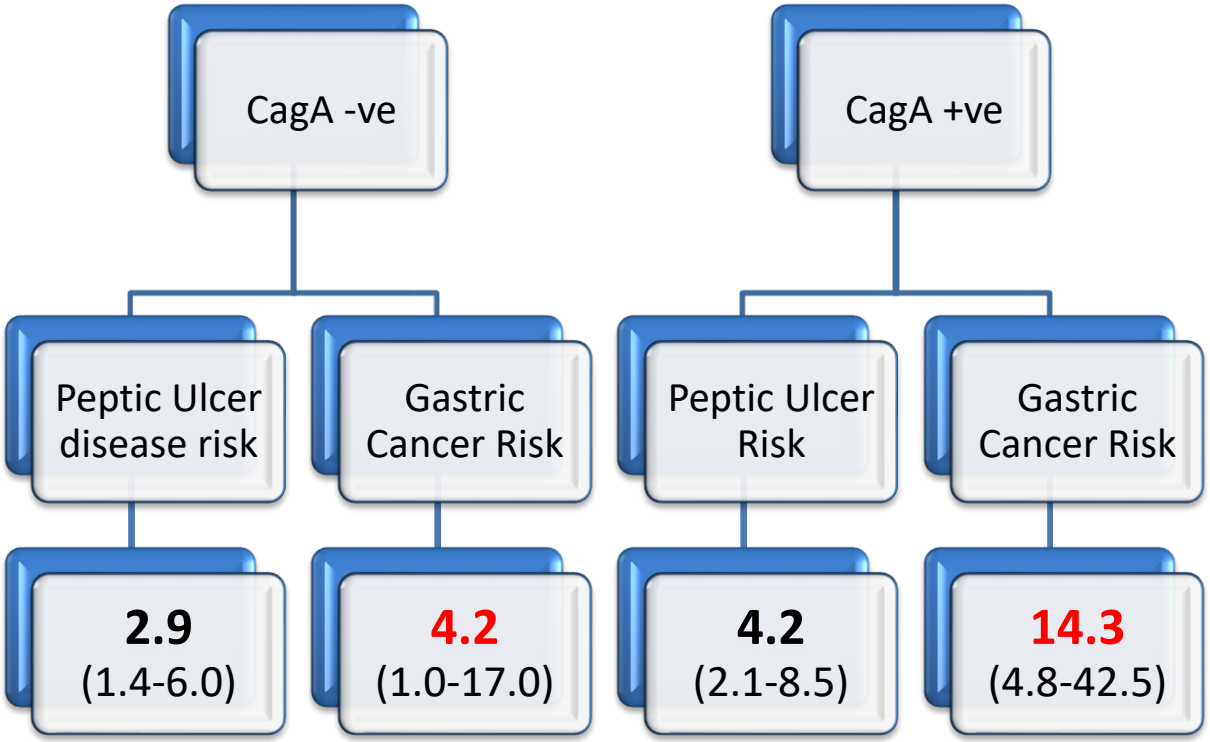
- Gastric cancer represents 10% of all global cancers (4th commonest, 2nd commonest cause of death)
- <2% of *H. pylori*-infected individuals ever develop gastric cancer
- Overall odds ratio for gastric cancer development in *H. pylori*–infected vs - uninfected persons of **3.8** (95% confidence interval [CI], 2.3–6.2).
- Attributable fraction of non-cardia gastric cancers due to *H. pylori* infection ~ 74-78%



Moss S. *CMGH* 2017; 3(2), P183-191

LO2

Helicobacter pylori virulence factors

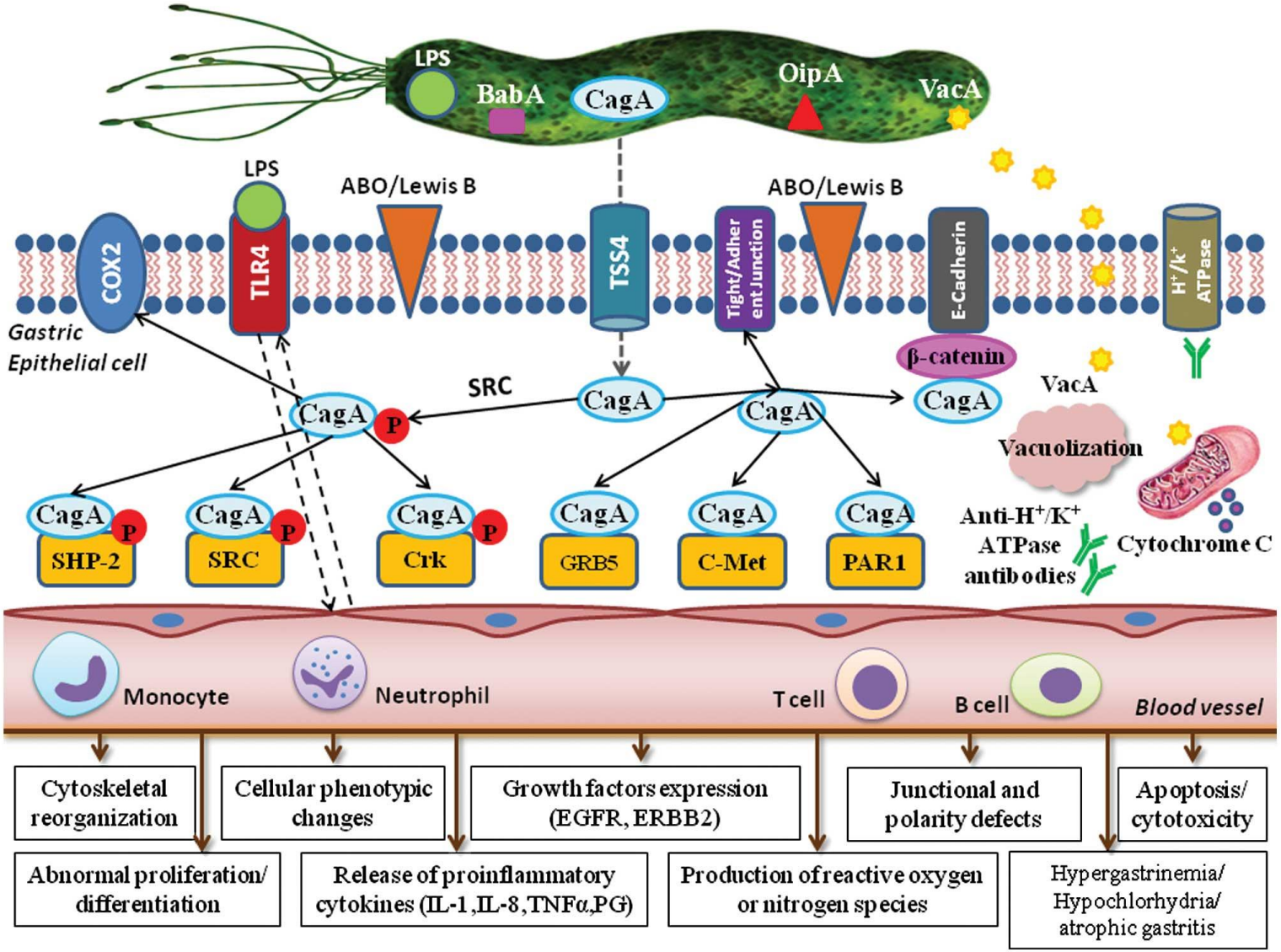


Risks shown as relative risk compared to uninfected control population (95% CI)

Nomura *et al. Am. J Epidemiol.* 2002; 155, 1054-9
Held *et al. Helicobacter* 2004; 9, 271-77

LO2

H. Pylori CagA cytotoxin

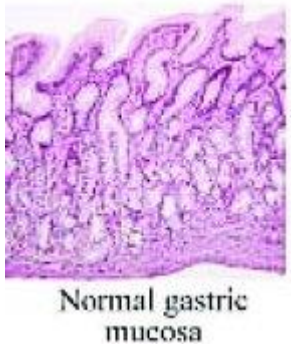


Conteduca *et al.* *Int J Oncology* 2013; 42, 5-18

LO2

Typical gastric carcinogenesis pathway

Normal gastric epithelium



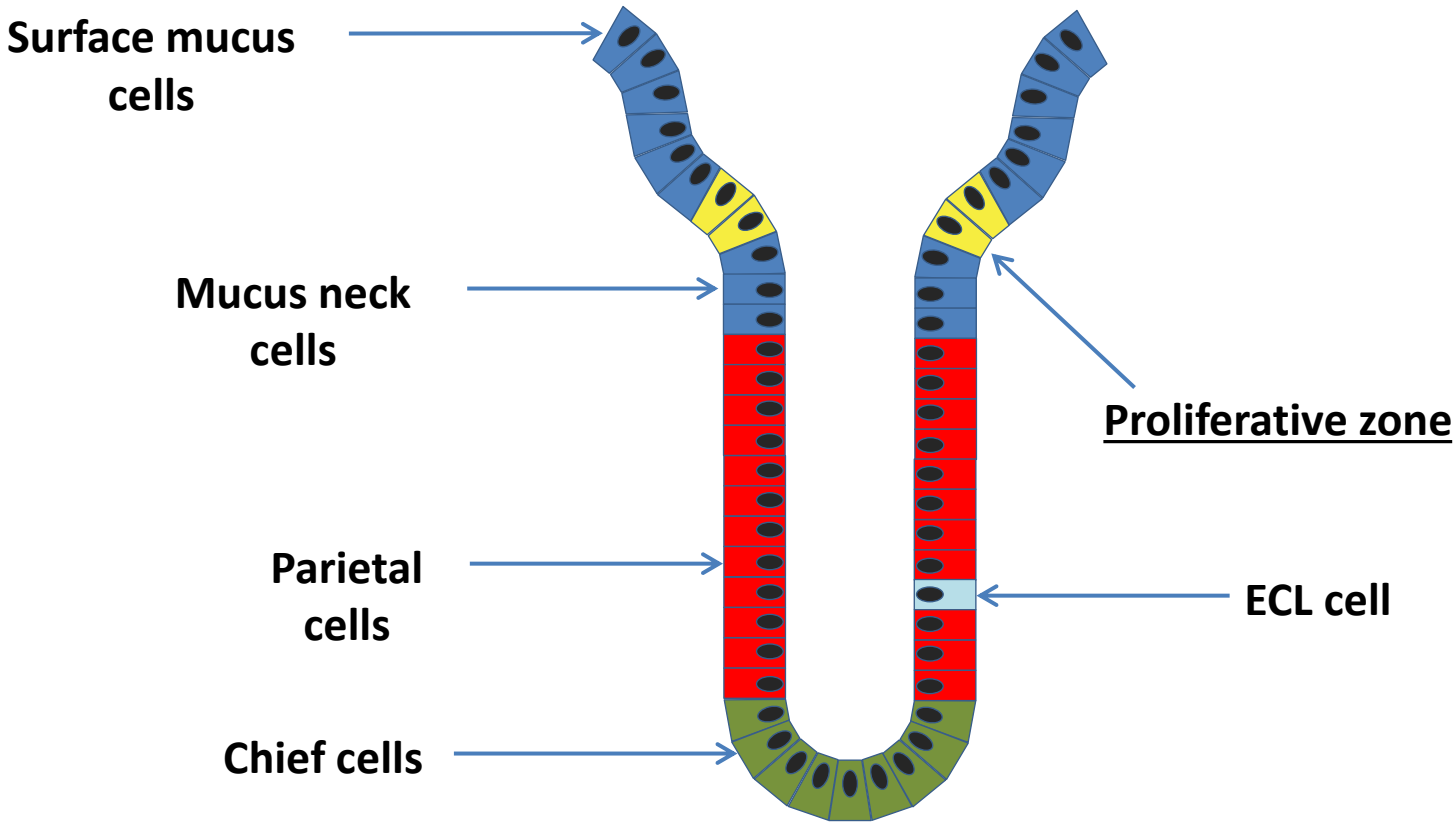
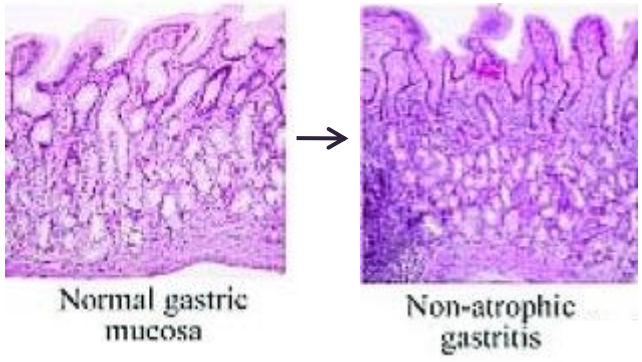


Image adapted from…Correa *et al.*, Scand. J. Surgery 2006; 95: 218–224

LO2

Typical gastric carcinogenesis pathway

Normal gastric epithelium → Superficial gastritis



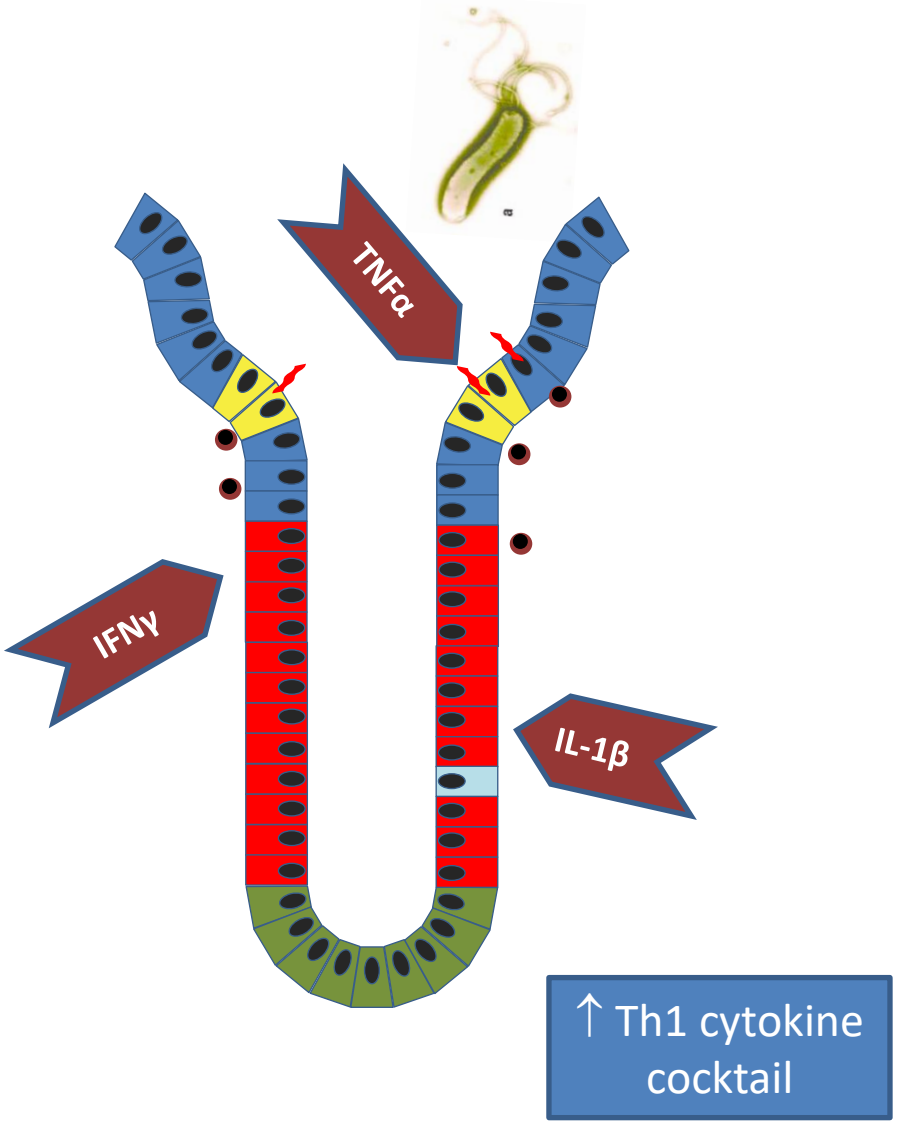


Image adapted from…Correa *et al.*, Scand. J. Surgery 2006; 95: 218–224

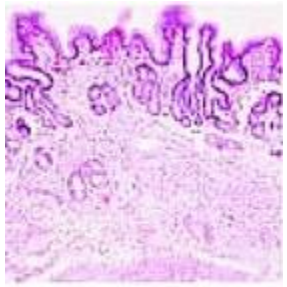
LO2

Typical gastric carcinogenesis pathway

Normal gastric epithelium → Superficial gastritis → Atrophic gastritis

hyperproliferation

Less numbers of Parietal cells



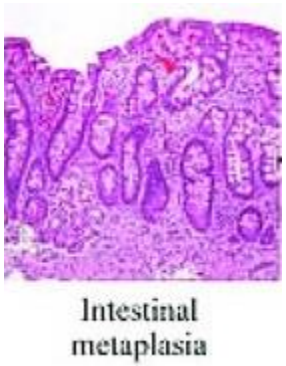
Multifocal atrophic gastritis

Image adapted from…Correa *et al.*, Scand. J. Surgery 2006; 95: 218–224

LO2

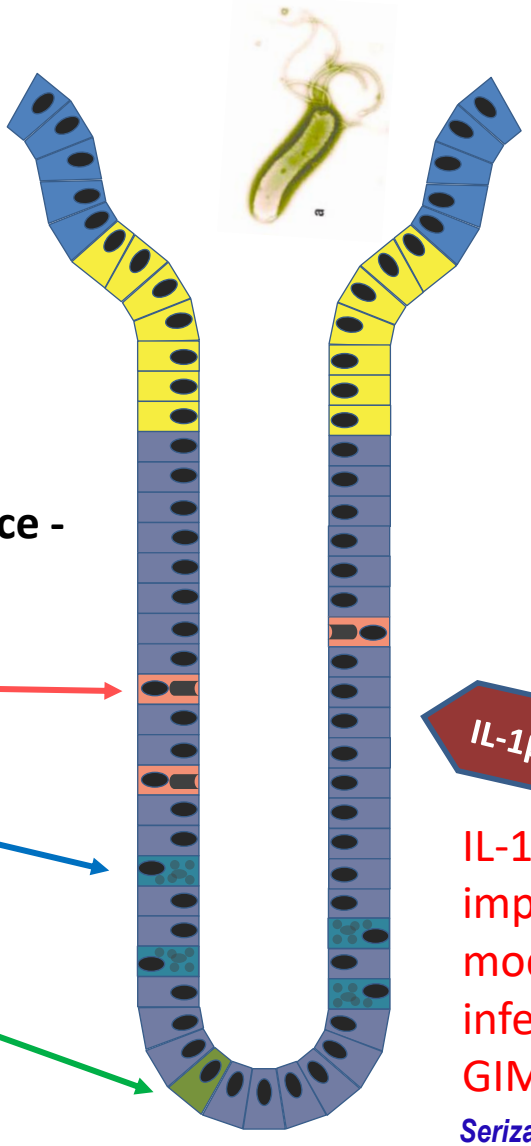
Typical gastric carcinogenesis pathway

Normal gastric epithelium → Superficial gastritis → Atrophic gastritis → Intestinal metaplasia


Intestinal metaplasia

Intestinal appearance - Presence of poorly aligned intestinal **GOBLET CELLS** and **PANETH cells**

Loss of chief cells



IL-1/IL1-R signalling important in mouse models of *H. pylori* infection-induced GIM

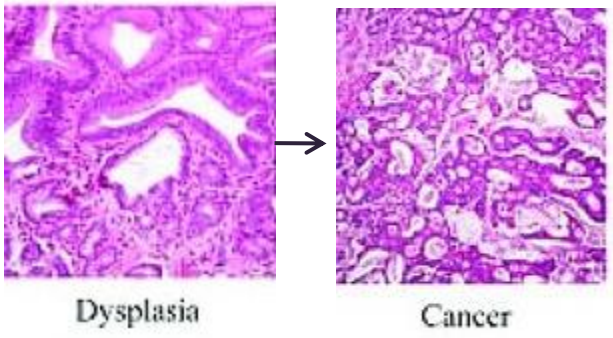
Image adapted from…Correa *et al.*, Scand. J. Surgery 2006; 95: 218–224

Serizawa et al. Infect Immun. 2016; 84;562

LO2

Typical gastric carcinogenesis pathway

Normal gastric epithelium → Superficial gastritis → Atrophic gastritis → Intestinal metaplasia → Dysplasia → Gastric Adenocarcinoma



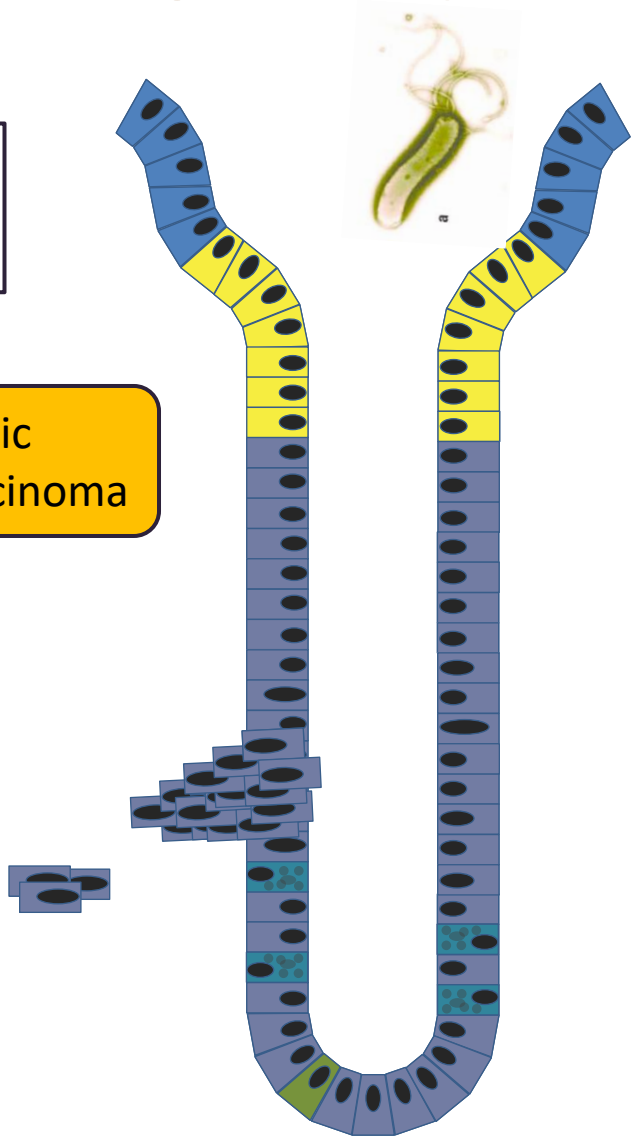
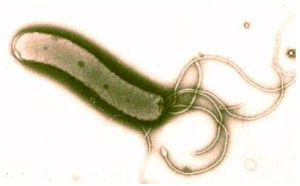


Image adapted from…Correa *et al.*, Scand. J. Surgery 2006; 95: 218–224

LO2



Helicobacter pylori colonisation

Diet (esp. High salt, pickled/smoked foods)

EXOGENOUS FACTORS

Bacterial virulence

Non-*Helicobacter* microbiota

Smoking

Normal gastric epithelium → Superficial gastritis → Atrophic gastritis → Intestinal metaplasia → Dysplasia → Gastric Adenocarcinoma

Hypergastrinaemia

Hypochlorhydria and hypergastrinaemia

Proliferation

Inflammation

GENOTYPE

Apoptosis

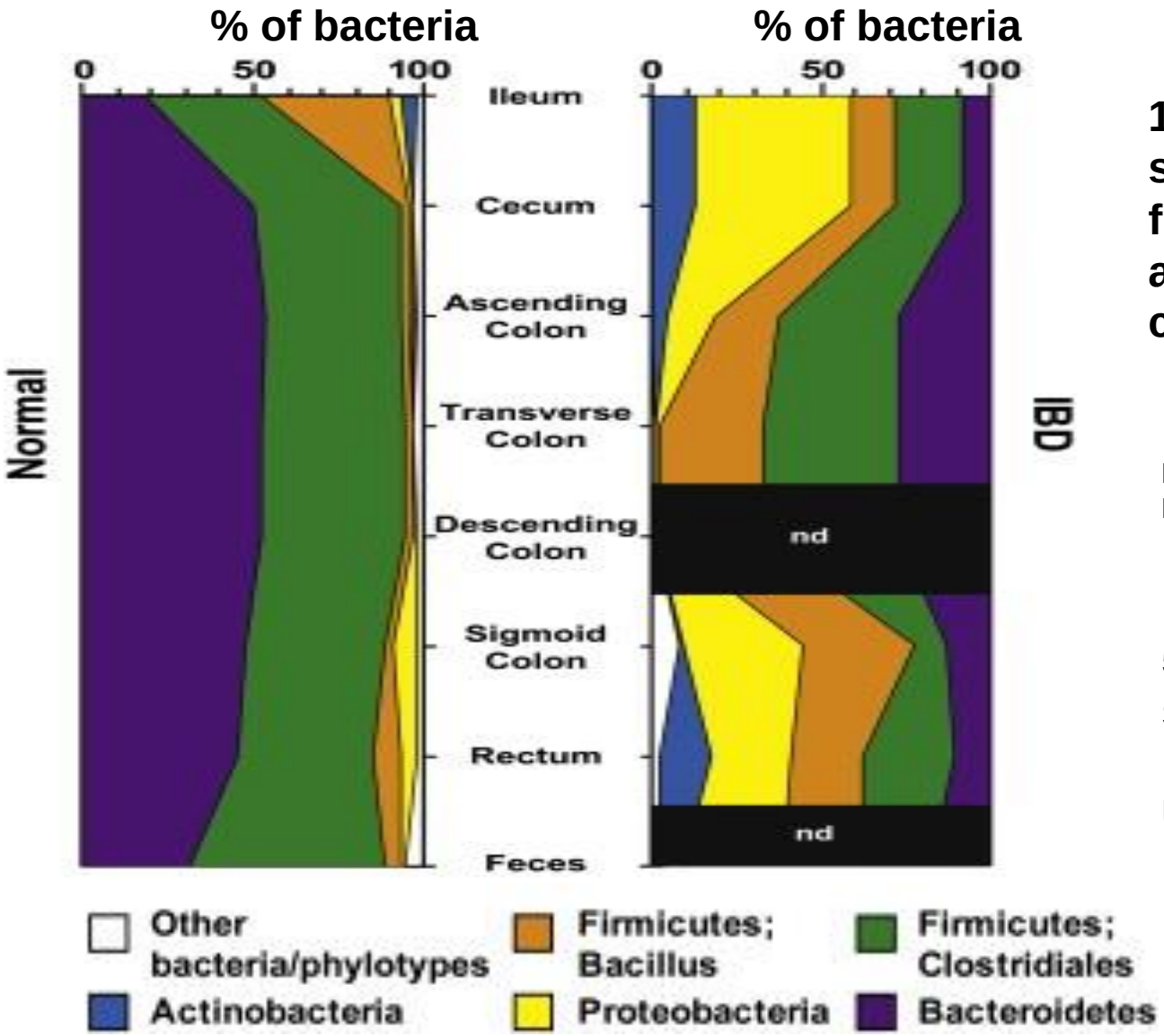
Stromal remodelling

ENDOGENOUS FACTORS

Adapted from …Burkitt M *et al.* World J Gastro. 2009. 15(1):1-16.

LO1

Inflammation re-shapes the intestinal microbiome



13,000 16S rRNA sequences analysed from healthy young adults and non-IBD controls.

Seven major phyla

Firmicutes
Bacteroidetes } **>90%**

5,405 16S rRNA sequences from patients with CD and UC.

Petersen *et al.* 2008 *Cell Host & Microbe* 3, 417-27

LO2

Dysbiosis: The first hit for digestive system cancer

Key increased mucosal bacteria include:

	Effects on carcinogenesis
Fusobacterium nucleatum →	Promotes CRC cell proliferation *in vitro* Increases tumor growth rates in patient-derived CRC xenografts in mice Increase levels of lymphocyte-attracting chemokines CCL5, CCL20, and CXCL11 Recruit other bacteria to form biofilms coating human CRCs
Strep. (bovis) gallolyticus	
Bacteroides fragilis →	Promote the development of precancerous lesions (i.e., adenomas) Induce a pro-carcinogenic Th17 response by recruiting M-MDSCs Induce DNA damage *via* promoting the inflammation and oxidative stress Induced the expression and secretion of CXCL1-ortholog IL-8 from epithelial cells *via* activation of NF-κB Enhance tumorigenesis in preclinical CRC models
Akkermansia mucinophilia	
Escherichia coli →	Produce the genotoxin colibactin and result in mutagenic DNA damage in colonic epithelial cells Induce intestinal stem cell mutations *in vitro* Increase levels of lymphocyte-attracting chemokines CCL5, CCL20, and CXCL11 Induce DNA damage *via* promoting inflammation and oxidative stress
Enterococcus faecalis	

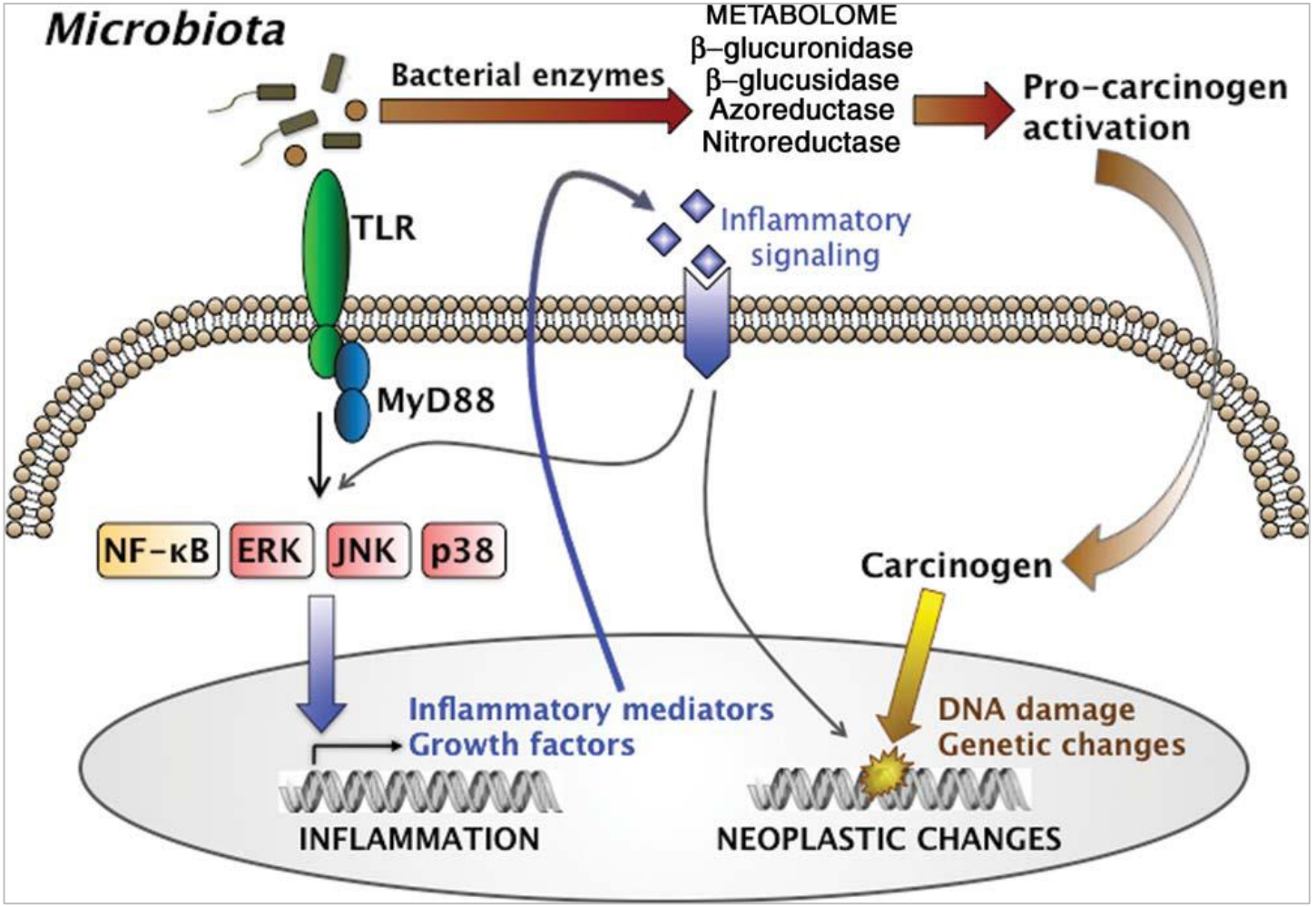
Note: Dysplastic mucosa has no overlying mucus and no glycocalyx and likely allows easy access for bacteria to contact the epithelial cells

M-MDSCs, monocytic-like myeloid-derived suppressor cells.

Mei *et al. Front. Physiol. 13: 1040991* *doi: 10.3389/fphys.2022.1040991*

LO1
LO2

Potential mechanisms by which bacteria promote inflammation-associated colorectal cancer

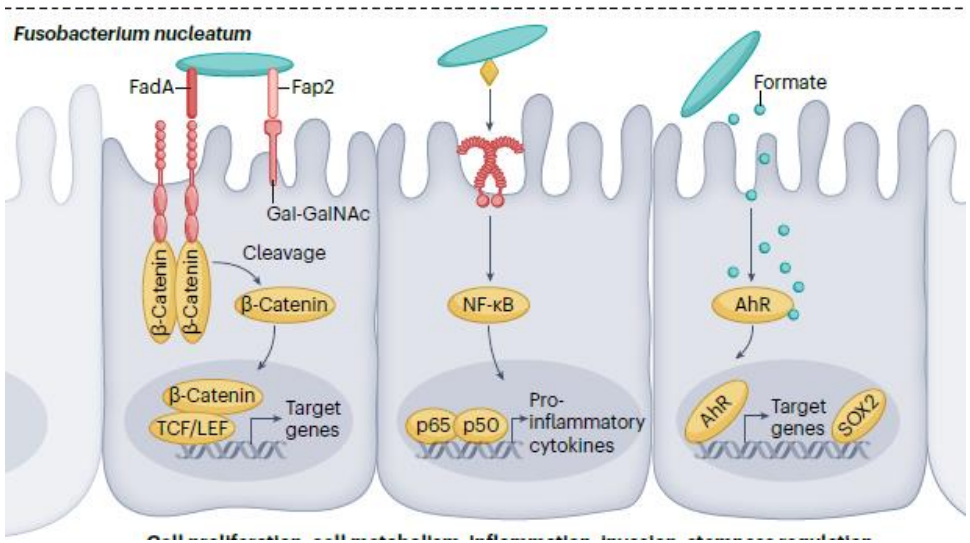


Arthur & Jobin. *IBD* 2011; 17: 396–409

LO2

Key bacteria in colon cancer initiation and progression

Fusobacterium nucleatum ➡



El Tekle & Garrett. Nature Reviews Cancer 2, 600-18 (2023)

A distinct *Fusobacterium nucleatum* clade dominates the colorectal cancer niche

A bacterial strain linked to colon cancer is pinpointed

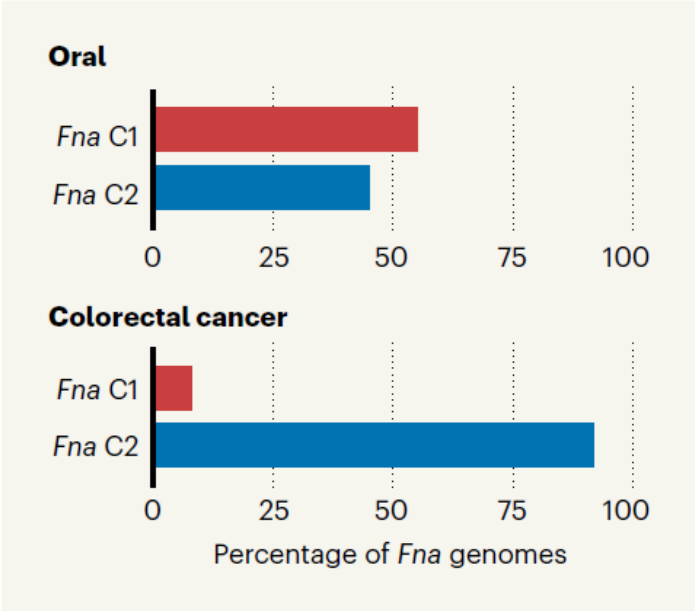


Figure 1 | *Fusobacterium nucleatum animalis* (*Fna*) microbes in the human mouth and in colorectal cancer samples. Zepeda-Rivera *et al.*[1] report that this bacterium associated with colorectal cancer can be divided into two clades. Clade 2 (C2) is highly prevalent in tumours. (Adapted from Fig. 2f of ref. 1.)

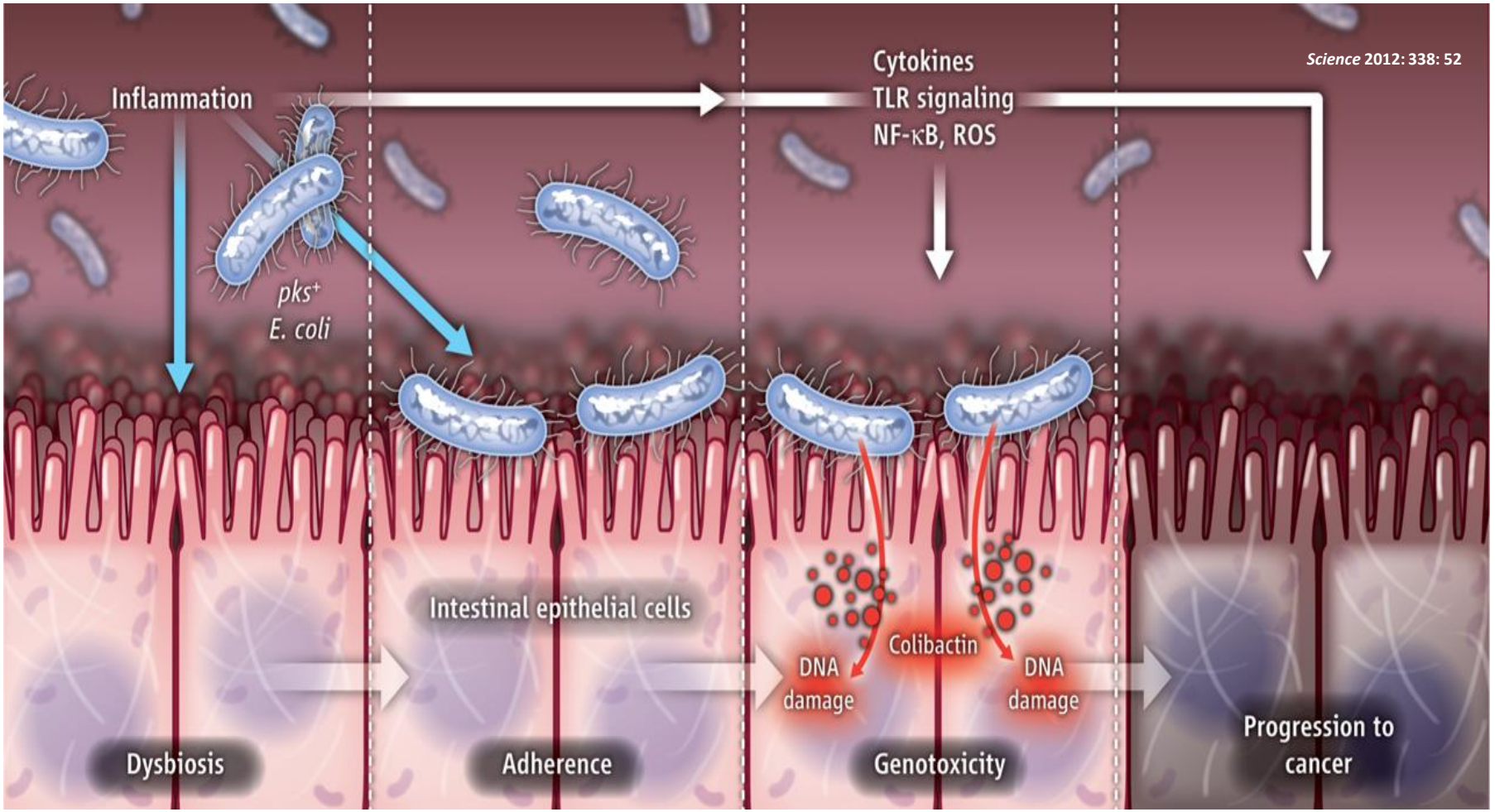
Sears & Queen. Nature 628, 275-276 (2024); Zepeda-Rivera et al. Nature 628, 424-32 (2024).

LO1
LO2

Intestinal Inflammation Targets Cancer-Inducing Activity of the Microbiota

Janelle C. Arthur,[1] Ernesto Perez-Chanona,[1] Marcus Mühlbauer,[1] Sarah Tomkovich,[1] Joshua M. Uronis,[1] Ting-Jia Fan,[1] Barry J. Campbell,[2] Turki Abujamel,[3,4] Belgin Dogan,[5] Arlin B. Rogers,[6] Jonathan M. Rhodes,[2] Alain Stintzi,[3] Kenneth W. Simpson,[5] Jonathan J. Hansen,[1] Temitope O. Keku,[1] Anthony A. Fodor,[7] Christian Jobin[1]*

Arthur *et al*. 2012. *Science* 338: 120-3



***pks* – polyketide synthase pathogenicity island**

LO1
LO2

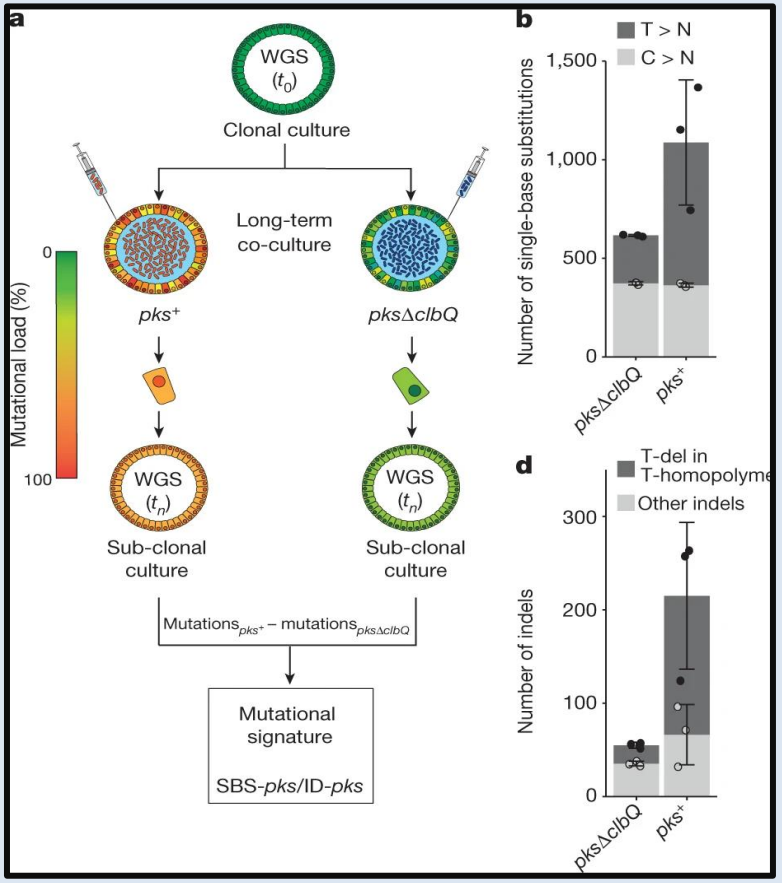
Mutational signature in colorectal cancer caused by genotoxic *pks+ E. coli*

- **Human intestinal organoids**
  exposed to genotoxic *pks+ E. coli* by repeated luminal injection over 5 months

- **Distinct mutational signature seen was absent from organoids injected with a mutant strain producing no active colibactin.**

- **The same mutational signature was detected in a subset of 5,876 human cancer genomes from two independent cohorts, predominantly in colorectal cancer.**

Pleguezuelos-Manzano *et al*.
***Nature* 2020; 580, 269**



Whole-genome sequencing confirms this:

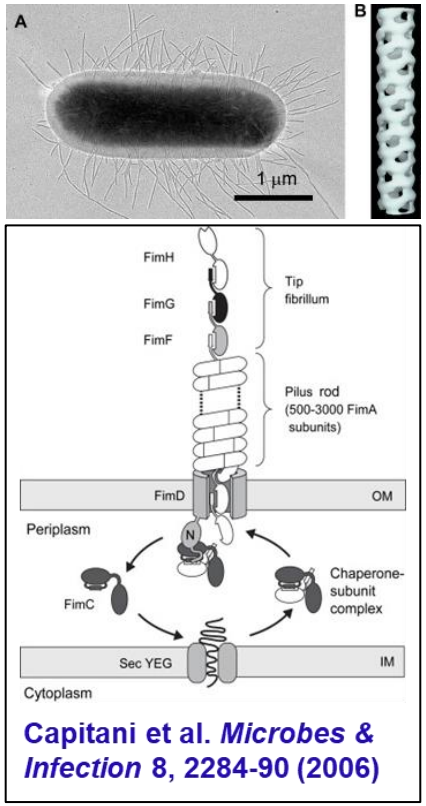
- **In contrast to healthy individuals, normal crypts of colon cancer patients have a high incidence of *pks* + *E.coli* mutational and indel signatures**

Chen *et al*. *Nat Commun* 2023; 14, 7827

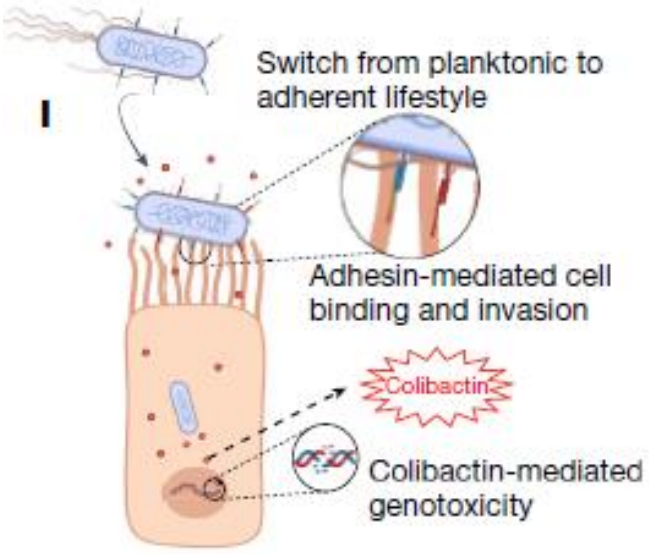
Colibactin-driven colon cancer requires adhesin-mediated epithelial binding

- Oncogenic potential of *pks*+ *E. coli* depends on adhesion to host epithelium, mediated by the type 1 pilus adhesin FimH and the F9 pilus adhesin FmlH

- Colibactin production in close proximity to host epithelial cells promotes DNA damage and drives CRC development.



Capitani et al. *Microbes & Infection* 8, 2284-90 (2006)



Blocking bacterial adhesion using a pharmacological FimH inhibitor attenuates colibactin-mediated genotoxicity and CRC exacerbation.

Promises new therapeutic avenues to reduce risk for developing CRC

Jans *et al*. *Nature 635*; 472 (14 Nov 2024)

LO2

Concept of colorectal cancer as a bacterial disease is gaining support

1. Bacteria-epithelial interaction may be crucial by:
   (i) stimulation of epithelial (Toll-Like) receptors and consequent inhibition of apoptosis, a process that clears damaged cells
   (ii) DNA-damaging (genotoxic) effects.

2. This interaction is much more likely once an adenomatous polyp is present

3. E. coli may be particularly important because they
   a) adhere and invade to epithelial cells,
   b) tolerate relatively high oxygen environment (microaerophilic)
   c) produce genotoxins (*pks* PAI $\rightarrow$ colibactin and cytolethal distending toxin as another causative agent associated with colorectal cancer)
   pro-inflammatory and pro-angiogenic

4. Adhesin interactions occur with carbohydrate receptors on epithelial cells, which may be preventable by dietary plant polysaccharides*

***A diet rich in fruit and vegetables decreases risk of CRC**

LO1

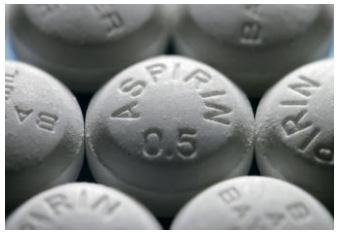
If genetic damage is the match that lights the fire, and inflammation is the fuel that feeds the flames…

Q. Can we then dampen inflammation, making it much harder for cancers to flourish?

- **Use of anti-inflammatory drugs (e.g. Aspirin)**
- **Reduce environmental exposure to damaging agents**
- **Modify diet:**
  - **Increase intake of anti-inflammatory, anti-oxidant, anti-genotoxic, prebiotic dietary components, including more soluble fruit, legume and vegetable fibre ('*5-a day*')**
  - **Decrease intake of saturated animal fat & processed meat/refined sugar/salt/additives**

LO1

Evolution of evidence on aspirin and colon cancer prevention



1991

Rosenberg *et al*

Case-control: NSAIDS and large-bowel cancer.

RR = 0.50

Thun *et al*

Observational: Aspirin and fatal colon cancer.

RR = 0.60 (men)
RR = 0.58 (women)

Baron and Greenberg

Editorial: "...controlled clinical trial...would be most useful..."

1994

Giovannucci *et al*

Observational: Aspirin and colorectal ad-enoma and cancer (men).

RR = 0.51 (total cancer)

1995

Giovannucci *et al*

Observational: Aspirin and colorectal cancer (women).

RR = 0.56 (total cancer)

1997

IARC

Working Group review: "limited evidence for the cancer-preventive activity of aspirin..."

IARC

Handbooks on Cancer Prevention: Non-Steriodal Anti-Inflammatory Drugs

2003

Baron *et al*

Randomized trial: Aspirin and colorectal ad-enomas.

RR = 0.59 (adv neoplasm)

Sandler *et al*

Randomized trial: Aspirin and colorectal adenomas (previous cancer)

RR = 0.65

2007

Flossman *et al*

Randomized trial: Aspirin and colorectal cancer

HR = 0.63

Review: "....consistent with findings from observational studies"

2010

Rothwell *et al*

Randomized trials: Aspirin and colorectal adenoma and cancer (20-year follow-up).

HR = 0.75

2011

Burn *et al*

Randomized trial: Aspirin and colorectal cancer (hereditary Lynch syndrome)

HR = 0.41

Chan and Lippman

Editorial: "..continuing uncertainty about dose and duration." "...results... arguably support more general recommendations..."

2014

Cuzick *et al*

Data review: Overall health benefits-harms of aspirin.

RR = 0.73 (colorectal cancer)

RR = 0.94 (net overall health benefit cancer/CVD)

Cuzick *et al (con't)*

Colorectal cancer accounts for 30 - 36% of aspirin's overall benefits.

"Prophylactic aspirin use...appears to have favourable benefit-harm profile"

LO1

If genetic damage is the match that lights the fire, and inflammation is the fuel that feeds the flames…

Q. Can we then dampen inflammation, making it much harder for cancers to flourish?

- **Use of anti-inflammatory drugs (e.g. Aspirin)**
- **Reduce environmental exposure to damaging agents**
- **Modify diet:**
  - **Increase intake of anti-inflammatory, anti-oxidant, anti-genotoxic, prebiotic dietary components, including more soluble fruit, legume and vegetable fibre ('*5-a day*')**
  - **Decrease intake of saturated animal fat & processed meat/refined sugar/salt/additives**

- **Target bad microbes: Use of antibiotics/probiotics/FMT/vaccines**

The microbiome, cancer & cancer therapy - Helmink *et al.* Nature Medicine 25, 377-88 (2019)

Targeting the gut & tumor microbiota in cancer - Park *et al.* Nature Medicine 28, 690-703 (2022)

UNIVERSITY OF LIVERPOOL

Thank you for your attention.

bjcampbl@liverpool.ac.uk

https://pcwww.liv.ac.uk/~bjcampbl/Inflammation and cancer.htm