Unexpected guests in tumor microenvironment: Human microbiota and cancer

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Outline

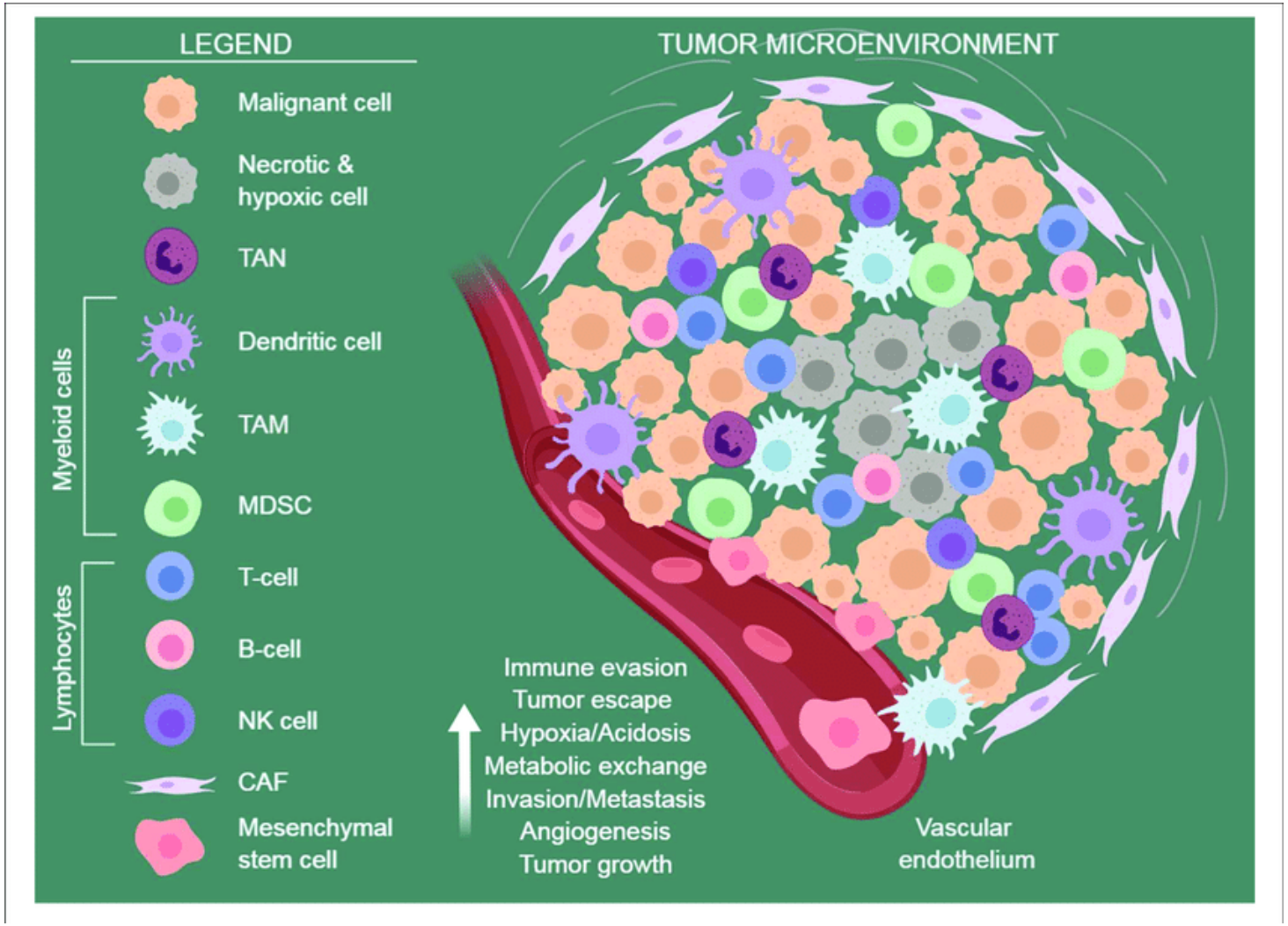
- Introduction to Tumor Microenvironment (TME) and human microbiome
- Human microbiome and TME : Case study
- Summary & Take-home message

What is Tumor Microenvironment (TME)?

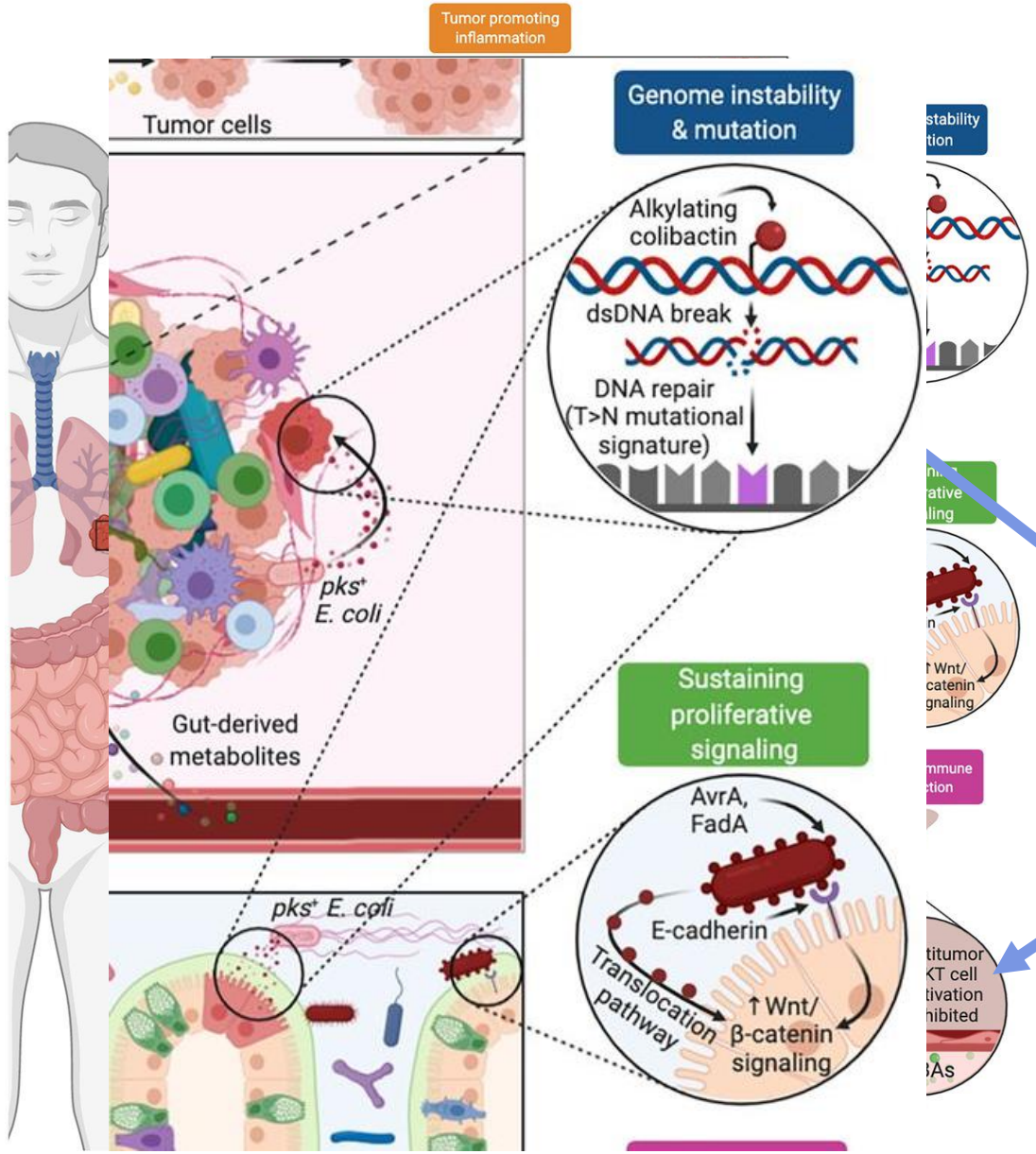
Tumor Microenvironment

- It is the comprehensive environment around a tumor.
- TME components can contribute both positive and negative signals to the tumor
- Assist cancers to evade immune sureveilance and destruction.

(Joyce, 2005)



(Figure 1, Audrito et al., 2019)



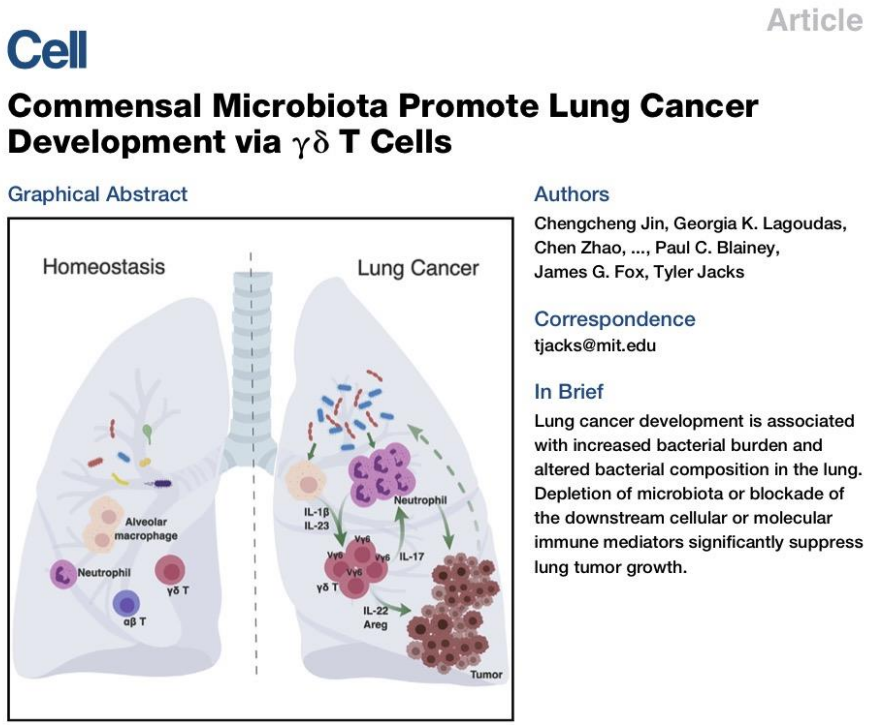
Human Microbiome can influence tumor through TME

- About 4×10^{13} microbial cells spanning ~3×10^{3} species inhabiting the human body.
  (Sender, Fuchs, & Milo, 2016)
- Potentially 'complicit' microbial function : promote carcinogenesis but are insufficient to cause cancer
- Some metabolites may be linked to the immune system' s role in solid tumorigenesis

(Figure 1, Sepich-Poore et al., 2021)

Core Paper

- Jin, C., Lagoudas, G. K., Zhao, C., Bullman, S., Bhutkar, A., Hu, B., ... & Jacks, T. (2019). Commensal microbiota promote lung cancer development via γ δ T cells. *Cell*, *176*(5), 998-1013.
- Ma, C., Han, M., Heinrich, B., Fu, Q., Zhang, Q., Sandhu, M., ... & Greten, T. F. (2018). Gut microbiome－mediated bile acid metabolism regulates liver cancer via NKT cells. *Science*, *360*(6391



Article

Cell

Commensal Microbiota Promote Lung Cancer Development via γδ T Cells

Graphical Abstract

Authors

Chengcheng Jin, Georgia K. Lagoudas, Chen Zhao, ..., Paul C. Blainey, James G. Fox, Tyler Jacks

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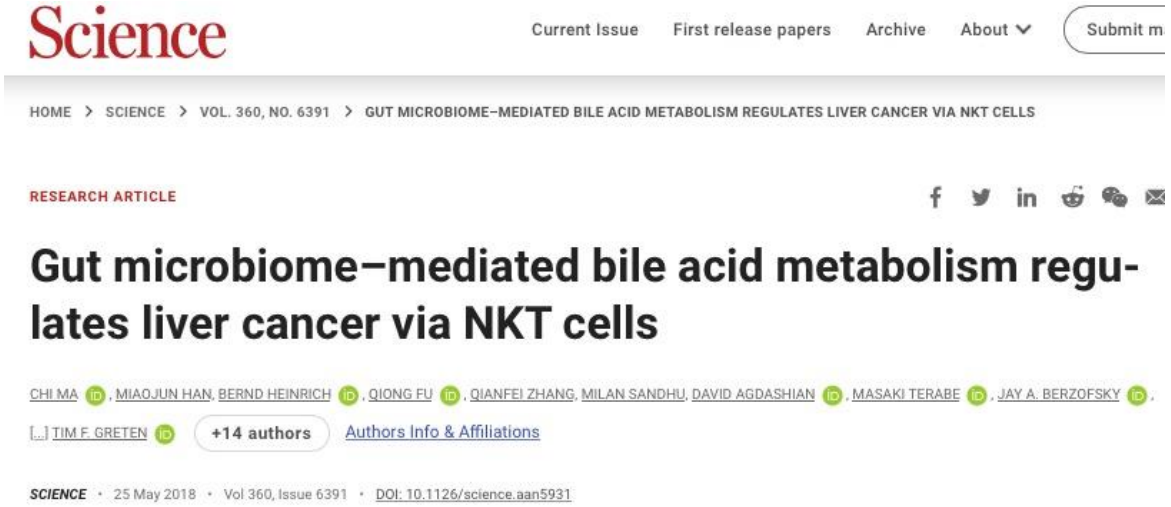
In Brief

Lung cancer development is associated with increased bacterial burden and altered bacterial composition in the lung. Depletion of microbiota or blockade of the downstream cellular or molecular immune mediators significantly suppress lung tumor growth.



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RESEARCH ARTICLE

Gut microbiome–mediated bile acid metabolism regulates liver cancer via NKT cells

CHI MA, MIAOJUN HAN, BERND HEINRICH, QIONG FU, QIANFEI ZHANG, MILAN SANDHU, DAVID AGDASHIAN, MASAKI TERABE, JAY A. BERZOFSKY, [...] TIM F. GRETEN +14 authors Authors Info & Affiliations

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Lung Cancer Study

Article

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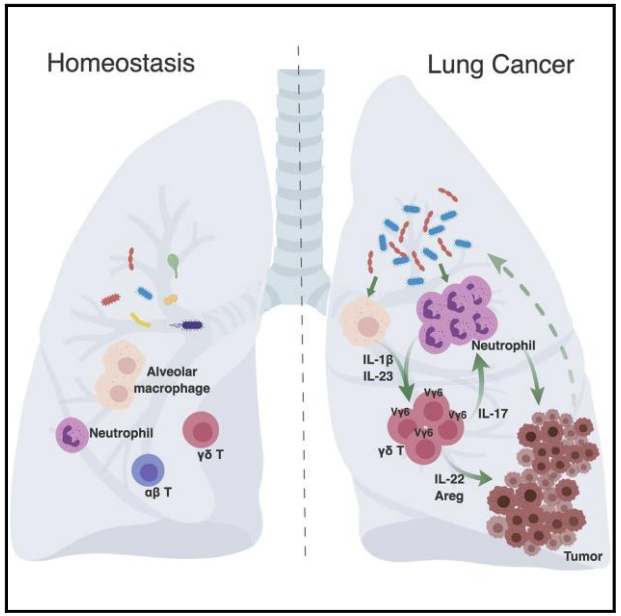

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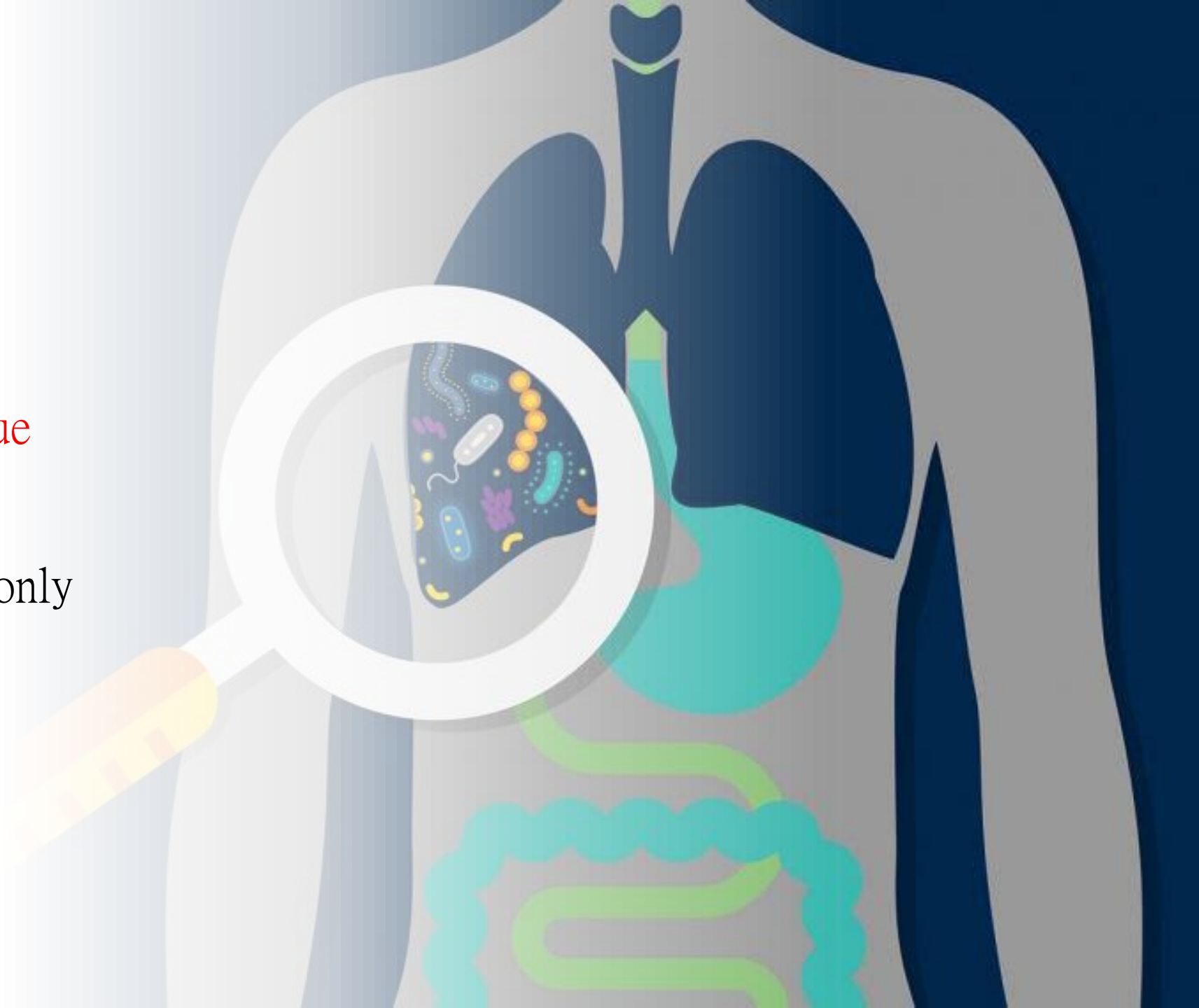
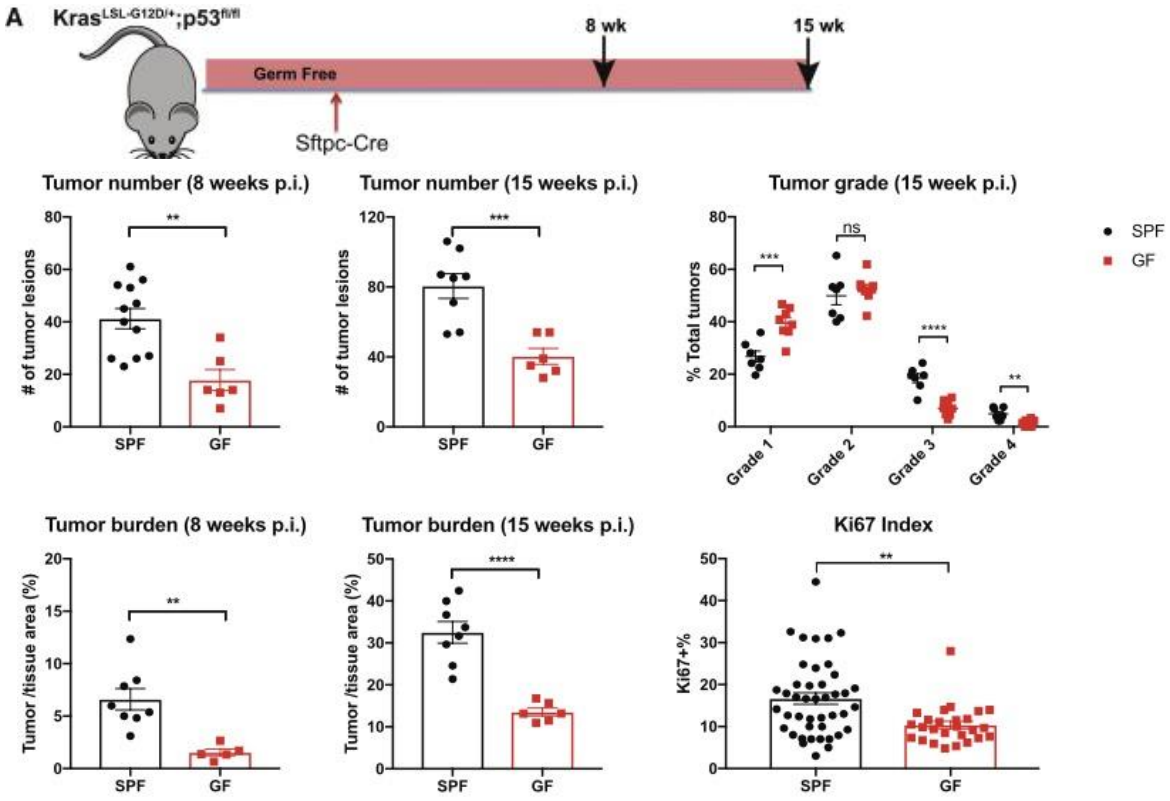
Lung Cancer

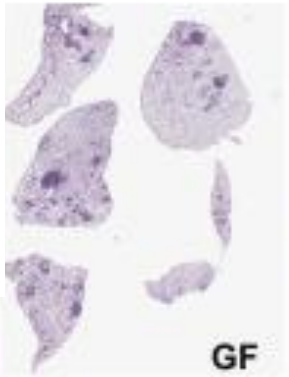
- The lung is a mucosal tissue colonized by a diverse bacterial community
- Bacterial infections commonly presenting in lung cancer patients could be linked to tumor development and clinical outcomes.

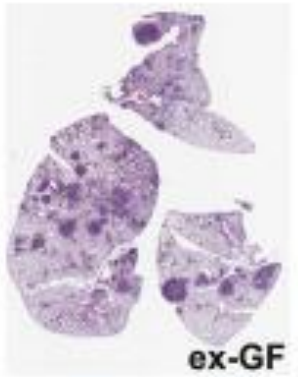


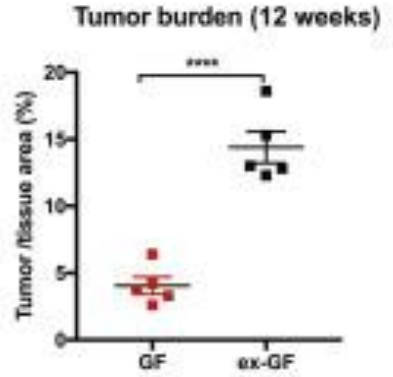
Animal model study

- Established mice model with oncogenic gene *KrasG12D* activation & tumor suppressor *p53* deletion
- Tumor development comparison: Germ-free (GF) mice VS. specific pathogen-free (SPF) mice
- Further co-housing GF mice with SPF mice (exposed to the microbiome)



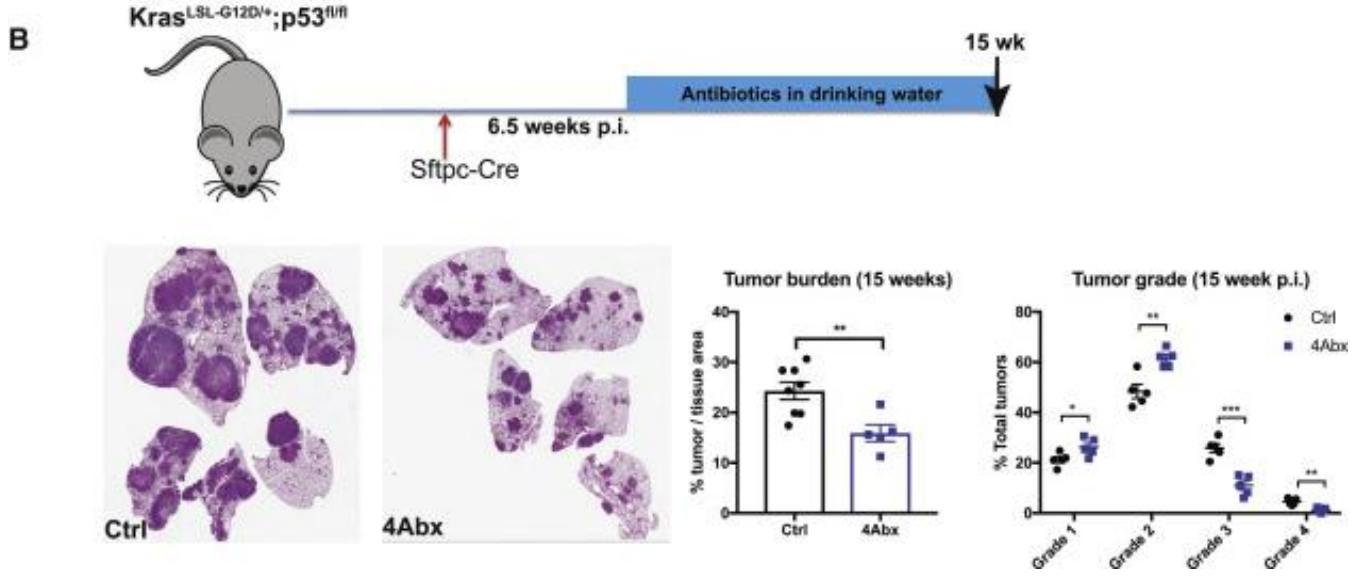






Animal model study

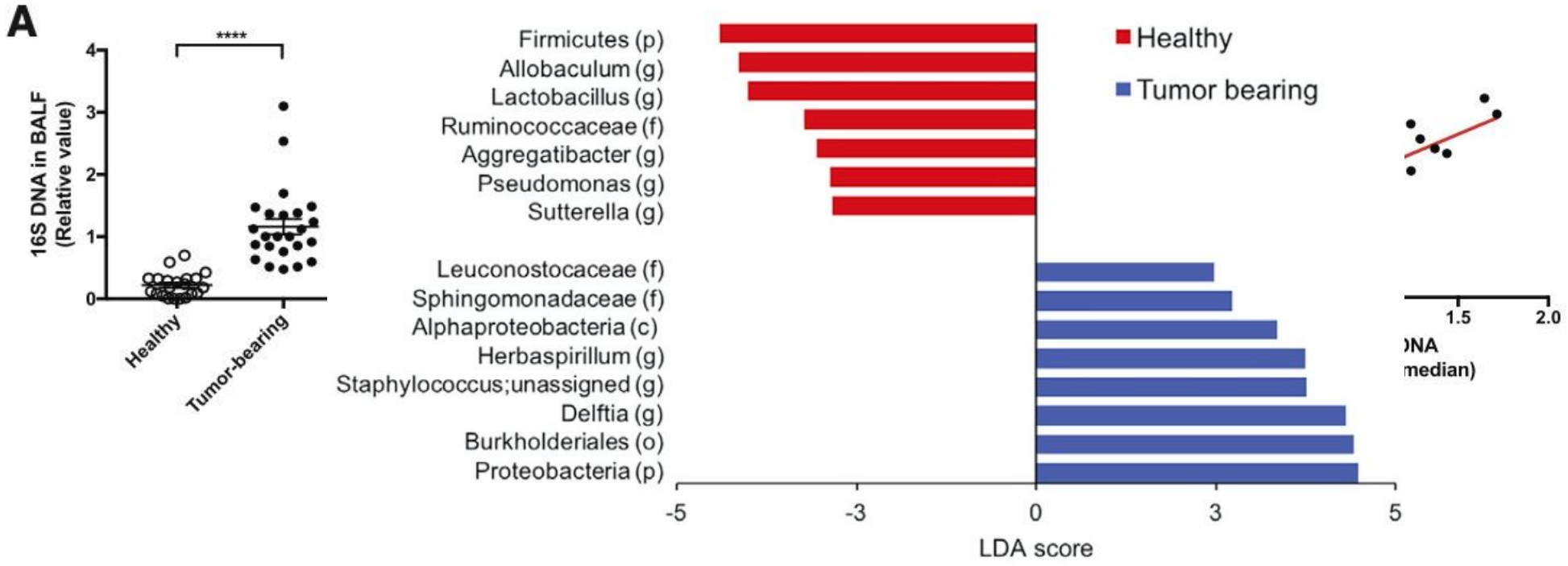
- Treated SPF KP mice with an antibiotic cocktail (4Abx) after tumor initiation



The exist of microbiota could significantly promote lung tumor development.

Microbiome and Bioinformatics analysis

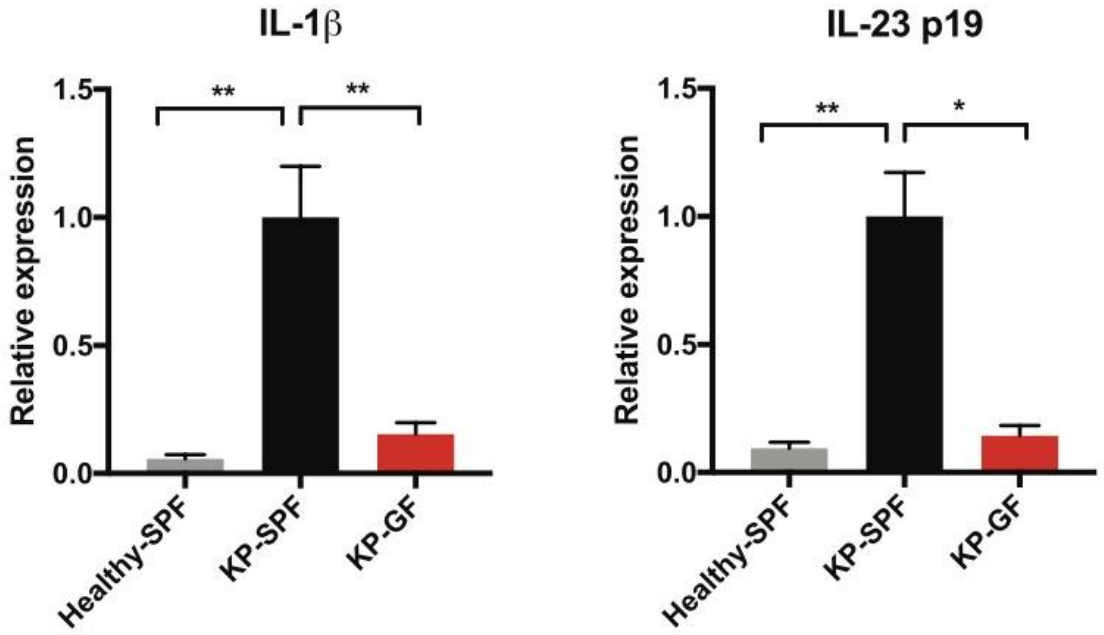
- 16S V1-V2 sequencing methods of bacteria abundance analysis in the bronchoalveolar lavage fluid (BALF).

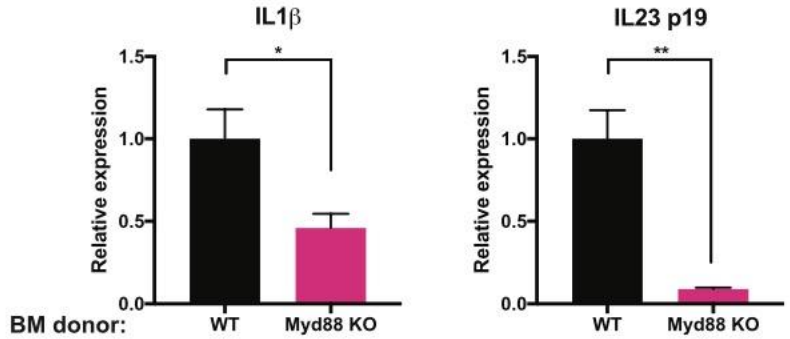


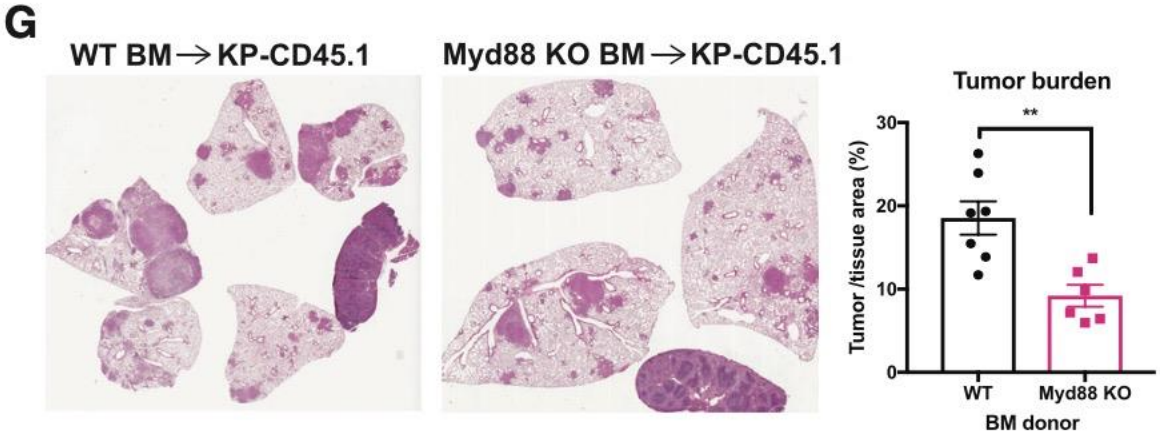
Gene expression study

Microbial products derived from intestinal microbiota are known to induce pro-inflammatory cytokines and mediate tumor-associated inflammation in colon cancer

- Real-time qPCR analysis of IL-1 β and IL-23 p19 mRNA expression in the lung tissue.

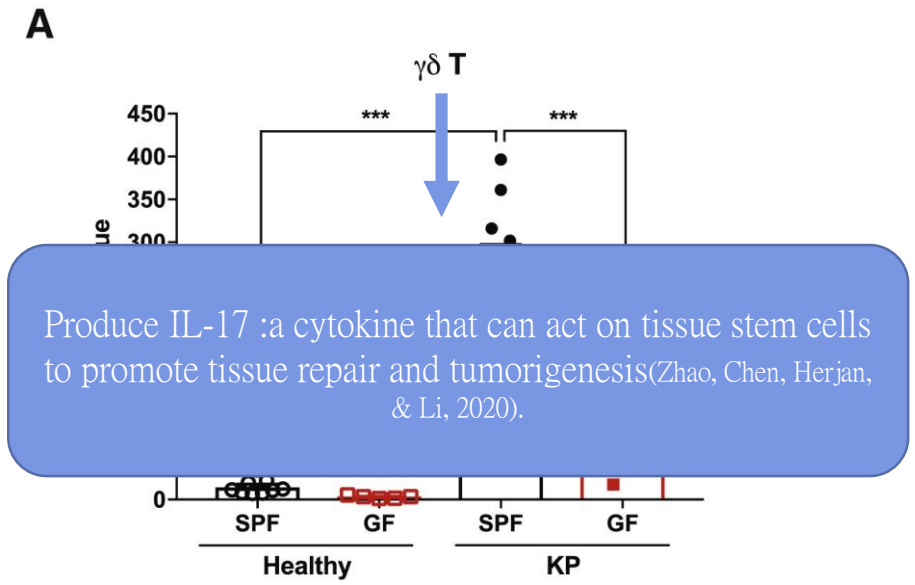


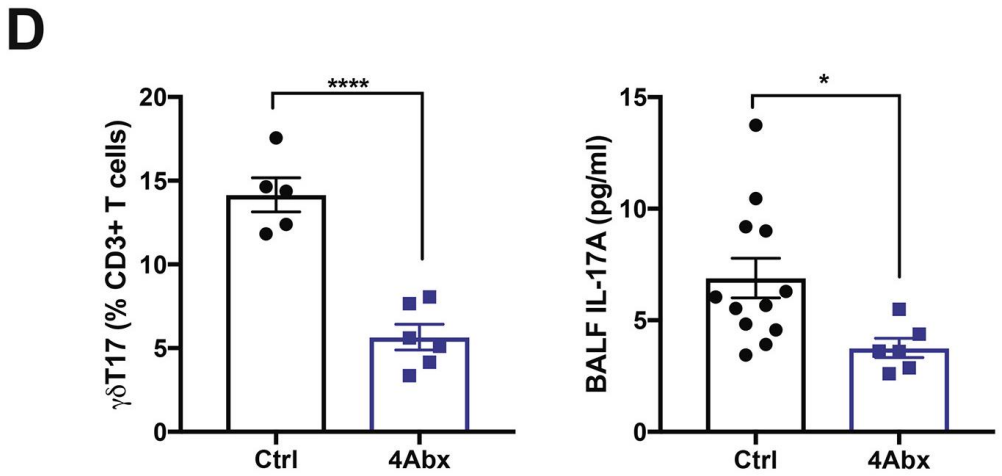




Target immune cells

- Analyse the immune cells in tumor-bearing lungs from GF and SPF mice by flow cytometry.
- gd T cell abundance was dominant in SPF tumor mice, but completely abrogated in the GF tumor mice
- Reducing the commensal microbiota with the 4Abx cocktail dramatically decreased the abundance of gd T17，resulting in lower IL-17A levels in the BALF or serum





Summary

- Lung cancer development is associated with local dysbiosis and inflammation
- Microbiota could drive proliferation and activation of gbT cells in lung cancer and result in the difference of related inflammatory factors.
- Depletion of commensal microbiota can suppress lung cancer development
- Did not give detail on the potentially functional bacteria species.

Liver Cancer Study

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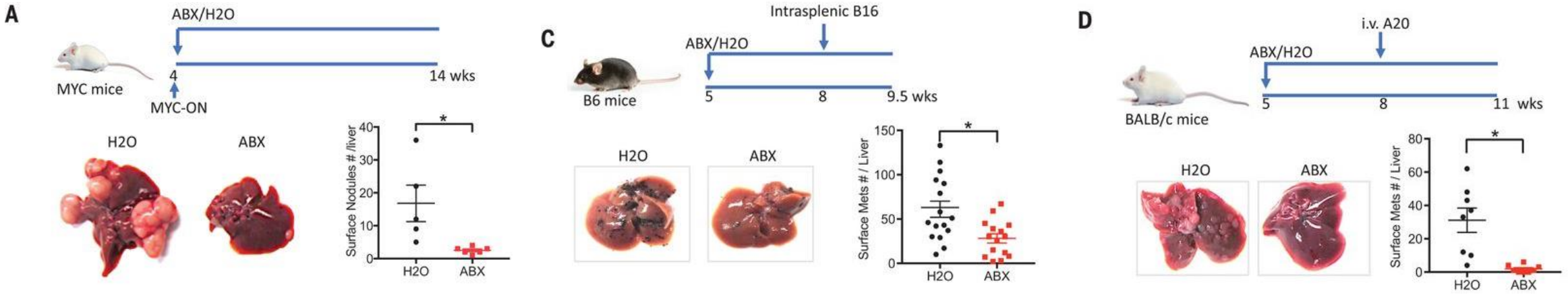
Animal model study

- Antibiotic cocktail treatment VS. H_2O control
- Modulating commensal bacteria can specifically modify growth kinetics of intrahepatic tumor.



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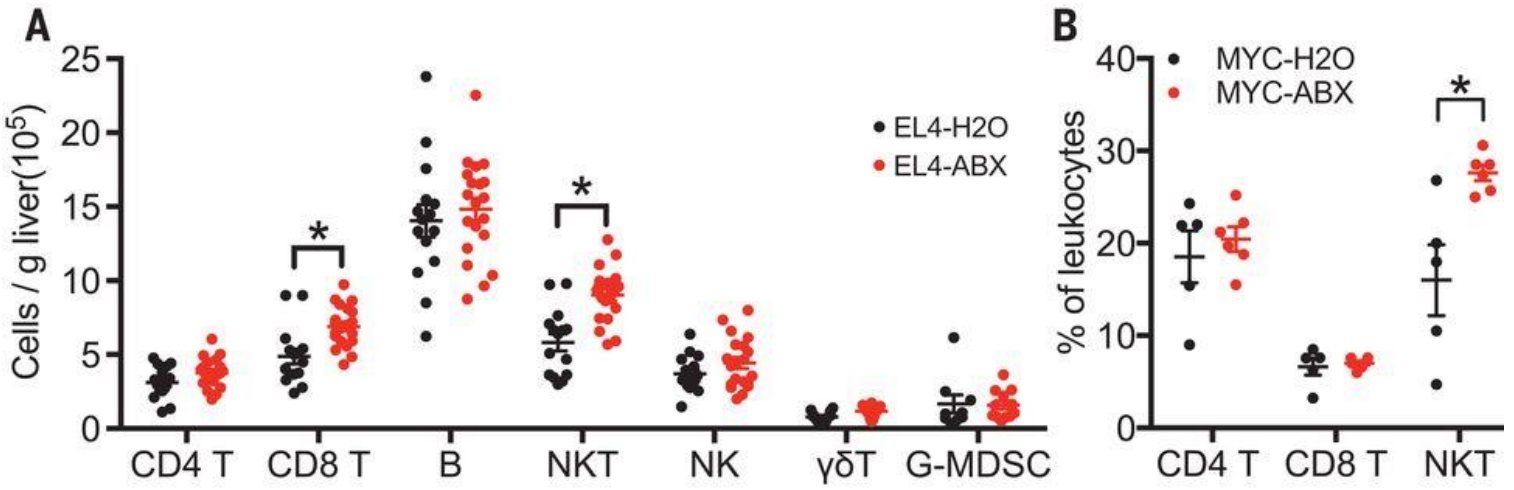
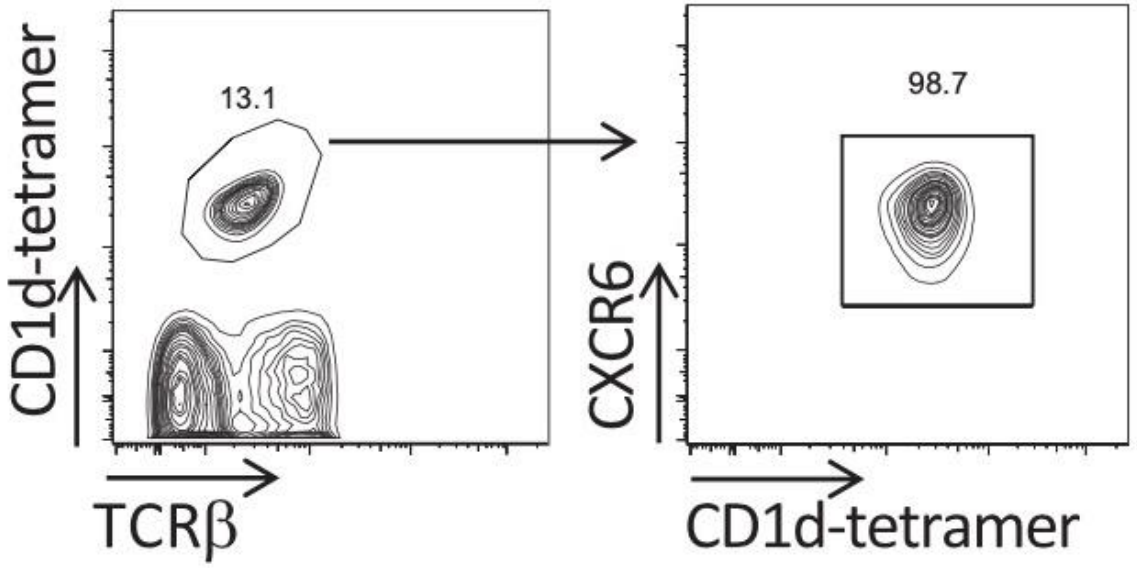
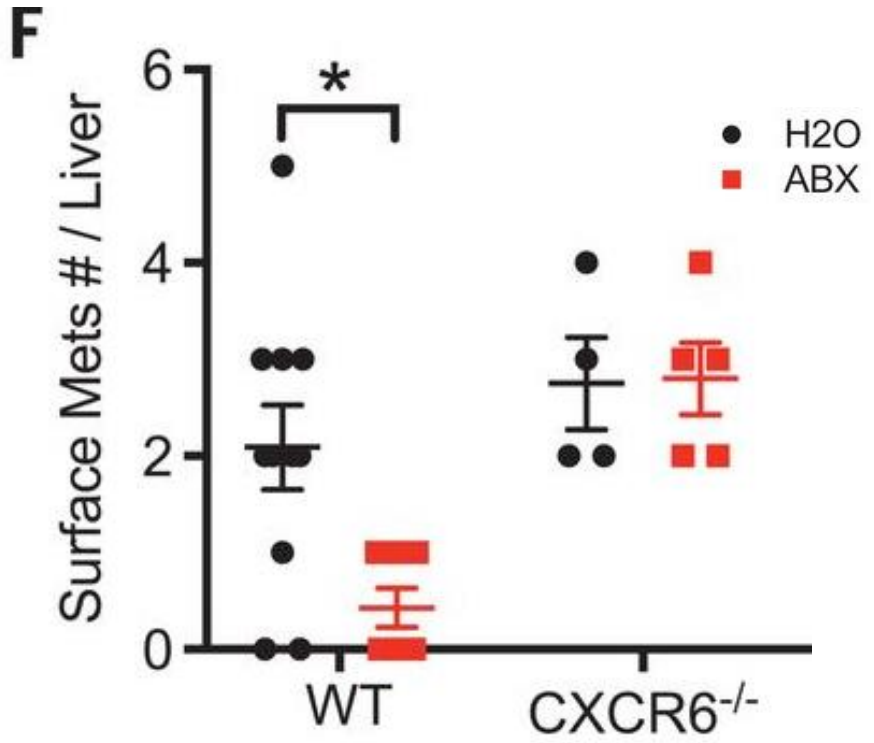


- Microbiota could mediate hepatic natural killer T cells accumulation in liver.

Target potential receptors on NKT

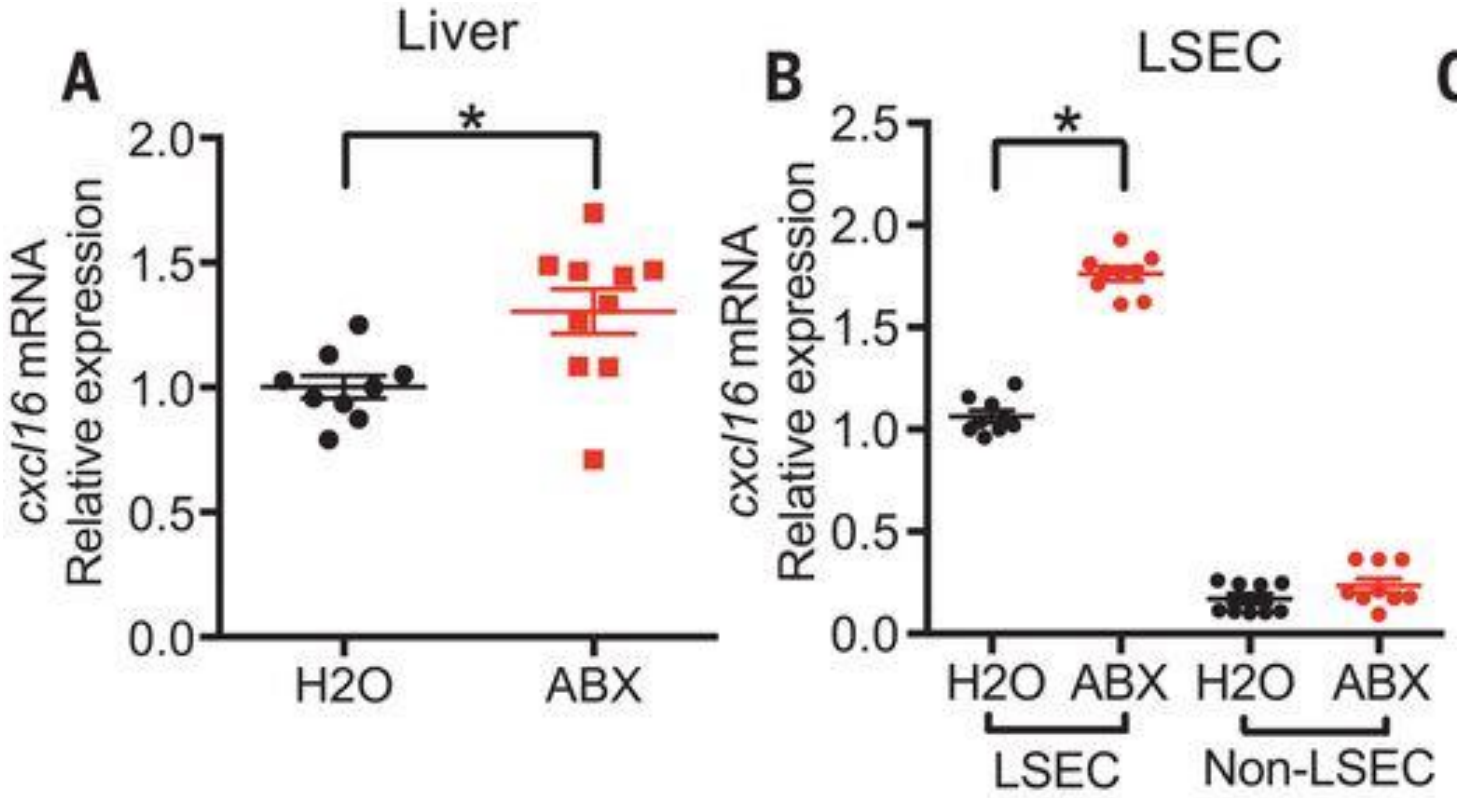
- All hepatic NKT cells expressed CXCR6.
- For CXCR6 knockout mice, no reduction in liver tumor could be found even receiving antibiotics treatments





Target corresponding bio-molecules

- CXCL16: solo ligand for CXCR6
- LSECs: the major source of CXCL16 production in the liver
- The concentration of CXCL16 could influence the accumulation of hepatic NKT through the binding of CXCR6 receptors.



Bile acid influenced the CXCL16 production level from LSECs

CXCL16 has both cell-surface and secreted forms; the cell-surface form has been identified to be involved in lipid metabolism

Bile acid is highly associated in the lipid metabolism among gut microbiome.

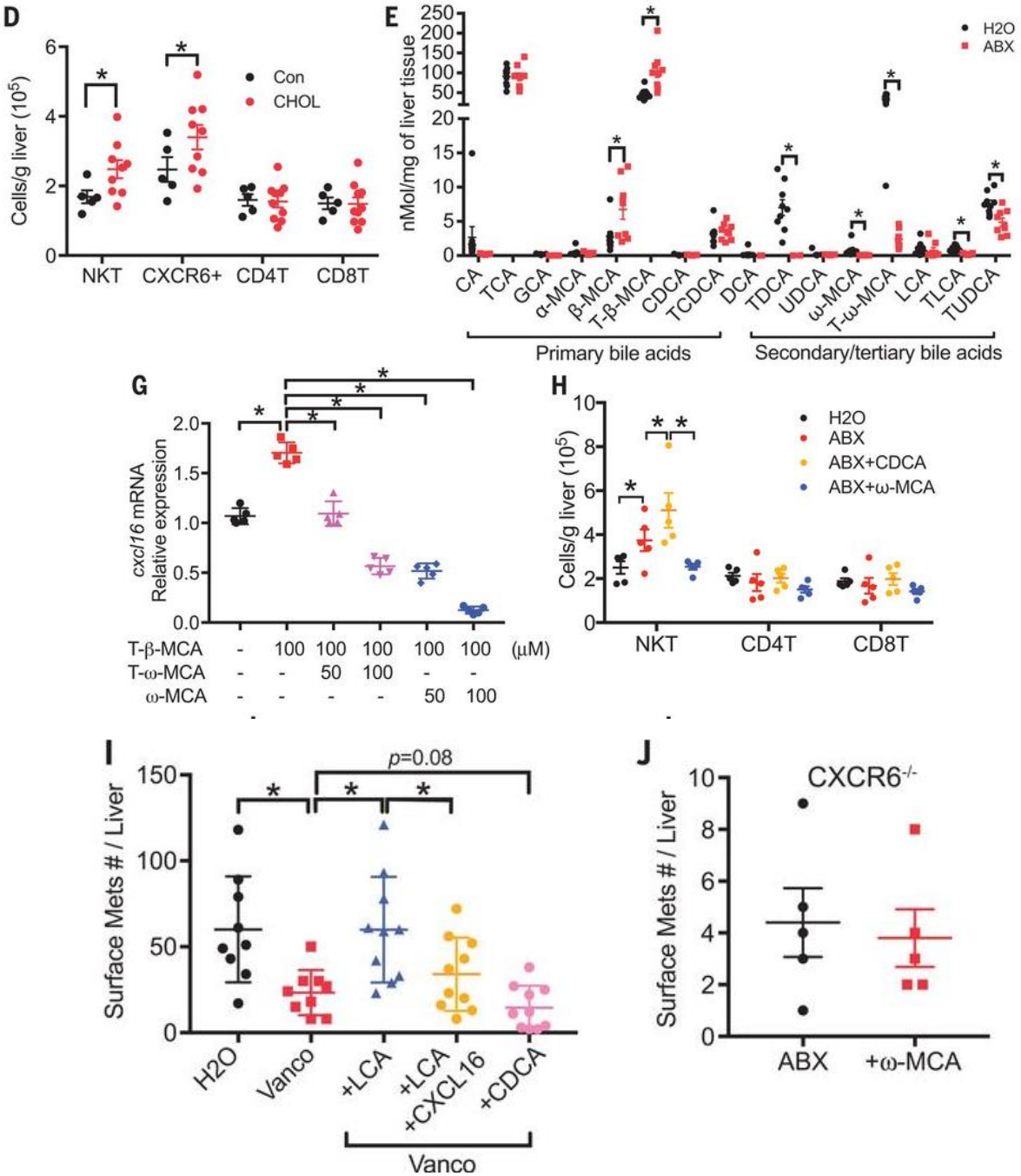
Primary bile acids could be synthesized by the liver.

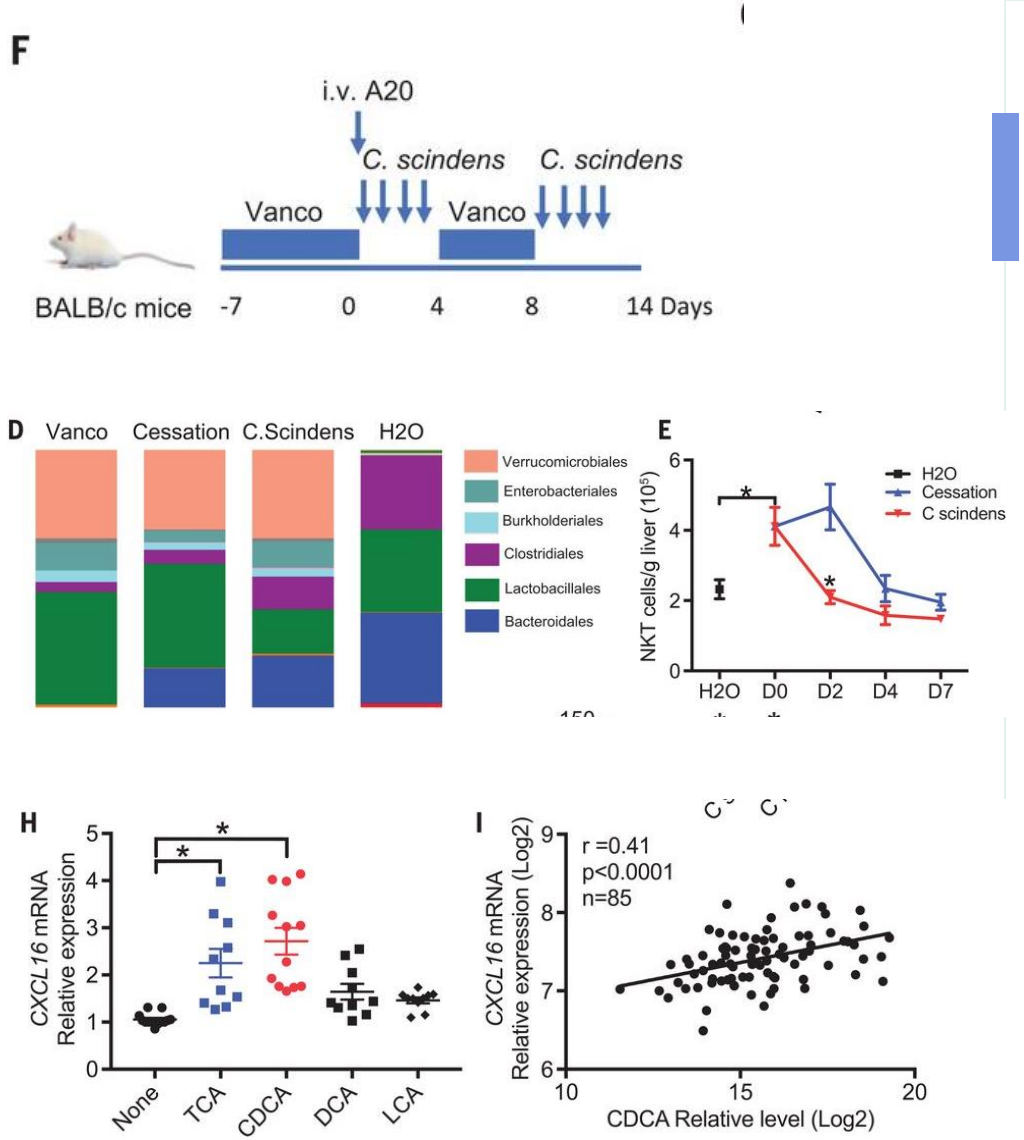
Secondary bile acids result from gut bacterial actions.

Metabolite bile acids: different combination treatments for analysis

- CHOL: cholestyramine, a chemical could reduce bile acid levels in liver

	CXCL16 gene	CXC6+ NKT cell	Liver tumor
Primary bile acid	↑	↑	↓
Secondary bile acid	↓	↓	↑

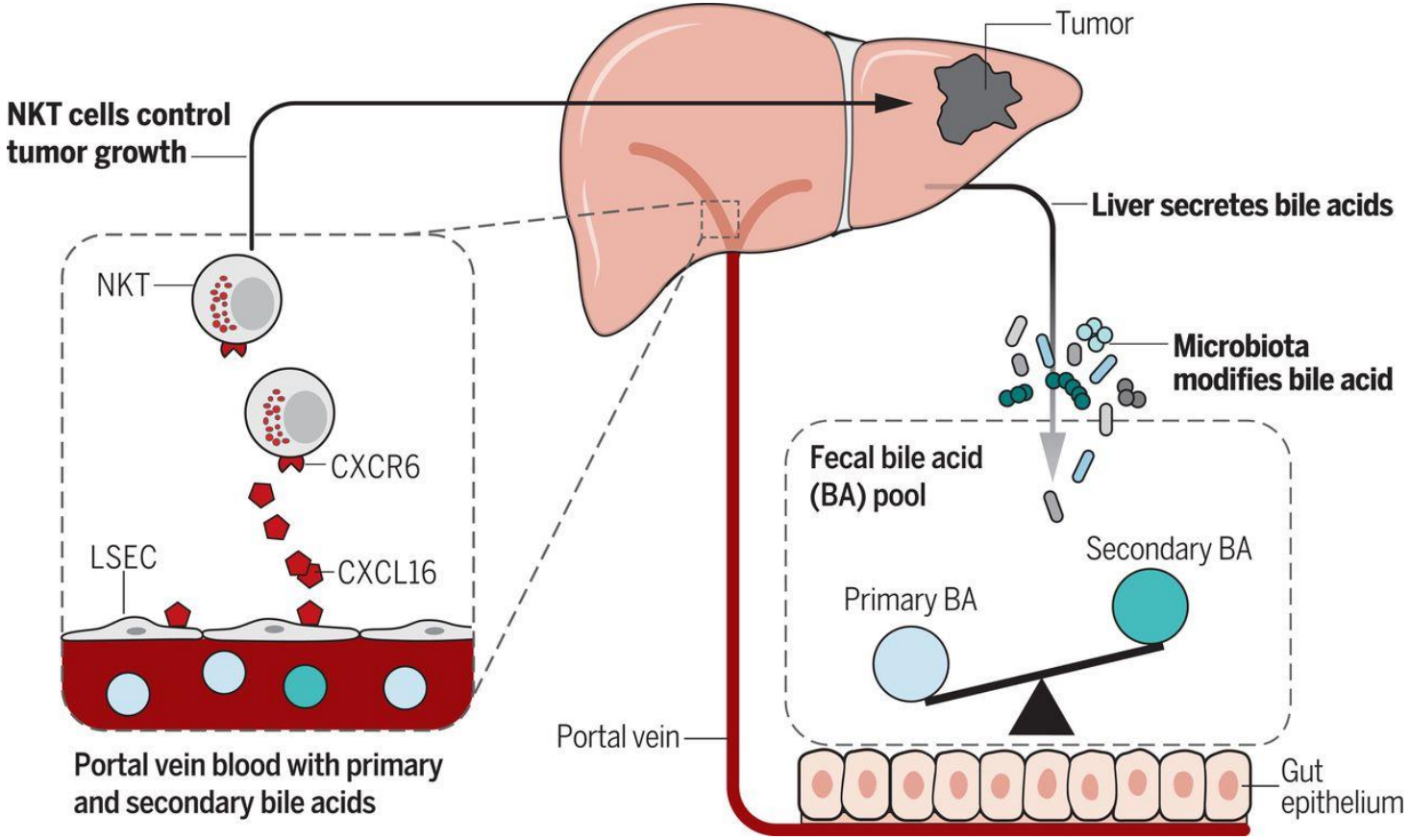




Target potential bacteria

- The secondary bile acids metabolites is highly related to gram-positive bacterium to Clostridium cluster XIV.
- Mice were fed with vancomycin for 1 week, then vancomycin was stopped, and the mice were given C. scindens or vehicle.
- In-vitro human LSECs study: Bile acids control liver CXCL16 expression in humans

Gut microbiome modulates liver cancer through bile acid – regulated NKT cells



Summary & Take-home message

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Human microbiota could influence tumorigenesis through TME indirectly.

Establish animal cancer model based on interested variable

Target specific cells and biomolecules(flow cytometry, ELISA, etc.)

Bioinformatic analysis and gene expression comparison (NGS, qPCR, RNA seq, etc.)

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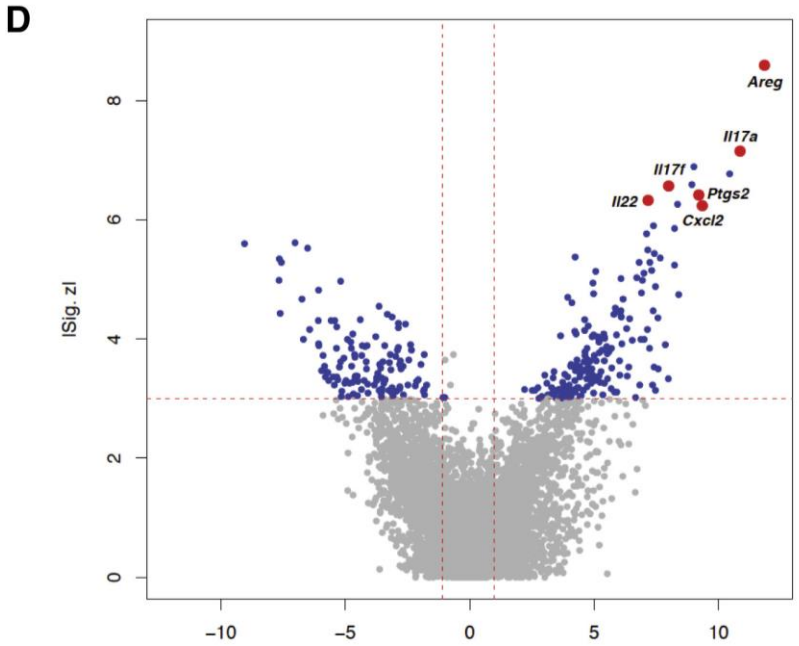
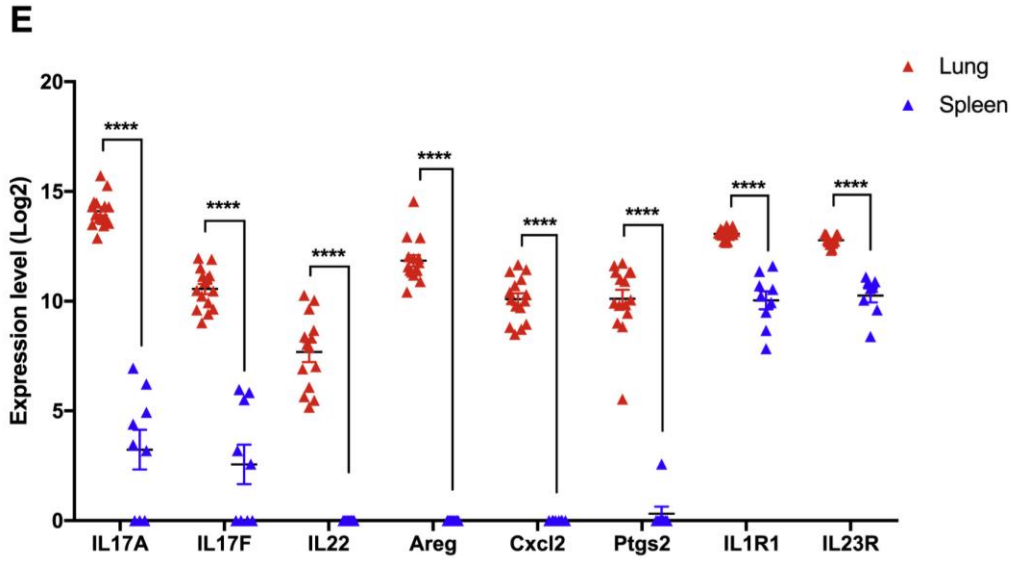
Q&A

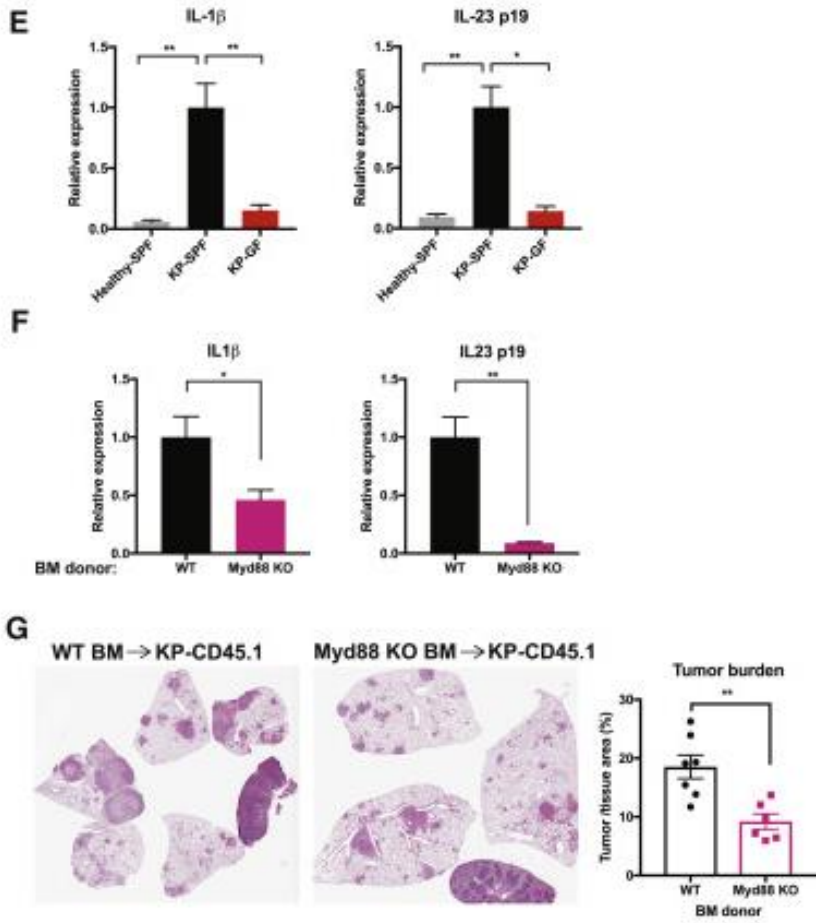
Method & Result (Bioinformatics analysis)

- Characterized the transcriptional profile in comparison to splenic gd T cells using RNA sequencing (RNA-seq).



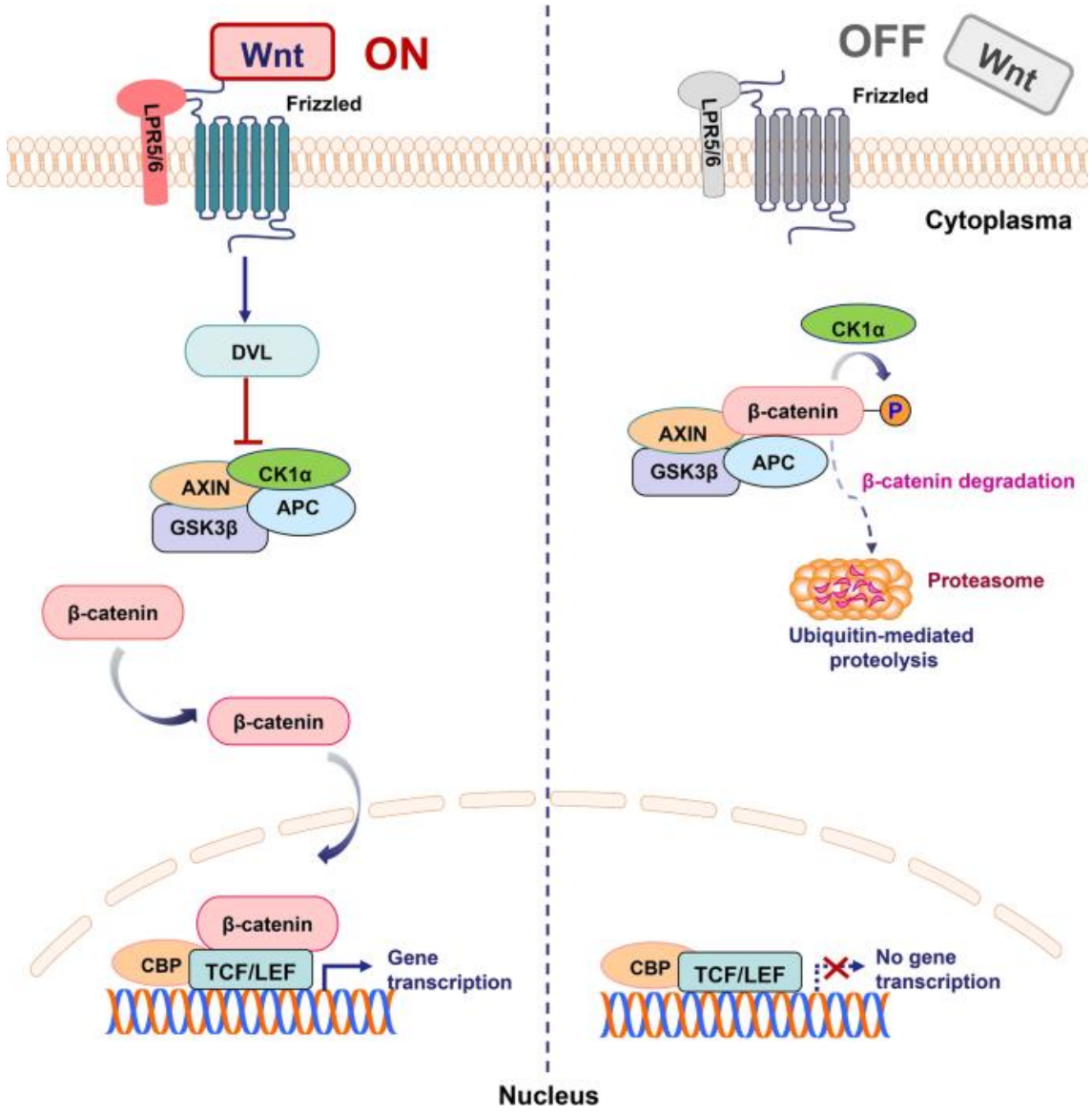


Method & Result



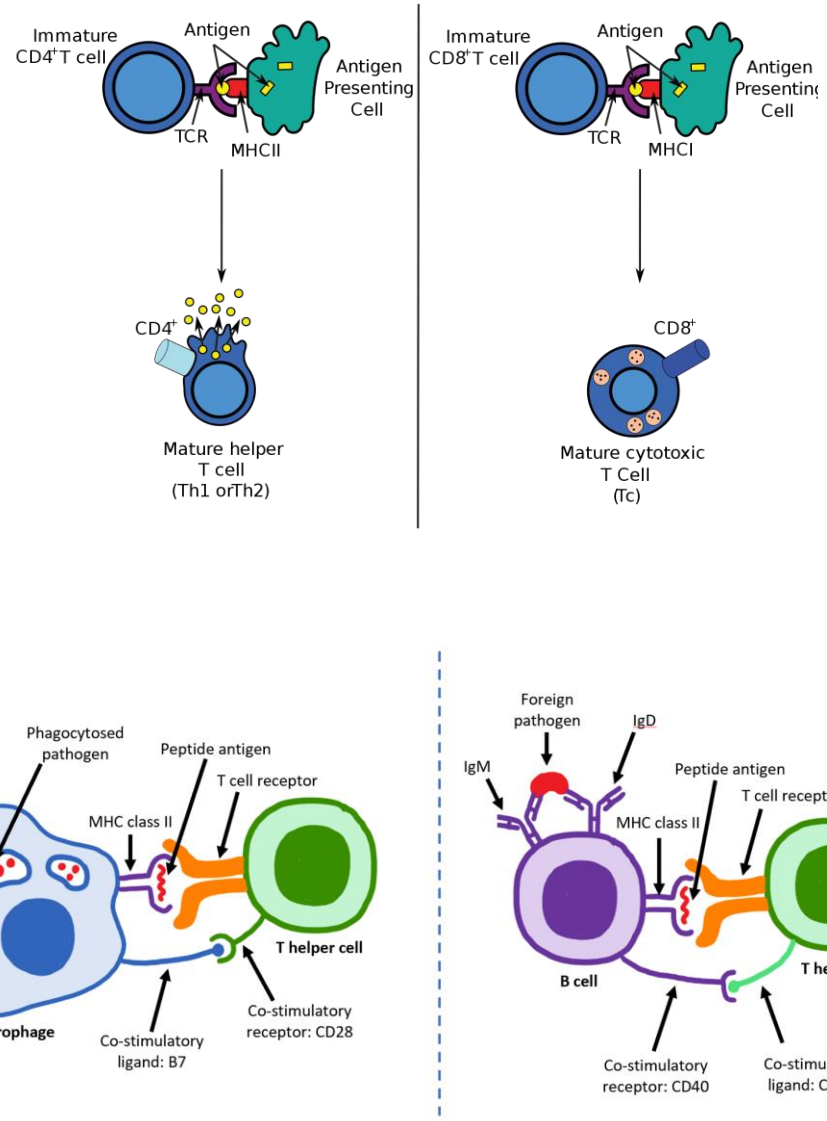
- IL-1b : is the member of the interleukin 1 family of cytokines. This cytokine is produced by activated macrophages and related to inflammation.
- IL-23: an inflammatory cytokine, which is a key cytokine for T helper cell (Th17) maintenance and expansion.

Wnt/beta-catenin pathway

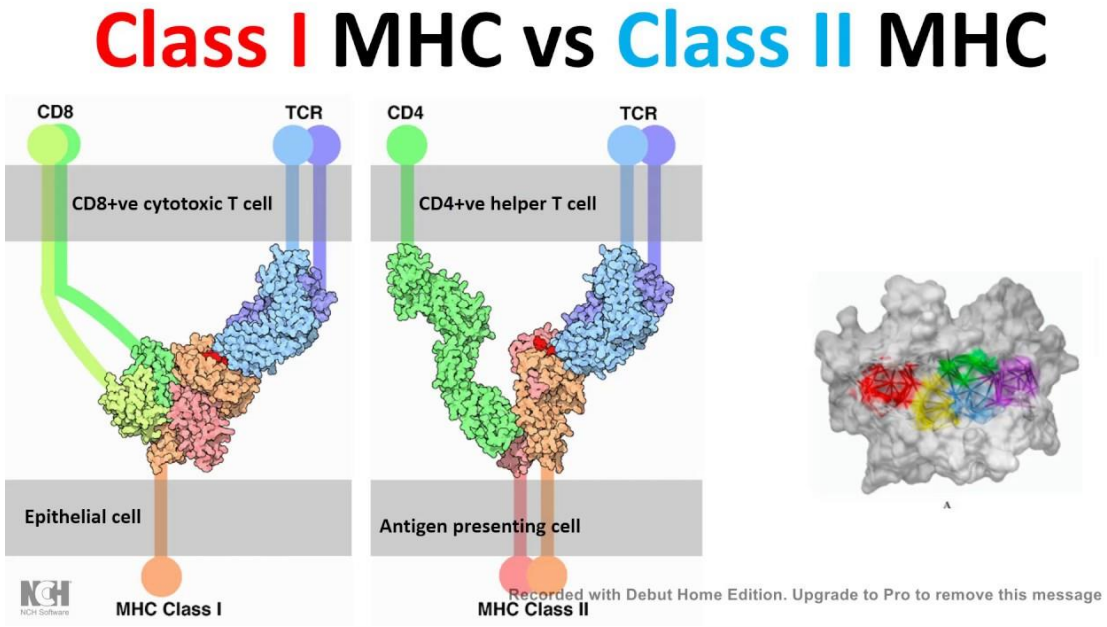


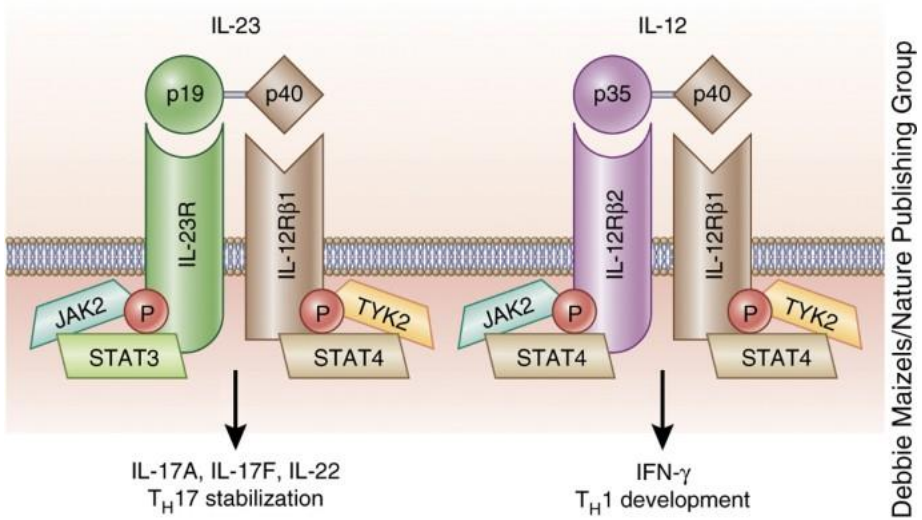
(Zhang & Wang, 2010)

- For T cell activation, there must be binding of the T cell receptor to both the antigen peptide and the MHC class II molecule on an antigen presenting cell (APC). Additionally, there must be binding of the two co-stimulatory molecules (B7 on the APC and CD28 on the T cell). For B cell activation, a pathogen must bind to the IgM and IgD antibodies in order to be internalized and presented on the MHC class II molecule of the B cell. Like T cell activation, there must be binding of the two co-stimulatory molecules (in this case CD40 with CD40L). Once a B cell is activated, it turns into a plasma cell which secretes antibodies.



IL-23 p19 & IL-1b





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