

# 병원기반 인간 마이크로바이옴 연구개발 사업 심포지엄

Human Microbiome and Clinical Application

2024.12. 3.(화) 8:30~17:00

노보텔 앰배서더(서울 동대문)



질병관리청  
국립보건연구원



보건복지부







# 병원기반 인간 마이크로바이옴 연구개발 사업 심포지엄

Human Microbiome and Clinical Application

## > 프로그램

시간	내용	발표자
08:30~09:20	등 록	
09:20~09:30	개 회 사	원장 박현영(국립보건연구원)
Session 1	Clinical application in diagnostics and therapeutics	좌장 이동우(연세대학교)
09:30~09:55	Establishment and results of clinical-based Inflammatory bowel disease cohorts for microbiome research	이창균(경희의료원)
09:55~10:20	Development of microbiome indicators: selection of indicators from microbiome and meta-data through artificial intelligence	최민혁(강남세브란스병원)
10:20~10:45	Development of microbiome solution based on precision classification platform	이제희(CJ Bioscience)
10:45~11:10	Development of microbiome treatment	임상균(고바이오랩)
11:10~11:35	Excavation and development of microbiome biomarkers applicable to clinical diagnosis	김지현(연세대학교)
11:35~13:10	점심시간 <b>패널토의</b> <b>좌장</b> 박현영(국립보건연구원) <b>패널</b> 이광준(국립보건연구원) / 김봉수(한림대학교) / 김병용(종근당건강) 설우준(중앙대학교) / 최준용(세브란스병원) / 안경진(서울경제신문)	
Session 2	Microbiome axis in health & disease	좌장 김봉수(한림대학교)
13:15~13:40	Understanding the host-microbiota interaction using an EAE animal model	이윤경(순천향대학교)
13:40~14:05	The gut-kidney axis in CKD: role of the microbiome	김지은(고려대학교구로병원)
14:05~14:30	Gut-lung axis: from epidemiology to therapeutic application	이세원(서울아산병원)
14:30~14:55	Gut-liver axis in health & disease	석기태(한림대학교춘천성심병원)
14:55~15:15	휴 식	






**Session 3****A guide to human microbiome research  
: Database & Bioinformatics analysis****좌장 정경태(국립보건연구원)**

15:15~15:40	Human microbiome reference and resource development for the clinical applications in diagnosis and therapeutics	이선재(광주과학기술원)
15:40~16:05	Analysis of microbiome genetic information using artificial intelligence	노미나(한양대학교)
16:05~16:30	Development of human virome analysis research methodology	이하나(고려대학교)
16:30~16:55	Approach of human microbiome data standardization : From sample collection to data analysis	이송희(국립보건연구원)
16:55~17:00	폐 회 사	이광준(국립보건연구원)





※ 상기 프로그램은 행사 진행 사정에 따라 변경이 될 수 있습니다.

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 이송희(국립보건연구원)



# 개회사



- ✓ 소속: 질병관리청 국립보건연구원
- ✓ 직위/직책: 원장
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1990	연세대학교 의학과 의학학사
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2002 ~ 2005	연세대학교 의과대학 부설연구소 연구조교수
2005 ~ 2005	질병관리청 국립보건연구원 유전질환과장
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2023 ~ 현재	질병관리청 국립보건연구원장



## **Session 1.**

# **Clinical application in diagnostics and therapeutics**



1. Establishment and results of clinical-based Inflammatory bowel disease cohorts for microbiome research
2. Development of microbiome indicators: selection of indicators from microbiome and meta-data through artificial intelligence
3. Development of microbiome solution based on precision classification platform
4. Development of microbiome treatment
5. Excavation and development of microbiome biomarkers applicable to clinical diagnosis







# 1부 좌장



- ✓ 소속: 연세대 생명공학과
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1997	연세대학교 생명공학과 공학학사
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2004 ~ 2010	펜실베니아대학교(Univ. of Pennsylvania) 박사후연구원
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2021 ~ 2024	연세대학교 교무처 교무부처장
2024 ~ 현재	국제극한환경미생물학회2026 조직위원장



01

## Establishment and results of clinical-based Inflammatory bowel disease cohorts for microbiome research



이창균  
(경희의료원)







## 1부 연자



- ✓ 소속: 경희대학교 의과대학, 경희대학교병원 소화기내과
- ✓ 직위/직책: 교수, 염증성장질환센터장, 임상시험센터장
- ✓ 성명: 이창균

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1998	경희대학교 의과대학 졸업
2003	의학석사, 경희대학교 대학원
2008	의학박사, 경희대학교 대학원

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2008 ~ 2010	순천향대학교 의과대학, 순천향대학교 천안병원
2015 ~ 2016	Cedars-Sinai Medical Center, US, 방문연구원
2011 ~ 현재	경희대학교 의과대학, 경희대학교병원 소화기내과
2018 ~ 2020	대한장연구학회 섭외홍보이사
2023 ~ 현재	대한장연구학회 학술이사

## 마이크로바이옴 연구를 위한 병원기반 염증성장질환 코호트 구축과 성과

복지부 병원기반 인간 마이크로바이옴 연구 장내과제 소개 및 마이크로바이옴 연구를 위한 염증성장질환 코호트 구축, 연구성과와 현황 염증성장질환 장마이크로바이옴을 이용한 진단적 유용성 연구 소개 등

# Hospital-Based Inflammatory Bowel Disease Cohort for Microbiome Research

**Chang Kyun Lee, MD, PhD**

Center for Crohn's and Colitis, Clinical Trials Center

Kyung Hee University Hospital, Seoul, Korea

2024-11-28

Human Microbiome and Clinical Application

1

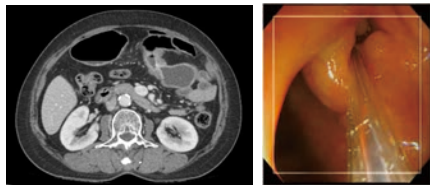
**Financial support:** This presentation was supported by a grant of the Korea Health Technology R&D Project through the Korea Health Industry Development Institute (KHIDI), funded by the Ministry of Health & Welfare, Republic of Korea (grant number : HI23C0661)

**COI:** Abbvie, Janssen, Takeda, BMS, Lilly, Celtrion, Samsung Bioepis, Pfizer, Esai, Yuhan Pharm.

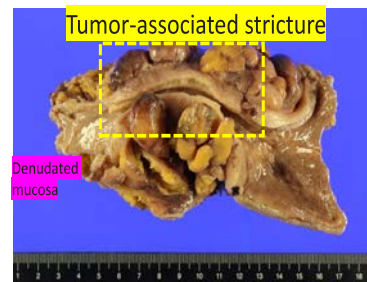
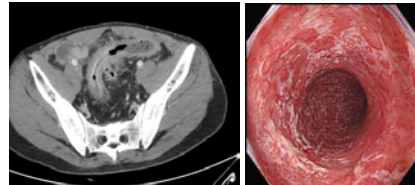
Human Microbiome and Clinical  
Application

2

## IBD is a progressive disease.



45 y-o male with Crohn's disease (CD), disease duration 11 y,  
A2,L3,B3p: Segmental SB resection after bowel perforation

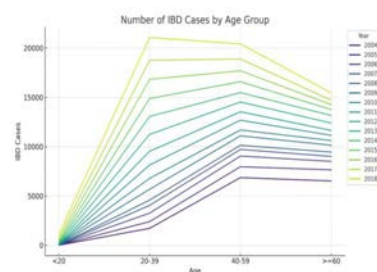
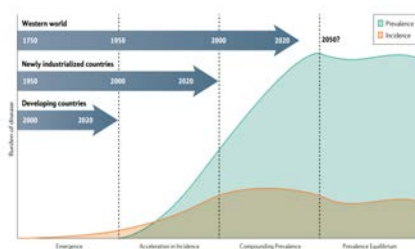


37 y-o male with ulcerative colitis (UC), disease duration 20 y, extensive colitis: advanced colon cancer presented with bowel perforation, stage IV

Human Microbiome and Clinical Application

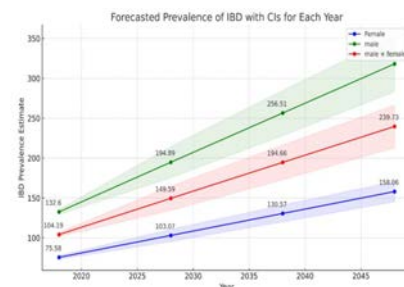
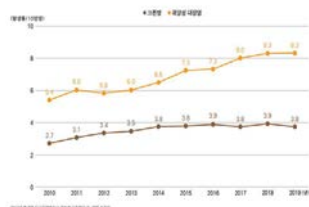
3

## Epidemiological burden of IBD



10년간 발생률

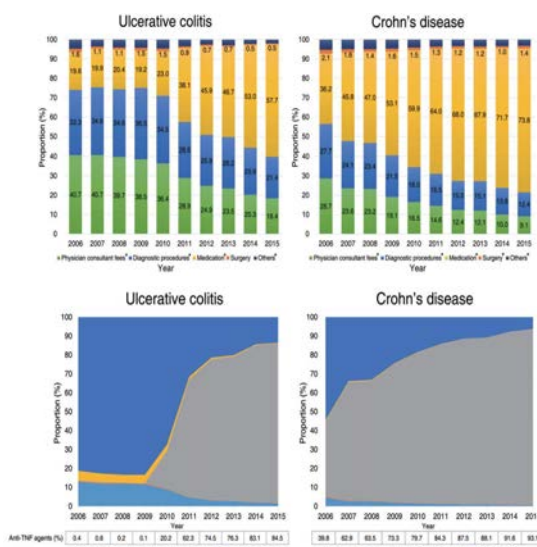
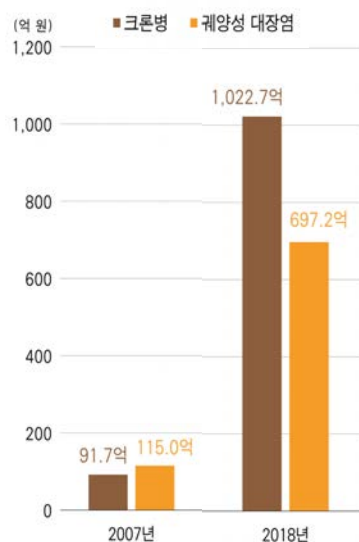
\* 조세공제 도입분의 합계율은 매년 증가하다가 세입의 대당인원 2018년까지, 조세율은 2014년까지 양자되는 것으로 보인다.



Samsung Bioepis Epiztek

4

## Economic burden

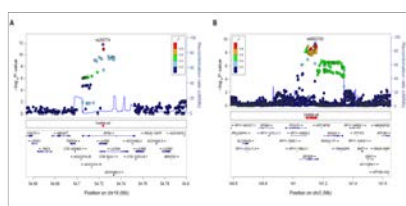


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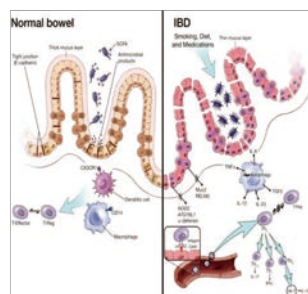
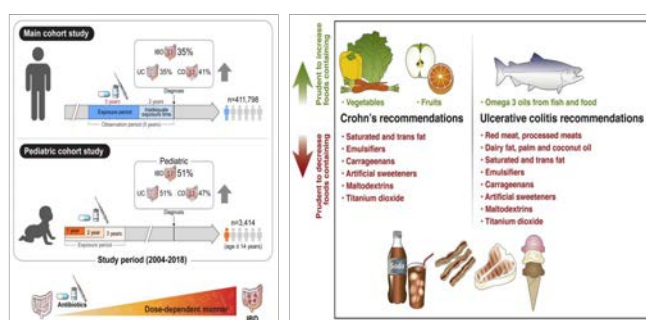
Kim JW, Lee CK et al. Scand J Gastroenterol. 2019 Apr;54(4):419-426; Kim JW, Lee CK et al. J Gastroenterol Hepatol. 2018 Apr;33(4):847-854; IBD fact sheets in Korea 2020;

## What triggers the development of IBD?



	Relative Risk	Absolute Risk
Lifetime risk for IBD	1x	1.3%
Familial aggregation		
IBD in first-degree relatives	4-15x	5.2-19.5%
Both parents affected	20-25x	30%
Genetic factors		
NOD2 variant <sup>1</sup>	2.1-3.0x	2.7-3.9%
Typical susceptibility variants	1.1-1.5x	1.4-2.0%
PRS—Individuals in the top 1%	3.9x	5.1%
Environmental factor		
Current smoking <sup>1</sup>	1.8x	2.3%

PRS, polygenic risk score. <sup>1</sup> Risk factor for Crohn's disease.



Human Microbiome and Clinical Application

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Lee CK. J Crohns Colitis. 2023 Jul 31;17(12):127-137; Lee HS et al. Cells. 2019 Jun 4;8(6):535; Oh SJ, Lee CK, AP&amp;T 2023 Jul;58(2):191-206; Levine A, et al. CGH 2020 May;18(6):1381-1392.

LETTER

doi:10.1038/nature21172

Precision editing of the gut microbiota ameliorates colitis

nature medicine

ARTICLES

https://doi.org/10.1038/nature21172

Corrected: Publisher Correction

Microbial network disturbances in relapsing refractory Crohn's disease

ARTICLE

OPEN

https://doi.org/10.1038/nature21172

Multi-omics of the gut microbial ecosystem in inflammatory bowel diseases

nature microbiology

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Article Published: 27 January 2022

Multi-omics analyses of the ulcerative colitis gut microbiome link *Bacteroides vulgatus* proteases with disease severity

Human Microbiome and Clinical Application 7

The abundance of nitrate-respiring oral microbe *Veillonella* is increased during inflammatory bowel diseases.

Opposing diet, microbiome, and metabolite mechanisms regulate inflammatory bowel disease in a genetically susceptible host

Human Microbiome and Clinical Application 8

## The Persistent Gap: Unmet Clinical Needs Despite Scientific Progress

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## The Persisting Dogma: No Gold Standard in IBD Diagnosis

Journal of Crohn's and Colitis, 2019, 144–164K  
doi:10.1093/ecco-jcc/jjy113  
Advance Access publication August 23, 2018  
ECCO Guideline/Consensus Paper

OXFORD

**ECCO Guideline/Consensus Paper**

**ECCO-ESGAR Guideline for Diagnostic Assessment in IBD Part 1: Initial diagnosis, monitoring of known IBD, detection of complications**

Christian Maaser,<sup>a</sup> Andreas Sturm,<sup>b</sup> Stephan R. Vavricka,<sup>c</sup> Torsten Kucharzik,<sup>d</sup> Gionata Fiorino,<sup>e</sup> Vito Annese,<sup>f</sup> Emma Calabrese,<sup>g</sup> Daniel C. Baumgart,<sup>h</sup> Dominik Bettenworth,<sup>i</sup> Paula Borralho Nunes,<sup>j,k</sup> Johan Burisch,<sup>k,l</sup> Fabiana Castiglione,<sup>m</sup> Rami Eliakim,<sup>m</sup> Pierre Ellul,<sup>n</sup> Yago González-Lama,<sup>o</sup> Hannah Gordon,<sup>o</sup> Steve Halligan,<sup>o</sup> Konstantinos Katsanos,<sup>p</sup> Uri Kopylov,<sup>m</sup> Paulo G. Kotze,<sup>q</sup> Eduards Krustins,<sup>q</sup> Andrea Laghi,<sup>r</sup> Jimmy K. Limdi,<sup>r</sup> Florian Rieder,<sup>r</sup> Jordi Rimola,<sup>s</sup> Stuart A. Taylor,<sup>t</sup> Damian Tolan,<sup>t</sup> Patrick van Rhee,<sup>u</sup> Bram Verstockt,<sup>bb</sup>, Jaap Stoker<sup>cc</sup>, on behalf of the European Crohn's and Colitis Organisation [ECCO] and the European Society of Gastrointestinal and Abdominal Radiology [ESGAR]

JCC

### Statement 1.1. ECCO-ESGAR Diagnostics GL [2018]

A single reference standard for the diagnosis of Crohn's disease [CD] or ulcerative colitis [UC] does not exist. The diagnosis of CD or UC is based on a combination of clinical, biochemical, stool, endoscopic, cross-sectional imaging, and histological investigations [EL5]



# How to identify patients who are most likely to benefit form specific agents?

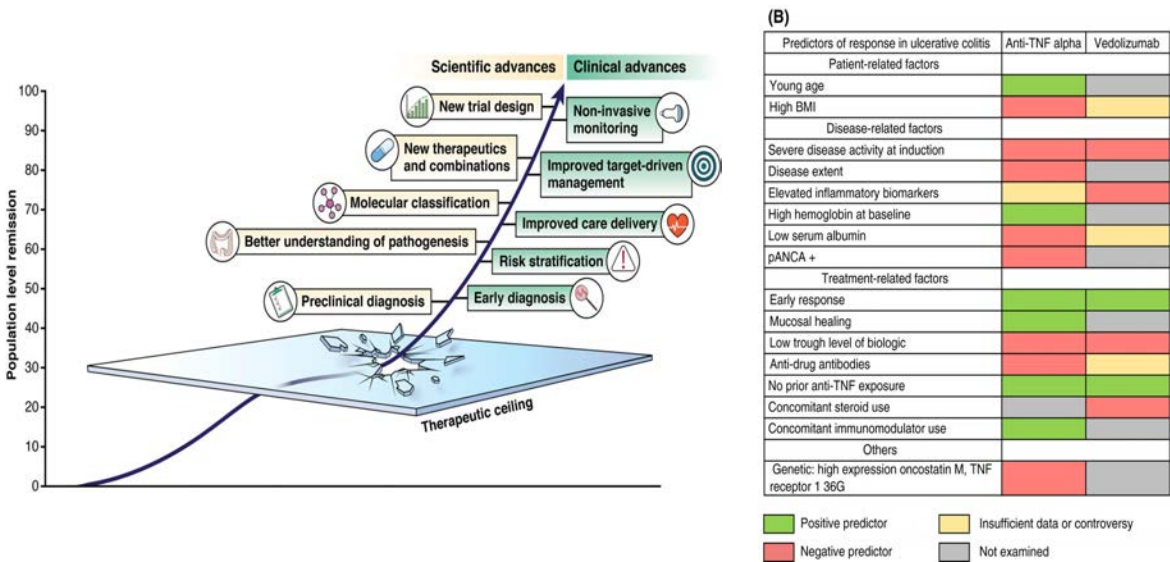
Disease severity variables	CD	UC	Disease activity variables*
Clinical			Clinical
Limited anatomic extent	X	X	Symptoms
Growth impairment (pediatric)	X	X	Biomarkers
Age at diagnosis <14 years	X	X	C-reactive protein
Age at diagnosis <40 years (adults)	X	X	Fecal calprotectin
Smoking history	X		Albumin
Perianal or severe rectal disease	X		Hemoglobin
Penetrating disease	X		Endoscopic
Strictureing disease	X		Disease activity scores
Multi- or long-segment ileal (>20 cm), disease proximal to T1	X		
Extensive bowel involvement	X	X	
Emergent diagnosis, hospitalization	X	X	
Delay in diagnosis	X	X	
Need for systemic steroids	X	X	
C difficile, cytomegalovirus infection		X	
Serologies			
ASCA (+)	X		
ANCA (+)	X	X	
Anti-Cbir (+) (UC; pediatric)	X	X	
Anti-GMCSF (+)	X		
Genetic			
NOD2 mutation	X		

X Mild  
X Severe

\* Based on data from Torres et al, Lancet, 2017 (CD) and Rubin et al, AJG, 2019 (UC)

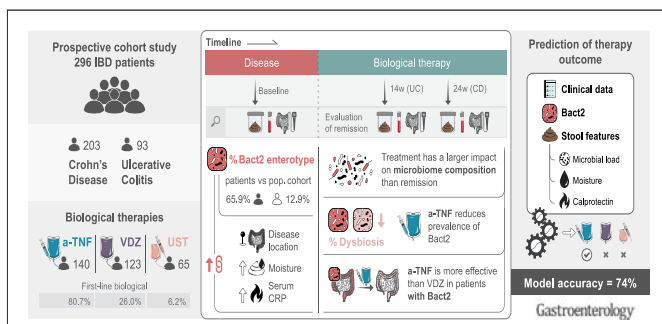
Agrawal M, et al. Gastro. 2021.

# The Therapeutic Ceiling



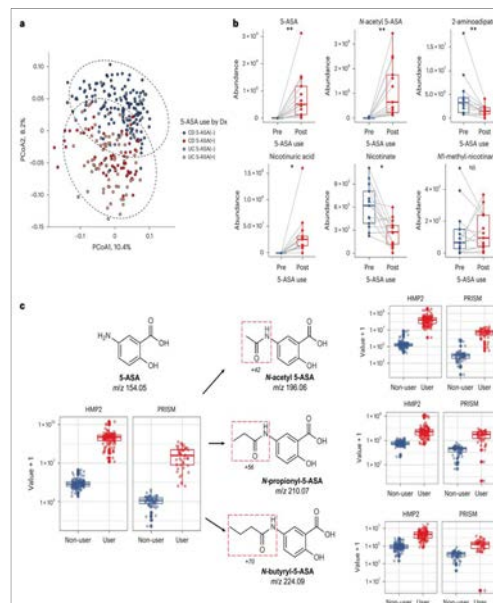


## Gut microbiota as a novel biomarker for predicting efficacy of advanced therapies in IBD



**The gut microbiome may be one solution, but it's not a complete answer on its own.**

**5-ASA directly impacts the fecal metabolome and undergoes biotransformation by the microbiome.**



Human Microbiome and Clinical Application

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Caenepeel C, et al. *Gastroenterology*. 2024 Mar;166(3):483-495. Mehta RS et al. *Nat Med*. 2023 Mar;29(3):700-709

## The Korean IBD Cohort: A Multi-omics Approach

Human Microbiome and Clinical Application

14

## Building a cohort is the foundation for further study.

**Gut Microbiome in Inflammatory Bowel Disease**

ClinicalTrials.gov ID **NCT03589183**

Sponsor **Kyunghee University Medical Center**

Information provided by **Chang Kyun Lee, Kyunghee University Medical Center (Responsible Party)**

Last Update Posted **2023-03-10**

**Study Details** | Researcher View | No Results Posted | Record History

**On this page**

- Study Overview
- Contacts and Locations
- Participation Criteria
- Study Plan
- Collaborators and Investigators
- Publications
- Study Record Dates
- More Information

**Study Overview**

**Brief Summary**

Inflammatory bowel disease (IBD) is a chronic inflammatory condition for gastrointestinal tract. There have been numerous studies to reveal dysbiosis of fecal/mucosal microbiome composition in IBD patients but actual trend of dysbiosis is strikingly different among patient's ethnicity.

In this background, the investigators have composed a prospective cohort of Korean IBD patients in a large academic referral IBD center. Using an integrated multi-omics bioinformatic analysis, the investigators aim to explore gut microbial signatures along with distinct clinical/genetic features, and their potential interplay in patients with IBD.

[Show less](#)

**Detailed Description**

This prospective cohort study aims to build a gut microbiome library for Korean IBD patients, and also aims to explore gut microbial signatures along with distinct clinical/genetic features and their potential interplay using the investigator's multi-

**Study Start (Actual)**  
2018-04-01

**Primary Completion (Estimated)**  
2028-03-31

**Study Completion (Estimated)**  
2028-03-31

**Enrollment (Estimated)**  
1500

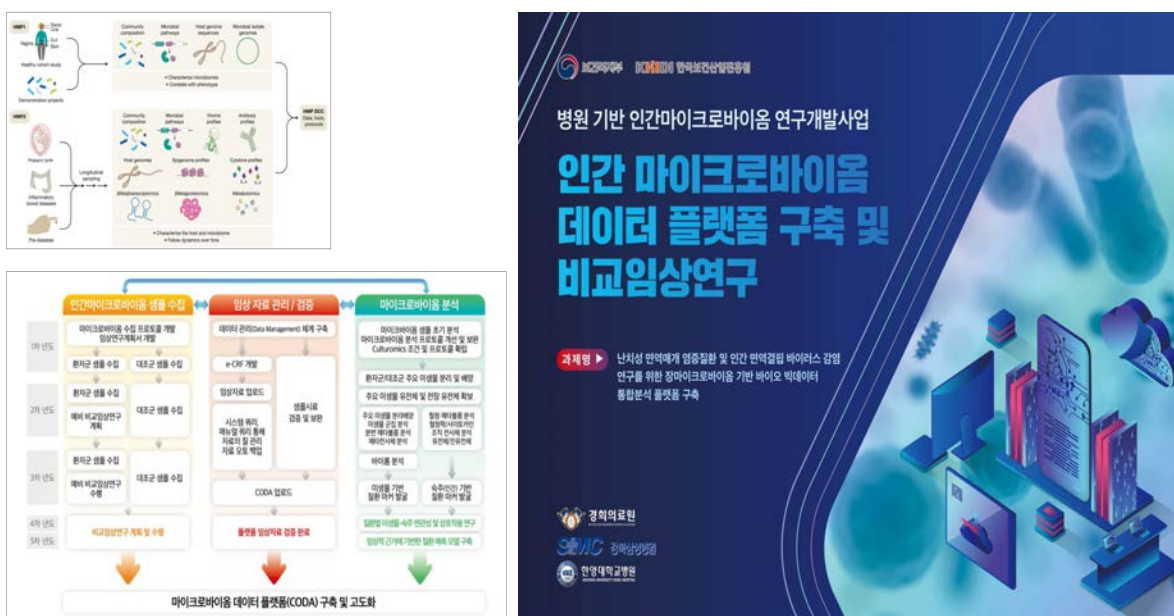
**Study Type**  
Observational

Outcome Measure	Measure Description	Time Frame
Taxonomic profiling of gut microbiome	Fecal and/or intestinal mucosal samples are obtained from enrolled subjects for metagenomic sequencing. After extracting fecal or mucosal DNA, taxonomic profiles associated with inflammatory bowel diseases and different disease phenotypes are analyzed from 16sRNA high-throughput sequencing or Shotgun method.	Up to 10 years
Finding of single nucleotide polymorphisms (SNPs)	Blood samples are obtained from enrolled subjects for genome-wide microarray. After extracting DNA, SNPs related to inflammatory bowel diseases and different disease phenotypes are explored (Genome-wide association study [GWAS] statistical significance threshold, $P < 5.00 \times 10^{-8}$ ).	Up to 10 years
Finding of serologic biomarkers	Blood samples are obtained from enrolled subjects for proteomic analysis. Serologic biomarkers implicating in inflammatory bowel diseases and disease phenotypes are explored.	Up to 10 years
Correlation between host genotyping and gut microbiome	Blood samples are obtained from enrolled subjects for genome-wide microarray. After extracting DNA, SNPs related to inflammatory bowel diseases and different disease phenotypes are explored (Genome-wide association study [GWAS] statistical significance threshold, $P < 5.00 \times 10^{-8}$ ). Fecal and/or intestinal mucosal samples are obtained from enrolled subjects for metagenomic sequencing. After extracting fecal or mucosal DNA, taxonomic profiles associated with inflammatory bowel diseases and different disease phenotypes are analyzed from 16sRNA high-throughput sequencing or Shotgun method. Taxonomic composition of gut microbiome are compared according to the sequencing data of host genomes.	Up to 10 years
Liquid biopsy biosignatures assessed by single cell RNA-Seq	Blood and intestinal mucosal biopsy samples are obtained from enrolled subjects, particularly those treated with biological drugs or small molecules for single cell analysis (RNA-Seq). Data obtained from single cell analysis will be compared across different time frame (for example, before versus after specific treatment) and across different disease phenotypes.	Up to 10 years

Human Microbiome and Clinical Application

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## The Hospital-Based Human Microbiome R&D Project



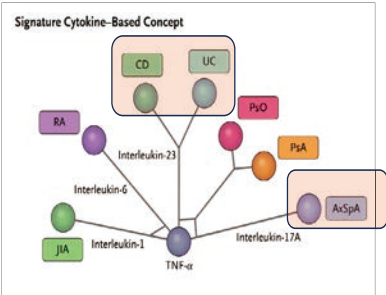
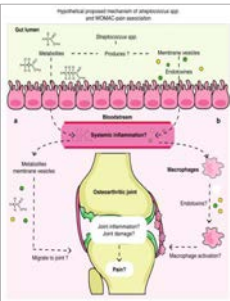
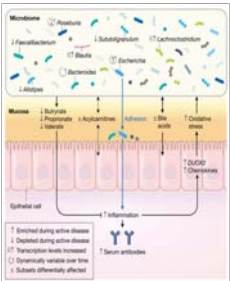
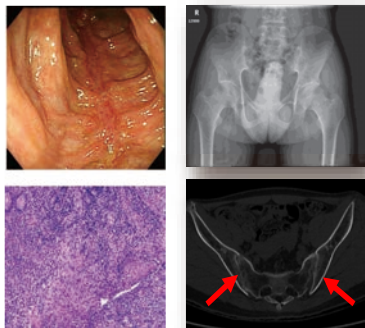
Human Microbiome and Clinical Application

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# The oral-gut microbiome axis in a Korean cohort with inflammatory bowel disease and ankylosing spondylitis (INTEGRATE): a prospective and observational study

All diseases begin in the gut.  
Immune-mediated inflammatory disorders (IMIDs)

19 y-o male with Crohn's disease and Ankylosing spondylitis



Nature. 2019 May;569(7758):641-648.

Human Microbiome and Clinical Application

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# The INTEGRATE study protocol

Table 2. Screening inclusion criteria for selecting participants<sup>1,2</sup>

Participants in the study must meet all of the following criteria <sup>1,2</sup>	
Patients with IBD <sup>1</sup>	
1. Males and females aged 13-85 years <sup>1</sup>	
2. Koreans diagnosed with CD according to the Crohn's disease diagnostic guideline (Korean J Gastroenterol 2009;53:161-176) <sup>1</sup>	
3. Koreans diagnosed with UC according to the ulcerative colitis diagnostic guideline (Korean J Gastroenterol 2009;53:145-160) <sup>1</sup>	
4. Participants who, after hearing and fully understanding the detailed explanation of this clinical trial, voluntarily decide to participate and agree in writing to follow the precautions (in the case of minors, written consent is obtained from a legal representative) <sup>1,2</sup>	
Patients with AS <sup>1</sup>	
1. Males and females aged 13-70 years <sup>1</sup>	
2. Koreans diagnosed with AS according to the American College of Rheumatology's Modified New York criteria and the 2010 spondyloarthritis diagnosis <sup>1</sup>	
3. Koreans aged 13 years or older and diagnosed with juvenile AS (diagnosis: juvenile ankylosing spondylitis, enthesitis-related arthritis) <sup>1</sup>	
4. Participants who, after hearing and fully understanding the detailed explanation of this clinical trial, voluntarily decide to participate and agree in writing to follow the precautions (in the case of minors, written consent is obtained from a legal representative) <sup>1,2</sup>	
Healthy population <sup>1</sup>	
1. Males and females aged 13-85 years <sup>1</sup>	
2. Participants judged by the researcher to be able to participate in the study based on screening tests (physical examination, clinical laboratory tests, questionnaires, and drug use history) <sup>1</sup>	
3. Participants who, after hearing and fully understanding the detailed explanation of this clinical trial, voluntarily decide to participate and agree in writing to follow the precautions (in the case of minors, written consent is obtained from a legal representative) <sup>1,2</sup>	
Families of patients with IBD <sup>1</sup>	
1. Males and females aged 13-85 years <sup>1</sup>	
2. Koreans who are first-degree relatives of the patient with IBD and share a living space with the patient <sup>1</sup>	
3. Participants who have not been diagnosed with IBD <sup>1</sup>	
4. Participants who, after hearing and fully understanding the detailed explanation of this clinical trial, voluntarily decide to participate and agree in writing to follow the precautions (in the case of minors, written consent is obtained from a legal representative) <sup>1,2</sup>	
Families of patients with AS <sup>1</sup>	
1. Males and females aged 13-70 years <sup>1</sup>	
2. Koreans who are first-degree relatives of the patient with AS and share a living space with the patient <sup>1</sup>	
3. Participants who have not been diagnosed with AS <sup>1</sup>	
4. Participants who, after hearing and fully understanding the detailed explanation of this clinical trial, voluntarily decide to participate and agree in writing to follow the precautions (in the case of minors, written consent is obtained from a legal representative) <sup>1,2</sup>	

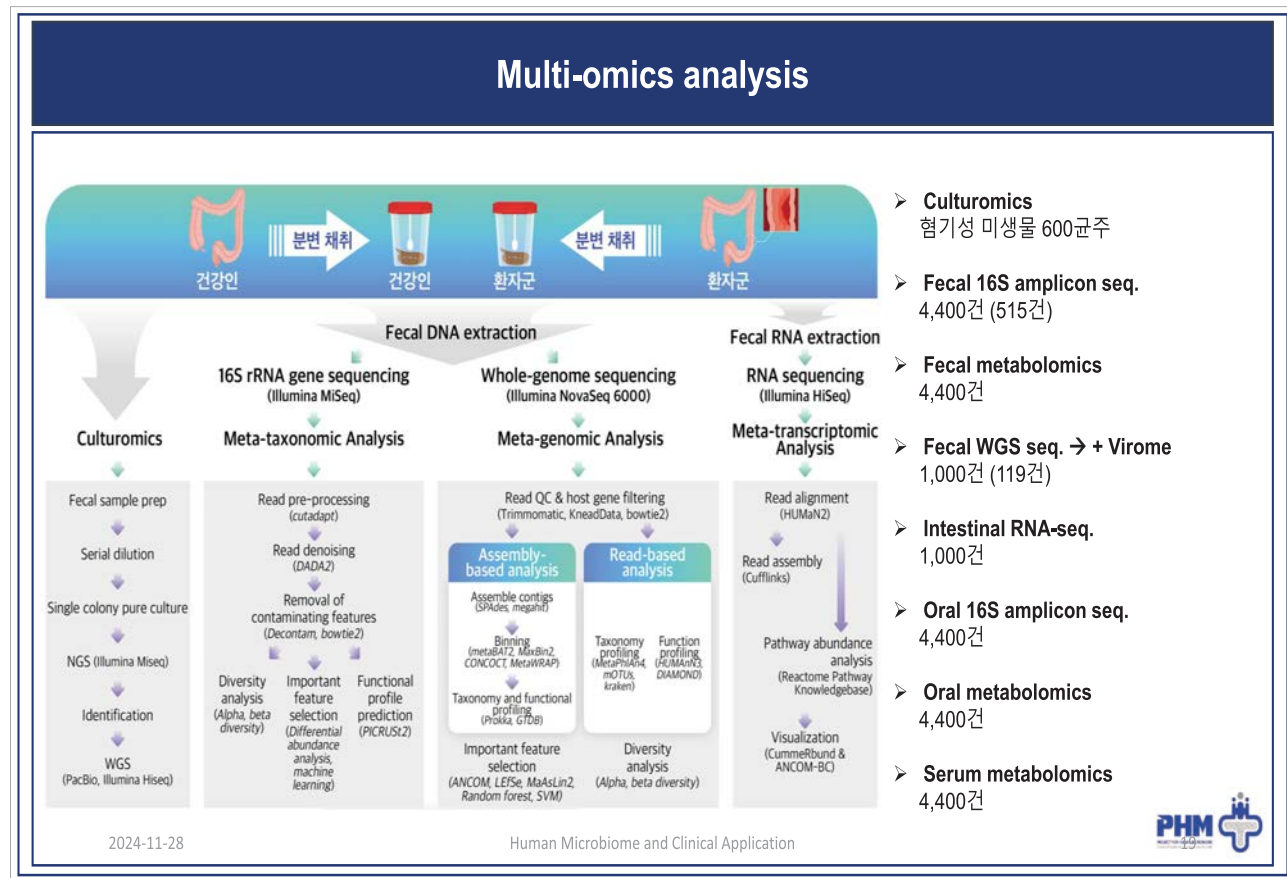
Table 4. Outline of study procedures<sup>1,2</sup>

Visit <sup>1</sup>	Patient with IBD-specific collection information <sup>1,2</sup>				Family of patient <sup>1,2</sup>
	Screening <sup>1</sup>	Visit 1 <sup>1</sup>	Visit 2 <sup>1</sup>	Visit 3 <sup>1</sup>	
Timepoint (Newly diagnosed) <sup>1</sup>	Day -30 to Day 0 <sup>1</sup> (Pre-enrollment) <sup>1</sup>	Day 0 <sup>1</sup> (Baseline) <sup>1</sup>	1 Year <sup>1</sup> (After diagnosis) <sup>1</sup>	3 Years <sup>1</sup> (After diagnosis) <sup>1</sup>	Day 0 <sup>1</sup> (Baseline) <sup>1</sup>
Timepoint (Drug response) <sup>1</sup>	Day -30 to Day 0 <sup>1</sup> (Pre-enrollment) <sup>1</sup>	Day 0 <sup>1</sup> (Before treatment) <sup>1</sup>	3 Month <sup>1</sup> (After treatment) <sup>1</sup>	1 Year <sup>1</sup> (After treatment) <sup>1</sup>	Day 0 <sup>1</sup> (Baseline) <sup>1</sup>
Obtain written consent <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>
Confirm consent <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>
Confirm selection criteria and assign screening number <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>
Demographic information <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>
Body measurements <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>
Vital signs <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>
Comorbidities and medical history <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>
Hospitalization history <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>
Surgical history <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>
Pedigree and familial history <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>
Concomitant medications and therapies <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>
Diagnosis <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>
Lifestyle habits survey <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>
Food intake survey <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>
Dietary supplement intake survey <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>
Clinical laboratory tests <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>
Stool specimen collection <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>
Stool specimen collection information <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>
Tissue biopsy specimen collection <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ (if necessary) <sup>1,2</sup>
Saliva specimen collection <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>
Blood specimen collection <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>	Ⓢ <sup>1</sup>
Patient with AS-specific collection information <sup>1,2</sup>					
Visit <sup>1</sup>	Screening <sup>1</sup>	Visit 1 <sup>1</sup>	Visit 2 <sup>1</sup>	Visit 3 <sup>1</sup>	Family of patient <sup>1,2</sup>

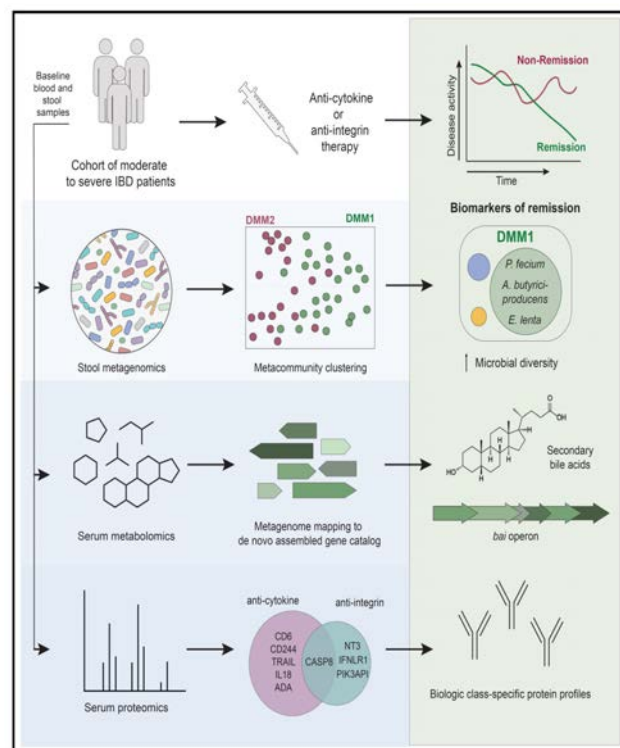
Developing an INTEGRated Gut Microbiome Data Analysis Platform and Conducting Comparative Clinical Studies in Korean Patients with Intractable Immune-Mediated Inflammatory Disorders and Healthy Volunteers (INTEGRATE)

Human Microbiome and Clinical Application

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Multi-omics analysis requires large amounts of high-quality samples.



## Nationwide Multicenter Prospective IBD Cohort in Korea

- 14 Academic Teaching Hospitals (As of Nov. 2024)

- 1) 경희대학교병원
- 2) 강동경희대학교병원
- 3) 강북삼성병원
- 4) 중앙대병원 (서울)
- 5) 한양대구리병원
- 6) 일산백병원
- 7) 중앙대광명병원
- 8) 단국대병원
- 9) 순천향대천안병원
- 10) 대전가톨릭대병원
- 11) 전남대병원
- 12) 원광대병원
- 13) 경북대병원
- 14) 해운대백병원
- 15) 계명대병원 - IRB 심의 중
- 16) 조선대병원 - IRB 심의 중

Human Microbiome and Clinical Application

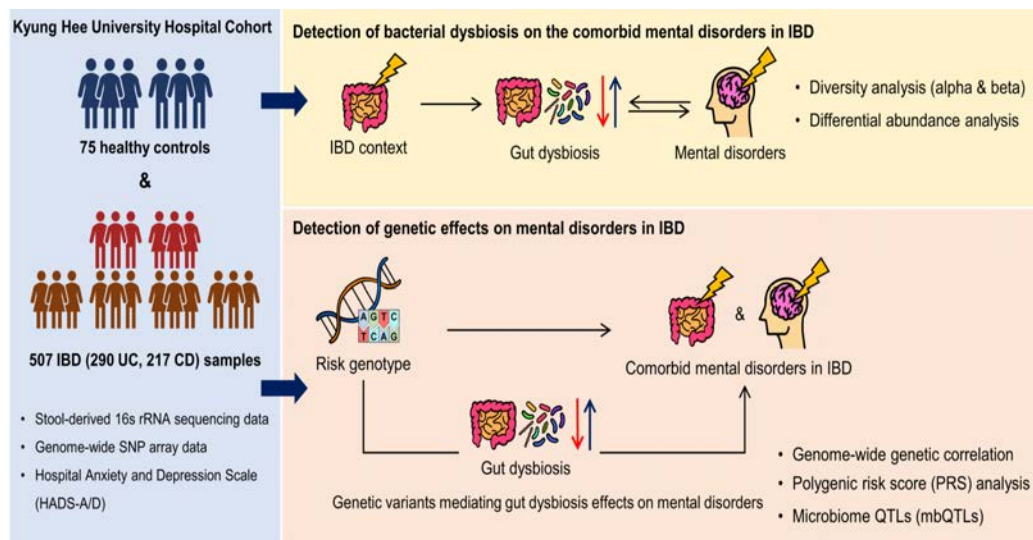


## Research in Progress: Current Investigations



## Gut microbial and human genetic signatures of inflammatory bowel disease increase risk of comorbid mental disorders.

**#Host-Gut Microbiome Interaction, #Brain-Gut Axis in IBD**



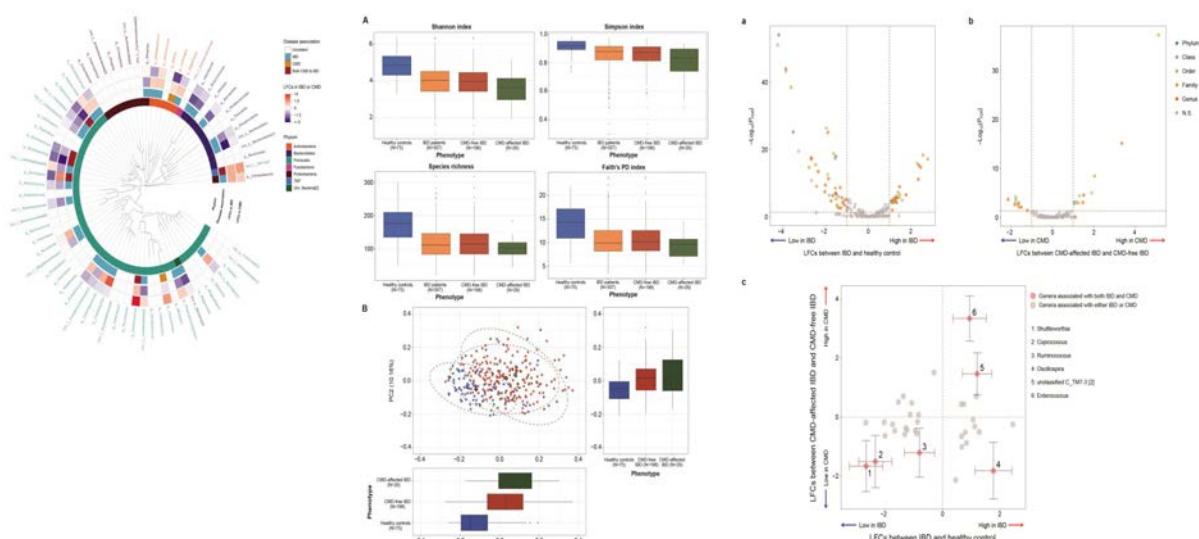
Human Microbiome and Clinical Application

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Lee J,...., Lee CK. *npj Genomic Medicine* 2024

**Gut microbial and human genetic signatures of inflammatory bowel disease increase risk of comorbid mental disorders.**

**#Host-Gut Microbiome Interaction, #Brain-Gut Axis in IBD**



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Lee J, et al., *npj Genomic Medicine* 2024

# Gut microbial and human genetic signatures of inflammatory bowel disease increase risk of comorbid mental disorders.

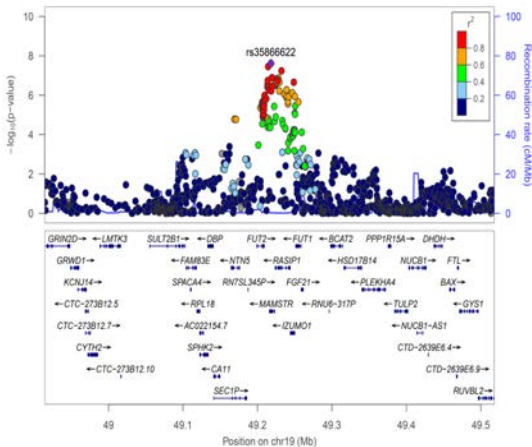
#Host-Gut Microbiome Interaction, #Brain-Gut Axis in IBD

Genome-wide correlation of genetic effects between IBD and mental disorders, estimated from previous large European GWASs

	Depression	Anxiety	CD	UC
Depression	-	-	-	-
Anxiety	0.64 (0.02) *	-	-	-
CD	0.11 (0.03) *	0.07 (0.03) *	-	-
UC	0.04 (0.04)	0.13 (0.03) *	0.63 (0.03) *	-
IBD	0.09 (0.03) *	0.11 (0.03) *	0.92 (0.01) *	0.90 (0.01) *

The table provides genetic correlation coefficients ( $r_g$ ) with the standard errors at parentheses.

\*, significant (FDR < 0.05).



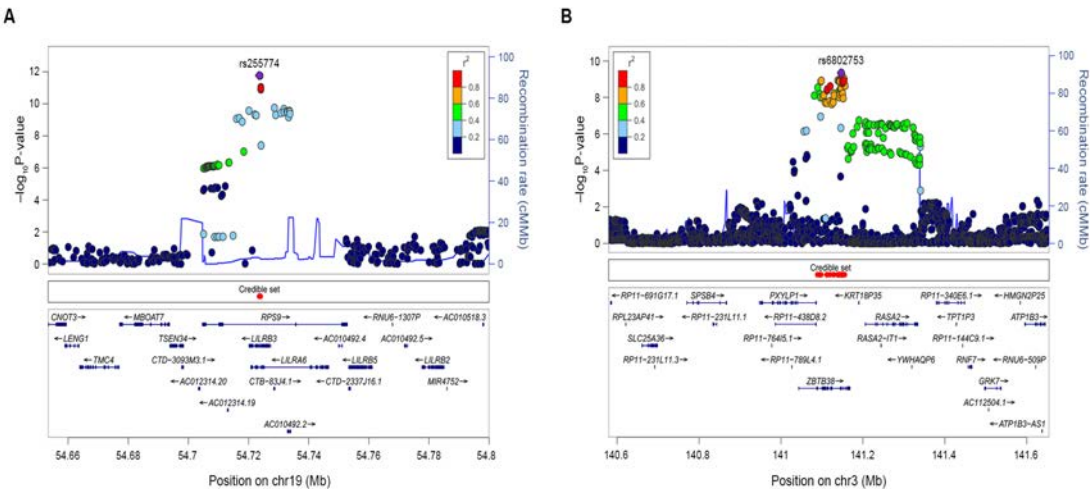
An IBD-associated mbQTL for the genus *Ruminococcus* (*torques* group)

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Lee J., ..., Lee CK. npj Genomic Medicine 2024

# Regional association plots of the novel susceptibility loci for Korean patients with Crohn's disease



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Kim K., ..., Lee CK. J Crohns Colitis. 2024

**Spatial Transcriptomics of Pre-treatment Biopsies Revealing Chronic Crypt Damage and Upregulated Inflammatory Process, Reflecting Histological Severity, as Predictors of Primary Responsiveness to TNF- Inhibitors in Bio-naïve Ulcerative Colitis Patients**

In draft

**Microbial inactivation of digestive proteases protects epithelial barrier integrity and colitis**



## **Shared and Distinct Features of the Oral-Gut Microbiome Axis in Immune-mediated Inflammatory Diseases: Initial Analysis from the INTEGRATE Cohort Study**

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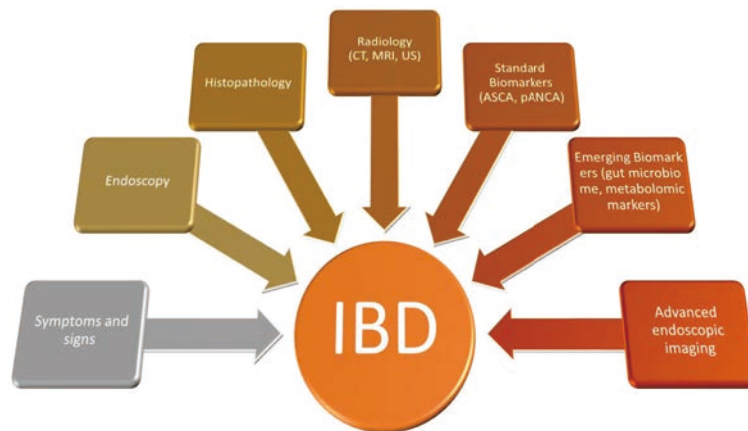
## **Lesson Learned and Future Tasks Following the Interim Analysis**

- High-resolution full-length 16S rRNA sequencing
- Clinical data standardization and quality control (missing data and human errors )
- Securing matched healthy controls
- Analysis of confounding factors after data supplementation
  
- Assessment of associations between clinical factors and data
- Development of experimental plans based on data-driven hypotheses
- Maximizing data size
- Integrating oral microbiome and metabolomics analyses

Human Microbiome and Clinical  
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## There is no single gold standard for diagnosis in IBD.



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## What about other markers?

Antibody	Antigen	Non-IBD (%) <sup>1</sup>	CD (%) <sup>1</sup>	UC (%) <sup>1</sup>	Korea <sup>2,3</sup>
ASCA	<i>Saccharomyces cerevisiae</i> cell wall	5–10	29–69	5–15	38.3-49.4% in CD
pANCA	Unclear	<5–48	2–28	45–82	44.2% in UC
Omp-C	<i>Escherichia coli</i> OMP	<5	24–55	5–28	unavailable
I2	<i>Pseudomonas</i> protein	15	36–60	42	unavailable
CBir-1	Bacterial flagellin	8–15	50–57	6–16	unavailable

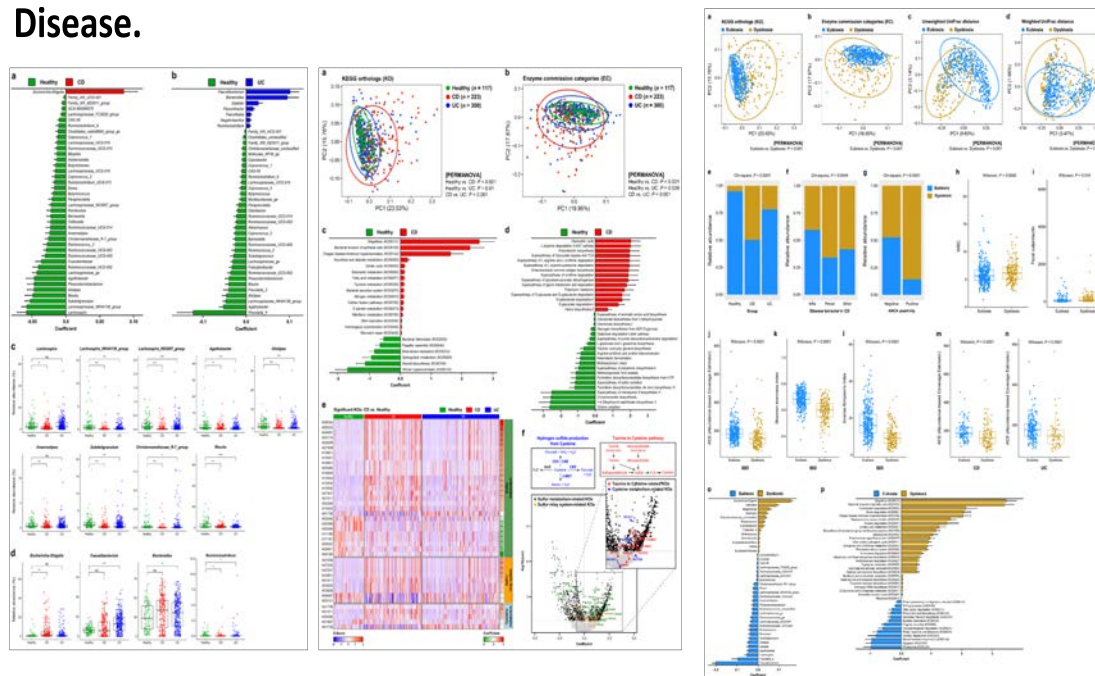
<sup>1</sup>Curr Opin Gastroenterol. 2014;30:359–64

<sup>2</sup>Dis Colon Rectum 2002;45:1062-1069.

<sup>3</sup>Dig Liver Dis 2007;39:610-616.

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## Dysbiotic signatures in Korean Patients with Inflammatory Bowel Disease.

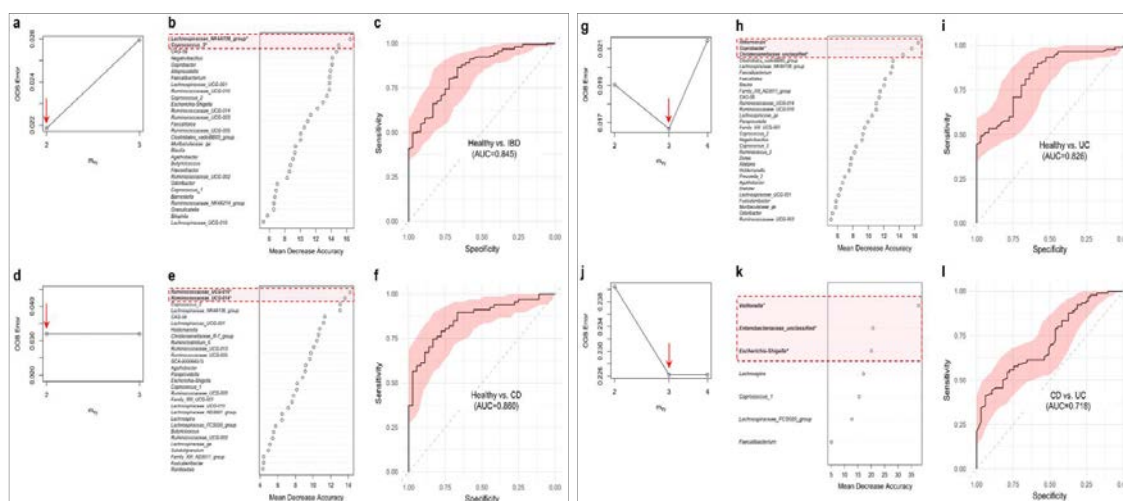


Kim HS, Kim BK, Park YK, ... JW Bae, Lee CK. Scientific Reports, 2024.

Human Microbiome and Clinical Application

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## Gut microbial markers precisely predict the subtypes of inflammatory bowel disease.



Kim HS, Kim BK, Park YK, ... JW Bae, Lee CK. Scientific Reports, 2024.

Human Microbiome and Clinical Application

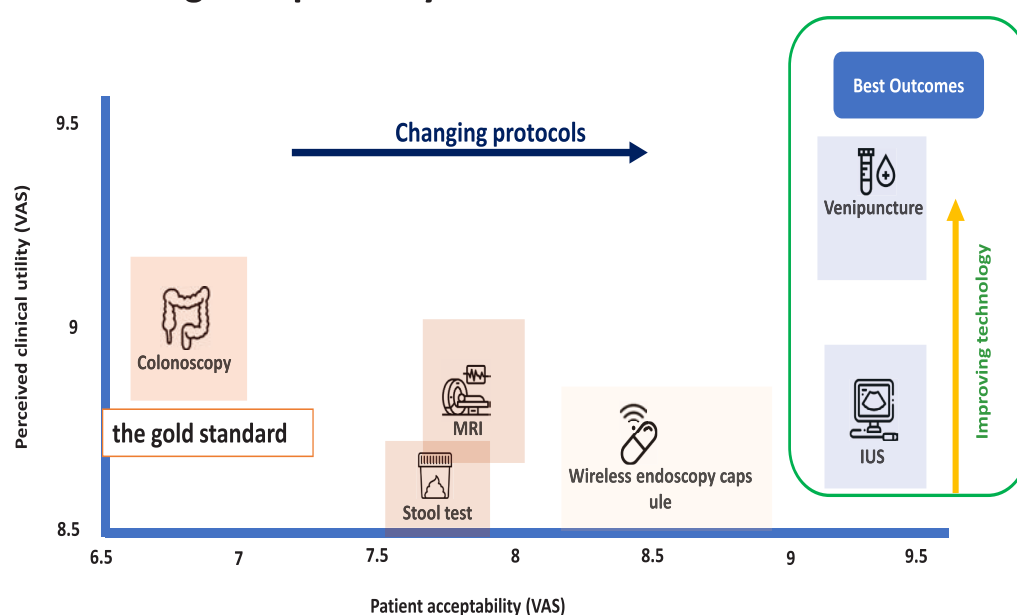
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## Multi-analytical Approaches Reveal Robust Gut Microbial Biomarkers for IBD Diagnosis: A Large-scale Cohort Study

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If we can't skip frequent tests to meet treatment goals, then considering acceptability will lead to better outcomes.



CD: Crohn's disease US: Intestinal ultrasound; MRI: Magnetic Resonance Imaging; VAS: visual analogue scale  
1. Buisson, A. (2017). *Inflammatory bowel diseases*, 23(8), 1425–1433, Modified from IUS primer module.

## Lessons Learned: Korean IBD Experiences

- **Multi-center cohorts are essential**

- (1) to achieve larger sample sizes for robust statistical analysis and
- (2) to encompass diverse patient populations, ensuring comprehensive and reliable IBD research outcomes.

- **Challenges**

- (1) Coordinating multiple centers can be a big challenge.
- (2) Ethical and regulatory considerations can be complex.
- (3) The hospital-based research environment for IBD studies is facing rapid and significant challenges.

Thank you.

[Changkyun.lee@khu.ac.kr](mailto:Changkyun.lee@khu.ac.kr)



# 02

## Development of microbiome indicators: selection of indicators from microbiome and meta-data through artificial intelligence



최민혁  
(강남세브란스병원)







# 1부 연자



- ✓ 소속: 연세의대 강남세브란스병원
- ✓ 직위/직책: 임상조교수
- ✓ 성명: 최민혁

## 🔍 학력사항

2013	연세원주의과대학 의학학사
2023	연세대학교 의과대학 의학박사 (석박통합)

## 🔍 경력사항

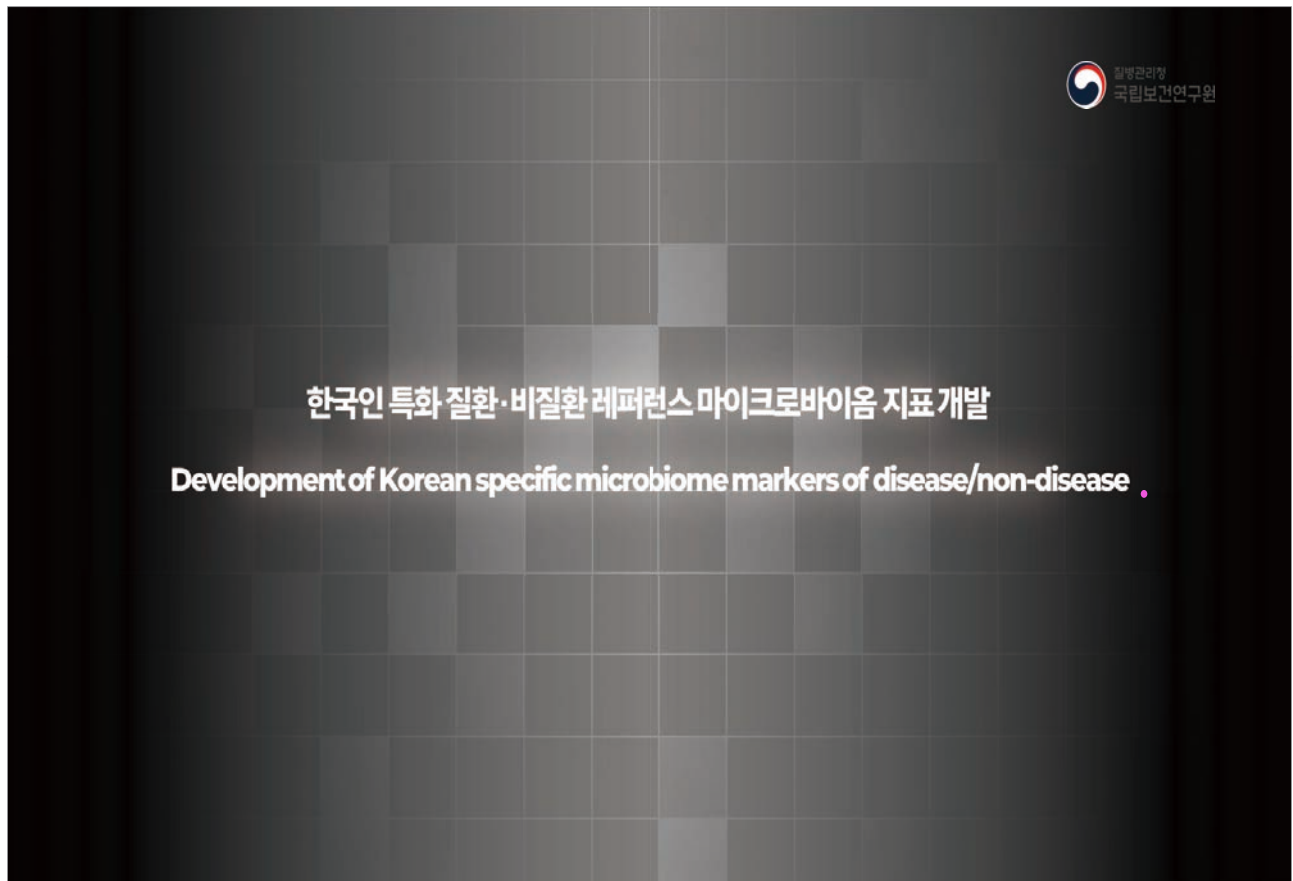
2013 ~ 2018	연세의대 세브란스병원 전공의
2021 ~ 2022	강남세브란스병원 임상강사
2022 ~ 2023	강남세브란스병원 진료교수
2024 ~ 현재	연세의대 진단검사의학교실 임상조교수

# 질환/비질환 레퍼런스 마이크로바이옴 지표 개발 및 진단 적용

병원에서 사용가능한 마이크로바이옴 지표 개발을 위한 코호트 및 분석 파이프라인 구축. Metagenomic assembled genome 기반의 파이프라인을 인공지능 분석을 통한 해석 시도.

2. Development of microbiome indicators: selection of indicators from microbiome and meta-data through artificial intelligence







**병원기반인간마이크로바이옴연구개발사업 심포지엄**  
Human Microbiome and Clinical Application

**신대림시에서이 무대**

**새로운 가치 창출**

**Conventional lab test**  
Serum creatinine

**Additional information**  
Estimated GFR

**Novel insights**  
AI models

Method	Formula
Cockcroft-Gault	Creatinine Clearance = $\frac{1.73 \text{ m}^2}{\text{m}^2} \times \left( \frac{140 - \text{age}}{72} \right) \times \left( \frac{\text{mg/dL}}{1.73} \right)$ (male)
MDRD-4 (simplified)	Estimated Glomerular Filtration Rate (mL/min/1.73 m <sup>2</sup> ) = $175 (\text{Serum Creatinine in mg/dL} \times 0.011312)^{-1.154} \times (\text{age in years})^{-0.203} \times (0.742 \text{ if female}) \times (1.212 \text{ if African American/black})$
CKD-EPI (2009)	Estimated GFR = $141 \times \text{min}(S_{Cr}/K, 1)^{\alpha} \times \text{max}(S_{Cr}/K, 1)^{-1.209} \times 0.990^{Age} \times 1.018 \text{ (if female)} \times 1.159 \text{ (if Black)}$ $S_{Cr}$ (standardized serum creatinine) = mg/dL, $K = 0.7$ (females) or $0.9$ (males), $\alpha = -1.320$ (female) or $-0.411$ (male), Min = indicates the minimum of $S_{Cr}/K$ or $1$ , max = indicates the maximum of $S_{Cr}/K$ or $1$ , Age = Years

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**인공지능분야기초연구**  
요로감염예측모델

Journal of Infection 2022

**Impact of urinary tract infection-causative microorganisms on progressing to bloodstream infection : a propensity score-matched analysis**

Adult patients with urinary tract infection (UTI) N = 84406

Patients with urinary tract-related bloodstream infection N = 5137

Propensity-score matching & Multivariable analysis

**UTI-causing microorganisms**

**요로감염 원인균의 변화**

*Escherichia coli* >>>  
*Enterococcus*, *Candida* species

**환자 고령화가 가장 큰 원인**

Journal of Infection 2022; 105: 1057-1065

Min Hyuk Choi<sup>1,2</sup>, Dohyun Kim<sup>1,2</sup>, Yongsang Park<sup>1</sup>, Seok Hoon Jeong<sup>1,2,3</sup>

**인공지능분야기초연구**  
요로감염예측모델

Journal of Clinical Microbiology 2024

**Predictive performance of urinalysis (UA) for urinary tract infections (UTIs) according to causative microorganism: an integrated analysis with artificial intelligence (AI)**

University hospitals & Commercial lab

A total of 360,376 suspected UTI cases were enrolled

Statistical model & Artificial intelligence model

**기준 장비 결과 활용**

**자동화요검사, 기본환자정보(환자나이,성별)**

**Performance of UA tests in predicting UTIs by causative microorganisms**

**Performance metrics of UA test combinations and AI models in predicting UTIs**

**Conclusions**

- As the prevalence of non-*Escherichia coli*-UTIs increases, the performance of UA in predicting UTIs could be compromised.
- The integration of UA and AI technologies showed a potential to improve predictive performance for UTIs.

인공지능분야기초연구

요로감염예측모델

강남세브란스병원

Severance Hospital

UTI-causing microorganisms

기타 검사 결과 활용

자동화요검사, 기본 환자정보, VS, Labs

Journal of Infection and Public Health 2024

Development and validation of artificial intelligence models to predict urinary tract infections and secondary bloodstream infections in adult patients

Adult patients with Urinary tract infection (UTI)  
N = 68,525

Patients with secondary bloodstream infection (BSI)  
N = 4507

Artificial intelligence (AI)  
analyses

The performance of AI-based UTI and secondary BSI prediction models

Critical variables with SHAP values for predicting UTI

Conclusion:

We found that AI models can improve the early prediction of urine culture positivity and secondary BSI by combining automated urinalysis with other clinical information

출원번호통지서

발행일자 2023.11.16  
 출제자명 김수영(서울 강남세브란스병원) (출제번호202311)  
 출제일자 2023.11.16 (출제번호202311)  
 출제번호 2023.11.16 (출제번호202311)  
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 출제일자 2023.11.16 (출제번호202311)  
 출제일자 2023.11.16 (출제번호202311)

출원번호통지서

Flask 기반 Web application

Prediction models for UTI and UT-BSI

Urinalysis results:

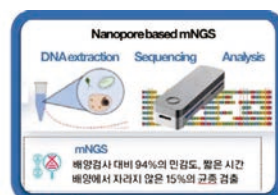
Routine laboratory findings:

Demographic data:

UTI Positive (33.24%) / Urinary tract-related BSI Positive (52.59%)

**The need to adopt a new diagnostic strategy.**

New insights



#### Metagenomic NGS

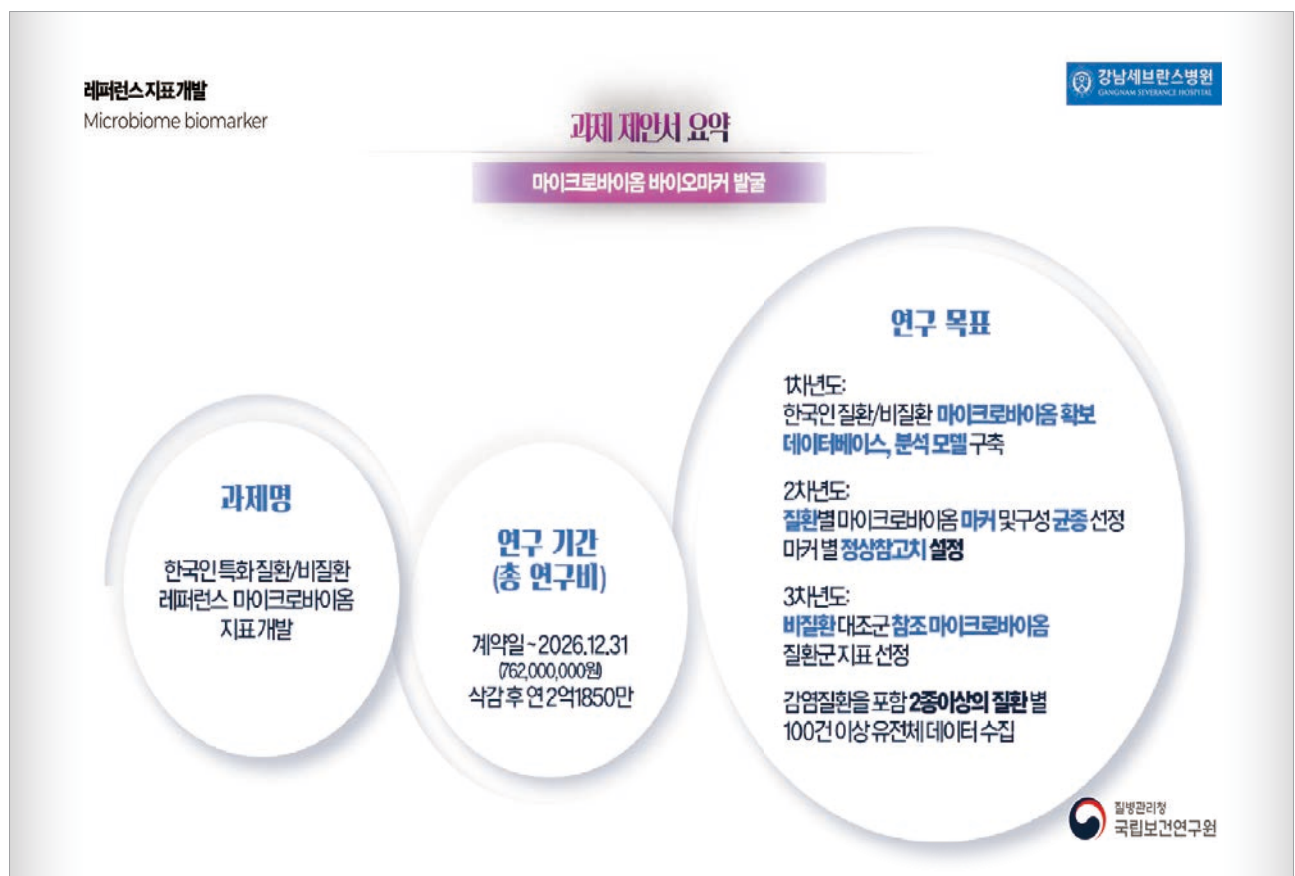
Shorten TAT and increase sensitivity for strains and resistances that are difficult to detect in traditional culture tests



#### Human microbiome

Hundreds of microbial OTUs, a variety of clinical information, microbiological factors, patient factors, and treatment factors all need to be considered.



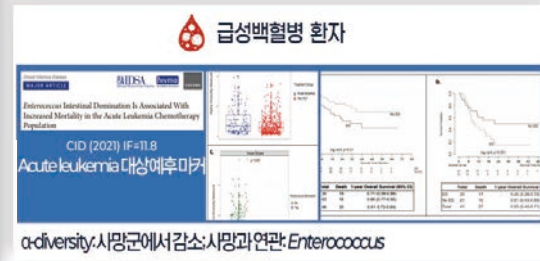
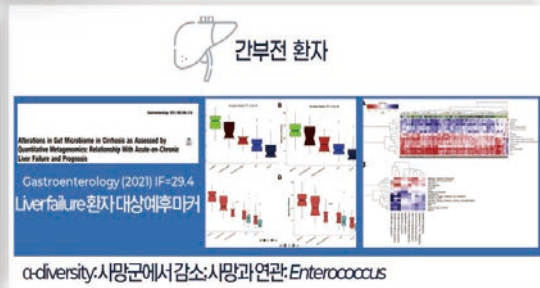
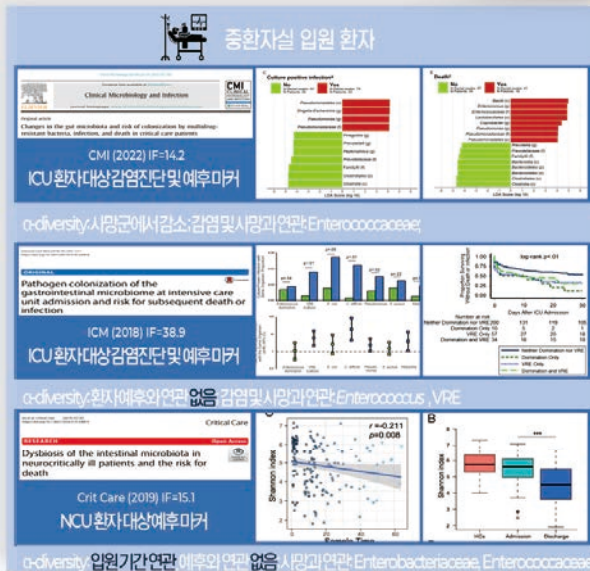


참고문헌

Previous studies

이전 연구 동향

마이크로바이옴 기반 환자 사망 예측 마커



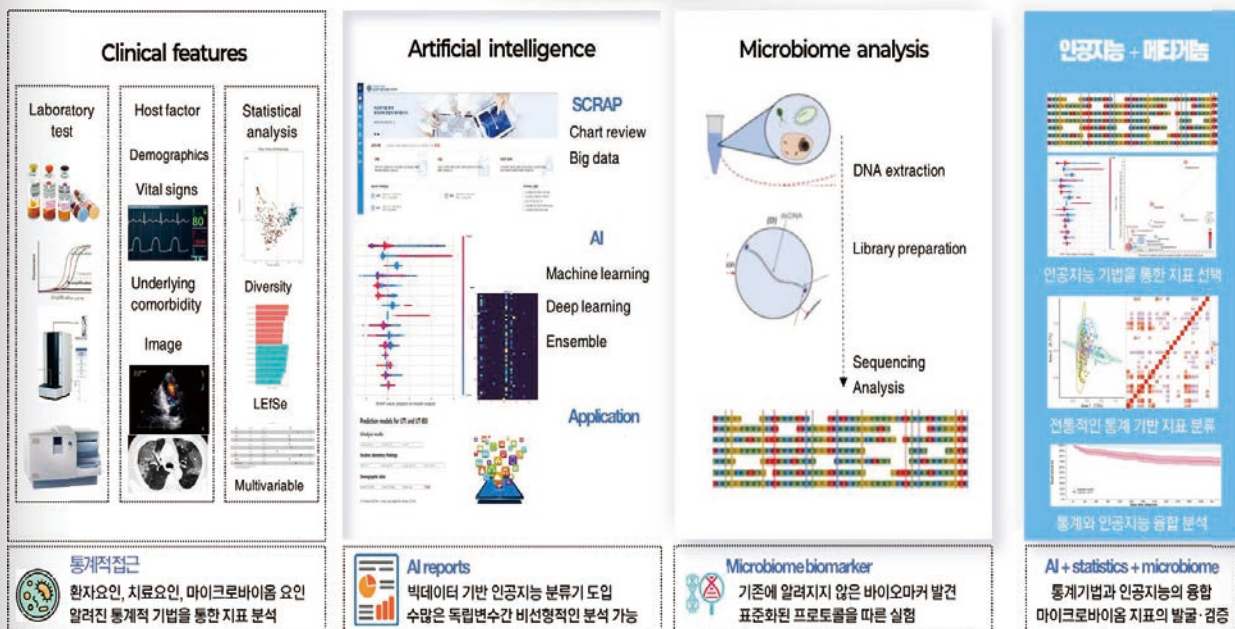
질병관리청  
국립보건연구원

융합연구

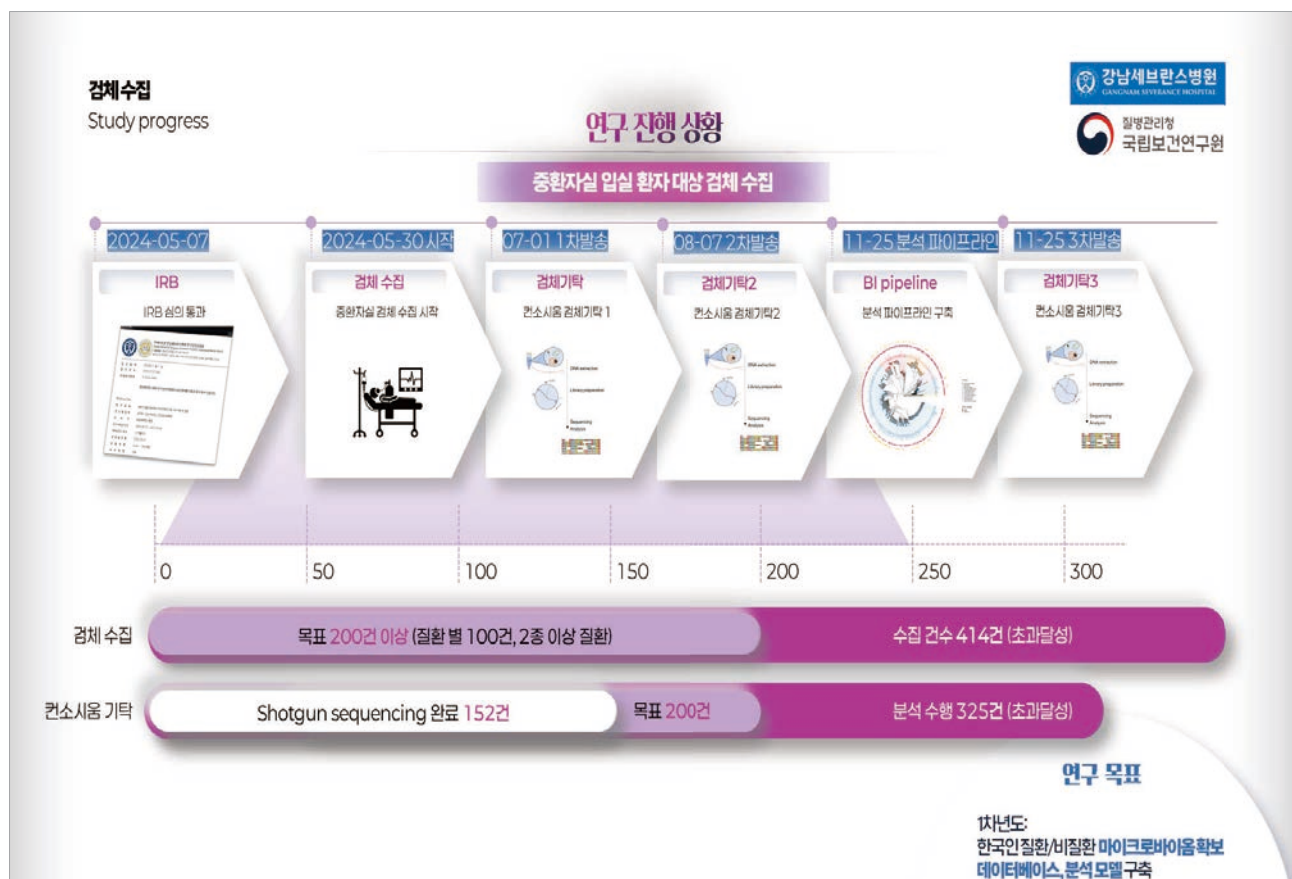
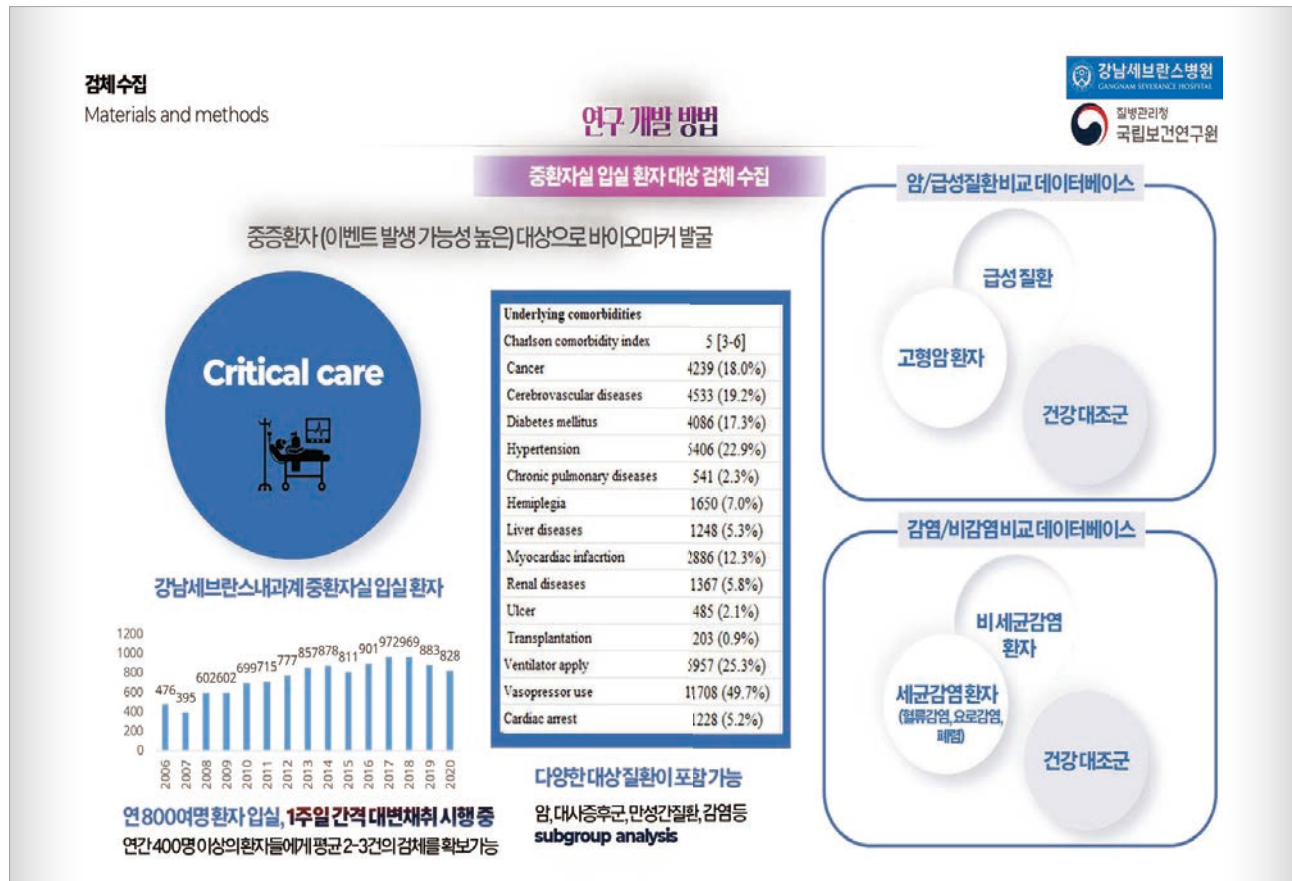
Objectives and contents

연구개발 목표 및 내용

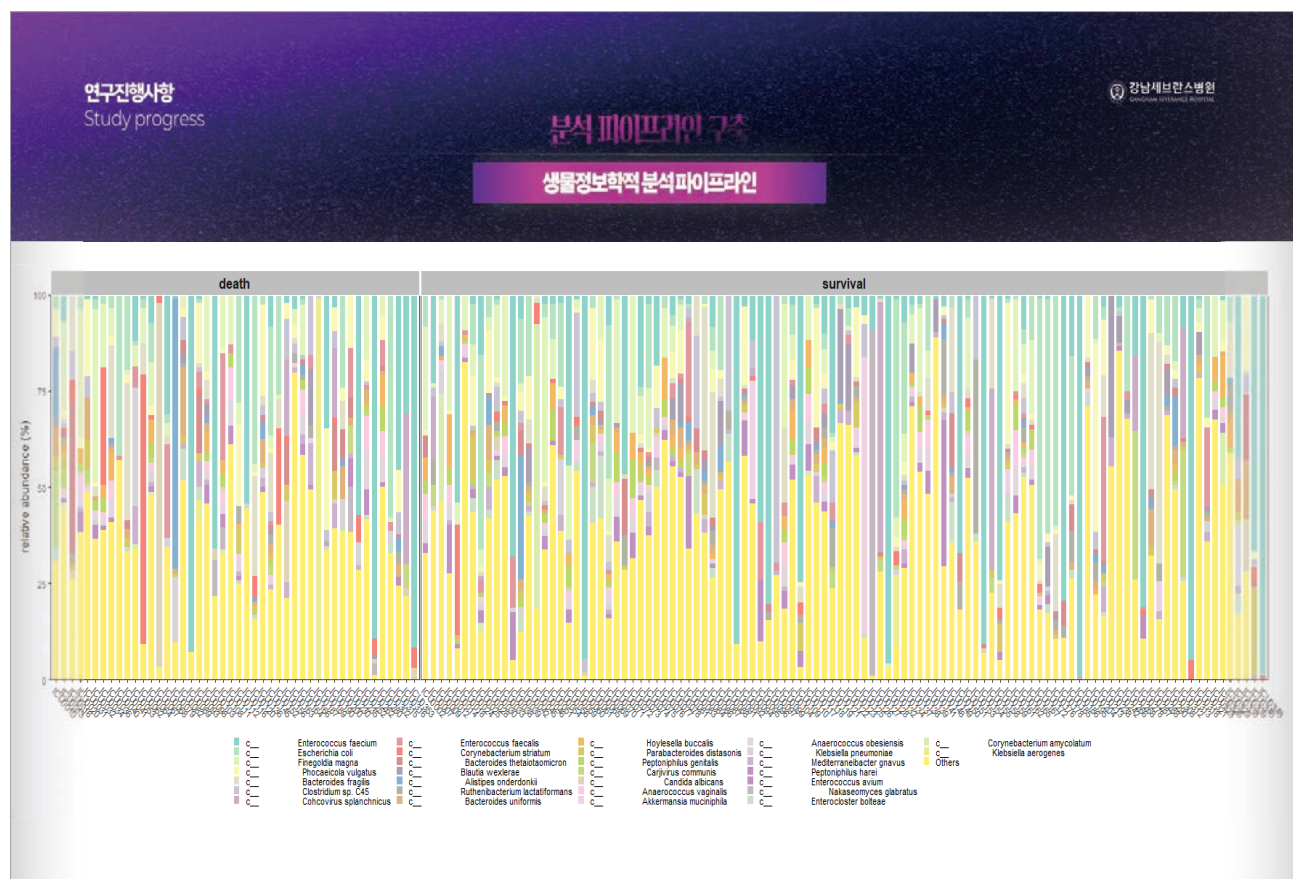
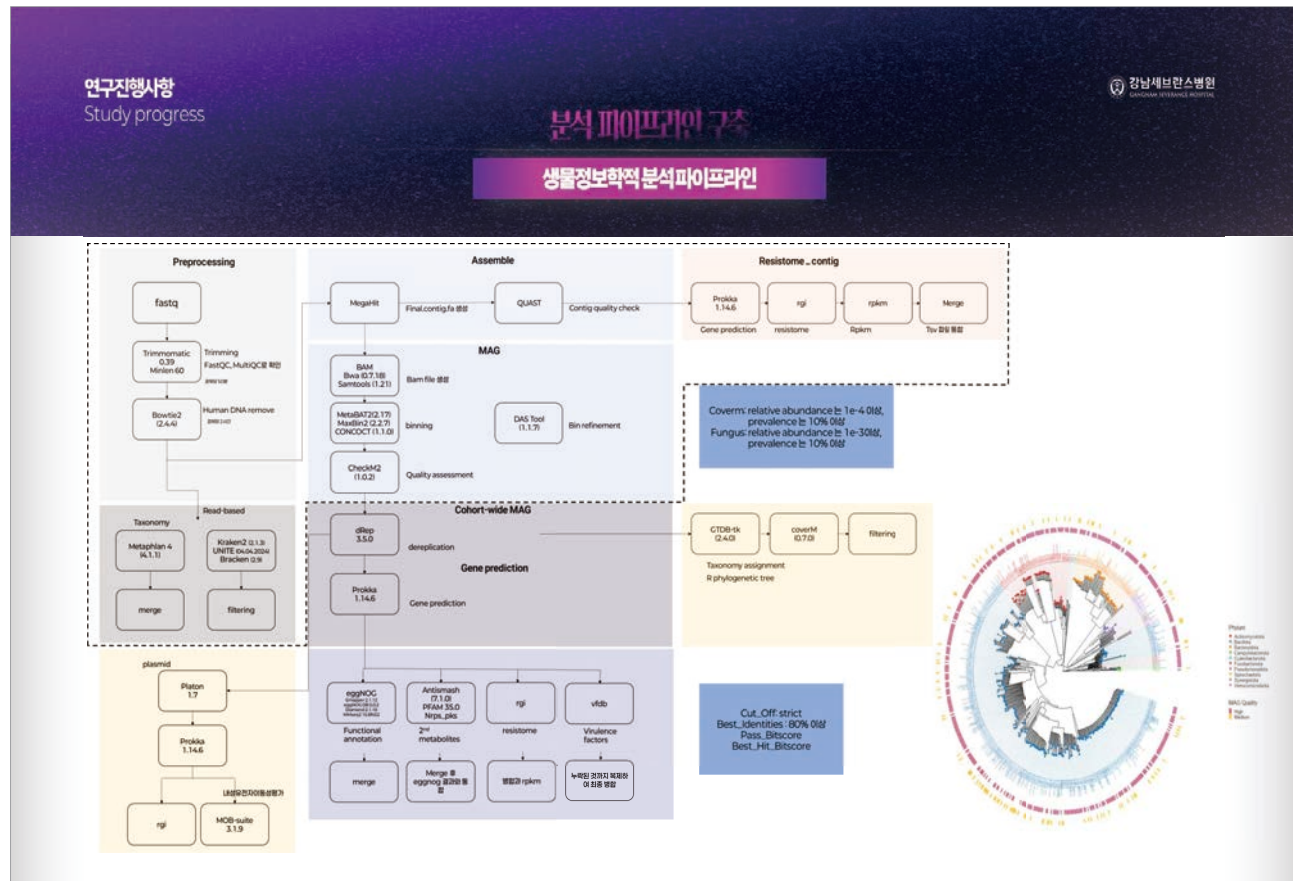
기존 바이오마커, 마이크로바이옴, AI



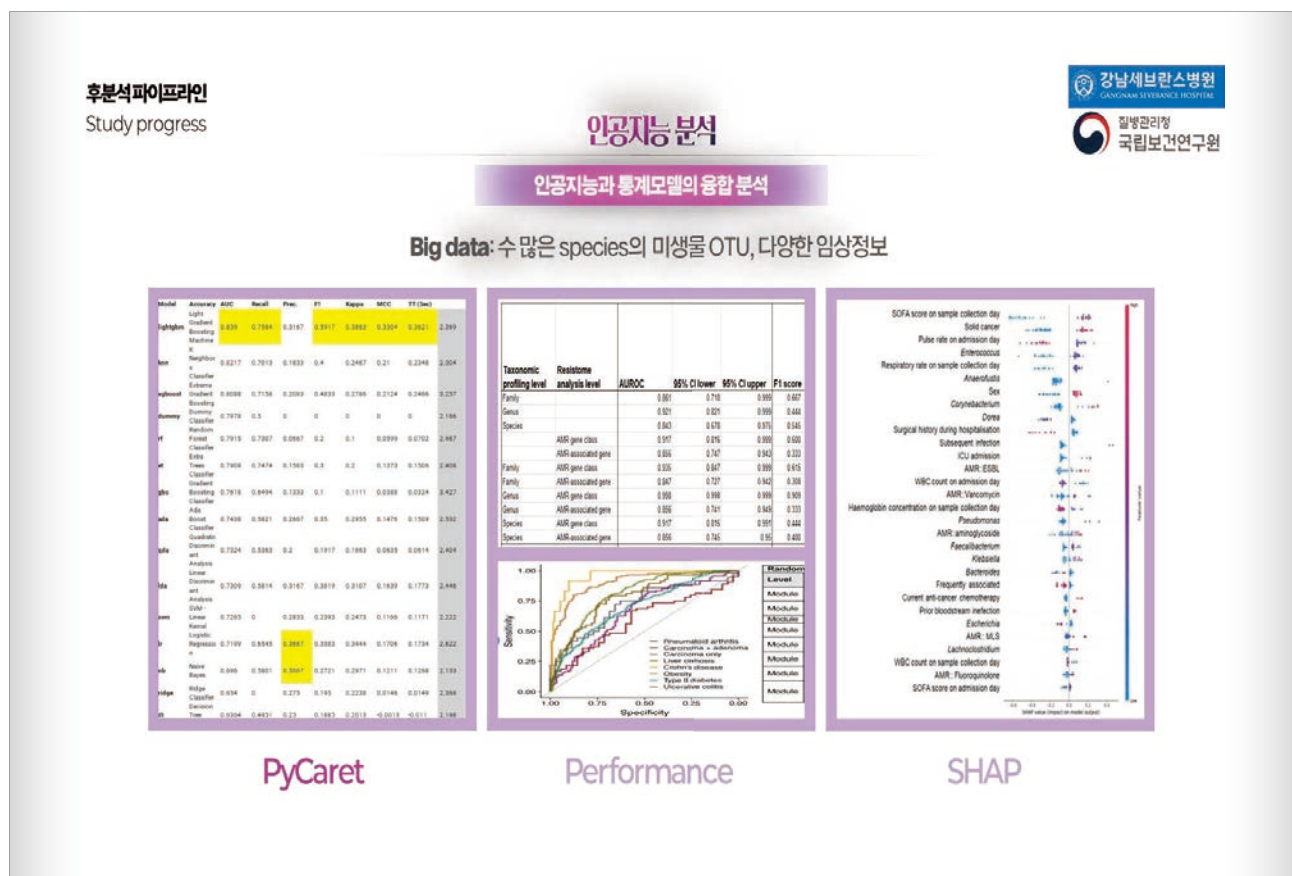
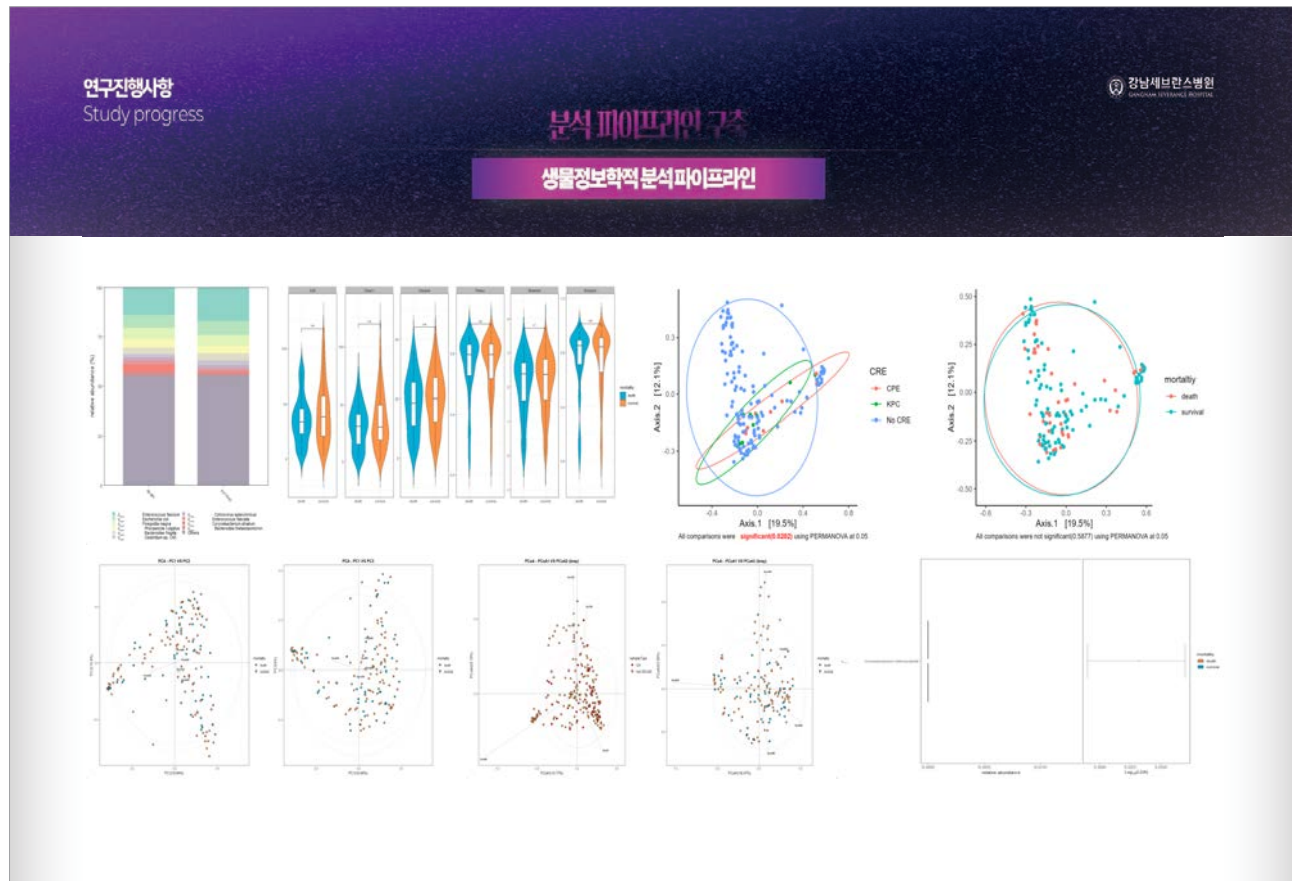
2. Development of microbiome indicators: selection of indicators from microbiome and meta-data through artificial intelligence







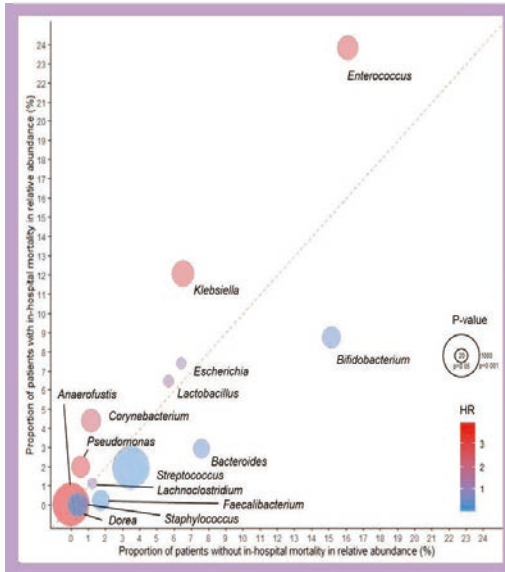
## 2. Development of microbiome indicators: selection of indicators from microbiome and meta-data through artificial intelligence



후분석파이프라인  
Study progress

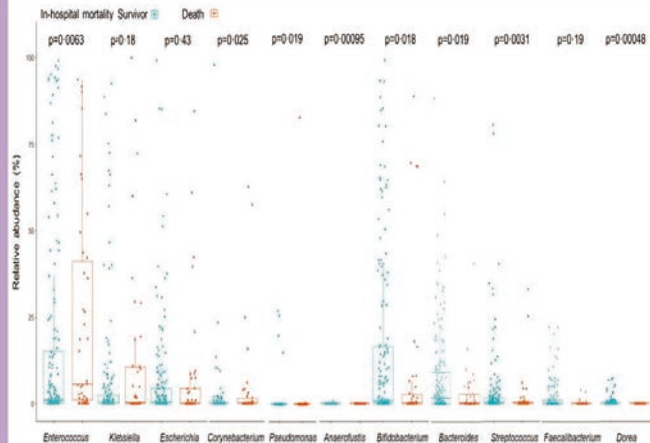
인공지능 분석

인공지능과 통계모델의 융합 분석



환자 후속 감염 및 병원내 사망의 마커 마이크로바이옴

좋은 예후 인자, 나쁜 예후 인자 발굴



후분석파이프라인  
Study progress

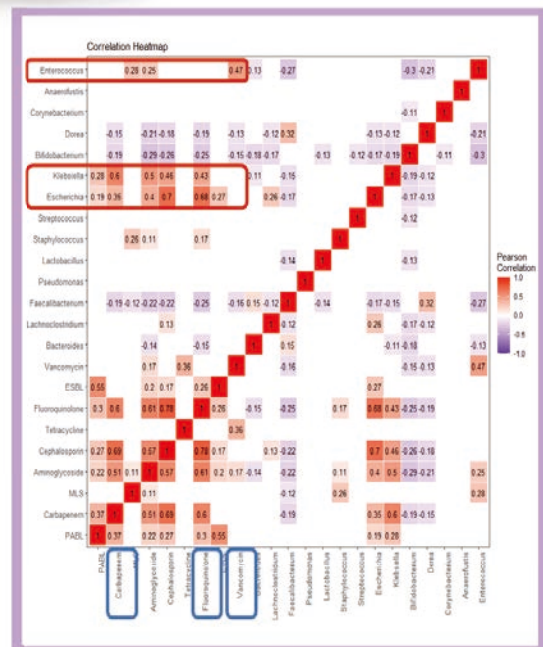
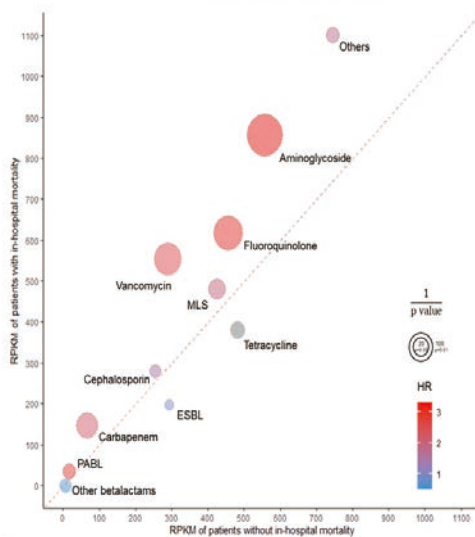
인공지능 분석

인공지능과 통계모델의 융합 분석



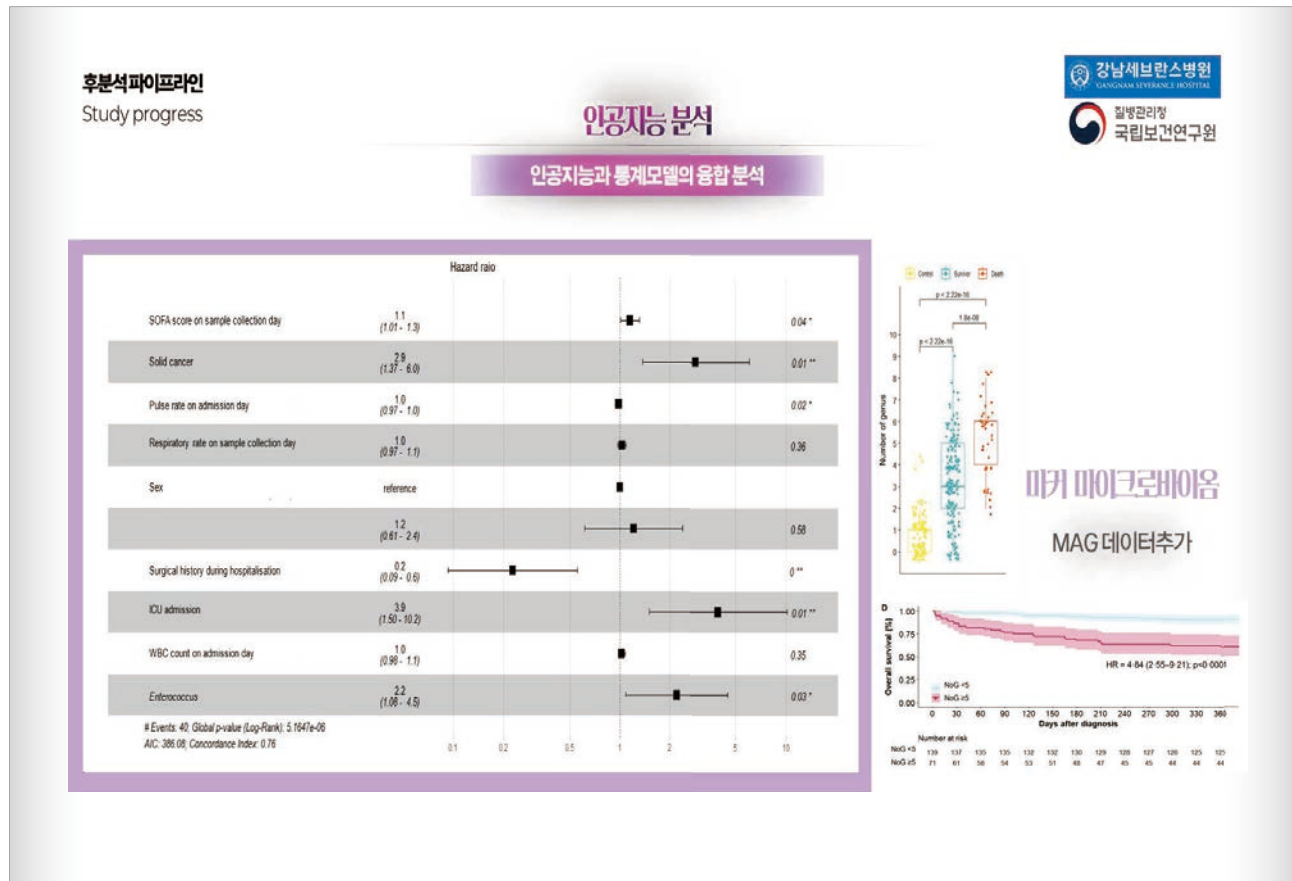
환자 후속 감염 및 병원내 사망의 마커 마이크로바이옴

예후 연관 레지스톰





## 2. Development of microbiome indicators: selection of indicators from microbiome and meta-data through artificial intelligence



**Diagnosis and Prognosis Prediction of  
Infectious Diseases Using Metagenomic  
Biomarkers and Artificial Intelligence .**

Min Hyuk Choi  
tcmhwd@yuhs.ac



# 03

## Development of microbiome solution based on precision classification platform



이제희  
(CJ Bioscience)





## 1부 연자



✓ 소속: CJ바이오사이언스

✓ 직위/직책: 수석연구원

✓ 성명: 이제희

### 🔍 학력사항

2002	서울대학교 자연과학대학 생명과학부 학사
2010	서울대학교 자연과학대학 생명과학부 박사

### 🔍 경력사항

2010 ~ 2011	국제백신연구소 분자미생물학과 Post doctor
2011 ~ 2020	Chunlab 신약개발본부 수석연구원
2021 ~ 현재	CJ바이오사이언스 Discovery 센터 수석연구원

# 정밀 분류 플랫폼 기반 마이크로바이옴 솔루션 개발

장내 분류 플랫폼을 통한 장내 미생물 분석 절차를 소개하고, 장내 미생물을 이용한 신약개발 전략에 대해 소개함



## 정밀 분류 플랫폼 기반 마이크로바이옴 솔루션 개발

Je Hee Lee



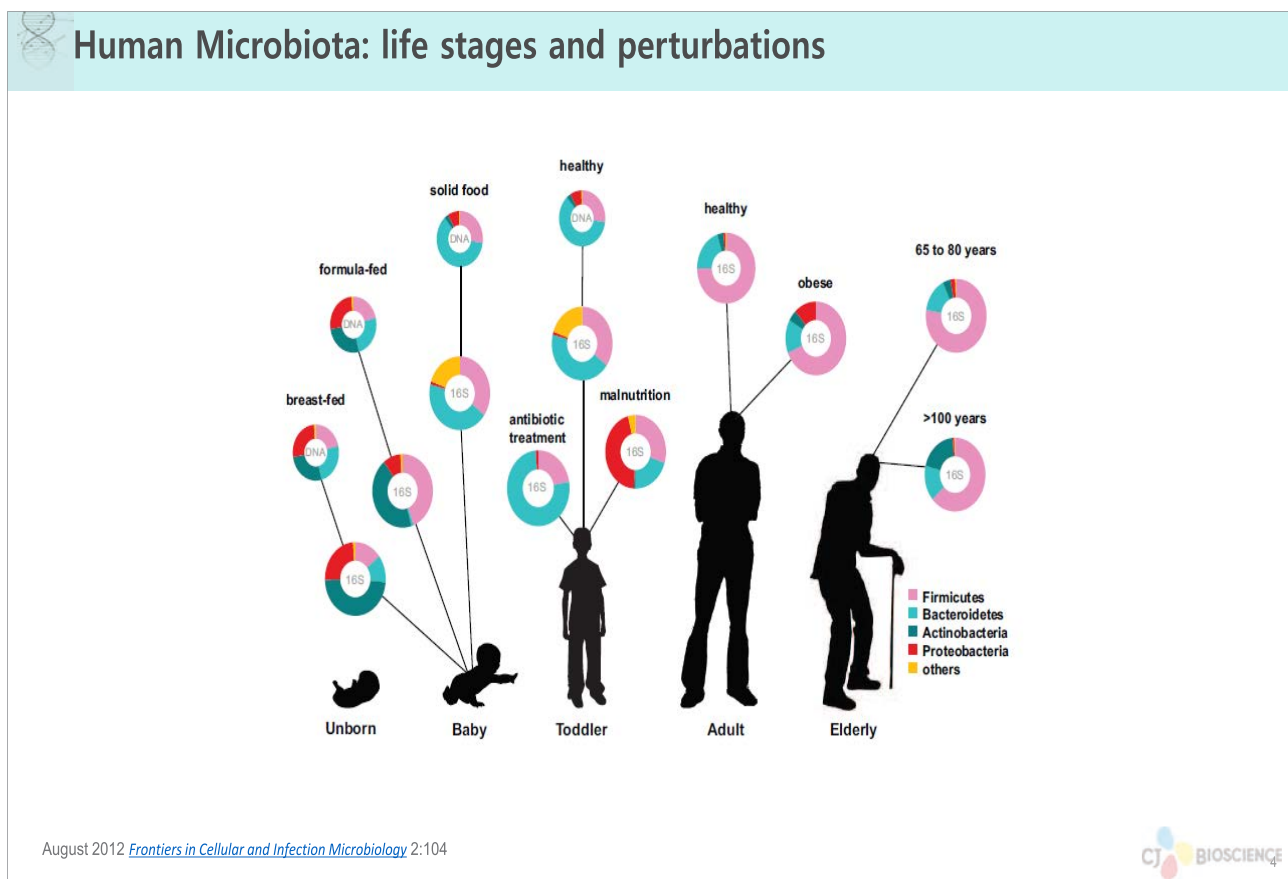
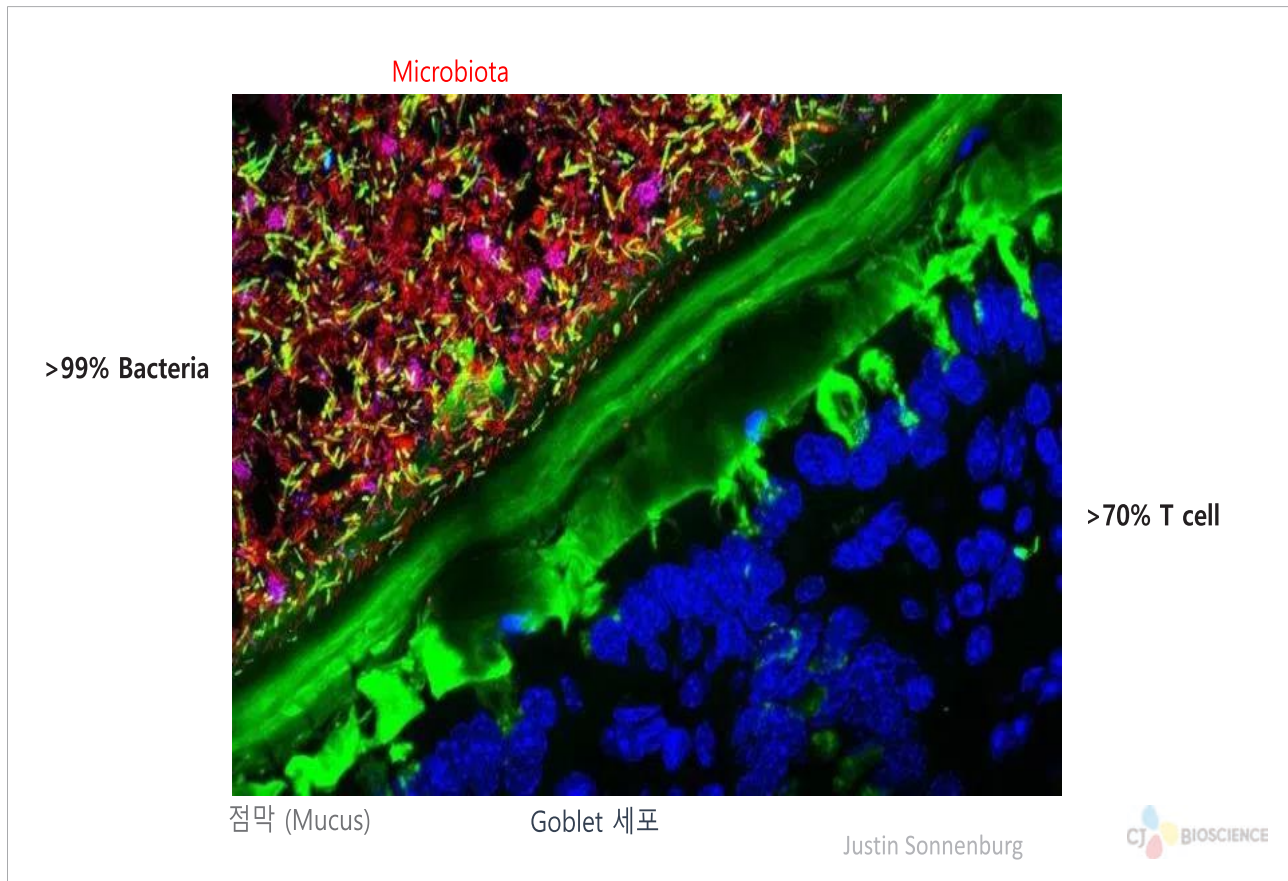
CJ바이오사이언스

### Microbe man

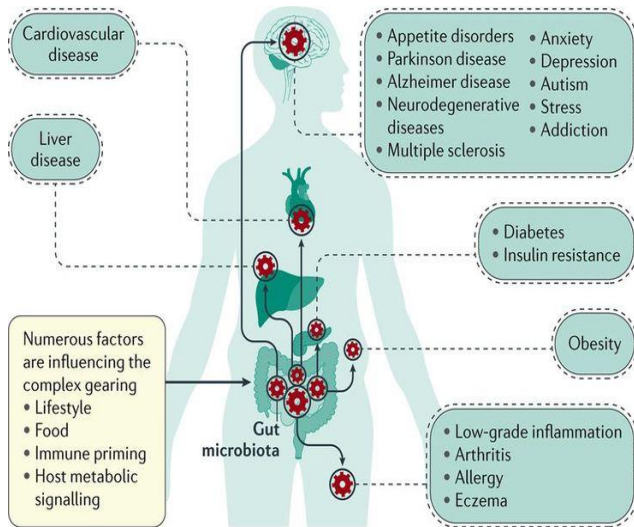


**Human =  
host + microbes**

- 인간 세포 수 약 30조
- 공생하는 미생물의 수 약 38조
- 인간 유전자 23,000
- 미생물 유전자 2백만 이상



## Many diseases are linked to human microbiome.



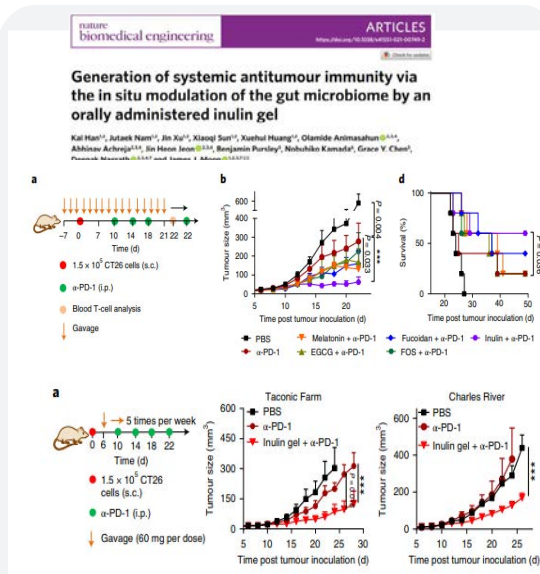
Nature Reviews | Gastroenterology & Hepatology



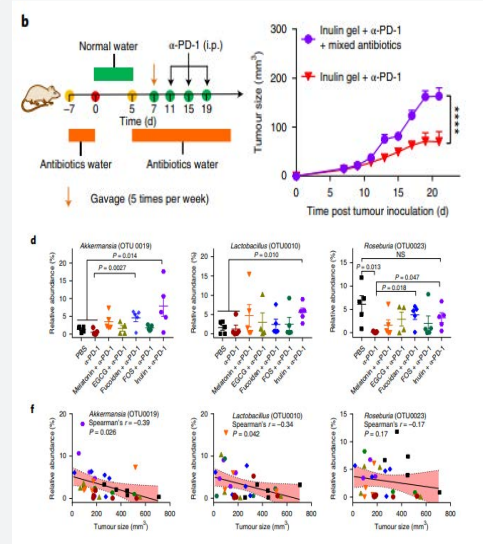
- 장내 미생물의 불균형은 다양한 질병에 연관됨
- 염증성 장질환
- 간질환
- 뇌질환
- 대사질환

## 식이섬유 (이눌린)과 암치료제와의 상관 관계 연구

- 이눌린을 통해 장내 세균을 변화 시키고, 이에 의해서 ( $\alpha$ -PD-1)의 항 종양 활성을 증폭시킴을 보고



- 이눌린 투입시 항종양 효과 확인
- 다양한 Mouse vender에서 재현성 데이터 확인



- 항생제 처리시 항암효과가 없어지는 것을 확인
- Akkermansia*, *Lactobacillus*, *Roseburia*가 증가 확인

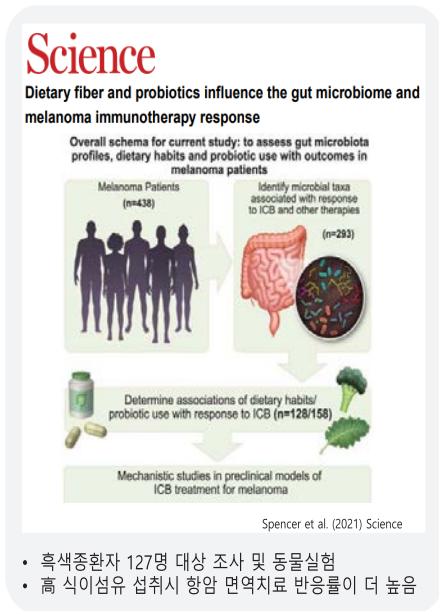


Nature Biomedical Engineering volume 5, pages1377–1388 (2021)



## 식이섬유와 암치료제와의 상관 관계 연구 (MD Anderson, Dr. Wargo 팀)

- 장내 마이크로바이옴은 약물 등에 의한 치료시 환자의 부작용을 완화하고 치료효과에 긍정적인 영향을 미칠 것으로 기대
- 치료 효과를 높일 수 있도록 식이를 이용한 장내 마이크로바이옴 변화 유도 및 효과 평가중



연구제목	Effect of Diet on the Immune System in Patients With Stage III-IV Melanoma Receiving Immunotherapy, DIET Study (DIET)
연구기관	M.D.Anderson Cancer Center
대상자	Pembrolizumab 또는 nivolumab 처방중인 흑색종(3기, 4기) 환자
내용	장내 마이크로바이옴 변화를 유발하는 식이(섬유질 많은 식단) 섭취 효과 확인 (11주간 섭취), 2상 진행 중
측정지표	마이크로바이옴, 면역, 대사, 삶의 질
임상시험 번호	NCT04645680

- 흑색종환자 대상 식이섬유 섭취시 치료효과 향상여부 연구
- 장내 마이크로바이옴 변화에 따른 효과 기대

### Precision nutrition\_Daytwo



- Cell, 2015 논문을 바탕으로 2015년 설립
- "Food-as-medicine"
- 알고리즘 기반 개인 맞춤형 식단 추천 서비스 제공

- B2B 사업 중심 -> 건강검진과 비슷한 개념으로 조직, 회사 단위로 서비스 제공
- Mayo clinic과 협업하여 임상시험 진행 -> JAMA network open, 2019. The American journal of clinical nutrition, 2019
- 미국에서 건강인을 대상으로 개인 맞춤형 식이를 제공했을 때 탄수화물 또는 칼로리 단독 보다 PPGR을 더 잘 예측했음
- 8만명 이상의 회원, fortune 500대 기업들이 service 이용 중
- \$499/1회, 매출 ~\$41M 추정

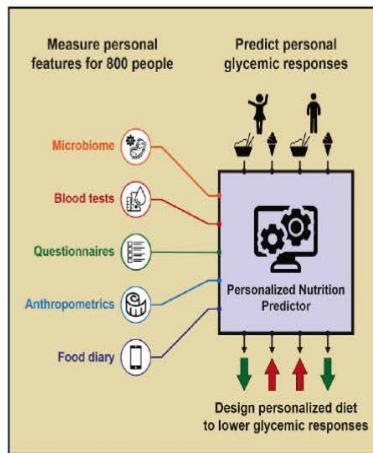


## Daytwo : 장내미생물-식이-혈당 DB를 통한 맞춤형 식단 제공

\*Observational Study of Blood Glucose Levels and Gut Microbiota in Healthy Individuals (13.04-15.04, Israel)

Personalized nutrition by prediction of glycemic responses, *Cell*, 2015 (2,218 citation)

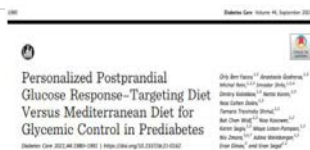
### Graphical Abstract



### Highlights

- High interpersonal variability in post-meal glucose observed in an 800-person cohort
- Using personal and microbiome features enables accurate glucose response prediction
- Prediction is accurate and superior to common practice in an independent cohort
- Short-term personalized dietary interventions successfully lower post-meal glucose

## Daytwo : 장내미생물-식이-혈당 DB를 통한 맞춤형 식단 제공



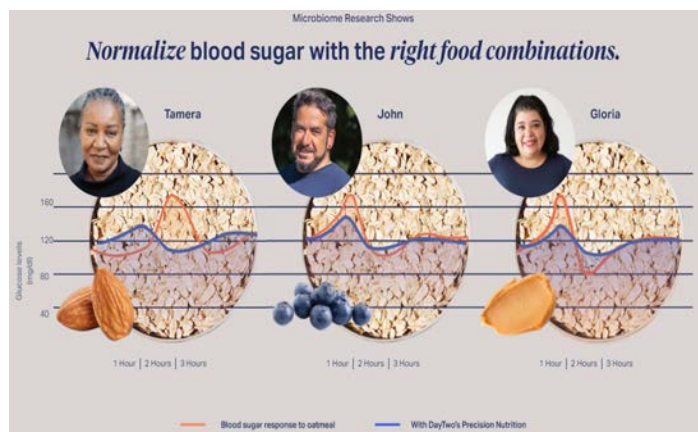
**OBJECTIVE**  
To compare the clinical effects of a personalized postprandial targeting (PPT) diet versus a Mediterranean (MED) diet on glycemic control and metabolic health in prediabetes.

**RESEARCH DESIGN AND METHODS**  
We randomly assigned adults with prediabetes ( $n = 32$ ) to follow a MED diet or a PPT diet for a 6-month dietary intervention and additional 6-month follow-up. The PPT diet relies on a machine learning algorithm that integrates clinical and microbiome features to predict personal postprandial glucose responses. During the intervention, all participants were counseled to consume glucose monitor (mg/dL) and self-reported dietary intake using a smartphone application.

**RESULTS**  
Among 32 participants randomized (24.7% women, mean  $\pm$  SD age  $59 \pm 7$  years, BMI  $32.3 \pm 3.8$  kg/m<sup>2</sup>, HbA<sub>1c</sub>  $5.8 \pm 0.5$ %,  $2$  h OGTT  $201 \pm 33$  mg/dL), fasting glucose  $128 \pm 12$  mg/dL,  $2$  h OGTT  $201 \pm 33$  mg/dL, and HbA<sub>1c</sub>  $5.8 \pm 0.5$ %, baseline characteristics were similar between the two groups. At baseline, all participants were counseled to consume glucose monitor (mg/dL) and self-reported dietary intake using a smartphone application.

**CONCLUSIONS**  
In this clinical trial in prediabetes, a PPT diet improved glycemic control significantly more than a MED diet as measured by daily time of glucose levels  $>140$  mg/dL (2.8 mmol/L) and HbA<sub>1c</sub>. These findings may have implications for dietary advice in clinical practice.

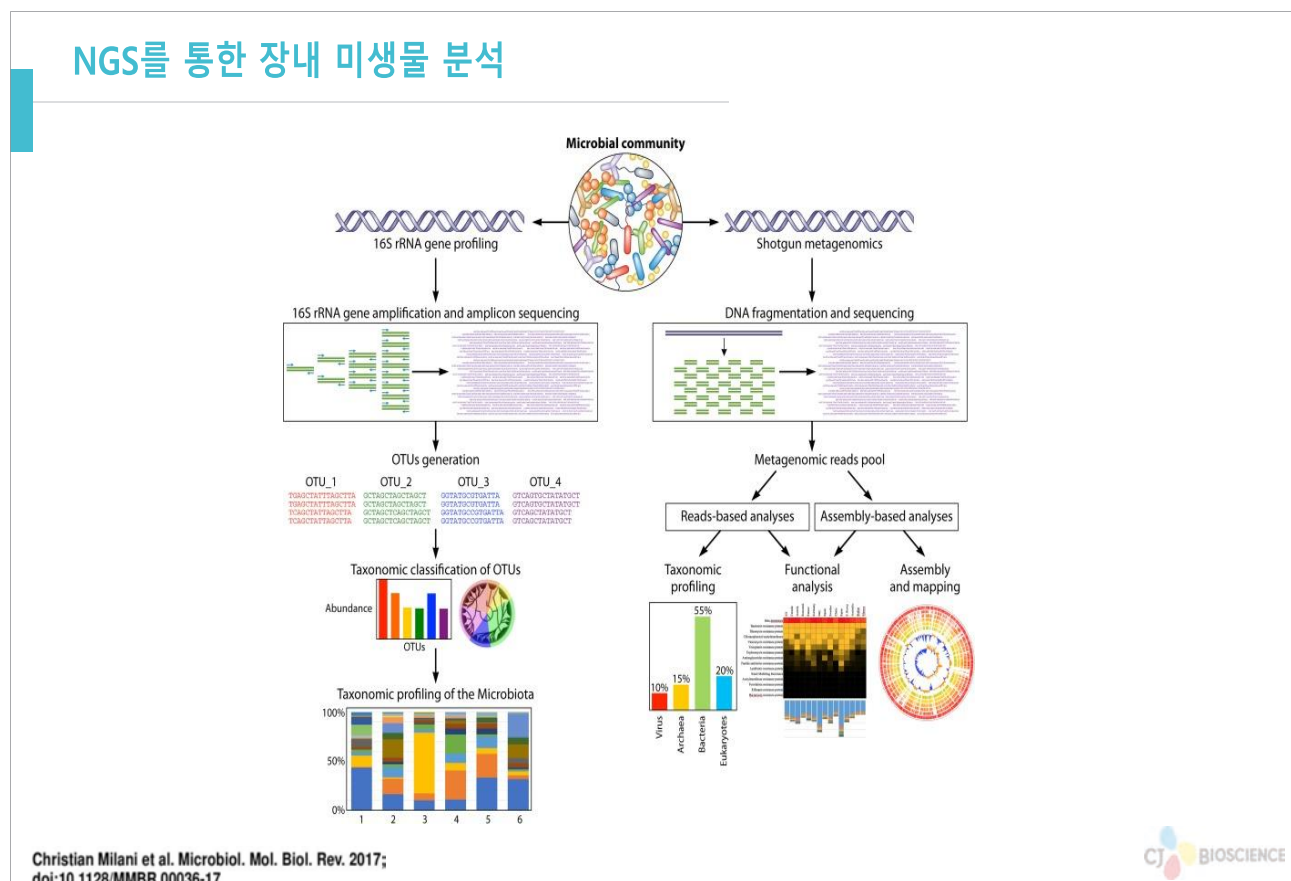
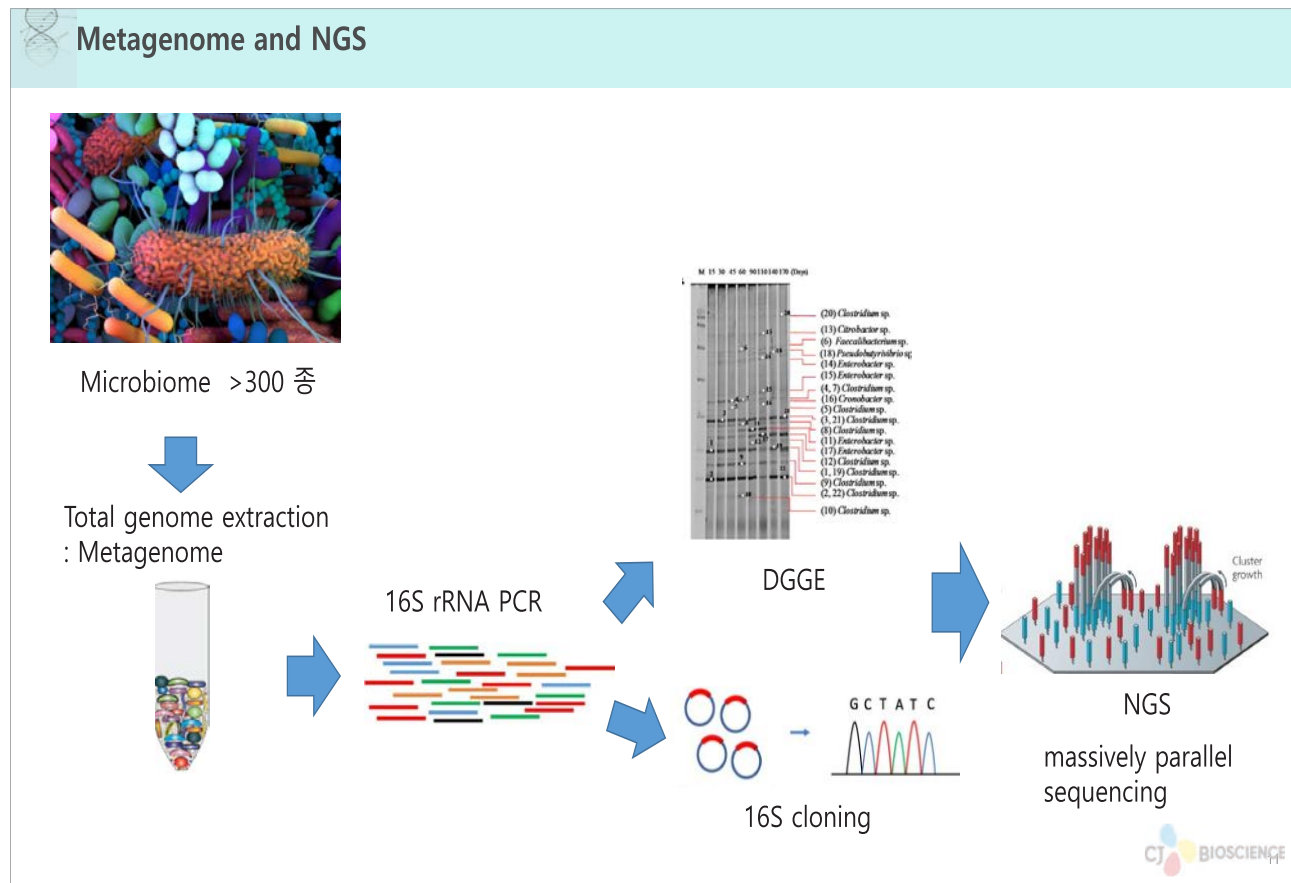
**TRANSLATION**  
Prediabetes is a leading risk factor for the development of type 2 diabetes and other metabolic abnormalities, such as cardiovascular and kidney disease (1,2). The progression of prediabetes in the adult population has increased dramatically in



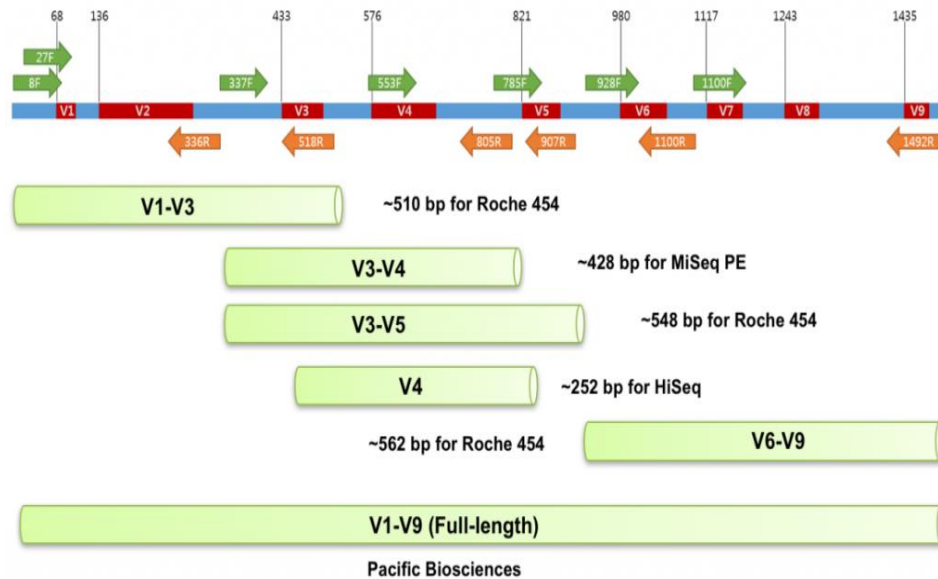
지중해식 식단 vs Daytwo solution 비교

개인별 맞춤형 식단 제공

→ Daytwo solution 에서 혈당 수치가 더 떨어짐



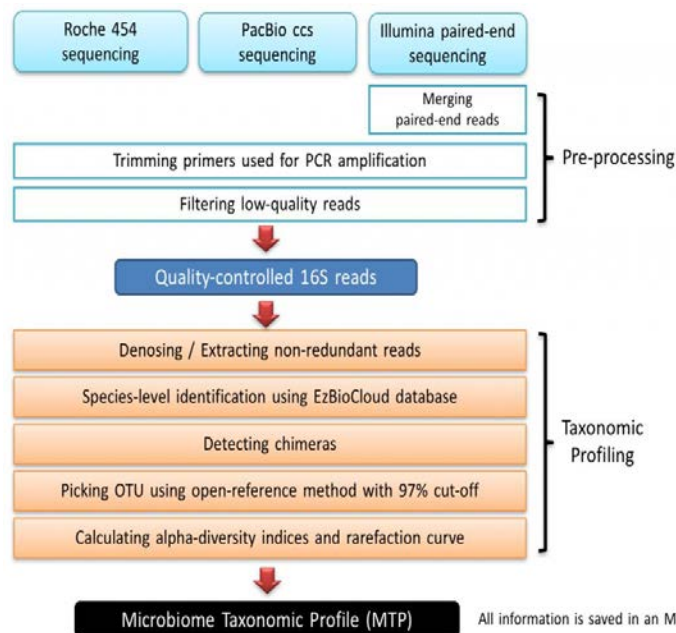
## PCR and sequencing of 16S rRNA gene



<https://help.ezbiocloud.net/16s-rna-and-16s-rna-gene/>



## Pipeline: 16S rRNA-based taxonomic profiling



<https://help.ezbiocloud.net/mtp-pipeline/>



## Limitations of Conventional Microbiome Research (16S rRNA analysis)

- 박테리아에서 가장 좋은 분류 마커임에도 불구하고, resolution의 한계
- 미생물의 종 정의에 따른 특정 마커의 한계
- Identification을 하기 위한 정확한 Database의 부재
- 난배양성 미생물 연구의 한계



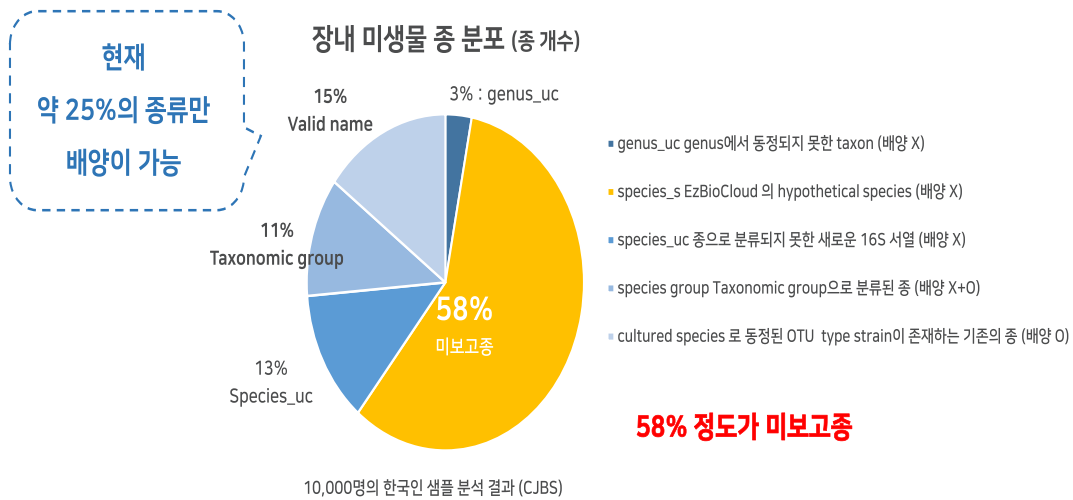
Trillion ( $10^{12}$ ) microbial species

Human gut  
> 1 Million

18,000  
prokaryotic  
species  
with valid  
names



## 장내 미생물 분석 : 난배양성 미생물



13



## 난배양성 분리 주요 후보군 선별

Microbiome 데이터베이스

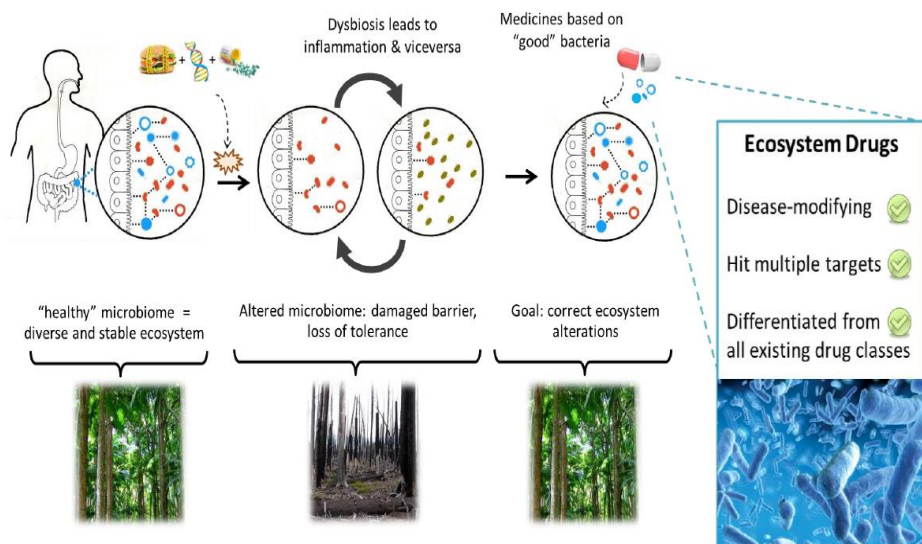
Genus	장내 비율 (%)	전체 종 수	미보고종 수 (EzBioCloud 기준)	난배양성 미생물 중 비율 (%)
<i>Bacteroides</i>	13.15	78	43	55.1
<i>Prevotella</i>	11.27	67	44	65.7
<i>Faecalibacterium</i>	9.09	10	9	90.0
<i>Blautia</i>	6.07	65	55	84.6
<i>Agathobacter</i>	2.23	9	7	77.8
<i>Lachnospira</i>	2.00	16	12	75.0
<i>Megamonas</i>	1.86	3	2	66.7
<i>Roseburia</i>	1.49	8	5	62.5
<i>Dorea</i>	1.25	6	3	50.0
<i>Ruminococcus</i>	1.14	38	36	94.7
<i>Alistipes</i>	0.69	33	23	69.7
<i>Collinsella</i>	0.68	8	4	50.0
<i>Alloprevotella</i>	0.63	13	10	76.9
<i>Coprococcus</i>	0.57	4	3	75.0
<i>Clostridium</i>	0.41	44	22	50.0
<i>Akkermansia</i>	0.37	3	2	66.7

10,000명의 한국인 샘플 분석 결과 (CJBioscience)



## Live Biotherapeutic Products

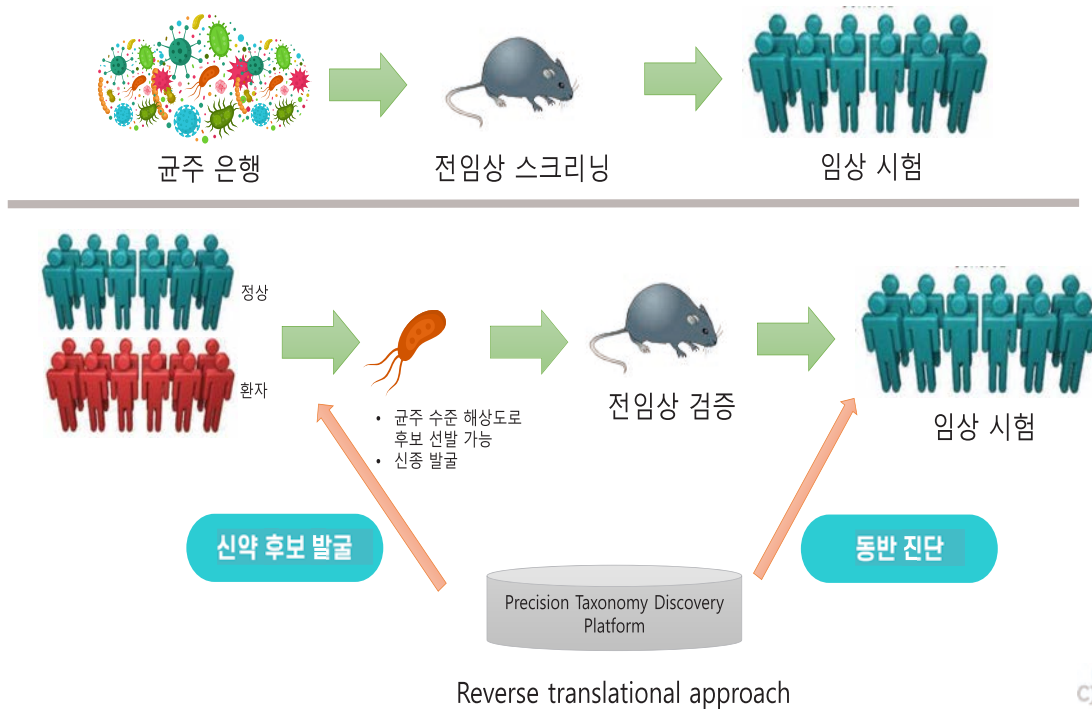
장내 불균형을 회복시키는 방법 : 살아있는 생균제를 통한 치료



CJ BIOSCIENCE

## Human data-driven rationally designed LBP

전통적인 치료제 개발

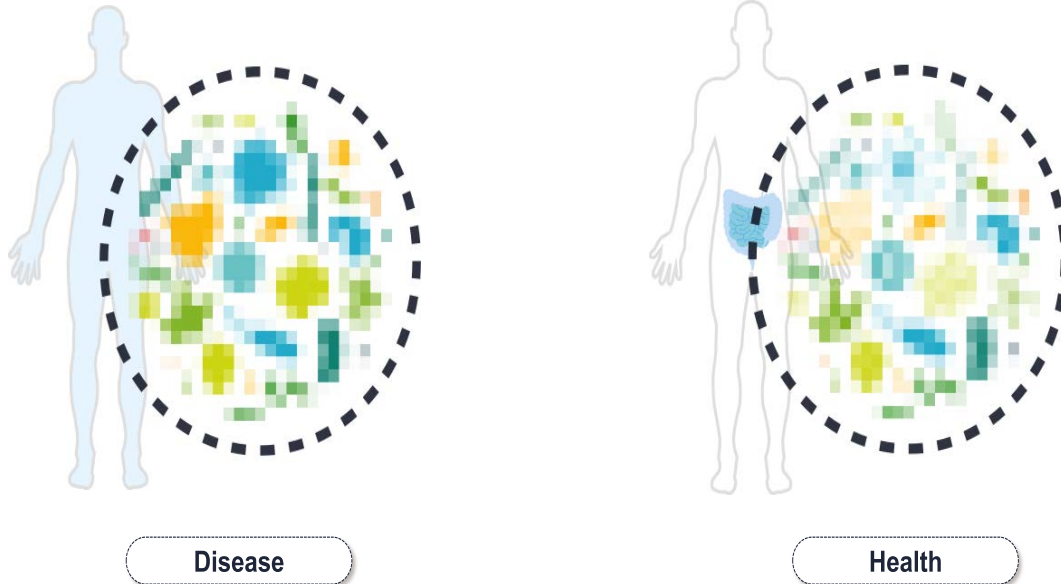


CJ BIOSCIENCE



## Limitations in Conventional Analysis Resolution

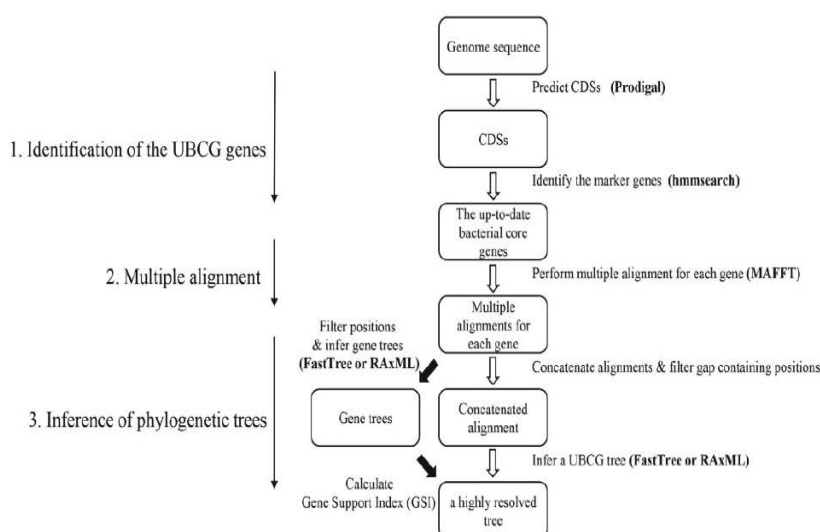
정확한 Reference database의 부재에 의해서 정확한 동정이 되기 힘들



**NO clear difference in health and disease**



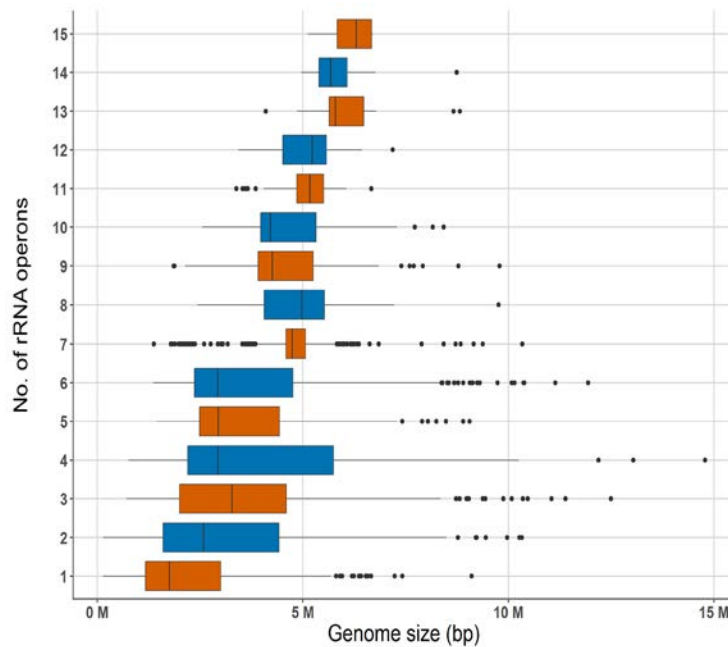
## UBCG : Up-to-date bacterial core gene set and pipeline for phylogenomic tree reconstruction



**Fig. 1.** The process of phylogenetic tree reconstruction using the UBCG pipeline. Each of UBCG genes is aligned separately before being concatenated into a single alignment. The pipeline generates 92 gene trees and one UBCG tree that is labeled with Gene Support Index (GSI) values. Externally executed software tools are indicated in the parentheses.



## Number of rRNA operons vs. Genome size



Based on the EzBioCloud database (4,131 complete genomes)



## 16S rRNA gene copy numbers of major gut microbiota

*Bacteroides fragilis* 6 copies

*Prevotella copri* 4 copies

*Escherichia coli* 7 copies

*Akkermansia muciniphila* 3 copies

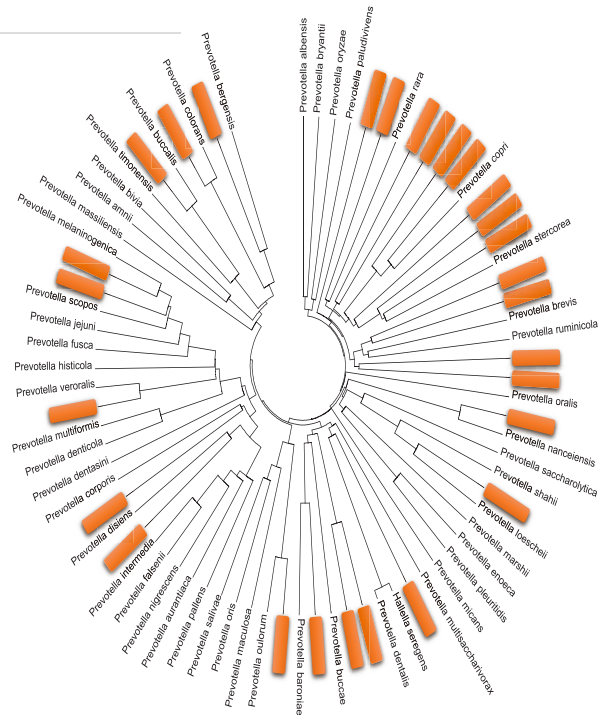
All data available from <https://www.ezbiocloud.net/>



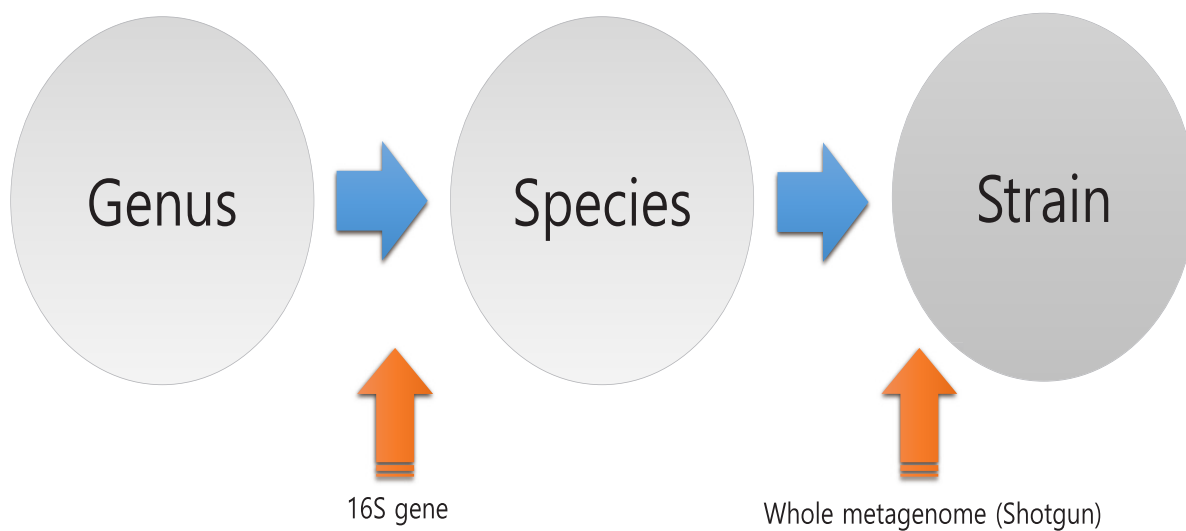


## The genus *Prevotella*

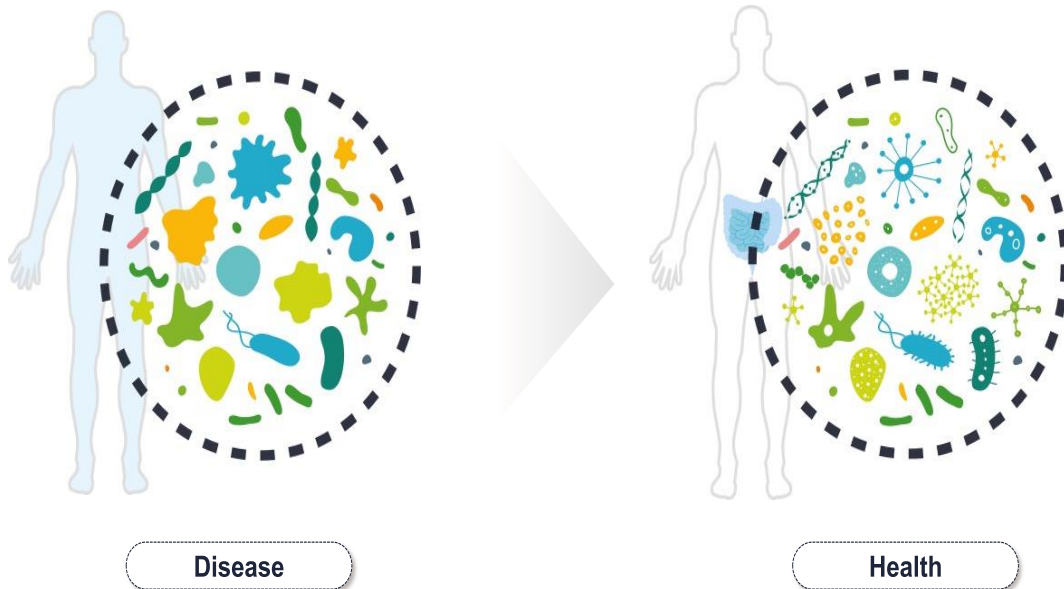
- Major genus of human gut microbiome
- 50 known species vs. 28 novel species



## Towards higher resolution

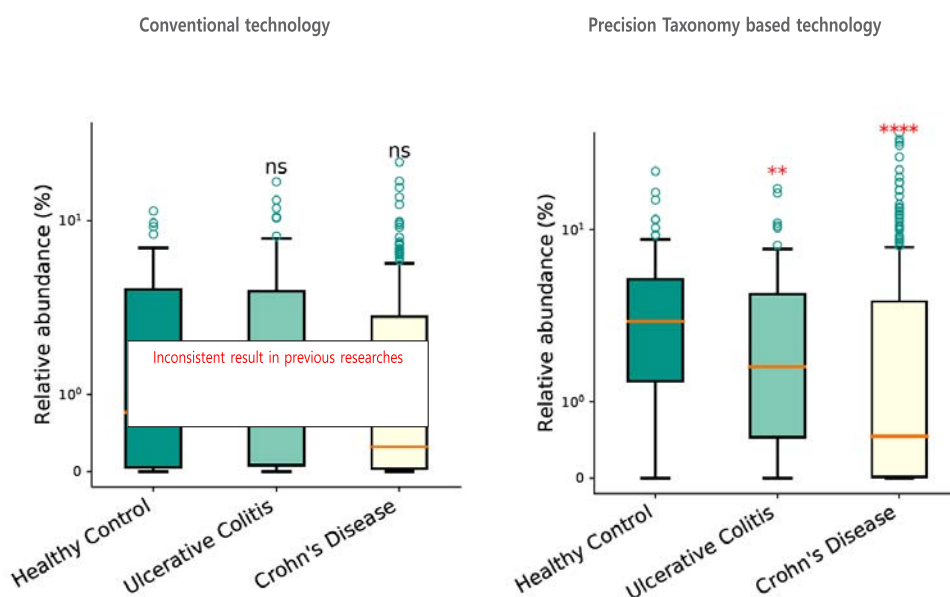


## Why Precision-Taxonomy Platform? – Highly accurate identification



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## Precision taxonomy 예시



CJ BIOSCIENCE

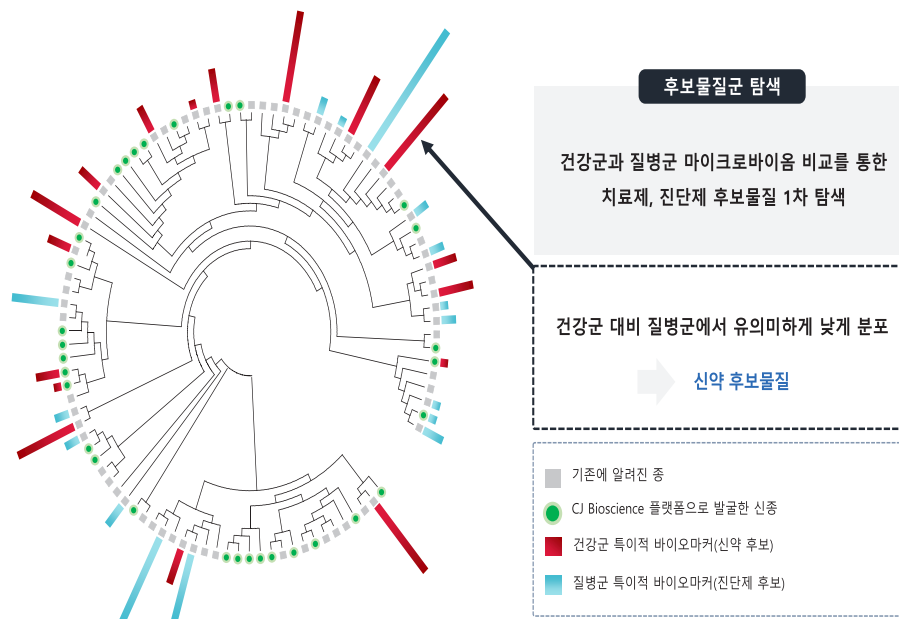
## LBP 치료제 기업



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## 치료제 후보물질 탐색

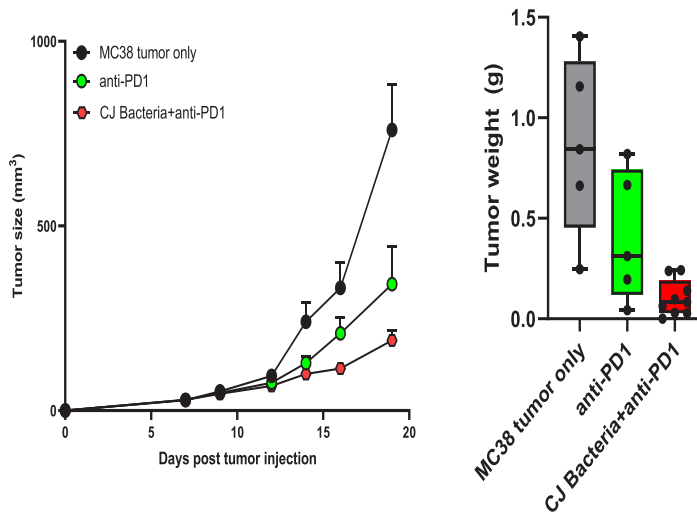
건강군과 질병군 마이크로바이옴 비교를 통한 치료제, 진단 후보물질 1차 탐색



CJ BIOSCIENCE

## Lead Pipeline\_Immuno-Oncology (CJRB-101)

- Promotes Anti-Tumor Immunity and Facilitates Anti-PD1 Efficacy (Therapeutic Model)



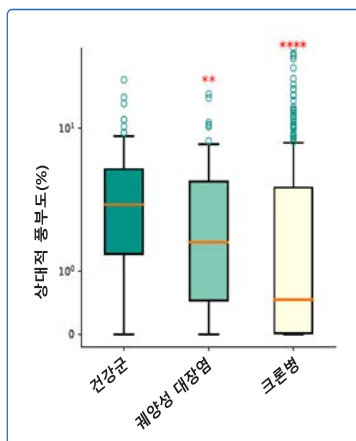
## Lead Pipeline\_IBD (CJRB-201)

- IBD와 높은 상관관계를 보이는 치료제 후보, CJRB-201 균주 발굴

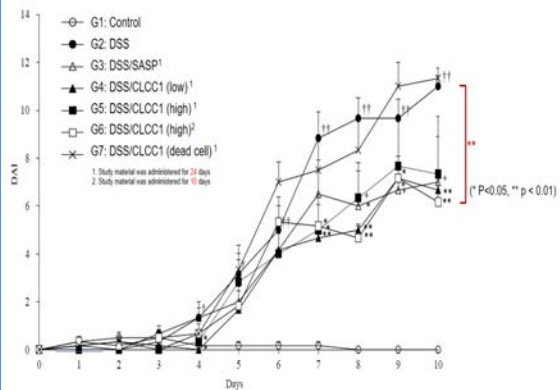
### CJRB-201 균주와 IBD의 상관관계

#### CJRB-201

정밀 분류 플랫폼을 통해 크론병, 궤양성 대장염 등 IBD 와 연관성을 보이는 균주 발굴



\* Ulcerative Colitis (궤양성 대장염), Crohn's Disease (크론병)



## 마이크로바이옴 신약 파이프라인

- 마이크로바이옴 관련 주요 질병 대상 치료제 개발 진행

	과제명	적응증	전임상	1상	2상	비고
면역항암	CJRB-101	고형암(Keytruda 병용)	IND 승인			
	CJRB-102	고형암	P1/2			MRx0518
	CJRB-103	고형암				MRx1299
소화기질환	CJRB-201	염증성 장질환(IBD)				CLP-105
	CJRB-202	염증성 장질환(IBD)				
	CJRB-203	염증성 장질환(IBD)				MRx1233
	CJRB-204	염증성 장질환(IBD)				MRx0001
	CJRB-205	과민성대장증후군(IBS)				MRx1234
중추신경계	CJRB-301	파킨슨(PD)	IND 승인			MRx0005
	CJRB-302	파킨슨(PD)	IND 승인			MRx0029
	CJRB-303	자폐증(ASD)				MRx0006
	CJRB-304	다발성 경화증(MS)				MRx0002
호흡기	CJRB-401	천식				
	CJRB-402	천식	P1/2			MRx0004
근골격계	CJRB-901	류마티스				MRx0006



## 장내미생물 치료제 방향성

Discovery



분석 방법 표준화  
(절대 정량)  
다양한 스크리닝 전략 개발

Chemistry  
Manufacturing  
and Control



LBP 물질 표준 측정 방법  
난배양성 균주의 대량 배양 고도화

임상



개인별 장내 미생물 분류/분석 방법 고도화





# 04

## Development of microbiome treatment



임상균  
(고바이오랩)







# 1부 연자



- ✓ 소속: (주)고바이오랩
- ✓ 직위/직책: 연구소장
- ✓ 성명: 임상균

## 🔍 학력사항

2001	서울대학교 미생물학과 학사
2006	위스콘신대학교 종양생물학 박사

## 🔍 경력사항

2007 ~ 2011	UT Southwestern, 박사후 연구원
2011 ~ 2016	UT Southwestern, 전임강사
2016 ~ 2018	Harvard Medical School, 책임연구원
2018 ~ 2019	VividBio (LLC), 수석연구원
2019 ~ 2023	카나프 테라퓨틱스, 상무이사
2023 ~ 현재	고바이오랩, 연구소장

## 마이크로바이옴 치료제 개발

다양한 질환들에서의 마이크로바이옴 연구들이 진행되어가며, 질환과 마이크로바이옴의 상관관계 뿐 아니라, 치료제로서의 개발가능성이 대두되었고, 실제 여러 질환 모델에서 마이크로바이옴 기반 신약 개발이 진행 중에 있음. 본 강연에서는 현재 마이크로바이옴 치료제 개발 현황을 리뷰하며, 타겟 질환들과 또 마이크로바이옴 기반의 다양한 modality 접근법에 대해 정리/발표될 예정이며, 이를 기반으로 한 하나의 예시로 장질환을 타겟하는 균주 개발 연구 과정에 대해 소개할 예정임.

# 마이크로바이옴 치료제 개발

2024-12-3

임상균, Ph.D.

(주)고바이오랩

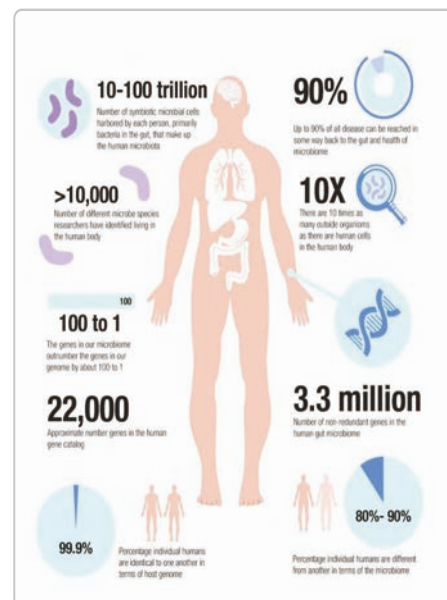
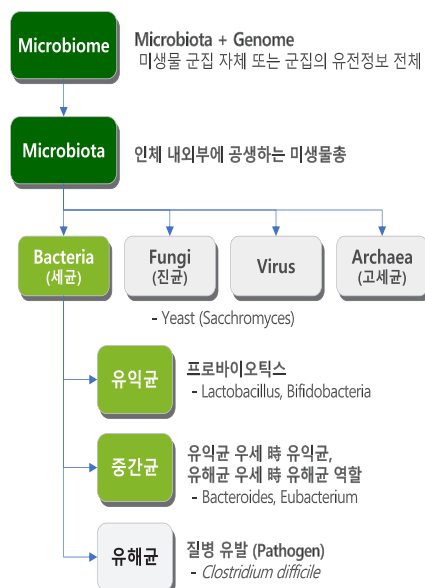


1

## Microbiome



**Microbiome is the microorganism community itself or the entire genetic information of microorganisms present in a specific environment**



Source) www.scdprobiotics.com/The-Importance-of-the-Microbiome-s/826.htm

Microbiome and Disease

KOBIOLABS

Human microbiota dysbiosis is implicated in various diseases

Heart disease

Hypertension

Atherosclerosis

Inflammatory bowel disease

Crohn's disease

Ulcerative colitis

Liver disease

Cirrhosis

Hepatitis

Chronic kidney disease

Brain disorders

Parkinson's disease

Alzheimer's disease

Depression

Diabetes

Type1

Type2

Gestational

Respiratory disease

Asthma

Bronchitis

Cancer

Lung cancer

Colorectal cancer

Pancreatic cancer

Oral cancer

Microbiota

Dysbiosis

Metabolites

Vitamin

SFAC

IL-6

Inflammatory Cytokines

LPS

TNF-α

Correlation

Function (Driver vs. Passenger)

Therapy

Hou K et al (2022)

Signal transduction and targeted therapy

Trends in Global Microbiome Therapeutics Development

KOBIOLABS

Indication<sup>1)</sup> & Development Stage (2024.1Q)

Pre-Clinical

Phase 1

Phase 2

Phase 3

IBS/IBD

10

8

3

21

Oncology

7

7

5

19

Metabolic Disorders

6

1

5

1

13

AD/Psoriasis

4

2

4

10

Infection

1

4

2

2

9

ASD

2

1

1

4

Microbiome Modality

Donor-derived Consortia & FMT

Rationally-designed Consortia

Single Strain

Engineered Bacteria

Bacterial Metabolite & Protein

60

76

27%

7

11

26

9

7

8

17

35

8

8

(12%)

(18%)

(43%)

(15%)

(12%)

(11%)

(22%)

(46%)

(11%)


(11%)

23.2Q


24.1Q

Note: 1) Major indications & therapeutic areas (IBS/IBD, oncology, ASD, metabolic disorders, infectious disease, atopic dermatitis, psoriasis) & development stage: pre-clinical, phase1, phase2, phase3


Source: KoBioLab's analysis (based on Pharamaproject, GlobalData)



## Microbiome Therapy – *C. difficile* infection (CDI)





**Rebyota® (FMT)**



**“The first FDA approved microbiome therapy”**

- ▶ **Key Technology**
  - By transplanting fecal matters from healthy donors, it restores gut microbial diversity and suppress CDI
  - Rectal enema administration (Single-dose)
- ▶ **Approval Status**
  - FDA approval: November 2022
  - First Launch (USA): February 2023
- ▶ **Phase 3 Results**
  - Phase III clinical evaluation criteria
    - CDI recurrence rate reduction with Rebyota®
    - Success rate (Week 8): 70.6%, compared to placebo 21.7% ↑






- Acquisition of company (Rebiotix) (Deal: Undisclosed value, April 2018)


Note: 1) *Clostridium difficile* infection  
 Sources: Press releases from each company, Evaluate Pharma


**Vowst® (Bacterial Spores)**



**“The first oral microbiome therapeutics marketed”**


- ▶ **Key Technology**
  - Oral version of FMT therapeutics, administering bacterial spores of ~86 species
  - Bacterial spores repopulates in gut and restores the gut microbiome's composition
- ▶ **Approval Status & Sales**
  - FDA approval: April 2023
  - First Launch (USA): June 2023
  - Expected Revenue ('23. 4Q) : \$22 million USD
- ▶ **Phase 3 Results**
  - Phase III clinical evaluation criteria
    - CDI recurrence rate reduction with VOWST®
    - Recurrence rate (Week 8): 12%, compared to placebo 27.4% ↓






- SER-109
- Co-commercialization license agreement
- US & Canada rights (2021, total \$525M)
- Outside US & Canada rights (2016, total \$1.9B)

5



## Hurdles in Developing Microbiome Therapy



LBP & Effector Molecule

- Discovery of **new LBP candidate strains**
- Need for defined **microbial components with distinct function**

Personalized Microbiome Therapy

- Tailored treatments to **microbiome profile, gene/metabolite analysis**

Synthetic Biology & Advanced Delivery

- **Engineered bacteria** to produce specific anti-inflammatory metabolites
- **Targeted microbiome/drug delivery systems**

Long-Term Safety

- Collection of long-term clinical data to ensure **safety**
- Establish new **regulatory standards**

Conducting more detailed research to **elucidate the exact molecular mechanisms** is ongoing.

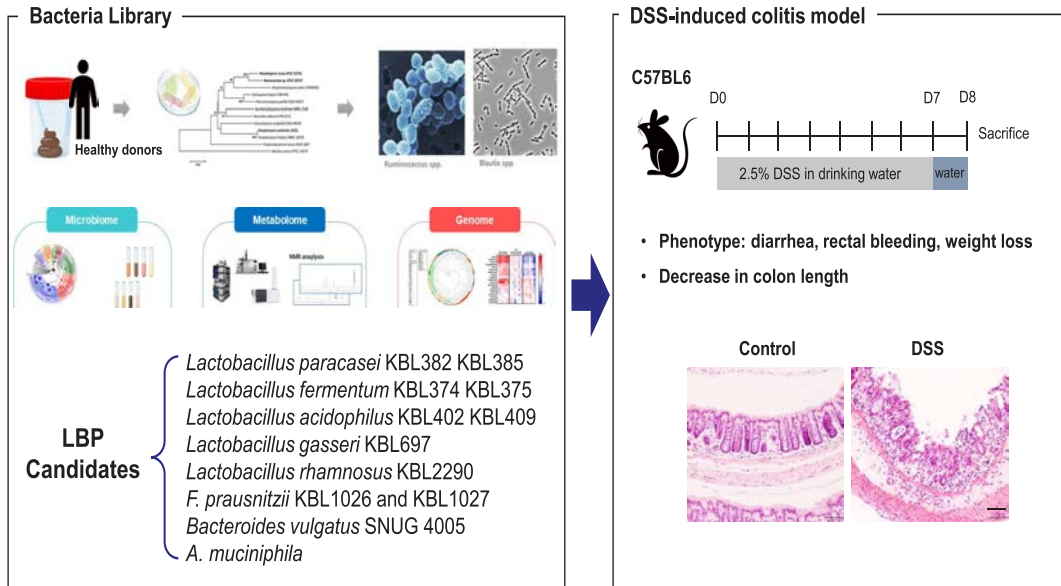
This approach includes **genomic, proteomic, and metabolomic analyses** to better understand the interactions between microbiota and host.

6

## Screening LBP Candidates – Irritable Bowl Disease (IBD)



KOBIO LABS

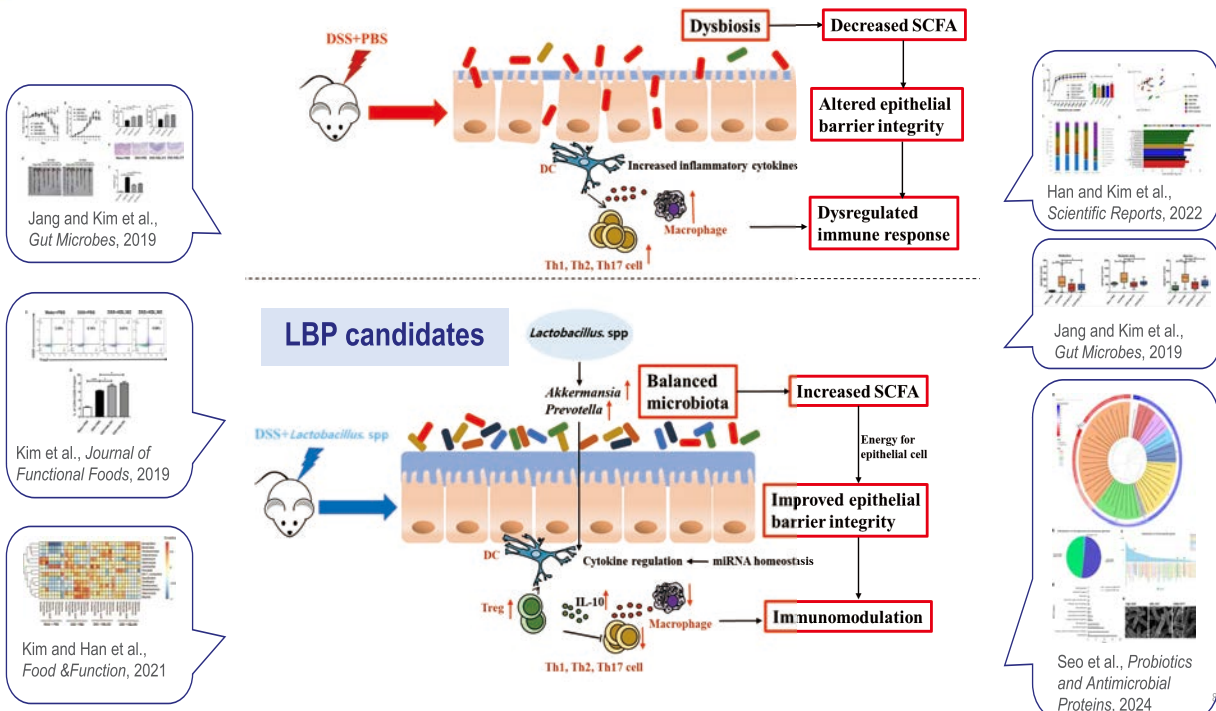


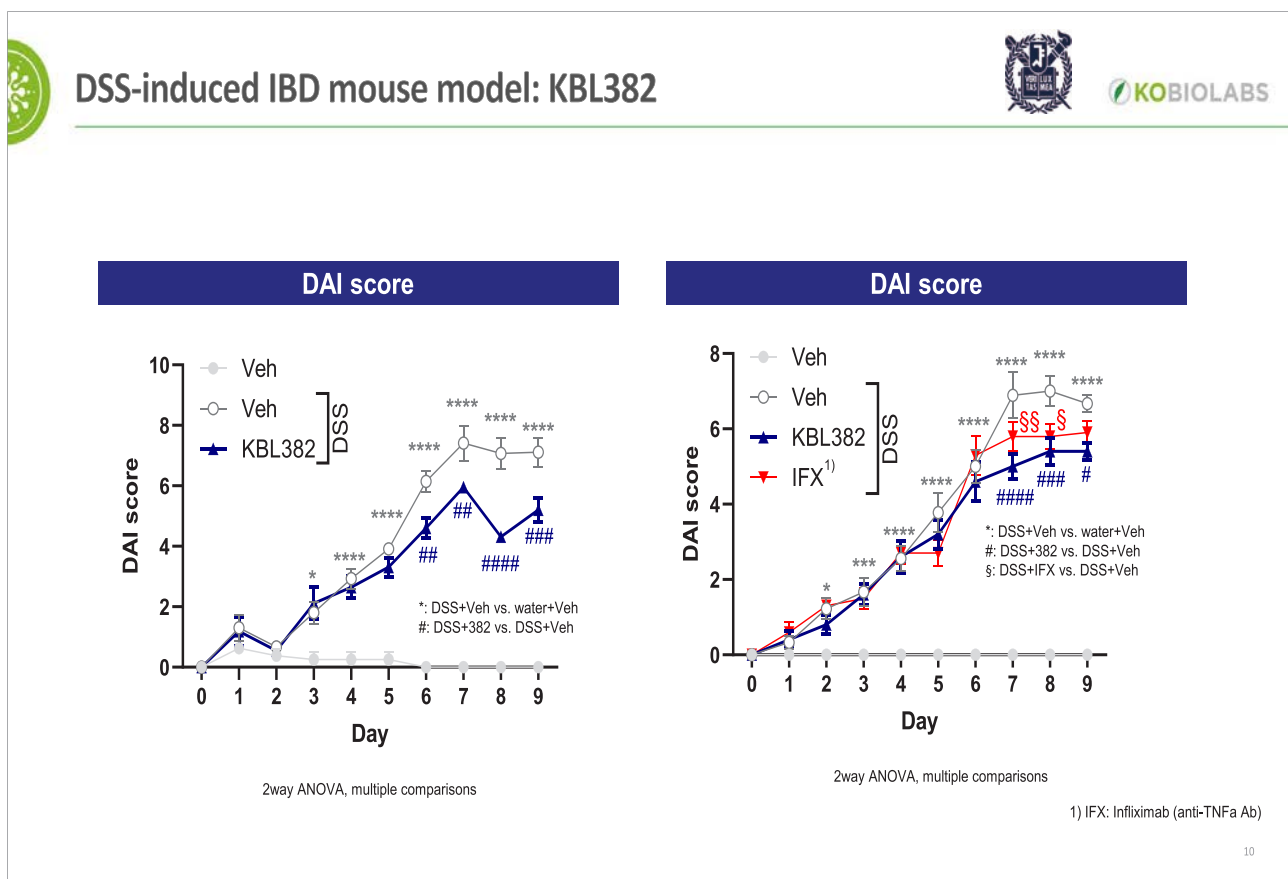
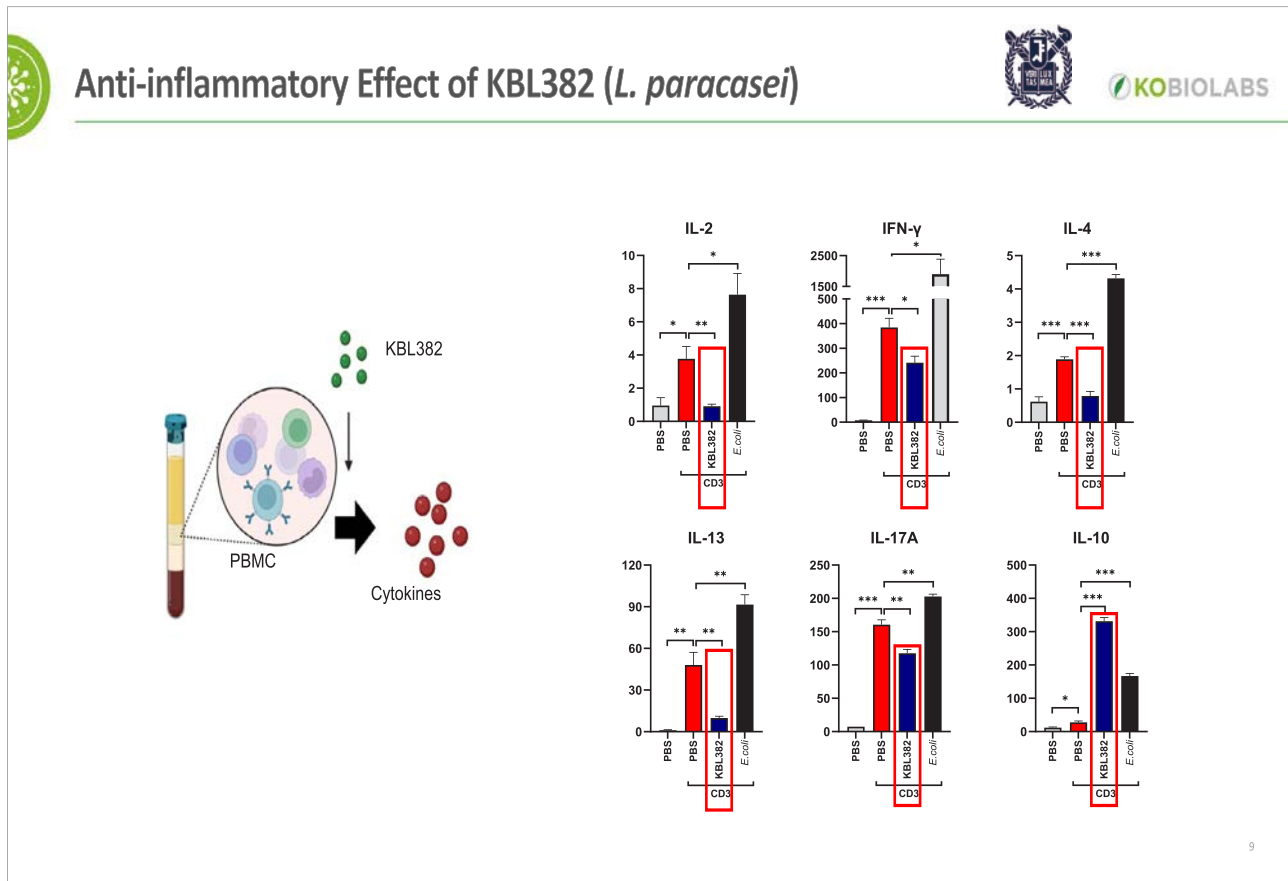
7

## Identification of LBP Candidates in IBD Mouse Model

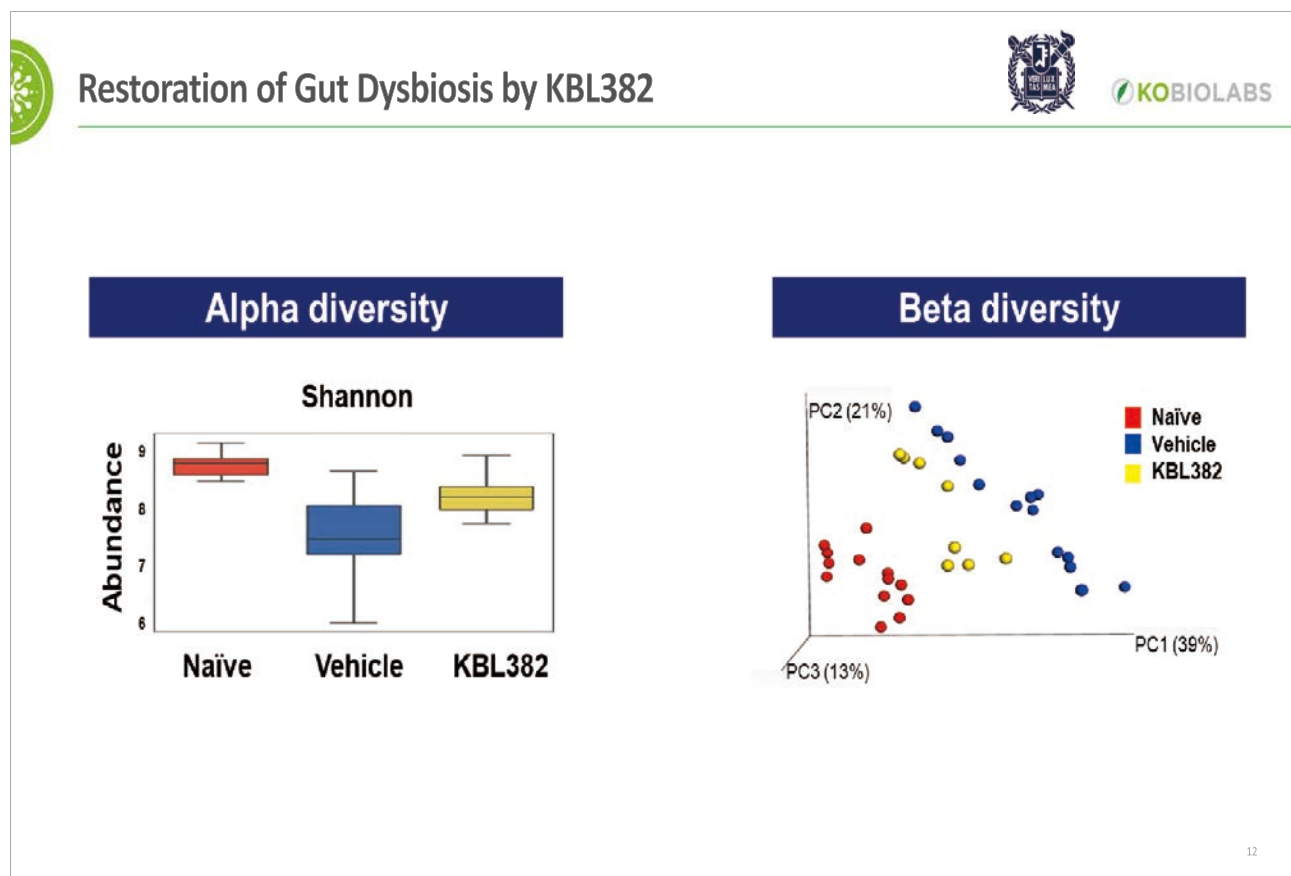
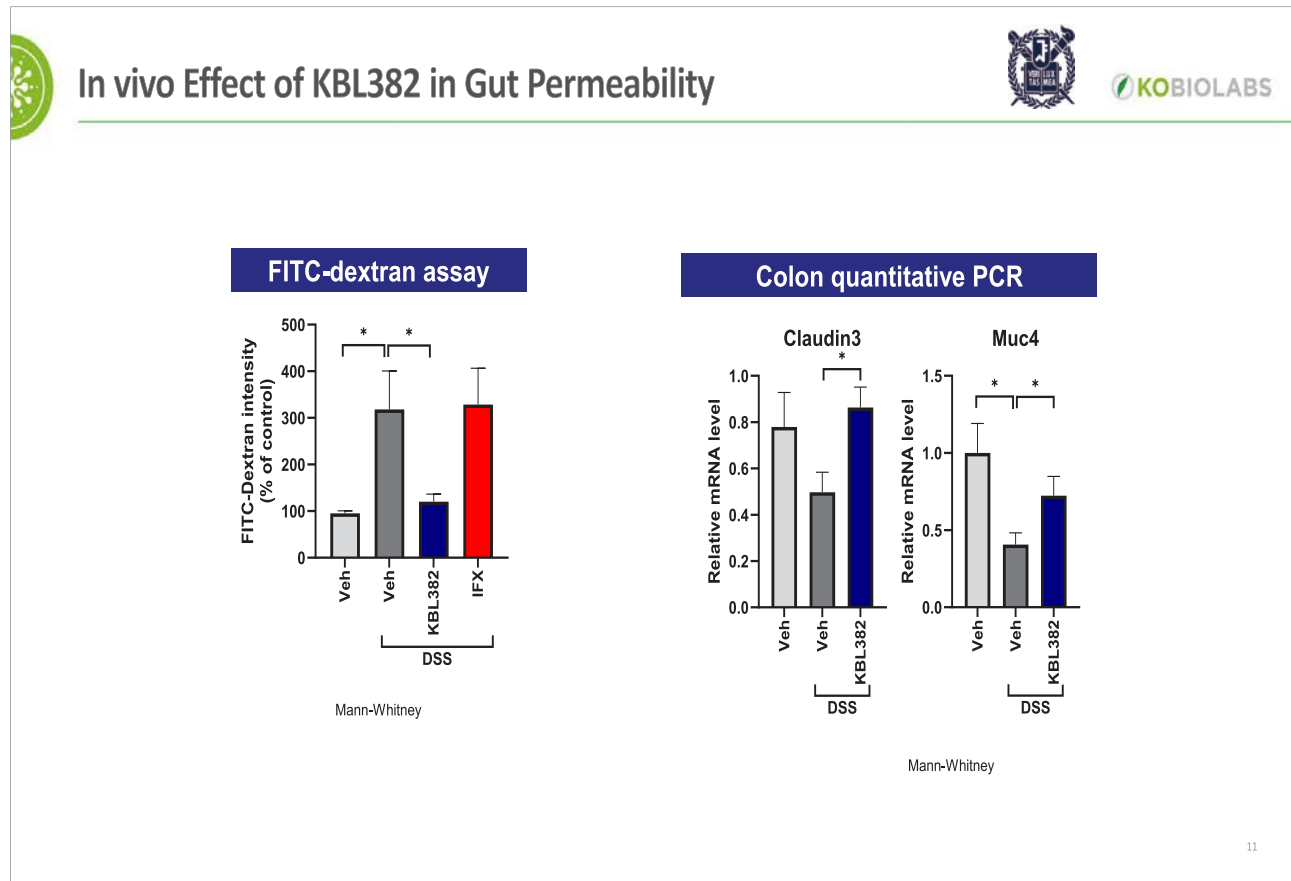


KOBIO LABS





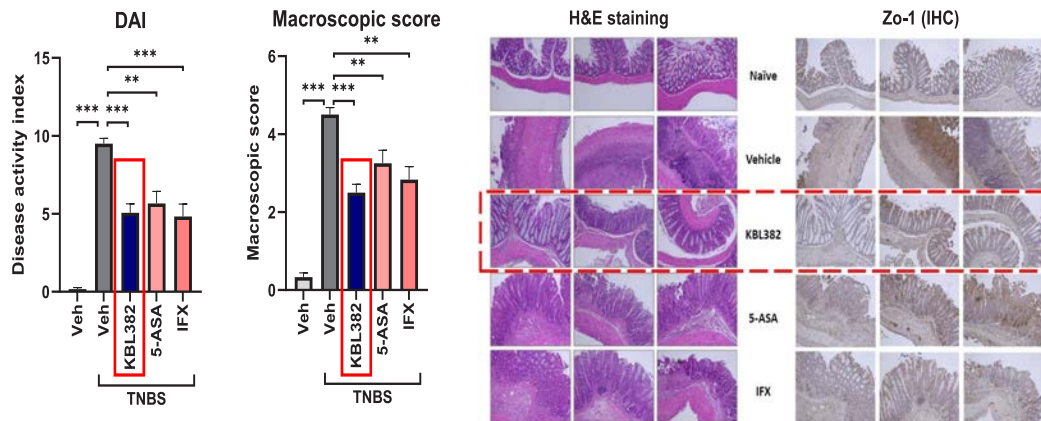




## TNBS-induced IBD mouse model: KBL382



KOBIOLABS



- Confirmation of mucosal healing effect by KBL382
- Better than or equivalent to 5-ASA or infliximab

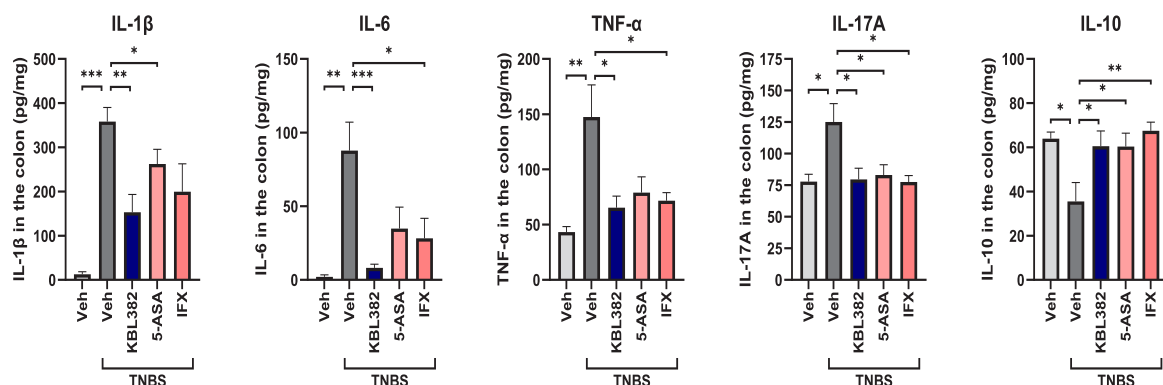
13

## TNBS-induced IBD mouse model: KBL382



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### Colonic cytokines



14



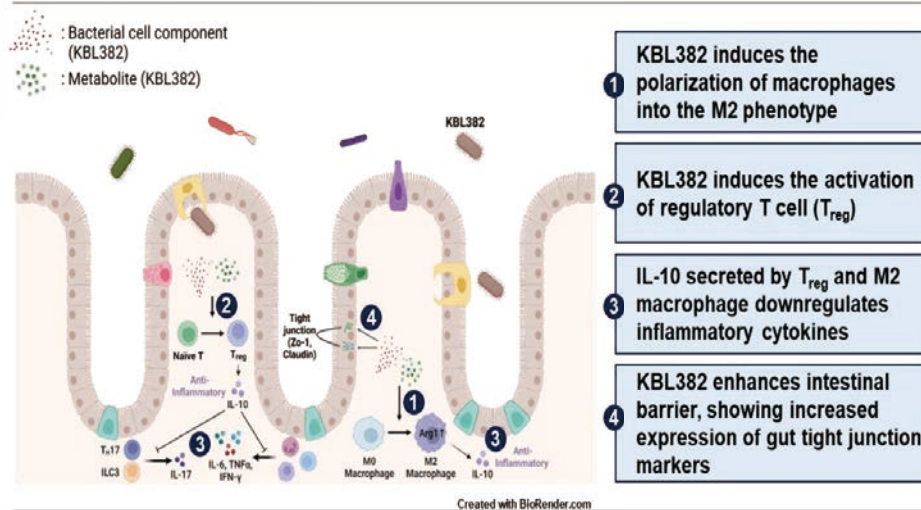
## Mechanism underlying the effect of KBL382



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**KBL382 improves IBD symptoms by activating M2 macrophage and  $T_{reg}$ , increasing IL-10 and reducing inflammatory cytokines**

### Mechanism of Action of KBL382<sup>1)</sup>



Note: 1) MOA study is in progress. M2 polarization of macrophage and induction of cytokines were studied and published (shown in the appendix)  
Source: D.H. Han, W.K. Kim, S.J. Park, Y.J. Jang, G.P. Ko, Lactobacillus paracasei treatment modulates mRNA expression in macrophages, Biochemistry and Biophysics Reports 23 (2020)

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## International Human Microbiome Consortium (IHMC)

M I C R O B I O M E  
C O N S O R T I U M

**IHMC 2026**

**June 3-5, 2026**  
**COEX, Seoul, Korea**

C O N G R E S S  
2 0 2 6







# 05

## Excavation and development of microbiome biomarkers applicable to clinical diagnosis



김지현  
(연세대학교)







# 1부 연자



- ✓ 소속: 연세대학교
- ✓ 직위/직책: 교수
- ✓ 성명: 김지현

## 🔍 학력사항

1989	서울대학교 학사
1991	서울대학교 석사
1997	Cornell University Ph.D.

## 🔍 경력사항

1997 ~ 2000	Cornell University Postdoctoral Associate
2000 ~ 2012	한국생명공학연구원 선임연구원/책임연구원/센터장
2012 ~ 현재	연세대학교 부교수/교수/학과장
2014 ~ 2022	다부처 유전체사업 미생물유전체전략연구사업단장
2016 ~ 현재	한국과학기술한림원 준회원/정회원/학부운영위원
2021 ~ 현재	연세 마이크로바이옴연구원장
2022 ~ 2024	연세대학교 연구처장/산학협력단장/기술지주 대표이사
2024 ~ 현재	연세 이윤재 펠로우
2024 ~ 현재	RECOMB 2025 조직위원장 / RECOMB-Microbiome 초대 원장

# 진단 적용 가능한 마이크로바이옴 바이오마커 개발

## Microbiome biomarkers for clinical diagnosis and prognosis of gastrointestinal cancers

Gastrointestinal cancers including gastric cancer (1.09 million cases and 769 thousand deaths in 2020) and colorectal cancer (1.93 million cases and 916 thousand deaths in 2020) are among the most common cancers and are leading causes of death worldwide. Although the gut microbiota is considered as a key player in the initiation and progression of gastrointestinal cancers, most retrospective or prospective studies have been focused on particular pathobionts such as *Helicobacter pylori* and *Fusobacterium nucleatum*. Here, we aimed to identify novel diagnostic or prognostic bacteria for gastric cancer and colorectal cancer by examining the gastric or colonic microbiota through 16S ribosomal RNA gene sequencing.

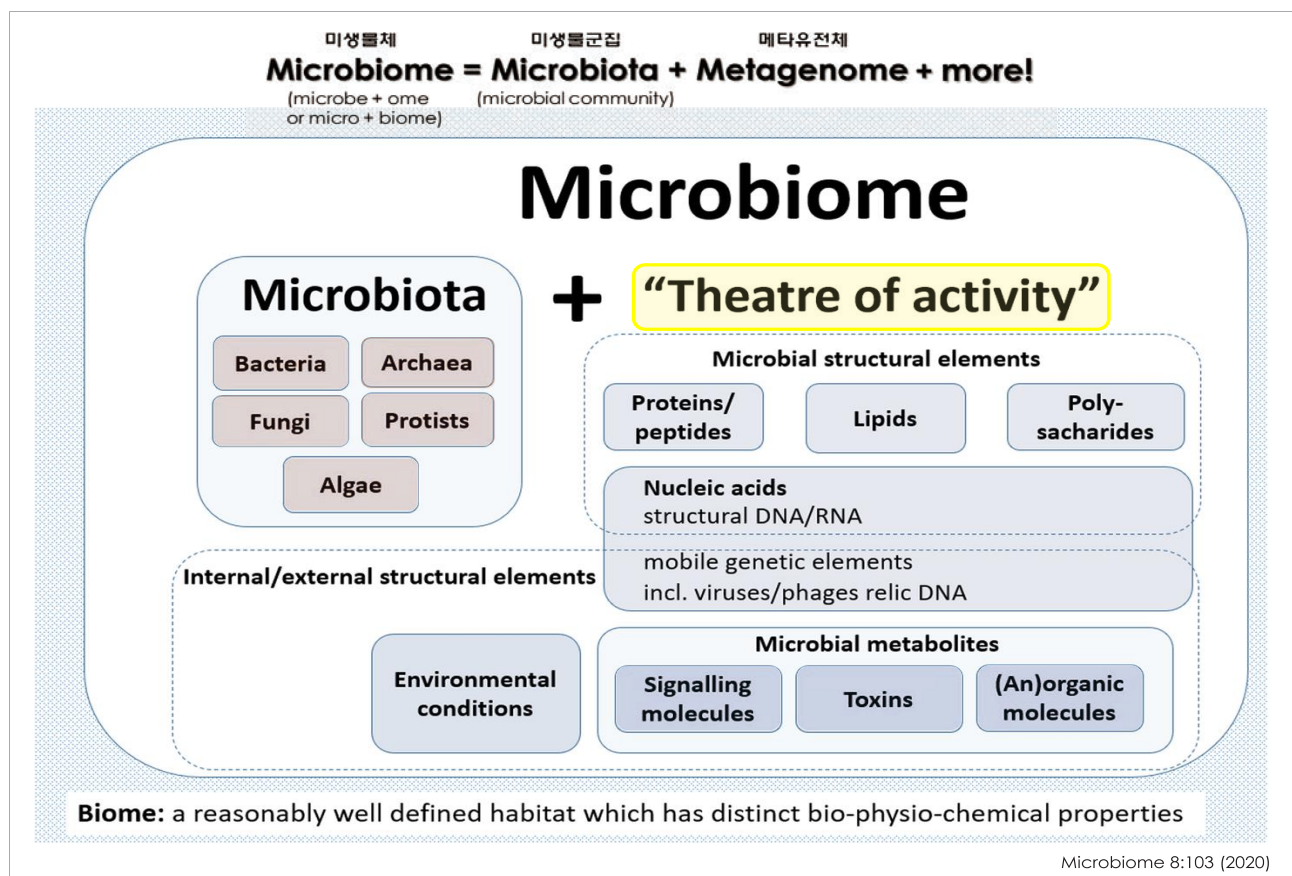
# 진단 적용 가능한 마이크로바이옴 바이오마커 개발

## Microbiome biomarkers for clinical diagnosis and prognosis of gastrointestinal cancers

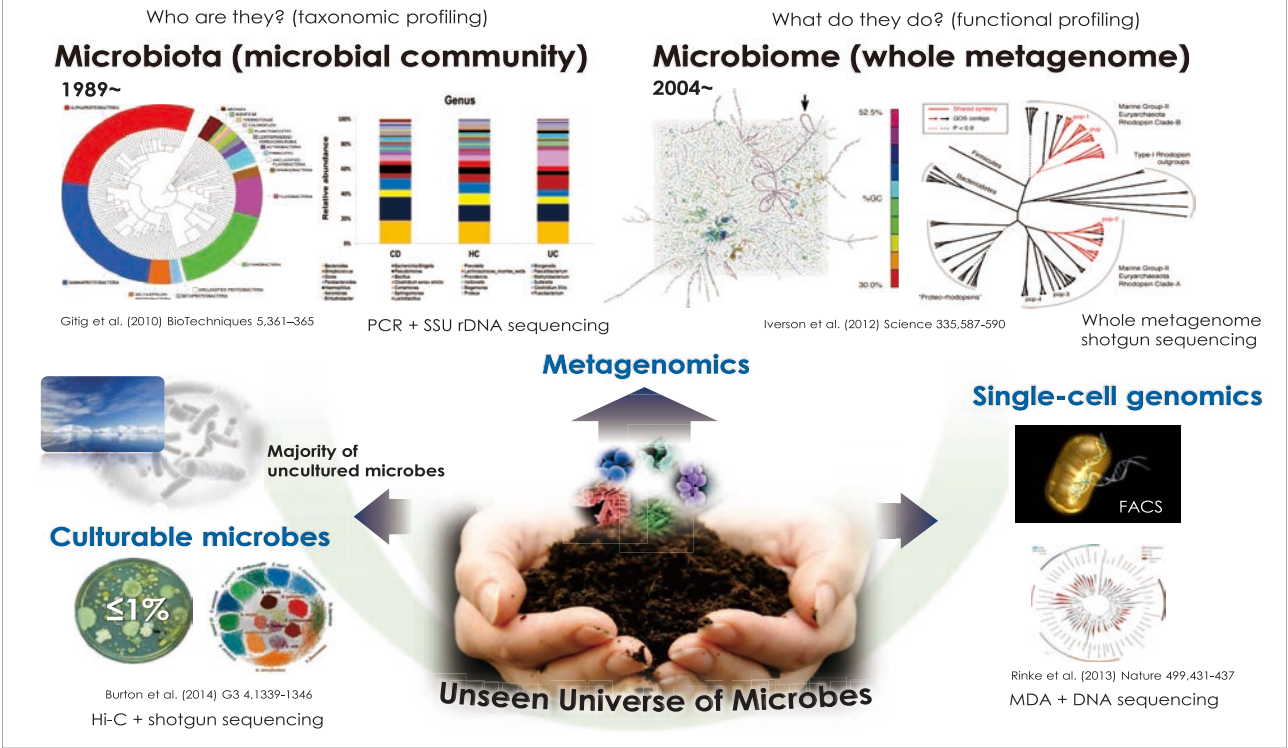
2024 병원기반 인간 마이크로바이옴  
연구개발 사업 심포지엄

Jihyun F. Kim

Yonsei University



Overview of microbiome studies



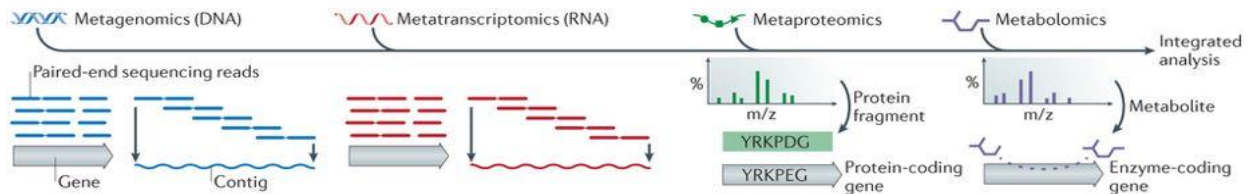
Pros and cons of metagenomic analyses

Method	Pros	Cons
Marker gene-based microbiota analysis	<ul style="list-style-type: none"><li>•Quick, simple and inexpensive sample preparation</li><li>•Correlates well with genomic content</li><li>•Amenable to low-biomass and highly host contaminated samples</li><li>•Large existing public data sets for comparison</li><li>•Many well developed tools available</li></ul>	<ul style="list-style-type: none"><li>• No live, dead or active discrimination</li><li>• Subject to amplification biases</li><li>• Choice of primers and variable region magnifies biases</li><li>• Requires a priori knowledge of microbial community</li><li>• Resolution typically limited to genus level at best</li><li>• Appropriate negative controls required</li><li>• Functional information is limited</li></ul>
Whole metagenome (including metagenome-assembled genomes) analysis	<ul style="list-style-type: none"><li>• Can directly infer the relative abundance of microbial functional genes; microbial taxonomic and phylogenetic identity to species and strains level is attainable for known organisms</li><li>• Does not assume knowledge of microbial community (that is, captures phages, viruses, plasmids, microbial eukaryotes, etc.)</li><li>• No PCR-related biases</li><li>• Can allow assembly of population-averaged microbial genomes</li><li>• Can be mined for novel gene families</li></ul>	<ul style="list-style-type: none"><li>• Relatively expensive, laborious and complex sample preparation and analysis</li><li>• Contamination from host-derived DNA and organelles may obscure microbial signatures</li><li>• Viruses and plasmids are not typically well annotated by default pipelines</li><li>• Deep sequencing depths are typically required relative to other methods</li><li>• No live, dead or active discrimination</li><li>• Population-averaged microbial genomes tend to be inaccurate owing to assembly artefacts</li></ul>

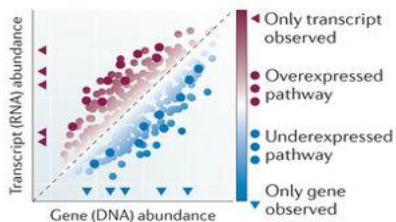
Knight et al. Nature Reviews Microbiology (2018)

## Integrating meta-omics data for deeper biological insights

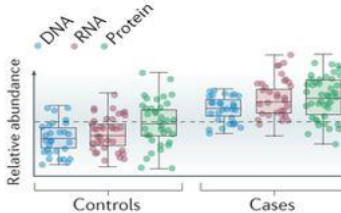
### a Multi-omics data types



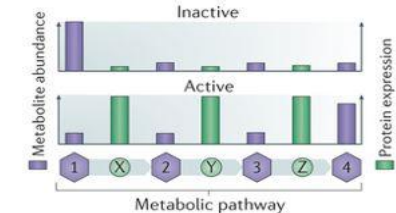
### b Normalization



### c Strengthening hypotheses

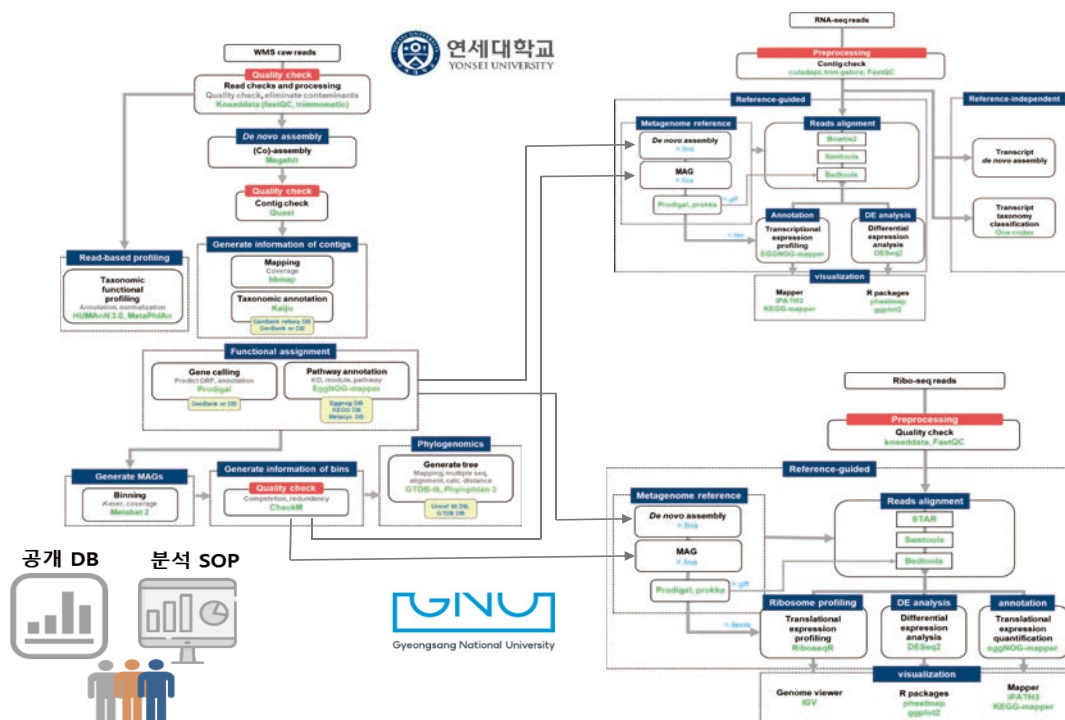


### d Descriptive modelling



Franzosa et al. (2015)  
Nature Reviews | Microbiology

## NGS-based meta-omics data analyses SOP





ABOUT US   PROTOCOLS   GENOME CATALOG   GENE CATALOG   BLAST   SAMPLES

MetaGenomics SOP

- I. Sample preparation-Sequencing
- II. Analysis
  - Installation
  - Preprocessing of the sequencing reads
  - Read-based profiling
  - De novo assembly
  - Taxonomic annotation of contigs
  - Functional annotation of contigs
  - Binning
  - Phylogenomic tree construction of the MAG

MetaTranscriptomics SOP

- I. Sample preparation-Sequencing
- II. Analysis
  - Installation
  - Preprocessing of the sequencing reads
  - Read alignment
  - Transcriptional expression analysis
  - Differential expression analysis
  - Reference-independent analysis
  - Transcript de novo assembly
  - Transcript taxonomy classification
  - Read-based profiling

MetaProteomics SOP

- I. Sample preparation-Sequencing
- II. Analysis
  - Installation
  - Preprocessing of the sequencing reads
  - Read alignment
  - Transcriptional expression analysis
  - Differential expression analysis
  - Reference-independent analysis
  - Small protein prediction
  - Translational quantification
  - Differential expression analysis
  - Read-based profiling

**PROTOCOLS Page**

**MetaGenomics SOP**

I. Sample preparation-Sequencing

II. Analysis

- Installation
- Preprocessing of the sequencing reads
- Read-based profiling
- De novo assembly
- Taxonomic annotation of contigs
- Functional annotation of contigs
- Binning
- Phylogenomic tree construction of the MAG

**MetaTranscriptomics SOP**

I. Sample preparation-Sequencing

II. Analysis

- Installation
- Preprocessing of the sequencing reads
- Read alignment
- Transcriptional expression analysis
- Differential expression analysis
- Reference-independent analysis
- Transcript de novo assembly
- Transcript taxonomy classification
- Read-based profiling

**MetaProteomics SOP**

I. Sample preparation-Sequencing

II. Analysis

- Installation
- Preprocessing of the sequencing reads
- Read alignment
- Transcriptional expression analysis
- Differential expression analysis
- Reference-independent analysis
- Small protein prediction
- Translational quantification
- Differential expression analysis
- Read-based profiling

**Sequencing**

**Biological samples**

- Fecal sample 1 g 당 500ul RNAlater (Ambion)를 처리하여 -80℃에 보관한다.

**RNA preparation**

**Reagents**

- RLT buffer (Qiagen), β-mercaptoethanol, Superscript III (Thermo Fisher Scientific), proteinase K (20mg/mL), 100% ethanol, 70% ethanol, 3M sodium acetate, phenol/chloroform/isoamyl alcohol 25:24:1 (pH 5.2), RNase-free water
- Kit: RNeasy mini plus kit (Qiagen), TURBO DNA-free™ (Ambion)

**Equipment**

FastPrep-24™ 50(MP), pipette, aerosol barrier pipette tips, microcentrifuge, 1mm zirconia/silica beads

Optional: Qubit fluorometer (Thermo Fisher Scientific), Fragment analyzer

**Stock solutions**

Stock solution A	volume
RLT buffer(Qiagen)	975 μL
β-mercaptoethanol	10 μL
Superscript-III, 20U/μL	15 μL
Total	1 mL

**RNA preparation**

- 12 screw-top tube에 fecal sample과 bead를 넣고 반응액을 처리한다.

Each tube	volume
Fecal sample	150mg
1.0mm zirconia/silica beads	~20
Stock solution A	600 μL

License & Citation

Contact

Version 1.0 Released 2023-09

**Metagenome analysis pipeline**

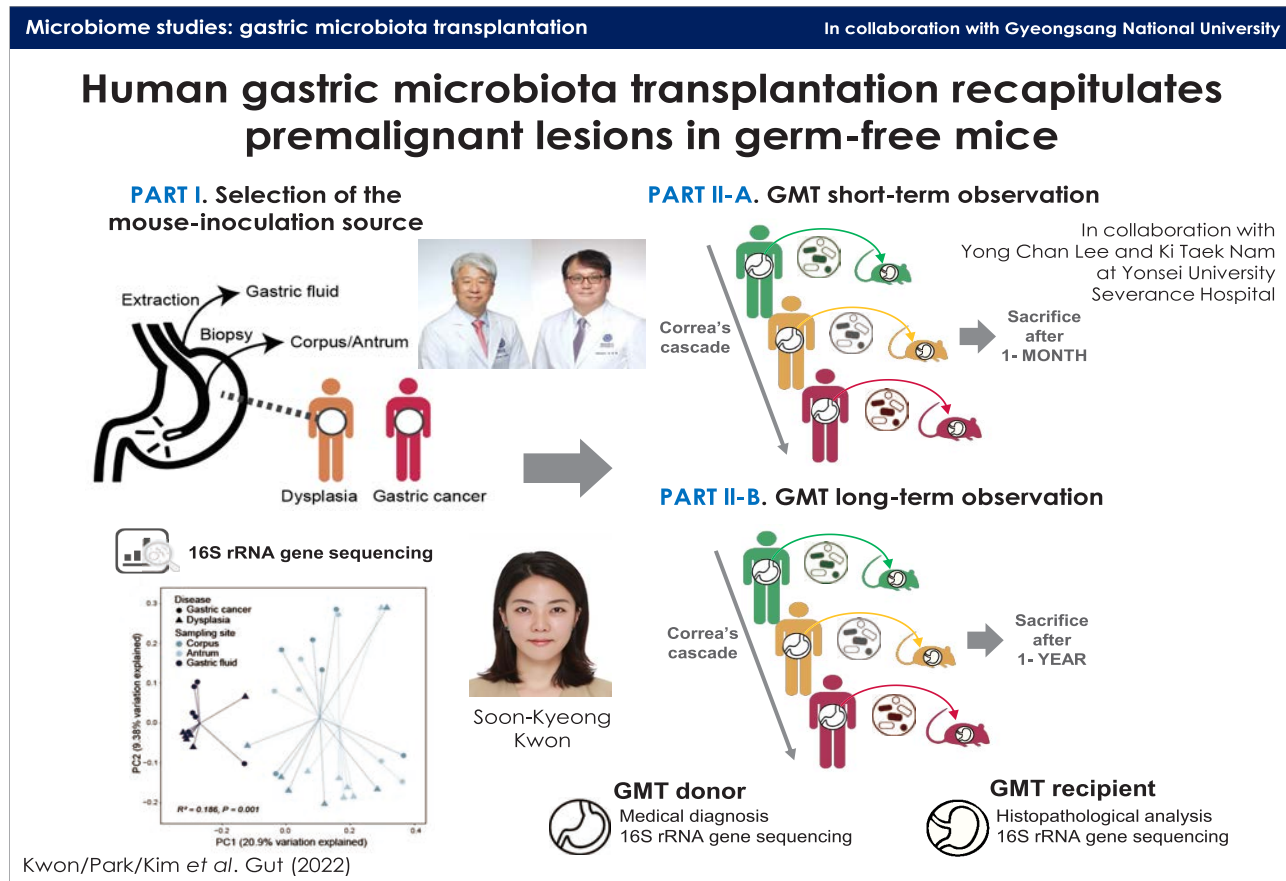
**metaFun: A genomic and metagenomic analysis pipeline for fast and unified functional searches**

Scalable and reliable analysis pipeline

Entirely containerized for painless operation

The diagram illustrates the metaFun workflow. It starts with 'Short paired-read metagenomic data' (RAWREAD\_QC) and 'Metagenome FASTQ' (Metadatas). These are processed through 'WMS\_FUNCTION' (Statistical analysis) and 'WMS\_TAXONOMY' (Statistical analysis) using 'HUMAN' and 'GTDB-based Kraken2' databases. The workflow then moves to 'GENOME ASSEMBLY' (Short-read FASTQ, Long-read FASTQ) and 'BIN\_ASSESSMENT' (Filtered genome, Metadata). The final step is 'COMPARATIVE\_ANNOTATION' (dbCAN, eggNOG, CARD, VFDB, KOfam) using 'MetaFun' and 'MetaFun' tools. The output is 'Genome' and 'Metadatas'.

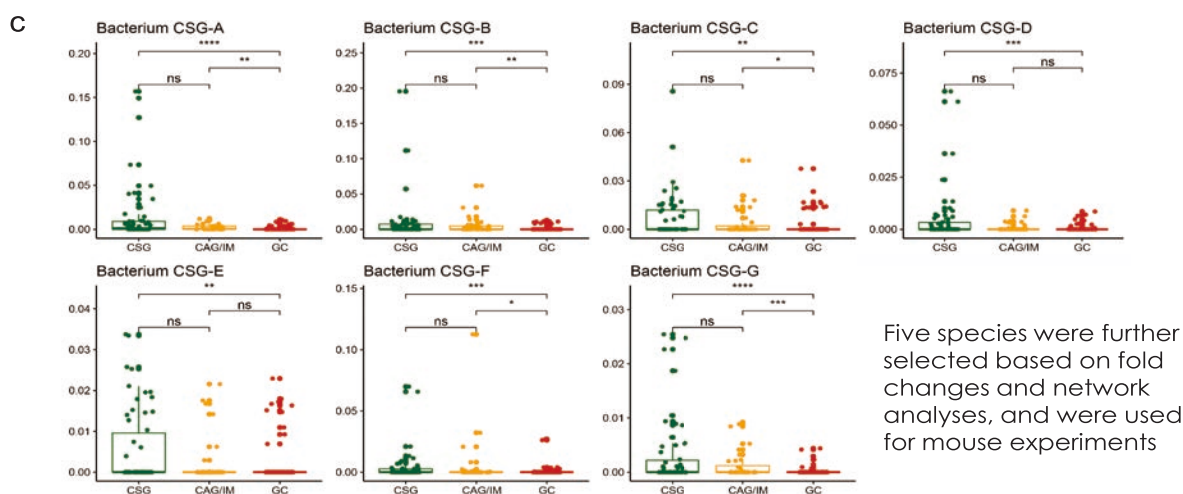
The screenshot shows the metaFun GitHub repository. It includes a 'Workflow Execution' section with a table of workflow runs and a 'Documentation' section with a 'Workflow Execution' section. A QR code is visible in the bottom right corner.



## Results

### Relative abundances of the species enriched in CSG between CSG, CAG/IM, and GC

Seven species enriched in CSG show differential abundances between CSG, CAG/IM, and GC

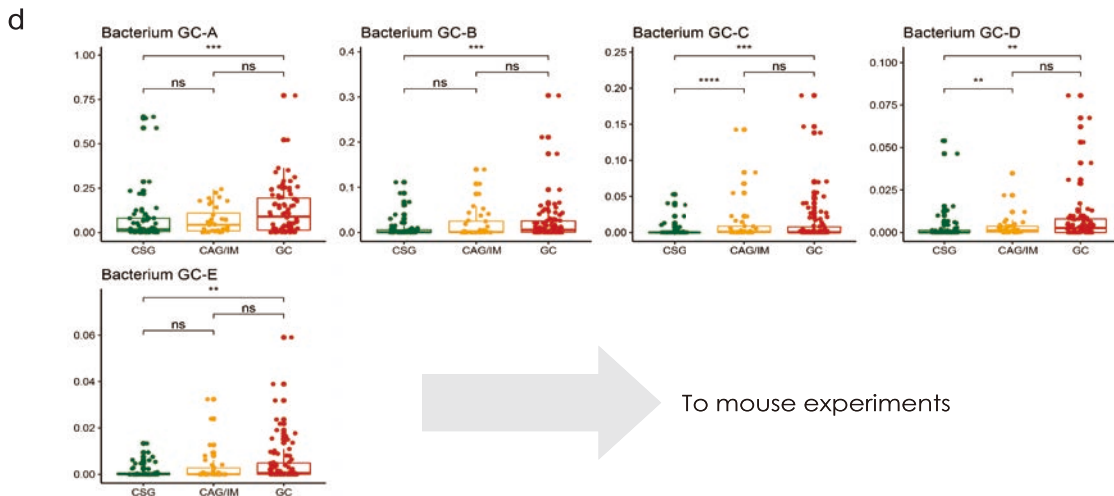


**Figure 1. Identification of bacterial species associated with disease states.** c, Relative abundance of seven selected bacterial species enriched in CSG. The species were shown in order of high LDA scores.

## Results

### Relative abundances of the species enriched in GC between CSG, CAG/IM, and GC

Five species enriched in GC show differential abundances between CSG, CAG/IM, and GC



**Figure 1. Identification of bacterial species associated with disease states.** d, Relative abundance of five selected bacterial species enriched in GC. The species were shown in order of high LDA scores.

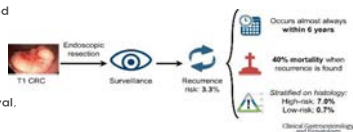
## Microbiome studies: prognosis of colorectal cancer

In collaboration with Seoul National University

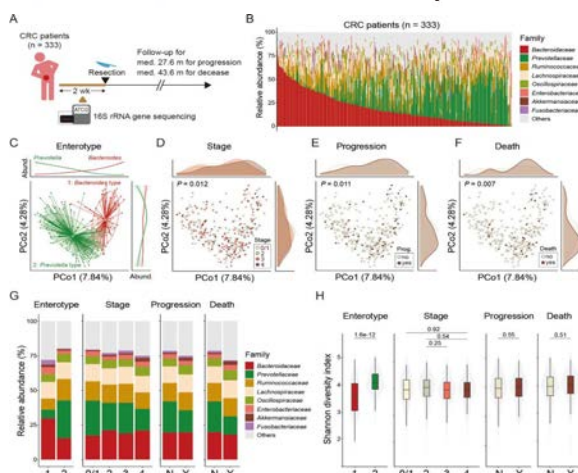
### Bacterial biomarkers in preoperative stool predict the clinical outcome of colorectal cancer

The third most common malignancy, and the second most common cause of cancer-related death worldwide; its incidence is steadily increasing in "Westernizing" countries

24% of patients with stage I-III CRC relapse within 5 years after tumor removal, and more than half of the cases occur within 2 years

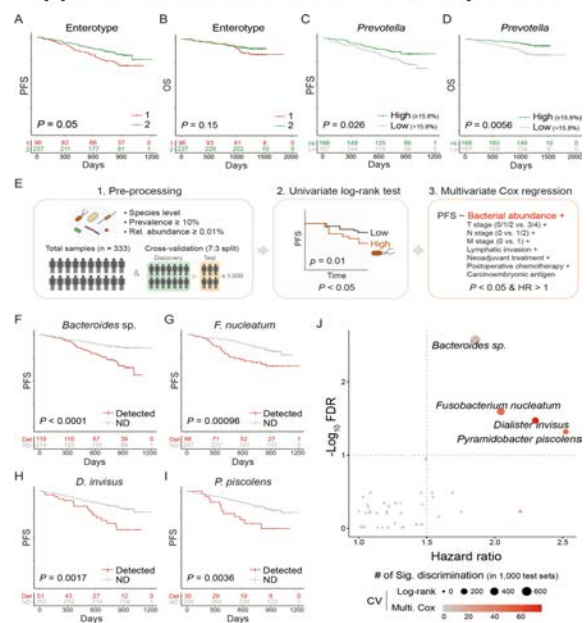


#### Preoperative gut microbiota of CRC patients is associated with the state of the patients



Huh *et al.* Microbiome (2022)

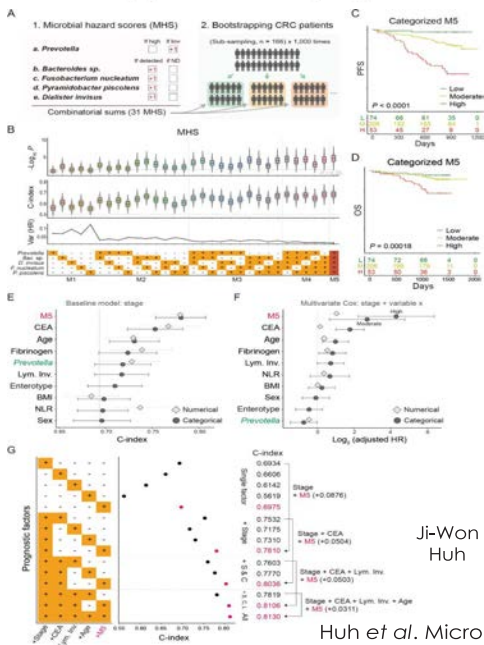
#### Enterotypical *Prevotella* and four opportunists indicate a state of CRC patients



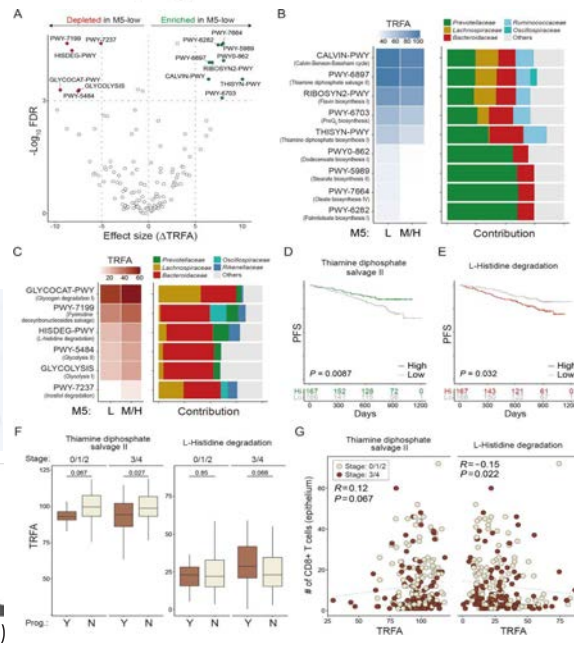


## Bacterial biomarkers in preoperative stool predict the clinical outcome of colorectal cancer

A gut microbiota-derived hazard score accurately predicts CRC prognosis



Microbial metabolism correlates with different prognoses



Huh et al. Microbiome (2022)

# Thanks to..



**Yonsei University**

Jaekyung Yoon  
Hyeongwon Lee  
Laeguen Jang  
Seolah Kim  
Yun Young Kim  
Dong Hyun Lee

**Prof. Yong Chan Lee**  
**Prof. Ki Taek Nam**



**Seoul National University**

**Prof. Ji Won Park**

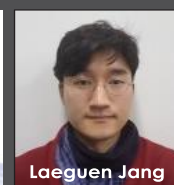
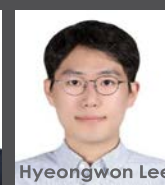
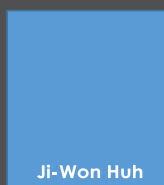


**Chungnam National University**

**Prof. Ji-Won Huh**

**Gyeongsang National University**

**Prof. Soon-Kyeong Kwon**





## **Session 2.**

# **Microbiome axis in health & disease**



1. Understanding the host-microbiota interaction using an EAE animal model
2. The gut-kidney axis in CKD: role of the microbiome
3. Gut-lung axis: from epidemiology to therapeutic application
4. Gut-liver axis in health & disease







## 2부 좌장



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2014 ~ 현재	한림대학교 생명과학과 교수

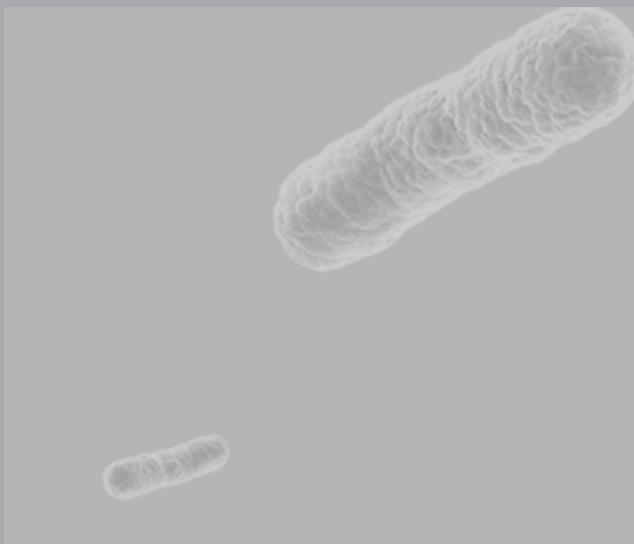


01

# Understanding the host– microbiota interaction using an EAE animal model



이윤경  
(순천향대학교)





## 2부 연자



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2009	University of Alabama at Birmingham, USA. Ph.D.

### 🔍 경력사항

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2015 ~ 현재	교수, 순천향대학교 순천향의생명연구원 의생명융합학과



# Understanding the host–microbiota interaction using an EAE animal model

The mammalian gastrointestinal tract contains a diverse, but well-balanced population of beneficial and potentially pathogenic microbes to maintain the complex gut ecosystem within the host. Recent understanding from numerous studies indicates that our health is highly dependent on the composition of intestinal microbiota. It has been suggested that alterations in the community and composition of microbiota contribute to the pathogenesis of extra-intestinal disease. Multiple Sclerosis (MS) is a devastating autoimmune disease leading to progressive deterioration of neurological function. Despite significant clinical and scientific efforts expended over decades, findings have not been sufficient to elucidate how the dysregulated holobiome in the gut correlates with the development and/or severity of MS. Here, we investigate the role of microbiota during the induction of experimental autoimmune encephalomyelitis (EAE), an animal model for MS. We observed that EAE mice harboring one of the human isolates show attenuated EAE symptoms and display an altered gut ecosystem. Furthermore, we reveal that gut microbiota-derived metabolites from the changed gut ecosystem by this human isolates reduce host susceptibility to central nervous system (CNS) autoimmunity using integrated metagenomics and metabolomics analysis.

SCH순천향대학교  
SOON CHUN HYANG  
UNIVERSITY

SCHSOONCHUNHYANG  
INSTITUTE OF MEDI-BIO SCIENCE

국립보건원  
12-03-2024

# Understanding the host-microbiota interaction using an EAE animal model

**Yun Kyung Lee**  
Department of Integrated Biomedical Science  
SoonChunHyang Institute of Medi-Bioscience (SIMS)  
SoonChunHyang University

## Pathogens vary greatly in size and lifestyle

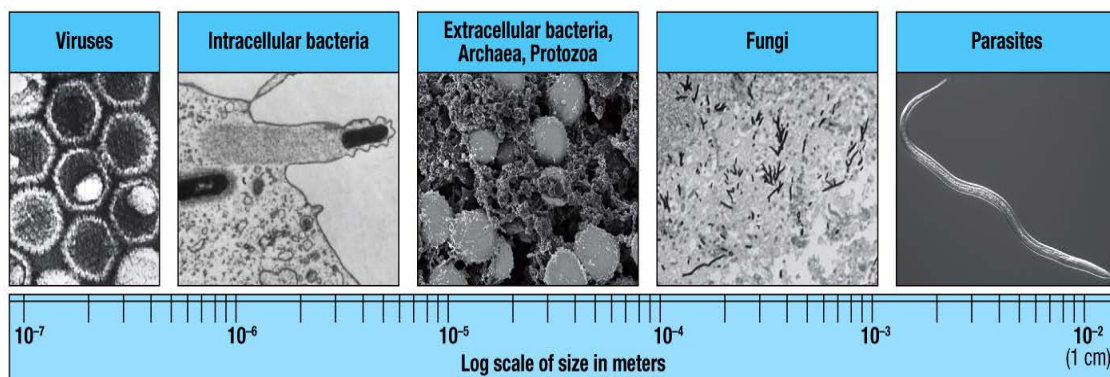


Figure 1.4 Janeway's Immunobiology, 9th ed. (© Garland Science 2017)

## The cells of the immune system

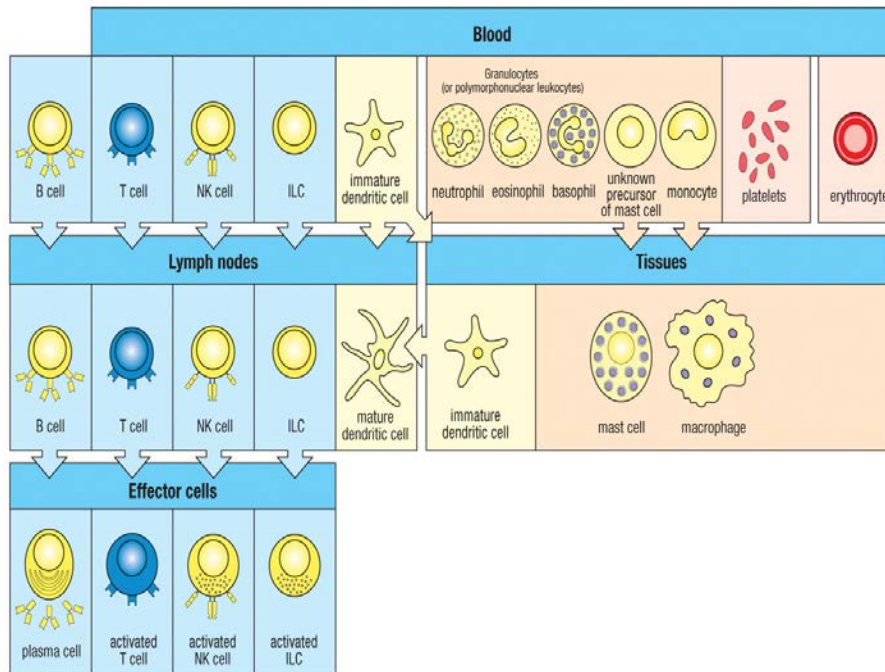
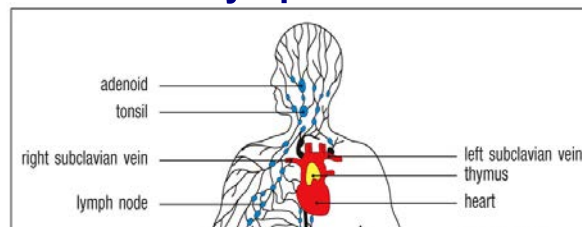


Figure 1.3 (part 2 of 2) Janeway's Immunobiology, 9th ed. (© Garland Science 2017)

## Immune System

### The distribution of lymphoid tissues in the body



**The core of the immune response is the ability to distinguish between “self” and “non-self”**

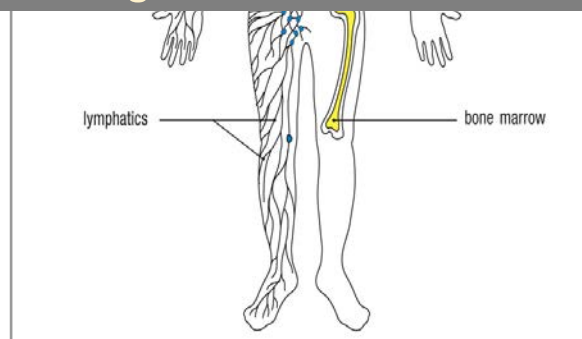
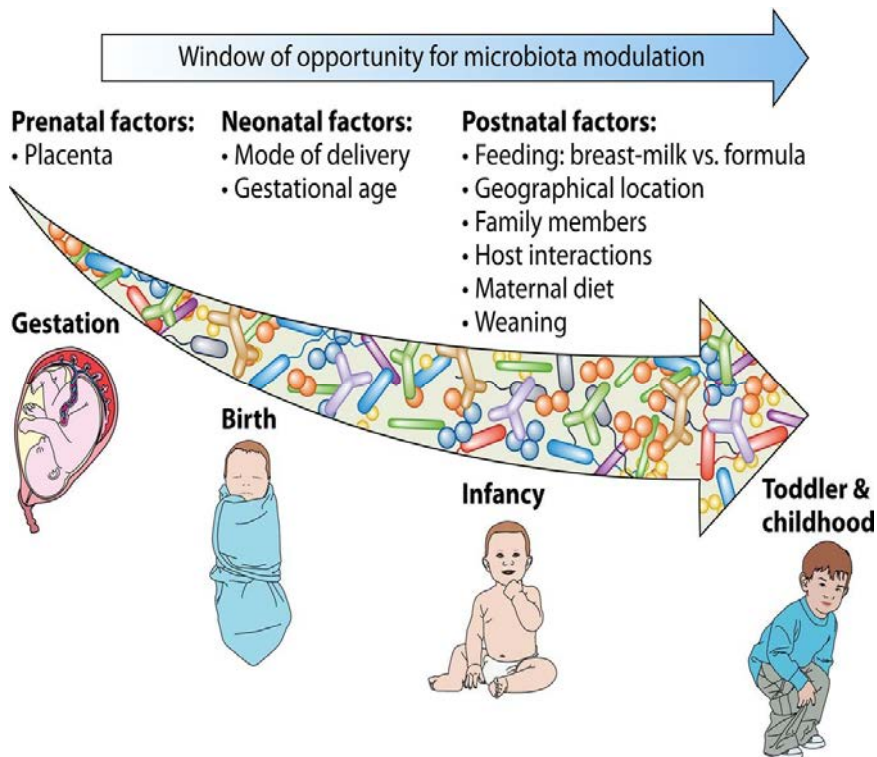


Figure 1.18 Janeway's Immunobiology, 9th ed. (© Garland Science 2017)

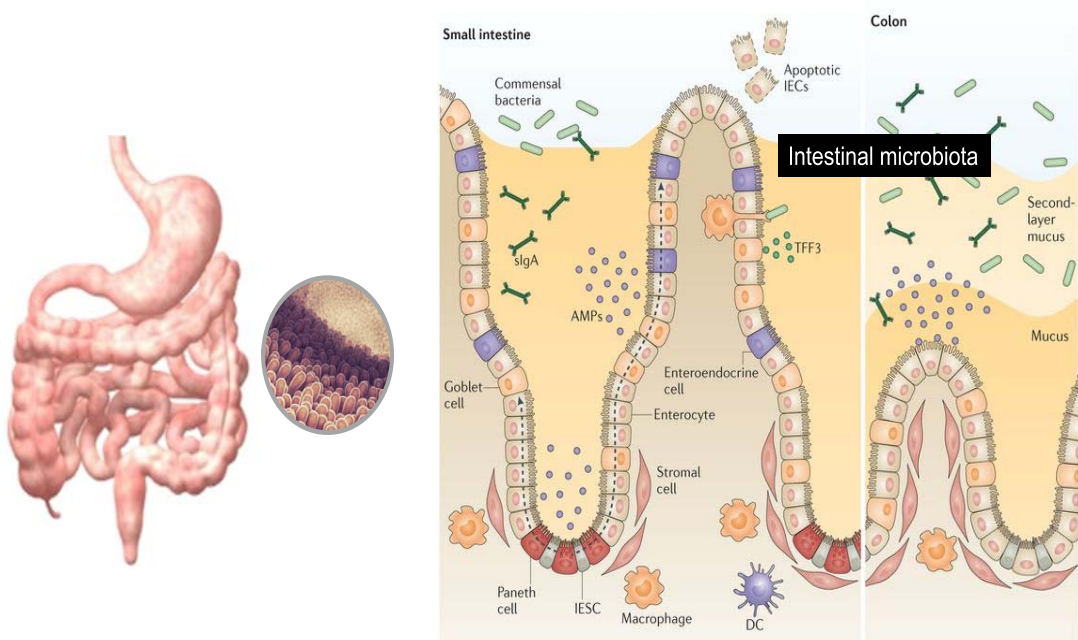


## The Microbial Colonizers of the human gut



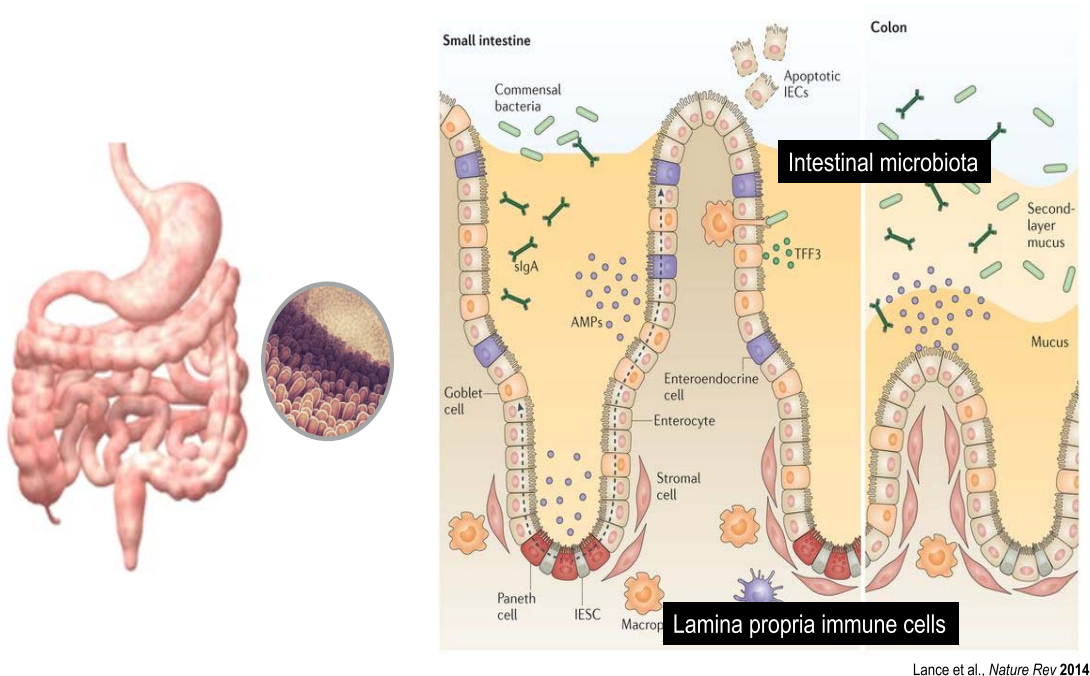
Milani C et al, *Microbio and Mol Biol Review*, 2017

## The Microbial Colonizers of the human gut



Lance et al., *Nature Rev* 2014

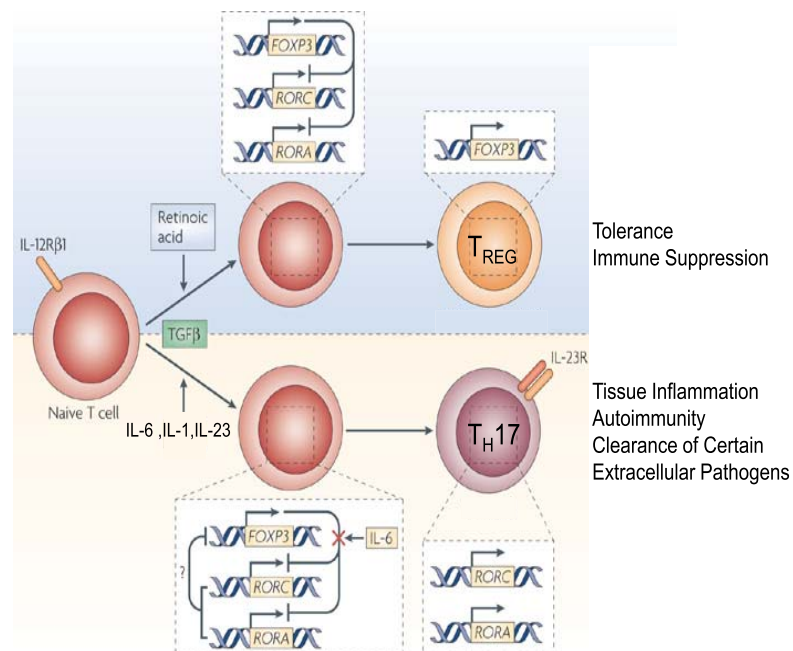
## Immune System in Microbial World?



Lance et al., *Nature Rev* 2014

## Mucosal immunity: Treg and Th17

The developmental link between the Treg and Th17 cell lineages



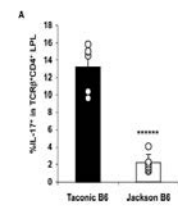
Nature Reviews | Immunology

Weaver CT, *Nat Rev Immunol* 2009

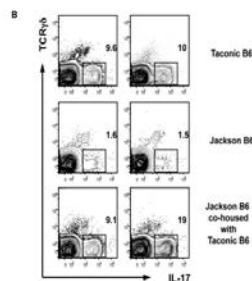
## Treg and Th17 by microbiota

### Specific Microbiota Direct the Differentiation of IL-17-Producing T-Helper Cells in the Mucosa of the Small Intestine

Ivaylo I. Ivanov,<sup>1</sup> Rosa de Llanos Frutos,<sup>5</sup> Nicolas Manel,<sup>1</sup> Keiji Yoshinaga,<sup>3,7</sup> Daniel B. Rifkin,<sup>3,4</sup> R. Balfour Sartor,<sup>6</sup> B. Brett Finlay,<sup>5</sup> and Dan R. Littman<sup>1,2,4</sup>

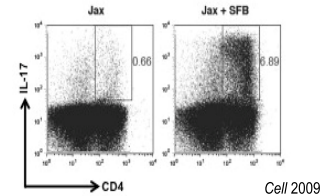


Cell host microbe 2008



### Induction of Intestinal Th17 Cells by Segmented Filamentous Bacteria

Ivaylo I. Ivanov,<sup>1,10</sup> Koji Atarashi,<sup>3,10</sup> Nicolas Manel,<sup>1,11</sup> Eoin L. Brodie,<sup>4,11</sup> Tatsuchihiro Shima,<sup>7,11</sup> Ulas Karaoz,<sup>4</sup> Dongguang Wei,<sup>5</sup> Katherine C. Goldfarb,<sup>4</sup> Clark A. Santee,<sup>4</sup> Susan V. Lynch,<sup>6</sup> Takeshi Tanoue,<sup>3</sup> Akemi Imaoka,<sup>7</sup> Kikui Itoh,<sup>8</sup> Kiyoshi Takeda,<sup>3</sup> Yoshinori Umesaki,<sup>7</sup> Kenya Honda,<sup>3,9,1</sup> and Dan R. Littman<sup>1,2,4</sup>



Cell 2009

### Segmented Filamentous Bacteria (SFB)

- Gram-positive anaerobic bacteria that colonizes the ileal epithelium
- SFB specifically induce the differentiation of effector T<sub>H</sub>17 cells in the lamina propria of small intestine



Ivanov II et al. *Mucosal Immunol* 2010

## Treg and Th17 by microbiota

### Induction of Colonic Regulatory T Cells by Indigenous *Clostridium* Species

Koji Atarashi,<sup>1\*</sup> Takeshi Tanoue,<sup>1\*</sup> Tatsuchihiro Shima,<sup>2</sup> Akemi Imaoka,<sup>2</sup> Tomomi Kuwahara,<sup>3</sup> Yoshihika Momose,<sup>4</sup> Genhong Cheng,<sup>6</sup> Sho Yamasaki,<sup>7</sup> Takashi Saito,<sup>7</sup> Yusuke Ohba,<sup>9</sup> Tadatsugu Taniguchi,<sup>1</sup> Kiyoshi Takeda,<sup>5</sup> Shohei Hori,<sup>8</sup> Ivaylo I. Ivanov,<sup>10</sup> Yoshinori Umesaki,<sup>2</sup> Kikui Itoh,<sup>4</sup> Kenya Honda<sup>1,11†</sup>

Science 2010

### *Clostridia* cluster-17 strains

:enriched in the chloroform-resistant fraction

### Inducible Foxp3<sup>+</sup> regulatory T-cell development by a commensal bacterium of the intestinal microbiota

PNAS 2010

June L. Round and Sarkis K. Mazmanian<sup>1</sup>

### A microbial symbiosis factor prevents intestinal inflammatory disease

#### *B. fragilis*

:The most commonly isolated indigenous gastrointestinal microbiota

Nature 2008

Sarkis K. Mazmanian<sup>1\*</sup>, June L. Round<sup>1\*</sup> & Dennis L. Kasper<sup>2,3</sup>

### The microbial metabolites, Short chain fatty acid (butyrate, propionate) Induces the differentiation of regulatory T cells

Science 2013 Smith PM, Garrett WS

Nature 2013 Furusawa Y, Ohno H

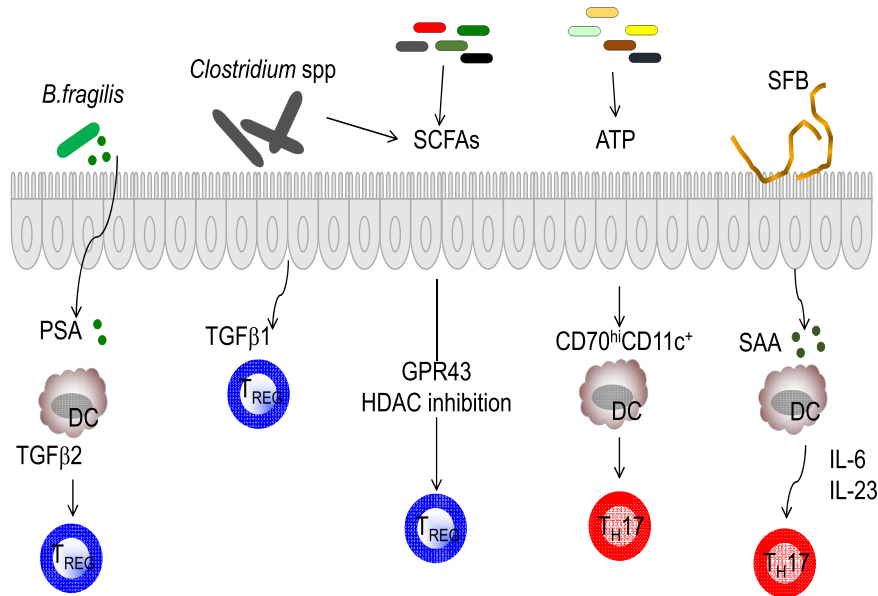
Nature 2013 Arpaia N, Rudensky AY

#### Propionate, Butyrate

:produced by commensal microorganisms during starch fermentation

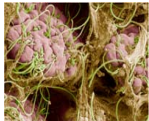


## The gut microbiota shapes CD4 T cell population



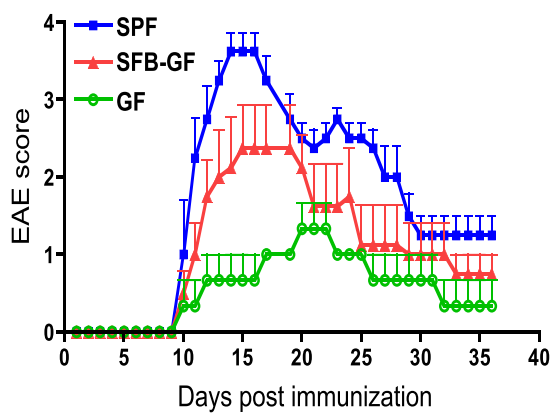
Mazmanian SK, Lee YK, *J Bacteriol & Virol* 2014

## SFB promote pro-inflammatory T cells response outside the gut during autoimmune diseases

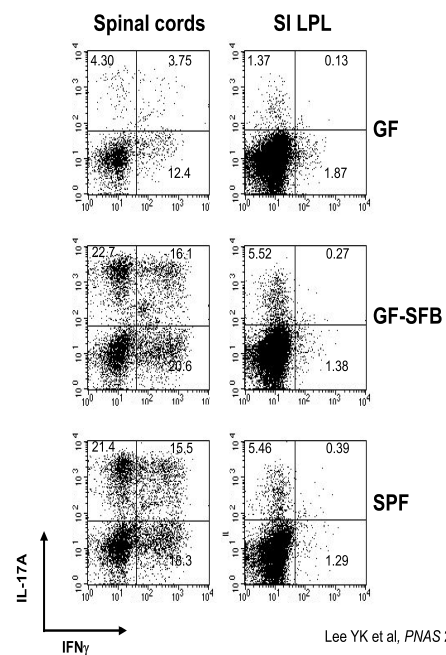


Ivanov II et al. *Mucosal Immunol* 2010

### Multiple Sclerosis (MS) Experimental Autoimmune Encephalomyelitis (EAE)



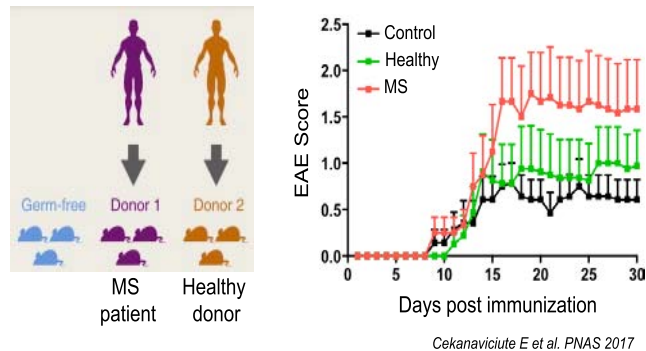
Intestinal colonization with SFB promotes EAE



Lee YK et al, *PNAS* 2011

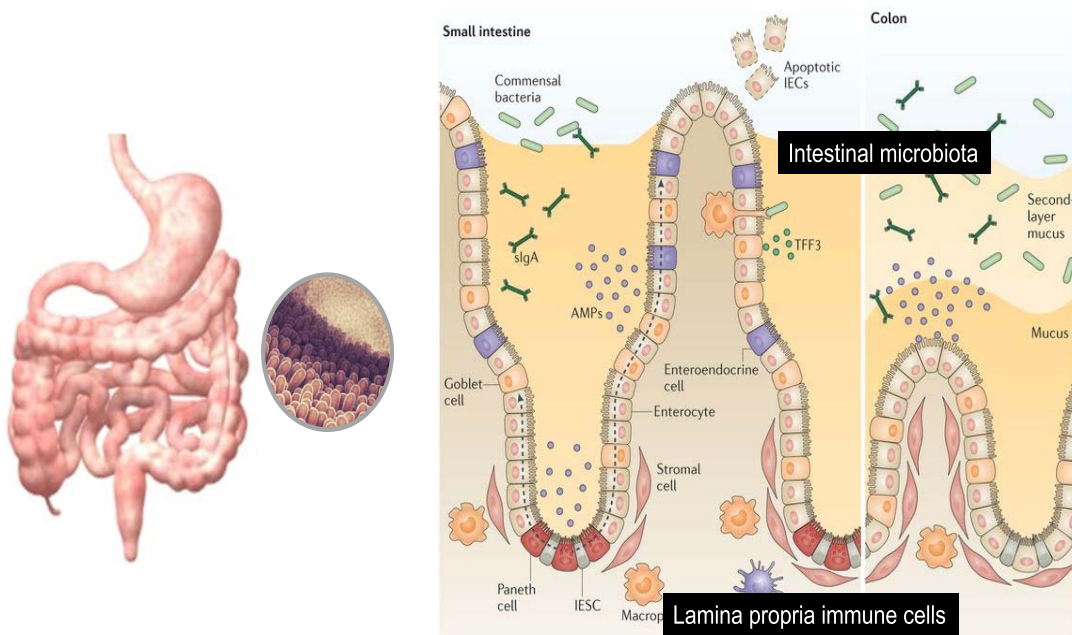
## Disease phenotype can be transmitted by human gut microbiota transplantation

-Colonization of germ-free animals with MS microbiota; humanized gut model



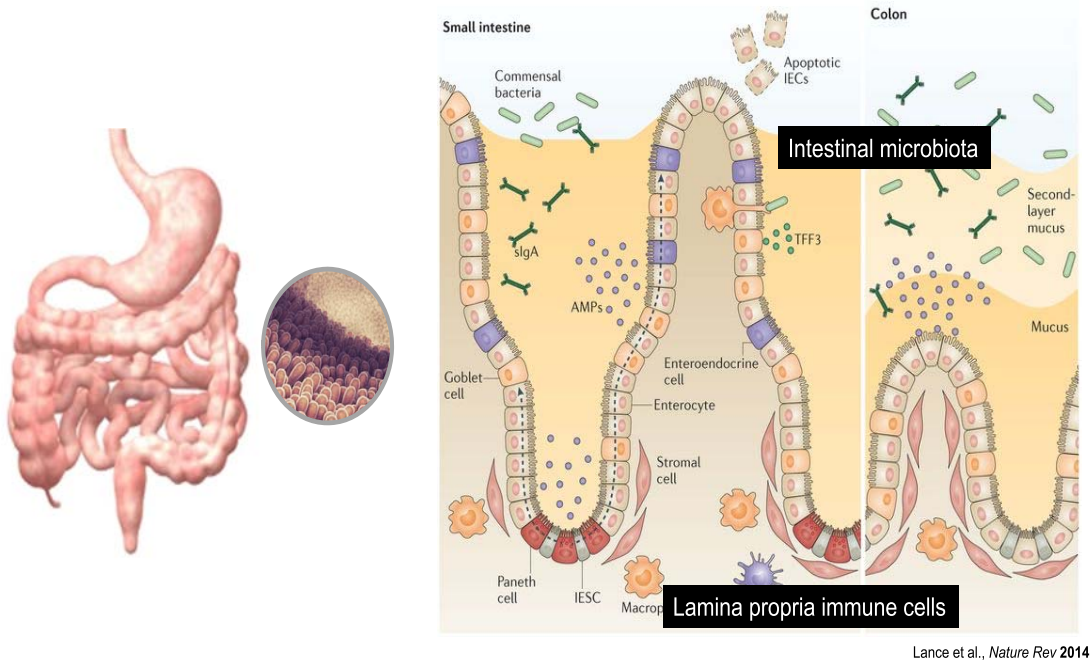
- Transplanting fecal microbiota from human donors of Multiple Sclerosis (MS)
- The capacity of their gut microbial communities to impact the development of MS

## Immune System in Microbial World?



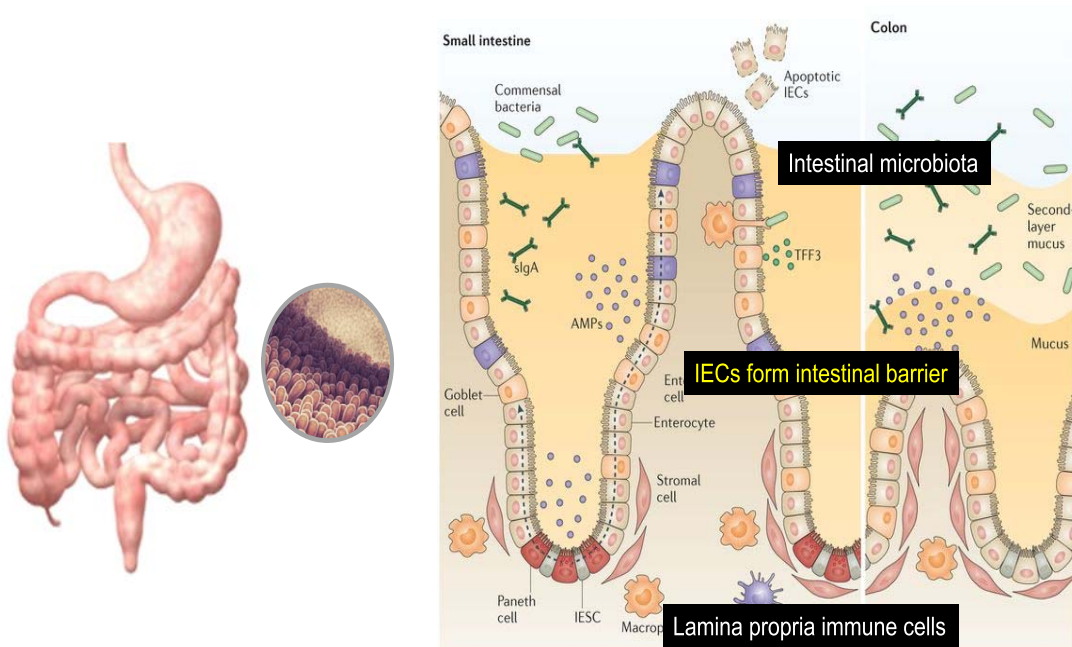
Lance et al., Nature Rev 2014

## Host-Microbiota Interaction



Lance et al., Nature Rev 2014

## Mucosal immunity: Intestinal Epithelial Cells

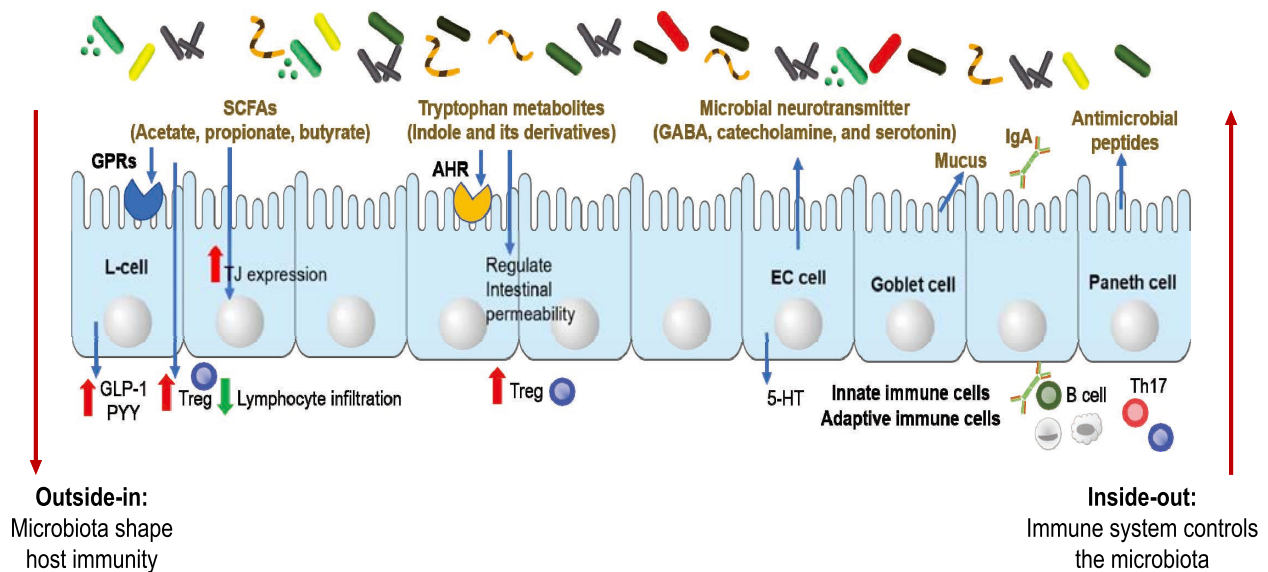


Lance et al., Nature Rev 2014

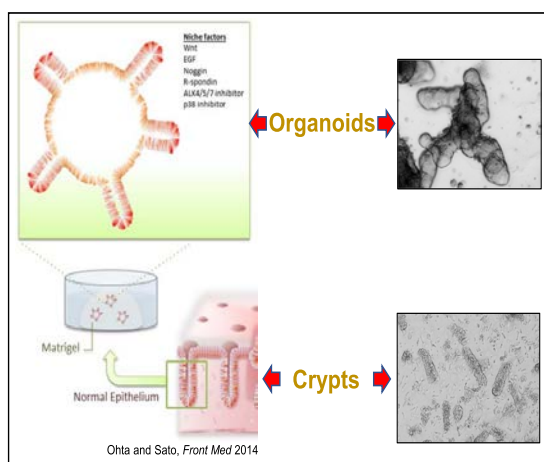
Mucosal immunity in intestine: Intestinal epithelial cells (IECs) and immune cells

## Intestinal epithelial cells as Barrier & Mediator

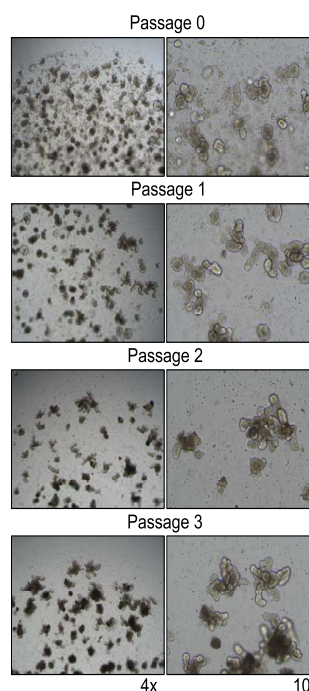
- Intestinal epithelial cells (IECs) sense the signals of intestinal environment and produce effective molecules to mediate the crosstalk between intestinal microbiota and host cells
- Cross-communication between intestinal microbiota and lamina propria immune cells
- Lamina propria immune cells sense microbial signals and produce cytokines, chemokines, immunoglobulin A (IgA)



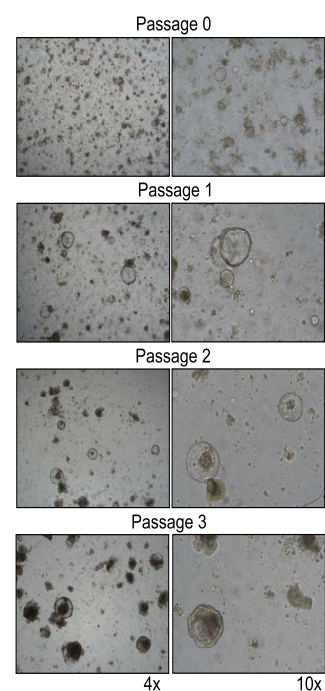
## The Intestinal organoid culture



Organoid culture-Small intestine

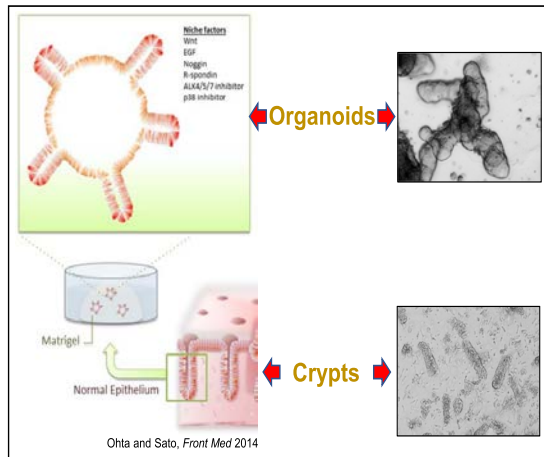


Organoid culture-Colon

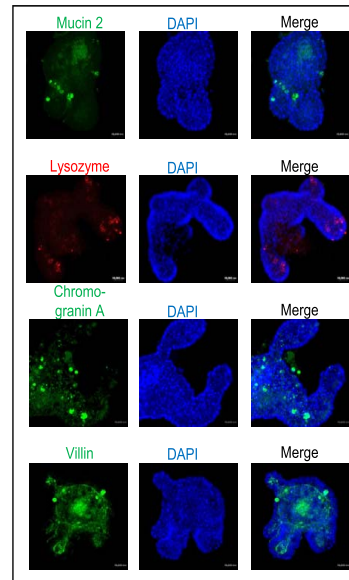
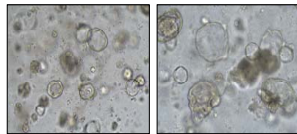




## The Intestinal organoid culture

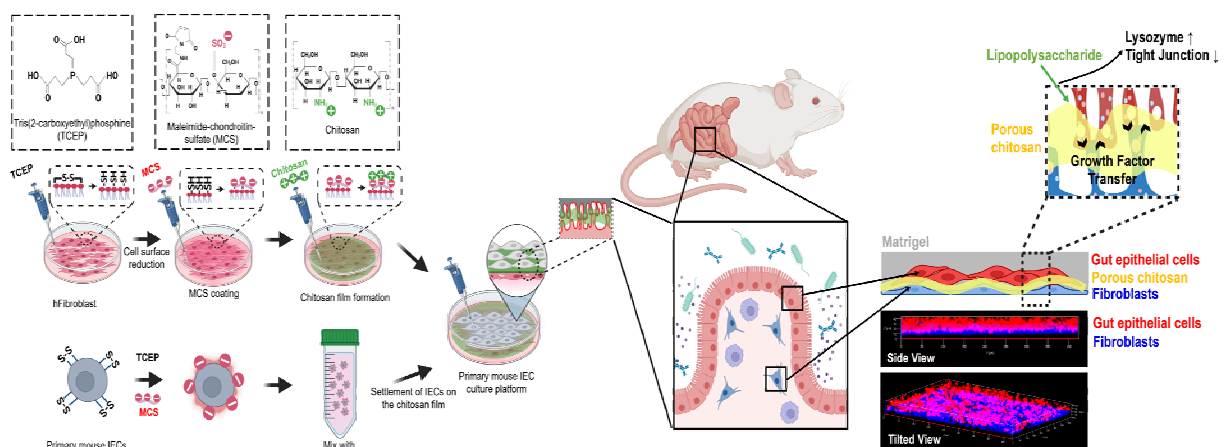


Human intestinal organoids



Muc2 : Goblet cell  
Lysozyme : Paneth cell  
ChrgA : Enteroendocrine cell  
Villin : Enterocyte

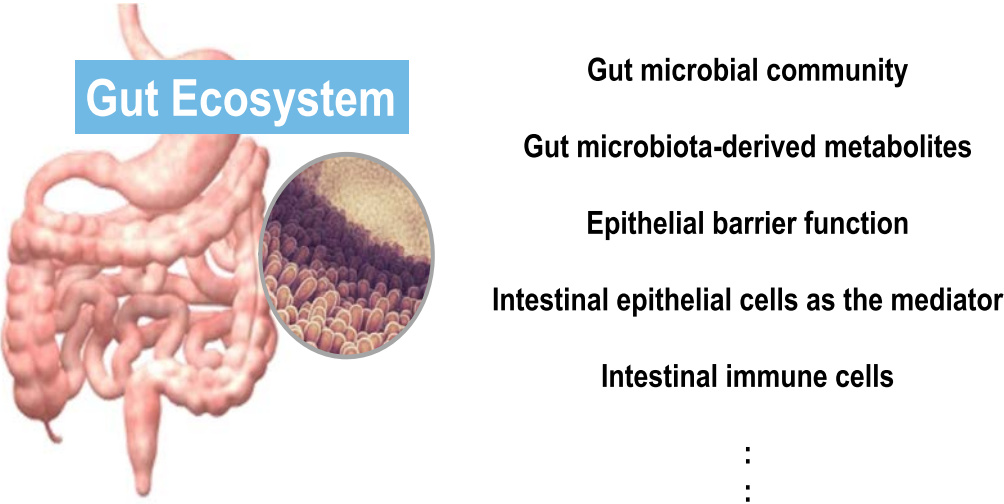
## Cell surface modification-mediated primary intestinal epithelial cell culture platforms for assessing host-microbiota interactions



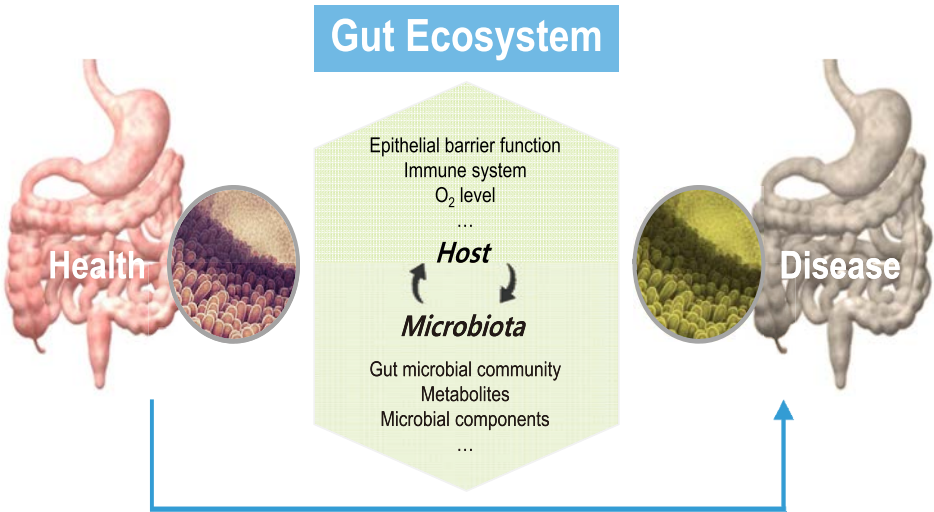
Sittipo P et al., Biomaterials Research 2024

- IEC culture systems will be extensively used to determine IEC function in response to stimuli, especially bacteria-derived molecules existing in the gastrointestinal tract

# Host-Microbiota Interaction



# Microbiota dysbiosis in diseases

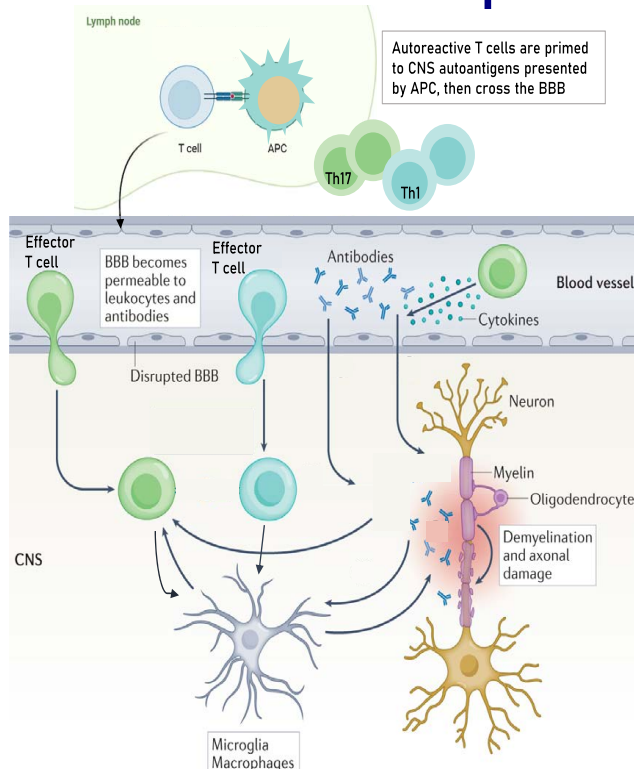


# The perturbation in gut ecosystem

## Gut microbiota dysbiosis is associated with all human diseases

- Inflammatory Bowel Diseases (IBD)-Ulcerative Colitis, Crohn's Disease
- Cancer-Colorectal cancer
- Autoimmune diseases-Multiple Sclerosis, Rheumatoid Arthritis
- Allergy-Atopic Dermatitis, Food allergy
- Metabolic diseases-Diabetes Mellitus, Atherosclerosis
- Neurodegenerative diseases-Parkinson disease, depression, Autism

## Multiple Sclerosis (MS)



An animal model of MS:  
Experimental Autoimmune Encephalomyelitis (EAE)

- MS is one of the inflammatory autoimmune diseases

- MS patients progress to physical and cognitive disability

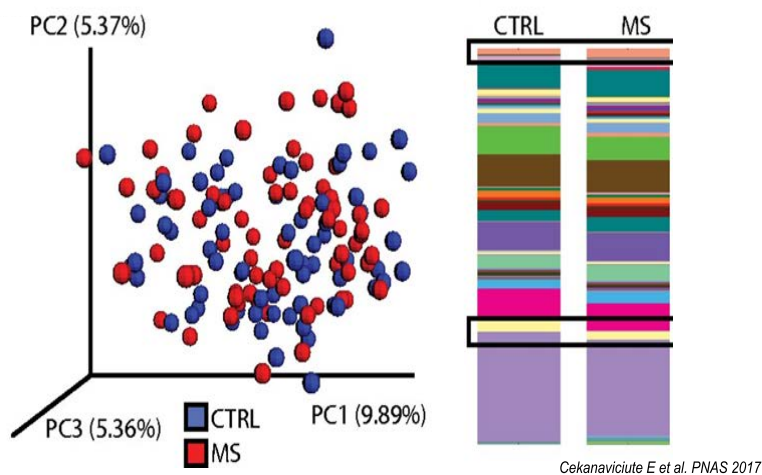
- The uncontrolled self-reactive T cells and additional cells of the immune system that infiltrate the CNS and attack the myelin sheath

Modified, Hoftberger R et al. Nat Rev Neurol 2022



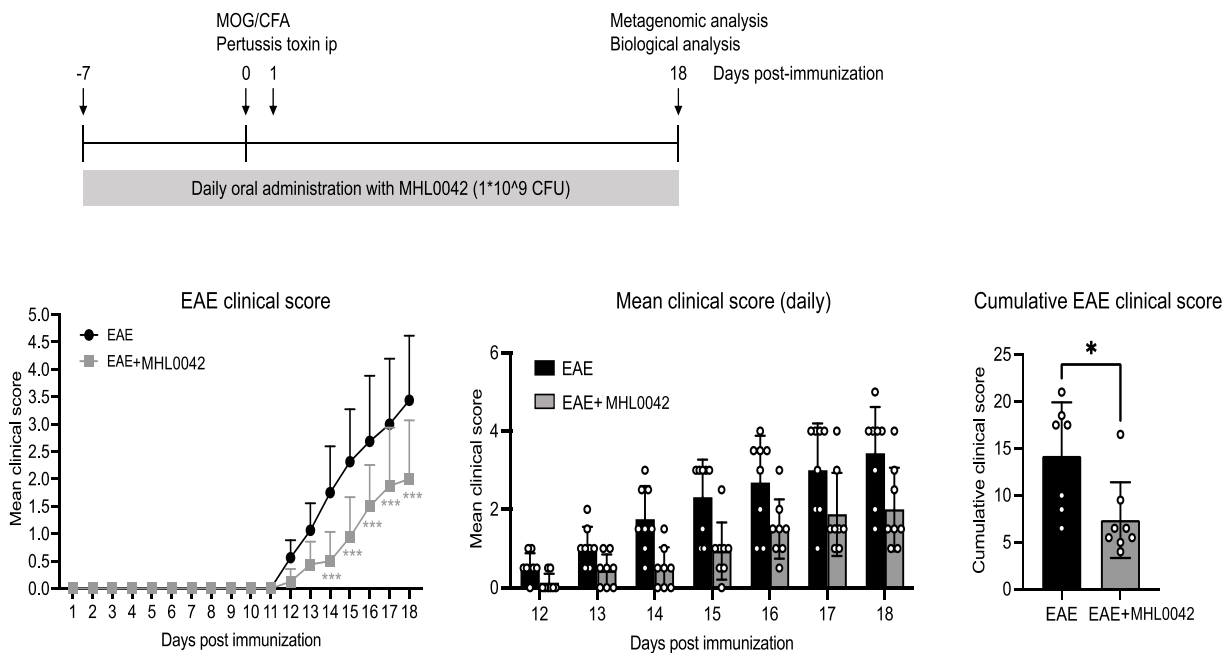
# The Multiple Sclerosis (MS) microbiome shows dysbiosis at the genus level

71 untreated relapsing-remitting MS patients  
71 healthy controls



MS-associated bacterial species reduce Tregs and increase Th1 lymphocyte differentiation *in vitro*

# Administration with MHL0042 reduces EAE severity





### Laboratory of Immune System in Microbial World

Panida Sittipo Ph.D.  
Eunike Tiffany  
Ara Oh  
Chanyoung Lee  
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NRF 2021년도 바이오의료기술개발사업  
(차세대 마이크로바이옴 기능 분석 시스템 및 활용기술)



# 02

## The gut–kidney axis in CKD: role of the microbiome



김지은  
(고려대학교구로병원)





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# The Gut-Kidney Axis in CKD: Role of the Microbiome

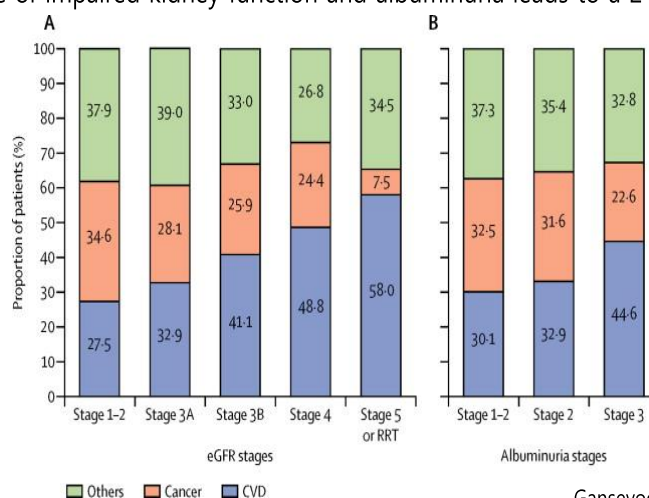
만성콩팥병에서 장내 마이크로바이옴 변화와 이에 따른 요독소 생성에 대한 병태생리를 설명하고, 만성콩팥병에서 마이크로바이옴 변화에 영향을 미치는 다양한 요인에 대해 소개함. 또한 현재까지 만성콩팥병에서 치료 전략으로써의 마이크로바이옴 연구 결과를 설명.

# The Gut-Kidney Axis in CKD: Role of the Microbiome

고려대학교 구로병원  
김지은

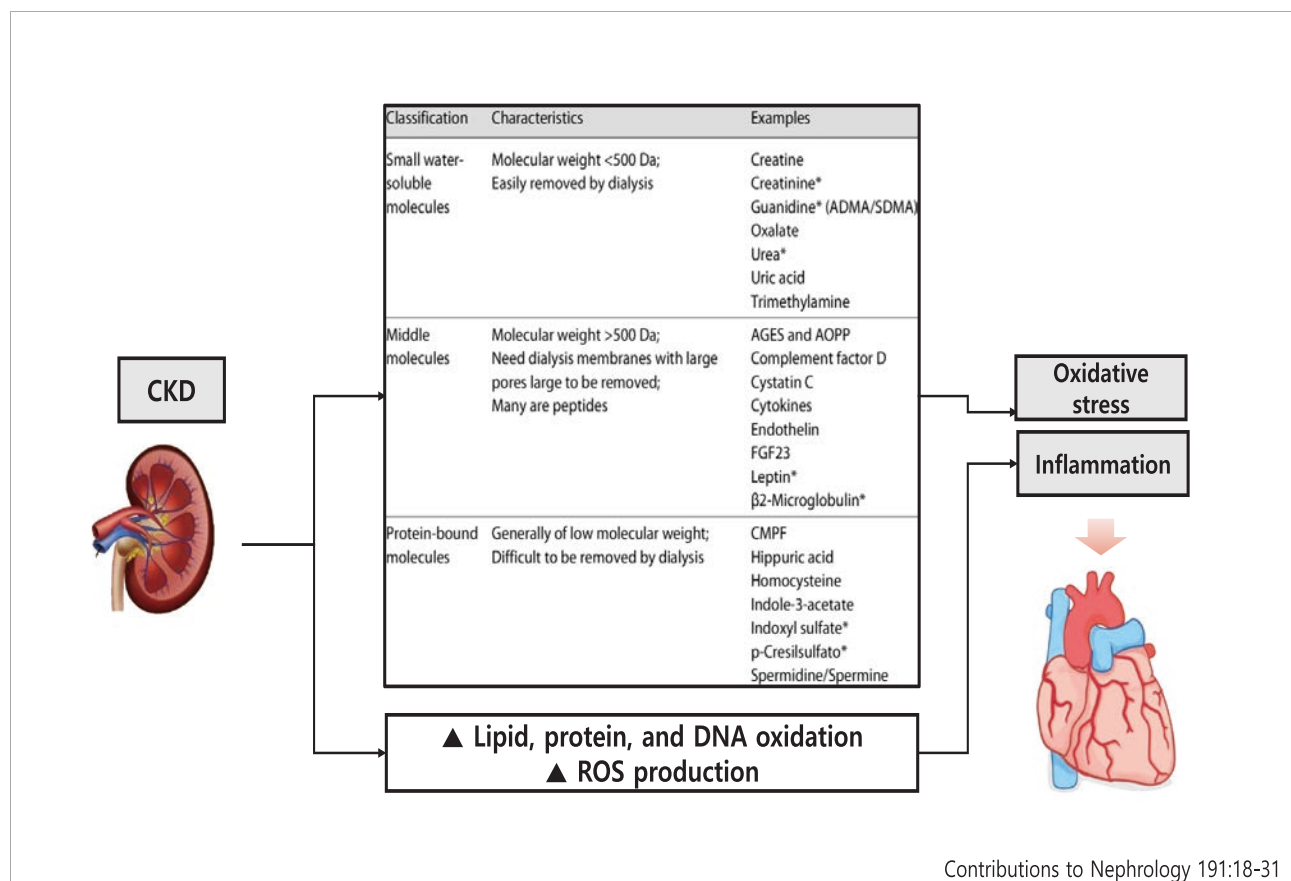
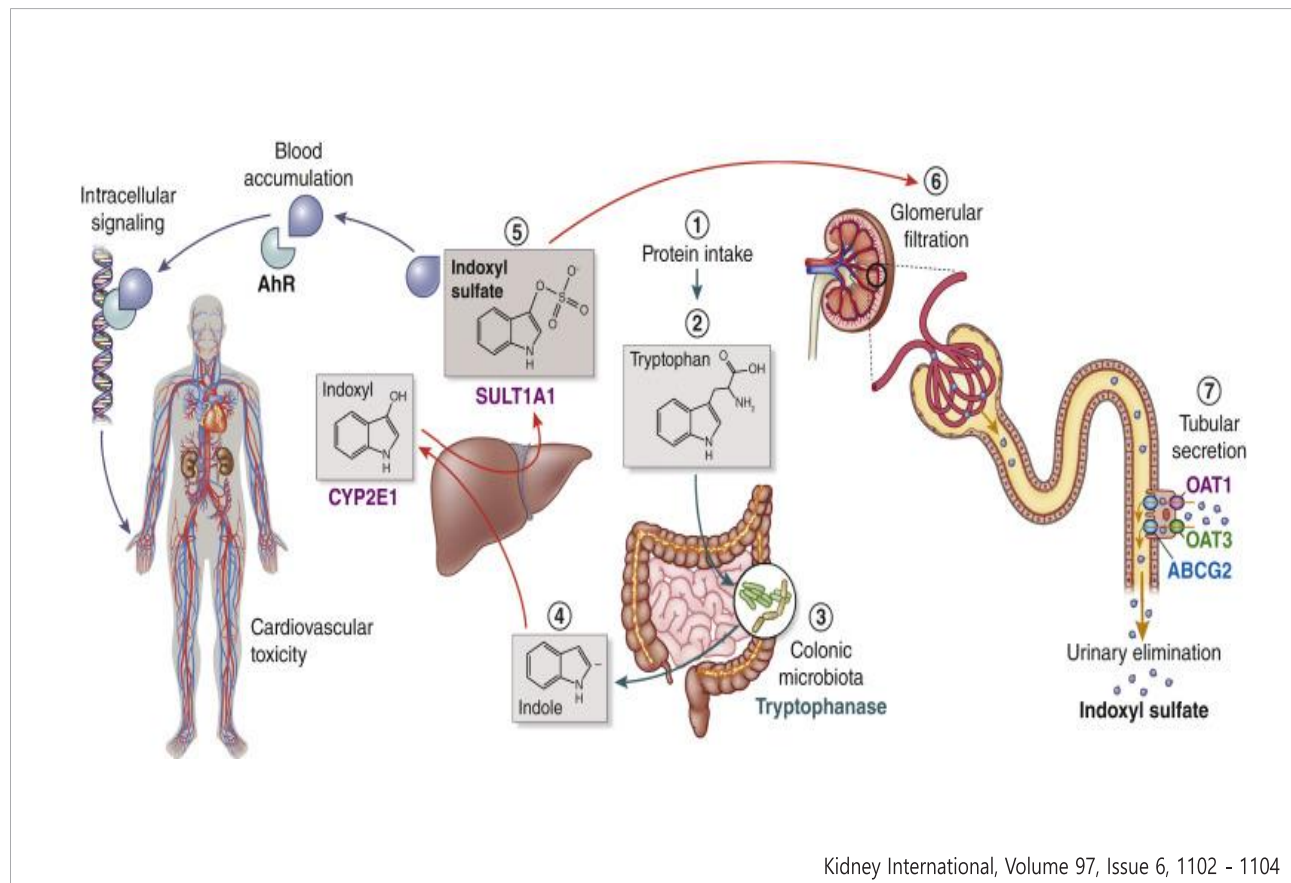
## Chronic kidney disease

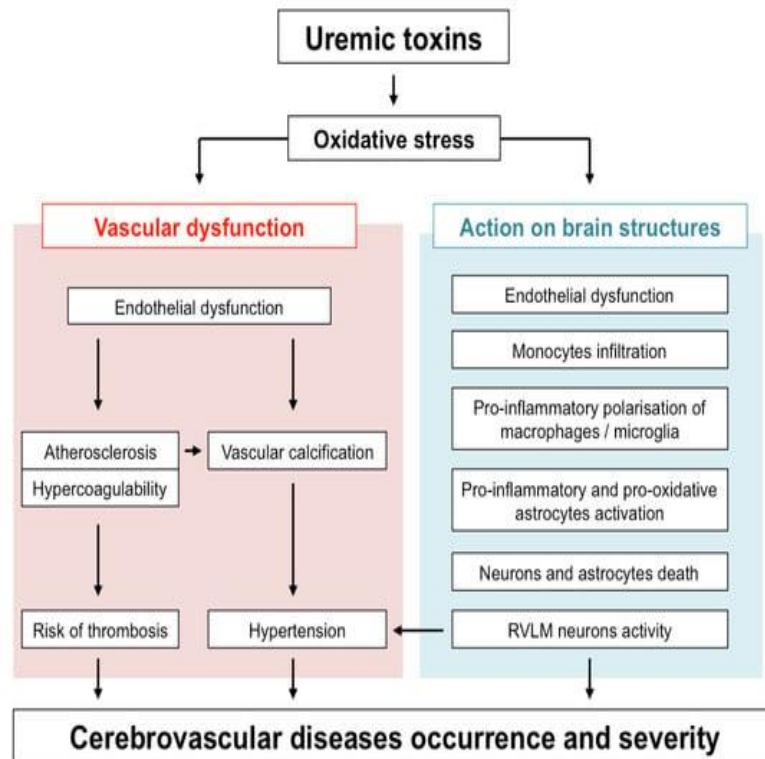
- The Global Burden of Disease Study 2017 reported a 9.1% global prevalence of CKD
- In 2017, CKD resulted in 1.2 million deaths worldwide.
- The presence of impaired kidney function and albuminuria leads to a 2-4 fold increase in the risk of CVD



Gansevoort, R. T. et al. Lancet 382, 339–352 (2013).

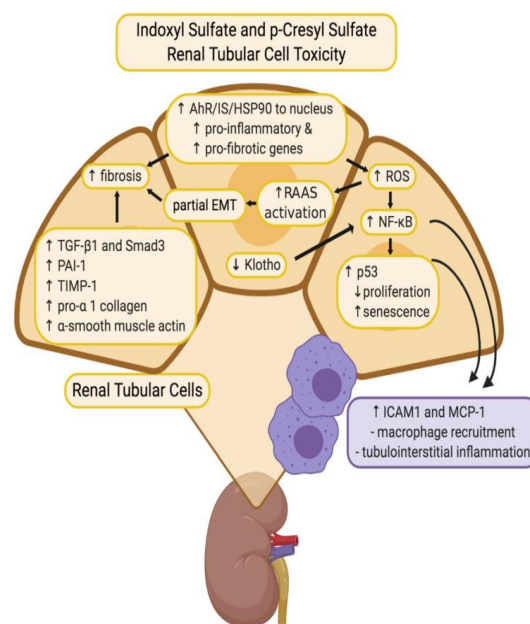






Toxins 2018, 10(7), 303

## Uremic Toxins in the Progression of Chronic Kidney Disease



Toxins (Basel). 2021 Feb 13;13(2):142.

# Protein-bound uremic toxins are not effectively removed by hemodialysis.

TABLE 2. Change of predialysis serum concentration of protein-bound compounds, water-soluble compounds, middle molecule,  $\beta_2$ M, and proteins

	Baseline	Week 4	Week 5	Week 9	Analysis of variance
Protein-bound total (mg/dL)					
Hippuric acid	4.07 ± 2.42	3.73 ± 2.46	3.14 ± 2.45	3.81 ± 2.74	ns
Indoleacetic acid	0.23 ± 0.17	0.24 ± 0.15	0.22 ± 0.13	0.21 ± 0.13	ns
Indoxylsulfate	1.65 ± 0.70	1.47 ± 0.68	1.47 ± 0.83	1.47 ± 0.73	ns
p-cresylsulfate	3.98 ± 1.51	3.44 ± 1.50	3.47 ± 1.52	3.17 ± 1.77*	0.04
cmpl	0.72 ± 0.52	0.70 ± 0.48	0.60 ± 0.48** <sup>†</sup>	0.64 ± 0.46*** <sup>††</sup>	<0.0001
Protein-bound free (mg/dL)					
Hippuric acid	2.19 ± 1.39	2.00 ± 1.50	1.69 ± 1.44	1.98 ± 1.56	ns
Indoleacetic acid	0.08 ± 0.06	0.08 ± 0.05	0.07 ± 0.05	0.07 ± 0.04 <sup>†</sup>	0.03
Indoxylsulfate	0.14 ± 0.07	0.14 ± 0.10	0.13 ± 0.08	0.12 ± 0.07	ns
p-cresylsulfate	0.25 ± 0.16	0.23 ± 0.21	0.22 ± 0.15	0.20 ± 0.19	ns
Water-soluble (mg/dL)					
Urea	127.5 ± 28.5	109.1 ± 25.1*	108.5 ± 29.3*	118.1 ± 28.6	0.03
Creatinine	8.1 ± 2.0	7.7 ± 2.5	7.6 ± 2.1	8.0 ± 2.5	ns
Uric acid	6.7 ± 1.4	6.5 ± 1.5	6.4 ± 1.3	6.5 ± 1.3	ns
Middle molecule (mg/L)					
$\beta_2$ M	24.7 ± 9.3	23.3 ± 9.5	22.7 ± 10.1	18.1 ± 6.7**	0.01
Proteins (g/L)					
Total protein	65.3 ± 3.6	64.2 ± 3.3	64.3 ± 2.9	63.4 ± 2.9	ns
Albumin	34.6 ± 6.2	36.0 ± 3.1	36.5 ± 4.6	35.9 ± 5.5	ns

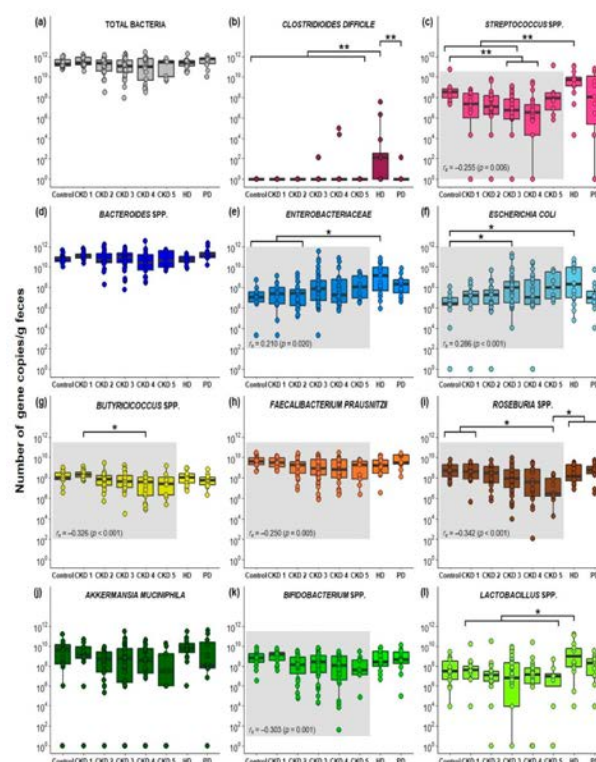
Meert, N. et al. *Artif. Organs* 34, 580–585 (2010).

# Targeting Uremic Toxins and Gut Microbiota in CKD Management

- One of the primary goals in CKD therapeutics is to develop treatments that can limit the accumulation of potential uraemic cardiotoxins, **preferably early** on in the course of the disease
- Recent insights indicate that gut microbial composition is altered in patients with CKD, which suggests that the intestinal tract and its **microbiota are potentially novel intervention targets**.

## Gut Microbiota and Uremic Toxins in CKD Progression

- Uremia-related factors can lead to alterations in gut microbiota composition
  - prolonged transit time, dietary restrictions and medication use
- Gut microbiota itself -> uremic toxin precursors -> contributes to CKD
- Uremic toxin accumulation in CKD alters gut microbiota further.
  - Inadequate excretion of toxin



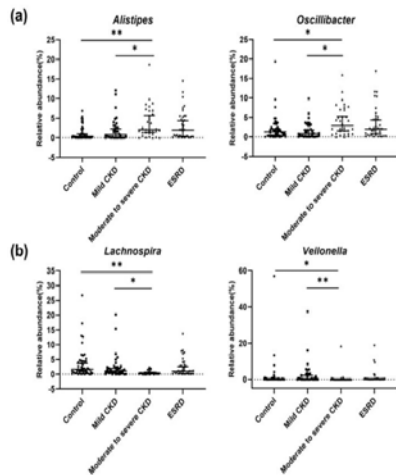
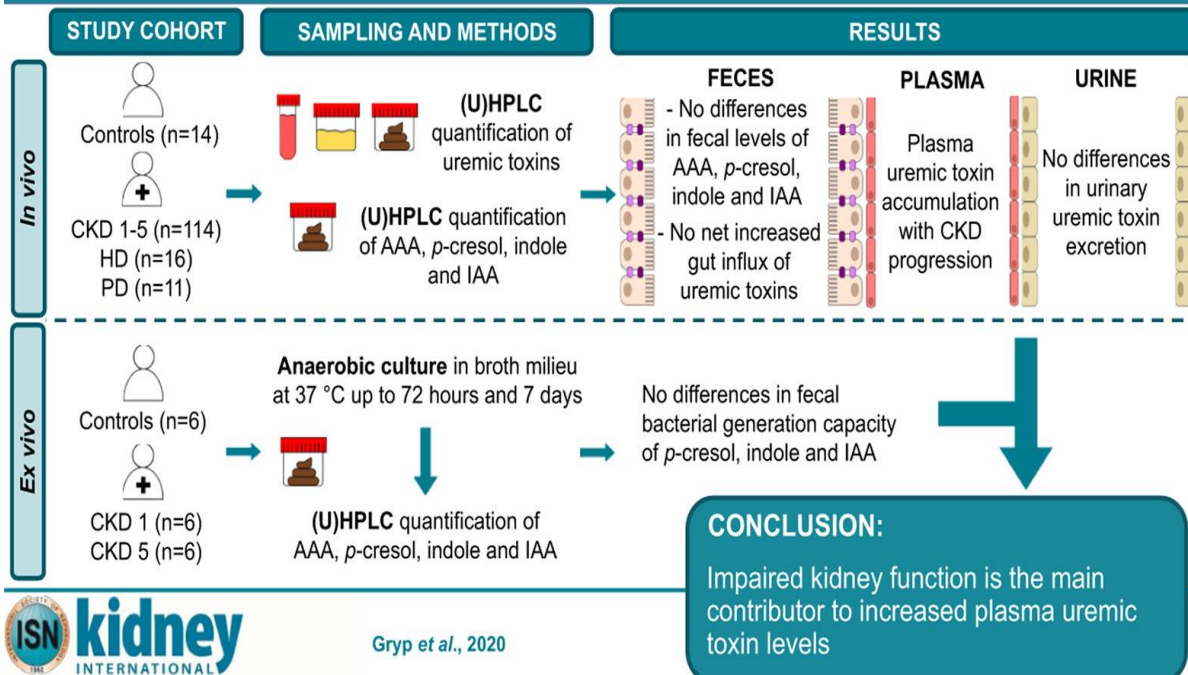


Table 2. Linear regression with each metabolite and major genera.

Predictors	Regression Coefficient	Standard Error	Adjusted R <sup>2</sup>	p	FDR
<b>p-cresyl sulfate (log)</b>					
<i>Alistipes</i>	0.207	0.050	0.100	<0.001	<0.001
<i>Oscillibacter</i>	0.238	0.045	0.155	<0.001	<0.001
<i>Lachnospira</i>	-0.105	0.043	0.033	0.016	0.039
<i>Veillonella</i>	-0.079	0.027	0.050	0.004	0.014
<i>Subdoligranulum</i>	0.234	0.085	0.042	0.007	0.023
<i>Megamonas</i>	-0.092	0.036	0.036	0.012	0.034
<b>p-cresyl glucuronate (log) *</b>					
<i>Prevotella</i>	-0.023	0.009	0.034	0.014	0.062
<i>Alistipes</i>	0.189	0.057	0.062	0.001	0.010
<i>Oscillibacter</i>	0.213	0.053	0.094	<0.001	0.001
<i>Lachnospira</i>	-0.104	0.049	0.024	0.034	0.103
<i>Subdoligranulum</i>	0.224	0.097	0.028	0.023	0.081
<b>Indoxyl sulfate (log)</b>					
<i>Alistipes</i>	0.126	0.043	0.048	0.004	0.035
<i>Oscillibacter</i>	0.112	0.041	0.043	0.007	0.037
<i>Lachnospira</i>	-0.091	0.036	0.034	0.014	0.058
<i>Subdoligranulum</i>	0.177	0.073	0.032	0.016	0.058
<b>TMAO (log)</b>					
<i>Prevotella</i>	-0.016	0.007	0.027	0.026	0.080
<i>Alistipes</i>	0.104	0.046	0.027	0.026	0.080
<i>Oscillibacter</i>	0.145	0.042	0.067	0.001	0.006
<i>Lachnospira</i>	-0.087	0.039	0.026	0.027	0.080
<i>Dialister</i>	-0.107	0.043	0.034	0.014	0.080

Kim JE et al. Microorganisms 2020, 8, 907

## Gut microbiota generation of protein-bound uremic toxins and related metabolites is not altered at different stages of chronic kidney disease

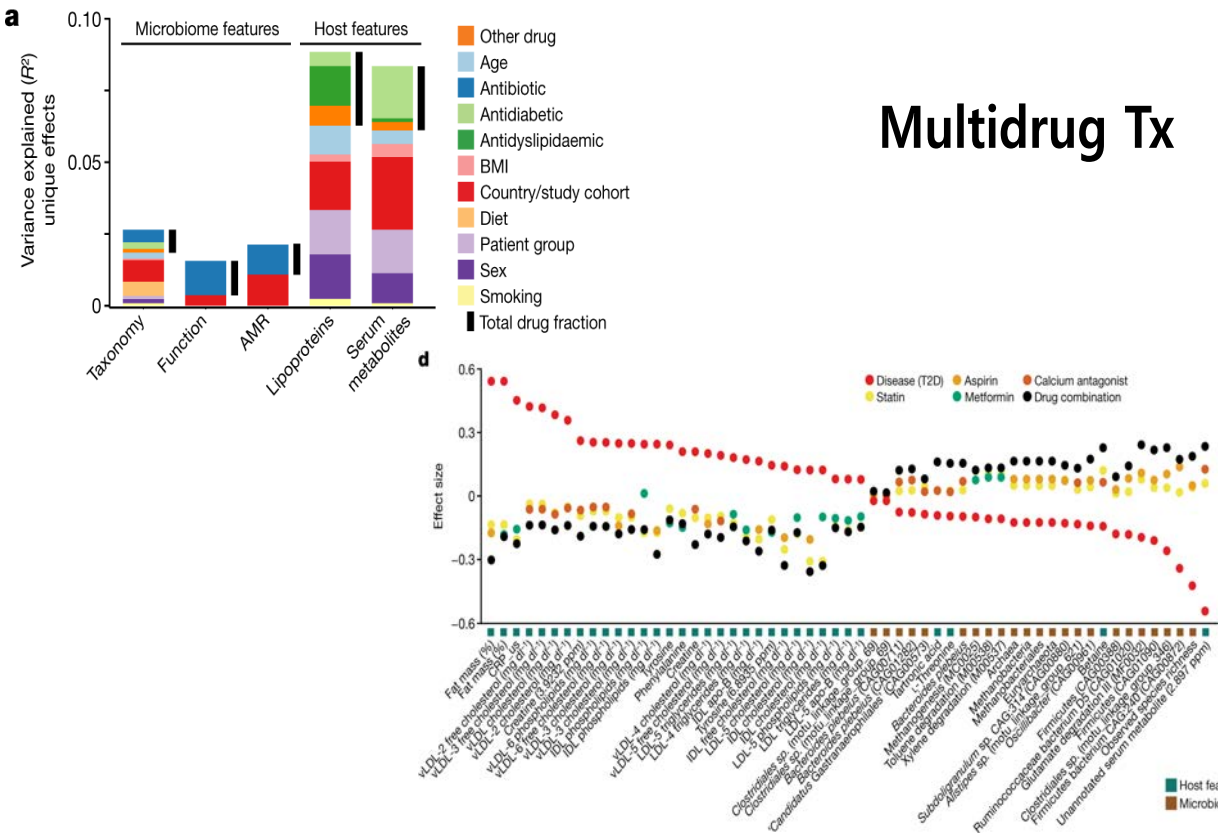




# Patient factors and the gut microbiome in CKD

- Challenges in CKD Microbiome Research

- multidrug therapy
- comorbidities
- uraemic milieu
- low-fibre diet
- limited mobility
- constipation and ageing

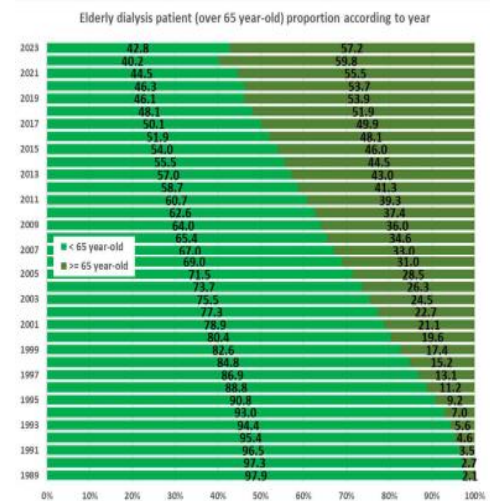
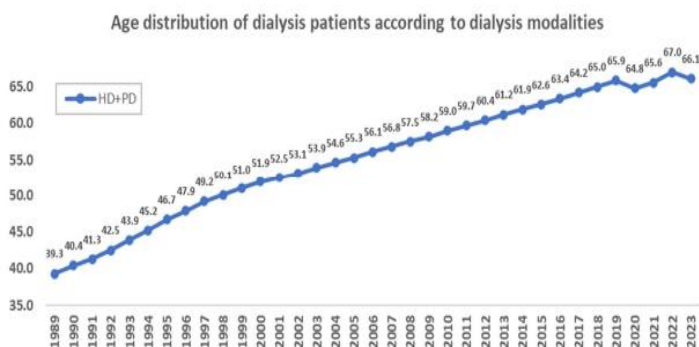


Nature 600, 500–505 (2021).



## Aging

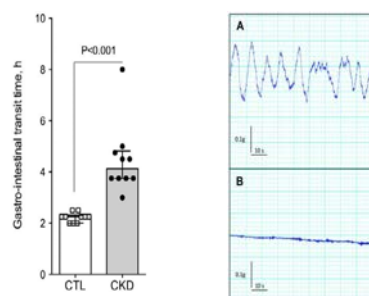
- The incidence of CKD is higher in the ageing population.
- Age-matched controls are crucial in studies.



Trends in epidemiologic characteristics of end-stage kidney disease from 2023 KORDS

## Prolongation of colonic transit time

- 56 HD vs. 63 CAPD patients, and 25 healthy control.
- Colonic transit time was significantly prolonged in HD patients ( $43.0 \pm 22.2$  versus  $32.7 \pm 13.7$  hours in CAPD patients and  $24.3 \pm 11.9$  hours in controls;  $P < 0.001$ ).
- Alteration in gastro-intestinal transit time in mice with CKD

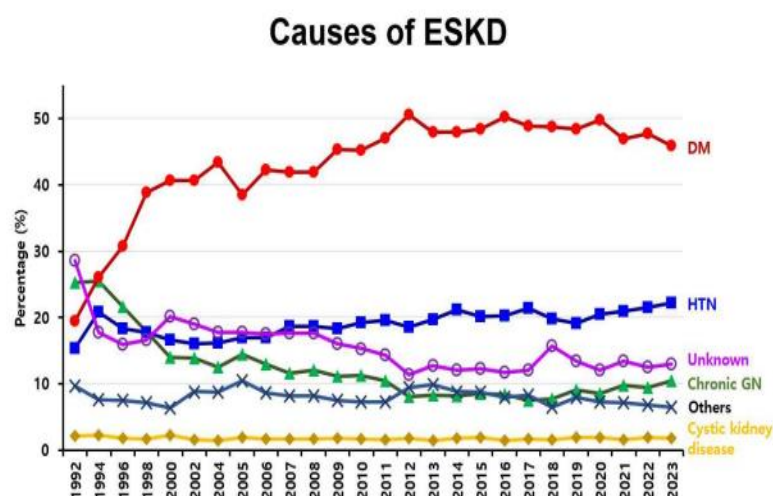


Am. J. Kidney Dis. 44, 322–327 (2004).  
Toxins (Basel). 2018 May 16;10(5):204

## Diet & Uremic milieu

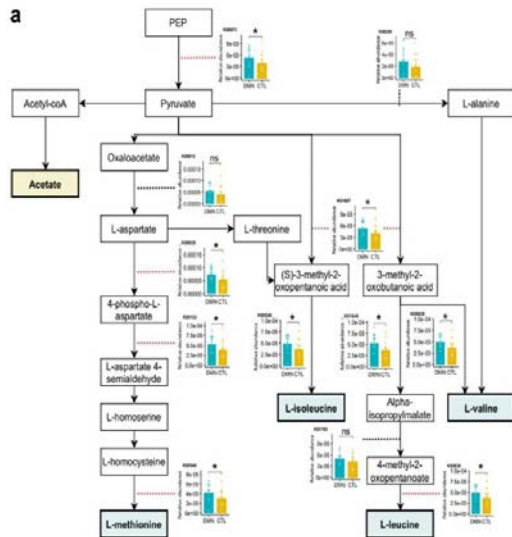
- To limit serum potassium levels, patients with CKD are often advised to **reduce fruit and vegetable intake**, resulting in a reduced fiber intake.
- **Urea and uric acid influx** boosts bacteria with urease, uricase, and toxin-generating enzymes while reducing butyrate producers.

## Disease Heterogeneity in CKD

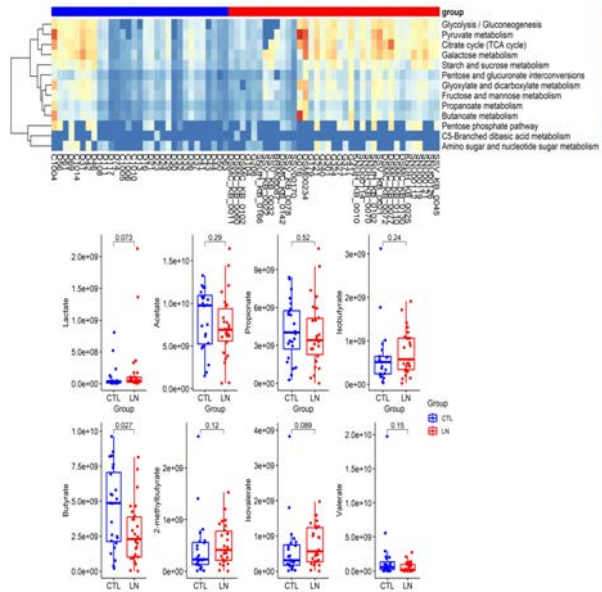


## Disease Heterogeneity in CKD

### Diabetic nephropathy



### Lupus nephritis



Microbiol Spectr. 2023 Mar 6;11(2):e0234422

Unpublished

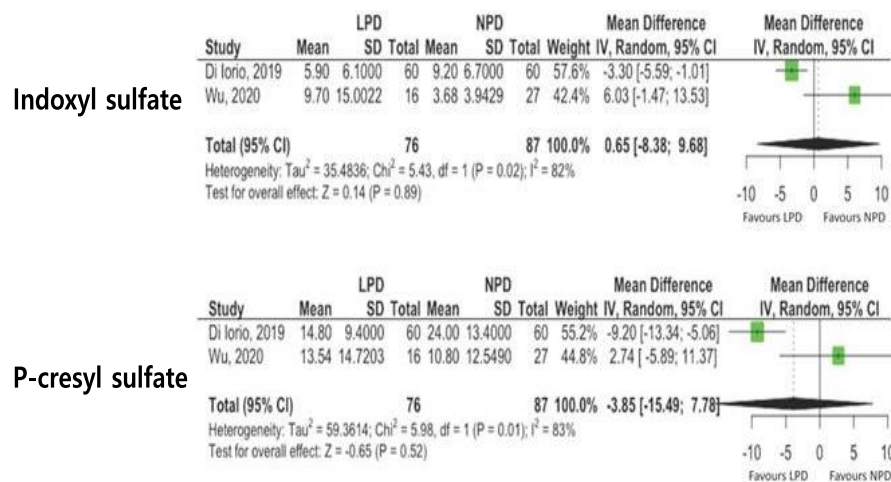
## Targeting uremic metabolites in CKD

- Treatment target : **decreasing levels of uremic toxins** and improving the outcomes of patients with CKD
- Various approaches...
  - Dietary interventions
  - Biotic interventions
  - Use of oral adsorbents

## Dietary interventions in CKD

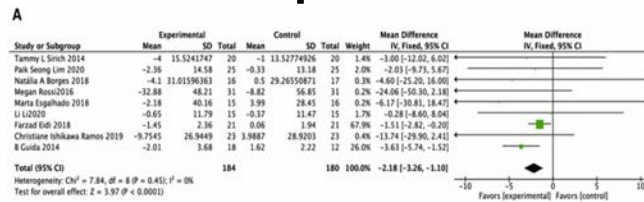
- Mediterranean diets (rich in vegetables, nuts, legumes, fruits and whole grains)
- Plant-based diets and low-protein diets (LPDs; reduced dietary protein intake but avoiding the complete absence of protein)
- **Decreasing bacterial proteolytic fermentation** by reducing the high protein consumption

## Effects of low protein diet on uremic toxins

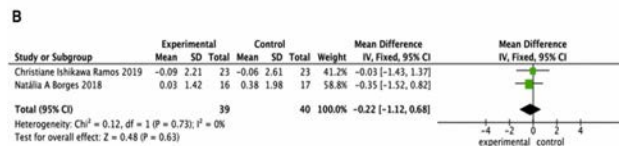


## Biotic interventions in patients with CKD

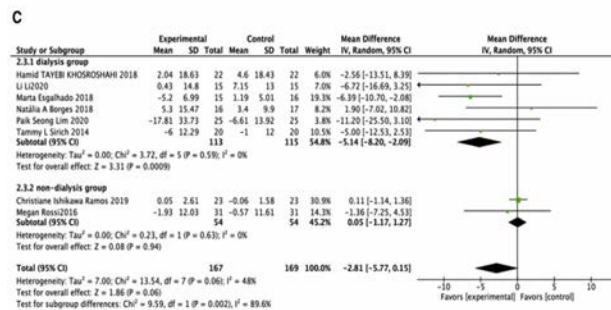
P-cresyl sulfate



Indole-3-acetic acid



Indoxyl sulfate



J. Ren. Nutr. 32, 10–21 (2022).

## Biotic interventions in patients with CKD

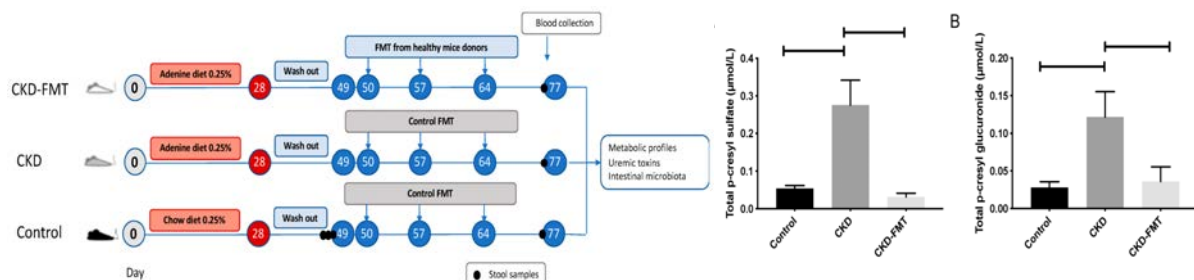
Table 2 | Biotic intervention studies in patients with CKD

Study design	Study duration (weeks)	CKD stage <sup>a</sup>	N	Supplementation	Uraemic toxin changes	Taxa changes post-intervention	Other remarks	Ref.
RCT, SC, DBP	18	4, 5	37	9 bacterial strains across <i>Bifidobacterium</i> , <i>Lactobacillus</i> and <i>Streptococcus</i> genera combined with inulin <sup>b</sup> , FOS <sup>b</sup> , GOS <sup>b</sup>	↓pCS ↓IxS <sup>b</sup>	↑ <i>Bifidobacterium</i> spp. ↑Lachnospiraceae ↑ <i>Faecalibacterium</i> spp. <sup>c</sup> ↓Clostridiales ↓Ruminococcaceae	↑Albuminuria	96
RCT, SC, DBP	26	5 (HD)	45	<i>Bifidobacterium longum</i> NQ1501, <i>Lactobacillus acidophilus</i> YIT2004 <i>Enterococcus faecalis</i> YIT0072	↓pCS <sup>d</sup>	↑Bacteroidaceae ↑Enterococcaceae ↓Ruminococcaceae ↓Halomonadaceae ↓Erysipelotrichaceae ↓Peptostreptococcaceae ↓Clostridiales family XIII	NA	82
RCT, DBP	38	5 (PD)	21	ITF <sup>b</sup>	None	↓Abundance of indole-generating species	↓Intestinal pH	94
RCT, DBP	52	3, 4	56	9 strains across <i>Bifidobacterium</i> , <i>Lactobacillus</i> , <i>Streptococcus</i> genera combined with HRSF <sup>b</sup>	None	↑ <i>Bifidobacterium</i> spp. ↑ <i>Blautia</i> spp.	↑Creatinine ↑eGFR	87
RCT, SC, SB	14	3, 4	59	β-glucan fibre <sup>b</sup>	↓Free pCS ↓Free IxS ↓pCG	Shift from <i>Bacteroides</i> 2 enterotype to <i>Prevotella</i> enterotype	↓LDL cholesterol	93

Nat Rev Nephrol. 2023 Feb;19(2):87–101.

## Fecal microbiota transplantation in CKD

- In mice with CKD, FMT improved p-cresol-derived uraemic toxin accumulation and reduced CKD complications by having a beneficial effect on gut microbiota



- FMT in CKD Patients: Unexplored

## Take home message

- The gut microbiome influences not only **CKD progression** but also the **development of cardiovascular disease (CVD)** through uremic toxin production and systemic inflammation.
- Diet, medications, uremic milieu, colonic transit time, aging, and comorbidities significantly alter gut microbiota, making **microbiome research in CKD inherently complex**.
- Although **biotic interventions**, including probiotics, prebiotics, and synbiotics, are being explored, **clear clinical implications have yet to be established**, highlighting the need for further research.





# 03

## Gut-lung axis: from epidemiology to therapeutic application



이세원  
(서울아산병원)





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2022 ~ 2024	울산대학교 의과대학 교육부학장
2023 ~ 현재	서울아산병원 호흡기내과 교수

# **Gut–lung axis: from epidemiology to therapeutic application**

The gut–lung axis represents a dynamic interplay between the gut and lung microbiota with profound implications for respiratory health and disease. Epidemiological studies have shown that dietary patterns significantly influence the risk of developing chronic respiratory diseases such as chronic obstructive pulmonary disease (COPD) and asthma. These findings suggest that diet-induced changes in the gut microbiota may play a crucial role in modulating systemic and pulmonary immune responses, thereby impacting respiratory disease pathogenesis. This lecture will delve into the mechanisms linking diet, gut microbiota, and respiratory health, with a focus on how dietary components influence the gut–lung axis. Emerging evidence suggests that interventions targeting the gut microbiota, including prebiotics, probiotics, postbiotics, and fecal microbiota transplantation (FMT), hold promise as therapeutic strategies for COPD. Early experimental studies have demonstrated that these approaches can improve pulmonary inflammation, enhance immune modulation, and potentially slow disease progression. We will examine the mechanisms underlying these effects, such as the production of beneficial metabolites like short-chain fatty acids (SCFAs) and the restoration of gut microbial diversity.

# **Gut-Lung axis**

## **From Epidemiology to Therapeutic Application**

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**Sei Won Lee, MD PhD**



University of Ulsan  
Asan Medical Center  
Pulmonology and Critical Care Medicine



## **Contents**

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- 1. Epidemiologic background in COPD**
- 2. Gut-Lung Axis**
- 3. Clinical application of microbiome in COPD**



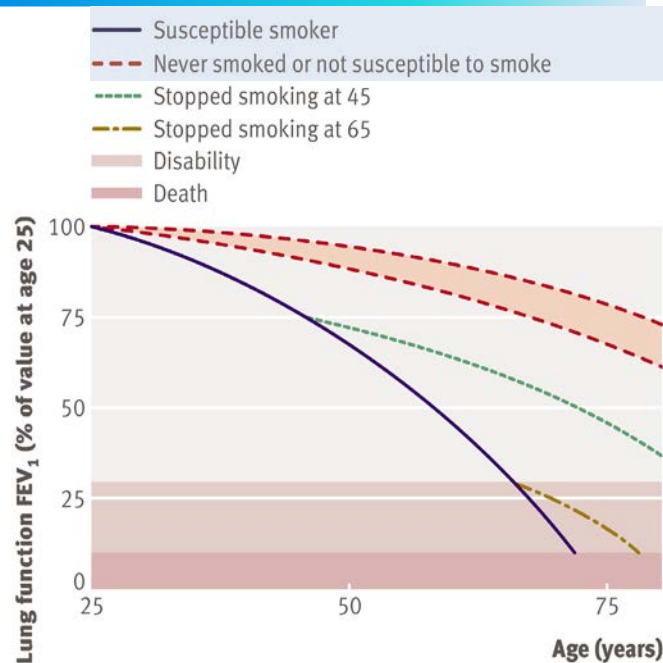
## COPD definition in GOLD 2023

“a heterogenous lung condition characterized by chronic respiratory symptoms (dyspnea, cough, sputum production and/or exacerbations) due to abnormalities of the **airways** (bronchitis, bronchiolitis) and/or **alveoli** (emphysema) that cause **persistent**, often **progressive**, **airflow obstruction**.”

*‘Cigarette smoking is a key environmental risk factor for COPD’*

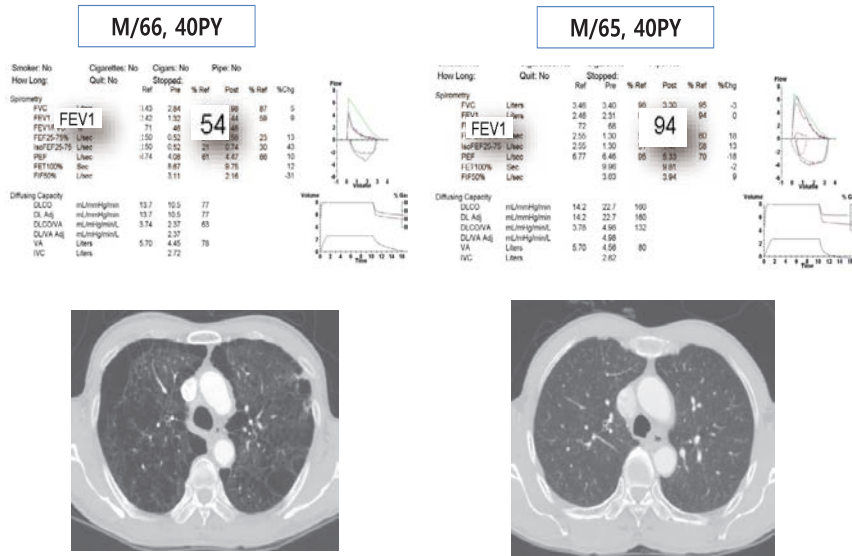
Global Strategy for the Diagnosis, Management, and Prevention of Chronic Obstructive Pulmonary Disease, Global Initiative for Chronic Obstructive Lung Disease (GOLD) 2023. Available from [www.goldcopd.com](http://www.goldcopd.com)


## Smoking: Key environmental risk factor



Fletcher et. al. , Br Med J 1977

## Smoking sensitivity decides disease developm



From a number of  studies, it has become apparent that there are “susceptible” smokers that will develop COPD, but **in most non-smokers and many smokers clinically significant airflow obstruction never develops.**

Fletcher C, Peto R, Tinker C, et al. The Natural History of Chronic Bronchitis and Emphysema. Oxford: Oxford University Press, 1976. European Respiratory Monograph, Vol.7. Sheffield, U.K., 1998. 41-72.

Smoking is considered the major risk factor for development of COPD and accounts for 89–90% of the cumulative risk. However, not all smokers will develop COPD. In fact, **only a small proportion (about 15%) will develop clinically relevant disease.**

Am Rev Respir Dis 1989;140:S82-4.  
Am Rev Respir Dis 1977;115(2):195-205.

## Prospective study of dietary fiber and COPD in US



- 111,580 US women and men (Nurses' Health Study and Health Professionals Follow-up Study), with 832 cases of newly diagnosed COPD being reported between 1984 and 2000.
- The cumulative average intake of total fiber and of fiber from specific sources (cereal, fruit, and vegetables) was calculated.

Am J Epidemiol 2010;171:776–784

## Fiber intakes reduced newly diagnosed COPD



	Quintile of Intake					P <sub>trend</sub>
	Quintile 1	Quintile 2	Quintile 3	Quintile 4	Quintile 5	
Current smokers						
Total fiber						
Median, g/day	10.8	14.0	18.1	21.1	25.3	
No. of cases	168	123	86	90	41	
Multivariate RR (95% CI) <sup>a</sup>	1.00	0.84 (0.66, 1.08)	0.70 (0.52, 0.93)	0.91 (0.67, 1.23)	0.56 (0.38, 0.85)	0.03
Multivariate RR (95% CI) <sup>b</sup>	1.00	0.85 (0.66, 1.08)	0.70 (0.54, 0.97)	0.96 (0.69, 1.29)	0.60 (0.40, 0.94)	0.09
Cereal fiber						
Median, g/day	2.1	3.4	4.6	6.1	9.0	
No. of cases	162	126	94	84	42	
No. of person-years	40,488	30,423	24,541	20,469	16,328	
Multivariate RR (95% CI) <sup>a</sup>	1.00	1.02 (0.80, 1.30)	0.94 (0.72, 1.24)	0.98 (0.73, 1.30)	0.63 (0.44, 0.91)	0.05
Multivariate RR (95% CI) <sup>b</sup>	1.00	1.02 (0.79, 1.30)	0.95 (0.73, 1.26)	0.99 (0.76, 1.36)	0.66 (0.46, 0.94)	0.09
Fruit fiber						
Median, g/day	1.2	2.5	3.7	5.0	7.6	
No. of cases	200	114	90	68	36	
No. of person-years	44,676	29,369	23,304	19,006	15,894	
Multivariate RR (95% CI) <sup>a</sup>	1.00	0.68 (0.49, 1.11)	0.93 (0.72, 1.21)	0.94 (0.70, 1.25)	0.65 (0.45, 0.95)	0.09
Multivariate RR (95% CI) <sup>b</sup>	1.00	0.90 (0.71, 1.14)	0.98 (0.75, 1.27)	1.00 (0.74, 1.35)	0.72 (0.49, 1.06)	0.33
Vegetable fiber						
Median, g/day	3.4	4.9	6.2	7.7	10.8	
No. of cases	133	97	107	101	70	
No. of person-years	32,290	28,263	25,770	23,385	22,541	
Multivariate RR (95% CI) <sup>a</sup>	1.00	0.80 (0.61, 1.04)	0.93 (0.72, 1.21)	1.02 (0.78, 1.34)	0.82 (0.60, 1.13)	0.74
Multivariate RR (95% CI) <sup>b</sup>	1.00	0.81 (0.62, 1.06)	0.97 (0.75, 1.27)	1.09 (0.82, 1.45)	0.92 (0.66, 1.28)	0.68

Am J Epidemiol 2010;171:776–784

## Prudent diet is associated with good lung function



- The relationships between dietary patterns and lung function and spirometrically defined COPD.
- 1,551 males and 1,391 females in Hertfordshire, UK.

Eur Respir J 2010; 36: 277–284

## Prudent diet is associated with good lung function



Eur Respir J 2010; 36: 277–284

	Subjects	Partially adjusted <sup>a</sup> coefficient	Fully adjusted <sup>a</sup> coefficient
<b>Males</b>			
Prudent diet score			
1 (lowest fifth)	470	2.69 ± 0.63	0.06 (0.01–0.13)
2	365	2.81 ± 0.62	0.16 (0.07–0.24)
3	265	2.96 ± 0.54	0.19 (0.10–0.28)
4	258	2.90 ± 0.59	0.13 (0.05–0.21)
5 (highest fifth)	196	3.01 ± 0.53	0.19 (0.10–0.28)
<b>Females</b>			
Prudent diet score			
3 (middle fifth)	118	1.88 ± 0.42	0.05 (0.03–0.07)
4	228	1.91 ± 0.42	0.05 (0.03–0.07)
5 (highest fifth)	324	1.93 ± 0.42	0.05 (0.03–0.07)
Fifths of prudent diet score as a trend	391	1.99 ± 0.42	0.04 (0.02–0.05)
		2.09 ± 0.38	0.04 (0.02–0.05)

“Prudent” pattern:

high consumption of fruit, vegetables, oily fish and wholemeal cereals

Eur Respir J 2010; 36: 277–284

## Prudent diet is associated negatively with new onset COPD



Eur Respir J 2010; 36: 277–284

TABLE 5 Relationship between “prudent” dietary pattern and chronic obstructive pulmonary disease (COPD) in males and females				
	Subjects	Subjects with COPD	Partially adjusted <sup>a</sup> OR	Fully adjusted <sup>a</sup> OR
<b>Males</b>		144 (30.6)		
Prudent diet score				
1 (lowest fifth)	470	72 (20.1)	0.67 (0.48–0.95)	0.54 (0.36–0.82)
2	359	45 (17.0)	0.61 (0.41–0.91)	0.61 (0.38–0.97)
3	265	45 (17.4)	0.57 (0.38–0.84)	0.62 (0.38–1.00)
4	258	45 (17.4)	0.49 (0.31–0.78)	0.46 (0.26–0.81)
5 (highest fifth)	196	28 (14.3)	0.84 (0.76–0.92)	0.86 (0.76–0.97)
<b>Females</b>				
Prudent diet score				
3 (middle fifth)	118	24 (20.3)	0.93 (0.51–1.69)	1.28 (0.64–2.56)
4	227	40 (17.6)	0.92 (0.52–1.65)	1.43 (0.72–2.85)
5 (highest fifth)	322	50 (15.5)	0.70 (0.38–1.30)	1.11 (0.54–2.28)
Fifths of prudent diet score as a trend	391	35 (10.6)	0.62 (0.34–1.13)	0.95 (0.47–1.94)
		38 (9.7)	0.88 (0.77–1.00)	0.95 (0.82–1.09)

Data are presented as n, n (%) or OR (95% CI), unless otherwise stated. <sup>a</sup>ORs are adjusted for age, sex, pack-years, <sup>b</sup>for age, height, smoking status, pack-years, smoke in home, age left education, home ownership status, number of children, social class, fat mass, activity score, energy intake, alcohol, dietary supplement use, birth weight, father's social class at birth, inhaled or oral steroids use, paracetamol use.

“Prudent” pattern:

high consumption of fruit, vegetables, oily fish and wholemeal cereals

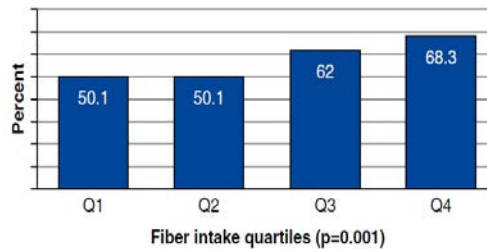
Eur Respir J 2010; 36: 277–284



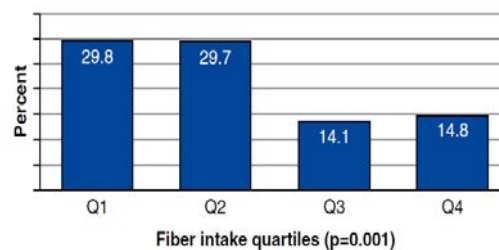
## Fiber intake correlates normal lung function



### Normal airflow pattern(%)



### Restrictive airflow pattern (%)

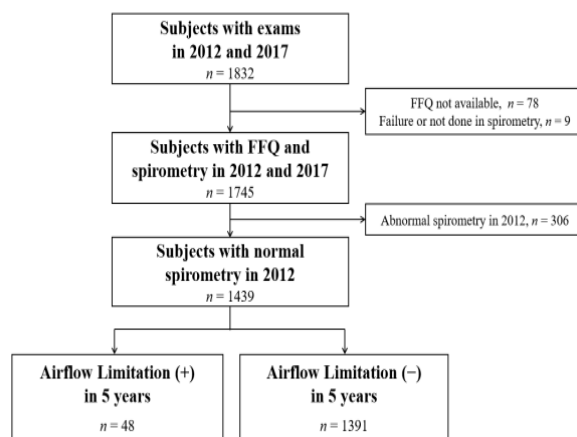


Ann Am Thorac Soc Vol 13, No 5, pp 643–650, May 2016

## Reduced fiber intake correlated with increased COPD incidence



Young Ju Jung MD, PhD



Jung YJ and Lee SW, et al. Nutrients 2021

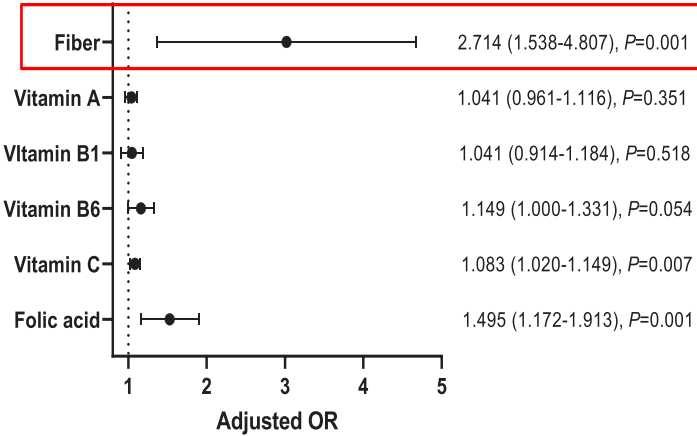
## Reduced fiber intake correlated with increased COPD incidence



Young Ju Jung MD, PhD

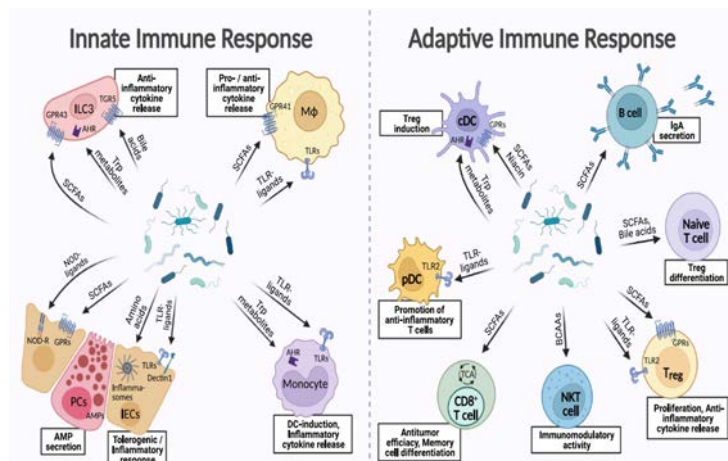
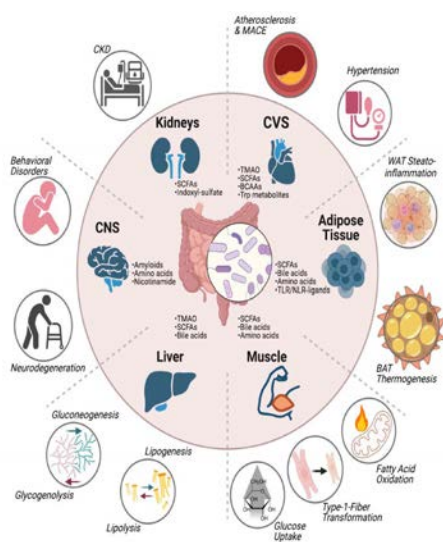


### Adjusted ORs



Jung YJ and Lee SW, et al. Nutrients 2021

## Systemic impacts of microbial products in colon



EMBO reports 23: e55664 | 2022

## Contents

1. Epidemiologic background in COPD

2. Gut-Lung Axis

3. Clinical application of microbiome in COPD

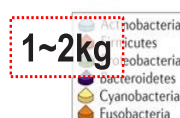
We are 90% microbe and 10% human: ~2kg

The main colonizers

- Firmicutes
- Bacteroidetes
- Proteobacteria
- Actinobacteria
- Fusobacteria
- Cyanobacteria

100,000,000,000,000 bacteria in intestine

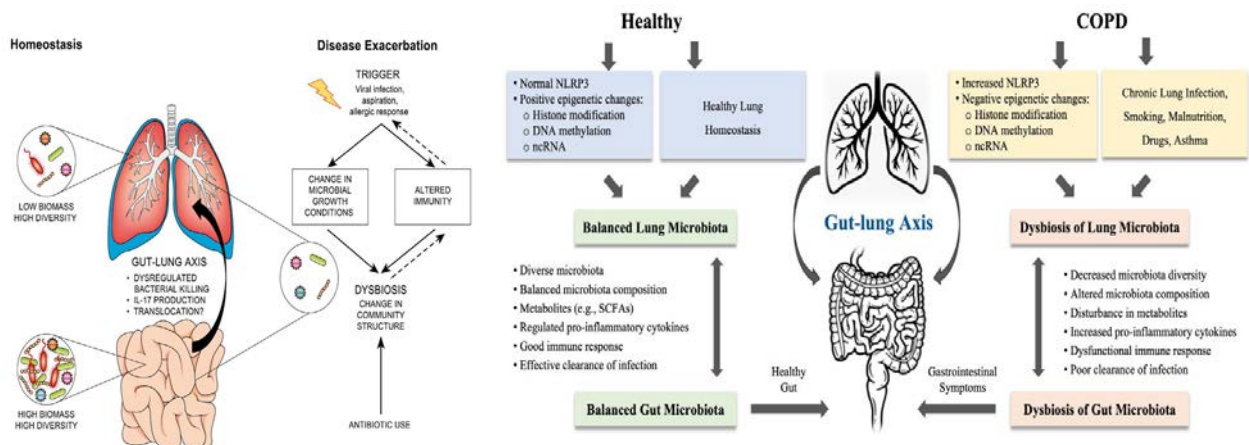
1~2kg



Nature Reviews | Genetics



## Gut-lung axis: Gut microbiota impacts lung health



J Immunol 2016; 196:4839-4847  
Front. Microbiol. 2022;13:868086

nature  
medicine

### ARTICLES

## Gut microbiota metabolism of dietary fiber influences allergic airway disease and hematopoiesis

Aurélien Trompette<sup>1</sup>, Eva S Gollwitzer<sup>1</sup>, Koshika Yadava<sup>1</sup>, Anke K Sichelstiel<sup>1</sup>, Norbert Sprenger<sup>2</sup>, Catherine Ngom-Bru<sup>2</sup>, Carine Blanchard<sup>2</sup>, Tobias Jun<sup>3</sup>, Laurent P Nicod<sup>1</sup>, Nicola L Harris<sup>4</sup> & Benjamin J Marsland<sup>1</sup>

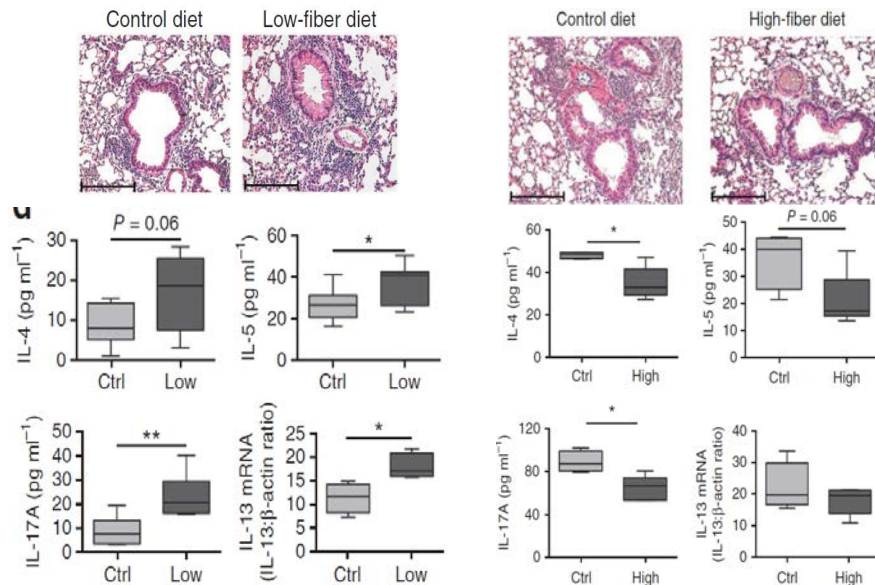
Metabolites from intestinal microbiota are key determinants of host-microbe mutualism and, consequently, the health or disease of the intestinal tract. However, whether such host-microbe crosstalk influences inflammation in peripheral tissues, such as the lung, is poorly understood. We found that dietary fermentable fiber content changed the composition of the gut and lung microbiota, in particular by altering the ratio of Firmicutes to Bacteroidetes. The gut microbiota metabolized the fiber, consequently increasing the concentration of circulating short-chain fatty acids (SCFAs). Mice fed a high-fiber diet had increased circulating levels of SCFAs and were protected against allergic inflammation in the lung, whereas a low-fiber diet decreased levels of SCFAs and increased allergic airway disease. Treatment of mice with the SCFA propionate led to alterations in bone marrow hematopoiesis that were characterized by enhanced generation of macrophage and dendritic cell (DC) precursors and subsequent seeding of the lungs by DCs with high phagocytic capacity but an impaired ability to promote T helper type 2 (T<sub>H</sub>2) cell effector function. The effects of propionate on allergic inflammation were dependent on G protein-coupled receptor 41 (GPR41, also called free fatty acid receptor 3 or FFAR3), but not GPR43 (also called free fatty acid receptor 2 or FFAR2). Our results show that dietary fermentable fiber and SCFAs can shape the immunological environment in the lung and influence the severity of allergic inflammation.

Nat Med 2014 Feb;20(2):159-66.

## High fiber diet attenuates allergic airway inflammation

Low Fiber aggravates airway inflammation

High Fiber improves airway inflammation

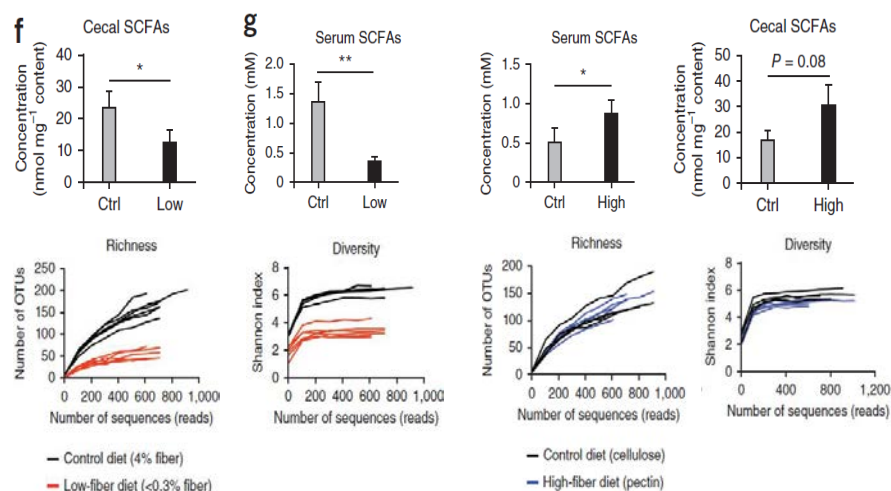


Nat Med 2014 Feb;20(2):159-66.

## High fiber diet increases cecal SCFAs

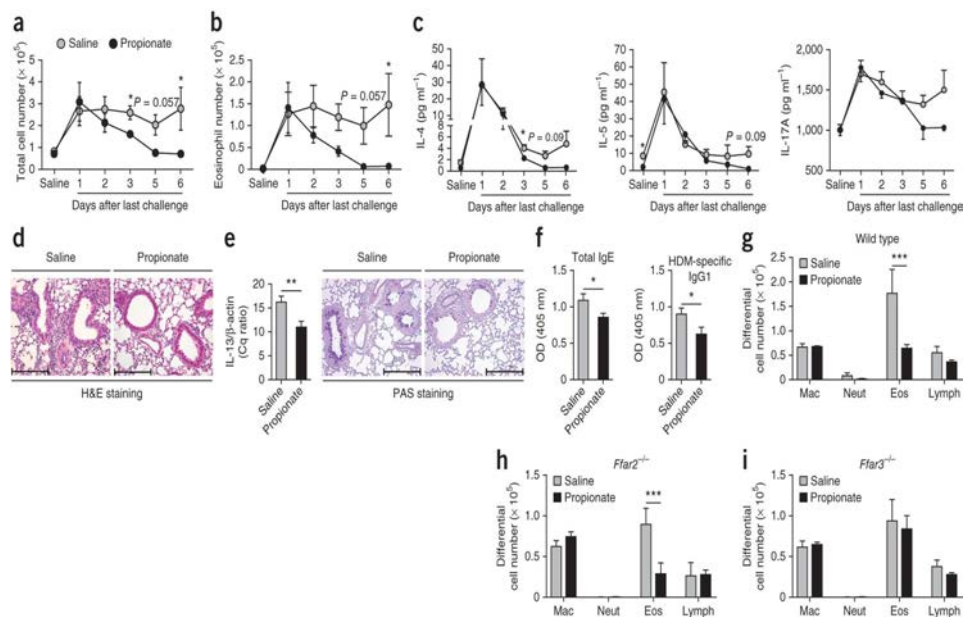
Low Fiber aggravates airway inflammation

High Fiber improves airway inflammation



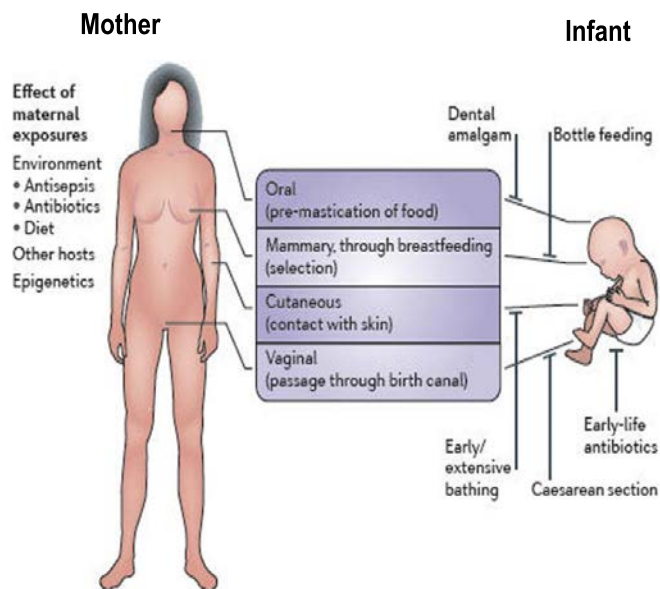
Nat Med 2014 Feb;20(2):159-66.

## Propionates reduced allergic airway inflammation



Nat Med 2014 Feb;20(2):159-66.

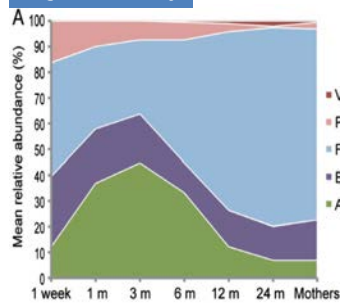
## Microbiota formation in early infant



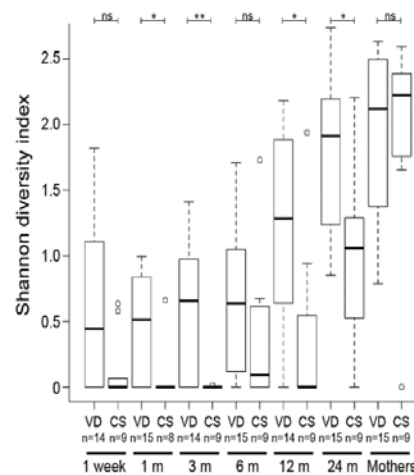
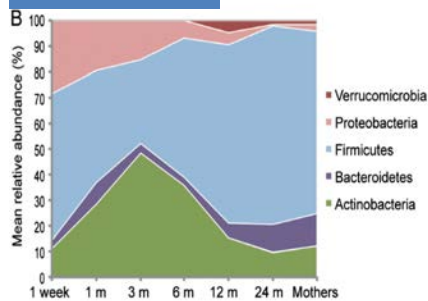
Cho I et. al. Nat Rev Genet. 2012; 13(4): 260–270.

## C-sec significantly reduce gut microbiota diversity

### Vaginal delivery

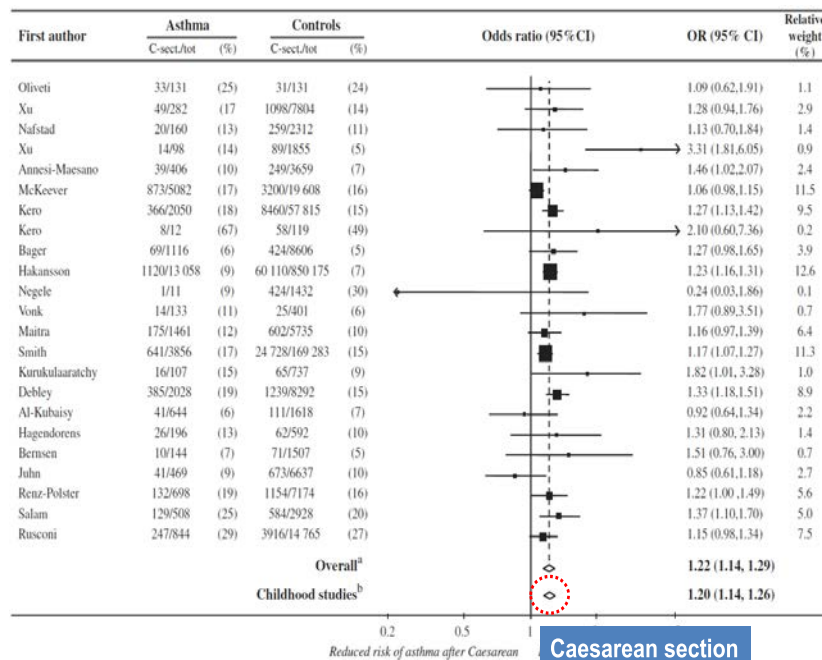


### Caesarean section



Gut 2014;63:559–566

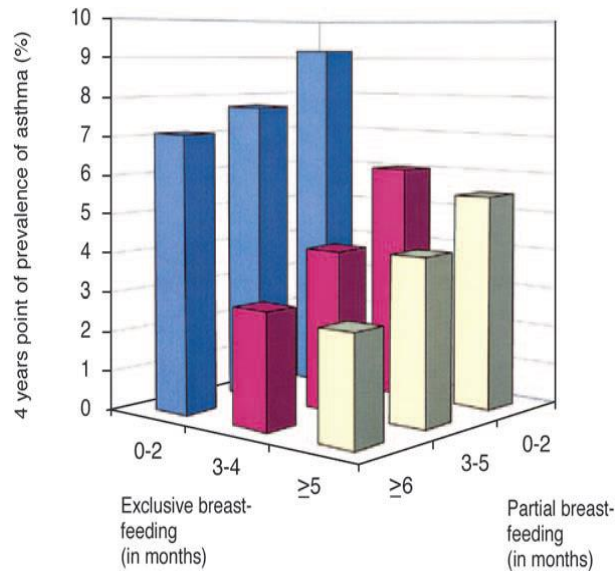
## C-sec increased asthma



Clinical and Experimental Allergy, 38, 629–633



## Breast Feeding is protective to Asthma



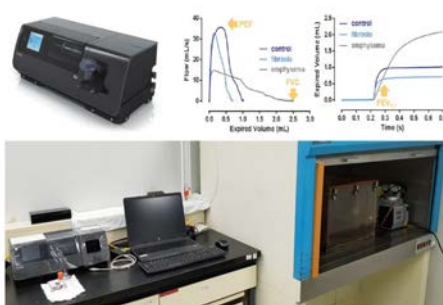
J Allergy Clin Immunol 2004;114:755-60

## 효과 검증의 핵심 - Human relevant animal model

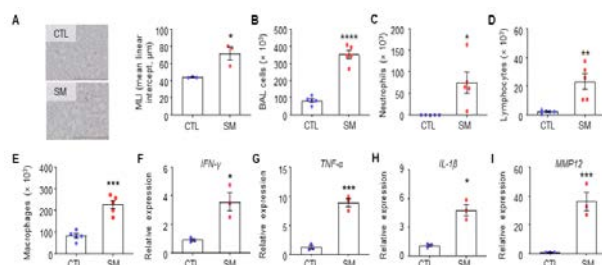
### 흡연 독성실



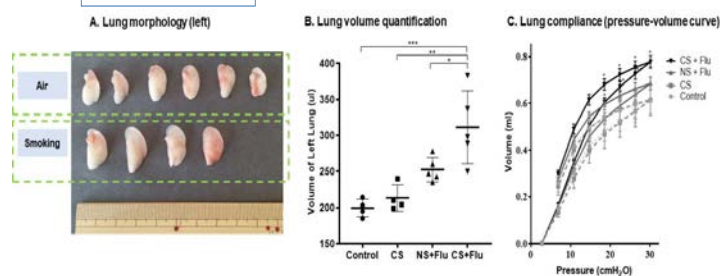
### 폐기능 분석 장비



### 병리 분자생물학적 분석



### 폐기능 분석



## How about COPD? – Nutritional recommendation

- Small amount, nutritious, and frequent meals
- Induce weight gain with enough calories
- Preparation should be easy (e.g., liquid nutritional food, microwaveable)
- Contains a variety of vitamins

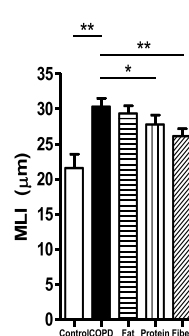
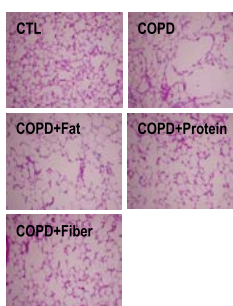
JOURNAL OF TRANSLATIONAL INTERNAL MEDICINE 2015

## High fiber diet attenuated emphysema in smoking exposed model



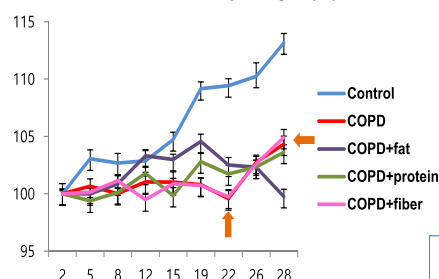
Yoon Ok Jang, PhD

Various diets in animal model



High fiber diet recovered body weight

Relative body weight (%)

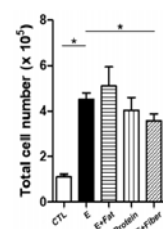


Smoking exposed emphysema model

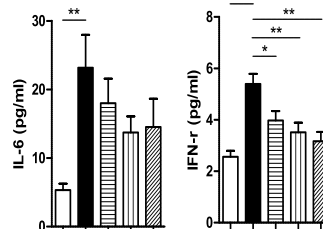


Inflammatory marker reduced in high fiber diet

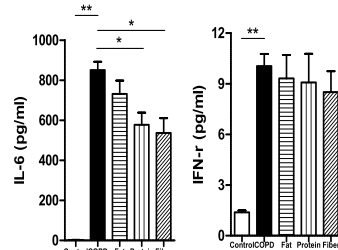
Cell count



Serum



BAL sup

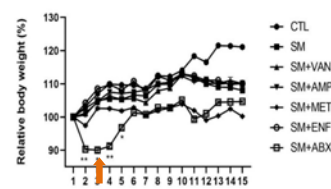
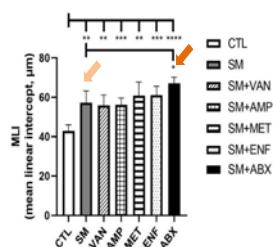
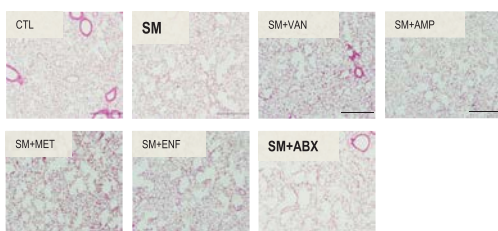
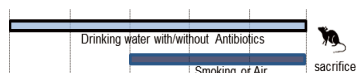


Jang YO, Lee SH & Lee SW et al. Exp Mol Med. 2020

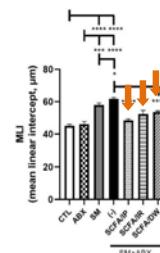
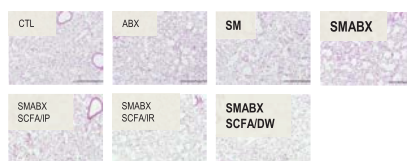
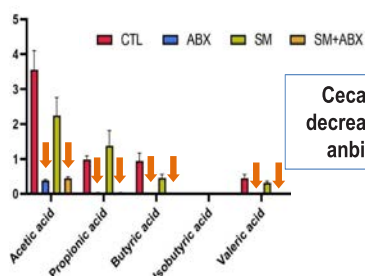


## Antibiotics induced emphysema aggravation: improved with SCFA

Emphysema  
aggravation with  
antibiotics



SCFA improves  
emphysema



Kim NH et. al. Exp Mol Med 2023



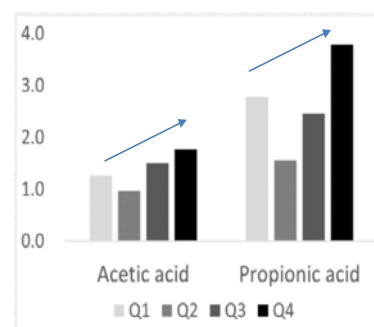
Na Hyun Kim, MS

## Healthy smoker/ emphysema: Gut microbiota & metabolome

Healthy smoker/ sever emphysema

	Asymptomatic smoker	Emphysema patients	P value
Age, yrs	66.20 $\pm$ 6.18	65.44 $\pm$ 6.11	0.93
Height, cm	165	165	0.060
Body weight, kg	80.75 [66.6-88.0]	52.25 [46.5-61.0]	0.0002
BMI, kg/m <sup>2</sup>	27.73 [23.0-30.7]	19.35 [17.7-20.5]	0.0003
Smoking status			
Pack-year	50 [37-60]	45 [35-55]	0.578
Current smoker	5 (100)	7 (43.7)	0.76
Ex-smoker	0	0	
mMRC	0.50 $\pm$ 0.70	3.19 $\pm$ 0.98	<0.0001
CAT score	N/A	26.43 $\pm$ 7.13 <sup>†</sup>	
Spirometry			
FVC measured	2.76 $\pm$ 0.41	2.7 $\pm$ 0.41	
FVC % predicted	2.63 [2.50-3.14]	0.81 [0.57-0.97]	
FEV <sub>1</sub> measured	0.8 $\pm$ 0.2	0.8 $\pm$ 0.2	
FEV <sub>1</sub> % predicted	86.80 $\pm$ 7.18	26.38 $\pm$ 9.44	
FEV <sub>1</sub> /FVC	28.1 $\pm$ 5.5	28.1 $\pm$ 5.5	
DLCO, ml/min/mmHg	82.10 $\pm$ 11.23	30.19 $\pm$ 9.35	<0.0001
DLCO % predicted			

Fiber intake and metabolite



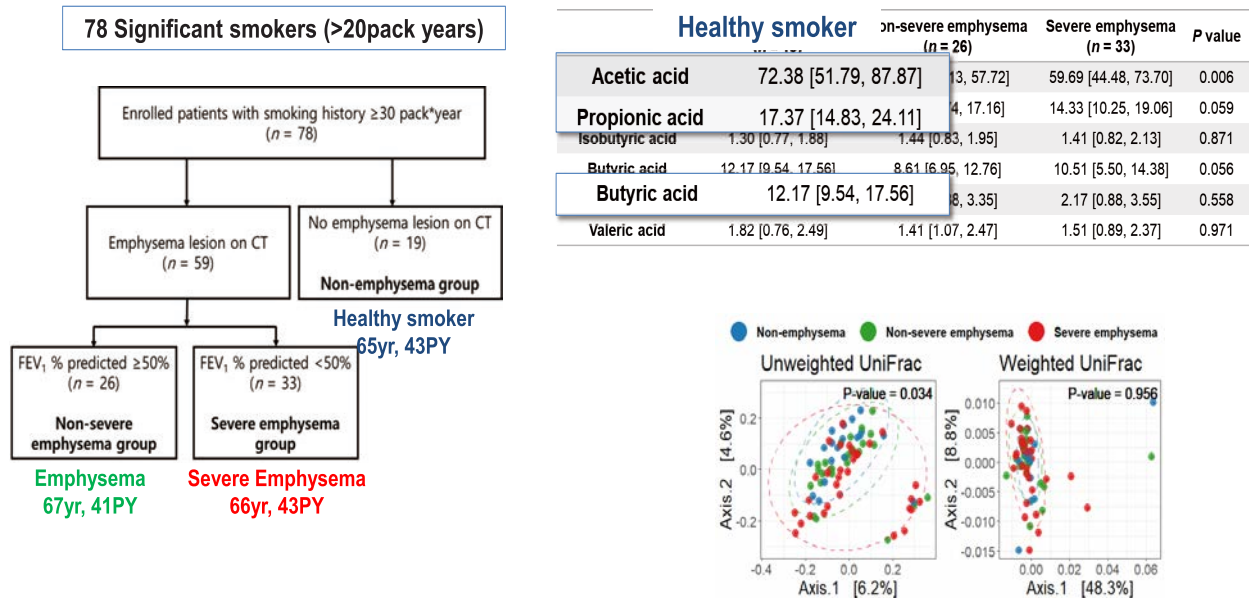
Lee SH, Kim JS & Lee SW et. al. Thorax 2023



Se Hee Lee, MD PhD

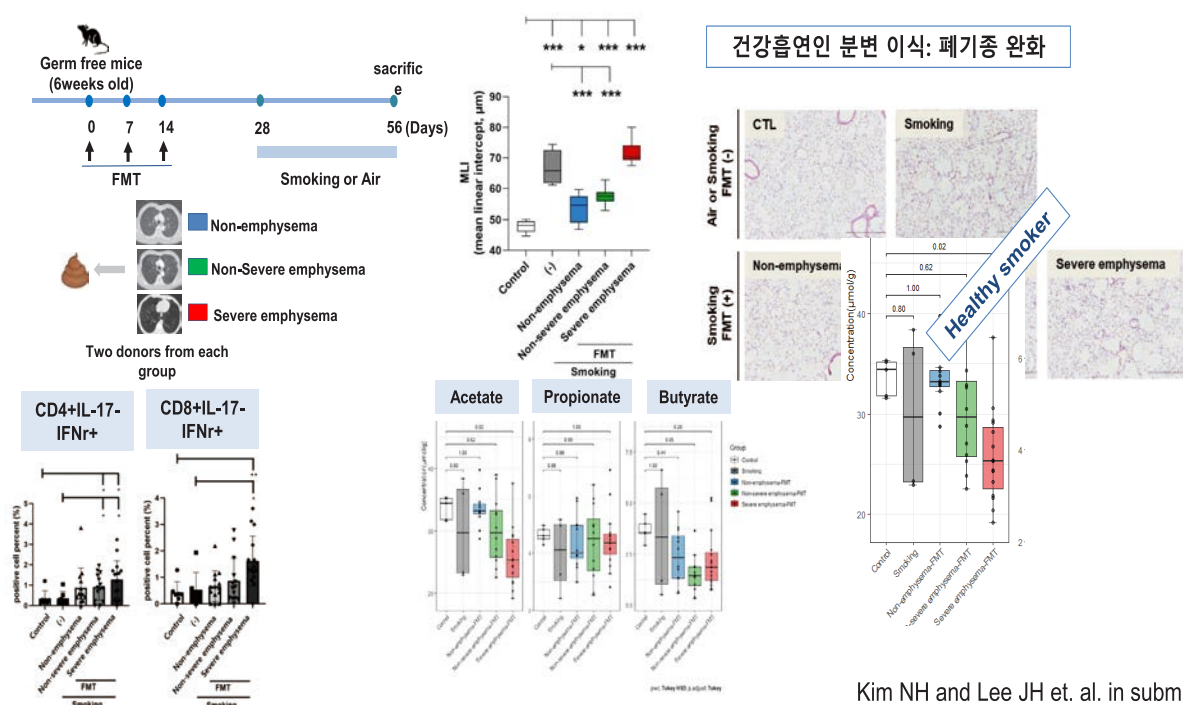
- 154 -

## Different gut microbiome & metabolome as emphysema in smokers



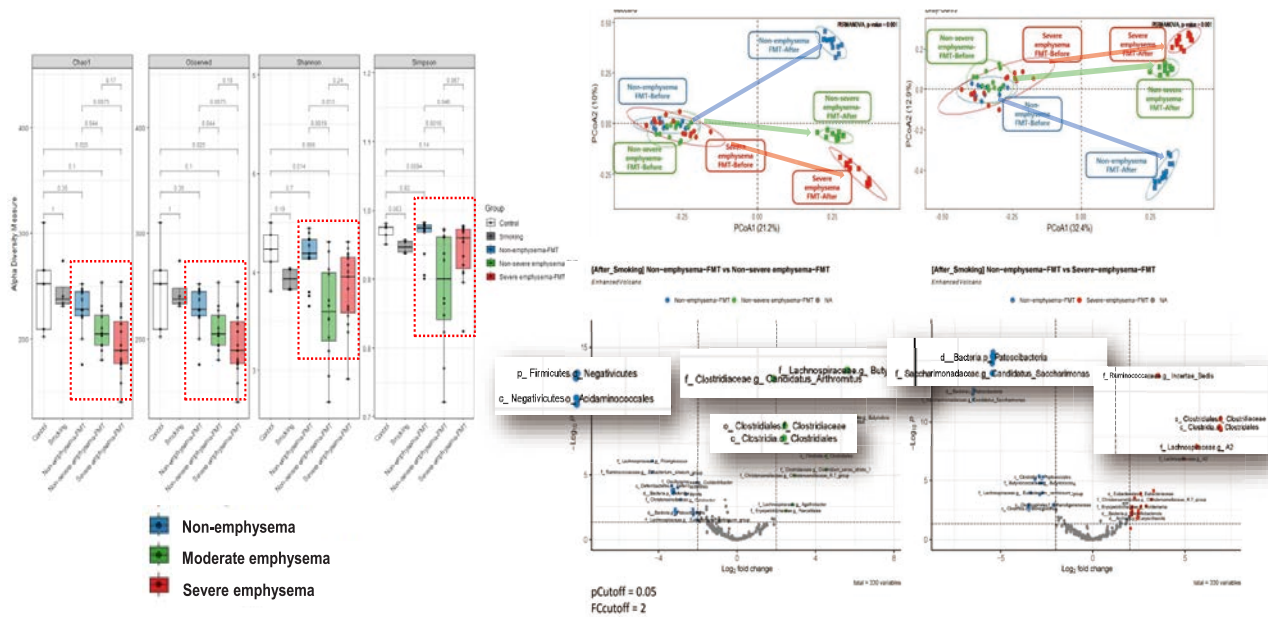
Kim NH and Lee JH et. al. in submission

## FMT from healthy smoker attenuated emphysema in animal model

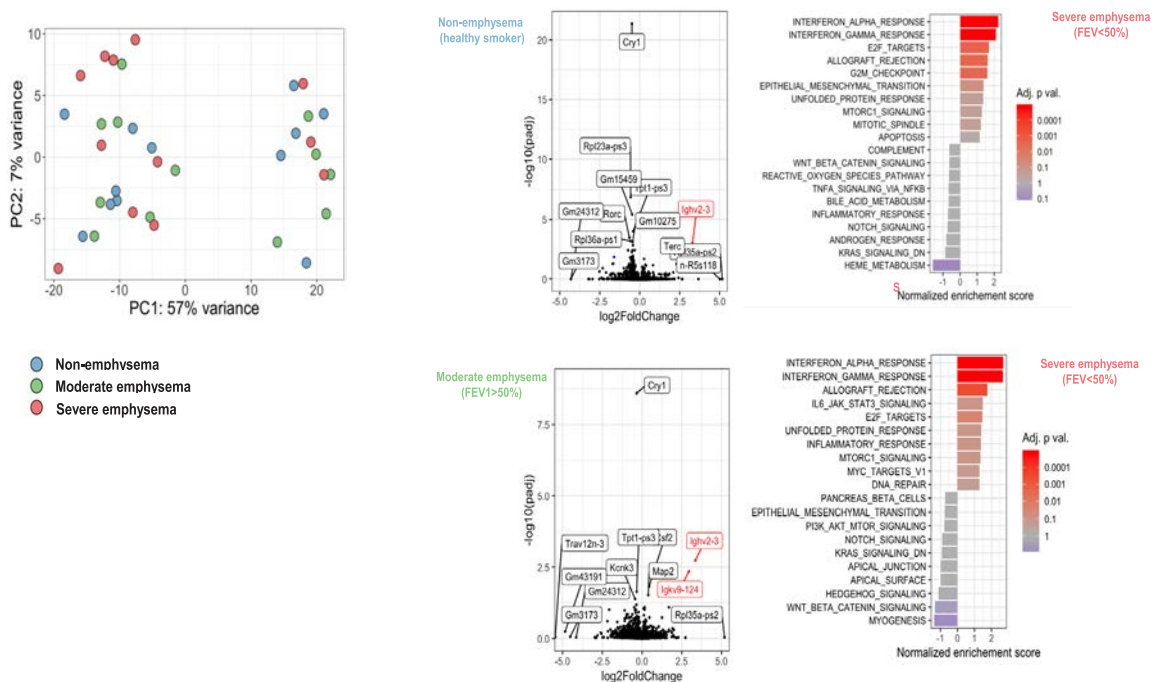


Kim NH and Lee JH et. al. in submission

## 건강흡연인/폐기종 분변 Germ-free mouse 분변 microbiome 분석



## Germ free mice RNA seq 기전 분석



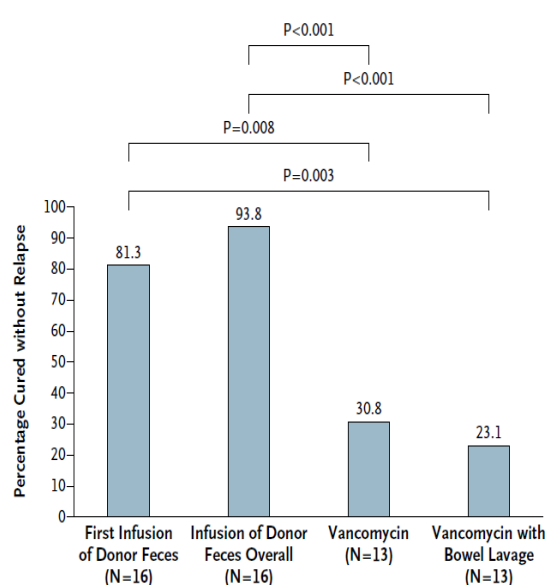
## Contents

### 1. Microbiome in COPD

### 2. Gut-Lung Axis

### 3. Clinical application of microbiome in COPD

## Duodenal Infusion of Donor Feces for Recurrent *C. difficile*

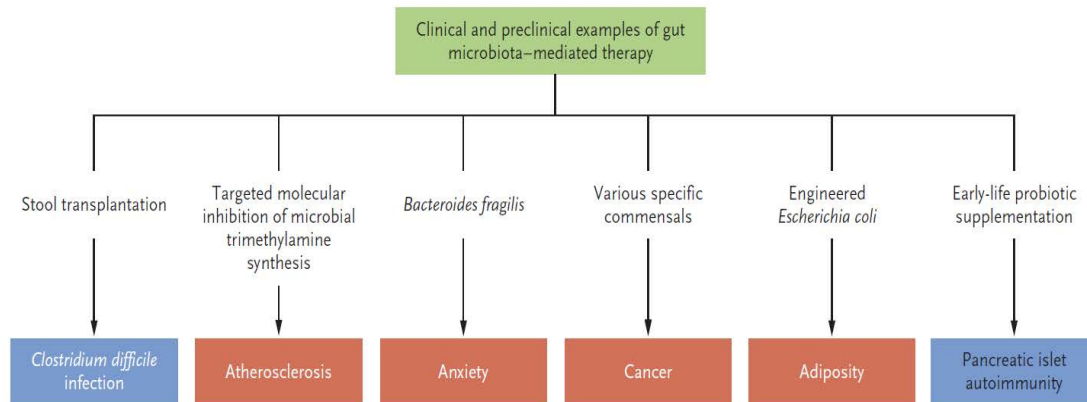


**Table 2.** Adverse Events in 16 Patients in the Infusion Group.\*

Adverse Event	On Day of Infusion of Donor Feces	During Follow-up
	no. of events	
Belching	3	0
Nausea	1	0
Vomiting	0	0
Abdominal cramps	5	0
Diarrhea	15	0
Constipation	0	3
Abdominal pain	2 (associated with cramping)	0
Infection	0	2†
Hospital admission	NA	1‡
Death	0	0
Other adverse event	1§	1‡



## Therapeutic application of Gut Microbiome



N Engl J Med 2016;375:2369-79.

## Four factors which can modulate Microbiome

### Prebiotics

**Food or food substrates**, which increases beneficial bacteria. Non-digestable fiber or sugar. Resistant to gastric acid and enzyme. Poorly absorbed

### Probiotics

**Beneficial Microorganism**, which survives gastric acid and pancreatic enzyme, competes with pathological bacteria.

### Postbiotics

**Non-viable bacteria product, metabolic byproduct of probiotic microorganism**, safer than probiotics

### FMT

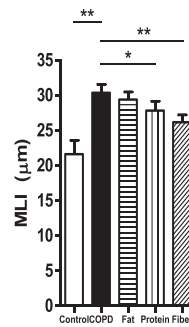
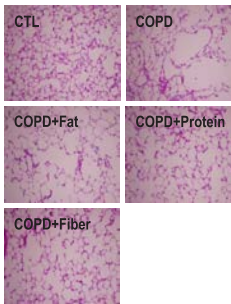
**Gut microbiota with fecal material.** Colonize more effectively than probiotics



## Prebiotics

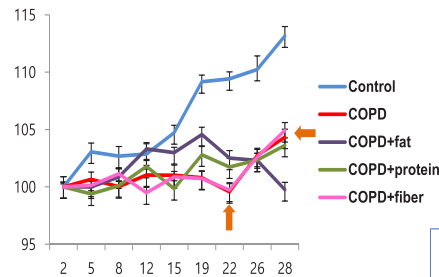
### High fiber diet attenuated emphysema in smoking exposed model

#### Various diets in animal model



#### High fiber diet recovered body weight

##### Relative body weight (%)

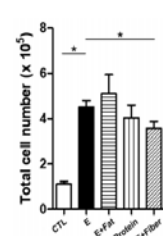


#### Smoking exposed emphysema model

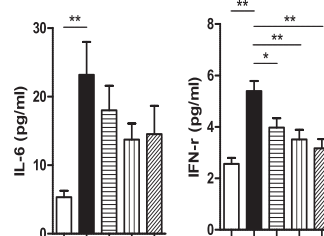


#### Inflammatory marker reduced in high fiber diet

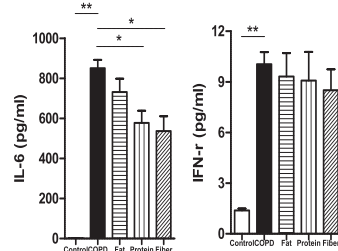
##### Cell count



##### Serum



##### BAL sup

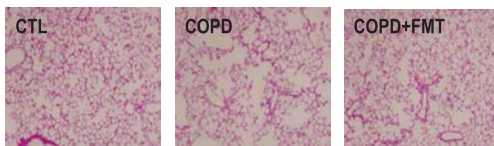


Jang YO & Lee SW et al. Exp Mol Med. 2020

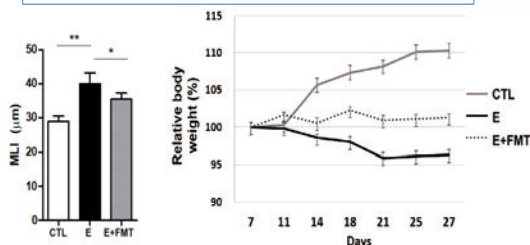
## FMT

### Emphysema improvement with FMT from healthy mice

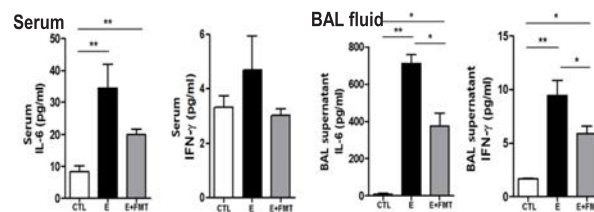
#### FMT from healthy mice improved emphysema



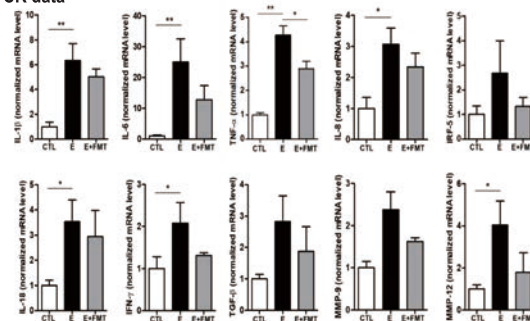
#### FMT from healthy mice improved weight reduction in emphysema model



#### FMT from healthy mice improved inflammatory markers



##### Real time PCR data

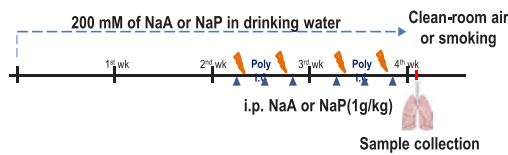


Jang YO & Lee SW et al. Exp Mol Med. 2020

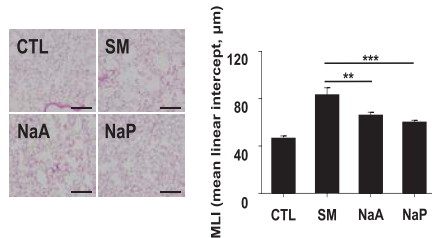
## Postbiotics

### Postbiotics improved emphysema

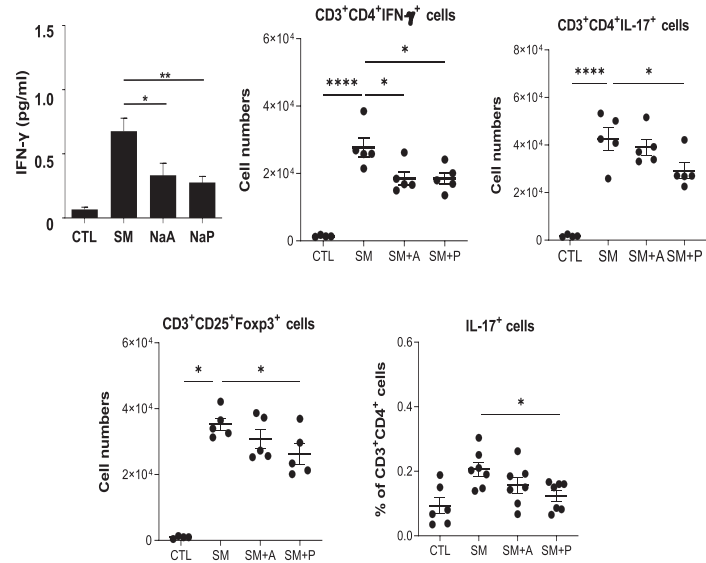
Experimental model:  
Acetate/Propionate



Postbiotics: Emphysema  
improvement



Postbiotics: Emphysema inflammatory  
marker reduced



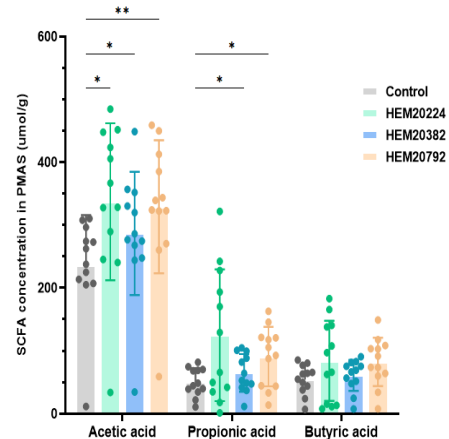
Lee SH, Kim JS & Lee SW. Thorax 2023

## Probiotics

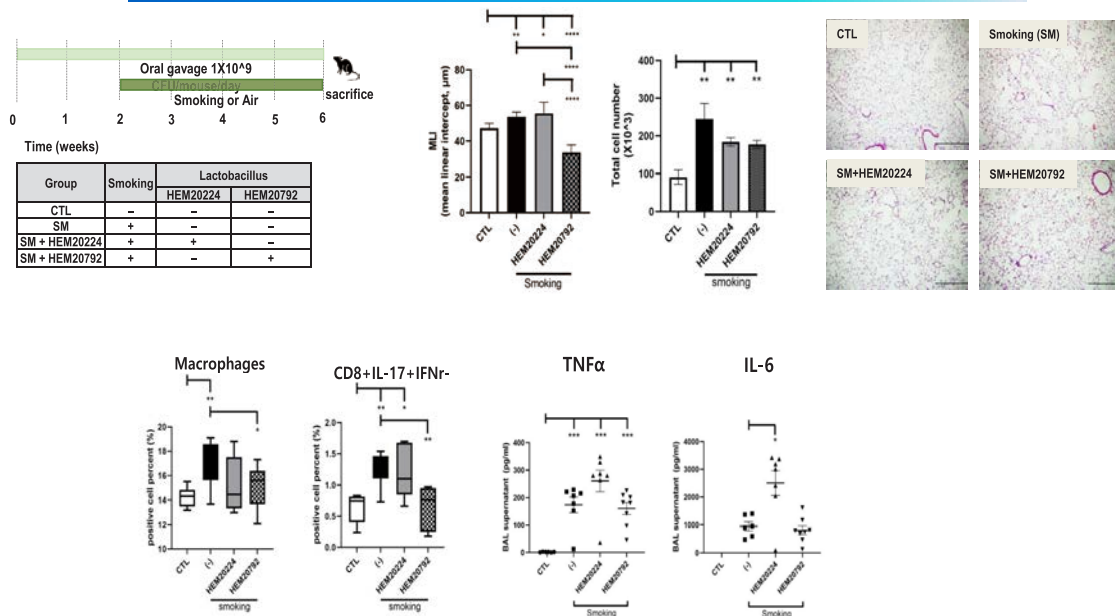
### PMAS 모델 통한 균주 screening



Feces donors for PMAS experiments (n = 12)	
Male, n (%)	12 (100.0%)
Age, years	68.3 $\pm$ 5.8
Body mass index, kg/m <sup>2</sup>	20.5 $\pm$ 2.6
Current smoker, n (%)	1 (8.3%)
Smoking amount, pack*years	53.7 $\pm$ 22.0
Pulmonary function test	
FEV <sub>1</sub> measured, L	1.21 $\pm$ 0.65
FEV <sub>1</sub> % predicted	37.8 $\pm$ 16.0
FVC measured, L	83.0 $\pm$ 11.9
FVC % predicted	33.3 $\pm$ 13.8
FEV <sub>1</sub> /FVC	6.5 $\pm$ 3.3
DL <sub>CO</sub> mL/min/mmHg	30.4 $\pm$ 11.6
Inhaler use	
LAMA+LABA	8 (66.7%)
Triple therapy	4 (33.3%)
n = 6	
Any exacerbation in the previous year	0 (0.0%)
Moderate to sev exacerbation in the previous year	0 (0.0%)
COPD assessment test score	16.0 $\pm$ 3.8

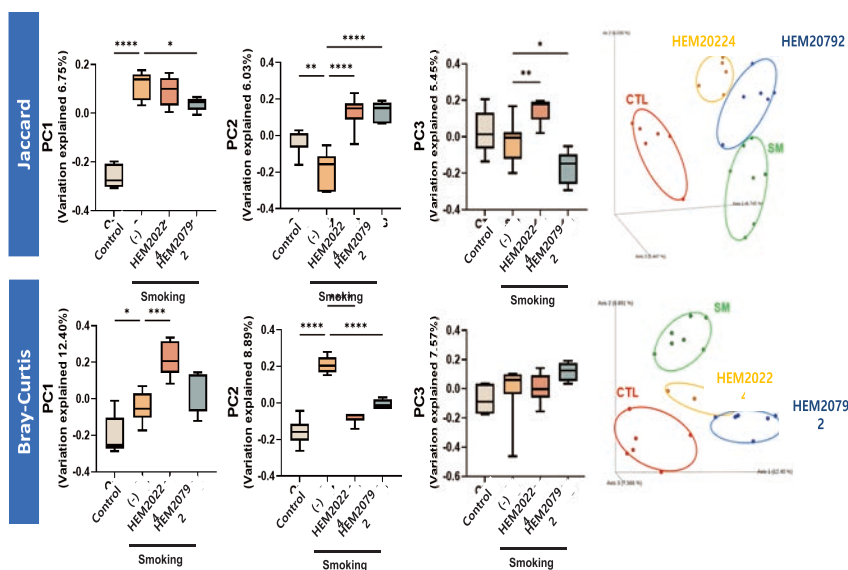


## LBP emphysema model에 투여

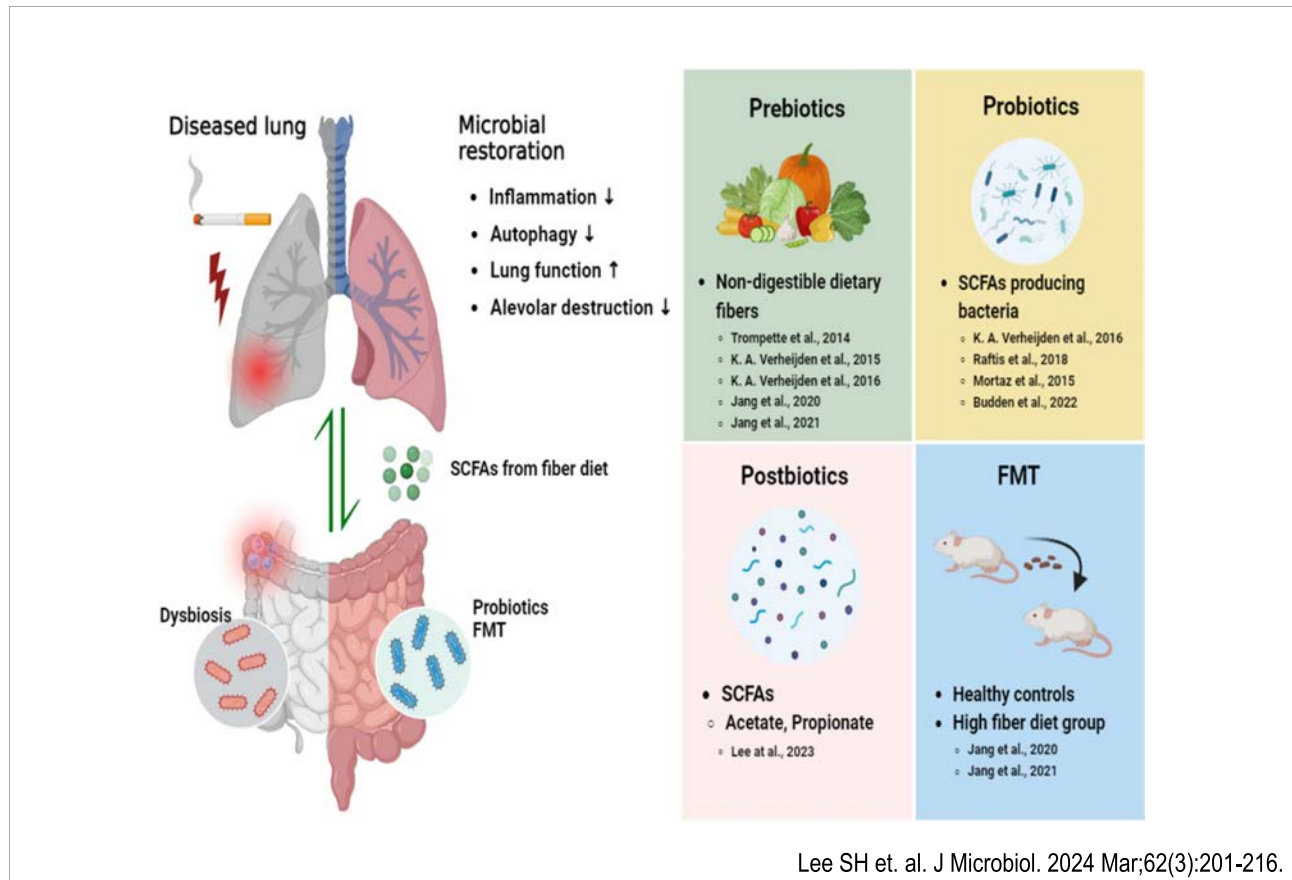
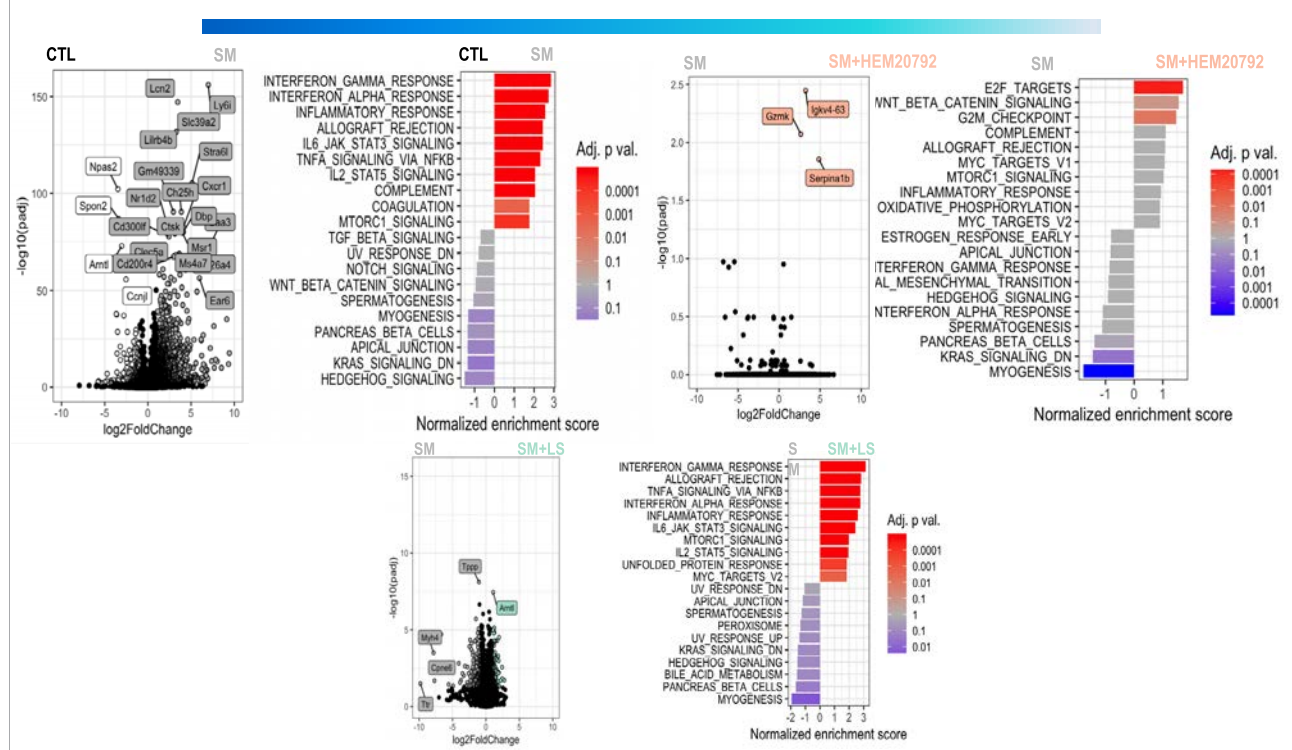


## PMAS 분석으로 제시된 LBP 투여의 microbiome 변화

LBP 후보 물질투여에 따른 microbiome 변화



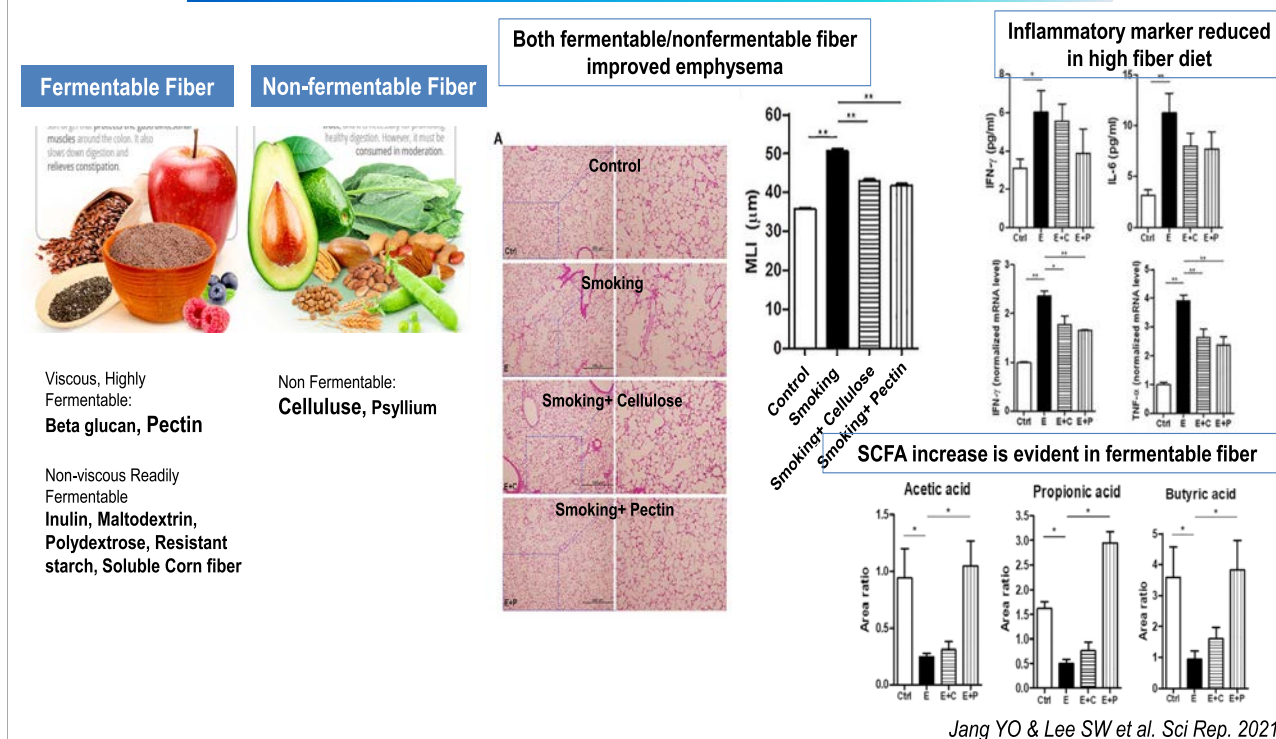
## LBP 투여 후 폐기종 호전의 기전 탐색: Bulk RNA seq



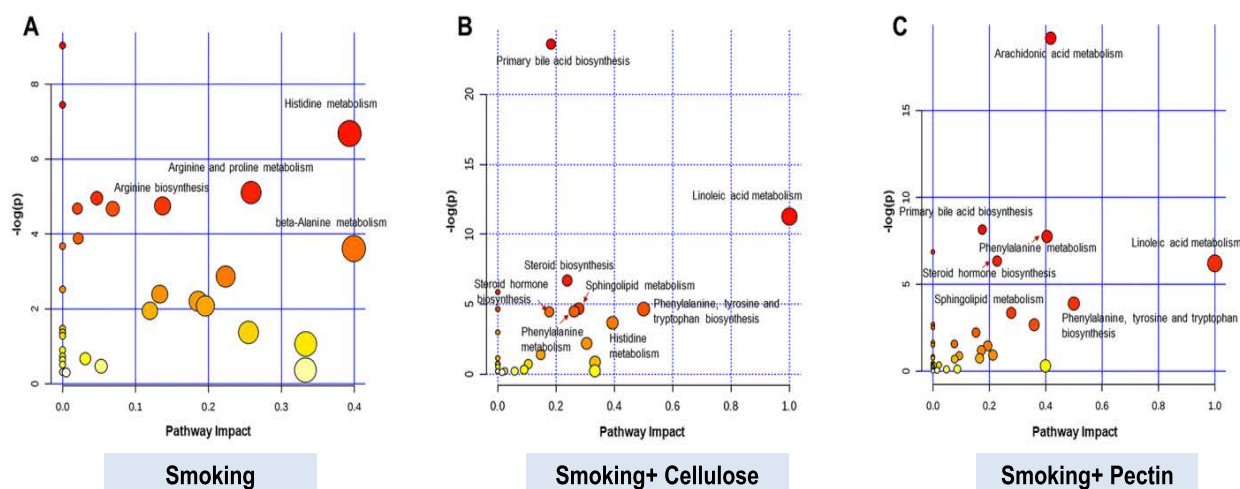
Lee SH et. al. J Microbiol. 2024 Mar;62(3):201-216.



## High fiber diet attenuated emphysema in smoking exposed model



## High fiber diet attenuated emphysema in smoking exposed model



## Summary

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- 1. High fiber diet has protective effect in both animal and human**
- 2. Gut microbiota affects respiratory diseases**
- 3. Recent development in microbiome technique showed potential in COPD treatment**



# 04

## Gut–liver axis in health & disease



석기태  
(한림대학교춘천성심병원)





## 2부 연자



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- ✓ 성명: 석기태

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2008년 3월-2010년 8월	연세대학교 의과대학 의학박사
2014년 9월-2016년 6월	COLUMBIA UNIV. POST-DOC, USA

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2009년~2009년	도쿄 토라노몬 병원	방문 의사
2009년~2010년	성지병원	내과장
2014년~2016년	COLUMBIA UNIV.	POST DOC research scholar
2010년 3월 1일~현재	한림대학교 춘천성심병원 과장 임상초음파학회 교육이사 대한간학회 강원지회 학술이사 한림대학교 소화기연구소 교수	

# GUT-LIVER-AXIS

Nonalcoholic fatty liver disease (NAFLD) is one of the most common types of liver disease worldwide. NAFLD includes a broad spectrum of disorders, including steatosis, nonalcoholic steatohepatitis (NASH), fibrosis and cirrhosis. Human gut microbiota is an ecological community comprised of commensal, symbiotic, and pathogenic microorganisms totaling 1–2 kg in weight. Gut microbiota are integral for immunological, hormonal, and metabolic homeostasis of the host. The close relationship between the gut and liver appears to be a crucial factor in liver injury. The liver receives most of its blood and nutritional supply from the gut through the portal vein and is the first organ to be exposed to gut-derived toxic factors, including bacteria, damage-associated metabolites (i.e., damage-associated molecular patterns), and bacterial products (i.e., pathogen-associated molecular patterns [PAMPs]). Dysbiosis is defined as quantitative and qualitative changes in intestinal bacteria, and small intestine bacterial overgrowth can both lead to an increase in intestinal permeability and translocation of endotoxins to the portal tract, which activates the signal pathways of a wide array of inflammatory cytokines in the liver.

Alcoholic liver disease (ALD) is responsible for approximately 25% of deaths resulting from alcohol consumption. Activation of Kupffer cells has been identified as an essential element in the pathogenesis of ALD. Alcohol induces bacterial overgrowth and the translocation of the endotoxin lipopolysaccharide (LPS) from the gut to the liver. Alcohol has been known to disrupt the gut barrier function, which consequently promotes the translocation of microbial LPS from the lumen of the intestines to the portal vein, where it travels to the liver. Kupffer cells and macrophages recruited to the liver can be activated by bacterial endotoxin such as LPS through toll-like receptor 4. Moderate alcohol consumption has also been identified as a strong risk factor for small intestinal bacterial overgrowth.

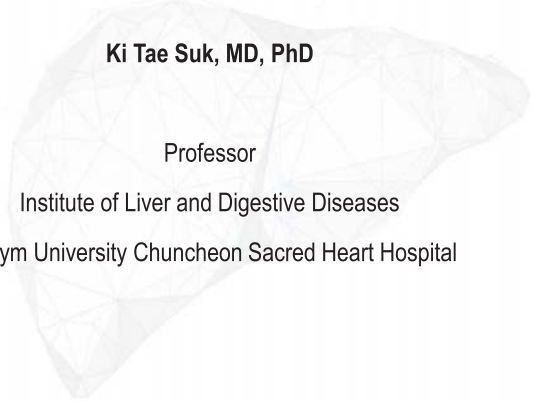
Recently, probiotics, prebiotics, antibiotics, fecal transplantation, and ETC. have been shown to be effective in reducing or preventing liver inflammation. A potential mechanism is transforming the composition of intestinal microbiota, which leads to reductions in dysbiosis, intestinal permeability, bacterial translocation, endotoxemia, and consequently, the development of inflammation.



# Gut-liver axis

Ki Tae Suk, MD, PhD

Professor  
Institute of Liver and Digestive Diseases  
Hallym University Chuncheon Sacred Heart Hospital



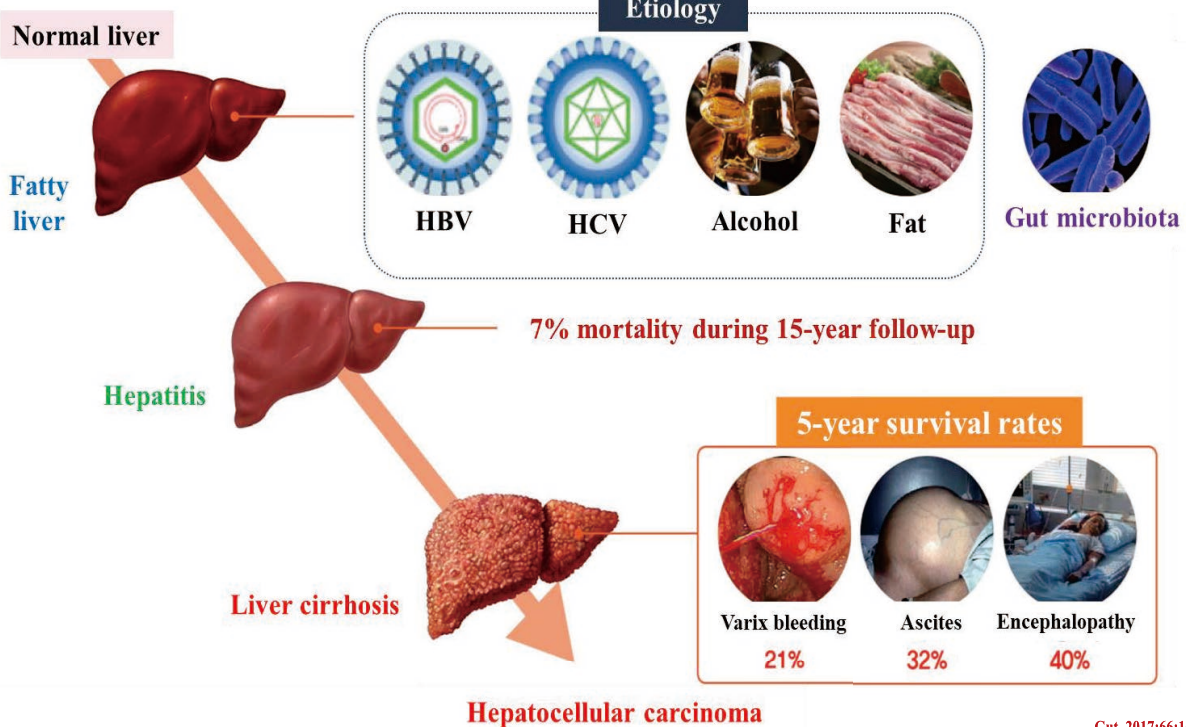
## **CONTENTS**

- **Liver disease**
- **Microbiome in Liver disease**
- **Microbial - precision medicine in Liver disease**
- **Conclusion**

# Liver disease



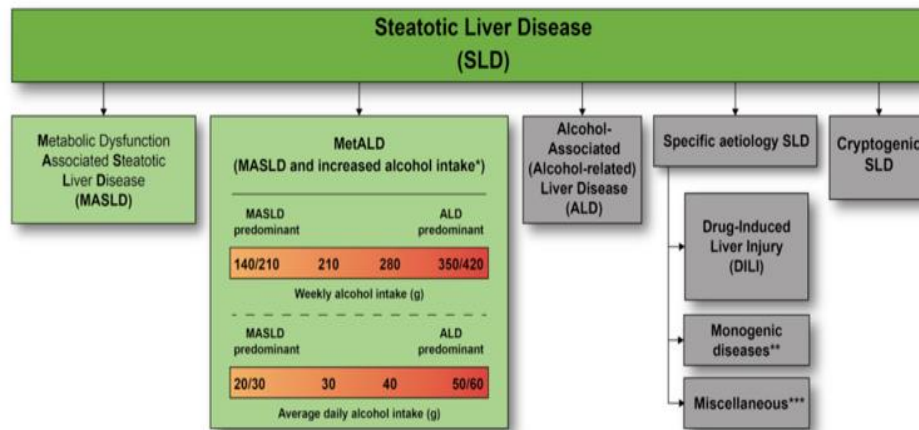
## Liver disease



Gut. 2017;66:1688-1696.  
Hepatology. 2016;64:954.



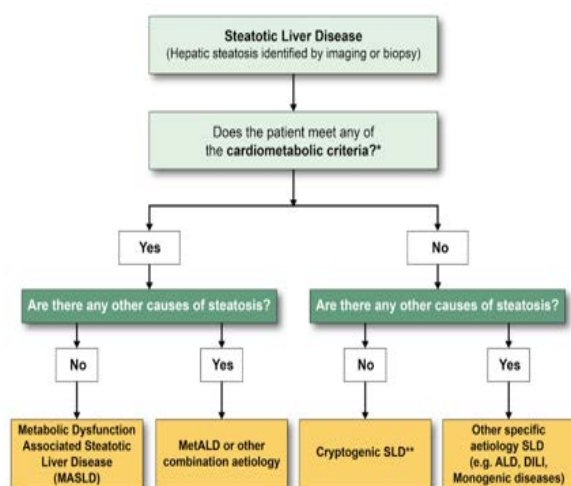
## Steatotic Liver Disease (SLD): new terminology



- The term **steatohepatitis** was felt to be an important pathophysiological concept that should be retained.
- NAFLD → metabolic dysfunction-associated steatotic liver disease (**MASLD**).
- NASH → metabolic dysfunction-associated steatohepatitis (**MASH**).
- **MetALD**: MASLD and increased alcohol intake (**male > 210 g/wk; female > 140 g/wk**).

<https://www.aasld.org/new-naflid-nomenclature/about-steatotic-liver-disease-sld>

## MASLD: cardiometabolic criteria

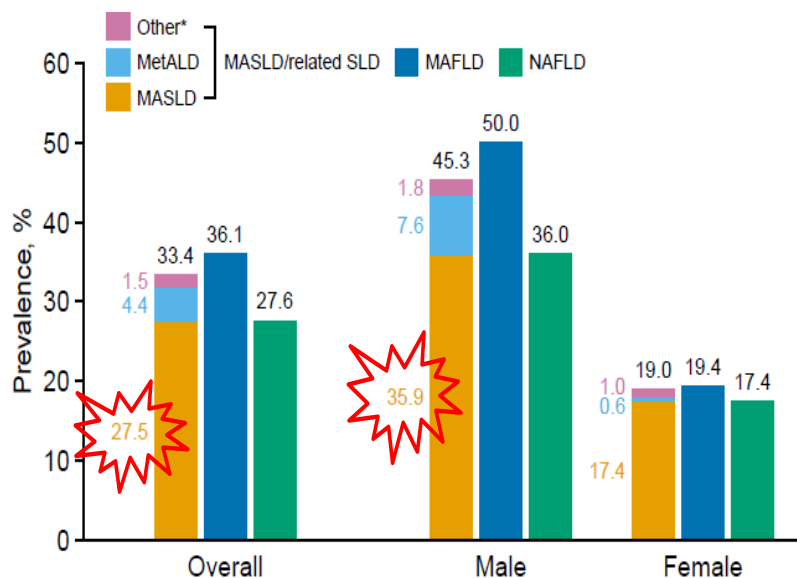


- (1) **BMI**  $\geq 25$  kg/m<sup>2</sup> ( $\geq 23$  kg/m<sup>2</sup> in Asian) or **waist circumference**  $> 94$  cm in men,  $> 80$  cm in women, or ethnicity adjusted
- (2) **Fasting serum glucose**  $\geq 100$  mg/dL ( $\geq 5.6$  mmol/L) or 2-hour post-load glucose level  $\geq 140$  mg/dL ( $\geq 7.8$  mmol/L) or **HbA1c**  $\geq 5.7\%$  or on specific drug treatment
- (3) **Blood pressure**  $\geq 130/85$  mmHg or specific drug treatment
- (4) **Plasma triglycerides**  $\geq 150$  mg/dL ( $\geq 1.70$  mmol/L) or specific drug treatment
- (5) **Plasma HDL cholesterol**  $< 40$  mg/dL ( $< 1.0$  mmol/L) for men and  $< 50$  mg/dL ( $< 1.3$  mmol/L) for women or specific drug treatment

SLD patients meet any of the cardiometabolic criteria → MASLD

J Hepatol. 2023 Jun 20:S0168-8278(23)00418-X.

## Prevalence of SLD: Korea



- Approximately, one-third of Korean adults are classified as having MASLD and related SLD
- The presence of MASLD and related SLD is associated with a higher risk of cardiovascular disease.

Lee HH, Lee HA, Lee HK., Kim SU. Gut 2023

## Microbiome in Liver disease

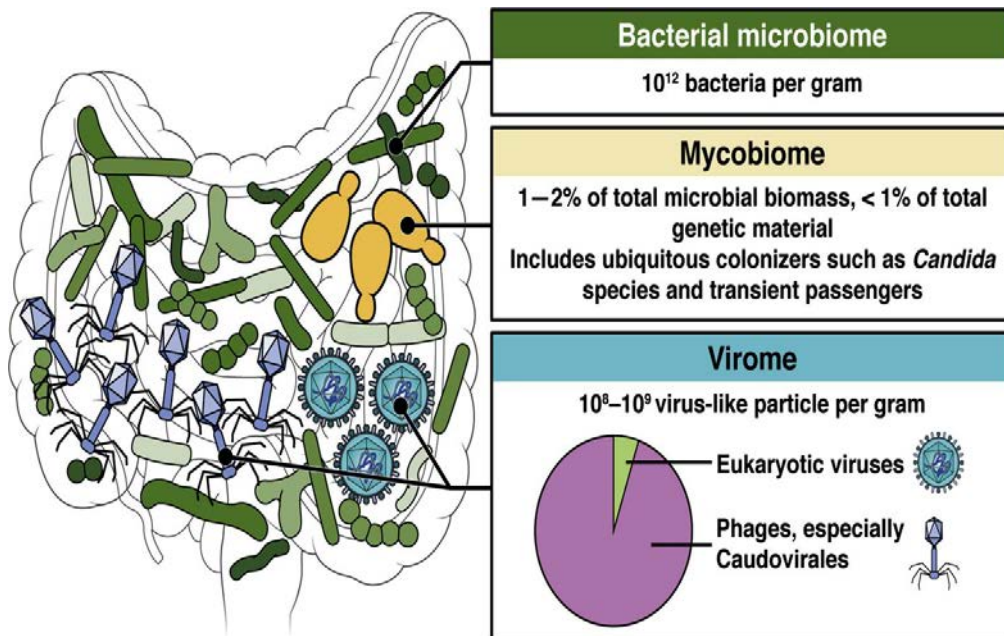


## Microbiome



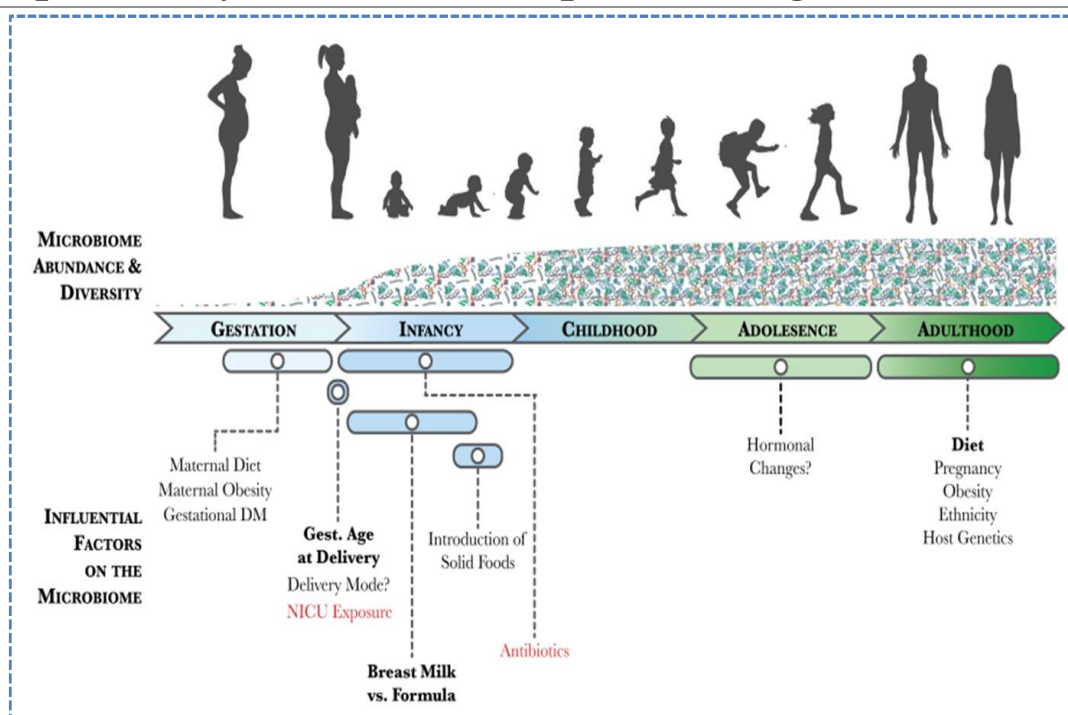
Community of microorganisms that can be found living together in any given habitat.

1988 by Whipps *et al.*



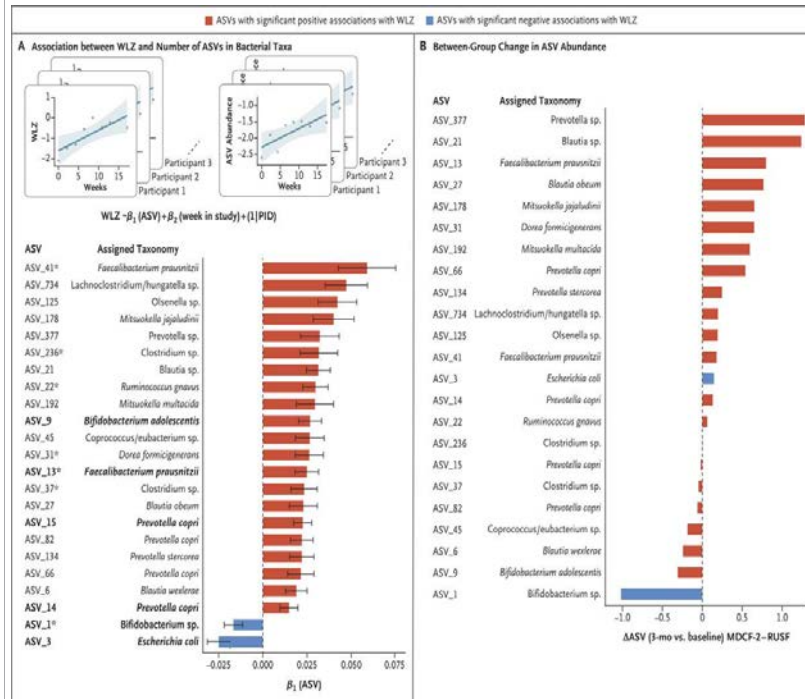
<https://doi.org/10.1053/j.gastro.2020.06.100>

## Complex ecosystem (>>1,400 species, >1kg)



Various factors affect to the microbial variation.

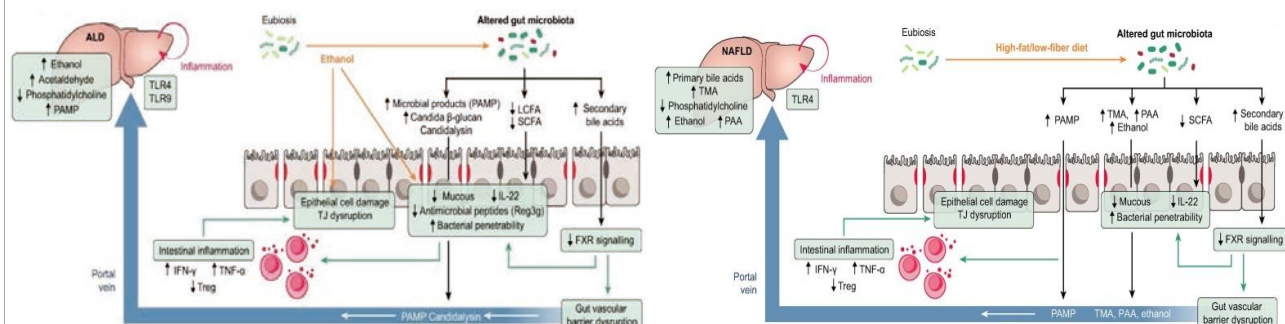
[Vincent B Young, 2017]



**RY Chen et al. N Engl J Med 2021;384:1517-1528.**



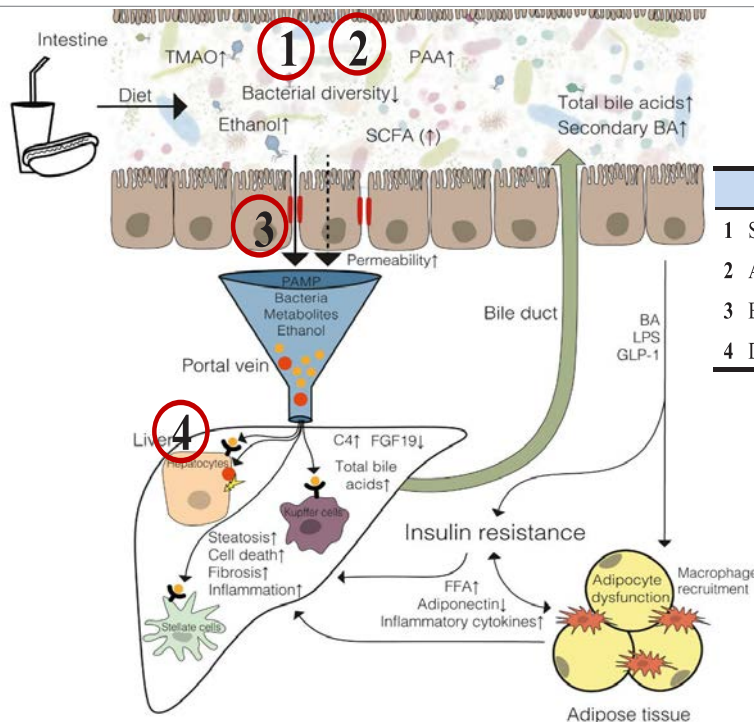
**Dysbacteriosis**, is characterized by a disruption to the microbiome resulting in an imbalance in the microbiota, changes in their functional composition and metabolic activities, or a shift in their local distribution



**J Hepatol. 2020 Mar;72(3):558-577.**

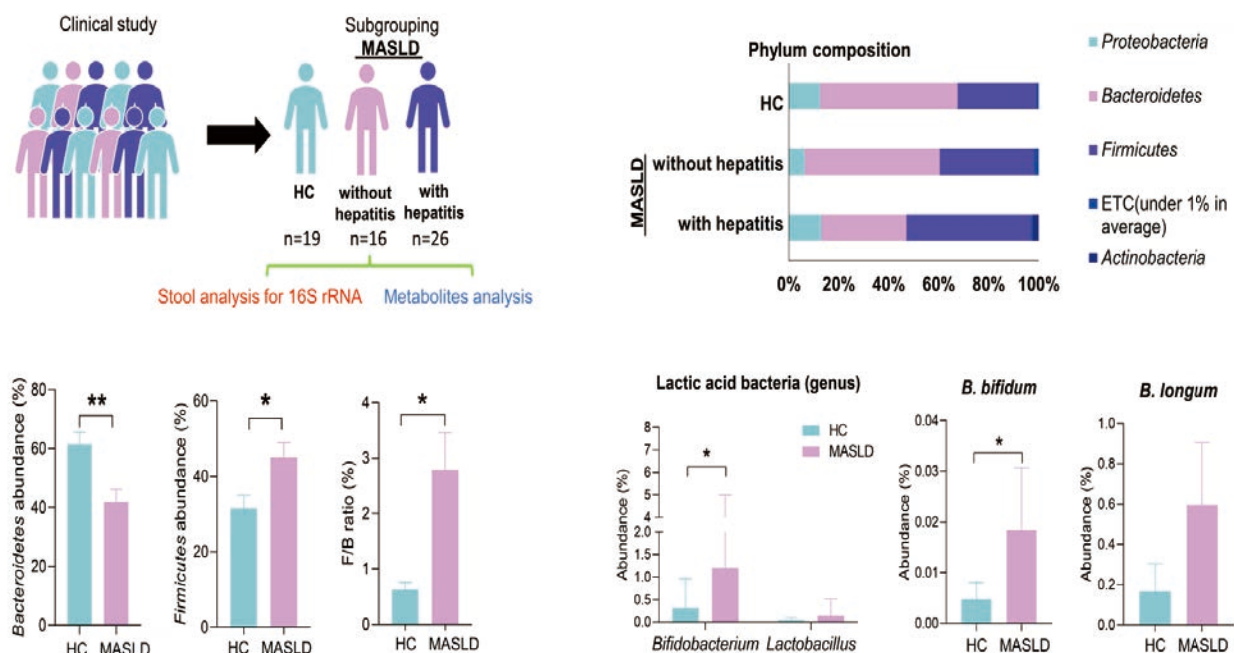


## Pathophysiology of MASLD



Bernd Schnabl. Cell Host Microbe 2020

## MASLD: microbiota diversity



Min BH, Suk KT Gut microbes 2024

## MTPSET composition species rank

### MASLD

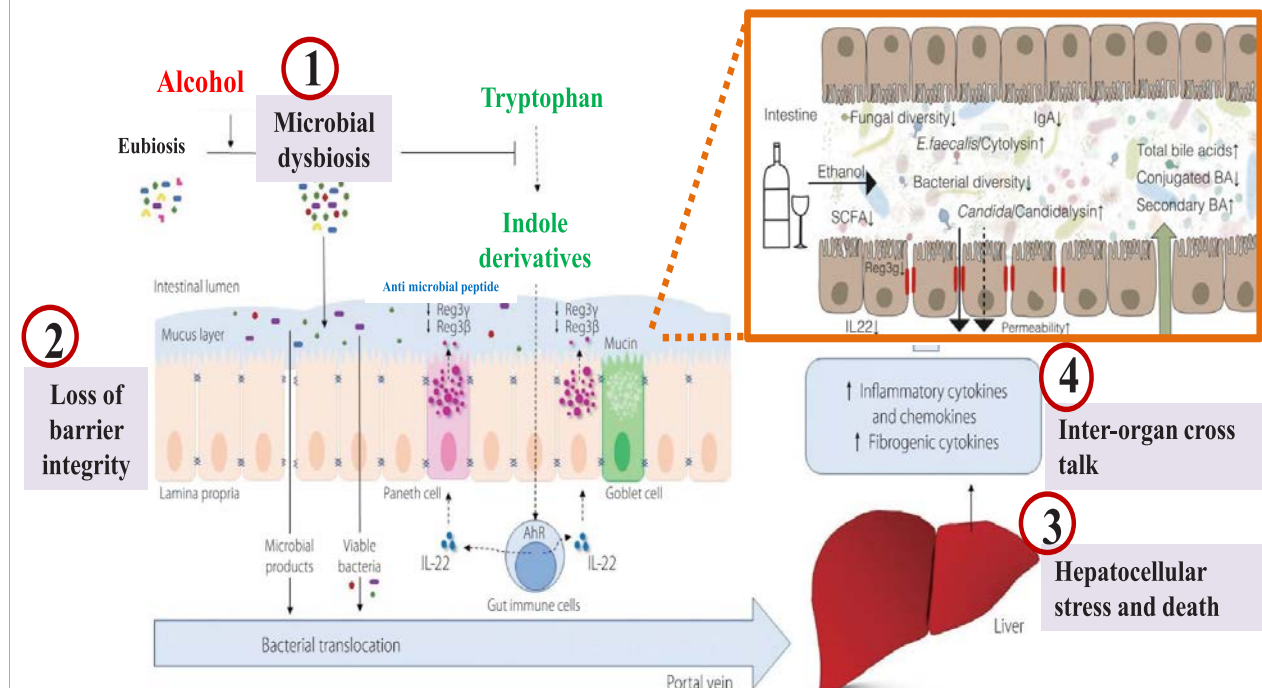
Disease progression

Normal		Fatty liver		MASH	
Taxon Name	Ratio	Taxon Name	Ratio	Taxon Name	Ratio
<i>Prevotella uc</i>	1.115	<i>Prevotella uc</i>	9.044	<i>Escherichia coli</i> group	5.560
<i>Escherichia coli</i> group	6.822	<i>Escherichia coli</i> group	8.236	<i>Faecalibacterium prausnitzii</i> group	5.457
<i>Bacteroides plebeius</i>	6.016	PAC001304 s	6.494	PAC001304 s	5.357
PAC001304 s	4.945	<i>Bacteroides vulgatus</i>	6.303	<i>Prevotella uc</i>	4.379
<i>Faecalibacterium prausnitzii</i> group	4.705	<i>Bacteroides plebeius</i>	5.109	<i>Bacteroides plebeius</i>	3.825
<i>Bacteroides vulgatus</i>	4.627	<i>Faecalibacterium prausnitzii</i> group	4.710	<i>Bacteroides vulgatus</i>	3.627
<i>Bacteroides uniformis</i>	3.305	<i>Bacteroides uniformis</i>	3.778	<i>Veillonella dispar</i>	2.179
<i>Bacteroides coprocola</i>	2.940	<i>Phascolarctobacterium faecium</i>	1.604	<i>Bacteroides uniformis</i>	2.139
<i>Megamonas rupellensis</i> group	1.676	<i>Dialister succinatiphilus</i>	1.589	<i>Phascolarctobacterium faecium</i>	2.131
<i>Phascolarctobacterium faecium</i>	1.259	<i>Bacteroides stercoris</i>	1.526	<i>Bacteroides dorei</i>	1.857
<i>Bacteroides dorei</i>	1.252	<i>Prevotella stercora</i>	1.439	<i>Phascolarctobacterium succinatutens</i> group	1.675
<i>Eubacterium eligens</i> group	1.203	<i>Phascolarctobacterium succinatutens</i> group	1.383	<i>Enterobacteriaceae</i> group	1.426
<i>Bacteroides fragilis</i>	1.199	<i>Bacteroides dorei</i>	1.217	<i>Bacteroides caccae</i>	1.399
<i>Phascolarctobacterium succinatutens</i> group	1.176	<i>Alistipes putredinis</i>	1.203	<i>Lactobacillus rogosae</i> group	1.356
<i>Prevotella stercora</i>	1.139	PAC001276 s	1.139	PAC001039 s	1.346
<i>Veillonella dispar</i>	1.114	<i>Bacteroides coprocola</i>	1.107	<i>Bacteroides coprocola</i>	1.267
<i>Alistipes putredinis</i>	1.114	PAC001046 s	1.018	<i>Dialister succinatiphilus</i>	1.262
<i>Parabacteroides merdae</i>	1.057	<i>Parabacteroides merdae</i>	0.882	<i>Bacteroides stercoris</i>	1.233
<i>Paraprevotella clara</i>	1.050	PAC001292 s	0.826	<i>Roseburia inulinivorans</i>	1.180
<i>Prevotella copri</i>	1.012	KQ968618 s group	0.825	<i>Dialister invisus</i>	1.178

- *Prevotella uc*, *B. plebeius* – good bacteria??
- *E. coli* group – bad bacteria??

Suk KT, Unpublished data

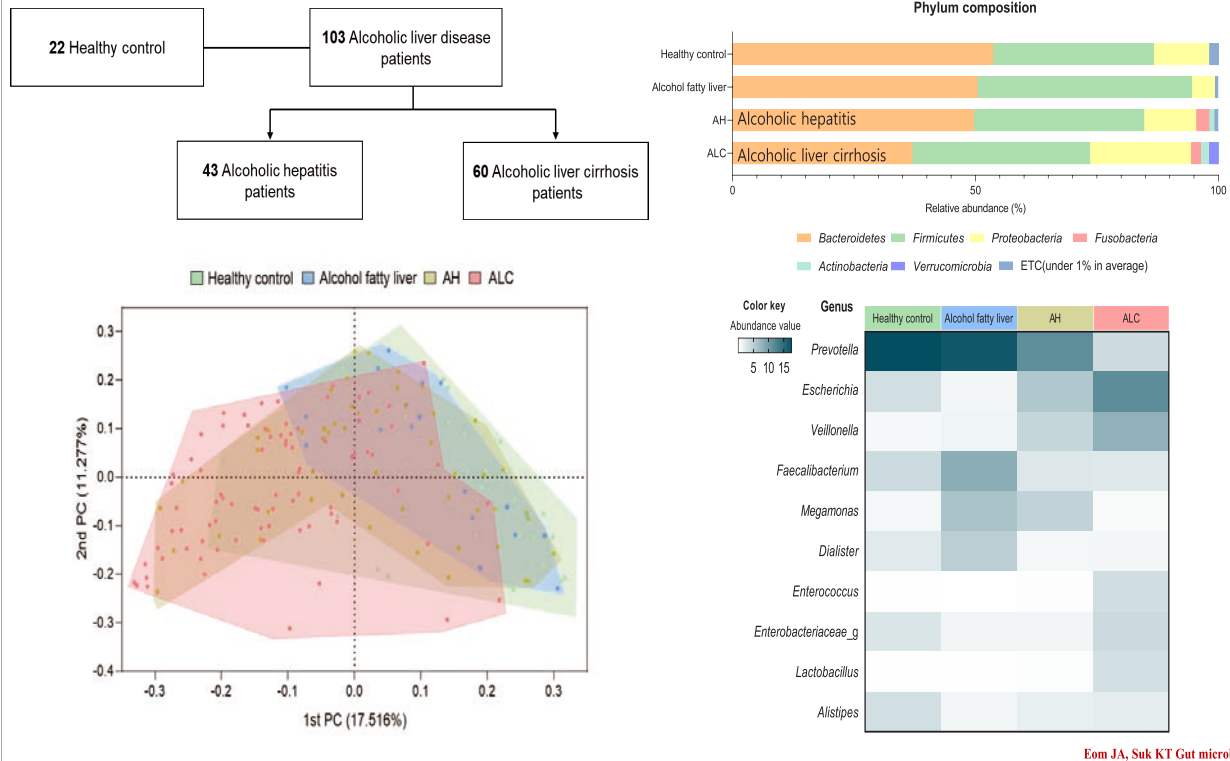
## Pathophysiology of ALD & MetALD



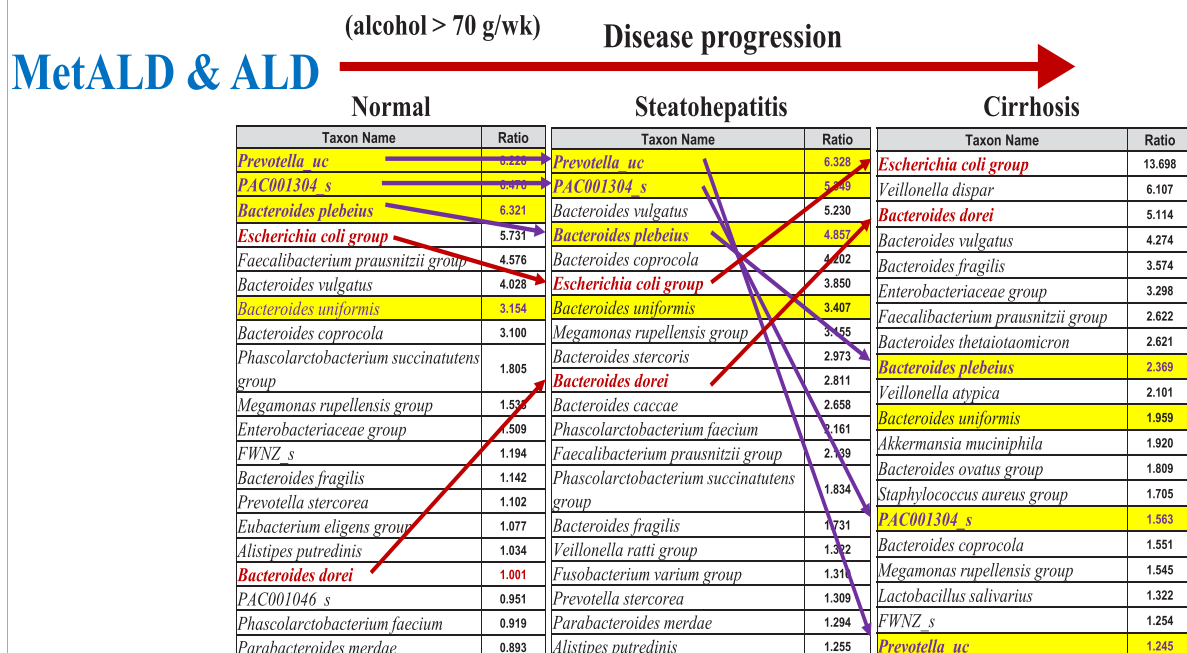
Bernd Schnabl, Clin Mol Hepatol 2020  
Bernd Schnabl, Cell Host Microbe 2020



## MetALD & ALD



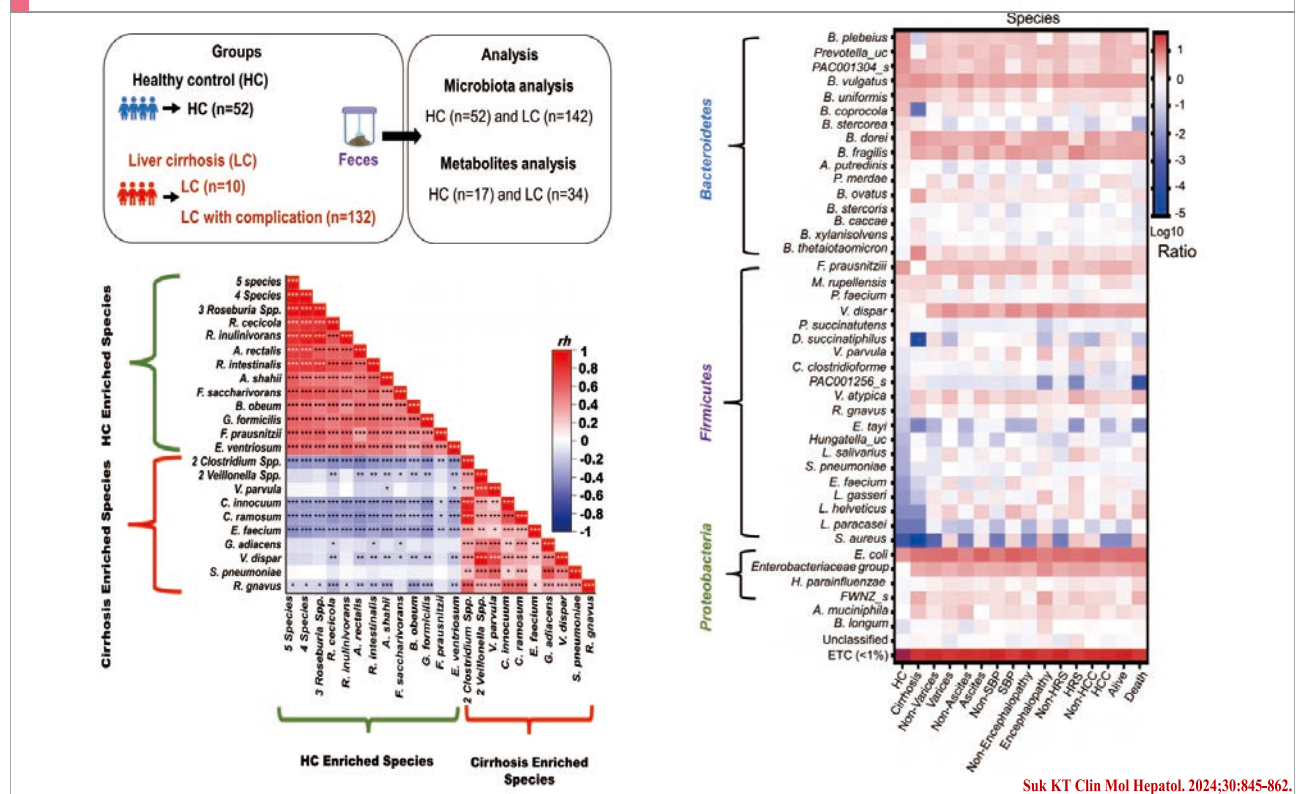
## MTPSET composition species rank



- *Prevotella uc*, *PAC001304*, *B. plebeius* – good bacteria??
- *E. coli* group and *B. dorei* – bad bacteria??

Suk KT, Unpublished data

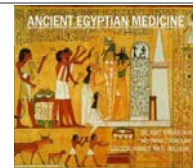
## Liver cirrhosis and complications



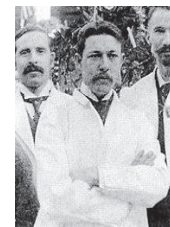
## History: cancer and microbiome

- Treatment for tumors: application of a poultice to the site followed by an incision, causing an infection

Ebers Papyrus, 2600 BC



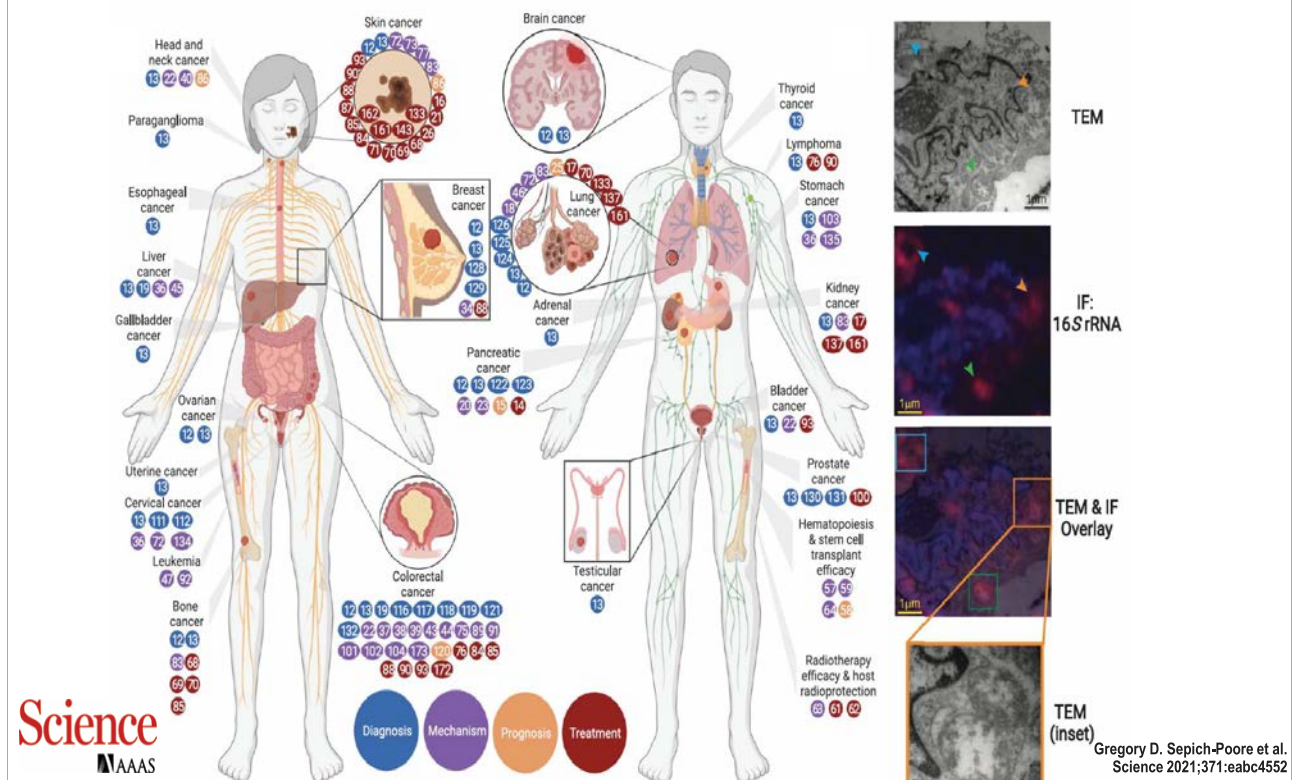
- **Peregrine Laziosi (1265–1345)**  
spontaneous regression of his septic, ulcerative tibial bone malignancy
- **William Coley (1862-1936) - 1<sup>st</sup> intentional demonstration of immunotherapy**  
*Streptococcus* & *Serratia* species on terminal cancer patients  
10-year disease-free survival: ~30%  
**No mechanical evidence, Irreproducible results, and Hazardous therapy**



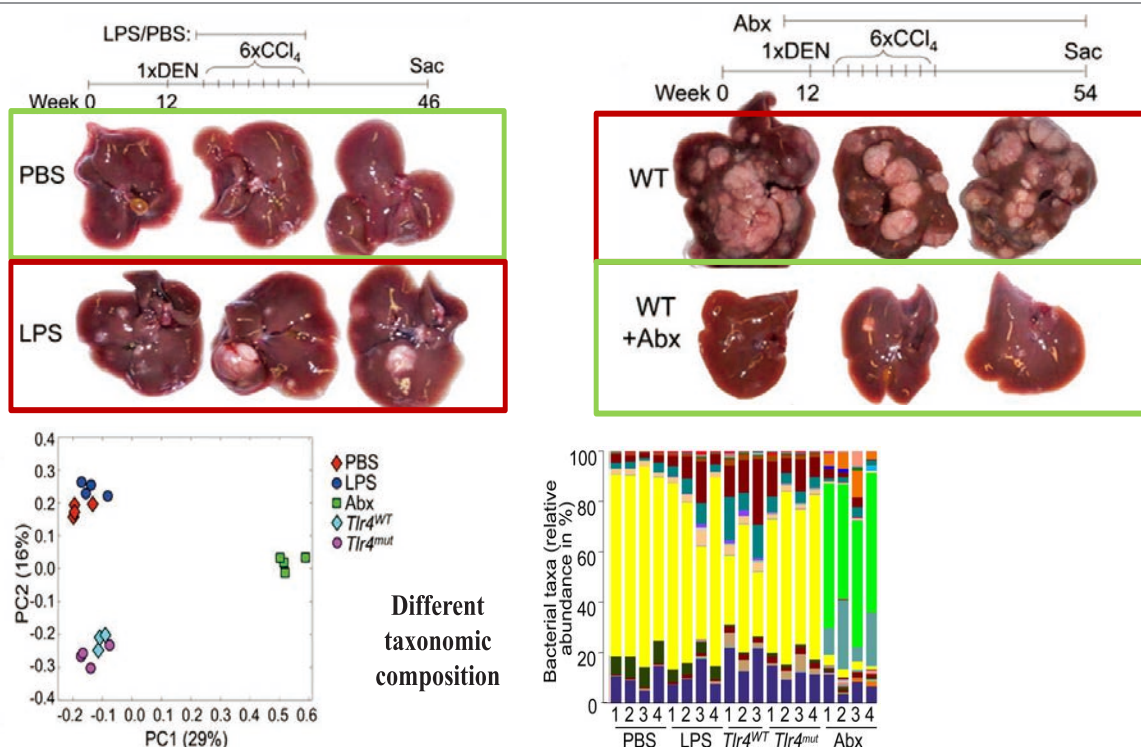
## Microbes may play a broad role in cancer diagnosis, pathogenesis, and treatment

-imaging, sequencing, and genetically engineered and germ-free mouse models-

## Current landscape of the cancer microbiome



## Microbiome is related with hepatocarcinogenesis



Dapito DH, Batailler R, Schwabe RF Cancer Cell 2012

## Summary



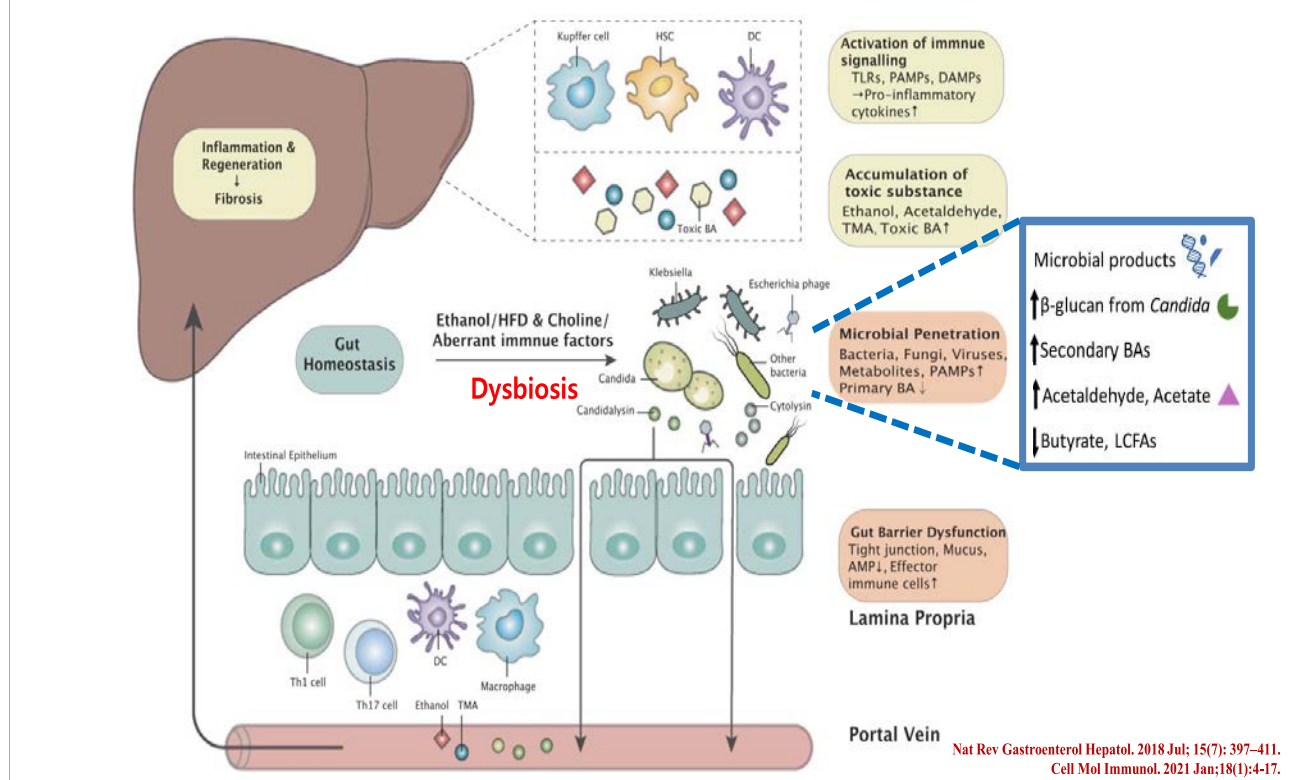
- The composition of microbiome depends on the type of liver disease.
  - normal population **vs.** fatty liver **vs.** hepatitis **vs.** liver cirrhosis
  - MASLD **vs.** ALD **vs.** MetALD
- Microbiome contributes to hepatocarcinogenesis
- Microbial differences can be used for the diagnosis and treatment of liver disease

## Metabolites in Liver disease

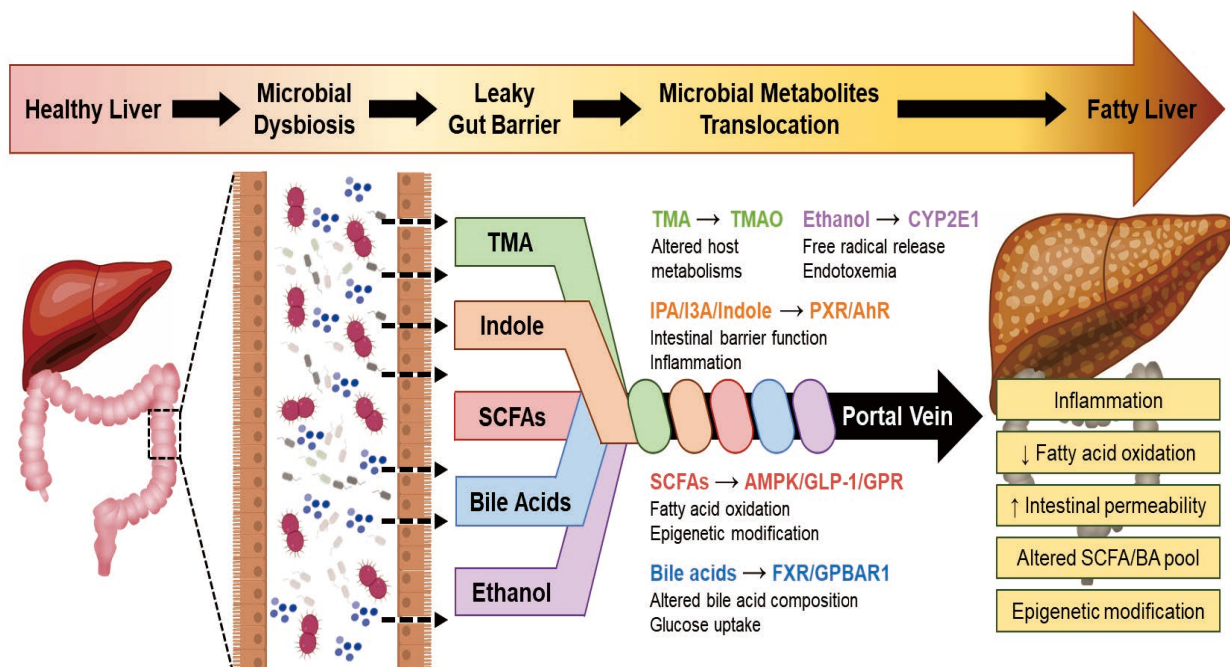




## Bidirectional communication between gut and liver



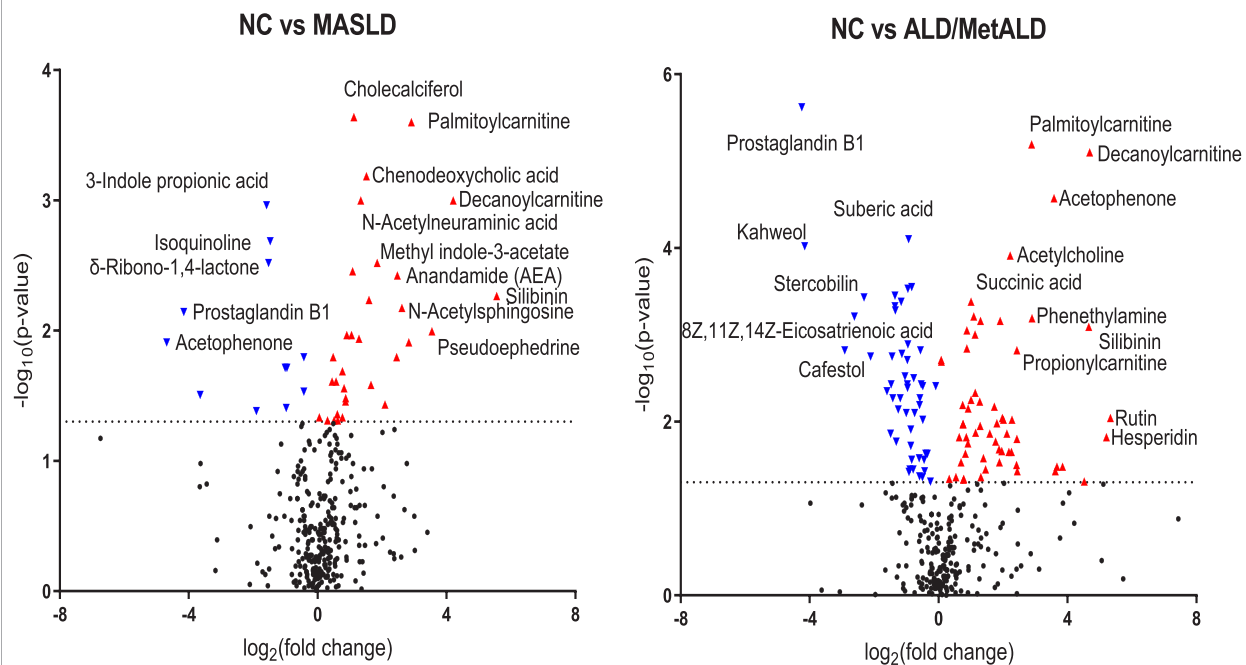
## Microbiota-derived metabolites



**Dysregulated microbiota-relating metabolites induce liver disease**

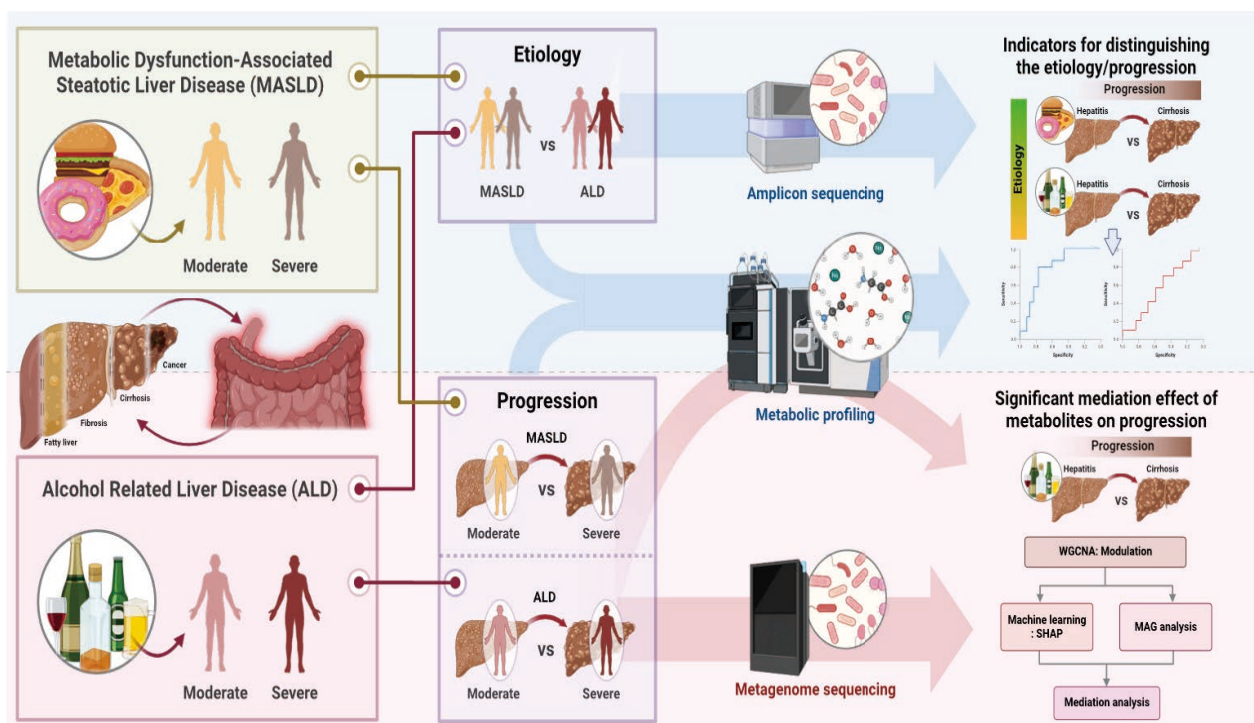
Suk KT, Kim DJ. Cells. 2021

## Metabolome data: MASLD & ALD/MetALD



Min BH, Suk KT, Gut Microbes. 2024;1:16(1)

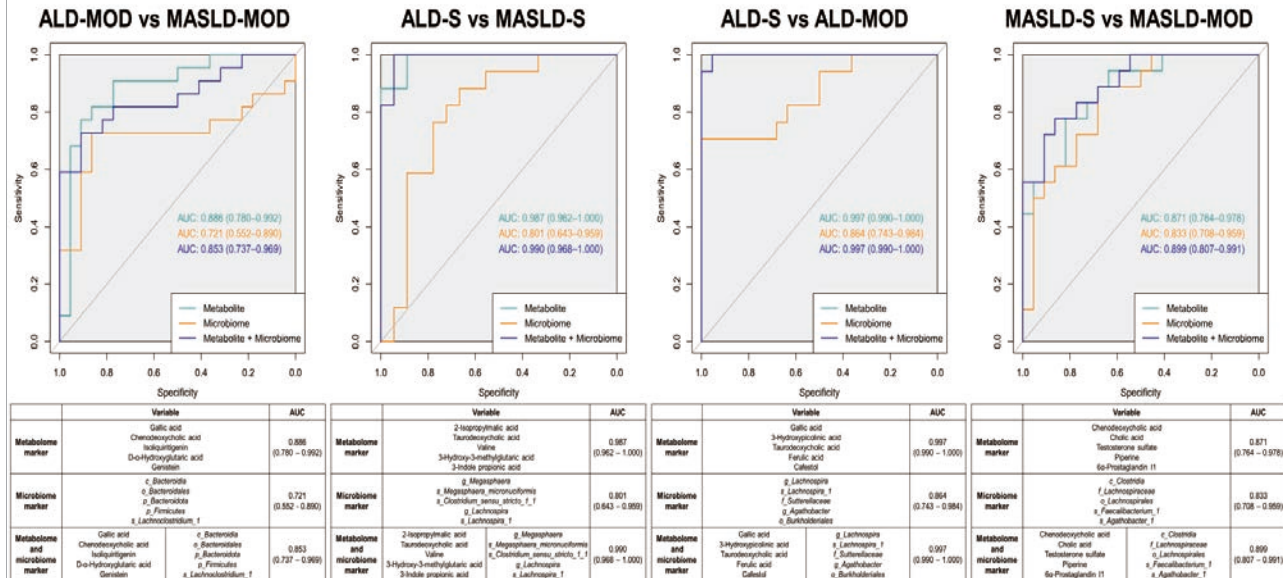
## Gut microbial and metabolomic features in liver disease



Lee DY, Suk KT, Unpublished data



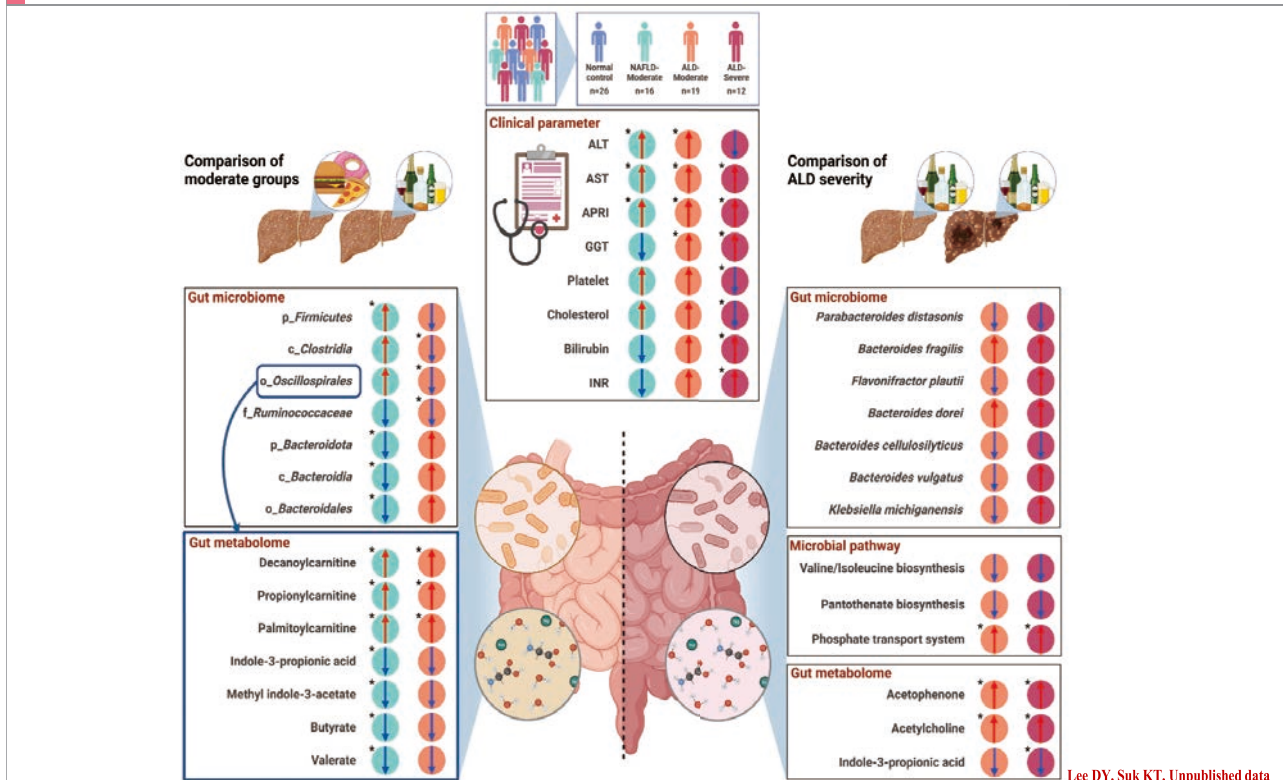
## Gut microbial and metabolomic features in liver disease



We can effectively diagnose liver diseases by utilizing microbiome and metabolite.

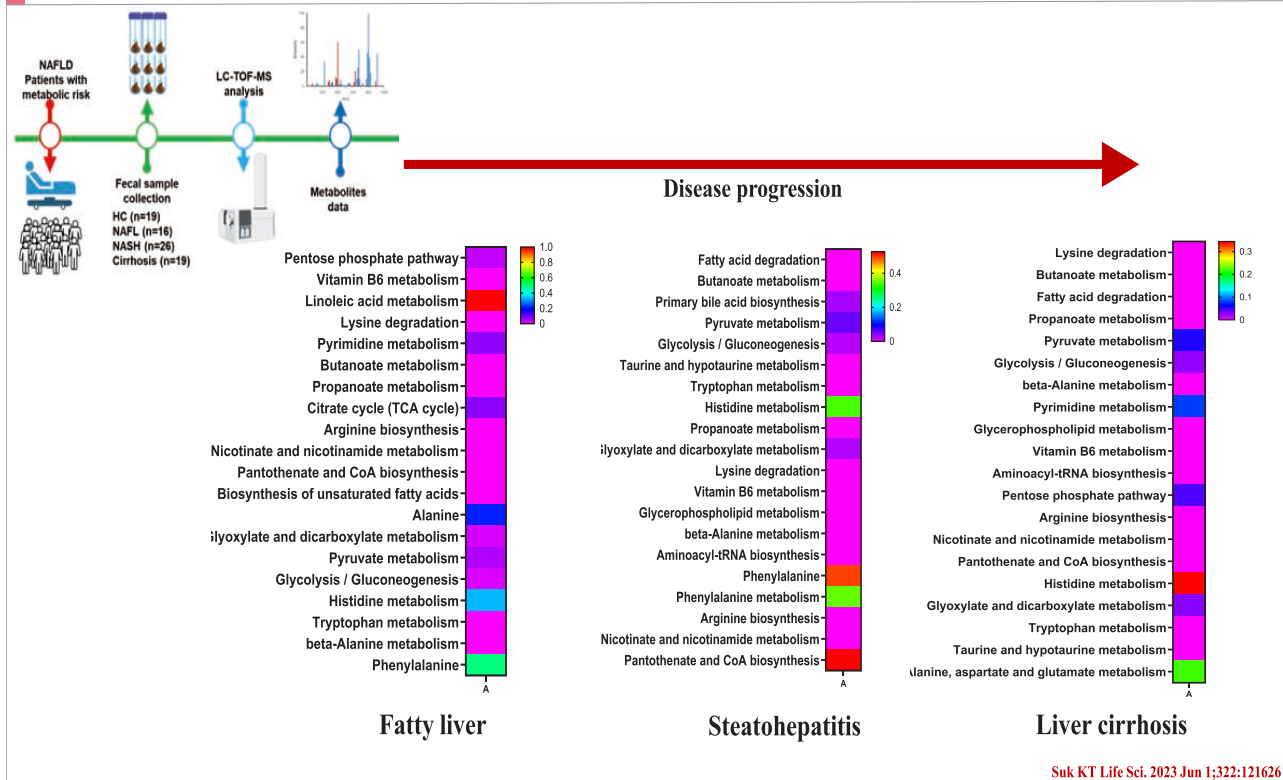
Lee DY, Suk KT, Unpublished data

## Gut microbial and metabolomic features in liver disease

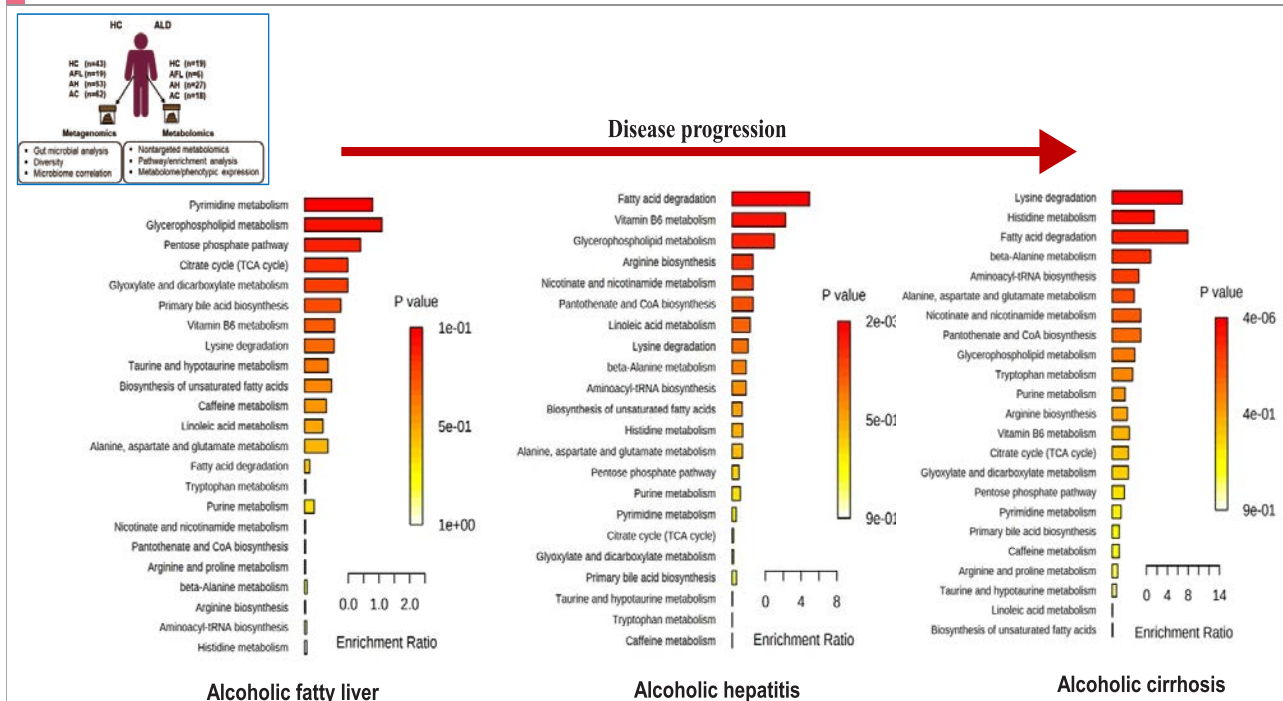


Lee DY, Suk KT, Unpublished data

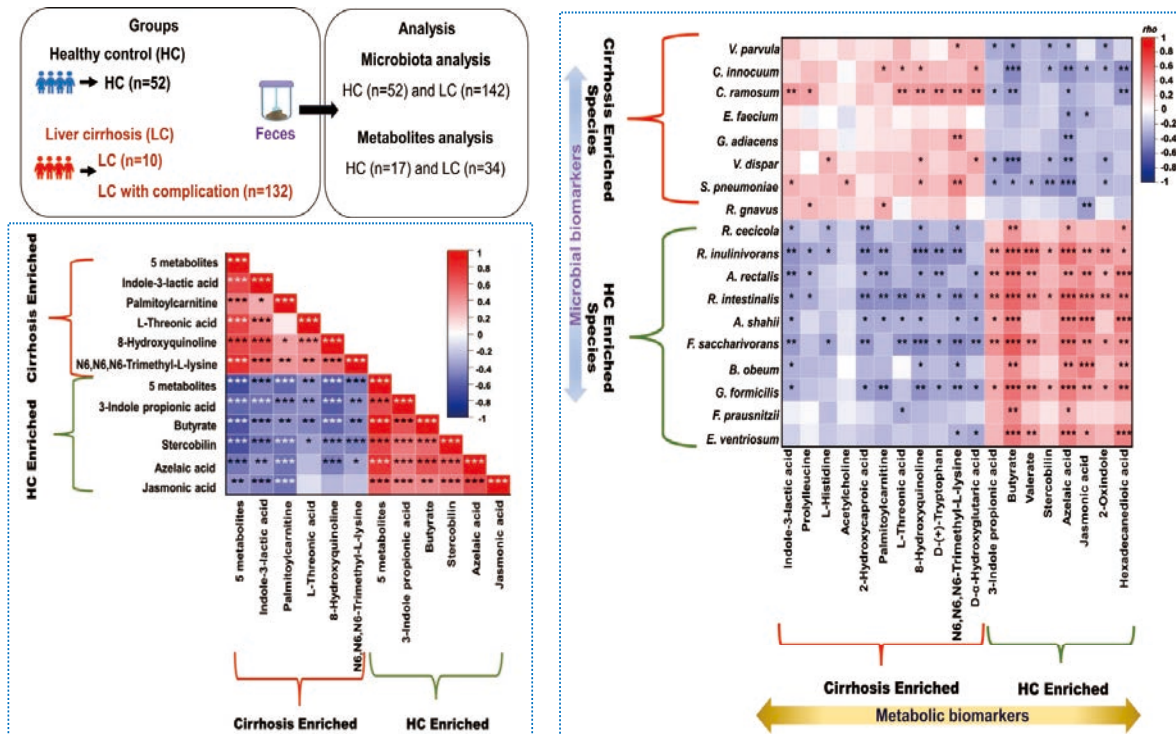
## Metabolic pathways of MASLD



## Metabolic pathways of MetALD/ALD



## Microbiota and metabolites in liver cirrhosis



Suk KT Clin Mol Hepatol. 2024;30:845-862.

## Summary

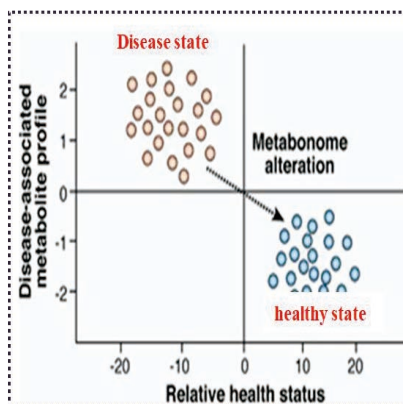


- Dysregulated microbial metabolites induce liver disease
- Microbial metabolites are associated with the severity of liver disease.
  - normal population **vs.** fatty liver **vs.** hepatitis **vs.** liver cirrhosis
- Metabolic pathway is different according to the liver disease
- Metabolic differences can be used for the diagnosis and treatment of liver disease

## Microbial - precision medicine in Liver disease

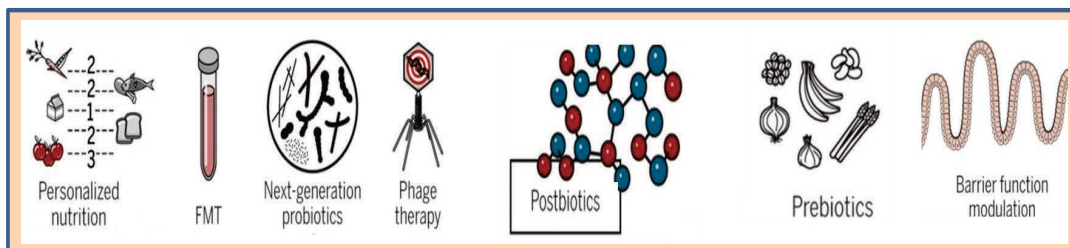


We cannot change our **genome**, we can change **microbiome**!



Modulation of gut-liver axis can cause metagenome alteration to healthy state.

**We can hypothesize that we can cure disease by modulation of microbiome!**

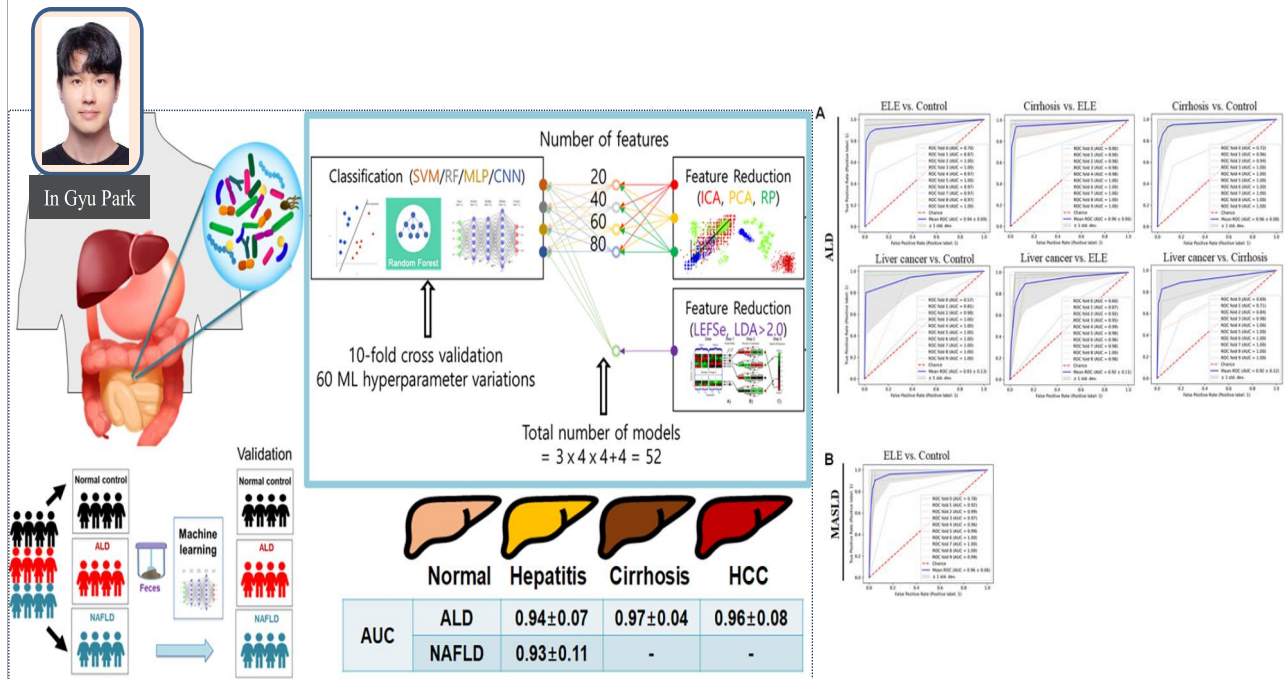


KT Suk Expert Rev Gastroenterol Hepatol. 2019  
Sci Transl Med 2019  
Bernd Schnabl. Cell Host Microbe 2020



## Gut microbiota-based machine-learning signature

Diagnostic marker



The gut microbiota-based ML strategy can be used for the diagnosis of ALD and MASLD!!

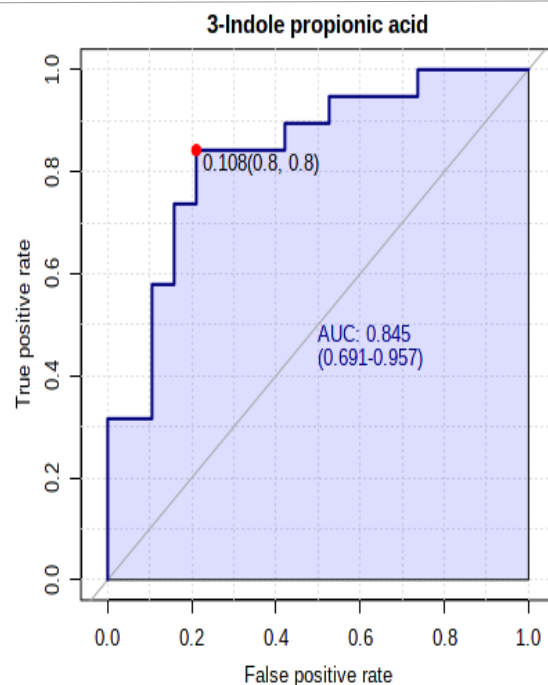
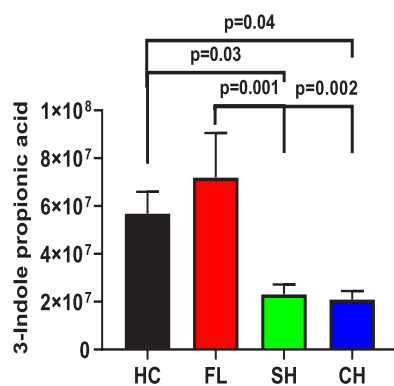
Suk KT Sci Rep. 2024 Jul 12;14(1):16122

## Biomarker of MASLD: metabolites

Diagnostic marker



Raja Ganesan



3-Indole propionic acid is biomarker for the diagnosis of MASLD.

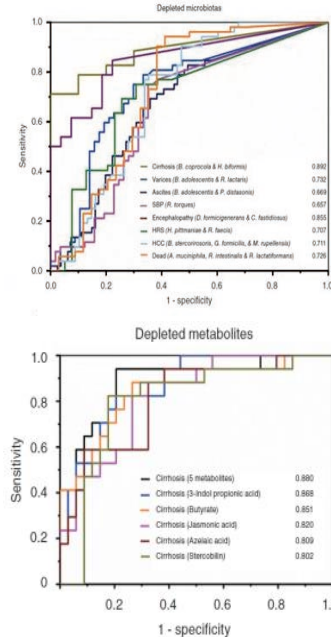
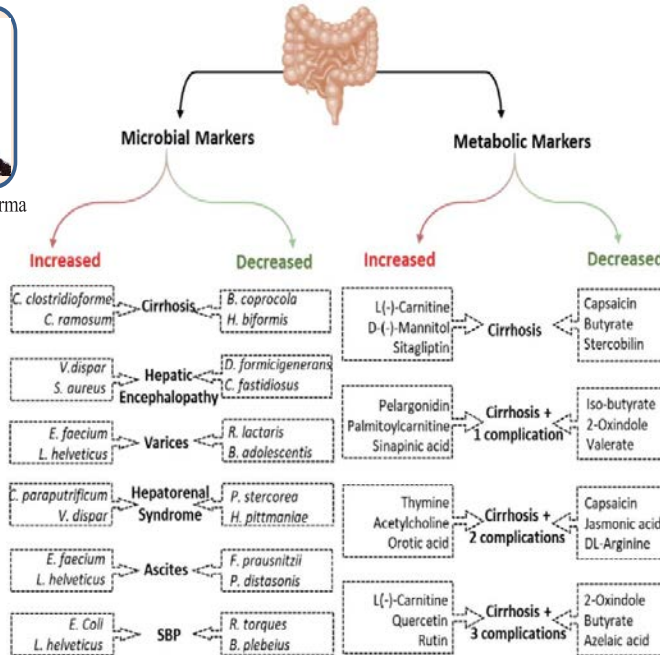
Suk KT Life Sci. 2023 Jun 1;322:121626

## Biomarkers of cirrhosis & complications

## Diagnostic marker



SatyaPria Sharma



Gut microbiota and metabolites can be used as noninvasive biomarkers for the diagnosis of cirrhosis and its complications.

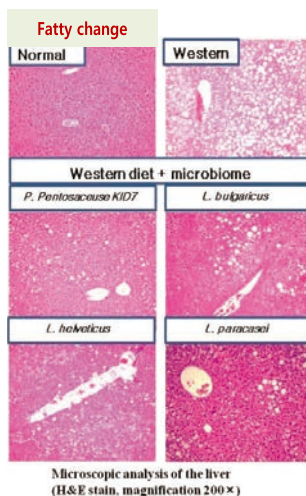
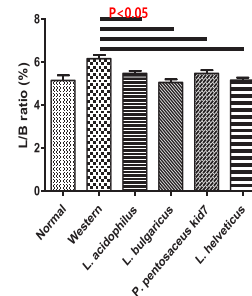
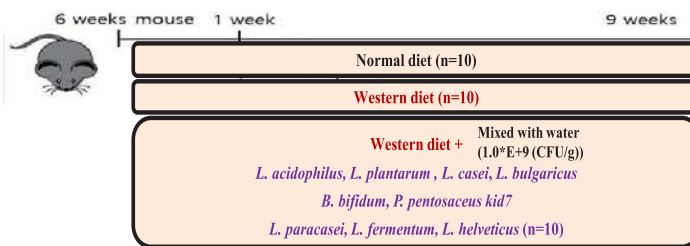
Suk KT Clin Mol Hepatol. 2024;30:845-862.

## MASLD animal model – 15 strains

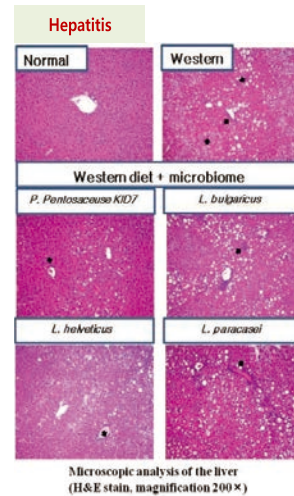
## Prevention



Jung A Eom Hee Jin Park



Strain
<i>B. bifidum</i>
<i>L. acidophilus</i>
<i>L. plantarum</i>
<i>L. casei</i>
<i>L. fermentum</i>
<i>L. bulgaricus</i>
<i>P. pentosaceus kid7</i>
<i>L. paracasei</i>
<i>L. helveticus</i>
Normal control
Western control

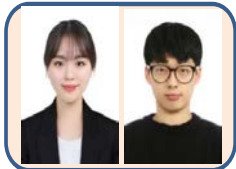


Strain
<i>L. acidophilus</i>
<i>L. plantarum</i>
<i>L. casei</i>
<i>L. fermentum</i>
<i>B. bifidum</i>
<i>L. bulgaricus</i>
<i>P. pentosaceus kid7</i>
<i>L. paracasei</i>
<i>L. helveticus</i>
Normal control
Western control

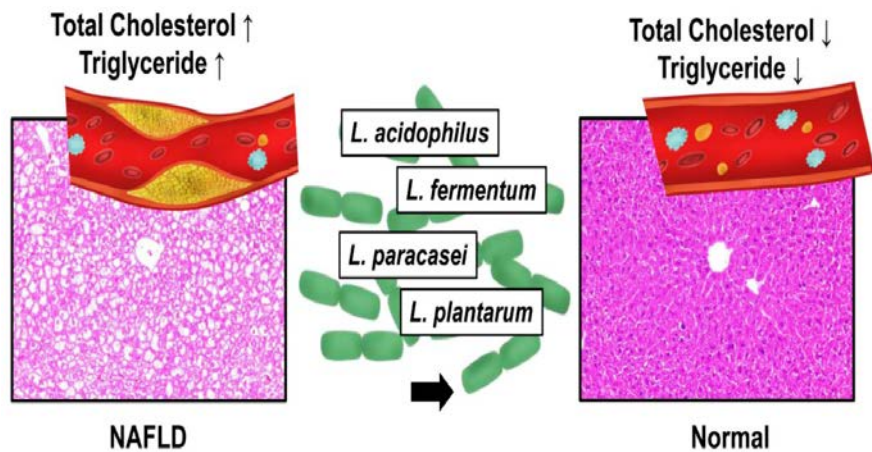
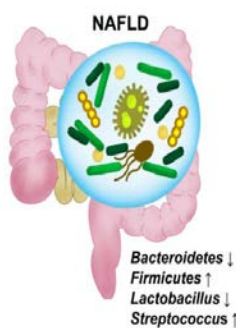


## *Lactobacillus* attenuates progression of MASLD by lowering cholesterol/steatosis

Prevention



Nayoung Lee Sang Jun Yoon



- Selected and administered *Lactobacillus* strains in NAFLD animal model.
- Intake of *Lactobacillus* lowered total cholesterol and triglyceride levels.
- *Lactobacillus* species strains may suppress fat production by modulating the microbiome for preventing obesity-induced diseases NAFLD.

Ingestion of *Lactobacillus* ameliorates the progression of SLD by lowering cholesterol.

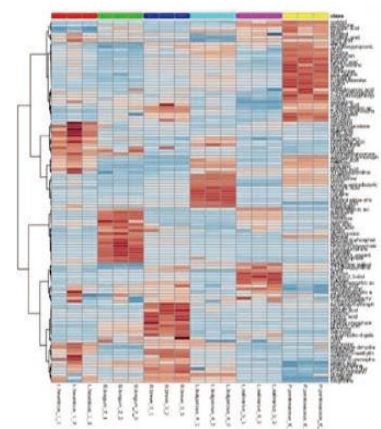
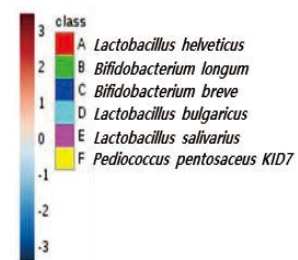
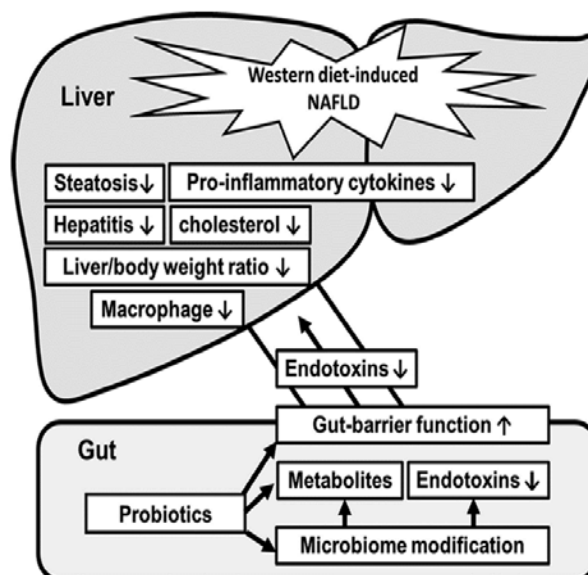
Suk KT. Clin Mol Hepatol. 2021 Jan

## *Lactobacillus*/Pediococcus on MASLD

Prevention



Nayoung Lee

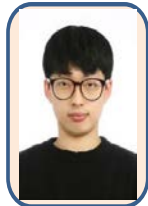


*L. bulgaricus*, *L. casei*, *L. helveticus*, and *P. pentosaceus* KID7 improve MASLD by modulating gut-microbiome.

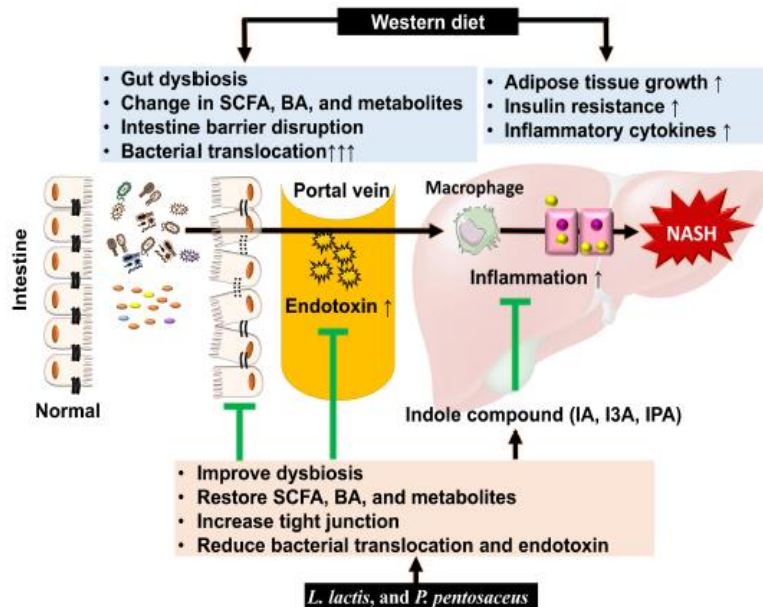
Suk KT. Gut Microbes. 2020 Jul

## Lactobacillus and Pediococcus

Prevention



Sang Jun Yoon

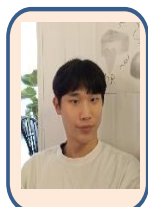


*L. lactis* and *P. pentosaceus* ameliorate NAFLD progression by modulating gut metagenomic and metabolic environment.

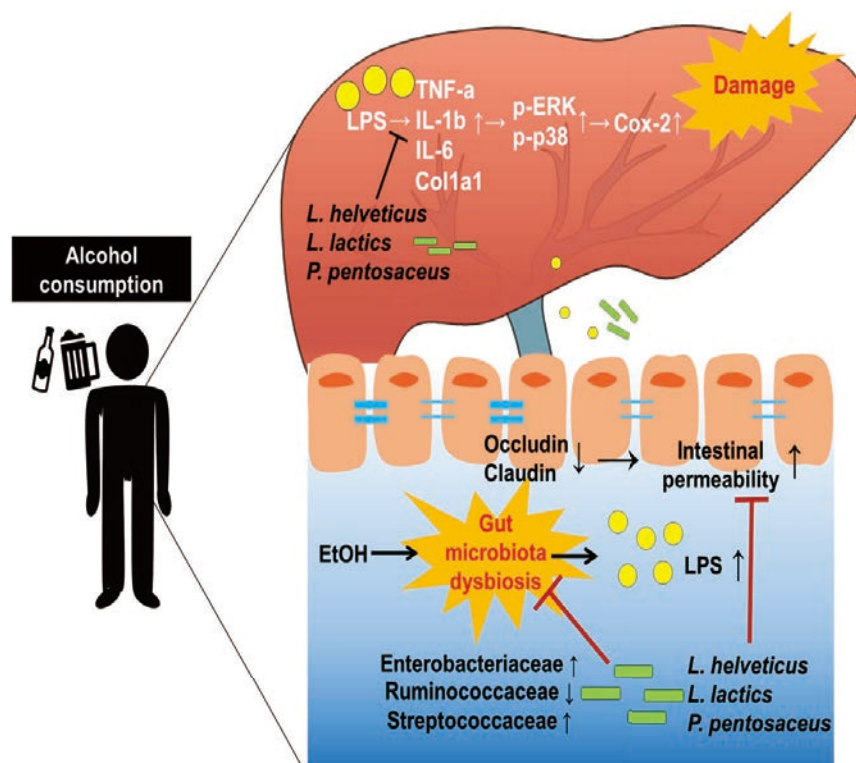
Suk KT, Lee DY. Clin Trans Med 2021

## Alcohol mouse model: probiotics

Prevention



Goo Hyun Kwon



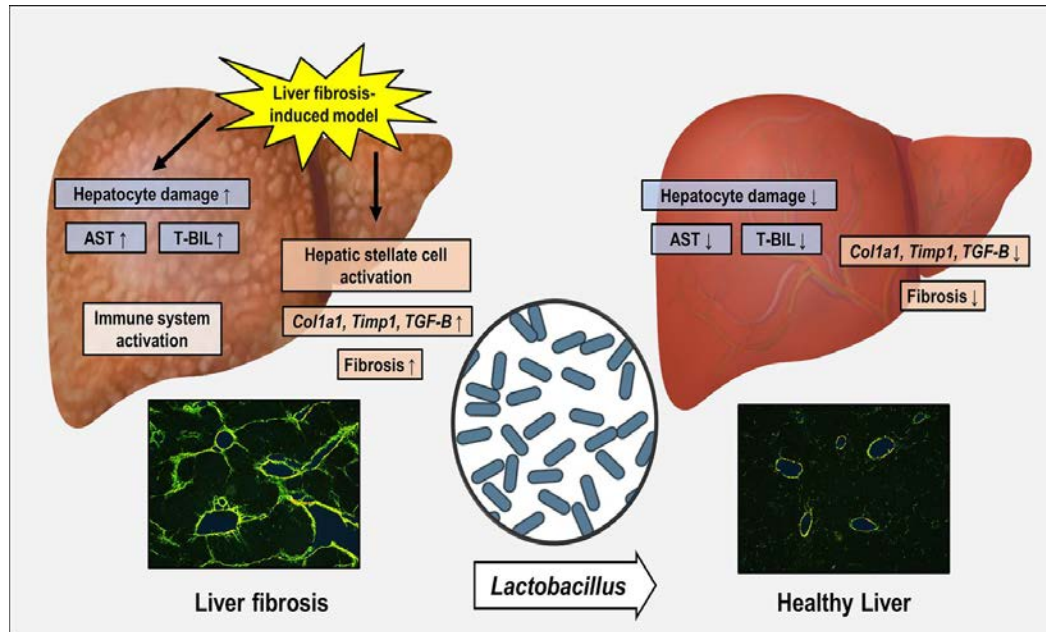
Kim HS, Suk KT, unpublished data

## Liver fibrosis and microbiotas

Prevention



Sung Min Won

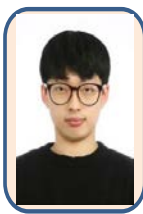


*L. lactis* and *L. rhamnosus* showed preventive/therapeutic effects on liver fibrosis.

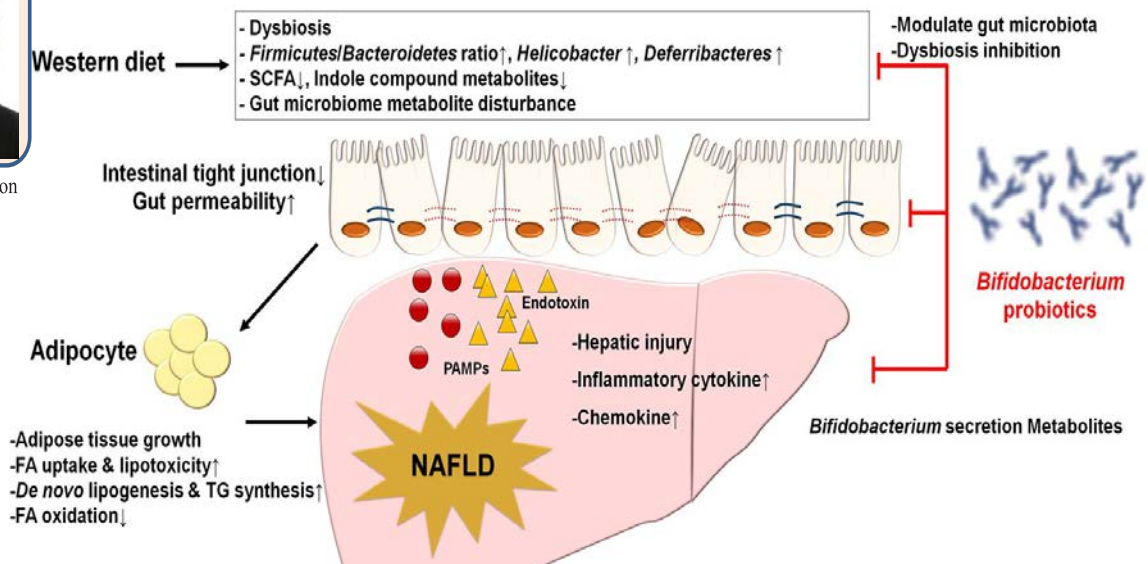
SM Won, KT Suk 2023, J Microbiology

## Bifidobacterium and NAFLD

Treatment



Sang Jun Yoon



*Bifidobacterium* attenuate MASLD by improving microbiome-associated factors of the gut-liver axis.

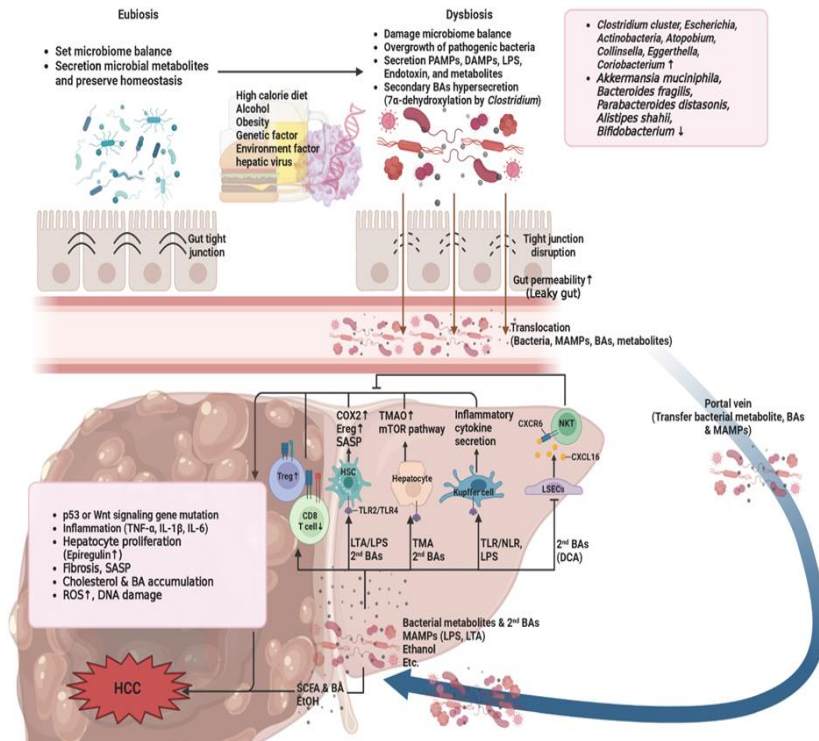
SJ Yoon, Suk KT, DJ Kim. Frontiers in microbiol 2023



# HCC and Microbiome



Sang Jun Yoon



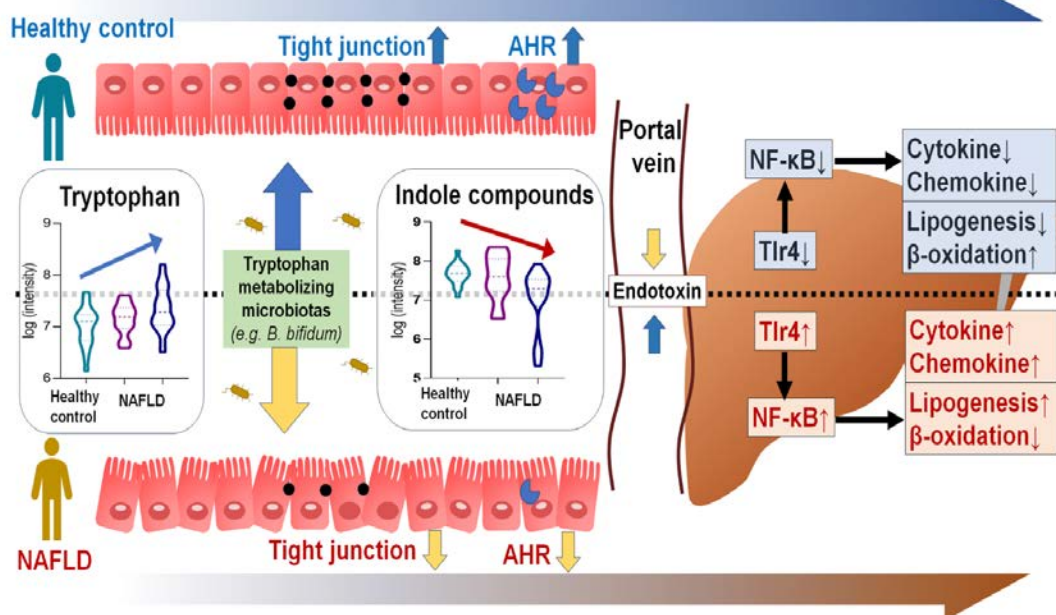
2024 review Critical Reviews in Microbiology

## NAFLD and Indole

## Prevention



Byung Hyun Min



**IPA and IAA derived from the gut microbiota have novel preventive or therapeutic potential for MASLD treatment.**

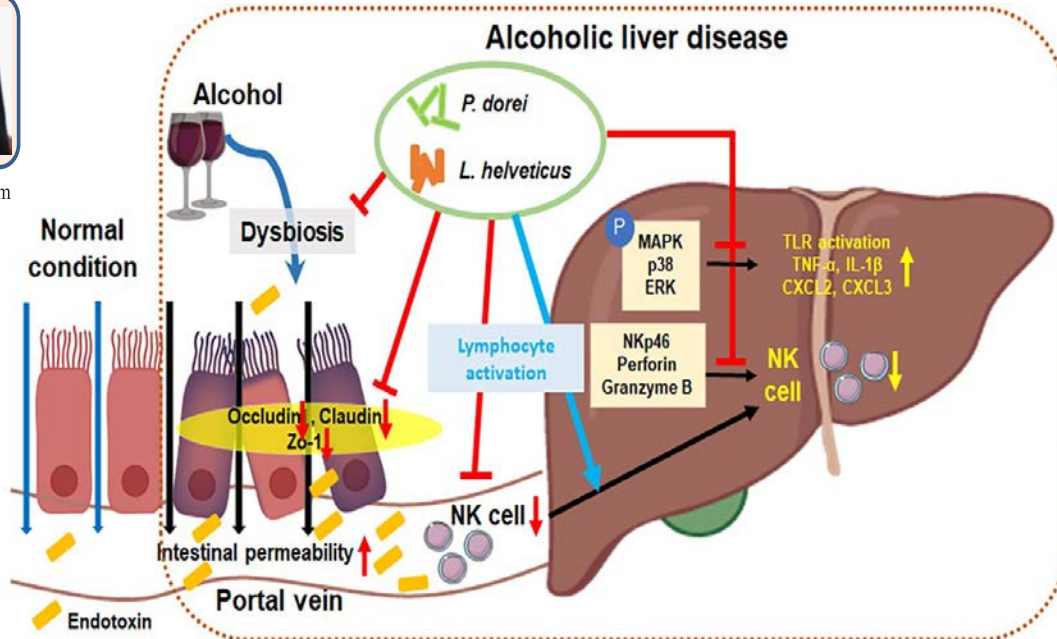
BH Min, KT Suk 2024 Gut microbes

## Immune and microbiome

Prevention



Jung A Eom



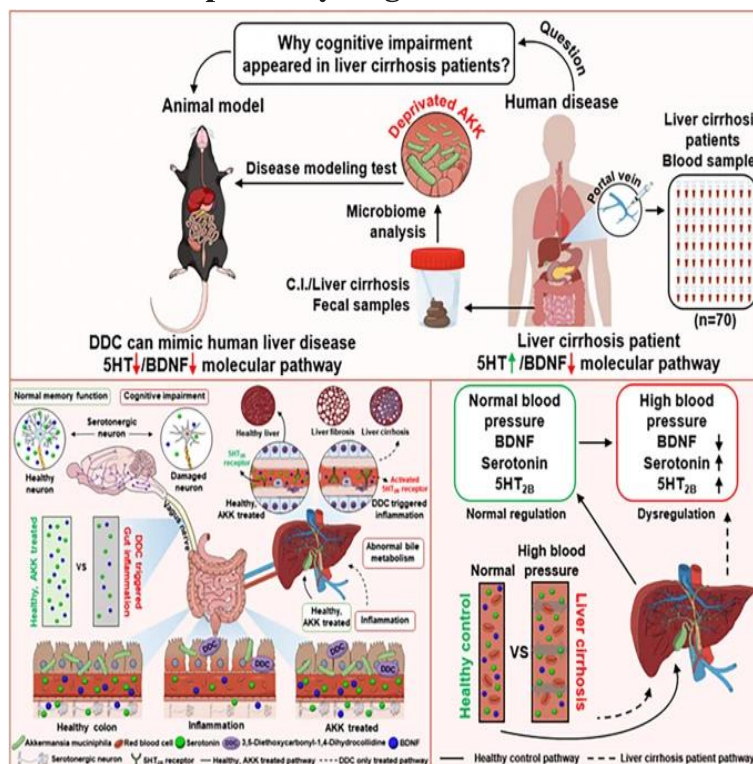
Gut microbiome ameliorate ALD by regulating immune cells.

JA Eom, KT Suk. 2024 Gut microbes

## Akkermansia muciniphila improve cognitive dysfunction by regulating BDNF and serotonin pathway in gut-liver-brain axis



Sung Min Won



*A. muciniphila*, a next-generation probiotic, is a therapeutic candidate for alleviating the symptoms of liver fibrosis and cognitive impairment.

SM Won, KT Suk 2024, Microbiome

## Alcohol clinical trial 1: probiotics

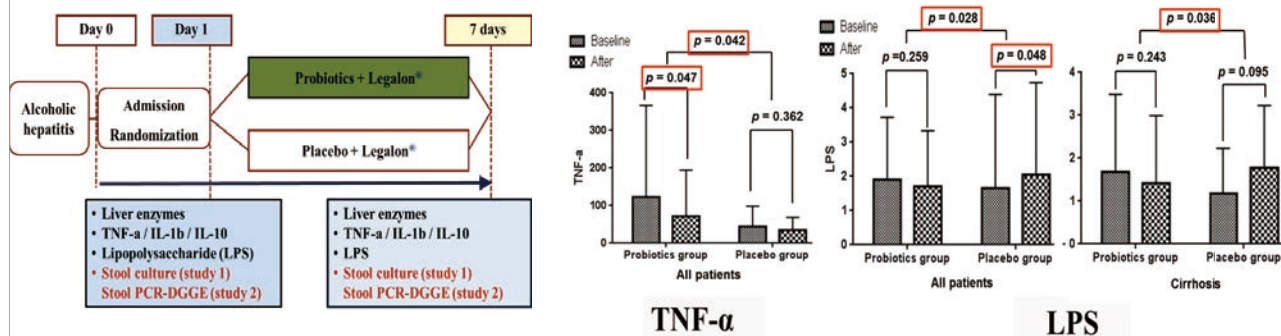


*Lactobacillus subtilis*/Streptococcus faecium

A multicenter, randomized, clinical trial at 4 university hospitals (Lactowel®, 1,500 mg/day)

September 2010 – September 2012

117 Patients with AH were recruited.



7 days of oral supplementation with probiotics is associated with improvement of TNF-α and LPS in AH patients, especially with alcoholic cirrhosis

Suk KT, Kim DJ. Eur J Gastroenterol Hepatol. 2015

## Alcohol clinical trial 2: probiotics



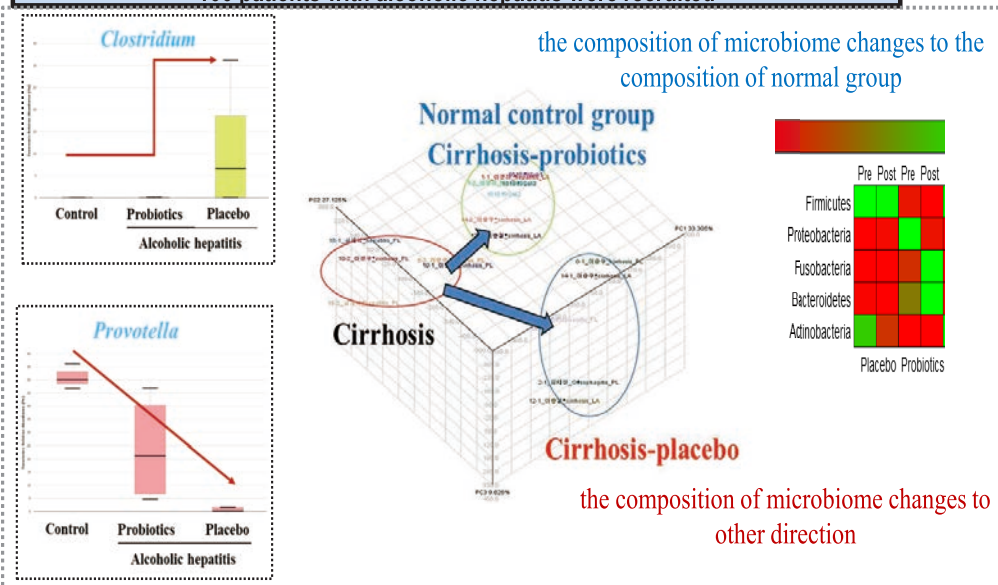
*Lactobacillus rhamnosus* R0011/acidophilus R0052

(Lacidophil®, 120 mg/day)

100 patients with alcoholic hepatitis were recruited



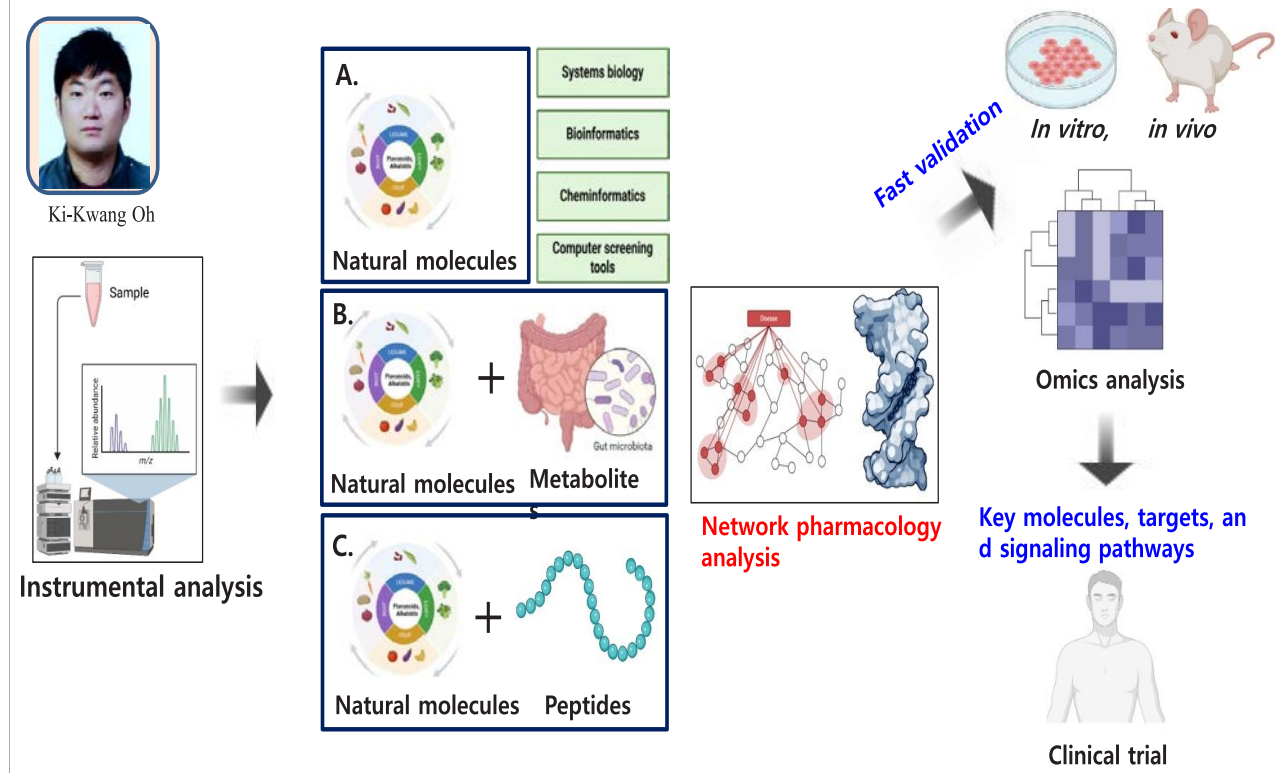
Haripriya Gupta



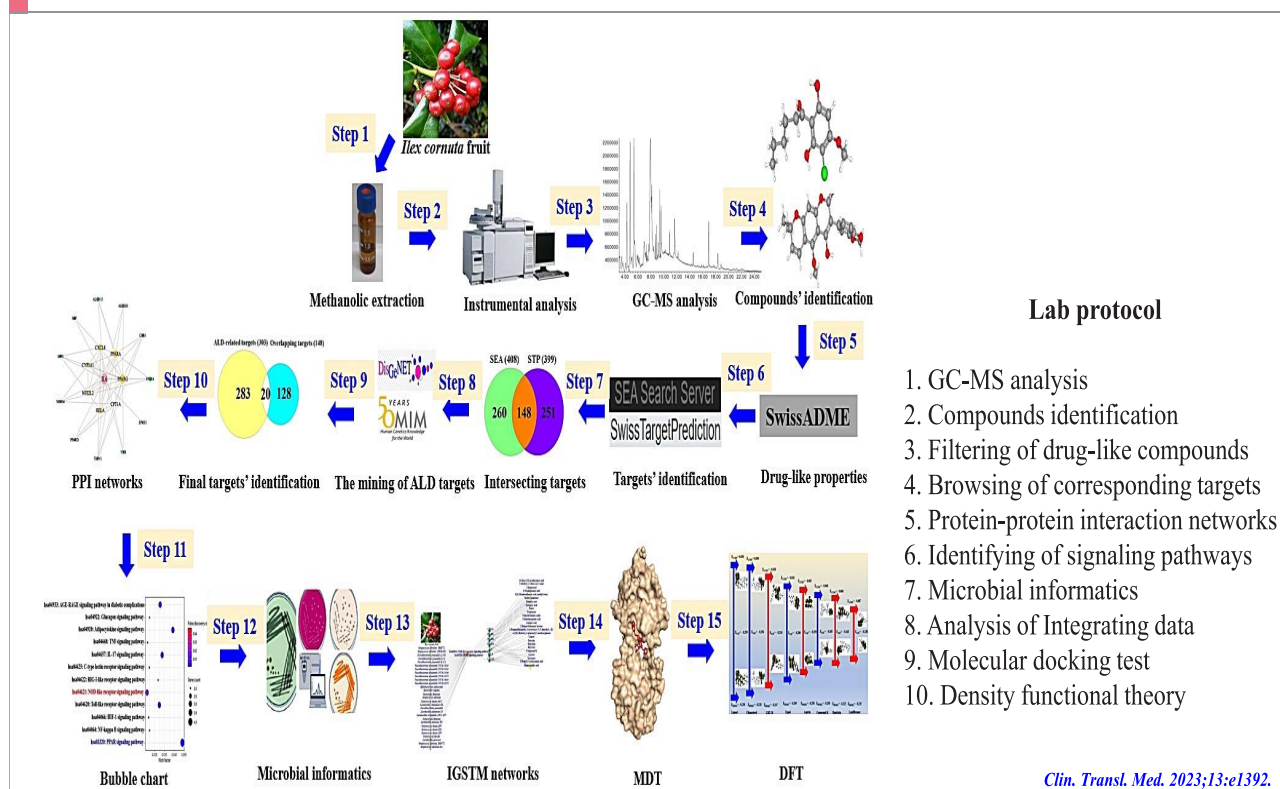
Haripriya Gupta Suk KT, Kim DJ et al, 2022 microorganisms



## Network pharmacology



## The juxtaposition of *Ilex cornuta* fruit and gut microbiota against alcoholic liver disease based on the integrated pharmacology via metabolomics



## A layout of the promising components for liver disease

### 53 gut microbes

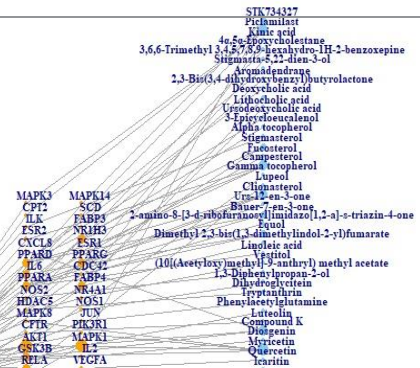
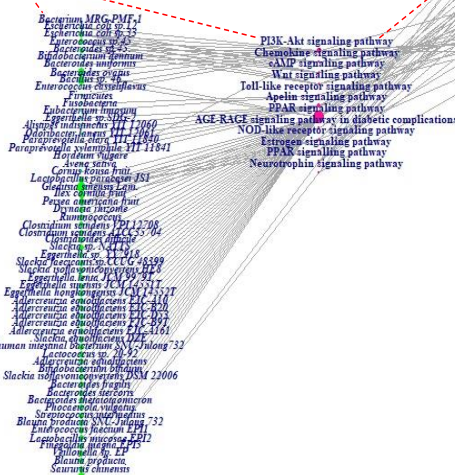
### 7 natural resources



<i>Gleditsia sinensis</i> Lam.	<i>Persea americana</i>	<i>Avena sativa</i> (*)	<i>natural resources or diet</i>
<i>Bacterium</i> MRG-PMF-1 <sup>TM</sup>	<i>Slackia</i> sp. NATTS		
<i>Escherichia coli</i> sp. 12	<i>Escherichia</i> sp. YY7918		
<i>Escherichia coli</i> sp. 33	<i>Slackia faecalis</i> sp. CCG 48399		
<i>Enterococcus</i> sp. 45	<i>Slackia isolavoniomicrobium</i> HES		
<i>Bacteroides</i> sp. 45	<i>Escherichia lenta</i> JCM 9979T		
<i>Bifidobacterium dentium</i>	<i>Escherichia sinensis</i> JCM 14551T		
<i>Bacteroides uniformis</i>	<i>Escherichia hongkongensis</i> JCM 1453		
<i>Bacteroides ovatus</i>	<i>Adlercreutzia equalifaciens</i> FJC-A10		
<i>Bacillus</i> sp. 46	<i>Adlercreutzia equalifaciens</i> FJC-B20		
<i>Enterococcus casseliflavus</i>	<i>Adlercreutzia equalifaciens</i> FJC-D53		
<i>Firmicutes</i>	<i>Adlercreutzia equalifaciens</i> FJC-B9T		
<i>Fusobacteria</i>	<i>Adlercreutzia equalifaciens</i> FJC-A161		
<i>Eubacterium limosum</i>	<i>Slackia equalifaciens</i> DZE		
<i>Escherichia</i> sp. SDG-2	<i>Intestinal bacterium</i> SNU-Julong 732		
<i>Alistipes indistinctus</i> YIT 12060	<i>Lactococcus</i> sp. 20-92		
<i>Odoribacter laneus</i> YIT 12061	<i>Adlercreutzia equalifaciens</i>		
<i>Paraprevotella clara</i> YIT 11840	<i>Bifidobacterium bifidum</i>		
<i>Paraprevotellaylaniphila</i> YIT 11841	<i>Slackia isolavoniomicrobium</i> DSM		
<i>Hordeum vulgare</i> (*)	<i>Bacteroides fragilis</i>		
<i>Avena sativa</i> (*)	<i>Bacteroides stercoris</i>		
<i>Cornus kousa</i> fruit (*)	<i>Bacteroides thetaiotaomicron</i>		
<i>Lactobacillus paracasei</i> JS1	<i>Phocaeicola vulgatus</i>		
<i>Gleditsia sinensis</i> Lam. (*)	<i>Streptococcus intermedius</i>		
<i>Ilex cornuta</i> fruit (*)	<i>Blautia producta</i> SNU-Julong 732		
<i>Persea americana</i> fruit (*)	<i>Enterococcus faecium</i> EPI1		
<i>Drynaria rhizome</i> (*)	<i>Lactobacillus mucosae</i> EPI2		
<i>Ruminococcus</i>	<i>Finegoldia magna</i> EPI3		
<i>Clostridium scindens</i> VPI 12708	<i>Veillonella</i> sp. EP		
<i>Clostridium scindens</i> ATCC35704	<i>Blautia producta</i>		
<i>Clostridioides difficile</i>	<i>Saururus chinensis</i>		

### 12 signaling pathways

PI3K-Akt signaling pathway	PPAR signaling pathway
Chemokine signaling pathway	AGE-RAGE signaling pathway in diabetic complications
cAMP signaling pathway	NOD-like receptor signaling pathway
Wnt signaling pathway	Estrogen signaling pathway
Toll-like receptor signaling pathway	PPAR signalling pathway
Apelin signaling pathway	Neurotrophin signaling pathway



30 targets

37 molecules

1. CIMB
2. Cells
3. Artificial cells, Nanomedicine, and Biotechnology
4. Scientific Reports
5. Journal of Translational Medicine
6. Clinical and Translational Medicine
7. Journal of Translational Medicine 10. Clinical and Translational Science
8. Food Chemistry

## Summary

- Dysbiosis and metabolites alteration has been varied according to the severity of liver diseases
- Deeper understanding of dysbiosis, metabolic stability, reaction phenotyping in SLD are needed
- Dysbiosis and microbiota-derived metabolite are related disease phenotype in SLD.

## Conclusion

**In the liver disease, gut microbiota and its metabolites played central role in the disease progression**

## Acknowledgements



한림대학교춘천성심병원



한림대학교광주성심병원



한림대학교경주성심병원



한림대학교동탄성심병원



한림대학교항강성심병원



한림대학교광주성심병원

**THANK YOU**

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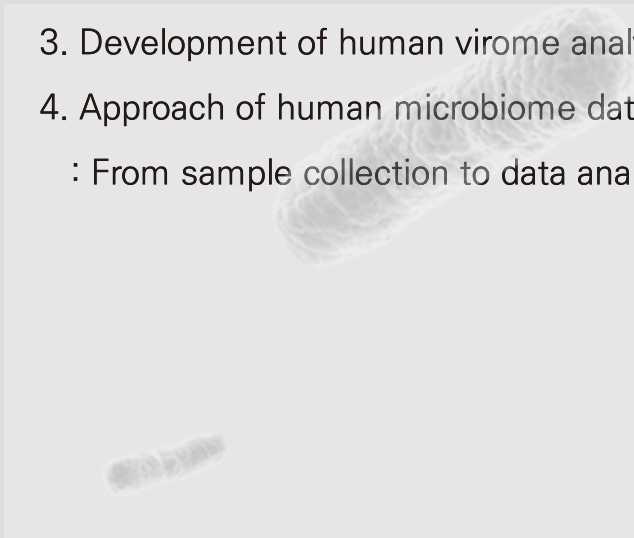


## **Session 3.**

# **A guide to human microbiome research: Database & Bioinformatics analysis**



1. Human microbiome reference and resource development for the clinical applications in diagnosis and therapeutics
2. Analysis of microbiome genetic information using artificial intelligence
3. Development of human virome analysis research methodology
4. Approach of human microbiome data standardization  
: From sample collection to data analysis







## 3부 좌장



- ✓ 소속: 질병관리청 국립보건연구원 감염병연구센터
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2017 ~ 2020	질병관리본부 백신연구과 과장
2020 ~ 현재	질병관리청 국립보건연구원 감염병연구센터장



01

# Human microbiome reference and resource development for the clinical applications in diagnosis and therapeutics



이선재  
(광주과학기술원)





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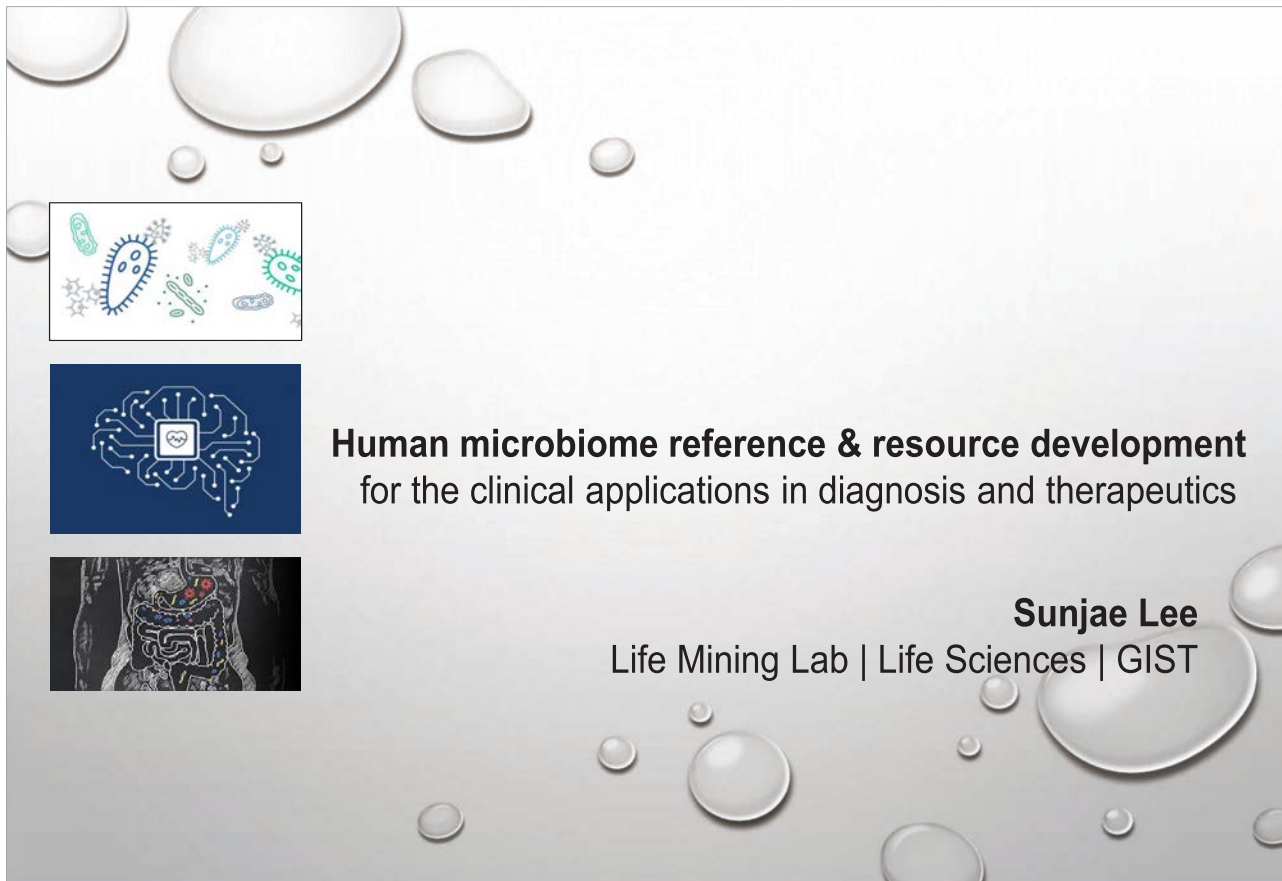
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2020~현재	광주과학기술원 생명과학부 교수

# Human microbiome reference and resource development for the clinical applications in diagnosis and therapeutics

최근 제2의 게놈으로써 인체 마이크로바이옴이 새롭게 규명되어지고 있으며, 마이크로바이옴 변화에 따른 인체 표현형의 변화, 질병 연관성 등의 다양한 실험적, 역학조사를 통해서 밝혀짐에 따라서, 인체 마이크로바이옴 기능 이해에 대한 중요도가 높아지고 있다. 이러한 인체 마이크로바이옴을 이해하기 위해서는 샷건 메타지놈 등의 유전체 데이터 생성을 통해서 마이크로바이옴 구성과 기능에 대한 이해가 필요하며, 또한 다양한 질병 코호트 연구를 통해서 건강한 개인과 질병 환자의 마이크로바이옴 구성 및 기능 차이에 대한 이해가 필요하다. 특히 inter-individual variation이 높은 인체 마이크로바이옴을 이해하기 위해서는 Big Data Scale의 대규모 샷건 메타지놈 분석이 필요로 하며, 이를 바탕으로 임상 현장에서 적용할 수 있는 진단 기법을 개발하고, 질병 예방/치료 전략을 수립할 수 있을 것으로 기대된다. 본 발표에서는 현재까지의 마이크로바이옴 유전체 레퍼런스들과 Big Data 기법을 통해 질병 마이크로바이옴의 특징을 확인하고, 이를 기반으로 임상 현장에 응용될 수 있는 마이크로바이옴 데이터 기반 예측 모델들 논의하도록 한다.



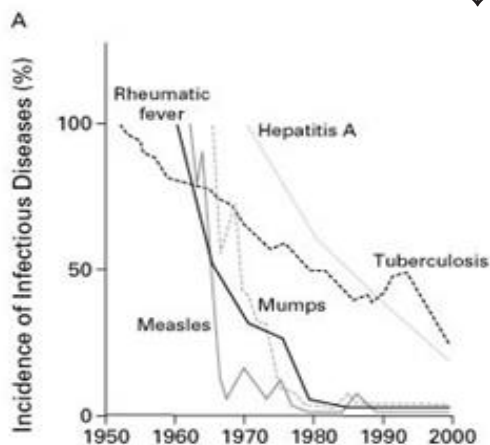


**Human microbiome reference & resource development**  
for the clinical applications in diagnosis and therapeutics

**Sunjae Lee**  
Life Mining Lab | Life Sciences | GIST

## Modern disease landscape

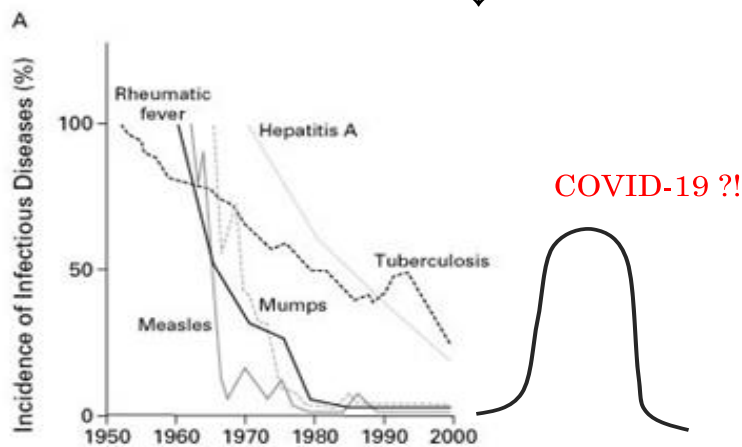
Infectious disease ↓



REF | Jean-Francois Bach, NEJM, 2002

## Modern disease landscape

Infectious disease ↓



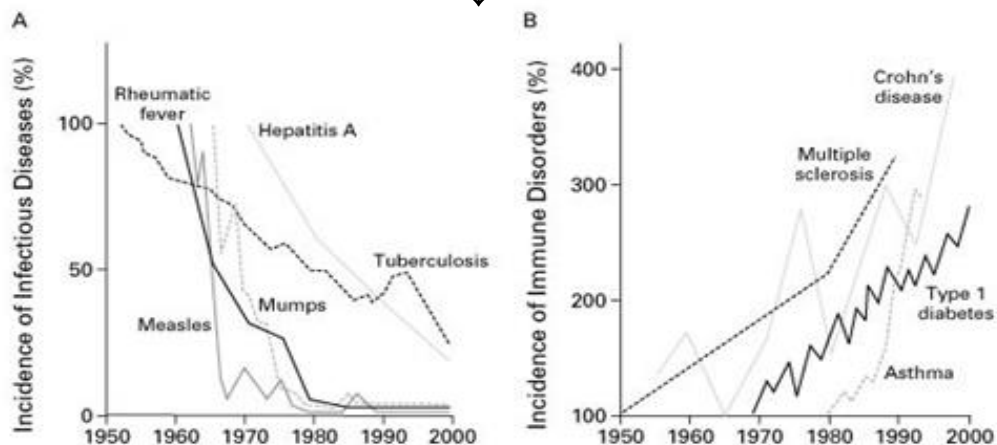
REF | Jean-Francois Bach, NEJM, 2002

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## Modern disease landscape

Infectious disease ↓

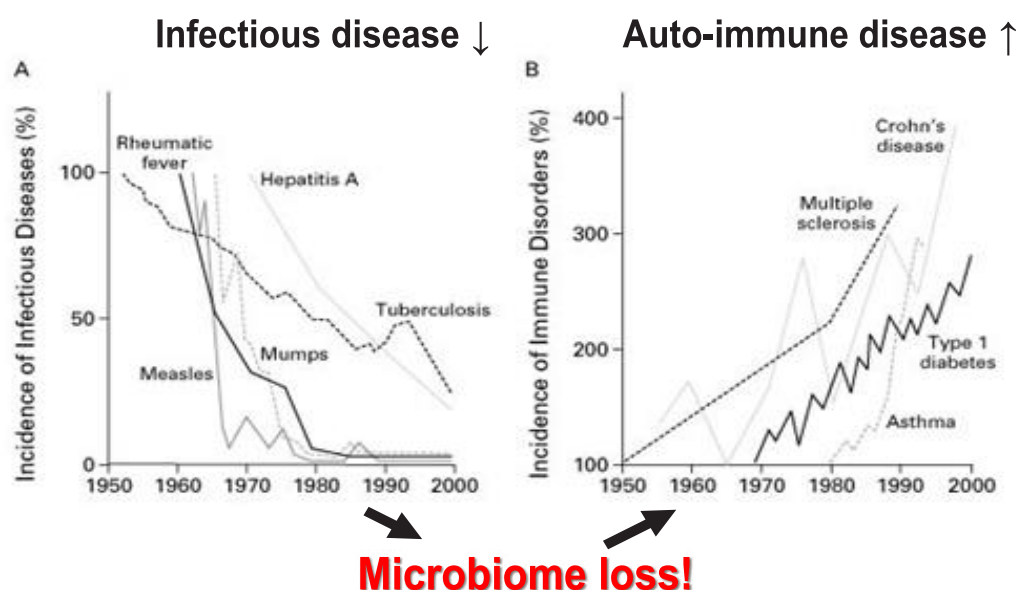
Auto-immune disease ↑



REF | Jean-Francois Bach, NEJM, 2002

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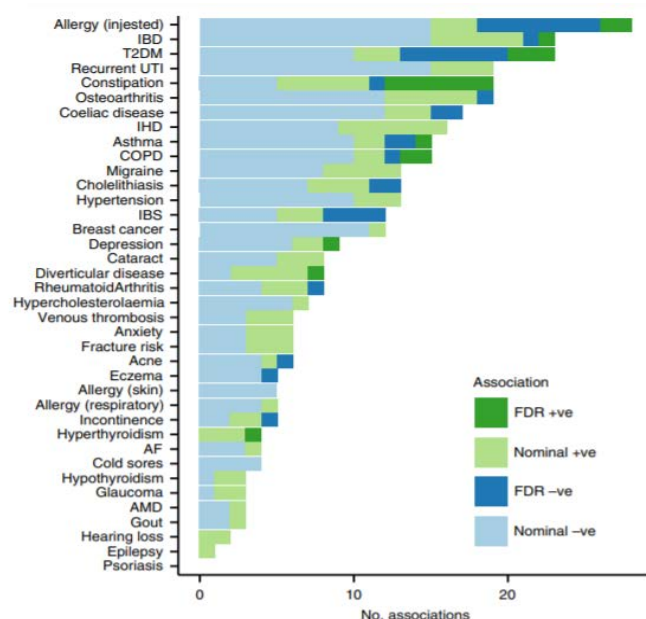
## Modern disease landscape



REF | Jean-Francois Bach, NEJM, 2002

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## Common diseases associated with microbiome changes



Allergy  
Constipation  
Migraine  
T2D  
Asthma  
Hypertension  
...

King's College  
London  
Twin Study

REF | Matthew A Jackson et al., Nature Communications (2018)

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## Preserving microbiome - Microbiota vault?

Maria G Mominguez Bello



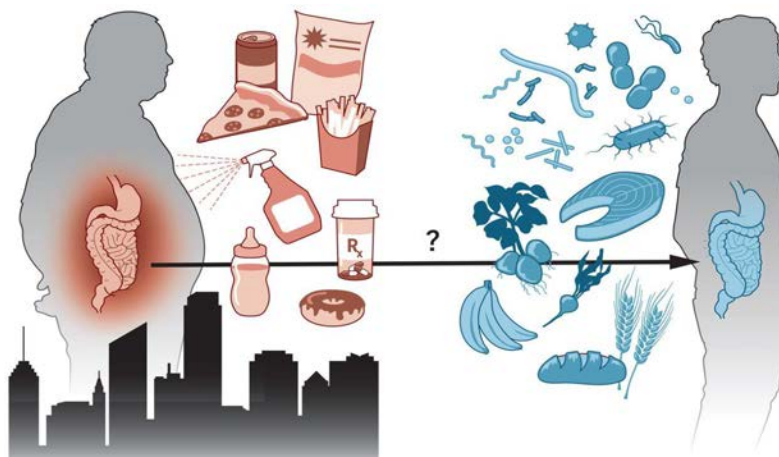
Martin J Blaser  
Author of Best-seller  
*"missing microbes"*



REF | Maria G Mominguez Bello et al., Science, 2018

7

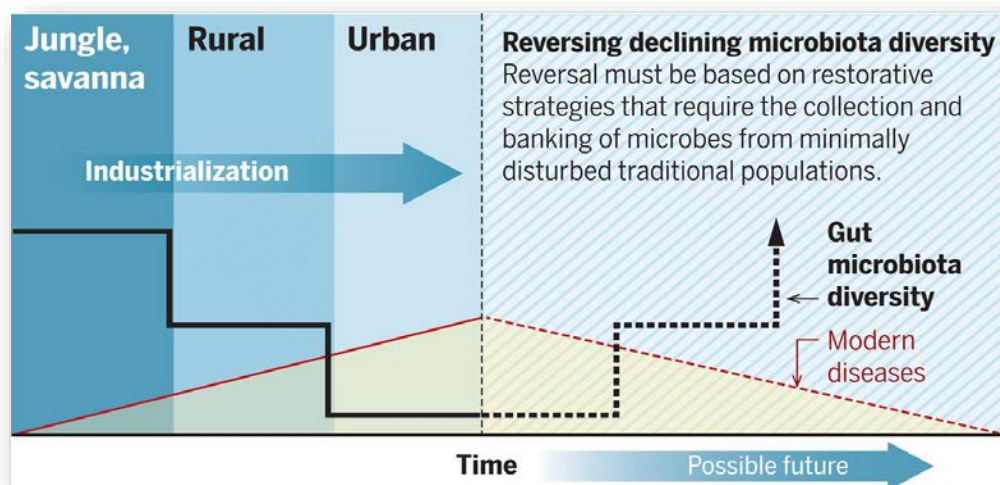
## We are losing our microbiome



Processed foods, food additives, high fat diets, low fiber intake, etc

8

## We are losing our microbiome



REF | Maria G Mominguez Bello et al., Science, 2018

9

**Then how can we find “healthy” microbiome?**

10

## Big Data Approaches !

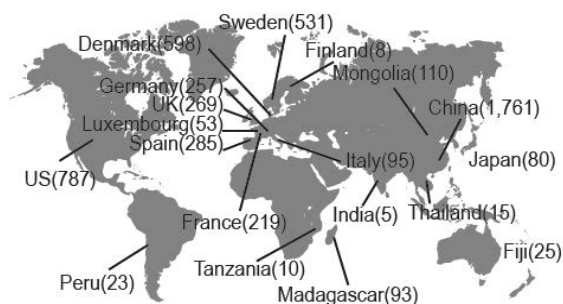
11

Big data analysis by  
microbiome reference & resource developments

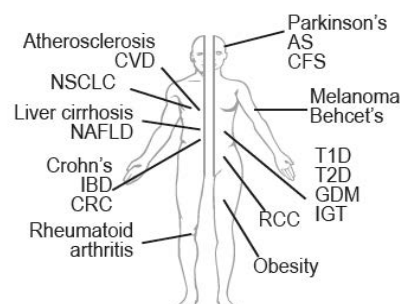
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## Big Data Analysis of Human Gut Microbiome



**19 countries**



**23 Diseases**

- Healthy samples from 18 countries
- Disease samples from 28 cohorts (e.g. liver diseases, Parkinson's)

**Sunjae Lee et al., Genome Research (2024)**

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## Microbiome Big Data Initiatives



2007-2016

**13,000 samples**  
\$170 million USD



2008-2012

**200 samples**  
3.3 million genes



2014-2016

**12,000 samples**  
Multi-omics




2020-present

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## Microbiome Big Data Initiatives

China National GeneBank CNGBdb

 **MMHP**

### MMHP: Million Microbiomes from Humans Project

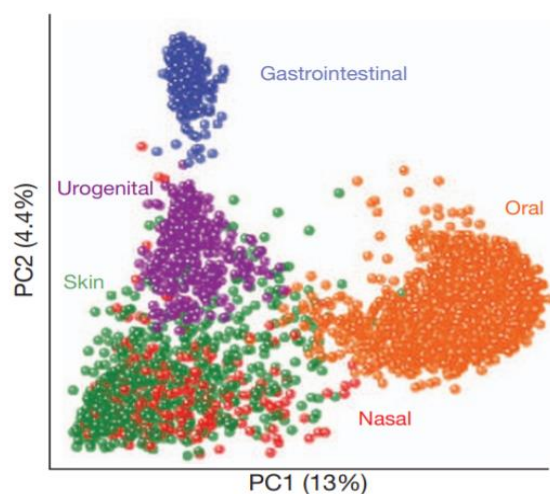
The Million Microbiomes from Humans Project (MMHP) was officially launched at the 14th International Conference on Genomics (ICG-14). Scientists from China, Sweden, Denmark, France, and Latvia agreed to collaborate on a large-scale microbial metagenomic project, aiming to sequence and analyze one million samples from the intestine, mouth, skin, reproductive tract, and other organs in the next three to five years construct a microbiome map of the human body and build the world's largest database of the human microbiome.

The White Paper now openly available for download below, provides an overview of this project.

[White paper](#)

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## Big data analysis reveals microbiome landscape

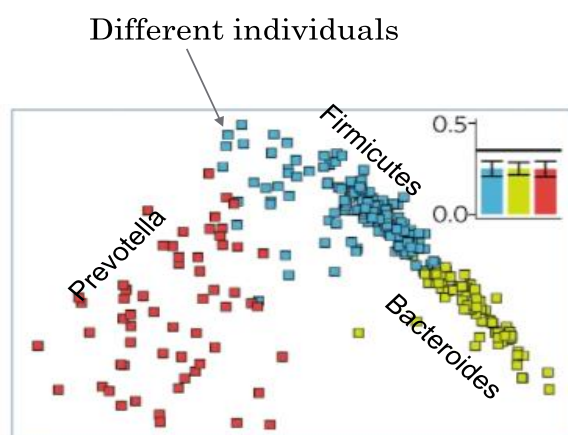


Landscape of human microbiome

REF | Human Microbiome Project Consortium, 2012, Nature

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## Big data analysis reveals microbiome landscape



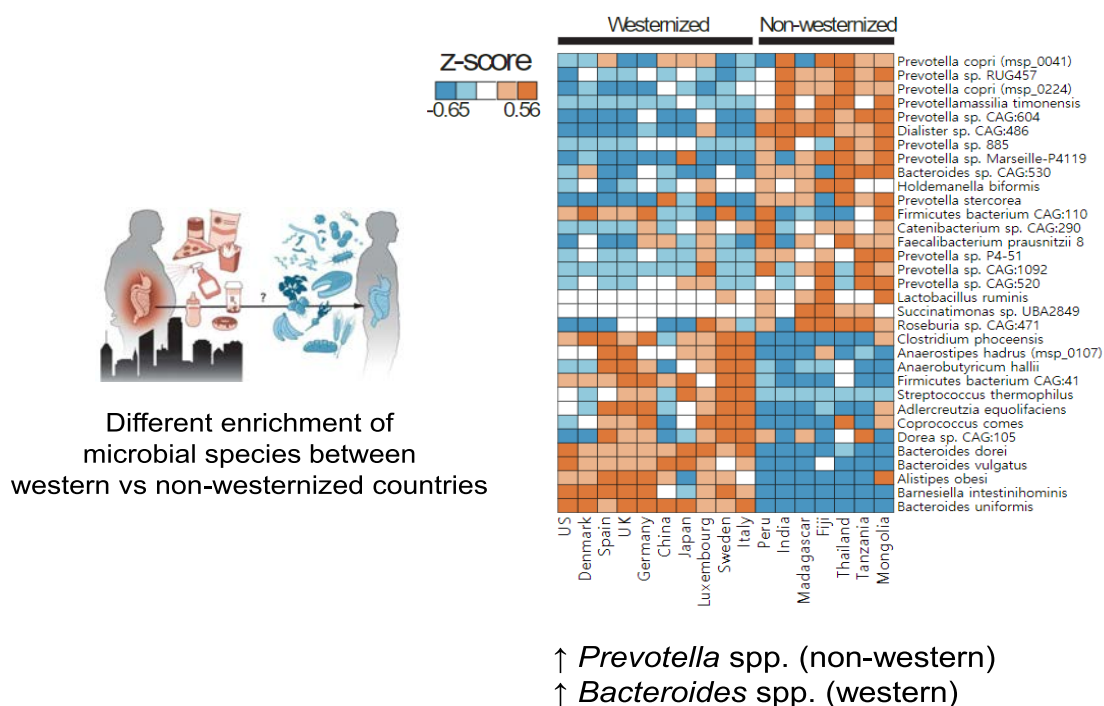
**ET Prevotella:** fiber-rich diet  
**ET Bacteroides:** fat-rich diet

Enterotypes

REF | Paul I Costea et al., Nature Microbiology, 2018

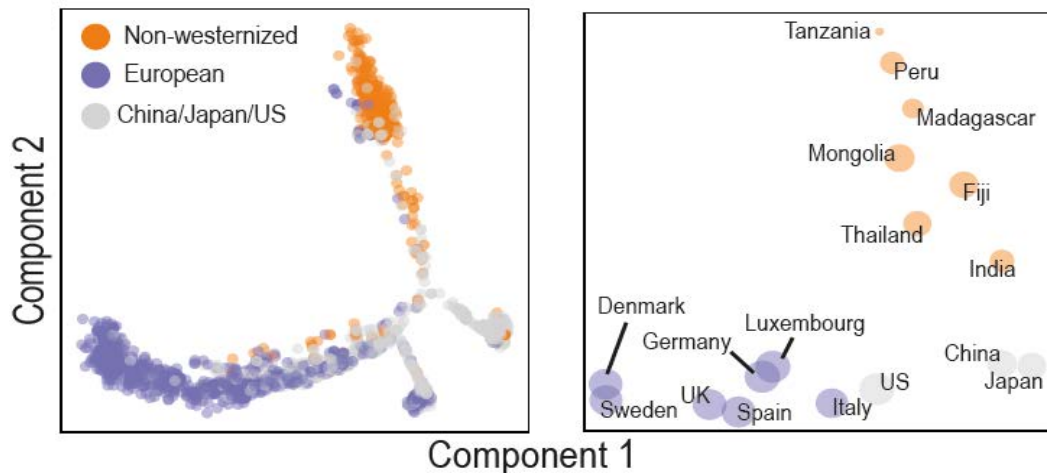
17

## Global healthy microbiome



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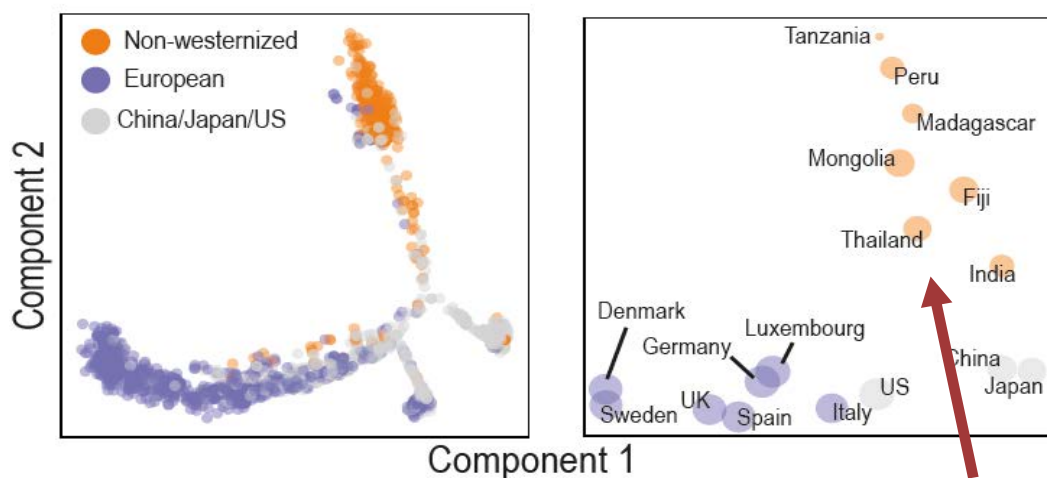
## Global healthy microbiome



Three geographical clusters were identified

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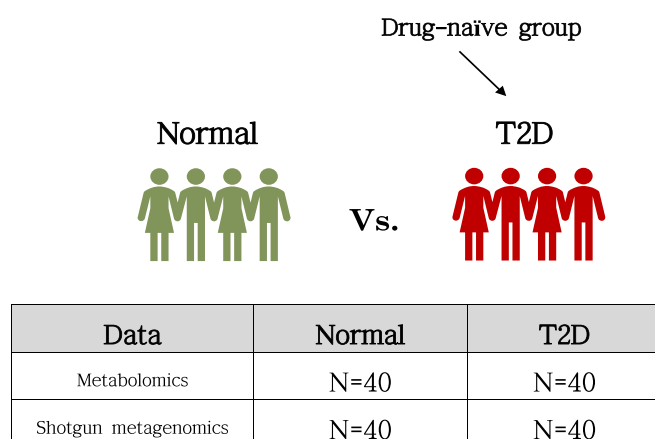
## Global healthy microbiome



Three geographical clusters were identified

20

## Korean microbiome data ?!



### KoGES cohort

**Shotgun metagenome:**  
Illumina NovaSeq 6000 150  
PE sequencing (10Gb)

**Metabolomics:**  
Biocrates MxP Quant 500  
(targeted metabolomics)



Namhan Cho Jang Won Son

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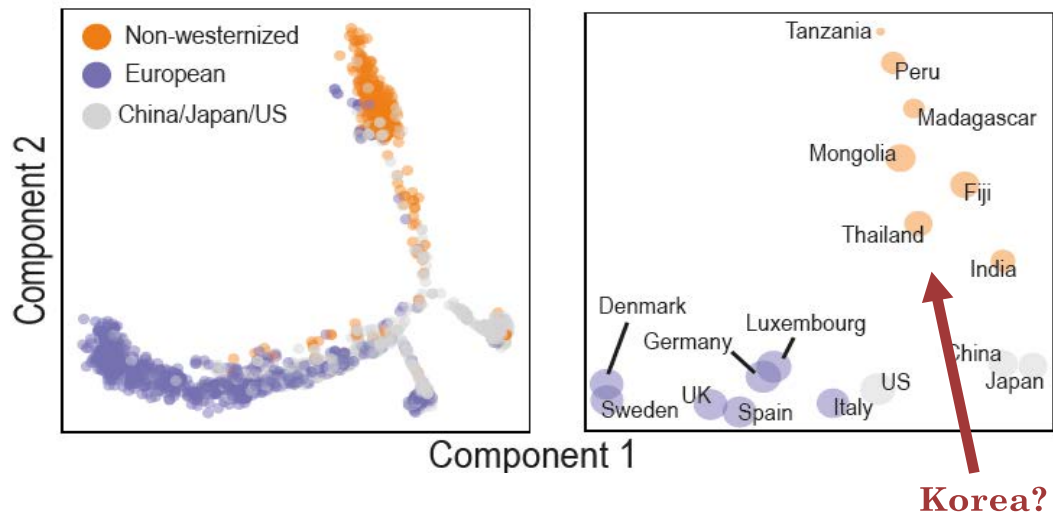
## Korean microbiome data ?!

Top 5 bacteria(species)	Name	percent
Control	<i>Prevotella copri</i>	24%
	<i>Bacteroides vulgatus</i>	4%
	<i>Bacteroides plebeius</i>	3%
	<i>Escherichia coli</i>	2%
	<i>Eubacterium rectale</i>	1.4%
DM(diabetes mellitus)	<i>Prevotella copri</i>	20%
	<i>Bacteroides vulgatus</i>	3.6%
	<i>Bacteroides uniformis</i>	2.5%
	<i>Prevotella stercorea</i>	2%
	<i>Bacteroides stercoris</i>	1.7%
Pre-DM	<i>Prevotella copri</i>	11%
	<i>Escherichia coli</i>	7%
	<i>Bacteroides vulgatus</i>	4.4%
	<i>Bacteroides plebeius</i>	3%
	<i>Prevotella copri-2</i>	2.6%

UNPUBLISHED

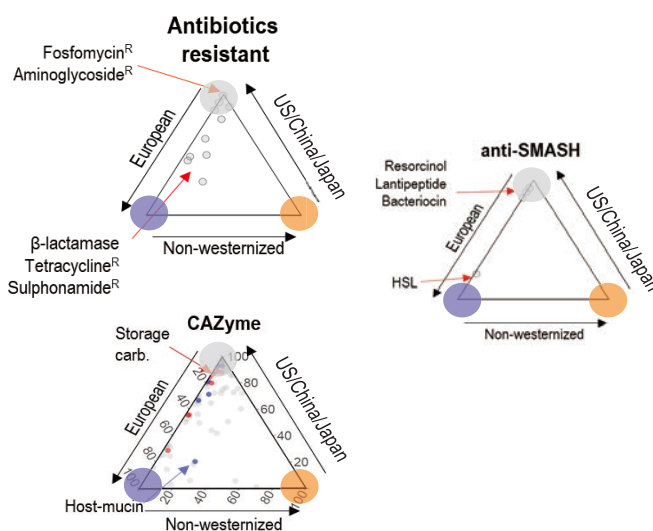
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## Global healthy microbiome



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## Functional enrichment in different geographical clusters



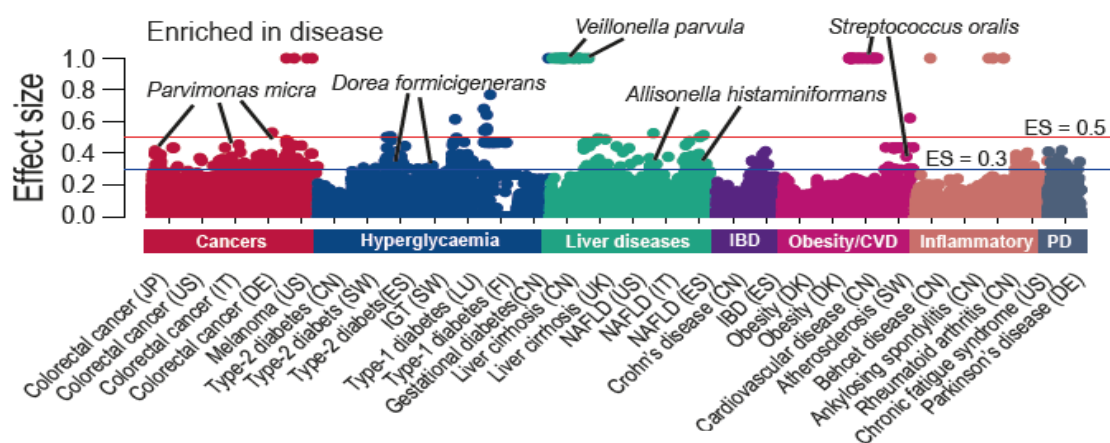
### Industrialized countries harbours

- antibiotic resistant genes
- mucin degrading enzymes
- secondary metabolites

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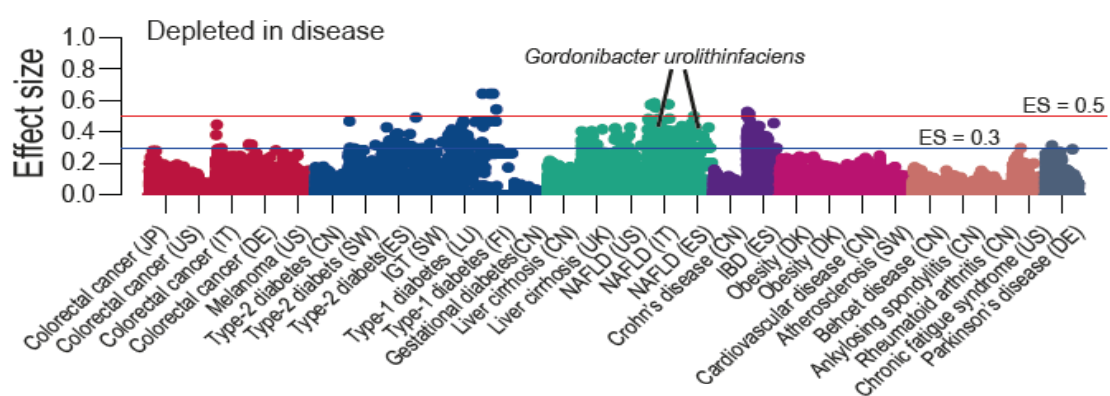


## Pan-metagenomics associations study (Pan-MGAS)



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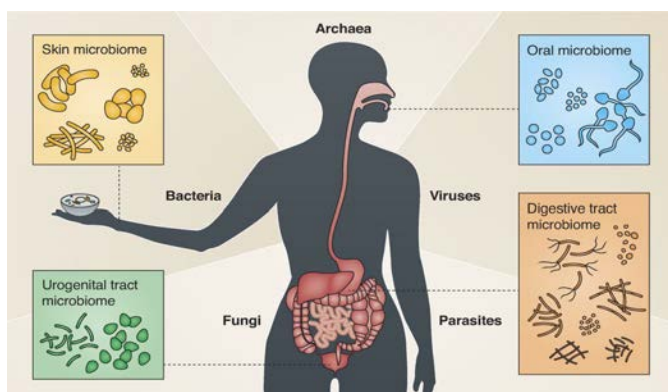
## Pan-metagenomics associations study (Pan-MGAS)



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## Human gut-oral microbiome axis



Wendy et al., 2015, JCB

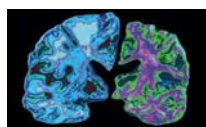
**Oral microbiome**  
- The second largest and diverse microbiota after gut

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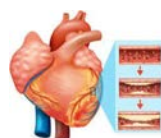
## Oral microbiome in diseases



Periodontitis



Alzheimer's



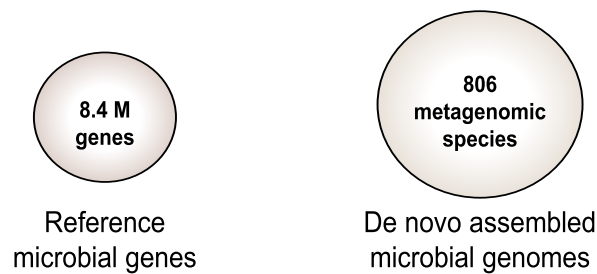
Cardiovascular



Liver cirrhosis

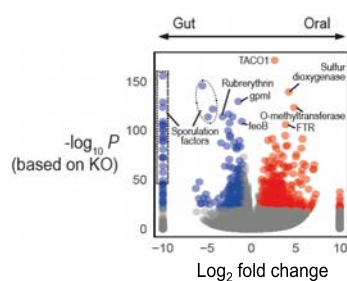
30

## Establishing human oral microbiome reference



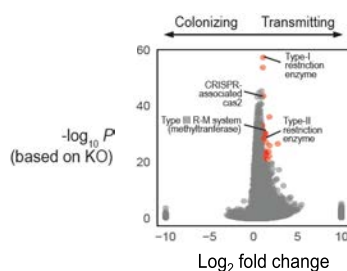
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## Contrasted functions of gut and oral microbiome



**Gut species:**  
endospore functions

**Oral species:**  
oxidative damage system  
(peptide methionine sulfoxide reductase)

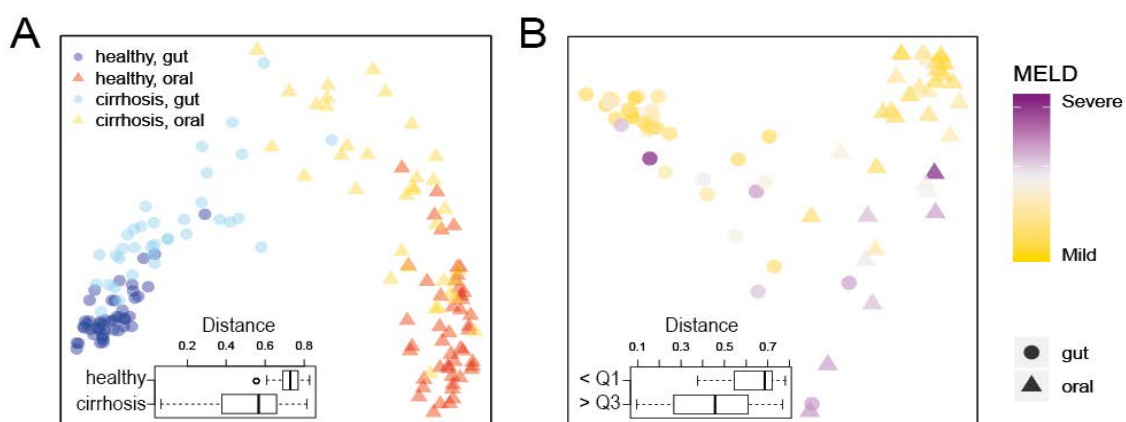


**Gut-oral transmitting species:**  
Bacterial immune systems such as  
CRISPR-cas, Restriction-modification system

Unpublished

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## Gut-oral microbiome axis in liver cirrhosis

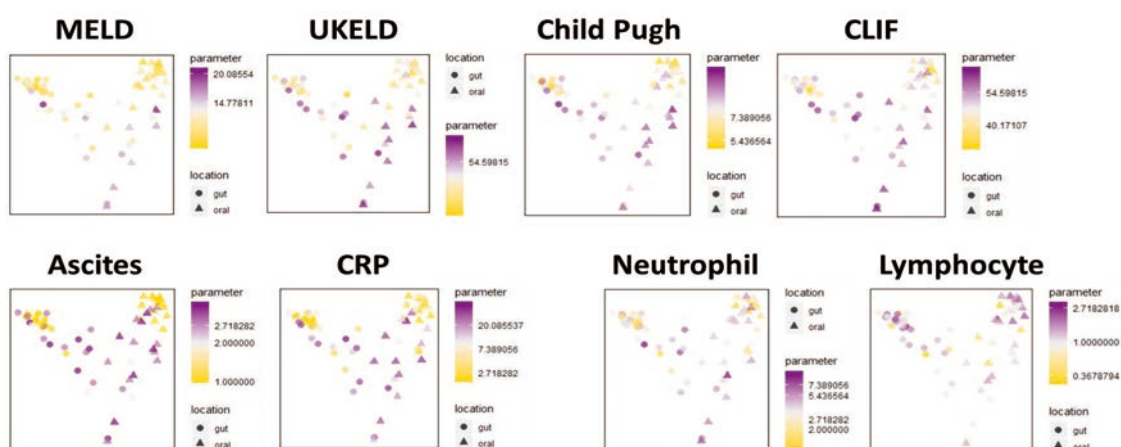


Gut and oral microbiome were shared as cirrhosis progressed

Unpublished

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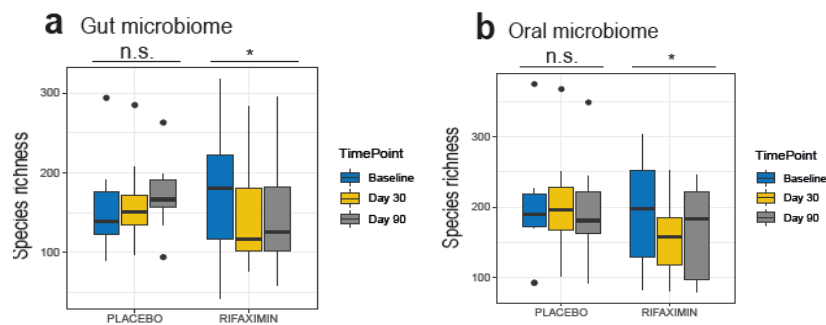
## Gut and oral microbiome in liver cirrhosis



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## Rifaximin intervention study

- suppressing gut-oral translocation and complications (hepatic encephalopathy)

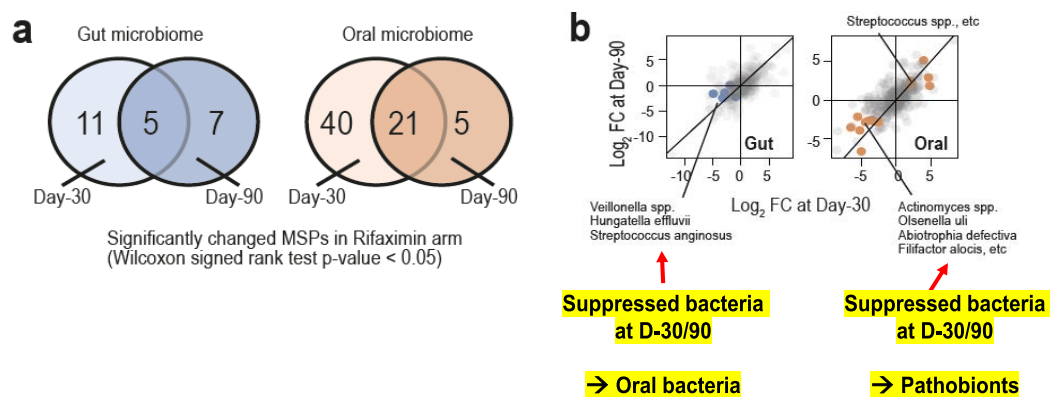


REF | Vishal C Patel, Sunjae Lee et al., J Hepatology (2021)

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## Rifaximin intervention study

- suppressing gut-oral translocation and complications (hepatic encephalopathy)



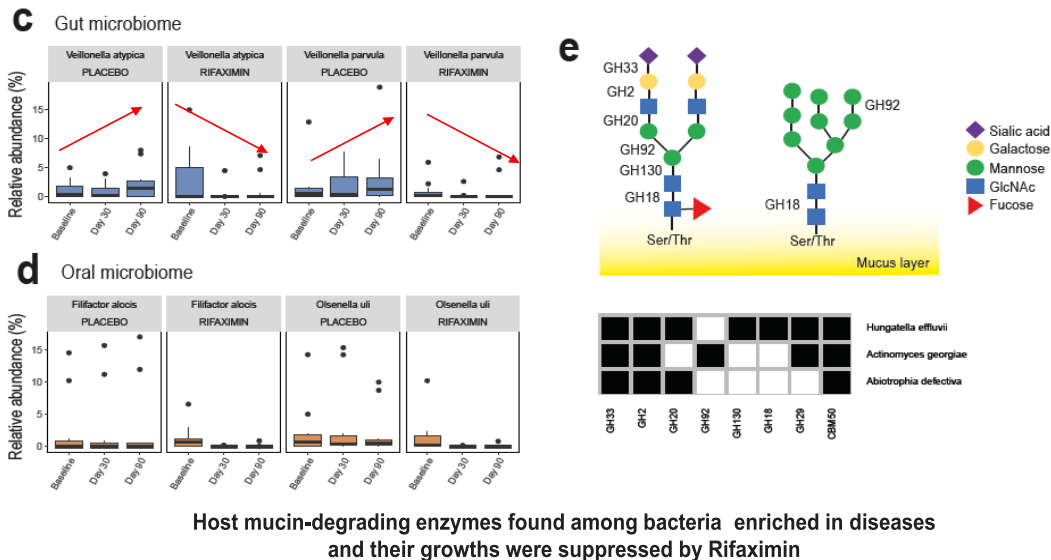
REF | Vishal C Patel, Sunjae Lee et al., J Hepatology (2021)

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# Rifaximin intervention study

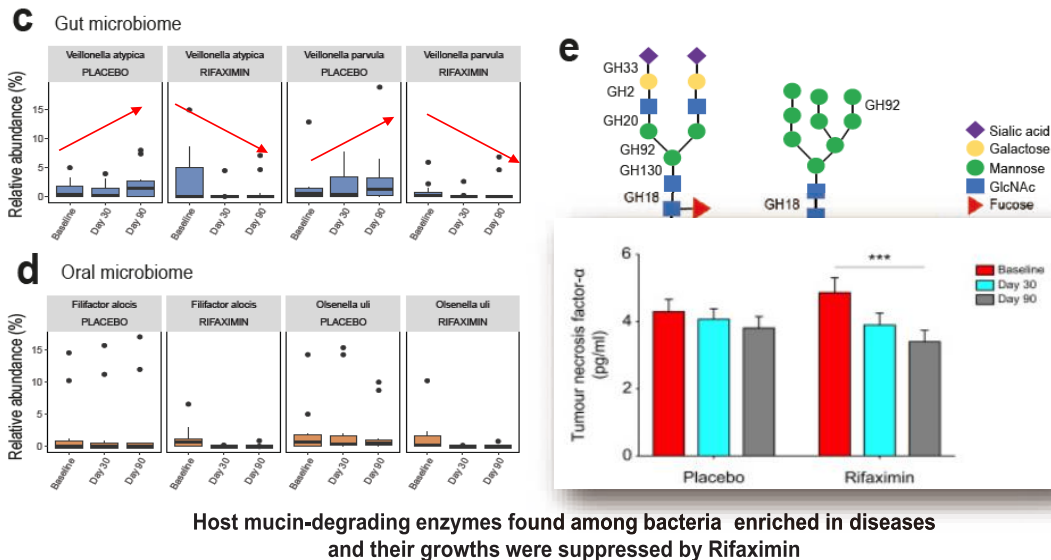
- suppressing oralization of gut bacteria and complications (hepatic encephalopathy)



REF | Vishal C Patel, Sunjae Lee et al., J Hepatology (2021)

# Rifaximin intervention study

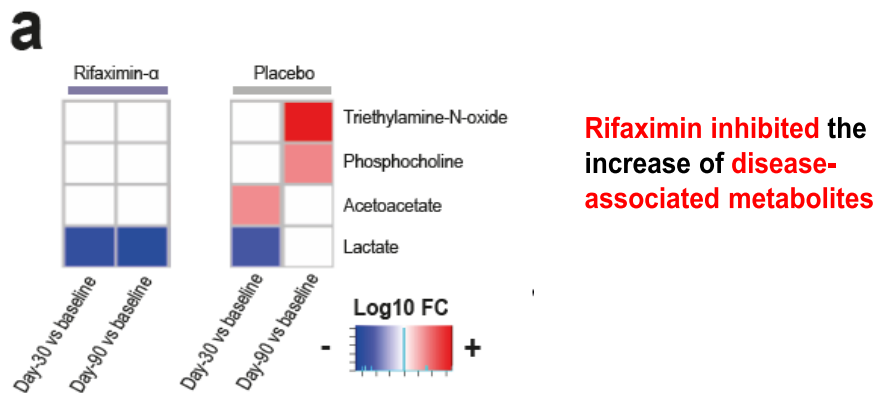
- suppressing oralization of gut bacteria and complications (hepatic encephalopathy)



REF | Vishal C Patel, Sunjae Lee et al., J Hepatology (2021)

## Rifaximin intervention study

- suppressing gut-oral translocation and complications (hepatic encephalopathy)

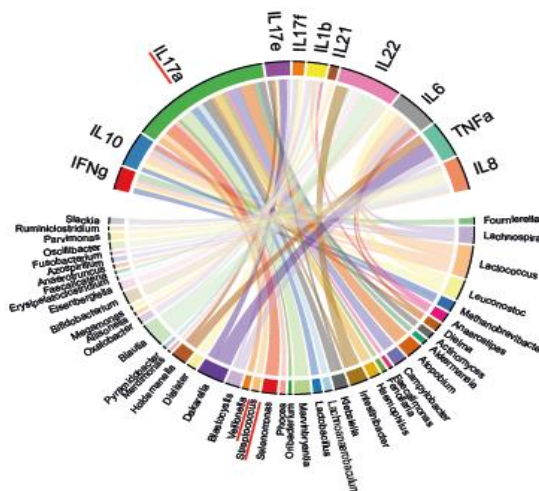


REF | Vishal C Patel, Sunjae Lee et al., J Hepatology (2021)

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## Rifaximin intervention study

- suppressing gut-oral translocation and complications (hepatic encephalopathy)



**Inflammatory cytokine (IL17a) associated with oral bacteria in the gut (Veillonella and Streptococcus)**

REF | Vishal C Patel, Sunjae Lee et al., J Hepatology (2021)

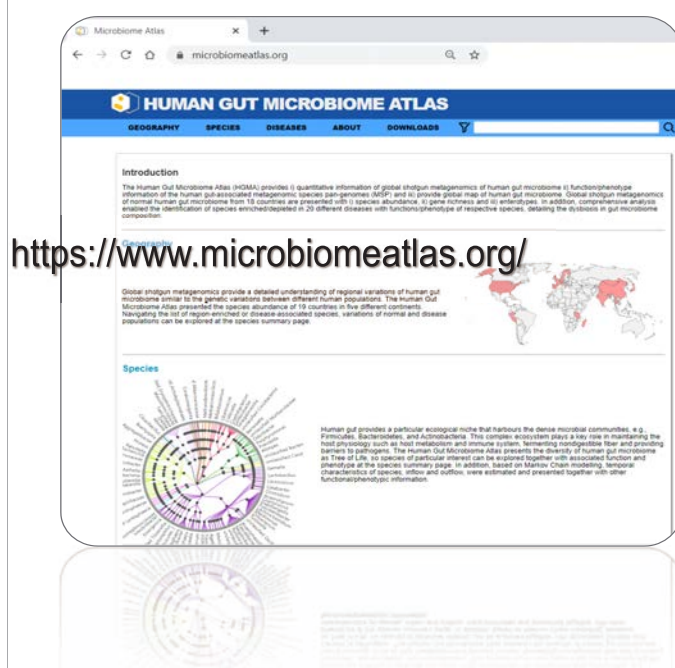
40

## Conclusions

**Exploration of systemic microbiome  
(i.e., gut & oral) is necessary for  
disease studies**

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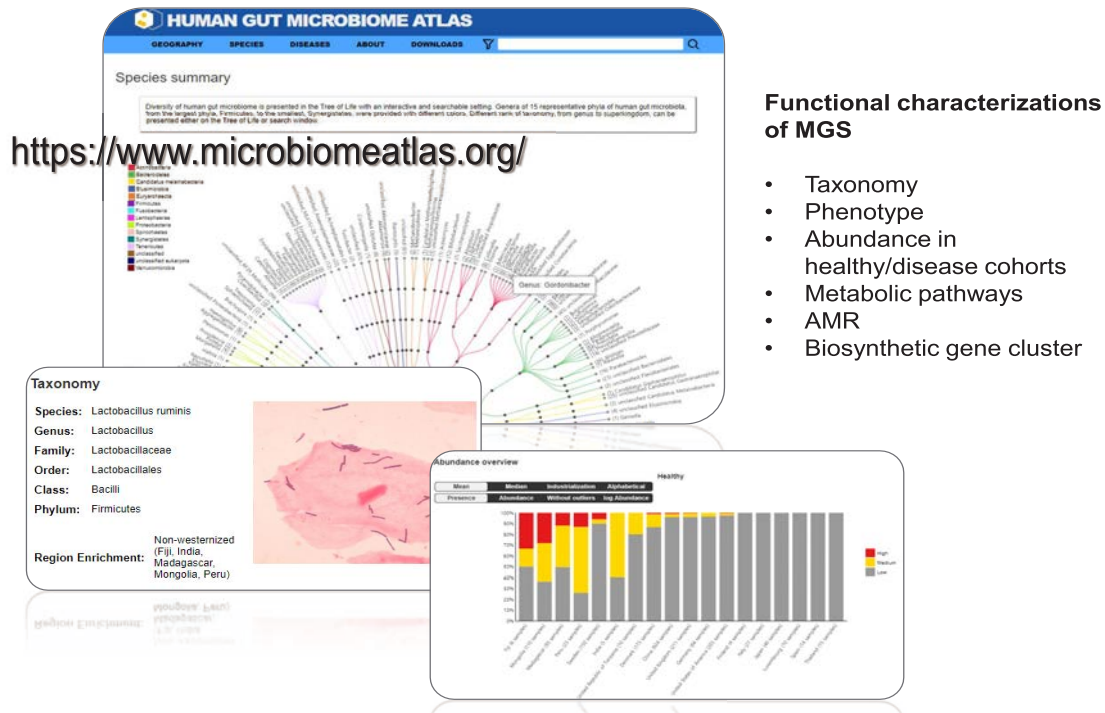
## Human Gut Microbiome Atlas (HGMA)



- New resource of human gut metagenome data was launched – April, 2021
- Total 1,989 gut metagenomic species (MGS) were profiled across 6,000 metagenomics samples
- Meta-analysis of metagenomics studies for the region/disease-enrichment of species

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## Human Gut Microbiome Atlas (HGMA)



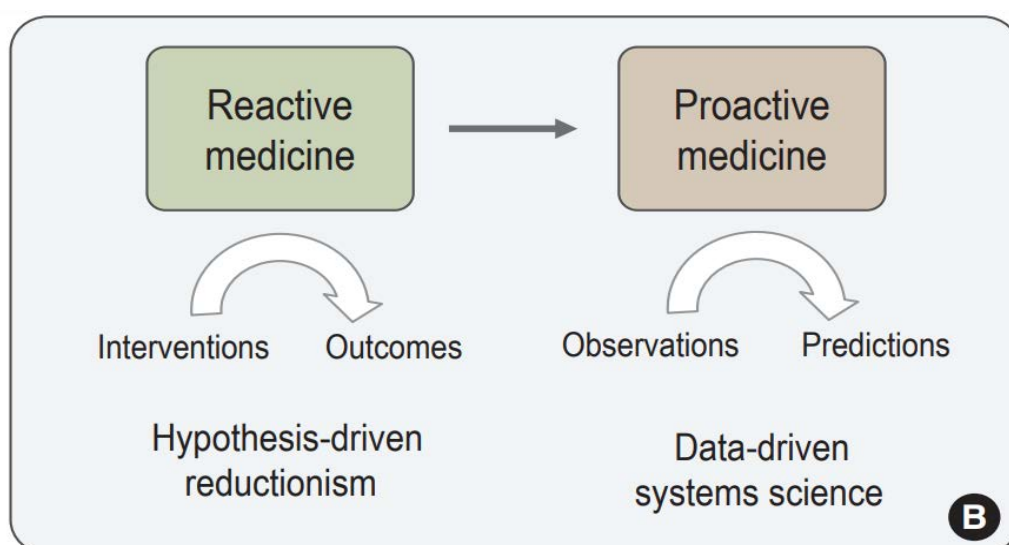
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## Human Gut Microbiome Atlas (HGMA)

Msp_0004	<i>Blastocystis</i> sp. subtype 2	<i>Blastocystis</i>	Peru, United Republic of Tanzania	Ankylosing spondylitis (ES=0.310262) Atherosclerosis (ES=0.301437) Colorectal cancer (ES=0.319117) Colorectal cancer (ES=0.33148) Crohn's disease (ES=0.311) NAFLD (ES=0.311) Type 1 diabetes (ES=0.667388) Type 1 diabetes (ES=0.349634) Type 2 diabetes (ES=0.460432)	Inflow (0.437) Outflow (0.379)
Msp_0005	<i>Escherichia coli</i>	<i>Escherichia</i>	India, Madagascar		
Msp_0006	<i>Blastocystis</i> sp. subtype 4	<i>Blastocystis</i>	European		
Msp_0017	<i>Roseburia intestinalis</i>	<i>Roseburia</i>	Germany, Luxembourg	(ES=0.310044) Atherosclerosis (ES=0.341434) Colorectal cancer (ES=0.375093) Colorectal cancer (ES=0.32) Crohn's disease (ES=0.31) Type 1 diabetes (ES=0.306258) Type 2 diabetes (ES=0.350805)	Inflow (0.388)
Msp_0018	<i>Parabacteroides</i>	<i>Parabacteroides</i>	European		

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## Future medicine



REF | Jang Won Son et al., Endocrinology and Metabolism, 2020

45

## Acknowledgement

**GIST**  
All lab members



**GARD cohort**  
**KOGES cohort**  
**CNU**  
**SNU**  
**Catholic Univ.**  
**POSTECH**  
**Gacheon Univ.**

**INRAe**  
S Dusko Ehrlich  
Mathieu Almeida  
Emmanuelle Le Chatelier  
...



**Thank you for your  
attention**

**King's College London**  
Saeed Shoale  
Gholamreza Bidkhori  
...

**SciLifeLab**  
Adil Mardinoglu  
Mathias Uhlen  
Jens Nielsen  
...

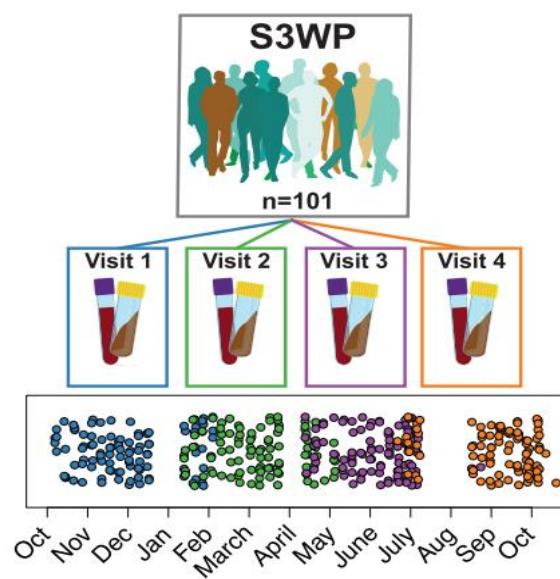


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## Appendix

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## Big data analysis of longitudinal microbiome

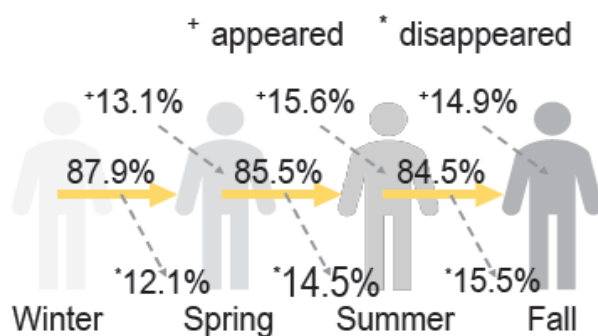


NPJ Biofilms & Microbiomes

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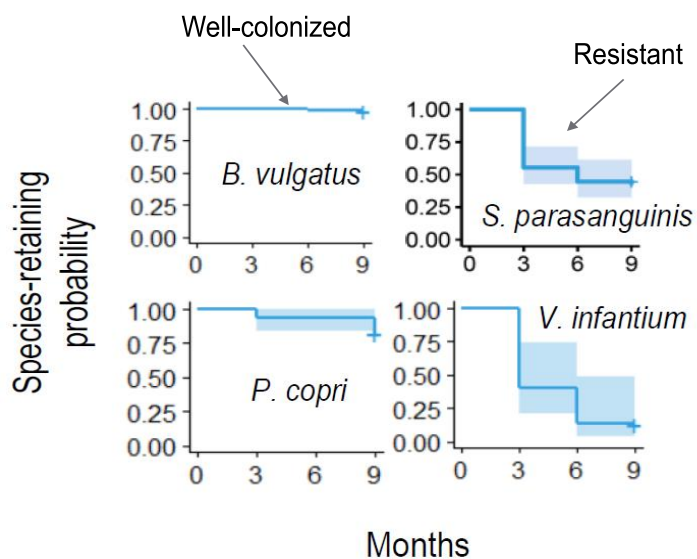
## Big data analysis of longitudinal microbiome



We ourselves are similar in microbiome by time (**~80%**)

49

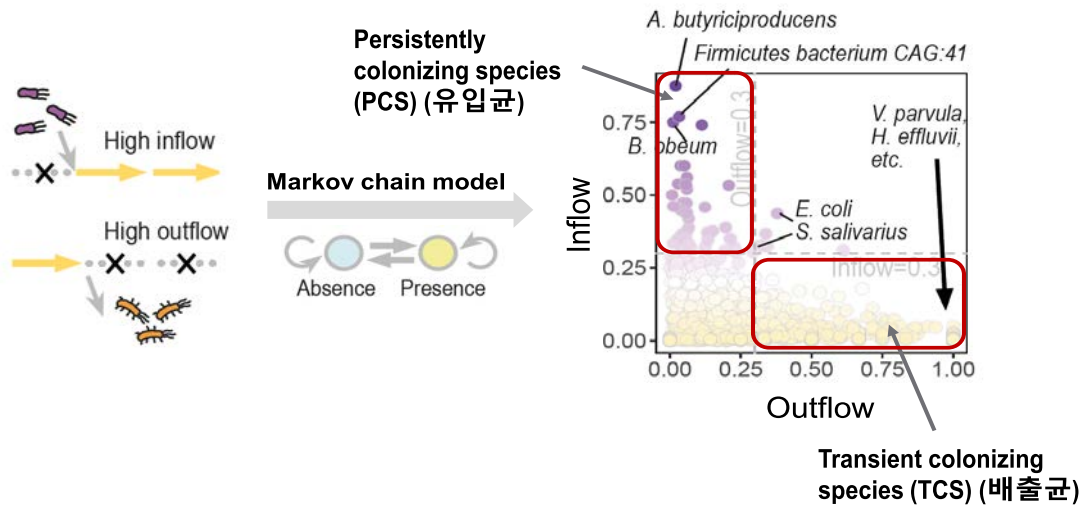
## Big data analysis of longitudinal microbiome



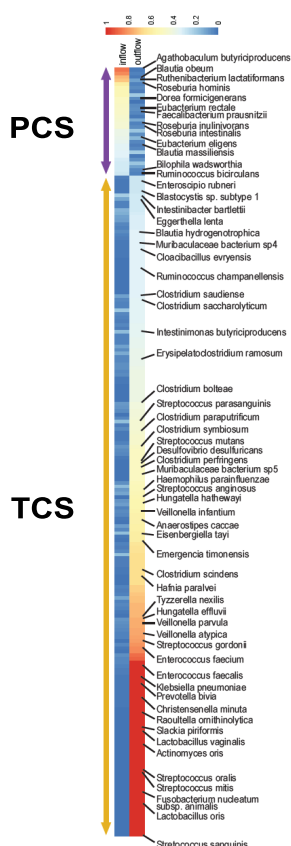
However, longitudinal behaviour of individual species are very distinct

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## Modeling of longitudinal microbiome changes



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**SCFA producers**  
(*Eubacterium rectale*, *Blautia obeum*, etc)

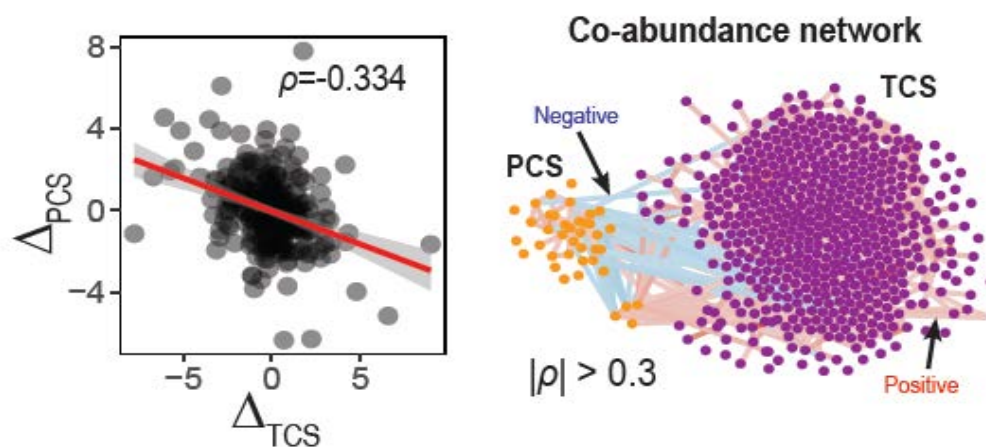
**Pathobionts** (*Enterococcus faecalis*,  
*Klebsiella pneumoniae*, *Clostridium* spp.  
*Erysipelatoclostridium* spp. etc)

**Oral bacteria** (*Streptococcus* spp.,  
*Veillonella* spp., etc)

**Lactobacillus** spp.

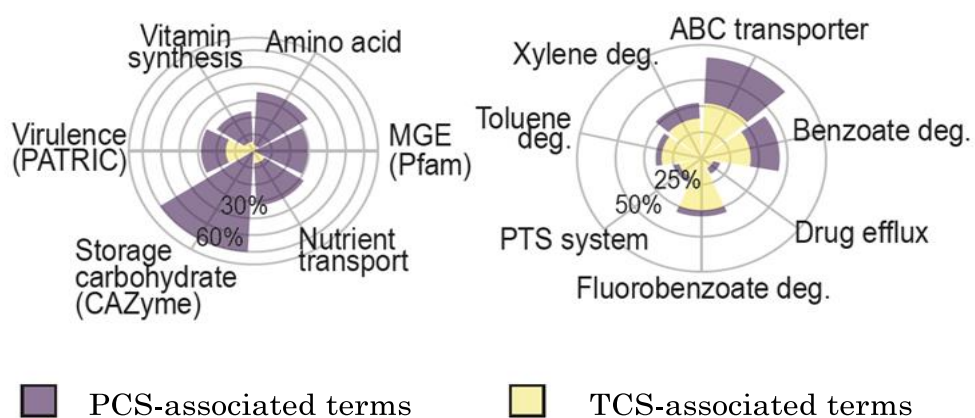
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## Negative correlations between PCS(유입균) & TCS(배출균)



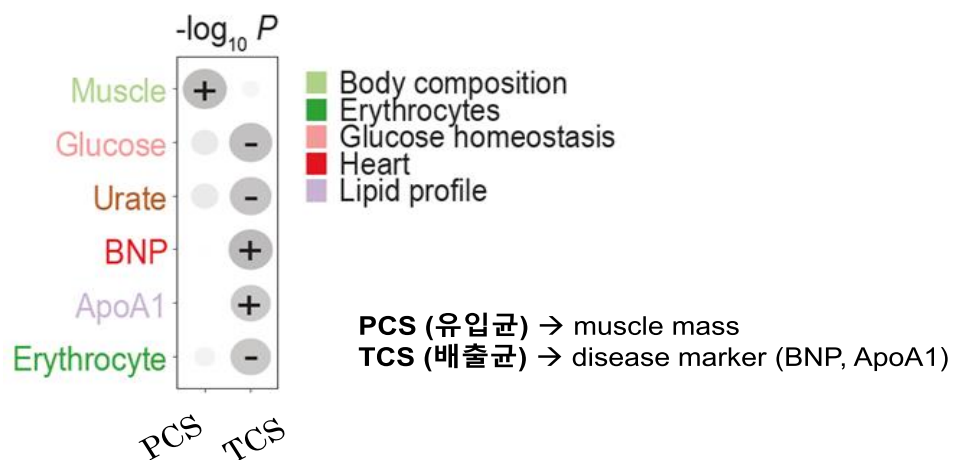
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## Different functional enrichment of PCS (유입균) vs TCS (배출균)



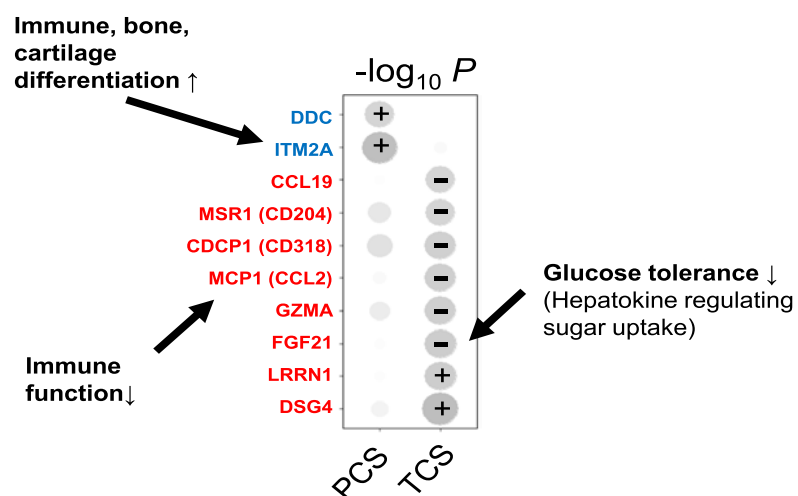
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## Correlations with clinical metadata



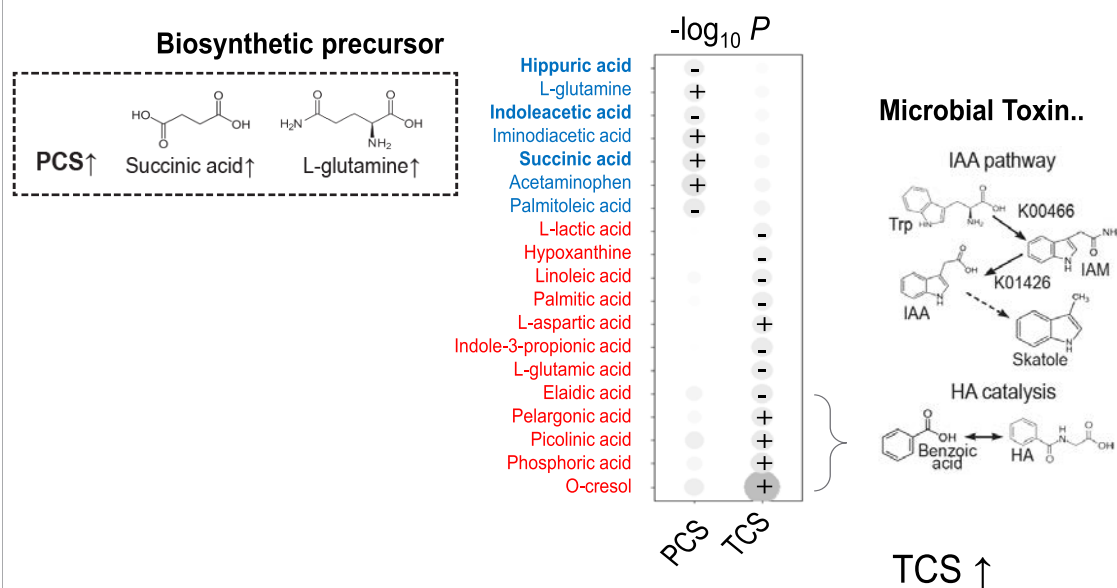
55

## Correlations with plasma proteomics & metabolomics



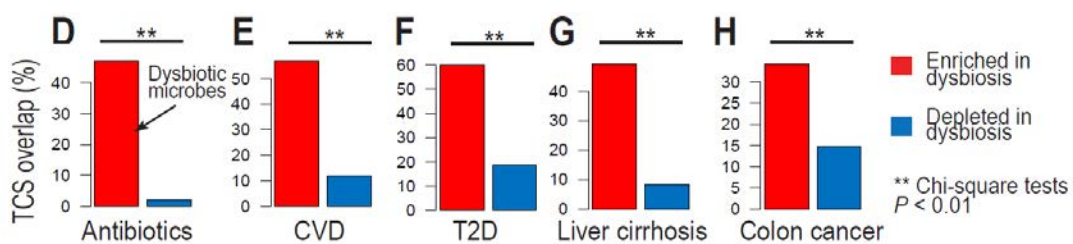
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## Correlations with plasma proteomics & metabolomics



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## Rebalancing TCS/PCS will be new therapeutic strategy

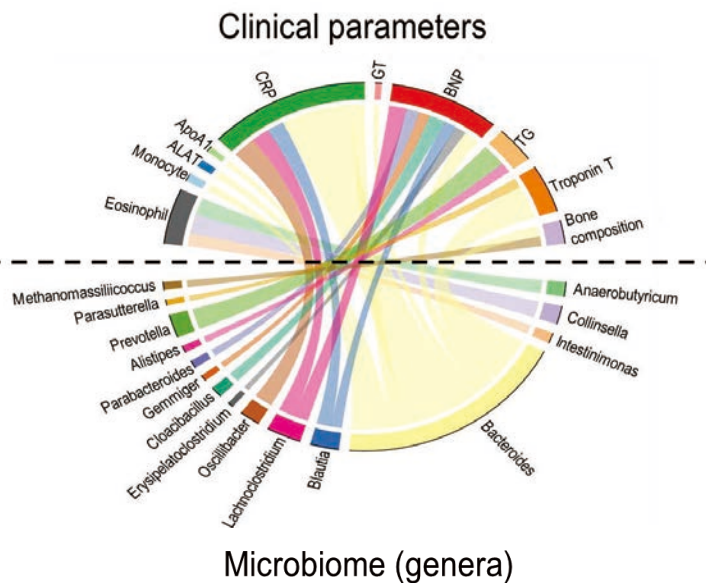
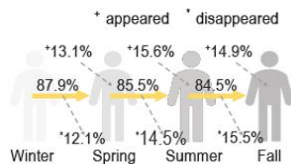


**Disease microbiome was mostly “TCS species” → suppressing TCS will be new therapeutic strategy**

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Host phenotypes explained by microbiome changes,  
even from **normal populations**

A year-long follow-up study  
of 86 healthy Swedish

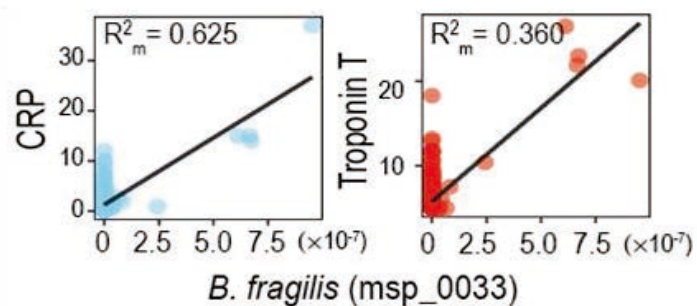


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Host phenotypes explained by microbiome changes,  
even from **normal populations**

Inflammation

Heart failure

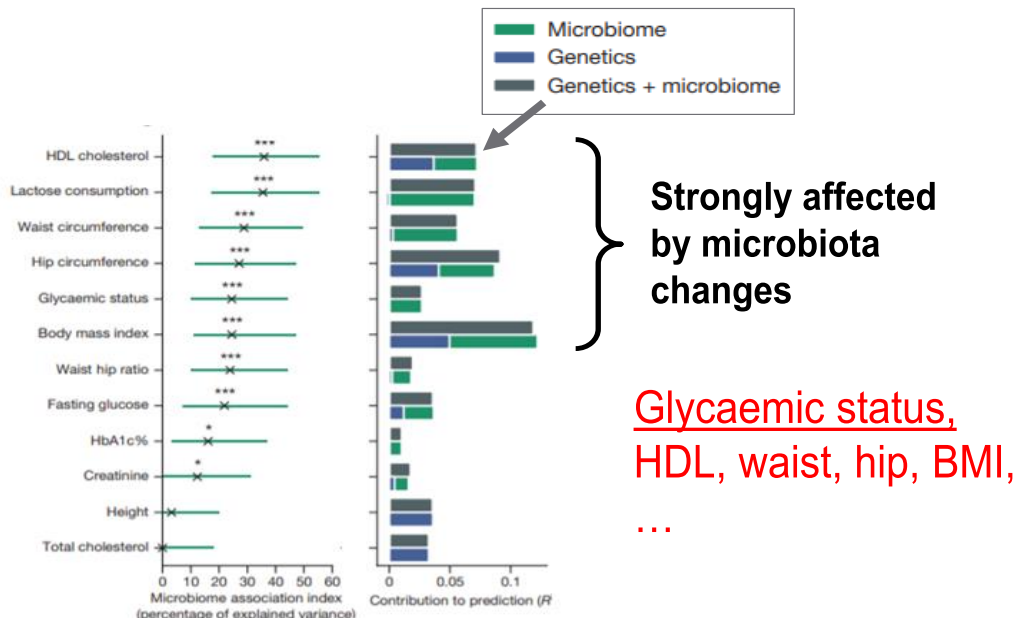


Unpublished

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## Host phenotypes explained by microbiome changes, even from **normal populations**



REF | Daphna Rothschild et al., Nature, 2018

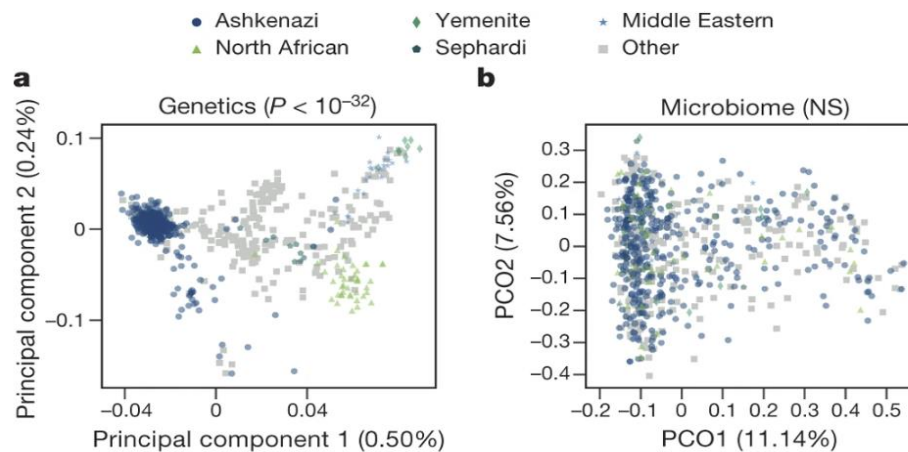
이스라엘 Eran Segal 그룹

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## Current findings of human microbiome

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## 1 Microbiome ≠ Genome ?

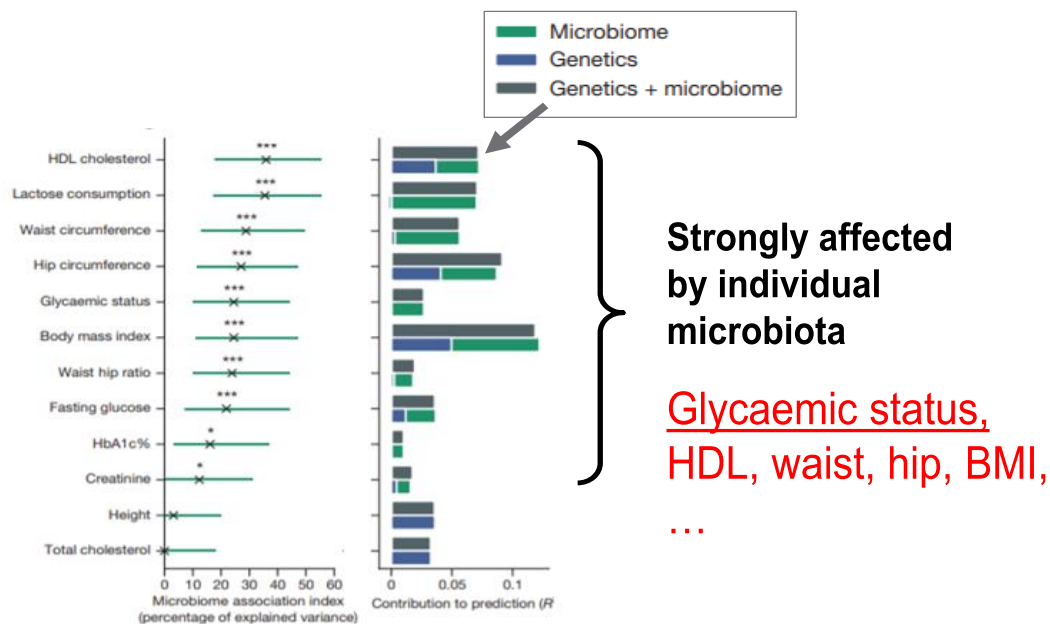


Microbiome ≠ Ethnicity (i.e. genetic factors)

REF | Daphna Rothschild et al., Nature, 2018

63

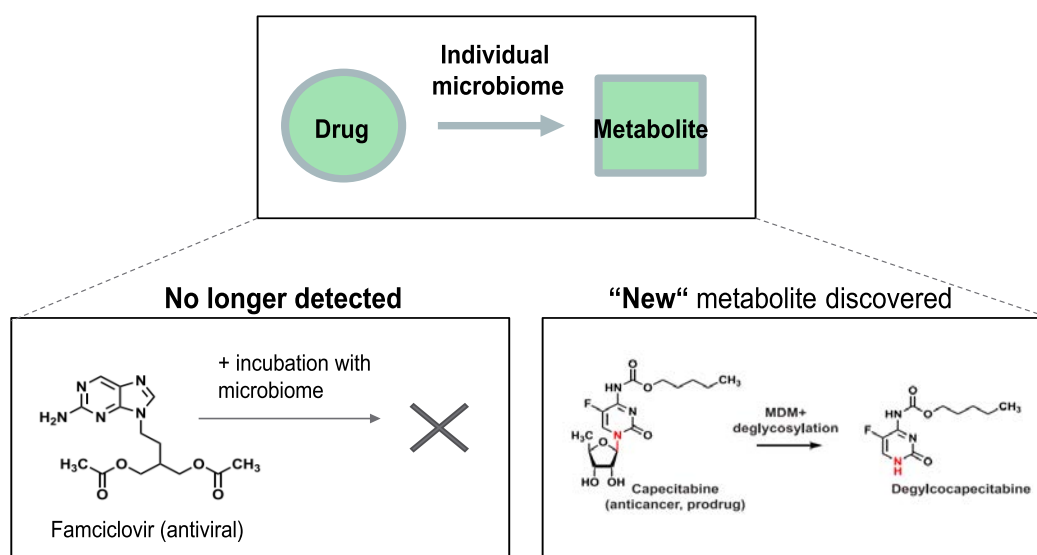
## 1 Microbiome >> Genome ?



REF | Daphna Rothschild et al., Nature, 2018

64

## 2 Microbiome affects individual drug response

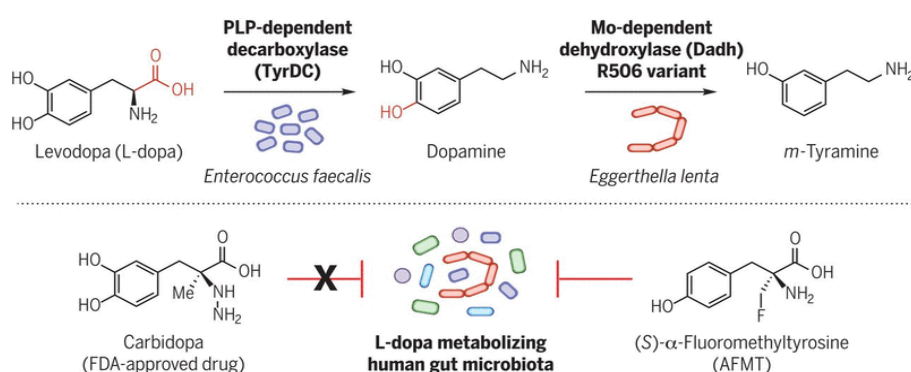


REF | Bahar Javdan et al., Cell, 2020

65

## 2 Microbiome affects individual drug response

### Microbiome affects Levodopa efficiency

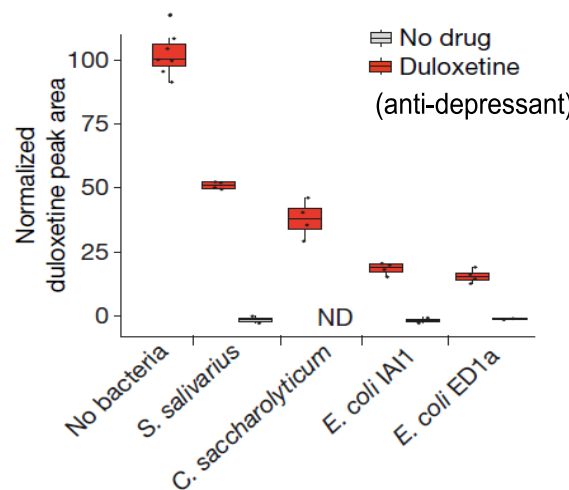


REF | VM Rekdal et al., Science 2019

66

## 2 Microbiome affects individual drug response

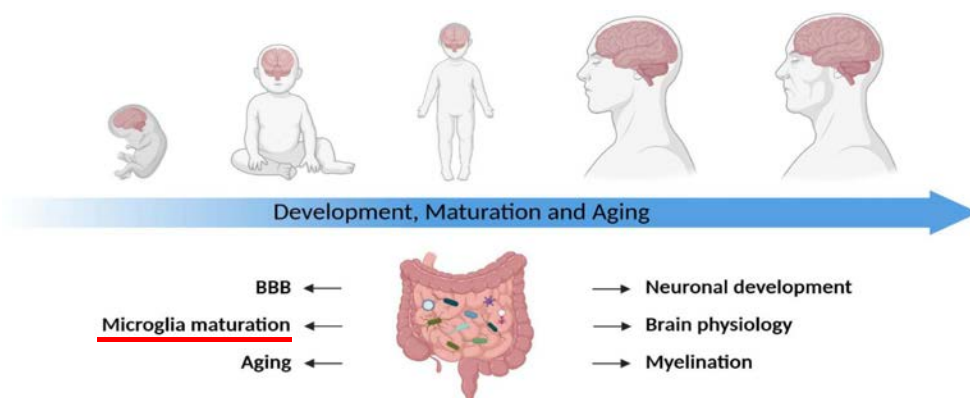
**Microbiome can bio-accumulate host-direct drugs**



REF | Martina Klünemann et al., Nature, 2021

67

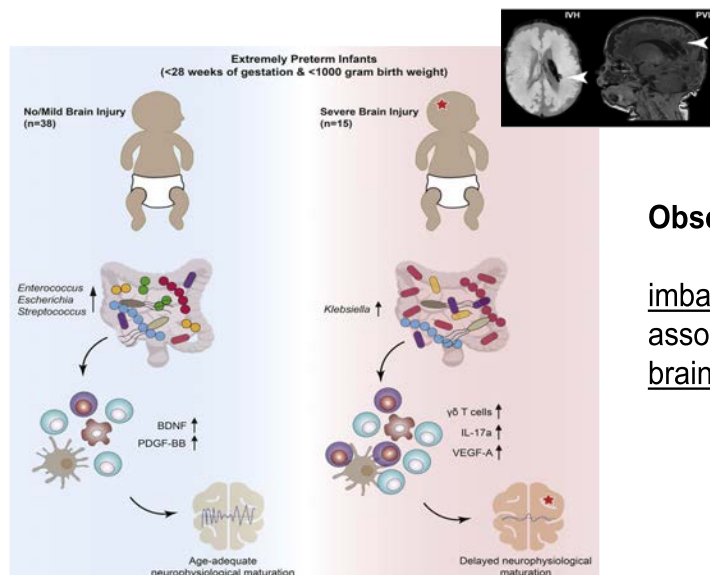
## 3 Microbiome affects brain development



**Gut Microbiome is important**

68

### 3 Microbiome affects brain development



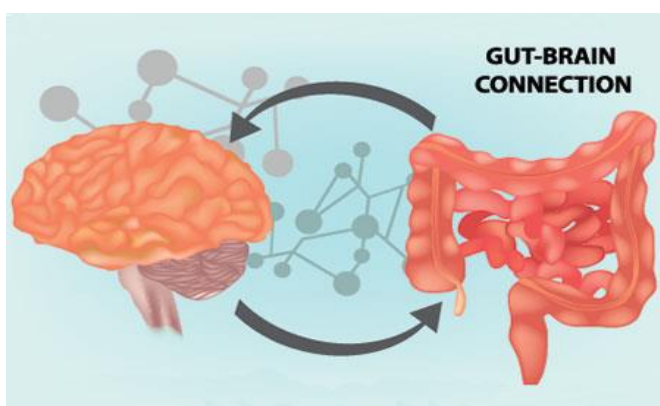
#### Observational evidence:

imbalanced microbiome was associated with brain injury of preterm baby

REF | David Seki et al., Cell Host & Microbe (2021)

69

### 4 Gut-Brain Axis



- **Bi-directional** communications between the **intestine** and **CNS** exist
- **Human gut microbiota** has a **key role** in signaling between gut and brain

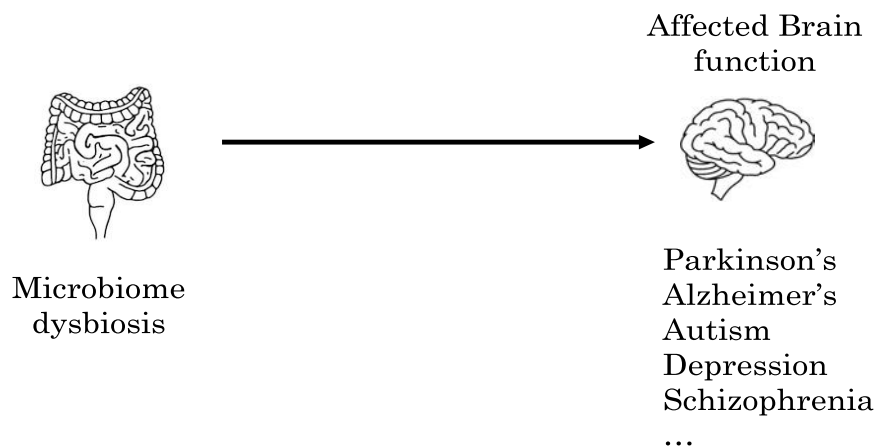
REF | Emeran A Mayer et al., Annual Review of Medicine (2022)

REF | Lisa D Blackmer-Raynolds & Timothy R Sampson, Cell Host & Microbe (2022)

REF | Livia H Morais et al., Nature Reviews Microbiology (2020)

70

## 4 Gut-Brain Axis



REF | Brittany D Needham et al., Nature Reviews Neuroscience (2020)

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## Integrated understanding of multi-omics



**What will happen?**

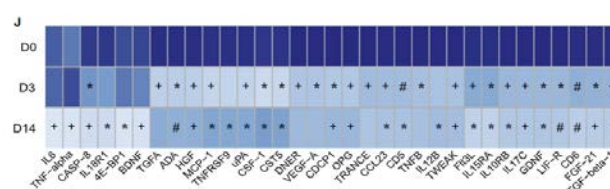
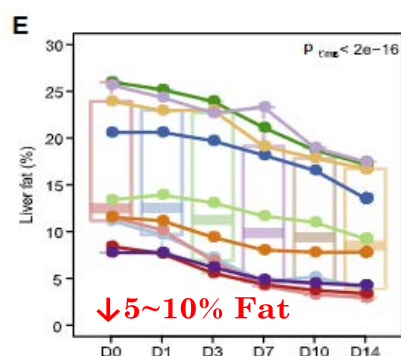
REF | Adil Mardinoglu et al., Cell Metabolism, 2018

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## Integrated understanding of multi-omics

### Low carb. diet for NAFLD patient (isocaloric)



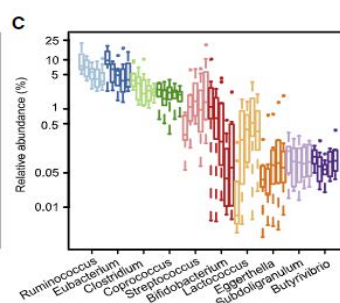
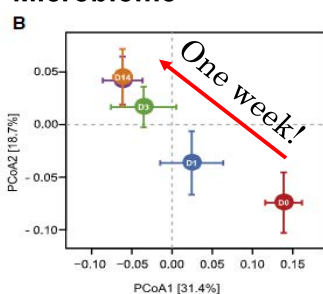
- Decreased liver fat
- Decreased inflammation

REF | Adil Mardinoglu et al., Cell Metabolism, 2018

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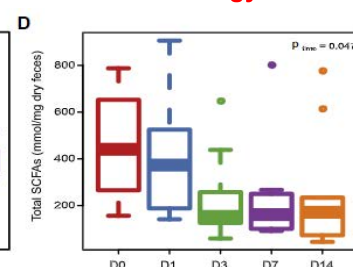
## Integrated understanding of multi-omics

### Microbiome



↓ Butyrate producers

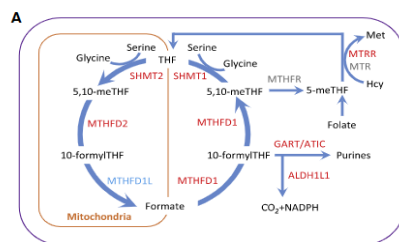
### Decreased energy harvest



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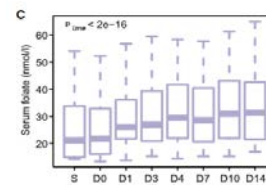
## Integrated understanding of multi-omics

## Liver transcriptome



## Glutathione metabolism

## Microbial metabolome



Increased folate synthesis

## Supporting fat burning

# 02

## Analysis of microbiome genetic information using artificial intelligence

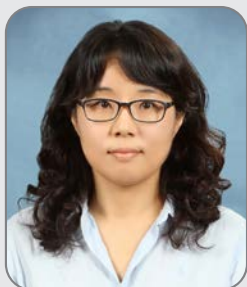


노미나  
(한양대학교)





## 3부 연자



- ✓ 소속: 한양대학교
- ✓ 직위/직책: 교수
- ✓ 성명: 노미나

### 🔍 학력사항

1992-1998	이화여자대학교 학사
1999-2001	Boston University 석사
2003-2009	Indiana University 박사

### 🔍 경력사항

2012-2013	Roswell Park Cancer Institute 조교수
2013-현재	한양대학교 조교수, 부교수, 교수

# 인공지능을 이용한 마이크로바이옴 유전정보 분석

The human microbiome plays a crucial role in health and disease, influencing processes such as digestion, immunity, and metabolism. Recent advancements in high-throughput sequencing have generated vast amounts of microbiome data, offering unprecedented opportunities to understand these complex ecosystems. However, the complexity of microbiome datasets pose significant analytical challenges. Artificial Intelligence (AI) has emerged as a transformative tool for microbiome analysis, offering the ability to uncover intricate patterns, predict functional interactions, and provide personalized insights. In particular, deep learning methods enable robust analysis of microbial diversity, functions, and interactions in the microbiomes. These approaches excel in integrating multi-omics data, modeling microbial networks, and identifying biomarkers for health and disease. Moreover, AI-driven predictive models are reshaping personalized medicine by linking microbial profiles to therapeutic responses. Despite its potential, AI-based microbiome analysis faces challenges such as data standardization, interpretability of models, and the need for high-quality annotated datasets. Addressing these challenges through interdisciplinary collaboration will unlock the full potential of AI to revolutionize microbiome research, fostering new frontiers in diagnostics, therapeutics, and precision health.

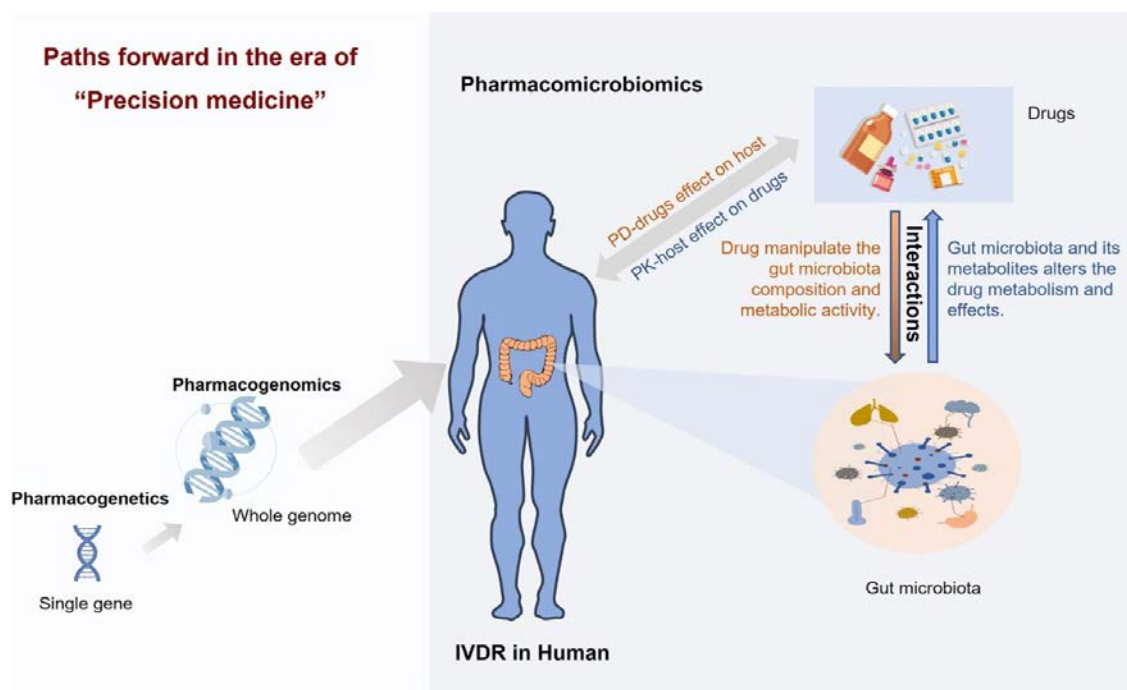


2024 병원기반 인간 마이크로바이옴 연구개발사업 심포지엄  
2024.12.03

## Analysis of microbiome genetic information using artificial intelligence

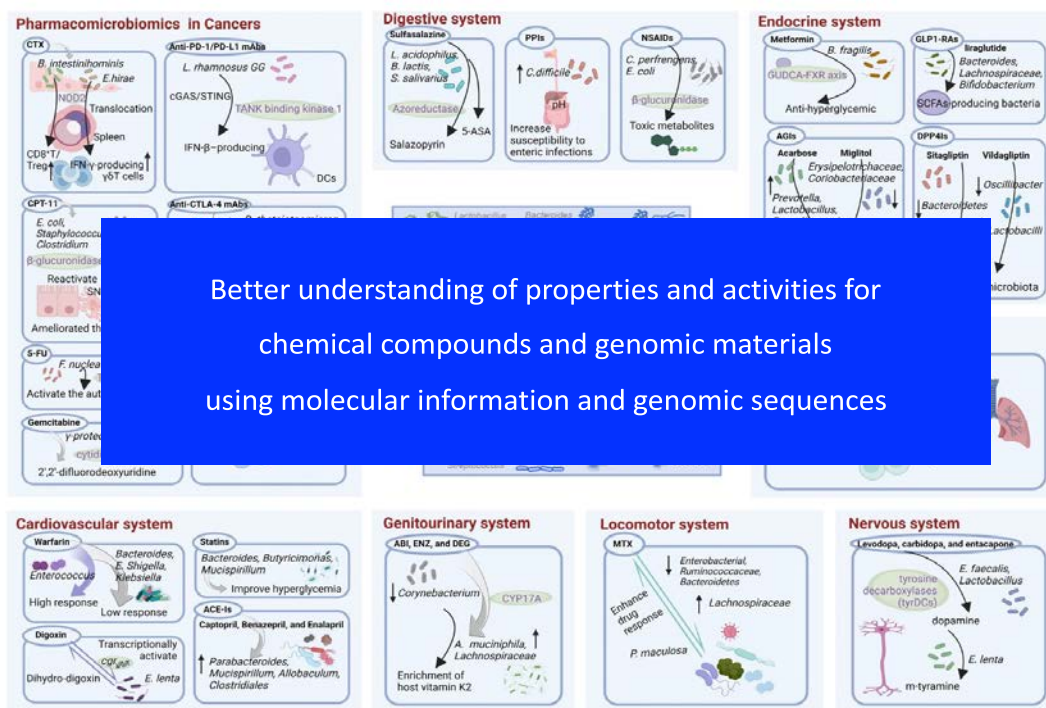
노미나  
한양대학교

### Pursuing personalized treatments



Sig Transduct Target Ther, 2023

## Pursuing personalized treatments



Sig Transduct Target Ther, 2023

**CAS**

A division of the American Chemical Society



201 million organic and inorganic substances, including alloys, coordination compounds, minerals, mixtures, polymers, and salts disclosed in publications since the early 1800s.

How big is the known chemical space?

279M published substances in CAS (2024)

230M commercial small molecules in ZINC15 (2022)

112M NIH depository of compounds (PubChem, 2022)

2.3M small molecules annotated w/ bioactivity data (ChEMBL, 2022)

1.1M small molecules annotated w/ bioactivity data (BindingDB, 2022)

11.9K small-molecule drugs (DrugBank, 2022)

4.2K approved drugs (DrugBank, 2022)



2.7K approved small molecule drugs (DrugBank, 2022)

<https://www.cas.org/cas-data/cas-registry>

<https://zinc15.docking.org/>

<https://go.drugbank.com/stats>

<https://pubs.acs.org/doi/10.1021/ar500432k>

 **InterPro**  
Classification of protein families

### How big is the known protein space?

- 248.8M entries in UniProtKB (2024)
  - 248.3M unreviewed TrEMBL
  - 0.5M reviewed as Swiss-Prot
- 22K entries in Pfam (2024)
- 19K entries in CDD (2024)
- 1K entries in SMART (2024)

## REVIEW

doi:10.1038/nature14539

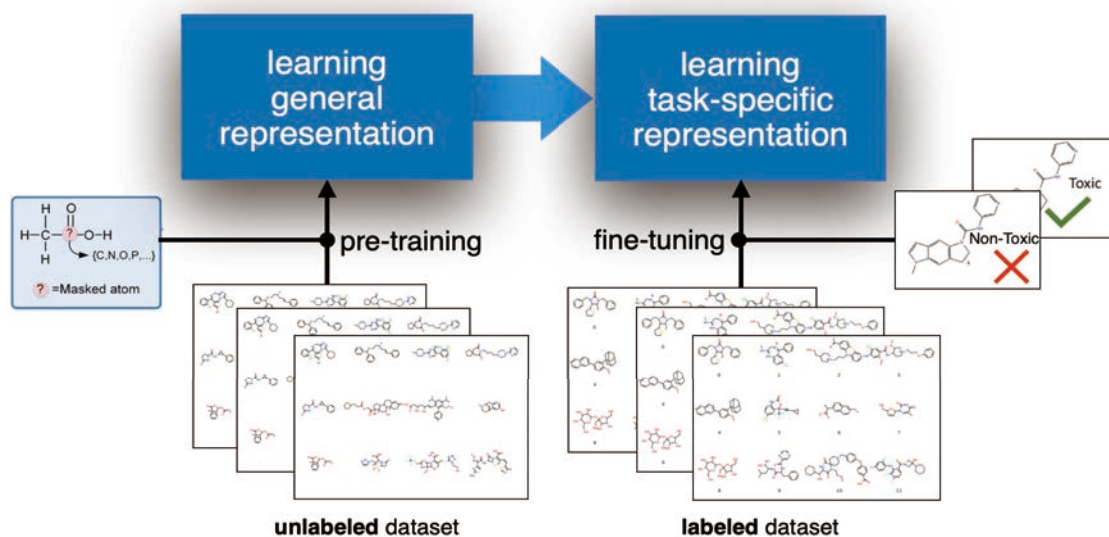
# Deep learning

Yann LeCun<sup>1,2</sup>, Yoshua Bengio<sup>3</sup> & Geoffrey Hinton<sup>4,5</sup>

Deep learning allows computational models that are composed of multiple processing layers to learn representations of data with multiple levels of abstraction. These methods have dramatically improved the state-of-the-art in speech recognition, visual object recognition, object detection and many other domains such as drug discovery and genomics. Deep learning discovers intricate structure in large data sets by using the backpropagation algorithm to indicate how a machine should change its internal parameters that are used to compute the representation in each layer from the representation in the previous layer. Deep convolutional nets have brought about breakthroughs in processing images, video, speech and audio, whereas recurrent nets have shone light on sequential data such as text and speech.

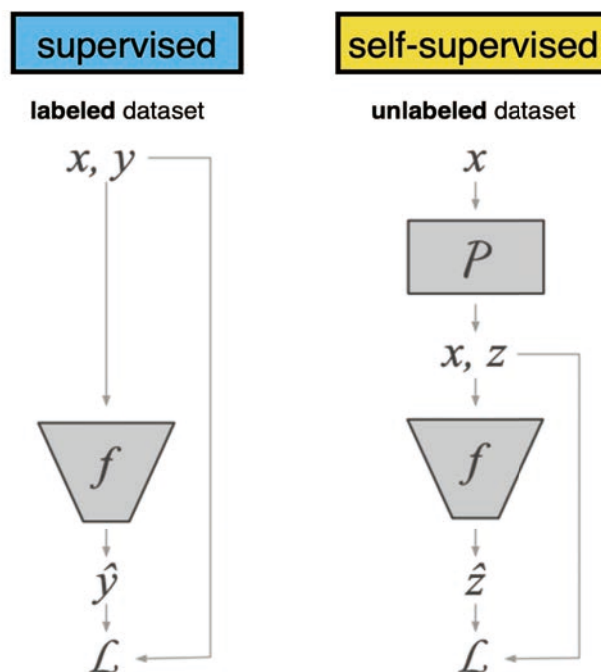
<sup>1</sup>Facebook AI Research, 770 Broadway, New York, New York 10003 USA. <sup>2</sup>New York University, 715 Broadway, New York, New York 10003, USA. <sup>3</sup>Department of Computer Science and Operations Research Université de Montréal, Pavillon André-Aisenstadt, PO Box 6128 Centre-Ville STN Montréal, Québec H3C 3J7, Canada. <sup>4</sup>Google, 1600 Amphitheatre Parkway, Mountain View, California 94043, USA. <sup>5</sup>Department of Computer Science, University of Toronto, 6 King's College Road, Toronto, Ontario M5S 3G4, Canada.

## Learning general and task-specific representations



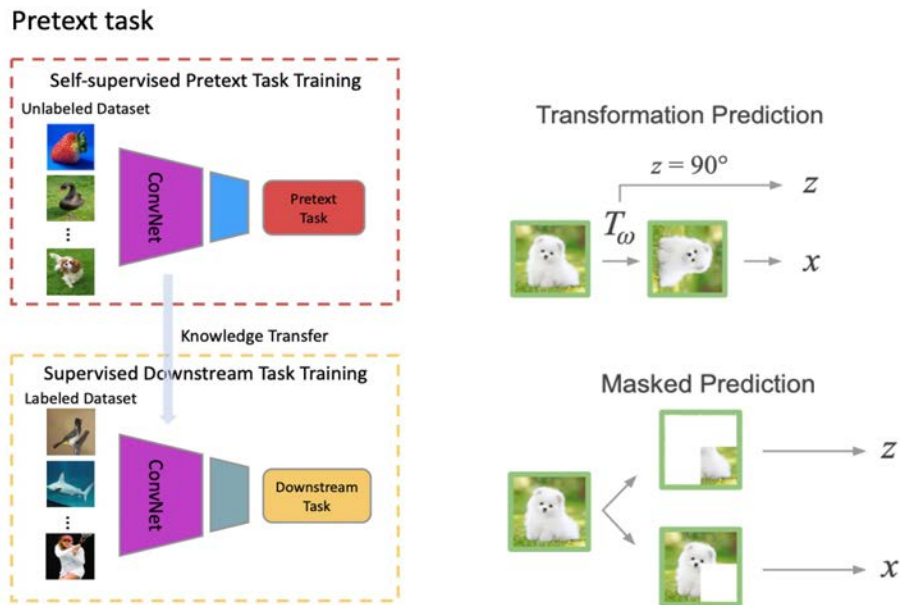
7

## Learning general representations with unlabeled data



8

## Learning general representations with unlabeled data



9

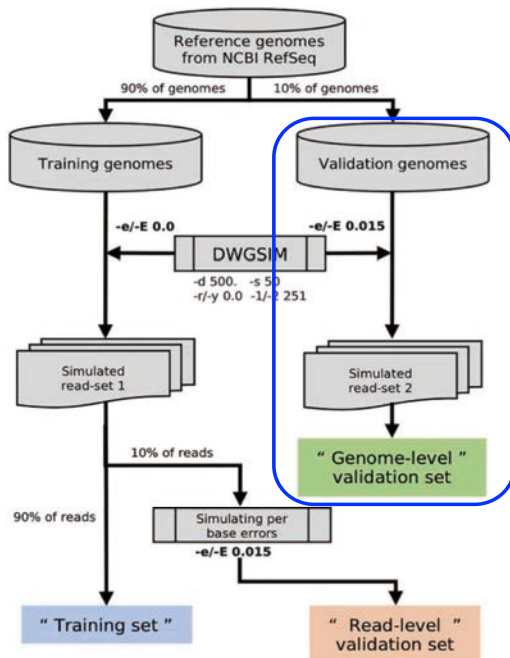
## Taxonomic classification from sequencing read sequence

- Marker-based virus identification is challenging
- Sequence homology-based search against databases of known virus genomes
  - good for known viruses, but low recall rate for unknown viruses
- To increase recall rates, expanding the repertoire of viruses is required
- What if we use metagenome shotgun sequencing data?
  - Simultaneous detection of a broad range of viruses can be possible
- Using sequence fragments of 150 -250 bps, the sequence should be assigned (classified) to one of the taxa at a certain taxonomy level
- Constructing generalized models is another way of increasing recall rates



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## Taxonomy classification from sequencing read sequence



10,119 viral genomes  
2,293 eukaryotic DNA viruses  
2,733 eukaryotic RNA viruses

5,529 bacterial genomes  
(one genome for each species)

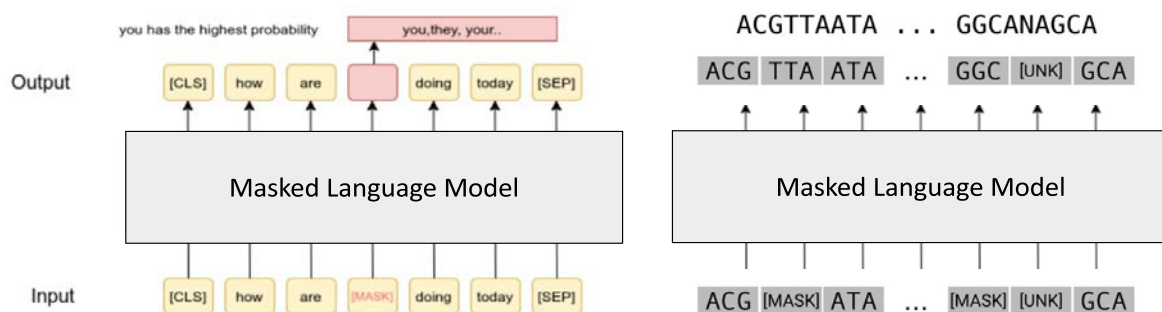
Eukaryote-related viruses were retained  
(Based on ICTV annotation)

The length of reads was set to 151 and 251  
Insert size from 300 to 800

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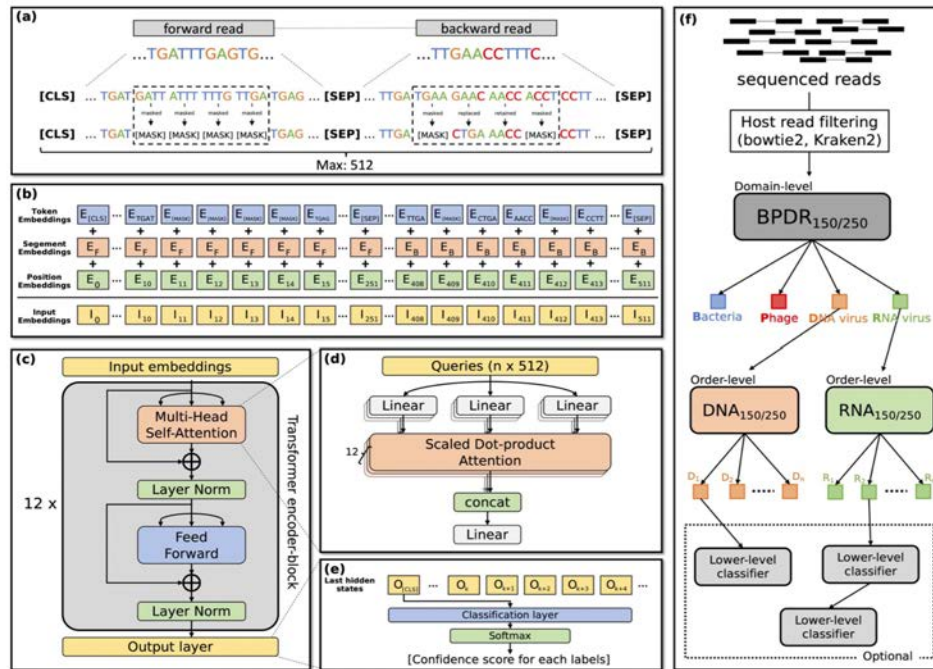
## Using language models for genomic sequence data

- It consists of a series of words  
Genome / Gene / Protein ----- Document / Sentence  
Nucleotides / Amino acids ----- Words
- Local relationship between words  
Motifs ----- Idioms / word order
- Remote relationship between words  
Co-evolution ----- Context





## VIBE: Taxonomy classification from sequencing read sequence



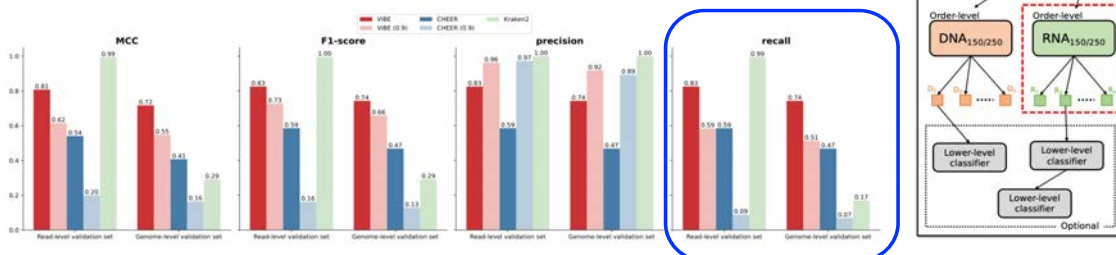
Brief Bioinfo, 2023

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## VIBE: Taxonomy classification from sequencing read sequence

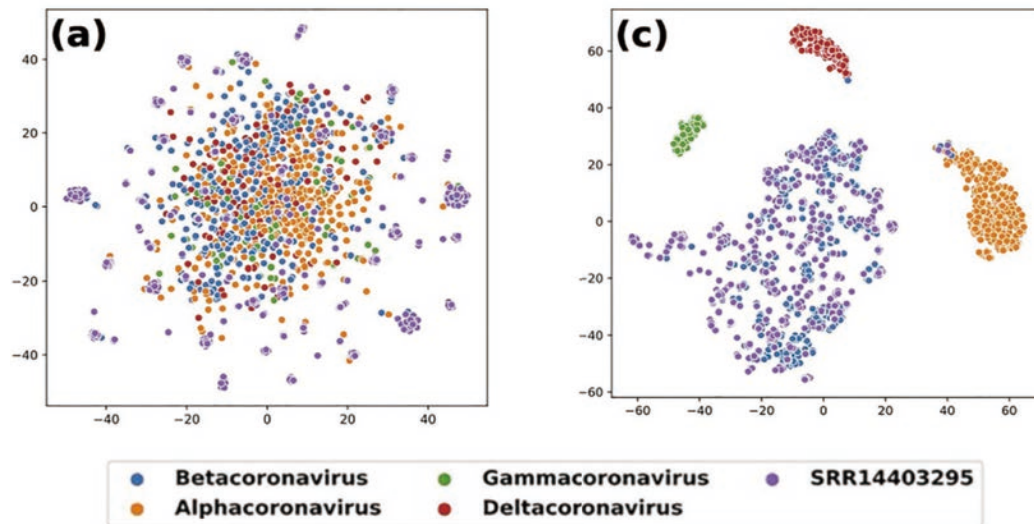
### Order-level classifier for RNA viruses

- SOTA methods:
  - CHEER / Skip-Gram + parallel CNN model
  - Kraken2 / k-mer homology method
- Test data:
  - Read-level validation set
  - Genome-level validation set



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## VIBE: Taxonomy classification from sequencing read sequence



- The ViBE was re-trained without the SARS-CoV-2 reference genome
- COVID-19 samples were tested with the model
- (A) 4-mer frequency, (C) embedded vector by fine-tuned model

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## VIBE: Taxonomy classification from sequencing read sequence

- Number of reads classified as virus from vaginal virome samples

Sample	colpitis01	colpitis02	colpitis03	colpitis04	normal01	normal02
No. of reads	233 241	83 093	807 297	162 205	240 385	944 500
DNA viruses	20 534	9667	68 926	12 418	14 057	36 388
Herpesvirales (o)	7223	2308	5848	3046	2319	3080
Herpesviridae (f)	6905	2075	4794	2893	2177	2725
Zurhausenvirales (o)	152	1203	24 020	618	1188	7637
Papillomaviridae (f)						
Alphapapillomavirus (g)	94	1013	22 576	496	1061	6738
Others (o)	1807	787	9983	1805	2120	9224
Unclassified (o)	11 352	5369	29 075	6949	8430	16 447

- 3.85–11.63% of reads and 1.72–4.8% of reads were DNA viruses and RNA viruses
- 0.46–32.75% of the virus reads were correctly predicted as *Alphapapillomavirus*, a genus of HPV
- [Herpesvirales](#), one of the prevalent pathogens infecting the vagina, were found
- We assembled a set of reads classified as Herpesvirales by ViBE and performed a BLASTX search against the ViPR database
- Among the 68 contigs (>500 bp), 30 contigs found significant hits for human herpesvirus (HHV) from the ViPR database

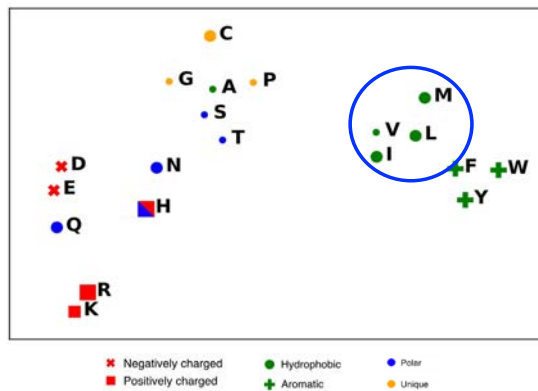
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## Amino acid embedding

Protein function is encoded in the amino acid sequence

Sequences can diverge during evolution while maintaining the same function



	C	S	T	A	G	P	D	E	Q	N	H	R	K	M	I	L	V	W	Y	F	
C	9																				C
S	-1	4																			S
T	-1	1	5																		T
A	0	1	0	4																	A
G	-3	0	-2	0	6																G
P	-3	-1	-1	-1	-2	7															P
D	-3	0	-1	-2	-1	-1	6														D
E	-4	0	-1	-1	-2	-1	2	5													E
Q	-3	0	-1	-1	-2	-1	0	2	5												Q
N	-3	1	0	-2	0	-2	1	0	0	6											N
H	-3	-1	-2	-2	-2	-2	-1	0	0	1	8										H
R	-3	-1	-1	-1	-2	-2	-2	0	1	0	0	5									R
K	-3	0	-1	-1	-2	-1	-1	1	1	0	-1	2	5								K
M	-1	-1	-1	-1	-3	-2	-3	-2	0	-2	-2	-1	-1	5							M
I	-1	-2	-1	-1	-4	-3	-3	-3	-3	-3	-3	-3	-3	1	4						I
L	-1	-2	-1	-1	-4	-3	-4	-3	-2	-3	-3	-2	-2	2	2	4					L
V	-1	-2	0	0	-3	-2	-3	-2	-2	-3	-3	-3	-2	1	3	1	4				V
W	-2	-3	-2	-3	-2	-4	-4	-3	-2	-4	-2	-3	-3	-1	-3	-2	-3	11			W
Y	-2	-2	-2	-2	-3	-3	-3	-2	-1	-2	2	-2	-2	-1	-1	-1	-1	2	7		Y
F	-2	-2	-2	-2	-3	-4	-3	-3	-3	-3	-1	-3	-3	0	0	0	-1	1	3	6	F
	C	S	T	A	G	P	D	E	Q	N	H	R	K	M	I	L	V	W	Y	F	

Scoring matrix BLOSUM

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## Amino acid embedding increases performance

Model	Architecture	Training Dataset	Embedding Quality	Key Application
ProtBERT	BERT	UniRef100	High-quality, bidirectional	Function prediction, localization
ESM	Transformer	UniRef50, larger for ESM-2	Captures evolutionary info	Mutational effect, structure prediction
ProtT5	T5	BFD100, UniRef50	Handles long-range dependencies	Protein engineering mutation effect
ProtTrans	BERT, T5, XLNet	UniRef50, BFD100	Versatile, multi-architecture	Structure prediction, PPI prediction
AlphaFold-inspired Transformers	Specialized transformer	Structure-focused datasets	3D-aware embeddings	Structure prediction
ProtXLNet	XLNet	UniRef50	Captures contextual variability	Functional site prediction

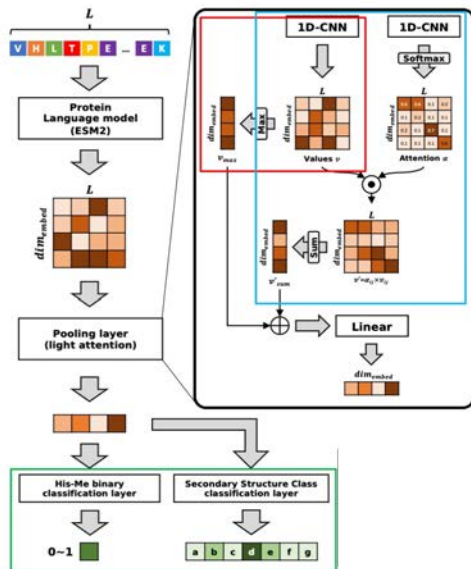
  

ProtT5	6.6±1.52	7.4±3.8	2.7±0.69	4.6±3.3	3.2±1.08	3.8±2.74	0.4±0.45	0.6±0.12
ESM2 8M*	4.9±0.5	15.8±3.11	6.5±1.36	1.5±3.98	2.0±0.86	3.9±2.11	1.8±0.79	0.8±0.18
ESM2 35M*	3.9±0.4	21.8±6.41	5.2±0.73	7.8±1.94	1.2±2.97	3.2±2.45	4.7±1.34	0.9±0.1
ESM2 150M*	5.2±0.33	18.9±2	4.6±1.14	-5.1±3.11	0.9±1.94	2.2±2.08	3.0±1.58	0.6±0.16
ESM2 650M*	4.0±0.51	31.2±5.04	2.2±1.31	7.9±7.56	0.8±2.03	0.9±2.57	0.0±0.78	0.8±0.08
ESM2 3B	4.9±0.22	8.1±1.21	2.7±0.99	5.2±1.12	2.2±1.55	3.6±1.4	0.6±0.97	0.8±0.1
Ankh base	3.2±0.35	17.6±5.22	1.8±1.62	5.3±6.17	3.5±1.95	2.2±1.31	-2.6±0.75	-0.4±0.08
Ankh large	2.5±0.49	11.7±2.84	3.4±1.68	11.3±6.64	-2.1±4.8	2.2±2.92	-1.1±0.8	-0.3±0.17
	GFP	AAV	GB1	Stability	Meltome	Subcellular location	Disorder	Secondary structure
	mutational landscape			diverse dataset				

Blue tiles mark statistically significant increase  
Yellow mark statistically insignificant change



## FuncPred: Function prediction from protein sequences



Capturing **conserved residues** and **secondary structure** is essential to classify His-Me finger nuclease

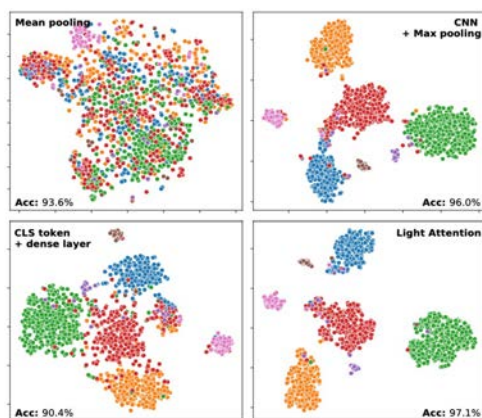
Pooling: light attention

- **1D-CNN + max pooling:** capturing secondary structure
- **Convolution x attention:** capturing conserved residues

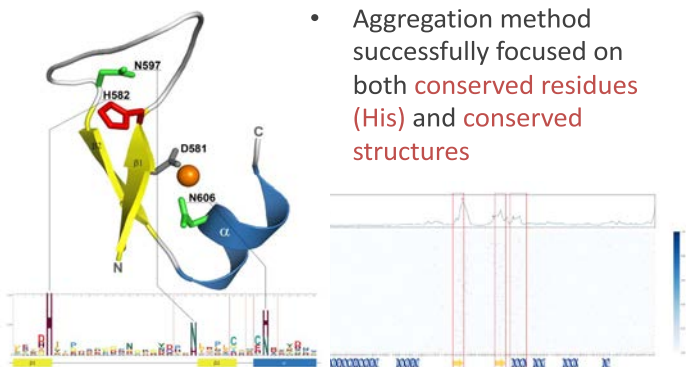
Using sequence-level representation, **binary classification** for detecting His-Me finger nuclease and **secondary structure class classification** were performed simultaneously.

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## FuncPred: Function prediction from protein sequences



- a: All alpha
- b: All beta
- c: A|B
- d: A+B
- e: Multi-domain
- f: Membrane and cell surface
- g: Small proteins



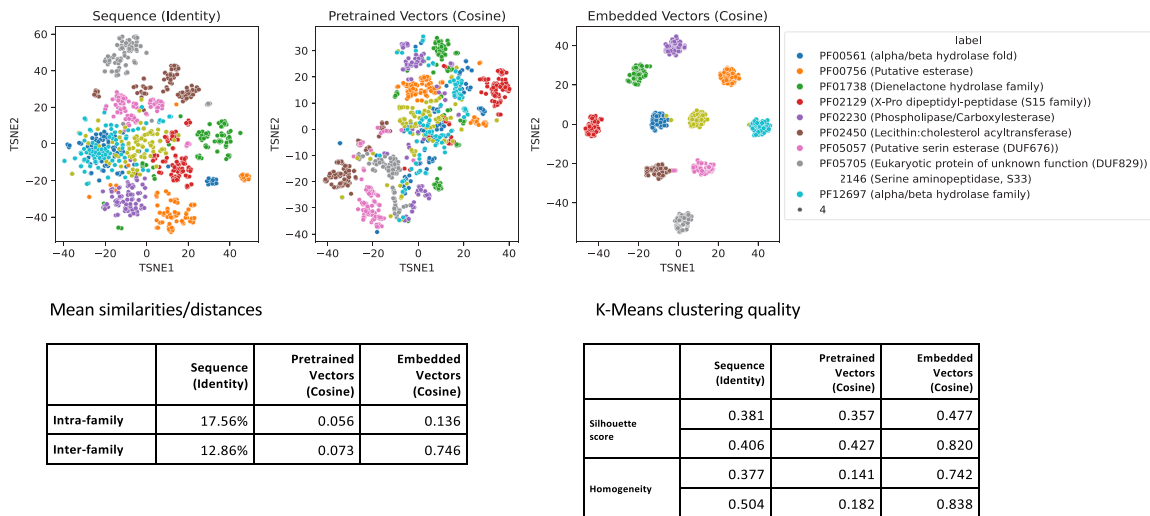
- Aggregation method successfully focused on both **conserved residues** (His) and **conserved structures**

query ID	subject ID	percent identity	coverage	query alignment length	query length	query start	query end	subject start	subject end	e-value
d1zm8a	NUCA_NOSS1/43-263	99.55	92.5%	221	239	8	228	1	221	6.42E-164
d4e3ya	A0A240BSN3_SERP/24-245	94.12	92.1%	221	240	1	221	2	222	2.27E-161
d1u3em1	B6V2J8_BPSP1/53-98	100.00	43.8%	46	105	53	98	1	46	1.59E-29
d1a73a	A0A1E3PUR8_LIPST/1-90	46.15	24.1%	39	162	88	126	16	54	4.E-06
d1e7la2	A0A2H4YFJ9_9CAUD/1-97	90.72	94.2%	97	103	1	97	1	97	1.15E-63
d2pu3a	Q5E7R4_ALIF1/26-232	91.30	100.0%	207	207	1	207	1	207	3.01E-146
d1v0da	A0A0H2UHU4_RAT/123-344	96.85	90.6%	222	245	19	240	1	222	7.69E-164
d4ogca2	F9PLJ4_9ACTO/560-606	97.87	29.2%	47	161	54	100	1	47	1.01E-29
d5axwa2	K9B5K9_9STAP/65-120	82.14	38.1%	56	147	51	106	1	56	4.05E-29
d4oo8a2	CAS9_STRP1/821-872	98.08	39.1%	52	133	47	98	1	52	1.52E-30
d6w0va	A0A1I6EC40_9FIRM/69-117	38.71	24.8%	31	125	94	124	15	45	2.E-04
d3qevd	A0A671YF54_SPAAU/33-134	98.02	80.8%	101	125	25	125	1	101	5.23E-73

Remote homologous proteins

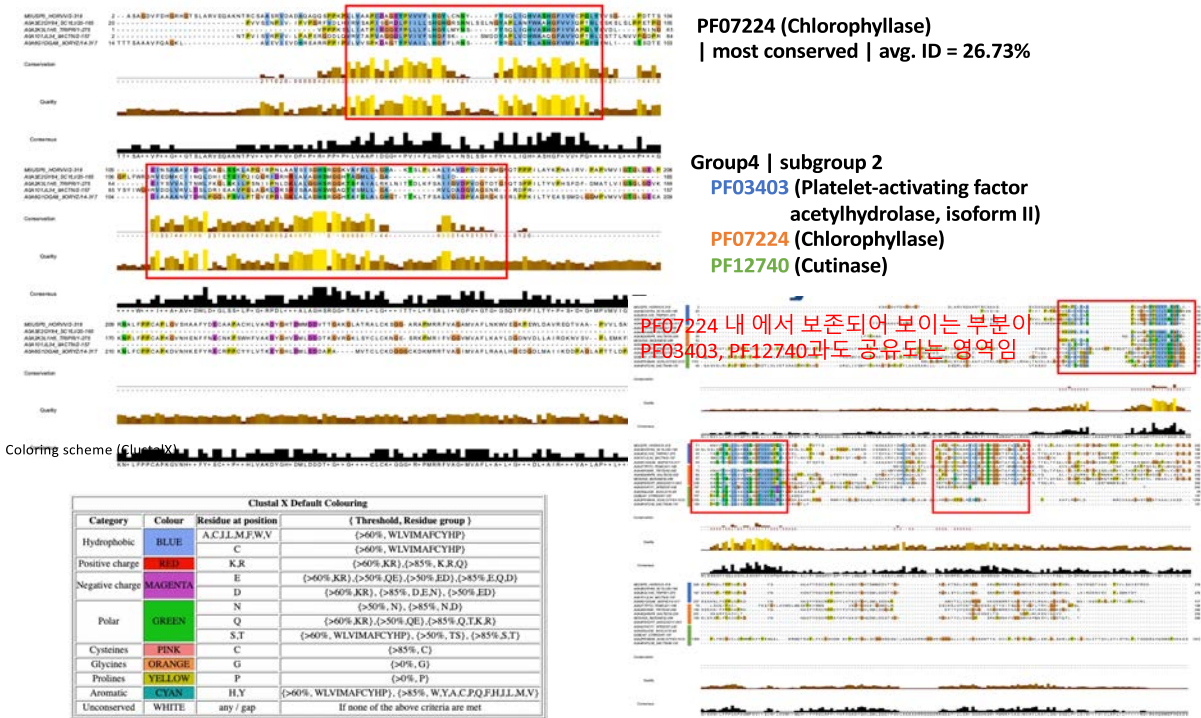
Highly homologous proteins, which was not reported as His-Me finger nuclease

Evaluation with the 10 families of lowest average intra-sequence similarity



23

Evaluation with the 10 families of lowest average intra-sequence similarity





### Take-home messages

- Deep learning models discover **multiple representations** for **genomic sequences and chemical compounds**
- **Generalization** and **specialization** can be achieved by **self-supervised learning** in the pre-training process and **task-specific learning** in the fine-tuning process
- Properties for **both local and global relationship** should be focused to understand more useful meaning of data



# 03

## Development of human virome analysis research methodology



이하나  
(고려대학교)





## 3부 연자



- ✓ 소속: 고려대학교
- ✓ 직위/직책: 교수
- ✓ 성명: 이하나

### 🔍 학력사항

2000	서울대학교 생물교육과 이학사
2007	서울대학교 생명과학부 이학박사

### 🔍 경력사항

2007 ~ 2009	University of Massachusetts, Amherst
2010 ~ 2012	서울대학교 유전공학연구소 연구조교수
2012 ~ 현재	고려대학교 보건과학대학 교수

## 인체 바이롬 분석 연구방법론 개발

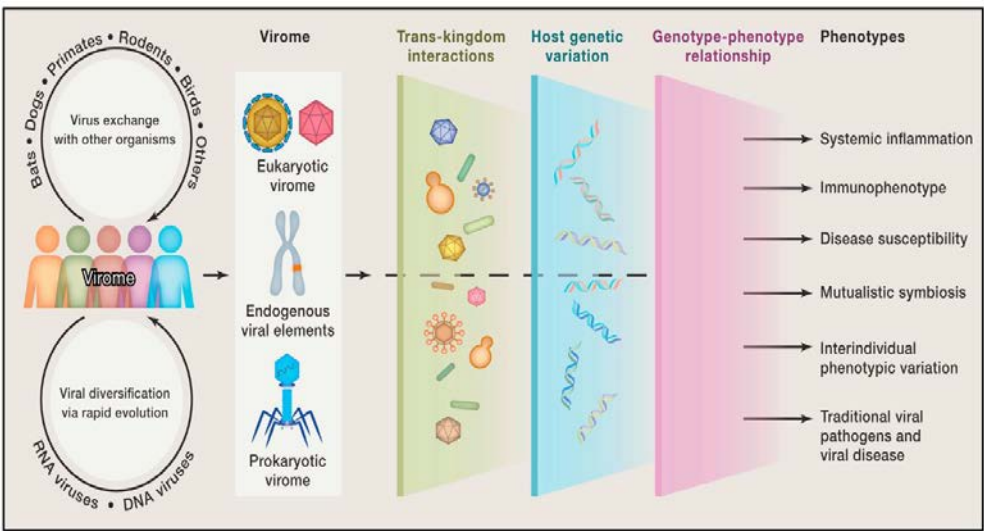
바이롬은 인체 마이크로바이옴의 일부로 박테리아 또는 인체와 상호작용하며 건강과 질병 상태에 중요한 역할을 한다. 그동안 박테리옴을 중심으로 마이크로바이옴 연구가 진행되어 의미 있는 결과들을 도출하고 상용화에 도달하고 있는 상황이지만, 아직까지 바이롬 연구는 상대적으로 미비한 상황이다. 아직 연구 초기 단계라고 할 수 있는 바이롬 연구를 성공적으로 수행하기 위해서는 실험 및 분석 방법론에 대한 고민이 선행되어야 한다. 성공적인 바이롬 연구를 위해 인지해야 할 연구방법론적 주의사항을 고찰하고 이를 바탕으로 표준화되고 효율적인 실험 및 분석 방법론을 개발할 필요가 있다.



# Development of methodologies for human virome study

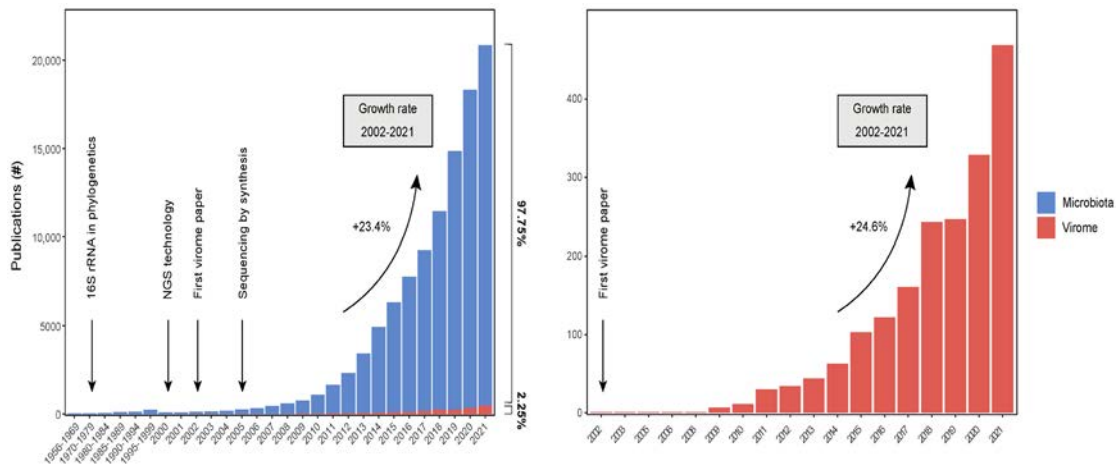
Hana Yi  
Korea University

## Virome



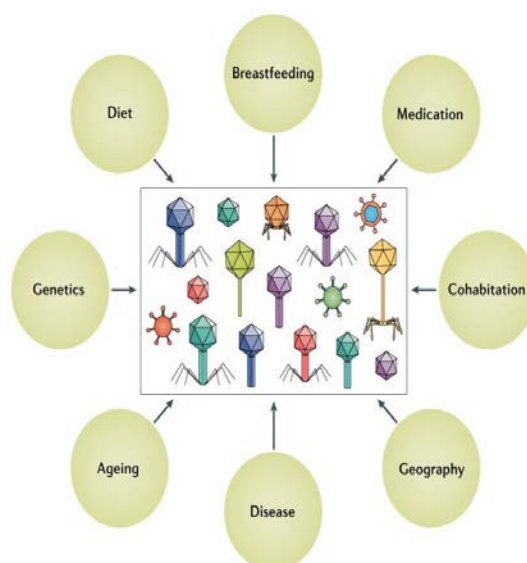
Virgin HW. The virome in mammalian physiology and disease. Cell. 2014;157:142-50.

## Publications



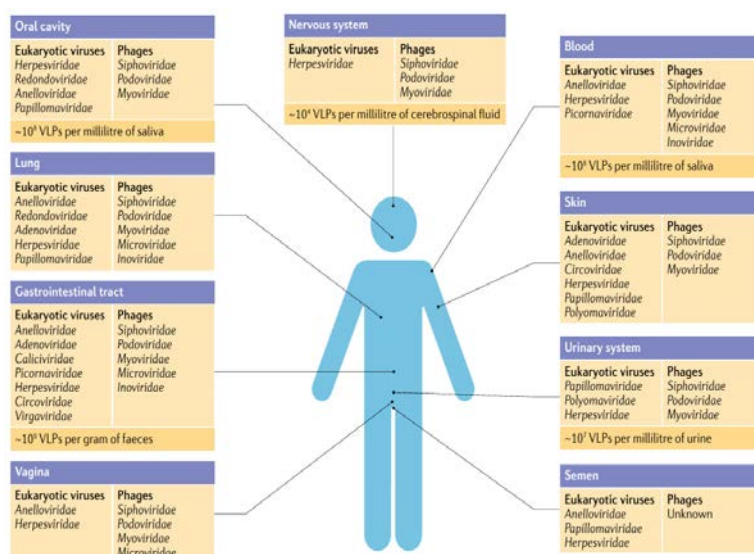
Jansen D. et al. The emerging role of the gut virome in health and inflammatory bowel disease: challenges, covariates and a viral imbalance. *Viruses*. 2023;173.

## Factors that shape the virome



Liang G, et al. The human virome: assembly, composition and host interactions. *Nat Rev Microbiol* 2021;19:514-27.

## Human virome at different body sites



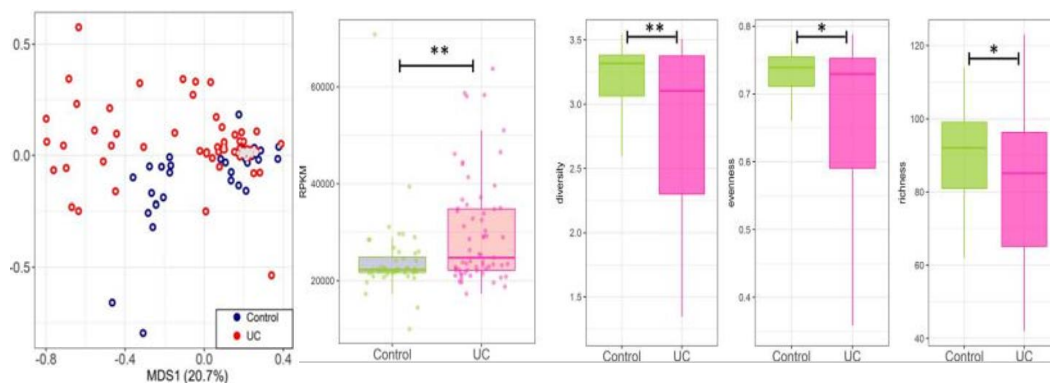
Liang G, et al. The human virome: assembly, composition and host interactions. Nat Rev Microbiol 2021;19:514-27.

## Virome alterations in human disorders

Human disease	Sample	Major virome alteration	Human disease	Sample	Major virome alteration
Severe acute malnutrition	Faeces	Reduced viral diversity	Bacterial vaginosis	Vaginal swabs	Viral population structures correlated with bacterial vaginosis
Crohn's disease and ulcerative colitis	Faeces	Increased <i>Caudovirales</i> richness	Early-diagnosed Crohn's disease and ulcerative colitis	Gut biopsies	Increased <i>Hepadnaviridae</i> and <i>Hepeviridae</i> ; reduced <i>Polydnaviridae</i> , <i>Tymoviridae</i> and <i>Virgaviridae</i>
Crohn's disease	Faeces and biopsies	Moderate alterations	Coeliac disease autoimmunity	Faeces	Increased enteroviruses
AIDS	Faeces	Increased enteric adenoviruses	Crohn's disease	Faeces	The virulent phages are replaced with temperate phages
Type 1 diabetes	Faeces	Reduced viral diversity	Ulcerative colitis	Gut biopsies	Increased <i>Caudovirales</i> , phage and bacteria virulence functions, and loss of viral-bacterial correlations
Hypertension	Faeces	<i>Erwinia</i> phage ΦEaH2 and <i>Lactococcus</i> phage 1706 may be associated with hypertension	HIV viraemia	Seminal fluid	Increased human cytomegalovirus
Type 2 diabetes	Faeces	Increased putative phage scaffolds	Very early-onset inflammatory bowel disease	Faeces	Increased ratio of <i>Caudovirales</i> to <i>Microviridae</i>
DOCK8 deficiency	Skin swabs	Increased skin virome, especially human papillomavirus	Haematopoietic stem cell transplantation	Faeces	Increased picobirnaviruses
Colorectal cancer	Faeces	Increased viral diversity			
Crohn's disease and ulcerative colitis	Faeces	Increased <i>Caudovirales</i> abundance			
Type 1 diabetes during pregnancy	Faeces	Increased picobirnaviruses and tobamoviruses			

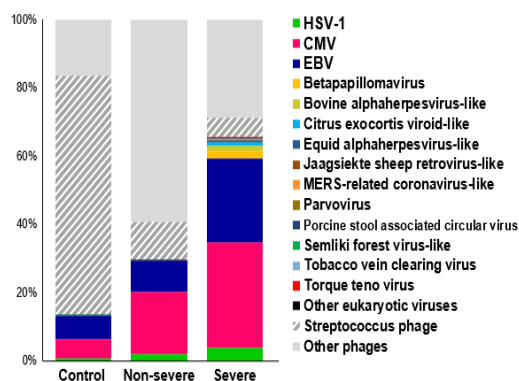
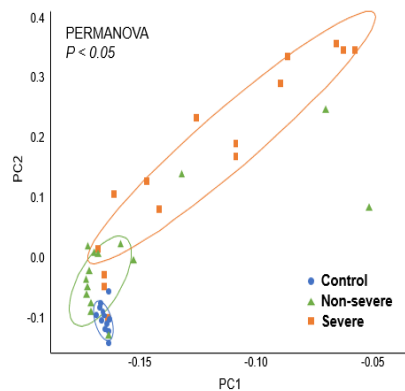
Liang G, et al. The human virome: assembly, composition and host interactions. Nat Rev Microbiol 2021;19:514-27.

## Gut mucosal virome alterations in UC

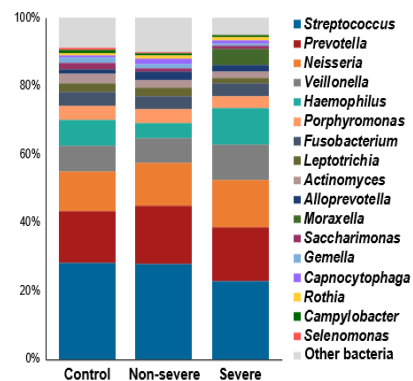
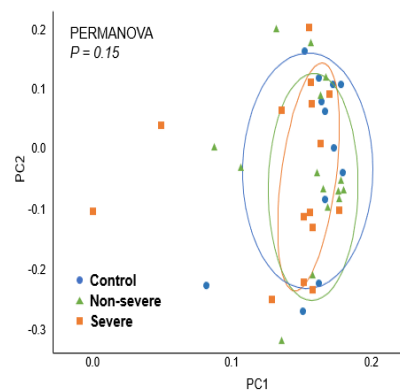


Zuo T. et al. Gut mucosal virome alterations in ulcerative colitis. Gut 2019;68:e1.

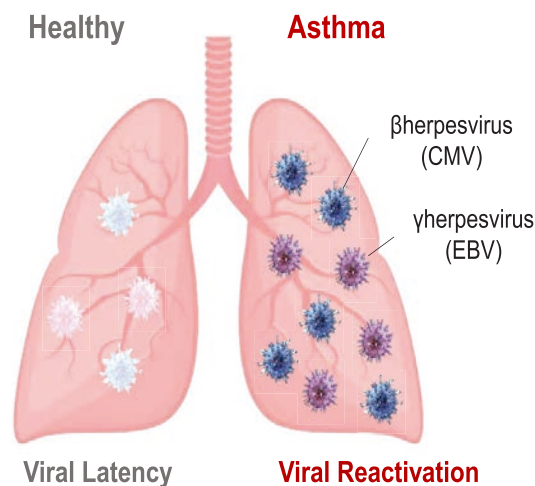
### Virome



### Bacteriome

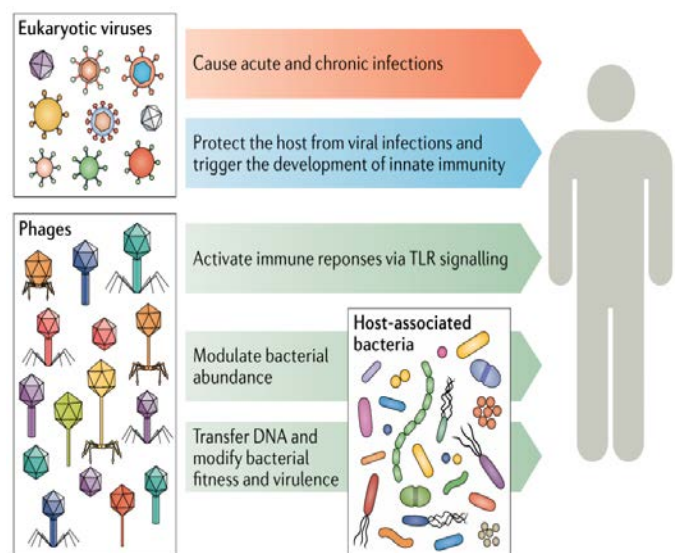


# Virome alterations in asthma



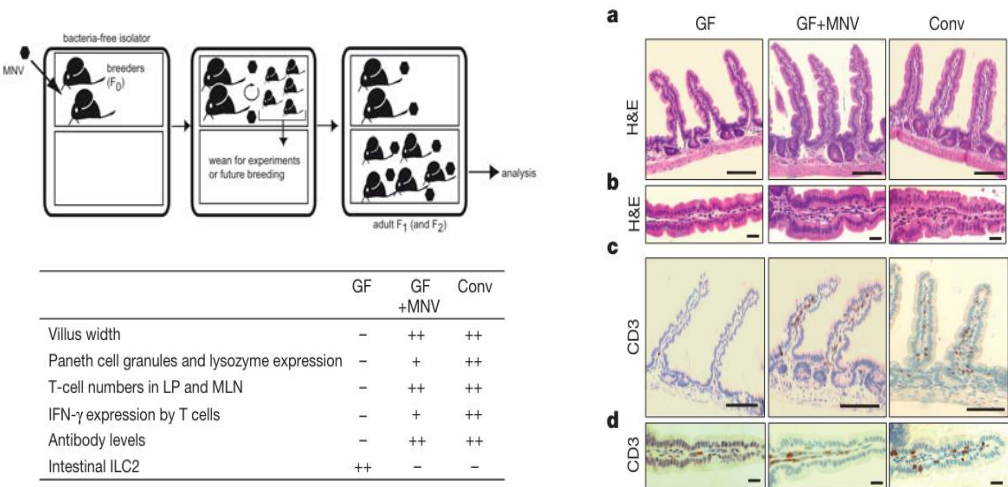
S Choi et al. Lung virome: New potential biomarkers for asthma severity and exacerbation. J Allergy Clin Immunol. 2021;148:1007-1015.

# Host-virome interactions



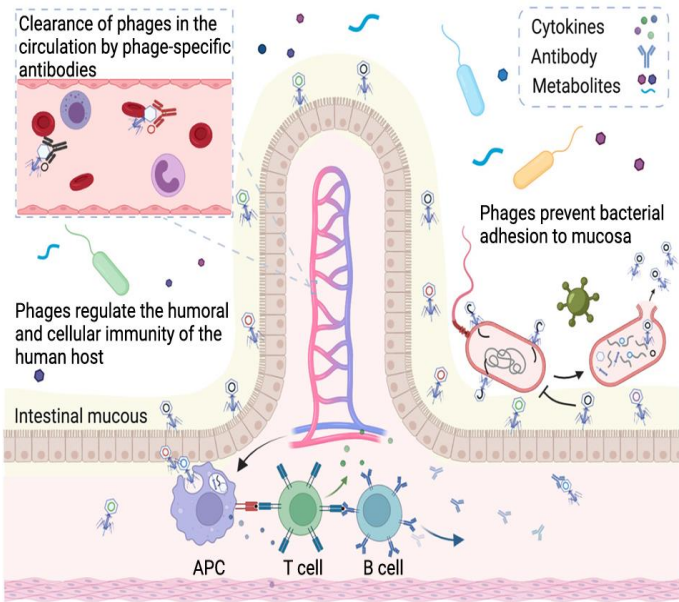
Liang G, et al. The human virome: assembly, composition and host interactions. Nat Rev Microbiol 2021;19:514-27.

## Murine noroviruses in mucosal immunity



Kernbauer E, et al. An enteric virus can replace the beneficial function of commensal bacteria. *Nature*. 2014;516:94-8.

## Interactions between phages and immune system



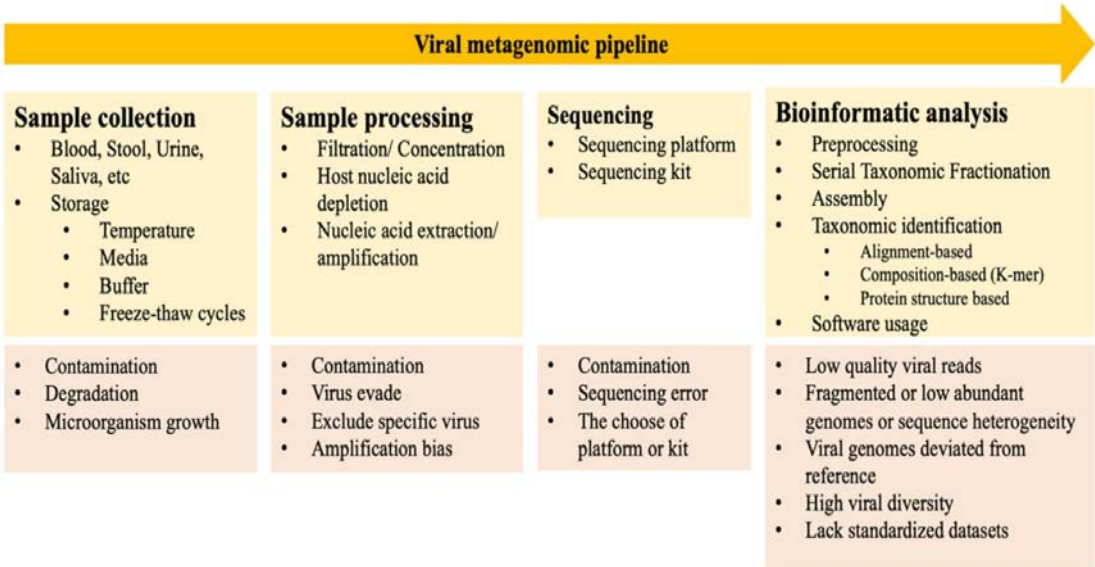
Cao Z. et al. The gut virome: a new microbiome component in health and disease. *eBioMedicine*, 2022;81:104113.



# Challenges in virome study

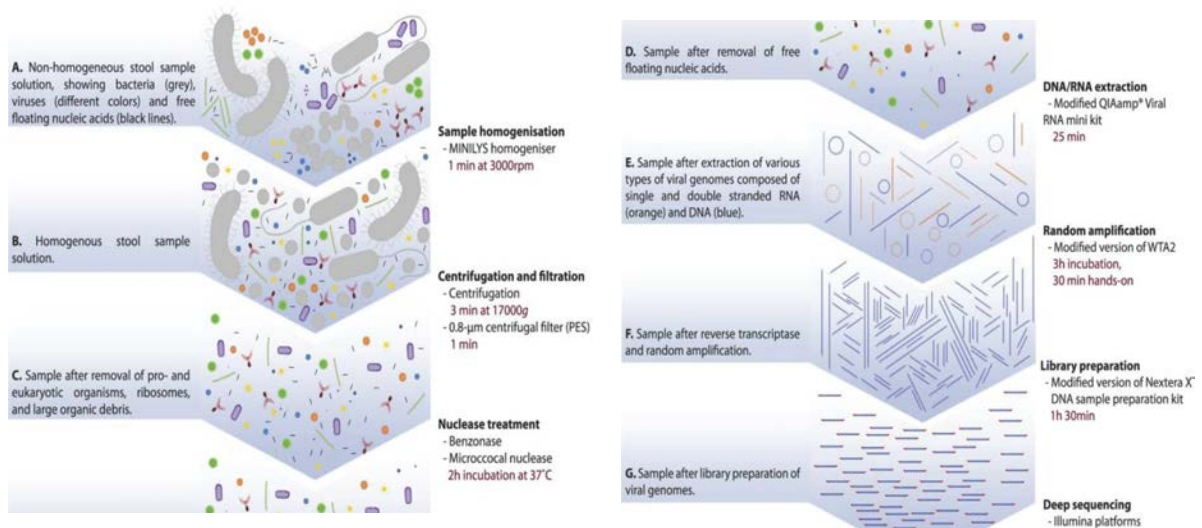
- Low biomass
- RNA/DNA genomes
- Absence of a universal viral gene
- Lack of standardized computational pipeline
- Lack of standardized viral metagenome database

# Drawbacks and potential biases



Bai G-H et al. The human virome: viral metagenomics, relations with human diseases, and therapeutic applications. Viruses. 2022;14:278.

## NetoVIR (Novel enrichment technique of VIRomes)



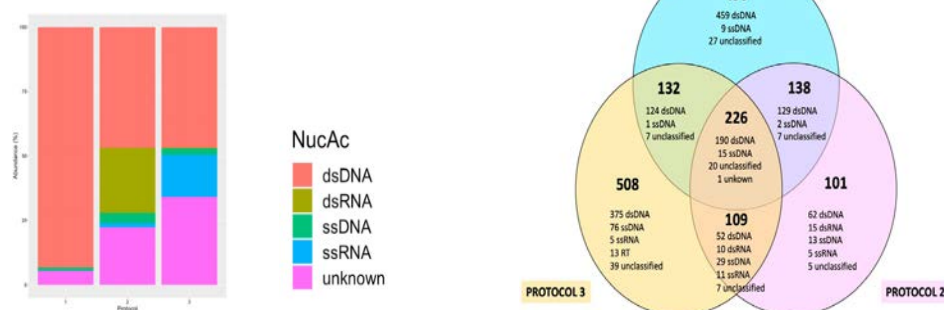
Conceição-Neto, N et al. Modular approach to customise sample preparation procedures for viral metagenomics: a reproducible protocol for virome analysis. Sci Rep 2015;5:16532.

## Experimental biases

Protocol 1: Bulk Metagenomics Strategy

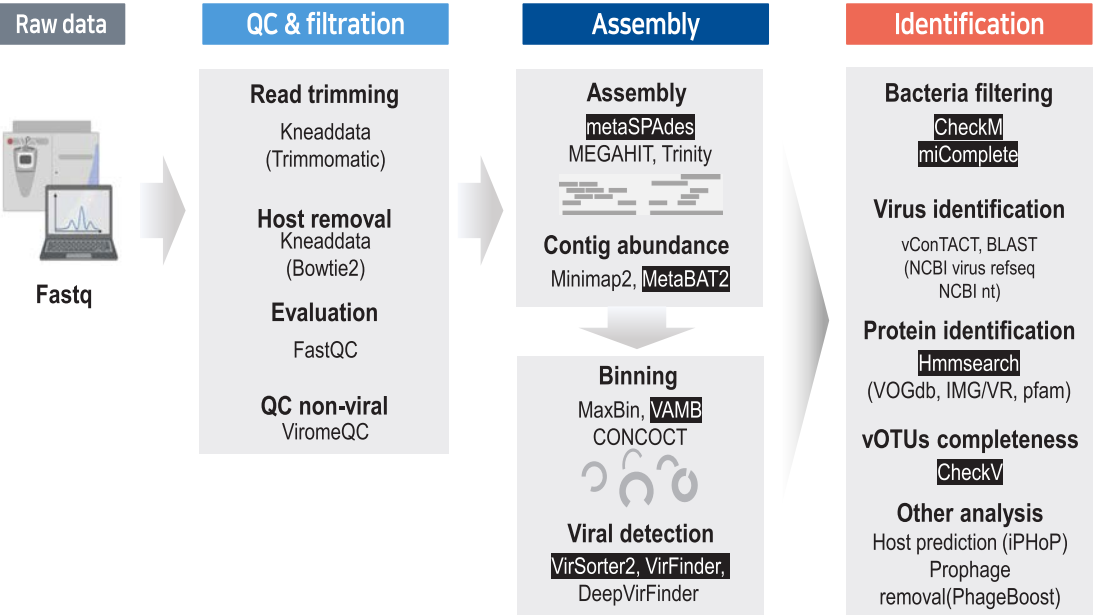
Protocol 2: VLP-Enrichment Strategy A (Modified NetoVIR)

Protocol 3: VLP-Enrichment Strategy B (Modified SISPA)



Soria-Villaba A. et al. Comparison of experimental methodologies bases on bulk-metagenome and virus-like particle enrichment. Microorganisms. 2024;12:162.

# Bioinformatic pipeline for virome



# Database issues

GOLDEN STANDARD SEQUENCE		
correct viral sequence	5' UTR [blue bar] 3' UTR	
TYPES OF ERROR		Examples (NCBI Acc. and species name)
empty sequence	N	1PFI_C Primolivicivirus Pf1
incomplete sequence	5' UTR [blue bar]	EU255989 Hepacivirus hominis
chimeric sequence	5' UTR [blue bar] 3' UTR [blue bar] FALSE	KY766069 Zika virus
misannotaed sequence	[orange bar] FALSE	AJ000888 Hepacivirus hominis
wrong orientation	3' UTR [blue bar] [orange bar] 5' UTR	KU059750 Orthomarburgvirus marburgense

Ritsch M. et al. Navigating the landscape: A comprehensive review of current virus database. Viruses. 2023;15:1834.

## Human Virome Program (HVP), USA

- Characterize the human virome
- Develop tools, models, and methods
- Elucidate the interactions
- Consortium Organization & Data Collaboration Center



<https://commonfund.nih.gov/humanvirome>

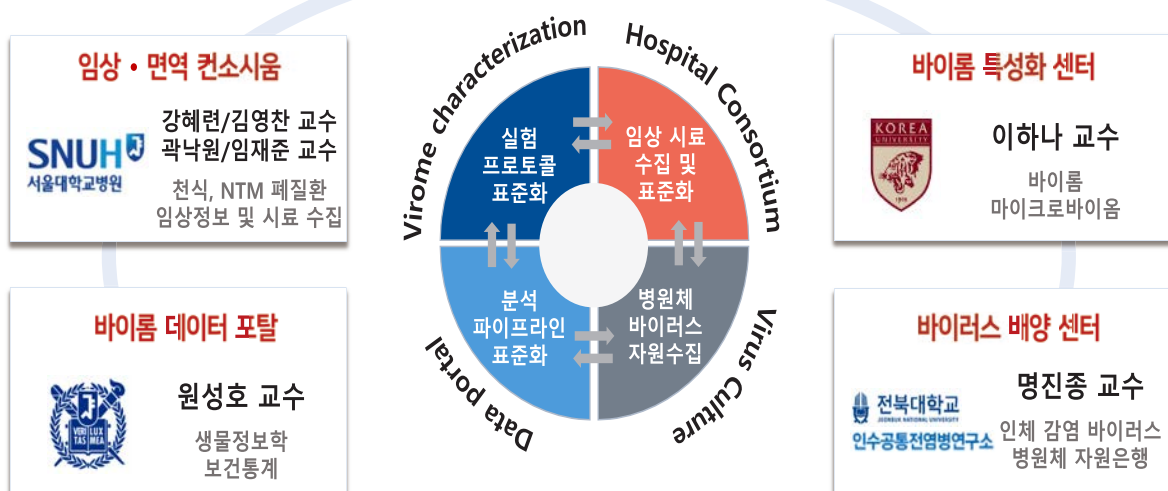
## 한국인 Virome 특성 및 질환 간의 상관관계 분석

최종목표	<b>01</b> 인체 바이롬 분석체계 구축 및 병원체 자원화 <b>02</b> 인체 바이롬 분포현황 파악 및 질환 간의 연관성 분석
연차별 세부 목표	<b>1차년도</b> 바이롬 분석 체계 구축 및 표준화된 바이롬 데이터 생산을 위한 가이드라인 제시 <b>2차년도</b> 가이드라인에 따라 확보된 검체를 대상으로 바이롬 분석 및 자원 확보 국내 건강인 대상 연령별 바이롬 분포 현황 확인 및 바이롬 자원 특성 분석 국내 질환군과 대조군(건강인) 바이롬 분포 비교 분석 인간 숙주와 바이롬 사이의 상호작용 규명 <b>3차년도</b> 질환군에서의 바이롬 및 마이크로바이옴 변화와 비교 분석 바이롬 분포와 질환과의 상관성 확인 질환군 바이롬의 병원성 잠재력 평가

## 인체 바이롬 분석체계 구축 및 병원체 자원화

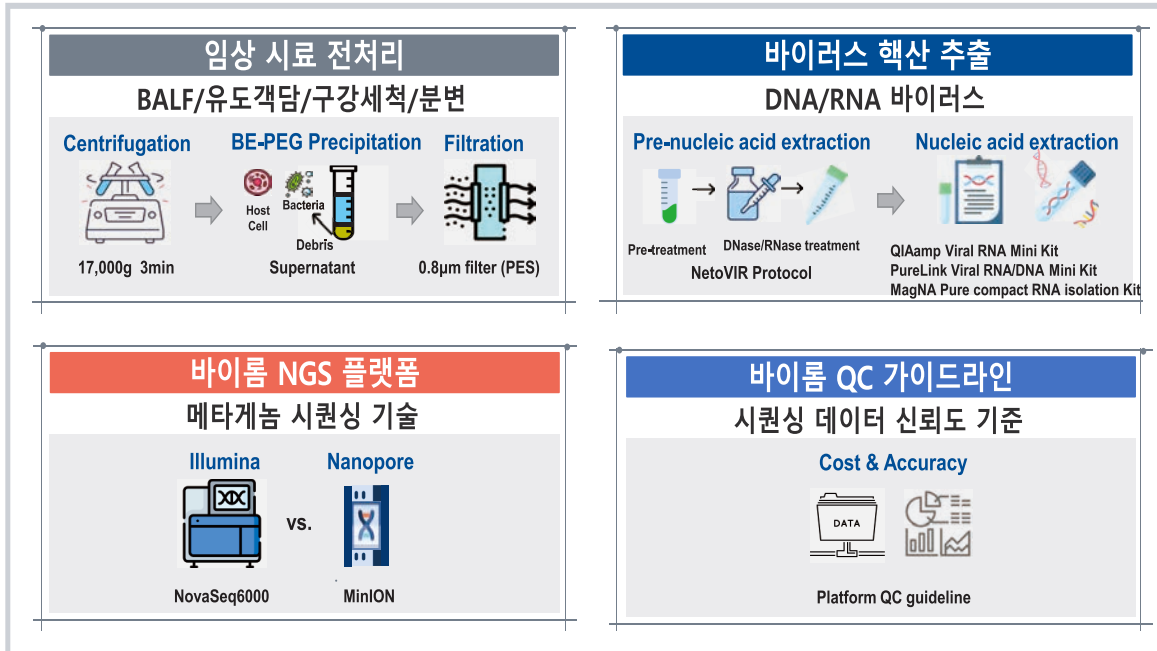


## Human Virome Project Platform



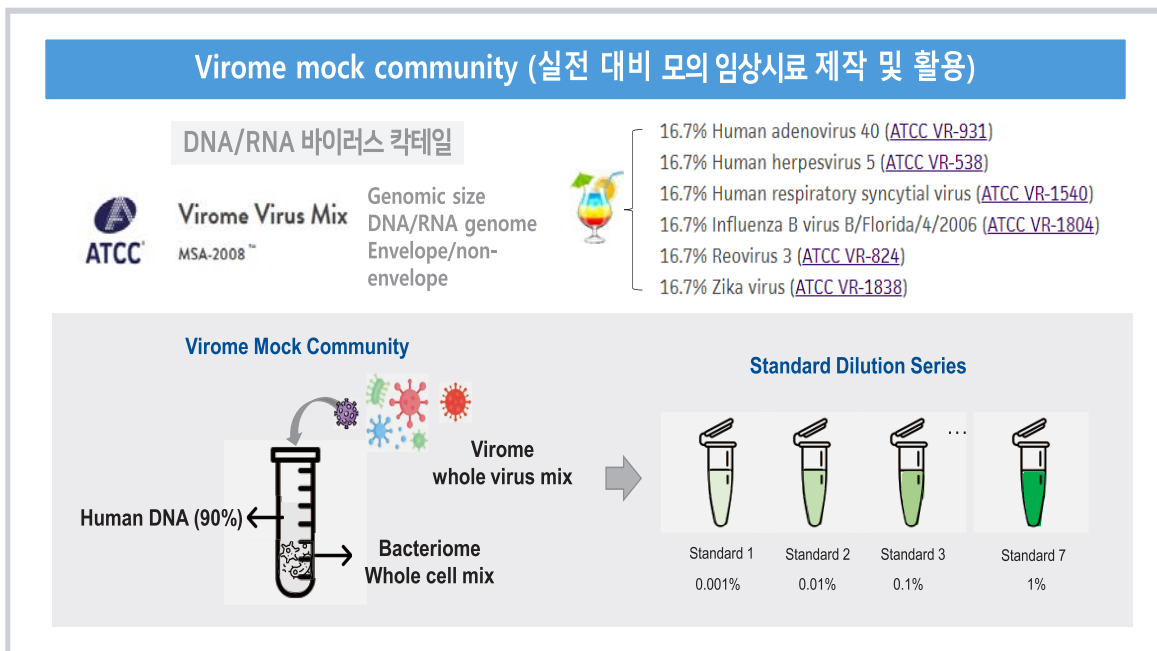
## 01 바이롬 분석 체계 구축 및 표준화된 바이롬 데이터 생산을 위한 가이드라인 제시 - 실험

법



## 01 바이롬 분석 체계 구축 및 표준화된 바이롬 데이터 생산을 위한 가이드라인 제시 - 실험

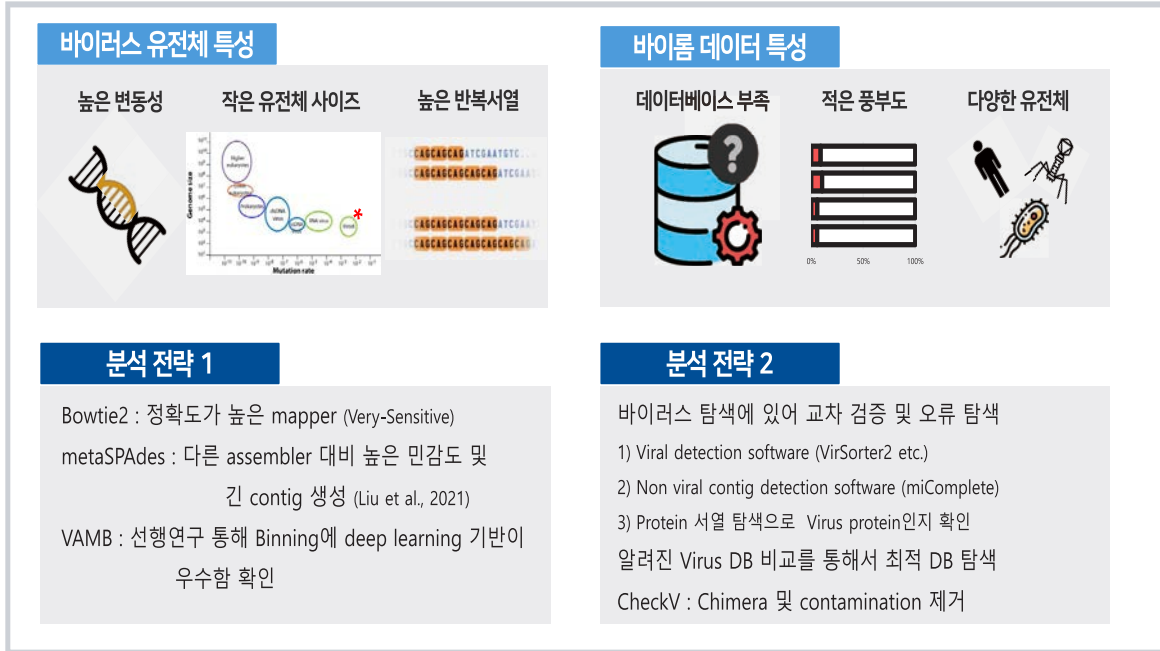
법





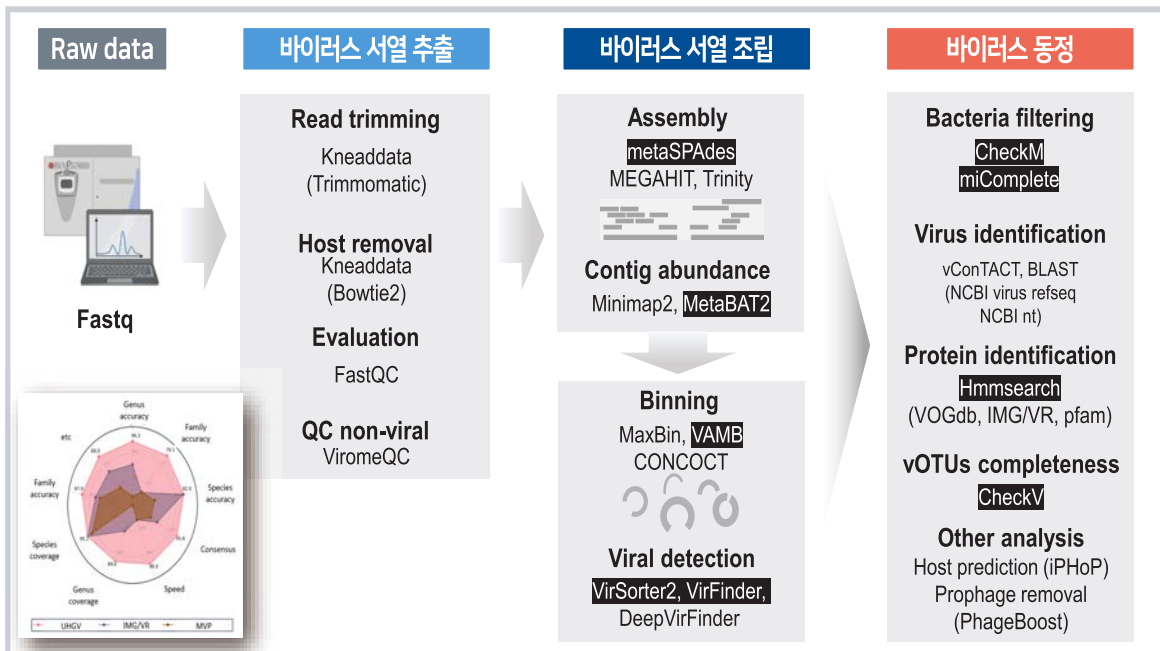
## 01 바이롬 분석 체계 구축 및 표준화된 바이롬 데이터 생산을 위한 가이드라인 제시 - 분석

본



## 01 바이롬 분석 체계 구축 및 표준화된 바이롬 데이터 생산을 위한 가이드라인 제시 - 분석

본



감사합니다

# 04

## Approach of human microbiome data standardization: From sample collection to data analysis



이송희  
(국립보건연구원)





## 3부 연자



- ✓ 소속: 질병청 국립보건연구원 인수공통감염연구과
- ✓ 직위/직책: 박사후연구원
- ✓ 성명: 이송희

### 🔍 학력사항

2019	을지대학교 임상병리학 (학사)
2021	을지대학교 시니어헬스케어학 (석사)
2024	을지대학교 임상병리학 (박사)

### 🔍 경력사항

2024 ~ 현재	질병관리청 국립보건연구원 박사후연구원
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# Approach of human microbiome data standardization: From sample collection to data analysis

국가 차원의 신뢰할 수 있는 마이크로바이옴 데이터 생산, 수집 및 통합 분석을 위해 마이크로바이옴 분석 표준화 필요

표준물질(mock community)를 활용하여 분석 표준지침 마련 연구수행 필요

mock community를 활용한 표준화 확립으로 편향적인 유전체 데이터 생산을 감소시키고 신뢰성 있는 데이터 생산을 증가시키고자 함.



A presentation slide with a dark blue background featuring glowing blue bacteria and a DNA double helix. The title is in English and Korean. The presenter's name and affiliation are listed in the center. The logo of the National Research Institute of Health is at the bottom center.

**Approach of human microbiome data standardization  
: From sample collection to data analysis**  
**인체 마이크로바이옴  
데이터 표준화 전략: 검체 수집부터 데이터 분석까지**

이승희  
인수공통감염연구과

 국립보건연구원  
National Research Institute of Health

1

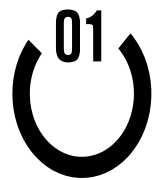
## 목차

### 01 병원기반 인간 마이크로바이옴 사업

- 마이크로바이옴 데이터 표준화 필요성
- 운영 가이드라인 고도화

### 02 미생물 군집 염기서열 분석 표준화 전략 연구

- Mock community 검증
- Wholemata shotgun 데이터의 표준화

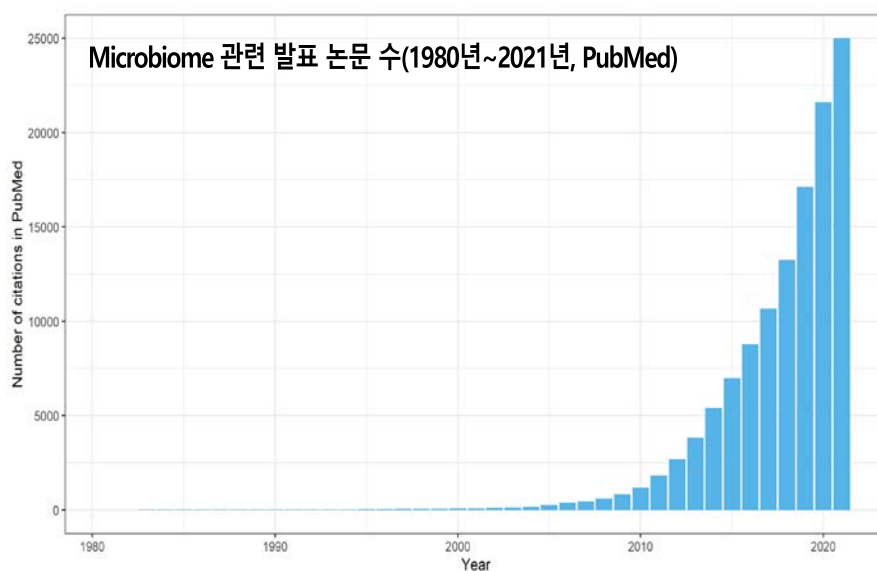


## 병원기반 인간 마이크로바이옴 사업



병원기반 인간 마이크로바이옴 사업

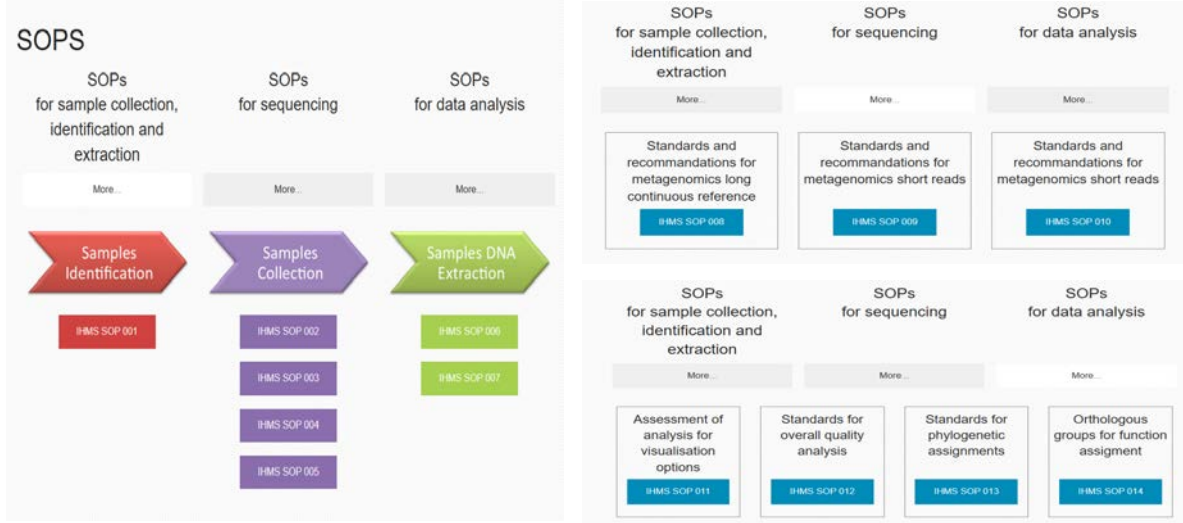
### 인체 마이크로바이옴 연구의 가속화



## 병원기반 인간 마이크로바이옴 사업

# 마이크로바이옴 데이터 표준화 필요성

- 마이크로바이옴 유전체 데이터는 실험 기술적으로 **검체 채취, 수송, 핵산 추출, 시퀀싱, 분석 방법** 등에 영향을 받음
- 많은 연구에서 서로 다른 실험실에서 생산된 데이터가 일관된 결과를 도출하지 않는 경우가 다수 발생
- 세계적으로 이러한 연구 제한점을 인지하고 국가 차원의 **표준화** 하기 위한 **다양한 프로젝트와 컨소시엄**을 구성

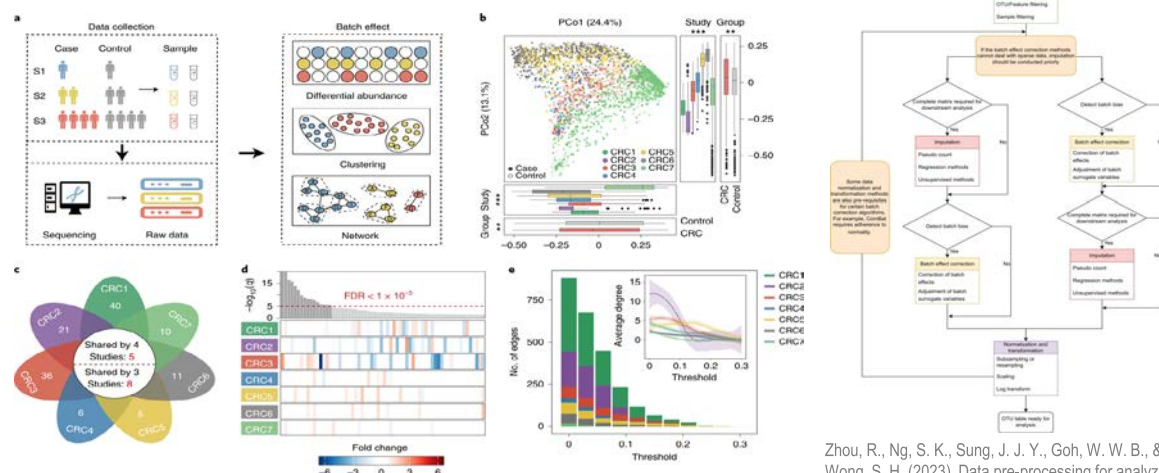


Raising standards in microbiome research. Nat Microbiol 1, 16112 (2016). <https://doi.org/10.1038/nmicrobiol.2016.112>

## 병원기반 인간 마이크로바이옴 사업

# 마이크로바이옴 데이터 표준화 필요성

- 마이크로바이옴 기반의 **새로운 진단 및 치료법 개발**을 위해서는 마이크로바이옴 **연구결과의 신뢰성 및 재현성**이 보장되어야 함
- 현재 국내 마이크로바이옴 연구는 데이터 표본 크기와 메타 데이터의 부족으로 **결과 편차가 심함**
- 표준화된 분석 방법의 부재로 인해 **연구 결과의 비교 및 통합의 어려움** 존재함



Xiao, L., Zhang, F., & Zhao, F. (2022). Large-scale microbiome data integration enables robust biomarker identification. *Nature Computational Science*, 2(5), 307-316.

Zhou, R., Ng, S. K., Sung, J. J. Y., Goh, W. W. B., & Wong, S. H. (2023). Data pre-processing for analyzing microbiome data—A mini review. *Computational and Structural Biotechnology Journal*, 21, 4804-4815.

## 병원기반 인간 마이크로바이옴 사업

# 운영 가이드라인 고도화

## 국가 마이크로바이옴 분석 표준화를 위한 표준지침 마련 ('23.3.)

CONTENTS	
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표준지침 개정을 위해 실험적 근거 마련 연구 필요

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## 병원기반 인간 마이크로바이옴 사업

# 인프라 구축 ( '23~ )



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## 병원기반 인간 마이크로바이옴 사업

## (1) 임상정보 수집

- 개인차에 의한 마이크로바이옴 조성 변화가 크기 때문에 임상/역학적 지표 연관분석이 필요
- 연관성 분석을 위한 표준화 된 공통 필수 임상정보, 질환 별 필수 지표제시 필수

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## PART 1 정보 수집 항목

- 임상정보
- 검체정보

## PART 2 검체 채취

- 소화기 검체
- 비노생식기 검체
- 호흡기 검체
- 구강 검체
- 피부 검체

## PART 3 검체, 핵산 보관 및 수송

- 검체 보관 및 수송
- 분석기관의 검체 보관 및 핵산 추출
- 핵산 보관 방법
- 분석기관의 검체, 핵산 폐기
- 핵산 추출액 기탁

## PART 4 정도관리

- 내부정도관리
- 외부정도관리
- Mock community

공통 임상정보 281 항목 + 질환군별 임상정보 111 항목

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## 병원기반 인간 마이크로바이옴 사업

## (2) 검체 채취 · 수송 및 보관

## CONTENTS

## PART 1 정보 수집 항목

- 임상정보
- 검체정보

## PART 2 검체 채취

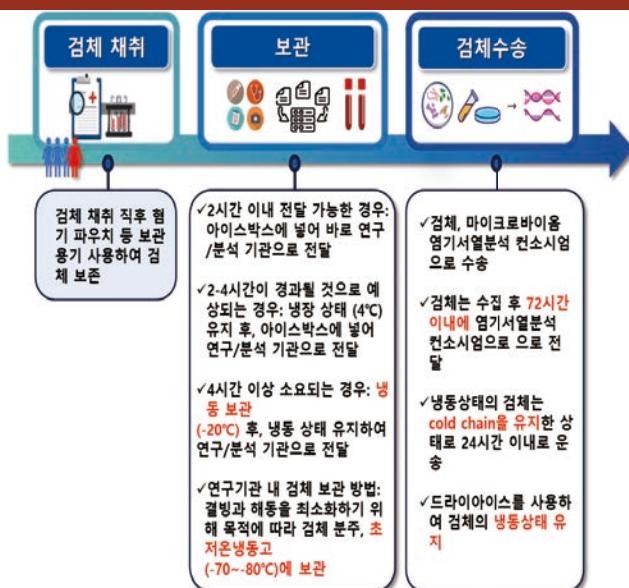
- 소화기 검체
- 비노생식기 검체
- 호흡기 검체
- 구강 검체
- 피부 검체

## PART 3 검체, 핵산 보관 및 수송

- 검체 보관 및 수송
- 분석기관의 검체 보관 및 핵산 추출
- 핵산 보관 방법
- 분석기관의 검체, 핵산 폐기
- 핵산 추출액 기탁

## PART 4 정도관리

- 내부정도관리
- 외부정도관리
- Mock community



병원기반 인간 마이크로바이옴 사업

### (3) 핵산 추출 방법

- 검체 추출 방법에 따른 미생물 구성 차이 확인을 위해 문헌에서 보고가 높은 2종의 핵산추출 키트 선정 및 비교 평가
- 평가 물질: Gut Microbiome Whole cell Mix (ATCC, MSA-2006)

#### CONTENTS

##### PART 6 핵산 추출

1. 공통사항
2. 수기법

##### PART 7 염기서열 분석 결과 유효성 평가

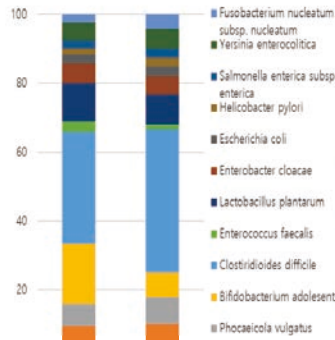
1. Amplicon sequencing
2. Whole metagenome sequencing

##### PART 8 데이터 분석

1. 시퀀싱 데이터 전처리
2. 마이크로바이옴 구성 분석
3. 마이크로바이옴 유래 세균 유전체 생성

#### [Gut]

##### Amplicon Sequencing (Miseq)



- 2종의 핵산추출 키트에서 동일하게 12 species가 검출

Species	ATCC no.	구성비 (%)
<i>Bacteriodes fragilis</i>	25285	8.3
<i>Phocaeicola vulgatus</i>	8482	8.3
<i>Bifidobacterium adolescentis</i>	15703	8.3
<i>Clostridioides difficile</i>	9689	8.3
<i>Enterococcus faecalis</i>	700802	8.3
<i>Lactobacillus plantarum</i>	BAA-793	8.3
<i>Enterobacter cloacae</i>	13047	8.3
<i>Escherichia coli</i>	700926	8.3
<i>Helicobacter pylori</i>	700392	8.3
<i>Yersinia enterocolitica</i>	27729	8.3
<i>Fusobacterium nucleatum subsp. nucleatum</i>	25586	8.3

[데이터 제공: 마이크로바이옴염기서열분석 컨소시엄]

병원기반 인간 마이크로바이옴 사업

### (4) 정도관리 (IPT, EQA)

- 내·외부 정도관리 프로트콜 개발

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##### PART 1 정보 수집 항목

1. 임상정보
2. 검체정보

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2. 비노생식기 검체
3. 호흡기 검체
4. 구강 검체
5. 피부 검체

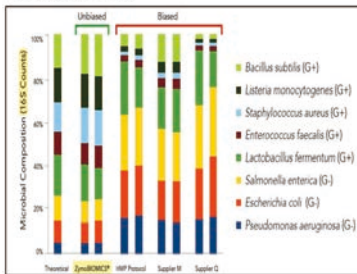
##### PART 3 검체, 핵산 보관 및 수송

1. 검체 보관 및 수송
2. 분석기관의 검체 보관 및 핵산 추출
3. 핵산 보관 방법
4. 분석기관의 검체, 핵산 폐기
5. 핵산 추출액 기탁

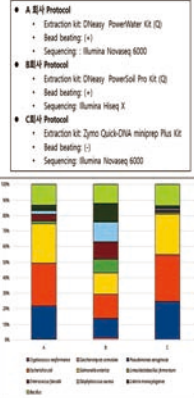
##### PART 4 정도관리

1. 내부정도관리
2. 외부정도관리
3. Mock community

#### Zymo mock community



Bacterial genus	Relative abundance range (%)
<i>Bacillus</i>	12.1 - 13.6
<i>Listeria</i>	7.4 - 9.2
<i>Staphylococcus</i>	12.0 - 13.8
<i>Enterococcus</i>	8.3 - 8.8
<i>Lactobacillus</i>	19.3 - 23.0
<i>Salmonella</i>	14.0 - 14.6
<i>Escherichia</i>	15.8 - 17.1
<i>Pseudomonas</i>	4.9 - 5.3



#### 마이크로바이옴 분석 허용 오차범위 설정

- ✓ DNA extraction 및 sequencing에서 발생할 수 있는 within-run bias 약 10%
- ✓ 분리된 run에서 추출되는 결과를 분석하여 between-run total bias를 제시
- ✓ Sequencing 과정에서 발생 가능한 오차 범위 제시

[데이터 제공: 병원기반 인간 마이크로바이옴연구개발사업 정도관리센터]



## 병원기반 인간 마이크로바이옴 사업

## (5) 염기서열 분석 플랫폼

## CONTENTS

## PART 6 핵산 추출

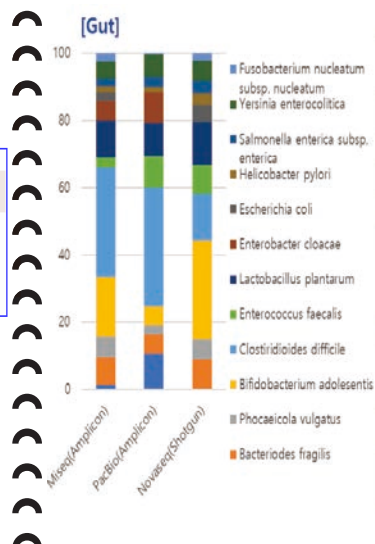
1. 공통사항
2. 수기법

## PART 7 염기서열 분석 결과 유효성 평가

1. Amplicon sequencing
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3. 마이크로바이옴 유래 세균 유전체 생성



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<i>Enterococcus faecalis</i>	700802	8.3
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<i>Enterobacter cloacae</i>	13047	8.3
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<i>Helicobacter pylori</i>	700392	8.3
<i>Yersinia enterocolitica</i>	27729	8.3
<i>Fusobacterium nucleatum subsp. nucleatum</i>	25586	8.3

[데이터 제공: 마이크로바이옴 염기서열분석 컨소시엄]

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## 병원기반 인간 마이크로바이옴 사업

## (6) 표준물질 (Mock community)

구분	시판 제품		국내 개발
	ATCC	Zymo	
구매비용 (1 set)	고비용 (>200만원)	저비용 (50만원 내외)	-
구매비용 (1 prep)			
구매절차	복잡 (수입허가필요)	간편	-
구매 소요시간	장시간 (>1개월)	단시간 (1주)	-
제품 종류	한정적	매우 한정적	신체부위별 다양화하여 개발 가능
구성 미생물	제한적	제한적	한국인 보유 주요 미생물로 구성된 맞춤형 물질 개발 가능

시판 mock community의 한정적인 종류 및 제한적인 구성 미생물 등으로  
한국 맞춤형 mock community 개발 및 확보 필요

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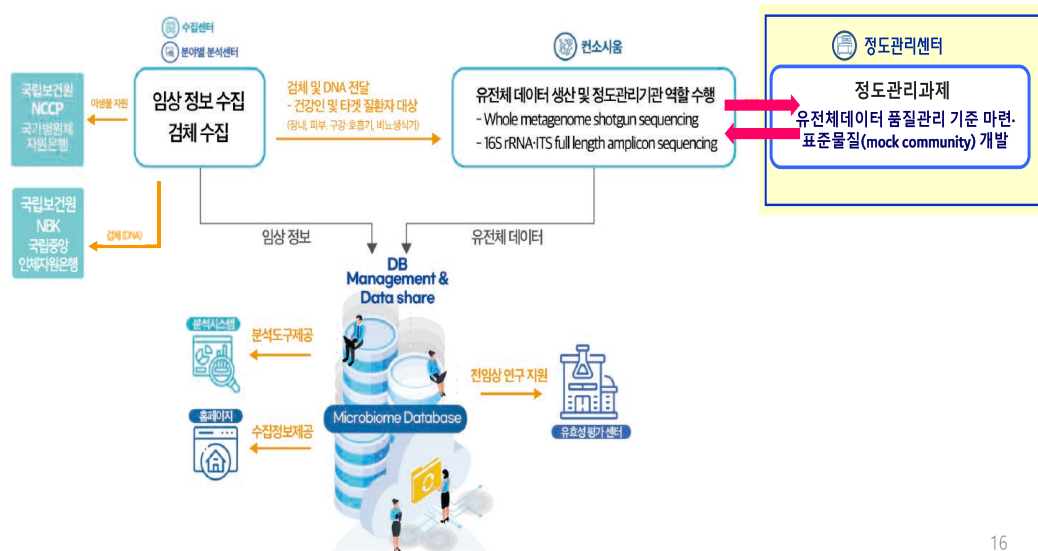
## 02

## 미생물 군집 염기서열 분석 표준화 전략 연구

### 미생물 군집 염기서열 분석 표준화 전략 연구

### 연구의 필요성

국가 차원의 신뢰할 수 있는 마이크로바이옴 데이터 생산·수집 및 통합 분석을 위해  
마이크로바이옴 분석 표준화 필요



## 미생물 군집 염기서열 분석 표준화 전략 연구

## 연구 개요

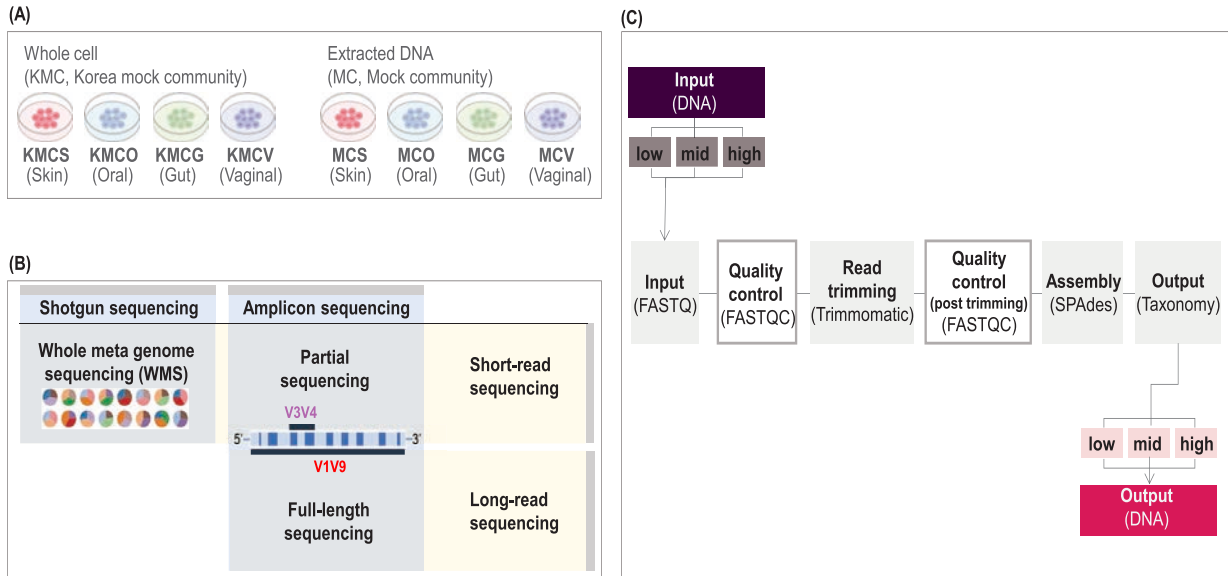


Figure 1. Study design.

(A) Sample type. (B) Technological platform. (C) Data analysis.

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## 미생물 군집 염기서열 분석 표준화 전략 연구

## 연구 결과

MCS			MCO			MCG			MCV		
Species	ATCC no.	Ratio (%)	Species	ATCC no.	Ratio (%)	Species	ATCC no.	Ratio (%)	Species	ATCC no.	Ratio (%)
<i>Acinetobacter johnsonii</i>	17909D-5	16.7	<i>Schaalia odontolytica</i>	17982D-5	16.7	<i>Bacteriodes fragilis</i>	25285D-5	8.3	<i>Gardnerella vaginalis</i>	14019D-5	16.7
<i>Corynebacterium striatum</i>	6940D-5	16.7	<i>Prevotella melaninogenica</i>	25845D-5	16.7	<i>Bacteroides vulgatus</i>	8482D-5	8.3	<i>Lactobacillus gasseri</i>	33323D-5	16.7
<i>Micrococcus luteus</i>	4698D-5	16.7	<i>Streptococcus mitis</i>	49456D-5	16.7	<i>Bifidobacterium adolescentis</i>	15703D-5	8.3	<i>Mycoplasma hominis</i>	23114D-5	16.7
<i>Cutibacterium acnes</i>	11828D-5	16.7	<i>Veillonella parvula</i>	17745D-5	16.7	<i>Clostridioides difficile</i>	9689D-5	8.3	<i>Prevotella bivia</i>	29303DQ	16.7
<i>Staphylococcus epidermidis</i>	12228D-5	16.7	<i>Haemophilus parainfluenzae</i>	33392D-5	16.7	<i>Enterococcus faecalis</i>	700802D-5	8.3	<i>Streptococcus agalactiae</i>	BAA-611DQ	16.7
<i>Streptococcus mitis</i>	49456D-5	16.7	<i>Fusobacterium nucleatum subsp. nucleatum</i>	25586D-5	16.7	<i>Lactobacillus plantarum</i>	BAA-793D-5	8.3	<i>Lactobacillus jensenii</i>	25258D-5	16.7
						<i>Enterobacter cloacae</i>	13047D-5	8.3			
						<i>Escherichia coli</i>	700926D-5	8.3			
						<i>Helicobacter pylori</i>	700392D-5	8.3			
						<i>Yersinia enterocolitica</i>	27729D-5	8.3			
						<i>Salmonella enterica subsp. enterica</i>	9150D-5	8.3			
						<i>Fusobacterium nucleatum subsp. nucleatum</i>	25586D-5	8.3			

Table 1. Bacterial composition of MC.

MC consists of MCS (MC-skin), MCO (MC-oral), MCG (MC-gut), and MCV (MC-vaginal).

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## 미생물 군집 염기서열 분석 표준화 전략 연구

### 연구 결과

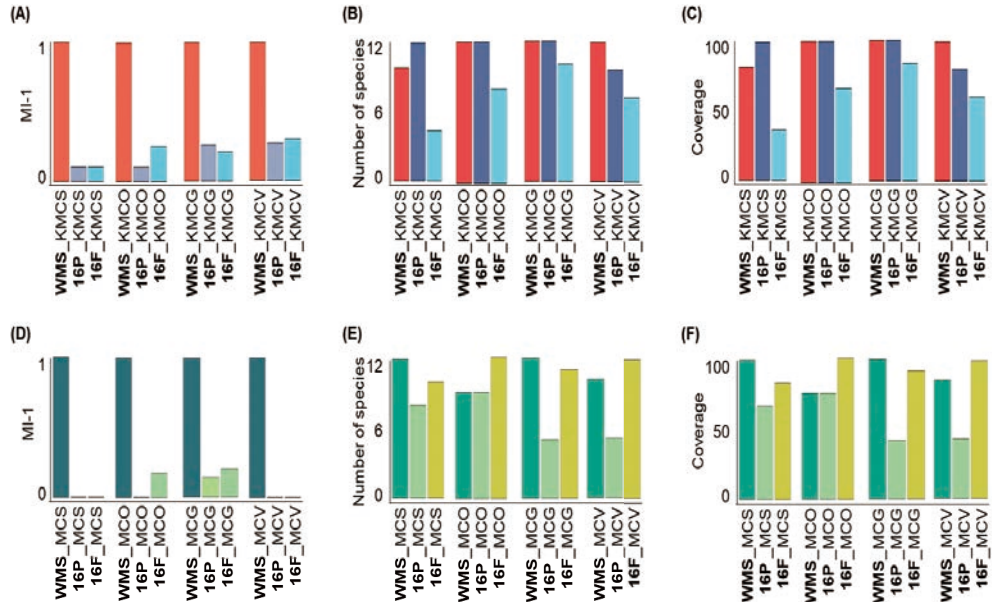


Figure 2. Comparison by Sequencing Platform.

(A-C) Comparison of sequencing platforms for KMC and (D-F) MC. (A, D) Qualitative reproducibility evaluation through MI (matching index)-1. (B, E) Observed number of species for each sequencing. (C, F) Library coverage for each sequencing.

## 미생물 군집 염기서열 분석 표준화 전략 연구

### 연구 결과

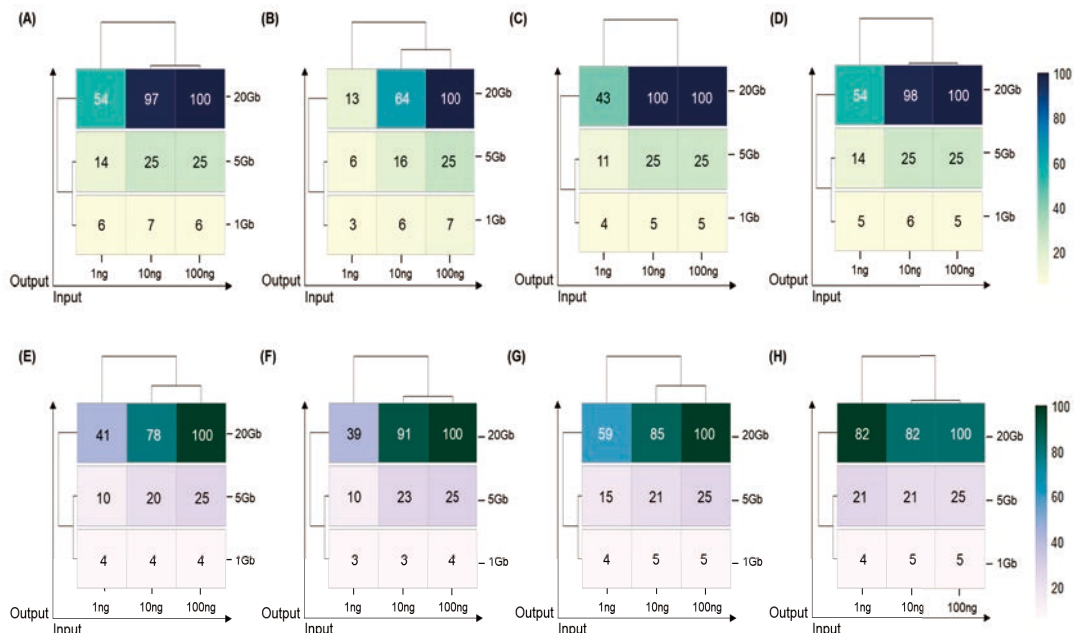


Figure 3. Resolution for shotgun sequencing data.

Analysis of optimal resolution in shotgun sequencing of WMS. Quantitative coverage evaluation was performed based on the high-score input and high-score output in shotgun sequencing through MI-2 (standard option). The resolution for (A-D) KMC and (E-H) MC was evaluated.

**감사합니다.**