



전자파와 장내 미생물 변화 연구

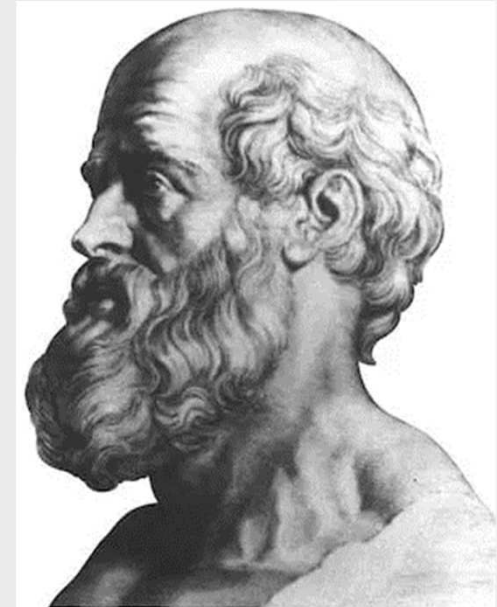
김혜선, 안영환
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Introduction: Microbiome

❖ Gut and health

“All Disease Begins in the Gut”

- Hippocrates
(BC 460-377)



Introduction: Microbiome and Research in RF

❖ International NTP validation project

- A 5-year animal experiment project
- 2-year life-long RF-EMF exposure
- 3 groups (cage-control, sham- and RF-exposed)
- 70 male rats per group

- GLP facilities of KIT (Korea) and DIMS (Japan)
- Reverberation chamber system
- Whole body SAR of 4W/kg

- As a part of **NTP⁺ study** in Korea, microbiome investigation designed

Introduction: Microbiome and Research in RF

❖ EMF and human bacteria

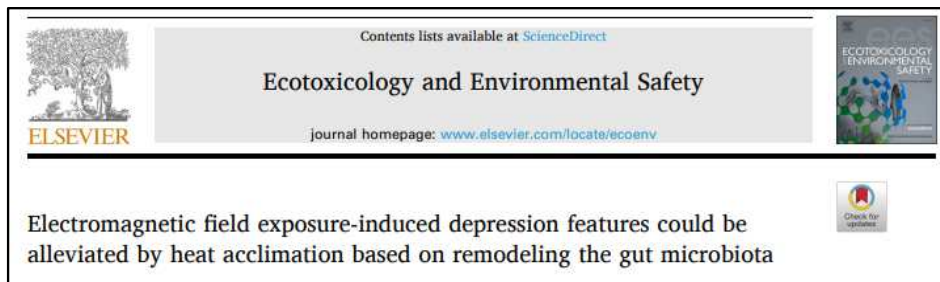


- Growth patterns of lab bacterial cultures under Static EMF were variable per different species
- Bacterial isolates of skin microbiota (n=4 different cell phone usage) showed inconsistent growth responses

Crabtree DPE et al (2016), J Microbiol

→ 1st EMF microbiome report

❖ RF-EMF and Gut microbiome



- RF-EMF (2.5W/kg SAR, 4h/d, 5wk) led to depression-like neurobehavioral disorders, gut microbiota, and metabolite imbalances

Luo X et al (2021), Ecotoxicol Environ Safety

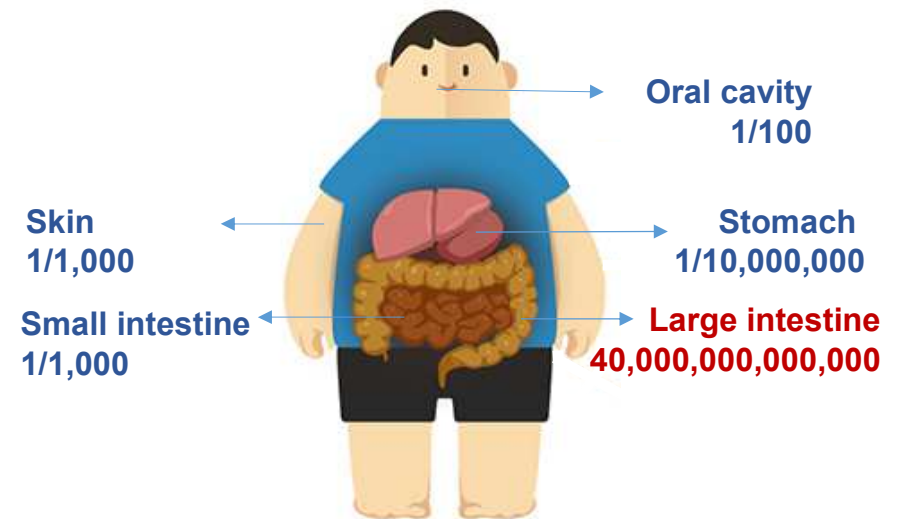
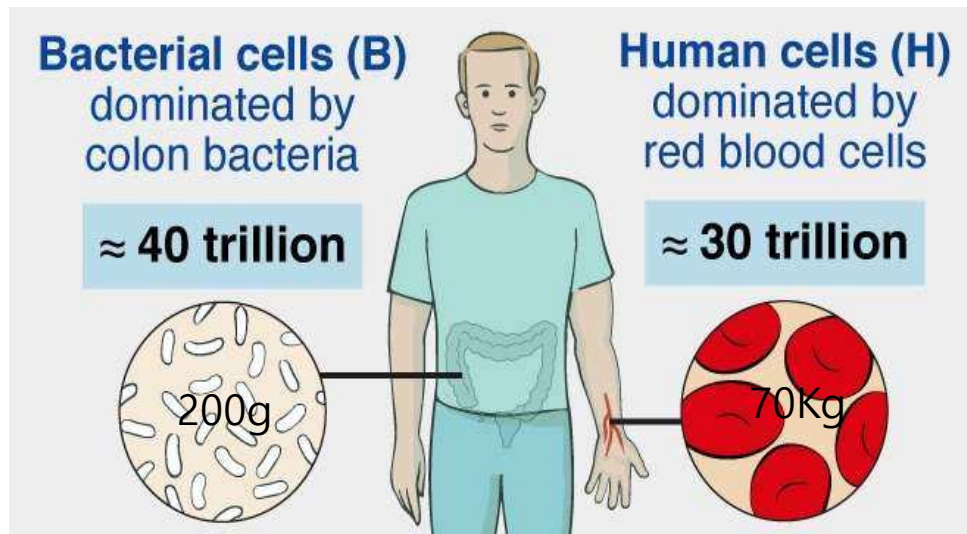
- A few studies examined RF-EMF exposure effects on biological processes
- Direct link to the altered microbiome and diseases remains inconclusive

Introduction: Microbiome

❖ Microbiota

- Living microorganisms found in a defined environment
- Bacteria, Archaea, Protozoa, Fungi, and Algae

❖ Microbiota in Numbers



Sender R et al (2016), ProS Biol.
<https://www.bioserendipity.com/cancers-with-a-possible-bacterial-contribution/>

Introduction: Microbiome

❖ Microbiota + **biome** = Microbiome (미생물 생태계)

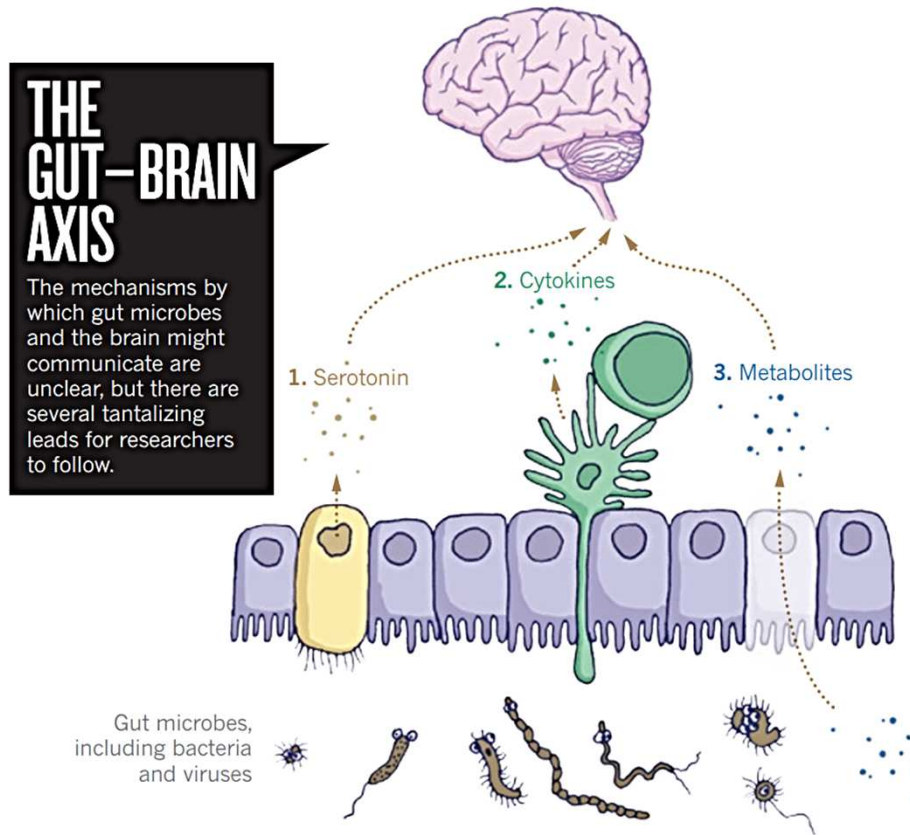
- Activities: **microbial structures**, **metabolites**, mobile genetic elements, and relic DNA embedded in the environmental conditions of the habitat
- 6 major phyla: *Firmicutes*, *Bacteroidetes*, *Actinobacteria*, *Fusobacteria*, *Proteobacteria*, *Verrucomicrobia*

❖ **Roles** of microbiome

- Regulation of **host immune response**
- Maintenance of **intestinal barrier integrity**
- Modulation of host-cell proliferation and vascularization
- Regulation of intestinal, **neurological**, endocrine functions
- **Energy source** (5-10% of daily host energy)
- **Synthesis of vitamins and neurotransmitters**

Introduction: Gut-Brain axis

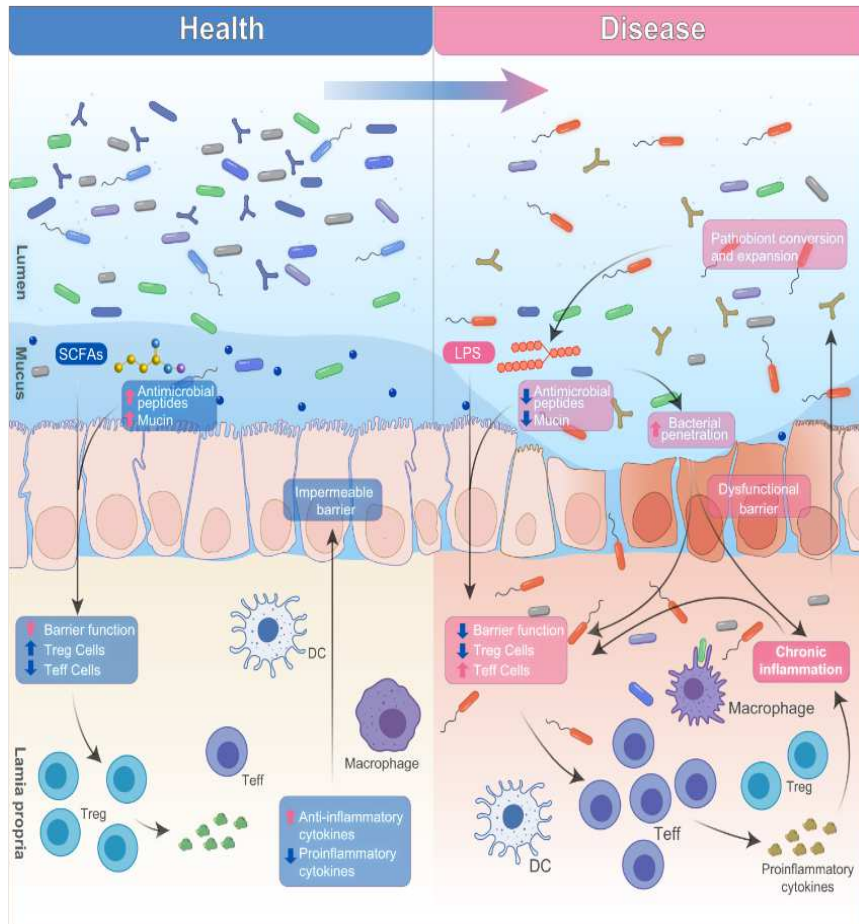
- ❖ Complex network involving multiple biological systems
- ❖ Maintain the balance of gastrointestinal, central nervous, and microbial systems.



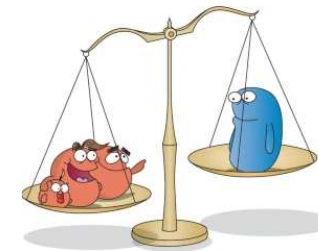
- ① **Chemical transmitters** facilitate direct communication between gut bacteria and brain
- ② **Nerve signals** relay information between gut and brain
- ③ **Immune cells and cytokines** mediate communication between gut and brain

Introduction: Microbiota Dysbiosis

❖ Composition of resident commensal communities and individual health



- Loss of beneficial microbial organisms
- Expansion of pathobionts or potentially harmful microorganisms
- Loss of overall microbial diversity

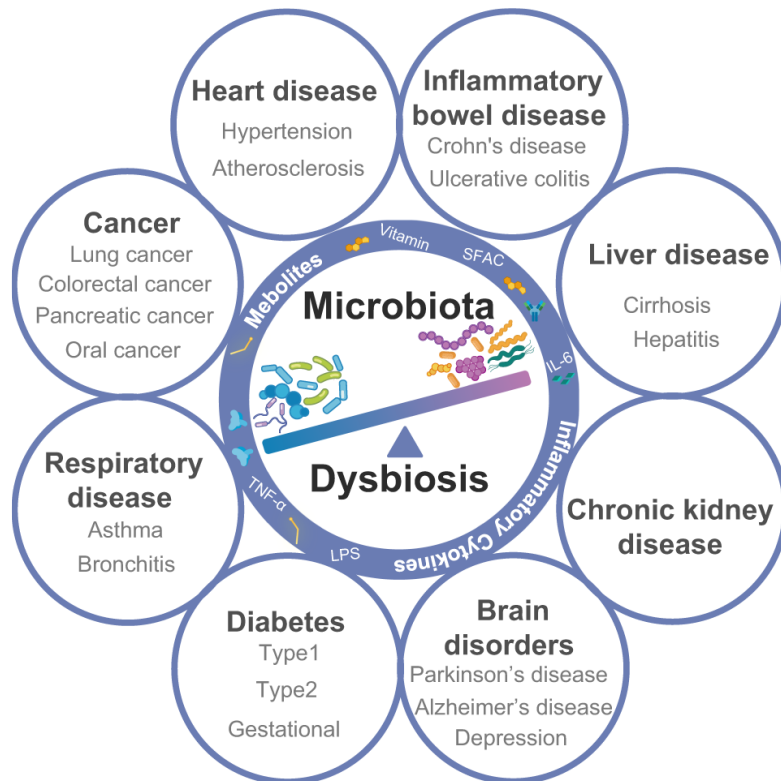


Petersen et al (2014), Cell Microbiol.

Introduction: Microbiota Dysbiosis

❖ High correlation between intractable diseases and gut microbiota dysbiosis

- Microbiota dysbiosis



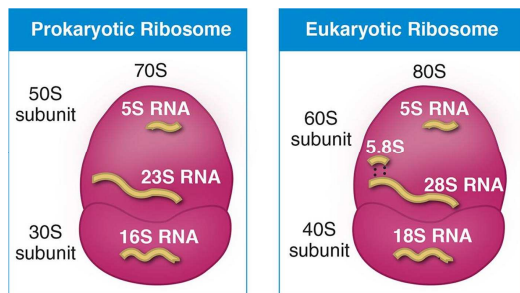
- Intractable Disease in the 21st Century

- ✓ Immune diseases
- ✓ Metabolic disease
- ✓ CNS disease
- ✓ Cancer

Introduction: Microbiome Research Methods

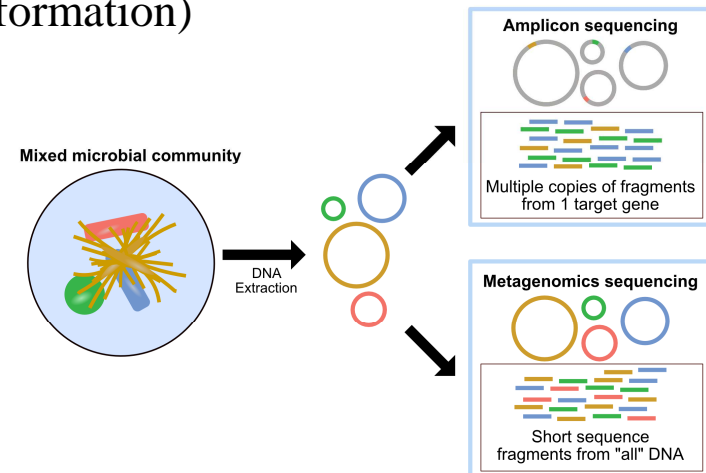
16S ribosomal RNA sequencing

- ❖ Targeted manner
- ❖ Hypervariable region (V1-V9) within bacterial 16S rRNA gene
- ❖ Specific bacterial taxa and their relative abundances (Taxonomic information) can be assessed by referencing public databases



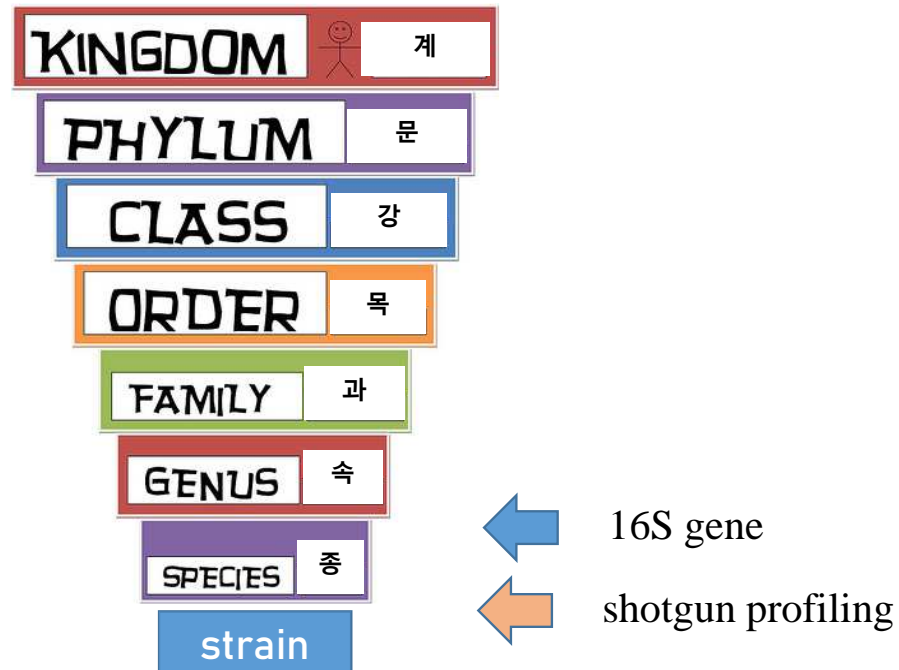
Metagenomic shotgun sequencing

- ❖ Untargeted manner
- ❖ High coverage (50-100x) of the unknown genome
- ❖ Can predict metabolic pathways that genes are involved (functional information)



Introduction: Microbiome Research Methods

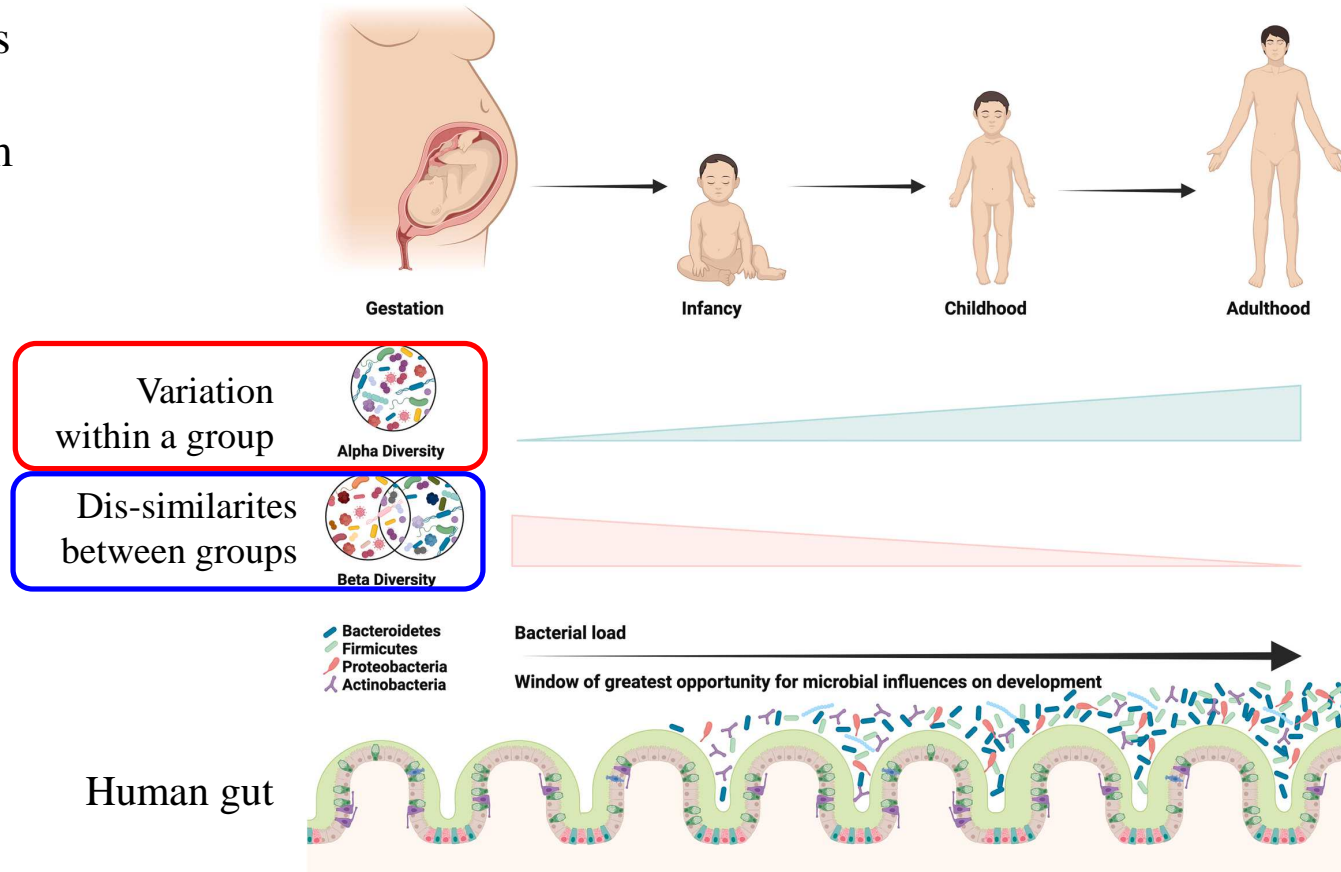
❖ Taxonomic resolution 16S gene and shotgun profiling



Taxonomic Hierarchy

Introduction: Microbiome Alpha and Beta-Diversity

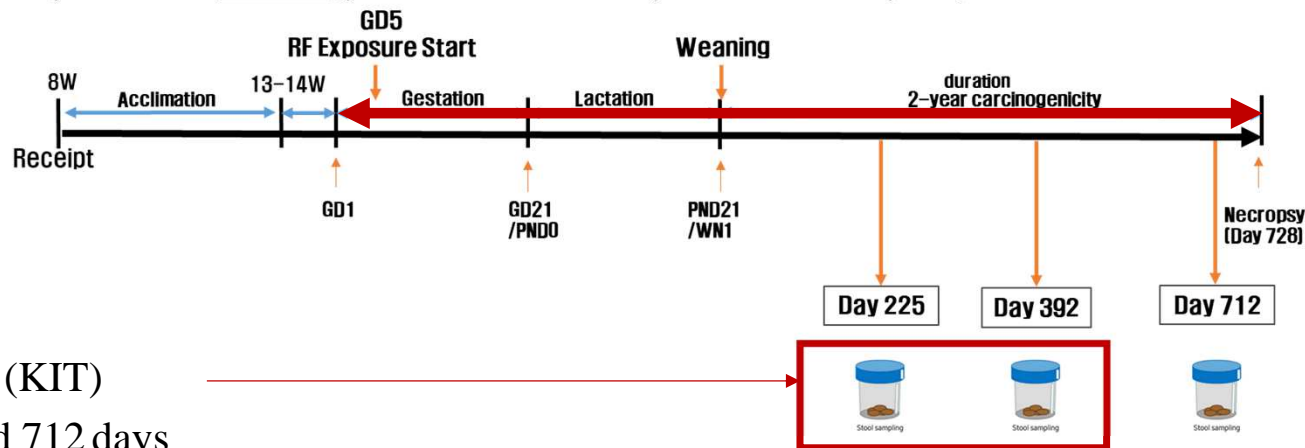
- ❖ Age-related changes in the human gut microbial ecosystem



- Healthy infant microbiome
 - **HIGH CONC.** : *Bifidobacterium* and *Lactobacilli*
 - **LOW CONC.** : *E. coli*, *Enterobacter*, and *Bacteroidetes*.

Study of Microbiome and RF-EMF : methods

- ❖ **Aim:** To investigate the **RF-exposure-induced changes** of microbiome compositions in rat
- ❖ **Reverberation chambers as RF exposure system** (KIT, Daejeon, Korea)
- ❖ **Experimental schemes**
 - Harlan SD male rat, Cage control- (n=29), Sham- (n=30) and RFR (n=30) group
 - 900 MHz CDMA (Wb SAR 4 W/kg) for 18 h 20 min (10 min on/off)/day



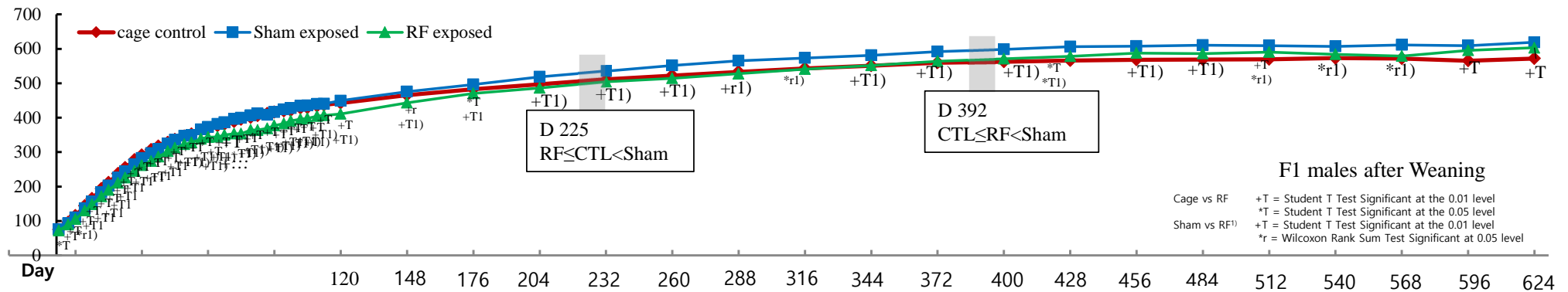
- Fecal sampling (KIT)
at 225, 392, and 712 days

- Microbiota analysis of fecal microbiota (16S rRNA amplicon sequencing)
- Interpretation of data

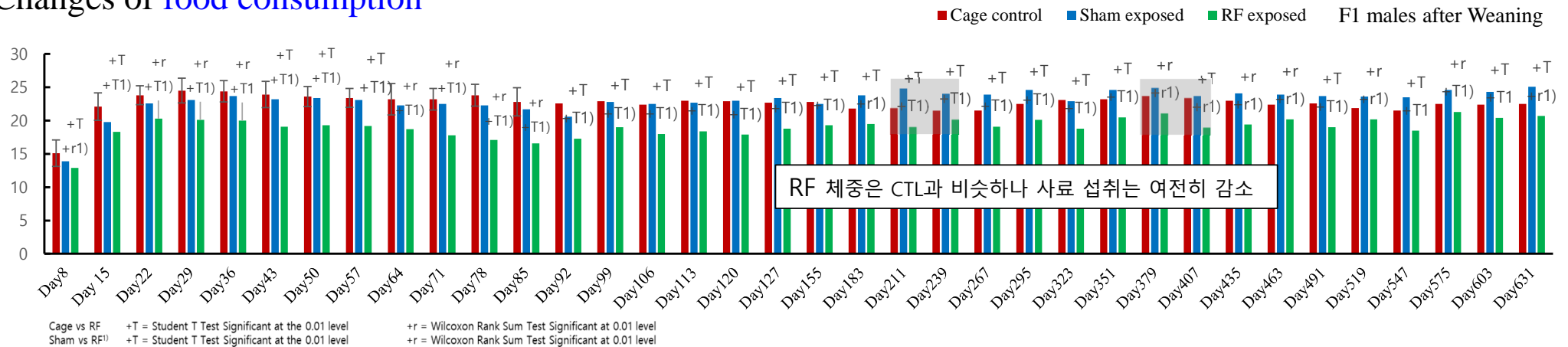
Data Source: KIT

Changes of Body weight and Food consumption during RF exposure

❖ Changes of body weight



❖ Changes of food consumption

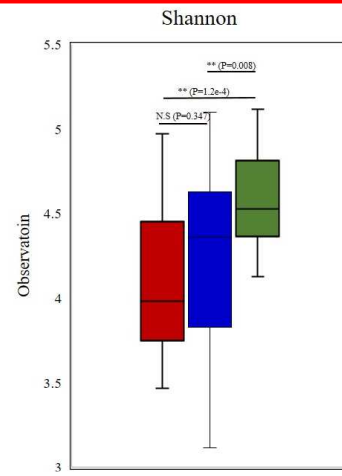
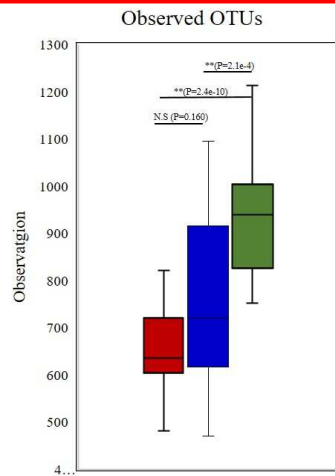


Data Source: KIT

Result: Microbiome Alpha-Diversity

❖ Microbiome index analysis : Observed OTUs and Shannon (D 225 + D 392)

D 225

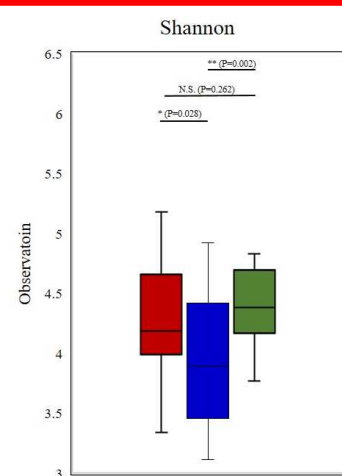
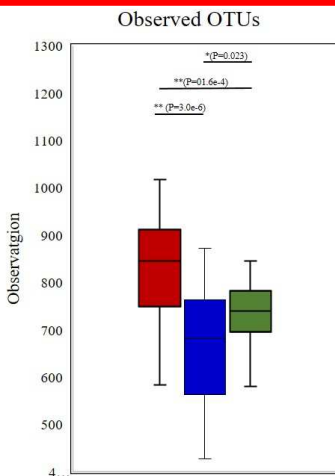


■ CTL (n=29)
■ Sham (n=30)
■ RF (n=30)

✓ RF group

- ✓ Higher richness
- ✓ Higher diversity
- ✓ Similar in pattern (2 indices)

D 392

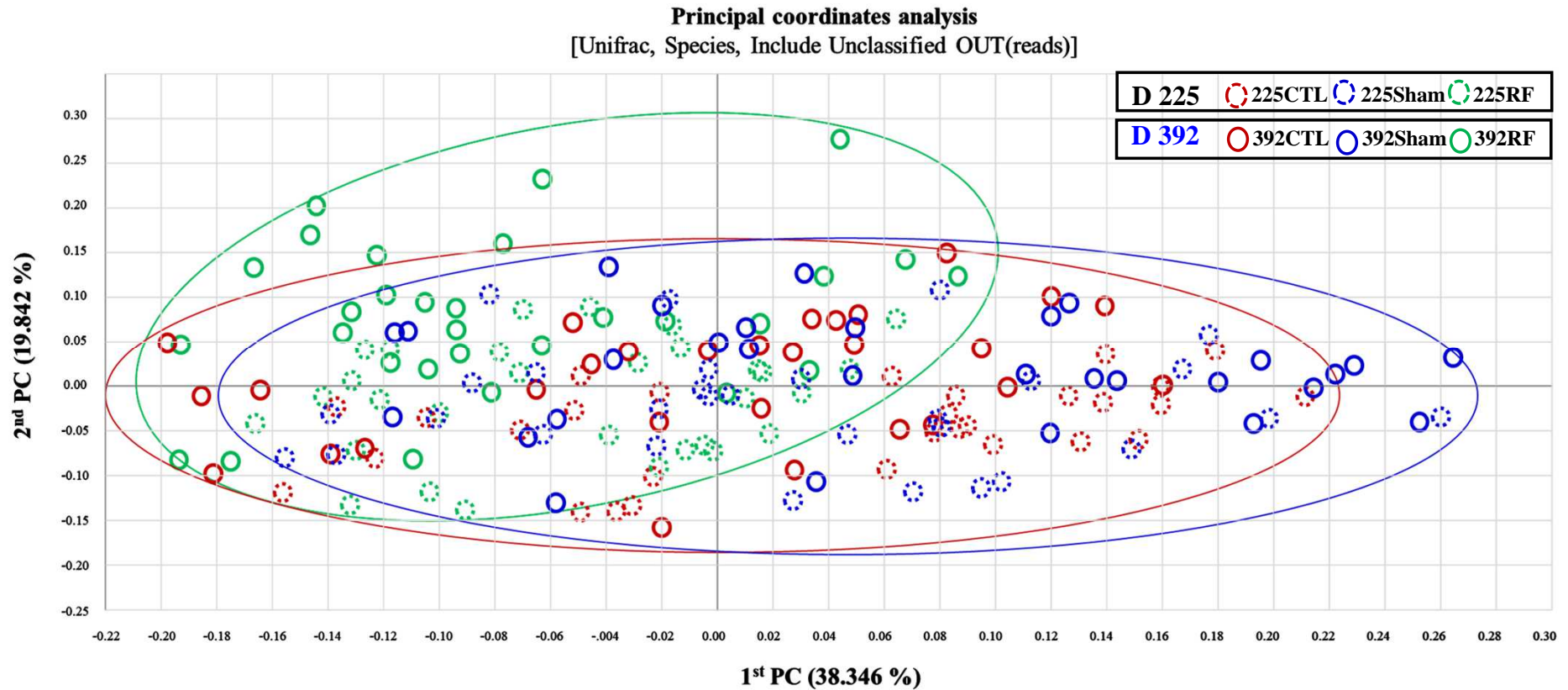


- ✓ Similar in pattern (2 indices)
- ✓ Changes in patterns are also similar overtime

- Observed OTUs: Number of species distributed / sample (richness)
- Shannon index: Diversity of the species present in the sample

Result: Microbiome Beta-Diversity

❖ Comparison by groups: CTL, Sham, and RF

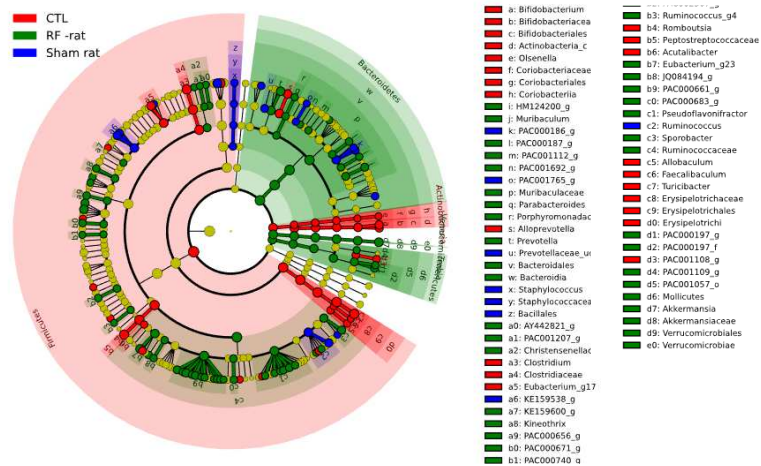


✓ Significant difference in beta-diversity between D 225 and D 392

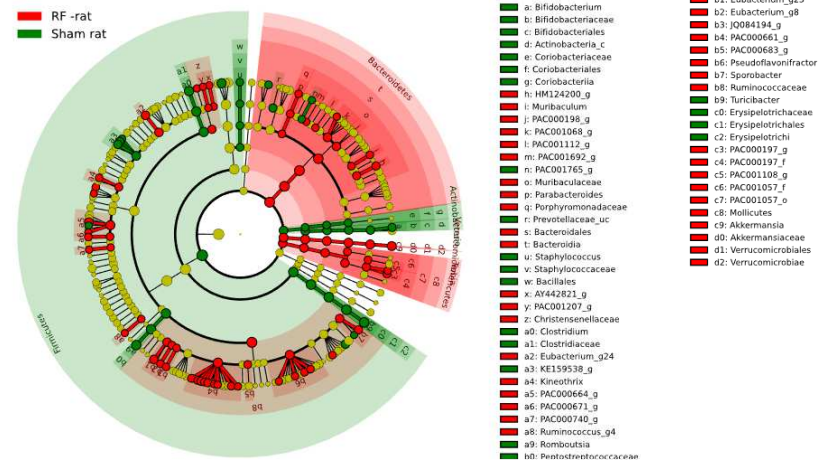
Result: Differentially Abundant Taxa of Microbiome

❖ Cladogram: Taxonomic changes in fecal microbiome (D 225)

- LefSe cladogram (Three-group comparison)



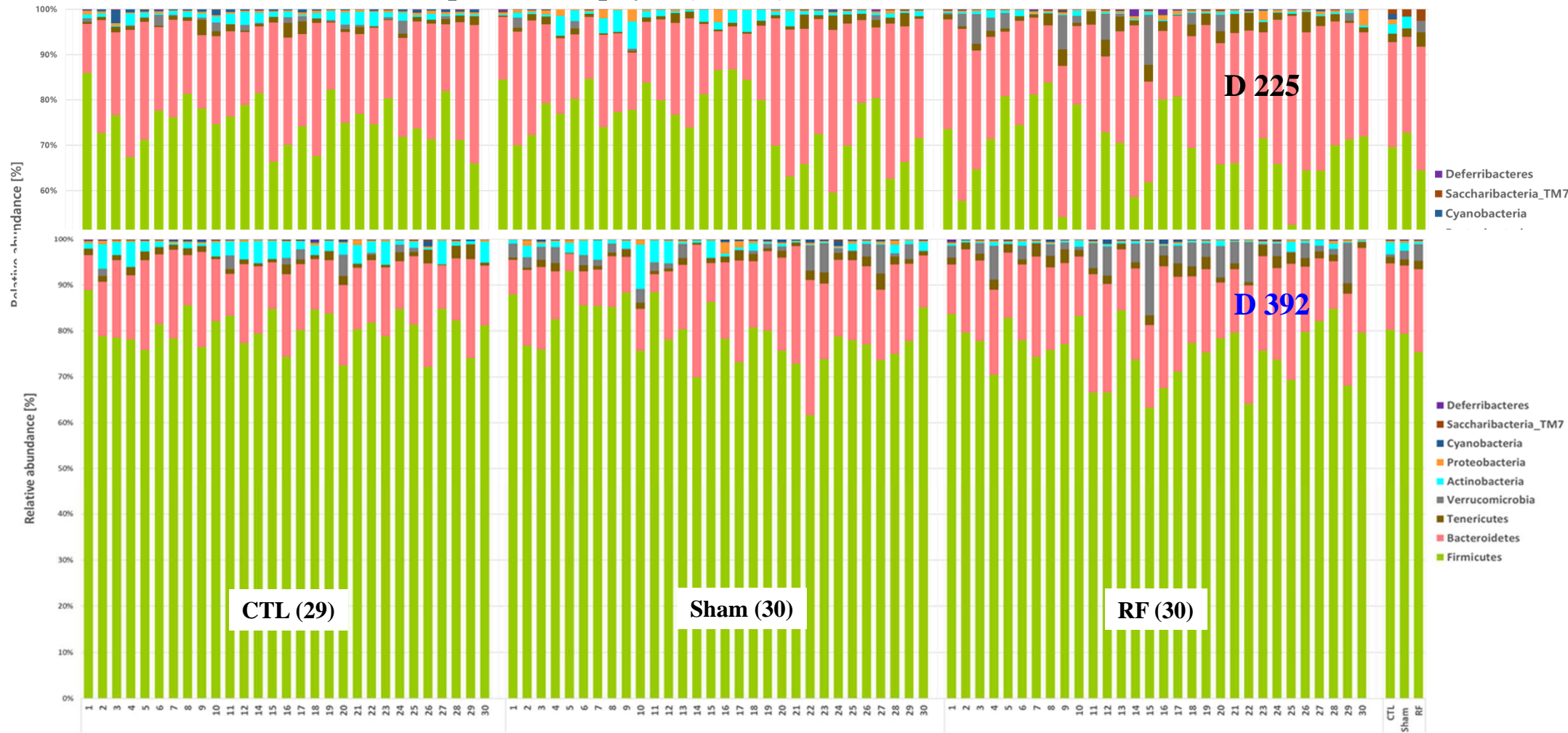
- LefSe cladogram (Two-group comparison)



Result: Taxonomic composition

❖ Relative abundances of the top bacterial phyla (D 392)

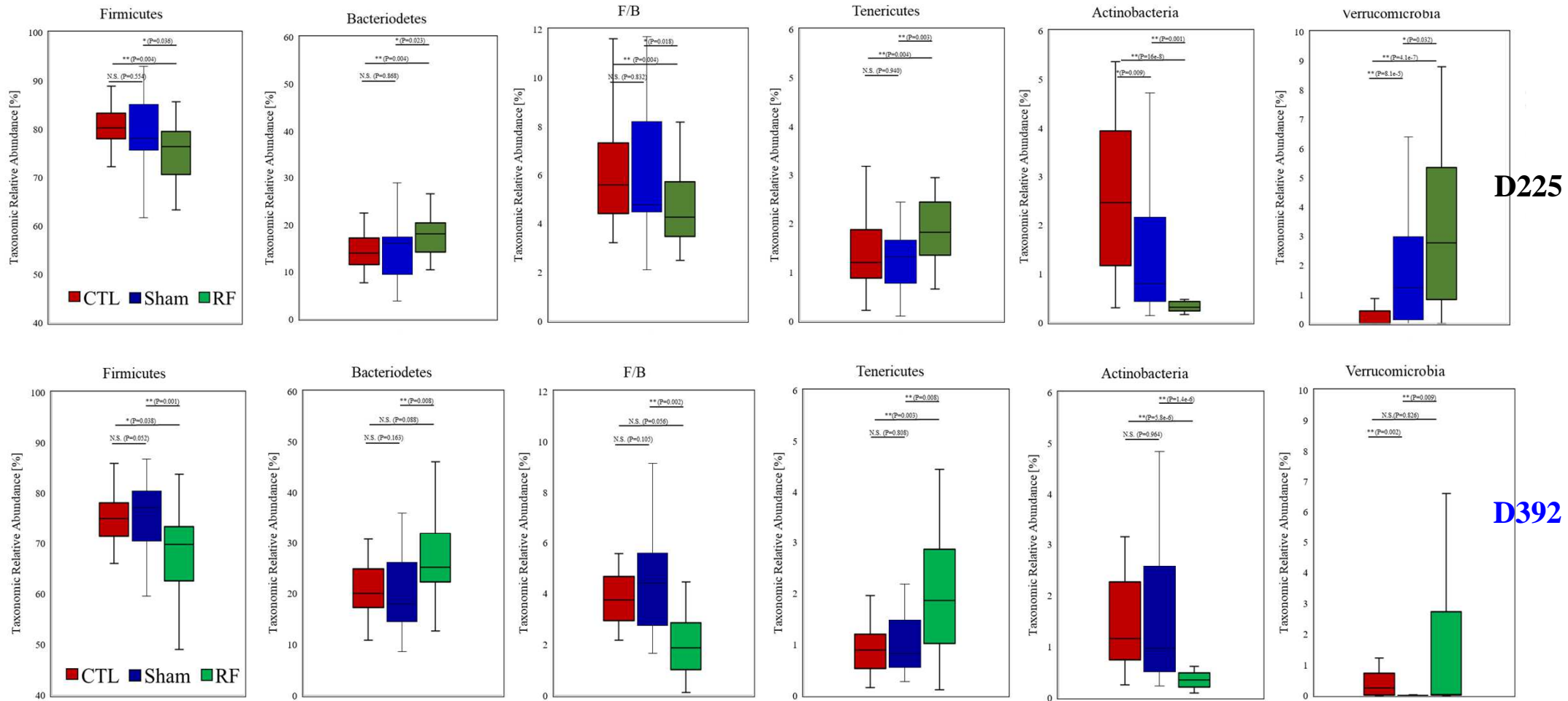
✓ Similar pattern



Result: Taxonomic composition

❖ Relative abundances of the top bacterial phyla (D 392)

✓ Similar pattern

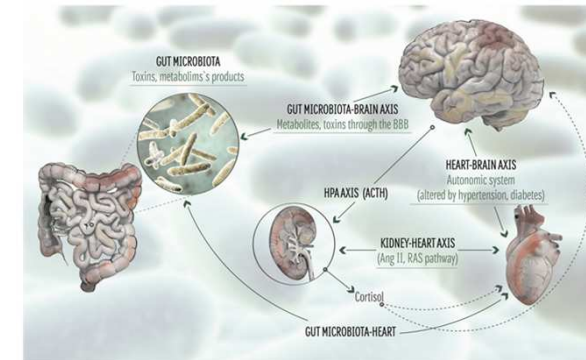


In Summary

- ❖ The fecal microbiome **composition** of the 3 groups was **heterogeneous**.
- ❖ **The major phyla** looked different in the RF-exposed group compared to cage-control and sham-exposed rats.

Take home messages

- ❖ RF-EMF **may cause changes of gut microbiome** in rat.
- ❖ This RF-exposure associated alteration of the gut microbiome **need to be studied further**



NTP+ Collaboration in KOREA

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ETRI

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- Kihwea KIM (RRA)
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- Hae-June Lee(KIRAMS)

KOREA steering committee (KSC)