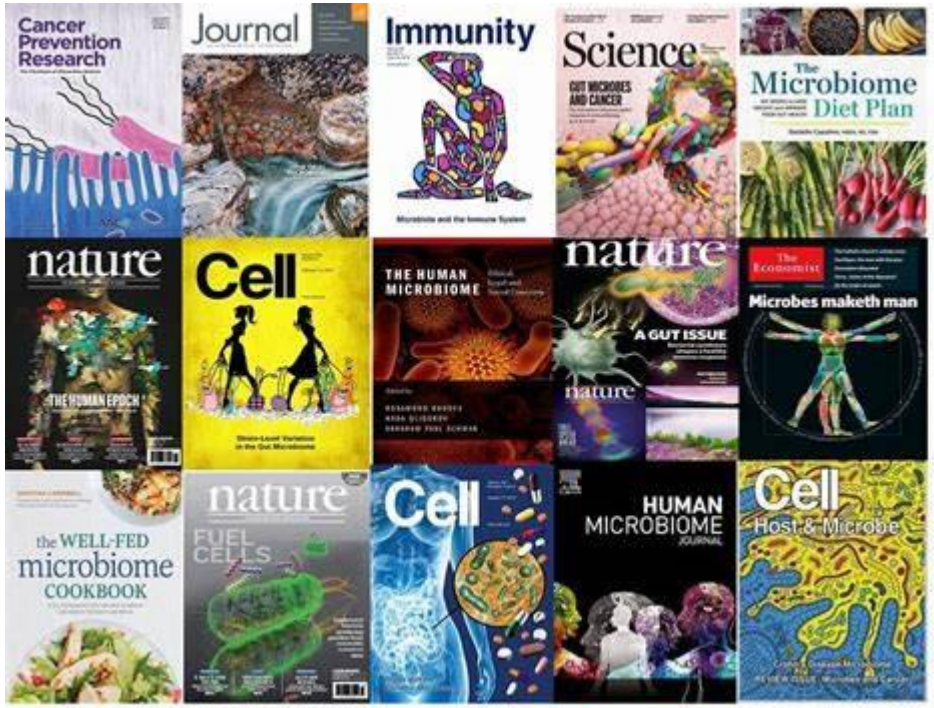
Investigarea prin metode independente de cultivare a dinamicii microbiomului in diferite patologii

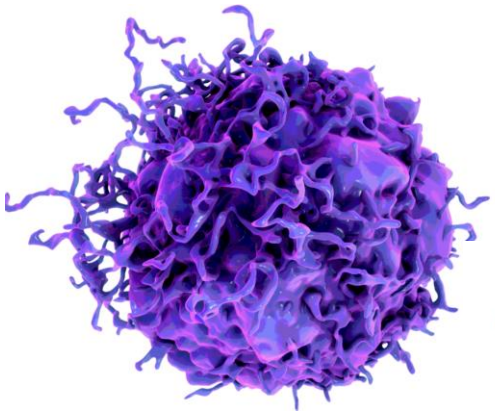
Dr. Gratiela Gradisteanu Pircalabioru, eBio-hub Research Center & ICUB

Era microbiomului…



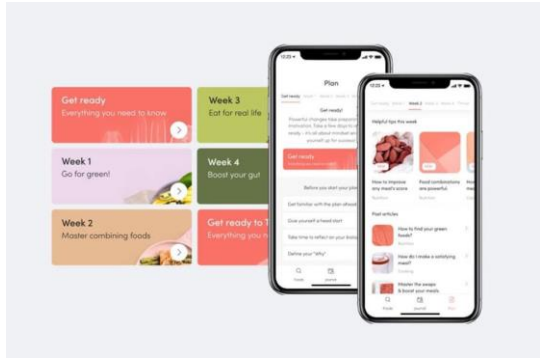
microbiotica

Translating Precision in Microbiome Science into Transformative Medicines and Biomarkers



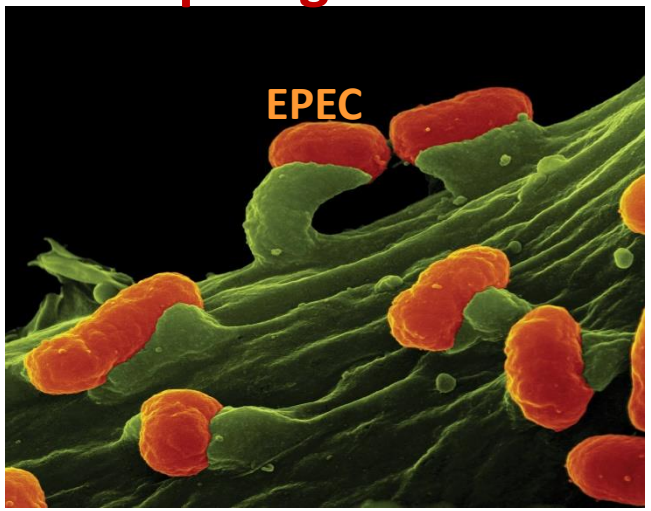
gut microbes against cancer

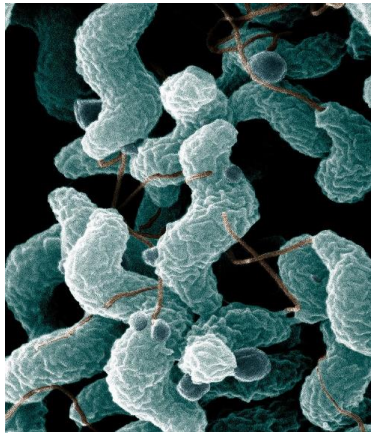
everIMMUNE



Short bio

Gazda-patogen 2011





2012

2016 –Rolul microbiotei in infectii

Cell Host & Microbe
Article

CellPress

Defensive Mutualism Rescues NADPH Oxidase Inactivation in Gut Infection

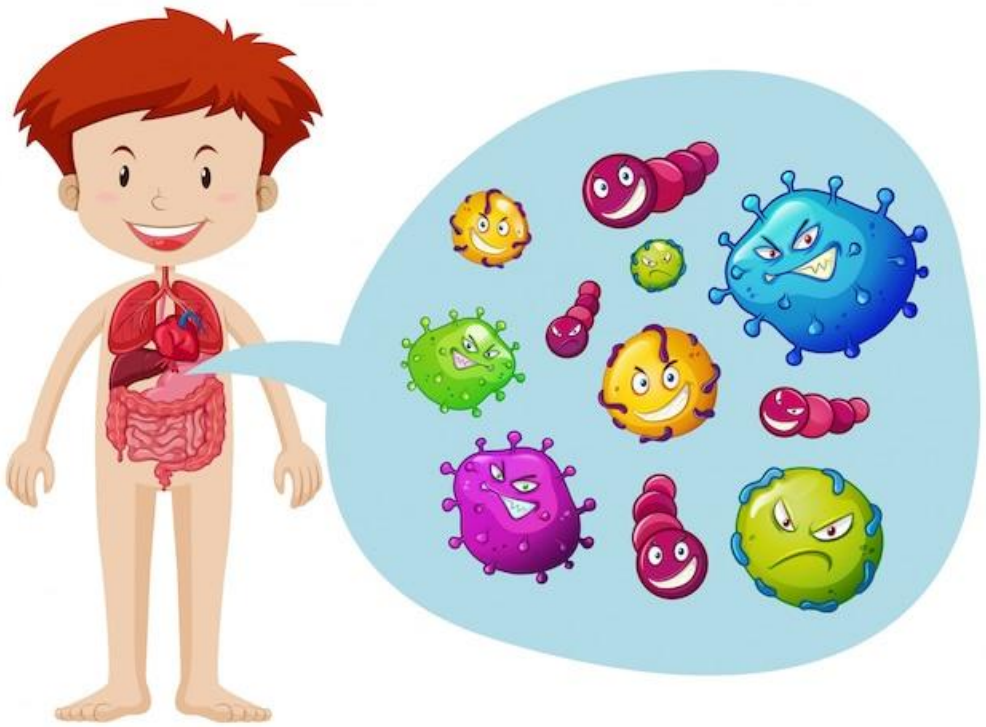
Gratiela Pircalabioru,[1] Gabriella Aviello,[2] Malgorzata Kubica,[1] Alexander Zhdanov,[3] Marie-Helene Paclet,[4] Lorraine Brennan,[5] Rosanne Hertzberger,[6] Dmitri Papkovsky,[3] Billy Bourke,[2] and Ulla G. Knaus[1,2,*]
[1]Conway Institute, School of Medicine, University College Dublin, Belfield, Dublin 4, Ireland
[2]National Children's Research Center, Our Lady's Children's Hospital Crumlin, Dublin 12, Ireland
[3]School of Biochemistry and Cell Biology, University College Cork, Cork T12 YN60, Ireland
[4]Institut de Biologie et Pathologie, Centre Hospitalier Universitaire de Grenoble, Grenoble 3809, France
[5]School of Agriculture and Food Science, University College Dublin, Dublin 4, Ireland
[6]Department of Molecular Microbiology, Washington University School of Medicine, St. Louis, MO 63110, USA
*Correspondence: ulla.knaus@ucd.ie
http://dx.doi.org/10.1016/j.chom.2016.04.007

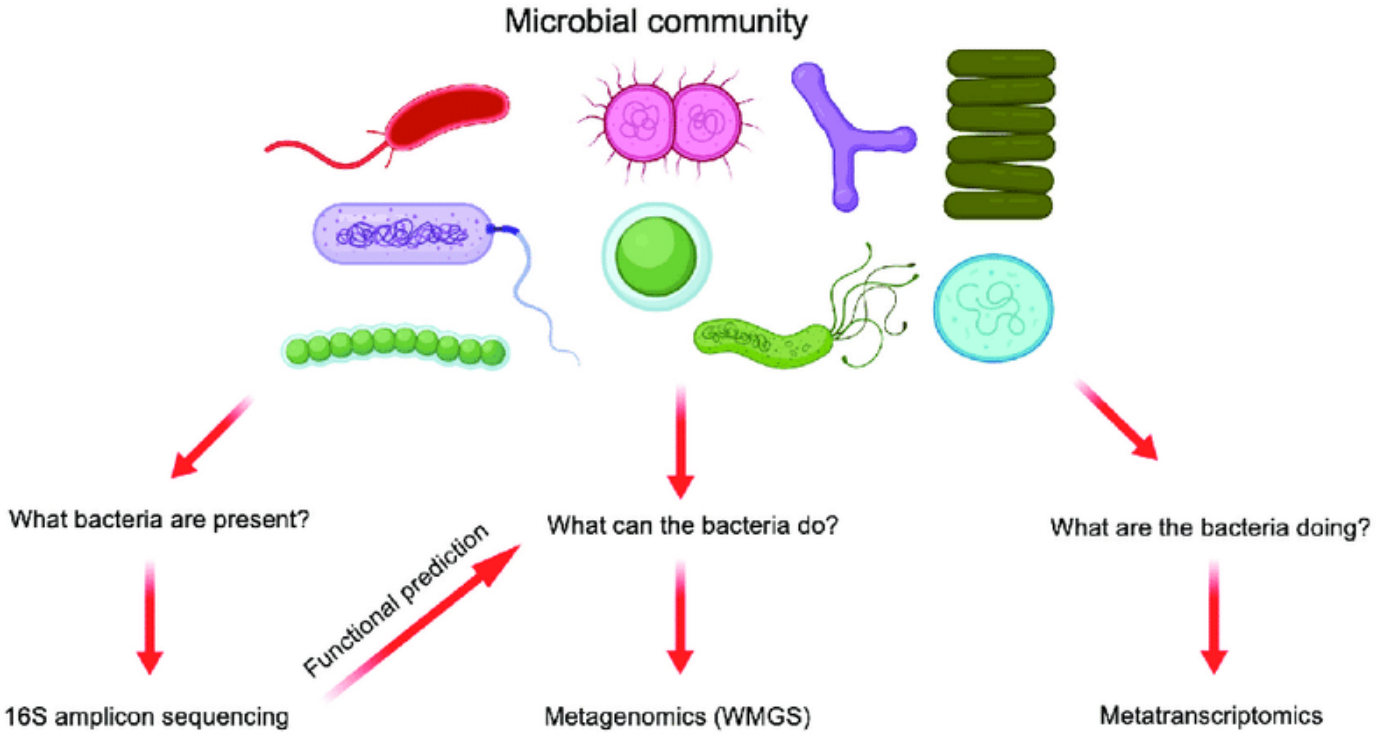
2019 – Reteaua Microbiome and Us

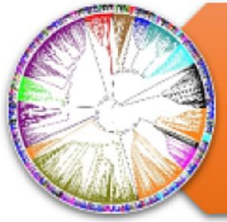
2021 –Setup NGS la ICUB

Cum analizam microbiomul?

Cum analizam microbiomul?







Gene Marker Analysis

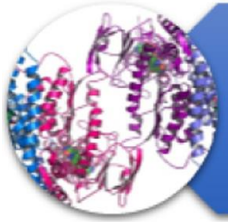
Technology Platform: Next Generation Sequencing
Common Software : QIIME, Mothur, VEGAN, phyloseq, DADA2
Pros: Cost-effective, analytical pipelines widely accepted
Cons: Lacks clear functional information, potential errors in differentiation of taxa

Shotgun Metagenomics

Technology Platform: Next Generation Sequencing
Common Software : IDBA-UD, SPAdes, MEGAHIT, MetaPhlAn2 , MG-RAST, HUMAnN2
Pros: Captures all microbial genomes present within a sample
Cons: Expensive, computationally demanding, no consensus on analytical pipelines

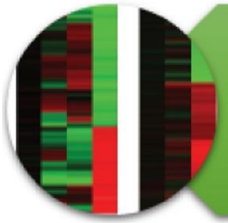
Metabolomics

Technology Platform: LC/GC-MS
Pros: allows for profiling of the metabolites microbiota produce, semi-quantitative
Cons: Origin of metabolite unknown



Metaproteomics

Technology Platform: LC/GC-MS
Pros: Allows for identification and quantification of the proteins within a sample
Cons: Origin of protein unknown

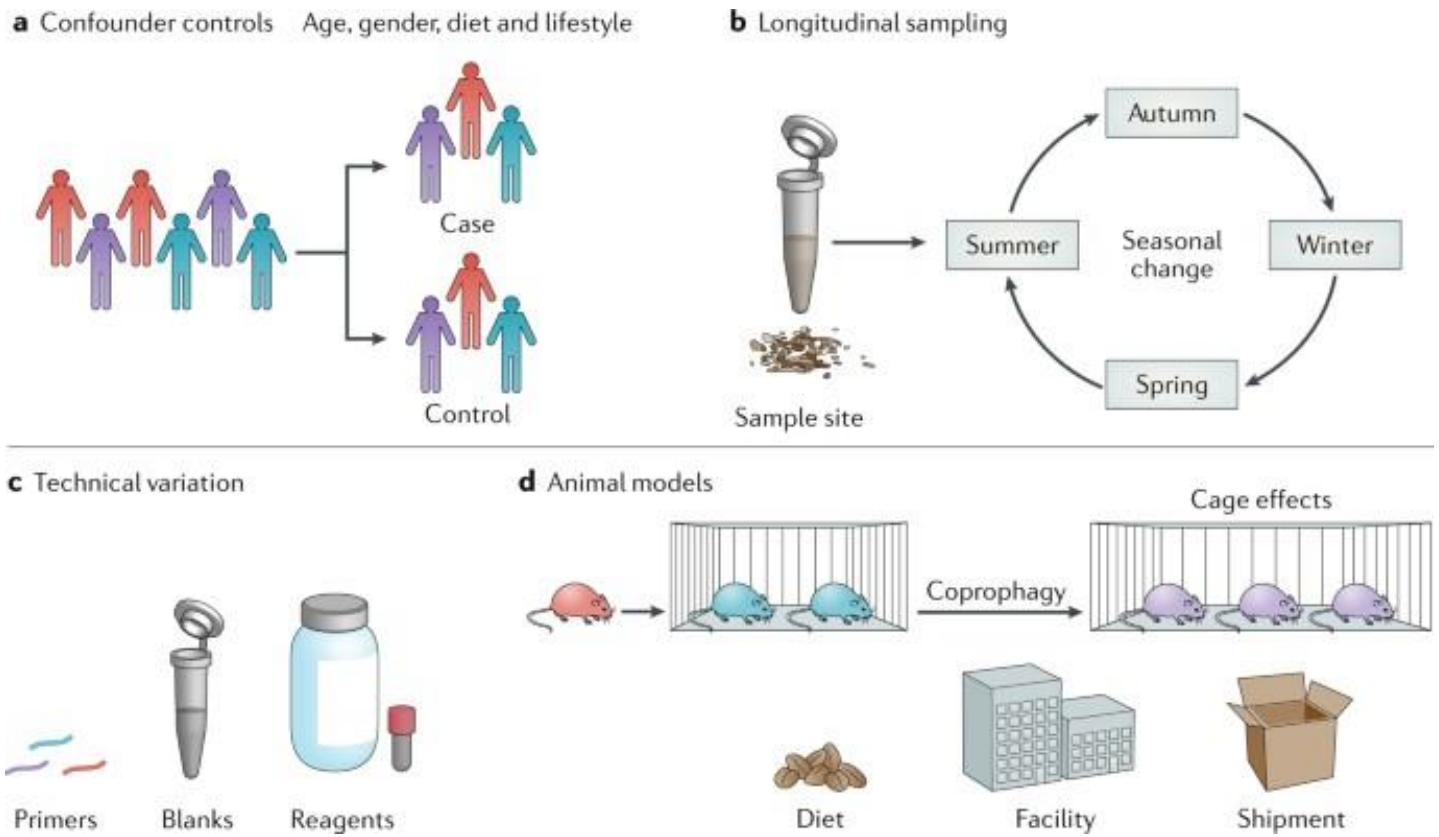


Metatranscriptomics

Technology Platform: Next Generation Sequencing
Common Software : SOAPdenovo
Pros: Allow assessments of gene expression
Cons: Protein expression may depend on translation and post-translational modifications

Cum analizam microbiomul?

Analiza microbiomului –Tips & Tricks



a Stratificarea subiectilor – varsta, sex, dieta, stil de viata, regim medicamentos

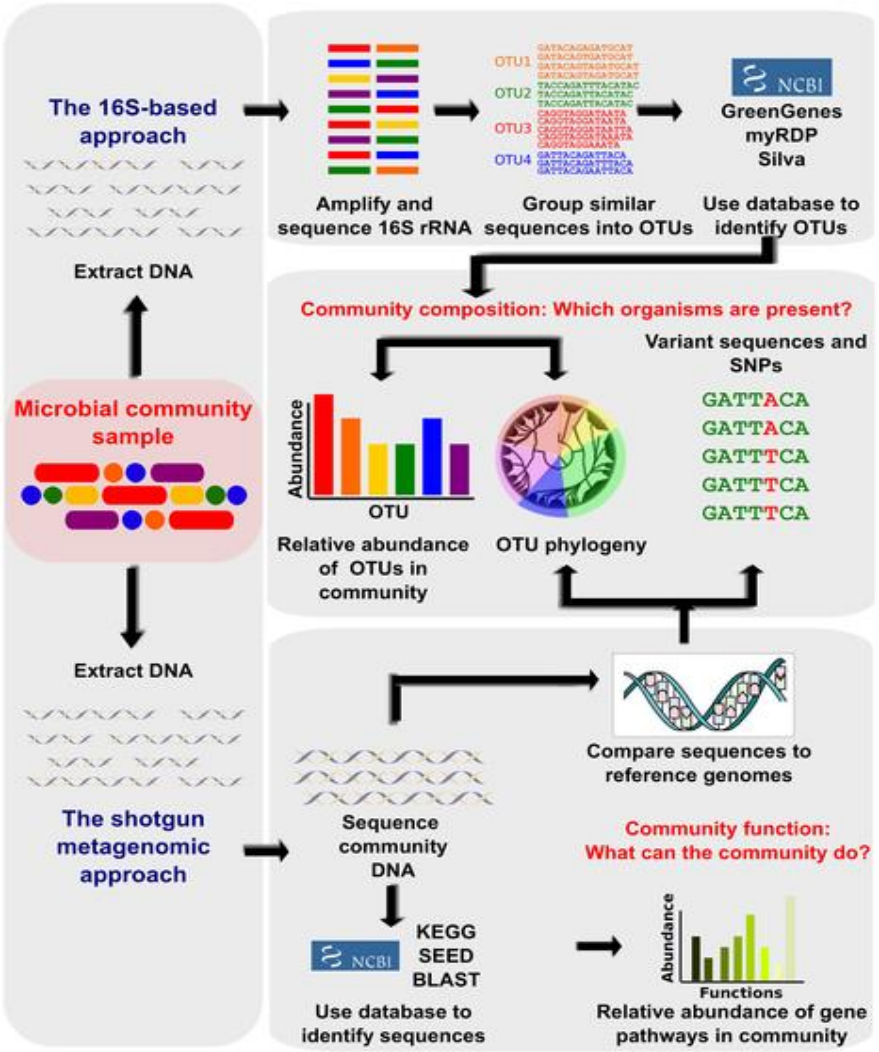
b Studii longitudinale

c Standardizarea factorilor de natura tehnica

d Studii pe model animal – impactul coprofagiei

Analiza ARNr 16S vs. Analiza Shotgun

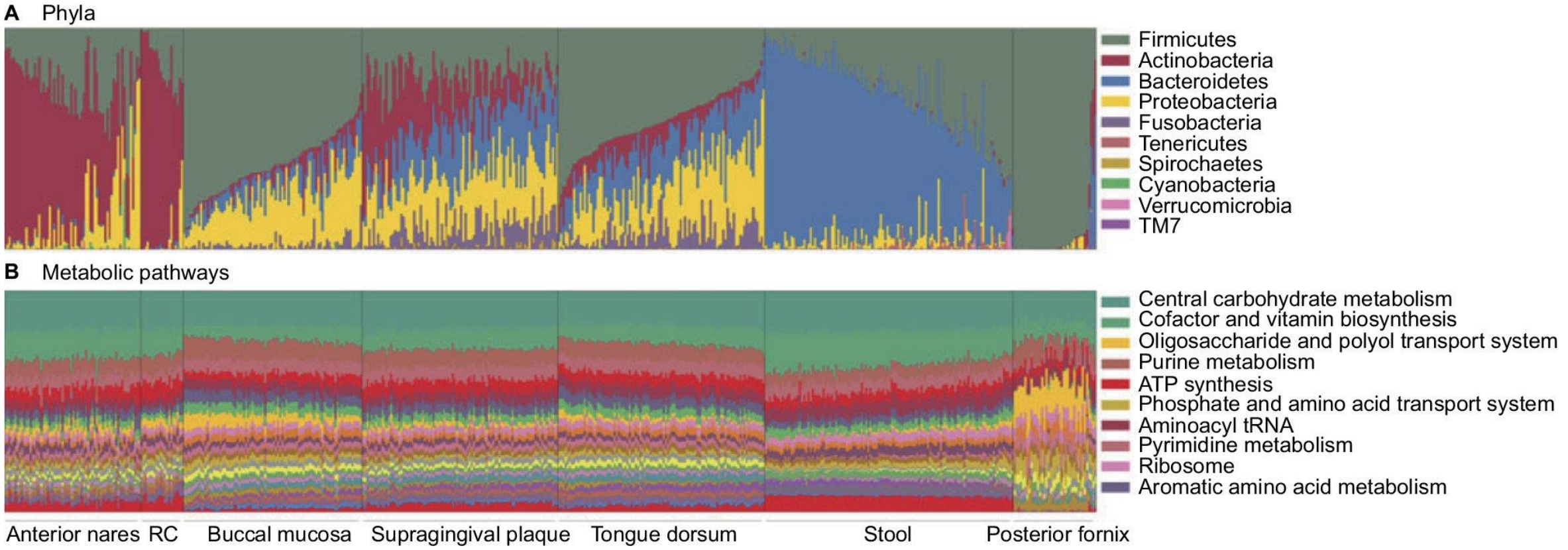
- **Diversitate taxonomica**
  - Ce comunitati microbiene exista in proba?
  - Operational Taxonomic Units (OTUs).
  - Diversitate microbiana
- **Metagenomica functionala**
  - Functiile biologice indeplinite de microorganismele identificate.



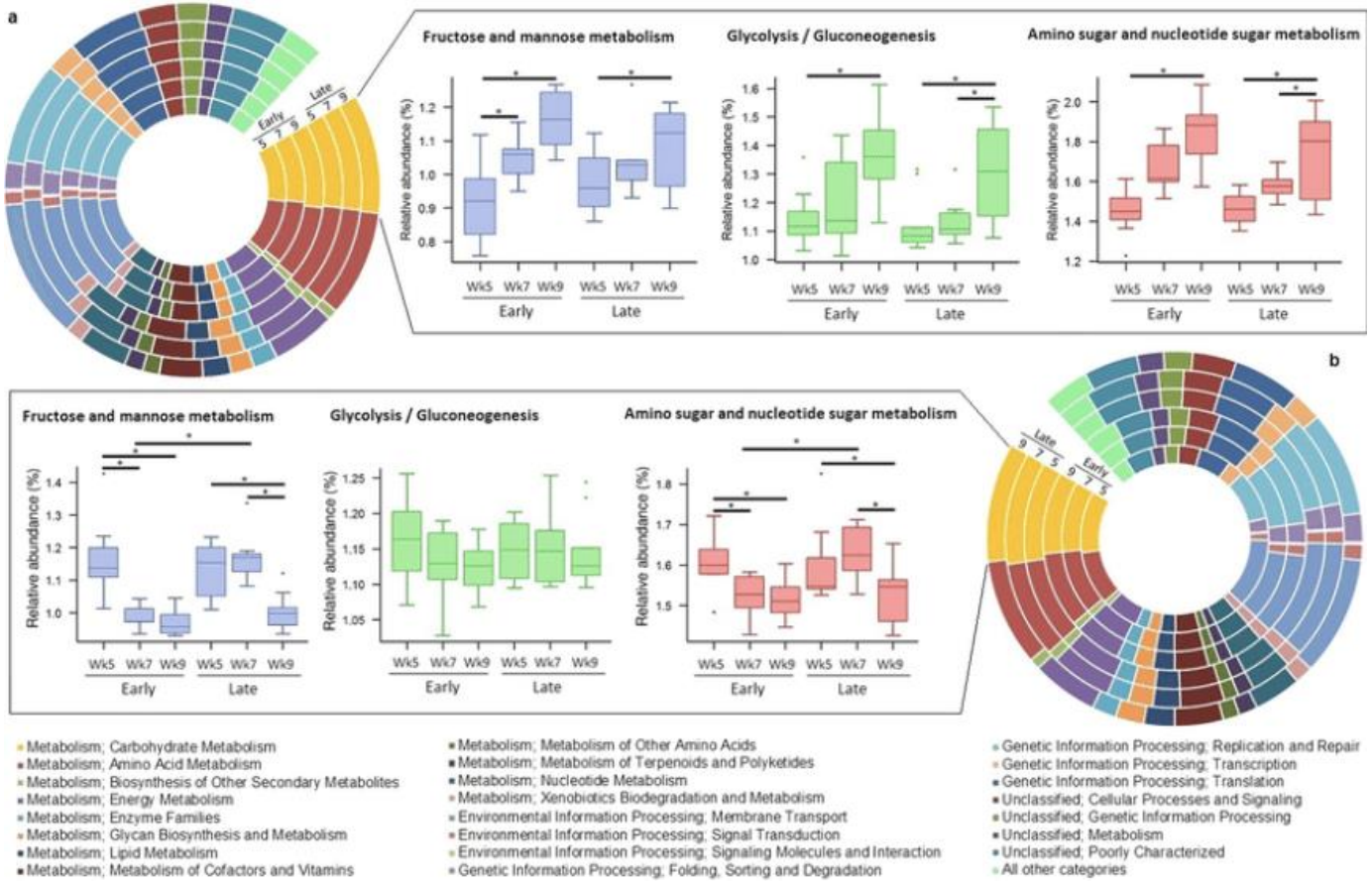
Morgan XC, Huttenhower C (2012) *PLoS Computational Biology e1002808*

Taxonomie vs. profil metabolic

A. Taxonomie –cien e acolo-20 USD
B. Functii biologice– ce fac? -300 USD



Predictia functiei pe baza informatiilor despre OTU



PICRUSt- Phylogenetic Investigation of Communities by Reconstruction of Unobserved States.

Tax4Fun -tool based on linking the 16S ribosomal RNA genes from all KEGG organisms with 16S rRNA gene sequences found in the SILVA ribosomal RNA database.

PAPRICA- a metagenome prediction tool based on placing input 16S rRNA gene sequences into a known phylogenetic tree based corresponding to reference genomes.

Metagenomica vs. Metatranscriptomica

MICROBIOME

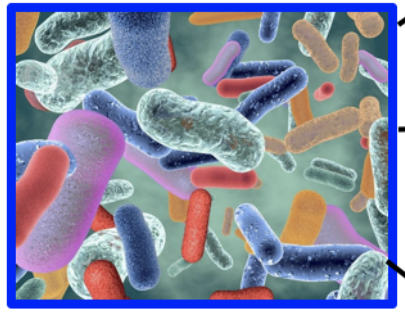
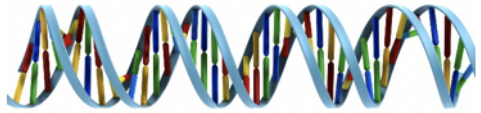


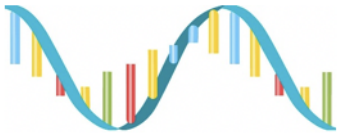
Photo Credit: Getty Royalty Free



METAGENOMICS

TAXONOMY

function



METATRANSCRIPTOMICS

TAXONOMY

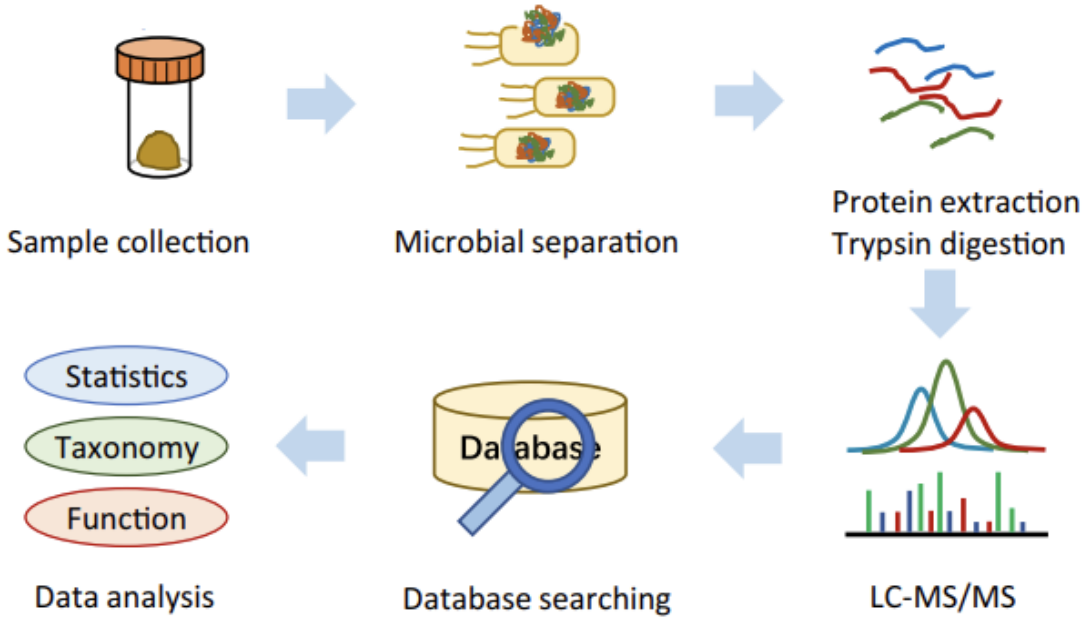
function

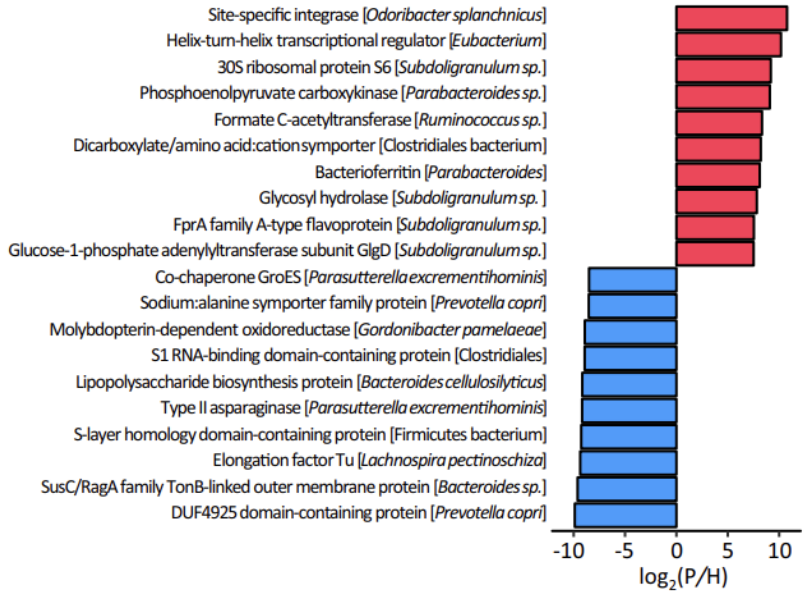
METAPROTEOMICS

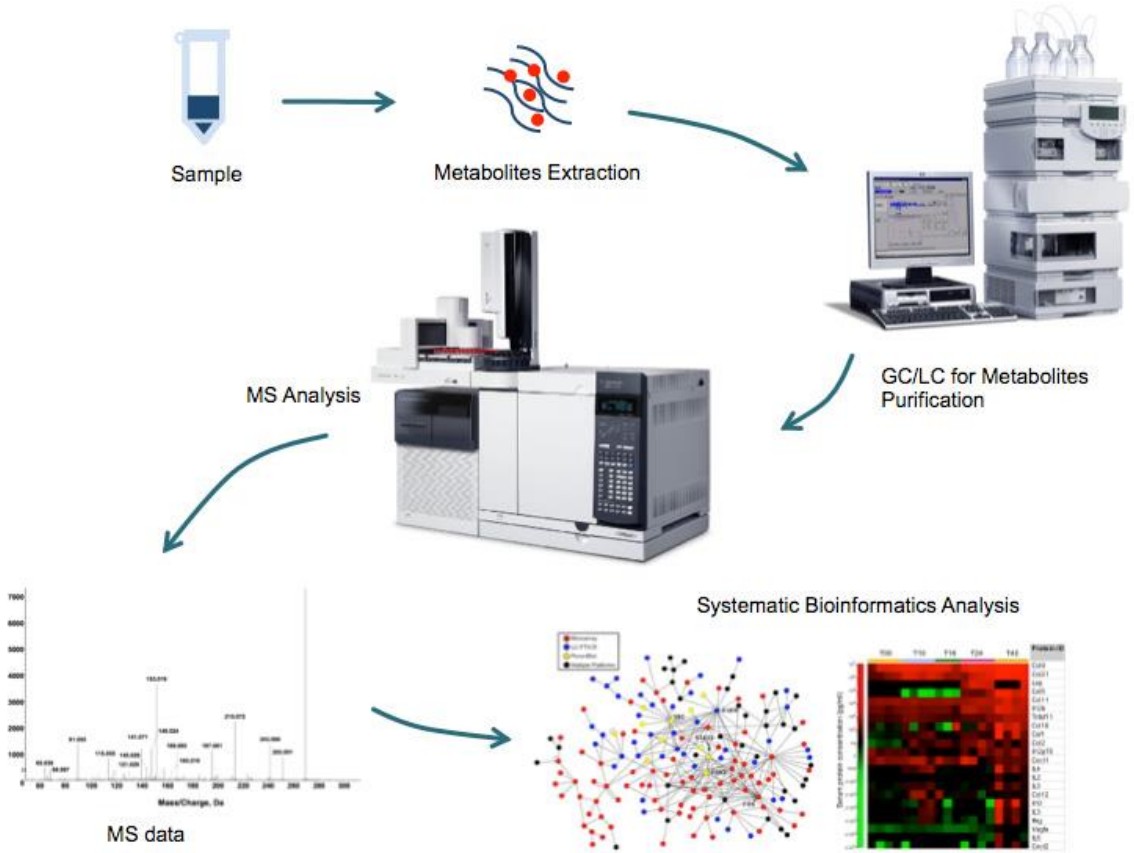
TAXONOMY

FUNCTION

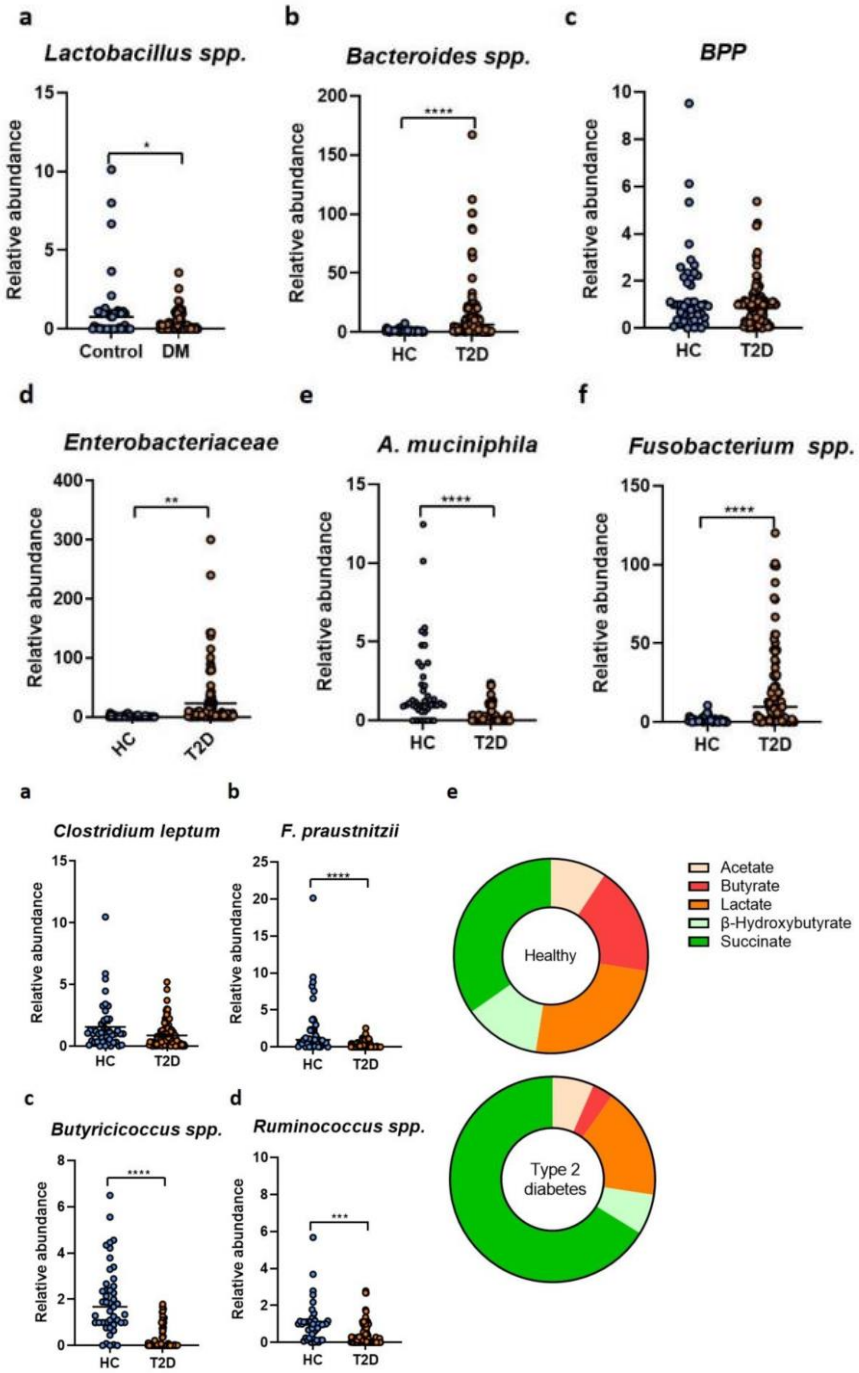
Metaproteomica & Metabolomica - workflow

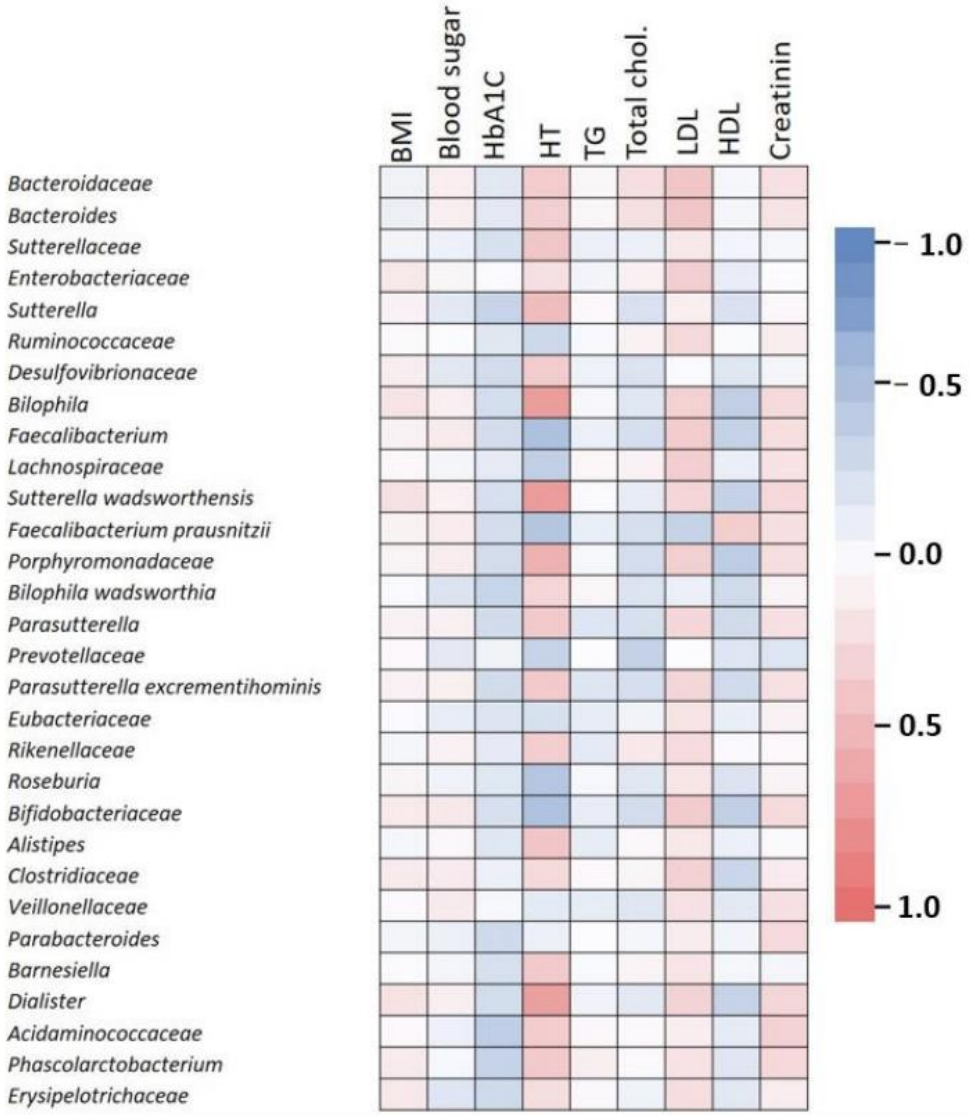






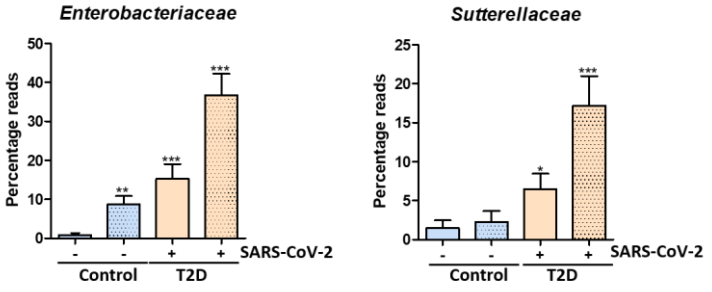
Microbiom & Metabolom in diabetul zaharat de tip 2

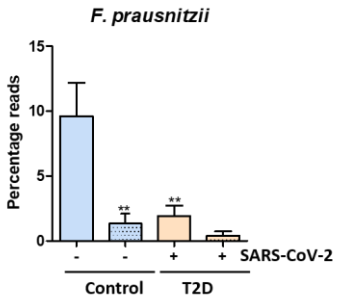


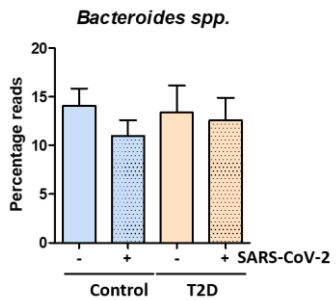


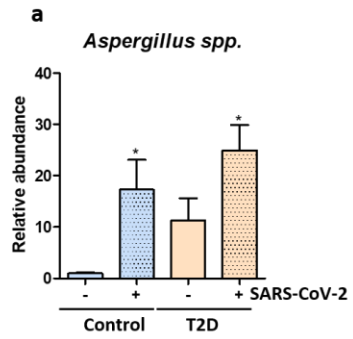
Gradisteanu Pircalabioru et al, 2022

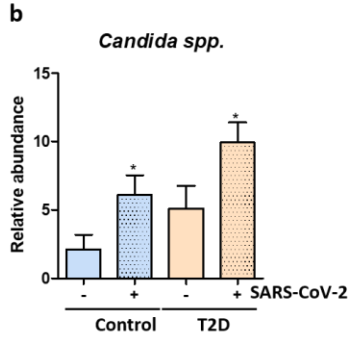
Microbiom in diabetul zaharat de tip 2 si COVID-19

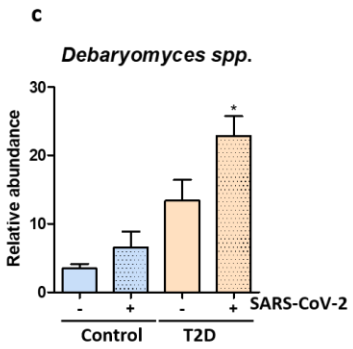


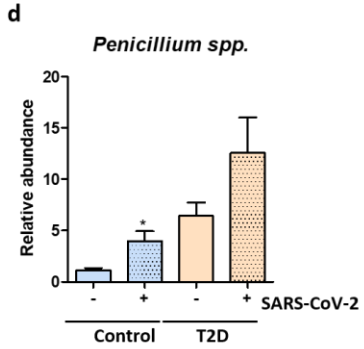


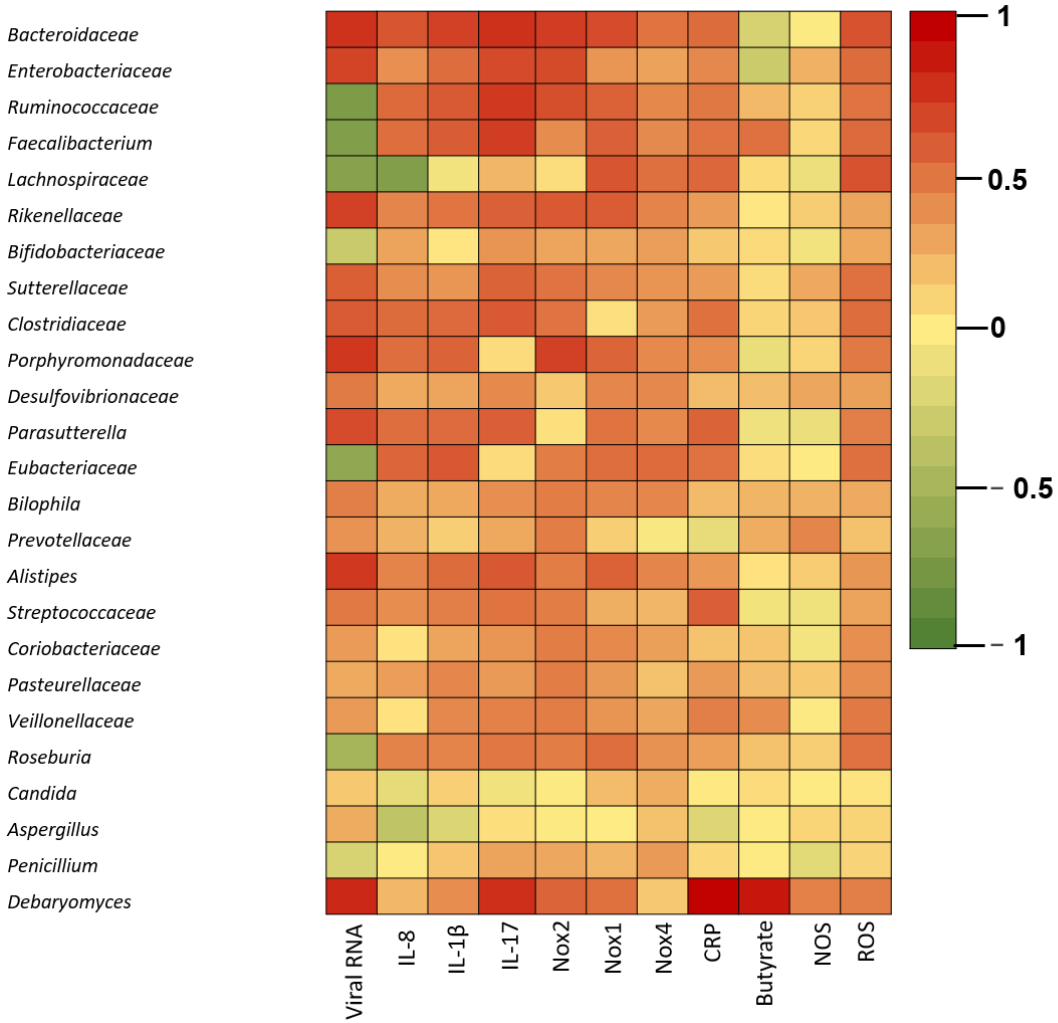






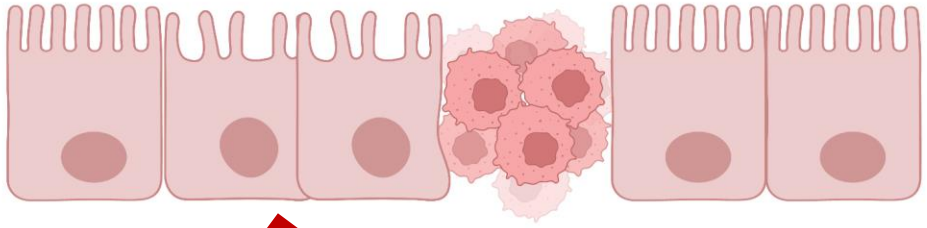




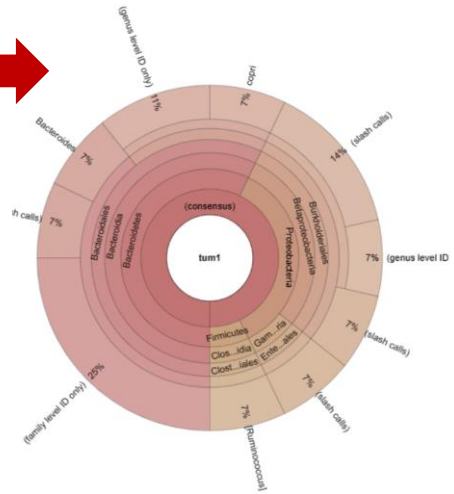


Gradisteanu Pircalabioru et al, 2023

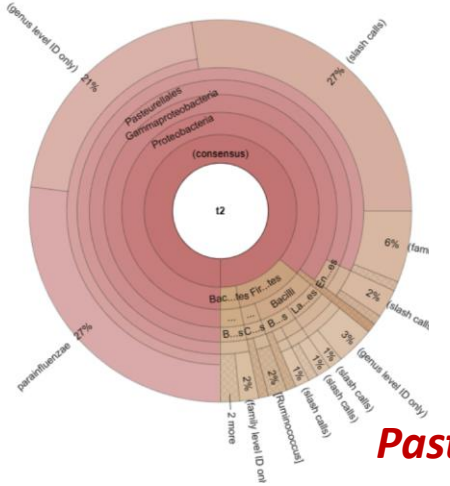
Microbiomul in cancerul colorectal

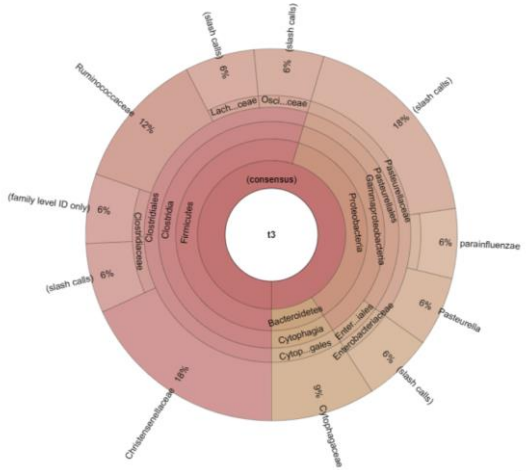


Expresie genica
Stres oxidativ

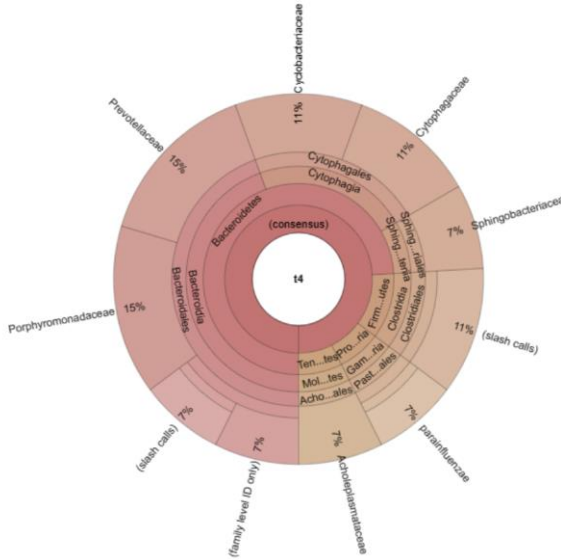


Bacteroides spp.

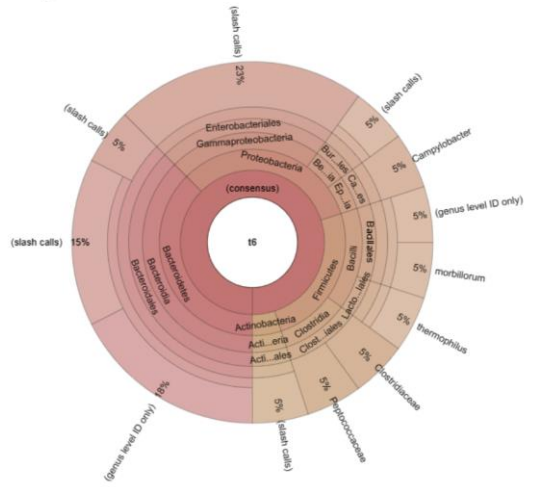




Pasteurellaceae

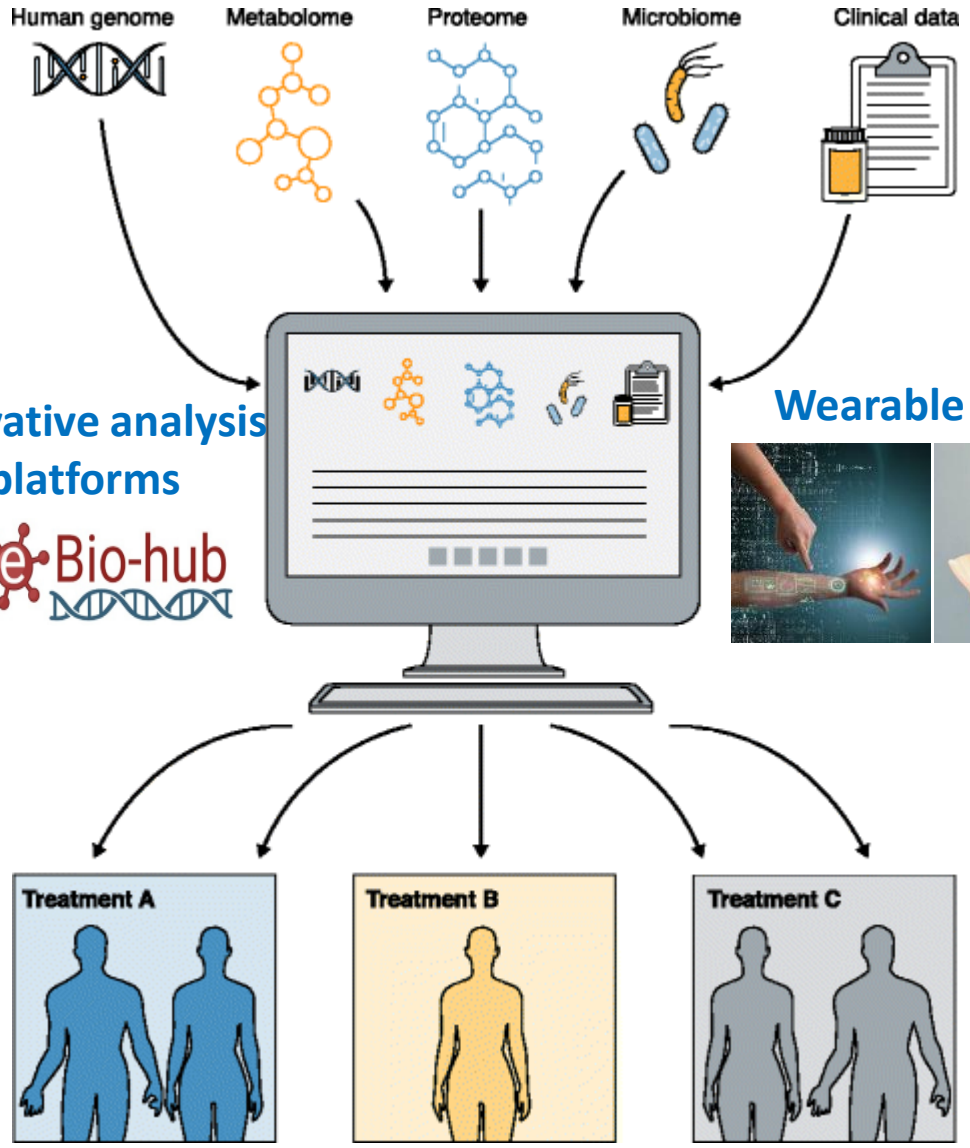


Cytophaga spp.



Enterobacteriaceae

Diagnostic molecular si medicina personalizata



- Detectie simultana de microorganisme, metaboliti
- Standardizare si automatizare a metodelor
- *Money, money, money*
- Considerente etice

Multumesc!

Prof. Dr. Carmen Chifiriuc
Dr. Ciprian Iliescu
Prof. Dr. Horia Iovu
Dr. Mădălina Mușat
Dr. Iulia Ilie
Dr. Anca Pantea-Stoian
Dr. Octavian Savu
Prof. Dr. Marieta Costache
Dr. Nicolae Corcionivoschi
Dr. Ozan Gundogdu

Project financed from the Horizon Europe Work Program 2021-2027,
Grant agreement 101087007-eBio-hub — HORIZON WIDERA-2022-TALENTS-01