



Genomics and Epidemiology for Gastric Adenocarcinomas (GE4GAC)

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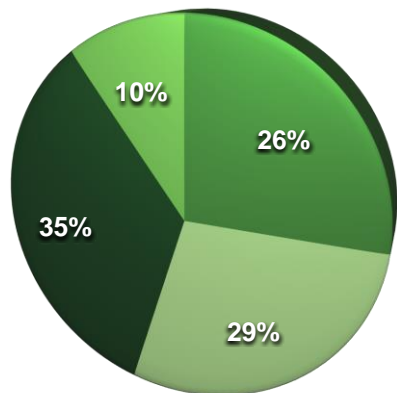


WHY GASTRIC CANCER?

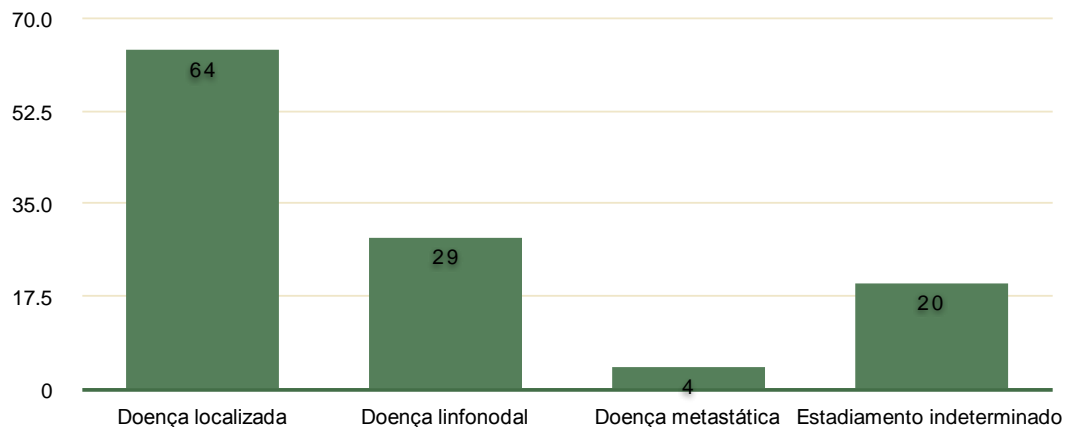
- 4th most common malignancy in the world
- 2nd most common cause of cancer-related death
- Important geographic variation:

High-risk areas: Japan, Korea, Latin America (Brazil, Peru), Russia

Low-risk areas: USA, Israel, Kuwait, Canada, UK



- Doença localizada
- Doença linfonodal
- Doença metastática
- Estadiamento indeterminado



In Brazil (INCa, 2016)

- 3rd more incident tumor in men
- 5th more incident tumor in women

- 20.5k new cases/year
- 14.2k deaths/year

Overall 5-year survival rates (all stages) (Jemal et al., 2010; Theuer et al., 2000)

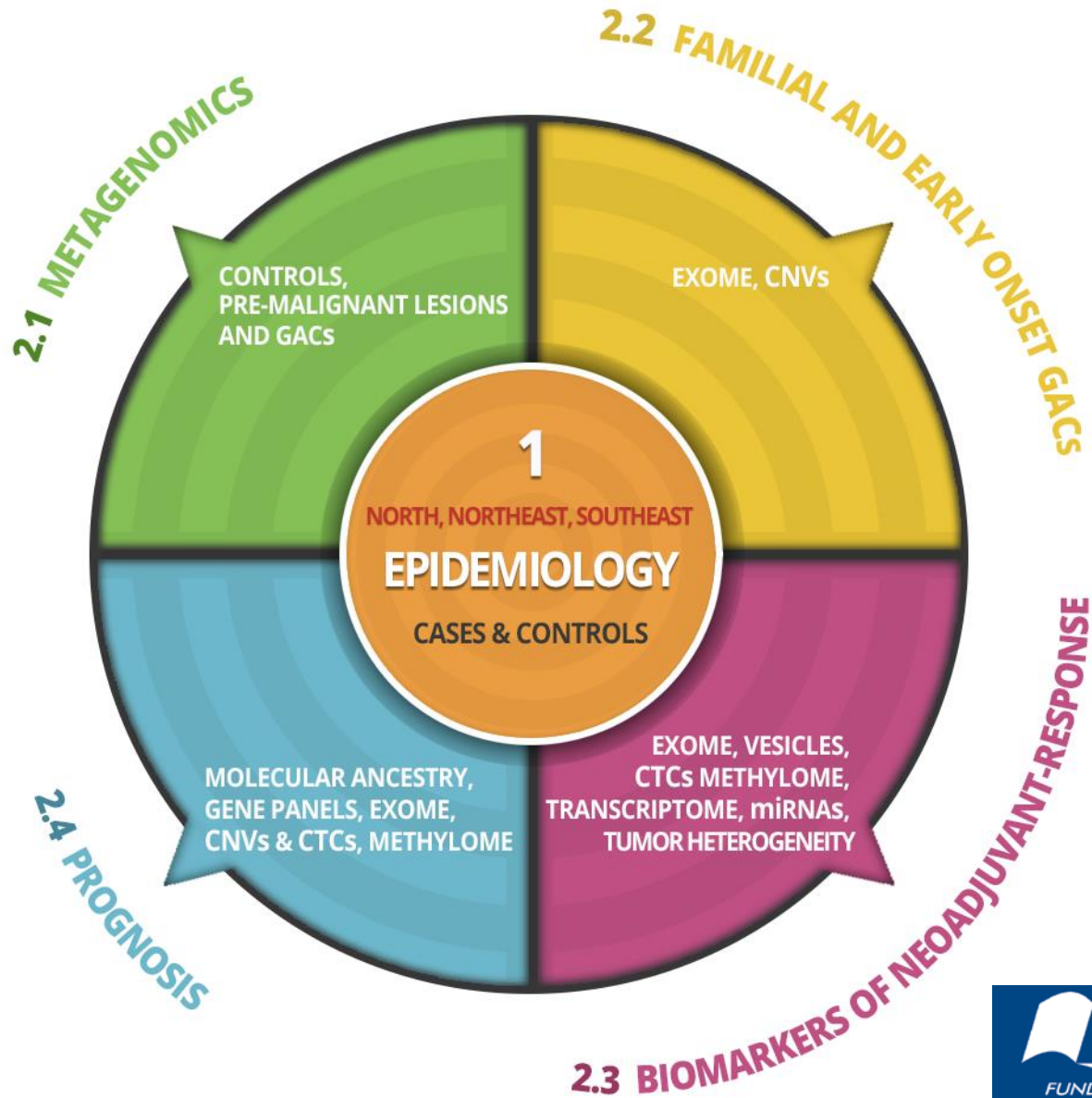
- ◆ 40-60% in Japan
- ◆ 15-30% in the USA

-Ethnicity and treatment response (Kim et al., 2010):

Group	Chemotherapy + Bevacizumab	Chemotherapy + placebo
Asians	13.9	12.1
Non-asians	11.5	6.5*

*p<0.01

Message: the disease is different according to the ethnical background!



Module 1 - Epidemiology

- To collect epidemiologic information from ~2,000 individuals:
 - Control 1** – Cancer prevention program: no cancer and no gastric complains.
 - Control 2** – Endoscopic controls (some gastric issues, no cancer)
 - Cases** – diagnosis of gastric adenocarcinoma
- Detailed socio-demographic information
- Data regarding diet, ethanol & tobacco consumption
- Use of drugs such as aspirin, anti-inflammatory agents, antibiotics, proton-pump inhibitors, sweeteners
- Brazil: Southeast, North, Northeast
- Peru?

Dynamic alterations in gastric cancer...

Intestinal type: ~54%

Presents as “glandular structures” in histology

Higher prevalence of *H. pylori* infections

Declining globally

Male:Female ratio = 2:1

Older patients

Better prognosis

Diffuse type: ~32%

Infiltrative lesions

Loss of *CDH1* (20%)

Male:Female ratio = 1:1

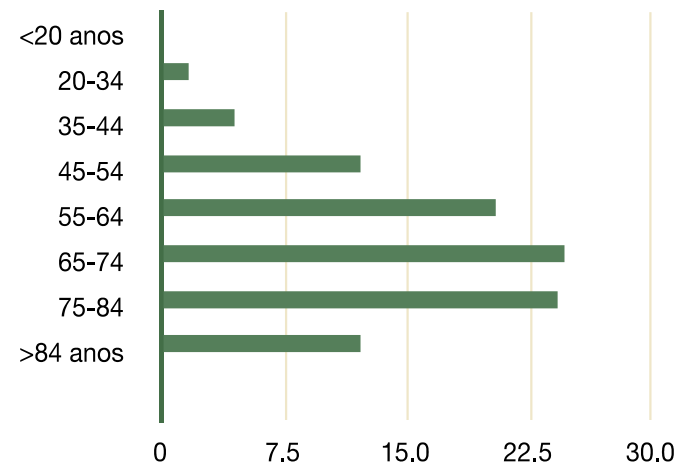
Incidence is not declining so fast (growing?)

Younger age

Worse prognosis

Mixed type: ~14%

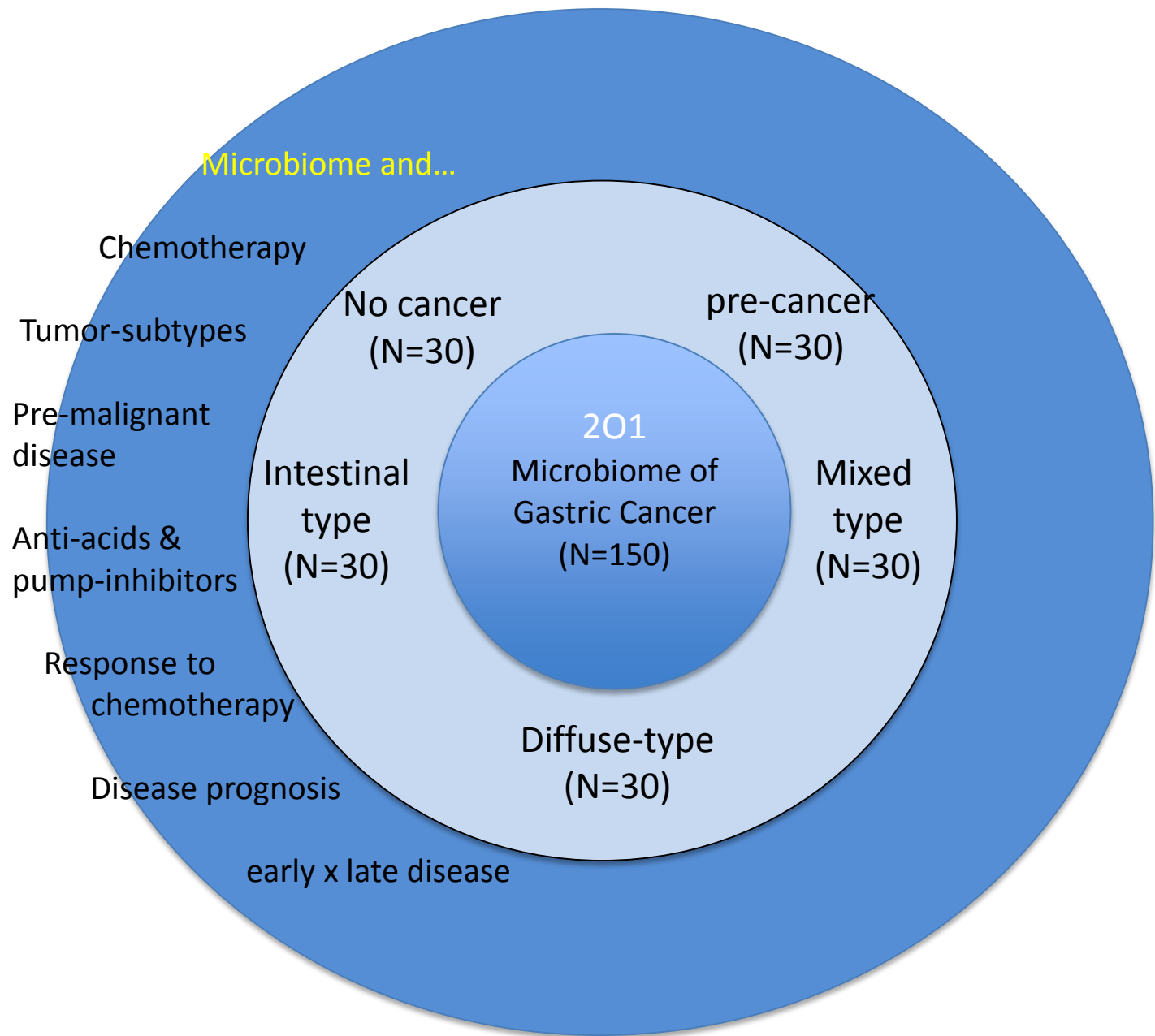
Idade ao diagnóstico	%
<20 anos	0.1
20-34	1.7
35-44	4.5
45-54	12.2
55-64	20.4
65-74	24.7
75-84	24.3
>84 anos	12.2



Module 2 – Molecular Biology

Full support from the epidemiology module

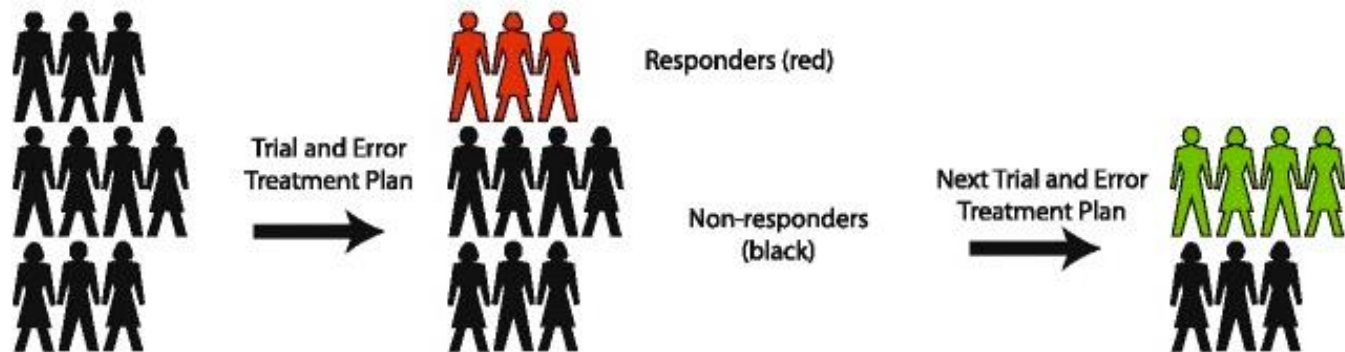
- 2.1 - Microbiome analysis
- 2.2 - Early Onset/Familial Gastric Cancer
- 2.3 - Markers of neoadjuvant chemotherapy response
- 2.4 - Markers of prognosis



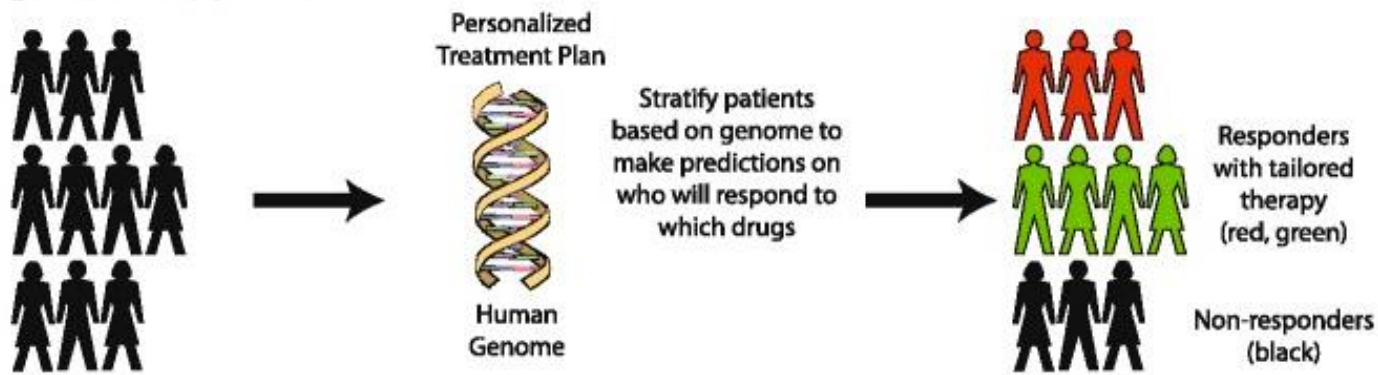
Module 2.1 – Microbiome Analysis

- Gastric juice collected from >150 patients
- Biopsies available for ~90%

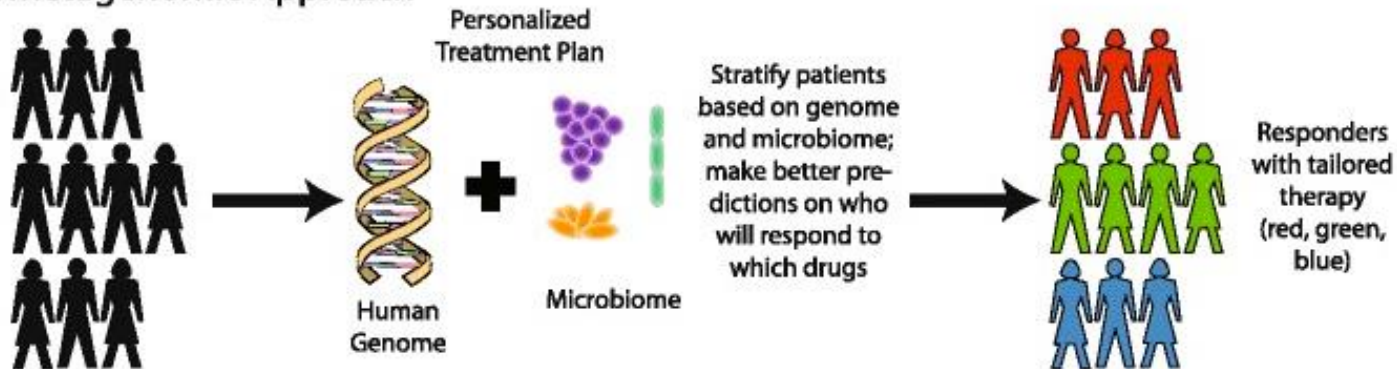
A Trial and Error Approach



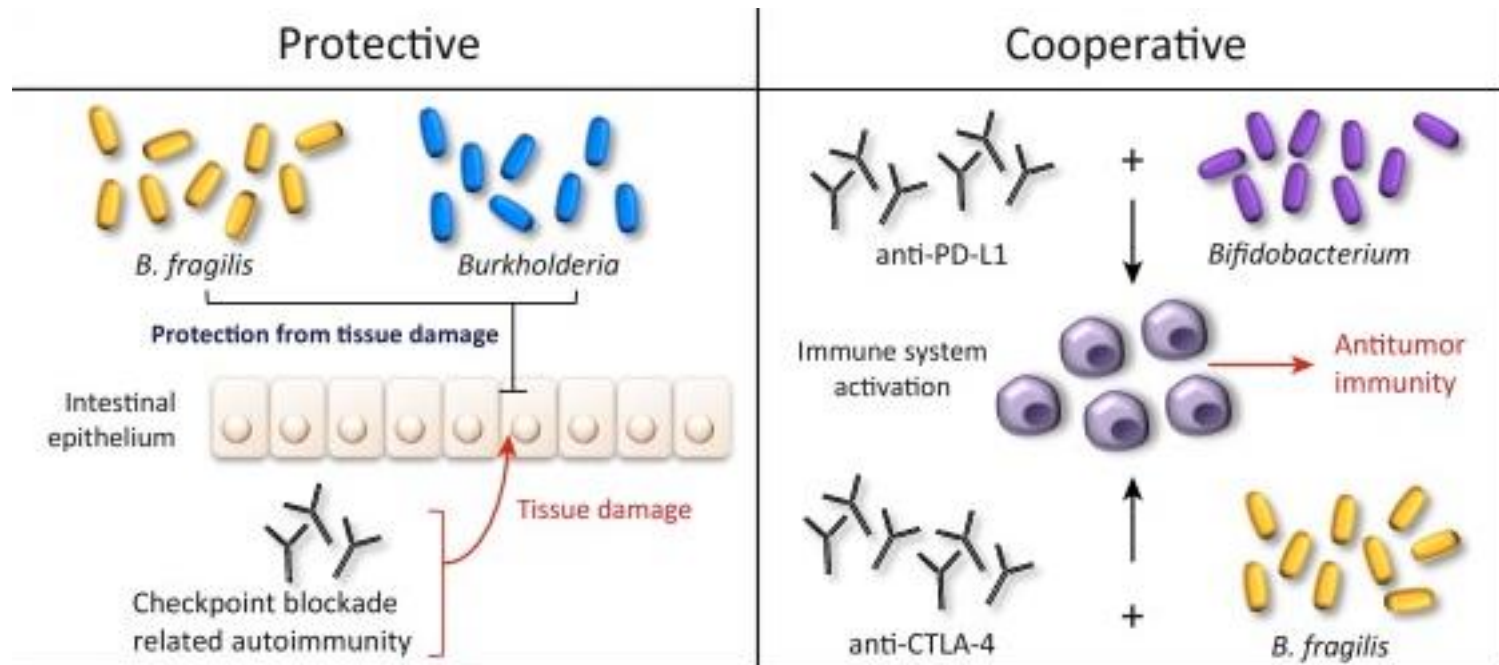
B Pharmacogenomic Approach



C Pharmacometagenomic Approach

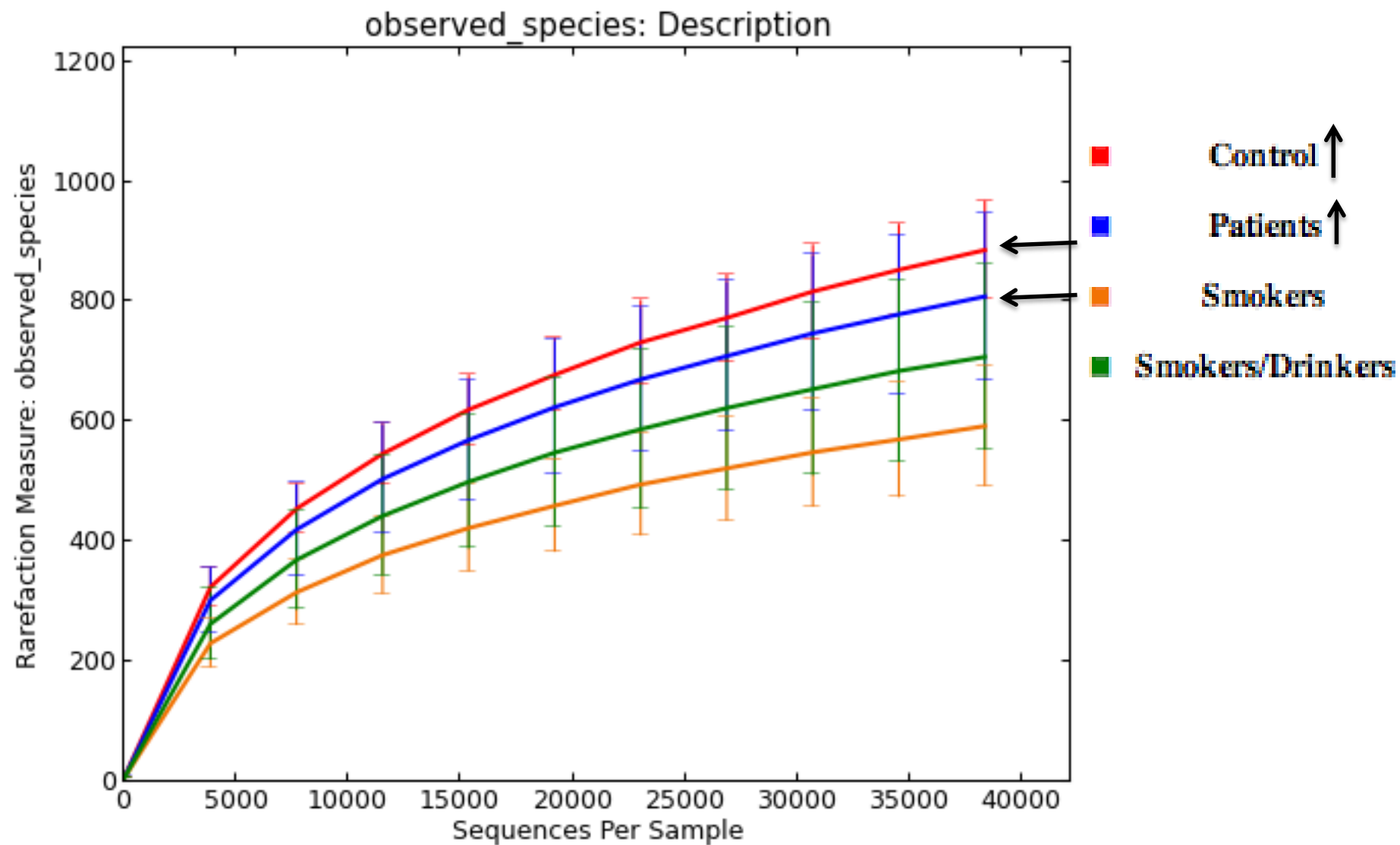


Microbiome and chemotherapy response?



Trends in Immunology

Oral Cancer - Species Richness

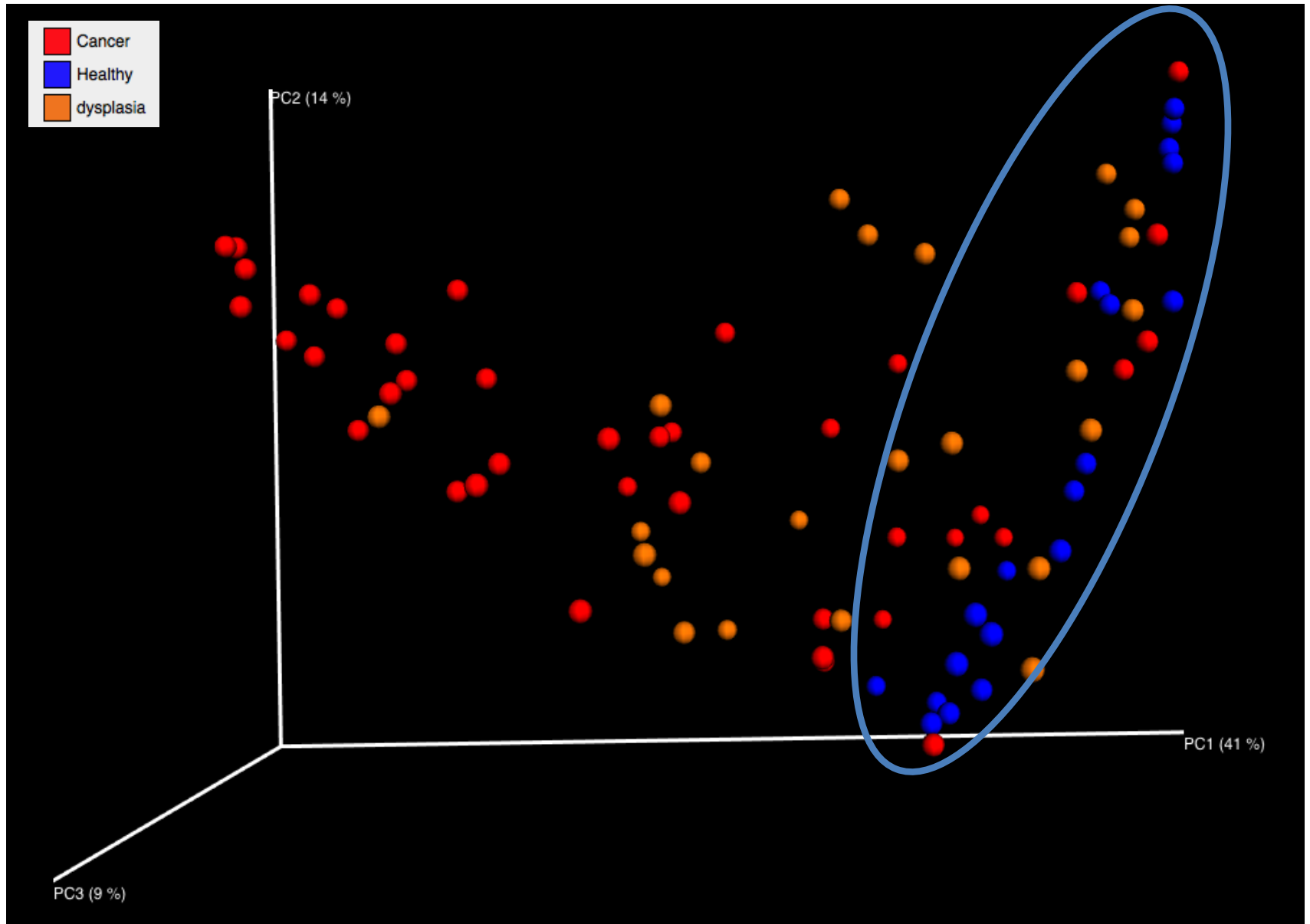


Thomas et al., 2014

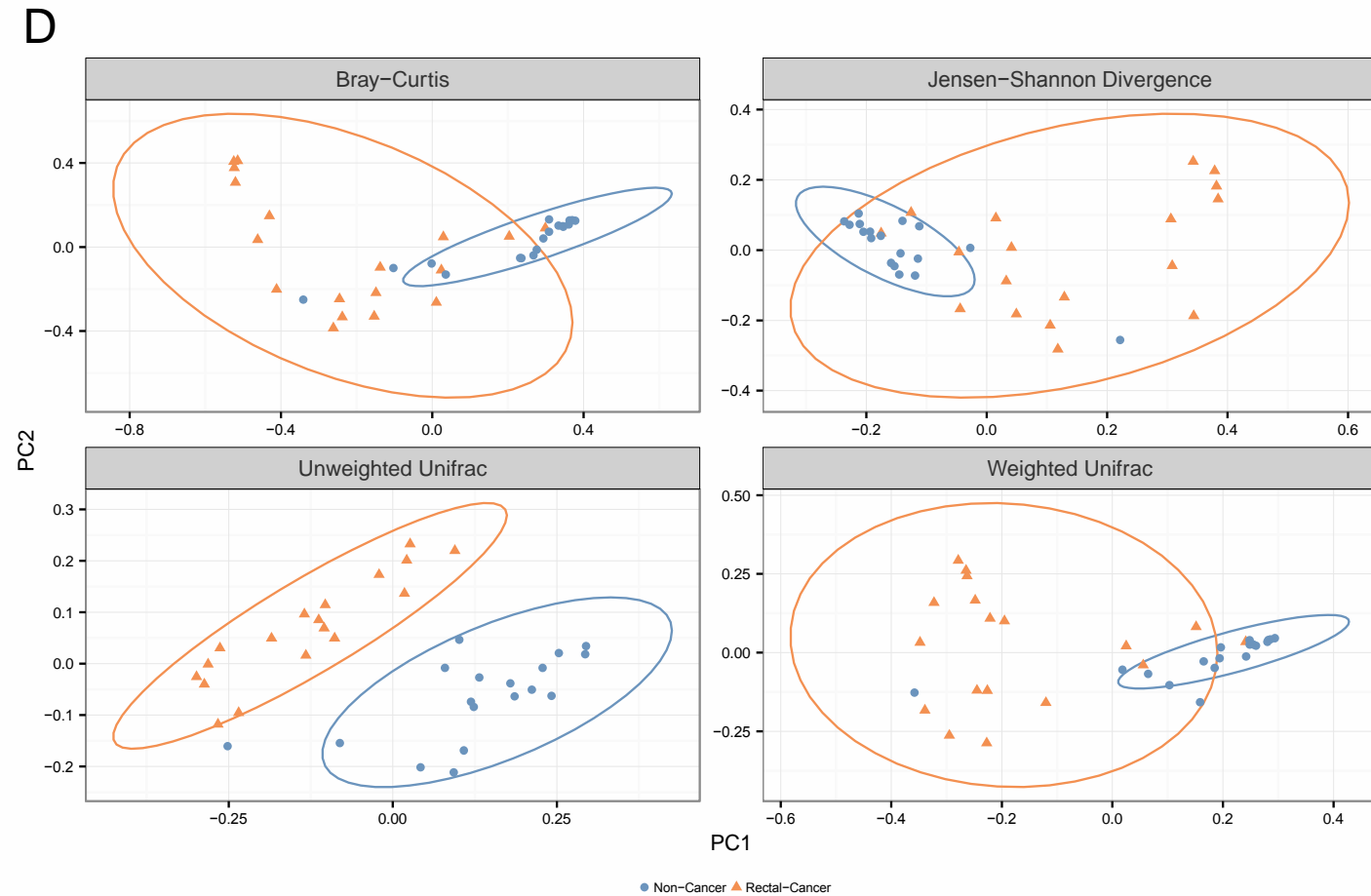
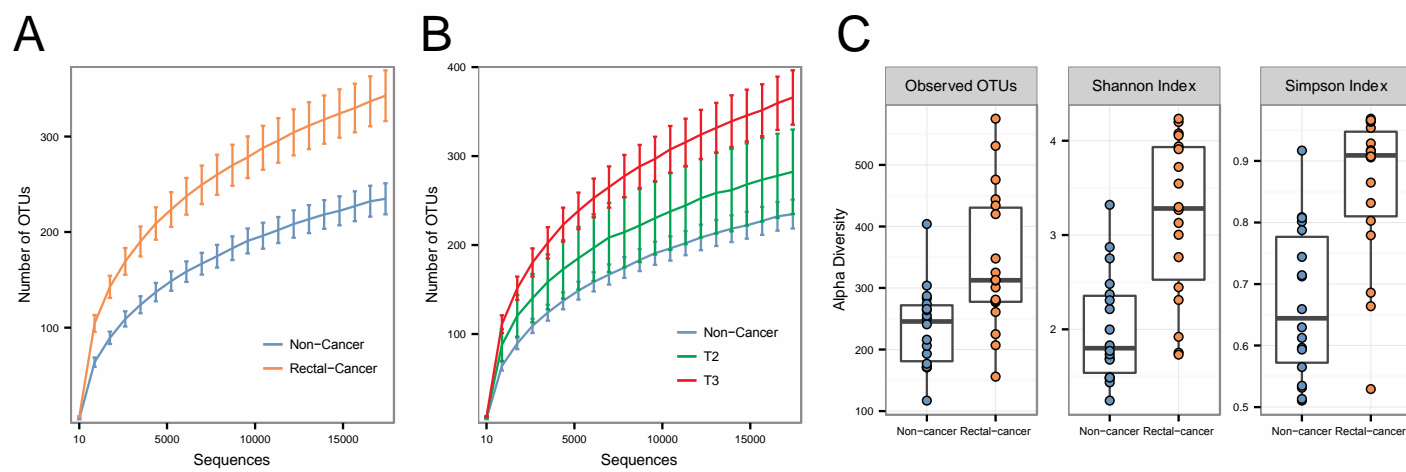
Thomas et al., in preparation

Best predictive genera

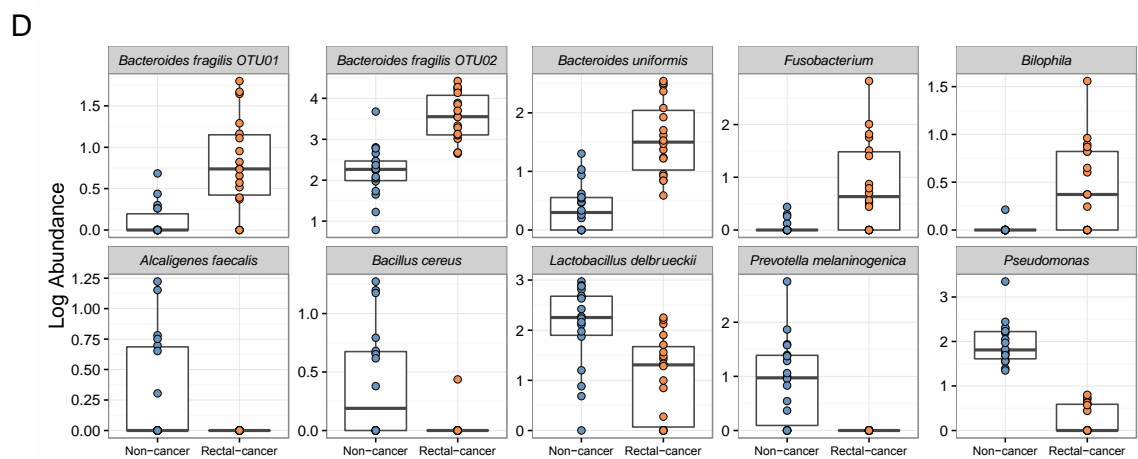
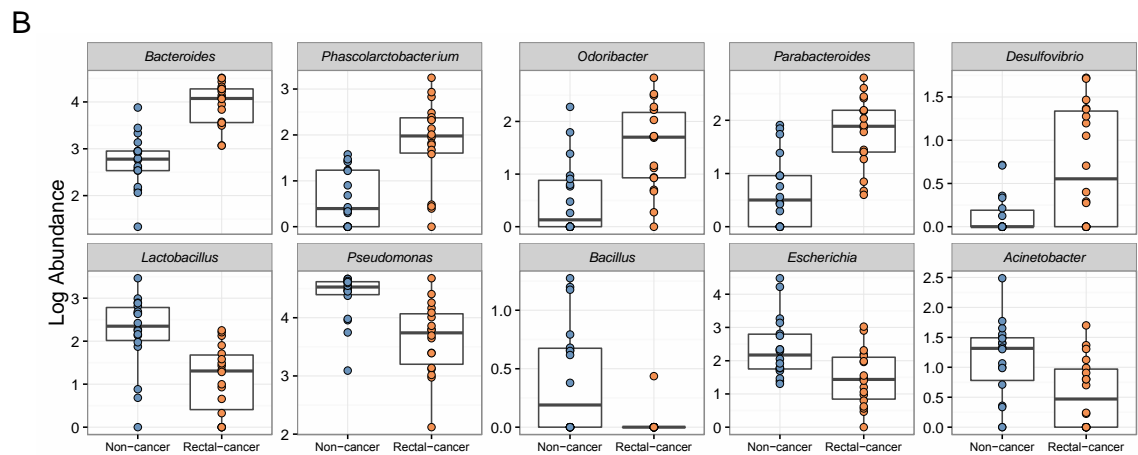
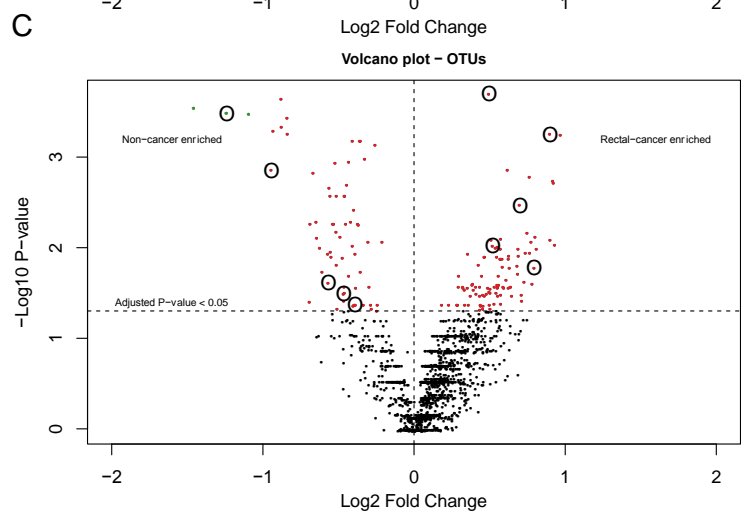
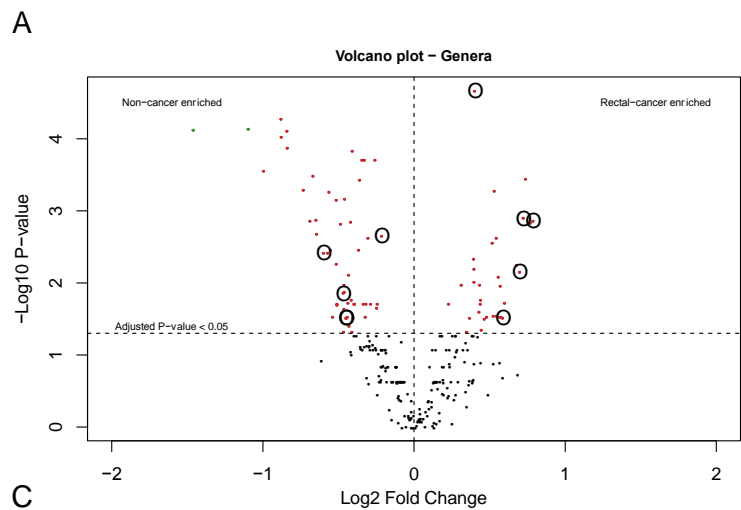
PCoA plot – public dataset

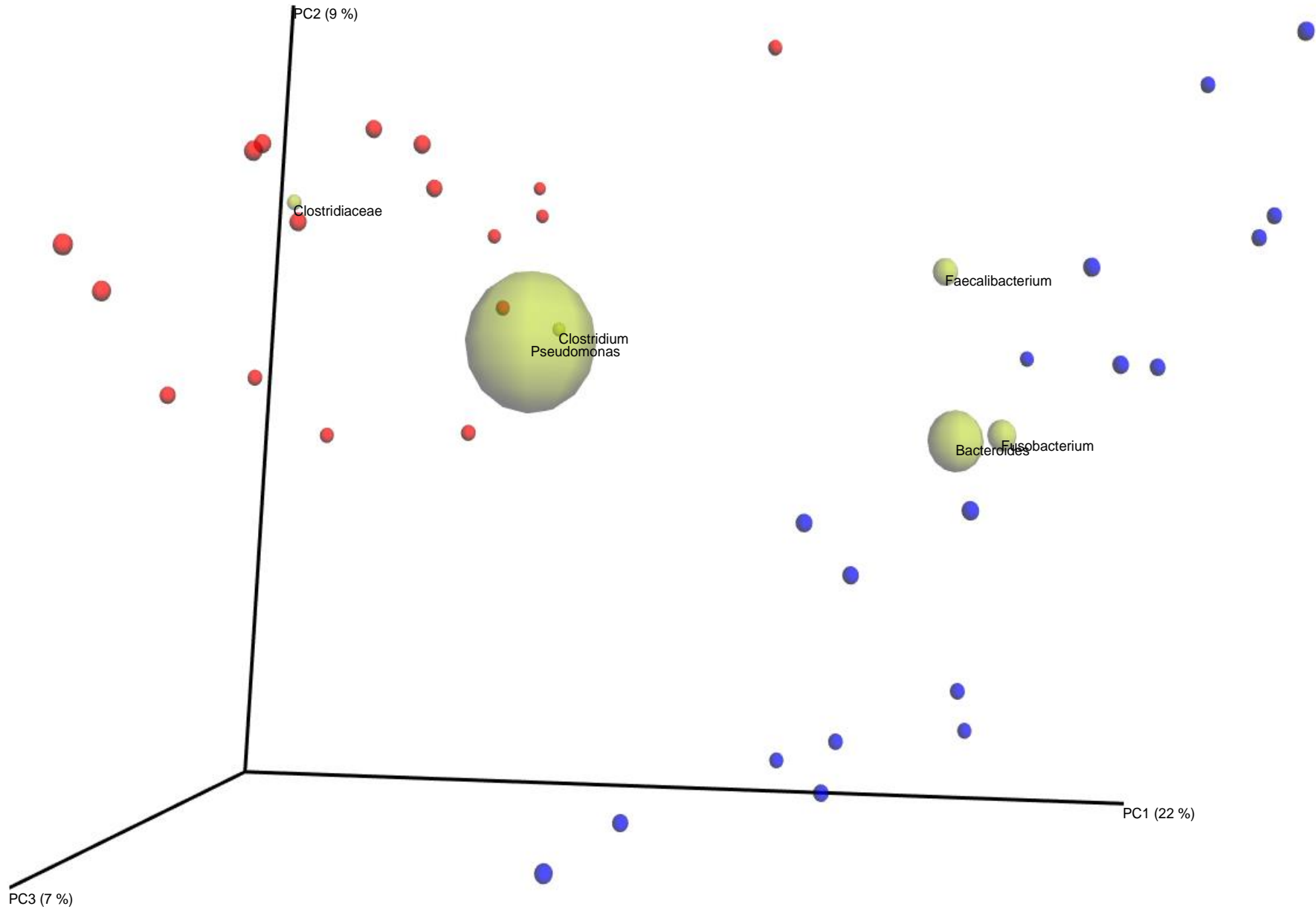


Rectal Cancer

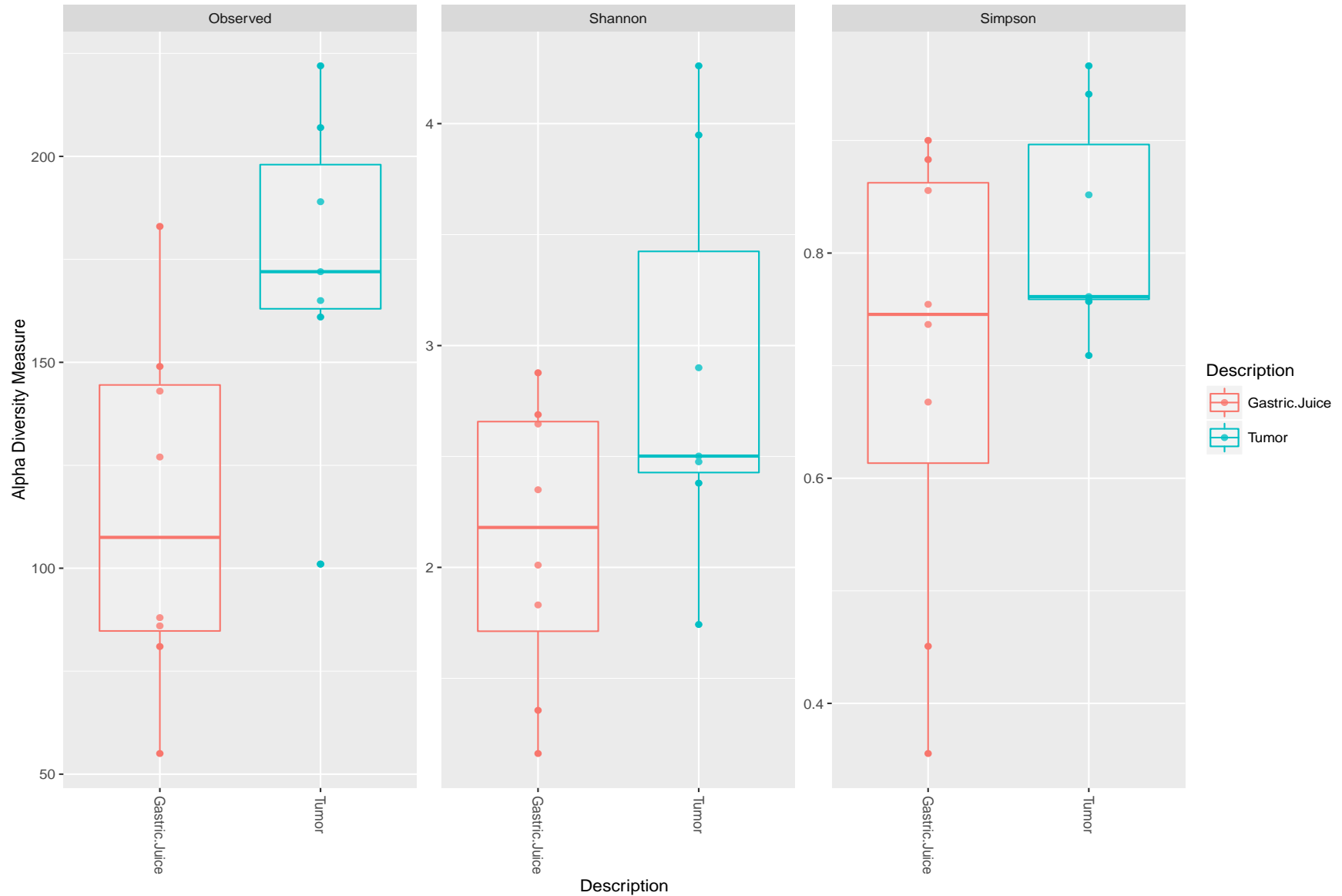


Thomas et al.,
in preparation

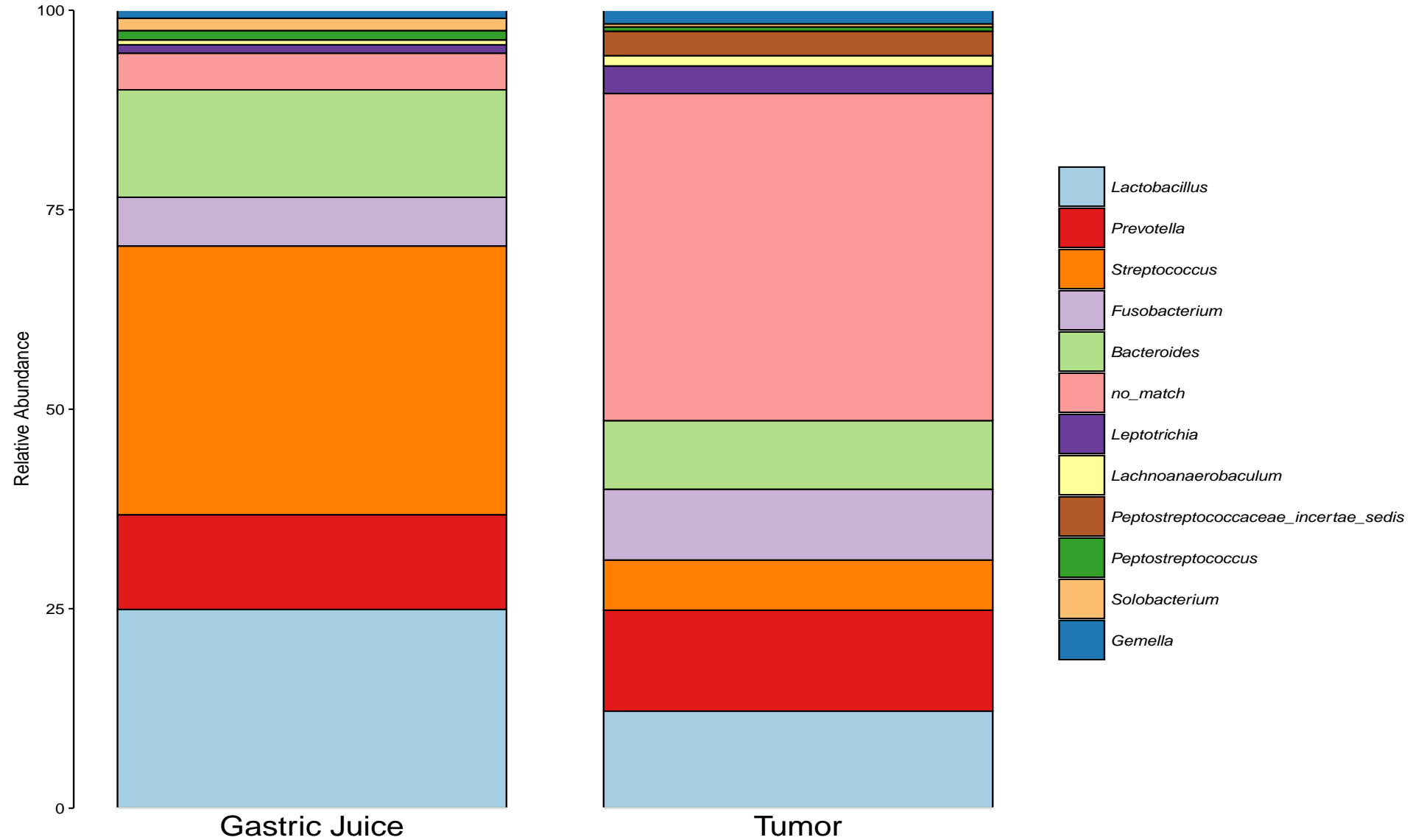




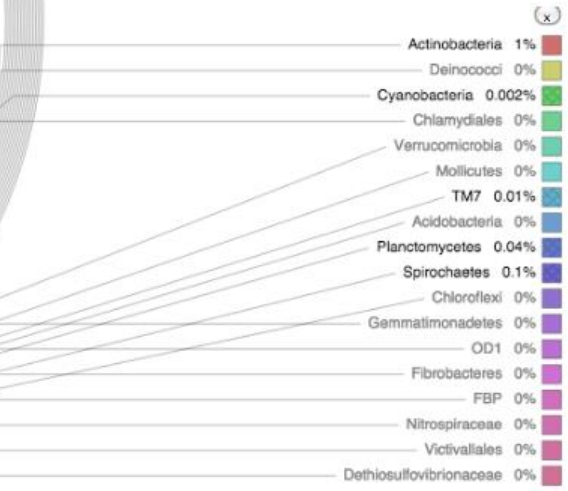
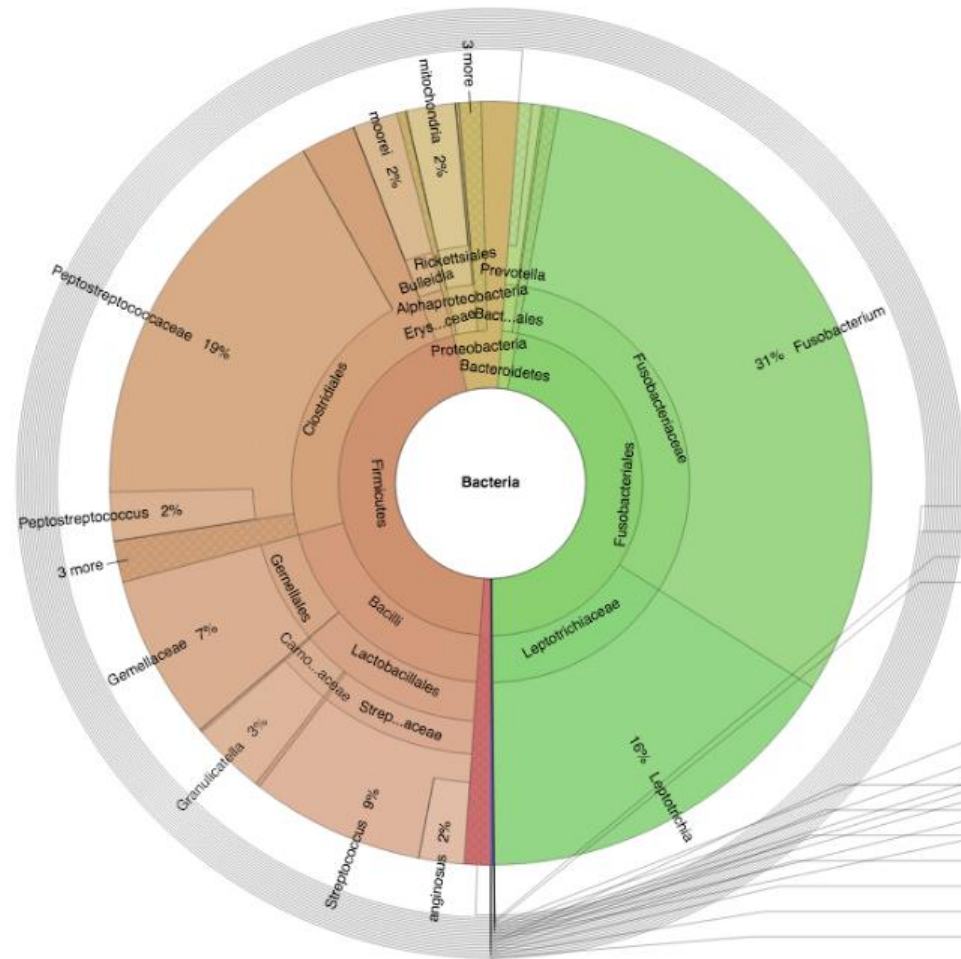
Gastric cancer - bacterial diversity



Abundance of distinct genera

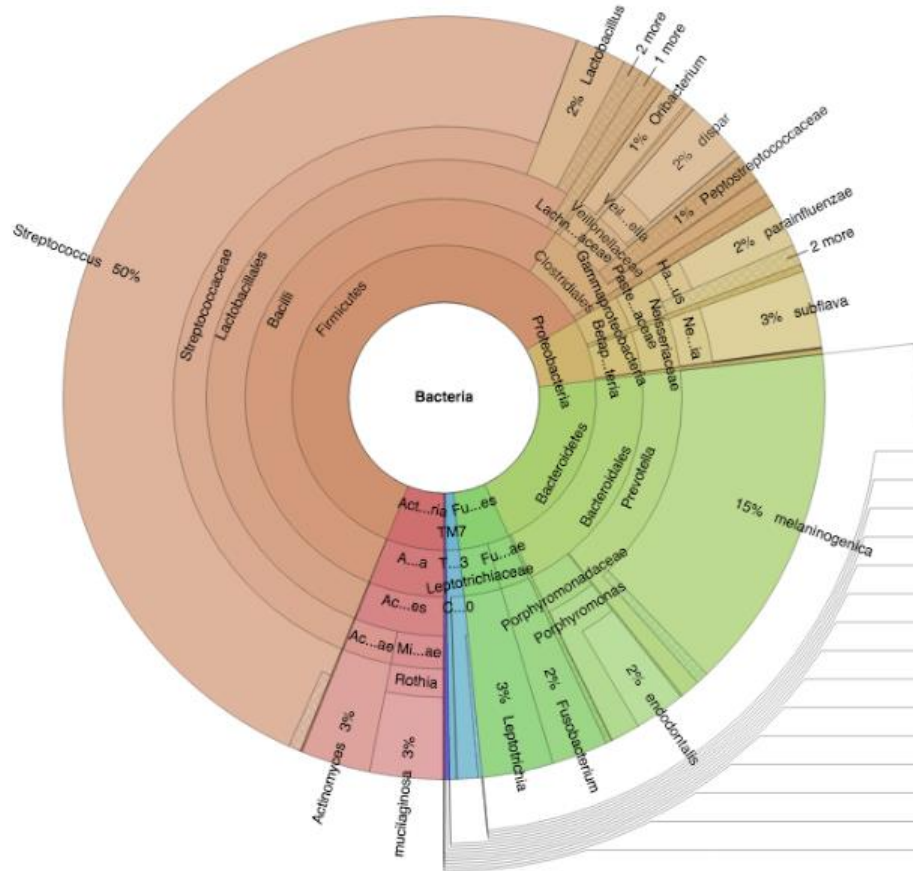


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Metro
 SG13
 SG15
 SG190
 SG6
 T29
 T31
 T33
 T34

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- Deinococci 0%
- Cyanobacteria 0%
- Chlamydiales 0%
- Verrucomicrobia 0%
- Mollicutes 0%
- Acidobacteria 0%
- Planctomycetes 0%
- Spirochaetes 0.2%
- Chloroflexi 0%
- Gemmatimonadetes 0%
- OD1 0%
- Fibrobacteres 0%
- FBP 0%
- Nitrospiraceae 0%
- Victivallales 0%
- Dethiosulfovibrionaceae 0.002%

Shotgun metagenomics?

Microbiome manipulation?

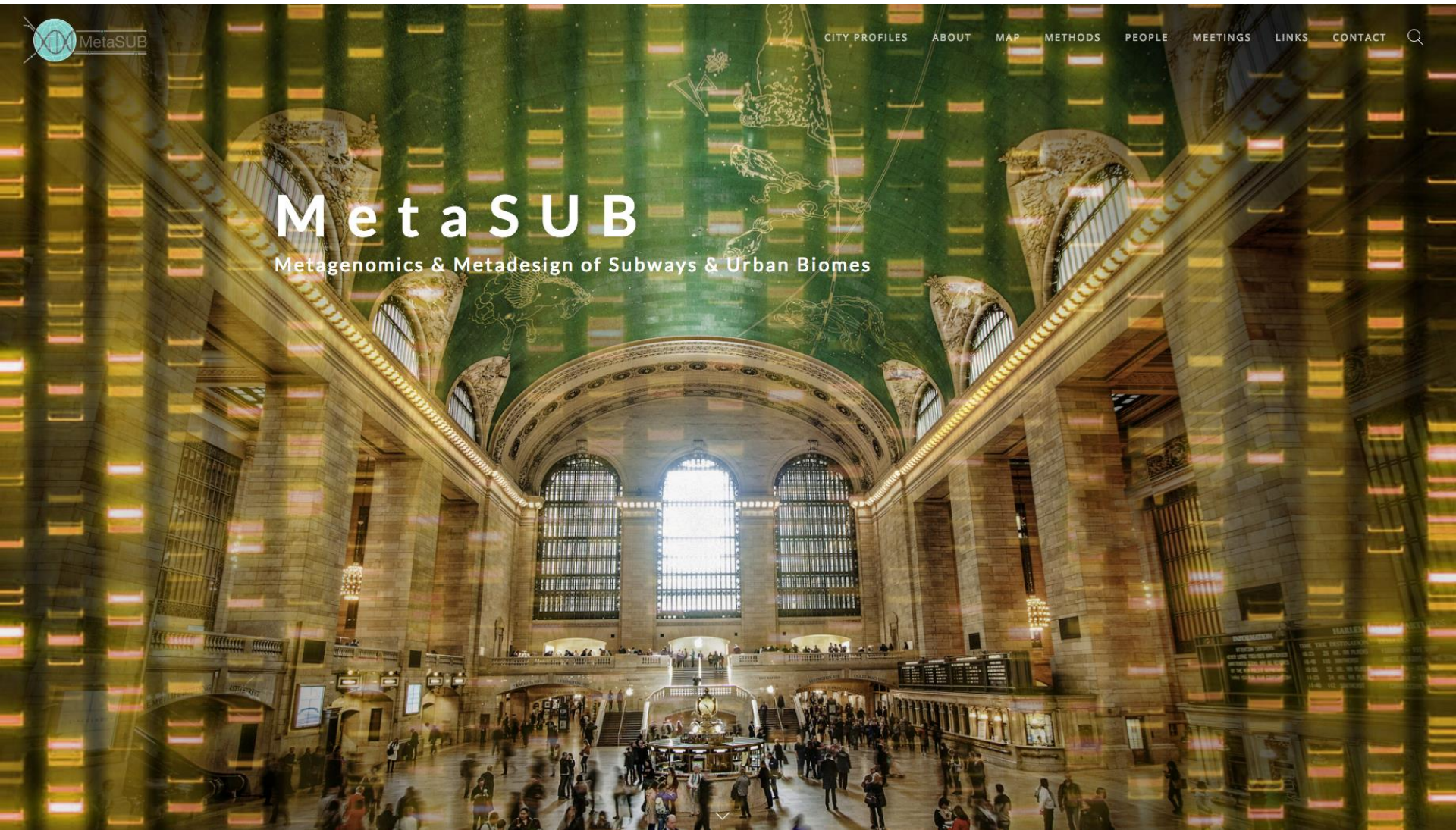
www.metasub.org

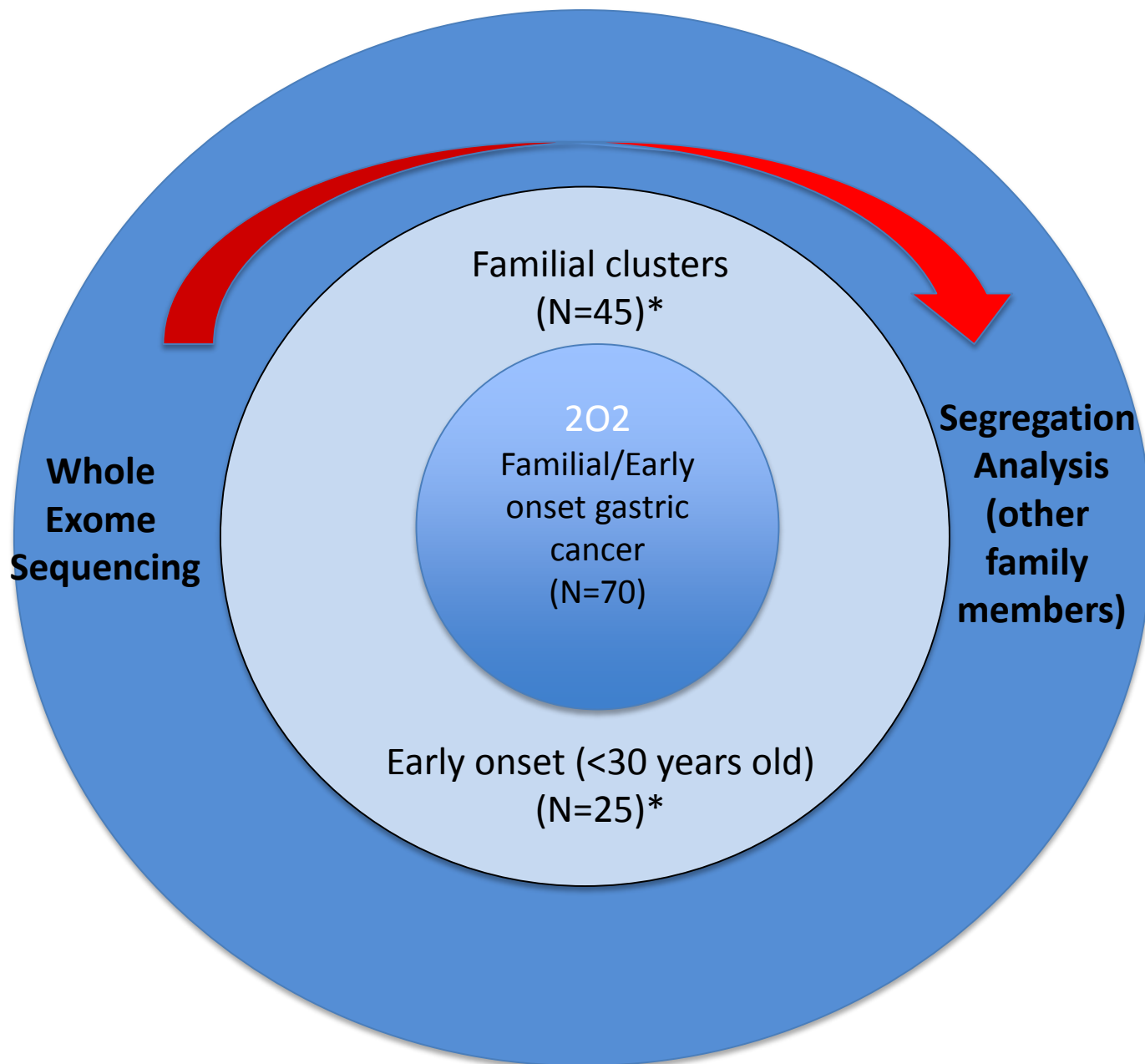


[CITY PROFILES](#) [ABOUT](#) [MAP](#) [METHODS](#) [PEOPLE](#) [MEETINGS](#) [LINKS](#) [CONTACT](#)

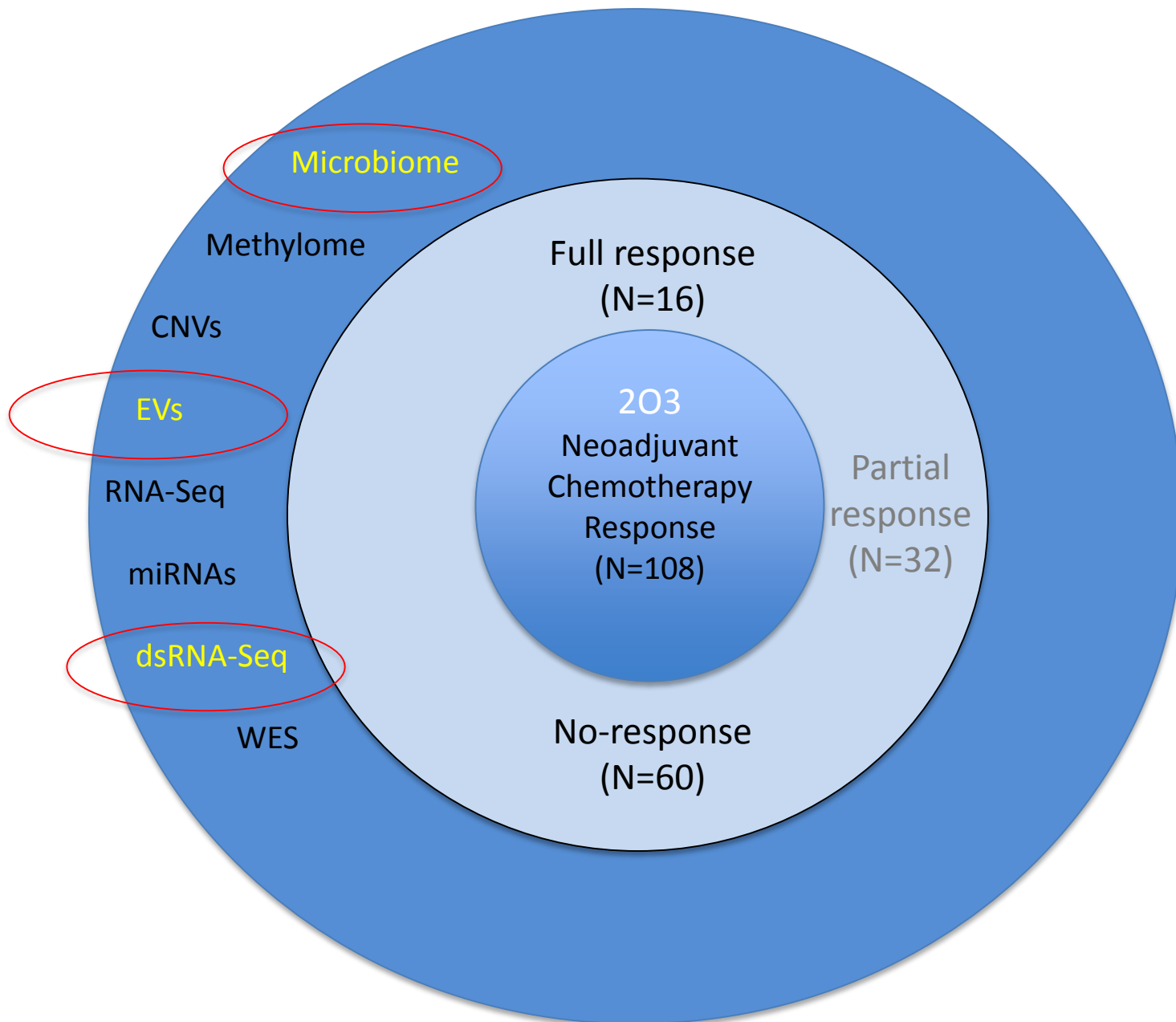
MetaSUB

Metagenomics & Metadesign of Subways & Urban Biomes





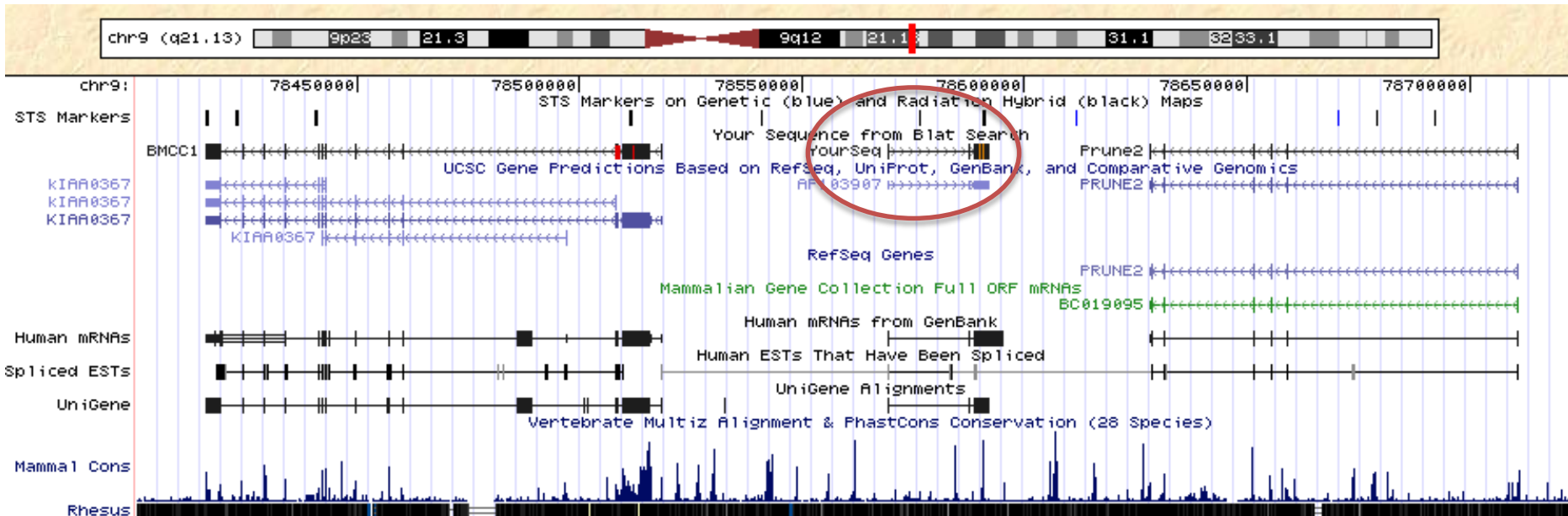
* Negative for *CDH1*



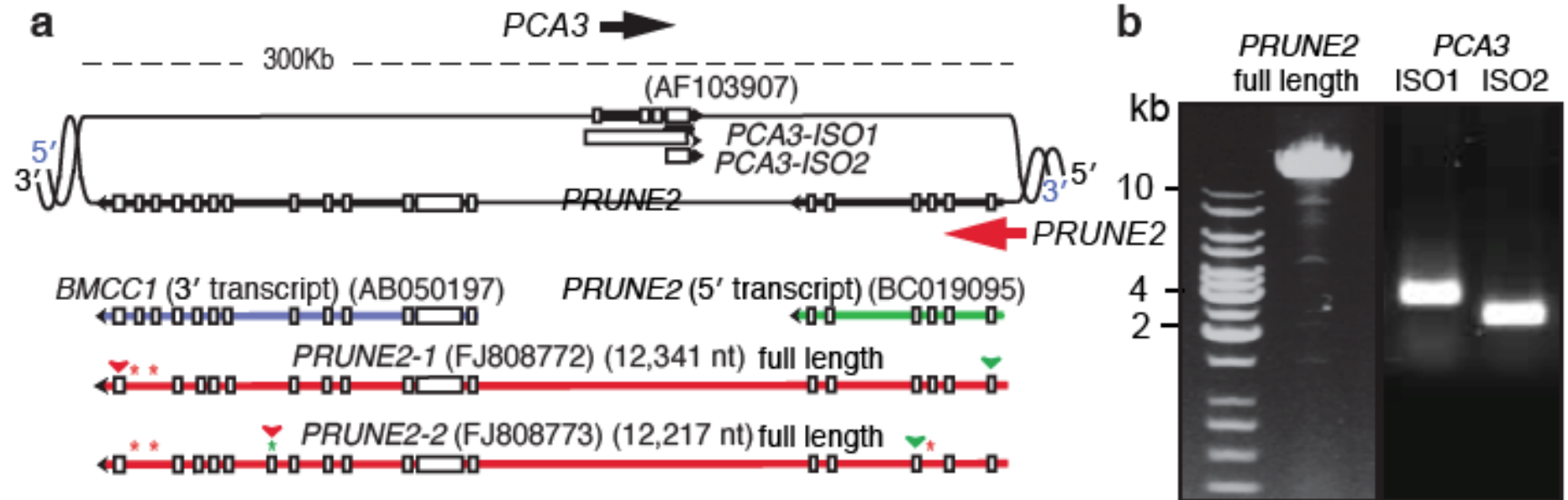
Why should we study the double-stranded transcriptome?

PROGENSA[®]

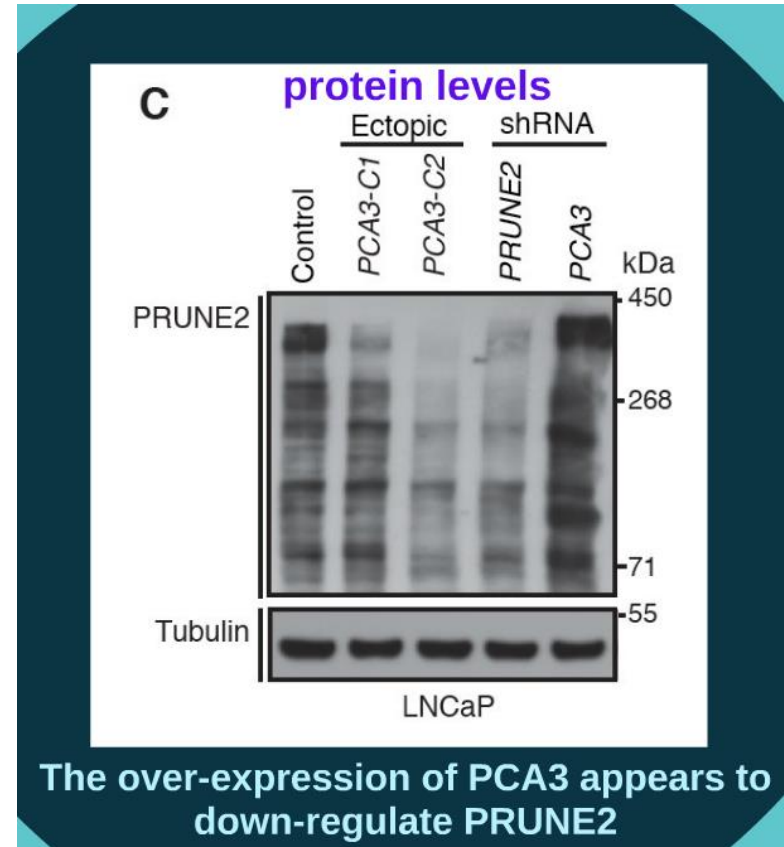
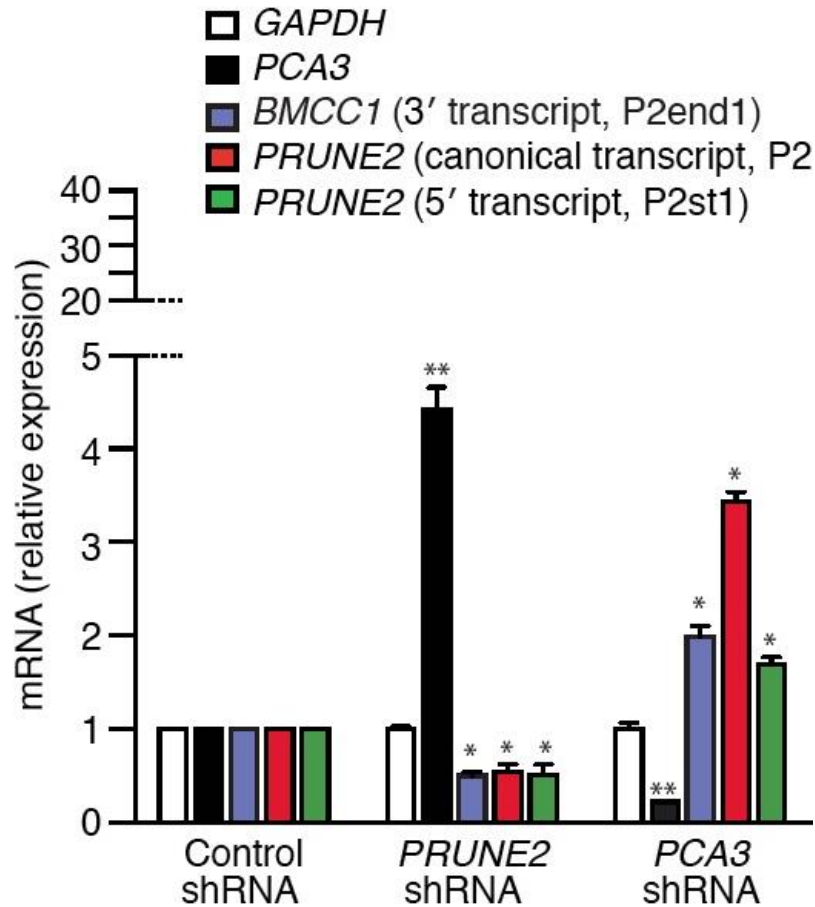
PCA3

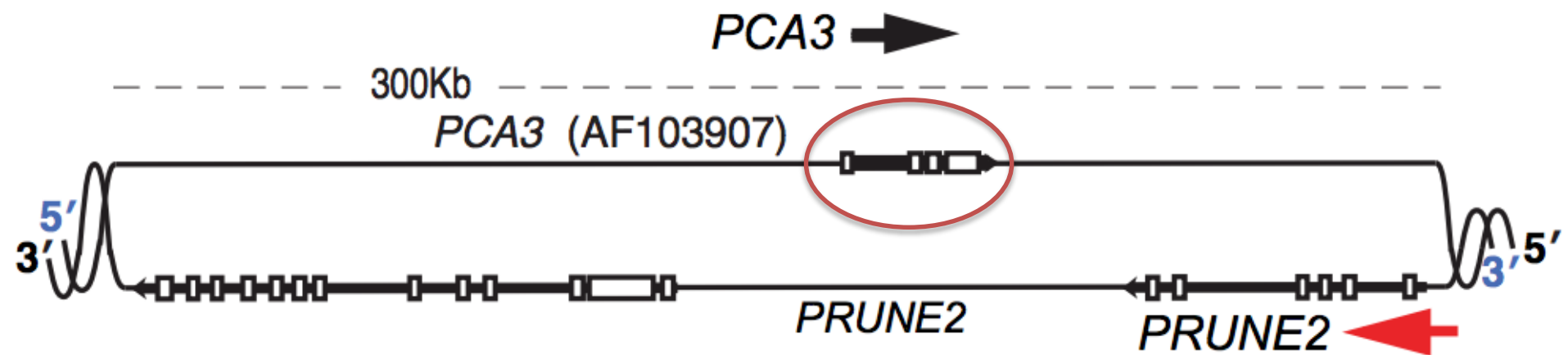


PCA3 is located in the intronic antisense strand of PRUNE2

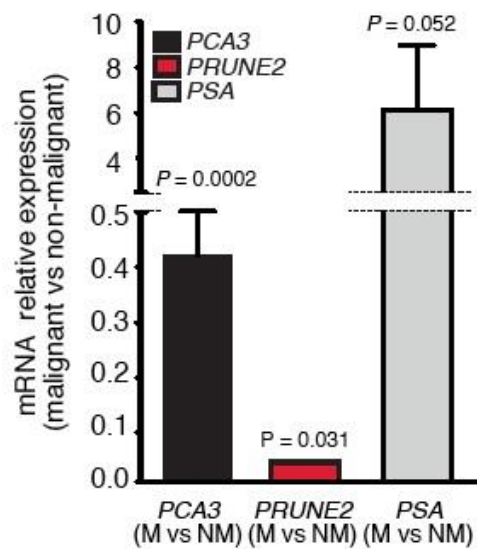


PCA3 regulates mRNA and protein levels of PRUNE2

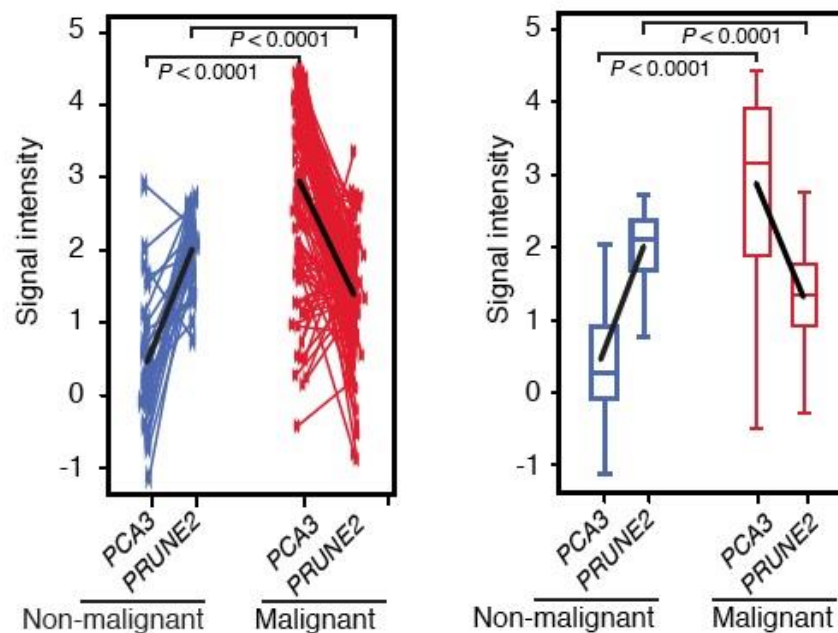


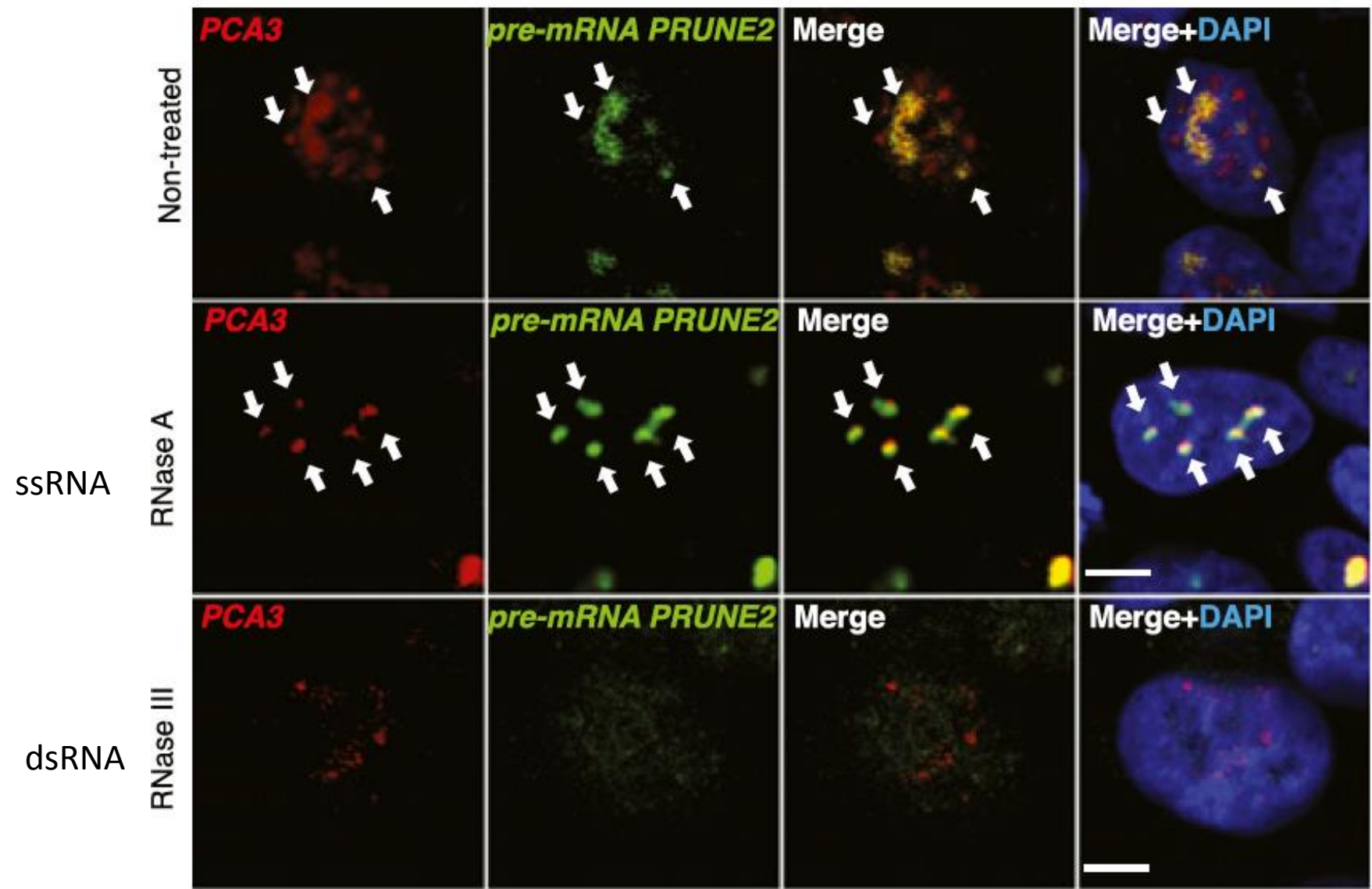


A



B



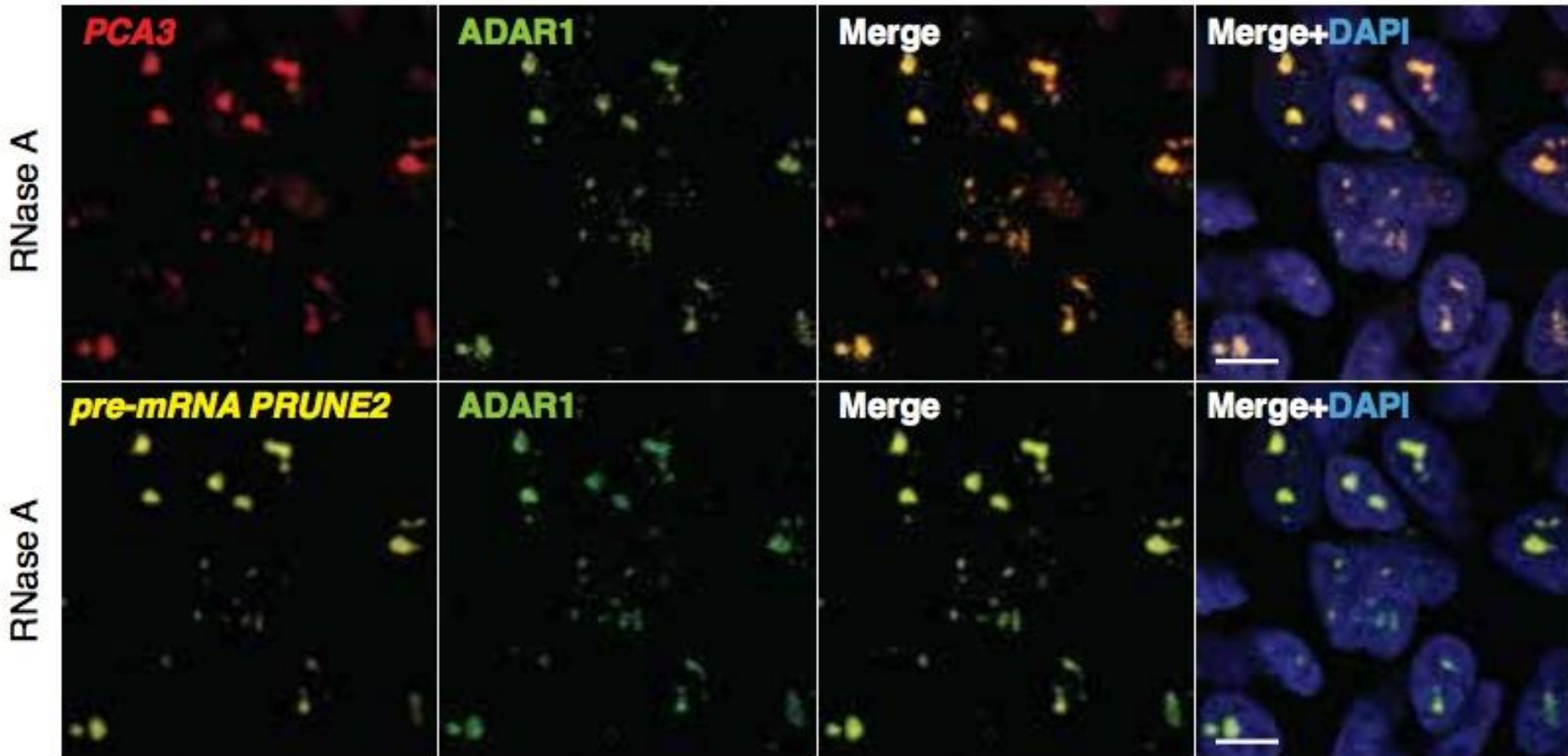


Mature PCA3 forms a dsRNA complex with immature PRUNE2

dsRNA – trigger anti-viral response?

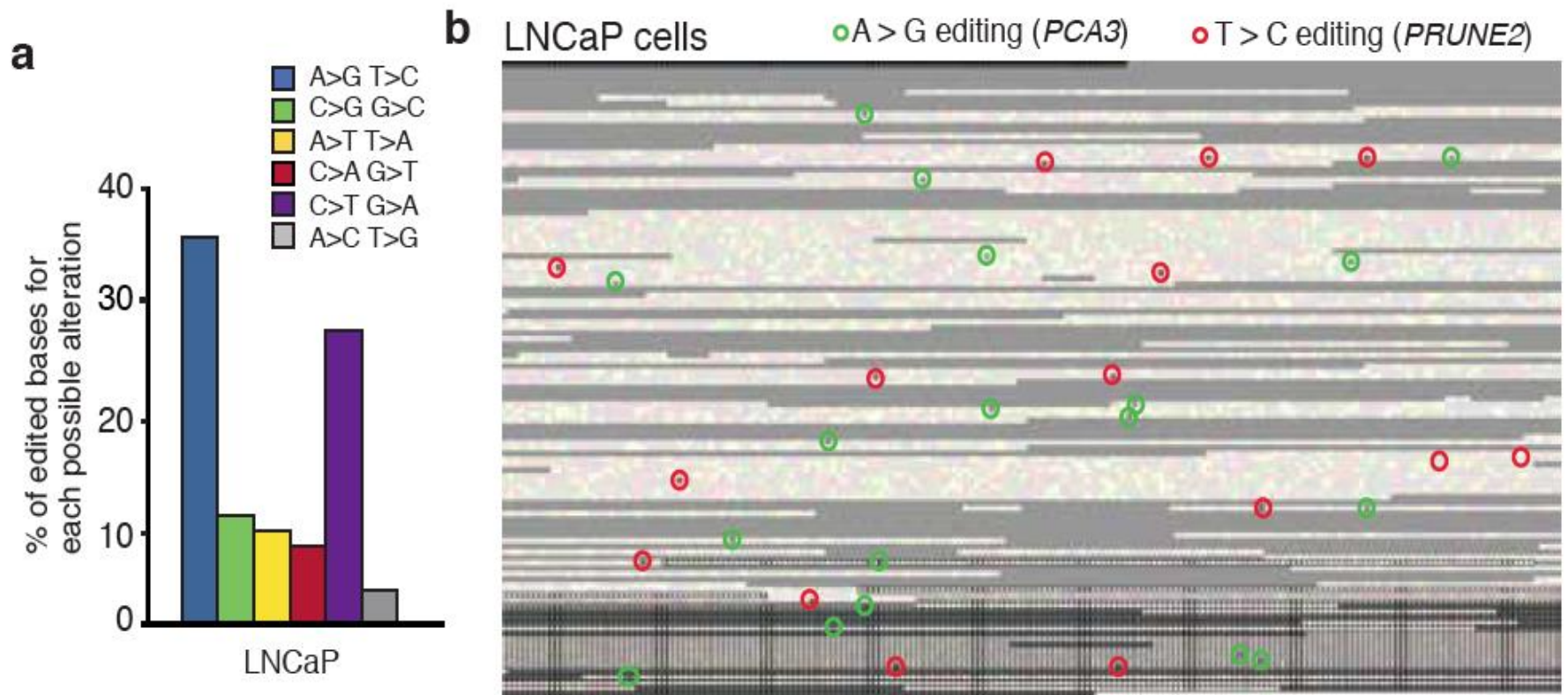
ADAR1 – Double-stranded RNA-specific deaminase

PCA3, PRUNE2 & ADAR1 co-localize in the cell nucleus and are resistant to RNaseA treatment

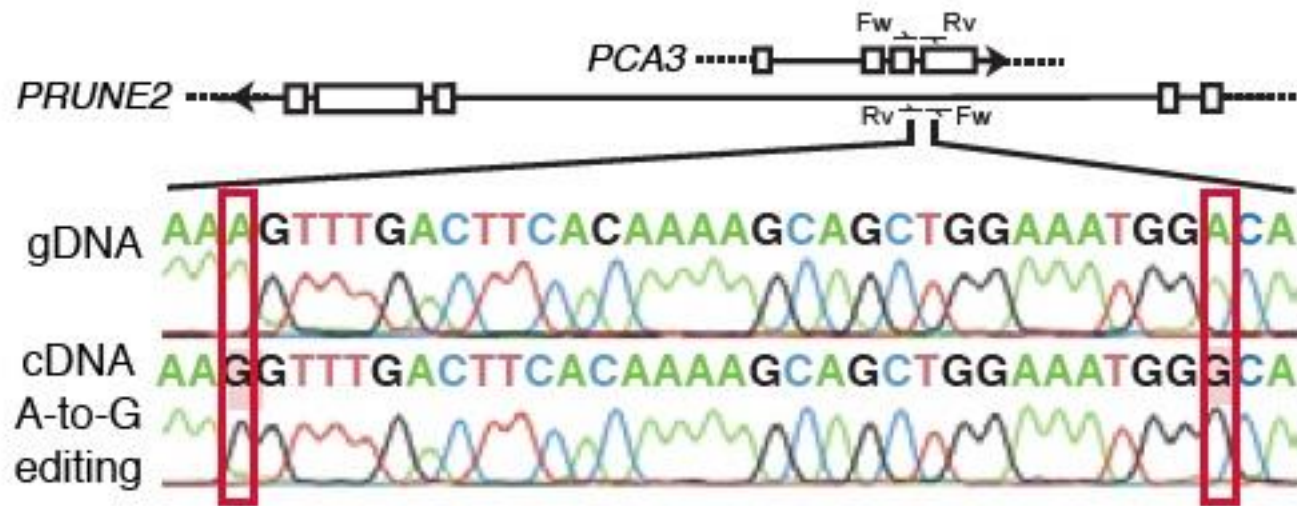


Sensitive to RNase-H treatment (dsRNA)

If the PCA3/PRUNE2/ADAR complex is functional, we should detect RNA editing

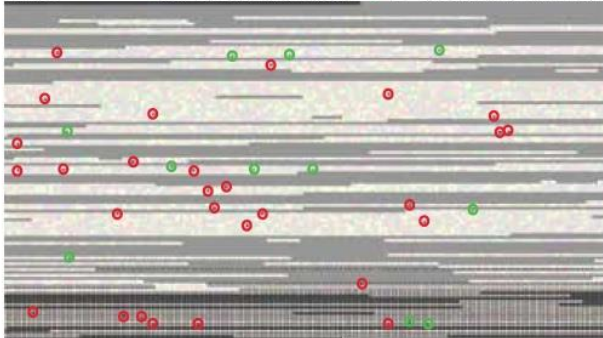


RNA editing was also observed in prostate cancer samples

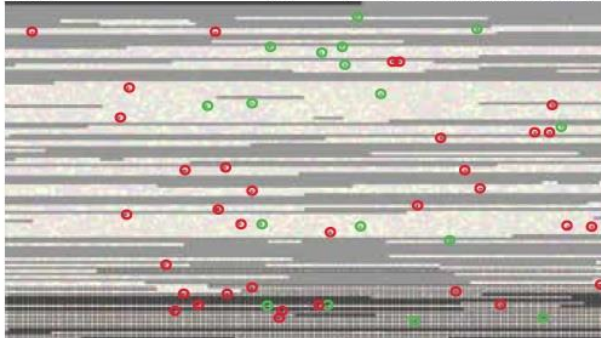


D

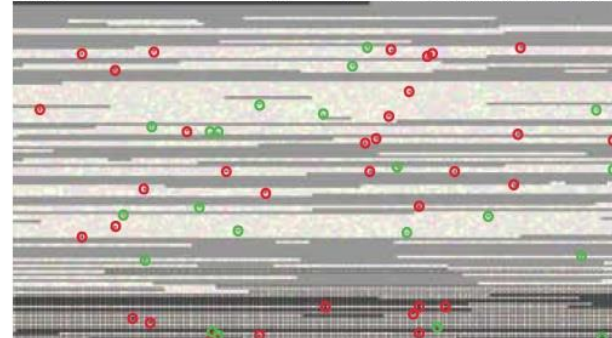
Pt #1 ● A > G editing (*PCA3*) ● T > C editing (*PRUNE2*)



Pt #2 ● A > G editing (*PCA3*) ● T > C editing (*PRUNE2*)

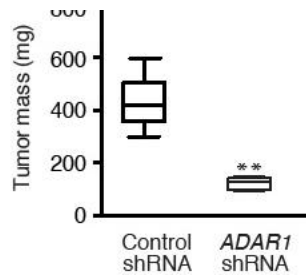
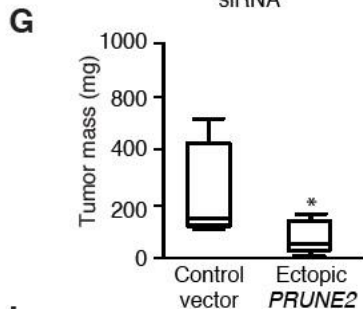
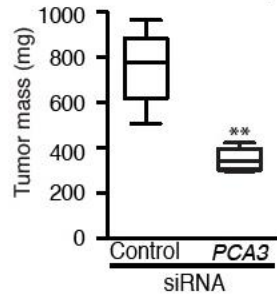
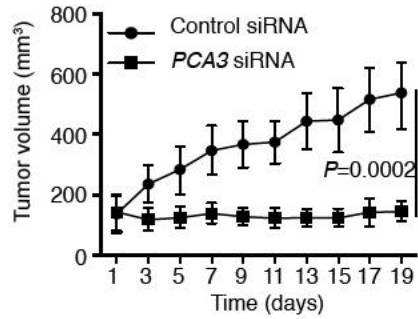


Pt #3 ● A > G editing (*PCA3*) ● T > C editing (*PRUNE2*)



**If PCA3 and ADAR1 act blocking PRUNE2,
we shall be able to reduce tumor growth
if we control these transcripts**

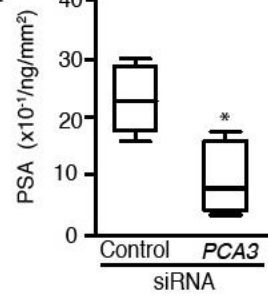
Reduction of tumor mass



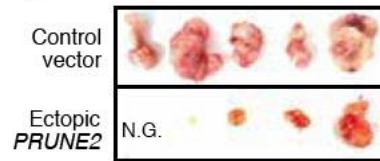
L



N



o



p



shRNA PCA3



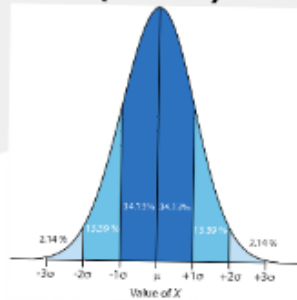
Ectopic PRUNE2



shRNA ADAR1

**The study of the ds-transcriptome
(dsRNA-Seq) in gastric cancer may
reveal novel actionable targets for this
malignancy**

overall survival
(N=200)



WES: 60 polar phenotypes

Gene Panel (V1)
- 89 genes
- 643,812 nt
- >2000X coverage

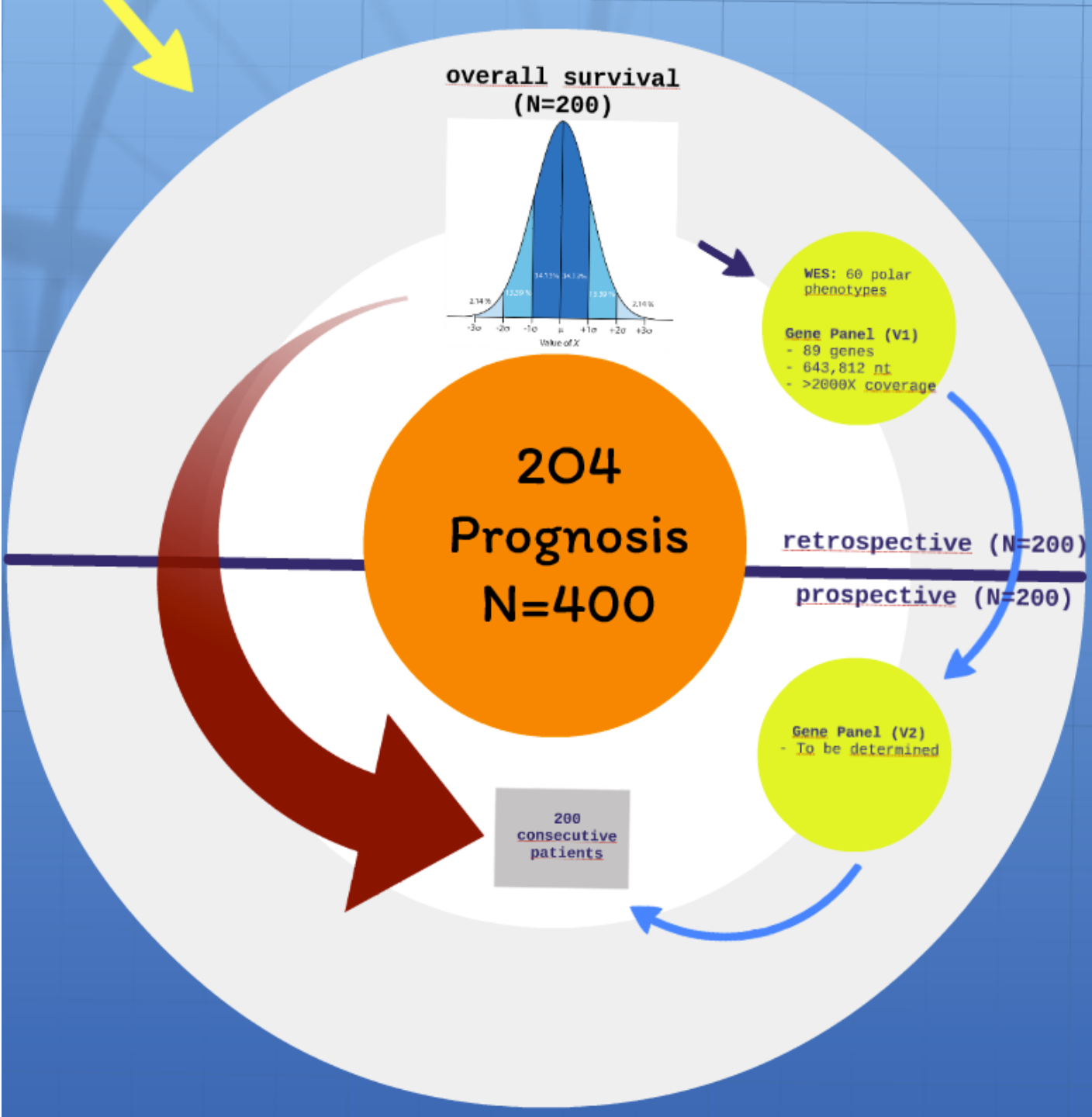
204
Prognosis
N=400

retrospective (N=200)

prospective (N=200)

Gene Panel (V2)
- To be determined

200
consecutive
patients



- **Gene panel (~160 transcripts) based in the 4 most recent comprehensive studies of gastric cancer genomics (200 cases)**
- **Evaluate the effect of cumulative mutations and prognosis**
- **WES for polar cases (disease free-survival)**
- **Gene panel v2.0**
- **Molecular Ancestry studies**

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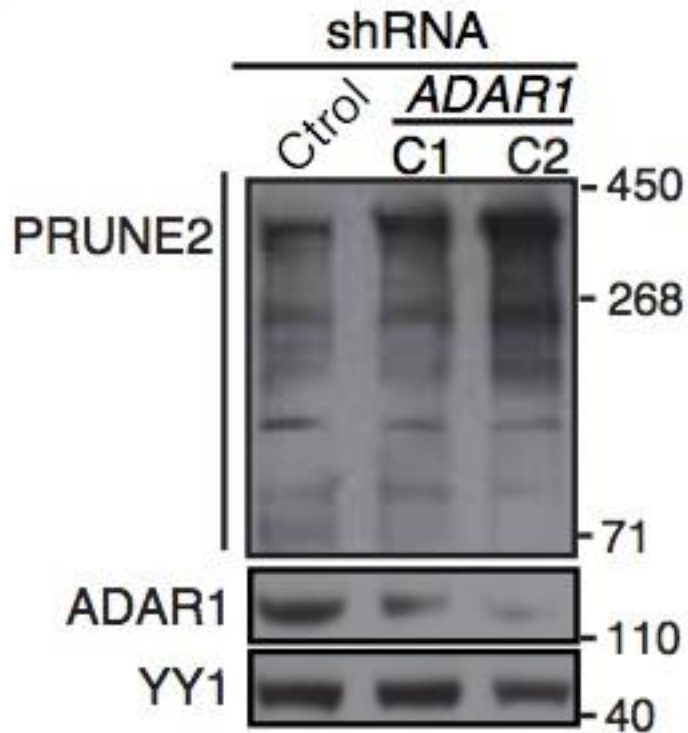
Renan Valieris



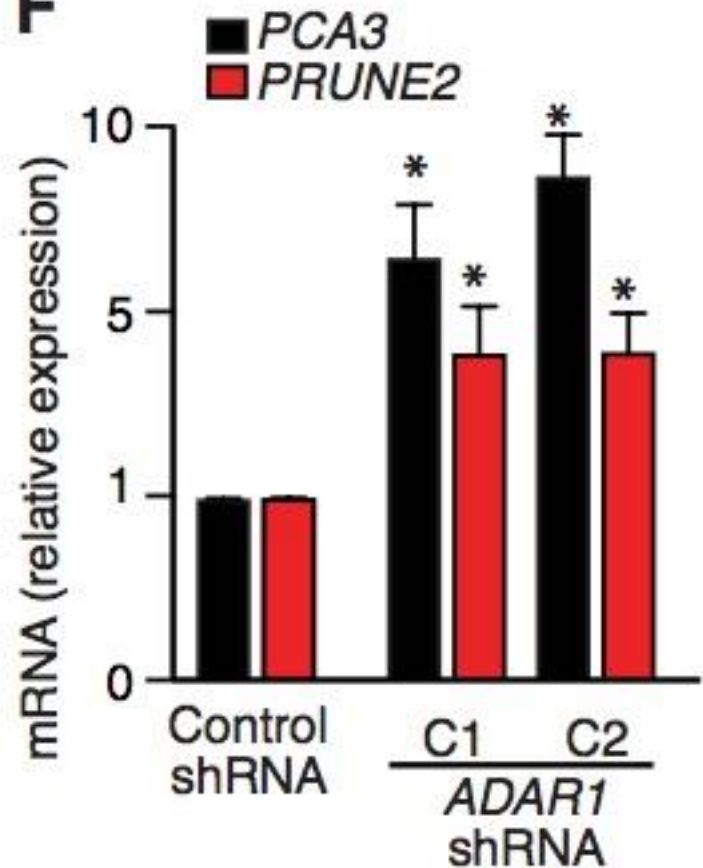
emmanuel@cipe.accamargo.org.br

shRNA ADAR1: increases PCA3 & PRUNE2 levels (protein and mRNA)

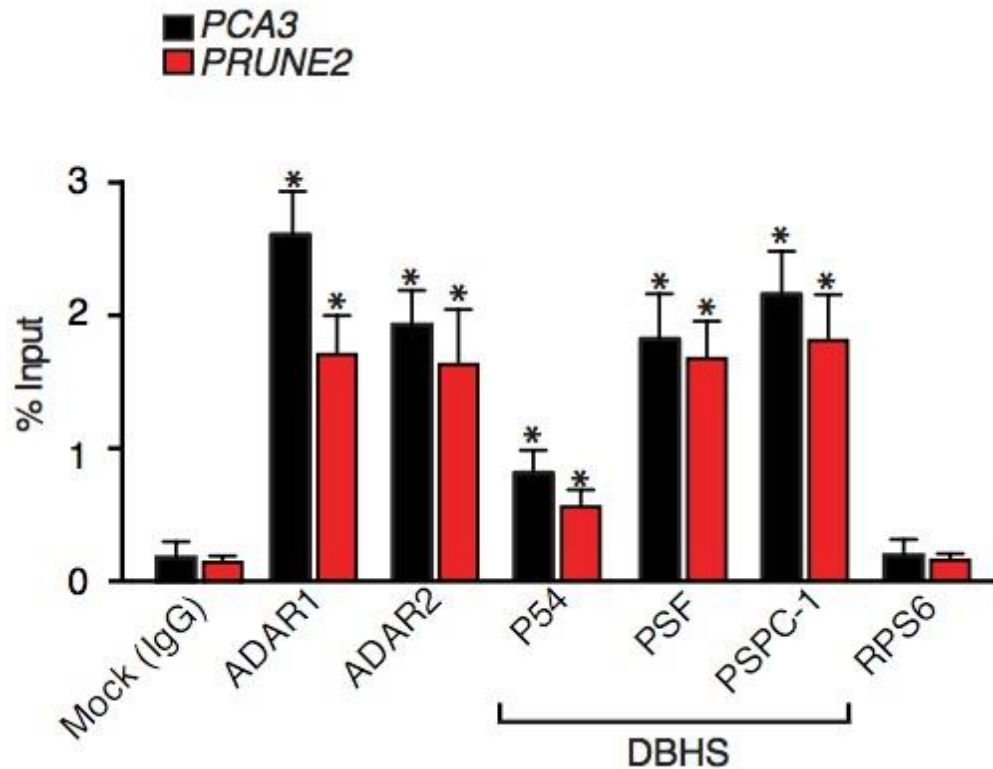
E



F



RNA-ChIP: ADAR1 and paraspeckle proteins are in the same complex as PCA3 & PRUNE2



Paraspeckle-proteins: nuclear retention/degradation

