GUT MICROBIOTA E PATOLOGIE CORRELATE

Dr GIAN MARCO GIORGETTI

DIRETTORE UO NUTRIZIONE CLINICA ASL ROMA2

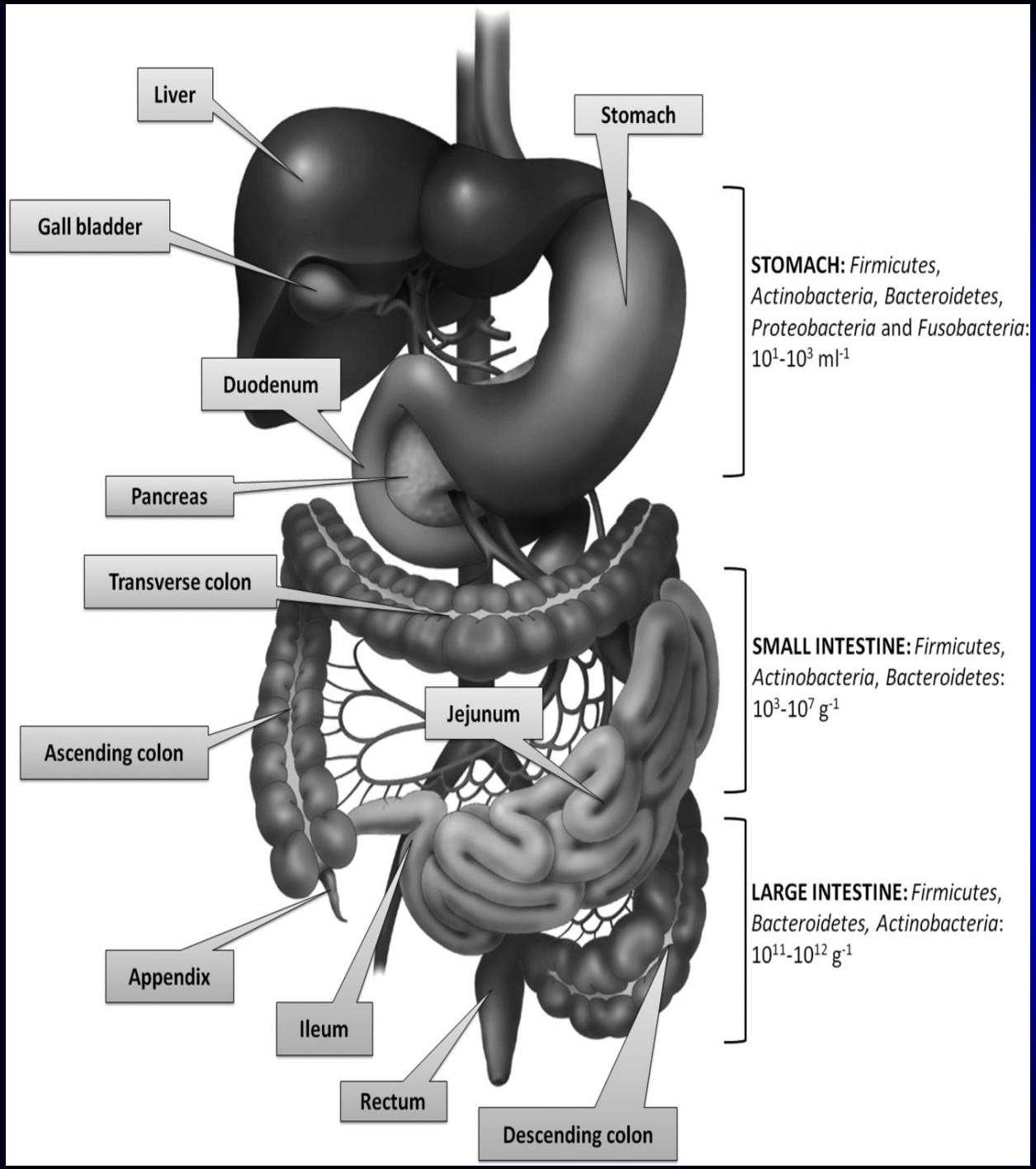
THE GUT MICROBIOTA COMPOSITION

TABLE. **Major Bacteria and Archaea Phyla and Genera Found in the Human Gut Microbiota**[a]

Phyla	Representative genera
Bacteria	
Firmicutes	*Ruminococcus*
	Clostridium
	Peptostreptococcus
	Lactobacillus
	Enterococcus
Bacteroidetes	*Bacteroides*
Proteobacteria	*Desulfovibrio*
	Escherichia
	Helicobacter
Verrucomicrobia[b]	
Actinobacteria	*Bifidobacterium*
Cyanobacteria[b]	
Synergistes[b]	
Archaea	
Euryarchaeota	*Methanobrevibacter*

Humans: Meta-organism

10-fold greater numbers of bacterial tha animal cells, matabolically and immunologically intagrated, with a biomass >1 Kg



$<10^{3}$ CFU/mL

10^{2}-10^{9} CFU/mL

10^{4}-10^{12} CFU/mL

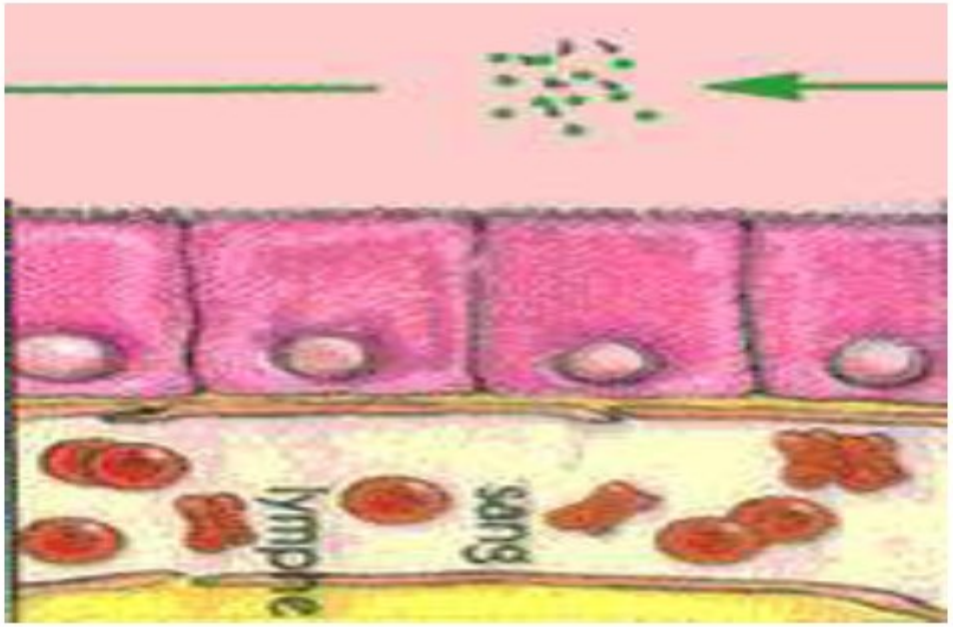
GUT MICROBIOTA: only bacteria?

Virome **Bacteriome** **Mycobiome**

Mucosal Barrier

Epithelial barrier

Endocrine system



Acquired and Innate immunity

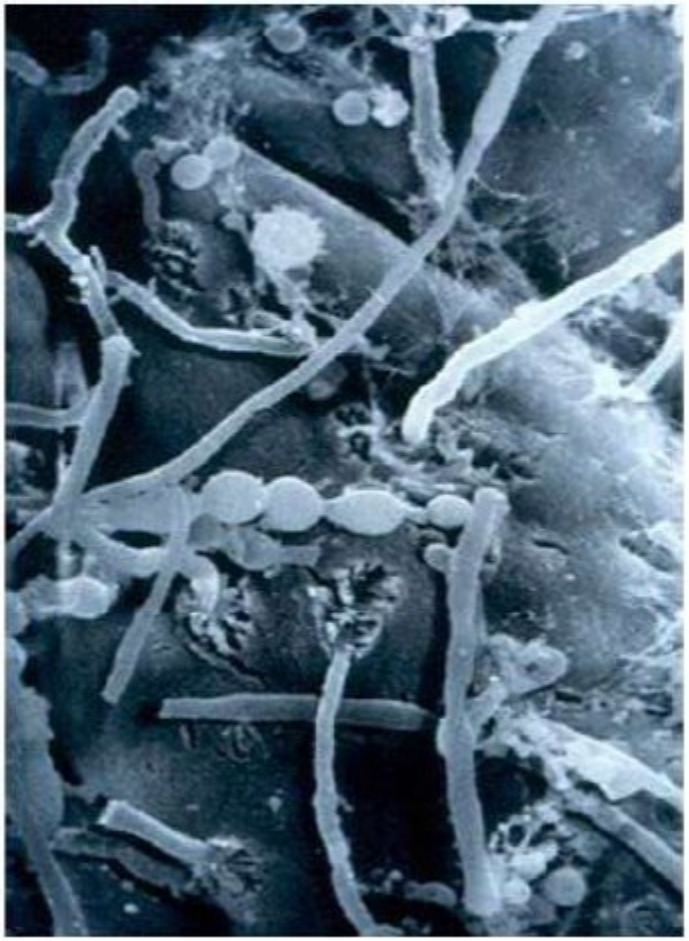
Vascular and lymphatic systems

Neuroenteric system

Digestive enzymes

What role for YEASTS?
Candida from commensal to pathogen

- **Yeasts are commensal to the gut at low concentrations**
- **Candida overgrowth is a consequence of disturbances in the host's defense systems:** ***antibiotic therapy and change in physiological gut microbiota, pH, partial CO2 pressure, amino acid availability, iron deficiency…***
- **Yeast genome can be modified by repeated point mutations** ***(«microevolution»)*** **in order to overcome host protective measures**



Thewes S, Mol Microbiol 2007

GUT BACTERIOME

700-800 kg of bacteria, >3.300.000 genes

8 bacterial divisions (superkingdoms)
95% genes identity

↓

>1100 species
Over 99% genes identity

↓

>15000 strains
100% genes identity

Microbiome ⟷ **Metabolome**

Leser et al, Environ Microbiol 2011

Qin et al, Nature 2011

GUT mutalistic bacteria characterization

Need fo a Molecular Microbiology

Microscopic counts on human faeces suggest that

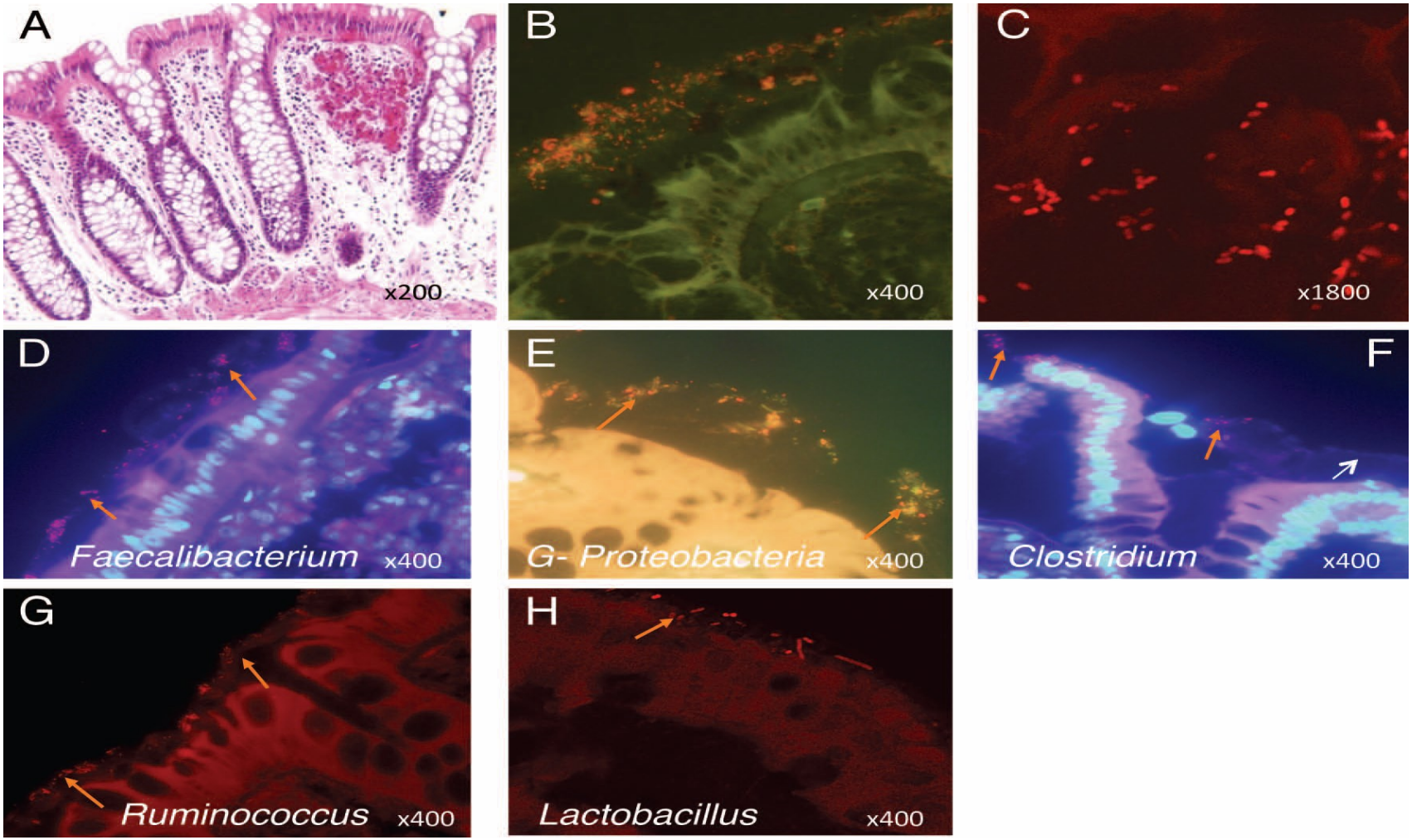
60 to 80% of the observable bacteria cannot be cultivated

Culture independent methods based on the analysis of the 16S rRNA gene reveals a *myriad of novel bacterial lineages within the human gut*

Suau et al, Appl Environ Microbiol 1999

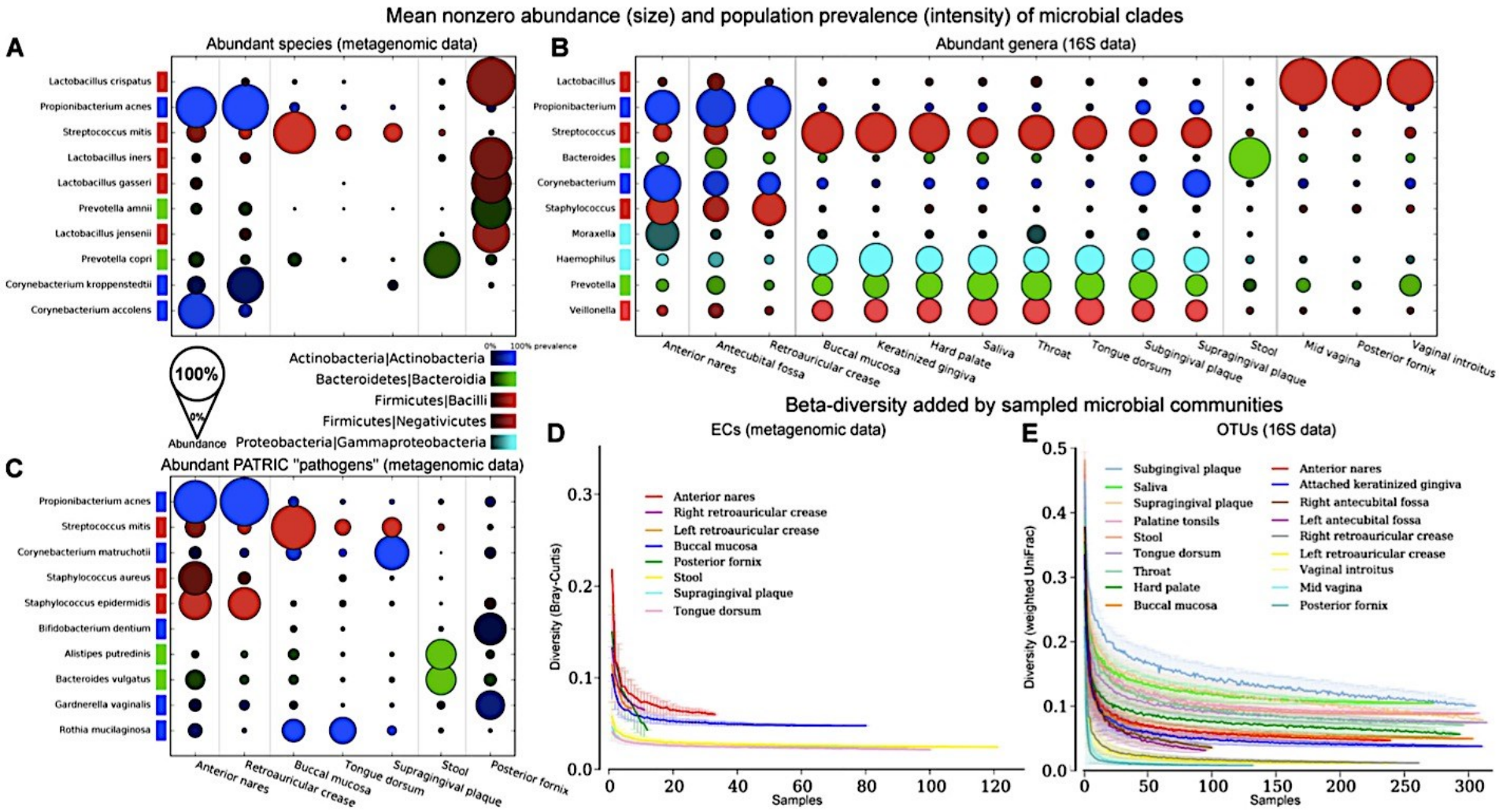
GUT MICROBIOTA

Fluorescence in-situ hybridization (FISH) using bacterial 16S rRNA probes showing bacteria localized to the mucus layer.



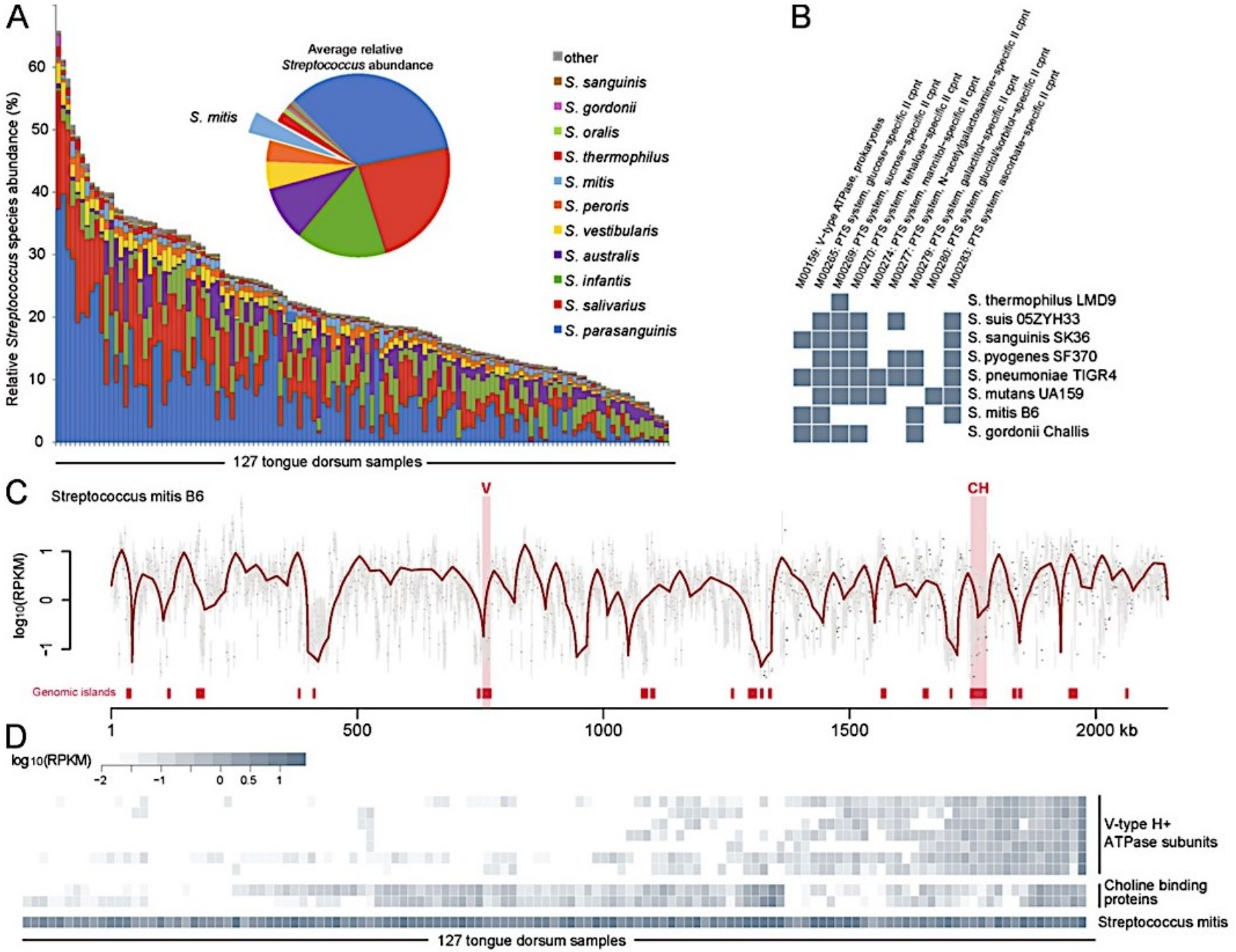
Xiang Jun Shen, Gut Microbes2010

Taxa prevalenti nel microbioma umano, che sono stati metagenomicamente e tassonomicamente ben definiti nella popolazione HMP.



Nature. Author manuscript; available in PMC 2013 February 05.

L’assetto del microbioma varia tra i soggetti al livello di specie e ceppi.



Nature. Author manuscript; available in PMC 2013 February 05.

At birth the human body is sterile

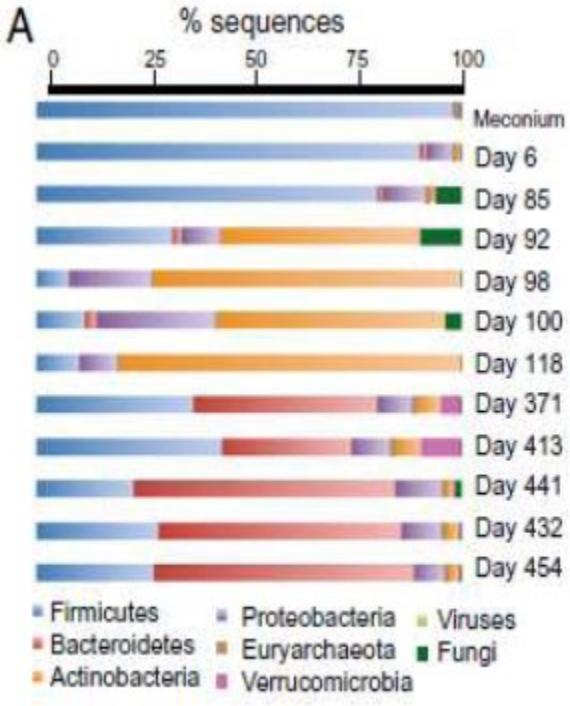
Vaginal microbiota (mother)

Fecal microbiota (mother)

Skin microbiota (mother/father/parents/ babysitter)

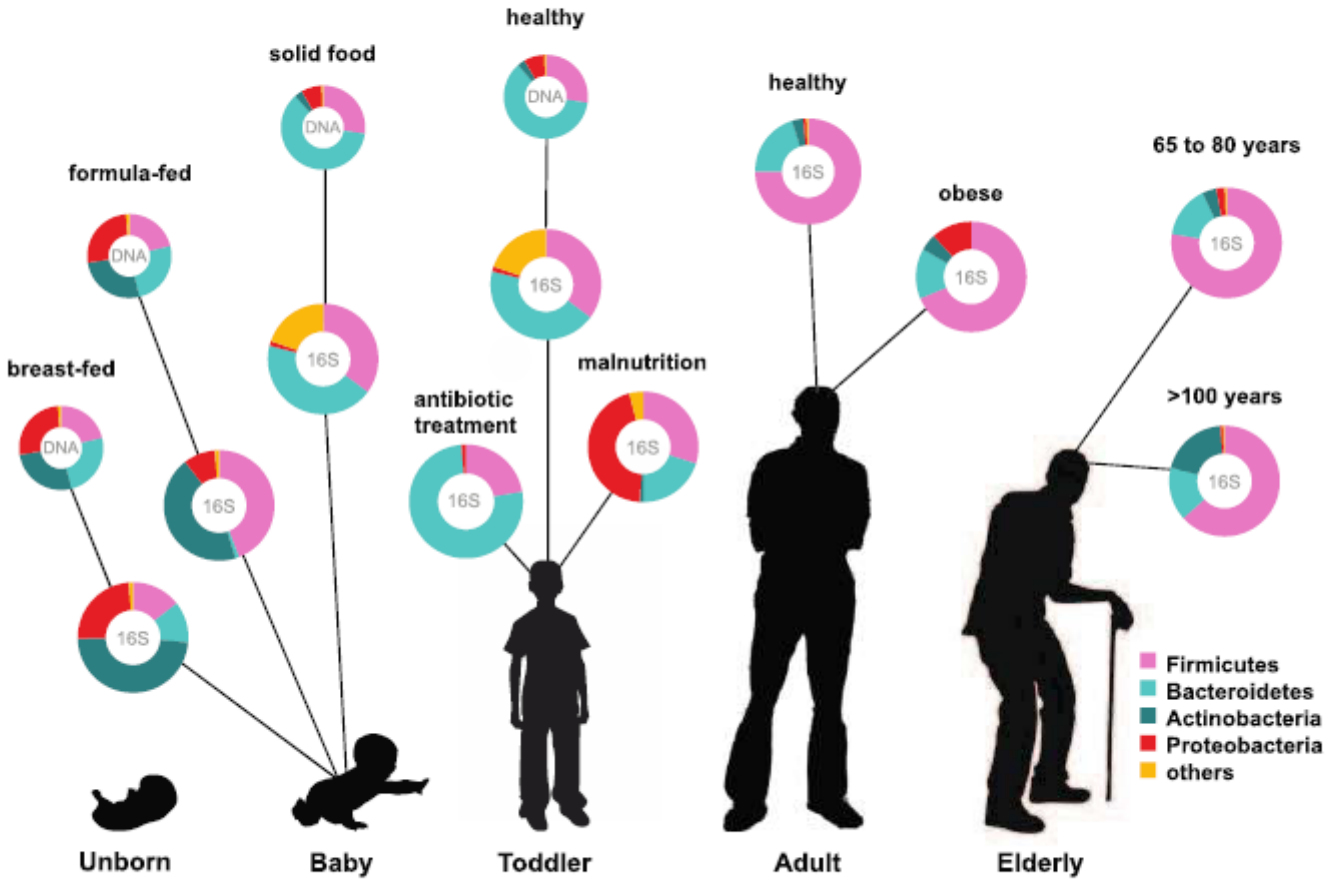
Diet

Ambient

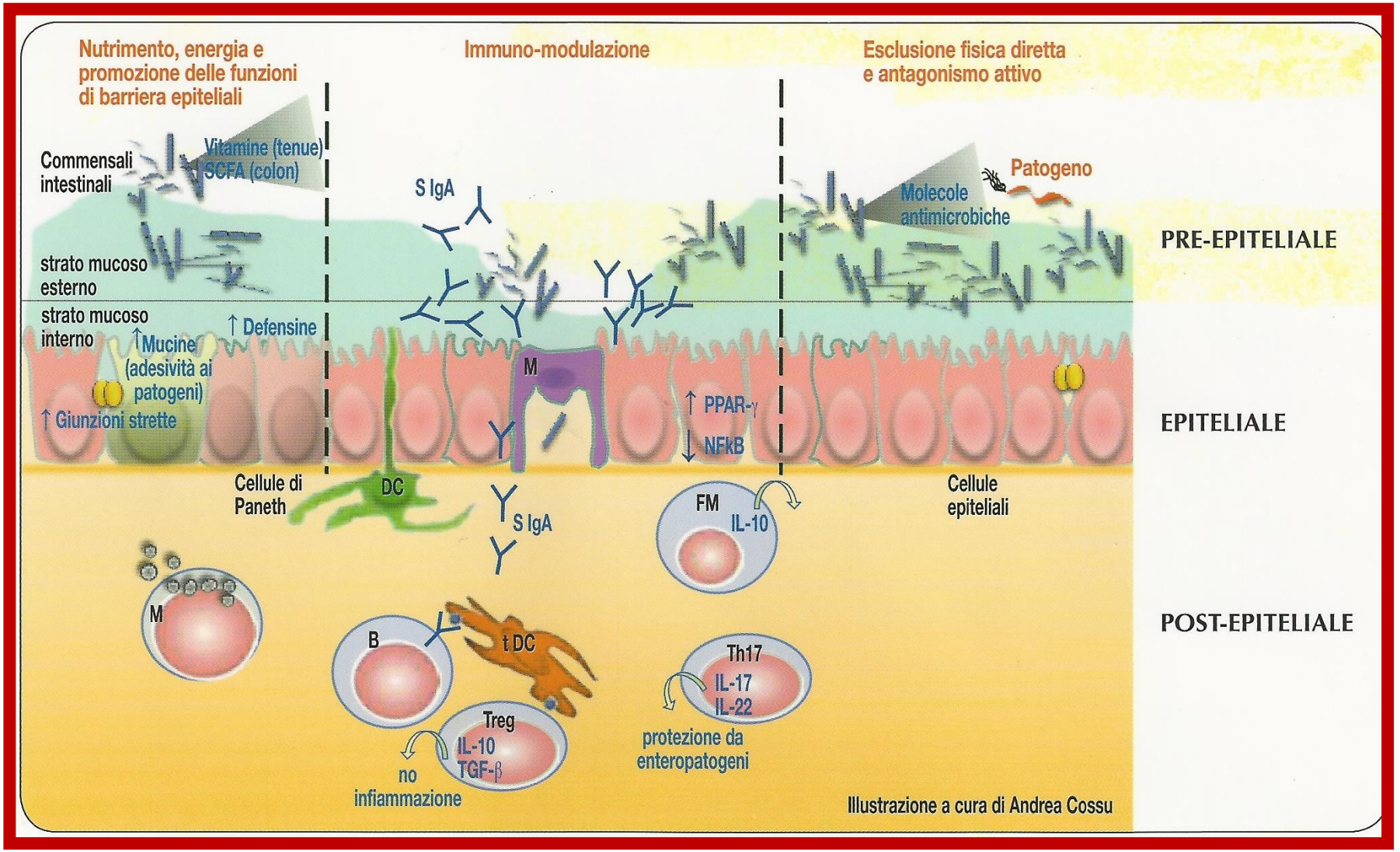


Native CORE microbiota (4-36 months of life)

Bacterial diversity increases with age



Ottmann N et al. Front Cell Infect Microb 2012


Nutrimento, energia e promozione delle funzioni di barriera epiteliali
Immuno-modulazione
Esclusione fisica diretta e antagonismo attivo
Commensali intestinali
Vitamine (tenue)
SCFA (colon)
S IgA
Patogeno
Molecole antimicrobiche
strato mucoso esterno
strato mucoso interno
↑ Defensine
↑ Mucine (adesività ai patogeni)
↑ Giunzioni strette
M
↑ PPAR-γ
↓ NFkB
PRE-EPITELIALE
EPITELIALE
POST-EPITELIALE
Cellule di Paneth
DC
S IgA
FM
IL-10
Cellule epiteliali
M
B
t DC
Th17
IL-17
IL-22
Treg
IL-10
TGF-β
no infiammazione
protezione da enteropatogeni
Illustrazione a cura di Andrea Cossu

Hindawi Publishing Corporation
Journal of Immunology Research
Volume 2015, Article ID 501361, 7 pages
http://dx.doi.org/10.1155/2015/501361

Hindawi

Review Article

Interactions between Innate Immunity, Microbiota, and Probiotics

GianMarco Giorgetti,[1] Giovanni Brandimarte,[2] Federica Fabiocchi,[3] Salvatore Ricci,[4] Paolo Flamini,[4] Giancarlo Sandri,[3] Maria Cristina Trotta,[5] Walter Elisei,[6] Antonio Penna,[7] Piera Giuseppina Lecca,[2] Marcello Picchio,[8] and Antonio Tursi[9]

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[9] *Gastroenterology Service, ASL BAT, Via Torino 49, 76123 Andria, Italy*

WHEN A DYSBIOSIS OCCURS?

Unbalanced DIET:

High fats

Low fibers

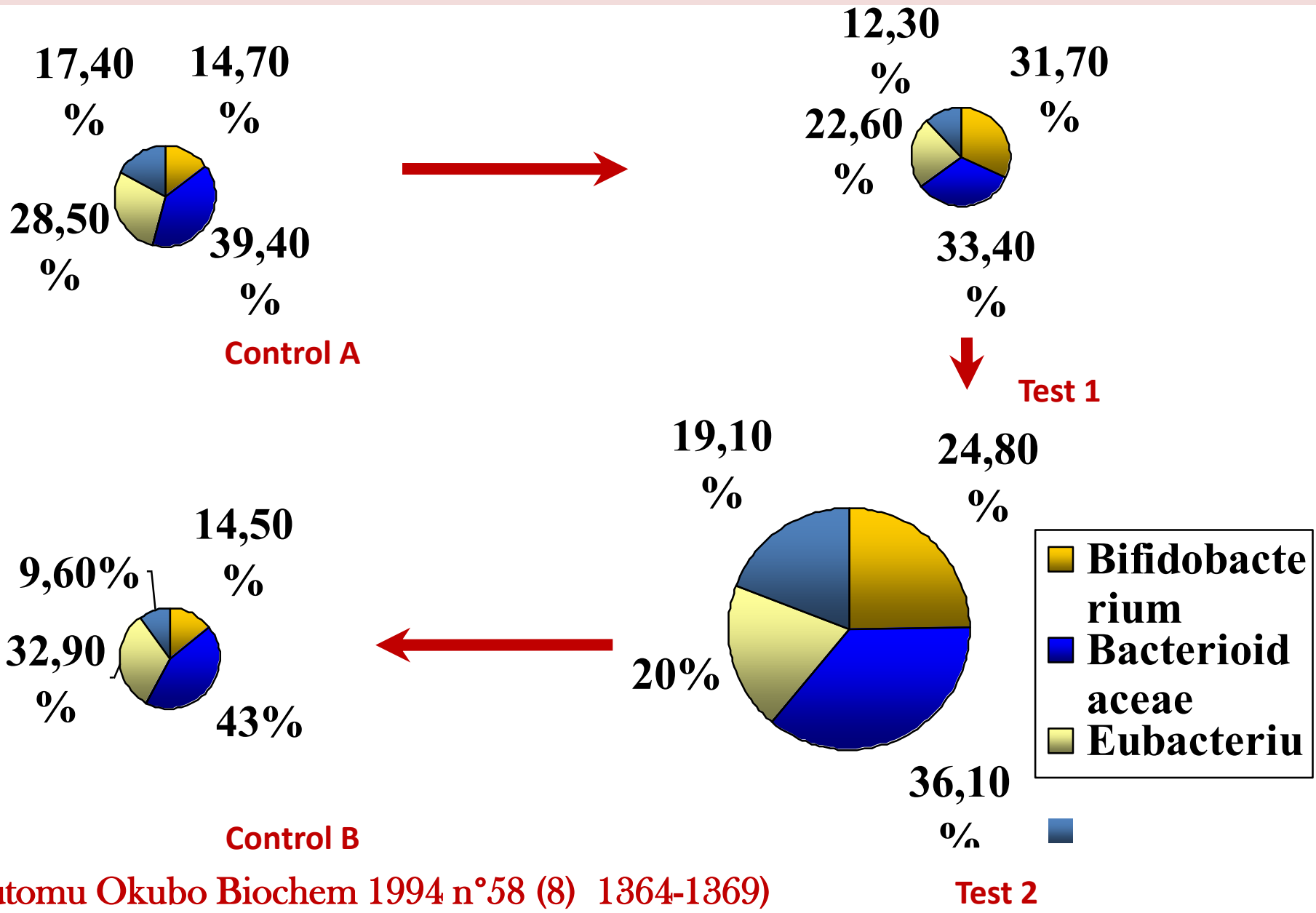
High calories

High meat content

….

DIETA

Percentage composition of Each Intestinal Microflora in Total Counts



(Tsutomu Okubo Biochem 1994 n°58 (8) 1364-1369)

nature COMMUNICATIONS

ARTICLE

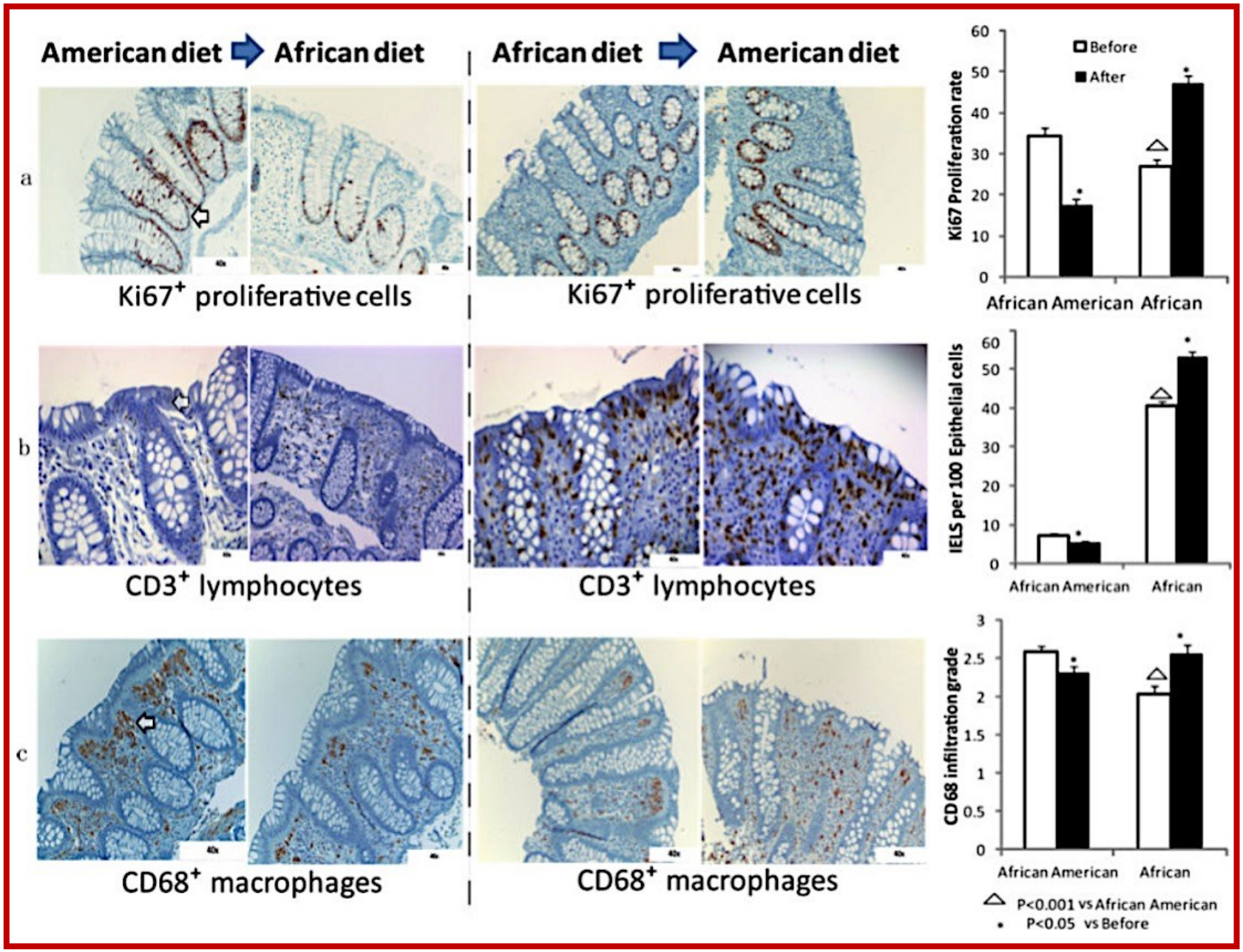
Received 23 May 2014 | Accepted 20 Jan 2015 | Published 28 Apr 2015

DOI: 10.1038/ncomms7342

Fat, fibre and cancer risk in African Americans and rural Africans

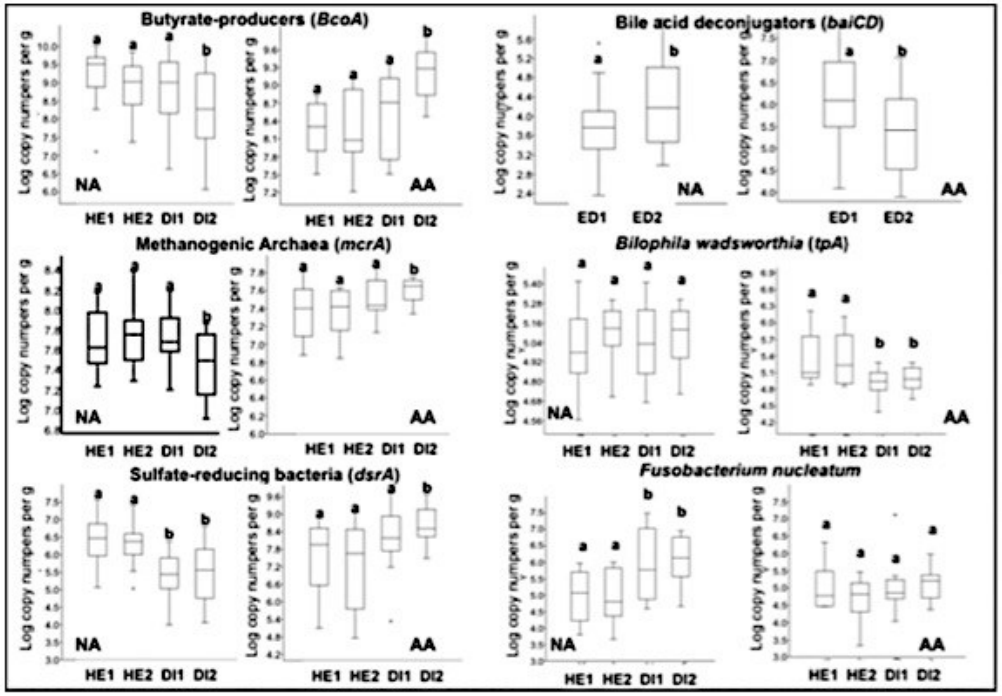
Stephen J.D. O'Keefe[1], Jia V. Li[2], Leo Lahti[3,4], Junhai Ou[1], Franck Carbonero[5,†], Khaled Mohammed[1], Joram M. Posma[2], James Kinross[2], Elaine Wahl[1], Elizabeth Ruder[6], Kishore Vipperla[1], Vasudevan Naidoo[7], Lungile Mtshali[7], Sebastian Tims[3], Philippe G.B. Puylaert[3], James DeLany[8], Alyssa Krasinskas[9], Ann C. Benefiel[5], Hatem O. Kaseb[1], Keith Newton[7], Jeremy K. Nicholson[2], Willem M. de Vos[3,4,10], H. Rex Gaskins[5] & Erwin G. Zoetendal[3]

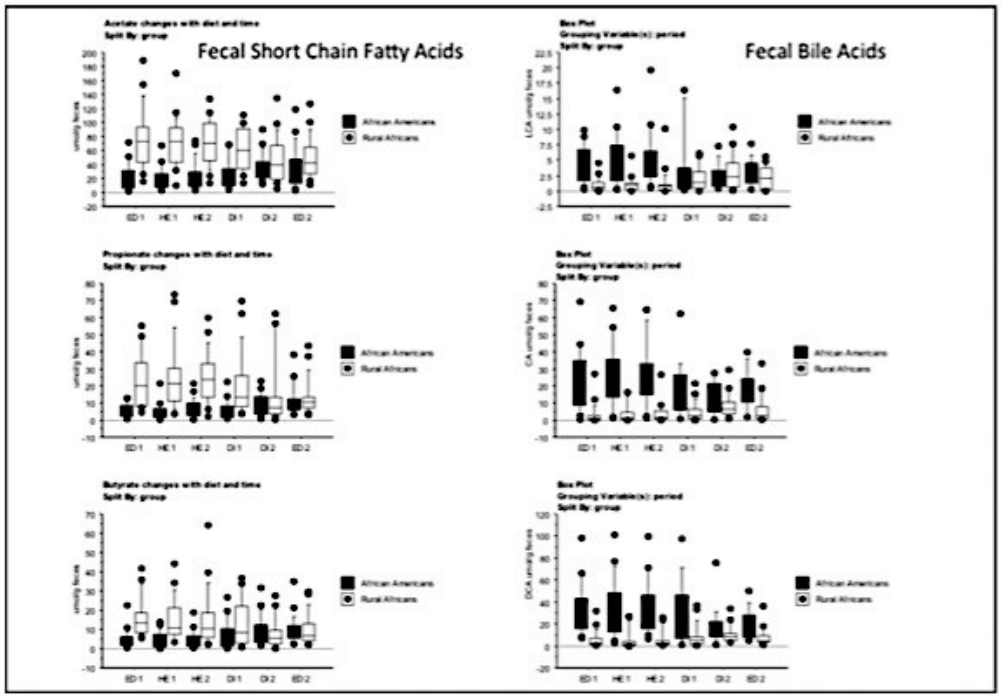
IMMUNOISTOCHIMICA DELLA MUCOSA DEL COLON DEI BIOMARKERS DI PROLIFERAZIONE E INFIAMMAZIONE



O'Keefe et al.; Fat, Fiber and Cancer Risk in African Americans and Rural Africans; *Nat Commun*. ; 6: 6342; 2015

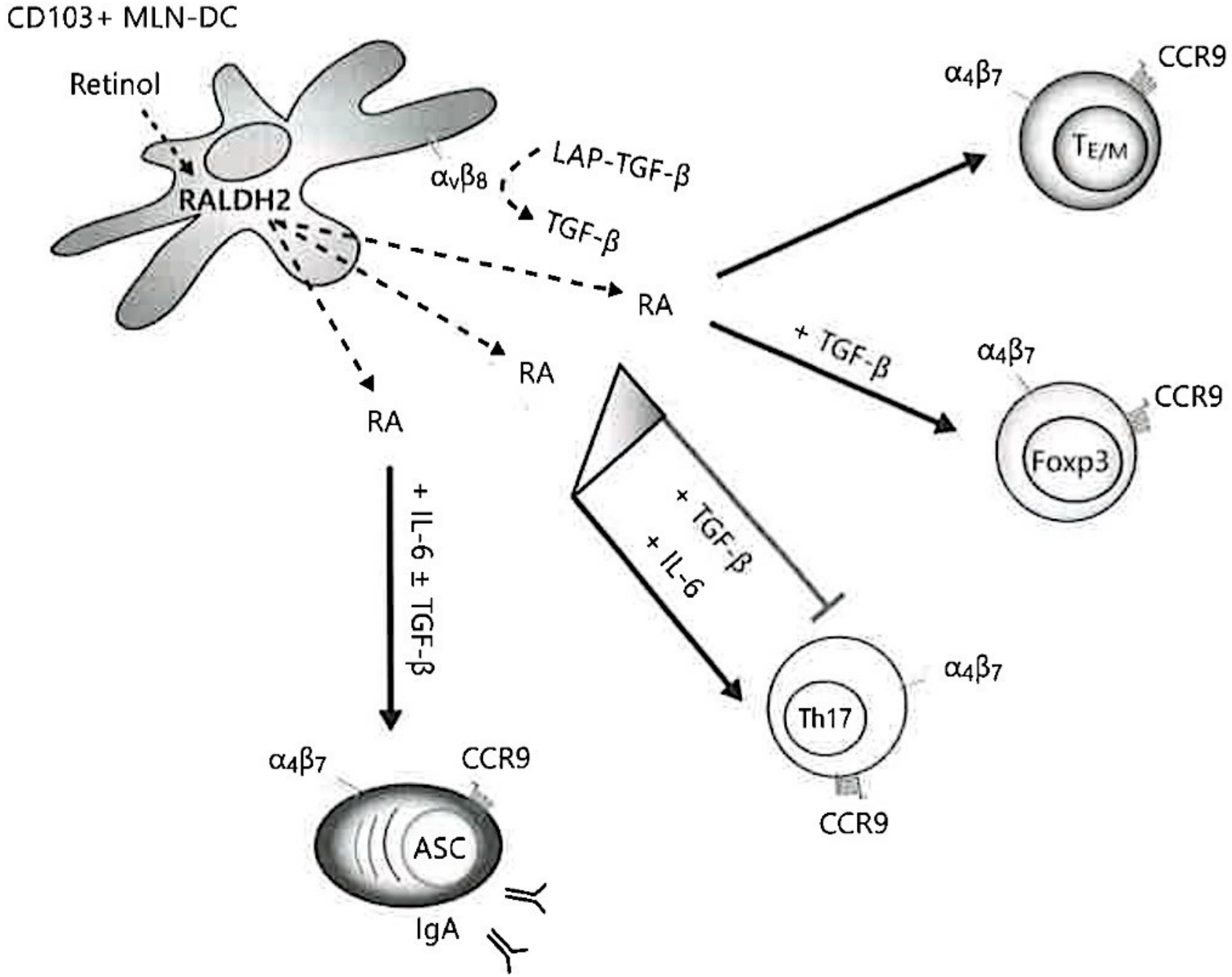
Analisi mirata dei geni funzionali delle specie microbiche di interesse specifico e dei loro metaboliti.



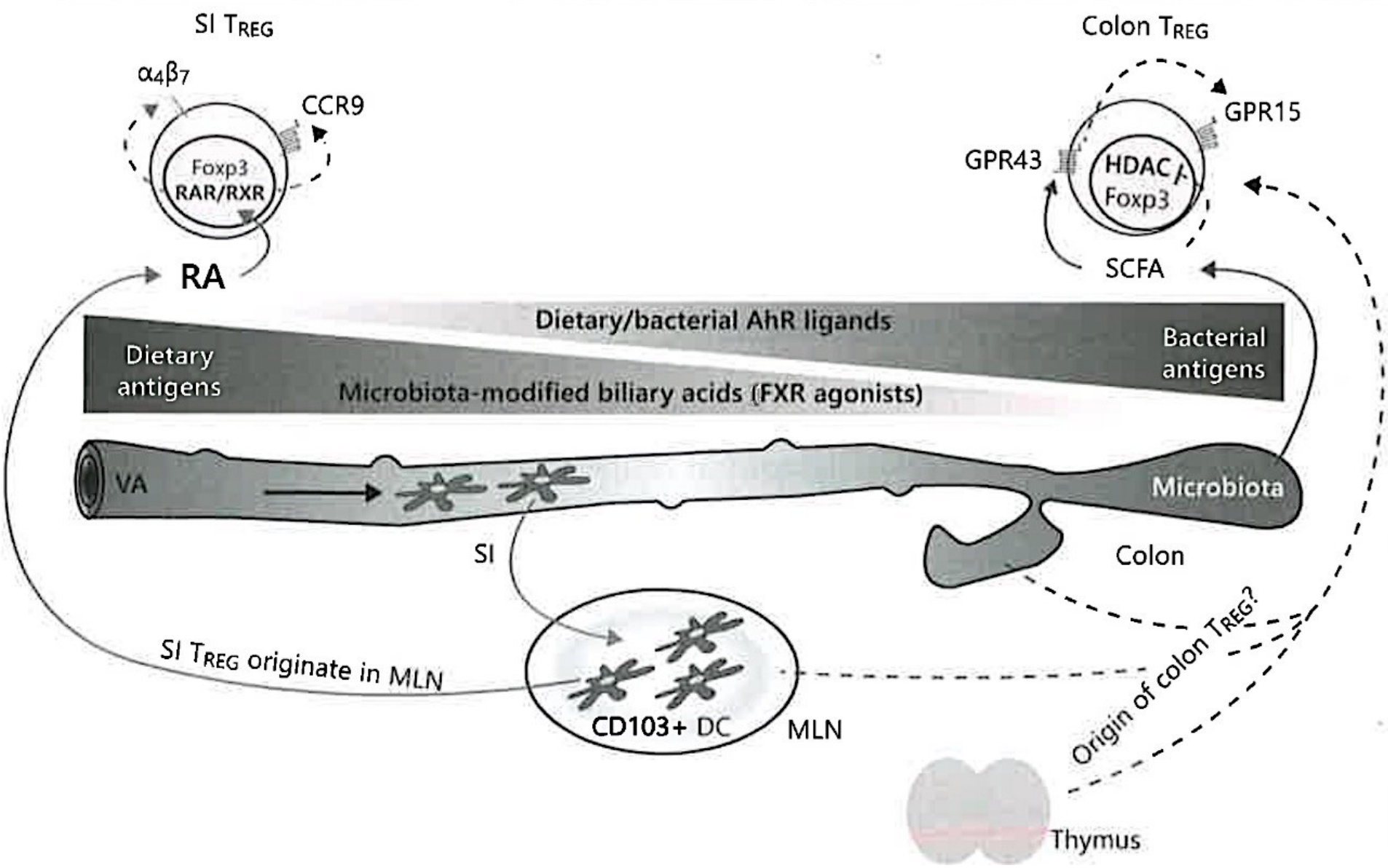


O'Keefe et al.; Fat, Fiber and Cancer Risk in African Americans and Rural Africans; *Nat Commun*. ; 6: 6342; 2015

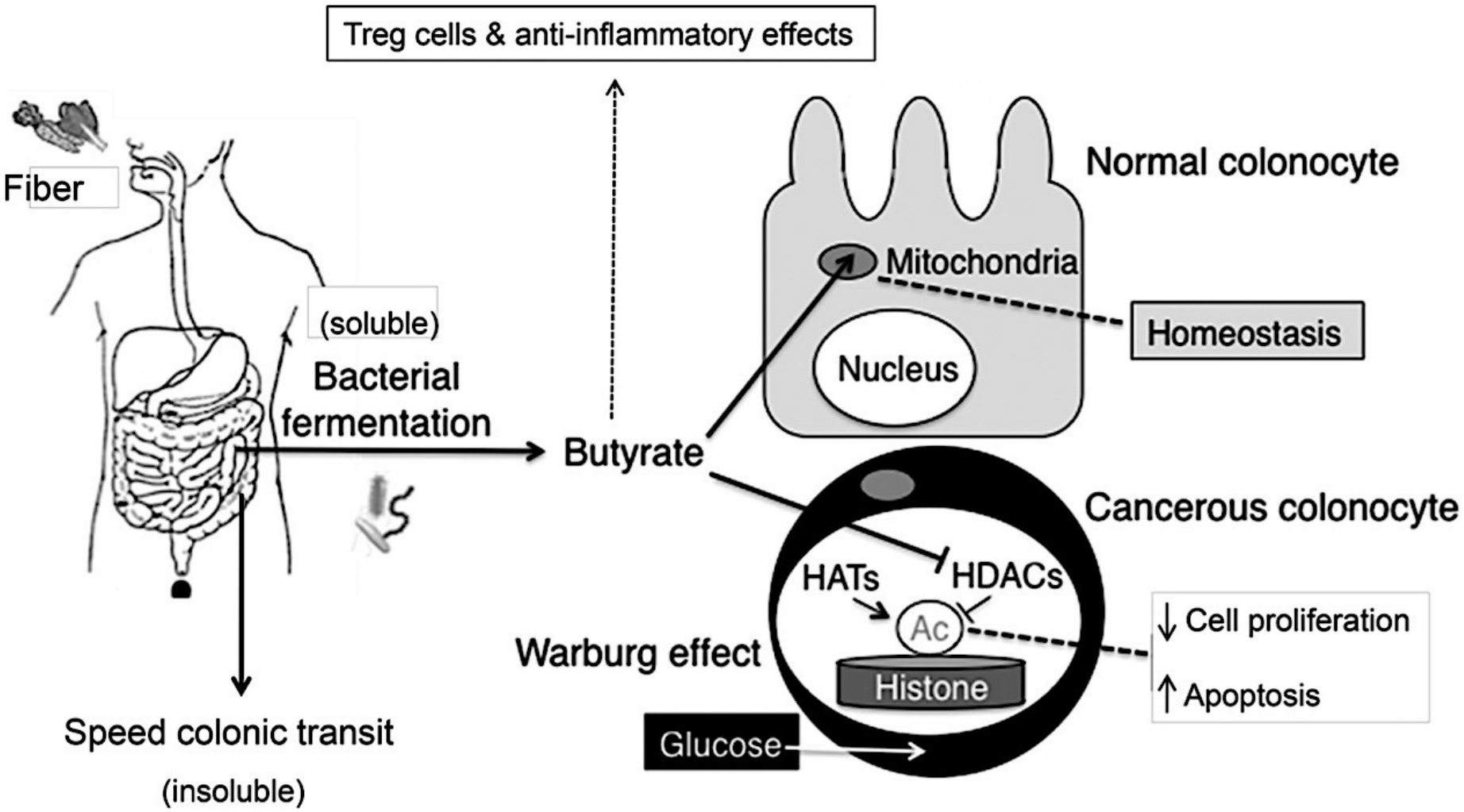
EFFETTI DEL RA SULL'*HOMING* E SULLA DIFFERENZIAZIONE DEI LINFOCITI.



RUOLO COMPLEMENTARE DEL RA E DEI SCFA NELLA TOLLERANZA IMMUNITARIA DEL PICCOLO INTESTINO E DEL COLON.

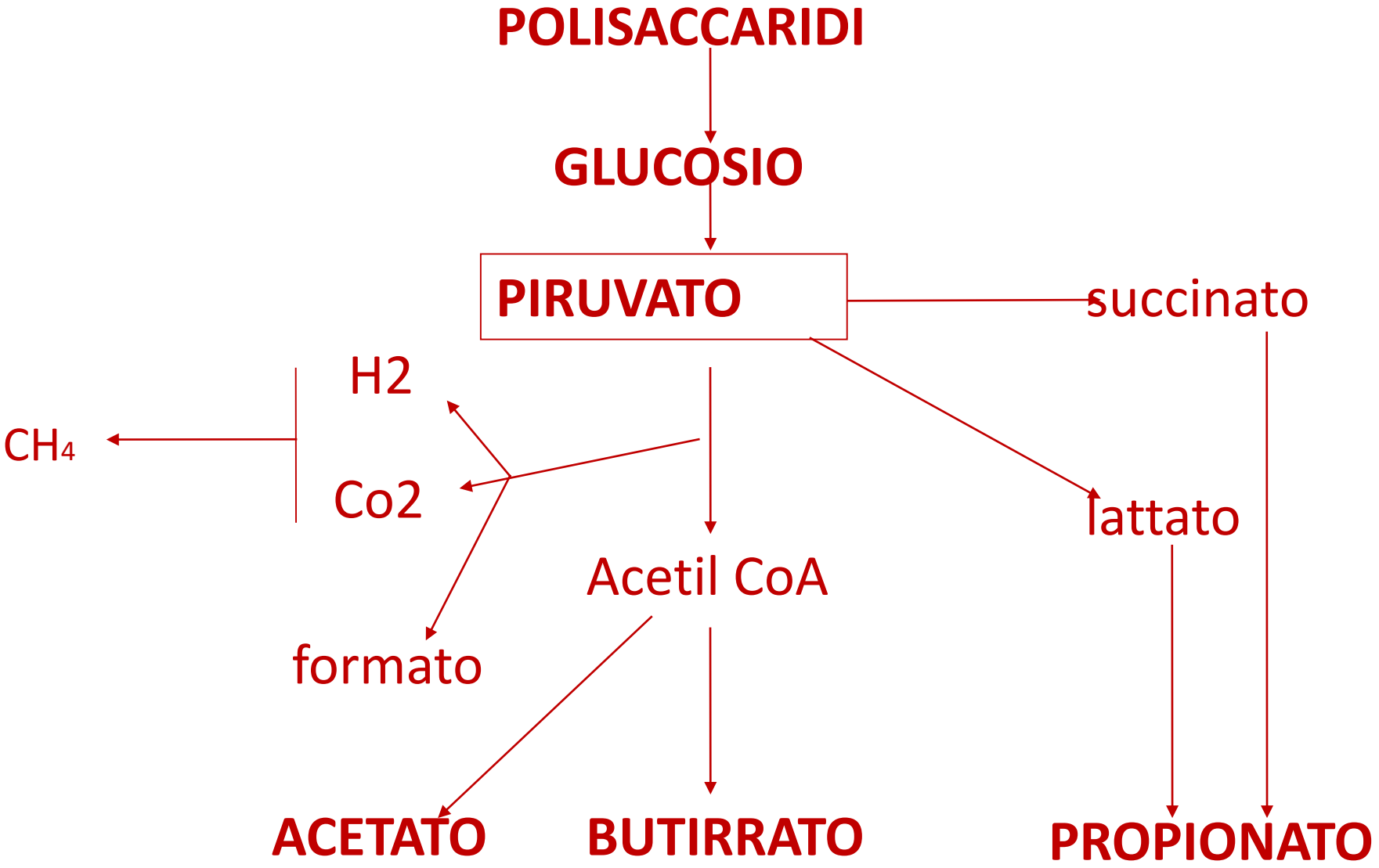


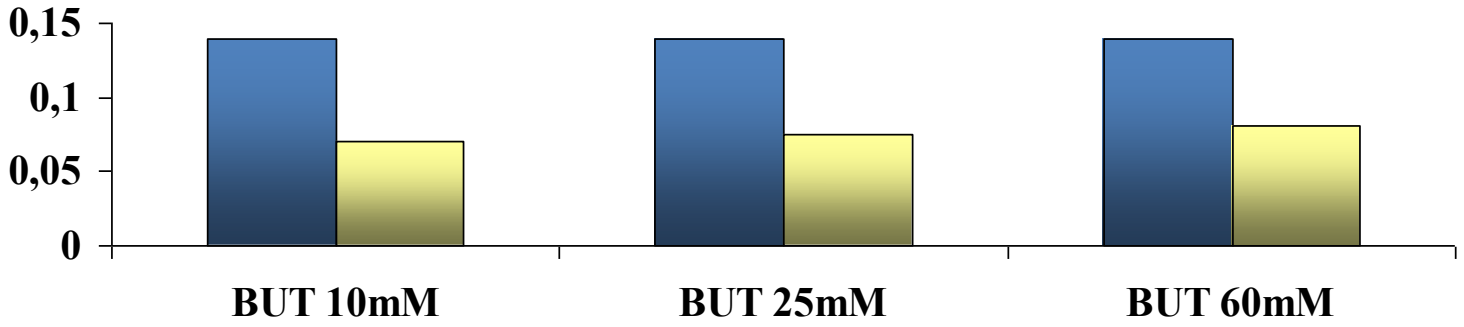
MECCANISMO DI SOPPRESSIONE DEL TUMORE FIBRE E BUTIRRATO-MEDIATA.

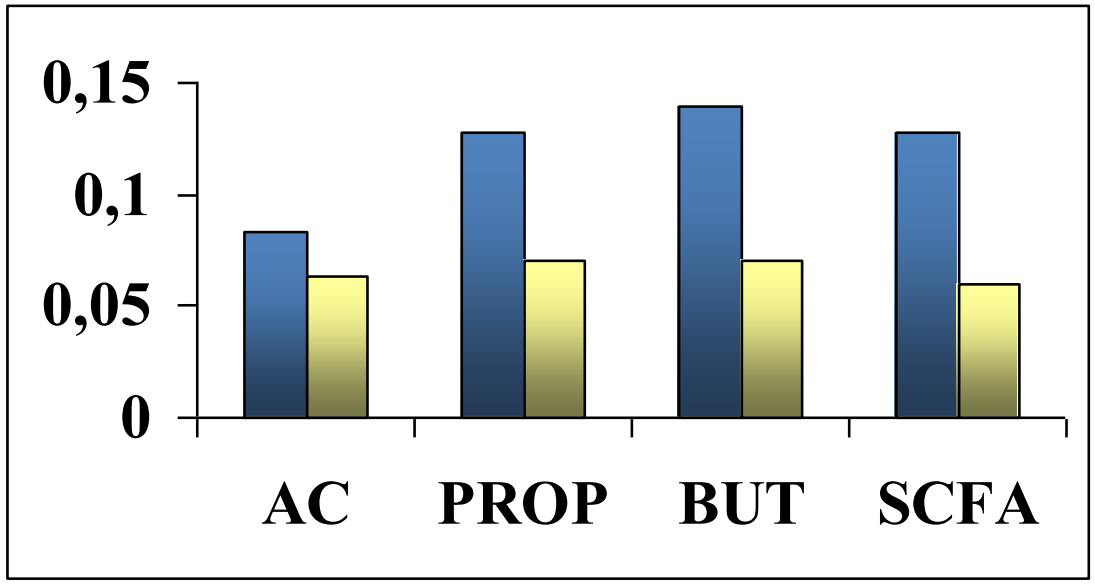


Scott J. Bultman; Interplay between diet, gut microbiota, epigenetic events, and colorectal cancer; Mol. Nutr. Food Res. 2016, 00, 1–12.

Fermentazione dei polisaccaridi nel colon dell'uomo: principali prodotti terminali

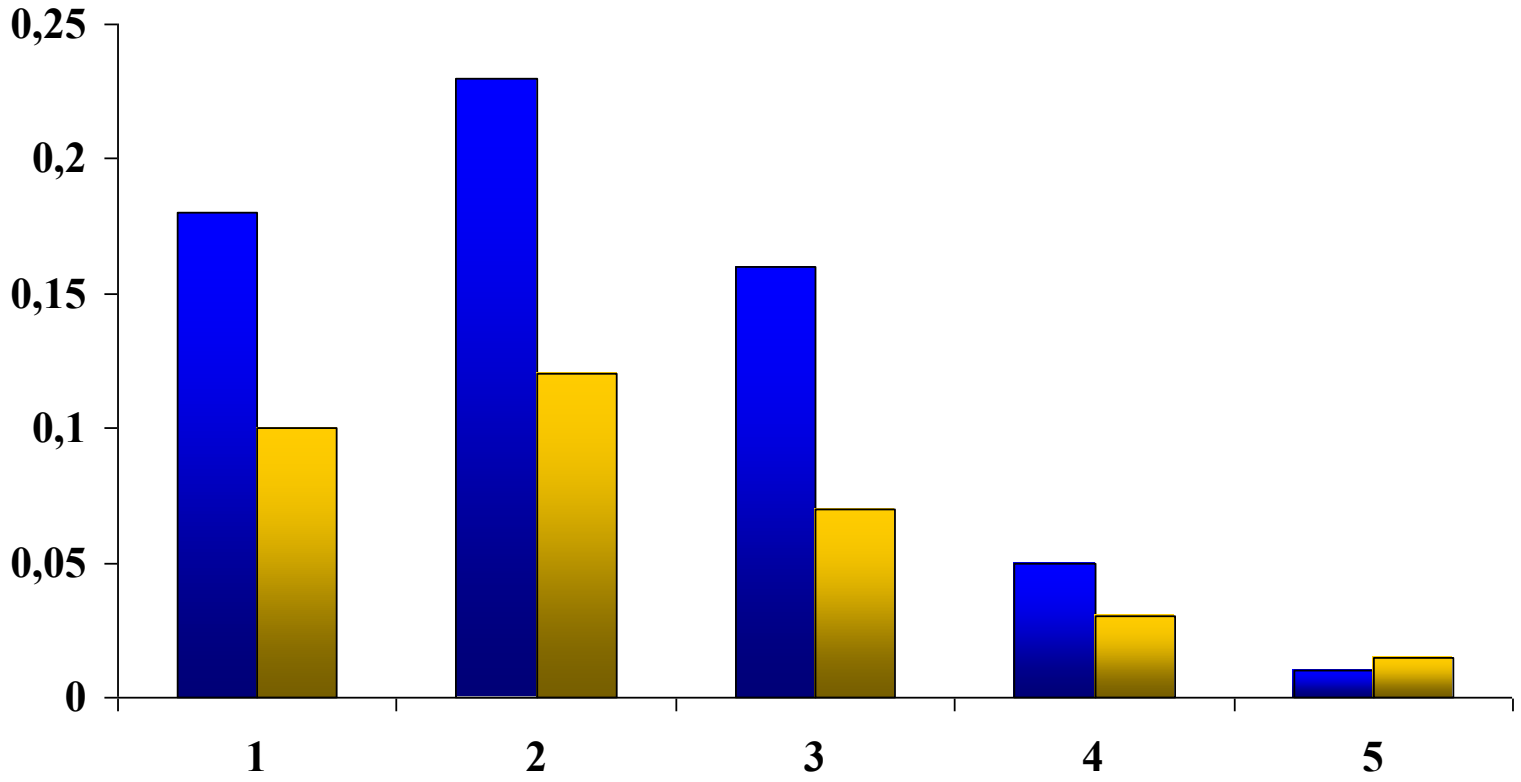






(W. Sheppach MD et al.: Journal of Parenteral and Enteral Nutrition Vol. 16 n°1 92 by ASPEN)

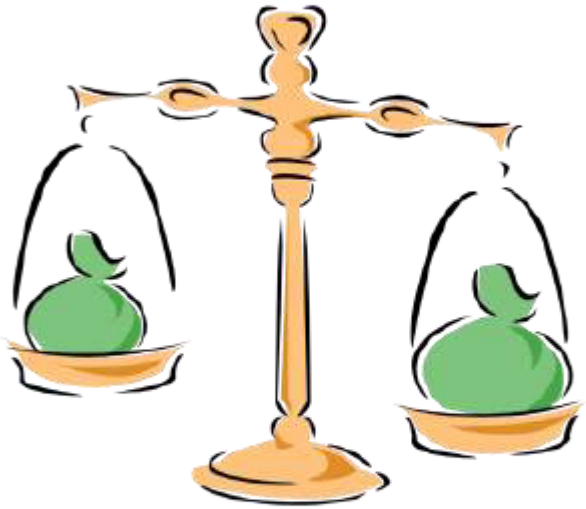
CRYPT COMPARTMENT



(W. Sheppach MD et al.:Journal of Parenteral and Enteral Nutrition Vol. 16 n°1 92 by ASPEN)

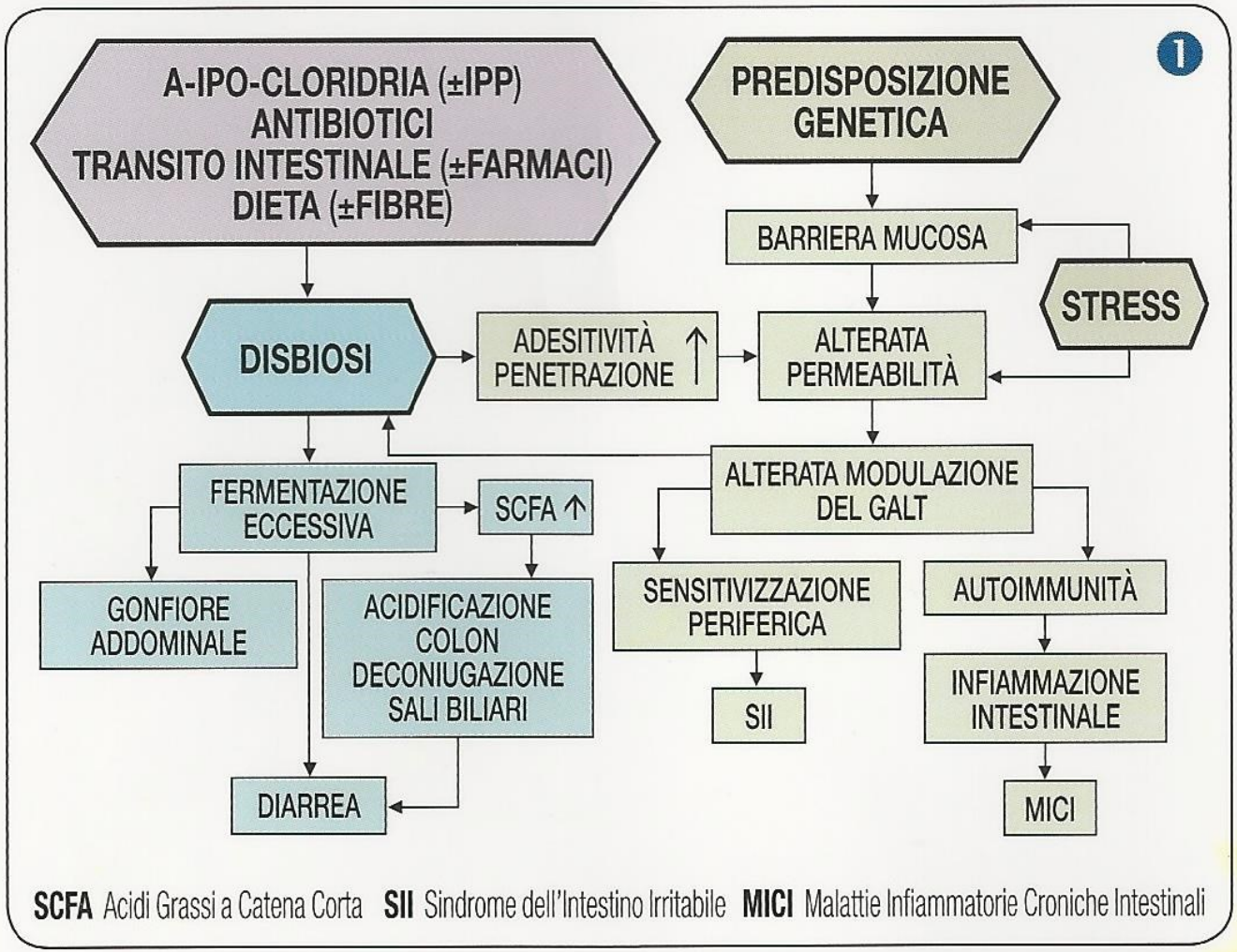
HOW THE HOST-GUT MICROBIOTA BALANCE IS MANTAINED?

- **Secretion of :**
  - ***Gastric acid***
  - ***Mucus***
  - ***Biliary salts***
  - ***Mucosal Ig***



- **Mucosal pH**
- **Mucosal barrier integrity**
- **Intestinal motility**
- **Local mucosal and systemic immunity**
- **Interactions among different bacteria species**

AZIONI DELLA FLORA INTESTINALE IN CONDIZIONI DI DISBIOSI E/O ALTERAZIONE DELLA BARRIERA MUCOSA



SCFA Acidi Grassi a Catena Corta **SII** Sindrome dell'Intestino Irritabile **MICI** Malattie Infiammatorie Croniche Intestinali

ANTIBIOTICI

Impact of antibiotics on the gut microbiota of critically ill patients

Journal of Medical Microbiology (2008), **57**, 1007–1014

Gaetano Iapichino,[1] Maria Luisa Callegari,[2] Silvia Marzorati,[1]

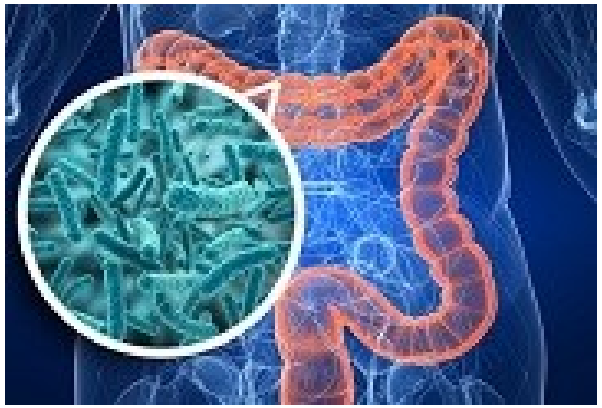
Table 3. Antibiotic therapies and microbiota variations in the patient cohort in this study

Results of RT-PCR are expressed as the amounts of bacterial target genomes of *Enterococcus* (g faeces)$^{-1}$.

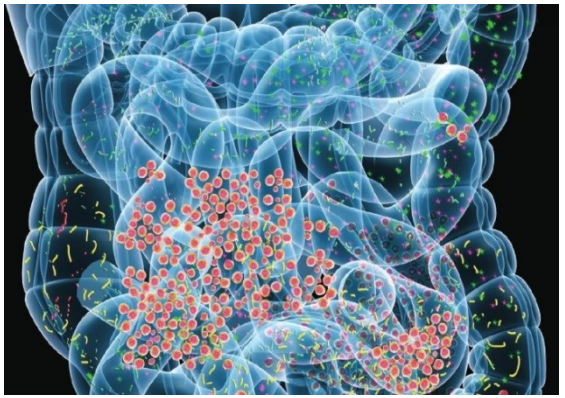
Patient	Antibiotics*	Enterococci in T0	Similarity of DGGE profiles T1 vs T0 (%)	Enterococci in T1	Similarity of DGGE profiles T2 vs T0 (%)	Enterococci in T2
1	CTX, CLI, SDD	3.0E+6	88.0	4.0E+9	–	
2†	AMP, CTX, CLI, SDD	1.5E+5	25.0	6.0E+10	–	
3	CRO, CLR	9.0E+7	75.0	4.45E+7	–	
4	CTX, CLR, SDD	6.7E+6	93.1	7.6E+6	–	
5†	CTX, CLR, SDD	2.0E+6	63.0	9.0E+10	–	
6†	CFZ, CLI	6.7E+6	62.9	1.3E+8	57.4	5.6E+8
7†	CRO, CLR	1.2E+7	34.4	3.4E+8	28.0	5.1E+9
8	CRO, CLR	4.4E+6	90.0	3.0E+6	–	
9	CRO, CLR	7.8E+6	68.0	2.6E+6	–	
10	CRO, CLI	5.0E+6	49.9	2.3E+8	–	
11	CRO, CLR, CLI, OXA, SDD	7.5E+6	33.3	3.0E+7	52.2	1.2E+9
12	AMP, CRO	1.2E+6	84.7	1.5E+6	39.4	3.0E+7
13	CFZ	1.2E+7	82.2	3.0E+7	51.2	2.0E+7
14	CRO, CLR, CLI	8.5E+7	3.8	4.5E+9	10.5	7.9E+10
15	CTX, CLI, SDD	1.2E+6	77.6	3.0E+5	48.7	6.0E+8

*AMP, ampicillin; CFZ, cefazolin; CLI, clindamycin; CLR, clarithromycin; CRO, ceftriaxone; CTX, cefotaxime; OXA, oxacillin; SDD, selective digestive decontamination.

†Died. All deaths were in the ICU.



GUT MICROBIOTA:
disbiosi e patologie correlate



Gut-microbiome and Host

IBD

Obesity

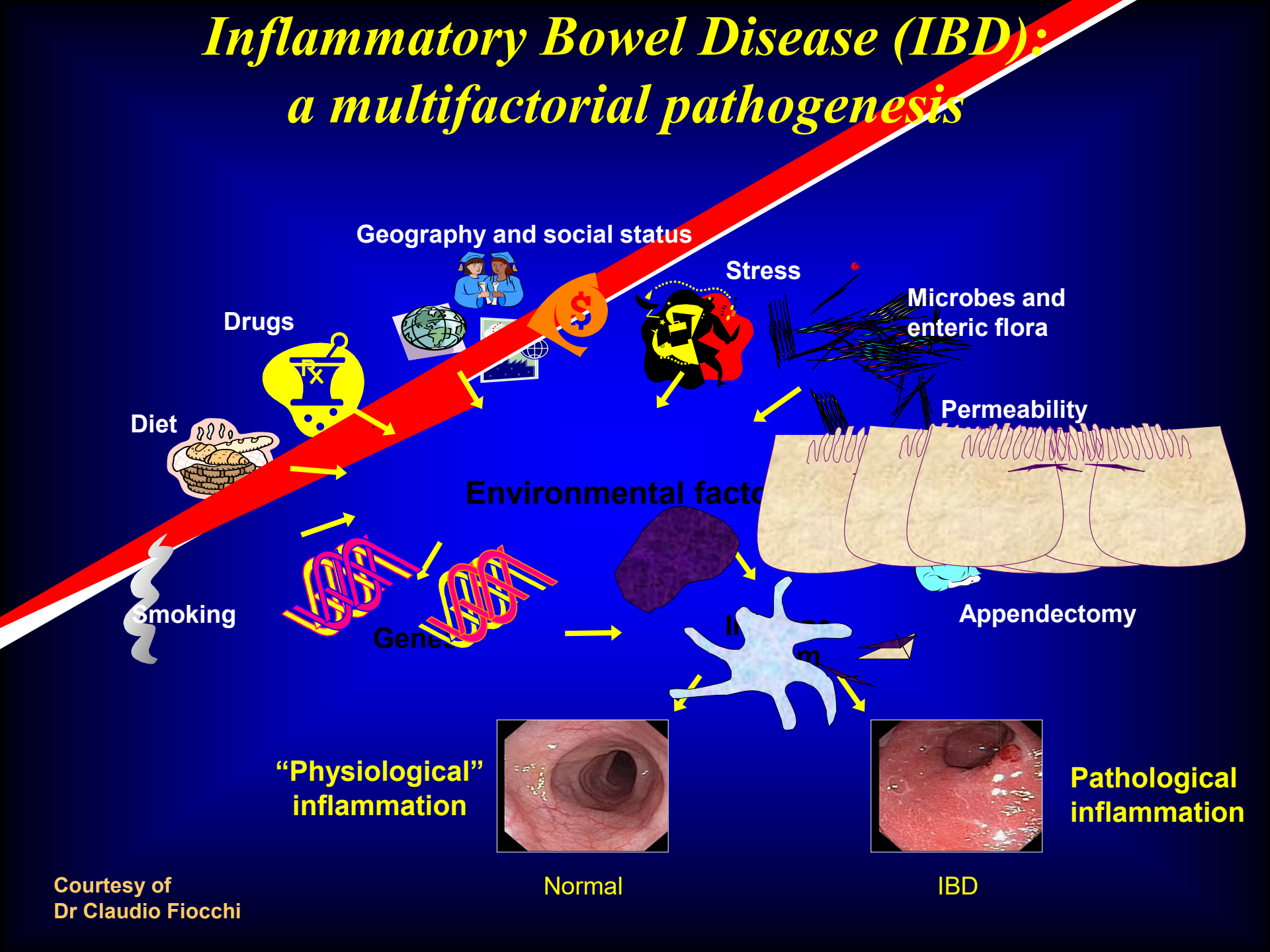
Diabetes Mellitus

NASH

Autoimmunity

Cancer

Inflammatory Bowel Disease (IBD): a multifactorial pathogenesis



Courtesy of
Dr Claudio Fiocchi

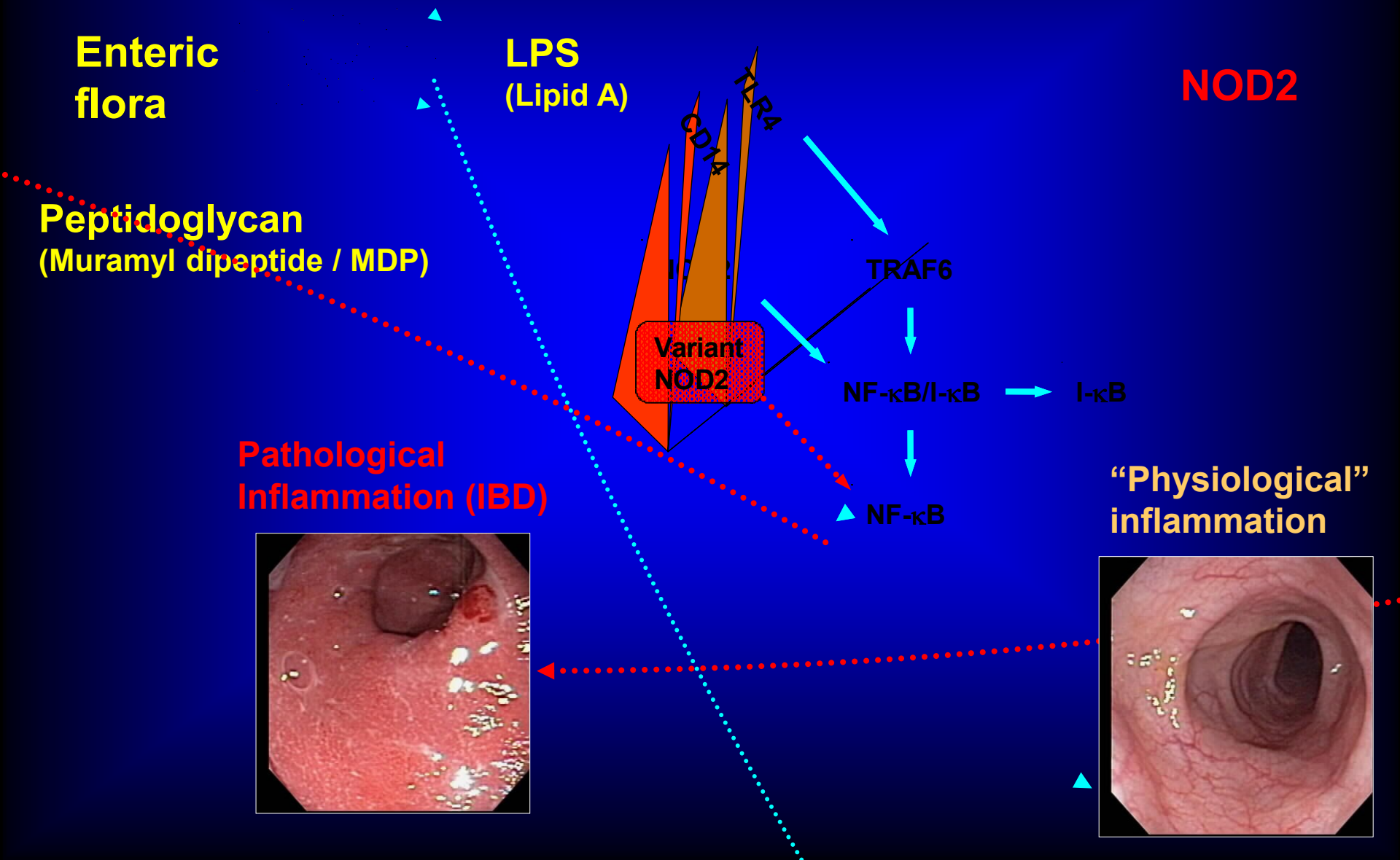
The crucial role of NOD2

Genetic studies confirm a role for the underlying microbial–host interaction in IBD pathogenesis. These include the genomic regions of nucleotide-binding oligomerisation domain containing 2 (NOD2), which is an intracellular receptor that recognizes proteins found in bacterial cell wall species.

Patients with CD demonstrate an association with the NOD2 gene including three polymorphisms that weaken the host peptidoglycan response. Carriers of NOD2 have a 1.75–4-fold increased risk of CD and are clinically more likely to undergo surgical gastrointestinal (GI) resection.

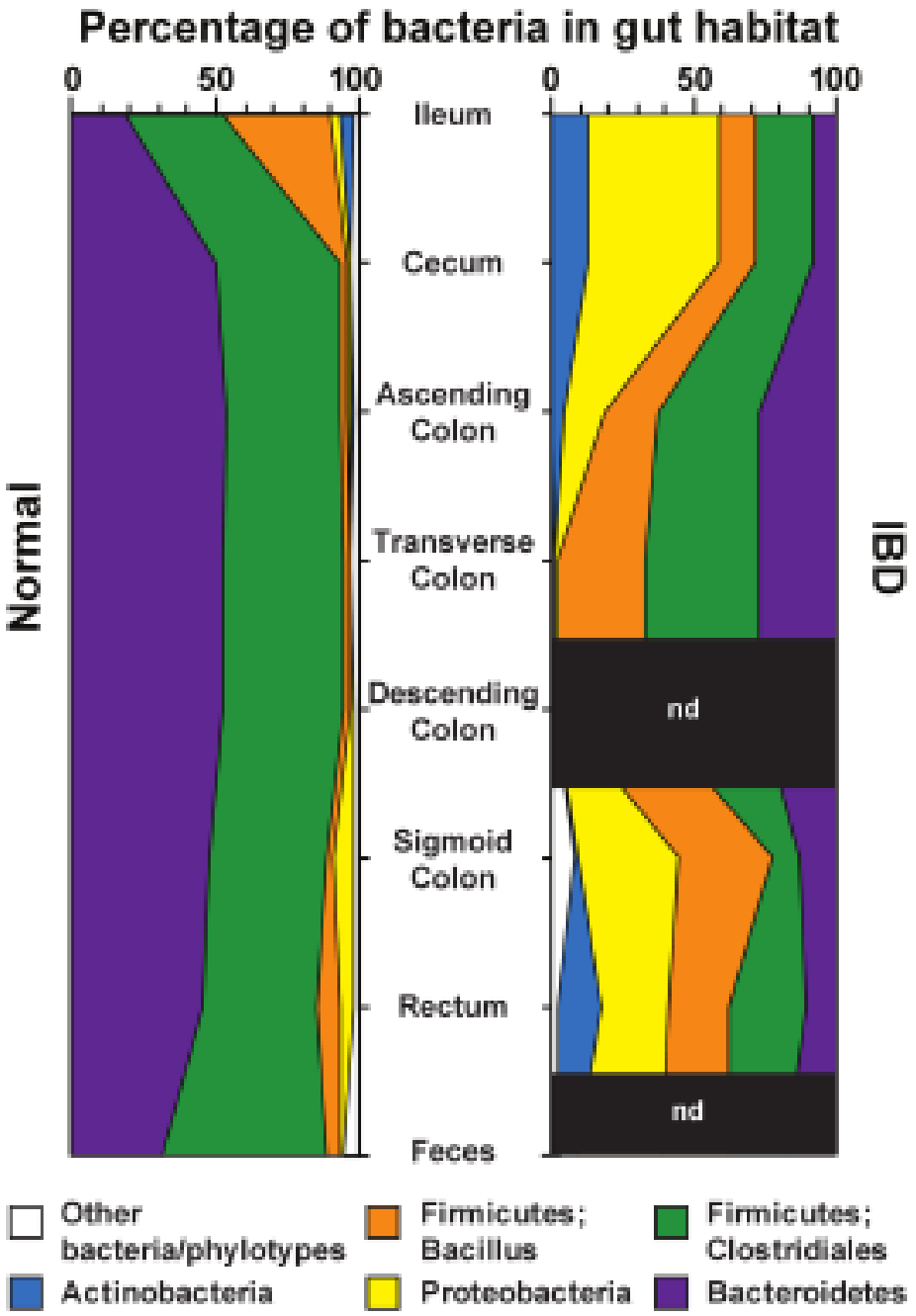
Holmes et al, Trends in Microbiology, July 2011

NOD2 DEFICIENCES INCREASE INFLAMMATORY RESPONSE TO LPS STIMULATION



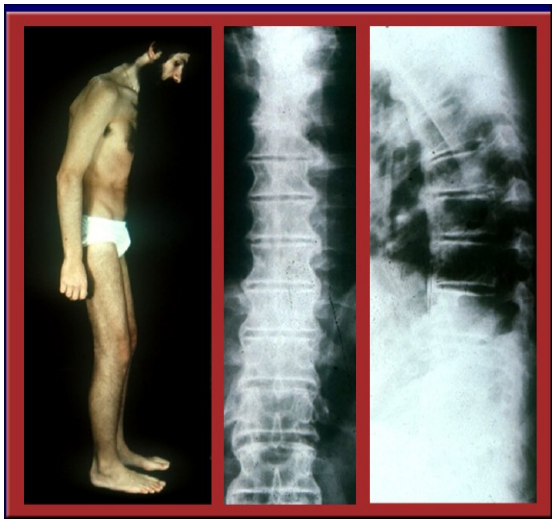
- **Decrease in beneficial Firmicutes (clostridia sp)**
  - Species able to indice immune tolerance and reduce colitis in animal models
  - Reduction may permit hyper-immune response

- **Increase in Proteobacteria**
  - Generally aerotolerant
  - Organisms able to manage oxidative stress
  - Injurious in animal models of intestinal inflammation



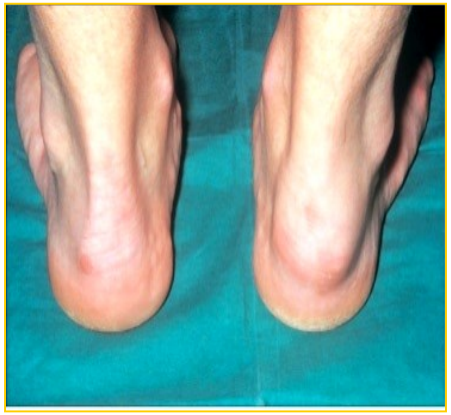
Cell Host Microbe. 2008 June 12; 3(6): 417–427. doi:10.1016/j.chom.2008.05.001.

Spondilite anchilosante



Immagini gentilmente concesse da J Braun, MD

Entesite

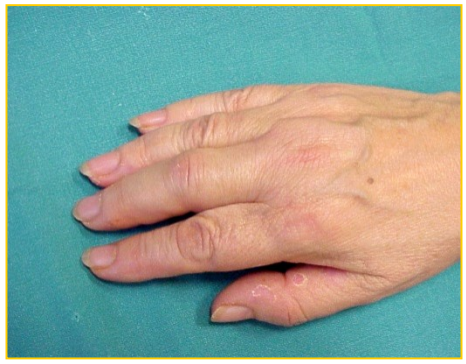


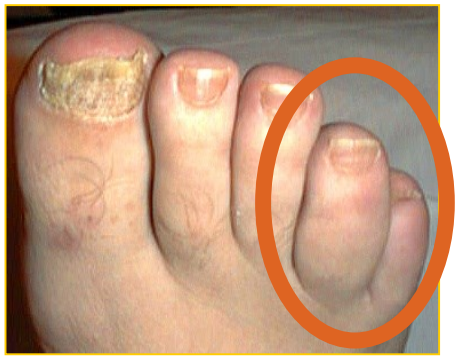
Questo è il segno distintivo delle spondiloartropatie

Frequenza di entesiti sintomatiche: 5-30% delle SpA

Predominante coinvolgimento entesale in alcuni casi

Dattilite





Fortemente associata a tenosinovite come evidenziato da MRI[1]

Due sottotipi

acuta: rigonfiamento morbido, forte dolore, rossore

cronica: rigonfiamento solido, dolore medio-moderato,

Frequenza: 5-30% delle SpA

Gut-microbiome and Host

IBD

Obesity

Diabetes Mellitus

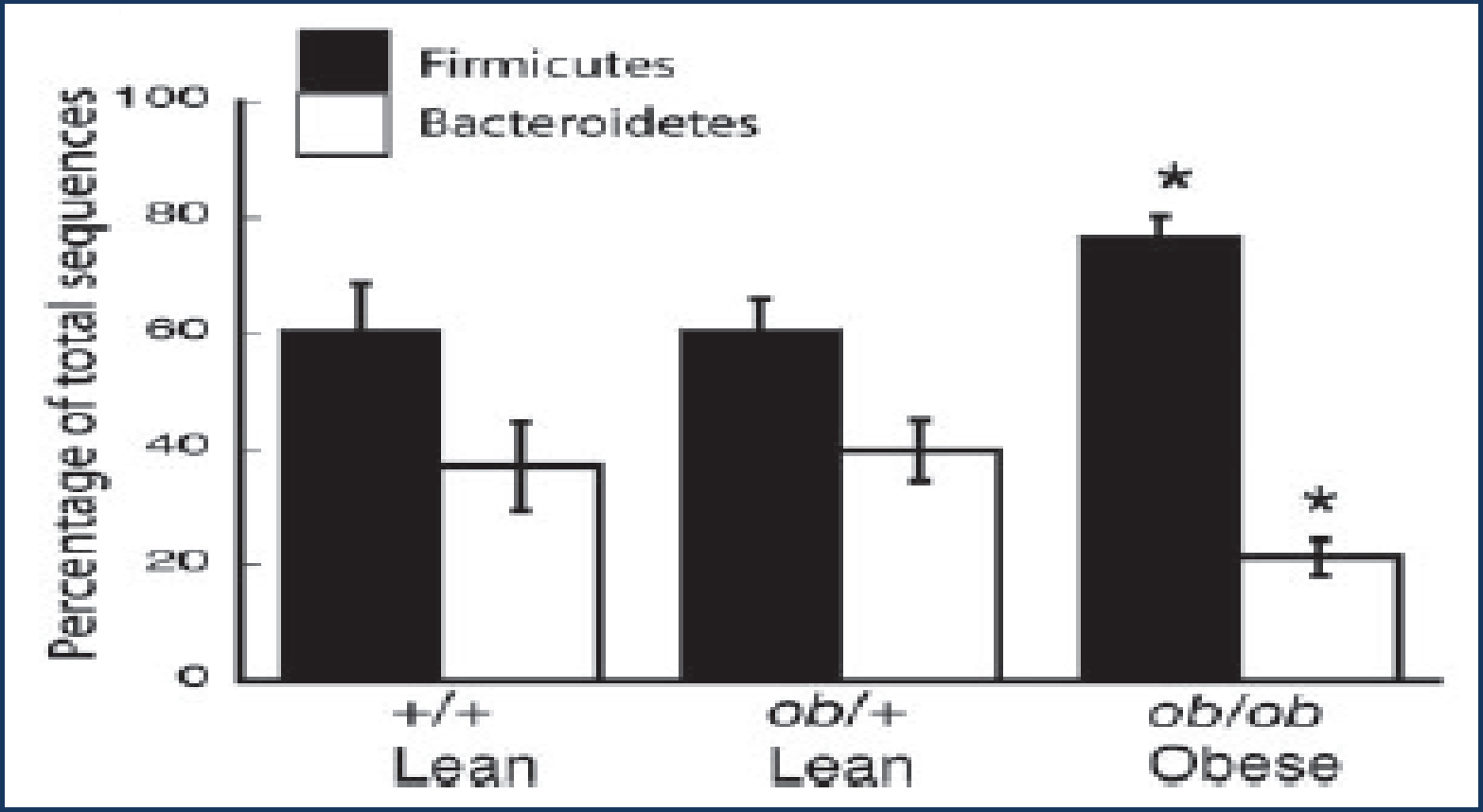
NASH

Autoimmunity

Cancer

GUT MICROBIOTA AND OBESITY: THE DYSBIOSIS CONCEPT

Obese animals have a 50% reduction of Bacteroidetes and a proportional increase in Firmicutes



Ley – PNAS 2005

Microbial metabolic production

✓ **Microbial signals also regulate Fiaf release from intestinal epithelial cells, which acts as an inhibitor of Lpl and thereby regulates peripheral fat storage**

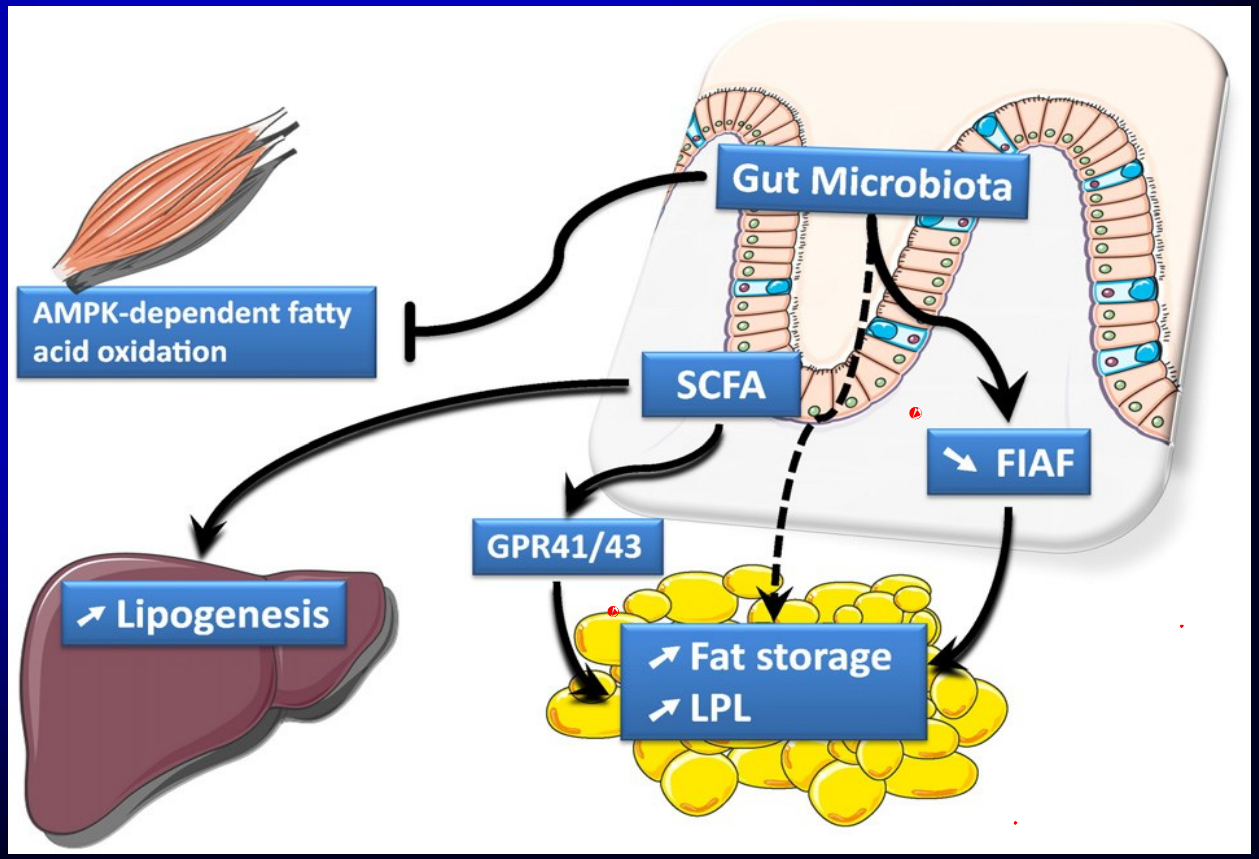
✓ **Through another unknown mechanism, the microbiota also regulates the energy gauge in the liver and muscle through the phosphorylation of Ampk. Glp2 ascertains epithelial barrier function**

✓ **A leaky barrier leads to exposure and activation of myeloid cells in response to microbial signals such as the Tlr4 ligand endotoxin.**

Tilg H. *J Clin Invest. 2011;121(6):2126–2132*

GUT MICROBIOTA & REGULATION OF ENERGY STORAGE

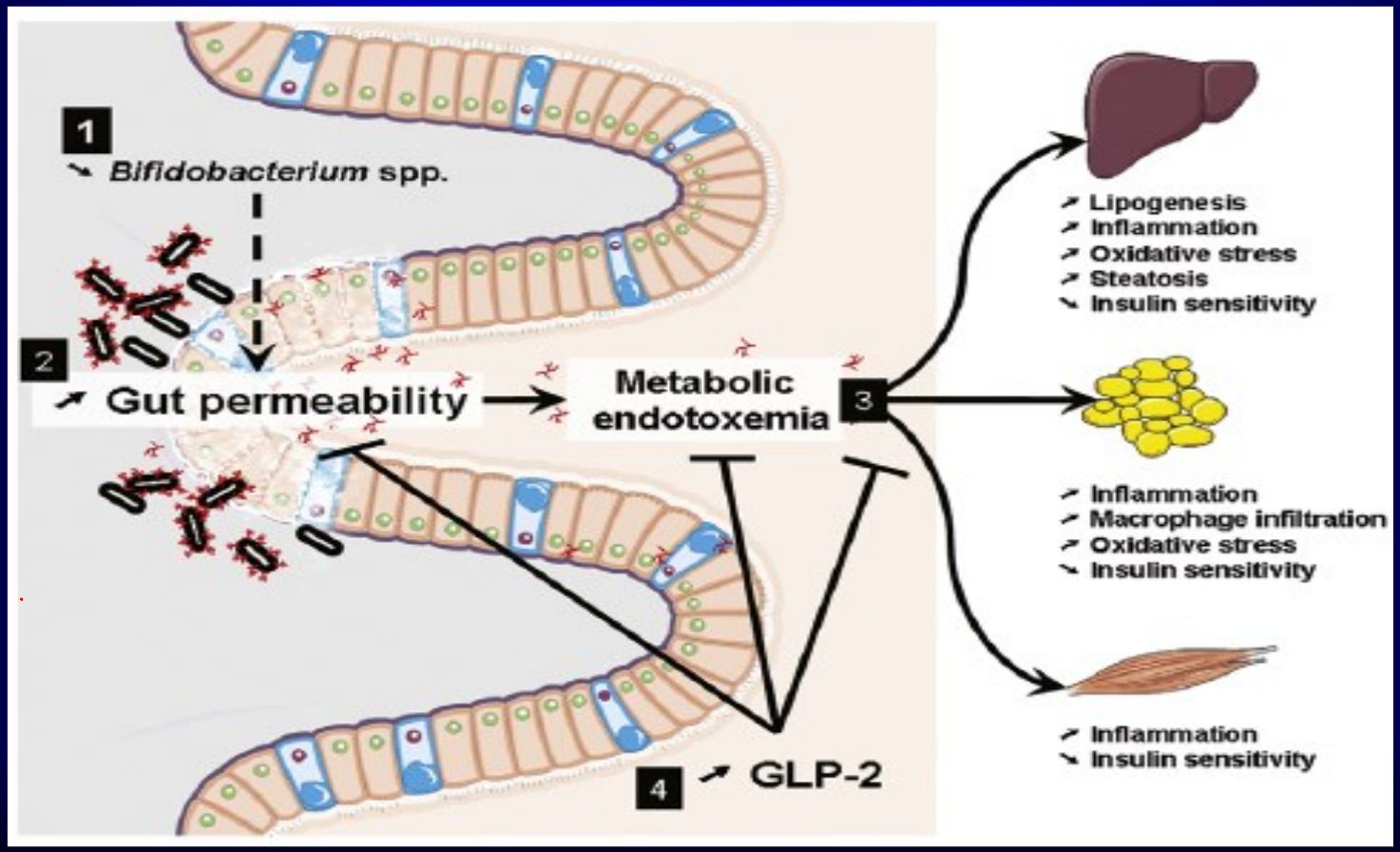
- ↓ *Suppression of Fast Induced Adipose Factor*
- *Stimulation LPL / inibition LPL inhibitor*
- ↑ *Increased hepatic lipogenesis and fat storage in adipocytes*



Backhed et al – PNAS 2004
Cani PD et al – Pharm Ther – in press

GUT MICROBIOTA and REGULATION OF ENERGY STORAGE

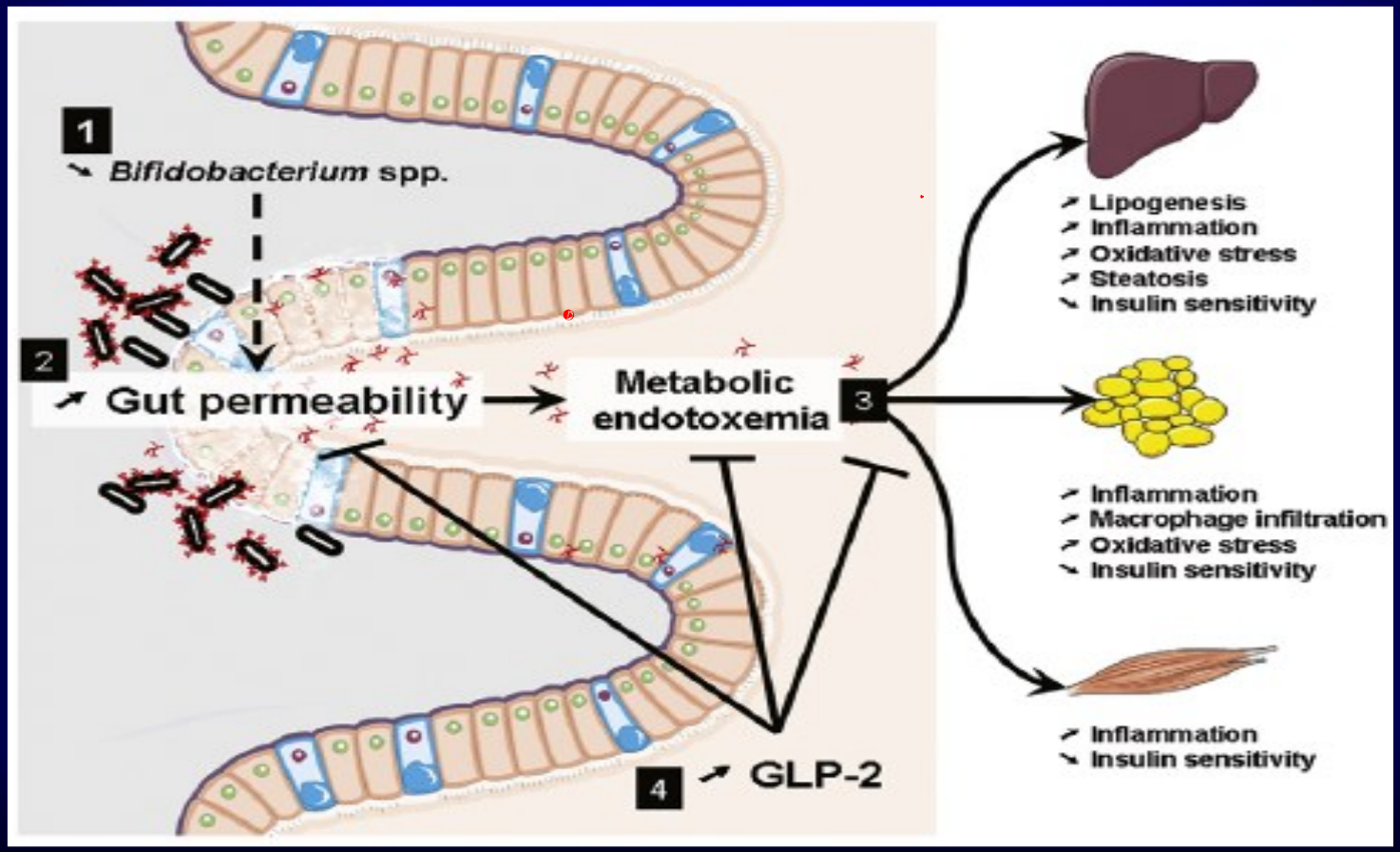
Changes the gut microbiota is associated with a higher gut permeability leading to a higher plasma LPS levels (metabolic endotoxemia)



Cani PD et al – Pharm Ther 2010

GUT MICROBIOTA and REGULATION OF ENERGY STORAGE

Metabolic endotoxemia promotes low-grade inflammation-induced metabolic disorders (insulin resistance, diabetes, obesity, steatosis, oxidative stress, and adipose tissue macrophage infiltration).

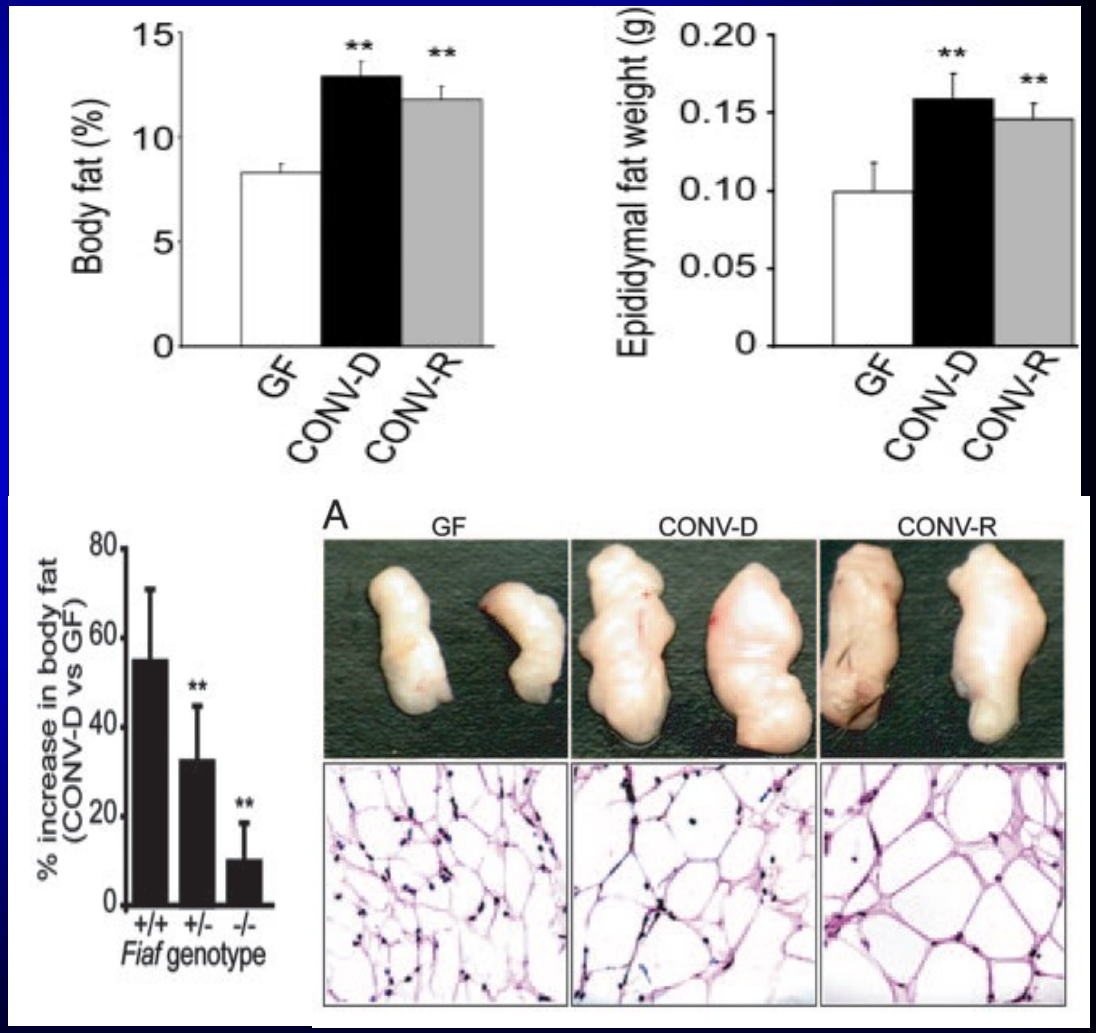


Cani PD et al – Pharm Ther 2010

GUT MICROBIOTA and REGULATION OF ENERGY STORAGE

- WT mice have 42% more total body fat and 47% more gonadal fat than germ-free (GF) mice, despite lower energy intake

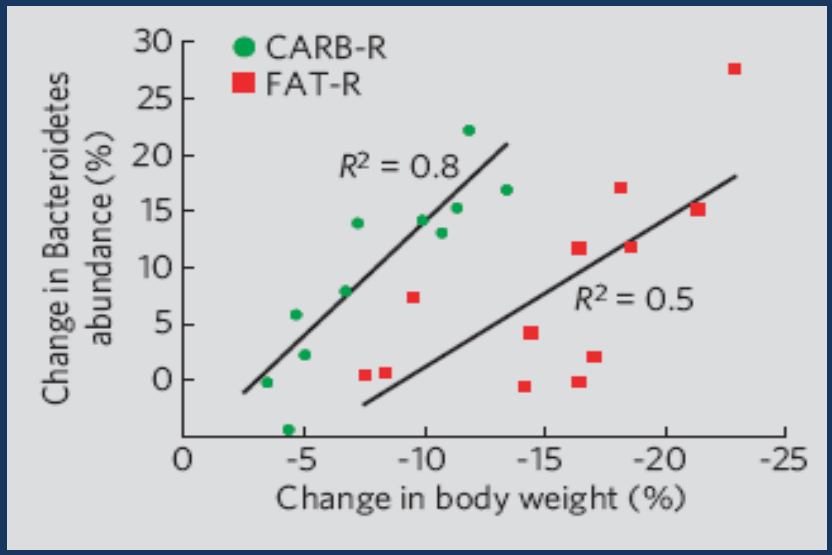
- Colonisation of GF mice with microbiota from control mice produces a 60% increase in body fat mass, associated with increased insulin resistance



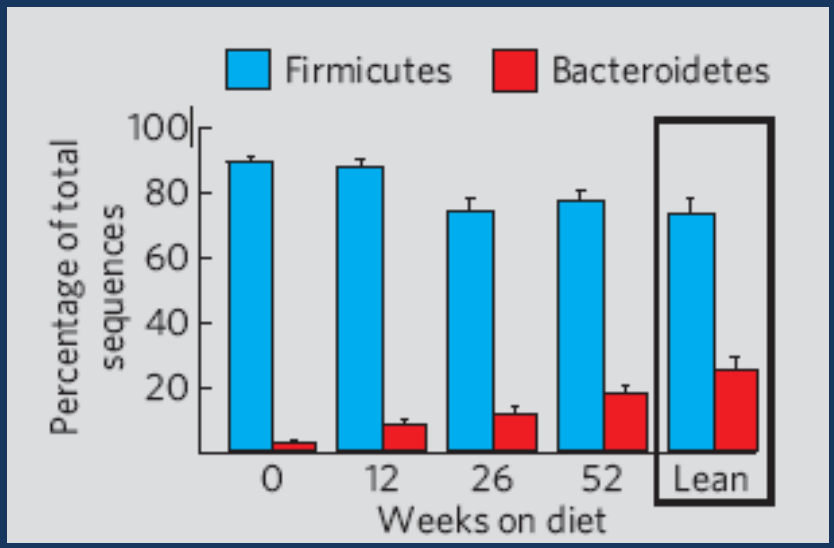
Backhed et al – PNAS 2004

GUT MICROBIOTA AND OBESITY: THE DYSBIOSIS CONCEPT

- Obese people had lower Bacteroides and more Firmicutes in distal gut flora than lean control



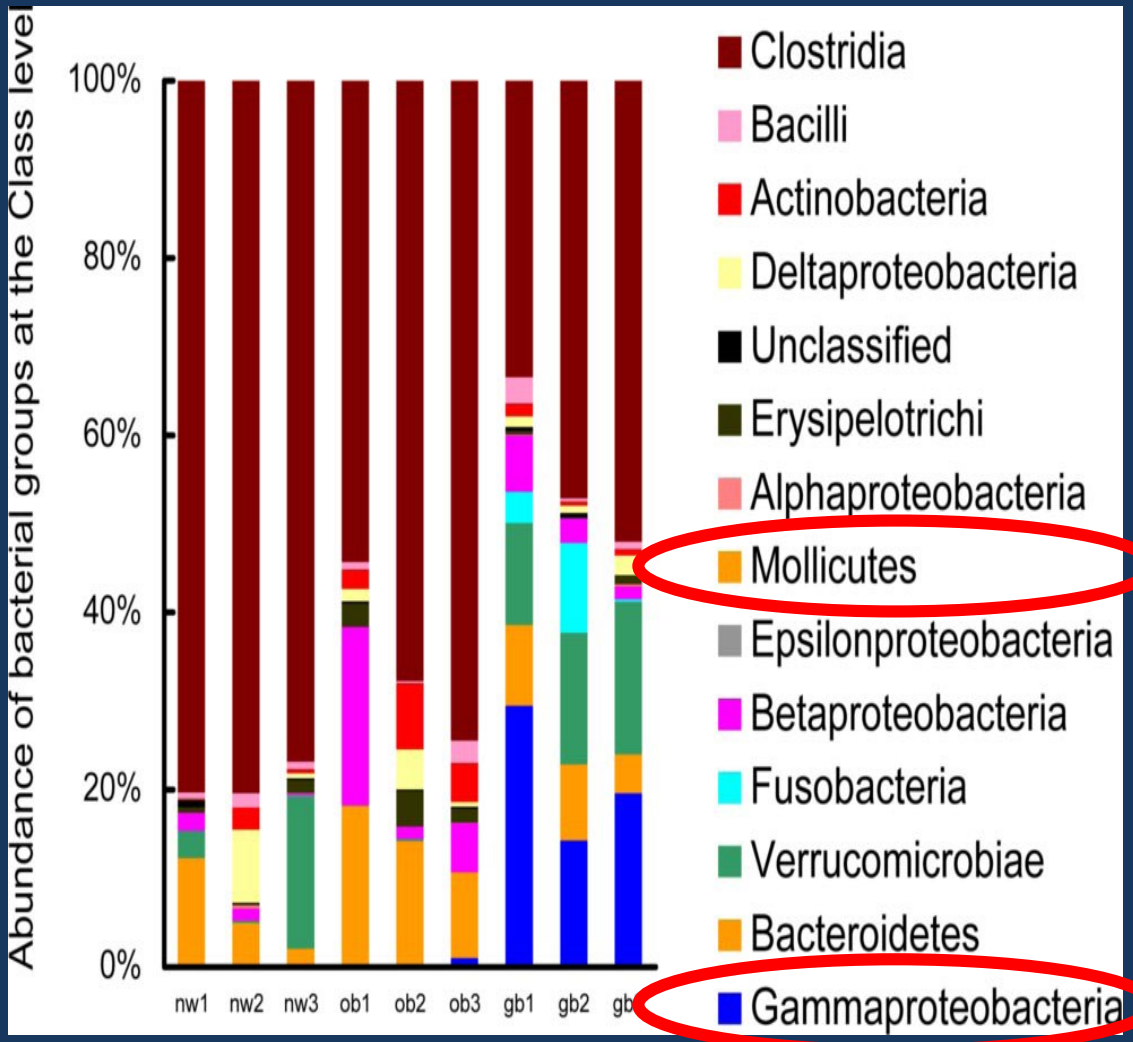
- Ratio of Bacteroidetes to Firmicutes approached a lean type profile after 52 weeks of diet-induced weight loss



Ley et al– Nature 2006

GUT MICROBIOTA AND OBESITY: THE DYSBIOSIS CONCEPT

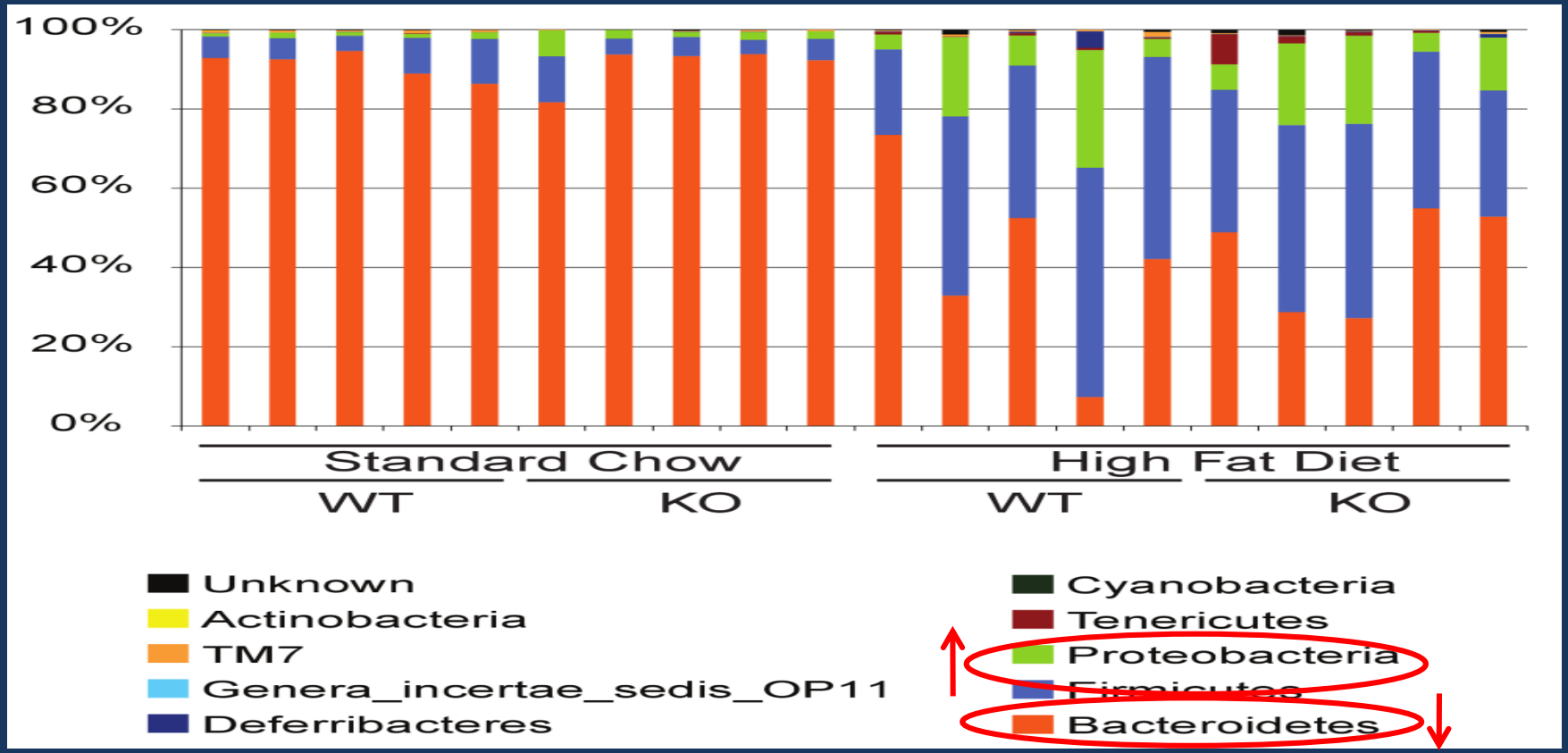
Increase in Gammaproteobacteria and proportional decrease in Firmicutes after bariatric surgery (gastric by-pass)



Zhang et al PNAS 2009

GUT MICROBIOTA AND OBESITY: THE DYSBIOSIS CONCEPT

Switching mice to a High-Fat-Diet drives to a decrease in Bacteroides and an increase in both Firmicutes and Proteobacteria indipendently of obesity



Hildebrand- Gastroenterology 2009

GUT MICROBIOTA AND OBESITY: THE DYSBIOSIS CONCEPT

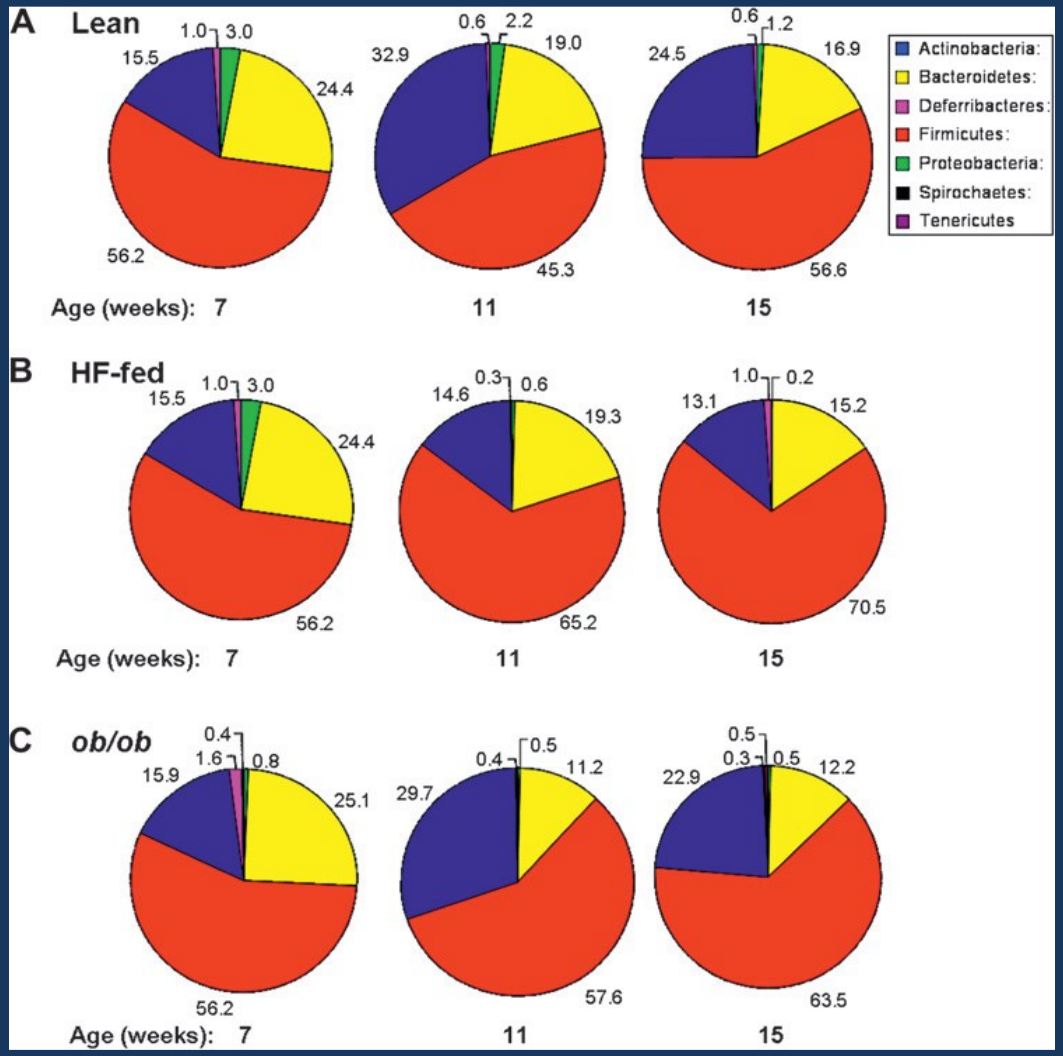
A- WT mice/Low Fat diet

B- WT mice/High Fat diet — **8 wks**

C- Ob/ob mice/ LF diet

➢Increase in Firmicutes in both HF-fed and ob/ob mice ($p<0.05$)

➢Reductions in Bacteroides in ob/ob mice ($p<0.001$)



Murphy – Gut 2010

THE CORE GUT MICROBIOME

•No single bacterial phylotype was detectable at an abundant frequency

•The hypothesis of a core human gut microbiome may be incorrect

• A core gut microbiome exists at the level of metabolic functions

•Understanding the underlying principles should provide insights about microbial adaptation to a wide range of environments

Turnbaugh – Nature 2009

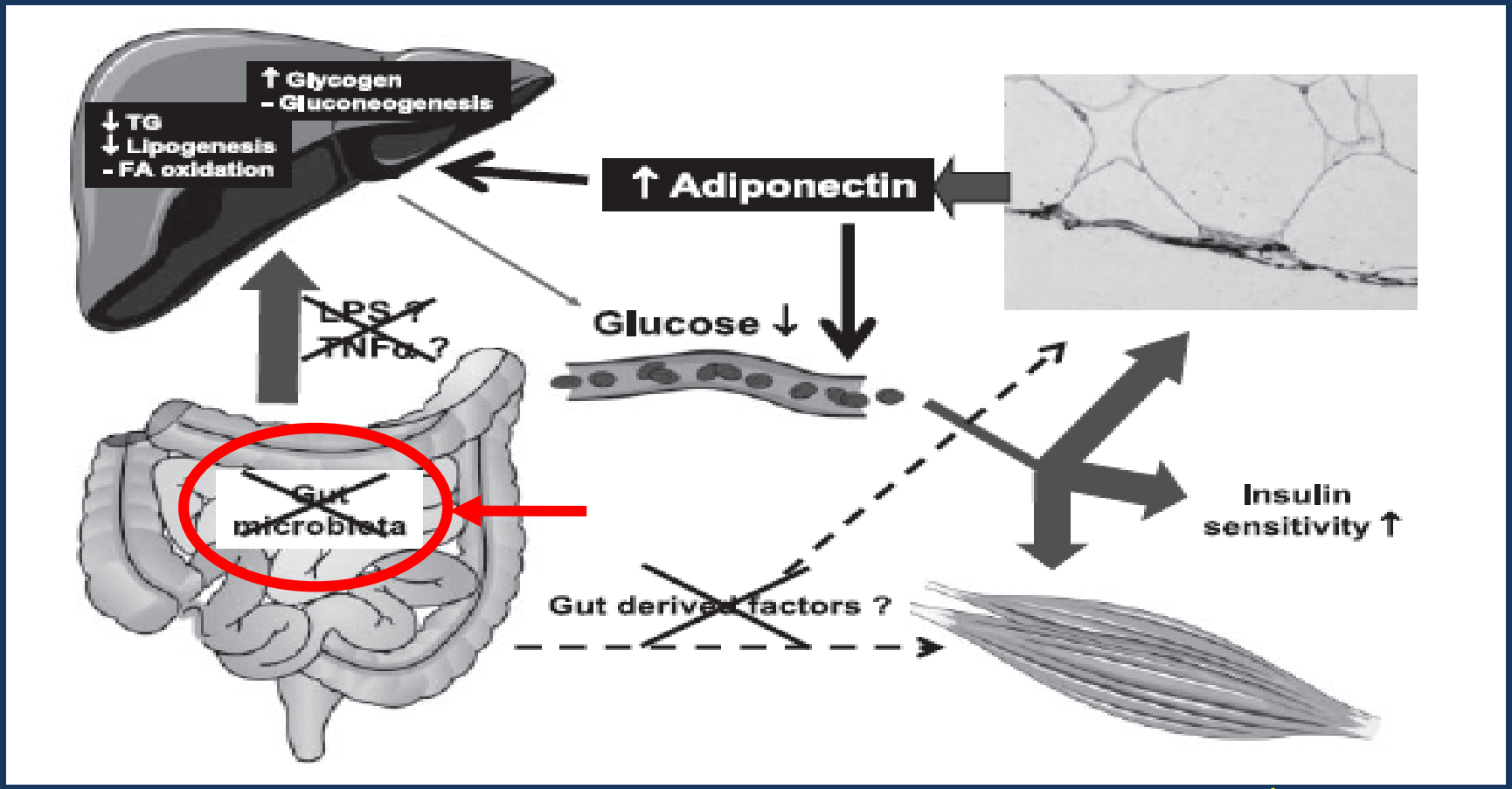
Gut-microbiome and Host

- ✓ IBD
- ✓ Obesity
- ✓ Diabetes Mellitus
- ✓ NASH
- ✓ Immune system
- ✓ Cancer

GUT MICROBIOTA in TYPE II DIABETES

Gut microbiota modulation improves glucose tolerance by :

reducing the population of gut microbiota

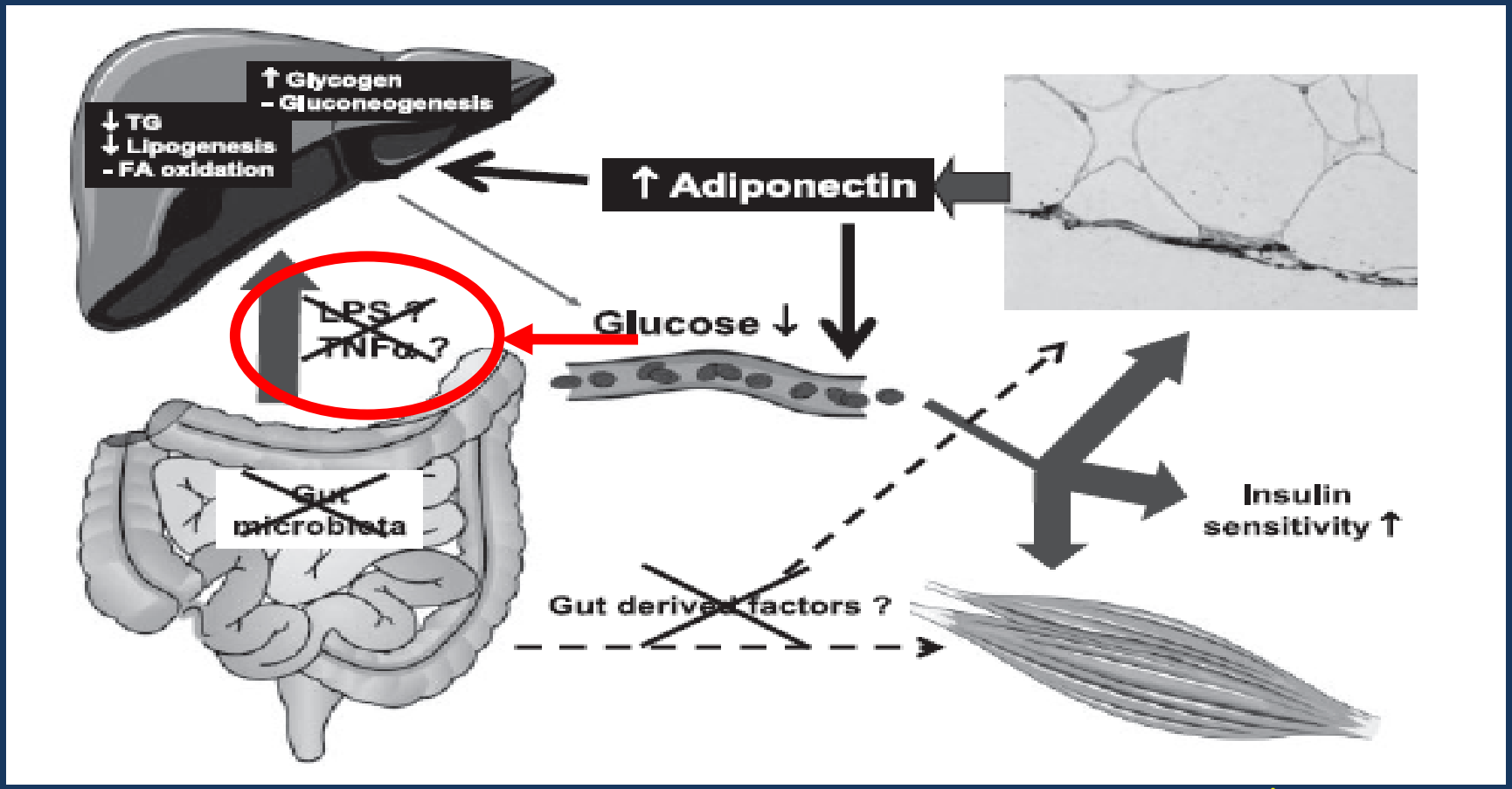


Membrez-FASEB 2008

GUT MICROBIOTA in TYPE II DIABETES

Gut microbiota modulation improves glucose tolerance by :

suppressing factors like TNF and LPS

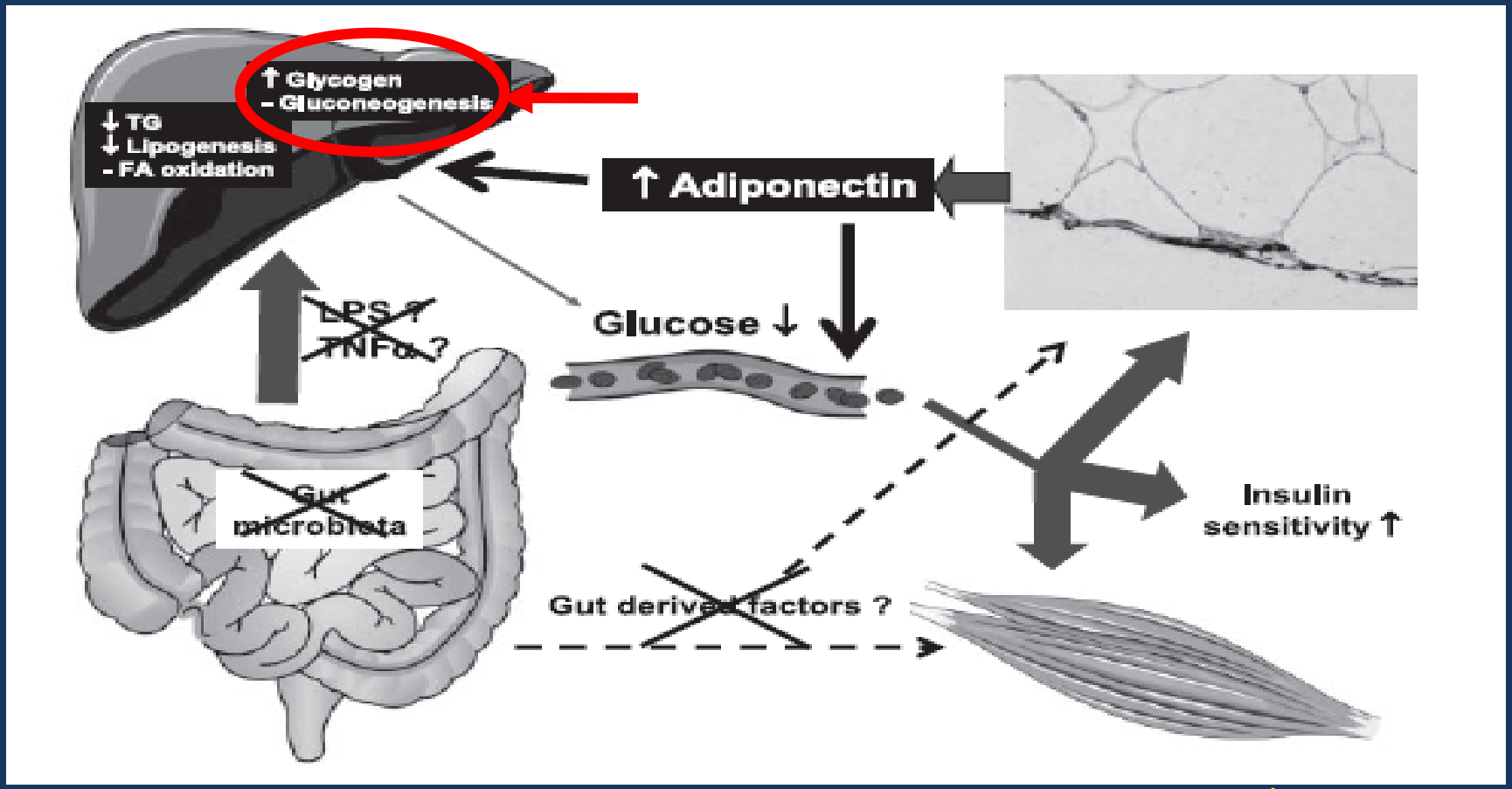


Membrez-FASEB 2008

GUT MICROBIOTA in TYPE II DIABETES

Gut microbiota modulation improves glucose tolerance by :

increasing glycogen storage

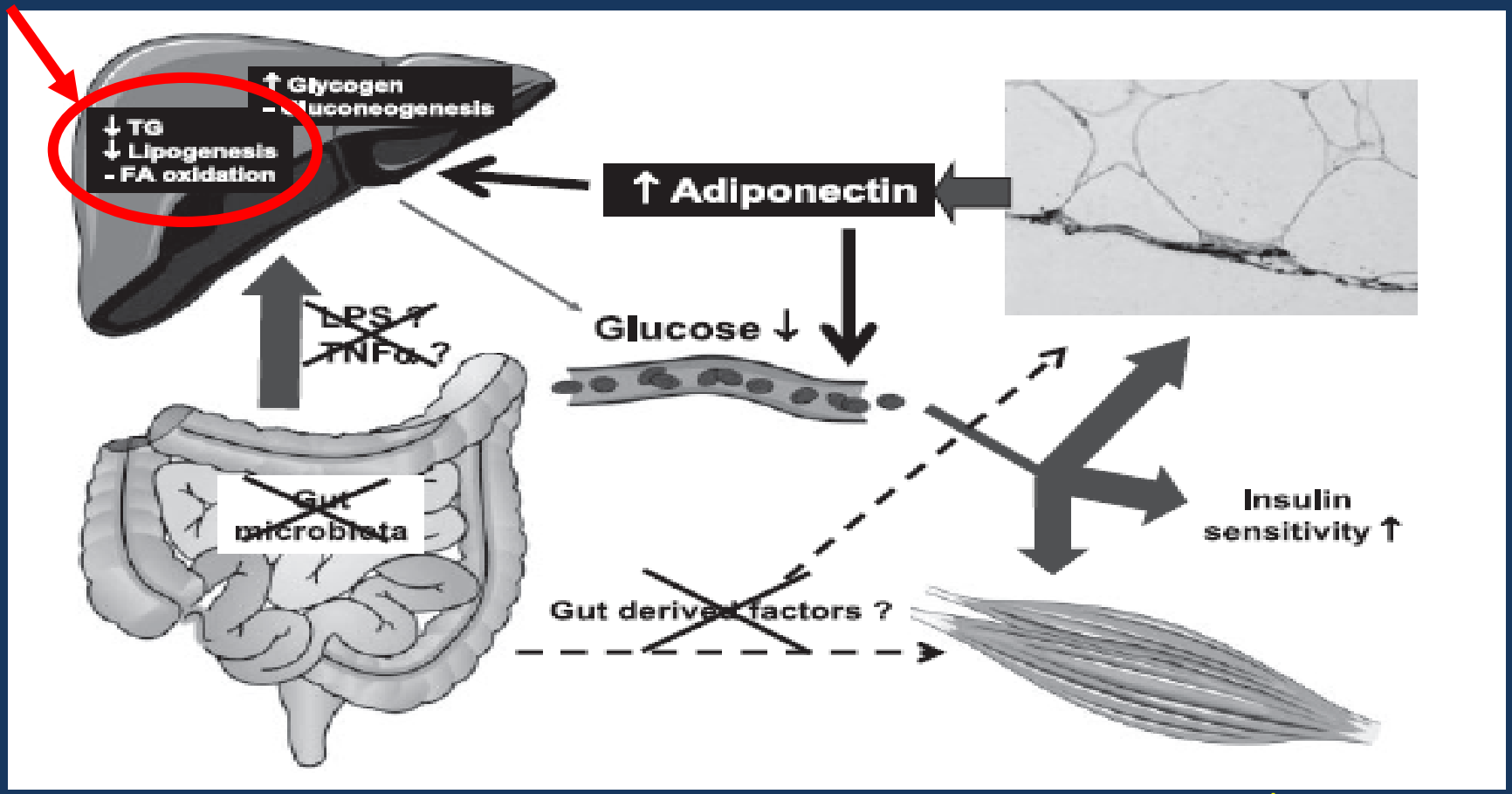


Membrez-FASEB 2008

GUT MICROBIOTA in TYPE II DIABETES

Gut microbiota modulation improves glucose tolerance by :

decreasing triglyceride accumulation by the liver

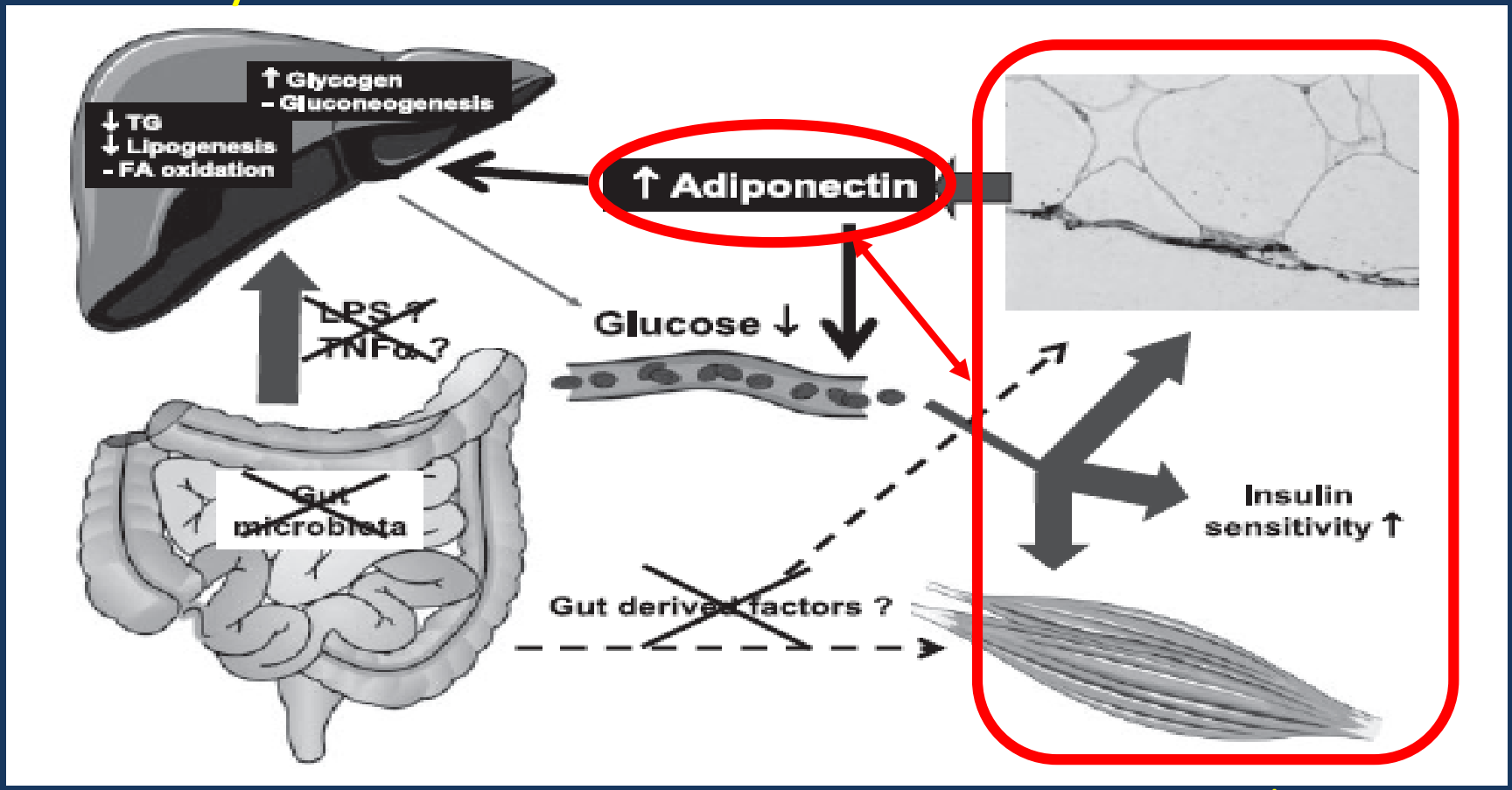


Membrez-FASEB 2008

GUT MICROBIOTA in TYPE II DIABETES

Gut microbiota modulation improves glucose tolerance by :

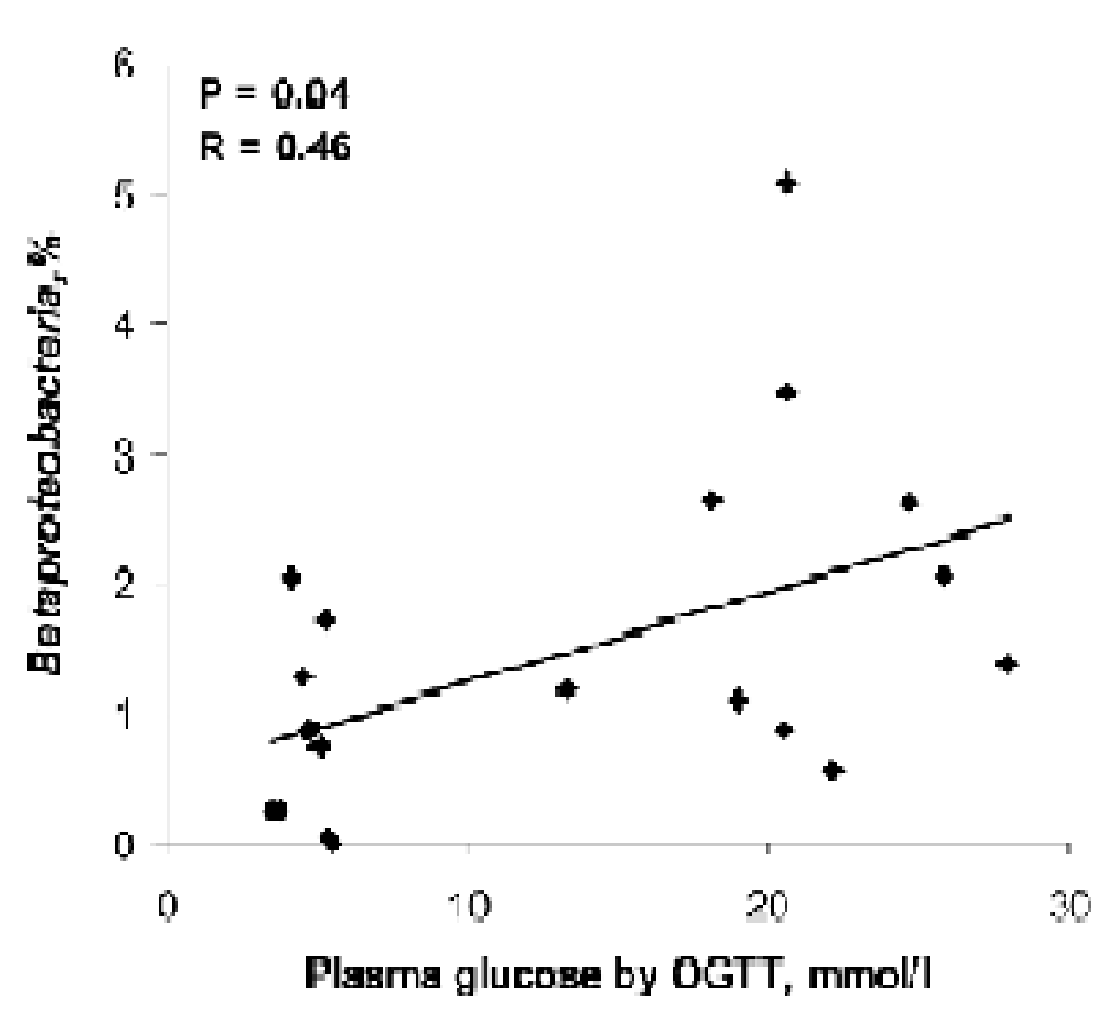
High adiponectin levels might further enhance the insulin sensitivity



Membrez-FASEB 2008

TYPE 2 DIABETES IS ASSOCIATED WITH COMPOSITIONAL CHANGES IN GUT MICROBIOTA.

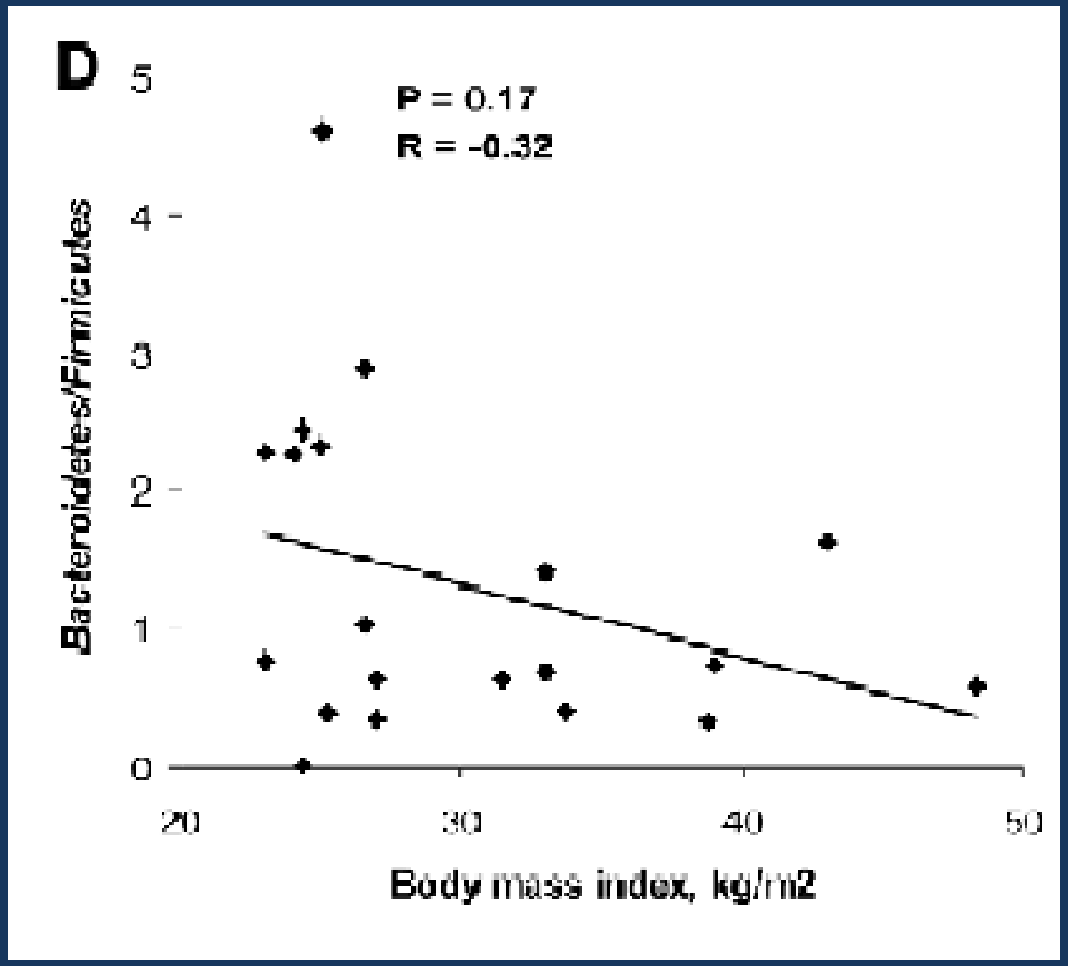
Betaproteobacteria was highly enriched in diabetic compared to non-diabetic persons (P = 0.02) and positively correlated with plasma glucose (P = 0.04)



Larsen et al PLOS ONE 2010

TYPE 2 DIABETES IS ASSOCIATED WITH COMPOSITIONAL CHANGES IN GUT MICROBIOTA.

Ratios of Bacteroidetes to Firmicutes as well as the ratios of Bacteroides-Prevotella group to C. coccoides-E. rectale group correlated positively and significantly with plasma glucose concentration (P = 0.04) but not with BMIs



Larsen et al PLOS ONE 2010

Gut-microbiome and Host

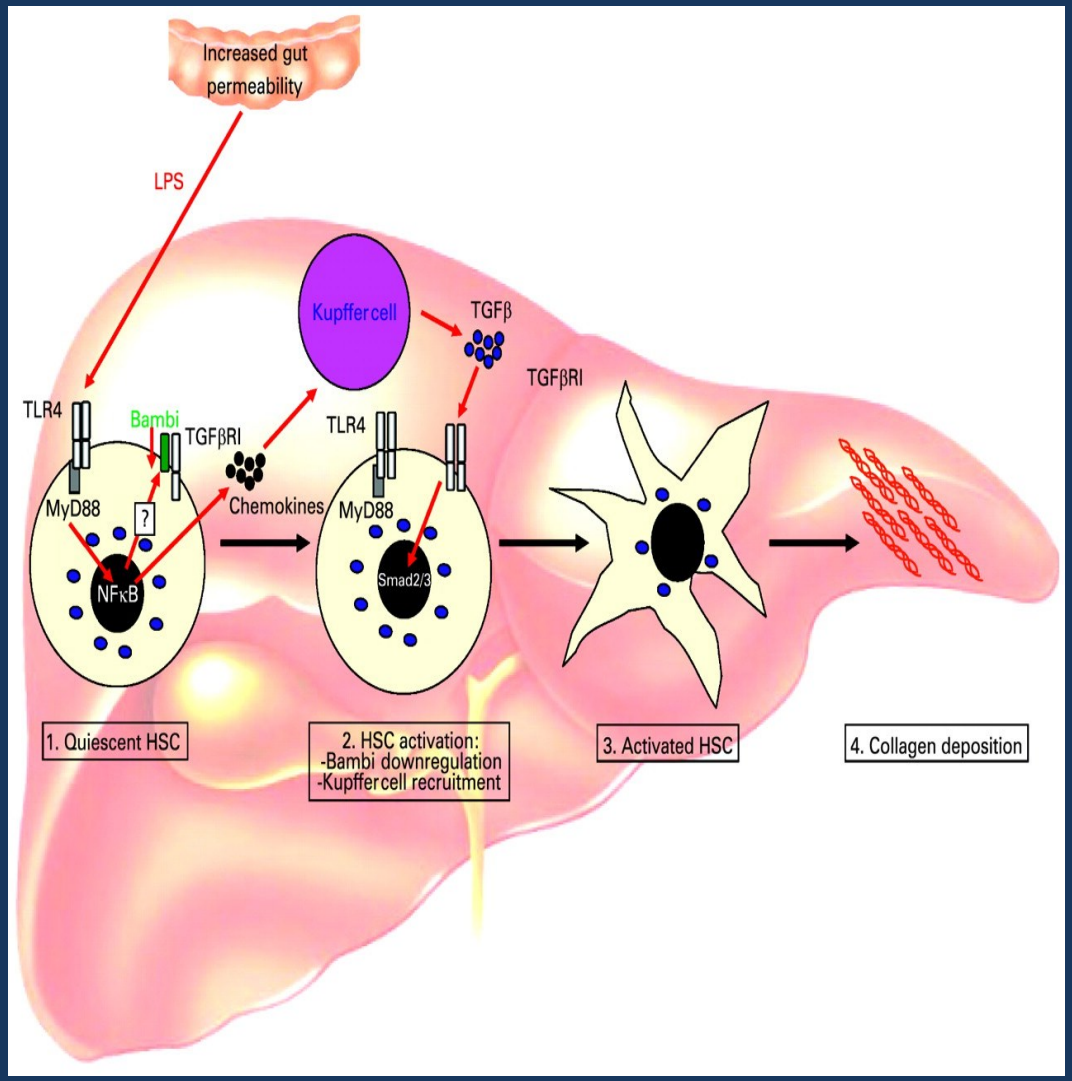
- ✓ IBD
- ✓ Obesity
- ✓ Diabetes Mellitus
- ✓ NASH
- ✓ Autoimmunity
- ✓ Cancer

Consequences of the altered microbial/metabolic balance

NAFLD and NASH

NAFLD and NASH are strongly linked to obesity, type 2 diabetes mellitus and the metabolic syndrome and, accordingly, have become common worldwide problems

Small intestinal bacterial overgrowth of Gram-negative organisms could promote insulin resistance, increase endogenous ethanol production and induce potential mediators of this association, lipopolysaccharide



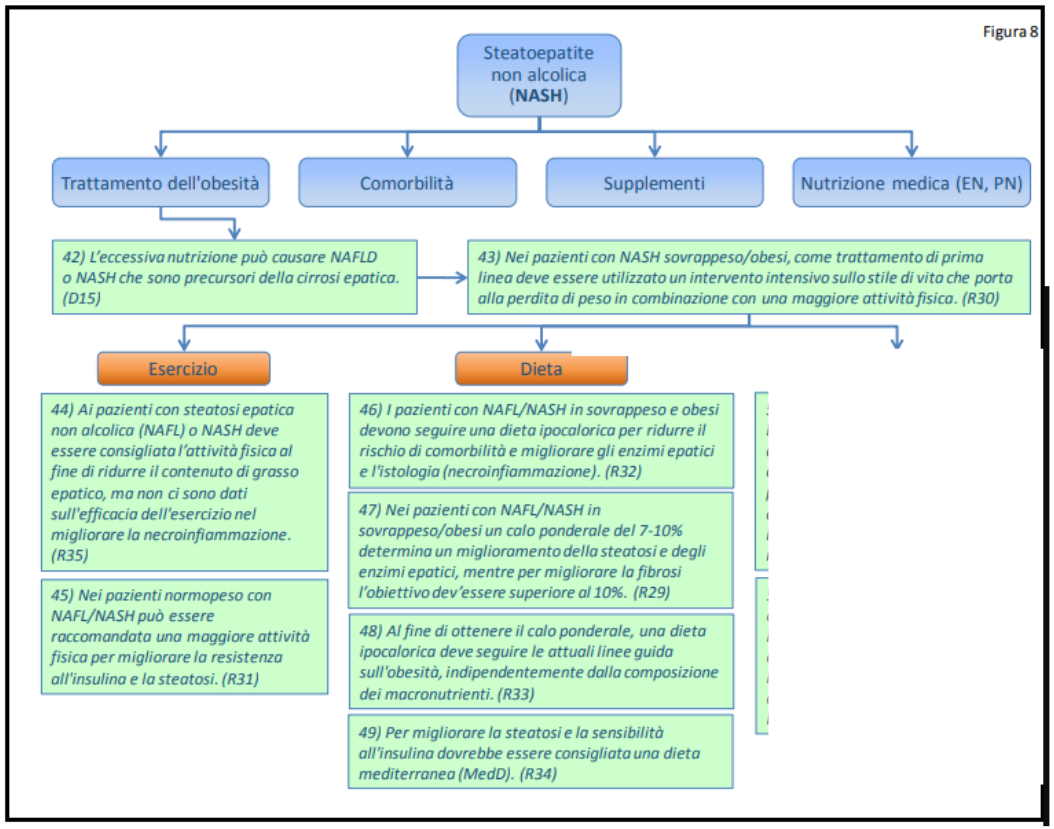
Abu-Shanab A, Nat Rev Gastroenterol Hepatol. 2010

ESPEN
European Society for Clinical Nutrition and Metabolism

SINPE

2.3 Steatoepatite non alcolica (NASH) (Figure 8 e 9)

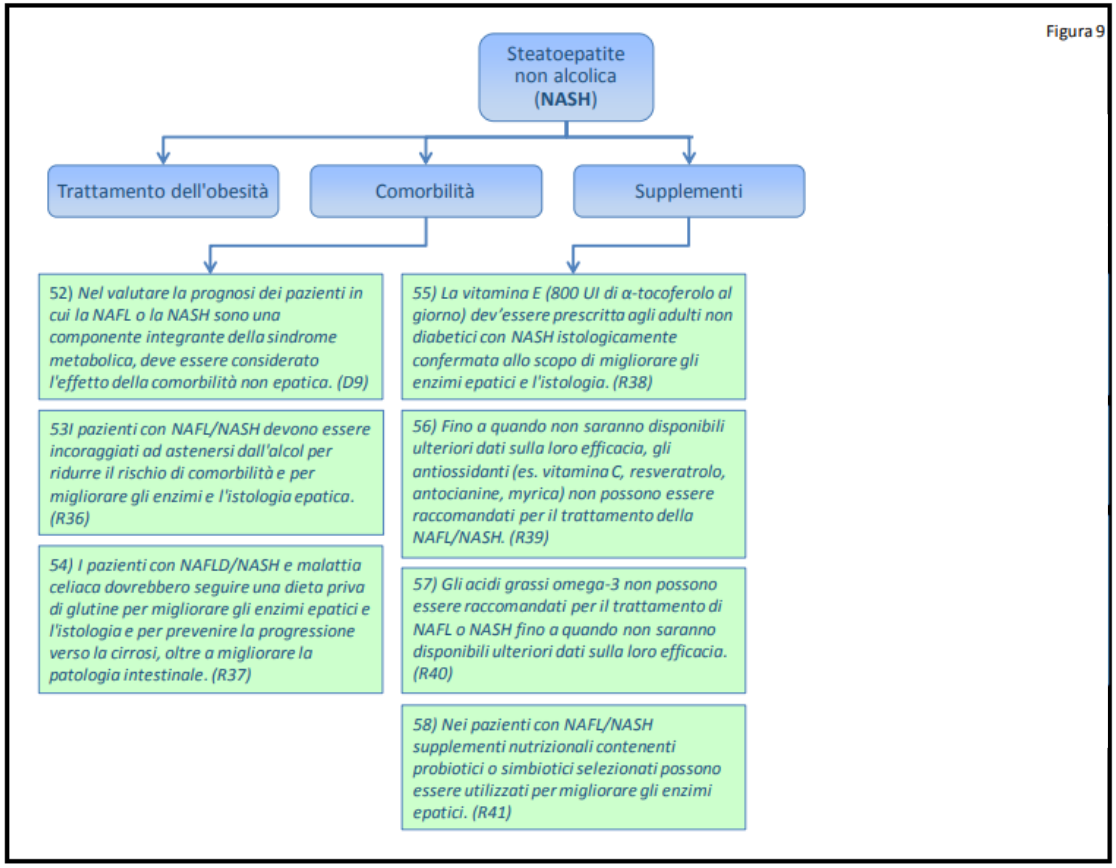
Figura 8



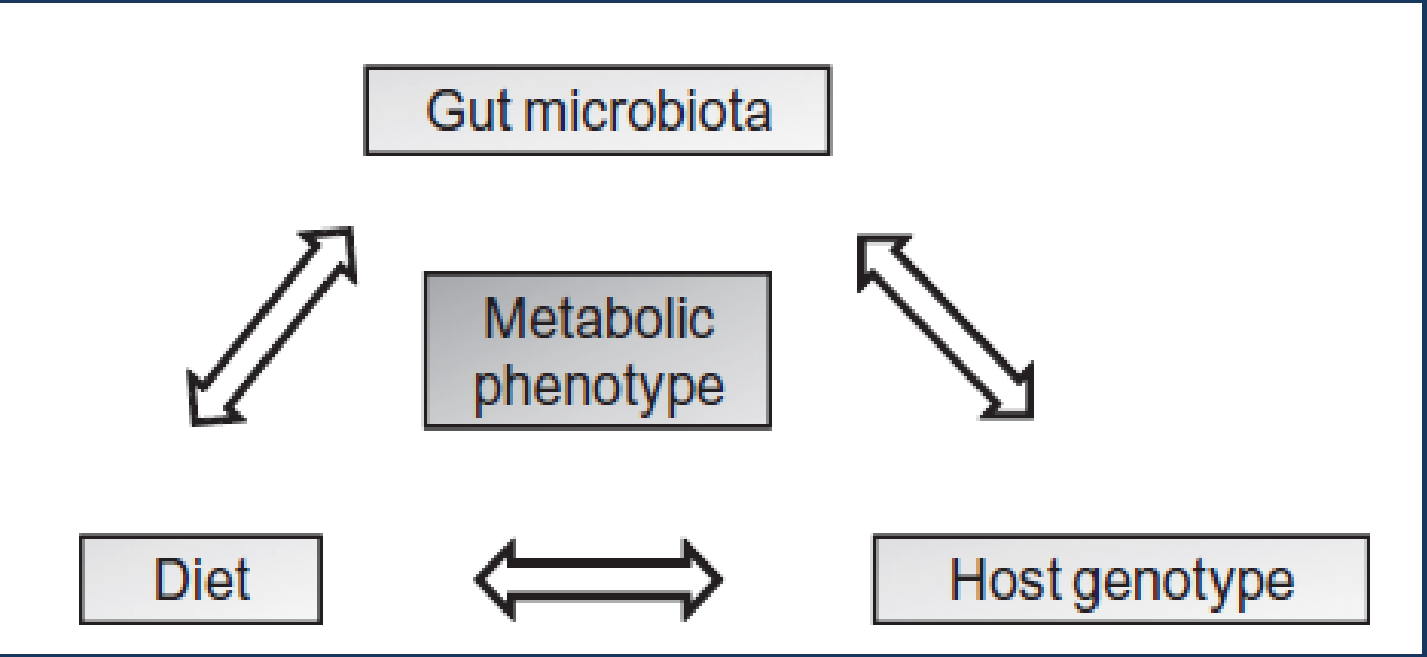
ESPEN
European Society for Clinical Nutrition and Metabolism

SINPE

Figura 9



A complex interaction …



Banked F. Nutrition Reviews® Vol. 70(Suppl. 1):S14–S17

Gut-microbiome and Host

- ✓ IBD
- ✓ Obesity
- ✓ Diabetes Mellitus
- ✓ NASH
- ✓ Autoimmunity
- ✓ Cancer

International Journal of
Molecular Sciences

MDPI

Review

Molecular Insight into Gut Microbiota and Rheumatoid Arthritis

Xiaohao Wu [1,2,†], Bing He [1,2,†], Jin Liu [1,2,†], Hui Feng [3,4,†], Yinghui Ma [3,4], Defang Li [1,2,5], Baosheng Guo [1,2,5], Chao Liang [1,2,5], Lei Dang [1,2], Luyao Wang [1,2], Jing Tian [4], Hailong Zhu [1,2,*], Lianbo Xiao [3,4,*], Cheng Lu [1,2,6,*], Aiping Lu [1,2,3,4,5,6,*] and Ge Zhang [1,2,4,5,*]

Table 1. Alterations of gut microbiota related with RA.

Studygroups	Sample Type	Technology Employed	Bacterial Taxa (↓low, ↑enriched)	Ref.
Early RA (51) *vs.* Fibromyalgia (50)	Stool	16S rRNA hybridization, and DNA-staining	↓*Bifidobacteria*, ↓*Bacteroides-Porphyromonas-Prevotella*, ↓*Bacteroides fragile*, ↓*Clostridium coccoides*	[24]
Early RA (15) *vs.* Healthy (15)	Stool	Quantitative real-time PCR	↑*Lactobacillus*	[25]
New-Onset RA (44) *vs.* Healthy (28)	Stool	16S rRNA gene and WGS sequencing	↑*Prevotella copri*, ↓*Bacteroidetes*	[26]
RA (30) *vs.*Healthy (30)	Stool	16S rRNA gene and WGS sequencing	↑*Enterococci*, ↑*Clostridia*, ↑*Colibacteria*, ↓*Lactobacteria*	[27]
Treatment-naïve RA (94) *vs.* Healthy (97)	Stool, Dental, Saliva	Metagenomic shotgun sequencing	↑*Lactobacillus salivarius*, ↑*Gordonibacter pamelaeae*, ↑*Clostridium asparagiforme*, ..., ↓*Haemophilus* spp.	[28]

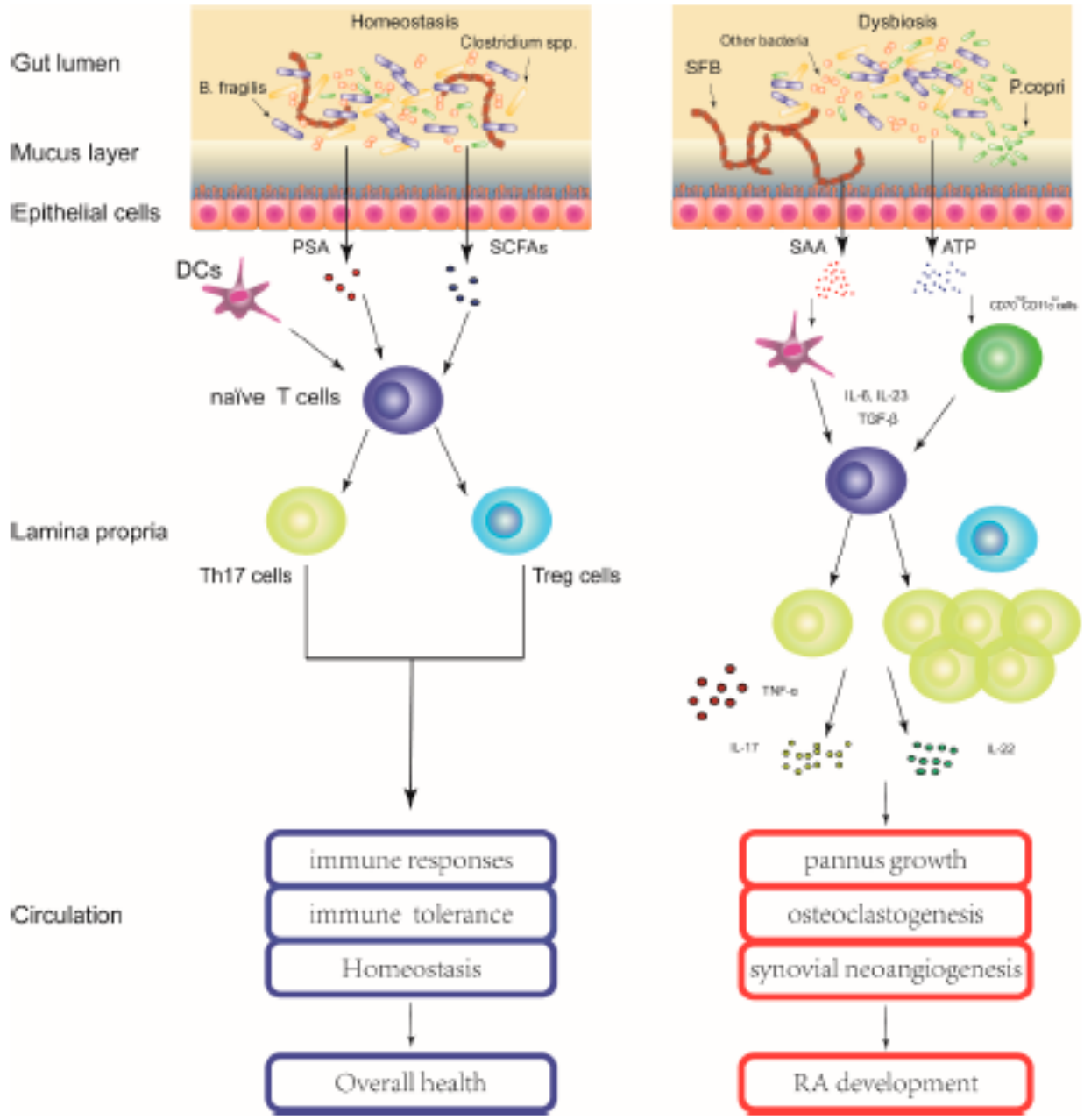
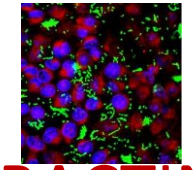


Figure 1. Gut microbiota contribute to the pathogenesis of RA. The healthy gut microbiota is in a homeostasis state that maintains integrity of the intestinal epithelial cell layer and has multiple symbiotic microbes to help in physiological functions. In genetically susceptible individuals, environmental factors can influence the gut microbiota causing changes in the types and abundance of microbiome (dysbiosis). The dysbiosis in gut microbiota, in association with genetic factors, may disrupt the innate and adaptive immune system and contribute to the development of RA via multiple molecular mechanisms. PSA, polysaccharide A; SCFAs, short-chain fatty acids; SAA, serum amyloid A; ATP, adenosine 5′-triphosphate; DCs, dendritic cells; IL, Interleukin; TGF-β, Transforming growth factor-beta; Treg cells, regulatory T cells; Th17 cells, T helper 17 cells.

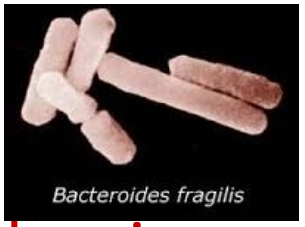
Gut-microbiome and Host

- ✓ **IBD**
- ✓ **Obesity**
- ✓ **Diabetes Mellitus**
- ✓ **NASH**
- ✓ **Autoimmunity**
- ✓ **Cancer**

- **ESCHERICHIA COLI AIEC**



Produce una genotossina COLIBACTINA che danneggia il DNA ed è essenziale per lo sviluppo dei tumori.



- **BACTEROIDES FRAGILIS**

Attiva attraverso una tossina (BACTERIOLISINA) che si interpone ad una proteina *suppressor* del tumore β.-caderina e attiva il sistema β-catenina che ↑VEFG, ↑COX_2 e quindi l'attecchimento e proliferazione del tumore.

- **FUSOBACTERIUM NUCLEATUM**

Presenta una adesina (FADA) che si trova sulla sua superificie, che si attacca e promuove l'inversione delle cellule tumorali intestinali.

Diet, GUT microbiota and Colonic Cancer

Diet

Modulate composition and metabolic activity of colon ecoflora

Clostridia and *Bacteroides* increase the tumor proliferative index

Lactobacilli and *Bifidobacteria* protective role against the cancerogenetic process

Hornie et al, Eur J Cancer Prev 1999

Diet, GUT Microbiota and carcinogens

Red meat

High dietary sulfur

Growth of sulfur-reducing bacteria
Desulfovibrio vulgaris

Free radicals

Hydrogen sulphide

Co-carcinogen

Impairs cytochrome oxidase

- Butyrate utilization
- Syntesis of mucus
- Methylation of DNA

Christl et al. Gut 1992

www.impactjournals.com/oncotarget/ Oncotarget, Vol. 7, No. 29

Research Paper

Could gut microbiota serve as prognostic biomarker associated with colorectal cancer patients' survival? A pilot study on relevant mechanism

Zhiliang Wei[1], Shougen Cao[1], Shanglong Liu[1], Zengwu Yao[2], Teng Sun[3], Yi Li[1], Jiante Li[1], Dongfeng Zhang[4], Yanbing Zhou[1]

[1]Department of General Surgery, Affiliated Hospital of Qingdao University, Qingdao, China

[2]Department of General Surgery, Yantai Yuhuangding Hospital, Yantai, China

[3]Department of General Surgery, Qingdao Municipal Hospital Group, Qingdao, China

[4]Department of Epidemiology and Health Statistics, Qingdao University Medical College, Qingdao, China

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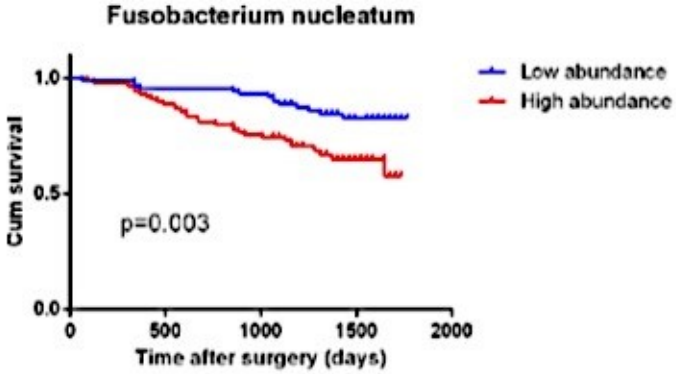
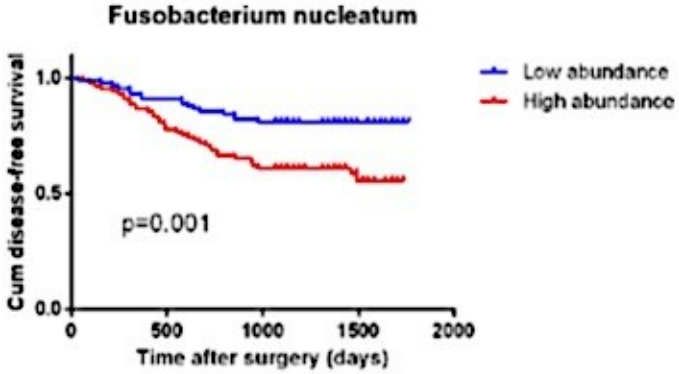
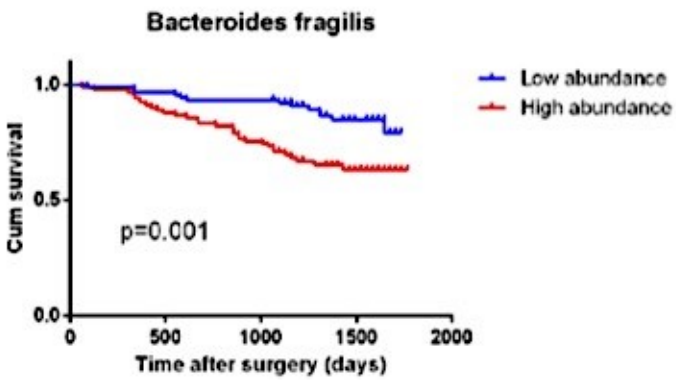
Keywords: *colorectal cancer, inflammation, intestinal microbiology, prognostic biomarker, prognosis*

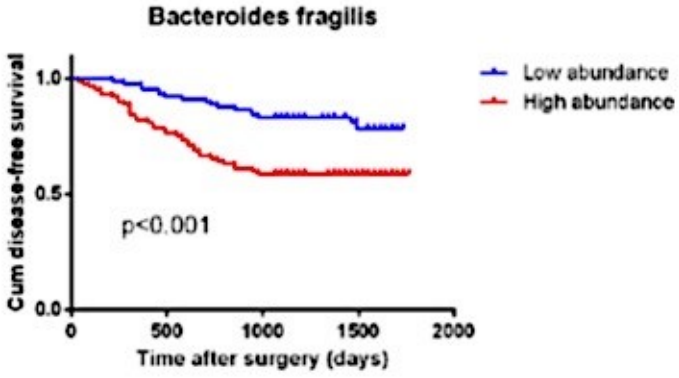
Received: *February 24, 2016* ***Accepted:*** *June 02, 2016* ***Published:*** *June 15, 2016*

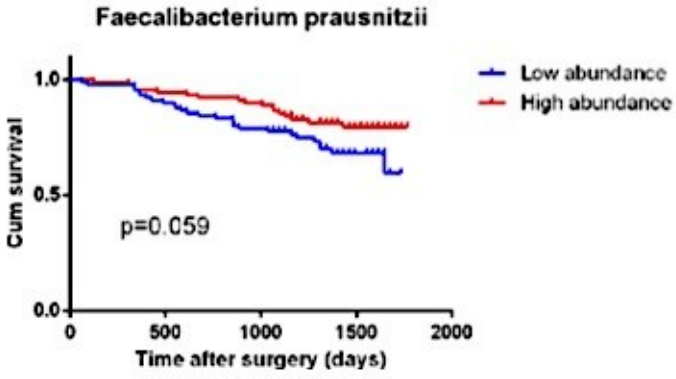
Curve di sopravvivenza di Kaplan-Meier per i sopravvissuti totale (OS) e isopravvissuti non malati(DFS) in 180 pazienti CRC.

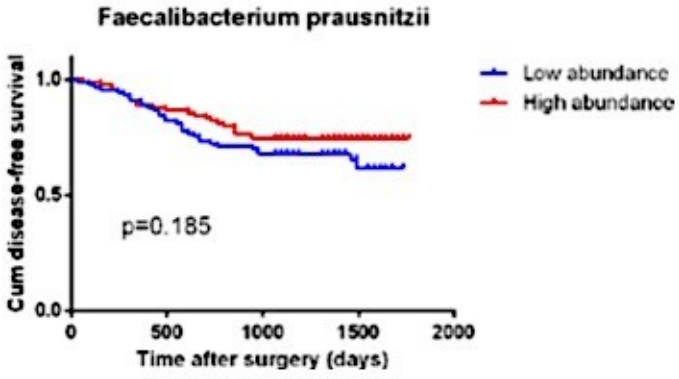






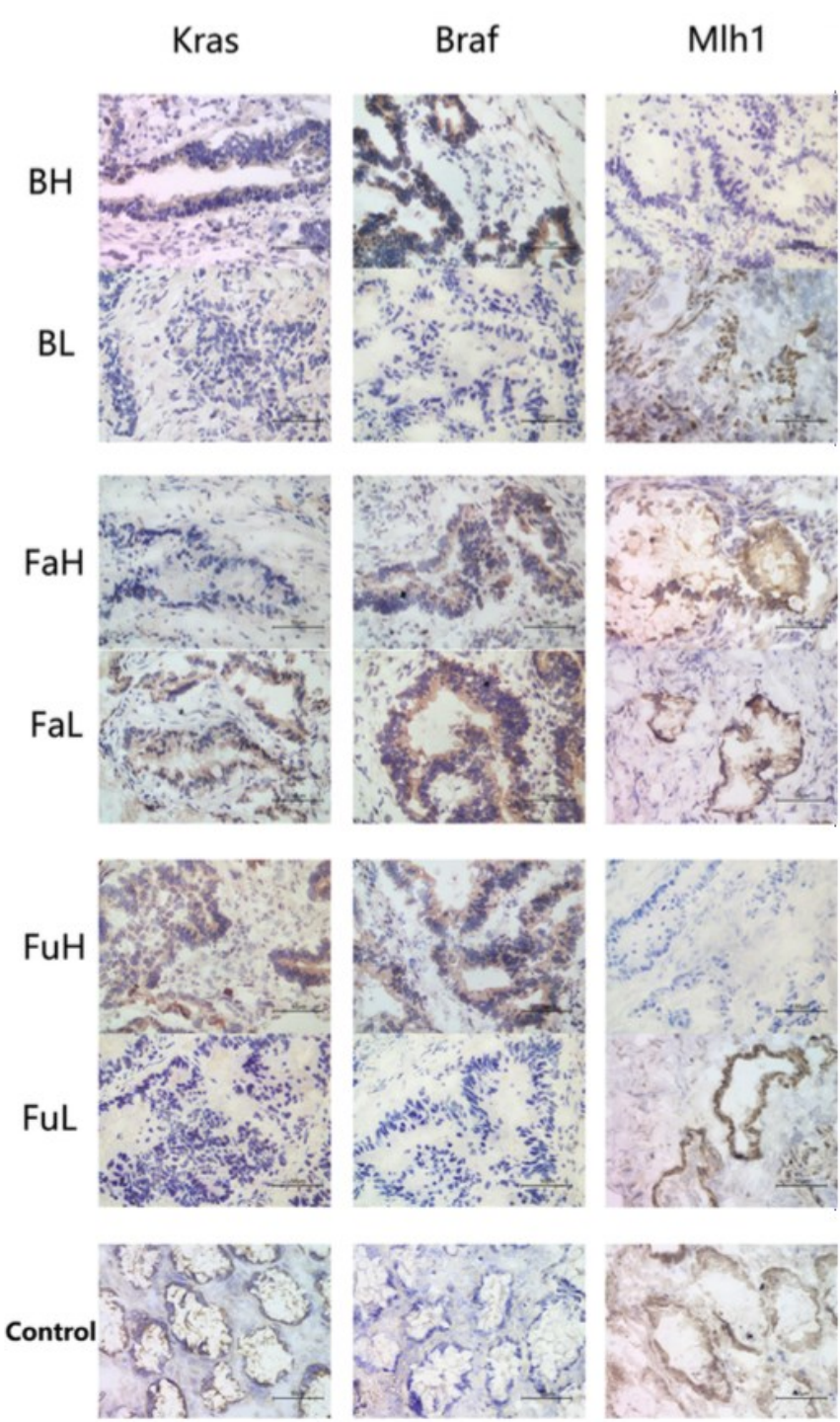






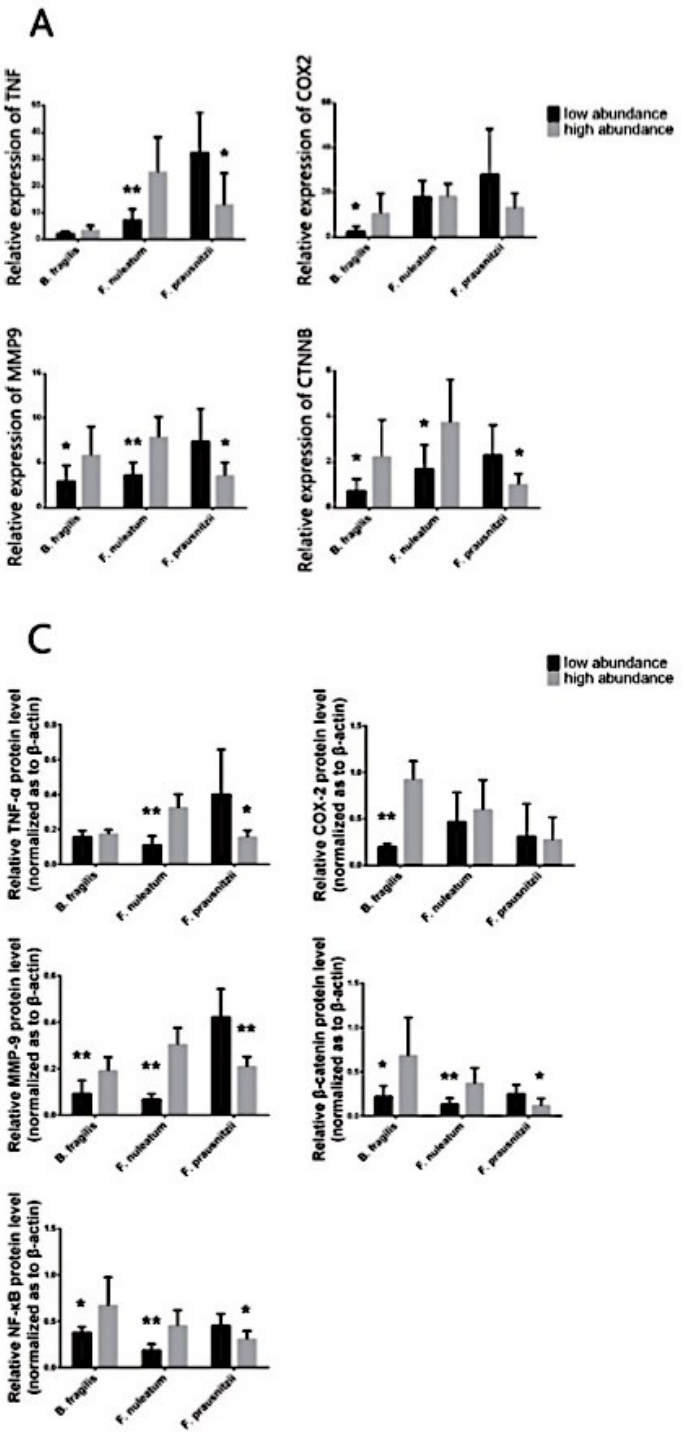
Wei Z et al; Could gut microbiota serve as prognostic biomarker associated with colorectal cancer patients' survival? A pilot study on relevant mechanism; Oncotarget, 2016.

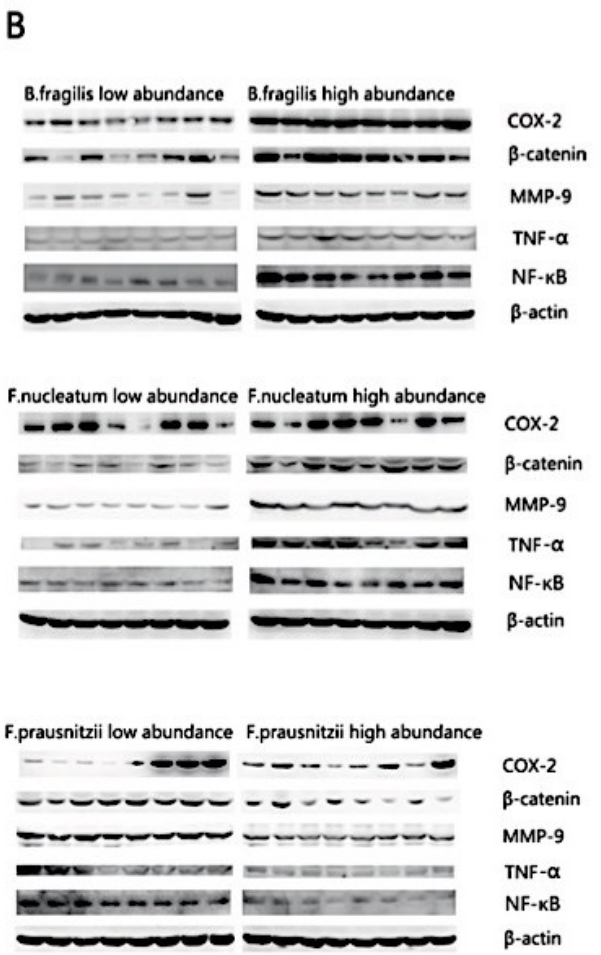
Immunoistochimica per l'analisi della espressione del gene KRAS, BRAF e MLH1 nei tessuti tumorali con diversa abbondanza rispettivamente di *B. fragilis, F. nucleatum e F.prausnitzii.*



Wei Z et al; Could gut microbiota serve as prognostic biomarker associated with colorectal cancer patients' survival? A pilot study on relevant mechanism; Oncotarget, 2016.

Livelli di espressione dell'mRNA del *TNF, COX2, MMP9* and *CTNNB* nei tessuti cancerosi con diversa concentrazione di *B. fragilis*, *F. nucleatum* e*F.prausnizii* analizzati con RT-PCR, e le proteine TNF-α, COX-2, MMP-9, β-catenina eNF-κB analizzati con western blot.

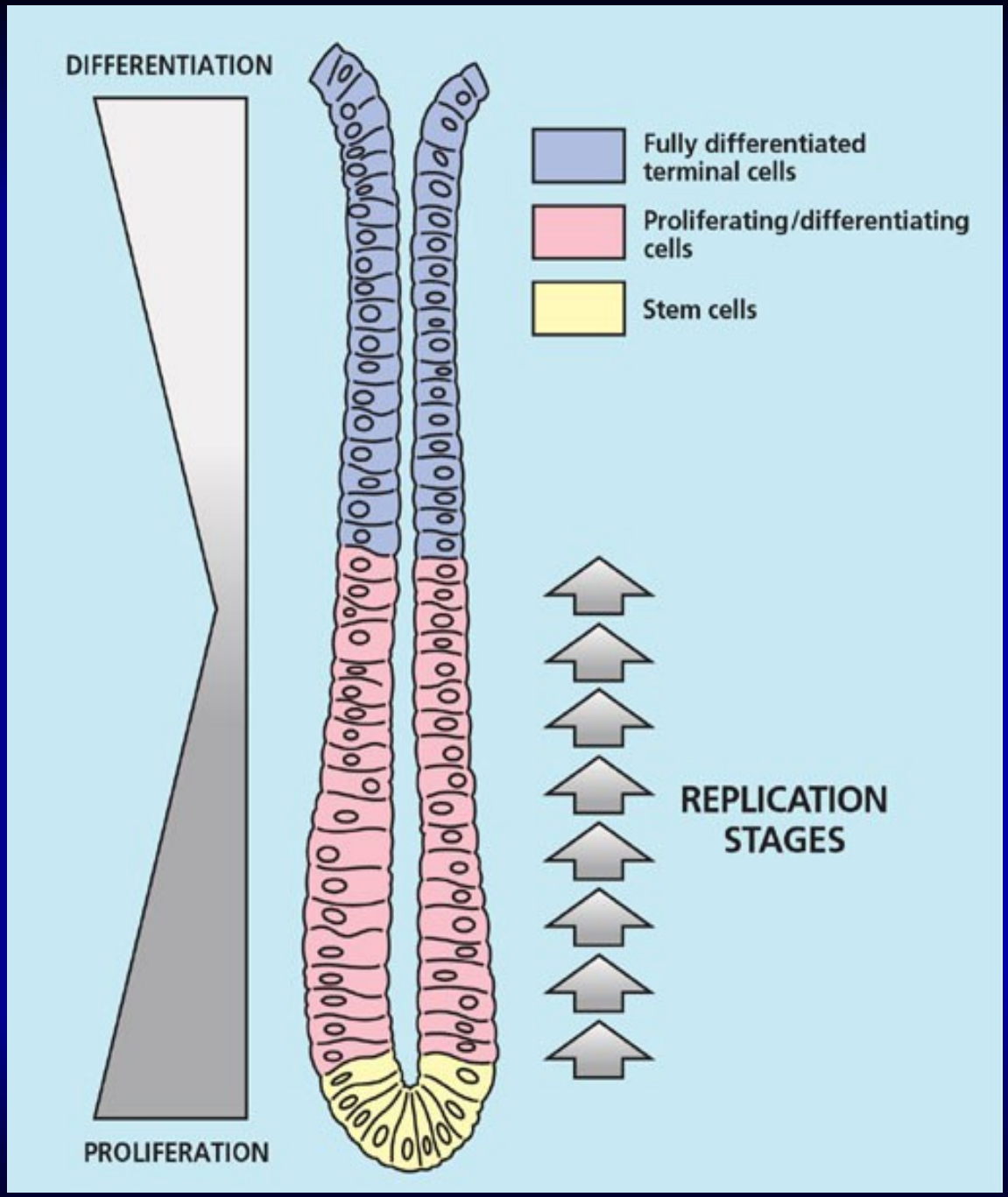




Wei Z et al; Could gut microbiota serve as prognostic biomarker associated with colorectal cancer patients' survival? A pilot study on relevant mechanism; Oncotarget, 2016.

Question 9

May GUT bacteria interfere with GI stem cells for regeneration?

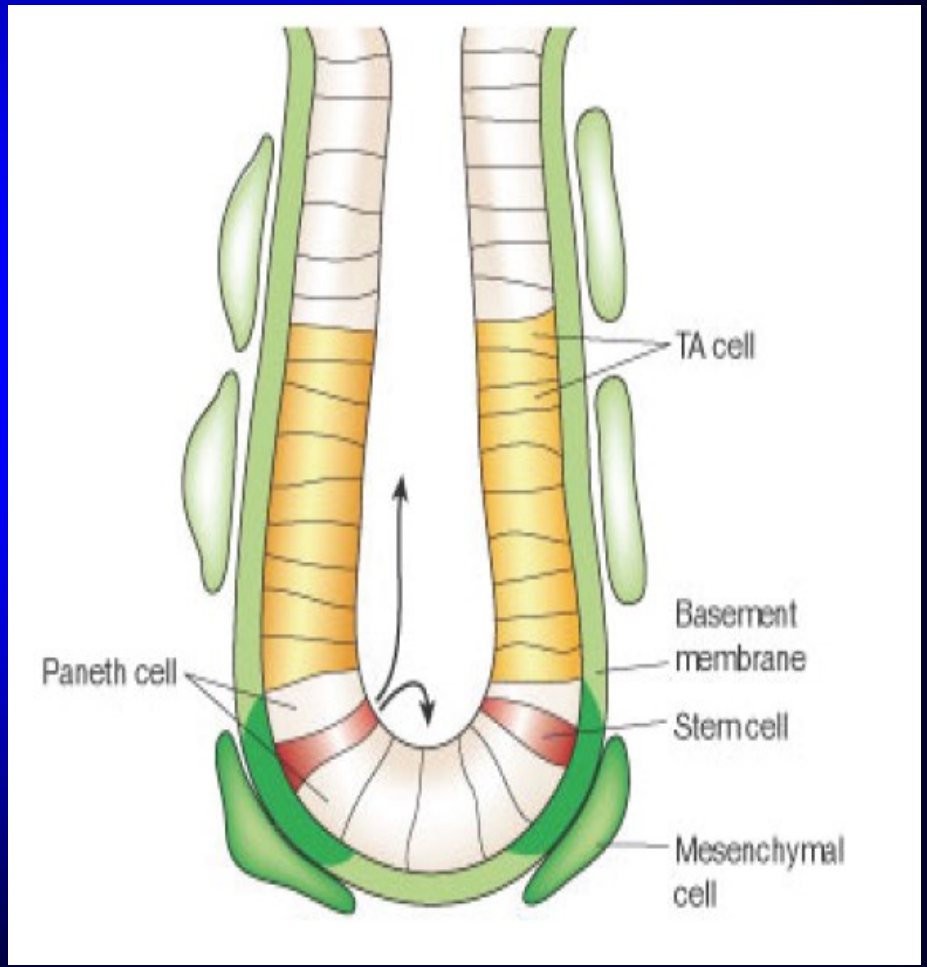

DIFFERENTIATION
PROLIFERATION
Fully differentiated terminal cells
Proliferating/differentiating cells
Stem cells
REPLICATION STAGES

GI TURNOVER

- The GI epithelium experiences continuous cell loss, also enhanced by the high rates of mechanical attrition

- Most of the epithelial cells are replaced every 2-5 days

- The GI renewal depends on a small population of multipotent GI-SCs situated within the intestinal crypts and the gastric glands (*unitarian hypothesis)*

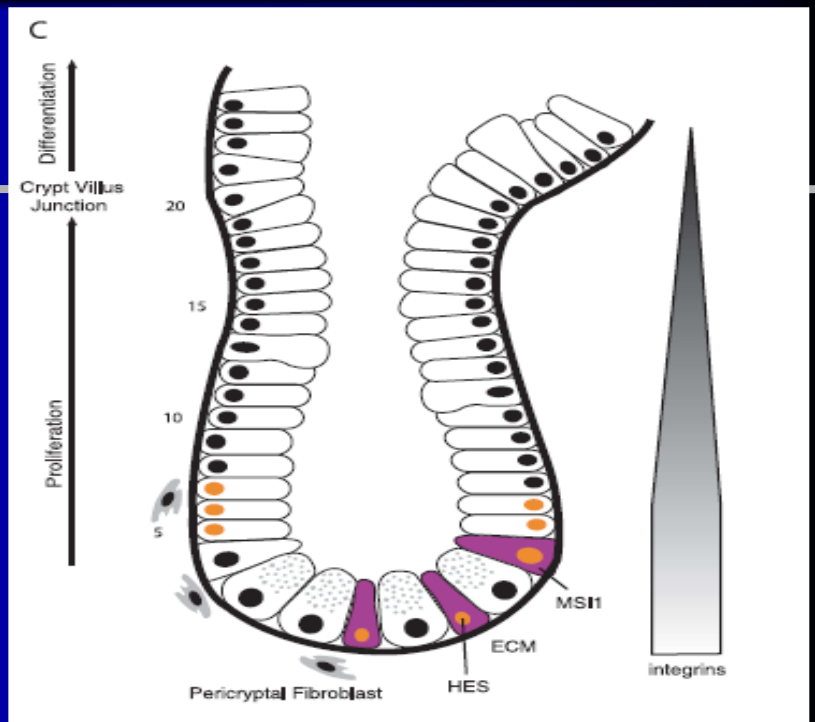


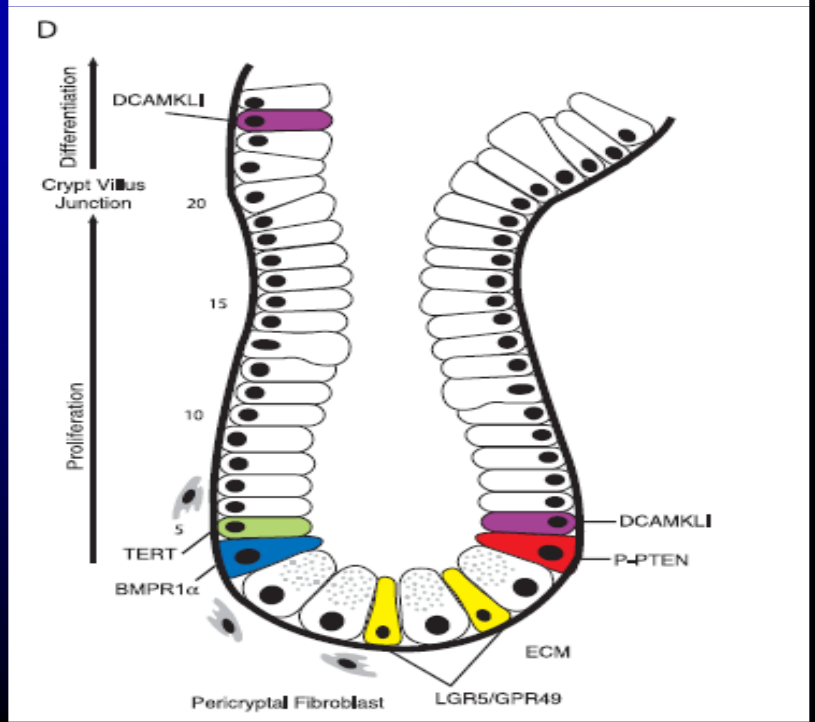
MARKERS OF GISCs

- Potential markers of GISCs:
- *Musashi-1 (Msi-1)*
- *Enhancer of Split Homolog-1 (Hes),*
- *side population,*
- *FoxP4,*
- *Eph,*
- *EphA6*

- Signaling pathways controlling GISCs fate:
- *Wnt/β-catenin*
- *T-cell factor (TCF)/lymphoid enhancing factor (LEF)*

Robert K. J. Anat (2008)



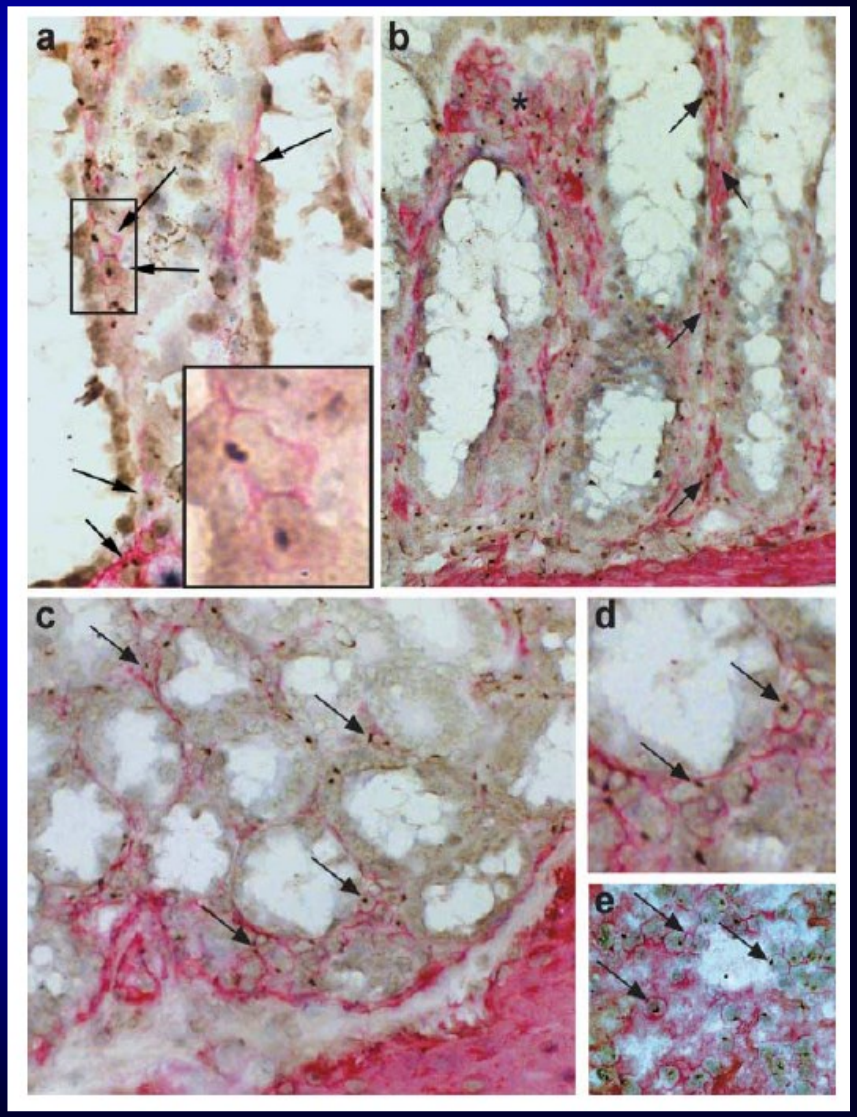


POSSIBLE ORIGINS OF GISCs

GISCs are mainly endogenous gut cells

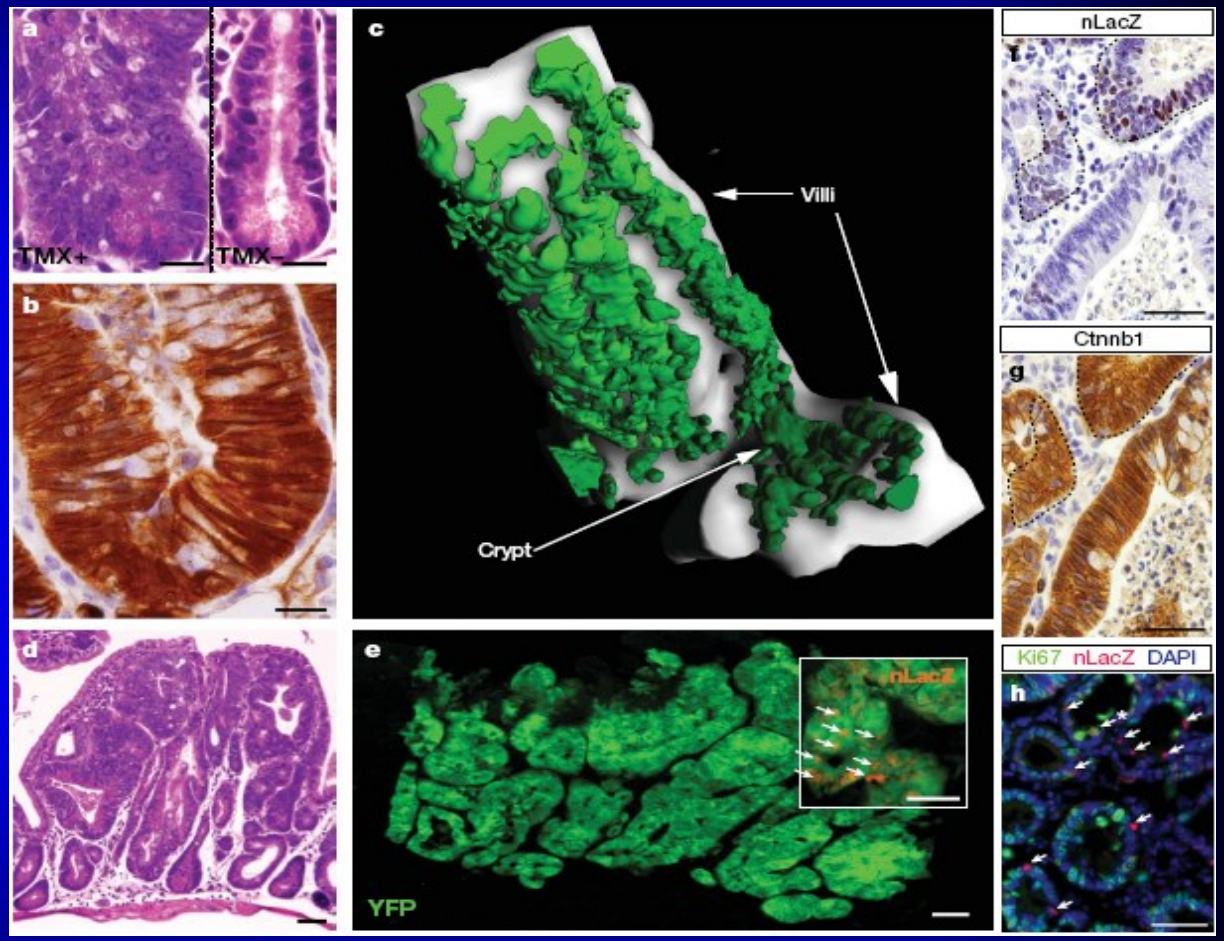
However, extra-intestinal cells (such as bone marrow SCs) can colonize the GI epithelium and contribute to the intestinal repopulation by:

- *giving rise to GISCs*
- *providing supporting elements (i.e. myofibroblasts, which play an important role in the regulation of GI-SC niche)*



Brittan M, Wright NA. Gut 2004

Activation of Wnt signalling pathway in CD133+ small bowel stem cells → neoplastic transformation of the small intestinal mucosa.



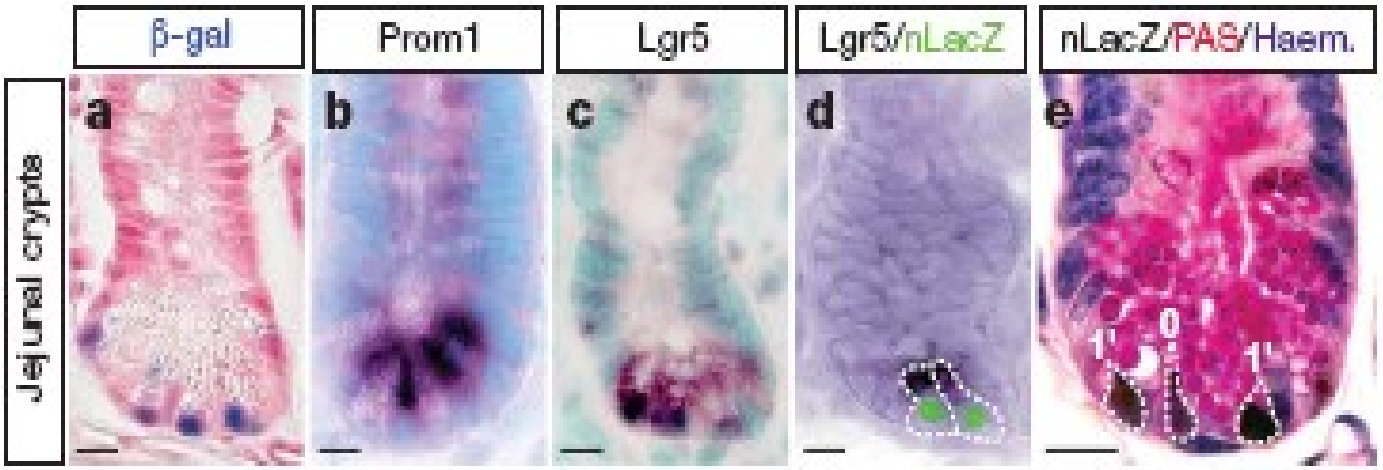
Zhu L Nature. 2008

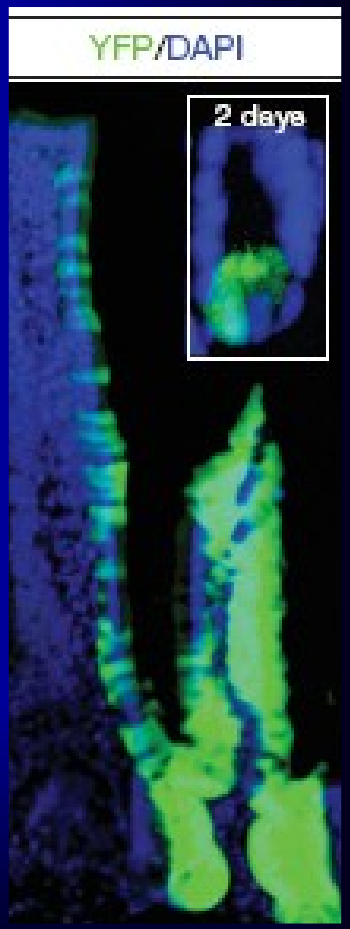
CD133+ stem cells give rise to small bowel tumors *via* Wnt activation

Prominin 1 marks intestinal stem cells that are susceptible to neoplastic transformation

Liqin Zhu[1], Paul Gibson[1], D. Spencer Currle[1], Yiai Tong[1], Robert J. Richardson[1], Ildar T. Bayazitov[1], Helen Poppleton[1], Stanislav Zakharenko[1], David W. Ellison[2] & Richard J. Gilbertson[1,3]

CD133 (Prominin1) is a marker of Lgr5+ stem cells in the small intestine





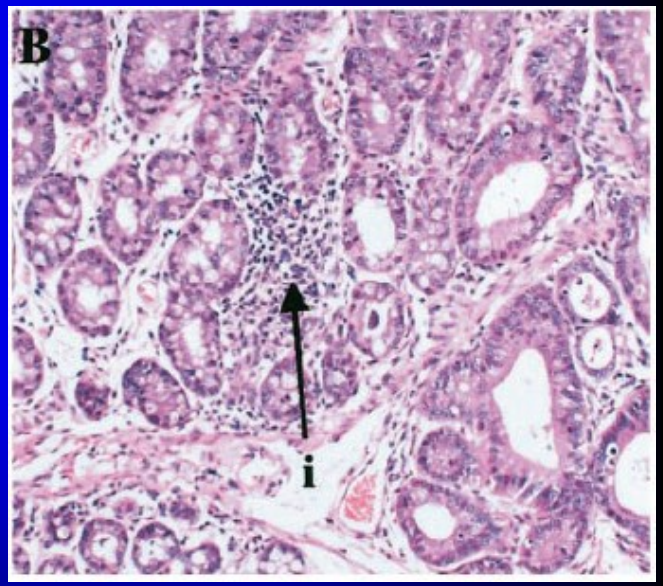
Zhu L Nature. 2008

Elimination of Colon Cancer in Germ-free Transforming Growth Factor Beta 1-deficient Mice

Sandra J. Engle, Ilona Ormsby, Sharon Pawlowski, et al.

Cancer Res 2002;62:6362-6366.

- Patients with ulcerative colitis are at risk for colon cancer and frequently have microsatellite instability, which is usually associated with inactivation of transforming growth factor (TGF) signaling.
- TGF-1 deficiency in mice can lead to colon cancer (precancerous lesions having submucosal inflammation and hyperplastic crypts)
- Germ-free TGF-1-deficient mice are free of inflammation, hyperplasia, and cancer, but when reintroduced into a Helicobacter hepaticus-containing specific pathogen-free room, these lesions reappear



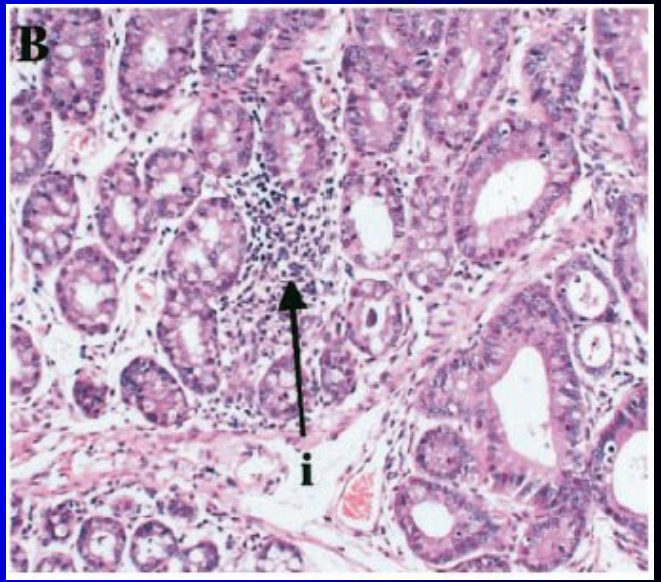
Elimination of Colon Cancer in Germ-free Transforming Growth Factor Beta 1-deficient Mice

Sandra J. Engle, Ilona Ormsby, Sharon Pawlowski, et al.

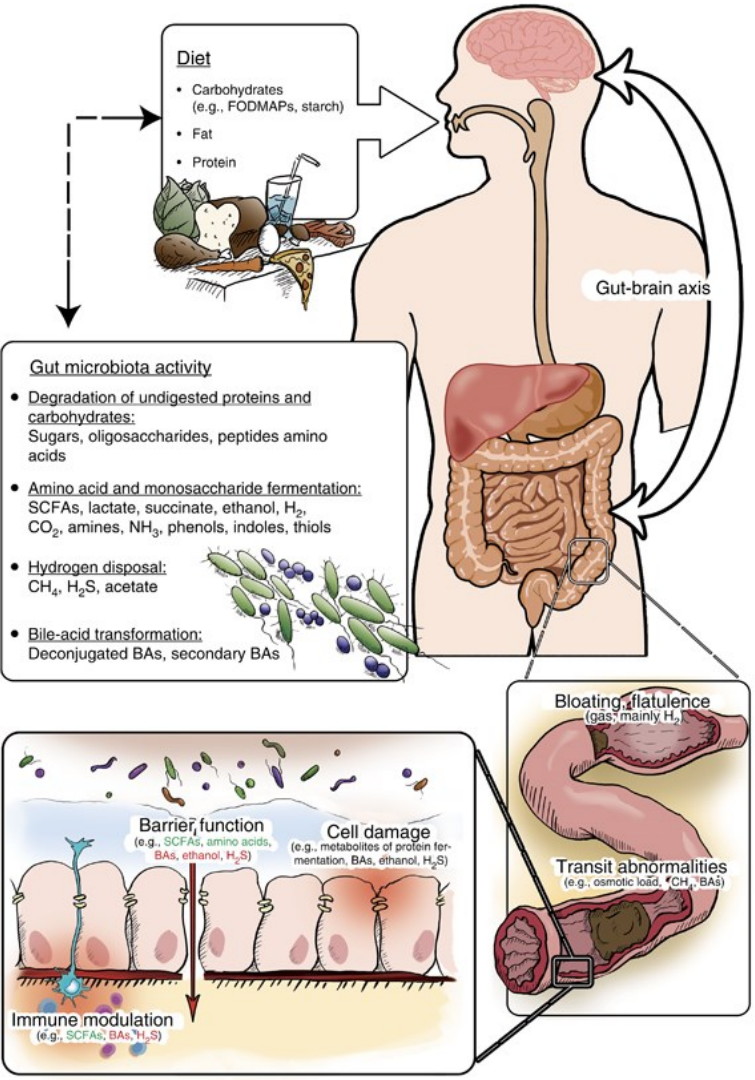
Cancer Res 2002;62:6362-6366.

-Because adenoma/carcinoma but not inflammation/hyperplasia is dependent on the genetic backgrounds tested, colitis is required, but not sufficient, for carcinogenesis.

- This animal model should provide insight into the protective role of TGF-1 in early stages of ulcerative colitisassociated human colon cancer



(Tgfb1)-null mice develop colonic cancer in the presence of conventional gut microbiota



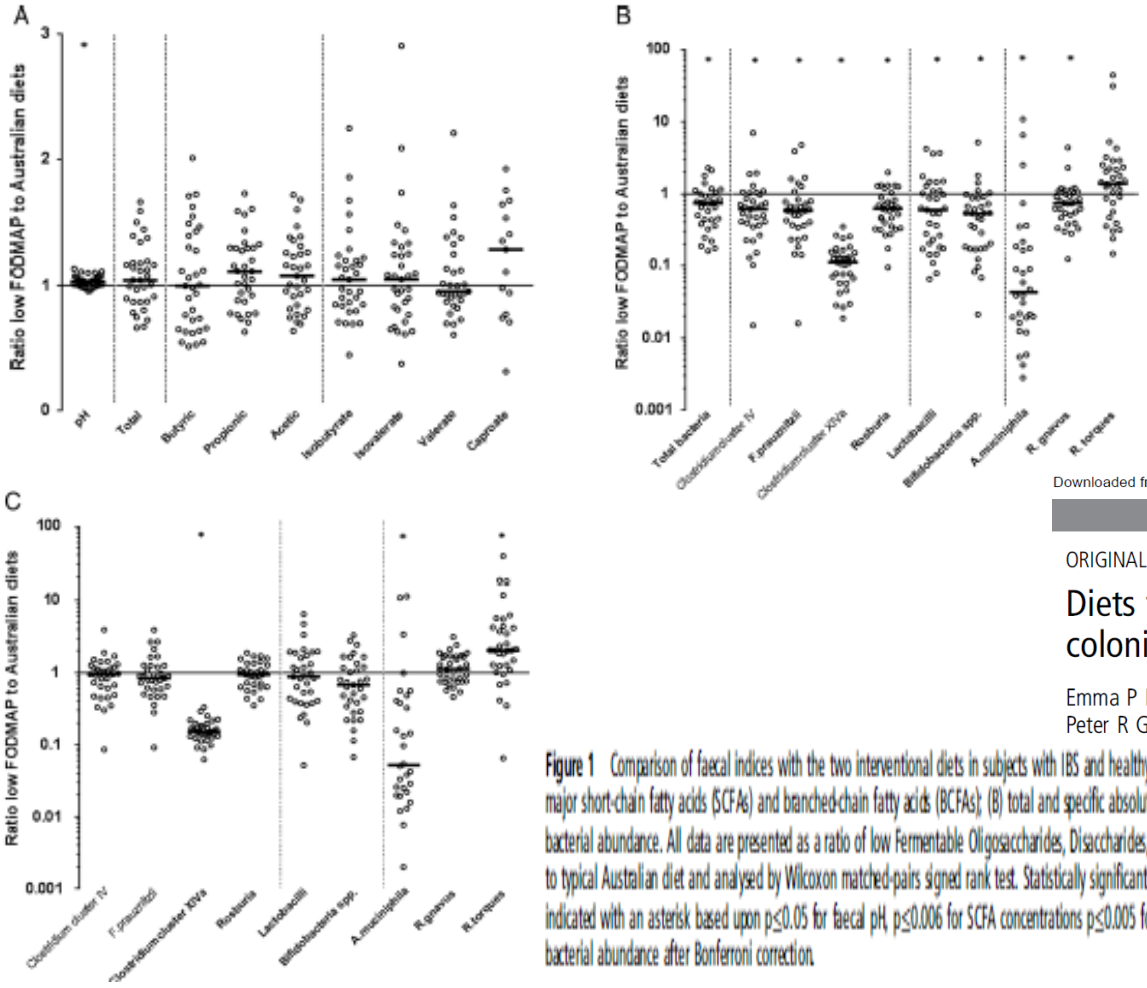
FODMAP

F: fermentabili
O: oligosaccaridi
D: disaccaridi (lattosio, fruttani, fruttosio, galattani)
M: monosaccaridi
A: and
P: polyols (sorbitolo, mannitolo, xilitolo, maltitolo, lattulosio)

- ✓ Osmoticamente attivi
- ✓ Scarsamente assorbiti nell'intestino
- ✓ Rapidamente fermentati nel tenue

DIETA FODMAP

	Fruttosio	Lattosio	Oligosaccaridi (fruttani-galattani)	Polioli
Fonti alimentari ad ALTO CONTENUTO di FODMAP	**Frutta:** mele, pere, pesche, mango, anguria **Miele** **Dolcificanti:** fruttosio, sciroppo di mais **Alte dosi di fruttosio:** concentrati di frutta, conserve di frutta, succhi di frutta, frutta secca	**Latte:** mucca, capra, percora, gelati **Formaggi:** morbidi e freschi (per es. ricotta)	**Verdure:** carciofi, asparagi, barbabietole, cavolini di Bruxelles, broccoli, cavoli, finocchio, aglio, gombo, cipolle, piselli, scalogno **Cereali:** Frumento e segale se consumati in grandi quantità (pane, pasta, couscous, crackers, biscotti) **Legumi:** ceci, lenticchie, fagioli, fave **Frutta:** anguria, pesche bianche, cachi	**Frutta:** mele, albicocche, ciliegie, pere, pesche, susine, prugna, anguria **Verdura:** avocado, cavolfiori, funghi, piselli **Dolcificanti:** sorbitolo, mannitolo, xilitolo e altri che terminano in -olo
Fonti alimentari a BASSO CONTENUTO di FODMAP	**Fruttosio**	**Lattosio**	**Oligosaccaridi (fruttani-galattani)**	**Polioli**
	Frutta: banana, mirtillo, pompelmo, uva, melone, kiwi, limone, mandarino, arancia, lampone, fragola **Sostitutivi del miele:** sciroppo d'acero **Dolcificanti:** tutti eccetto i polioli	**Latte:** latte delattosato, di soia, di riso **Formaggi:** formaggi duri e stagionati **Sostituti del gelato:** sorbetti **Burro**	**Verdure:** germogli di bambù, sedano, peperoni, melanzane, fagiolini, lattuga, erba cipollina, zucca, cipolla verde, pomodoro **Cereali:** prodotti senza glutine e farro	**Frutta:** banana, mirtillo, pompelmo, kiwi, mandarino, limone, arancia, uva, lampone, fragola **Dolcificanti:** zucchero (saccarosio), glucosio, dolcificanti che non terminano in -olo



ORIGINAL ARTICLE

Diets that differ in their FODMAP content alter the colonic luminal microenvironment

Emma P Halmos,[1,2] Claus T Christophersen,[3] Anthony R Bird,[3] Susan J Shepherd,[1] Peter R Gibson,[1,2] Jane G Muir[1,2]

Figure 1 Comparison of faecal indices with the two interventional diets in subjects with IBS and healthy subjects. (A) Changes in pH, total and major short-chain fatty acids (SCFAs) and branched-chain fatty acids (BCFAs); (B) total and specific absolute bacterial abundance; and (C) relative bacterial abundance. All data are presented as a ratio of low Fermentable Oligosaccharides, Disaccharides, Monosaccharides And Polyols (FODMAP) to typical Australian diet and analysed by Wilcoxon matched-pairs signed rank test. Statistically significant differences between the diets are indicated with an asterisk based upon $p \leq 0.05$ for faecal pH, $p \leq 0.006$ for SCFA concentrations $p \leq 0.005$ for absolute and $p \leq 0.006$ for relative bacterial abundance after Bonferroni correction.


… ed il microbiota ha un suo ruolo nella malattia celiaca?

Intestinal Microbiota and Probiotics in Celiac Disease

Luís Fernando de Sousa Moraes, Lukasz Marcin Grzeskowiak, Tatiana Fiche de Sales Teixeira, Maria do Carmo Gouveia Peluzio
Department of Nutrition and Health, Federal University of Viçosa, Viçosa, Minas Gerais, Brazil

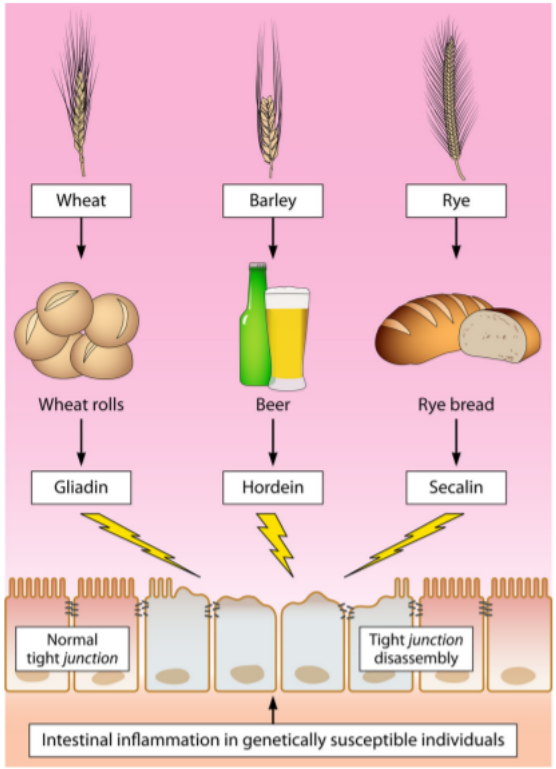


FIG 1 Different cereal-derived products and intestinal inflammation in CD subjects. Consumption of food-derived products containing wheat, barley, and rye by individuals genetically susceptible to CD leads to villous atrophy, intestinal inflammation, and disassembly of tight junctions.

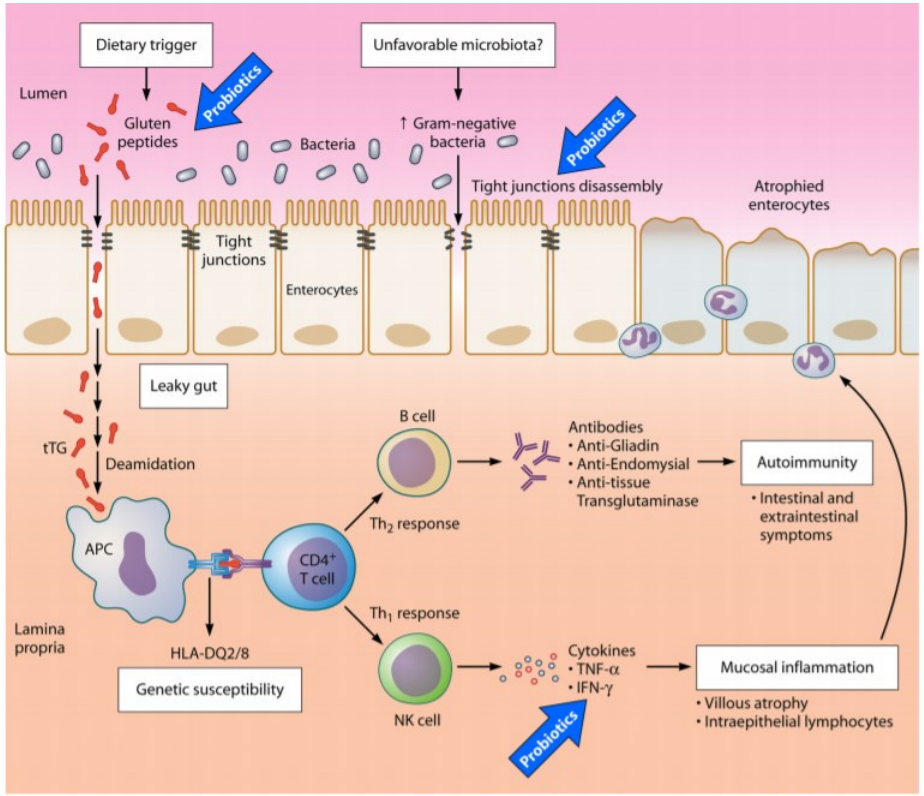


FIG 2 Inflammation process and possible routes of probiotic action in the maintenance of CD. In CD patients, increased epithelial tight junction permeability ("leaky gut") favors the entrance of non-well-digested gluten peptides from the lumen to the lamina propria. Once there, they are deamidated by the tissue transglutaminase (tTG) enzyme and presented to $CD4^+$ T immune cells by the human leukocyte antigen (HLA) in antigen-presenting cells (APCs), which in CD patients is often of the haplotypes DQ2 and DQ8. Thereafter, Th1 and Th2 immune responses are triggered, resulting in autoimmunity, mucosal inflammation, and the growth of unfavorable microbiota, worsening the prognosis of disease. Three large arrows indicate where probiotics could act.

Clinical and Microbiological Effect of a Multispecies Probiotic Supplementation in Celiac Patients With Persistent IBS-type Symptoms

A Randomized, Double-Blind, Placebo-controlled, Multicenter Trial

Ruggiero Francavilla, MD, PhD, Maria Piccolo, PhD,† Antonio Francavilla, MD, PhD,‡ Lorenzo Polimeno, PhD,* Francesco Semeraro, MD,§ Fernanda Cristofori, MD,* Stefania Castellaneta, MD,‖ Michele Barone, MD, PhD,¶ Flavia Indrio, MD,* Marco Gobbetti, PhD,# and Maria De Angelis, PhD†*

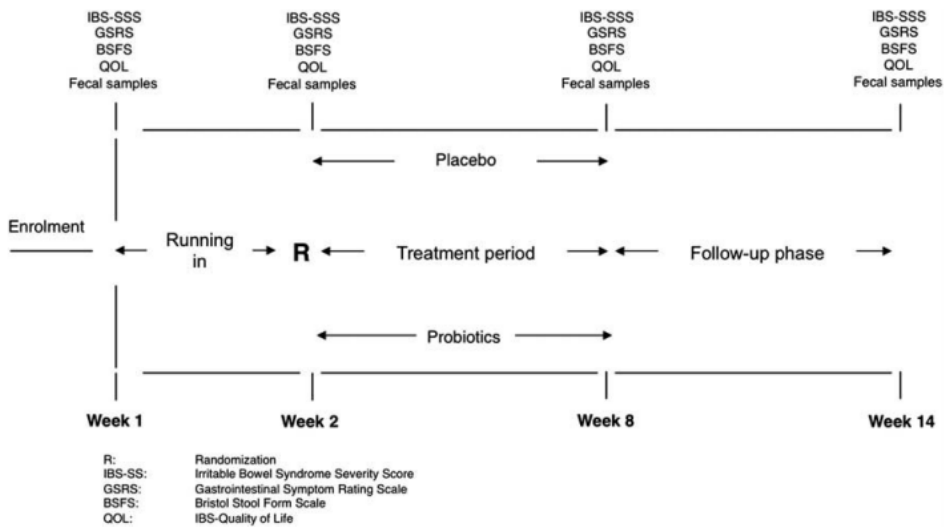


FIGURE 1. Study design. BSFS indicates Bristol Stool Form Scale; GSRS, Gastrointestinal Symptom Rating Scale; IBS-SSS, Irritable Bowel Syndrome Severity Scoring System; QOL, Quality of Life; R, randomization.

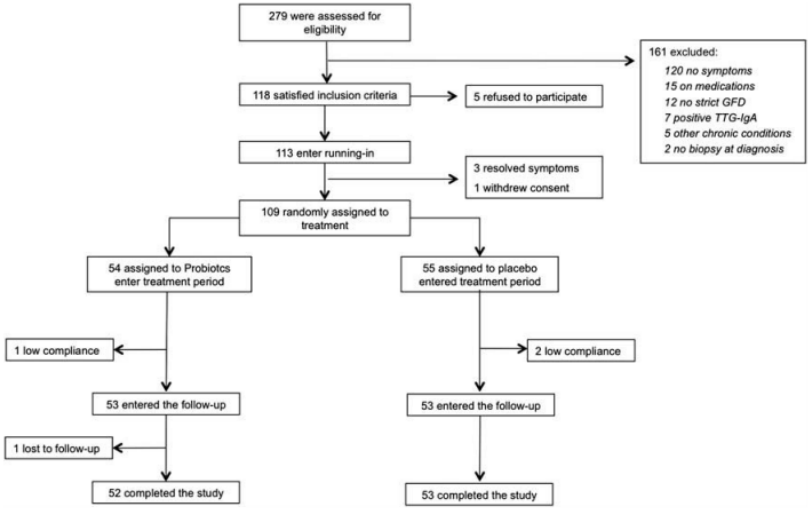


FIGURE 2. Flow diagram of patients in the trial from eligibility to the end of follow-up. GFD indicates gluten-free diet; TTG-IgA, tissue transglutaminase immunoglobulin-A.

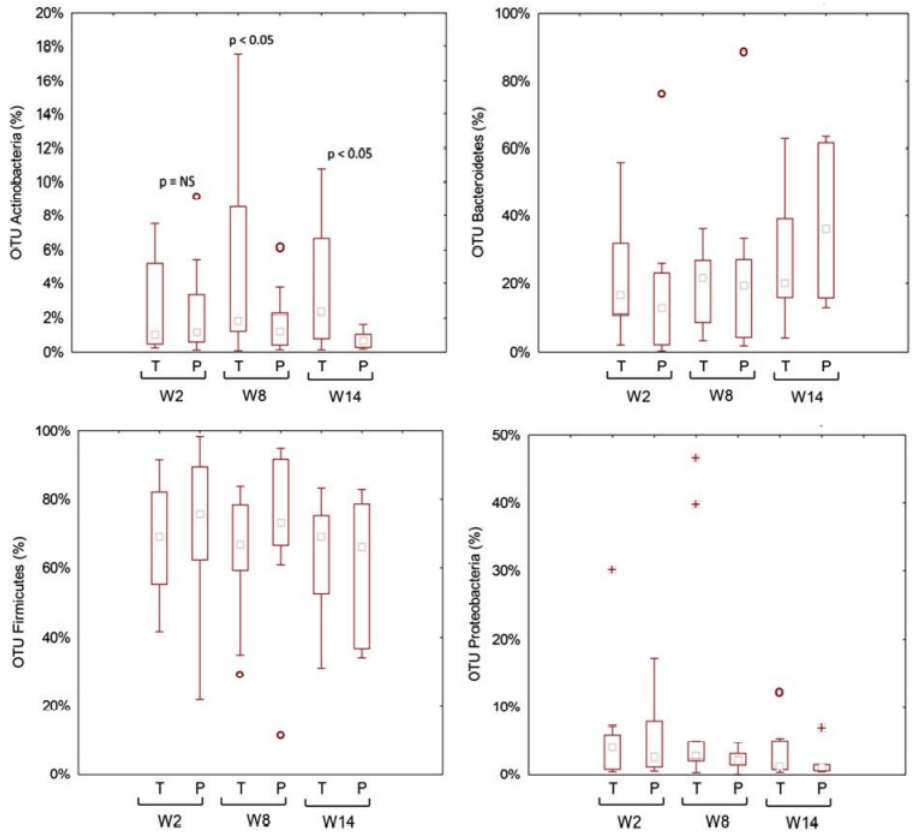


FIGURE 3. Relative abundance of the most relevant metabolically active bacterial phyla found in feces of the fecal samples of celiac disease patients with irritable bowel syndrome at baseline (W2), after 6 weeks (W8) of treatment with probiotics or placebo, and at the end of follow-up (W14). NS indicates not significant; OTU, operational taxonomic unit; P, placebo; T, treated.

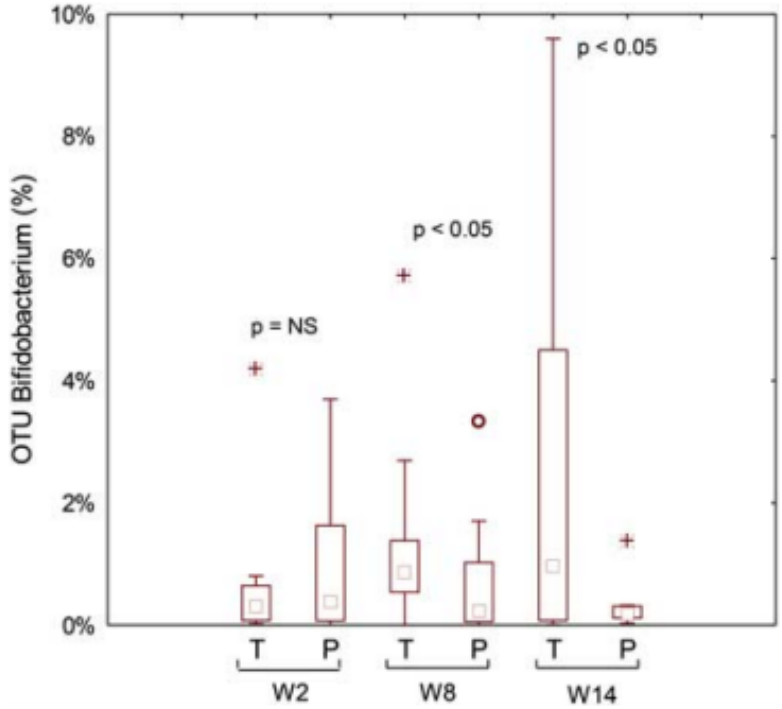
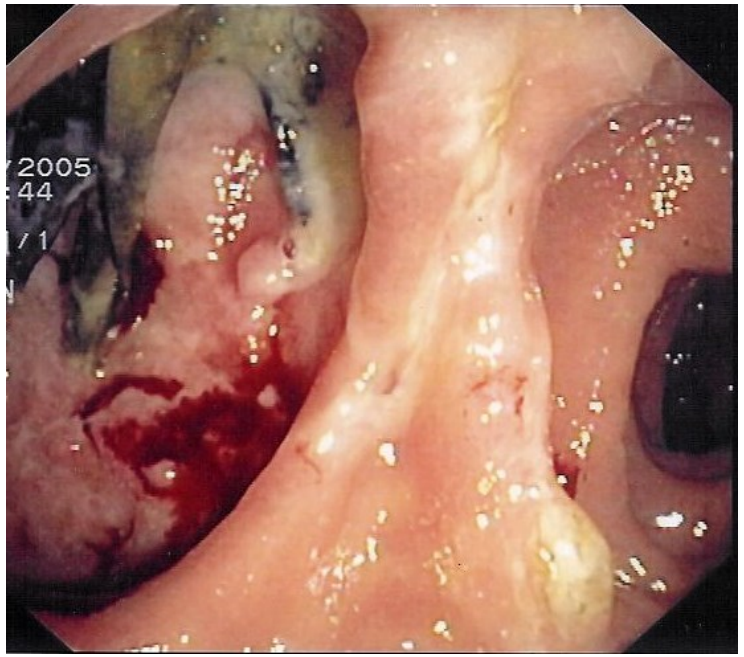
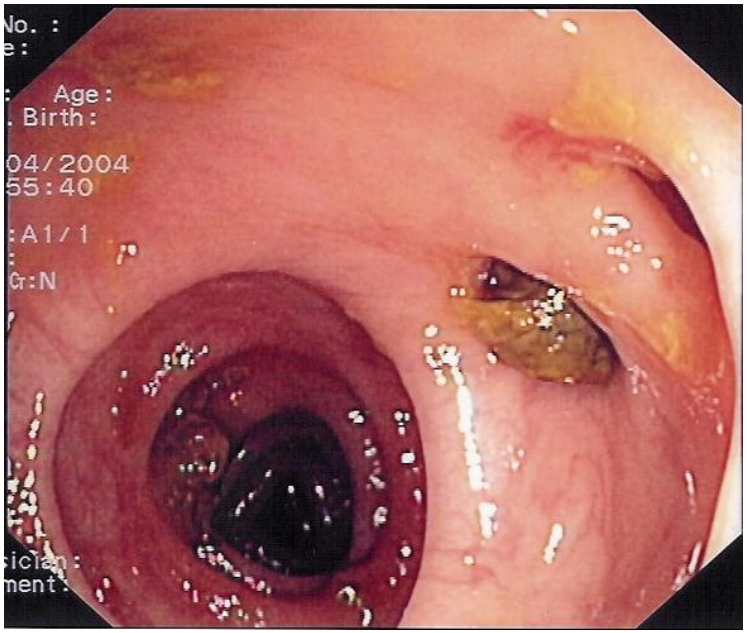
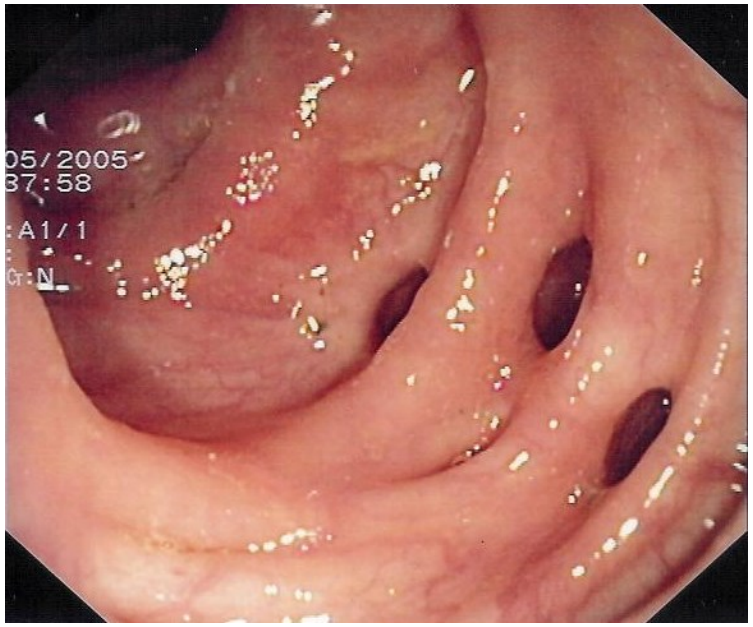
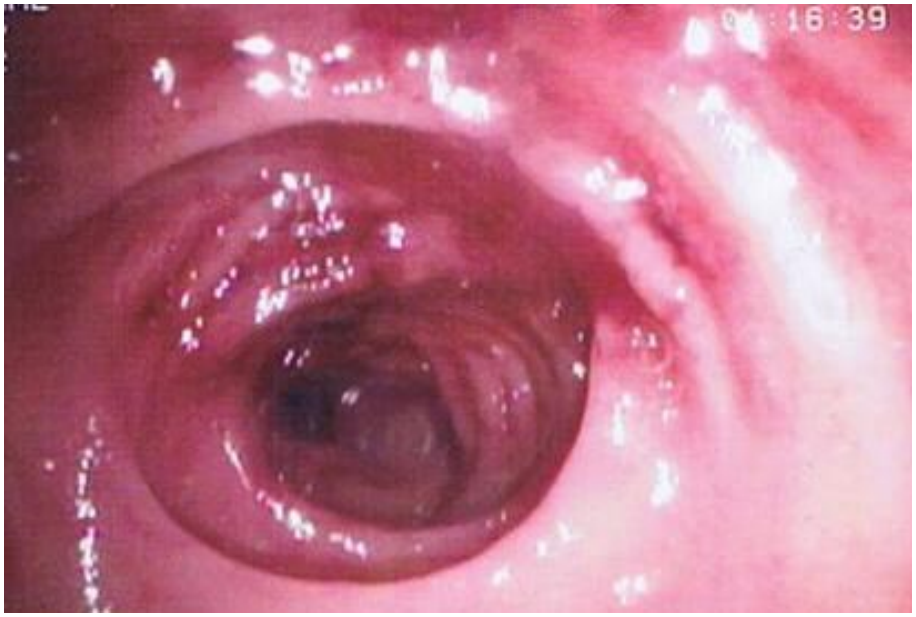


FIGURE 4. Metabolically active *Bifidobacterium* genus found in feces of the fecal samples of celiac disease patients with irritable bowel syndrome at baseline (W2), after 6 weeks (W8) of treatment with probiotics or placebo, and at the end of follow-up (W14). NS indicates not significant; OTU, operational taxonomic unit; P, placebo; T, treated.





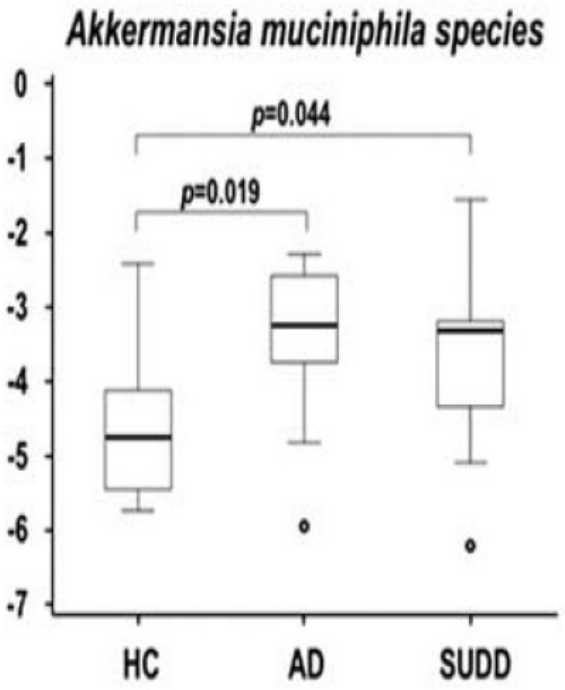


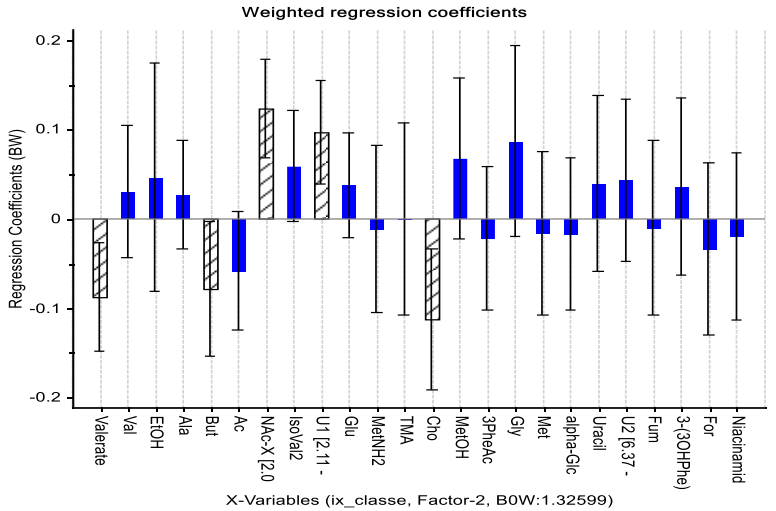


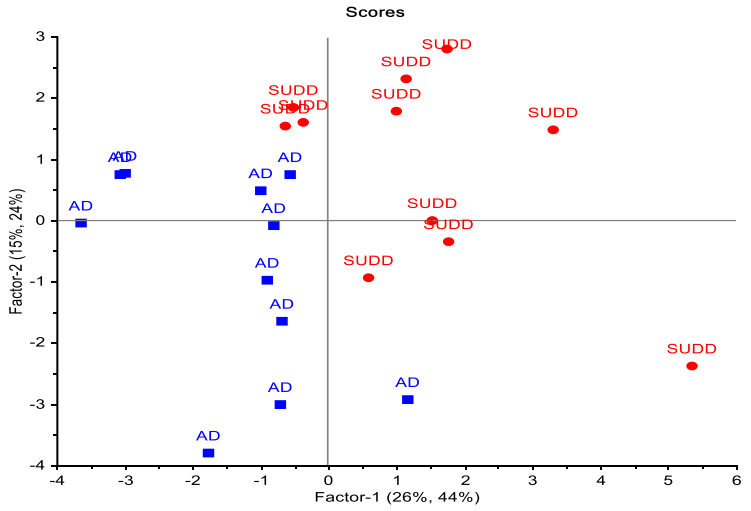
Endoscopic finding of diverticular inflammation in patients with symptomatic diverticular disease

Assessment of Fecal Microbiota and Fecal Metabolome in Symptomatic Uncomplicated Diverticular Disease of the Colon

Antonio Tursi, MD, Paola Mastromarino, MD,† Daniela Capobianco, BSci,† Walter Elisei, MD,‡ Alfredo Miccheli, MD,§ Giorgio Capuani, PhD,§ Alberta Tomassini, PhD,§ Giuseppe Campagna, MS, PhD,‖ Marcello Picchio, MD,¶ GianMarco Giorgetti, MD,# Federica Fabiocchi, MD,# and Giovanni Brandimarte, MD***







Akkermansia mucinphila

J Clin Gastroenterol 2016;50:S9–S12

(Giorno 0)

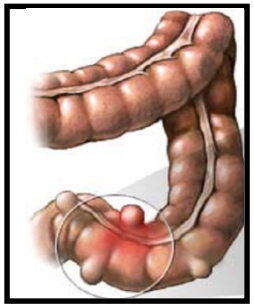
T0

2 settimane di Trattamento con:

Probiotico (DSF)® (P) o Rifaximina (R) o Mesalazina (M) o Fibre (F)

T1

(Giorno14)



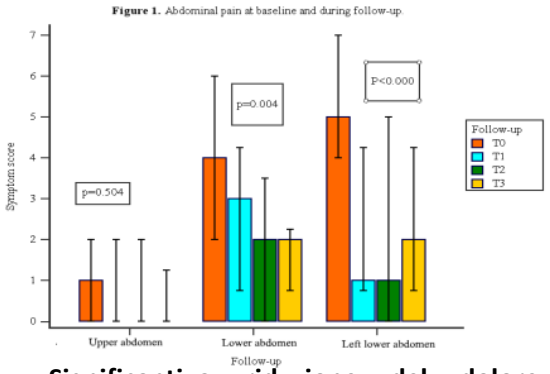
T2

(Giorno 44)

T3

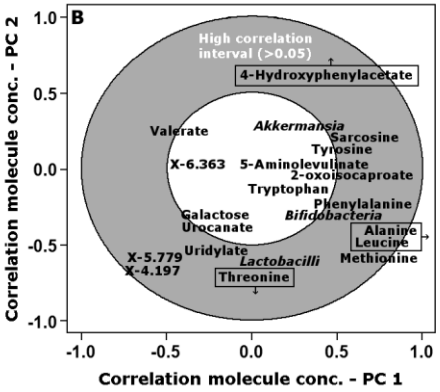
(Giorno 74)

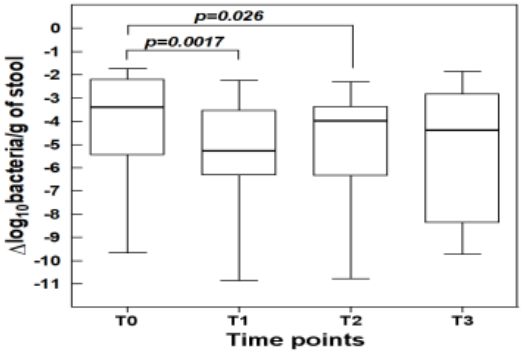
Il Microbiota ed il Metaboloma cambiano sotto trattamento nei pazienti con SUDD, confermando il possibile ruolo della disbiosi/dismetabolismo nella patogenesi della malattia



Significantiva riduzione del dolore addominale in fossa iliaca sinistra durante il follow-up (p<0.0001), ma con nuovo incremento a T3.

Il MODELLO DI PCA DI T0 vs T1 DIFFERISCE SIGNIFICATIVAMETNE COME BATTERI E METABOLITE





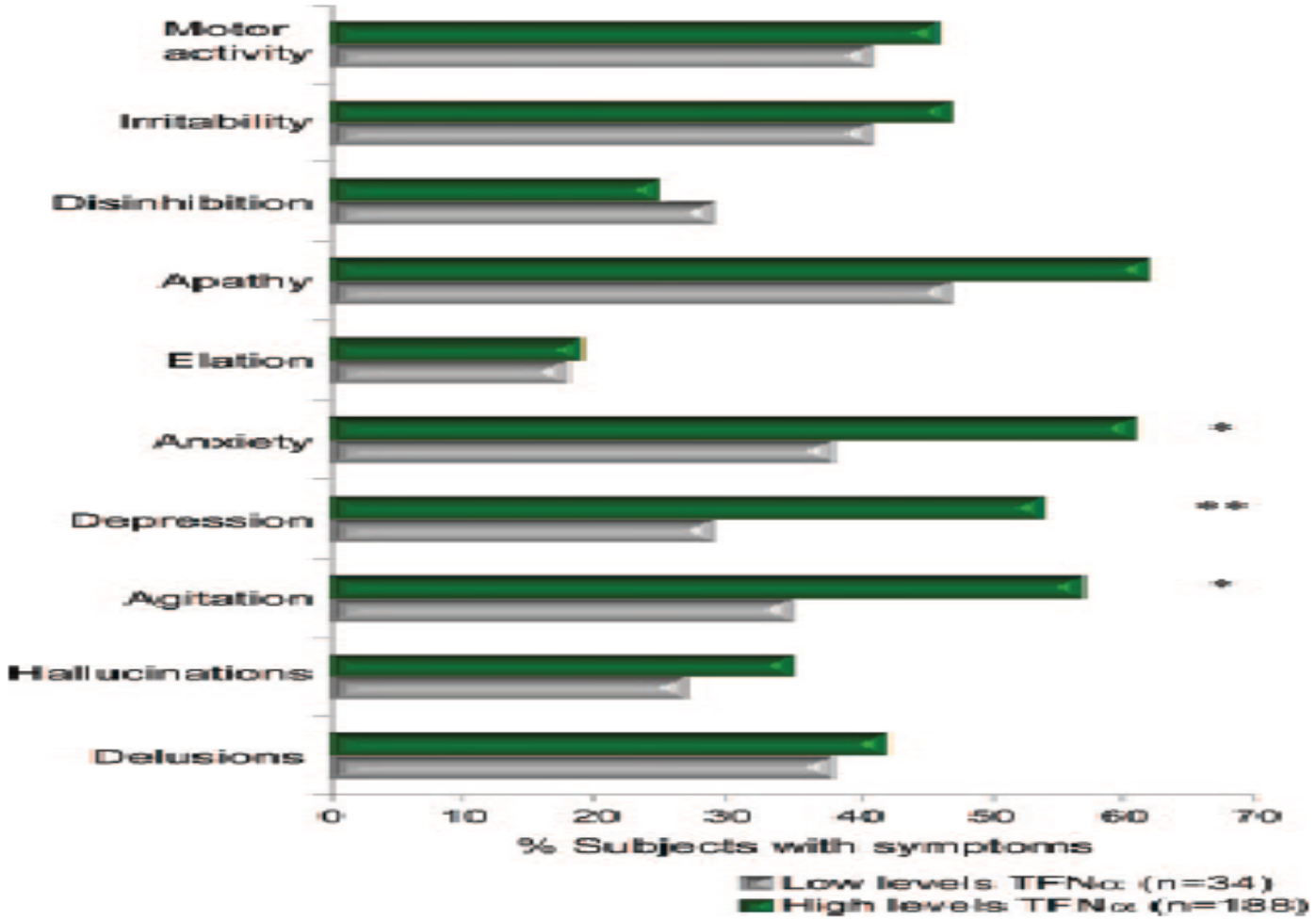
La quantità totale di *Akkermansia muciniphila* si riduceva a T1 (p=0.017) e T2 (p=0.026), mentre a T3 tornava simile ai livelli di T0 (p=0.09).

Laghi L. J Biol Regul Homeost Agents 2018;32: 1421-1432

MA GUT MICROBIOTA AGISCE SULLA BARRIERA ENCEFALICA DELL‘ANZIANO??

Proinflammatory cytokines, sickness behavior, and Alzheimer disease

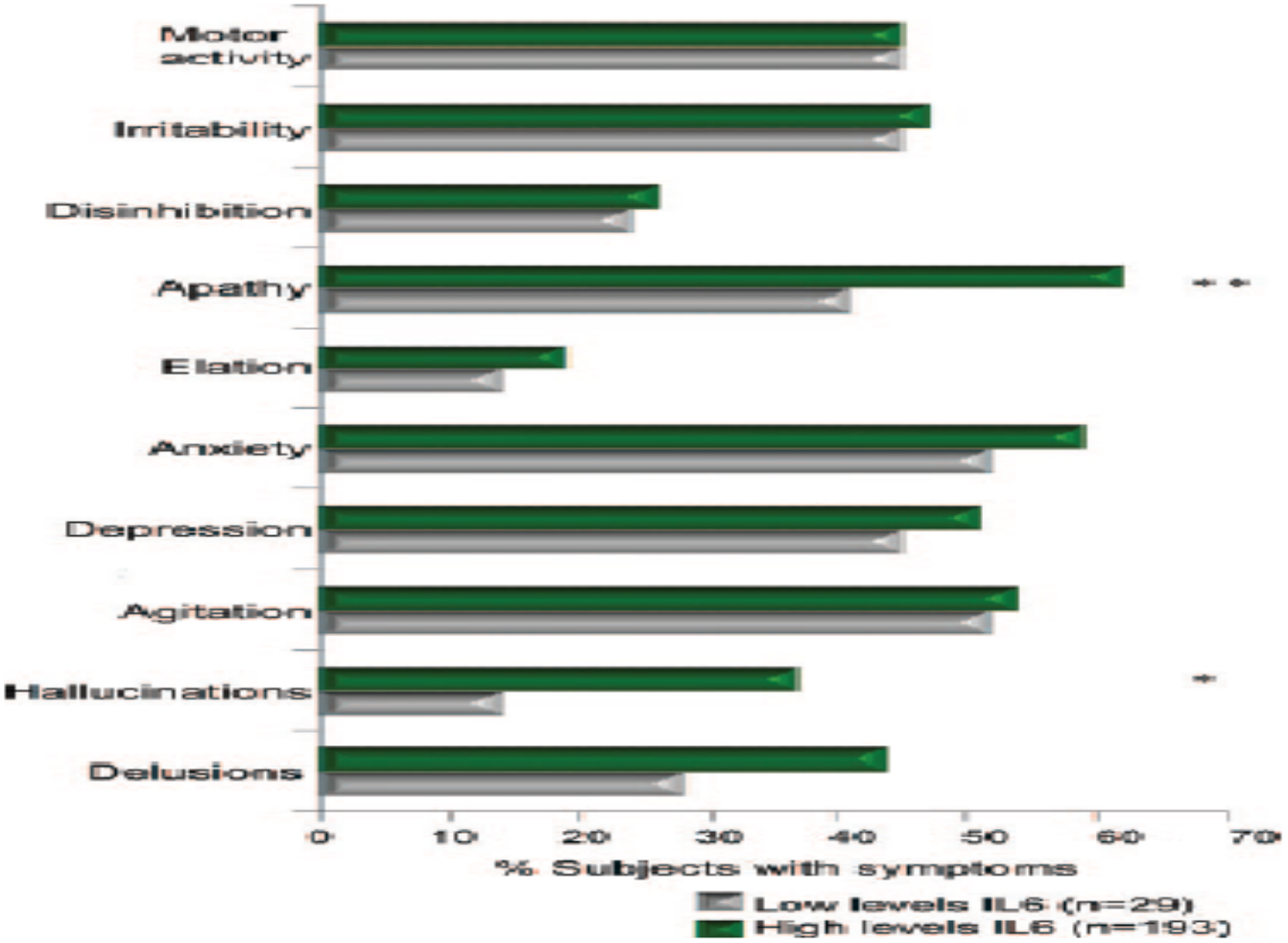
Figure 1. Frequency distribution of neuropsychiatric features in subjects with Alzheimer disease over the 6-month follow-up period by low or high levels of tumor necrosis factor-α (TNFα)



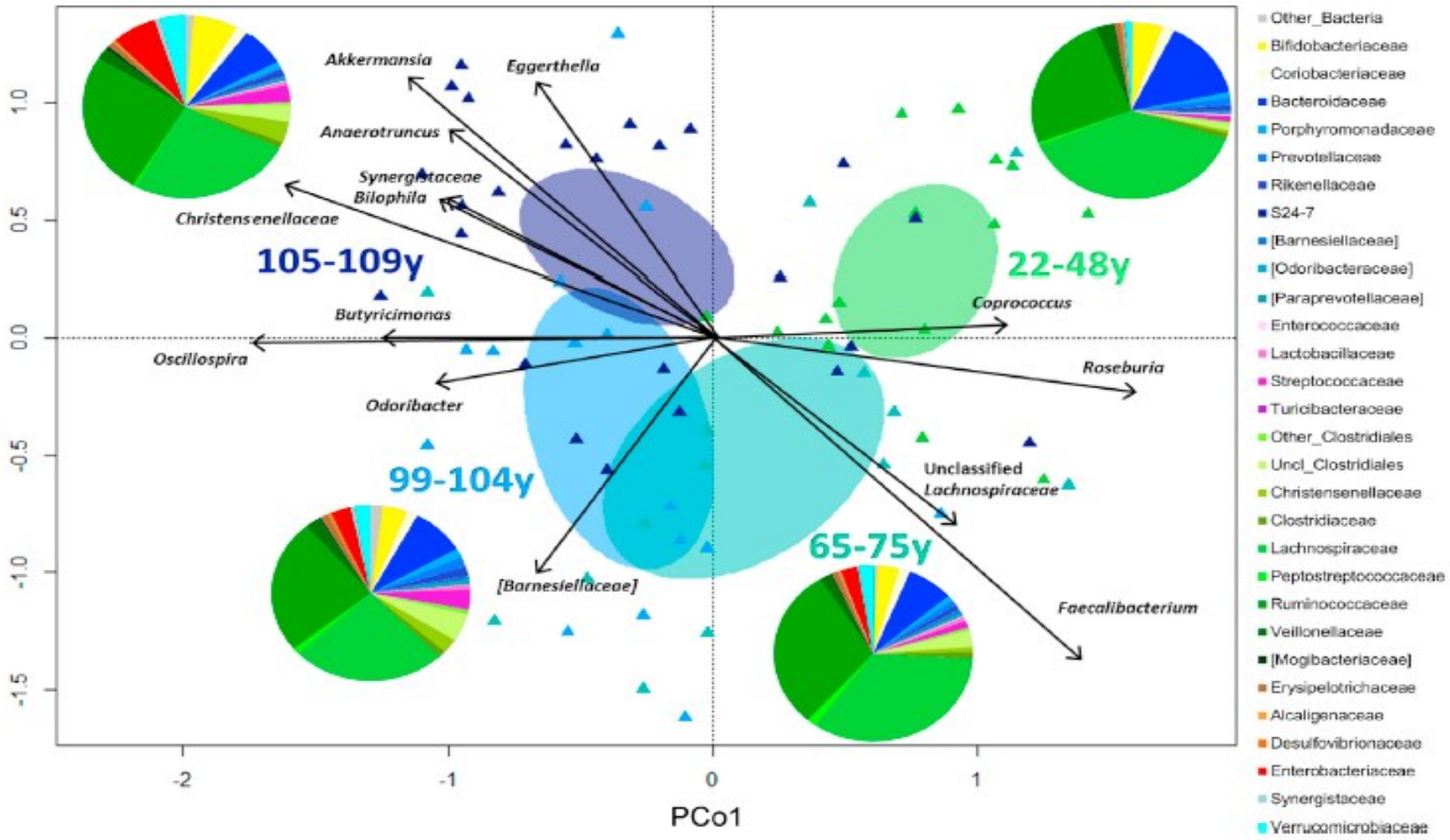
*p ≤ 0.05, **p ≤ 0.01, adjusted for baseline age, gender, Alzheimer's Disease Assessment Scale Cognitive subscale score, and presence of delirium during follow-up.

Proinflammatory cytokines, sickness behavior, and Alzheimer disease

Figure 2 Frequency distribution of neuropsychiatric features in subjects with Alzheimer disease over the 6-month follow-up period by low or high levels of interleukin-6 (IL-6)



*p ≤ 0.05, ** p ≤ 0.01, adjusted for baseline age and gender.



Current Biology 26, 1–6, June 6, 2016 ª 2016 Elsevier Ltd.

« …. La morte si trova nel viscere …
e la cattiva digestione
è la radice di ogni male….»

grazie dell’attenzione!!!!

gianmarcogiorgetti@hotmail.com