

8[^] edizione
Top Ten
IN GASTROENTEROLOGIA

17-18 MARZO 2017

ISEO (BS)

Iseo Lago Hotel - Via Colombera, 2

Patrizia Brigidi

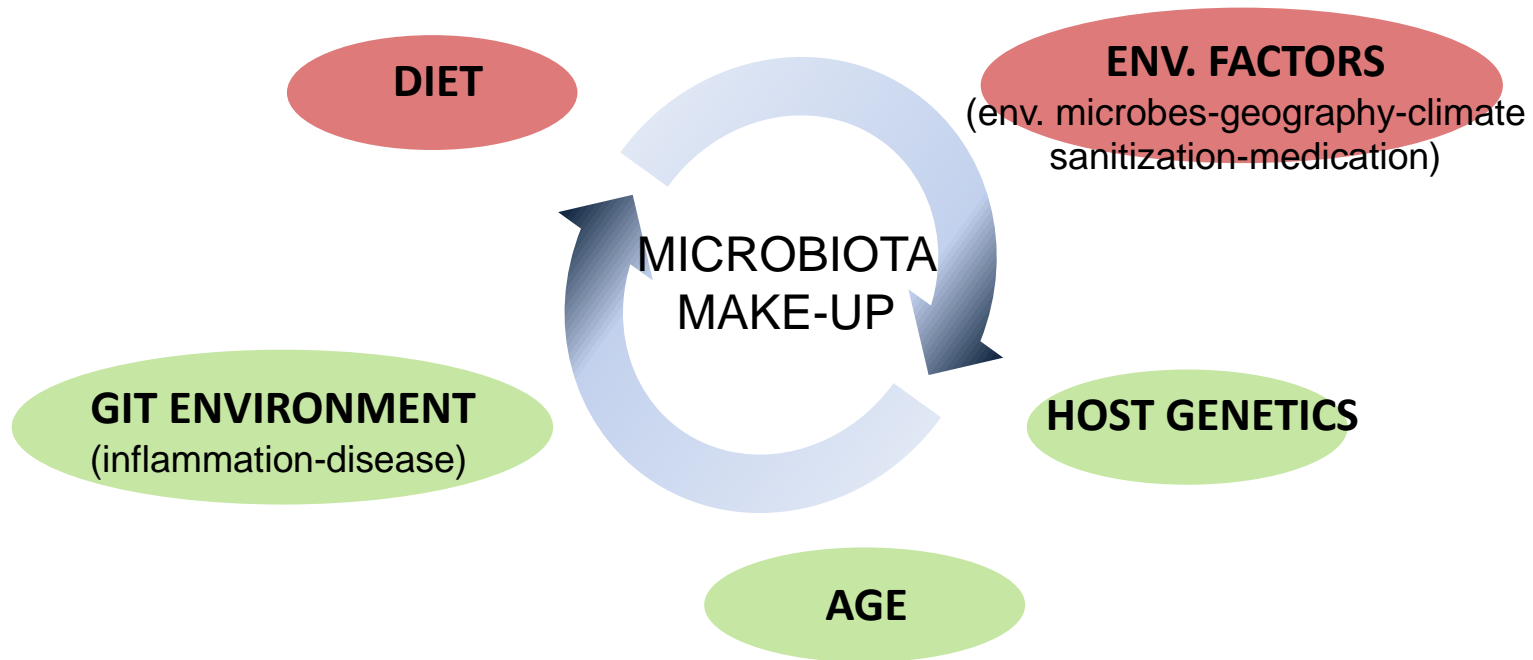
LA DISBIOSI INTESTINALE

Cosa è, come diagnosticarla come
curarla



MICROBIOTA PLASTICITY

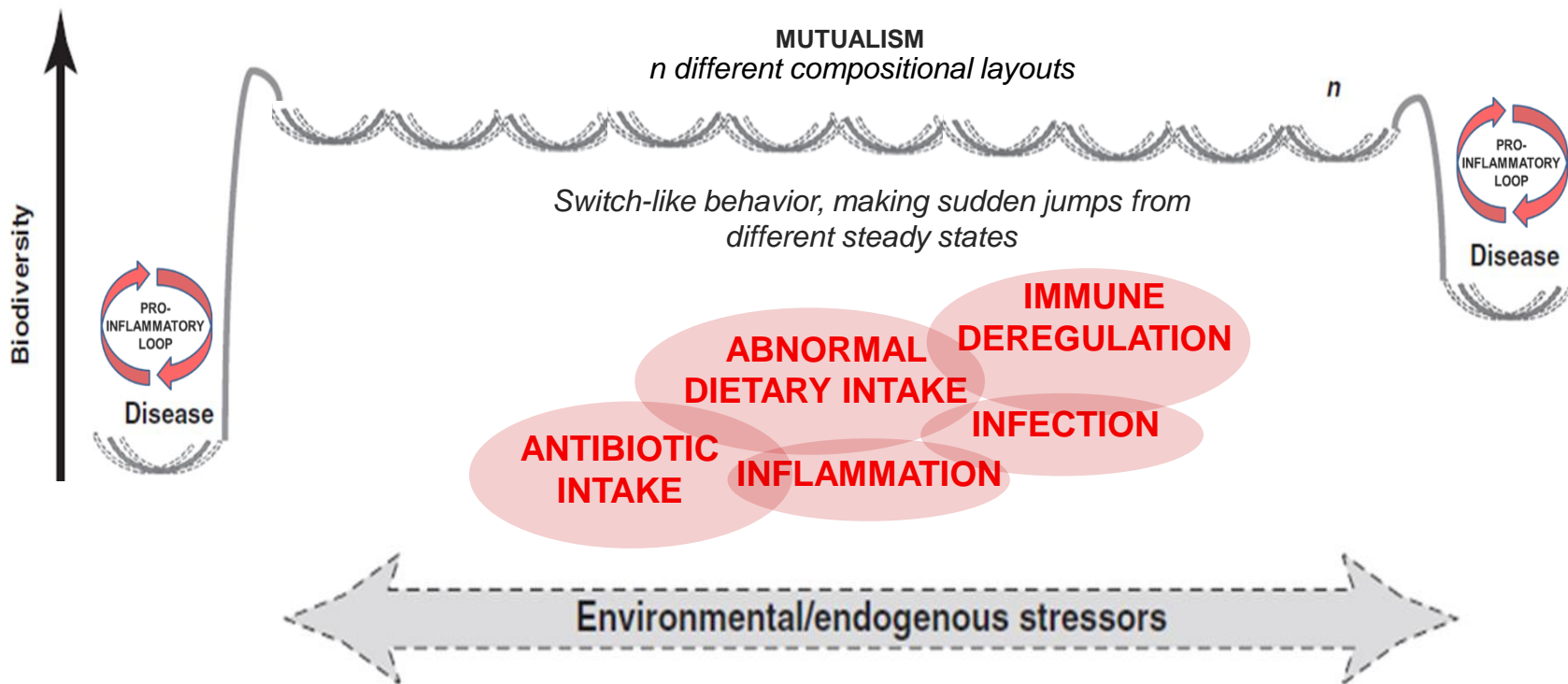
THE INDIVIDUAL MICROBIOTA COMPOSITION CONTINUOUSLY CHANGES IN RESPONSE TO **EXTRINSIC** AND **INTRINSIC** VARIABLES



IN A MUTUALISTIC CONTEXT, THE PLASTICITY OF THE HUMAN MICROBIOTA GUARANTEES A RAPID ADAPTATION OF THE SUPER-ORGANISM IN RESPONSE TO DIET CHANGES, AGE, ETC
there is a strong selection towards a readily changeable individual microbiome profile

MUTUALISM BREAKDOWN

the GM is a multistable system with a variable fraction at 40% of the total community

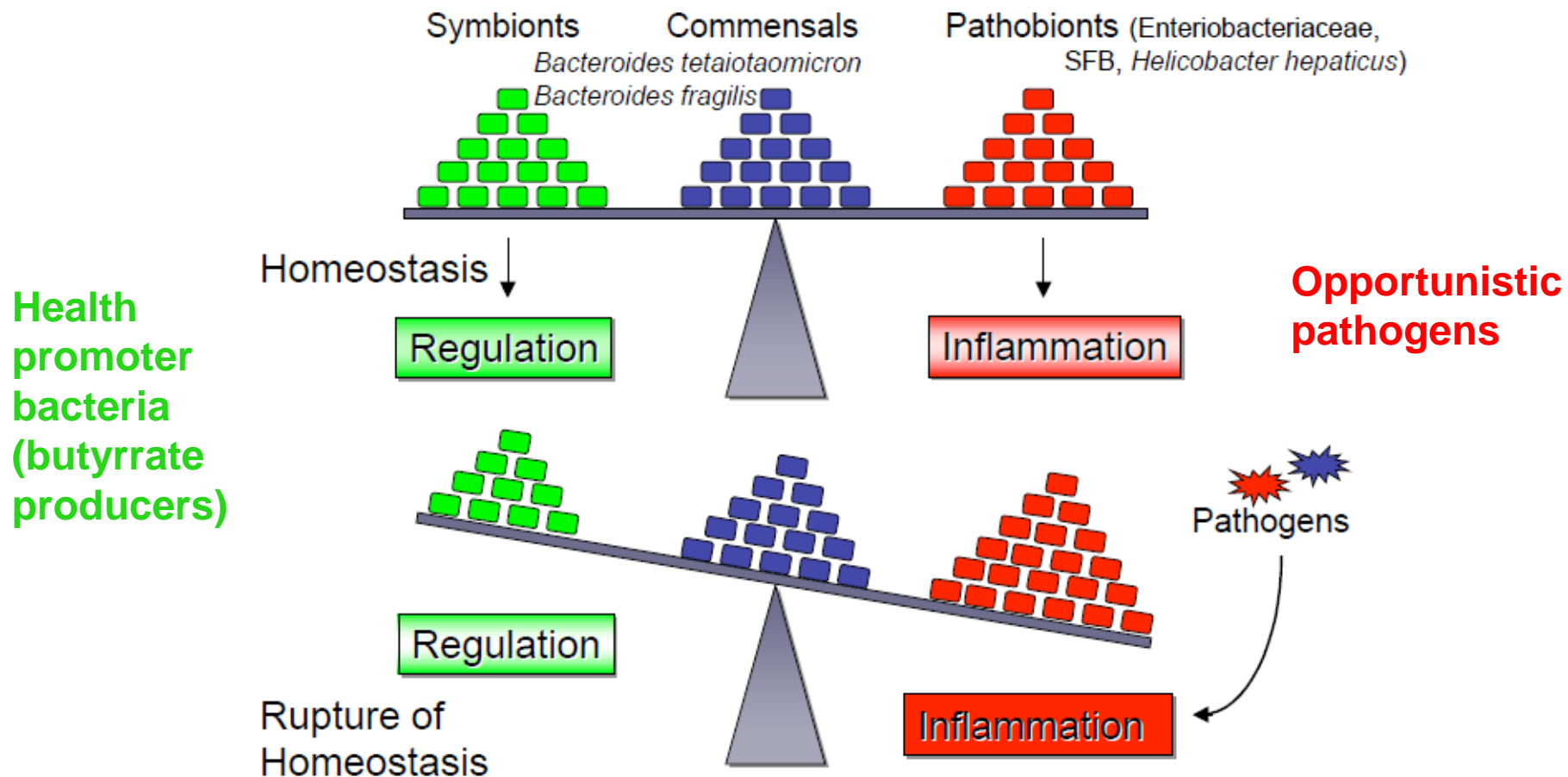


RUPTURE OF THE GM-HOST MUTUALISTIC AGREEMENT AND COMPROMISED HOST ENERGY BALANCE AND IMMUNE HOMEOSTASIS

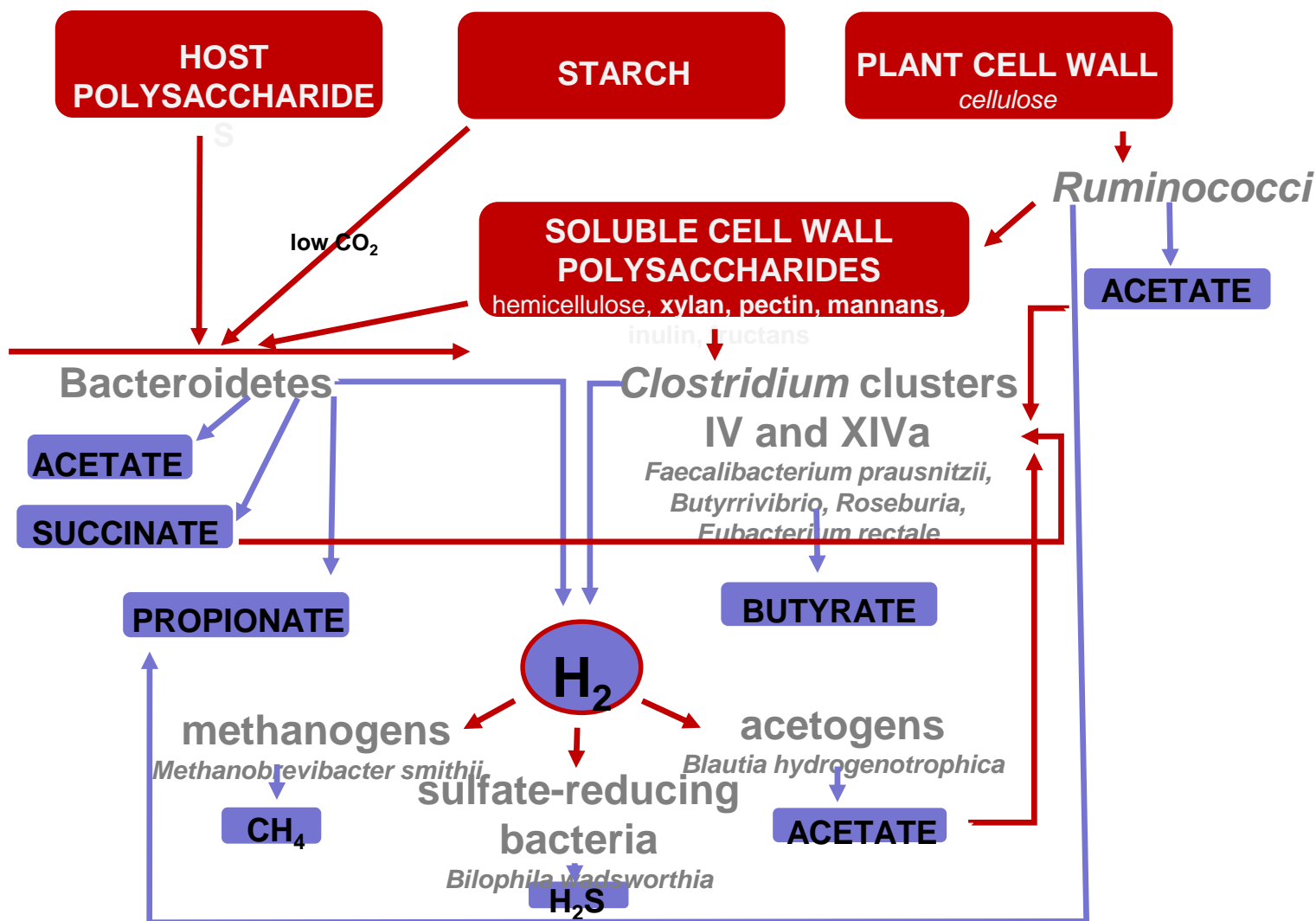
COMMON TRAITS OF DYSBIOSIS

- Reduction of SCFA producing bacteria (butyrate producers such as *Faecalibacterium*, *Roseburia*, *Lachnospiraceae*, *Eubacterium*)
- Increased mucus degradation potential by abnormal mucin degraders that displace *Akkermansia*
- Reduced hydrogen and methane production combined with increased hydrogen sulphide production. H₂S is toxic for the epithelium
- Increase in abundance of bacteria with LPS endotoxins (Proteobacteria) that can drive inflammation
- Increased potential to manage oxidative stress, i.e. microbes became able to proliferate in close vicinity to the epithelium

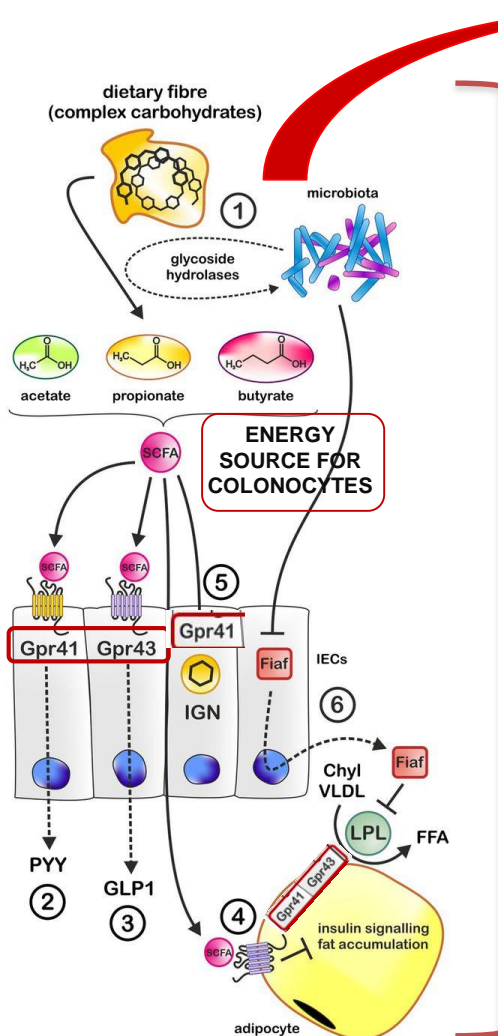
IMMUNOLOGICAL DISREGULATION ASSOCIATED WITH MICROBIOTA DYSBIOSIS



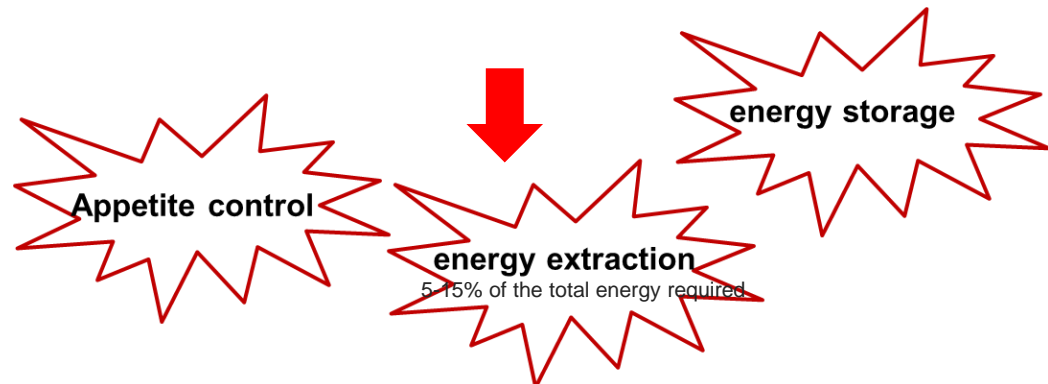
SCFA PRODUCTION: SYNTROPHIC MICROBIAL NETWORKS



SCFA POSSESS A KEY MULTIFACTORIAL ROLE IN METABOLIC HOMEOSTASIS

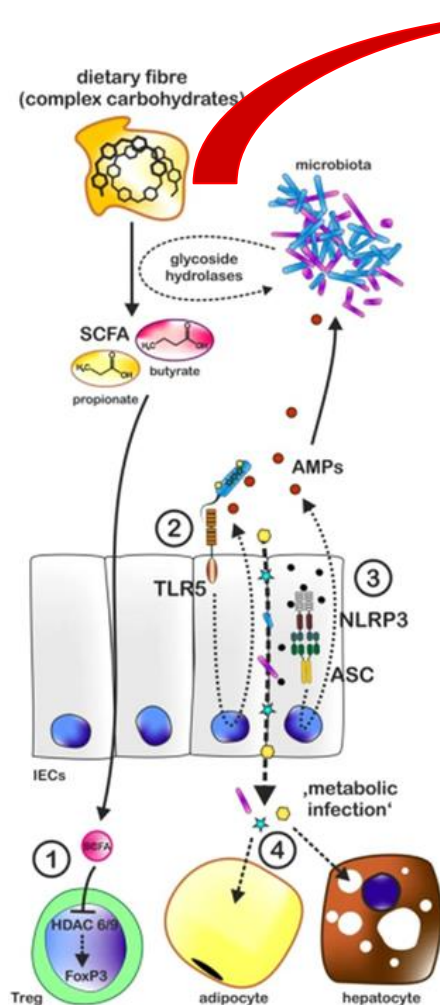


- ✓ **peptide Pyy expression (2):** inhibition of gut motility; increase of intestinal transit rate; reduction of energy harvest from diet
- ✓ **glucagon-like peptide 1 expression (3):** increase of insulin sensitivity
- ✓ **intestinal gluconeogenesis activation (5):** favors glucose control
- ✓ **expression of fasting-induced adipose factor (6):** favors fat storage
- ✓ **suppression of insulin signaling in adipose tissue (4)**

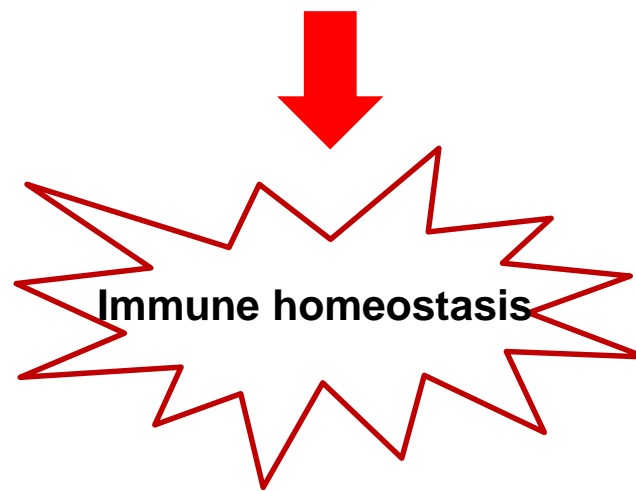


Tilg *et al.*, Gut 2014, Koh *et al.*, Cell, 2016

SCFA POSSESS A KEY MULTIFACTORIAL ROLE IN REGULATION OF THE HOST IMMUNE FUNCTION



- ✓ development of colonic and extrathymic Treg
- ✓ regulation of bone marrow hematopoiesis
- ✓ regulation of dendritic cell function



Tilg *et al.*, Gut 2014, Koh *et al.*, Cell, 2016

SIDE PRODUCTS FROM GM PROTEIN FERMENTATION

AMINO ACIDS

DETRIMENTAL FOR THE HOST HEALTH

proteolytic
clostridia and
Bacteroidetes

(*Alistipes*)

SCFA

BCFA

**PHENOLIC AND
INDOLIC
METABOLITES**

METHYLAMINES

**IMPACT ON
HOST
PHYSIOLOGY**

**AROMATIC
AMINO ACIDS**

PHENOLIC AND INDOLIC
METABOLITES
(indoles; p-cresol; phenols;
phenylacetic acid)

**LIVER
STEATOSIS,
OBESITY AND
DIABETES**

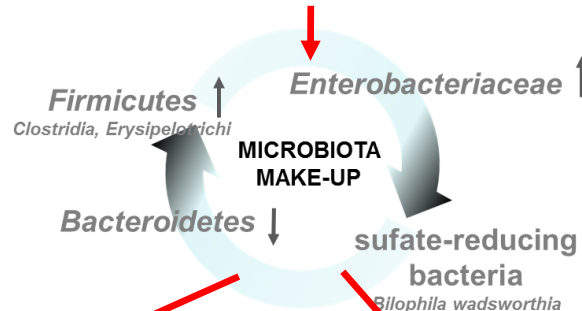
**ALIPHATIC
AMINO ACIDS**

METHYLAMINES

**IMMUNE
ACTIVATION
AND
DIABETES**

IMPACT ON HOST PHYSIOLOGY OF THE GM ADAPTATION TO FAT

DIETARY FATS → BILE ACIDS



BIOACTIVE COMPOUNDS

SECONDARY BILE ACIDS

reabsorption and

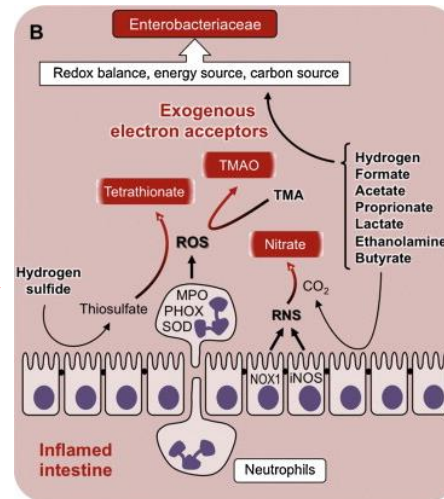
MODULATION OF FXR AND TGR5 RECEPTORS

REGULATION OF DIETARY FAT ABSORPTION

OBESITY, TYPE II DIABETES

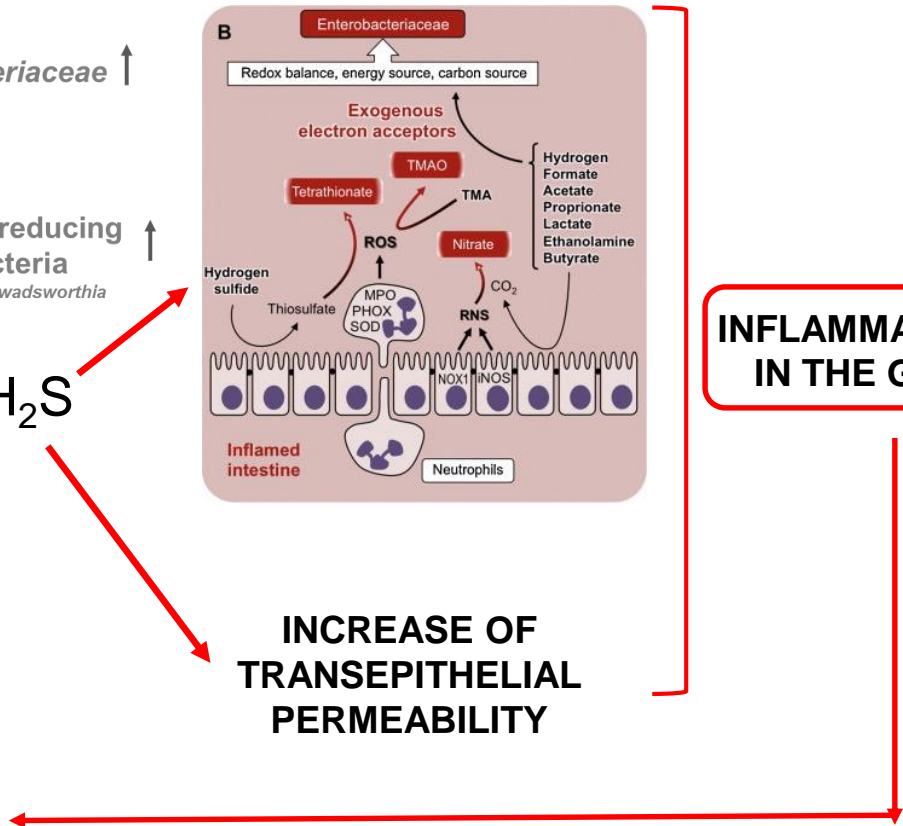
H₂S

PRO-INFLAMMATORY DYSBIOSES OF GM



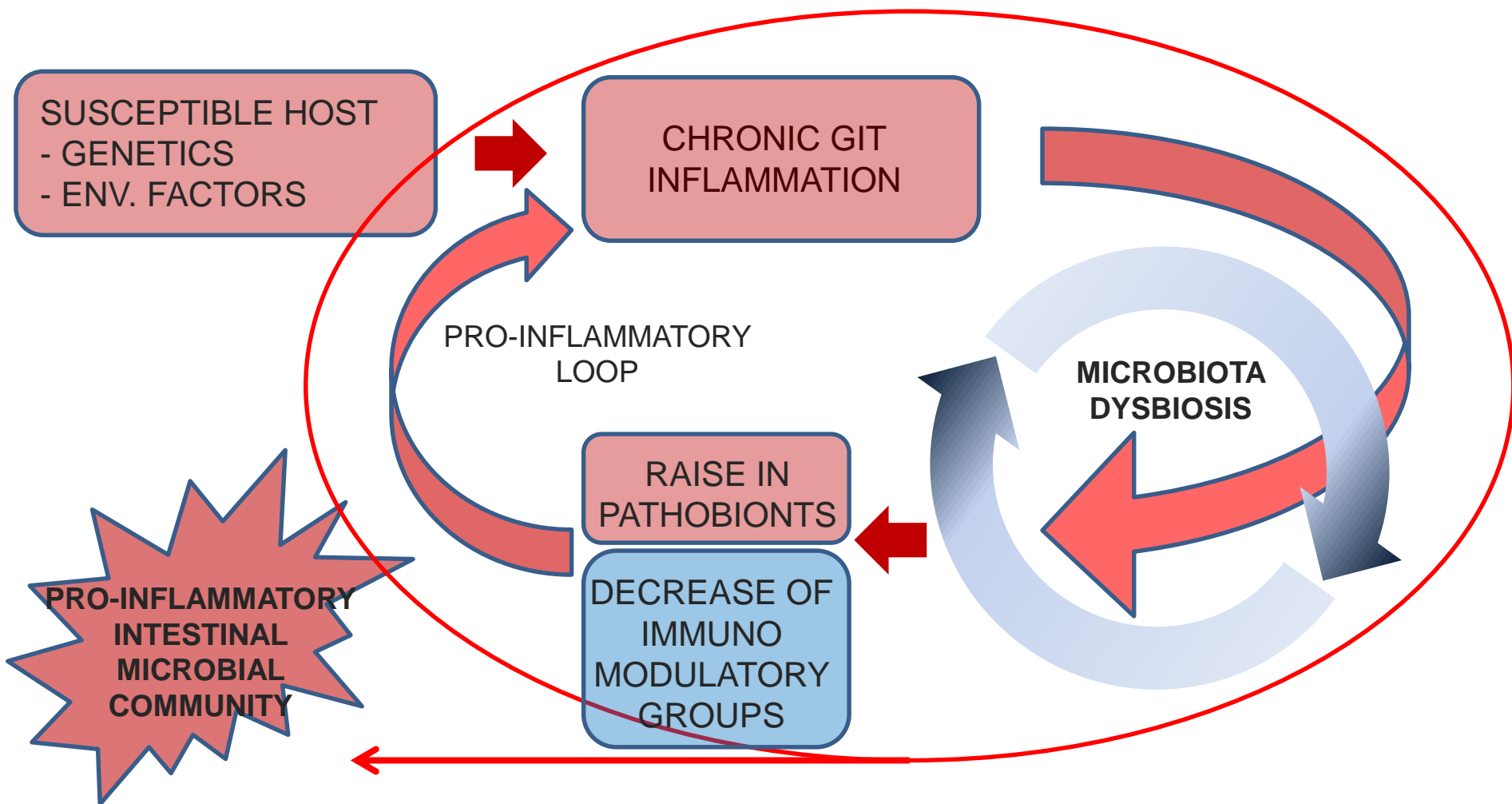
INFLAMMATION IN THE GUT

INCREASE OF TRANSEPITHELIAL PERMEABILITY



INFLAMMATION AND DYSBIOSIS

A non-controlled pro-inflammatory pathway can dramatically impact on the composition of the intestinal microbiota



GUT MICROBIOTA-ASSOCIATED DISORDERS

- **Nutrition related disorders** (obesity, type 2 diabetes, metabolic syndrome)
- **Inflammatory bowel diseases** (UC and CD)
- **Functional bowel disorders** (IBS)
- **Systemic complications of decompensated liver disease**
- **Cardio-vascular diseases**
- **Atopy/allergy**
- **Colo-rectal Cancer**
- **Neuro-developmental conditions** (autism, depression)
- **Arthritis**

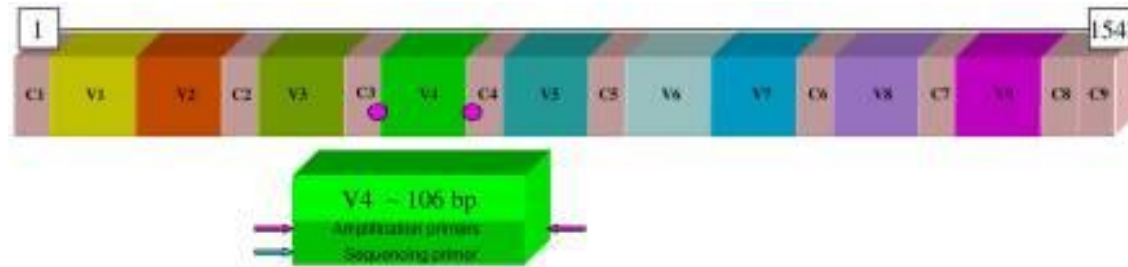
HOW TO ANALYZE THE GUT MICROBIOTA?

FIRST STEP OBTAIN TOTAL BACTERIAL DNA



DNA from all bacteria present in a sample (*feces, but also ileal content or biopsy*) need to be recovered

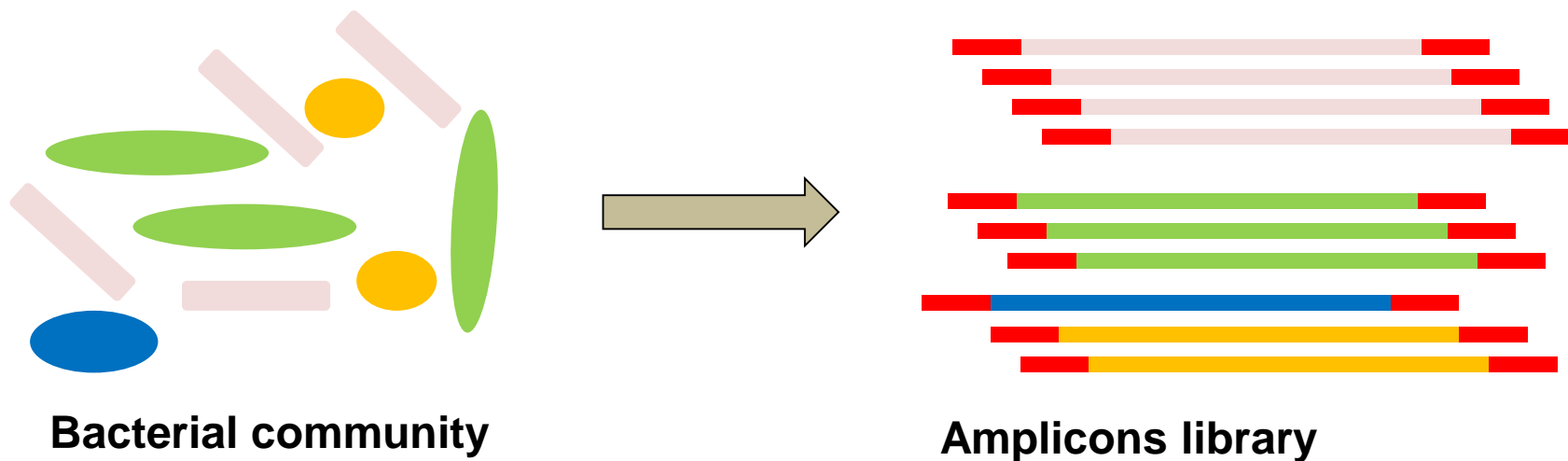
16S BACTERIAL SMALL SUBUNIT rRNA GENE-BASED SURVEY OF THE GUT MICROBIOTA



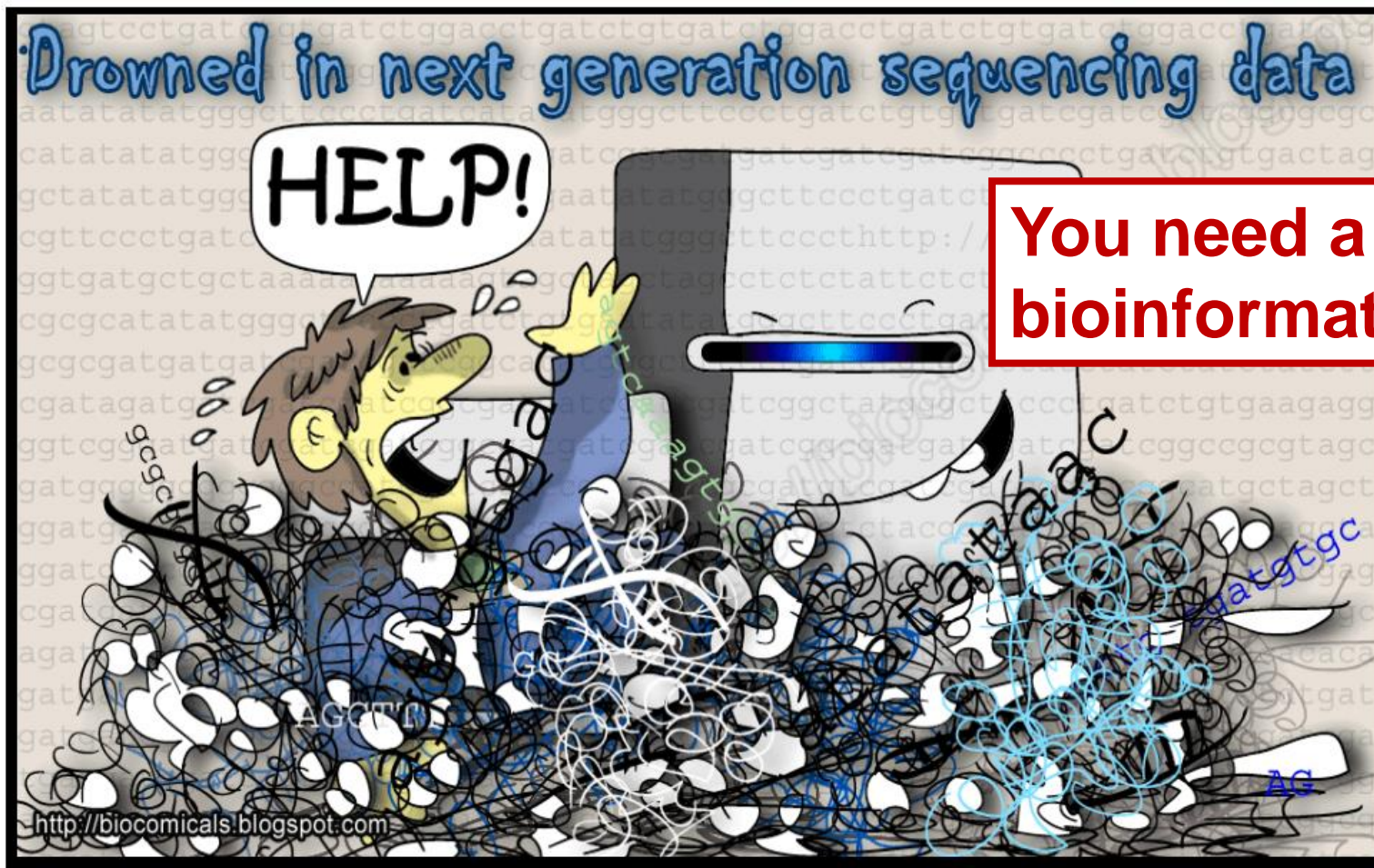
- Conserved regions allow the amplification of the 16S gene of ALL present bacteria with a unique PCR reaction
- Hypervariable regions allow to trace the species from whom they were amplified.

LAST 5 YEARS:
WIDESPREAD (AND PROGRESSIVELY DECREASING COST)
OF NEXT GENERATION SEQUENCING

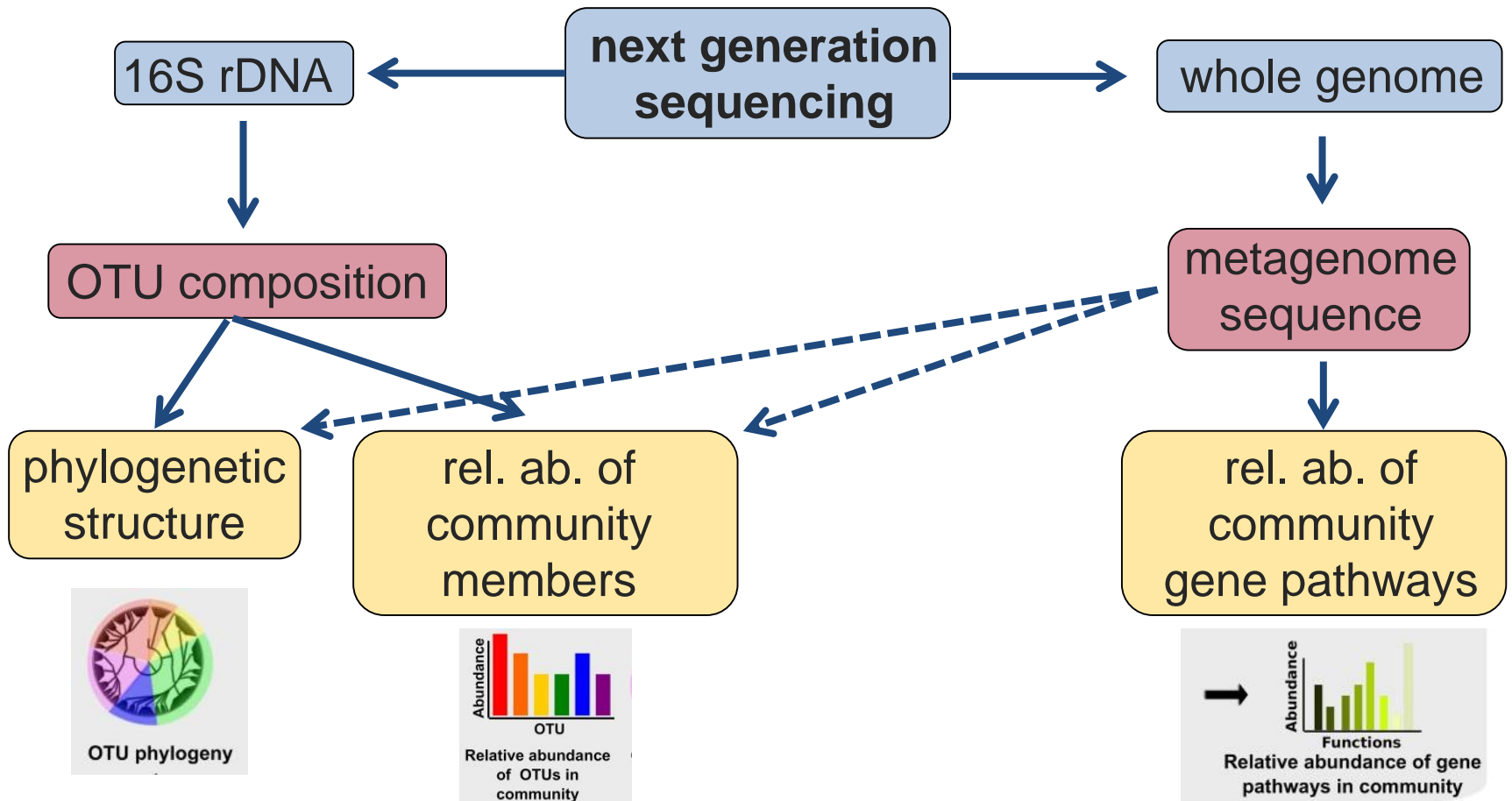
NEXT GENERATION SEQUENCING



«SEQUENCING» MEANS TO OBTAIN THE SEQUENCES OF ALL THE DNA MOLECULES IN THE AMPLICON LIBRARY

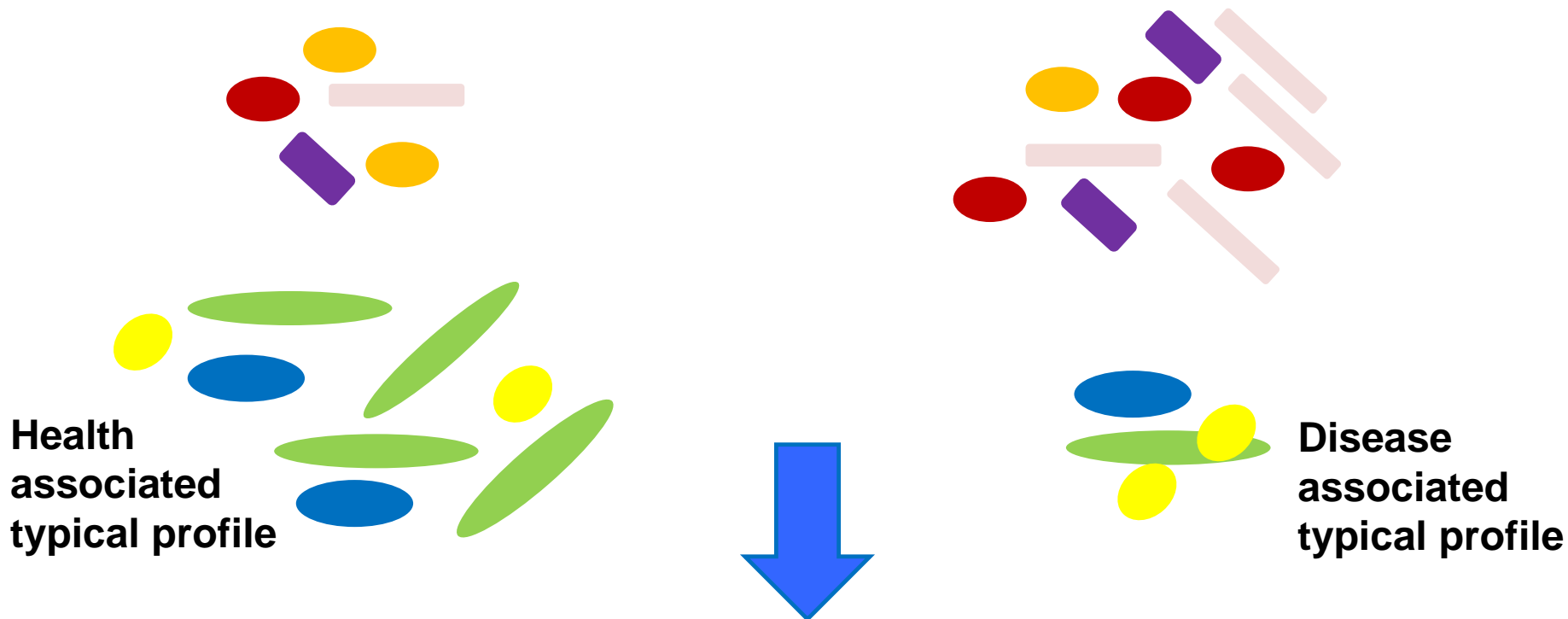


MICROBIOTA MOLECULAR ASSESSMENT



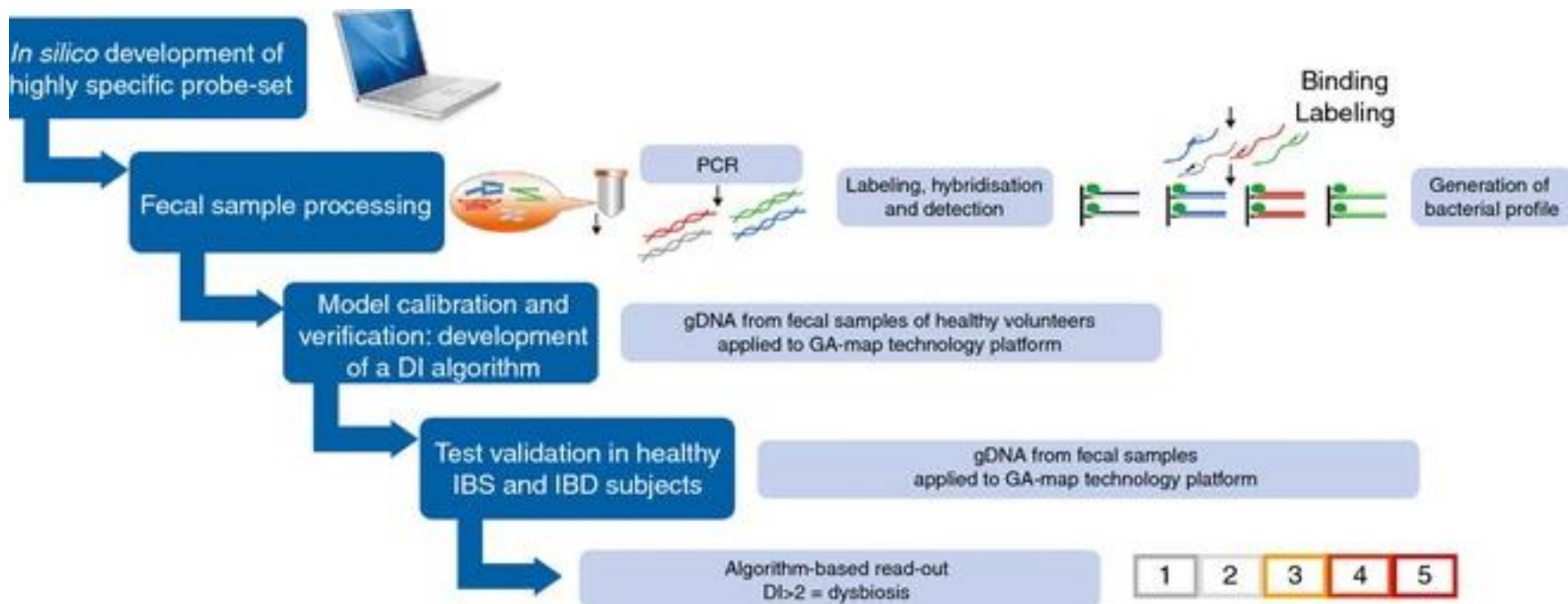
**STANDARDIZED DIAGNOSTIC
TOOLS TO PROFILE THE
MICROBIOTA AND CHARACTERIZE
DYSBIOSIS:
DEVELOPMENT OF A DYSBIOSIS
INDEX SCORE**

IDENTIFICATION OF MICROBIOTA SIGNATURES



MATHEMATICAL (AND STATISTICAL) ELABORATION OF INDEXES TO CLASSIFY THE CONTRIBUTION OF A SPECIFIC PROFILE TO THE RISK OF DEVELOPING/CONSOLIDATING DYSBIOSIS

GA-MAP DYSBIOSIS TEST



Deviations in human gut microbiota: a novel diagnostic test for determining dysbiosis in patients with IBS or IBD

FROM COMPOSITION TO FUNCTION (NGS)

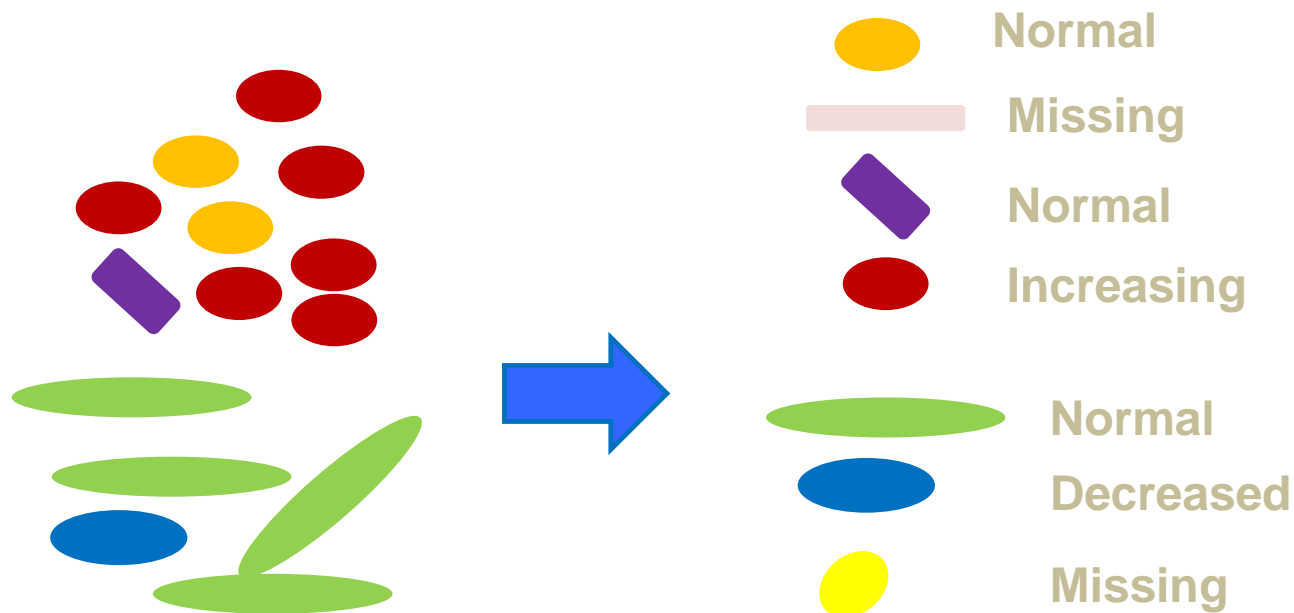
REDUCE COMPLEXITY

FOCUSING ON ESSENTIAL FUNCTIONS

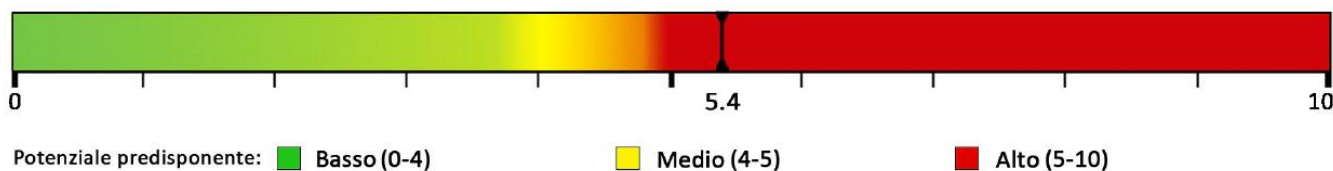
- **Short chain fatty acid production**
- **H₂S production**
- **LPS production**
- **Proteolytic activity**
- **Mucolysis**

DYSBIOSIS FEATURES AND RISK

Patient profile



CALCULATION OF THE SPECIFIC INDEX CONTRIBUTION TO DYSBIOSIS



I seguenti indici associano il profilo dell'ecosistema batterico analizzato con le **principali funzioni fisiologiche** dell'ospite in cui il microbiota intestinale è coinvolto. Tali indici sono calcolati tenendo conto delle abbondanze relative di quei gruppi batterici coinvolti in ognuna delle funzioni fisiologiche indicate.

Immunomodulazione

Il valore indica la potenzialità del microbiota di favorire il corretto funzionamento del sistema immunitario prevenendo l'insorgere di stati infiammatori causati da una scorretta attività del sistema immunitario stesso ed è calcolato sulla base dell'abbondanza relativa di gruppi batterici immunomodulanti.



Potenziale antinfiammatorio e immunomodulante: ■ Basso (0-5) ■ Medio (5-6.4) ■ Alto (6.4-10)

Malattie Infiammatorie Intestinali

Il valore indica la potenzialità del microbiota di favorire l'instaurarsi o consolidarsi di malattie infiammatorie intestinali come colite ulcerosa, morbo di Crohn, diverticolite ed è calcolato sulla base dell'abbondanza relativa di quei gruppi batterici coinvolti in questo processo.



Potenziale predisponente: ■ Basso (0-3.2) ■ Medio-Basso (3.2-4) ■ Medio-Alto (4-5) ■ Alto (5-10)

Permeabilità Intestinale ("Leaky Gut")

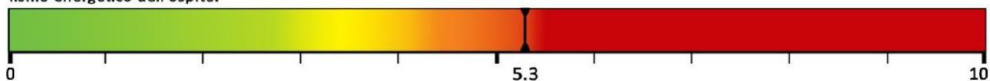
Il valore indica la potenzialità del microbiota di favorire la permeabilità intestinale calcolato sulla base dell'abbondanza relativa di gruppi batterici in grado di avere effetti negativi o positivi sull'integrità della mucosa intestinale. Un'eccessiva permeabilità intestinale può facilitare il passaggio di tossine dal lume intestinale ai tessuti circostanti, favorendo l'insorgenza di condizioni infiammatorie.



Potenziale predisponente: ■ Basso (0-4) ■ Medio (4-5) ■ Alto (5-10)

Obesità, Diabete di Tipo 2 e Sindrome Metabolica

Il valore indica la potenzialità del microbiota di favorire l'instaurarsi o il consolidarsi di disordini metabolici calcolato sulla base dell'abbondanza relativa di gruppi batterici obesogenici e anti-obesogenici, o in grado di avere un impatto sull'equilibrio del metabolismo energetico dell'ospite.



Potenziale predisponente: ■ Basso (0-3.4) ■ Medio-Basso (3.4-4.5) ■ Medio-Alto (4.5-5.5) ■ Alto (5.5-10)

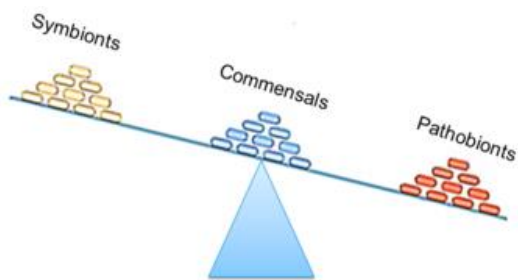
Invecchiamento

Il valore indica la potenzialità del microbiota di agire come fattore predisponente nei confronti di disordini tipici dell'invecchiamento come l'immunosenescenza e l'inflammaging (stati infiammatori tipici dell'invecchiamento) contribuendo a compromettere le normali funzioni immunologiche e metaboliche dell'individuo adulto sano.

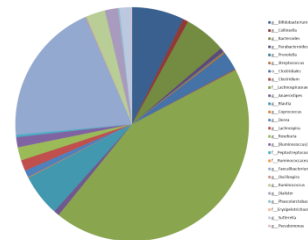


Indice d'invecchiamento: ■ Basso (0-1.9) ■ Medio-Basso (1.9-3.6) ■ Medio-Alto (3.6-4.9) ■ Alto (4.9-10)

WHAT CAN WE DO FOR A DYSBIOTIC MICROBIOTA?



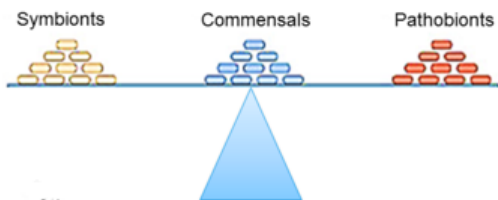
Altered profile



Phylogenetic characterization



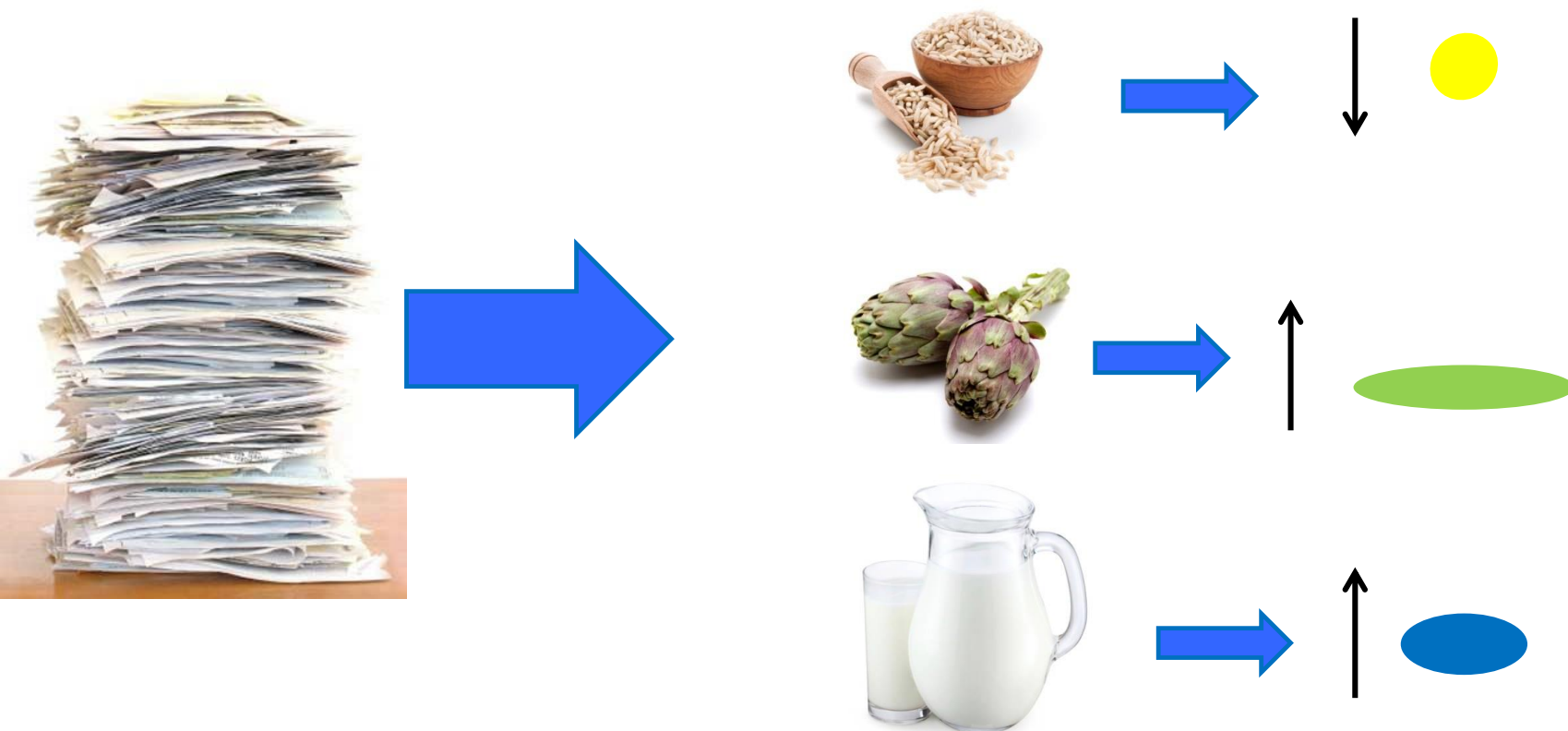
Personalized intervention strategy



Changes towards a healthy-like profile

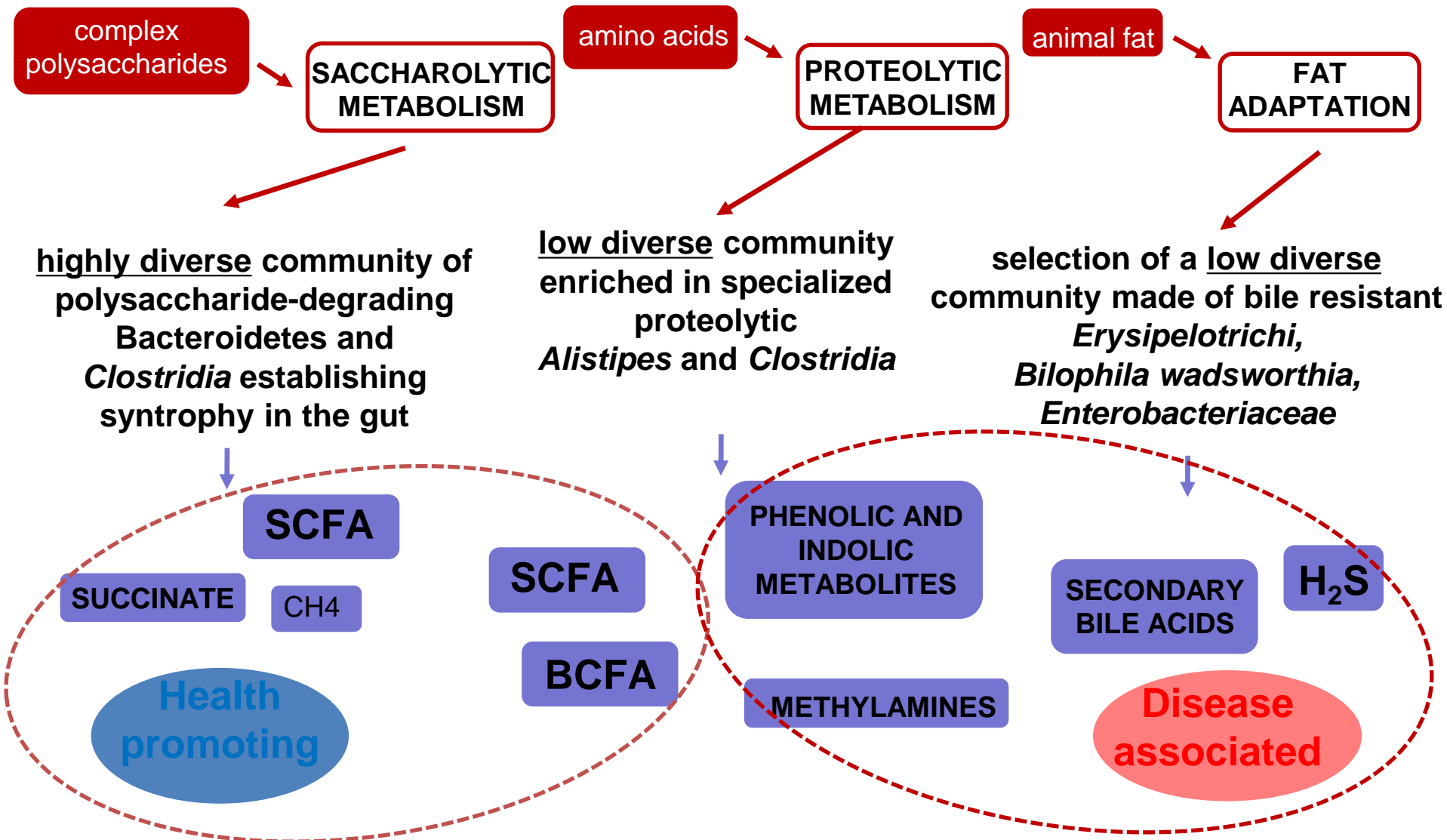
IDENTIFICATION OF APPROPRIATE DIETARY INTERVENTION

**Bacterial groups associated to increased/decreased
consumption of certain foods are identified**



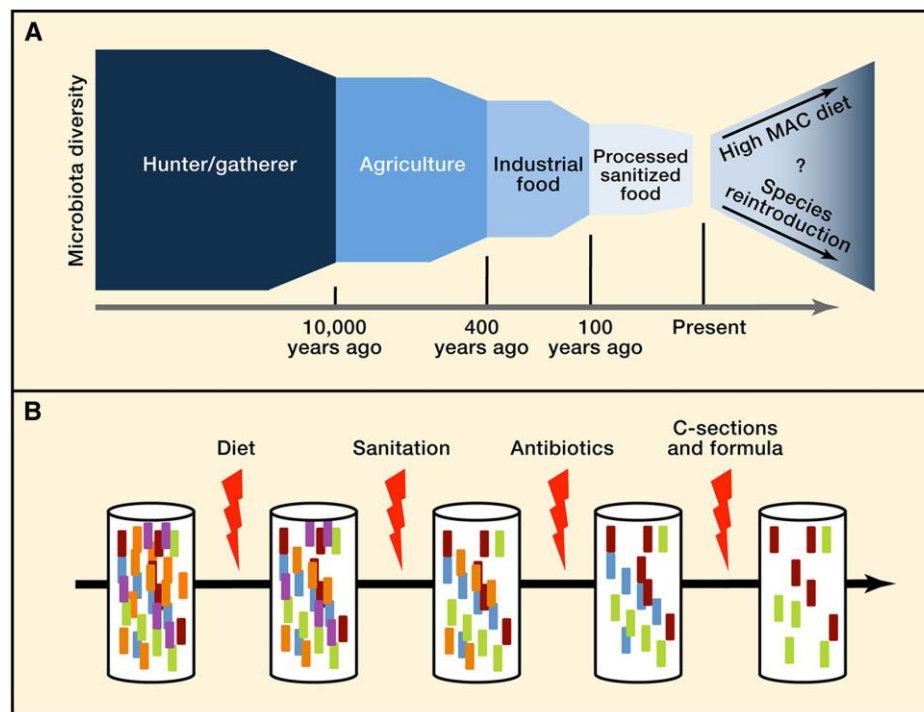
DIETARY INTERVENTION TO COUNTERACT DYSBIOSIS

diet regulates microbiota composition and metabolic output with a final impact on host physiology



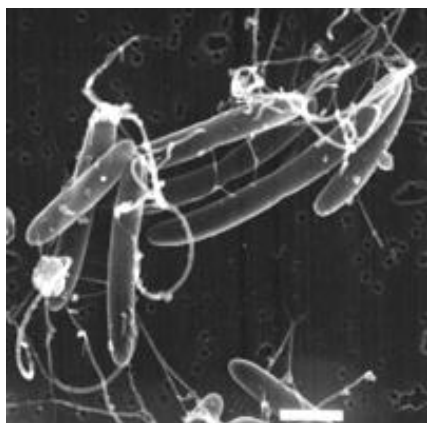
DIET AND DYSBIOSIS: THE MULTIPLE HIT HYPOTHESIS

The transition from rural communities to modern urban societies involve the progressive loss of gut microbiome species



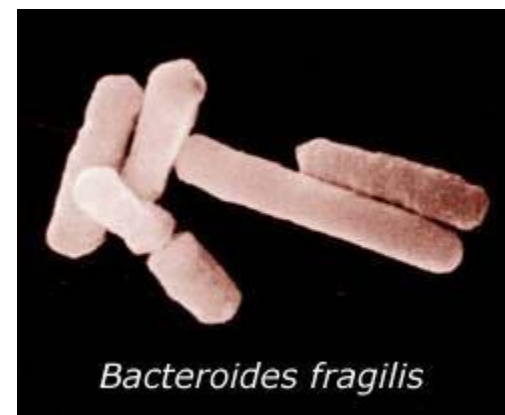
NEXT GENERATION PROBIOTICS

Faecalibacterium prausnitzii



Roseburia

Bacteroides



PREBIOTICS

‘Non digestible food ingredients that selectively stimulate a limited number of bacteria in the colon, to improve host health’

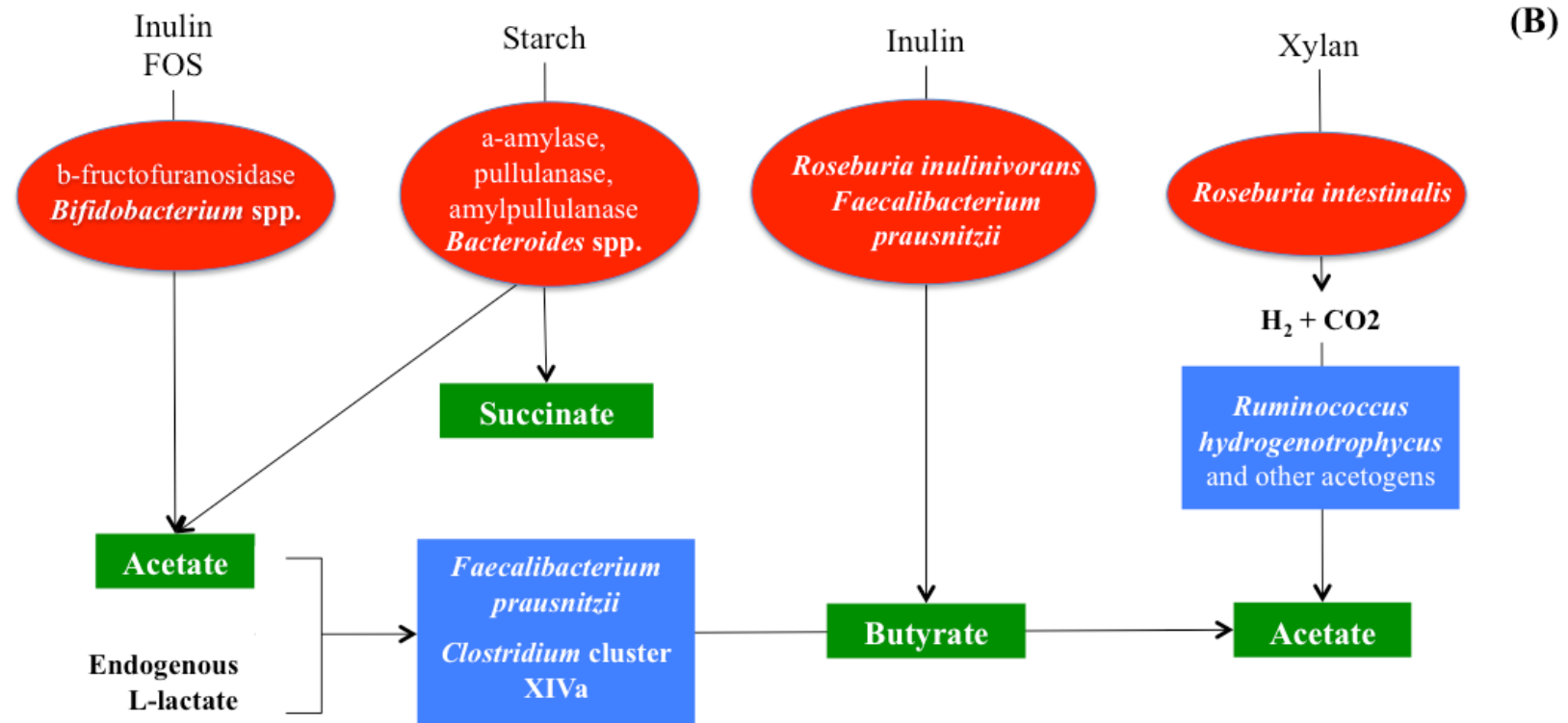
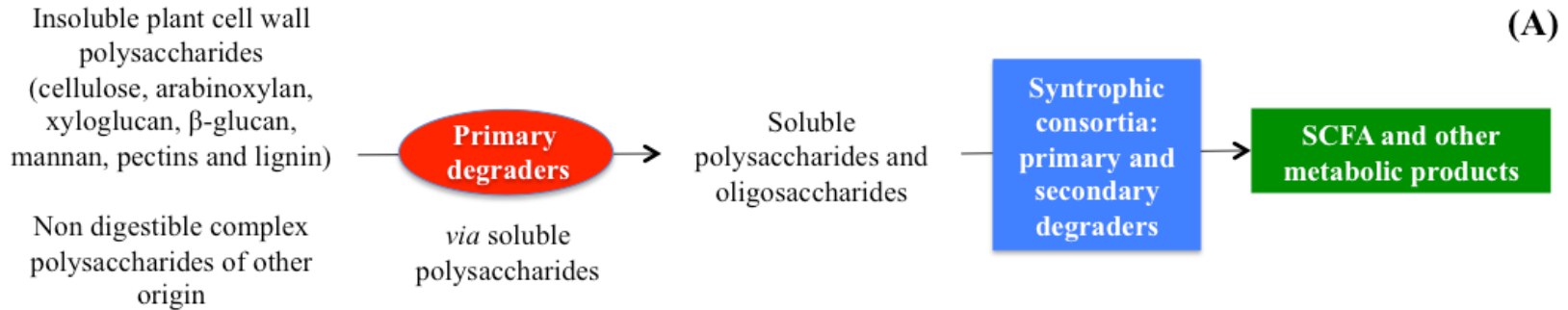
- Fructooligosaccharides (tested in humans)
- Lactulose (tested in humans)
- Trans-galactooligosaccharides

Present in: bananas, artichoke, leeks, onions, garlic, asparagus, chicory

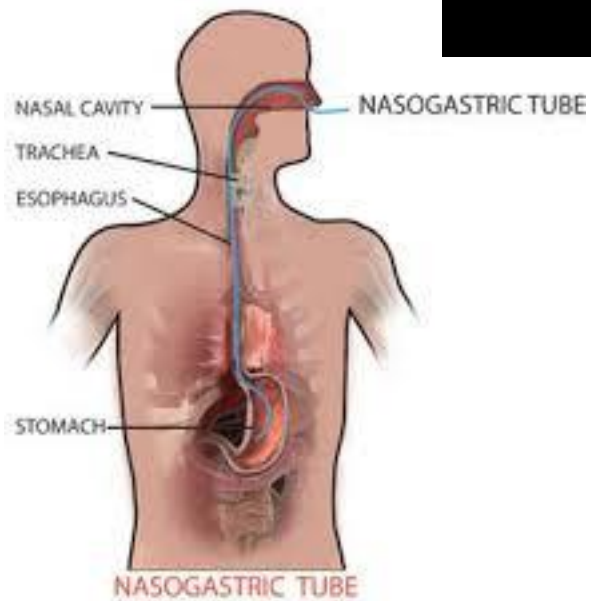
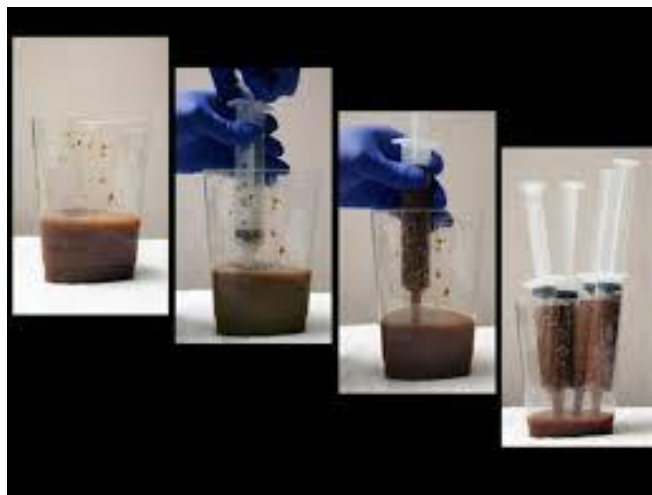
Application of prebiotics: Beverages and fermented milks, Health drinks, Infant formulae and weaning foods, Cereals, Biscuits, Confectionery, Cakes, Food supplements, Pet food, Farm animals



PREBIOTICS FERMENTATION IN HUMAN GUT



FECAL TRANSPLANTATION



MAIN GM RESEARCH CHALLENGES

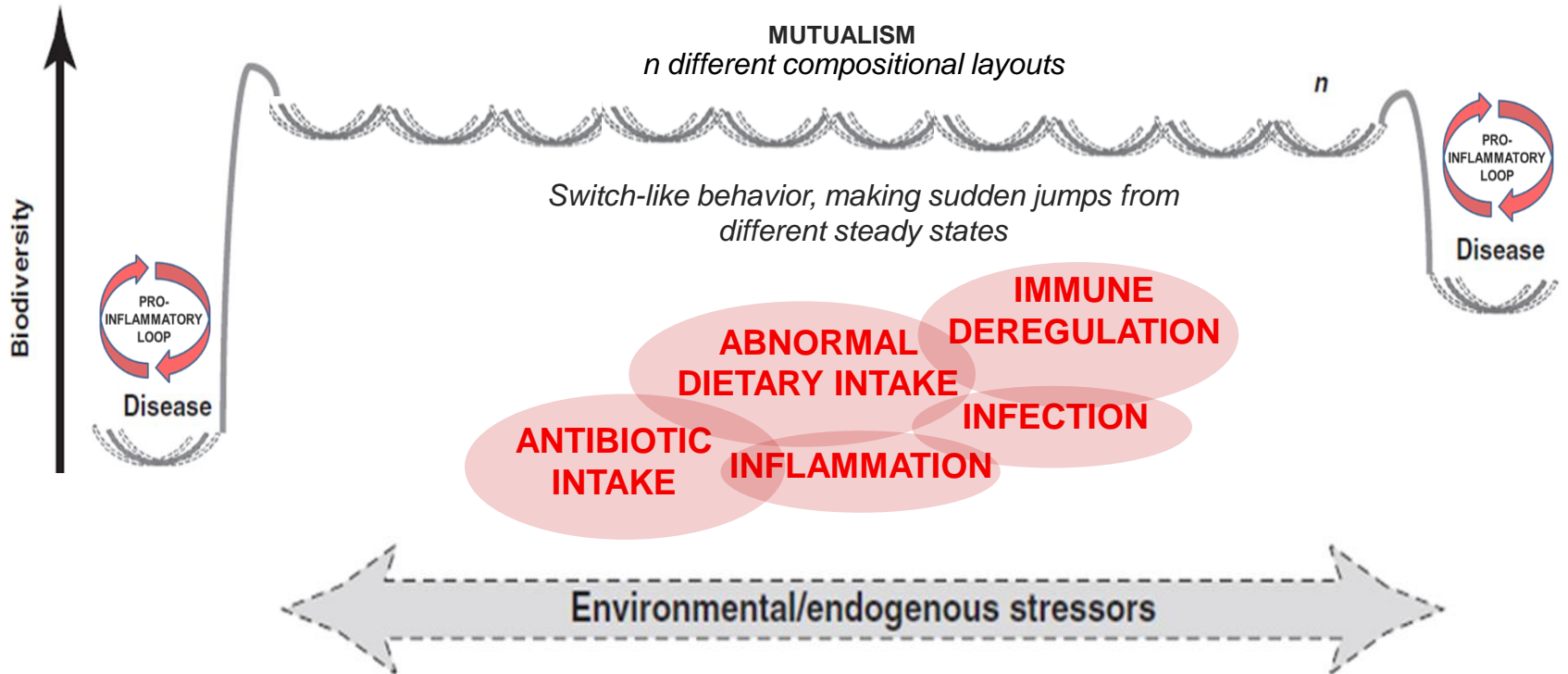
- ✓ **Variation in clinically relevant population**
- ✓ **Temporal variation and stability of biomarkers**
- ✓ **Effect of host genetics**
- ✓ **Effect of environment**
- ✓ **Microbiota-drug associations**
- ✓ **Clinical end points for pre-/pro-/ syn-biotics, functional foods, nutraceutical interventions**

THANKS FOR YOUR ATTENTION

<i>Marco Candela</i>			<i>Silvia Turrone</i>			
	<i>Matteo Soverini</i>		<i>Patrizia Brigidi</i>		<i>Sara Quercia</i>	
<i>Andrea Castagnetti</i>					<i>Simone Rampelli</i>	
			<i>Elena Biagi</i>			<i>Monica Barone</i>

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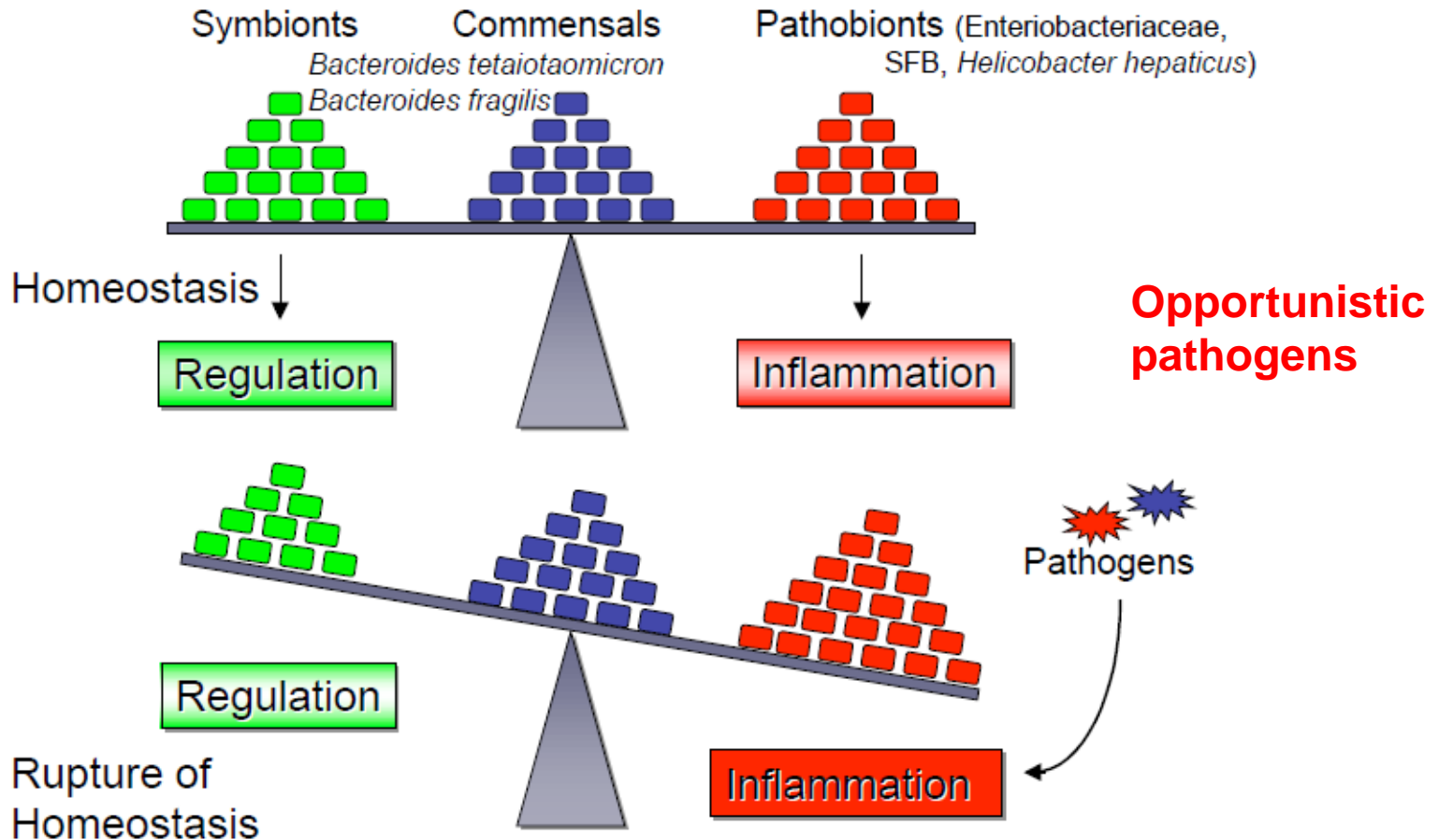
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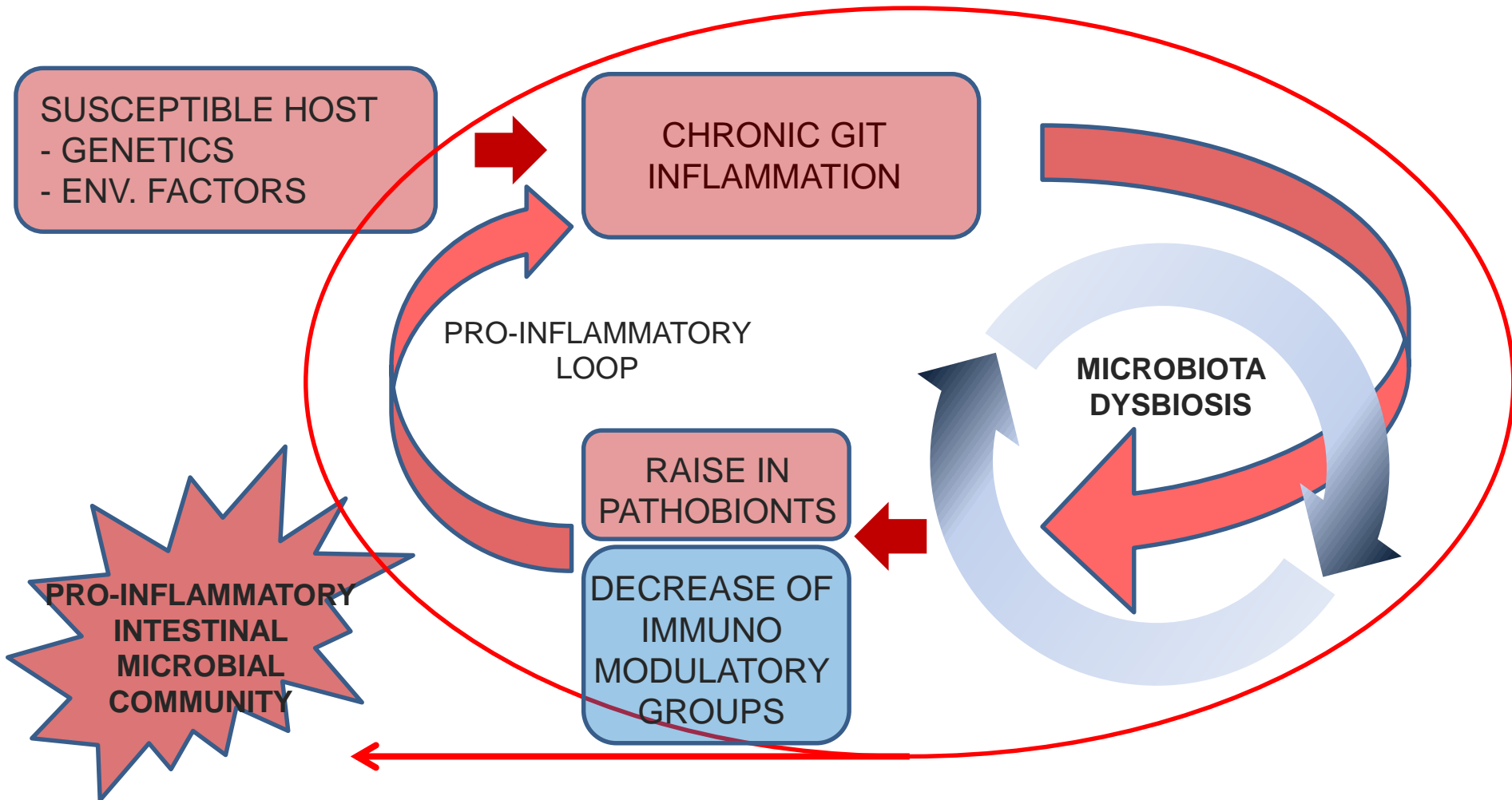
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IMMUNOLOGICAL DISREGULATION ASSOCIATED WITH MICROBIOTA DYSBIOSIS

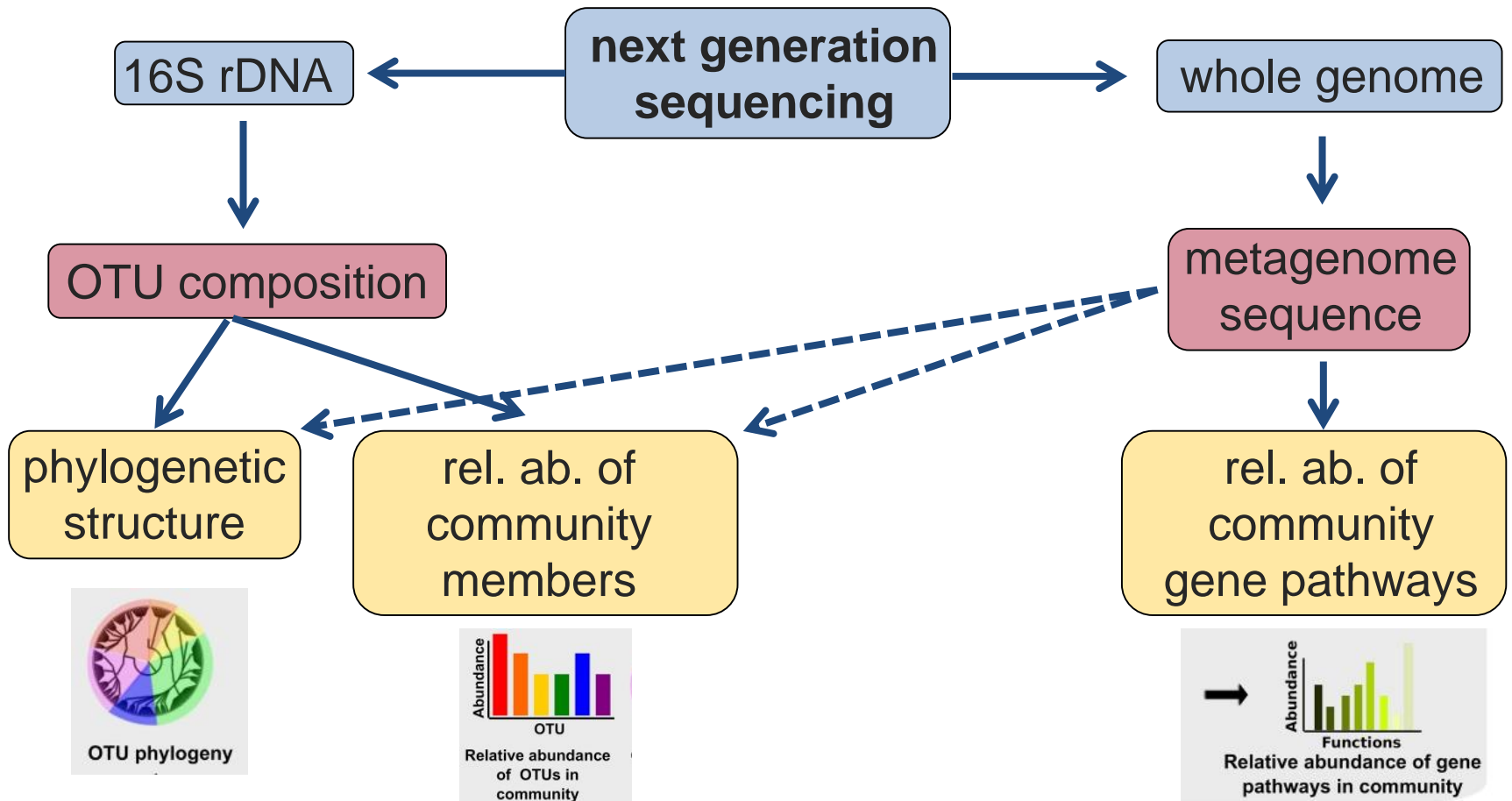
Immuno-modulatory bacteria (butyrate producers)



A non-controlled pro-inflammatory pathway can dramatically impact on the composition of the intestinal microbiota



MICROBIOTA MOLECULAR ASSESSMENT



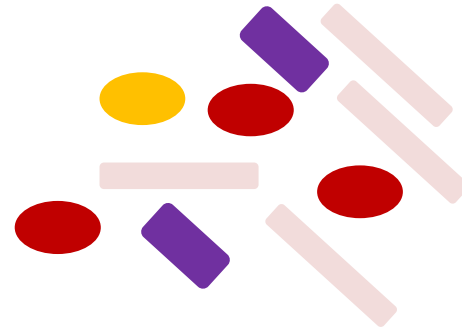
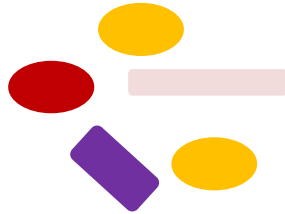
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GUT MICROBIOTA-ASSOCIATED DISORDERS

- **Nutrition related disorders** (obesity, type 2 diabetes, metabolic syndrome)
- **Inflammatory bowel diseases** (UC and CD)
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- **Neuro-developmental conditions** (autism, depression)
- **Arthritis**

7

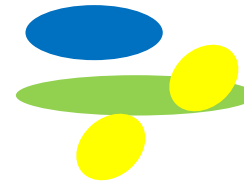
IDENTIFICATION OF MICROBIOTA SIGNATURES



Health
associated
typical profile



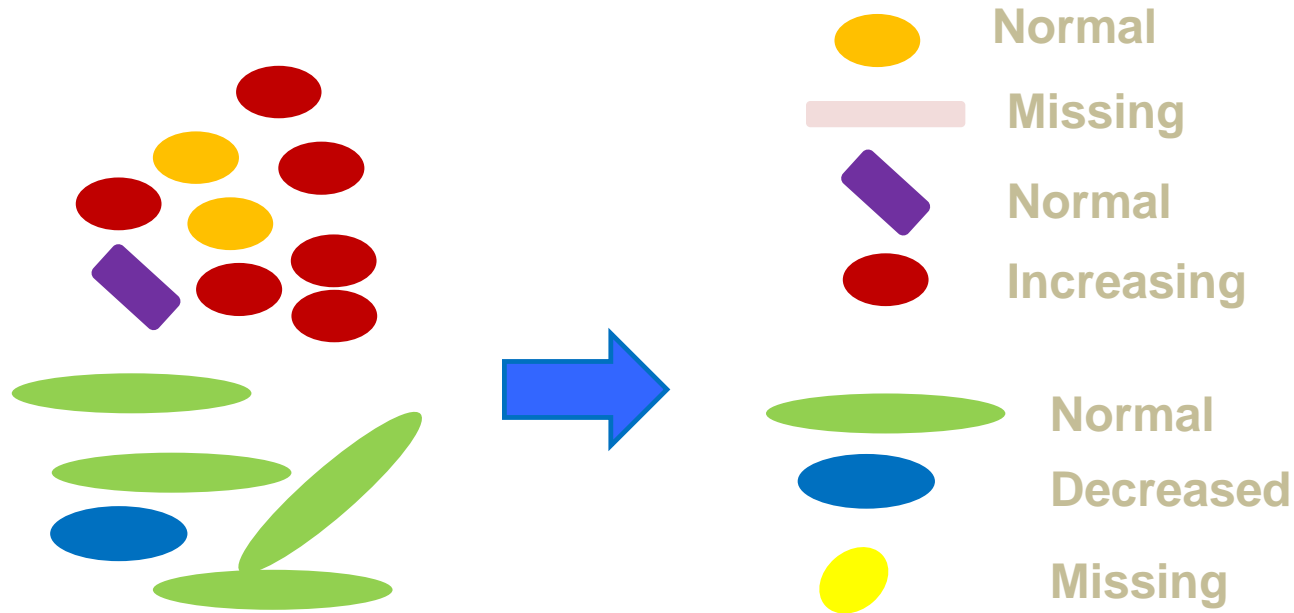
Disease
associated
typical profile



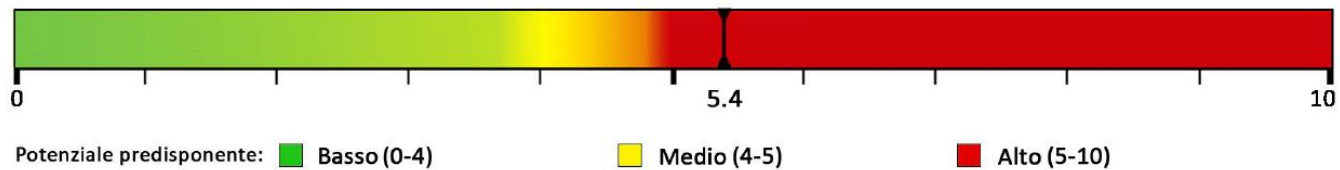
MATHEMATICAL (AND STATISTICAL) ELABORATION OF INDEXES TO CLASSIFY THE CONTRIBUTION OF A SPECIFIC PROFILE TO THE RISK OF DEVELOPING/CONSOLIDATING DYSBIOSIS

DYSBIOSIS FEATURES AND RISK

Patient profile

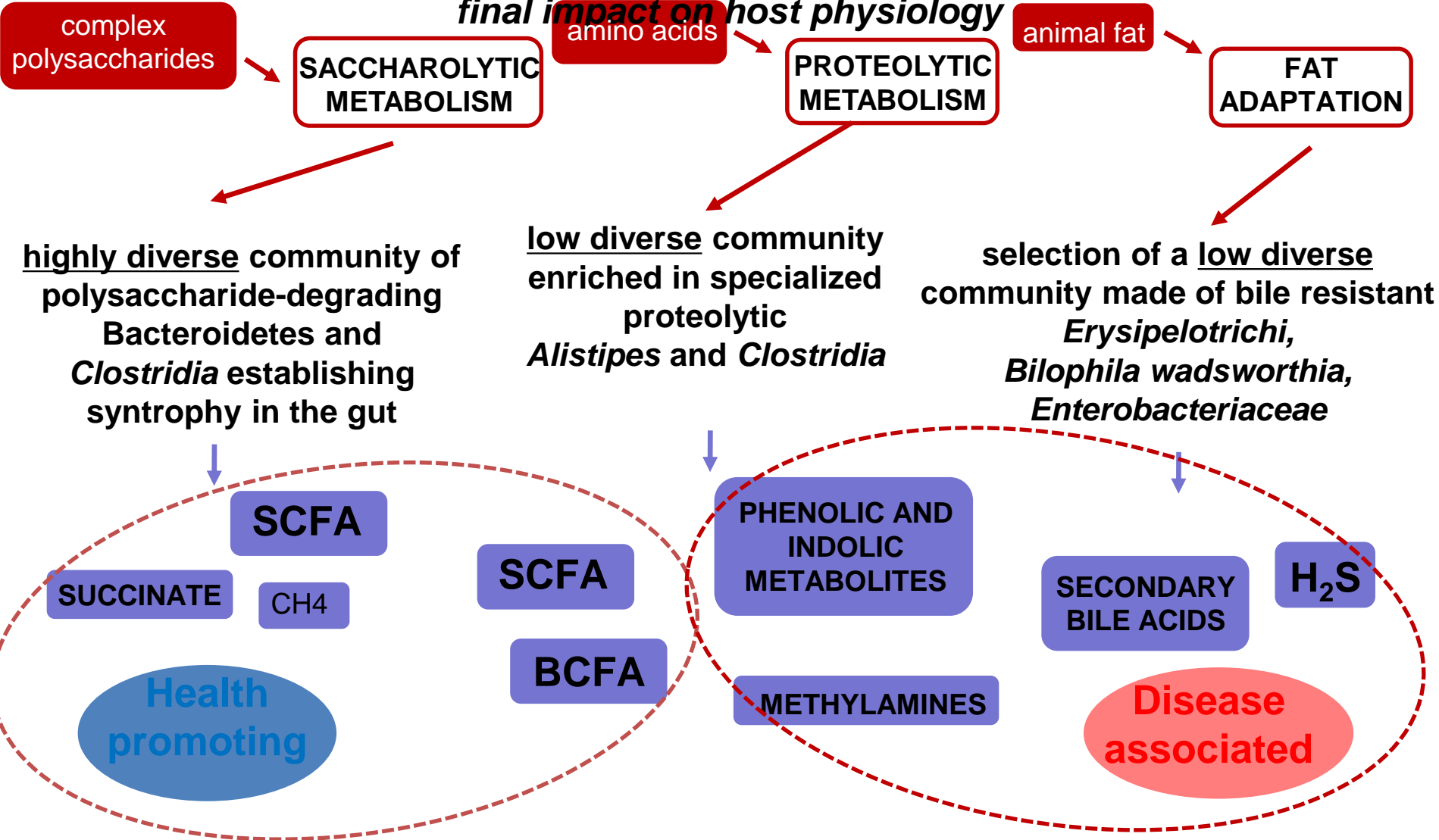


CALCULATION OF THE SPECIFIC INDEX CONTRIBUTION TO DYSBIOSIS



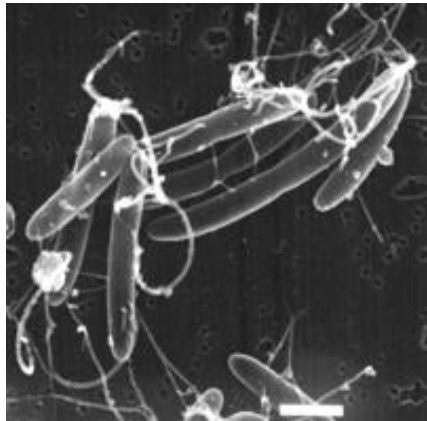
DIETARY INTERVENTION TO COUNTERACT DYSBIOSIS

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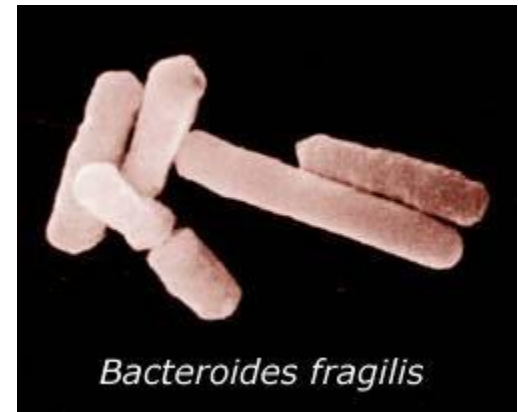
NEXT GENERATION PROBIOTICS

Faecalibacterium prausnitzii



Roseburia

Bacteroides



Bacteroides fragilis