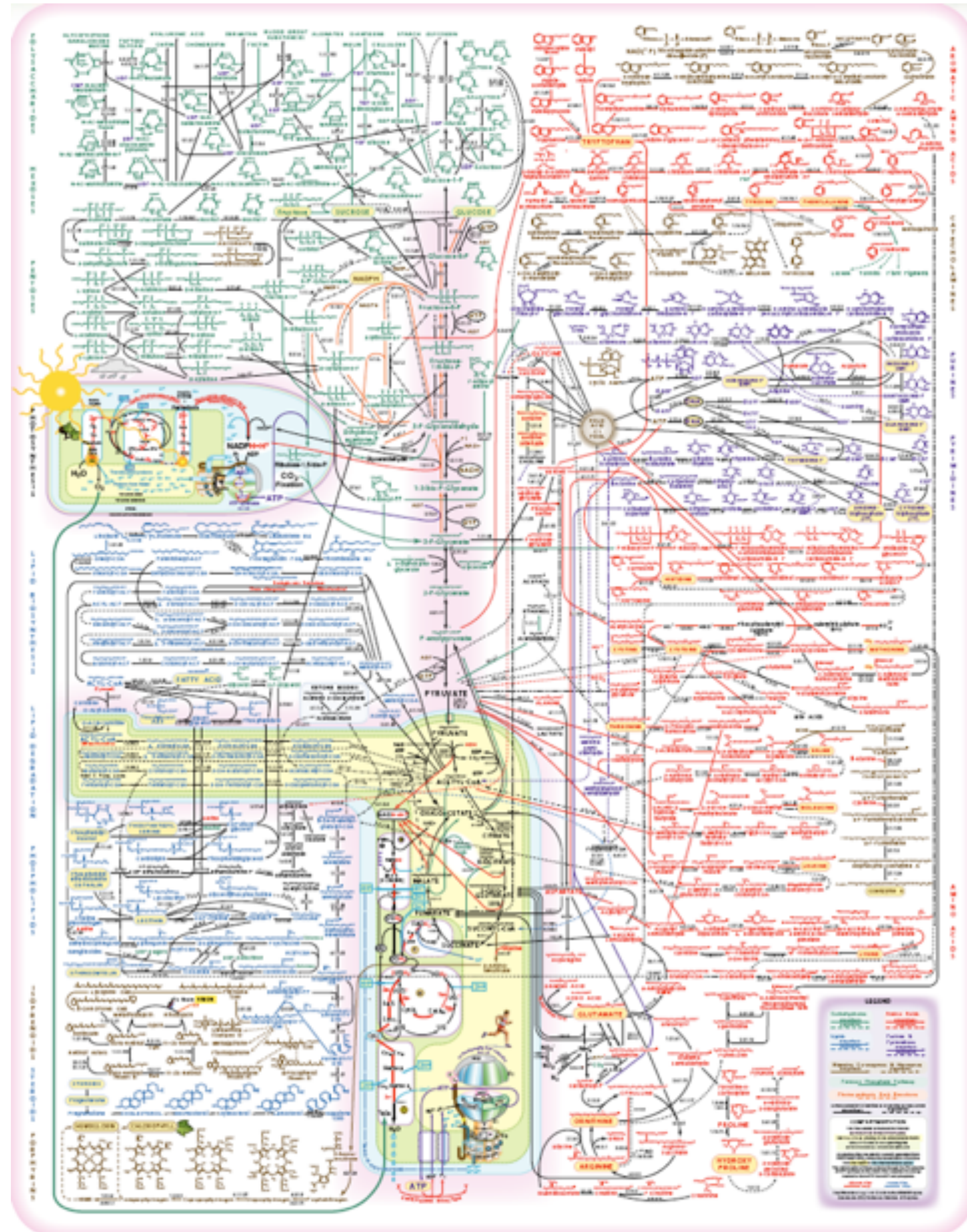
Ciências ULisboa

Faculdade de Ciências da Universidade de Lisboa



Mestrado
em
Microbiologia Aplicada

FRM

Fisiologia e Regulação Microbiana

IFRM

Introdução à Fisiologia e Regulação Microbiana

Ano Lectivo 2017/2018

1. **Introduction**
    Microbial physiology: problems and prospects
2. **Metabolic diversity. Metabolic processes of energy transduction**
    Diversity and adaptability: the energetic basis of microbial life
3. **Overview of central pathways of heterotrophic metabolism**
    Glycolysis, pentose phosphate pathway, Entner-Doudoroff pathway, glyoxylate cycle, the citric acid cycle and its modifications
4. **Physiology of growth in extreme environments**
    Adaptations to extreme conditions. Temperature, pH and osmotic homeostasis
5. **Membranes and solutes transport**
    Diversity of transport mechanisms
    A functional-phylogenetic classification system for transmembrane solute transporters
6. **Metabolic regulation**
    Transcription in Prokaryotes
    Enzyme induction
    Repression by catabolite
    Repression and attenuation by final metabolites
    Termination and anti-termination
7. **Physiological adaptation and homeostasis**
    General introduction to the systems of signal transduction
    Redox control and oxidative stress
    Hierarchical regulation of transport and utilization of carbon sources
    Regulation of gene expression associated with transition aerobic / anaerobic
    Photo-regulation in prokaryotes
8. **Nitrate reduction and nitrogen cycle in archaea**
9. **Methanogenic archaea, methane and carbon cycle**
10. **"Omics" technology and Systems Microbiology**

Bibliography

1. **D. White 2007. *The Physiology and Biochemistry of Prokaryotes* - 3rd ed.** Oxford University Press, Inc.
2. **B.H. Kim & G.M. Gadd 2008. *Bacterial Physiology and Metabolism*.** Cambridge University Press.
3. **L.L. Barton 2005. *Structural and Functional Relationships in Prokaryotes*.** Springer.
4. **Stephen Spiro & Ray Dixon 2010. *Sensory Mechanisms in Bacteria: Molecular Aspects of Signal Recognition***
5. **D.G. Nicholls & S.J. Ferguson 2002. *Bioenergetics3*.** Academic Press

✤ reviews and articles focused on specific topics in class

Evaluation

Two optional frequency (F1 and F2, each corresponding to 50% of final grade)
Final written examination (E) to be held at the time of exams
Two exam dates, according to the norms of FCUL

Final classification (CF): $CF = E$ or $CF = 0.5 \times F1 + 0.5 \times F2$
Approval: grade ≥ 10
Fail: note <10
The rounding is done directly to the units

Mestrado em Microbiologia Aplicada
Ano Letivo 2017/18 - 1º Semestre

Aulas Teóricas: Sala 2.4.16 [Edif. C2]

Aulas Teórico-Práticas/Práticas: Sala 2.2.15+ Sala 2.4.16 + Lab 2.4.39 [Edif. C2] + Lab Microbiologia-BioISI [Edif.TecLabs]

Semana	Data	SEG [4 h]		TER [4 h]	QUA [2 h]	QUI [3 h]	SEX [2 h]
1	18 set - 22 set	VMM (V+MM)	EAM T1	-	DVM T1	-	FRM T1
2	25 set - 29 set	VMM - V T1	EAM T2	LM1 - TP1 + P1 Técnicas Lab Microbiologia [AR, LC]	DVM T2	LM1 - P2 Técnicas Lab Microbiologia [AR, LC]	FRM T2
3	2 out - 6 out	VMM - V T2	EAM T3	LM1 - TP2 + P3 [FRM & DVM 4 h - AT, FA]	DVM T3	Feriado	FRM T3
4	9 out - 13 out	VMM - V T3	EAM T4	LM1 - TP3 + P4 [FRM & DVM 4 h - AT, FA]	DVM T4	LM1 - P5 [FRM & DVM 3 h - AT, FA]	FRM T4
5	16 out - 20 out	VMM - V T4	EAM T5	LM1 - TP4 + P6 [FRM & DVM 4 h - AT, FA]	DVM T5	LM1 - P7 [FRM & DVM 3 h - AT, FA]	FRM T5
6	23 out - 27 out	VMM - V T5	**EAM Teste 1**	LM1 - TP5 + P8 [FRM & DVM 4 h - AT, SC]	**DVM Teste 1**	-	**FRM Teste 1**
7	30 out - 3 nov	**VMM Teste 1**	EAM T6	LM1 - TP6 + P9 [VMM - V 4 h - FC, LC]	Feriado	LM1 - P10 [VMM - V 3 h - FC, LC]	FRM T6
8	6 nov - 10 nov	VMM - MM T6	EAM T7	LM1 - TP7 + P11 [VMM - V 4 h - FC, LC]	DVM T6	LM1 - P12 [VMM - V 3 h - FC, LC]	FRM T7
9	13 nov - 17 nov	VMM - MM T7	EAM T8	LM1 - TP8 + P13 [EAM 4 h - FD, AR]	DVM T7	LM1 - P14 [DVM & FRM 3 h - AR, LC]	FRM T8
10	20 nov - 24 nov	VMM - MM T8	EAM T9	LM1 - TP9 + P15 [VMM - MM 4 h - RSL, CV]	DVM T8	LM1 - P16 [DVM & FRM 3 h - AR, LC]	FRM T9
11	27 nov - 1 dez	VMM - MM T9	EAM T10	LM1 - TP10 + P17 [VMM - MM 4 h - RSL, CV]	DVM T9	LM1 - P18 [DVM & FRM 3 h - AR, LC]	Feriado
12	4 dez - 8 dez	VMM - MM T10	**EAM Teste 2**	LM1 - TP11 + P19 [VMM - MM 4 h - RSL, CV]	DVM T10	LM1 - P20 [DVM & FRM 3 h - AR, RT]	Feriado
13	11 dez - 15 dez	**VMM Teste 2**	-	LM1 - TP12 + P21 [VMM - MM 4 h - RSL, CV]	**DVM Teste 2**	**Revisões**	FRM T10
14	18 dez - 22 dez	-	**FRM Teste 2**	**Avaliação LM1**	-		

VMM: Virologia e Microbiologia Molecular [RSL]; EAM: Evolução e Adaptação Microbiana [FD]; DVM: Diversidade Microbiana [MB]; FRM: Fisiologia e Regulação Microbiana [AT]; LM1: Laboratório de Microbiologia I [AR]

Evaluation

F1: 27/10

F2: 18/12

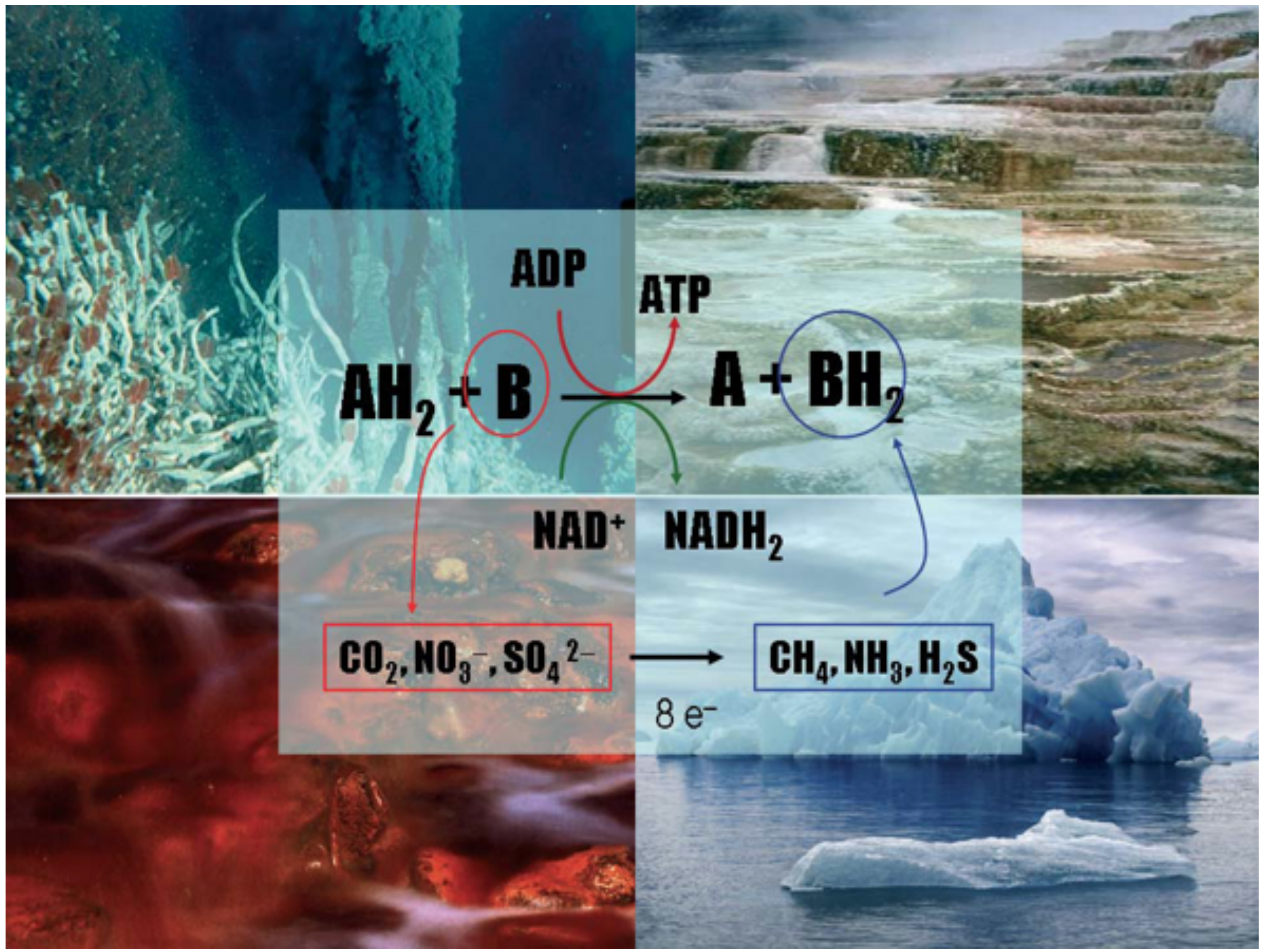
The Microbe's Contribution to Biology A. J. Kluyver and C. B. van Niel (Harvard University Press, 1956)

Table 1. *The Microbe's Contribution to Biology.*

Chapter	Main contributions described in the chapter
1. Microbial metabolism and the energetic basis of life	• Microbes open a new vision on the almost infinite capacity of life, for adapting itself to the immense variety of external conditions realized on Earth. • These tiny unicellular organisms cannot be considered as mere curiosities for life's terrestrial activities. • Many microorganisms are ubiquitous on Earth. • Various microbial types, surprisingly, have enormous diversity regarding their nutritional requirements.
2. Microbial metabolism; further evidence for life's unity	• The general equation of metabolism is: $AH_2 + B \rightarrow A + BH_2$ • This conversion will yield compounds with an "energy-rich" phosphate bond. • The continuous flow of hydrogen atoms appears to be essential for the maintenance of life. • Life has diverse ways of solving the problem for obtaining energy.
3. Phototrophic bacteria; key to the understanding of green-plant photosynthesis	• The absorbed radiant energy in photosynthesis permits the conversion of CO_2 into organic compounds. • O_2 produced during oxygenic photosynthesis must come entirely from H_2O. • Only one special pigment is involved in photosynthesis (chlorophyll *a*, in green plants; bacteriochloropyll *b* in purple bacteria, and bacteriochloropyll *e* in green sulfur bacteria. • Red sulfur bacteria are photosynthetic as well as chemosynthetic.

R. Guerrero and M. Berlang (2006). Life's unity and flexibility: the ecological link International Microbiology 9:225-235 Research Review

The Microbe's Contribution to Biology A. J. Kluyver and C. B. van Niel (Harvard University Press, 1956)



The Microbe's Contribution to Biology A. J. Kluyver and C. B. van Niel (Harvard University Press, 1956)

Table 1. *The Microbe's Contribution to Biology.*

Chapter	Main contributions described in the chapter
4. Life's flexibility; microbial adaptation	• The study of the microbe has made another important contribution to biology by casting a clear light on the flexibility of the living cell with regard to its metabolism. Microbial species are not characterized by specific and fixed metabolic patterns. • Changing the enzymatic equipment allows a cell to better fit its new environment. Environmental factors may induce (induced enzyme synthesis) the appearance of a new characteristic in a genetically constant cell.
5. Trial and error in living organisms; microbial mutations	• Studies carried out in the 1940s showed that mutations in bacteria are spontaneous, that bacteria can exchange genetic material, and that this material is DNA. These studies provided evidence for the unity of genetics in all living organisms. • A variation in properties may also result from sudden and random changes in genotype, with subsequent selection by environment of those modified microorganisms that can successfully compete with the original type. This illustrates the operation of the "trial-and-error" principle in the world of microbes.
6. Evolution as viewed by the microbiologist	• The same general concepts are also applicable to plants and animals. • The apparent "simplicity" of microbes suggests that they occupy a position somewhere near the base of the evolutionary scale, but the problem of the manner in which microorganisms came into being is still unresolved. • Life is a special property of matter at a certain stage of complexity.

R. Guerrero and M. Berlang (2006). Life's unity and flexibility: the ecological link International Microbiology 9:225-235 Research Review

Think like a bacterium

"We must, albeit reluctantly, dismiss the naive but comforting myth of the bacterial cell as a simple loner: a self-contained bag of uncompartmentalized enzymes, so exquisitely uncomplicated as to be understood outright and considered passé in this new millennium — a biological beaker for examining the more complicated eukaryotic versions of life."

"Bacteria cooperate in both uniform and mixed societies, sabotage the relatively mammoth creatures that they colonize, modify their own living spaces, and anticipate predictable changes in their surroundings. They achieve these feats with a clear sense of time and place, both within and outside their small but well organized bodies, and send and receive messages across comparatively vast space."

"To fully understand them, perhaps we should think of 'bacterial culture' not in the sense of something in a flask, but rather as a dynamic and cosmopolitan life style shared by creatures of the microbial world, into which we have been allowed to peer."

Meeting Report - Think like a bacterium - Conference on Bacterial Neural Networks
EMBO (European Molecular Biology Organization) Vol 4 | Nº1 | 2003

Horizon 2020 at a glance

FUNDING WILL BE FOCUSSED ON THE FOLLOWING CHALLENGES:

- Health, demographic change and wellbeing;
- Food security, sustainable agriculture, marine and maritime research, and the bio-economy;
- Secure, clean and efficient energy;
- Smart, green and integrated transport;
- Climate action, resource efficiency and raw materials;
- Inclusive, innovative and secure societies.

Horizon 2020 at a glance

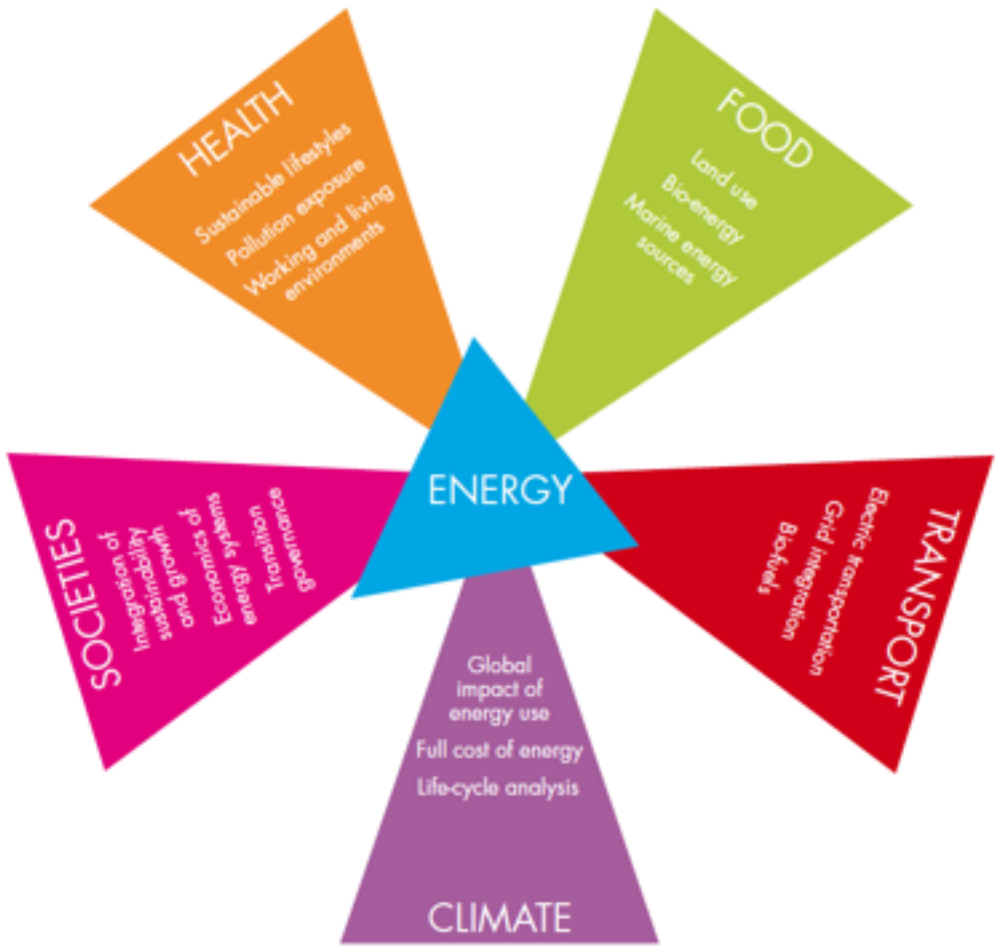
FOOD SECURITY, SUSTAINABLE AGRICULTURE, MARINE AND MARITIME RESEARCH AND THE BIO-ECONOMY

The complexity of the challenges related to food, feed and biomass production and the associated multi-directional value chain from primary production to the needs and opportunities from climate change and its mitigation, available resources, environment, new biomaterials, energy, food, feed, health, transport, and new bio-materials perspectives calls for an interactive and multidisciplinary cross-cutting research and innovation approach that embraces the other five defined societal challenges. It is critical that research and innovation in this area are addressed in a fully integrated manner,

Horizon 2020 at a glance

SECURE, CLEAN AND EFFICIENT ENERGY

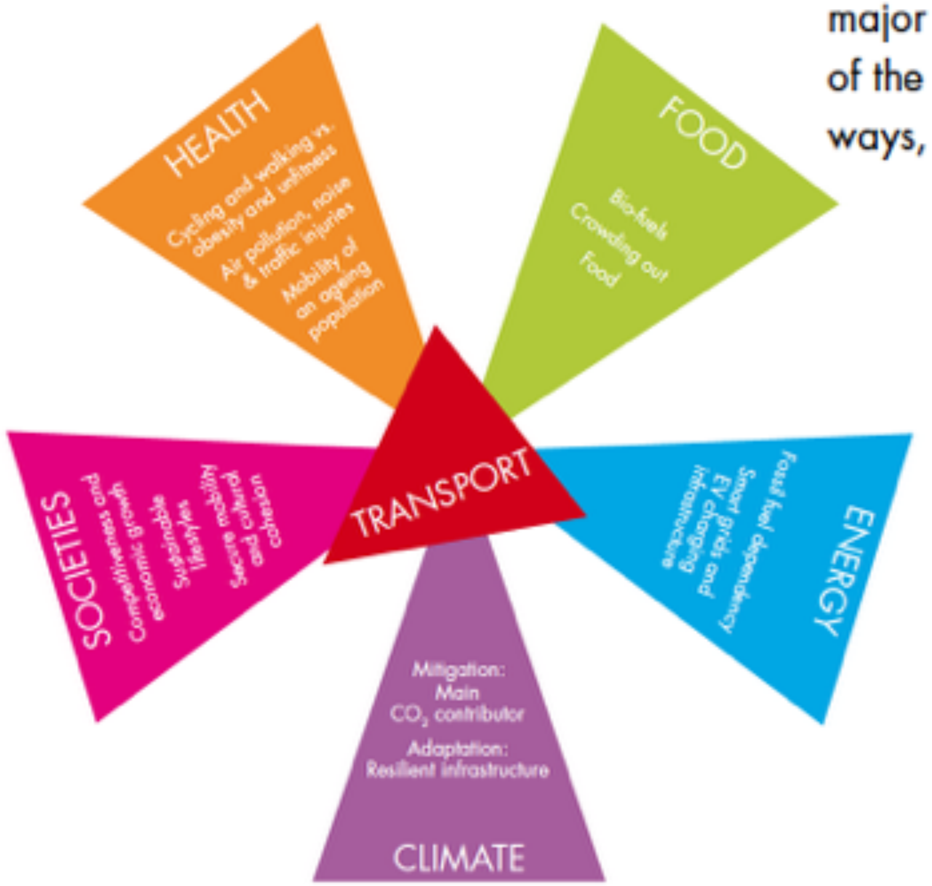
The transition towards carbon-free energy solutions calls for a more integrated approach combining technological, economic, social and cultural aspects, and for better cooperation between policy and research. Sustainable energy solutions need two types of approaches: Highly innovative technology research plus a new approach to systemic research and innovation challenges.



Horizon 2020 at a glance

SMART, GREEN AND INTEGRATED TRANSPORT

Passenger and freight transport is derived from the spatial dispersion of people's activities and production of goods and as such an essential and integral element in our lifestyle and consumption pattern. Transport research therefore should be (and have been) using multidisciplinary approaches ranging from several branches of engineering to various disciplines in social sciences, such as economics, sociology, psychology, geography and political science. The major transport (sub-)challenges also relates closely to the themes of the five other societal challenges, and the relationship goes both ways, clearly calling for cross-cutting research cooperation:



Horizon 2020 at a glance

CLIMATE ACTION, RESOURCE EFFICIENCY AND RAW MATERIALS

Climate-related research, climate action and increasing resource-use efficiency are cross-cutting challenges which cannot be considered on their own but need to be addressed as a part of the overall challenge of developing a sustainable path for society. They must, therefore, be embedded in the remaining challenges, and cannot be carried out independently from, for example, the energy challenge, food security or the development of inclusive and equal societies. Ideally, all six challenges should be incorporated into the overarching challenge of achieving sustainable development. With

This Position Paper, developed by the Marine Board Working Group on Marine Biotechnology, presents a shared vision for European Marine Biotechnology whereby:

By 2020, an organised, integrated and globally competitive European Marine Biotechnology sector will apply, in a sustainable and ethical manner, advanced tools to provide a significant contribution towards addressing key societal challenges in the areas of food and energy security, development of novel drugs and treatments for human and animal health, industrial materials and processes and the sustainable use and management of the seas and oceans.

This 2020 Vision for European Marine Biotechnology will only be achieved through a coordinated implementation in a joint effort with active support and involvement from all relevant stakeholders, of the following high level recommendations:

- RECOMMENDATION 1: Create a strong identity and communication strategy to raise the profile and awareness of European Marine Biotechnology research.
- RECOMMENDATION 2: Stimulate the development of research strategies and programmes for Marine Biotechnology research and align these at the national, regional and pan-European level.
- RECOMMENDATION 3: Significantly improve technology transfer pathways, strengthen the basis for proactive, mutually beneficial interaction and collaboration between academic research and industry and secure access and fair and equitable benefit sharing of marine genetic resources.
- RECOMMENDATION 4: Improve training and education to support Marine Biotechnology in Europe.

Executive Summary Box A **Marine Biotechnology research priorities to address key societal challenges**	
Target research area for development	**Research priorities and objectives**
Food: Development of food products and ingredients of marine origin (algae, invertebrates, fish) with optimal nutritional properties for human health	- Develop innovative methods based on -omics and systems biology for selective breeding of aquaculture species; - Develop biotechnological applications and methods to increase sustainability of aquaculture production, including alternative preventive and therapeutic measures to enhance environmental welfare, sustainable production technologies for feed supply, and zero-waste recirculation systems; - Integration of new, low environmental impact feed ingredients to improve quality of products and human health benefits.
Energy: Development and demonstration of viable renewable energy products and processes, notably through the use of marine algae	- Produce an inventory of microalgae resources for biofuel production to support optimisation of the most appropriate strains; - Improve knowledge of basic biological functions, tools for steering the metabolism and cultivation methods of marine microalgae to improve the photosynthetic efficiency, enhance lipid productivity and obtain microalgae with optimum characteristics for mass cultivation (mixed & mono cultures), biofuel production and biorefinery; - Develop efficient harvest, separation and purification processes for micro- and macroalgae.
Health: Development of novel drugs, treatments and health and personal care products	- Increase the focus on the basic research (taxonomy, systematics, physiology, molecular genetics and chemical ecology) of marine species and organisms from unusual and extreme environments to increase chances of success in finding novel bioactives; - Improve the technical aspects of the biodiscovery pipeline, including the separation of bioactives, bio-assays that can accommodate diverse material from marine sources, dereplication strategies and structure determination methods and software; - Overcome the supply problem to provide a sustainable source of novel pharmaceutical and healthcare products through scientific advances in the fields of aquaculture, microbial and tissue culture, chemical synthesis and biosynthetic engineering.

Executive Summary Box A Marine Biotechnology research priorities to address key societal challenges	
Target research area for development	**Research priorities and objectives**
Environment: Development of biotechnological approaches, mechanisms and applications to address key environmental issues	- Develop automated high-resolution biosensing technologies allowing *in situ* marine environmental monitoring to address coastal water quality, including prediction and detection of Harmful Algal Blooms and human health hazards; - Develop cost-effective and non-toxic antifouling technologies combining novel antifouling compounds and surface engineering; - Consolidate knowledge on DNA-based technologies for organism and population identification and support the development of commercial tools and platforms for routine analysis.
Industrial Products and Processes: Development of marine-derived molecules exploitable by industry including enzymes, biopolymers and biomaterials	- Develop enabling technologies for high throughput enzyme screening and for the expression of marine proteins and enzymes through dedicated hosts; - Produce marine biopolymers as novel competitive commercial products in food, cosmetics and health.

People are not just people. They are an awful lot of microbes, too

4 9 2 | N AT U R E | VO L 4 6 8 | 2 5 N O V E MBE R 2 0 1 0

Jillian Banfield - microbial ecologist

Michael Morowitz - neonatal surgeon

David Relman - medical researcher

Morowitz takes faeces samples almost daily from birth, isolates DNA, sends it to a high throughput sequencing centre, then passes the data and clinical information to **Banfield** and **Relman**. Relman analyses the 16S sequences at every time point to get a census of the species and their abundance; Banfield then selects a few time points for more extensive sequencing and genome analysis to identify the species, strains and genes present.

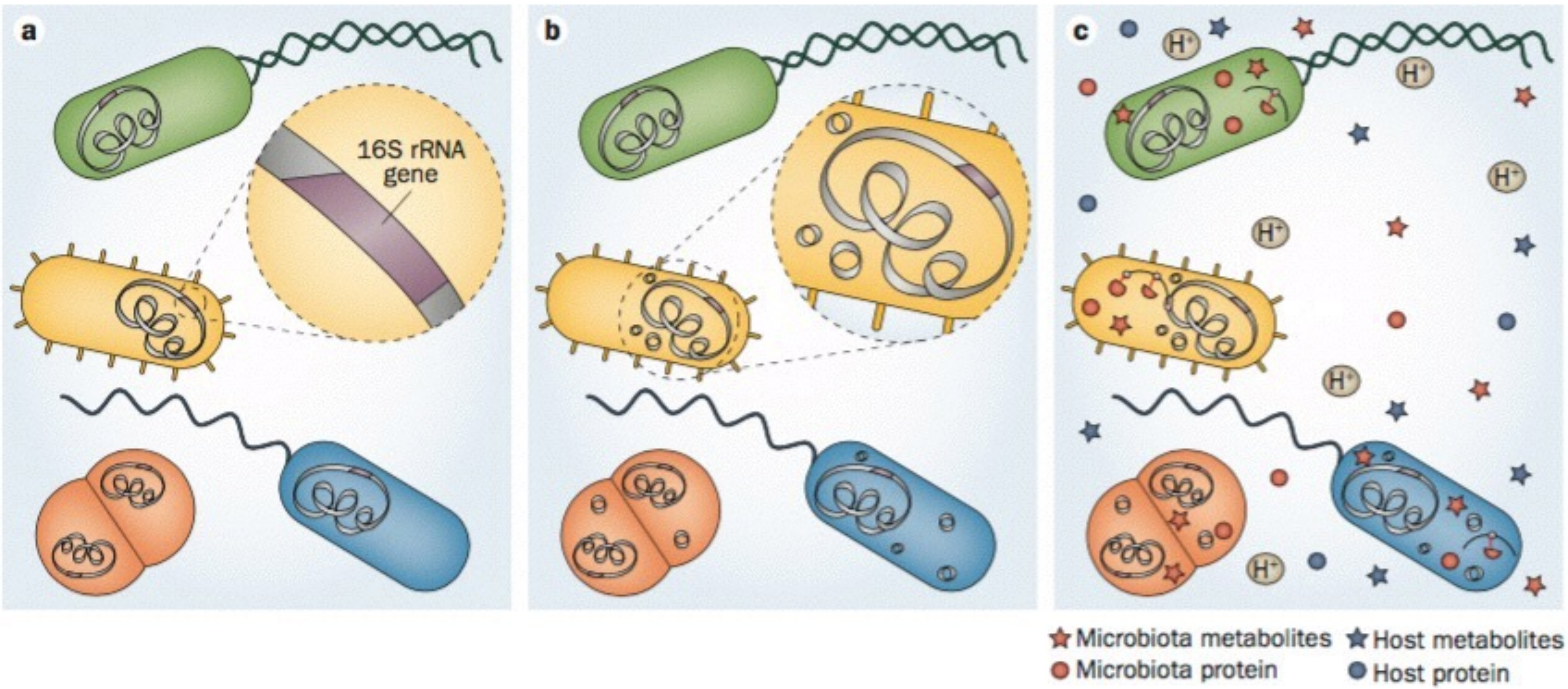


Figure 1 | Definition of the microbiota, metagenome and microbiome as used in this Review. Each image represents the same population; however, different approaches to define the population provide different information. **a** | Microbiota: 16S rRNA surveys are used to taxonomically identify the microorganisms in the environment. **b** | Metagenome: the genes and genomes of the microbiota, including plasmids, highlighting the genetic potential of the population. **c** | Microbiome: the genes and genomes of the microbiota, as well as the products of the microbiota and the host environment. Abbreviation: rRNA, ribosomal RNA.

EXPLORING THE SUPERORGANISM

Big questions about the microbial multitudes inside.

How stable is the microbial community?
After populations of intestinal microbes — the microbiota — are established, it's unclear how they change with age, shifts in diet, activity level, co-habitation or after a blast of antibiotics. One study[7] revealed that two courses of the antibiotic ciprofloxacin wreaked havoc in the gut microbes of healthy people, and that the communities never fully recovered.

Can the microbiota be used for diagnosis?
Analysis of an individual's microbes and their metabolites may reveal those directly associated with disease and those that serve as biomarkers for a wider spectrum of conditions. It may prove possible to tailor treatments to the microbiota.

Can the microbiota be changed?
Studies show a tight relationship between diet and microbiota, so it may be possible to fine-tune diet to support the most beneficial community. Prebiotics — ingesting particular foods high in fibre or vitamins that promote the growth of specific microbes — are now receiving attention from the food industry. Probiotics — microbes in food or a pill — are already common, although evidence that they have any benefits is so far equivocal.

A more radical approach is faecal transplants, and there are anecdotal accounts of such a procedure, used as a last resort, benefiting recipients whose microbial community was highly disturbed. But microbes that are innocuous in one individual could be pathogenic in another.

***Helicobacter pylori*: just one of the residents of the human gastrointestinal tract.**

Rather than relying on transplants of foreign faeces, "It might be wiser to bank your poop, like you do your bone marrow," says David Relman of Stanford University in California, to repopulate the intestine in the event of disease. "But you can't propagate faecal matter, so you'd be stuck with the dose that was originally frozen."

Does the microbiota affect behaviour?
A number of conditions that affect behaviour, such as autism and schizophrenia, have been associated with digestive problems, and symptoms are often reported to be connected to diet. Microbes produce a range of compounds that can potentially affect brain activity. Some studies have suggested that the structure of microbial communities in children with autism differ from those in children without the condiiton[8]. No causal relationship has been demonstrated, but some researchers think that analysis of the microbial community may allow clinicians to diagnose these conditions and begin therapy before the symptoms even start.

How does modern life affect the microbiota?
Microbes that humans come into contact with and support might be changing dramatically, particularly in wealthier, developed countries, thanks to lavish use of antibiotics, high hygiene standards, more Caesarean sections, less breastfeeding and processed-food diets. One version of the 'hygiene hypothesis' proposes that the increasingly sterile environment in which humans are raised may alter the microbial community in the body, preventing normal development of the immune system and potentially leading to increasing prevalence of conditions such as allergies, asthma, Crohn's disease and even autism. Some now call it the 'microbiota hypothesis'. On the other hand, it may become possible to adjust microbes to help deal with modern environmental toxins.

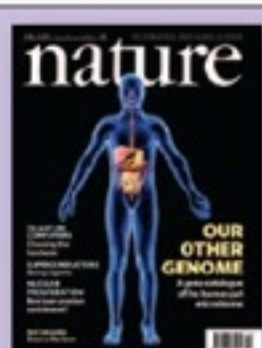
Can the microbiota be used in forensics?
It might be possible to learn about and track criminals by analysing their microbiota — collected from body fluids or fingerprints. This might reveal what a suspect eats, where they live, how active they are and whether they have certain pets. There are numerous caveats, though, including how much the microbial community changes with time, or after someone else touches the same spot. L.B.

NEWS FEATURE

A snapshot of the microbiome field

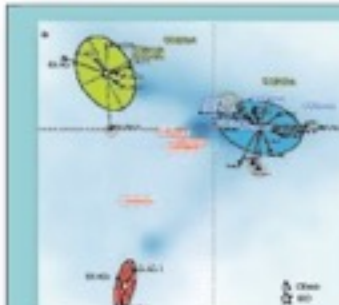
A survey of leading scientists from academia and industry highlights notable papers that have garnered momentum in microbiome research.

Ranked 1st



Qin, J. *et al.* A human gut microbial gene catalogue established by metagenomic sequencing. *Nature* **464**, 59–65 (2010).

"The first gene catalog of the human microbiome, demonstrating that the [gut microbiome] metagenome contains 150-fold more genes than our own genome." **Frederik Bäckhed**, *University of Gothenburg, Sweden.*



Arumugam, M *et al.* Enterotypes of the human gut microbiome. *Nature* **473**, 174–180 (2011).

Ranked 2nd

"Stirred up discussions but indicates high-level clustering that will be useful to provide further segmentation." **Willem de Vos**, *Wageningen University, The Netherlands.*

NEWS FEATURE

A snapshot of the microbiome field

A survey of leading scientists from academia and industry highlights notable papers that have garnered momentum in microbiome research.

Ranked 3rd

Turnbaugh, P.J. *et al.* An obesity-associated gut microbiome with increased capacity for energy harvest. *Nature* **444**, 1027–1031 (2006).

"Demonstrated in rodents that an obese phenotype could be transferred or reversed by fecal transplant of the microbiota." **Peter DiStefano**, *Second Genome, San Bruno, California, USA.*

"The authors showed that the microbiome could actively shape disease susceptibility. All the ingredients of microbiome research (germ-free mice, next-generation sequencing and fecal transplantation) are included in this paper. The study has trail-blazed this field of research and provided a template for investigation of the impact of the microbiome on diseases and health such as cancer, diabetes, behavior and colitis." **Christian Jobin**, *University of North Carolina at Chapel Hill, USA.*

VOLUME 31 NUMBER 4 APRIL 2013 NATURE BIOTECHNOLOGY

Ranked 4th

Yatsunenko, T. *et al.* Human gut microbiome viewed across age and geography. *Nature* **486**, 222–227 (2012).

"Demonstrates differences in microbiomes between the US, South America and Africa. Shows that microbiota of many human populations still need to be studied." ***Erwin Zoetendahl****, Wageningen University, The Netherlands.*

Ranked 5th

Eckburg, P.B. *et al.* Diversity of the human intestinal microbial flora. *Science* **308**, 1635–1638 (2005).

Huttenhower, C. *et al.* Structure, function and diversity of the healthy human microbiome. *Nature* **486**, 207–214 (2012).

"Its impact stemmed from integration of emerging insights and approaches [in environmental microbiology and bacterial diversity, concepts in ecology] and long-standing interest among clinicians and clinical microbiologists in the possible roles of the human microbiome in health and disease. This near–full-length 16S-rRNA gene–based survey of the healthy human distal gut was the most extensive sequence-based survey of any environment at that time (to my knowledge)." ***David Relman****, Stanford University and the Veterans Affairs Palo Alto Health Care System, Palo Alto, California, USA.*

"Crucial, as (together with other papers), it laid the foundations for mapping the phylogenetic and genetic makeup of the human microbiome." ***Jeroen Raes****, Flanders Institute for Biotechnology, Belgium.*

Published online: August 6, 2015

Science & Society

EMBO reports

Better tools, new problems

New technologies help to advance research in the life sciences, but the quantities of data generated are proving hard to manage and interpret

Philip Hunter

> *"As the sequencing of whole genomes becomes faster and cheaper, data production is running ahead of the ability to make sense of it..."*

> *"The problems with data accuracy and data analysis are particularly relevant for the application of NGS in the clinic..."*