Short and long term road map for the development of a robust mechanistic and dynamic model of the MELiSSA C1 compartment based on microbial community characterization

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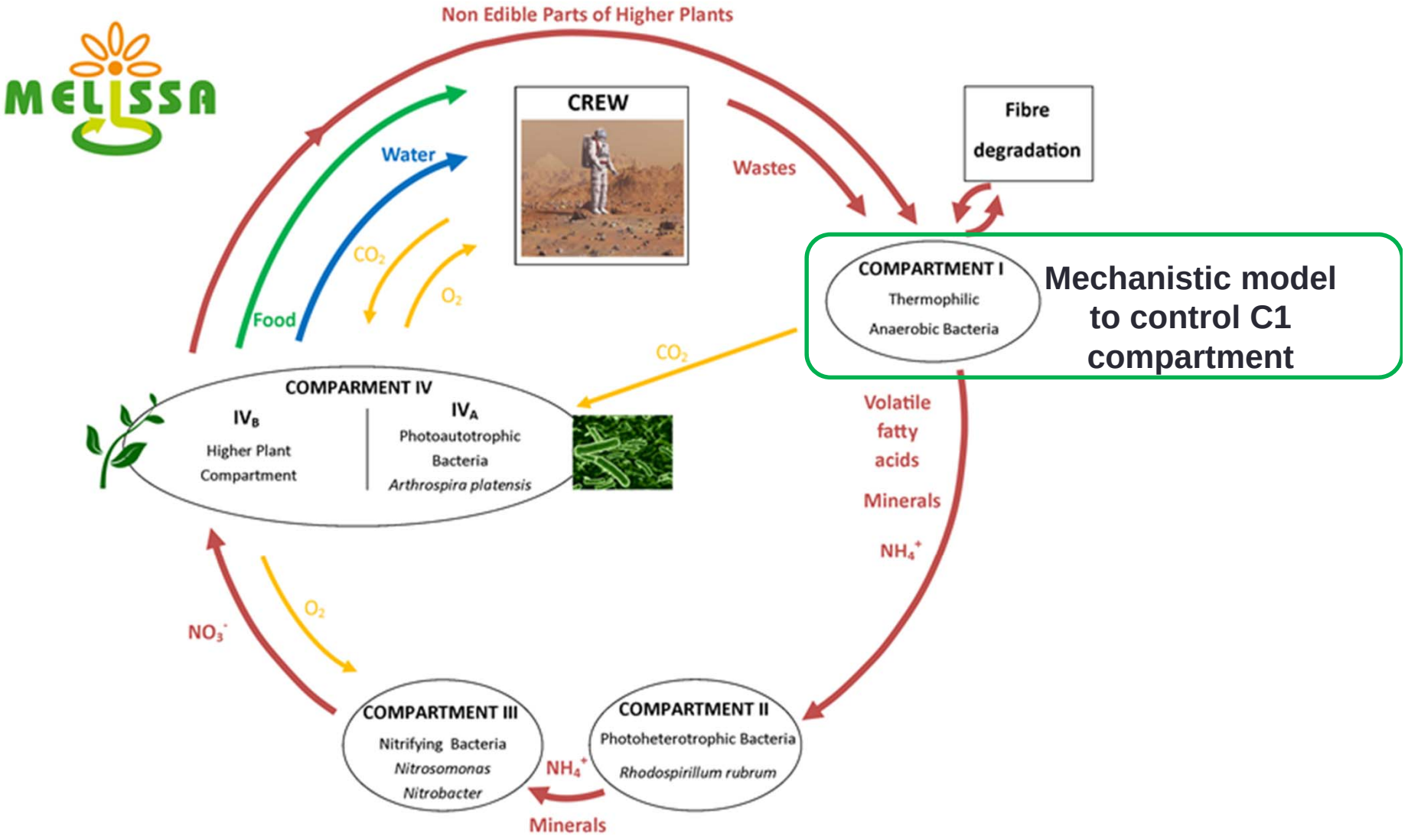
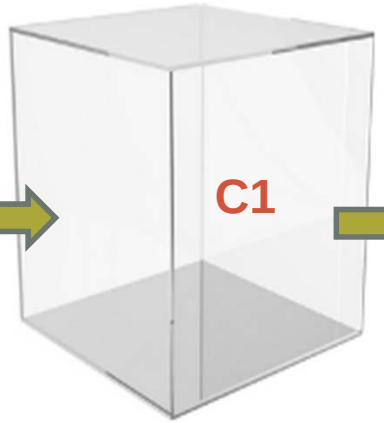
KU LEUVEN

MELiSSA workshop. Lausanne.
June 8th 2016

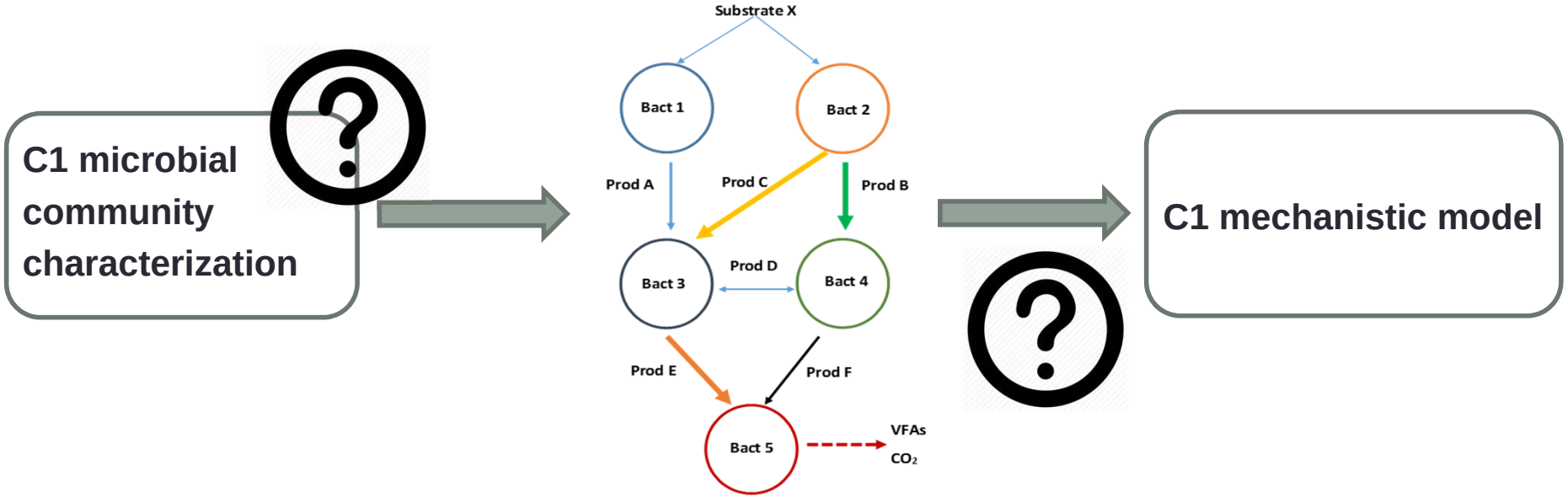

Non Edible Parts of Higher Plants
CREW
Fibre degradation
Wastes
Water
Food
CO_2
O_2
COMPARTMENT I
Thermophilic Anaerobic Bacteria
Mechanistic model to control C1 compartment
CO_2
COMPARMENT IV
IV_B
Higher Plant Compartment
IV_A
Photoautotrophic Bacteria
Arthrospira platensis
Volatile fatty acids
Minerals
NH_4^+
NO_3^-
O_2
COMPARTMENT III
Nitrifying Bacteria
Nitrosomonas
Nitrobacter
COMPARTMENT II
Photoheterotrophic Bacteria
Rhodospirillum rubrum
NH_4^+
Minerals

TOMORROW

MELiSSA feed → 

→ CO_2, VFA, NH_4^+ → **C1 mechanistic model**

- Complete microbial information
  - Taxonomy
  - Function
  - Metabolome
- Reaction rates/kinetics
- Interactions



Interactive metabolic model of C1 community

- **Deep Knowledge / understanding of C1 microbial community**
- **Correct experimental design / approach**
- **Which tools/techniques to use**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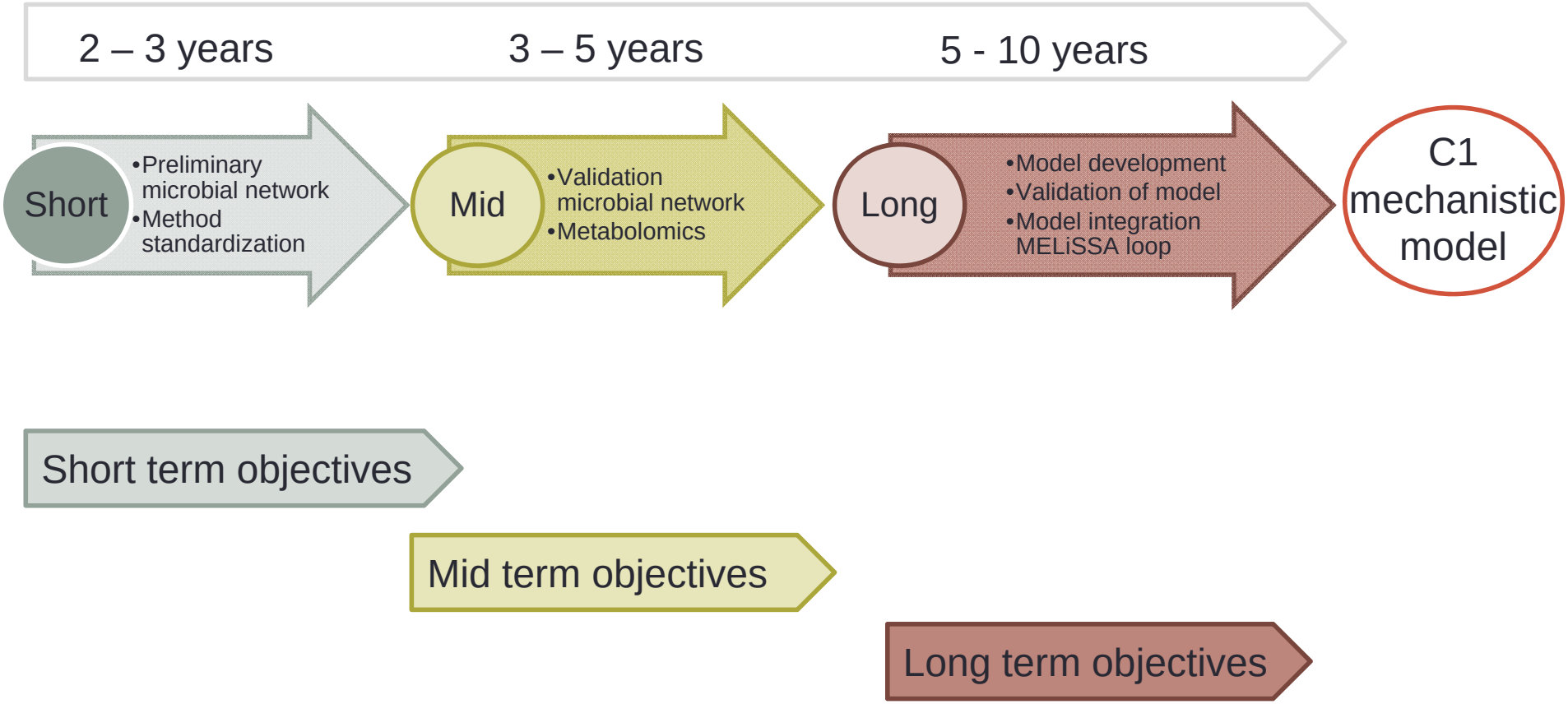
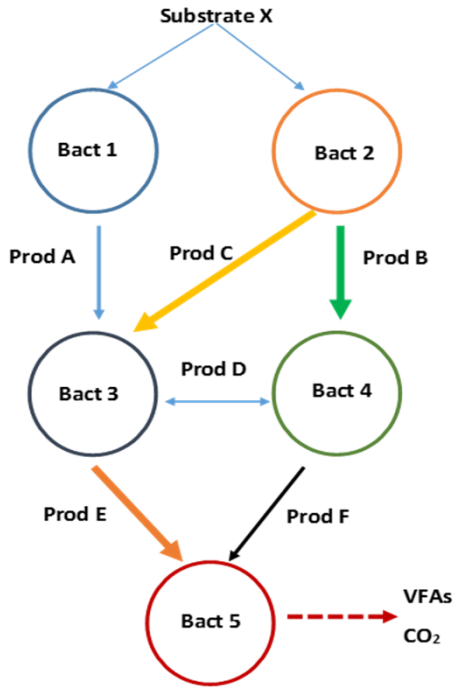
Road map!


2 – 3 years
3 – 5 years
5 - 10 years
Short
•Preliminary microbial network
•Method standardization
Mid
•Validation microbial network
•Metabolomics
Long
•Model development
•Validation of model
•Model integration MELiSSA loop
C1 mechanistic model
Short term objectives
Mid term objectives
Long term objectives

WHAT?

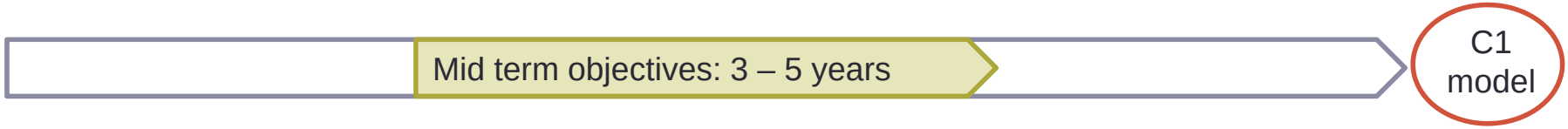
- Preliminary microbial network and microbial interactions based on a stable and well-functioning microbial community.
- Identification of crucial bacterial species and biomolecular makers involved in correct functioning of C1.

Interactive metabolic model of C1 community Essential functions/organisms



HOW?

- Standardization of analysis methods for omics:
  - DNA, RNA, protein extraction
  - Bioinformatic analysis
- Time point study
- Meta-omics (DNA; RNA; Proteins)
  - Determination of what is “stable/constant” (in time) community composition.
    - 16S targeted amplicon
    - Metagenomics
  - Determination of what is “stable/constant” (in time) functionality of microbial community.
    - Metatranscriptomics
    - Metaproteomics



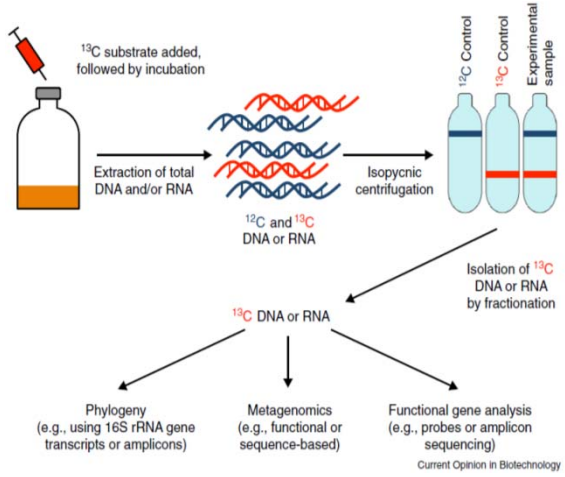
WHAT?

- Validation of microbial network proposed in short term phase.
- Study the crucial bacterial species/functions:
  - Individual species/functions
  - Selected microbial groups/functions
- Relevant perturbations to the C1 system to be able to identify differences in:
  - variations in active microbial community composition.
  - variations in the functions of the active microbial community.
- Begin with
  - Metabolomic analysis of C1 microbial community.
  - Flux analysis of selected metabolites

Network validation - HOW?

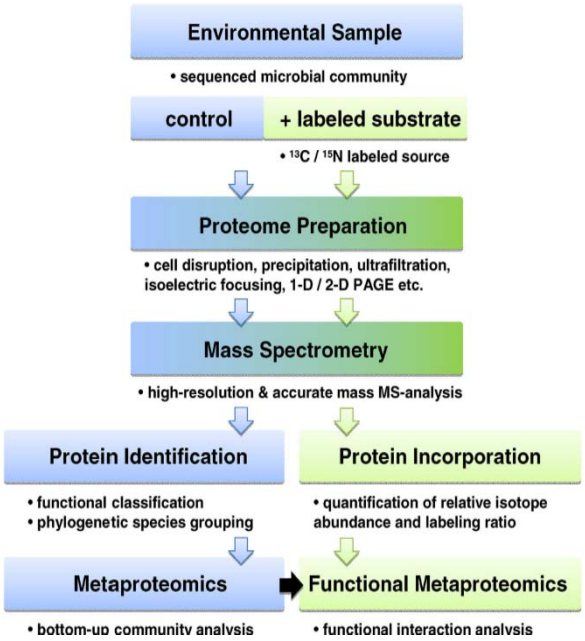
- SIP with different and relevant substrates to validate the microbial network.
- Identify/validate the crucial species/functions.

DNA/RNA - SIP



Coyotzi *et al.,* (2016), *Curr. Opin. Biotechnol.* 41, 1–8

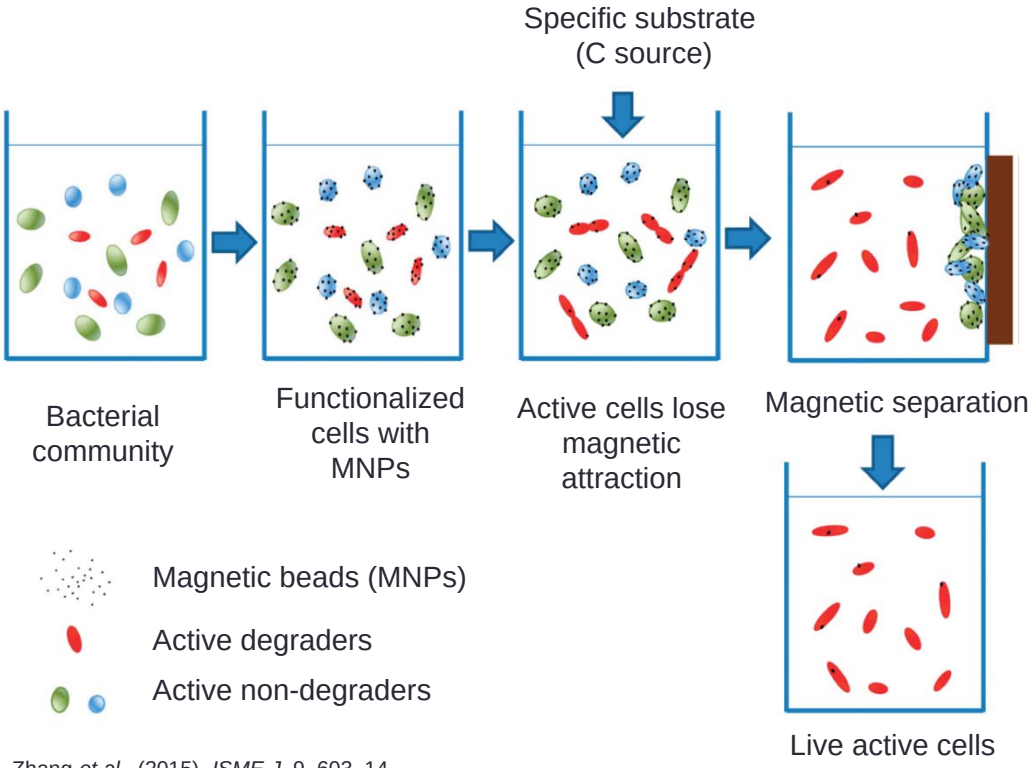
Protein - SIP



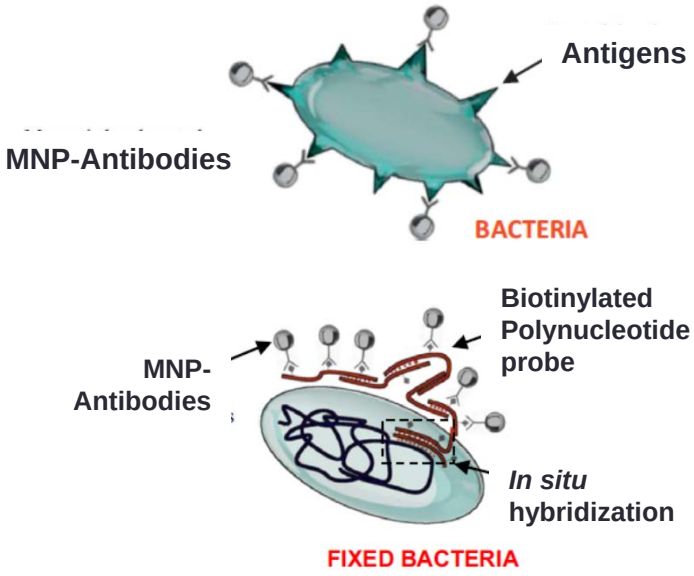
Seifert et *al.,* (2012), *Mass Spectrom. Rev.*, 683–697

Single cells / specific populations/functions - HOW?

- Separation by magnetic nanoparticles and similar.



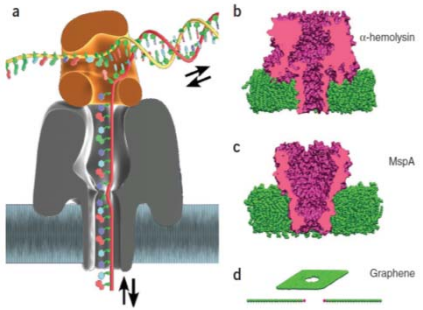
Zhang *et al.*, (2015). *ISME J.* 9, 603–14



Pivetal *et al.*, (2015). *J. Magn. Magn. Mater.* 380, 72–77.

Single cells - HOW?

- Sequencing of:
  - Single cell of relevant bacteria.
  - Metagenomic selected bacterial populations.
- New sequencing technologies:



Schneider & Dekker (2012). *Nat. Biotechnol.* 30, 326–328.

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Long term objectives; 5 – 10 years

C1 model

WHAT?

- Kinetics of selected bacteria/functions.
- Kinetics of entire C1 microbial community.
- Metabolic network of the C1 microbial community.

- Integrate information for model development.

- Development technologies/assays to follow, monitor and quantify the crucial selected species, functions or biomolecular markers.

- Validation of final proposed mechanistic model.
- Integration of C1 model in entire MELiSSA loop.

Long term objectives; 5 – 10 years

C1 model

HOW?

- Kinetics/ metabolomics of:
  - Selected bacterial populations/functions
  - Single cell
- Model validation with:
  - C1 perturbations/changes.
  - MELiSSA loop perturbations/changes.

- “Miniaturized” single cell omics
- Real-time single cell/community omics
- Real-time metabolomics

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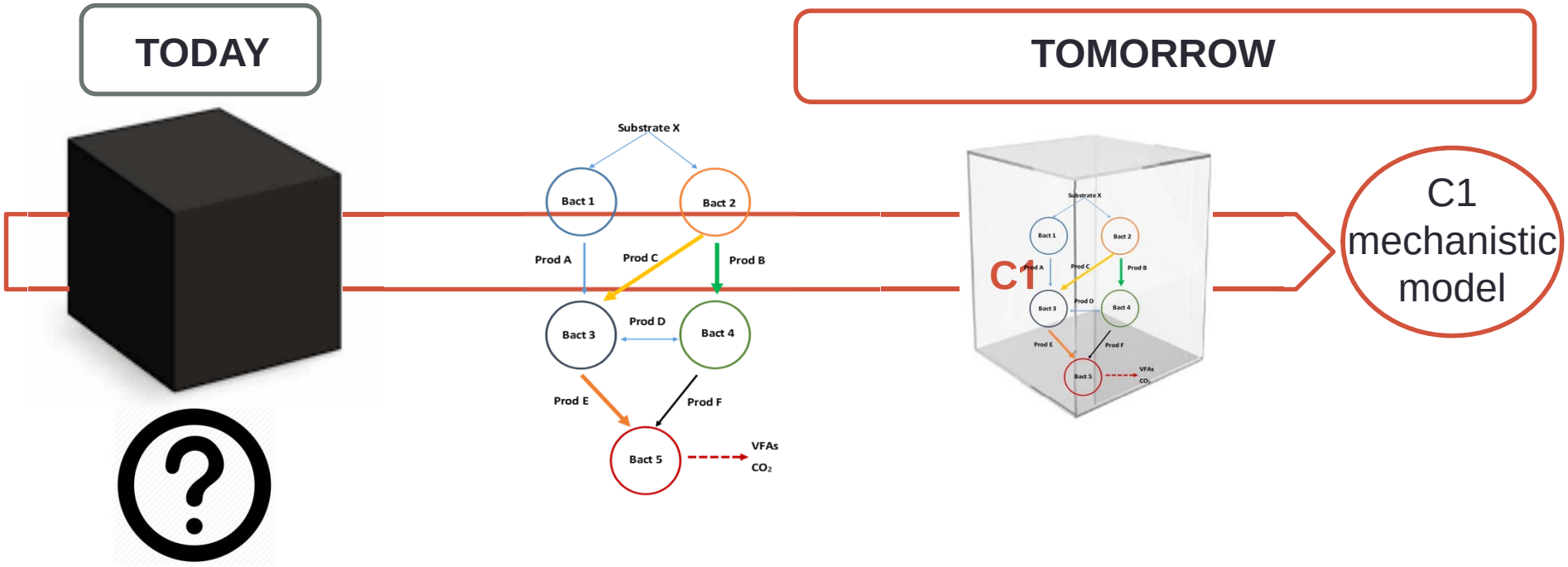
‘SmidgION’

Summary


TODAY
TOMORROW
Substrate X
Bact 1
Bact 2
Prod A
Prod C
Prod B
Bact 3
Prod D
Bact 4
Prod E
Prod F
Bact 5
VFAs
CO_2
C1
C1
mechanistic
model

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