Report and recommendations

European Plant Science Organisation
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EPSO Workshop
Implementing a Plants and Microbiomes Strategy in Europe
Cologne, October 2018

Brussels, 18.10.2019

1. Background

The aim of this second EPSO workshop on Plants and Microbiomes was to assess progress in implementing the recommendations from the 1st workshop and to update the needs in research and innovation among the microbiome actors in order to provide advice to the then discussed EU Horizon 2020 Work Programme 2020 and now its successor "Horizon Europe", particularly the Strategic Programming.

Overall, the meeting intended to increase collaborations between the working group members through e.g. a COST action, Coordination & Support Actions and initiate more bi- and multi-lateral collaborations.

2. Recommendations – updated from the 1st workshop

We now added the current state of the art and we suggest actions for the topics if necessary.

Main message:

1-Diverse crops with diverse microbiomes for diverse diets for human and animal health and resilient production systems

►Diversity matters increasingly, this suggestion should be most welcome and supported with high priority to achieve the UN Sustainable Development Goals, e.g. SDG 2 'Zero hunger', SDG 3 'Health and wellbeing', SDG 12 'responsible consumption and production', SDG 13 'Climate action', SDG 15 'Life on land'.

2-The term plant microbiome / plant microbiota comprises all microorganisms being associated with the plant including human/animal/plant pathogens

There is some controversy how to interpret the term plant microbiome. The participants clearly recommend to include all microorganisms (fungi, bacteria, archaea, viruses, protists) living at least part of their time in a sphere, which is influenced by the plant (i.e. rhizosphere, endosphere, phyllosphere etc.), whereby different plant compartment and tissues host distinct microbiota. The concept of the core microbiota is useful to distinguish ubiquitous and persistent commensals from microbial tourists of plants. As insects are not microbial organisms and serve as hosts of a dedicated insect microbiota, they should not be considered as integral part of the plant holobiome.

√ The term is well defined, no need for further adjustment. A definition paper will be published as an outcome of the MicrobiomeSupport project around early 2020.

3-Moving from correlation to causation under lab, greenhouse and field conditions

We currently base our understanding mostly on correlations (e.g. microbiome diversity with certain plant traits or functions) and need additional understanding on causality. Therefore, we need to develop and utilize a four-component system (synthetic communities + gnotobiotic plants + defined cultivation matrix + defined growth conditions) for the main model crops in Europe to change only one component and trace its impact.

This has to be applied in a stepwise manner at laboratory, greenhouse and open field conditions.
Genetics and thus the use of genetic modified organisms in research are important to increase mechanistic understanding. Performing field experiments with such GMOs for solely experimental purposes would support efforts to advance our understanding of plant-microbe interactions under more natural conditions.
In particular, the following topics need further understanding: i) functions of the microbiome and interactions among microbiota members including aspects such as signal exchange, ii) plant responses to microbiota, iii) functional aspects, including plant nutrition N, P, of the microbiome and iv) linking between the microbiome and plant traits. Two additional aspects are becoming more important to investigate: The role of epigenetics within the plant microbiome and in plant-associated pathogens / beneficials, and the mobile elements of microbes such as plasmids and transposons (called the mobilome), which spread resistance and are of special interest for plant and human health.
►Continue effort to elucidate functions of microbiomes. Decipher mechanisms underlying invasion and persistence of commensal (additional) microbes in standing heterogeneous communities, learn from medical research (e.g. microbiome transplants). Refocus: in / from the lab, to greenhouse, to field.

4-More understanding on the complexity of the ecosystem-plant-microbiome system is needed

Finally, we have to take ecosystem complexity into consideration. Ultimately, microbiome understanding will lead to new aspects to be integrated in precision farming.
Multidisciplinary research is needed to understand the relevance of plant microbiomes in an ecosystem context. (Plant) Microbiomes are highly complex and we have to understand ecology and functioning at a multi-trophic level. This includes also a holobiont approach considering all organisms interacting as one biont and aspects such as parallel evolution, adaptation and transmission routes are to be considered.
The topics for understanding equal those at lab and field approach (see previous point).
►Complex longer – term issue, discuss at next meeting.

5-Plant mechanisms to attract / interact with microbiota require understanding

Limited understanding exists on how plants respond to beneficial microorganisms besides few well investigated examples such as rhizobia or mycorrhizae. The identification of plant (genetic) markers correlating with a beneficial plant response or being responsible for the interaction with specific microorganisms could open new avenues for plant breeding.
Recent literature pointed at traits implicated in mineral uptake as master switches for the assembly of the microbiota thriving at the root-soil interface. Likewise, specific plant secondary metabolites have been implicated as recruitment cues for at least some members of the microbiota in both model and crop plants.
►Encourage more research on "plant candidate traits attracting beneficial microbiota" combined with bottom-up genetic association studies considering microbiota composition an "external plant phenotype". This will be instrumental to identify plant genes underpinning microbiota recruitment and molecular markers associated to them to expand the breeders' toolkit for the development of microbiota-ready crops for sustainable agriculture.

6-Proposed reference plants for Europe include barley, potato, tomato, pea and strawberry

As model crops barley (for cereals, monocots), potato (dicots), tomato (for vegetables), pea (for legumes) and strawberry (for fruits) are proposed as they all are agronomic relevant in Europe. Tomato and barley are model crops, which are already in use in plant research. It should be considered that particularly for the understanding of mechanisms it is important that mutants and breeding lines are available and that research with well-established model plants such as *Arabidopsis* and *Medicago* should be further pursued.
►Extend to crops and their wild relatives (barley, tomato, legumes..), add rapeseed (Brassicaceae) and trees e.g. poplar to better cover the plant kingdom; continue (Arabidopsis).

7-Precompetitive research should address the identification of microbiome-based plant health and resilience indicators and microbiome understanding needed by the industry

The industry is interested to provide solutions in form of microbial products for sustainable crop production. However, the question remains if such solutions can be provided at local or global scales. Since abiotic factors, edaphic factors and crop species can greatly influence the microbiome, it is important to define a 'healthy microbiome', which necessitates the identification of plant health indicators. It is also important to identify if a 'core microbiome' or 'key species in the microbiome' is associated with healthy plants. This requires in depth investigation of crop-plant microbiomes as it is done in the human microbiome project. This type of research will lead to the identification of diverse sets of microbiomes as resilience indicators. This will contribute to the goal of diverse crops with diverse microbiomes.

►Not really implemented yet, urgently needed. Projects that target a specific feature in the agricultural practice can be tailored to the needs of industry, however projects that target mechanistic understanding would not necessary require a demand from the industry.
For instance, disease-suppressive soils are ecosystems where plants are protected from root pathogens due to antagonistic activities of the root microbiota. Suppressive soils have been described for various economically relevant soil-borne pathogens such as bacteria, fungi, nematodes and oomycetes and can be induced by continuous cultivation of a susceptible host plant. Disease suppressiveness can be transplanted to non-suppressive soils, which is conceptually analogous to fecal transplants in humans. Pre-competitive research on disease-suppressive soils is timely to explore this natural resource for the development of rational plant probiotics. Similarly, it is crucial to better understand the key microbial features underlying soil health being able to develop relevant soil health indicators. We also require advanced understanding on the ecology and activity of introduced microbial strains or consortia in the field, which factors (including host-microbe and microbial interactions) drive microbial establishment, colonization behavior and the expression of microbial activities to improve microbial applications in a knowledge-driven manner.

8-More understanding is needed to understand the interaction of plants (e.g. secondary metabolites) with the animal and human microbiomes, which is likely to affect animal/human health

In addition to the direct effects between the plant microbiome (e.g. endophytic microorganism) on the human gut microbiome, there are crucial indirect effects through the plants: plant metabolites (bioactives produced by the plant and /or by microbial endophytes living within the plant), and effects deriving from the interaction between plants and microbes, influence animal/human microbiomes and thereby impact human/animal health. To better understand the interaction of plant metabolites with the human microbiome we will have to consider two perspectives: (i) Identification of derived colonic metabolites with potential health benefits and (ii) The role of plant metabolites in animal / human microbiota modulation as a possible mechanism by which they may exert their effect. These are crucial to support the application of plant metabolites in the human diet and will be a component of clinical trials to further assess their bio-kinetics.
In addition, we can apply concepts, basic understanding and methodologies to different microbiomes.

► Not really implemented yet, urgently needed.

9-Plants may host human/animal pathogens, which should be considered in applications as well as in food safety assessment

Microorganisms belonging to pathogenic phyla or being at least related to human pathogens are commonly found in plant environments. A better understanding on the ecology of such pathogens in agricultural systems is needed to warrant food safety from plant produce.

►Discuss at next meeting with the coordinator of an on-going COST action on the issue.

10-International (beyond Europe) cooperation is highly recommended

International cooperation avoids research duplication and increases impact from funding coming from different countries, thereby allowing effective use of public funding. Sharing of databases and culture collections (comprising not only single strains but also consortia) as well as experiments, protocols, standardized procedures, testing environments would help make results more widely understandable and usable.
In addition, research collaborations across countries would allow testing and exchange of microbiomes across geo-climatic zones, leading to diverse global agricultural systems. Similarly, to sharing plant genetic resources, for Australian crop microbiomes may benefit southern European systems by providing crop-microbiome adaptations.

►There is a great interest for aligning EU with national programmes at a global level. Currently initiatives are scattered (e.g. 'Rice endophytes', 'N2 Africa', 'Back to roots'), the US initiated programme 'Phytobiomes Alliance' has an opening for international collaborative projects.
Funding non-EU partners from EU-funded projects or EU partners in other international projects is a current bottleneck. The CSA MicrobiomeSupport, which includes support to the IBF Microbiome, is an example of international cooperation.

11-Early and wide communication of plant and microbiome science and applications is recommended

To avoid experiences made e.g. with the public perception of GM plants or synthetic biology, it is recommended to address stakeholders at the local level and communicate clear and balanced messages, possibly formed into stories and illustrated by examples. Local stakeholders should include the public, NGOs, farming communities including organic and conservation farmers, retailers like SMEs and supermarkets, wholesale buyers, schools and regulators. Communication should also allow the public to see and experience plants and microbiomes, e.g. by including microbes in activities of the EPSO Fascination of Plants Day or in natural sciences museums. It is important to educate the public that most (plant) microbiome members are beneficial and very important for ecosystems and our well-being, but that some microorganisms can be / are pathogenic.

►Suggest professionals in social media / communication at EU level (e.g. in the EC) to gather and disseminate information on microbiome science and specific details of individual funded projects. In addition, the CSA MicrobiomeSupport contributes to communication of microbiome science and applications. However, appropriate communication is extremely important and should go in each microbiome project, beyond to that performed within the CSA.

12-The industry needs personnel trained in classical microbiology and modern microbiome skills

To improve current university education programmes, we propose to organize an international, interdisciplinary Master programme on plant microbiomes including classical microbiology, soil science, plant physiology, plant molecular biology, microbial ecology and bioinformatics under the Erasmus Plus scheme.

►As there is no 'dedicated' microbiome curricula, "boundaries" between experimental and computational biology curricula need to be removed to train experimental scientists in classical microbiology and modern microbiome skills incl. a broader understanding of bioinformatics and data analysis**.** We suggest wider use of existing platforms (e.g., ERASMUS plus, COST actions…) to strengthen relationships among Universities and provide more focused education and training in plant microbiome research.

13-Regulation of microbial products requires improvement to support European bioeconomy and make best use of the plant microbiomes potential

Current regulatory demands are prohibitive for SMEs to bring microbial products to the market. Regarding biocontrol products (against pests and pathogens), the regulatory process is too long. We recommend focusing the registration on safety and efficacy and introducing a "fast-track" procedure for organisms, which are not registered pathogens. This would support SMEs as often such products have only a limited market potential.

Regarding biofertilisers, currently different regulation requirements exist in different European countries. This is under revision and we suggest aiming for one regulation across Europe.

In addition, the regulation for bi-functional microbiome products with biocontrol as well as biofertiliser effects currently requires adhering to regulations for both uses. This prevents such products, or they are only marketed for one of the two uses they actually have.

SMEs in the microbiome product development suggest introducing a voluntary code of conduct among companies to state the active ingredient(s) and their proven effects. Currently several poorly defined microbial products are on the market which could cause a negative effect for the entire sector.

Microbiome applications might have various specifics from a regulatory point of view, which should be discussed between scientists and (EU) regulatory bodies. In addition, IP issues such as patenting of genes, which are likely to massively derive from microbiome sequencing, should be discussed as well.

►No progress yet, urgent action on details R13 needed, including streamlining the current procedure. Registering microbiome-derived products remains too long and complex. The CSA MicrobiomeSupport addresses this topic which is only a starting point, as various discussions and actions at various levels are required (see Annex p. 5).

14-Public programmes should focus on the lower TRLs, leaving the differentiation to companies themselves

Companies are best to take existing / proven knowledge and technologies closer to the market themselves to differentiate their products from their competitors. Therefore, the industry suggests that public funding supports development of basic proven knowledge and technologies towards solutions and may join such collaborative projects in the early stages of the research and innovation cycle (lower TRLs). Companies could be partner or member of a user group in such projects to facilitate guidance and early up-take by the industry. Companies will then undertake themselves up-scaling and validation in an economy-based context.

It will be important to apply the holistic and multidisciplinary approach - considering the complex interactions between soil, plant, environment and the microbiome - in the early as well as later stages of R&D to result in microbiome-based discoveries and innovations.

Industry is interested in a short time to market, increasing yield and thereby profit, while academia aims at open access and publication. This has to be discussed to support both sectors at the highest level. Model consortium agreements will be an important tool.

►The level of the applicability needs to be realistic: Horizon 2020 calls are too ambitious in this respect (only high TRLs). We urge linking to and funding basic research, then funding translation of knowledge from model species to crops, from laboratory to field, from small scale to large scale production.

15-Open access databases integrating (plant) microbiome and meta-data are required

There is currently a severe lack of capacity in sequence data storage and processing in Europe. This applies mainly to metagenome and other -omics data and related analyses. As research is moving from amplicon-based analyses to metagenomics-based research, the situation is likely to worsen in (the near) future. This includes a current tremendous dependency on US-based open access databases, which are largely overloaded, as well as limited available storage space for the deposition of raw sequence data needed for publication. A solution would be the establishment of a European (plant) microbiome database, enabling data deposition as well as data analyses. Ideally, the database is combined with other microbiome database resources in Europe, enabling utilization of e.g. annotation data from human, animal or environmental metagenomes. This database should be curated, manned and maintained by European resources. Metagenomic (DNA/RNA) based data should be accompanied with continued (accelerated) accumulation of genomic data from well characterized isolates from plant associated bacteria, but also from archaea, fungi, viruses and protists, which are currently underrepresented in the genomic databases. This is important for better and more accurate annotation of meta-omic data in future.

►Remind all to apply the FAIR principles: Findable, Accessible, Interoperable, and Reusable. This topic is addressed in general terms within a CSA MicrobiomeSupport workshop and recommendations will be provided by end of 2020.

16-Standards - best practices in plant microbiome research need to be implemented

Current lack of standards in sequence data (processing/deposition) makes meta-analyses comparing data from different experiments/research groups difficult and inaccurate. There is a need for introducing standards for minimum number of biological replicates, sampling procedures, sample treatment, recording of metadata, analytical pipelines and bioinformatic analysis.

►The technology is not yet enough standardized. Therefore: Define terms; Define which plant compartments are sampled (& separation methods); Use the same primer sets to sequence microbiota (to develop for oomycetes, protists), include viruses; Raw data deposition encouraged; Apply Minimum standardisation for reconstitution experiments. Recommendations will be elaborated within the CSA MicrobiomeSupport and made available by the end of 2020.

17-European infrastructure recommended for plant microbiome research

Infrastructure related to database development and maintenance was considered highly important. The establishment of a European Microbiome Competence Center (similar to the Joint Genome Institute in the US) would highly strengthen European Microbiome science. Such a center, not necessarily limited to plant microbiomes, should unify all aspects related to microbiomics. Such a center would act as a platform to unify and integrate all types of microbiome data. It is recommended to use already existing infrastructures in Europe to build a true collaborative infrastructural network. Existing plant phenotyping facilities in EU can be well integrated in and are highly useful for plant microbiome research.

►Challenge for the use of Microbiota in large phenotyping facilities (contamination between experiments) – suggestion to fund simpler, mobile and less costly equipment. Joint genotyping at EU level. Cultured collections / isolate collections should contain defined metadata and genome sequence, establish European (distributed) cultured collections – best dedicate a session at the next workshop on how to establish and fund facilities and collections. It is presently challenging to culture all plant-associated microbes from diverse natural ecosystems across Europe. Establishment of a European 'plant microbiota vault' should be considered as complementary long-term microbial resource for plant health. In such a vault plant specimen with their microbial assemblages can be preserved for future generations of humankind. This initiative could be coordinated with a similar approach undertaken to preserve microbial diversity for human health (see http://www.microbiotavault.org/).

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This 'Report with recommendations' as well as the 'Annex to the report' are available at https://epsoweb.org/working-groups/plants-and-microbiomes/ .

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Useful links

MicrobiomeSupport, a CSA project funded under Horizon 2020, 1.11.2018 – 31.10.2022
www.microbiomesupport.eu/

Report from 1st EPSO Workshop on Plants and Microbiomes, 23.3.2017 https://epsoweb.org/epso/epso-report-plants-and-microbiomes-from-1st-workshop-in-vienna-january-2017/2017/03/23/

EPSO Working Group Plants and Microbiomes:
https://epsoweb.org/working-groups/plants-and-microbiomes/

EPSO position on FP9, 19.9.2017:
https://epsoweb.org/wp-content/uploads/2018/11/17_09_19_EPSO_Position-on-FP9.pdf

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