Viewpoint

Building microbial synthetic communities: get inspired by the design of synthetic plant communities

Summary

In the last decade, the generation of host-associated microbial culture collections has allowed the fine disentangling of complex relationships between commensal microbes and their hosts, and within-microbiota interactions. Specifically, these culture collections have been used to construct microbial synthetic communities (SynComs), which allow the reconstruction of host microbiota in laboratory conditions. In three recent perspective publications, the importance of this tool has been highlighted, and the ground rules of utilization and designing of such SynComs have been laid out. It is important to note that although microbial SynComs are used to understand intricate ecological interactions occurring in natural conditions, the intraspecific genetic diversity present in natural microbial communities has been seldom considered in the design of interspecific microbial SynComs so far. In this Viewpoint, we therefore argue that designing microbial SynComs could benefit from recent developments in the design of synthetic plant communities, or plant SynComs. For instance, considering intraspecific plant genetic diversity and its effects on intra- and interspecific plant–plant interactions appears essential to better understand and predict highly productive and stable plant communities. Therefore, considering genetic diversity within microbial species undoubtedly represents an exciting opportunity to design innovative microbial SynComs.

Bridging the knowledge between laboratory observations and ecological processes in natural conditions is a long-standing aim of host-microbiota research. Constructing microbial SynComs in laboratory conditions has been a potent tool to address this disconnection and increase complexity under controlled conditions. In the context of this next generation of plant–microbiota studies, three perspective articles have been recently published, which set the tone for future work with SynComs. First, Northen *et al.* (2024) presented a framework to design reference microbial SynComs accessible by the plant microbiota research community. The authors highlighted the relevance of generating comprehensive microbial culture collections that mimic those of natural communities as the first requirement. Furthermore, they emphasized the importance of the public availability of such culture collections and their metadata. Finally, they listed a series of state-of-the-art methodologies to study and track microbial communities in SynCom experiments, which will allow further disentangling of microbe–microbe–host interactions. Similarly, Mehlferber *et al.* (2024) streamlined both dry and wet methodologies to study host–microbiota interactions, including plant–microbiota interactions. In addition, in this article, the authors argued for hypothesis-driven designs when building and evaluating microbial SynComs, tailored to the purpose of the study. Finally, the authors reminded us that, if this work is meant to be applied in clinical trials or natural environments, it is paramount to also keep in mind ethical considerations when moving to applications. By describing a more global approach to studying microbial communities, Chesneau *et al.* (2025) introduced the concept of synthetic ecosystems (SynEco), where both microbial SynComs and their related abiotic and biotic environments are manipulated and studied, at complementary organizational levels from genes to communities, through individuals (and vice versa).

Given the high microbial diversity found within natural communities at the interspecific level, maximizing microbial SynCom interspecific diversity, at both the taxonomic and functional levels, was also advocated in these three articles. In a complementary way, we argue that looking at the role of genetic and functional diversity within microbial species is critical for a better understanding of the functioning of microbial natural communities and/or interspecific SynComs (Fig. 1a). Indeed, several experimental studies reported the role of intraspecific diversity on, among others, the success of invaders (Jousset *et al.*, 2011; Eisenhauer *et al.*, 2012), plant pathogen protection (Hu *et al.*, 2016; Wang *et al.*, 2022), microbiota transmission from inocula to seeds to seedlings (Simonin *et al.*, 2023), and host performance (Fields *et al.*, 2021).

We also argue that the design of microbial SynComs would benefit from recent developments in the design of synthetic plant communities, hereafter named plant SynComs. These developments have been recently identified and listed by the international PLANTCOM network that gathers scientists from complementary disciplines (molecular and cellular biology, functional genetics, ecology, evolutionary biology, quantitative genetics, agronomy, and modeling) working on the ecologically relevant genetics of plant–plant interactions (Becker *et al.*, 2023). In particular, understanding and predicting highly productive and stable plant communities requires considering intraspecific plant genetic diversity and its effects on intra- and interspecific plant–plant interactions (Becker *et al.*, 2023). The necessity to consider both plant species and genotypic diversity and composition to predict complex plant assemblages was primarily based on the observation of intra- and interspecific variances of plant functional traits that

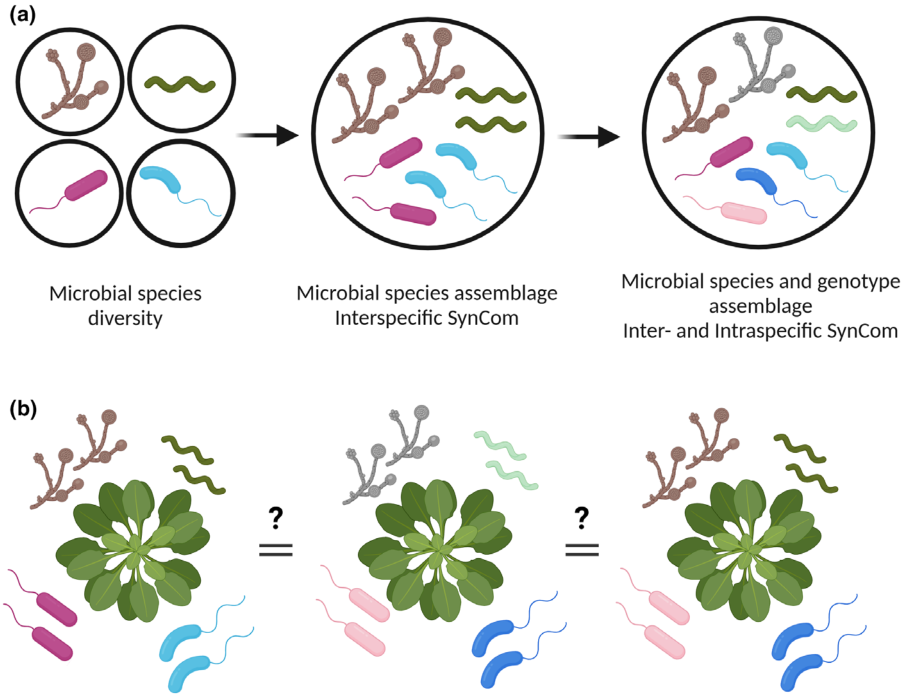

Fig. 1 Introducing intraspecific genetic diversity in the design of microbial synthetic communities (SynComs). To understand the functioning of natural microbial communities, the generation of comprehensive culture collections at the interspecific level and building SynComs has become the gold standard in the field of plant microbiota research. Here, we raise the following level of complexity, where the intraspecific genetic variation across microbial members is included (a). By doing so, we would be able to study whether microbial functions in association with their hosts depend on their specific genotype and whether SynComs equivalent at the species level but different at the genotypic level have an equivalent impact on their host and microbial community structure (b). This figure was created in BioRender (BioRender.com/o43v154).

often largely overlap in natural plant communities at a local geographic scale (Violle *et al*., 2012). This suggests that the function of one particular genotype of a plant species can be covered by one genotype of another plant species, that is, functional redundancy at the interspecific genotypic level. Interestingly, such a functional substitution of inter- and intraspecific diversity was also found in: (1) a mixed-species biofilm community composed of the three bacterial species *Pseudomonas aeruginosa*, *Pseudomonas protegens*, and *Klebsiella pneumoniae* (Lee *et al*., 2016); and (2) pathogen communities of the soft rot *Pectobacteriacea* (Barny *et al*., 2024). Thereafter, several empirical studies evidenced the importance of intraspecific plant genetic diversity in promoting local plant species diversity and co-existence. For instance, in managed grasslands, while multispecies assemblages were more productive than monocultures, increasing the number of genotypes per plant species increased the temporal stability of productivity, thereby demonstrating the complementary effects of species and genotypic diversity on proxies of performance of plant communities (Prieto *et al*., 2015). In addition, the design of plant SynComs each containing seven forage crops but differing in the level of genotypic diversity (represented by different numbers of cultivars per species) revealed a positive effect of genotypic diversity on yield, yield stability, and the equilibrium of species abundance in the mixtures over a five-year field trial (Meilhac *et al*., 2019). Finally, at a local scale, intraspecific genetic diversity can facilitate the adaptation of a particular plant species to strong environmental stress in complex plant assemblages, as demonstrated by a resurrection study conducted on a highly genetically polymorphic local natural population of the model plant *Arabidopsis thaliana*, which successfully adapted to climate warming over a period of 8 years while inhabiting a native plant community composed of almost 20 plant species (Frachon *et al*., 2017). In those empirical studies, the benefits of intraspecific genetic diversity on productivity and temporal stability of plant communities were linked to both spatial and temporal niche complementarity and niche expansion, which mainly resulted from Genotype × Genotype interactions and Genotype × Genotype × Environment interactions within and among plant species (Becker *et al*., 2023). However, we should stress that the effects of including intraspecific genetic diversity within multispecies plant assemblages might highly depend on the number of genotypes and genotypic composition per plant species. Indeed, as previously observed for crops, the magnitude and direction of the performance of variety mixtures compared with monocultures can strongly depend on both the number of varieties and the identity of the varieties included in a mixture (Reiss & Drinkwater, 2018; Alsabbagh *et al*., 2022; Montazeaud *et al*., 2022).

Whether intraspecific microbial genetic diversity leads to similar effects in the functioning of interspecific microbial communities and also affects plant–microbiota interactions remains an open question but certainly merits closer examination, notably in the design of microbial SynComs when studying interactions between a host and its microbiota (Fig. 1b). For instance, a large genetic diversity of strains has been reported at the population level, even at the individual level, for many pathogenic microbes in natural populations of *A. thaliana*, including bacterial pathogens such as *Pseudomonas syringae*, *Xanthomonas arboricola*, and *Xanthomonas campestris* (Bartoli *et al*., 2018; Karasov *et al*., 2018; Wang *et al*., 2018). The presence of genetically differentiated strains in a given local natural population of *A. thaliana* was also recently reported for some of the main bacterial commensals of the phyllosphere of *A. thaliana*, such as *Paraburkholderia fungorum* and a bacterial species belonging to the genus *Methylobacterium* (Ramírez-Sánchez *et al*., 2022). Altogether, these studies suggest the co-existence of numerous strains of a given bacterial species, potentially over several hundred thousand years (Karasov *et al*., 2018), within a complex plant microbiota. In line with the intraspecific genetic diversity observed at a very small organizational scale (e.g. within an individual plant), extensive genomic variation was observed among strains of a particular bacterial species interacting with *A. thaliana* in natural habitats, both in terms of single nucleotide polymorphisms/short insertion–deletion variants in the core genome and gene presence–absence polymorphisms in the accessory genome (Karasov *et al*., 2018; Wang *et al*., 2018). For instance, the *de novo* long read-based genome sequencing of six strains of the commensal bacteria *Pseudomonas siliginis*, the sixth most abundant species of the bacterial communities of 163 natural populations of *A. thaliana* located in the southwest of France (Bartoli *et al*., 2018) revealed that 16.9% of the orthogroups correspond to the accessory genome (Ramírez-Sánchez *et al*., 2022). In addition, by inoculating 162 genotypes of *A. thaliana* with 13 strains of seven commensal bacterial species of the phyllosphere of *A. thaliana*, a recent genome-wide association study conducted in field conditions revealed that the genetic architecture of the host plant response to a native commensal species was highly dependent on the strain identity of a given commensal bacterial species (Ramírez-Sánchez *et al*., 2024). This suggests that studying plant response to a particular microbial SynCom might be strongly affected by the identity of the native strains used to build the SynComs.

It is important to note that the lack of studies on intraspecific diversity in microbial communities is largely due to technical reasons, as the most common detection methods in diverse consortia, such as amplicon sequencing or quantitative polymerase chain reaction, generally do not allow for the study of genetically close strains (i.e. 16S rRNA resolution limited to the genus level). One example of improving bacterial identification was using highly degenerated primers for conserved marker genes in amplicon sequencing, such as a species-specific marker based on a fragment of the *gyrase B* gene (*gyrB*) (Barret *et al*., 2015). Nevertheless, further developments in this area are still needed to detect closely related strains. First, it is required to establish large collections of strains for the most dominant species in the microbiota (Northen *et al*., 2024; Mehlferber *et al*., 2024), as initiated for the phyllosphere microbiota of *A. thaliana* (Ramírez-Sánchez *et al*., 2022). These efforts will be facilitated by recent developments in streamlined workflows informed by mass spectrometry, cell sorting, strain profiling, and whole-genome sequencing (Northen *et al*., 2024). Monitoring within-species diversity in the microbial SynComs has greatly benefited from recent technological developments, such as genome-editing SynCom members (e.g. chromosomal integration of barcodes) (Daniel *et al*., 2024; Ordon *et al*., 2024) or through *in silico* analyses (e.g. by performing shotgun metagenomics and tracking strain-specific marker genes (Blanco-Míguez *et al*., 2023), or by sequencing amplicons of within-species variable regions).

Considering intraspecific genetic diversity, therefore, represents an exciting opportunity to design innovative microbial SynComs. This, in turn, may enhance our ability to understand and predict the functioning of microbial communities in natural and agroecosystems, and host–microbiota interactions. In particular, including intra- and interspecific diversity in microbial SynComs might help to understand the ecological mechanisms promoting: (1) microbial diversity maintenance, in particular in pathogen communities (Fierer & Lennon, 2011; Bartoli *et al*., 2018); (2) stability of the microbial communities by studying functional redundancy and complementarity (Puente-Sánchez *et al*., 2024); and (3) robustness to environmental perturbations (Johns *et al*., 2016). From an evolutionary point of view, it may also help to identify patterns in microbial population genomics (VanInsberghe *et al*., 2020), track co-evolutionary trajectories among microbiota members (Mesny *et al*., 2023), and uncover diffuse coevolution between a host and the key members of its microbiota (Karasov *et al*., 2014). Additionally, despite most microbes and plants largely differing in several life-history traits such as generation time, motility, and mating systems, testing whether similar community patterns and processes related to the effects of intraspecific genetic diversity emerge across both microbial and plant SynComs offers a lofty goal for establishing general rules in community ecology between kingdoms.

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Author contributions

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