Provisional Title

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Abstract

The abstract goes here.

1 Introduction

This is an example cite [1, 2]

2 Results

3 Discussion

4 Methods

4.1 Stabilization of environmental communities in simple synthetic environments

Communities were stabilized *ex situ* as described in [4]. In short, environmental samples (soil, leaves...) within one meter radius in eight different geographical locations were collected with sterile tweezers or spatulas into 50mL sterile tubes (Fig. **[missing ref(s)]**). One gram of each sample was allowed to sit at room temperature in 10mL of phosphate buffered saline (1×PBS) containing 200µg/mL cycloheximide to suppress eukaryotic growth. After 48h, samples were mixed 1:1 with 80% glycerol and kept frozen at -80°C. Starting microbial communities were prepared by scrapping the frozen stocks into 200µL of 1×PBS and adding a volume of 4µL to 500µL of synthetic minimal media (1×M9) supplemented with 200µg/mL cycloheximide and 0.07 C-mol/L glutamine or sodium citrate as the carbon source in 96 deep-well plates (1.2mL; VWR). Cultures were then incubated still at 30°C to allow for re-growth. After 48h, samples were fully homogenized and biomass increase was followed by measuring the optical density (620nm) of 100µL of the cultures in a Multiskan FC plate reader (Thermo Scientific). Communities were stabilized [4] by passaging 4µL of the cultures into 500µL of fresh media (1×M9 with the carbon source) every 48h for a total of 12 transfers at a dilution factor of 1:100, roughly equivalent to 80 generations per culture (Fig. **[missing ref(s)]**). Cycloheximide was not added to the media after the first two transfers.

4.2 Isolation of dominant species

For each community, the most abundant colony morphotype at the end of the ninth transfer was selected, resuspended in 100µL 1×PBS and serially diluted (1:10). Next, 20µL of the cells diluted to 10^{-6} were plated in the corresponding synthetic minimal media and allowed to regrow at 30°C for 48h. Dominants were then inoculated into 500µL of fresh media and incubated still at 30°C for 48h. After this period, the communities stabilized for eleven transfers and the isolated dominants were ready for the competition experiments (Fig **[missing ref(s)]**) at the onset of the twelfth transfer.

4.3 Simulations

We used the Community Simulator package [3] and included new features for our simulations. In the package, species are characterized by their resource uptake rates ($c_{i\alpha}$ for species i and resource α), and they all share a common metabolic matrix $\mathbf{D}$. The element $D_{\alpha\beta}$ of this matrix represents the fraction of energy in the form of resource α secreted when resource β is consumed. Here we implemented a new operation mode in which species can secrete different metabolites (and/or in different abundances) when consuming a same resource. Experimental observations support the idea of distinct species producing different sets of byproducts when growing in the same primary resource **[missing ref(s)]**. We call $D_{i\alpha\beta}$ to the fraction of energy in the form of resource α secreted *by species* i when consuming resource β —note that now $D_{i\alpha\beta}$ need not be equal to $D_{j\alpha\beta}$ if $i \neq j$, unlike in the original Community Simulator. In the package's underlying Microbial Consumer Resource Model [4, 5], this just means that the energy flux $J^{\text{out}}_{i\beta}$ now takes the form

$$J^{\text{out}}_{i\beta} = \sum_{\alpha} D_{i\beta\alpha} l_{\alpha} J^{\text{in}}_{i\alpha} \tag{1}$$

The documentation for the Community Simulator contains detailed descriptions of the model, parameters and package use. For the updated package with the new functionality, see Data & code availability.

For our simulations, we first generate a library of 660 species (divided into three specialist families of 200 members each and a generalist family of 60 members) and 30 resources (divided into three classes of 10 members each). We split this library into two non-overlapping pools of 330 species each. We randomly sample 50 species from each pool in equal ratios to seed 100 resident and 100 invasive communities respectively. We then grow and dilute the communities serially, replenishing the primary resource after each dilution. We repeat the process 20 times to ensure generational equilibrium is achieved [4]. We then perform the *in silico* experiments by using the generationally stable communities to seed 100 coalesced communities that we again stabilize as described previously. Similarly, we identify the dominant (most abundant) species of every resident and invasive community to carry out pairwise competition and single invasion simulations. Most parameters are set to the defaults of the original Community Simulator package. Table **[missing ref(s)]**shows those that are given non-default values to ensure enough variation in the primary communities.

5 Data & code availability

Experimental data and code for the analysis, as well as code for the simulations and the updated Community Simulator package with instructions to use the new features can be found in `github.com/jdiazc9/coalescence`.

6 Figures

7 References

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