

The Anna Karenina Principle in the microbiome: How *Pilidium lythri* destabilizes bacterial communities in soil

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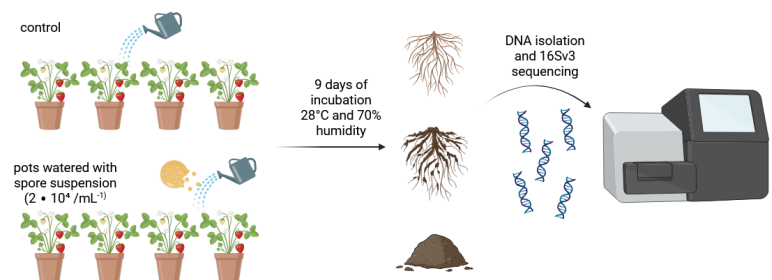
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INTRODUCTION

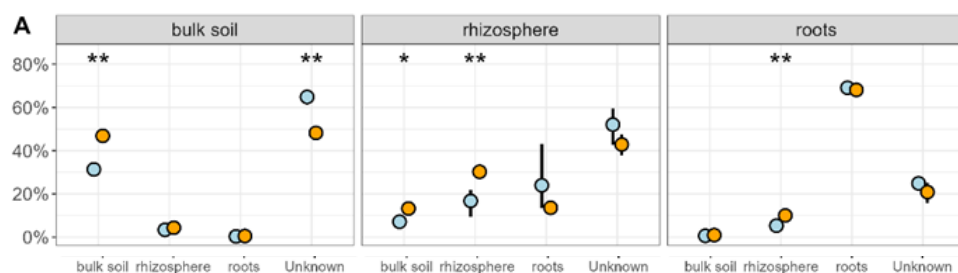
- ⇒ Global warming causes spread of thermotolerant fungal pathogens to new areas
- ⇒ *Pilidium lythri*—phytopathogen affecting many agriculturally important plants (olive, cornelian cherry, European plum, grapevine) and many ornamental flowers; with **strawberries as the primary host**
- ⇒ First recognised as a strawberry pathogen in Iran and China; now **expanding geographical distribution** to temperate climate, with recent identification in organic strawberry plantations in Poland
- ⇒ **Non-host specific**, capable of infecting both wounded and intact fruits
- ⇒ Despite its **accelerated spread due climate change**, significant knowledge gap remains regarding its influence on native microbiome structure

METHODS



- ⇒ DNA was isolated with EURx GeneMATRIX Soil DNA Purification Kit
- ⇒ Libraries were sequenced on Illumina MiSeq with Reagent Kit v3 (2x300)
- ⇒ Downstream analyses: QIIME2, Silva 138, microeco, SourceTracker

RESULTS



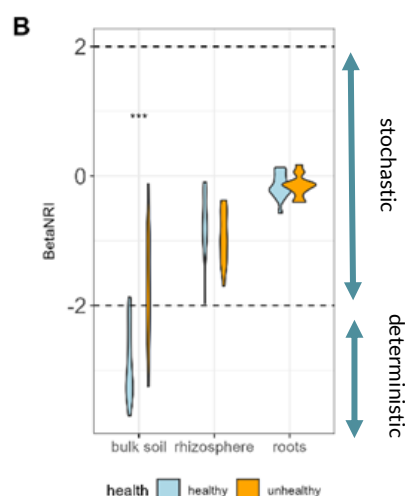
To elucidate the influence of *Pilidium lythri* on microbial migration between the niches, we used SourceTracker. Samples from sampling at 0 days were used as a source, and samples from sampling at 9 days as sinks. In bulk soil, bacterial community revealed significantly higher share of microorganisms in bulk soil and a lower portion from unknown source in infected samples (Fig. A). Furthermore, we observed migration of bacteria from bulk soil to rhizosphere in infected communities, but also from rhizosphere to rhizosphere for bacteria in infected communities. Finally, analysis revealed increased migration from rhizosphere to roots in infected samples in bacterial community.

SourceTracker is a Bayesian tool used to estimate the proportions of microbial communities that come from various known sources (like soil, rhizosphere, roots, etc.). It utilizes Gibbs sampler. (doi: 10.1038/nmeth.1650)

Beta Net Relatedness Index (β NRI) is a phylogenetic beta-diversity metric. It measures how different two communities are in terms of the evolutionary relationships among the species they contain. It tells us whether two microbial communities are made up of species that are more or less closely related than expected by chance. If the communities are made up of very distantly related species, β NRI will be higher than |2| (suggesting different environmental filtering or evolutionary histories). (doi: 10.1038/ismej.2013.93)

"All healthy microbiota are alike; each disease-associated microbiota is sick in its own way." - Anna Karenina Principle

Arnault, G., Mony, C., & Vandenkoornhuysen, P. (2023). Plant microbiota dysbiosis and the Anna Karenina Principle. Trends in Plant Science, 28(1), 18–30



The β NRI results revealed a dominance of stochastic processes in both healthy and infected rhizosphere and root samples of strawberry (betaNRI values between -2 and 2). Furthermore, we observed differences in community assembly processes between healthy and infected bulk soil, with a shift from deterministic processes (characterized by lower turnover) in healthy bulk soil to a dominance of stochastic processes in infected soil (Fig. B).

CONCLUSIONS

- ⇒ In healthy bulk soil, microbial communities were shaped by deterministic processes — suggesting similar, stable, and predictable communities across replicates. This aligns with the AKP's idea of "all healthy microbiomes are alike."
- ⇒ In infected bulk soil, we observed a shift toward stochastic processes, meaning more random and variable communities — a hallmark of "each dysbiotic microbiome is dysbiotic in its own way."
- ⇒ This shift implies that *Pilidium lythri* infection disrupts environmental filtering, leading to less control over which microbes establish — causing greater divergence between infected samples.
- ⇒ The increased microbial migration between niches in infected plants may further contribute to dysbiosis and the individualized trajectories of each infected microbiome.

Pilidium lythri infection introduces chaos into the microbial community structure, echoing the Anna Karenina principle: while healthy soils are predictable, infected ones fall apart each in their own microbial way.

more info

