

Soil microbes metabolize the same polyphenols found in chocolate, wine

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Lead author Bridget McGivern at Ohio State University in June 2018, shortly after setup of the soil experiment. Credit: Colorado State University

Fruits, vegetables, red wine and chocolate are all rich in polyphenols, natural plant compounds that double as cancer-fighting antioxidants. We can access these foods' health benefits because the microbes in our guts happily feast on them, breaking them down into smaller chemical components.

Microbiome scientists at Colorado State University wanted to know if microbes can also break down those same polyphenols in systems outside the human body, including the microbial wild west of soils.

A research team led by Kelly Wrighton, associate professor in the College of Agricultural Sciences' Department of Soil and Crop Sciences, has uncovered new insights into the role of polyphenols in the [soil microbiome](#), known as a black box for its complexity. They proffer an updated theory that soils—much like the human gut—can be food sources for the microbes that live there. Their results could upend a long-held theory that, under certain conditions, soil microbes can't access polyphenols and could thus be used as carbon traps to reduce greenhouse gases in the atmosphere.

"Our study opens the door for further study of polyphenol metabolism in the field, and how it fits into natural carbon cycles," said soil and crop sciences Ph.D. candidate Bridget McGivern, first author on the paper describing the work.

Published in *Nature Communications*, the research involved precisely monitored soil samples subjected to a palette of very high-resolution analytical chemistry in the lab. The researchers set out to prove the concept that microbes in soil, under oxygen-free conditions, do in fact break down polyphenols, likely releasing carbon dioxide.

These experiments fly in the face of the longstanding "enzyme latch" theory that soil microbes do not metabolize polyphenols when oxygen

isn't freely available in places like flooded wetlands and peatlands. If it were true that polyphenols remained undigested in oxygen-free soils, it would mean loading up soils with these fibrous compounds could be an easy carbon-trapping sink.

"We know that polyphenols are really sticky, and so people thought that, by being in the soil, not only they themselves wouldn't be broken down, but they'd stick to other carbons and enzymes in the soil and prevent further breakdown of everything else," McGivern said. "So, if you had an already broken-down system like a degraded wetland, you could go in and add wood chips to the system, flood it again, and lock up all the carbon."

Thinking about soil

Wrighton first started thinking about how polyphenols behave in soil systems while a faculty member at Ohio State University; she joined the CSU faculty in 2018.

She couldn't shake the fact that this enzyme latch theory in soils just didn't make sense, based on what we know about how polyphenols are broken down in our guts, which are also oxygen-free environments. Wrighton at that point had become an expert in the gut microbiomes of moose, ruminants which have evolved over millions of years to take full advantage of the polyphenolic compounds in their herbivorous diets.

"The breakdown of polyphenols has to happen in the gut for us to access those antioxidants, and to appreciate chocolate and red wine and all their benefits," said Wrighton, who received a National Science Foundation CAREER award several years ago to study polyphenols. "But then we would go into this other ecosystem, which had a totally different paradigm of how these compounds behaved. We couldn't rationalize that."

Of course, it could be that the inherent complexity of the soil environment provided some other reason for polyphenols to behave differently there, Wrighton surmised. With her hypothesis in hand, and the right high-resolution chemistry tools available, she and McGivern set out to test whether microbes in soils could, indeed, break down polyphenols in oxygen-free conditions.

Their controlled laboratory experiments, bolstered by a bevy of experts who helped them analyze what they were seeing, showed they were right.

Chemical maze

Wrighton and McGivern teamed up with soil polyphenolic expert Ann Hagerman of Miami University in Ohio to tackle the problem.

Collaborators at the Department of Energy's Joint Genome Institute performed metagenomic sequencing of the CSU team's soil microbiome, giving the team a snapshot of every gene found in the microbial community. Other collaborators at the DOE Environmental Molecular Sciences Laboratory performed metaproteomics, which provided information about which proteins were being expressed by which genes in the samples.

The research team, with critical contributions by Trent Northen at Lawrence Berkeley National Laboratory and Malak Tfaily at University of Arizona, drilled down even further with three types of high-resolution, metabolite-identifying chemistry, all with the goal of showing how the large, complex polyphenol compounds were being pulled apart by microbes into their small-molecule components. And they had to keep straight which molecules originated from the organic polymers and which were background noise from the soil matrix.

"It was just this chemical maze, tracking these compounds," Wrighton said.

Next steps: permafrost

Now that the team has shown, in a lab setting, that polyphenols are food sources for soil microbes in oxygen-free zones, the next logical step is to show the same behavior in a field setting. The team was recently awarded a grant to test their theories within a permafrost system in Sweden, along with another team of collaborators. Many believe thawing permafrost holds the greatest potential for greenhouse gas reductions via carbon lockdowns, so understanding how such [soil](#) systems work is critical, according to Wrighton and McGivern.

But first, McGivern will work on building a computational infrastructure that better categorizes and identifies the different enzymes associated with microbial polyphenolic metabolisms—information that's not widely available in public databases.

"My next project is basically building a polyphenol module to insert into an existing annotation infrastructure in our lab, so that if anybody goes in and annotates their genome, they'll start to see [polyphenol](#) metabolisms," McGivern said.

And the test case she's going to use to build that system? The human gut microbiome.

More information: Bridget B. McGivern et al, Decrypting bacterial polyphenol metabolism in an anoxic wetland soil, *Nature Communications* (2021). [DOI: 10.1038/s41467-021-22765-1](https://doi.org/10.1038/s41467-021-22765-1)

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