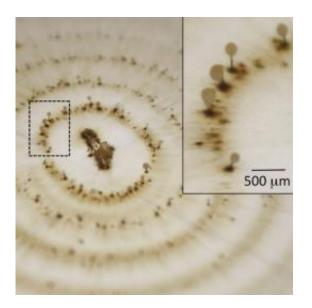


To clean up the mine, let Ascomycete fungus reproduce

July 16 2012



This colony of the fungus *S. aciculosa* shows manganese oxide deposits at the base of the reproductive bodies. Researchers have shown that superoxide transforms manganese into its environmentally useful mineral state. Credit: Photo courtesy of Colleen Hansel

Harvard-led researchers have discovered that an Ascomycete fungus that is common in polluted water produces environmentally important minerals during asexual reproduction.

The key chemical in the process, superoxide, is a <u>byproduct</u> of <u>fungal</u> <u>growth</u> when the organism produces <u>spores</u>. Once released into the environment, superoxide reacts with the element manganese (Mn),



producing a highly reactive mineral that aids in the cleanup of <u>toxic</u> <u>metals</u>, degrades carbon substrates, and controls the <u>bioavailability</u> of nutrients.

The results, which will inform a wide range of future studies in microbiology, environmental chemistry, <u>developmental biology</u>, and geobiology, were published online this week in the <u>Proceedings of the National Academy of Sciences</u> (*PNAS*).

Manganese is a versatile element, existing in multiple <u>oxidation states</u> and phases. Naturally occurring in the Earth's crust, it plays essential roles in <u>carbon sequestration</u>, <u>photosynthesis</u>, and the transport and fate of nutrients and contaminants.

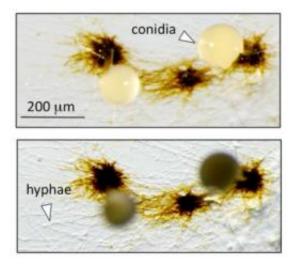
It can be an especially important reactant in polluted water, such as the runoff from <u>coal mines</u>. When the ion Mn(II) is converted to higher oxidized states, Mn(III) and Mn(IV), it forms a reactive mineral that is extremely useful in getting other pollutants—like arsenic, cadmium, and cobalt—under control and out of the water.

"If you can get manganese to oxidize, then it forms these really active minerals, manganese oxides, which are environmental sponges that will clean up the water," explains principal investigator Colleen Hansel, a faculty associate and former associate professor of environmental microbiology at the Harvard School of Engineering and Applied Sciences (SEAS). She is currently an associate scientist at Woods Hole Oceanographic Institution. "A lot of coal mine drainage remediation relies on getting bacteria and fungi to oxidize manganese to make these minerals."

"One problem with in situ remediation is that if you don't know how and why processes are occurring, you can't stimulate the organisms to do it. That's been a big problem with the remediation of coal mine drainage



sites. To stimulate microbial activity, the approach has been to provide complex carbon sources like corn cobs and straw and let the 'bugs' go to town, but it frequently doesn't work."



The fungus *S. aciculosa* produces spores (called conidia) on stalks during asexual reproduction. The brown color at the base of these structures is the natural color of the manganese oxides that precipitate there following oxidation of reduced manganese by superoxide. The hyphae are not involved in oxidation. Credit: Photo courtesy of Colleen Hansel

It turns out that the common fungus *Stilbella aciculosa* only produces the necessary ingredient, superoxide, during cell differentiation (an aspect of growth and development)—specifically, during the formation of asexual reproductive structures. The finding implies that adding excessive nutrients to <u>polluted water</u> may not necessarily contribute to remediation, unless it is designed to induce fungal reproduction.

For the fungus, superoxide appears to serve as cellular signal that moderates cell differentiation. The chemical's subsequent role in oxidizing environmental manganese so rapidly and efficiently may just



be a useful coincidence, beneficial to humans but of little consequence to the fungus.

All of the manganese-oxidizing bacteria and Ascomycete fungi known to date are heterotrophs; like humans, they eat carbon and breathe oxygen.

"They're not eating manganese the way some organisms eat other metals like iron," says Hansel. "This has been an enigma, in the field of metal biogeochemistry. According to evolutionary theory, organisms usually perform a process for a reason. But for decades no one has understood why or how bacteria and some groups of fungi (the Ascomycota) were oxidizing manganese, because they weren't doing it to gain energy."

Still, Hansel suggests that there may be more to the process than meets the eye.

"It looks like an accidental side reaction, but we don't really know, because manganese oxides are very reactive and could therefore provide some indirect benefits to the organism," she says. "The manganese oxides could, for instance, degrade recalcitrant carbon and thus feed the fungi new carbon sources that they can metabolize better. Maybe they are 'purposely' doing it. We want to address these biochemical questions and the evolutionary implications, as well as determine the larger relevance of superoxide-based metal cycling. How important is this process in terms of the biogeochemistry of other metals like iron and mercury? How significant is its impact on the ecology of microbial ecosystems?"

With co-authors Carolyn A. Zeiner (a graduate student at SEAS), Cara M. Santelli (a former postdoc, now at the Smithsonian National Museum of Natural History), and Samuel M. Webb (Stanford Synchrotron Radiation Lightsource), Hansel identified the biochemical mechanism that leads to the oxidation of manganese, including the class of enzymes



(NADPH oxidases) that spur the process.

The team's discovery that <u>superoxide</u> is the key player in fungal oxidation of <u>manganese</u> is especially exciting because some bacteria actually do it the same way, even using the same enzymes. The idea that prokaryotes and eukaryotes developed this homology raises intriguing questions in the history of evolution.

"We're traveling down a whole new avenue in biogeochemistry," says Hansel. "It's exciting right now to be one of the people sitting in the front seat."

Provided by Harvard University

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