



# Lichen takes more than two to tango

## Educator guide

### PAPER DETAILS

**Original title:** Basidiomycete yeasts in the cortex of ascomycete macrolichens

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**Authors:** Toby Spribille, Veera Tuovinen, Philipp Resl, Dan Vanderpool, Heimo Wolinski, M. Catherine Aime, Kevin Schneider, Edith Stabentheiner, Merje Toome-Heller, Göran Thor, Helmut Mayrhofer, Hanna Johannesson, John P. McCutcheon

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**Annotator(s):** Anne Farrell, Shannon Soucy, and Olga Zhaxybayeva

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## DISCUSSION QUESTIONS

1. Besides secondary metabolite (vulpinic acid) production, what other important functions might be performed in the cortex layer by this newly discovered symbiont? Think of where lichen live, and how they make a living.

(Note to educators: If you live in a place with access to lichen, providing real, local examples of different types and structures of lichen would help make discussion more interactive.)

2. Describe three pieces of evidence that the paper uses to support the conclusion that *Cyphobasidium* is a necessary member of lichen symbiosis? How do these pieces support each other? Did the order of experimental methods matter in arriving at the conclusion? If the order of analyses had changed, would the authors have made this discovery?
3. The researchers knew from prior studies that the two different lichens were composed of the same symbiont species. How did researchers initially explain why one lichen produced vulpinic acid? What features of DNA supported this initial hypothesis? What can cause two cells or organisms with the same DNA to have phenotypic differences? What results prompted scientists to look for another explanation?
4. For Figure 1, discuss the interpretation of the phylogenetic trees. Given the scale bar on each phylogeny and the “tightness” of clusters in each phylogeny, which phylogeny has more diversity among members—the fungus(A) or the alga(B)? How do you know? What does the scale of (A) and (B) tell you? What can you infer from the fungus tree? What can you infer from the alga tree?
5. In Figure 1, panels C,D,E: How many genes are significantly differentially regulated in alga associated with two phenotypes? How do data in Figures 1C and 1E lead authors to conclude that “little differential transcript abundance [is] associated with [the] phenotype?” Do plots on Figures 1C and 1E represent results the authors expected to see?
6. How would you test if there are any other undiscovered symbionts involved in lichen formation?
7. Why is understanding lichen symbiosis important? What are the broader impacts that this research might have on other biological research? Other research in other disciplines?

### LEARNING STANDARDS

LS1.A

LS2.A

Structure and Function

EK2.D.1

EK2.E.3

SEP4

SEP8

RST.11-12.5

RST.11-12.6

SP4

SP5

EK1.A.4

EK.1.B.2

LS3.B

Cause and Effect

EK1.B.1

SEP4

Stability and Change

SP5

SP6

EK.1.B.2

EK.1.A.4

EK.1.B.1

LS1.A

LS3.B

Cause and Effect

Structure and Function

RST.11-12.8

EK.1.B.1

Cause and Effect

SP4

Nature of Science

SEP6

Nature of Science

## ACTIVITIES FOR INTERACTIVE ENGAGEMENT

### Writing an abstract

Students write a new abstract for the article at a grade-appropriate reading level.

### Locating this study in the larger field

Students use the annotated list of references to explain how this research builds on the published work of at least one other independent group of scientists. Students will evaluate whether data from this research supports or contradicts previous conclusions, and reflect on the statement that scientific knowledge is a “community effort.”

### Science in the news

Students explore news stories in the Related Resources tab and evaluate the stories for tone, accuracy, missing information, etc. Students may then write their own popular press article, at a grade appropriate level, about lichen tri-symbiosis. They should be sure to address any weaknesses they identified in previously published pieces.

### Make parallels with other systems

Lichen is defined by symbiosis, particularly mutualism. These interactions affect the structure of an ecosystem. To introduce the idea of symbioses and draw parallels between lichens and other mutualistic relationships, use the following resource—developed with an emphasis on marine ecosystems.

<https://www.nationalgeographic.org/activity/ecological-relationships/>

Define and discuss the types of symbioses: mutualism, commensalism, and parasitism. How do the biological mutualisms in this activity compare to the symbiotic relationships found in lichens?

### Understanding phylogenetic trees

The techniques used by the researchers included many phylogenomic methods. To help students interpret the figures, they should be able to understand how scientists build and interpret evolutionary trees. Science in the Classroom’s partner, HHMI BioInteractive, provides a lesson on biodiversity and evolutionary trees, including an activity where students can look at differences in real DNA sequences and learn how this information is used to create trees. This would likely be a multiday activity for a class that is focused on learning more about the evidence for evolution.

<https://www.hhmi.org/biointeractive/biodiversity-and-evolutionary-trees>

<http://www.hhmi.org/biointeractive/creating-phylogenetic-trees-dna-sequences>

### The next steps

Students design a follow-on experiment to this study that either addresses flaws or unanswered questions in the research at hand, or builds on it to explore a new question.

#### LEARNING STANDARDS

**RST.9-10.2**

**RST.11-12.2**

**Nature of Science**

**RST.9-10.8**

**RST.11-12.8**

**Nature of Science**

**SEP4**

**SEP8**

**RST.9-10.5**

**RST.9-10.6**

**RST.11-12.1**

**RST.11-12.5**

**SP7**

**LS1.A**

**LS2.A**

**Structure and Function**

**RST.11-12.9**

**EK2.D.1**

**EK2.E.3**

**SEP4**

**LS3.B**

**Stability and Change**

**RST.11-12.9**

**EK1.A.1**

**EK1.A.4**

**EK1.B.1**

**EK1.B.2**

**Nature of Science**

**SEP1**

**RST.11-12.6**

**SP3**

**SP4**

**Nature of Science**

## ARTICLE OVERVIEW

### Article summary (recommended for educator-use only)

Lichens are one of the oldest known examples of a symbiotic organism. Lichens are made up of a fungus and an alga. The fungus provides the structure and “body,” and the alga provides energy through photosynthesis. However, attempts in the laboratory to synthesize a lichen using only these two partners were never successful. In this work, the researchers discover a third symbiont that builds the outer cortex of the lichen.

### Importance of this research

As stated above, lichens are one of the oldest examples of a symbiotic organism. Yet, the ubiquity and importance of symbiosis in living systems has only recently been realized. By combining new technologies in genomic and molecular methods, researchers detected a novel symbiont. This research is a great example of how scientists can use good questions and modern tools to discover something new that had been there all along.

### Experimental methods

- Metatranscriptomics
  - Transcriptome
  - Transcriptome abundance mapped against Ascomycota
  - Transcriptome abundance mapped against all fungi
- Polymerase Chain Reaction (PCR) with primer specific to the newly discovered symbiont
- Phylogenetics
  - 349 loci phylogeny
  - rDNA phylogeny
  - Molecular Clock
    - Fossil calibrations
- Microscopic imaging
  - Scanning Electron Microscopy (SEM)
  - Fluorescent *in situ* Hybridization (FISH)

### Conclusions

- There is a third partner that contributes to cortex formation of lichens. This tripartite symbiosis evolved long ago.
- All members of this newly discovered group of yeasts (*Cyphobasidiales*) are associated with lichens.
- Each *Cyphobasidiales* is specific to a species of lichen, and their presence accounts for phenotypic differences in lichen species.

## LEARNING STANDARDS ALIGNMENT

The following tables provide an overview of the learning standards covered by this article, including the A Framework for K-12 Science Education (Framework), Common Core State Standards English Language Arts-Literacy (CCSS ELA), Common Core State Standards Statistics and Probability (CCSS HSS), AP Science Practices, and Vision and Change for Undergraduate Education. Where applicable, activities and information will be marked with specific standards to which they are linked.

A Framework for K-12 Science Education		
Science and Engineering Practices	Disciplinary Core Ideas	Crosscutting Concepts
<p><b>Asking questions (SEP1)</b> Evaluate questions and challenge the premise(s) of an argument, the interpretation of a data set, or the suitability of a design.</p> <p><b>Analyzing and interpreting data (SEP4)</b> The student can perform data analysis and evaluation of evidence.</p> <p><b>Constructing explanations (SEP6)</b> Evaluate a solution to a complex real-world problem, based on scientific knowledge, student-generated sources of evidence, prioritized criteria, and tradeoff considerations.</p> <p><b>Obtaining, evaluating, and communicating information (SEP8)</b> Evaluate the validity and reliability of and/or synthesize multiple claims, methods, and/or designs that appear in scientific and technical texts or media reports, verifying the data when possible.</p>	<p><b>LS1.A: Structure and function</b> Systems of specialized cells within organisms help them perform the essential functions of life.</p> <p><b>LS2.A: Interdependent relationships in ecosystems</b> Ecosystems have carrying capacities, which are limits to the numbers of organisms and populations they can support. These limits result from such factors as the availability of living and nonliving resources and from such challenges such as predation, competition, and disease. Organisms would have the capacity to produce populations of great size, were it not for the fact that environments and resources are finite. This fundamental tension affects the abundance (number of individuals) of species in a given ecosystem.</p> <p><b>LS3.B: Variation of traits</b> The set of variations of genes present, together with the interactions of genes with their environment, determines the distribution of variation of traits in a population.</p>	<p><b>Cause and effect: Mechanism and explanation</b> Events have causes, sometimes simple, sometimes multifaceted. A major activity of science is investigating and explaining causal relationships and the mechanisms by which they are mediated. Such mechanism can be tested across given contexts and used to predict and explain events in new contexts.</p> <p><b>Structure and function</b> The way in which an object or living thing is shaped and its substructure determine many of its properties and functions.</p> <p><b>Stability and change</b> For natural and built systems alike, conditions of stability and determinants of rates of change or evolution of a system are critical elements of study.</p>

Common Core State Standards English Language Arts-Literacy		
Key Ideas and Details	Craft and Structure	Integration of Knowledge and Ideas
<p><b>RST.9-10.1</b> Cite specific textual evidence to support analysis of science and technical texts, attending to the precise details of explanations or descriptions.</p> <p><b>RST.9-10.2</b> Determine the central ideas or conclusions of a text; trace the text’s explanation or depiction of a complex process, phenomenon, or concept; provide an accurate summary of the text.</p> <p><b>RST.11-12.1</b> Cite specific textual evidence to support analysis of science and technical texts, attending to important distinctions the author makes and to any gaps or inconsistencies in the account.</p> <p><b>RST.11-12.2</b> Determine the central ideas or conclusions of a text; summarize complex concepts, processes, or information presented in a text by paraphrasing them in simpler but still accurate terms.</p>	<p><b>RST.9-10.4</b> Determine the meaning of symbols, key terms, and other domain-specific words and phrases as they are used in a specific scientific or technical context relevant to grades 9-10 texts and topics.</p> <p><b>RST.9-10.5</b> Analyze the structure of the relationships among concepts in a text, including relationships among key terms (e.g., force, friction, reaction force, energy).</p> <p><b>RST.9-10.6</b> Analyze the author’s purpose in providing an explanation, describing a procedure, or discussing an experiment in a text, defining the question the author seeks to address.</p> <p><b>RST.11-12.4</b> Determine the meaning of symbols, key terms, and other domain-specific words and phrases as they are used in a specific scientific or technical context relevant to grades 11-12 texts and topics.</p> <p><b>RST.11-12.5</b> Analyze how the text structures information or ideas into categories or hierarchies, demonstrating understanding of the information or ideas.</p> <p><b>RST.11-12.6</b> Analyze the author’s purpose in providing an explanation, describing a procedure, or discussing an experiment in a text, identifying important issues that remain unresolved.</p>	<p><b>RST.9-10.8</b> Assess the extent to which the reasoning and evidence in a text support the author’s claim or a recommendation for solving a scientific or technical problem.</p> <p><b>RST.9-10.9</b> Compare and contrast findings presented in a text to those from other sources (including their own experiments), noting when the findings support or contradict previous explanations or accounts.</p> <p><b>RST.11-12.8</b> Evaluate the hypotheses, data, analyses, and conclusions in a science or technical text, verifying the data when possible and corroborating or challenging conclusions with other sources of information.</p> <p><b>RST.11-12.9</b> Synthesize information from a range of sources (e.g., texts, experiments, simulations) into a coherent understanding of a process, phenomenon, or concept, resolving conflicting information when possible.</p>

AP Science Standards	
AP Science Practices	AP Biology Content Standards
<p><b>Science Practice 3 (SP3)</b> The student can engage in scientific questioning to extend thinking or to guide investigations within the context of the AP course.</p> <p><b>Science Practice 4 (SP4)</b> The student can plan and implement data collection strategies appropriate to a scientific question.</p> <p><b>Science Practice 5 (SP5)</b> The student can perform data analysis and evaluation of evidence.</p> <p><b>Science Practice 6 (SP6)</b> The student can work with scientific explanations and theories.</p> <p><b>Science Practice 7 (SP7)</b> The student is able to connect and relate knowledge across various scales, concepts, and representations in and across domains.</p>	<p><b>Essential knowledge 1.A.4 (EK1.A.4)</b> Molecular, morphological, and genetic information of existing and extinct organisms add to our understanding of evolution. Biochemical and genetic similarities, such as DNA nucleotide and protein sequences, provide evidence for evolution and ancestry.</p> <p><b>Essential knowledge 1.B.1 (EK1.B.1)</b> Organisms share many conserved core processes and features that evolved and are widely distributed among organisms today. (EK1.B.1a) Structural and functional evidence supports the relatedness of all domains. DNA and RNA are carriers of genetic information through transcription, translation, and replication.</p> <p><b>Essential knowledge 1.B.2 (EK1.B.2)</b> Phylogenetic trees and cladograms are graphical representations (models) of evolutionary history that can be tested.</p> <p><b>Essential knowledge 2.D.1 (EK2.D.1)</b> All biological systems from cells and organisms to populations, communities, and ecosystems are affected by complex biotic and abiotic interactions involving exchange of matter and free energy.</p> <p><b>Essential knowledge 2.E.3 (EK2.E.3)</b> Cooperative behavior within or between populations contributes to the survival of the populations, i.e. mutualistic relationships.</p>

Connections to the Nature of Science	
Vision and Change for Undergraduate Biology Education Core Competencies and Disciplinary Practices	A Framework for K-12 Science Education Understandings About the Nature of Science
<p><b>Ability to tap into the multidisciplinary nature of science</b> Solid grounding in the sciences, including computer science and social science, can advance the practice and comprehension of biology. Accordingly, all students should have experience applying concepts and subdisciplinary knowledge from within and outside of biology in order to interpret biological phenomena.</p> <p><b>Ability to understand the relationship between science and society</b> Identify social and historical dimensions of biology practice: evaluating the relevance of social contexts to biological problems, developing biological applications to solve societal problems, evaluating ethical implications of biological research.</p>	<p><b>Scientific investigations use a variety of methods</b> New technologies advance scientific knowledge. Scientific investigations use a variety of methods, tools, and techniques to revise and produce new knowledge. Scientific inquiry is characterized by a common set of values that include: logical thinking, precision, open-mindedness, objectivity, skepticism, replicability of results, and honest and ethical reporting of findings.</p> <p><b>Scientific knowledge is open to revision in light of new evidence</b> Scientific argumentations is a mode of logical discourse used to clarify the strength of relationships between ideas and evidence that may result in revision of an explanation.</p>