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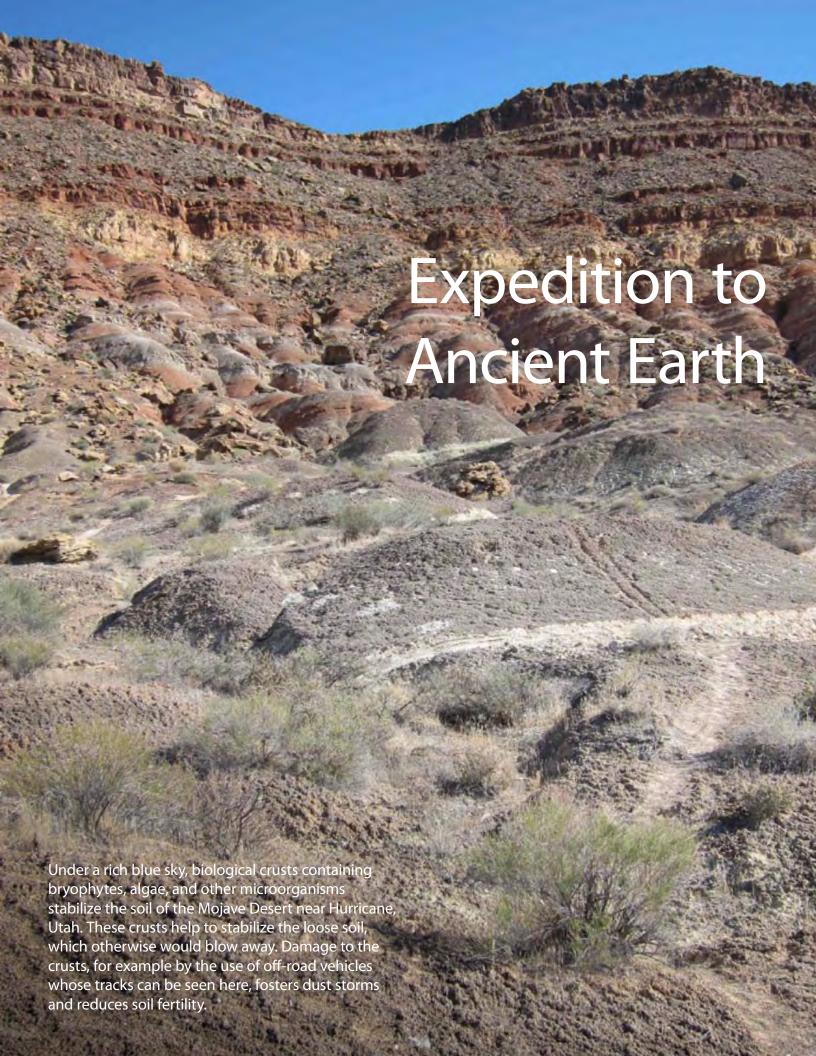
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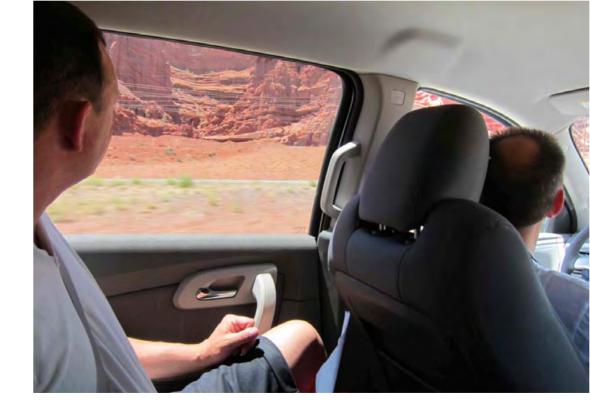


Text and photographs by Linda E. Graham

landscapes like before the rise of the first land plants? Could tough algae and simple plants like those occurring in arid regions today provide clues? To look for answers, in 2012 a research team composed of experts on algae and ancient environments explored diverse sites in the Mojave Desert of the US.

Professors Charles Wellman of Sheffield University in the UK and Wilson Taylor of the University of Wisconsin-Eau Claire study rare and hard-to-interpret microscopic fossils from terrestrial deposits that range from hundreds of millions to a billion years old. Thinking that some of those fossils might be ancient algae that had adapted to life on land, these paleobiologists teamed up with two algae experts. Prof. Louise Lewis (University of Connecticut) uses DNA sequences to decipher the evolutionary history of algae, focusing on species living or within sand and rocks of arid lands, and University of Wisconsin-Madison Prof. Linda Graham employs harsh chemical processes to discover which parts of modern algae might be able to survive post-mortem degradation long enough to become fossilized.





Team members Wilson Taylor and Charles Wellman provided expert commentary on stunning geological formations as the team drove from one Mojave site to the next.

Close-up view of biological crusts on gypsum-rich Mojave soil illustrates how bryophytes, algae, and other microorganisms hold together the otherwise loose soil.



During a previous expedition to the Australian outback, the team had found several kinds of algae in extremely arid regions. Surprisingly, certain common green algae had tough cell walls that survived harsh chemical treatment and closely resembled ancient fossils. So the team was anxious to learn how Mojave algae might compare.

Meeting in Riverside, CA, the team traveled by car for six days, stopping along the way to confer with local experts, check algae-containing biological soil crusts that are common in arid lands, and look for algae along the banks of desert streams and rivers. Algae were plainly visible in these and also some unexpected places, including the famously hot and arid Death Valley. The expedition proceeded smoothly until the final day, when a tire went flat in a remote area of Utah. But a consequent change in route took the team to the site of spectacular petroglyphs that they would otherwise have missed.

One amazing result from the expedition was the team's finding that desert stream Vaucheria, a filamentous member of the yellow-green (stramenopile) algae that also grows on soil, resisted rotting during several days of unrefrigerated transport. By contrast, other algae in the sample rotted, to the great disgust of travelers forced to sit next to the scientist whose backpack contained the smelly sample. Back in the lab, the cell walls of this alga also seemed unaltered by treatment with harsh chemicals and high temperatures. This result supported—though does not prove—a previous but contentious identification of a similar billion-year-old fossil. The team reported this and other results of their desert expeditions at the 2012 meetings of the Phycological Society of America and the Botanical Society of America. The team is planning other expeditions to environments extreme enough to serve as models of ancient Earth.





Growths of algae were visible along the muddy, organic-rich banks of the San Raphael River in Utah. Shortly after finding these algae, the team discovered a flat tire, forcing a precipitate return to civilization.

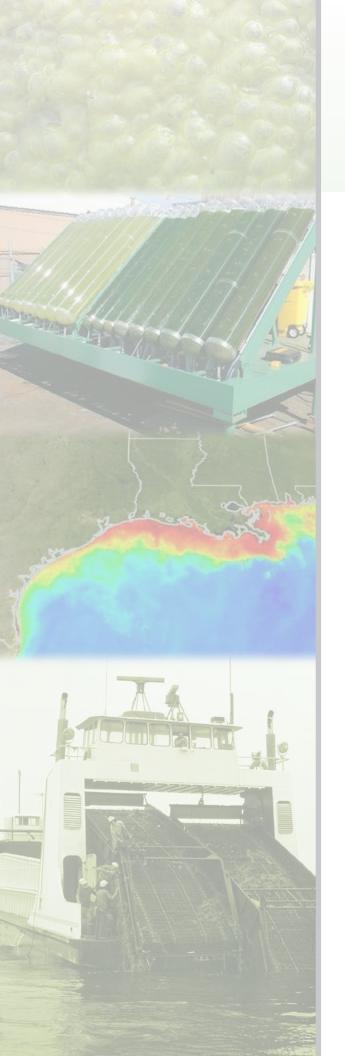




Not only did the team discover algae tough enough to fossilize, thereby aiding the interpretation of ancient fossils, their work also applies to major societal concerns. The expedition's results help to explain how Earth's earliest land photosynthesizers likely influenced their local habitats and how their modern descendants still do so today. For example, the erosion of soils in arid lands by wind and flooding constrains the ability of millions of modern humans to achieve a sustainable life. Yet the tough algae, lichens, bryophytes, and microbes present in arid lands fertilize the soil and help hold

it in place. So understanding how these organisms survive the stresses of their environments and provide key ecological services is valuable to mankind.

Further, because the past is often key to the present and future, understanding how ancient terrestrial photosynthesizers affected atmospheric chemistry and climate is fundamental to a complete understanding of modern climate change. Someday humans will explore other planets, seeking to understand when and how life takes hold. At that time, what we learn now about how life first conquered land on planet Earth will help to answer such cosmic questions.

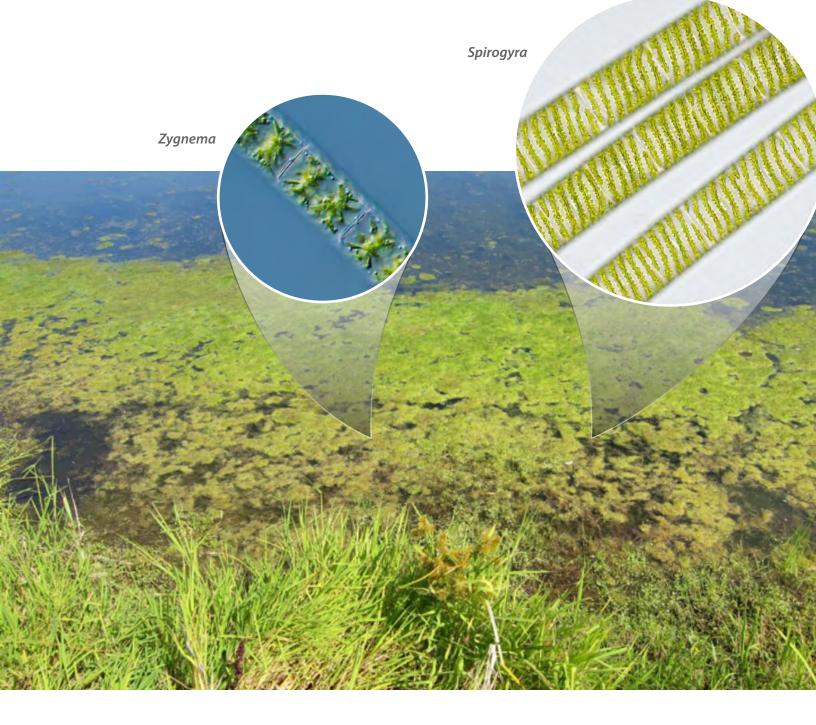


## Algae & Society



**Ponds** are popular features of municipal landscaping projects as well as gardens large and small. You'll see ponds everywhere, in cities as well as suburban and rural backyards. In many places, retention ponds are constructed as a way to prevent erosion, flooding, and harm to nearby lakes or streams. Whether ornamental or functional, ponds attract both people and wildlife.

Ponds contain diverse types of beautiful microscopic algae that are beneficial, and indeed essential to other pond life. That's because algal photosynthesis produces the oxygen and organic food needed by fish and other aquatic organisms, just as land plants generate oxygen and food humans and other terrestrial animals require. If the mineral fertilizer content of a pond is relatively low, you might never know those helpful algae are there.



But if too many mineral nutrients wash into ponds from recently fertilized fields or lawns, the algae will grow to conspicuous large populations called "algal blooms." In some cases the blooming algae are harmless to people and the environment, but in other cases algal blooms harm people, pets, livestock, and wildlife.

Examples of harmless pond algae are frothy leaf-green growths whose oxygen bubbles float them to the surface. These algae are *Spirogyra* and close relatives that are also closely related to land plants; they even reproduce using colorful structures that are the algal versions of flowers. You can verify their presence by touch because they feel slimy. Such frothy, leaf-green, slimy surface algae are not known to harm people or wildlife in any way. In fact, take a deep breath of the oxygen they so abundantly produce and appreciate their beauty.

A pond with harmless frothy, leaf-green, surface algae that are not only related to the grasses in the foreground, but should likewise be appreciated for their beauty and beneficial properties.



Microcystis

Unfortunately, many people don't appreciate the benefits and beauty of such harmless pond algae. Under the mistaken idea that blooms of any type are harmful in some way, people may be tempted to add chemical products to kill the algae. Such chemicals—known as algicides—work similarly to the commercial herbicides people might use to control weed growth in crop fields or lawns. Though using algicides may generate pleasingly clear waters at first, algicide use, which is expensive, can also worsen algal problems in the long run. Here's why:

When algicides kill algae, cell contents spill out into the water, adding to pond fertility. As soon as the algicides have been rendered ineffective by microbial action, algae will become abundant again, perhaps even more so, and the replacement species may not be so harmless.

Ponds that are rich in the mineral nutrients that boost algal populations have a greater tendency to contain blue-green algae, also known as cyanobacteria. Blue-green algal blooms, which give water a bluish-green or olive cast, are known to produce harmful toxins. When blue-green algae are abundant, so are the toxins they release into the water. These toxins can sicken people, but can be lethal to dogs, which are very sensitive. To be on the safe side, dogs

When abundant enough to make water murky, certain bluegreen algae (cyanobacteria) can injure the health of humans and other animals. The cells of these two toxin-producing cyanobacteria appear black rather than blue-green because the algal cells contain structures that help them float, but also refract light.



This urban retention pond, whose edges have been planted with attractive native flowering plants, does not harbor blooms of potentially toxic algae. The plants sponge up fertilizer that would otherwise run off nearby grassy slopes and into the pond. Because the fertilizer in run-off has been intercepted, it does not foster harmful algal blooms. Many types of algae occur in this pond, but they are of beneficial types that do not harm humans or wildlife.

should not be allowed to play in or drink water that has a conspicuous algal bloom.

Fortunately, easy steps can be taken to improve the appearance of ponds while also reducing the potential for toxic algal

blooms to develop. One attractive method is simply to plant native flowering plants around pond edges. These plants' roots will intercept fertilizers that would otherwise wash into the pond. The extra fertilizer boosts the production of attractive flowers that benefit butterflies and other wildlife. Since the plants take up the excess fertilizer, the pond water will not be over-fertilized, which makes algal blooms of any type less likely to form.

You say you want to get close to the water's edge? Build a walkway that allows you to walk through the planting to the water. Then you will be able to watch frogs, dragonflies and other wildlife that will be attracted to your edge-planted pond.

Other measures that will decrease the growth of algae in ponds include fertilizing nearby lawns or fields when rain is not predicted. This action increases the odds that grass or crops will take up most of the fertilizer before it can wash into the pond. It's better to use expensive fertilizers to boost grass or crop growth than to overfeed pond algae!

A microscopic view of algae from the edge-planted, urban retention pond shows harmless, leaf-green species that provide oxygen and other benefits to pond inhabitants. The radiating, flower-shape is Coleochaete, which is so closely related to plants that it is used as a laboratory research model for plants. The filament at right is Oedogonium. When this wiry alga grows too abundantly, there is no need for algicides, because Oedogonium can easily be removed from ponds by using a leaf rake.



## **Techniques: Imaging Algae**

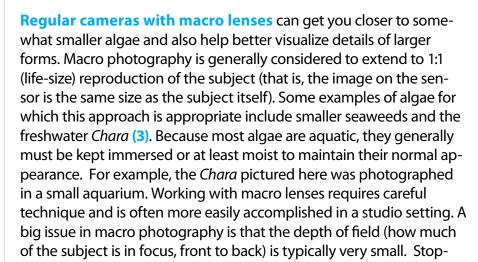
## Visualizing and photographing algae

Text and photographs by Lee W. Wilcox

Algae encompass an amazingly wide variety of growth forms that also cover an extremely large size range—from single-celled organisms barely visible with high-powered microscopes to multicellular marine seaweeds (kelps) that can reach 50 m in length. Accordingly, different approaches are necessary if you wish to view and photograph such disparately sized algae, but microscopes invariably prove useful in studying fine details of algae, even the largest species.

We'll take a brief look at some of the techniques used to examine and photograph algae, moving from larger to smaller scales.

Regular cameras with normal lenses are the best choice for photographing kelps and other large seaweeds. Smaller marine and freshwater algae that are barely, if at all, visible by themselves may form larger growths that are easily seen with the naked eye and photographed (1). Seaweeds often wash up on beaches and, if not yet dried or degraded, can make excellent photographic subjects (2). For photographing seaweeds underwater, watertight housings (often quite expensive) are available for any number of popular cameras. To photograph submersed seaweeds from above the surface, a polarizing filter is helpful in reducing reflections and glare.





Ulothrix, a green alga



Costaria, a kelp





The green alga *Coleochaete* growing on a rush stem



A research-grade stereoscope



A research-grade compound light microscope

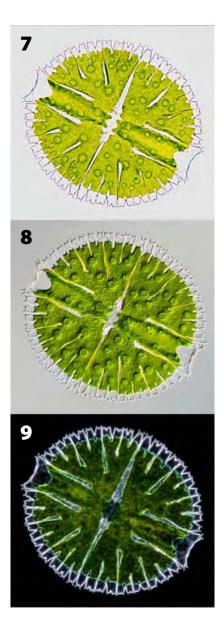
ping down the lens (using an f-stop with a bigger number) will gain you some depth of field but at the cost of somewhat reduced resolution. In recent years, software has become available that allows you to do "focus stacking," a technique that blends together a number of images shot with different planes of the specimen in focus. By combining the in-focus portions of the individual photos, a greater depth of field can be obtained in the final, composite image. This powerful technique, which also works well with microscopes, will be detailed more extensively in a later issue of this publication.

An alternative to traditional macro photography, but somewhat limited in terms of the types of algae that can be visualized, is to use a flatbed scanner. Care must be taken that specimens don't dry out or become deformed when placed on the scanner (leaving the top up, which yields a dark background, often works best). It should go without saying that electronic equipment and water don't mix well, so caution must also be exercised for that reason as well!

Dissecting microscopes or stereoscopes allow for a moderately large magnification of the subject (4), meaning that an image of the subject on the sensor is larger than its actual size. At this point, we enter the realm of "micro photography" or "photomicrography." Stereoscopes can be obtained at often modest cost through online auctions or surplus outlets. High-end research-grade instruments (5), on the other hand, easily cost several thousand dollars. Because separate optics are used for each eye, as with binoculars, you see subjects in stereo, which can provide striking views of even fairly small algae. Cameras (dSLRs and point and shoot models) can be fitted to one of the eyepieces or to a separate port on a "trinocular head." This is a more convenient arrangement, since you can easily switch between observation and photography. Most stereoscopes either have a series of discrete magnifications or a continuous range of magnifications—like a zoom lens on a camera.

An inexpensive alternative to traditional stereo microscopes has become available in recent years that connects directly to a computer (typically via a USB port). These instruments (essentially webcams with a built-in LED light source) are hard to beat for convenience and can do a decent job in many cases.

Compound light microscopes are probably what most people think of when the term "microscope" is mentioned (6). Compound microscopes can also be acquired for reasonably modest amounts but full-blown research models can range to many tens of thousands of dollars (or more), depending on features. Compound microscopes may have either one (monocular) or two (binocular) eyepieces or a trinocular head. Unlike stereoscopes, however, you



The single-celled green alga, *Micrasterias*, imaged with brightfield (the most basic approach) (7), differential interference contrast (8), and darkfield (9) optics. Note that different features are emphasized with each technique.

do not see the subject in stereo—the two viewing eyepieces each provide the same image—it's more comfortable for long-term viewing to use both eyes, rather than just one. In most cases, light passes through the specimen, which is then imaged with a combination of an "objective" lens, nearest the specimen, and the eyepiece ("compound" refers to such an arrangement). The most important consideration when purchasing a compound light microscope is the type of objective lenses that will be used. Not only are lenses available with different magnifications (the most common range from 4x to 100x), there are also different grades of objective lenses, which account for differences in resolution, contrast, color fidelity, and flatness of field (that is, whether objects at the edges of the field of view are in focus when those in the center are). Discussion of microscope lenses and microscope optics in general can be found on any number of websites, including <a href="http://www.microscopyu.com">http://www.microscopyu.com</a>.

Low-end objective lenses can be had for \$10s of dollars, whereas premium quality lenses range well into the thousands. Even relatively inexpensive lenses can still provide decent images and having a microscope with inexpensive lenses is better than not having one at all! It's generally the case that higher-quality lenses can be swapped in for a cheaper ones down the road and thus a microscope can be upgraded over time.

In a later article, further information on some of the different types of optical techniques—brightfield, darkfield, phase-contrast, fluorescence, etc.—that are employed with the compound light microscope will be provided along with some practical "how-to" advice in setting up a microscope to achieve the best results. Examples of three such techniques are shown in (7–9). Modern algal research often couples microscopy with molecular biology techniques such that, for example, DNA that contains particular genes can be localized to particular cells or regions within cells.

**Electron microscopes** are much more exotic instruments that use electrons rather than visible light to produce images of microscopic specimens. Some algae are so small that even the most powerful light microscopes reveal little useful detail. Electron microscopes can, however, resolve detail at a much finer scale. The scanning electron microscope (SEM) is typically used to examine surface details of specimens while the transmission electron microscope (TEM) is mostly used to examine internal structures of cells. Specimens are chemically treated to preserve their integrity, embedded in plastic resin, and cut into exceedingly thin sections, which permits the electrons to pass through, much like light passes through specimens on the compound light microscope. The cost and complexity of these instruments generally confines their use to large business, academic, and research settings.