

Evidence of Global Chlorophyll d

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We chemically detected chlorophyll d (Chl d) and its derivatives [e.g., pheophytin d (Phe d) and pyropheophytin d (PPhe d) (fig. S1)] directly from sedimentary samples, demonstrating that Chl d synthesis is widespread in oceanic and lacustrine environments covering a range of temperatures and salinities. Briefly, surface sediments were collected from (i) temperate coastal basins (Otsuchi Bay, Sagami Bay, Tokyo Bay, and Uchiura Bay, Japan), (ii) subarctic to Arctic pelagic basins (Bering Sea and Arctic Sea), (iii) Antarctic saline lakes (Lake Hunazoko and Lake Suribati), and (iv) a temperate freshwater lake (Lake Biwa, Japan) (tables S1 and S2). All samples contained pristine Chl d, Phe d, and

PPhe d, as well as more polar equivalents (possibly chlorophyllone d and its analogs) and a less polar equivalent with an undetermined structure (fig. S1) lacking pyrochlorophyll d (PChl d) (Fig. 1 and fig. S1). This composition of degraded products parallels that of other chlorophylls, including Chl a (Fig. 1 and fig. S1), and indicates that Chl d experiences similar chemical and biochemical regimes. Concentrations of Chl a, Phe a, Chl d, and Phe d were measured on the basis of absorption chromatograms at 660, 660, 702, and 696 nm (width = 2 nm), respectively (table S1).

Among these samples, concentrations of Chl d and Phe d relative to Chl a and Phe a, respectively, are high, reaching up to 4% (table S1). The relative

abundance of pigments in the surface sediment estimates the relative production rates between Chl a and Chl d [see supporting online material (SOM) text for methods and controls]. These production rates in turn are proportional to the rates of photosynthetic production of the respective chlorophylls because the quantum requirement of Chl d–based photosynthesis is equivalent to that of Chl a–based photosynthesis (1). Although Chl d derivatives can be hemisynthetically derived from Chl a derivatives in the laboratory (2), this conversion requires strong oxidants (e.g., NaIO₄) and is highly unlikely to occur in natural environments. Thus, these Chl d–related compounds appear to be derivatives produced by phototrophic organisms in the water column.

Our study extends the potential habitat of Chl d–producing phototrophs. Although *Acaryochloris marina* has only been reported from benthic habitats in littoral settings [e.g., (3–6)], our data indicate that Chl d–producing phototrophs also dwell offshore in hemipelagic to pelagic oceans, as well as in saline to freshwater lakes, and potentially thrive in most aquatic environments on Earth that receive near-infrared light.

Sediment core samples from Lake Biwa demonstrate marked variations in relative Chl d production over time (0.3 to 1.1%), whereas saline Antarctic lakes produced between 1.9 to 3.0% Chl d. These values are comparable to reported average Chl d abundances that were about 1% of Chl a in underwater seaweed beds (table S3). Therefore, a similar order of Chl d–based aquatic primary production is observable regardless of geographic and ecological setting. The presence of these organisms may be attributed either to *Acaryochloris*, with its metabolic adaptations to diverse conditions (7), or to unknown taxa producing Chl d. Regardless of what organism produces Chl d, Chl d–based photosynthesis needs to be properly re-evaluated in estimating global primary production.

References

1. S. Miyachi, K. Strassdat, H. Miyashita, H. Senger, *Z. Naturforsch. Teil C* **52**, 636 (1997).
2. T. Mizoguchi et al., *Photochem. Photobiol. Sci.* **5**, 291 (2006).
3. H. Miyashita et al., *Nature* **383**, 402 (1996).
4. A. Murakami, H. Miyashita, M. Iseki, K. Adachi, M. Mimuro, *Science* **303**, 1633 (2004).
5. M. Kuhl, M. Chen, P. J. Ralph, U. Schreiber, A. W. D. Larkum, *Nature* **433**, 820 (2005).
6. S. R. Miller et al., *Proc. Natl. Acad. Sci. U.S.A.* **102**, 850 (2005).
7. W. D. Swingle et al., *Proc. Natl. Acad. Sci. U.S.A.* **105**, 2005 (2008).

Supporting Online Material

www.sciencemag.org/cgi/content/full/321/5889/658/DC1

Materials and Methods

SOM Text

Figs. S1 to S4

Tables S1 to S3

References and Notes

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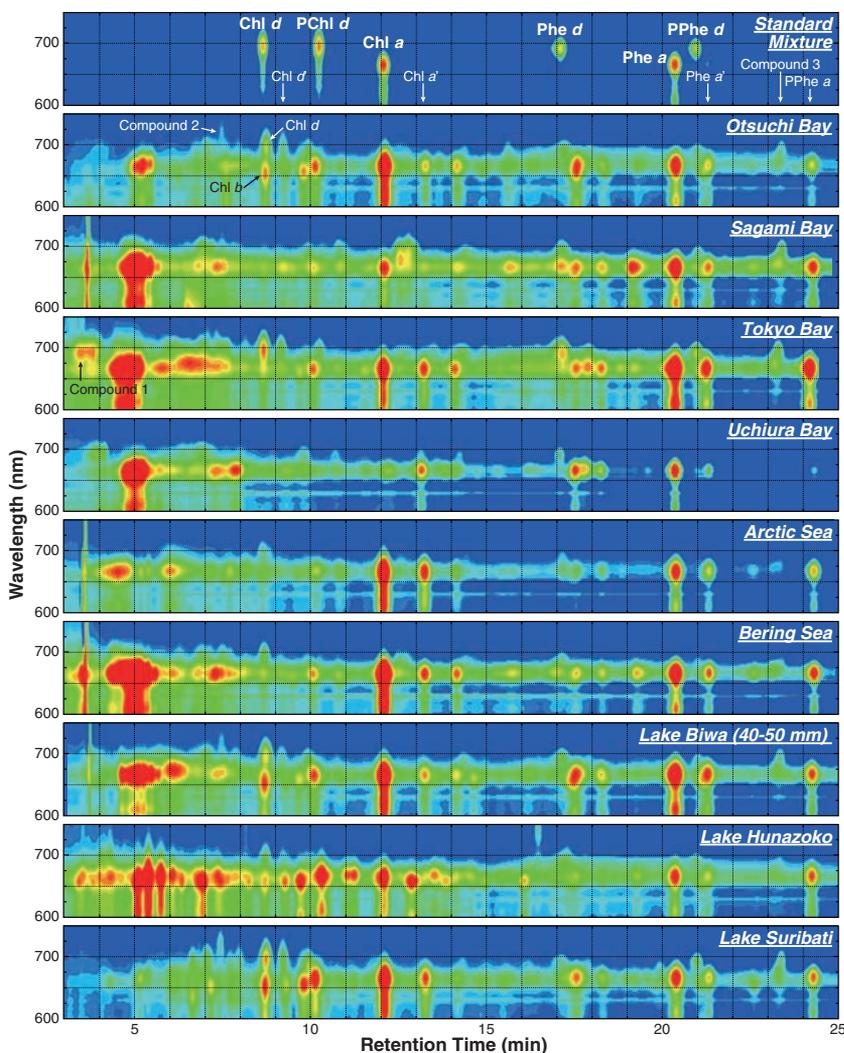


Fig. 1. Three-dimensional chromatograms of pigment analysis. Retention times and absorption of Chl a, Chl d, PChl d, Phe a, Phe d, and PPhe d and respective isomers Chl a', Chl d', and Phe a' are indicated by the chromatogram of the standard mixture. Retention times of other major pigments are also indicated. Note that Chl b and Chl d are co-eluted but are distinguished by absorption peaks. Compounds 1 to 3 have absorption maxima around 690 to 700 nm and might also be derivatives of Chl d. Compound 1 was tentatively assigned on the basis of its mass spectrum (fig. S2).