LIMNOLOGY and OCEANOGRAPHY: METHODS

Estimating pelagic primary production in lakes: Comparison of ^{14}C incubation and free-water O₂ approaches

Noah R. Lottig [ID],[1]* Joseph S. Phillips,[2] Ryan D. Batt,[3] Facundo Scordo [ID],[4,5] Tanner J. Williamson [ID],[6] Stephen R. Carpenter [ID],[1] Sudeep Chandra,[4] Paul C. Hanson [ID],[1] Chistopher T. Solomon [ID],[7] Michael J. Vanni [ID],[6] Jacob Zwart [ID][8]

[1]University of Wisconsin-Madison, Center for Limnology, Madison, Wisconsin
[2]Department of Aquaculture and Fish Biology, Hólar University, Hólar, Iceland
[3]Rutgers University, New Brunswick, New Jersey
[4]Global Water Center and the Department of Biology, University of Nevada, Reno, Reno, Nevada
[5]CONICET – Instituto Argentino de Oceanografía, Bahía Blanca, Buenos Aires, Argentina
[6]Department of Biology, Miami University, Oxford, Ohio
[7]Cary Institute of Ecosystem Studies, Millbrook, New York
[8]U.S. Geological Survey, Integrated Information Dissemination Division, South Bend, Indiana

Abstract

Historically, estimates of pelagic primary production in lake ecosystems were made by measuring the uptake of carbon-14 (^{14}C)-labeled inorganic carbon in samples incubated under laboratory or in situ conditions. However, incubation approaches are increasingly being replaced by methods that analyze diel changes in high-frequency in situ data such as free-water dissolved oxygen (O₂). While there is a rich literature on the comparison of approaches for estimating primary production using incubations (e.g., ^{14}C and O₂ bottle experiments), as well for approaches using high-frequency data (e.g., diel O₂ and CO₂ metabolism models), there are few direct comparisons of ^{14}C incubations and free-water O₂ approaches for estimating primary production. We used 20 lake-years of concurrent measurements of primary production quantified from high-frequency free-water O₂ data and ^{14}C incubations in four different lakes (4–7 years per lake) to compare these different approaches. Across all lakes, 61% of the ^{14}C production estimates were within the 95% credible intervals of the free-water O₂ production estimates. Error-in-variable regressions support the assumption that ^{14}C methods estimate a production value between gross primary production and net primary production and the bottle effect is constant across the entire range of production values considered here. There was little evidence that daily pelagic, epilimnetic estimates of primary production differed substantially based on the selection of free-water O₂ or ^{14}C approaches in these lakes during summer stratified conditions.

Primary production, the production of organic matter by autotrophs, is a fundamental ecosystem process and, combined with allochthonous inputs, determines the amount of energy available to higher trophic levels. In the pelagic regions of lakes and reservoirs, oxygenic primary production is typically determined using variants of two basic techniques: measuring the uptake of dissolved inorganic carbon or the production of dissolved oxygen (O₂) (Hall et al. 2007, but *see* Peeters et al. 2016, 2019 as examples of other approaches). These techniques can be applied either in situ or in laboratory incubations and can include measurements in bottles or open water.

Primary production estimates based on uptake of inorganic carbon usually involve labeling a sample of lake water with a known amount of inorganic carbon-14 (^{14}C) tracer, incubating the labeled sample under known temperature and light conditions (either in the lake or in the laboratory), and measuring the amount of labeled carbon that is fixed into organic forms by phytoplankton via photosynthesis during the incubation (Fee 1973; Peterson 1980). This ^{14}C technique has multiple advantages. It can be used in low production systems because the abundance of ^{14}C can be measured precisely (Hall et al. 2007). Samples can be incubated at multiple light levels allowing calculation of production vs. light relationships that can be used to estimate pelagic primary production at multiple depths in the lake. Repeated measurements within a year

*Correspondence: nrlottig@wisc.edu

Additional Supporting Information may be found in the online version of this article.

allow for the estimation of annual primary production and dynamics of primary production over time. For these reasons many research programs have compiled long time series of production measurements based on this technique.

The assumptions underlying the ^{14}C approach pose challenges for interpretation of the data and inferences drawn about ecosystems. First, this approach estimates production only of the water placed in the bottle under the environmental conditions in which it is incubated. Consequently, extrapolating production to a region of the lake or the whole ecosystem entails assumptions about the representativeness of those incubations for the region to which they are being extrapolated. Furthermore, because some carbon fixed during the incubation can be respired (Vollenweider et al. 1974; Peterson 1980), the technique is commonly presumed to underestimate pelagic gross primary production (GPP) and may be closer to net primary production (NPP). The magnitude of the underestimation is dependent on incubation time and algal turnover rates (Hall et al. 2007), although there are approaches to account for this underestimation (Legendre et al. 1983). In addition, the costs associated with specialized training and certifications to handle isotopes, management of radioactive waste, and the time-consuming nature of the incubations constrain the extent to which the methods are applied in most research programs. Nonetheless, this technique has been the standard by which all other approaches have been compared (Peterson 1980; Marra 2002), although free-water techniques are quickly becoming more popular (Staehr et al. 2010).

Over the past two to three decades, free-water O_2 techniques (Cole et al. 2000; Staehr et al. 2010; Vachon et al. 2020) have emerged as a common approach to estimate production of aquatic ecosystems because the data can be readily obtained at high frequencies using in situ sensors. While O_2 techniques have long been used to estimate production in aquatic systems (Sargent and Austin 1949; Odum 1956; Staehr et al. 2010), the advent of automated sensors capable of making in situ, high-frequency measurements of O_2 greatly reduced the labor associated with this technique along with providing opportunities to gather the data during difficult sampling conditions, such as storm events or ice breakup. Several models can be used to estimate metabolism from sensor data (Cole et al. 2000; Hanson et al. 2008; Batt and Carpenter 2012; Holtgrieve et al. 2013; Solomon et al. 2013; Phillips 2020). These approaches all assume that biological production and atmospheric exchange drive changes in oxygen (Odum 1956). A major advantage of the free-water O_2 approach is that it allows multiple components of metabolism (GPP, ecosystem respiration [R], and net ecosystem production [NEP]) to be estimated simultaneously. In addition, free-water O_2 metabolism estimates can integrate across habitats (e.g., benthic and pelagic production) when the sensor is located in a well-mixed parcel of water that is in contact with these habitats (Van de Bogert et al. 2007). Because of the relative ease of measurement using this technique, many

researchers and research groups, such as the Global Lake Ecological Observatory Network (Weathers et al. 2013), have adopted this technique (Solomon et al. 2013). However, estimates based on the free-water O_2 approach can be difficult to interpret because metabolic rates exhibit substantial vertical (Staehr et al. 2012*b*) and horizontal (Van de Bogert et al. 2012) heterogeneity within a lake, and movement of water parcels past the sensor can cause oxygen levels recorded by the sensor to change even in the absence of biological processes (Rose et al. 2014). Furthermore, along with spatial differences in processes, how models account for atmospheric gas exchange (Dugan et al. 2016) can lead to noisy high-frequency observations (Batt and Carpenter 2012) or to large and significant changes in estimated metabolism rates between days (Solomon et al. 2013). Therefore, heterogeneity complicates the interpretation of the results and potentially compromises their accuracy at short temporal scales.

As the free-water O_2 and other approaches based on high-frequency in situ data continue to gain popularity over the ^{14}C incubations, much is yet to be learned about how estimates of GPP compare between the two approaches. There is a long history of comparing ^{14}C incubations to O_2 production from light/dark bottle incubations to determine production levels in marine and aquatic environments (Williams et al. 1983; Bender et al. 1987; Gazeau et al. 2007). Similarly, studies in marine ecosystems have compared ^{14}C incubations to steady-state, sample-based oxygen methods such as ^{18}O labeling, triple-isotope, 17Δ, O_2/Ar, et al, and have generally found that the ^{14}C methods produce lower estimates (Juranek and Quay 2005; Quay et al. 2010; Hamme et al. 2012; Regaudie-de Gioux et al. 2014). To our knowledge, no direct comparison of the free-water O_2 and bottle ^{14}C methods across multiple lakes and years have been made (but *see* Lauster et al. 2006 for free-water and O_2 bottle comparisons).

Here, we use 20 lake-years of data from four lakes that differ in trophic status to assess how similar in situ free-water O_2 pelagic epilimnetic production estimates are to concurrent pelagic epilimnetic estimates made using ^{14}C incubations. Given the commonly presumed bias of ^{14}C to slightly underestimate GPP (Peterson 1980; Hall et al. 2007), we expect free-water O_2 approach to yield higher estimates than the ^{14}C approach, but they would have proportionally similar results (e.g., regression slope would be 1 between the two methods).

Materials and procedures

Study lakes

Daily lake ^{14}C pelagic production, high-frequency O_2, water temperature, and meteorological data were collected as part of ongoing long-term research projects in northern Wisconsin (North Temperate Lakes [NTL] Long-Term Ecological Research Program[1]; Trout and Sparkling Lakes), California (Castle Lake

---

[1]lter.limnology.wisc.edu

Table 1. Study lake characteristics. Nutrient and chlorophyll *a* values (standard deviation) are epilimnetic values averaged over the study period.

	Location (lat, lon)	Area (ha)	Mean Depth (m)	Total nitrogen (mg L^{-1})	Total phosphorus (mg L^{-1})	Chlorophyll *a* (μg L^{-1})
Trout	46.03, −89.67	1565.1	14.6	0.20 (0.07)	0.004 (0.004)	1.4 (1.3)
Sparkling	46.01, −89.70	63.7	10.9	0.21 (0.04)	0.005 (0.004)	1.2 (0.5)
Castle	41.23, −122.38	19	11.4	0.15 (0.09)	0.007 (0.003)	0.8 (0.2)
Acton	39.58, −84.76	232	3.9	2.70 (1.70)	0.102 (0.026)	70.0 (20.9)

Environmental Research and Education Program[2]; Castle Lake), and Ohio (Center for Aquatic & Watershed Sciences[3]; Acton Lake). Sparkling and Trout Lakes are embedded in a landscape that is predominantly a mix of deciduous and coniferous forest (54%), lakes (13%), and wetlands (28%; Magnuson et al. 2006). Both NTL study lakes are oligotrophic/mesotrophic with relatively low nutrient and chlorophyll concentrations (Table 1). Castle Lake is a meso-oligotrophic, subalpine (1646 m. a.s.l.) lake (Vander Zanden et al. 2006) located in northern California with similar nutrient and chlorophyll concentrations as Trout and Sparkling Lakes (Table 1). Acton Lake is a hypereutrophic reservoir (Table 1) that was created in 1957 by damming a creek for recreational use. Watershed landuse is primarily row crop agriculture (> 80%, Vanni et al. 2001).

Data analyzed in this study were all collected at the "deep hole" primary sampling site in each respective lake and were limited to observations collected in June, July, and August, which represent the time period that characterizes summer stratification, to ensure that both approaches were estimating pelagic primary production. As a consequence of restricting sampling to the summer stratified period, at no time during the study were any of the lakes mixed or iso-thermal.

^{14}C production methods

The approaches for estimating primary production in the study lakes using ^{14}C incubations differed slightly between the three research programs, but each estimated daily epilimnetic pelagic production (mmol C m^{-3} d^{-1}). In NTL lakes, integrated samples of water from the surface of the lake to the bottom of the epilimnion were collected between 2007 and 2013 using a 1.5-in. sampling tube approximately every 2 weeks during the open-water season (first described in these lakes by Adams et al. 1993). Samples were labeled with inorganic ^{14}C in the form of $NaHCO_3$ and then incubated in the laboratory for 3-h across a range of light intensities with additional dark bottles to correct for non-uptake sorption of ^{14}C at ambient epilimnetic water temperature. The resultant photosynthesis–irradiance (P–I) data were used to derive P–I

curves by fitting a three-parameter photosynthesis light-inhibition model (Platt et al. 1980) to these data. The P–I curves were coupled with concurrent, high-frequency photosynthetically active radiation (μmol m^{-2} s^{-1}; PAR) measurements and water column light extinction data (m^{-1}) to estimate daily primary production (mmol C m^{-3} d^{-1}) in both Sparkling and Trout Lake. Over this time period, the availability of data for ^{14}C production varied due to sporadic sample contamination and equipment failures. Light profiles were quantified in NTL lakes every 2 weeks using a LI-COR LI-192 light sensor to estimate light extinction coefficients, and PAR was continuously measured (1–10-min intervals) using a LI-COR LI-190 light sensor.

At Castle Lake, vertical water collections were made from 13 depths between the surface and 30 m; duplicate light samples and one dark bottle sample from each depth were labeled with inorganic ^{14}C in the form of $NaHCO_3$ and then incubated in situ at the depth of collection for 4 h. Detailed methods of ^{14}C production estimates are described in Goldman et al. (1963) and Goldman (1968). Total daily incident solar radiation was measured throughout the summer with a LI-COR Li-200 pyranometer. Light profiles at the height of the solar day were measured using a Biospherical Instruments 2104P radiometer. Daily phytoplankton productivity rates were calculated by scaling productivity measured during the incubation period by the fraction of the total daily PAR received during the incubation.

Methods for ^{14}C incubations in Acton Lake were similar to those in NTL lakes. Integrated samples were collected from the euphotic zone (usually equal to the epilimnion) and incubated in the laboratory for 1–2 h with $NaH^{14}CO_3$ at a range of light intensities (including dark bottles; Fee 1990). Incubations were usually done every 2 weeks (23 of 55 experiments over the 4 years) or more frequently (24 experiments); only eight experiments were done at intervals >2 weeks. As in NTL lakes, P–I curves were coupled with high-frequency PAR measurements, and water column light extinction data collected at weekly intervals. Light extinction coefficients were derived from weekly light profiles (Li-193) and surface PAR was measured continuously at an associated weather station on Acton Lake. Detailed methods for how ^{14}C production was estimated are described in Knoll et al. (2003).

---

[2]aquaticecosystemslab.org/projects/castlelake

[3]https://actonltreb.blog/

Free-water O$_2$ metabolism methods

The same approach was used to estimate pelagic primary production (mmol C m^{-3} d^{-1}; GPP) in all lakes using in situ time series of O$_2$ data. Free-water O$_2$ production estimates were based on high frequency measurements of O$_2$ (mg L^{-1}), water temperature (°C), PAR (μmol m^{-2} s^{-1}), wind speed (m s^{-1}), and barometric pressure (mbar). Data frequencies varied from 1 to 15 min based on the research program and the year data were collected. The raw, high-frequency time series of O$_2$ and water temperature were filtered to remove outliers by excluding values that were greater than 3 and 5 standard deviations, respectively, from a 7-d running average (Supporting Information Figure S11A,B; sensu Phillips 2020). The choice of sampling frequencies has implications for the processes influencing O$_2$ patterns and the amount of data needed to characterize those processes (Staehr et al. 2010). In general, frequencies between 30 min and 3 h are optimal for capturing changes driven by biological processes (Staehr et al. 2010). Thus, we extracted hourly time series (i.e., 1 measurement every 60 min) for all high frequency data by averaging observations (mean value) on the hour of observation ($n = 4$–60 depending on frequency of raw data) centered on the hour (Phillips 2020) for use in metabolism models.

Epilimnetic depth (m) was quantified from either high-frequency thermistor string data (Trout, Sparkling, and Acton Lakes) or discrete temperature profiles (Castle Lake). The high-frequency data were filtered for outliers as outlined above and epilimnetic depth determined using the rLakeAnalyzer package (Read et al. 2011; Winslow et al. 2019) at the temporal frequency of the raw data. Hourly aggregate data were then extracted based on a 1-d running average to reduce the significant amount of noise that existed in these estimates (Supporting Information Figure S1C). rLakeAnalyzer was also used to quantify epilimnetic depth from bi-monthly water temperature profile data in Castle Lake and linearly interpolated at hourly time steps between observations.

Exchange of dissolved gas with the atmosphere is a critical component of metabolism models, and, while there are several different models for estimating piston velocities in lentic ecosystems (Dugan et al. 2016), the model proposed by Vachon and Prairie (2013) is robust across multiple different types of lakes (Dugan et al. 2016). Piston velocities (m hr^{-1}) were calculated using the LakeMetabolizer R package (Winslow et al. 2016) and the parameterization proposed by Vachon and Prairie (2013). Light extinction coefficients (m^{-1}), which were typically quantified bi-monthly in all lakes, were linearly interpolated at hourly time steps between observations, and combined with epilimnetic depth and PAR to estimate the integrated light levels within the epilimnion of each lake for use in the metabolism model (Staehr et al. 2012a, Phillips (2020).

The data described above were used to generate daily estimates (mmol O$_2$ m^{-3} d^{-1}) of GPP, R, and NEP using a time-varying ecosystem metabolism model (Phillips 2020). This model differs from many of the more commonly used metabolism models (Winslow et al. 2016) in that the model is not fit iteratively over a daily time scale, but rather characterizes changes across all time periods (hourly measurements across 4–7 years of data) for a given lake in a single model fit, as well as constraining GPP and R to positive and negative values respectively (i.e., ecologically feasible ranges; Phillips 2020). This takes advantage of the fact that the physical and biological processes governing ecosystem metabolism and other aspects of DO dynamics are autocorrelated through time, which means that this shared information can be used to inform the parameter estimates across all time points. Furthermore, this method is statistically unified because it uses all data to fit a single model, which facilitates characterizing the uncertainty in the ecosystem metabolism estimates (Phillips 2020).

The equation used in this metabolism model to represent the relationship between light and GPP differs slightly from that presented in Phillips (2020) in that we used a photo-inhibition P–I curve (Steele 1962) to describe GPP (sensu Staehr et al. 2016) instead of a light saturating curve:

$$P_I = P_{\max} \frac{I}{I_{\mathrm{opt}}} \exp\left(1 - \frac{I}{I_{\mathrm{opt}}}\right),$$

where P_I is the production rate at light intensity I, $P_{\max}$ is the maximum production rate, and I_{opt} is the optimal light intensity. This photoinhibition model was chosen because recent work by Staehr et al. (2016) found that photoinhibition often occurred in lakes, and where photoinhibition does not occur, there appears to be little difference in metabolism model predictive performance among model formulations (Hanson et al. 2008). The model by Steele (1962) is one of the simplest photoinhibition models (two parameters), and regardless of the P–I curve formulation chosen, it is often difficult to distinguish significant differences in model fits between different models (Aalderink and Jovin 1997). Simple models are often better than their more complex counterparts (Peters et al. 2004; Downing 2009) and decreasing the number of parameters that needed to be fit by the Bayesian metabolism model increased the ease at which the model converged. Both $P_{\max}$ and I_{opt}, along with the model coefficient associated with R (see Phillips 2020), were allowed to vary through time at a daily time scale. The degree of auto-correlation in the parameters through time was constrained by hierarchical variance parameters in the random walk components of the model. Attempts to fit these parameters were unsuccessful, which is unsurprising as hierarchical variances often have poor identifiability. Thus, the random walk variances were treated as a "tuning parameters" and were selected manually such that the model converged while producing meaningful temporal smoothing in the parameters of the photoinhibition curve.

Observed O$_2$ time series were fit to all years (Trout Lake: 2007–2010, 2012; Sparkling Lake: 2007–2013; Castle Lake: 2014–2017; Acton Lake: 2010–2012, 2014) simultaneously for

each lake individually (i.e., lake-specific metabolism model fitting). Missing values in the model input data time series left some days with fewer than 24 observations. Although the metabolism model is robust to missing data because it fits the entire time series simultaneously instead of in discrete daily time steps, we did not estimate metabolism parameters for an individual day if more than 2 h of data were missing for that

day (Phillips 2020). The model was fit via Stan (Stan Development Team 2020) run in R (R Core Team 2020) using the rstan package as described in Phillips (2020). Posterior median values were used for daily GPP values along with the 0.025 and 0.975 quantiles of the posterior values to characterize the 95% credible intervals. Model fits were validated by checking effective sample size, $\widehat{R}$, tree depth, energy Bayesian Fraction
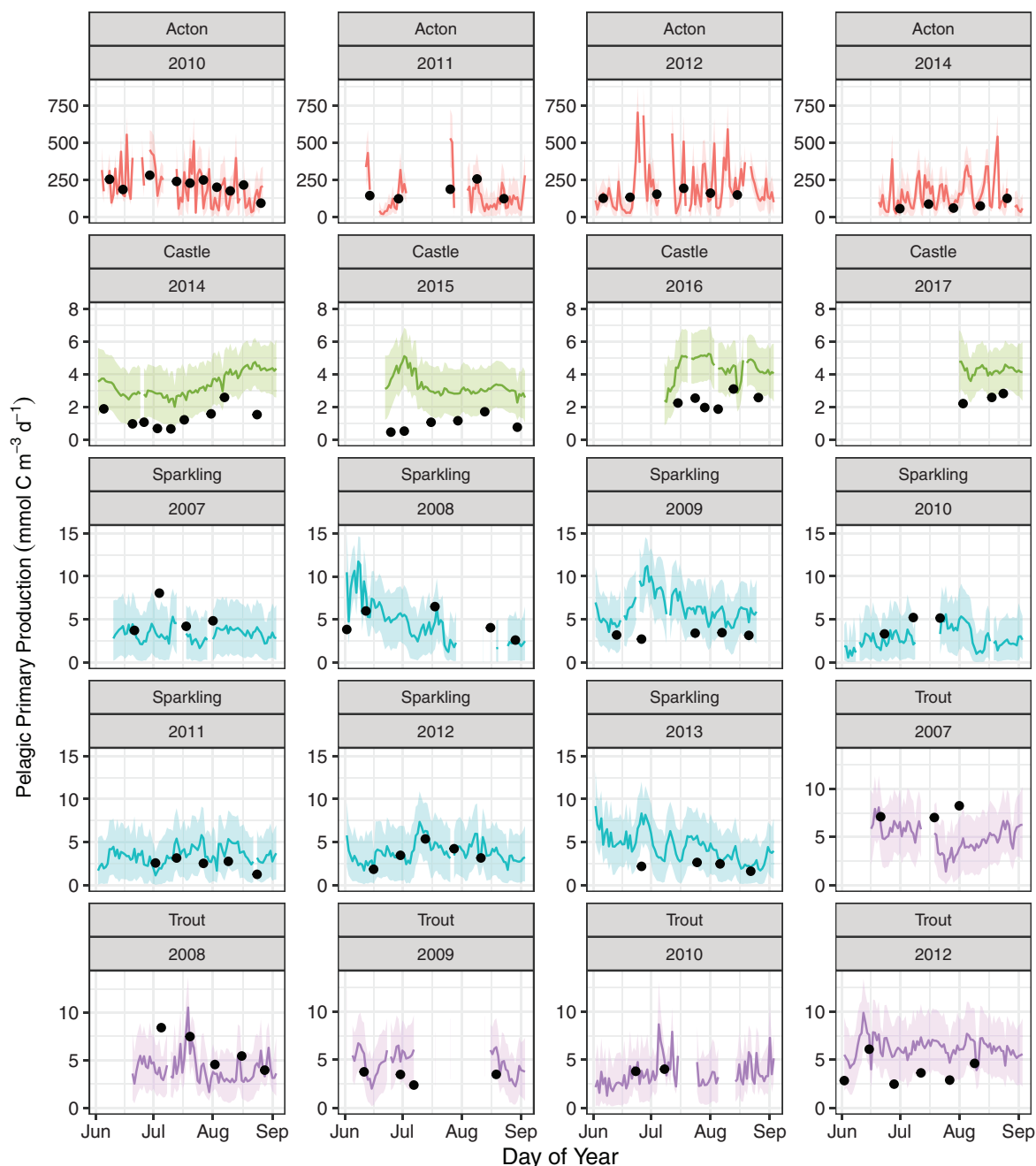

Fig. 1. Time series of pelagic epilimnetic primary production determined from high frequency in situ dissolved oxygen data (color) and discrete measurements of epilimnetic primary production determined from ^{14}C incubations (black) in four lakes that range in trophic status from ologitrophic to hypereutrophic. Light colored shaded areas represent the 95% credible interval of the free-water O_2 estimate.
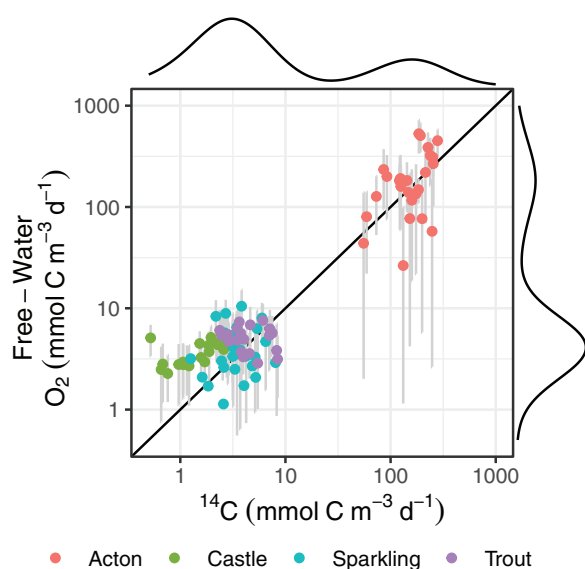
Fig. 2. Comparison of ^{14}C and free-water O_2 production (mmol C m^{-3} d^{-1}) estimates from concurrent observations in four lakes of varying trophic status. Error bars are 95% credible intervals from Bayesian metabolism model. Marginal plots are density distributions. Solid black line is line of equality (1:1).

of Missing Information, and divergence (*see* Betancourt 2007). Metabolism parameters were not estimated when the epilimnetic depth was shallower than the O_2 sensor (< 0.5 m in Trout, Sparkling, and Acton Lakes; < 3 m Castle Lake; 4% of all observations).

We recognize that prior research has identified a range in values of photosynthetic quotient for converting production estimates to units of carbon. The most direct evidence for photosynthetic quotient in the lakes analyzed as part of this study comes from prior research on NTL Lakes (Hanson et al. 2003). Given the differing scales (bottle vs. ecosystem) of ^{14}C vs. free-water O_2 measurements and the range of productivity observed in the study, we did not feel justified in estimating a value directly from the data used in this study. Thus, gross primary production values (mmol O_2 m^{-3} d^{-1}) were converted to units

of carbon (mmol C m^{-3} d^{-1}) assuming a photosynthetic quotient (O_2:CO_2) of 1.25 that was independently determined for NTL lakes (Hanson et al. 2003) and within the range (1.2–1.29) of other studies (Bott 1996; Hanson et al. 2003; Wielgat-Rychert et al. 2017).

Data and code (Lottig et al. 2021) associated with the analyses included in this manuscript are available for download through the Environmental Data Initiative (https://environmentaldata initiative.org).

Assessment

The goal of the analyses presented here is to compare ^{14}C and free-water O_2 daily primary production estimates to determine how interchangeable these two approaches are. We specifically tailor our analyses to identify two potential biases. First, given the commonly presumed bias of ^{14}C to slightly underestimate GPP (Peterson 1980; Hall et al. 2007), we wanted to know if there are constant differences in the magnitude of daily production values between the two approaches (i.e., we expected free-water O_2 approach to yield higher estimates than the ^{14}C approach). Second, we wanted to know if there were any fixed biases (i.e., intercept of linear regression different from zero) and/or proportional biases between the two methods (i.e., slope of linear regression different from 1). Our assumption was that free-water O_2 estimates of GPP would be slightly higher than ^{14}C (i.e., fixed bias) but the two methods were expected to yield proportionally similar results. If there were no significant fixed or proportional biases, we interpret the results to mean that the methods are interchangeable for the lakes considered in this study. Because both ^{14}C and O_2 estimates contain measurement errors (Macedo 2001; Pemberton et al. 2006; Solomon et al. 2013), we used robust error-in-variables regression (Passing and Bablok 1983) implemented in the mcr R package (Manuilova et al. 2014). Error-in-variable regression approaches, such as this, account for errors in both the Independent and Dependent variables instead of assuming that error only exists in the Dependent variable and the independent variable is known

Table 2. Error-in-variable regression results for all lakes as well as lakes separated by productivity class. Standard error (SE) and lower and upper 95% confidence intervals (CI) are also reported. Regressions using data from all lakes were log transformed prior to analysis.

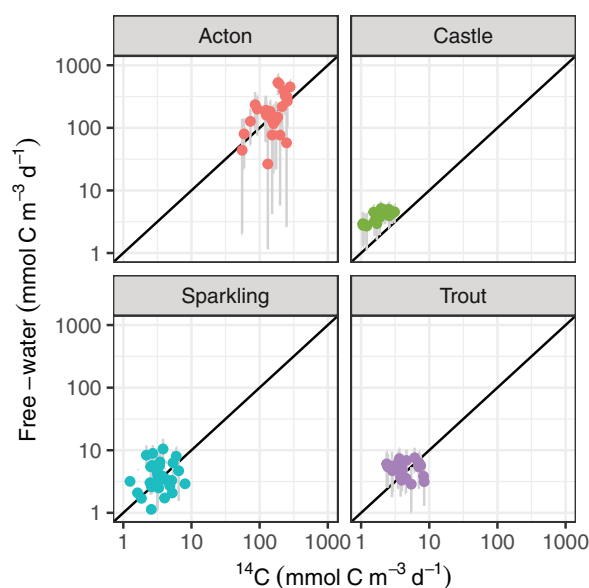|	Estimate	SE	Lower CI	Upper CI
All lakes				
Intercept	0.201	0.039	0.103	0.257
Slope	0.926	0.041	0.849	1.003
High productivity lake				
Intercept	−194.6	266.4	−765.8	68.14
Slope	2.445	1.691	0.987	6.215
Low productivity lakes				
Intercept	1.464	0.736	−0.071	2.311
Slope	1.019	0.276	0.636	1.559

Fig. 3. Discrete lake point estimate comparisons of ^{14}C and free-water O_2 production estimates from concurrent observations in four lakes of varying trophic status. Error bars are 95% credible intervals from Bayesian metabolism model. Solid black line is line of equality (1:1).

Table 3. Error-in-variable regression results for each individual lake. Standard error (SE) and lower and upper 95% confidence intervals (CI) are also reported.

	Estimate	**SE**	**Lower CI**	**Upper CI**
Acton Lake				
Intercept	−194.6	266.4	−765.8	68.14
Slope	2.445	1.691	0.987	6.215
Castle Lake				
Intercept	1.671	0.376	1.089	2.381
Slope	1.115	0.251	0.640	1.717
Trout Lake				
Intercept	8.157	3.260	5.689	16.66
Slope	−0.735	0.794	−2.928	−0.137
Sparkling Lake				
Intercept	4.846	9.591	−45.54	1.210
Slope	2.817	2.889	0.783	16.17

with certainty. Failure to account for errors in the independent variable can lead to regression dilution (i.e., bias slopes towards zero). Analyses that included all lakes across the entire productivity gradient were conducted on log10 transformed data, while the remaining regression analyses were conducted using non-transformed data.

Across the four lakes included in this study, we had 20 lake-years of concurrent ^{14}C and free-water O_2 pelagic, epilimnetic primary production estimates (Acton Lake: 4 yr, Castle Lake: 4 yr, Sparkling Lake: 7 yr, Trout Lake: 5 yr, Fig. 1). Direct comparisons between production estimates were available on 101 discrete days. In most cases (61% for all lakes; 75% excluding Castle Lake), ^{14}C estimates of production were contained within the 95% credible intervals of the free-water O_2 estimates and the seasonal patterns were similar between the two approaches (Fig. 1, but *see* Castle Lake).

A strong linear relationship was observed between the two approaches for estimating in-lake production across the approximately 200 mmol C m^{-3} d^{-1} pelagic epilimnetic production gradient observed in this study (Fig. 2). Error-in-variable regression using all discrete observations ($n = 101$) included the line of equality indicating across large gradients of pelagic epilimnetic production, there was no proportional difference between the two approaches for measuring production in the lakes examined here (Table 2). The slight rightward shift (Fig. 2, Table 2) in the distribution of free-water O_2 as well as an intercept significantly greater than 0 in the error-in-variables regression is consistent with the general assumption that ^{14}C production methods typically quantify a value lower than GPP (Peterson 1980, Hall

et al. 2007). Confidence intervals of nontransformed error-in-variable regression for Acton Lake (high productivity) and low productivity lakes included the line of equality (Table 2). While the residuals around the 1:1 line are distributed similarly for both the oligotrophic and hypereutrophic systems here (Fig. 2), we lacked data for mesotrophic systems and thus it is unknown if the same pattern would be observed in these systems as well.

The linear relationships between ^{14}C and free-water O_2 within lakes were not as strong as the relationships observed both across lakes and across wide gradients in pelagic epilimnetic production (Fig. 3, Table 3). For example, in Trout Lake there is a significant negative relationship as the credible intervals do not include zero (Table 3). The lack of a strong 1:1 linear relationship in lakes that have a limited range of ^{14}C estimates is likely due to the limited range of observed production values within a given lake combined with the uncertainty of both ^{14}C (not quantified) and free-water O_2 production estimates (quantified). Despite the narrow range of ^{14}C production observed in the different lakes, most of the points cluster around the 1:1 line and a majority (61%; 75% excluding Castle Lake) of the 95% credible intervals of the free-water O_2 estimates intersect the 1:1 line. We note that Castle Lake is unique among lakes in this study in that the data indicate a consistently lower production (about 1.7 mmol C m^{-3} d^{-1} offset) value estimated with the ^{14}C approach relative to the free-water O_2 approach (Table 3). We believe there are a few potential reasons for this including unique methodological considerations for this long-term dataset (*see* Discussion section).

Discussion

Our results indicate that the pelagic, epilimnetic ^{14}C and free-water O_2 production approaches examined here can be

interpreted similarly for the lakes considered in this study during summer stratified conditions. Across gradients in production from oligotrophic to hypereutrophric systems, both of these approaches provide production estimates that are very similar in magnitude. Comparison of results between both methods indicated no statistically significant deviation from the 1:1 relationship, although there is evidence that, as expected, ^{14}C estimates may be slightly lower than free-water O_2 estimates of pelagic epilimnetic production.

A priori, we anticipated that free-water O_2 estimates would be proportional to ^{14}C estimates and that ^{14}C estimates would be, on average, slightly lower than free-water O_2, because ^{14}C estimates tend to lie between GPP and NPP (Peterson 1980). In general, the results confirmed our expectations. The lack of strong statistical evidence when comparing the relationship across a wide gradient in of productivity (i.e., all lakes in the study) of lower ^{14}C relative to O_2 estimates in our study may reflect the considerable uncertainty in both estimates, which we account for in our analysis via the error-in-variables approach. In addition, the research programs responsible for generating the ^{14}C production estimates specifically targeted short incubation periods to generate estimates that closely approximated GPP (Hall et al. 2007). Thus, even though we observed slightly lower ^{14}C production estimates relative to the free-water O_2 estimates, the lack of strong statistical significance across all analyses is not necessarily surprising given approaches used by the research programs collecting the ^{14}C production data.

The specific results for Castle Lake are an exception to the conclusions based on results drawn from the full dataset spanning all lakes. There is strong evidence that ^{14}C estimates are significantly lower than free-water O_2 estimates for this lake alone, even though the two approaches are proportionally similar. We consider two potential reasons for this pattern in Castle Lake. First, the degree to which ^{14}C production estimates approximate GPP relative to NPP is influenced by incubation time (Hall et al. 2007), whereby shorter incubations tend to estimate a value closer to GPP, represented in this study as free-water O_2 estimates. Castle Lakes incubations were the longest ($\sim$ 4 h) of any of the three programs that collected ^{14}C data, and thus it might be expected that the relative difference between the approaches was greatest for this program compared to the other two programs that collect ^{14}C data. The other potential issue relates to how the ^{14}C data from Castle Lake were generated (*see* Methods above). Briefly, samples were incubated in situ for 4 h from 10:00 to 14:00 h (time period of maximum solar insolation), and the relationship between production and solar insolation (i.e., P–I curve) was assumed to be linear; whereas, a relationship that includes photoinhibition is likely more accurate (Huovinen 1999). Because the incubations were conducted when solar insolation was near maximal and not across a range of light intensities (*see* Lizon and Lagadeuc 1998), this approach has the potential to substantially underestimate production rates at lower light

levels regardless of the shape of the true P–I curve. Nearly, all of the lower ^{14}C epilimnetic production estimates in Castle Lake were in samples that were incubated directly at the lake's surface where solar insolation is much greater relative to insulation deeper in the water column. Samples incubated at deeper depths had higher production estimates and would be consistent with P–I curves characterized by strong photoinhibition. Thus, while not influencing the overall patterns across all lakes, Castle Lake serves as a reminder that it will be important to account for potential differences in how both ^{14}C and free-water O_2 production data are generated and understand the limitations of the methods employed for either approach.

While we suggest that free-water O_2 and ^{14}C epilimnetic daily pelagic production approaches are largely interchangeable when comparing across large gradients in productivity, it is important to emphasize that there is still substantial variability between the methods. It can be difficult to reliably fit free-water metabolism models in some systems, especially low productivity systems, because physical processes influencing O_2 often dominate the O_2 temporal patterns and/or errors in accounting for these physical processes influencing O_2 result in unrepresentative results. While physical processes are less of a concern for incubations, there is a suite of other concerns (Hall et al. 2007). At the hypereutrophic end of the productivity spectrum, high variability in daily free-water O_2 estimates are common (Williamson et al. 2020), and bottle incubations for ^{14}C production may miss important temporal and/or spatial variability that is captured by the free-water O_2 approach. The variability in free-water O_2 estimates also has implications for within lake comparisons between methods when historical ^{14}C estimates are characterized by a very constrained range of values. In many cases in this study, the free-water O_2 credible intervals were equal to the range of ^{14}C estimates observed within a single lake. Given the lack of explicit uncertainty estimates associated with up scaling ^{14}C estimates to the ecosystem scale along with the sensitivity of the method (Marra 2002), it is unclear if small differences between methods within a lake are real or an artifact of the scale at which the estimates were generated (i.e., ecosystem vs. bottle). Nonetheless, it is clear that the two approaches yield close agreement across a wide range of productivity values.

Estimating metabolism parameters, including primary production from free-water O_2 data can be challenging in low productivity systems (McNair et al. 2015; Richardson et al. 2017; Honti and Istvánovics 2019) where ^{14}C is generally considered optimal (Hall et al. 2007). Given that a majority of the lakes in this study are characterized by low productivity, it is likely that the ability of the Phillips (2020) model to leverage all possible data across multiple years to fit the models contributed to strong agreement between the free-water O_2 and ^{14}C estimates. While not the focus of this study, an exploration of how different free-water O_2 models perform may lead

to a better understanding of when and where different model formulations could be leveraged (McNair et al. 2015; Honti et al. 2016; Staehr et al. 2016). Similarly, most ^{14}C production estimates that we are aware of in freshwater systems do not propagate uncertainty in the production estimates that are scaled to the ecosystem scale. Thus, while we can account for uncertainty in free-water O_2 estimates, it is unclear how uncertainty in free-water O_2 estimates compare to ^{14}C and how quantifying uncertainty in ^{14}C estimates may alter our understanding about the relationships between these two approaches of estimating pelagic primary production in lake ecosystems.

Multisite comparisons like this study are critical for gaining a better understanding of lake daily production measurements generated by these two widely used methods, and to help guide limnologists on which methods to use and how to interpret estimates. Each method has unique advantages and disadvantages that may influence the choice of methods for particular research applications. For example, we would expect large differences between free-water and bottle estimates in lakes where littoral and benthic production contribute substantially to total metabolism, such as in shallow lakes, lakes with small surface areas, lakes with large surface area to volume ratios, or during time periods when lakes are not stratified (Lauster et al. 2006; Van de Bogert et al. 2007). Thus, it is likely that both methods will continue to be used and there will be an ongoing need to compare results across methods, during different seasons, and across a variety of different lake types. Analyses conducted here provide little evidence of systematic differences in estimates of epilimnetic, pelagic lake daily production based on free-water O_2 or ^{14}C methods across a wide gradient in lake trophic status.

References

Aalderink, R., and R. Jovin. 1997. Estimation of the photosynthesis/irradiance (P/I) curve parameters from light and dark bottle experiments. J. Plankton Res. **19**: 1713–1742. doi:10.1093/plankt/19.11.1713

Adams, M. S., T. W. Meinke, and T. K. Kratz. 1993. Primary productivity of three Wisconsin LTER lakes, 1985–1990. SIL Proc. **1922-2010**: 406–410. doi:10.1080/03680770.1992.11900149

Batt, R. D., and S. R. Carpenter. 2012. Free-water lake metabolism: Addressing noisy time series with a Kalman filter. Limnol. Oceanogr. Methods **10**: 20–30. doi:10.4319/lom.2012.10.20

Bender, M., and others. 1987. A comparison of four methods for determining planktonic community production. Limnol. Oceanogr. **32**: 1085–1098. doi:10.4319/lo.1987.32.5.1085

Betancourt, M. 2007. Robust statistical workflow with RStan. https://betanalpha.github.io/assets/case_studies/rstan_workflow.html.

Bott, T. 1996. Primary productivity and community respiration, p. 533–556. *In* Methods in stream ecology, 2nd ed. Academic Press.

Cole, J. J., M. L. Pace, S. R. Carpenter, and J. F. Kitchell. 2000. Persistence of net heterotrophy in lakes during nutrient addition and food web manipulations. Limnol. Oceanogr. **45**: 1718–1730. doi:10.4319/lo.2000.45.8.1718

Downing, J. A. 2009. Global limnology: Up-scaling aquatic services and processes to planet earth. Verh. Int. Verein. Limnol. **30**: 1149–1166. doi:10.1080/03680770.2009.11923903

Dugan, H. A., and others. 2016. Consequences of gas flux model choice on the interpretation of metabolic balance across 15 lakes. Inland Waters **6**: 581–592. doi:10.1080/IW-6.4.836

Fee, E. J. 1990. Computer programs for calculating in situ phytoplankton photosynthesis. Canadian technical report of fisheries and aquatic sciences. Technical Report of Fisheries and Aquatic Science, No. 1740. Department of Fisheries and Oceans Winnipeg, Manitoba.

Fee, E. J. 1973. A numerical model for determining integral primary production and its application to Lake Michigan. J. Fish. Res. Board Can. **30**: 1447–1468. doi:10.1139/f73-235

Gazeau, F., J. Middelburg, M. Loijens, J. Vanderborght, M. Pizay, and J. Gattuso. 2007. Planktonic primary production in estuaries: Comparison of ^{14}C, O_2 and ^{18}O methods. Aquat. Microb. Ecol. **46**: 95–106. doi:10.3354/ame046095

Goldman, C. R. 1968. The use of absolute activity for eliminating serious errors in the measurement of primary productivity with C^{14}. ICES J. Mar. Sci. **32**: 172–179. doi:10.1093/icesjms/32.2.172

Goldman, C. R., D. T. Mason, and B. J. B. Wood. 1963. Light injury and inhibition in Antarctic freshwater phytoplankton. Limnol. Oceanogr. **8**: 313–322. doi:10.4319/lo.1963.8.3.0313

Hall, R., S. Thomas, and E. Gaiser. 2007. Measuring freshwater primary production and respiration, p. 175–203. *In* T. Fahey and A. Knapp [eds.], Principals and standards for measuring primary production. Oxford Univ. Press.

Hamme, R. C., and others. 2012. Dissolved O_2/Ar and other methods reveal rapid changes in productivity during a lagrangian experiment in the Southern Ocean. J. Geophys. Res. Oceans. **C00F12**: 117. doi:10.1029/2011JC007046

Hanson, P. C., D. L. Bade, S. R. Carpenter, and T. K. Kratz. 2003. Lake metabolism: Relationships with dissolved organic carbon and phosphorus. Limnol. Oceanogr. **48**: 1112–1119. doi:10.4319/lo.2003.48.3.1112

Hanson, P. C., S. R. Carpenter, N. Kimura, C. Wu, S. P. Cornelius, and T. K. Kratz. 2008. Evaluation of metabolism models for free-water dissolved oxygen methods in lakes. Limnol. Oceanogr. Methods **6**: 454–465. doi:10.4319/lom.2008.6.454

Holtgrieve, G. W., and others. 2013. Patterns of ecosystem metabolism in the Tonle Sap Lake, Cambodia with links to

capture fisheries. PLoS One **8**: e71395. doi:10.1371/journal.
pone.0071395

Honti, M., V. Istvánovics, P. A. Staehr, L. S. Brighenti, M. Zhu,
and G. Zhu. 2016. Robust estimation of lake metabolism by
coupling high frequency dissolved oxygen and chlorophyll
fluorescence data in a Bayesian framework. Inland Waters
6: 608–621. doi:10.1080/IW-6.4.877

Honti, M., and V. Istvánovics. 2019. Error propagation during
inverse modeling leads to spurious correlations and misin-
terpretation of lake metabolism. Limnol. Oceanogr.:
Methods **17**: 17–24. doi:10.1002/lom3.10293

Huovinen, P. 1999. Temporal and vertical dynamics of phyto-
plankton net growth in Castle Lake. Calif. J. Plankton Res.
21: 373–385. doi:10.1093/plankt/21.2.373

Juranek, L. W., and P. D. Quay. 2005. In vitro and in situ gross
primary and net community production in the north
pacific subtropical gyre using labeled and natural abun-
dance isotopes of dissolved O_2. Global Biogeochem. Cycles
19: GB3009. 10.1029/2004GB002384

Knoll, L. B., M. J. Vanni, and W. H. Renwick. 2003. Phyto-
plankton primary production and photosynthetic parame-
ters in reservoirs along a gradient of watershed land use.
Limnol. Oceanogr. **48**: 608–617. doi:10.4319/lo.2003.48.2.
0608

Lauster, G. H., P. C. Hanson, and T. K. Kratz. 2006. Gross pri-
mary production and respiration differences among littoral
and pelagic habitats in northern Wisconsin lakes. Can.
J. Fish. Aquat. Sci. **63**: 1130–1141. doi:10.1139/f06-018

Legendre, L., S. Demers, C. M. Yentsch, and C. S. Yentsch.
1983. The ^{14}C method: Patterns of dark CO_2 fixation and
DCMU correction to replace the dark bottle. Limnol.
Oceanogr. **28**: 996–1003. doi:10.4319/lo.1983.28.5.0996

Lizon, F., and Y. Lagadeuc. 1998. Comparisons of primary pro-
duction values estimated from different incubation times
in a coastal sea. J. Plankton Res. **20**: 371–381. doi:10.1093/
plankt/20.2.371

Lottig, N. R., S. Chandra, E. H. Stanley, M. Vanni, F. Scordo &
W. Tanner 2021. Pelagic, epilimnetic production estimates
in Sparkling, Trout (Wisconsin), Acton (Ohio), and Castle
(California) lakes (USA) calculated using 14C and free-water
O2 metabolism methods, 2007–2017 ver 1. Environmental
Data Initiative. doi:10.6073/pasta/14a201367db1429e8acbe
b7e5a7341b8

Macedo, M. F. 2001. Annual variation of environmental vari-
ables, phytoplankton species composition and photosyn-
thetic parameters in a coastal lagoon. J. Plankton Res. **23**:
719–732. doi:10.1093/plankt/23.7.719

Magnuson, J. J., T. K. Kratz, and B. J. Benson [eds.]. 2006.
Long-term dynamics of lakes in the landscape: Long-term
ecological research on North Temperate Lakes. Long-Term
Ecological Research Network Series. Oxford Univ. Press.

Manuilova, E., Schuetzenmeister, A., and Model, F. 2014. mcr:
Method comparison regression. https://CRAN.R-project.
org/package=mcr.

Marra, J. 2002. Approaches to the measurement of plankton
production. Blackwell Science, p. 78–108.

McNair, J. N., M. R. Sesselmann, S. T. Kendall, L. C. Gereaux,
A. D. Weinke, and B. A. Biddanda. 2015. Alternative
approaches for estimating components of lake metabolism
using the free-water dissolved-oxygen (FWDO) method.
Fundam. Appl. Limnol. **186**: 21–44. doi:10.1127/fal/2015/
0626

Odum, H. T. 1956. Primary production in flowing waters.
Limnol. Oceanogr. **1**: 102–117. doi:10.4319/lo.1956.1.2.
0102

Passing, H., and W. Bablok. 1983. A new biometrical procedure
for testing the equality of measurements from two different
analytical methods. Application of linear regression proce-
dures for method comparison studies in clinical chemistry,
Part I. J. Cin. Chem. Clin. Biochem. **21**: 709–720. doi:10.
1515/cclm.1983.21.11.709

Peeters, F., D. Atamanchuk, A. Tengberg, J. Encinas-Fernández,
and H. Hofmann. 2016. Lake metabolism: Comparison of
lake metabolic rates estimated from a diel CO_2- and the
common diel O_2-technique. PLoS One **11**: e0168393. doi:
10.1371/journal.pone.0168393

Peeters, F., H. Hofmann, and J. E. Fernández. 2019. On the cal-
culation of lake metabolic rates: Diel O_2 and 18/16O tech-
nique. Water Res. **165**: 114990. doi:10.1016/j.watres.2019.
114990

Pemberton, K., K. Clarke, and I. Joint. 2006. Quantifying
uncertainties associated with the measurement of primary
production. Mar. Ecol. Prog. Ser. **322**: 51–59. doi:10.3354/
meps322051

Peters, D. P. C., J. E. Herrick, D. L. Urban, R. H. Gardner, and
D. D. Breshears. 2004. Strategies for ecological extrapola-
tion. Oikos **106**: 627–636. doi:10.1111/j.0030-1299.2004.
12869.x

Peterson, B. J. 1980. Aquatic primary productivity and the
^{14}C-CO_2 method: A history of the productivity problem.
Annu. Rev. Ecol. Syst. **11**: 359–385. doi:10.1146/annurev.
es.11.110180.002043

Phillips, J. S. 2020. Time-varying responses of lake metabolism
to light and temperature. Limnol. Oceanogr. **65**: 652–666.
doi:10.1002/lno.11333

Platt, T., C. Gallegos, and W. Harrison. 1980. Photoinhibition
of photosynthesis in natural assemblages of marine phyto-
plankton. J. Mar. Res. **38**: 687–701.

Quay, P. D., C. Peacock, K. Björkman, and D. M. Karl. 2010.
Measuring primary production rates in the ocean: Enig-
matic results between incubation and non-incubation
methods at Station ALOHA. Global Biogeochem. Cycl. **24**:
GB3014. doi:10.1029/2009GB003665

R Core Team. 2020. R: A language and environment for statis-
tical computing. R Foundation for Statistical Computing.
https://www.R-project.org/.

Read, J. S., D. P. Hamilton, I. D. Jones, K. Muraoka, L. A.
Winslow, R. Kroiss, C. H. Wu, and E. Gaiser. 2011.

Derivation of lake mixing and stratification indices from high-resolution lake buoy data. Environ. Model. Software **26**: 1325–1336. doi:10.1016/j.envsoft.2011.05.006

Regaudie-de Gioux, A., S. Lasternas, S. Agustí, and C. M. Duarte. 2014. Comparing marine primary production estimates through different methods and development of conversion equations. Front. Mar. Sci. **1**: 19. doi:10.3389/fmars.2014.00019

Richardson, D. C., C. C. Carey, D. A. Bruesewitz, and K. C. Weathers. 2017. Intra- and inter-annual variability in metabolism in an oligotrophic lake. Aquat. Sci. **79**: 319–333. doi:10.1007/s00027-016-0499-7

Rose, K. C., L. A. Winslow, J. S. Read, E. K. Read, C. T. Solomon, R. Adrian, and P. C. Hanson. 2014. Improving the precision of lake ecosystem metabolism estimates by identifying predictors of model uncertainty: Lake ecosystem metabolism uncertainty. Limnol. Oceanogr. Methods **12**: 303–312.

Sargent, M. C., and T. S. Austin. 1949. Organic productivity of an atoll. Trans. Am. Geophys. Union **30**: 245. doi:245. 10.1029/TR030i002p00245

Solomon, C. T., and others. 2013. Ecosystem respiration: Drivers of daily variability and background respiration in lakes around the globe. Limnol. Oceanogr. **58**: 849–866. doi:10.4319/lo.2013.58.3.0849

Staehr, P. A., L. Baastrup-Spohr, K. Sand-Jensen, and C. Stedmon. 2012a. Lake metabolism scales with lake morphometry and catchment conditions. Aquat. Sci. **74**: 155–169. doi:10.1007/s00027-011-0207-6

Staehr, P. A., D. Bade, M. C. Van de Bogert, G. R. Koch, C. Williamson, P. Hanson, J. J. Cole, and T. Kratz. 2010. Lake metabolism and the diel oxygen technique: State of the science. Limnol. Oceanogr. Methods **8**: 628–644. doi:10.4319/lom.2010.8.0628

Staehr, P. A., L. S. Brighenti, M. Honti, J. Christensen, and K. C. Rose. 2016. Global patterns of light saturation and photoinhibition of lake primary production. Inland Waters **6**: 593–607. doi:10.1080/IW-6.4.888

Staehr, P. A., J. P. A. Christensen, R. D. Batt, and J. S. Read. 2012b. Ecosystem metabolism in a stratified lake. Limnol. Oceanogr. **57**: 1317–1330. doi:10.4319/lo.2012.57.5.1317

Stan Development Team. 2020. RStan: The R interface to Stan. R package version 2.21.2, http://mc-stan.org.

Steele, J. H. 1962. Environmental control of photosynthesis in the sea. Limnol. Oceanogr. **7**: 137–150. doi:10.4319/lo.1962.7.2.0137

Vachon, D., and others. 2020. Paired O_2–CO_2 measurements provide emergent insights into aquatic ecosystem function. Limnol. Oceanogr. Lett. **5**: 287–294. doi:10.1002/lol2.10135

Vachon, D., and Y. T. Prairie. 2013. The ecosystem size and shape dependence of gas transfer velocity versus wind speed relationships in lakes. Can. J. Fish. Aquat. Sci. **70**: 1757–1764. doi:10.1139/cjfas-2013-0241

Van de Bogert, M. C., D. L. Bade, S. R. Carpenter, J. J. Cole, M. L. Pace, P. C. Hanson, and O. C. Langman. 2012. Spatial heterogeneity strongly affects estimates of ecosystem metabolism in two north temperate lakes. Limnol. Oceanogr. **57**: 1689–1700. doi:10.4319/lo.2012.57.6.1689

Van de Bogert, M. C., S. R. Carpenter, J. J. Cole, and M. L. Pace. 2007. Assessing pelagic and benthic metabolism using free water measurements: Benthic and pelagic metabolism. Limnol. Oceanogr. Methods **5**: 145–155. doi:10.4319/lom.2007.5.145

Vander Zanden, M. J., S. Chandra, S. K. Park, Y. Vadeboncoeur, and C. R. Goldman. 2006. Efficiencies of benthic and pelagic trophic pathways in a subalpine lake. Can. J. Fish. Aquat. Sci. **63**: 2608–2620. doi:10.1139/f06-148

Vanni, M. J., W. H. Renwick, J. L. Headworth, J. D. Auch, and M. H. Schaus. 2001. Dissolved and particulate nutrient flux from three adjacent agricultural watersheds: A five-year study. Biogeochemistry **54**: 85–114. doi:10.1023/A:1010681229460

Vollenweider, R. A., J. F. Talling, and D. F. Westlake. 1974. A manual on methods for measuring primary production in aquatic environments. IBP handbook, v. **12**, 2nd Edition. Blackwell Scientific.

Weathers, K. C., and others. 2013. The global lake ecological observatory network (GLEON): The evolution of grassroots network science. Limnol. Oceanogr. Bull. **22**: 71–73. doi:10.1002/lob.201322371

Wielgat-Rychert, M., K. Rychert, Z. Witek, and M. Zalewski. 2017. Calculation of the photosynthetic quotient (PQ) in the Gulf of Gdansk (southern Baltic). Baltic Coastal Zone **21**: 51–60.

Williams, P. J.l. B., K. R. Heinemann, J. Marra, and D. A. Purdie. 1983. Comparison of ^{14}C and O_2 measurements of phytoplankton production in oligotrophic waters. Nature **305**: 49–50. doi:10.1038/305049a0

Williamson, T. J., M. J. Vanni, and W. H. Renwick. 2020. Spatial and temporal variability of nutrient dynamics and ecosystem metabolism in a hyper-eutrophic reservoir differ between a wet and dry year. Ecosystems **24**: 68–88. doi:10.1007/s10021-020-00505-8

Winslow, L., Read, J., Woolway, R., Brentrup, J., Leach, T., Zwart, J., Albers, S., and Collinge, D. 2019. rLakeAnalyzer: Lake physics tools. https://CRAN.R-project.org/package=rLakeAnalyzer.

Winslow, L. A., J. A. Zwart, R. D. Batt, H. A. Dugan, R. I. Woolway, J. R. Corman, P. C. Hanson, and J. S. Read. 2016. LakeMetabolizer: An R package for estimating lake metabolism from free-water oxygen using diverse statistical models. Inland Waters **6**: 622–636. doi:10.1080/IW-6.4.883