Faster CO_2 Response Curves with the LI-6800: RACiR and Steady-state parameter estimate comparison

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Introduction

- Portable gas exchange systems, including the LI-6400XT and the LI-6800 are often used to perform CO_2 response curves. These data are often used to partition limitations of photosynthesis and to calculate parameters of the FVcB model (Farquhar *et al.,* 1980).
- Traditional CO_2 Response Curves measure Assimilation (A) and inter-cellular CO_2 (Ci) at a series of steady-state CO_2 concentrations. Practical recommendations for proper parameter estimation include 5 points per limiting region (Long & Bernacchi, 2003). At a typical measurement time of ~ 2 minutes per data point, each curve can take 30 – 40 minutes.
- The Rapid A – Ci Response (RACiR) approach (Stinziano *et al*., 2017) is an instrument non-steady-state approach that rapidly changes in-coming CO_2. This approach has the potential for faster response curves
- Here we compare parameter estimation between steady-state and RACiR CO_2 response curves, as well as compare different rates of altering CO_2 using the RACiR method.

Methods

CO_2 Response Curves steady-state Comparisons

Steady-state comparisons were performed near mid-day on field-grown *Helianthus spp*. Ambient temperature was ~30°C. Chamber conditions were matched as closely as possible between the instruments: LI-6400XT: constant H2OS (starting VPDleaf of 1.5 kPa), LI-6800: VPDleaf 1.5 kPa. In both instruments, leaf temperature was 30 °C, light intensity was 1500 µmol m^{-2} s^{-1}. Leaves were acclimated at 400 µmol mol^{-1} until steady-state A and gsw were achieved.

Steady-state [CO_2] were 400, 300, 200, 150, 100, 50, 400, 400, 600, 800, 1000, 1200, 1400, 1600, 1800, 2000. Data was logged if slope A and CO2_s was < 0.5 or at 120 seconds. RACiR curves used same conditions and ramped CO_2 in linear fashion at 100 umol mol^{-1} $minute^{-1}$. Five leaves per each treatment were analyzed.

RACiR ramp rate comparisons

Comparisons were made on field-grown *Helianthus spp.* Similar chamber and acclimation conditions as above were used. CO_2 was linearly ramped to complete a 2000 µmol mol^{-1} ramp in 20, 15 and 10 minutes.

Parameter Estimation

All parameter estimation was performed using the R plant ecophysiology package (Duursma, 2015) using all default values. Values reported are corrected to 25°C.

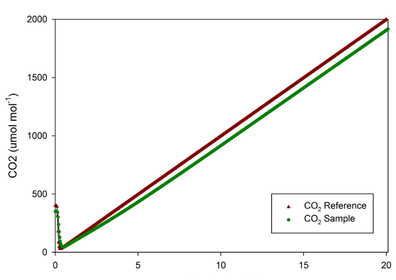

Figure 1: Example of a ramp in CO_2 at 100 µmol mol^{-1} min^{-1} using the RACiR technique. This example is with an active leaf in the chamber. The method requires post-correction with empty chamber data

Results

RACiR and steady-state comparison

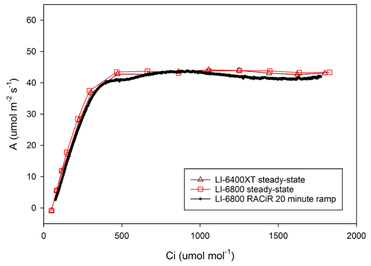

Figure 2: Example CO_2 Response Curves for steady-state (LI-6400XT and LI-6800) and RACiR (LI-6800, 20 minute ramp).

Table 1: Parameter estimates (mean ± S.E.) from CO_2 Response Curves, n = 5 leaves per curve type. See methods for description of curve fit.

	$V_{c,max}$	J_{max}	V_{TPU}
LI-6400XT	127.5 ± 3.4	234.6 ± 10.5	14.7 ± 0.4
LI-6800 steady-state	123.5 ± 3.1	219.8 ± 9.9	14.1 ± 0.7
LI-6800 (RACiR)	121.5 ± 5.8	234.4 ± 20.8	13.6 ± 1

RACiR ramp rate comparison

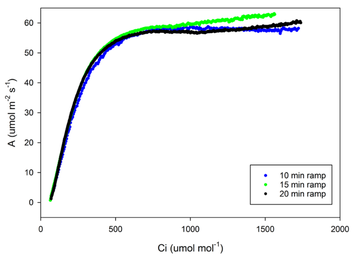

Figure 3: Example CO_2 Response Curves for 10 – 2010 umol mol^{-1} RACiR ramps with rates of 100, 133, and 200 µmol mol^{-1} min^{-1} for total time of 20, 15 and 10 minutes, respectively.

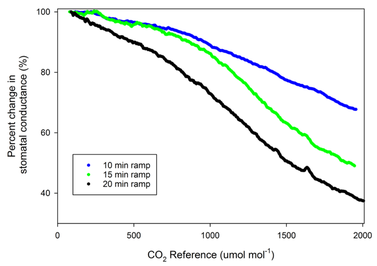

Figure 4: Change in stomatal conductance, reported as % change from starting conductance value. Values shown are the mean of n = 3 leaves.

Table 2: Parameter estimates (mean ± S.E.) from RACiR CO_2 Response Curves, n = 3. See methods for description of curve fit

	$V_{c,max}$	J_{max}	V_{TPU}
20 minute ramp (100 umol mol^{-1} min^{-1})	143.7 ± 9.9	325.4 ± 11.5	20.1 ± 0.2
15 minute ramp (133 umol mol^{-1} min^{-1})	151.1 ± 2.9	337.2 ± 6.7	19.8 ± 0.5
10 minute ramp (200 umol mol^{-1} min^{-1})	143.7 ± 5.4	344.6 ± 14.0	20.0 ± 0.4

Results

Parameter Estimation using segments of the response Curve

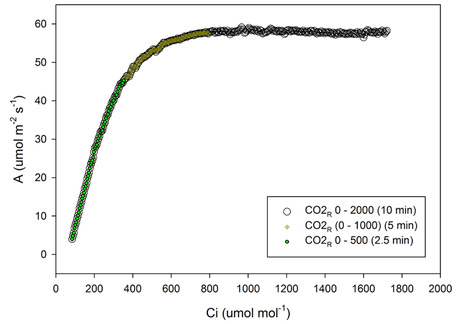

Figure 5: Example CO_2 Response Curve for 10 – 2010 umol mol^{-1} RACiR ramp showing the different portions used for parameter estimation in Table 3.

Table 3: Parameter estimates (mean ± S.E.) from RACiR 10 minute CO_2 Response Curves, using different portions of the curve, n = 3. See methods for description of curve fit. N.A. is not applicable as V_{TPU} was not attempted to be fit with the 0 -500 data.

	Total Time (minutes)	$V_{c,max}$	J_{max}	V_{TPU}
10 minute ramp 0- 2000 µmol mol^{-1}	10	143.7 ± 5.4	344.6 ± 14.0	20.0 ± 0.4
10 minute ramp 0- 1000 µmol mol^{-1}	5	143.9 ± 3.4	337.8 ± 11.5	20.0 ± 0.4
10 minute ramp 0- 500 µmol mol^{-1}	2.5	146.4 ± 3.3	336.6 ± 15.1	N.A.

Conclusions

- Faster CO_2 Response curves at high CO_2 limit stomatal response during the measurement.
- In this data-set, on a single species, important physiological parameters calculated from RACiR CO_2 response curves are not different to those calculated from steady-state CO_2 response curves.
- Depending on species, growth conditions and parameter of interest, RACiR response curves may be completed in as little as 2.5 minutes.

References

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