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Influences of light and humidity on carbonyl sulfide-based estimates of photosynthesis

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Understanding climate controls on gross primary productivity (GPP) is crucial for accurate projections of the future land carbon cycle. Major uncertainties exist due to the challenge in separating GPP and respiration from observations of the carbon dioxide (CO2) flux. Carbonyl sulfide (COS) has a dominant vegetative sink, and plant COS uptake is used to infer GPP through the leaf relative uptake (LRU) ratio of COS to CO2 fluxes. However, little is known about variations of LRU under changing environmental conditions and in different phenological stages. We present COS and CO2 fluxes and LRU of Scots pine branches measured in a boreal forest in Finland during the spring recovery and summer. We find that the diurnal dynamics of COS uptake is mainly controlled by stomatal conductance, but the leaf internal conductance could significantly limit the COS uptake during the daytime and early in the season. LRU varies with light due to the differential light responses of COS and CO2 uptake, and with vapor pressure deficit (VPD) in the peak growing season, indicating a humidity-induced stomatal control. Our COS-based GPP estimates show that it is essential to incorporate the variability of LRU with environmental variables for accurate estimation of GPP on ecosystem, regional, and global scales.

Carbonyl sulfide (COS) follows the same diffusion pathway into the leaf chloroplasts as CO2 and is consumed by the enzyme carbonic anhydrase (CA) (1, 2). The hydrolysis of COS via CA is irreversible (3), such that no respiration-like COS flux is evident under ambient conditions. Consequently, the atmospheric drawdown of COS above an ecosystem reflects the uptake of COS by plants, provided that other sources and sinks in the ecosystem are negligible or known. The dominant vegetative sink of COS was therefore recognized as a way to separate net terrestrial photosynthesis, which cannot be directly measured at scales greater than the leaf level. The accuracy of COS-based estimates of gross primary production (GPP) depends on how we relate the COS uptake to that of CO2. This study shows that COS-based GPP estimates will be significantly overestimated if the different environmental responses of COS and CO2 uptake are not taken into account. These findings are relevant for studies that rely on COS to quantify ecosystem to regional scale GPP, and support the use of a COS-based approach to constrain ecosystem flux partitioning. Moreover, the strong stomatal control on COS uptake shown in this study makes COS a suitable tracer for stomatal diffusion.

Author contributions: L.M.J.K., W.S., J.A., K.-M.E., K.M., U.S., T.V., I.M., and H.C. designed research; L.M.J.K., W.S., J.A., K.-M.E., K.M., and H.C. performed research; L.M.J.K., W.S., J.A., K.-M.E., and H.C. analyzed data; and L.M.J.K., W.S., J.A., K.-M.E., U.S., T.V., I.M., and H.C. wrote the paper.

The authors declare no conflict of interest.

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Data deposition: The data used in this work are available from https://zenodo.org/record/1211481#.XB4PStIzbIU. The dataset includes branch fluxes and mole fractions of COS and CO2, LRU, stomatal conductance, internal conductance, eddy-covariance fluxes of COS and CO2, and meteorological information. The code used to calculate the chamber fluxes from measured mole fractions is available from https://zenodo.org/record/1197330#.XB4O1NizbiU. The code used to generate the main results in this work is available from https://zenodo.org/record/1211499#.XBAPr5zbiU.

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behind due to its light dependence. The peak of FCOS is typically over a short period of time were confined to laboratory conditions or field measurements to test the applicability of findings from previous studies over different phenological stages. This dataset allows us to under field conditions and investigate if FCOS and FCO2 respond similarly to environmental changes. We performed continuous COS and CO2 branch chamber measurements over 5 mo during spring recovery and early summer in 2017 in a boreal forest in Finland, making this a study investigating FCOS at the branch level over different phenological stages. This dataset allows us to test the applicability of findings from previous studies—which were confined to laboratory conditions or field measurements over a short period of time—to different phenological stages and environmental conditions. With the different components of FCOS (ecosystem, soil, and branch fluxes) being characterized at the site, we are able to derive COS-based GPP estimates and test the effect of the variability of LRU on GPP.

Results and Discussion

Responses of FCOS and FCO2 to Light and Stomatal Conductance. Both FCOS and FCO2 show a strong diurnal cycle with a sink during the daytime (Fig. 1A). The increase of COS uptake (more negative FCOS) early in the morning coincides with the increase of stomatal conductance (gs,COS), whereas the increase of FCO2 lags behind due to its light dependence. The peak of FCOS is typically 1 h earlier than that of FCO2 (Fig. 1C), which was also observed by Geng and Mu (24) in a Chinese deciduous forest. Unlike FCOS, FCO2 Shows continued uptake during nighttime of 1.44 ± 0.95 pmol m\(^{-2}\)·s\(^{-1}\) (median ± SD) in May–July (Fig. 1C). The different responses of FCOS and FCO2 to light is also evident from Fig. 2A and B; FCO2 increases with the photosynthetically active radiation (PAR) up to 700 μmol m\(^{-2}\)·s\(^{-1}\), whereas FCOS increases up to a PAR value of 200 μmol m\(^{-2}\)·s\(^{-1}\). The light dependence of FCOS is caused by two distinct processes: (i) carbon fixation depends on the light reactions in the photosystems (25) and (ii) stomatal apertures, which controls the intercellular CO\(_2\) available for fixation, increases with light as a strategy to optimize carbon gain against water loss (26, 27). In contrast to CO\(_2\), the COS biochemical reactions are light independent (14, 21, 22), but FCOS responds to light solely due to the light response of stomatal conductance.

FCOS and FCO2 peak early in the morning when VPD is still low and gs,COS is high (Fig. 1A and C), which confirms a shared stomatal control on both fluxes. We find strong correlations of FCOS with gs,COS at all light levels and even during night (Fig. 2E and F). This is strong evidence that FCOS could provide a means to constrain stomatal conductance—during both day and night—and therefore links to both the carbon and water cycles (23).

In contrast, we also find that, at high light levels, the increase of FCOS with gs,COS is smaller than at low light levels (Fig. 2E and F). This suggests that during the daytime FCOS is colimited by nonstomatal resistances, which will be further discussed in the next section.

In the correlation with PAR, we find a decrease of COS uptake (less negative FCOS) toward higher light levels (Fig. 2A and B) that is consistent with a decrease of gs,COS (SI Appendix, Fig. S2), while on average FCO2 remains constant. This is in line with the hypothesis that the stomatal closure would affect FCO2 more than it would affect FCOS because the stomatal conductance is a more dominant component for FCOS than it is for FCO2 (17). This may also explain why the peak of FCOS occurs earlier than that of FCO2 (Fig. 1C); FCOS becomes more limited as VPD increases and gs,COS is limited (Fig. 1C), whereas FCO2 can continue to increase due to increasing PAR.

LRU varies largely over a day, which reflects the fact that COS uptake is light independent, whereas CO2 uptake is restricted under low light conditions, e.g., around sunrise and sunset (Fig. 1B). Therefore, LRU decreases exponentially toward high PAR (Fig. 2C and D), which is similar to the findings in Stimler et al. (14) and Sun et al. (17). The variation of LRU with PAR largely explains the variation of daytime LRU between days (SI Appendix, Fig. S1). Moreover, LRU does not become constant toward high light conditions (Fig. 2C and D, Inset), which was also observed by Sun et al. (17) for vegetation in a freshwater marsh. At high light levels we find a correlation between LRU and VPD (P < 0.01) and between LRU and gs,COS (P < 0.05) in the peak of the growing season (SI Appendix, Fig. S3), which is likely due to the different responses of FCOS and FCO2 to gs,COS. These findings support that differential stomatal limitations on FCOS and FCO2 drive LRU variation.

The light-saturated LRU (for PAR > 700 μmol m\(^{-2}\)·s\(^{-1}\)) is on average 1.1, which is on the lower end of LRU values reported in previous studies (see ref. 18 for an overview). Note that previous LRU measurements could have been affected by the dependence of LRU to PAR. LRU values have not always been determined at high light levels, which would have lead to underestimated LRU. For
mated the internal conductance to COS (gi,COS), which is a largely controlled by gs,COS. Furthermore, gi,COS has a relatively smaller limiting role on daytime gi,COS is smaller than gs,COS (see SI Appendix, Fig. S5). The fact that the LRU–VPD correlation follows the progression of the growing season is associated with the increase of daytime VPD. Early in the season FCOS and FC02 are not solely limited by stomatal conductance but rather by low temperatures, as is shown in SI Appendix, Fig. S5. The low temperatures suppress enzyme activities or mesophyll diffusion and therefore gi,COS has a relatively larger limiting effect on FCOS than gs,COS early in the season. In the course of the growing season, the limiting role of VPD on stomatal conductance becomes stronger, which manifests in the LRU–VPD relationship. This emphasizes that the LRU–gi,COS correlation (SI Appendix, Fig. S3) only applies when both FCOS and FC02 are controlled by stomatal conductance; i.e., at high temperatures and high light conditions.

**Seasonal Variation of LRU Influenced by Environmental Variables.** Fig. 3 shows the light-saturated LRU per month binned by VPD. The monthly median LRU decreases by 0.2 from April to July. No significant correlation between LDIR and VPD can be detected before June, whereas a significant decrease of LRU with VPD is observed in June and July (indicated by the significance levels in Fig. 3). The fact that the LRU–VPD correlation follows the progression of the growing season is associated with the increase of daytime VPD. Early in the season FCOS and FC02 are not solely limited by stomatal conductance but rather by low temperatures, as is shown in SI Appendix, Fig. S5. The low temperatures suppress enzyme activities or mesophyll diffusion and therefore gi,COS has a relatively larger limiting effect on FCOS than gs,COS early in the season. In the course of the growing season, the limiting role of VPD on stomatal conductance becomes stronger, which manifests in the LRU–VPD relationship. This emphasizes that the LRU–gi,COS correlation (SI Appendix, Fig. S3) only applies when both FCOS and FC02 are controlled by stomatal conductance; i.e., at high temperatures and high light conditions.

**Light and Humidity-Dependent LRU Required for Accurate COS-Based GPP Estimates.** In Fig. 4 we compare COS-based GPP estimates (GPP,COS) from COS ecosystem fluxes (determined from eddy-covariance measurements and subtracted estimates of the soil respiration (7, 15, 29)) and GPP from a traditional flux-partitioning method based on extrapolating nighttime respiration to the daytime (28) (GPP,SEE). GPP,COS is determined using different parameterizations of LRU: (i) a fit of the measured LRU (averaged over chambers 1 and 2) against PAR, which captures the decrease of LRU toward simultaneously increasing VPD and PAR (GPP,COS; see SI Appendix, Fig. S6 for the LRU–PAR relationship) and (ii) LRU fixed at 1.1 (the average LRU that we find at high light levels) and 1.6 (similar to what has been frequently used in other literature (7, 15, 29)), where the latter is shown in Fig. 4. The GPP,COS values from the GPP estimates represent the uncertainty based on Monte Carlo sampling of all parameters contributing to the GPP calculations (Methods). The GPP,COS uncertainty is larger than that of GPP,SEE, partly because the relative uncertainty of COS mole fraction measurements (∼1.7% of a typical ambient level of 450 ppm) is greater than that of CO2 mole fraction measurements (∼1.06% of a typical value of 400 ppm) (30). Still, Fig. 4 shows that the accuracy of GPP,COS is sufficient to detect differences between GPP,COS and GPP,SEE. We also calculated GPP,COS with the measured hourly LRU to determine to what extent uncertainty in the LRU–PAR function adds uncertainty to GPP,COS-fit. The uncertainties did not decrease with measured LRU values compared with the LRU–PAR function, implying that the empirical function captures the variability of LRU over the measurement period well.

With the constant LRU, the earlier peak of FCOS leads to an earlier peak in GPP,COS-const compared with GPP,SEE. The peak of ecosystem FCOS, and thus that of GPP,COS-const, is 2 h later than the peak of FCOS measured at the branch level at the top of the canopy. The reason for the delay between the FCOS peak from branches and ecosystem is that the diurnal pattern of the bulk canopy conductance is more symmetric, because light rather than gi is limiting CO2 assimilation in the lower canopy, in contrast to the top of the canopy (31). When GPP,COS is calculated with the average LRU that we find at high light levels (1.1), we find GPP,COS(13.4 ± 1.3 g C m⁻²d⁻¹; daytime data only) to

**Fig. 2.** Responses of FCOS, FC02, and LRU to light and of FCOS to g0,COS. Average FCOS, FC02 (A and B) and LRU (C and D) versus PAR, and FCOS versus g0,COS (E and F) from 18 May to 13 July for chambers 1 (Left) and 2 (Right). Data are plotted as the median of 15 equal-sized bins in the x range. The error bars represent the 25th and 75th percentiles of data in each bin. For the correlation of FCOS with g0,COS (E and F) the different colors represent different light conditions: blue (nighttime), green (daytime with low light conditions (PAR < 150 and 100 μmol m⁻²s⁻¹ for chambers 1 and 2, respectively; green); orange). A transition phase between low and high PAR is neglected. The coefficient of determination (R²), slope (sl), significance level (P), and number of data (n) are given for a linear regression through the median values (E and F).

**Internal Conductance of COS Limits FCOS During Daytime.** We estimated the internal conductance to COS (gi,COS), which is a combination of nonstomatal conductance terms, and find that during daytime gi,COS is smaller than g0,COS (see SI Appendix, Fig. S4 and the accompanying explanation). The ratio of gi,COS over g0,COS determines the relative importance of the two conductances on FCOS and thereby also on LRU (see equation S8 in ref. 12). The fact that we find a relatively low gi,COS compared with g0,COS during the daytime implies that g0,COS has a relatively large control on FCOS. Wehr et al. (23) estimated that the biochemical conductance (the CA activity) was of similar magnitude as g0,COS and therefore g0,COS has a relatively larger effect on day–night differences of FCOS than gi,COS has. This means that the diurnal change of FCOS is largely controlled by g0,COS. Furthermore, g0,COS has a relatively larger limiting role on FCOS during daytime than during nighttime (SI Appendix, Fig. S4). This variable role of g0,COS over a day explains why the relation between FCOS and gi,COS is different between different moments of the day, as depicted by different light levels in Fig. 2 E and F. If FCOS is used to determine g0,COS, and the limiting role of g0,COS on FCOS is ignored, this would lead to underestimation of daytime g0,COS. When the FCOS–g0,COS relationship is assumed to be the same for daytime and nighttime (following the blue curve in Fig. 2 E and F, gi,COS would be equal to 0.012 and 0.020 μmol m⁻²s⁻¹ for chambers 1 and 2, respectively, at FCOS of ~3 μmol m⁻²s⁻¹ (the average FCOS at high light levels). These values are, respectively, 46% and 48% smaller than what is actually observed (following the orange curve in Fig. 2 E and F). Therefore, ignoring the role of g0,COS would lead to a substantial underestimation of g0,COS.

**Example.** Kesselmeier and Merk (9) determined LRU at a light level of 300 μmol m⁻²s⁻¹ and Sandoval-Soto et al. (8) also measured LRU in Scots pine but at a light level of 600 μmol m⁻²s⁻¹ where FC02 is not PAR saturated.

The GPP,COS values from the GPP estimates represent the uncertainty based on Monte Carlo sampling of all parameters contributing to the GPP calculations (Methods). The GPP,COS uncertainty is larger than that of GPP,SEE, partly because the relative uncertainty of COS mole fraction measurements (∼1.7% of a typical ambient level of 450 ppm) is greater than that of CO2 mole fraction measurements (∼0.16% of a typical value of 400 ppm) (30). Still, Fig. 4 shows that the accuracy of GPP,COS is sufficient to detect differences between GPP,COS and GPP,SEE. We also calculated GPP,COS with the measured hourly LRU to determine to what extent uncertainty in the LRU–PAR function adds uncertainty to GPP,COS-fit. The uncertainties did not decrease with measured LRU values compared with the LRU–PAR function, implying that the empirical function captures the variability of LRU over the measurement period well.

With the constant LRU, the earlier peak of FCOS leads to an earlier peak in GPP,COS-const compared with GPP,SEE. The peak of ecosystem FCOS, and thus that of GPP,COS-const, is 2 h later than the peak of FCOS measured at the branch level at the top of the canopy. The reason for the delay between the FCOS peak from branches and ecosystem is that the diurnal pattern of the bulk canopy conductance is more symmetric, because light rather than gi is limiting CO2 assimilation in the lower canopy, in contrast to the top of the canopy (31). When GPP,COS is calculated with the average LRU that we find at high light levels (1.1), we find GPP,COS(13.4 ± 1.3 g C m⁻²d⁻¹; daytime data only) to
be overestimated by 72% compared with GPP_{COS-fit}. A fixed LRU value of 1.1 is not even sufficient for peak daytime values of GPP_{COS}, because cloudy days would have lower PAR, leading to higher LRU and lower GPP_{COS}. When GPP_{COS-const} is based on the LRU value that is frequently used in other studies (1.6), GPP_{COS} is underestimated at high light levels, but overestimated in the early morning and late afternoon (Fig. 4) with the daytime sum being 7% overestimated compared with GPP_{COS-fit} (daytime data only). These comparisons demonstrate that a constant LRU does not capture the variability of GPP. To the contrary, the diurnal cycles of GPP_{NEE} and GPP_{COS-fit} track closely in the early morning and late afternoon. The sum of daily GPP estimates differ by 13% (6.8 ± 0.3 and 7.8 ± 0.9 g C m⁻²·d⁻¹ for GPP_{NEE} and GPP_{COS-fit}, respectively). If LRU is held constant at a too-high value toward high PAR—when the different response of F_{COS} and F_{CO2} to stomatal closure is ignored—this would lead to an underestimation of GPP during daytime.

Ideally, COS would be used to validate other flux-partitioning methods and to assess assumed relations, such as the relation between respiration and temperature that is used to determine methods and to assess assumed relations, such as the relation between GPP_{COS-fit} and GPP_{NEE} around noon that is diurnal cycles of GPP_{NEE} and GPP_{COS-fit} track closely in the early morning and late afternoon. The sum of daily GPP estimates differ by 13% (6.8 ± 0.3 and 7.8 ± 0.9 g C m⁻²·d⁻¹ for GPP_{NEE} and GPP_{COS-fit}, respectively). If LRU is held constant at a too-high value toward high PAR—when the different response of F_{COS} and F_{CO2} to stomatal closure is ignored—this would lead to an underestimation of GPP during daytime.

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The Implications in Large-Scale GPP Estimates. The application of COS as a GPP tracer in terrestrial biosphere models can make use of LRU to scale the COS vegetation fluxes to those of CO₂. The relationships of LRU with PAR and VPD that are presented in this study are needed to make sure that the diurnal and seasonal variability of LRU is accounted for in terrestrial biosphere models (36–38). With accurate representation of LRU, the use of COS could potentially help improve the representation of diurnal variations of GPP in such models (29, 38). Furthermore, a time-integrated LRU is a useful measure to make the link between plant COS uptake and gross CO₂ uptake when large-scale biosphere models do not resolve diurnal cycles. For the months May–July we find that the time-integrated ratio of COS and CO₂ deposition velocities (based on branch measurements) is 1.6 (daytime only); including the months February–April the value is equal to 2.0 due to lower light conditions early in the season (SI Appendix, Fig. S7). Taking into account the nighttime data, the time-integrated F_{COS} to F_{CO2} ratio is 1.9 and 5.3 for May–July and February–July, respectively. These values apply to the branch level, in this case particularly to branches at the top of the canopy. For an accurate conversion of F_{COS} to GPP on the ecosystem and regional scales one needs to account for vertically varying PAR levels within the canopy. Furthermore, for upscaling to regional and global scales, the LRU–PAR relationship would have to be determined for other plant species in different ecosystems, given that large variability in LRU has previously been found between plant species (8, 12, 14–16, 39). For example, nighttime COS uptake by vegetation was shown to be small in a Mediterranean climate (39), leading to lower variation of LRU with PAR than what we found in this study for a boreal forest. These differences in LRU response in different climatic regions show that results from LRU dependencies in a single biome cannot be generalized across different climatic regions.

Moreover, the strong relationship between F_{COS} and g_s that was shown in this study can be used in process-based modeling studies to constrain the CO₂ diffusion pathway. Large improvements can be made particularly on the extent of nighttime stomatal opening, which is otherwise poorly quantified.

Conclusion. The different responses of F_{COS} and F_{CO2} to environmental variables, especially light, should not be ignored when COS flux measurements (either at leaf, ecosystem, regional, or global scales) are used to interpret changes in photosynthetic CO₂ uptake. Our findings show that the strong variability of LRU
for COS and 0.23 parts per million for CO2 (30). More information about the measurements and measurement precision was determined to be 7.5 parts per trillion on the branch chambers along with half-hourly cylinder measurements for the branch chambers (41). The sample tubing outside the instrumentation cabin was heated to prevent condensation on the tubing small holes in the chamber housing (41). The sample tubing outside the instrumentation and the calibration method can be found in Kooijmans et al. (42).

with environmental variables and phenological stages must be incorporated to obtain accurate estimates of GPP from COS measurements. The LRU–PAR relationship found in this study can help to scale up LRU to ecosystem, regional, and global scales. Furthermore, the close relationship between FCO2 and g0 that we observed can provide additional constraints to both the carbon and water cycles. With recent efforts to characterize sources and sinks of COS in ecosystems, accurate COS-based GPP estimates are now within reach and will allow testing and validation of other flux-partitioning methods.

Methods

Branch Measurements. Measurements were performed at the Station for Measuring Ecosystem–Atmosphere Relations II (SMEAR II) in Hyytiälä, Finland (61°51’N, 24°17’E, 181 m above sea level), which is dominated by Scots pine (Pinus sylvestris L.) (40). Four automated gas-exchange chambers were installed at the top of the canopy in two Scots pine trees between February 16 and July 17, 2017; details of those chamber measurements are provided in SI Appendix. PAR was measured by quantum sensors (Li-Cor LI-190) inside and outside the chambers. Temperature sensors (thermocouples and PT100) were placed inside the chambers. During measurements, the chambers were closed for 4 min and each chamber was measured once every hour. Air was pumped through a 4-mm (inner) diameter Synflex (Decabon) tube of 65-m length from the branch chambers to a quantum cascade laser spectrometer (QCLS) (Aerodyne Research Inc.) with a flow of 1–1.5 L min⁻¹ which was constantly recorded with Honeywell flowmeters (AWMS101VN). No active supply flow was provided, but ambient air could enter the chamber through small holes in the chamber housing (41). The sample tubing outside the instrumentation cabinet was heated to prevent condensation on the tubing walls. The QCLS measured CO2, CO, H2O mole fractions (1 Hz) from the branch chambers along with half-hourly cylinder measurements for calibration. We corrected for the spectral water vapor interference of COS (30). The overall uncertainty including scale transfer, water vapor corrections, and measurement precision was determined to be 7.5 parts per trillion for COS and 0.23 parts per million for CO2 (30). More information about the instrumentation and the calibration method can be found in Kooijmans et al. (30) and the deployment of the instrument at the SMEAR II station in Kooijmans et al. (42).

Fluxes were calculated from the change of molar concentrations within the chamber during chamber closure through the following mass balance equation:

\[
\frac{dC}{dt} = FA + q(C_a - C) \tag{2}
\]

where C is the molar concentration of each species inside the chamber (mol m⁻³), C_a the ambient molar concentration (mol m⁻³), V the chamber volume (m³), F the uptake or emission rate (mol m⁻² s⁻¹), A the leaf area (m²), and q the flux rate (mol s⁻¹). The measured mole fractions of the gas species (mol mol⁻¹) are converted to molar concentrations using the ideal gas law with average temperature during chamber closure and pressure measurements at the site. The fluxes were calculated from least-square fit of the time series of molar concentrations inside the chamber and by solving Eq. 2. C_a was determined from open chamber measurements during a few minutes before chamber closure.

We measured fluxes in empty chambers (called “blank” measurements) to test and correct for gas exchange by the chamber and possibly by tubing materials. We measured blanks for all chambers in July and during a few days in March, May, and June. The fluxes were corrected for the blank emissions as is further described in SI Appendix.

In SI Appendix we also discuss the effect of leaf mitochondrial respiration on FCO2 and LRU. Since we do not have the means to quantify diurnal changes of leaf mitochondrial respiration we approximate leaf-level LRU with the observed FCO2.

Stomatal Conductance. With transpiration measurements (FH2O) available, we would ideally calculate stomatal conductance to water vapor (gH2O) from FH2O normalized by VPD, where FCO2 is simultaneously determined along with FCO2 and FCO2 from the branch chamber measurements. However, in chamber measurements, transpiration is underestimated at high relative humidity (RH) levels because the transpired water vapor can get adsorbed on the chamber walls. Measurements of FCO2 therefore may not provide reliable gH2O estimates at high humidity levels. Therefore, we determined gH2O from the Ball-Berry model where the empirical slope (m) and intercept (g0) parameters are determined from gH2O which is determined with FCO2 and VPD under low-humidity conditions. We use a threshold for RH (70%) to avoid the effect of condensation on the chamber walls. The Ball-Berry model describes gH2O as function of FCO2, RH and the atmospheric CO2 mole fraction (43):

\[
g_{\text{H2O}} = m \cdot F_{\text{CO2}} + \frac{RH}{\text{a}_{\text{CO2}}} \cdot g_0 \tag{3}
\]

The model parameters m and g0 are determined through linear regression with an R² of 0.98 and 0.99 for chambers 1 and 2, respectively. With the regression being linear, we do not expect that using the Ball–Berry model rather than the measured gH2O leads to a bias in the results. As the Ball–Berry model does not allow for gH2O estimates in the dark when there is no photosynthesis, we determined the nighttime gH2O based on FCO2 normalized by VPD (for RH < 70%). The leaf temperature used for VPD was calculated from a leaf energy balance model that incorporated heating by incoming shortwave radiation and cooling by transpiration and sensible heat transfer (44). The RH used for VPD calculations was determined from heat vapor mole fractions in the open chamber a few minutes before chamber closure.

GPP Estimates. We determined GPP from NEE and extrapolated nighttime respiration following the traditional flux-partitioning method in Reichstein et al. (28). In addition to these NEE-based GPP estimates, we calculated GPP through Eq. 1 using different representations of LRU: with a PAR-dependent fit to the measured LRU (SI Appendix, Fig. S6) and with LRU fixed at 1.1 and 1.6. Vegetative COS fluxes were determined from eddy-covariance (EC) measurements in 2017 and soil COS fluxes that were characterized at the site in 2015 (45). The EC measurements of COS fluxes were made with a second QCLS of the same make at 10-Hz frequency together with a sonic anemometer (Solent Research HS1199, Gill Ltd.) at 23-m height. EC fluxes of COS were calculated from COS mixing ratios (corrected for water vapor in air) using the EddyUH software package developed at the University of Helsinki (46). Storage fluxes were estimated from mole fractions at 18 m assuming a constant height profile. More details about the flux and storage calculation procedure can be found in Kooijmans et al. (42). Data with low friction velocity (<0.3 m s⁻¹) were filtered out. Soil COS fluxes were measured in 2015 at the Hyytiälä site and showed no seasonal or diurnal cycle (45). An average

Fig. 4. GPP estimates based on COS and standard methods. Diurnal cycles of GPP between 18 May and 13 July 2017 based on NEE partitioning (GPPNEET, red) and observed COS ecosystem fluxes (using EC measurements with soil flux estimates subtracted (45)) following Eq. 1 (GPPCOS). For GPPCOS, LRU is determined with branch-dependent representation: from a PAR-dependent fit (SI Appendix, Fig. S6) of LRU based on the continuous branch measurements, where the average of chambers 1 and 2 is taken (GPPCOS-1; blue); LRU fixed at 1.6 (GPPCOS-const; green), which is similar to what has been frequently used in other literature (7, 15, 29). The averages (thick lines) and uncertainties (shaded areas) are based on 1,000 subsamples of Monte Carlo simulations that include uncertainties of all contributing components in the GPP calculation in each bin (Methods). We calculated the summed GPP from the average diurnal cycle for each representation (for daytime data only), which is shown in the top right corner.

| Time of day, UTC+2 [h] | GPP [µmol m⁻²s⁻¹] |
|------------------------|-------------------|
| 0                      | 6.8 ± 0.3         |
| 6                      | 7.2 ± 0.9         |
| 12                     | 8.4 ± 1.3         |

GPPNEET
GPPCOS-1
GPPCOS-const
GPPCOS-mean
soil flux of −2.7 pmol m⁻² s⁻¹ was subtracted from the ecosystem fluxes such that the remaining flux represents the vegetative COS exchange. The averages and uncertainties shown in Fig. 4 are based on 1,000 subsamples of Monte Carlo simulations that include uncertainties of all contributing components in the GPP calculation. That is, the SE of ECOS-EC and NEE; the COS soil flux uncertainty of 1.1 pmol m⁻² s⁻¹ (45); the SE of the fitting parameters of the LRU-PAR relation (using the median PAR in the calculation), or no uncertainty in LRU in the case of a constant LRU; the uncertainties of COS and CO₂ molar fractions of 0.60 pm and 0.13 ppm respectively (30), and the range of respiration calculations from figure 11 in ref. 47.

Meteorological Data: In addition to the temperature and PAR sensors installed at the branch chambers we use the data that are made available through the Meteorological Data.

Statistical Tests: The significance of correlations is tested with two-sided t tests of the significance levels (P) are reported. To reduce the effect of outliers we test the linear correlation of data based on bin-averaged medians, where the bins are of equal size. The number of samples of the original data are reported for each t test. The number of bins are mentioned in figure legends.

Code and Data Availability: Data and code related to this paper are available in refs. 48–50.

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