

Importance of the method of leaf area measurement to the interpretation of gas exchange of complex shoots

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Summary

Net CO₂ uptake in full sunlight, total leaf area (TLA), projected leaf area of detached leaves (PLA), and the silhouette area of attached leaves in their natural orientation to the sun at midday on June 1 (SLA) were measured for sun shoots of six conifer species. Among species, TLA/SLA ranged between 5.2 and 10.0 ($\bar{x} = 7.3$), TLA/PLA ranged between 2.5 and 2.9 ($\bar{x} = 2.7$), and PLA/SLA ranged between 2.0 and 3.7 ($\bar{x} = 2.2$). These ratios were reflected in the ratios of net photosynthesis computed on the basis of the three measures of leaf area. The much smaller values for TLA/PLA compared with the values for TLA/SLA indicate that leaf orientation effects, or shading, or both, caused more variation in the interception of solar radiation than did variation in leaf geometry (i.e., cross-section). Silhouette leaf area of lodgepole pine, (*Pinus contorta* spp. *latifolia*) and subalpine fir (*Abies lasiocarpa*) shoots measured at the summer solstice varied almost 2-fold with diurnal changes in solar altitude and azimuth. Sun shoots of both species and shade shoots of lodgepole pine had the greatest SLA during the early morning and late afternoon. The midday decline in SLA was related to the relatively upright orientation of needles of subalpine fir sun shoots and the relatively upright orientation of both sun and shade shoots of lodgepole pine. Shade shoots of subalpine fir reached a maximum in SLA at midday and this was related to the near horizontal orientation both of the shoots and the needles on them.

Introduction

Traditionally, photosynthesis and transpiration measurements on thin-leaved plants have been expressed according to the amount of leaf area (one side) intercepting direct beam sunlight (see Šesták et al. 1971, Zelawski and Walker 1976, and Lange et al. 1982 for reviews; also Körner et al. 1979). This area is usually referred to as projected leaf area (PLA), and in thin flat leaves is approximately one-half the total leaf area (TLA). In thin flat leaves in direct sunlight and oriented perpendicular to the solar beam, the interception of solar radiation depends only on PLA and radiant flux density measured with a flat sensor parallel to the leaf surface. However, if a shoot has leaves that are not planar, or that have various orientations, or have a uniform orientation that is not perpendicular to the solar beam, photosynthesis measured on a PLA basis may give a poor indication of the photosynthetic efficiency of the leaf tissue (Leverenz and Jarvis 1979, Carter and Smith 1985, Oker-Blom 1985, Smolander et al. 1987, Leverenz 1988). In such cases, photosynthesis measured on the basis of the silhouette area of leaves attached to the shoot in its natural orientation to the sun (SLA) may provide a better indication of photosynthetic capacity (Carter and Smith 1985).

The aim of the present study was to investigate the ways in which photosynthetic rates computed on a leaf area basis are affected by the measure of leaf area adopted. For this purpose measurements were made of net photosynthesis, TLA, PLA, and SLA of sun shoots of six conifer species, which provided a wide range of needle shapes, lengths, and orientations (van Gelderen and van Hoey Smith 1986). A study was also made of the diurnal variation in SLA of both sun and shade shoots of two conifer species. The results show that a much better understanding of the photosynthetic capacity of a shoot under natural conditions is possible if photosynthetic measurements are made in conjunction with all three measures of leaf area. In particular, ratios of the three measures of leaf area provide information about effects of leaf geometry, orientation, and shading on the interception of light. They show also that diurnal measurements of SLA provide additional important information about effects of leaf orientation and shading on light interception.

Materials and methods

All gas exchange measurements were made on shoots of trees growing in natural conditions at a subalpine site (2872 m elevation) in the Medicine Bow Mountains of southwestern Wyoming, USA (41°21' N, 106°10' W). Carbon dioxide and water vapor exchange for three to four shoots each from a different tree, were measured during midday (1000–1400 h) for six conifer species (*Pinus contorta* Dougl. ex Loud, ssp. *latifolia* (Engelm.) Critchfield (lodgepole pine), *Pinus ponderosa* Dougl. (ponderosa pine), *Pinus flexilis* James (limber pine), *Pseudotsuga menziesii* spp. *glauca* Franco (Douglas-fir), *Abies lasiocarpa* Nutt. (subalpine fir), and *Picea engelmannii* Parry (Engelmann spruce)) with a Li-Cor model 6250 portable photosynthesis system (4-liter cuvette). Individuals sampled were saplings ranging in height from 2.5 to 5.7 m. Measurements were taken under clear skies between 1045 and 1230 h on June 1, 1989. Photon flux densities, which ranged from 1474 to 1690 $\mu\text{mol m}^{-2} \text{s}^{-1}$, were measured with a quantum sensor (Li-Cor model 190s) oriented in the same plane as the main axis of the shoot. Cuvette air temperatures varied from 15.4 to 17.6 °C, and the vapor pressure deficit varied from 3.1 to 3.3 kPa. The terminal portion of a sun-exposed (south-facing) shoot from mid-canopy was enclosed in the cuvette in its natural orientation to the sun. Only the previous year's shoot increment and needles were used.

Total leaf area was determined by the glass-bead technique described by Thompson and Leyton (1971). Both PLA for individual needles and SLA for attached needles on a given shoot were measured with a video leaf area meter (Decagon Instruments, Pullman, Washington). For determination of PLA, all leaves were detached and laid flat beneath the camera. The SLA was determined by orienting individual shoots with attached leaves to the video camera to simulate the shoot's natural orientation to the sun (inclination and azimuth) at the time of the cuvette measurements. The video camera was adjusted to maximize the depth of the focus field so that an accurate measurement of SLA was possible for even the relatively long leaves of ponderosa pine. Calibrations were performed across the entire focus

width of the shoot using small cylinders of known area to simulate the needle-like leaves of these conifers. These error estimates were less than $\pm 5\%$ at all positions. In all cases, the silhouette area of the non-photosynthetic stem was subtracted from the final determination of SLA. The SLA measurement was not corrected for possible stem shading of leaves, although this error was estimated visually to be less than 5% of PLA. Photosynthetic CO_2 exchange was also computed on a dry weight basis after drying leaves for 72 h at 80 °C.

Three sun and three shade shoots were collected from the bottom third and south-side of the leaf canopy from three mature *P. contorta* and three mature *A. lasiocarpa* trees (dbh = 26 cm to 34 cm) for determination of SLA variation during the summer solstice (June 21–22). Three additional sun shoots were collected from the top of the same three subalpine fir trees. Shoots were rotated under the video area meter to simulate the movement of the sun on June 21–22 (List 1968). Variations in SLA were determined at each hour of the day for each shoot positioned in its natural orientation on the tree (azimuth angle and angle from horizontal).

Results

Mean values of net photosynthetic CO_2 uptake measured on June 1, which are shown in Table 1, were within the range reported for numerous conifer species (e.g., Larcher 1980, Smith 1985, Smith and Knapp 1989). Net photosynthesis expressed on a unit dry weight basis ranged from a low of $14.9 \mu\text{mol g}^{-1} \text{s}^{-1}$ in *Pseudotsuga menziesii* to a high of $23.4 \mu\text{mol g}^{-1} \text{s}^{-1}$ in *Pinus flexilis*. On a TLA basis, computed photosynthesis ranged from a low of $1.55 \mu\text{mol m}^{-2} \text{s}^{-1}$ in *P. menziesii* to a high of $2.95 \mu\text{mol m}^{-2} \text{s}^{-1}$ in *P. ponderosa*. Photosynthesis computed on either a PLA or an SLA basis differed from that computed on a TLA basis in direct proportion to the ratios of TLA/PLA and TLA/SLA, respectively (Table 1 and Figure 1). The greatest difference between photosynthesis computed on an SLA basis and photosynthesis computed on a TLA basis was observed in *Pinus flexilis* (25.2 versus $2.45 \mu\text{mol m}^{-2} \text{s}^{-1}$, respectively), whereas the smallest difference was measured in *Abies lasiocarpa* (7.91 versus $1.56 \mu\text{mol m}^{-2} \text{s}^{-1}$, respectively).

Table 1. Mean CO_2 uptake rates measured on June 1 and expressed on a total leaf area (A^T , $\mu\text{mol m}^{-2} \text{s}^{-1}$), projected leaf area (A^P , $\mu\text{mol m}^{-2} \text{s}^{-1}$), silhouette leaf area (A^S , $\mu\text{mol m}^{-2} \text{s}^{-1}$), and dry weight (A^W , $\mu\text{mol g}^{-1} \text{s}^{-1}$) basis. In each column the largest 95% confidence interval is shown ($n = 3-4$, each value being the mean of three measurements).

Species	A^W	A^T	A^P	A^S	A^P/A^T	A^S/A^T	A^S/A^P
<i>Pinus flexilis</i>	23.4 ± 4.4	2.45	6.65	25.2 ± 3.9	2.7	10.3	3.8
<i>Pinus ponderosa</i>	19.3	2.95 ± 0.57	7.89 ± 1.3	23.4	2.7	7.9	3.0
<i>Pinus contorta</i>	22.1	2.23	6.28	17.2	2.8	7.7	2.7
<i>Pseudotsuga menziesii</i>	14.9	1.55	4.19	10.4	2.7	6.7	2.5
<i>Abies lasiocarpa</i>	16.2	1.56	3.94	7.91	2.5	5.1	2.0
<i>Picea engelmannii</i>	16.9	2.07	5.74	12.2	2.8	5.9	2.1

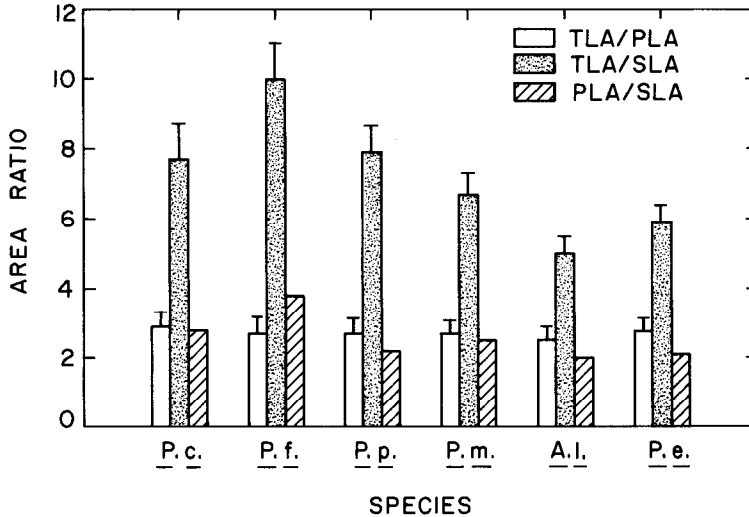


Figure 1. Quantitative comparisons of projected (PLA), silhouette (SLA), and total leaf area (TLA). These values also represent the proportional differences in calculated photosynthetic flux density for CO_2 uptake that would occur depending on which leaf area measurement was used in the computation. Vertical bars denote 95% confidence intervals.

Diurnal changes in SLA at the time of the summer solstice for a long-needled pine species (*Pinus contorta* spp. *latifolia*) and a short-needled, spruce-fir type (*Abies lasiocarpa*) are shown in Figure 2. Substantial reductions in SLA (up to 50%) occurred at midday in sun shoots of both species. The duration of the midday decline in SLA in lodgepole pine was almost twice that in subalpine fir (Figure 2). Variation in SLA of subalpine fir shade shoots showed the opposite trend to that for sun shoots, with SLA increasing to a maximum at midday and then decreasing. In lodgepole pine the diurnal variation in SLA of shade shoots was qualitatively similar to that in sun shoots, although the midday decline was much smaller.

Discussion

The photosynthetic capacity of plants is often measured on a leaf area basis. However, the interception of solar radiation is not directly related to total leaf area (TLA), but depends on many factors including leaf cross-section, orientation, and shading. The influence of leaf cross-sectional shape on the interception of sunlight, can be assessed by comparing TLA and projected leaf area (PLA). In laminar leaves, $\text{TLA/PLA} = 2.0$. For all other shapes TLA is more than twice PLA (e.g., for cylindrical leaves and stems $\text{TLA/PLA} \approx \pi$).

Projected leaf area (PLA) usually exceeds the area of unshaded foliage. Moreover, even without mutual shading, PLA overestimates light interception unless leaves are oriented perpendicular to the incident radiation. Use of silhouette leaf area (SLA), takes account of the effects of leaf geometry, orientation and shading on the interception of direct (collimated) solar radiation. Thus, the influence of mutual shading and

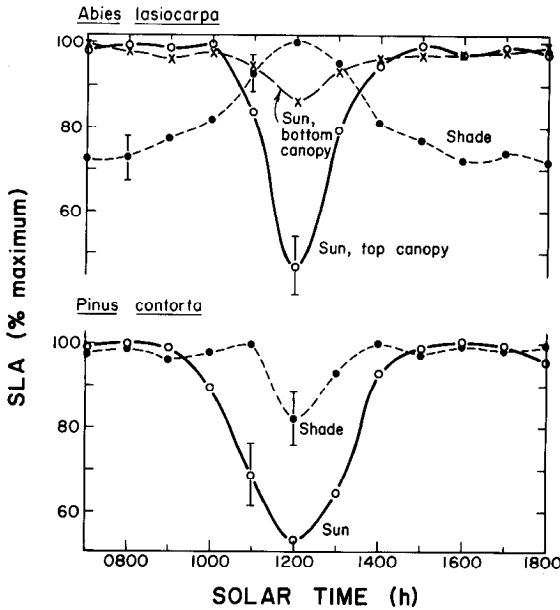


Figure 2. Variation in SLA throughout the day in subalpine fir (*Abies lasiocarpa*) and lodgepole pine *Pinus contorta* on June 21–22 (summer solstice). All shoots were collected from the south side of the tree; sun and shade shoots were arbitrarily selected based on shoot position and appearance. Each shoot was oriented in its natural azimuth orientation (170° to 191°) and angle from horizontal ($A.I. = 23\text{--}29^{\circ}$; $P.c. = 50\text{--}61^{\circ}$). The sun's altitude (video camera angle) above the horizon at a given time of day was simulated by rotating the shoot to duplicate the natural solar angles provided by List (1968) for June 21–22. The worst confidence intervals are indicated for the means of the nine shoots (three shoots from each of three trees) sampled for each curve.

leaf orientation on light interception can be evaluated by comparing PLA with SLA. For example, if all leaves on a shoot are thin, flat, and oriented with their adaxial plane perpendicular to the incident (collimated) light and do not shade one another, PLA equals SLA. Higher PLA/SLA values indicate that either leaf orientation or shading must be taken into account when considering the dependence of photosynthesis on leaf area.

For the conifers studied here, TLA/PLA ranged from 2.5 to 2.9, whereas TLA/SLA ranged from 5.2 to 10.0 (Figure 1). These values are similar to those reported by Carter and Smith (1985) and expressed as the silhouette-to-total area ratio (STAR). Differences in TLA, PLA, and SLA were reflected in differences in photosynthetic rates computed on the basis of the different measures of leaf area. Expressing photosynthesis on a PLA basis rather than a TLA basis increased computed CO_2 uptake rates almost 3-fold in all species. Expressing photosynthesis on an SLA basis rather than a TLA basis increased computed CO_2 uptake rates from 5- to 10-fold, depending on species.

When expressed on an SLA basis, the net photosynthetic rates observed in this study were among the highest reported for any species, and are in the range of values commonly reported for agricultural and herbaceous heliophyte species (Larcher

1980; Table 34). Other investigators have reported comparable rates of photosynthesis per unit leaf area for conifer shoots, but only in saturating diffuse light (Zelawski et al. 1973, Szanjawski et al. 1978, Leverenz and Jarvis 1979, Oker-Blom 1985). However, under such conditions, (e.g., in an integrating sphere) influences of leaf geometry, orientation, and shading on light interception, which are important under natural illumination, are absent.

Silhouette leaf area (SLA) varies diurnally and seasonally with changes in solar azimuth and altitude. The study of these changes can provide insight into the physiological implications of shoot structure. The midday decline that we observed in the SLA of sun shoots of lodgepole pine and subalpine fir and in shade shoots of lodgepole pine (Figure 2) can be related to the relatively upright orientation of the needles of sun shoots of subalpine fir, and the relatively upright orientation of both sun and shade shoots of lodgepole pine (see Carter and Smith 1985, for illustrations). Shade shoots of subalpine fir have a much more planar arrangement of non-overlapping needles than sun shoots. This, together with the relatively horizontal orientation of the shoots themselves, accounts for the midday increase in the SLA of subalpine fir shade shoots. In contrast, shade shoots of lodgepole pine have similar needle orientations to sun shoots. Shoot orientation in lodgepole pine is much less variable throughout the crown than in subalpine fir, which explains the similarity between sun and shade shoots of this species in the diurnal variation in SLA (Figure 2).

A better understanding of the photosynthetic capacity of shoots is possible if photosynthetic measurements are made in conjunction with measurements of SLA, PLA, and TLA. The ratios of these parameters provide information about leaf cross section, shading, and orientation. The same leaf area parameters could also be used to quantify shoot structural characteristics such as the degree of leaf clumping, which has potentially important effects on the thermal and aerodynamic properties of shoots (Hadley and Smith 1987, Smith and Carter 1988). However, without a more comprehensive and systematic evaluation of leaf area effects on the gas exchange of shoots, estimates of photosynthetic performance per unit leaf area will continue to be ambiguous.

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