ELSEVIER

Environmental Pollution 119 (2002) 163–175

ENVIRONMENTAL
POLLUTION

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A statistical approach to estimate O_3 uptake of ponderosa pine in a mediterranean climate

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Received 3 July 2001; accepted 9 November 2001

"Capsule": *Statistical models were developed to estimate stomatal conductance of ponderosa pine trees in sites differing in ozone exposure.*

Abstract

In highly polluted sites, stomatal behavior is sluggish with respect to light, vapor pressure deficit, and internal CO_2 concentration (Ci) and poorly described by existing models. Statistical models were developed to estimate stomatal conductance (gs) of 40-year-old ponderosa pine at three sites differing in pollutant exposure for the purpose of calculating O_3 uptake. Gs was estimated using julian day, hour of day, pre-dawn xylem potential and photosynthetic photon flux density (PPFD). The median difference between estimated and observed field gs did not exceed 10 mmol H_2O m^{-2} s^{-1}, and estimated gs within 95% confidence intervals. O_3 uptake was calculated from hourly estimated gs, hourly O_3 concentration, and a constant to correct for the difference in diffusivity between water vapor and O_3. The simulation model TREGRO was also used to calculate the cumulative O_3 uptake at all three sites. O_3 uptake estimated by the statistical model was higher than that simulated by TREGRO because gas exchange rates were proportionally higher. O_3 exposure and uptake were significantly correlated ($r^2 > 0.92$), because O_3 exposure and gs were highly correlated in both statistical and simulation models. © 2002 Elsevier Science Ltd. All rights reserved.

Keywords: Ozone uptake; ponderosa pine; Stomatal conductance; TREGRO

1. Introduction

There is no one model that fully explains stomatal behavior. The optimization hypothesis first proposed by Cowan (1977) suggests that stomata respond to environmental conditions to minimize water loss and maximize CO_2 uptake, mediated by substomatal CO_2 concentration (C_i). Under laboratory conditions, the resulting model (Farquhar and Sharkey, 1982) is generally supported. Under field conditions, the model is less reliable (e.g. Williams, 1983; Fites and Teskey, 1988). One poorly defined source of variability may be background O_3 exposure: it directly modifies stomatal behavior (Matyssek et al., 1995; Heath and Taylor, 1997). At low to moderate O_3 exposures,[1] a decline in assimilation is proportional to a decline in stomatal conductance (gs) in ponderosa pine (*Pinus ponderosa* var. *ponderosa* Dougl. ex Laws; Weber et al., 1993) and in sugar maple (*Acer saccharum* Marsh., Tjoelker et al. (1995). At higher O_3 exposures, or in combination with other environmental stressors, there appear to be irregularities in stomatal behavior. Sluggish stomatal response has been found in conifers with respect to increasing CO_2 (Keller and Häsler, 1987), vapor pressure deficit (VPD; Patterson and Rundel, 1989; Grulke, 1999), and photosynthetic photon flux density PPFD (Reich and Lassoie, 1984; Barnes et al., 1990; Weiser and Havranek, 1993; Grulke, 2000). Although the maximum rate of carboxylation (V_{cmax}) can be derived from A/C_i responses, external CO_2 concentrations greater than 800 ppm appear to suppress, not stimulate, assimilation of ponderosa pine at a high pollution site during late summer drought stress (Grulke, 1999).

These stomatal aberrations are not accounted for in the Farquhar model, and as such, are not incorporated into current physiological process models. In Martin et

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[1] "Moderate" as defined using cumulative ozone exposures in Table 1.

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PII: S0269-7491(01)00338-4

al.'s (2001) modification of the Farquhar model, aspen response to O_3 is activated after a critical exposure is achieved and defense systems are overloaded. Despite all the chamber exposure and field studies on ponderosa pine, a single threshold cannot be assigned for the species, and may vary between stands due to differing environmental stressors and underlying genetic structure. Although development of a related statistical model has allowed such a threshold to be defined for each site (Grulke et al., 2002), it could not be determined a priori. Also, existing physiological models simulate response to moderate O_3 exposures (<260 ppm h). The O_3 exposure at two of the three sites described in this paper is greater than any other forested site in the United States (300–345 ppm h accumulated over the growing season). Stomatal behavior at the two most polluted sites used in this study is further compounded by high nitrogen deposition, which has been found to modify defense systems (Polle, 1998), complicating Martin et al.'s (2001) approach.

The best way to determine canopy O_3 uptake is by direct measurement (Amthor et al., 1994; Goldstein et al., 2000), but this is not possible for the majority of stands, through time, for which estimates of O_3 uptake are needed. For these reasons, a statistical model was applied to estimate field responses of gs and thus calculate O_3 uptake throughout the growing season at each of three sites across a pollution gradient for hours and days when measurements were not made. A simple approach was taken: gs was estimated using julian day, hour of day, pre-dawn xylem potential, PPFD, and site-specific gs response to PPFD. O_3 uptake was calculated from estimated daily gs, hourly O_3 concentration, and a constant to correct for the difference in diffusivity between water vapor and O_3. The statistical model was developed from 1995 field data, and was tested by comparing predicted versus observed gs during a specific hour and day in two growing seasons with average- (1993) and below-average precipitation (1994).

We also used TREGRO, a carbon-balance simulation model, to calculate O_3 uptake at the three sites in the same years. This model incorporates a module for handling O_3 exposure via reductions in mesophyll resistance and/or respiration (Weinstein et al., 1991), as well as a routine for calculating O_3 uptake (Constable and Retzlaff, 2000; Retzlaff et al., 2000). We used this simulation model because it was already parameterized with the environmental and physiological data set for the same trees used in the data presented here (Constable et al., 1996; Grulke and Retzlaff, 2001), and had the above capabilities.

2. Methods

2.1. Research sites

Three sites differing in pollutant exposure were chosen for study within the mixed conifer zone (sensu Barbour, 1988), in the San Bernardino Mountains east of Los Angeles, California. The sites are ordered from west to east across the mountain range, from high (Crestline), to moderately high (Strawberry Peak), to moderate pollution exposure (Barton Flats, Table 1). This region

Table 1
Summary of differences in pollution exposure, microenvironment, and soil characteristics between sites across the pollution gradient

	Crestline	Strawberry Peak	Barton Flats
Pollution load	"high"	"moderately high"	"moderate"
Elevation (m)	1800	2240	1830
Latitude	$34^\circ 14''05'$N	$34^\circ 14''00'$N	$34^\circ 09''42'$N
Longitude	$117^\circ 19''12'$W	$117^\circ 08''112'$W	$116^\circ 51''00'$W
Average O_3 concentrations (ppb)[a]	79–80[b]	69–76	62–64
N deposition (kg ha^{-1})[c]	20–40	n.d.	6–9
Soil N (%)[d]			
A_1	0.20	0.11	0.08
B_1	0.09	0.07	0.04
Pre-dawn ψ (MPa)[d]	-1.0 to -1.5	-0.9 to -1.6	-1.1 to -1.7
Annual precipitation (cm)[e]	98	96	90
Cum degree days (°C)[f]	2650–2840	1980–1820	1310–1230

[a] Summarized from Grulke (1999).

[b] When two numbers are listed, the first number represents a year of average precipitation (1993) and the second number represents a year of 20% lower than average precipitation (1994).

[c] Values for N deposition are broad due to variations in canopy structure (summarized from Kieffer and Fenn, 1997).

[d] Summarized from Grulke et al. (1998).

[e] Annual precipitation was averaged over the hydrologic years 1980 through 1997 (San Bernardino County Water Resources).

[f] Summarized in Grulke and Balduman (1999).

east of the Los Angeles air basin has received significant pollutant deposition over the last 50 years (Miller and McBride, 1999).

Gas exchange was measured in 1993, 1994, and 1995. In 1993 and 1995, above average precipitation was received (208 and 134 cm, respectively), of which 5 and 3% fell during the summer from 15 May through 30 September. In 1994, 80% (73 cm) of the 115 year regional average of 93 cm was received, of which 1% fell during the summer (San Bernardino County Water District). The 1993 field season was preceded by a greater than average precipitation year. In both mesic and xeric years, the upper soil horizons dry to $<2\%$ soil moisture outside of the canopy drip line by mid July (Grulke, 1999). Higher soil moisture has been reported directly under the tree canopy (Temple and Miller, 1998) presumably due to hydraulic lift (Dawson, 1994). However, this phenomenon was not observed for 40-year-old trees at these sites. Trees may draw water from sources deep in the weathered bedrock in late summer (Hubbert, 1999), which is inaccessible for routine measurement. For this reason, pre-dawn xylem potential (from Grulke et al. 1998) was used to indicate degree of drought stress experienced by trees on site.

2.2. Environmental monitoring

Microenvironmental and edaphic conditions of these three sites have been described in detail elsewhere (Grulke et al., 1998; Grulke, 1999; Grulke and Balduman, 1999; summarized in Table 1). Ozone concentrations (Model 1008, Dasibi Environmental Corporation, Glendale, CA) were measured hourly throughout the year, but presented here for 15 April through 15 October in 1993, 1994, and 1995. Sample air was drawn from approximately 3 m above the ground, at approximately 2 m from the buildings where instrumentation was housed. Dust was filtered through a teflon filter prior to passing the sample air through the analysis tube. Ozone monitors were calibrated once per month during the summer months (May through October) with a tertiary transfer standard. The calibration of the monitors passed external audits annually, using a device certified by the US Environmental Protection Agency.

PPFD was measured hourly over the growing season in 1993, 1994, and 1995 (Model 190s, LiCor Instruments) in an open area. Data were recorded every 5 min, and hourly average values were recorded on a data logger (Model 21×, Campbell Scientific). The time of dawn (>2 μmol quanta $m^{-2}s^{-1}$) and sunset (<2 μmol quanta $m^{-2}s^{-1}$) were determined from these measurements. PPFD levels were also monitored on each of the two primary branches used for gas exchange measures, on each of six trees per site in 1995. PPFD sensors (4 mm diameter, Hammamatsu Corp.) were installed just above the middle of each live needle age class on each of two primary branches. The average PPFD over the live foliage was monitored every 1 min and recorded as a 5 min average for each primary branch on days of diurnal measurements. These data were used in conjunction with gs measurements to determine site-specific response to PPFD.

2.3. Selection of trees

At each site, a regionally typical, multi-age stand was chosen, and a plot was established to include at least 40 trees in the 21- to 60-year-old tree age class. For each tree in the plot, bole diameter, tree height, percent chlorotic mottle of previous year needles, number of needle age classes retained, and the distance to the nearest intra- and interspecific tree as a measure of inferred competition were measured. Six (1995) to twelve (1993, 1994) trees were selected for intensive measurements that had morphological attributes closest to that of the average of that tree age class. Although this is a broad tree age class, there were no significant differences between 21–40 and 41–60-year-old tree age classes with respect to gas exchange, growth rates, and phenological patterns (Grulke, 1999). At Crestline and Strawberry Peak, sampled trees were widely separated and had little canopy or root interaction. Barton Flats was characterized by clumps of trees, separated by open areas, typical of droughty environments. Individual trees were used as within-site replications.

2.4. Stomatal conductance

Gas exchange was measured on clear days, at monthly intervals, at three sites in the San Bernardino Mountains east of Los Angeles, California for three growing seasons. Diurnal gas exchange measured in 1995 was used to construct site-specific, empirical models of gs. Field observations of maximum daily gs in 1993 and 1994 were compared directly against model estimates of gs during the same hour and julian day. The data for the model were taken in a year of average precipitation (relative to a 115 year record maintained by the San Bernardino County Water District). Precipitation in 1994 was 20% below average, sufficient to cause significant drought stress (Table 1) and a reduction in elongation growth (Grulke and Balduman, 1999).

Stomatal conductance was measured with a closed gas exchange system (Model 6200, LiCor Instruments) using a modified technique. The standard methodology described for Model 6200 optimizes for measurement of CO_2 exchange rate: chamber vapor pressure is matched as carefully as possible with that of ambient by passing a portion of the air flow through the desiccant, and data were logged when a steady rate of change in CO_2 was observed. In this method, relatively little leaf area was placed in the cuvette and a short equilibration time was used to minimize the depletion of CO_2 in the closed system. However, the detection of changes in cuvette humidity is slower than that of CO_2, and errors result at both low and high leaf gas exchange rates. The standard technique was modified (described below) to optimize for measurement of gs. Calibration of the humidity sensor was checked several times a growing season on site with a dew point generator (Model 610, LiCor Instruments).

Three fascicles on an adjacent branch were inserted into the cuvette and dry air was added as necessary to minimize VPD ($\leqslant$0.01 kPa change). Air flow through the desiccant was switched off, 1-year-old needles were placed in the cuvette, and VPD was monitored. When transpiration began to humidify the cuvette, dry air was switched on to the pre-set flow, minor adjustments were made if necessary to maintain a constant VPD, and data were logged. By the time the cuvette was in equilibrium with respect to H_2O, CO_2 concentration in the closed system was often lower than that optimal for photosynthetic measures. At CO_2 concentrations <305 ppm, data were discarded because C_i was lowered sufficiently to increase gs. Repeat measurements of gs were generally within 2% for the same needles. Gs measured with this modified technique was 4–6 versus 3–4 mmol $H_2O\ m^{-2}s^{-1}$ as measured with an open gas exchange system (Model 6400, LiCor Instruments). At values of $gs > 15$ mmol $H_2O\ m^{-2}s^{-1}$, data were comparable between the two instruments.

Gs was reported on a needle surface area basis, determined from a geometric model constructed of fascicle diameter ($\leqslant$0.02 mm, Mityoto electronic calipers) and needle length (mm). Gs was measured on two primary branches per tree, in the lower third of the canopy, on the most open aspect of the tree (usually south-facing). Foliage on the same branches was measured monthly from the end of April through mid October. The primary branches used for gas exchange measurements received at least 4 h per day of direct sun ($\geqslant$1800 μmol quanta $m^{-2}s^{-1}$) in May, and at least 3 h per day of saturating light ($\geqslant$1000 μmol quanta $m^{-2}s^{-1}$) in all months of the growing season. The maximum daily gs for each tree was obtained between 08:30 and 13:30 in May and June, and between 08:30 and 11:30 in the remaining months at ambient CO_2, leaf temperature, VPD, and under saturating PPFD ($\geqslant$1400 μmol quanta $m^{-2}s^{-1}$ for at least 20 min prior to and during measurement). Diurnal gs was measured at ambient conditions from pre-dawn until dark, at approximately 1.5 h intervals on each tree.

2.5. Estimation of diurnal course of gs

We used a statistical approach to estimate the 90th percentile of field observations of diurnal gs instead of boundary line analysis (Webb, 1972). Site-specific estimates of gs were calculated from diurnal measures of six trees per site, taken monthly. The 90th percentile (henceforth referred to as "maximum") gs was determined instead of average response because any one primary branch was not in full sun for the entire day. The lighted portion of the canopy influences gs throughout the canopy, but gs of foliage that is not in direct light is depressed. Collating the response of all the trees on the site assured that some proportion of the foliage measured during any one hour would be exposed to non-limiting light on that sampling date. The fitted curves for the compiled tree diurnals were produced by the nonparametric scatterplot smoother called loess (Cleveland and Devlin, 1988). The technique uses locally linear or quadratic regression to estimate a relationship between the predictor, ×(hour of day), and the response, y (gs in mmol $H_2O\ m^{-2}s^{-1}$): the data suggest the shape of the relationship. The maximum diurnal gs response was estimated for times between sampling dates (and for times and dates in different years) from the nonparametric regression models. The empirical model assumed $gs=0$ before dawn and after dusk. The hourly values of gs were adjusted for fog or cloudy conditions based on hourly PPFD measurements, and the site-specific response between gs and PPFD.

2.6. Calculations for O_3 uptake

Ozone uptake for trees at each site was calculated from hourly estimates of gs in 1993 and 1994, hourly O_3 concentrations measured at each site, and a correction for the difference in diffusivity between O_3 and water vapor (Laisk et al., 1989). Because foliage was sampled between 1.5 and 2 m from the ground, and O_3 concentrations were measured at 3 m, no adjustments were made to measured O_3 concentrations. For calculated O_3 uptake, dawn was rounded down and dusk was rounded up to the nearest hour. Because gs and O_3 concentrations were both low at these times, the effects of rounding are believed to be negligible.

2.7. TREGRO description

TREGRO is a physiological simulation model of the carbon, water, and nutrient fluxes of an individual tree (Weinstein et al., 1991). TREGRO was developed to analyze the response of trees to multiple environmental conditions, such as temperature, drought, nutrient deficiency, and exposure to pollutants. In the model, the tree is divided into the following compartments: a canopy of leaves grouped by age class, branches, stem, and coarse and fine roots in three soil horizons. In each compartment, the model keeps track of three carbon pools: structure (living, respiring tissue), wood (the non-respiring tissue), and total non-structural carbohydrate (TNC). The parameter file defines species-specific characteristics including (but not limited to) maximum photosynthetic rate, rates of maintenance and growth respiration, specifics of nutrient uptake kinetics, phenological patterns of growth, growth rates of individual tree compartments, and carbon partitioning. The model calculates the carbon assimilation (Farquhar et al., 1980) of the entire tree each hour as a function of ambient environmental conditions and the availability of light in the canopy, water, and nutrients. In this simulation, a 40-year-old ponderosa pine parameter file from a previous TRE-GRO simulation exercise was used initially (Grulke and Retzlaff, 2001). For each site, the original tree was re-parameterized to reproduce growth (Grulke and Balduman, 1999) with special attention to match the gas exchange characters and leaf mass growth so that total tree CO_2 and O_3 flux would be calculated properly.

The interaction between tree growth and the environment in TREGRO was achieved by linking separate data files. The meteorological file defines the site-specific hourly environmental conditions including air temperature (°C), relative humidity (%), rainfall (mm), photosynthetic photon flux density (μmol quanta $m^{-2}s^{-1}$) and O_3 concentration (ppb). In this simulation, meteorological data were used from the three field sites described previously (Grulke, 1999). On an hourly basis, O_3 uptake was calculated from CO_2 and thus H_2O flux, hourly atmospheric O_3 concentration, and a constant to correct for the difference in diffusivity between water vapor and O_3. Cumulative O_3 uptake was calculated for the growth period 15 April through 15 October at each of the three sites, for 1993 and 1994. The water flux module of TREGRO was evoked to simulate years of average and 20% below-average precipitation.

3. Results

3.1. Ozone exposure at the three sites

Cumulative O_3 exposure increased linearly at all three sites in the 2 years of study (Fig. 1). The rank order of pollutant exposure at the three sites in the San Bernardino Mountains, from moderate, to moderately high, to high, from east to west across the San Bernardino Mountains did not differ between the 2 years of differing total annual precipitation (Table 1). Higher O_3 concentrations were expected in a drier year with higher spring temperatures (1994). However, O_3 concentrations were not greater at all sites in 1994. One of the sites, Strawberry Peak, had lower O_3 concentrations in 1994 probably due to greater incidence of morning fog in early summer at that site. Trees at the most polluted site experienced nearly identical O_3 exposure between the two years. Thus, the pattern of pollutant transport into the forest or its degradation was altered in a below-average precipitation year.

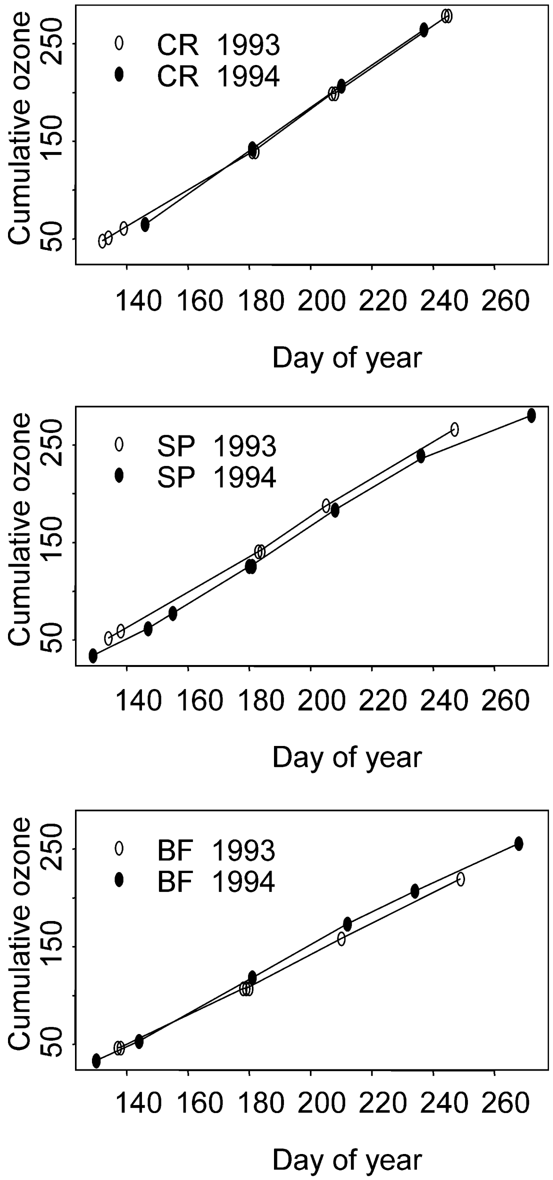

Fig. 1. Cumulative O_3 exposure (in ppm h) in 1993 and 1994 at the three sites across a pollution exposure gradient in the San Bernardino Mountains, east of Los Angeles, CA.

3.2. Seasonal changes in diurnal gs

The western two sites had similar diurnal gs patterns over the growing season except for August, where Strawberry Peak had half the maximum gs than that at Crestline (Fig. 2). Gs of trees at the eastern end of the pollution gradient, Barton Flats, was consistently lower in each month of the growing season than that of trees at the western sites, especially in August and September. However, predawn xylem potential was significantly lower at this site only in August, 1994 (Grulke et al., 1998). Because the response of six trees were collated,

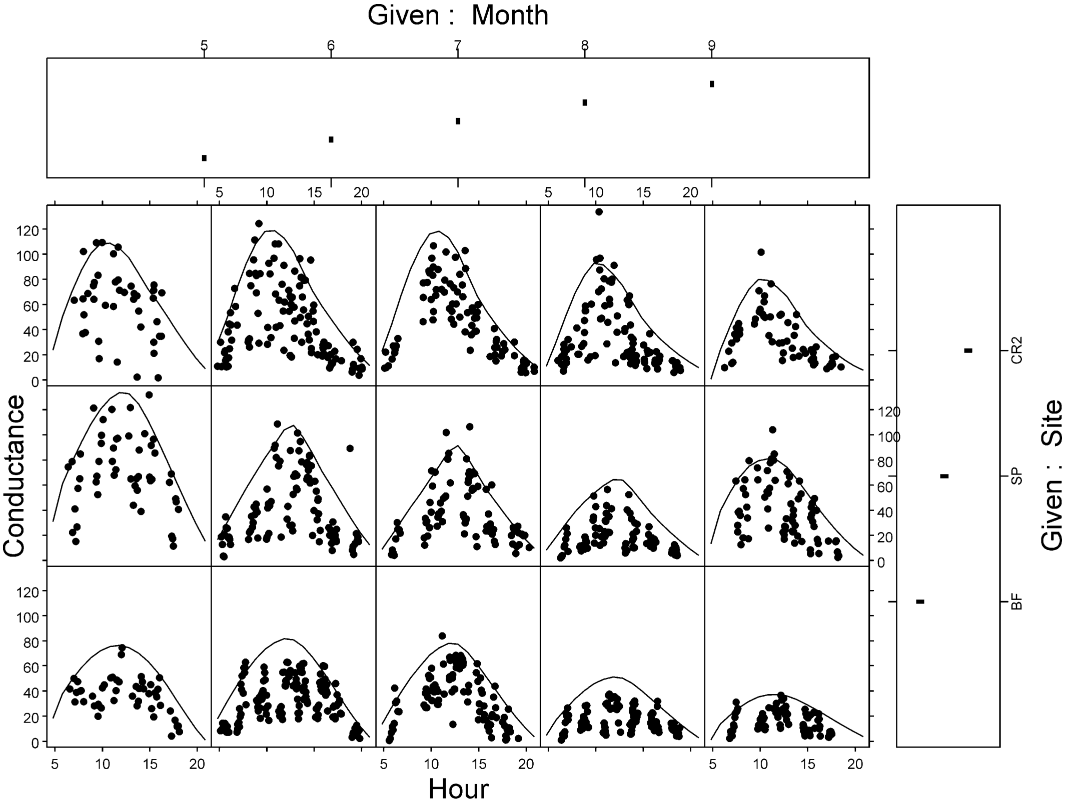

Fig. 2. Diurnal course of stomatal conductance (gs) in mmol H_2O $m^{-2}s^{-1}$ measured monthly (May–September) at the three sites across the pollution gradient. The solid line represents a statistically fit 90th percentile of the response of six 40-year-old trees measured at each site in 1995.

there was no evidence of midday depression in any of the diurnals: gs declined gradually from a midday maximum.

3.3. Interannual differences in daily maximum gs

Of all three years, maximum daily gs values were highest in spring of 1994 (Fig. 3), probably due to higher temperatures that spring (Grulke, 1999). At the end of the growing season, maximum daily gs values were lowest for all sites in 1994, the driest year. At Strawberry Peak and Barton Flats, late summer gs in 1995 was intermediate that in 1994 and 1993. The seasonal course of maximum daily gs was nearly monotonic at the western-most site in 1995. Maximum daily gs was about 15% lower on any one sampling date at Barton Flats relative to the two western sites. Pre-dawn xylem potential did not drop below−1.2 MPa in the years of average precipitation (1993, 1995) at the three sites. In 1994, pre-dawn xylem potential was <-1.2 MPa by mid July at Barton Flats, and by early August at Strawberry Peak and Crestline. Although pre-dawn xylem potential did not differ significanty across the three sites by early September (Table 1 and Grulke et al., 1998), it was a significant explanatory ($P = 0.05$) at Barton Flats for late summer gas exchange in 1994.

3.4. Light response of gs

Because gs was measured on clear days, it was necessary to determine the response of gs to PPFD to modify the estimated gs during cloudy or foggy periods. Using boundary line analysis (Webb, 1972), a line was fit to the upper 90% of the data taken over the entire 1995 growing season, determined separately for each site. Gs response was linearized with a natural logarithm transformation of PPFD (Table 2). Although the maximum observed gs was similar for the fitted lines at all three sites, the most polluted site required higher PPFD to elicit the same degree of stomatal opening. This was especially true for early morning stomatal opening.

3.5. Independent test of measured vs. fitted gs

Field measurements of gs in 1993 and 1994 were compared to estimated gs in the same hour and julian day (Fig. 4). Relatively few comparisons fell outside of the 95th percentile (dotted lines) of the 1:1 relationship between observed and predicted gs (solid line). The difference between observed and fitted gs was represented by box plots, presented separately for the three sites in each of the two years (Fig. 5). In all cases, the median difference between field observations and fitted gs did

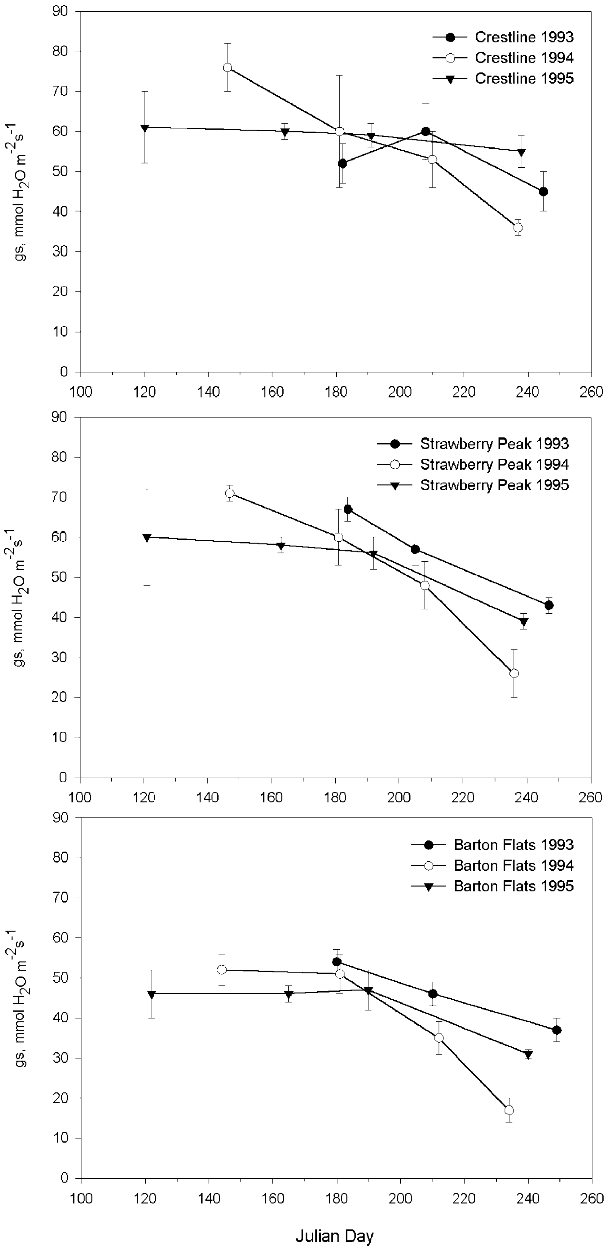

Fig. 3. Seasonal course of mean daily maximum stomatal conductance (in mmol H_2O $m^{-2}s^{-1}$) for 12 trees at each site in 1993 and 1994, and six trees at each site in 1995.

Table 2
Stomatal conductance (in mmol H_2O m^{-2} s^{-1}) response to PPFD[a] (in μmol quanta $m^{-2}s^{-1}$) for the three sites across the San Bernardino Mountains

Site	Light response	Adjusted r^2	P
Barton Flats	gs = 9.79 ln (PPFD) + 10.22	0.960	0.0001
Strawberry Peak	gs = 12.49 ln (PPFD) + 17.28	0.873	0.0001
Crestline	gs = 15.65 ln (PPFD)−10.35	0.826	0.001

[a] PPFD, photosynthetic photon flux density.

not exceed 10 mmol H_2O $m^{-2}s^{-1}$. For all sites combined, field observations in 1993 were slightly higher than fitted gs (by 4 mmol H_20 $m^{-2}s^{-1}$), and field observations in 1994 were slightly lower (by 5 mmol H_2O $m^{-2}s^{-1}$). When all data were combined within a site, the best fit was for the high pollution site (observed gs was 3 mmol H_2O $m^{-2}s^{-1}$ lower than that estimated), followed by the moderate pollution site (10 mmol H_2O m^{-2} s^{-1} higher than that estimated); the moderately high pollution site had the poorest fit, where observed gs was 12 mmol H_2O $m^{-2}s^{-1}$ lower than that estimated). At Barton Flats, field observations of gs were higher than those estimated for that site in both 1993 (by 10 mmol H_2O $m^{-2}s^{-1}$) and 1994 (by 2 mmol H_2O $m^{-2}s^{-1}$). At Strawberry Peak, field observations of gs were lower than those estimated for that site in both 1993 (by 11 mmol H_2O $m^{-2}s^{-1}$) and 1994 (by 3 mmol H_2O $m^{-2}s^{-1}$). At Crestline, field observations were higher than those estimated for that site in 1993 (by 4 mmol H_2O $m^{-2}s^{-1}$) and lower than those estimated for that site in 1994 (by 10 mmol H_2O $m{-}^2s^{-1}$).

Regressions between field observations and fitted gs were constructed to estimate the predictive capability of the statistical model estimates. The best fit was for Barton Flats, where the adjusted r^2 in 1993 was 0.44 ($P=0.0001$) and in 1994 was 0.75 ($P=0.0001$). At the higher pollution sites, the ability to predict gs was poorer. At Strawberry Peak, the adjusted r^2 (adjusted for sample size) was 0.14 in 1993 ($P=0.0001$) and 0.46 in 1994 ($P=0.0001$). At Crestline, the r^2 was 0.14 in both 1993 ($P=0.02$) and 1994 ($P=0.03$). Despite the difference in variation explained between 1993 and 1994 at each site, the regressions for the two years did not differ significantly, and fell within the 95% confidence intervals.

3.6. Estimated ozone uptake at the three sites

Cumulative O_3 uptake (calculated statistically) had the same rank order as cumulative O_3 exposure across the pollution gradient. The site with the greatest O_3 exposure, Crestline, also had the greatest O_3 uptake by trees on site (Table 3; Fig. 6). Conversely, the site with moderate O_3 exposure, Barton Flats, had the lowest estimated O_3 uptake. In the drier year, Barton Flats and Strawberry Peak had lower estimated O_3 uptake than in the year of average precipitation. Ozone uptake and cumulative O_3 exposure at Crestline was nearly identical in both 1993 and 1994.

Cumulative O_3 uptake as calculated by TREGRO (Fig. 6) was lower than that estimated with the statistical model. In a dry year, the rank order of sites followed that of O_3 exposure. In a year of average precipitation, the rank order changed: Strawberry Peak had the lowest O_3 uptake, and Crestline had the highest O_3 uptake. Ozone exposure was highly correlated with O_3 uptake as calculated by the statistical model, or

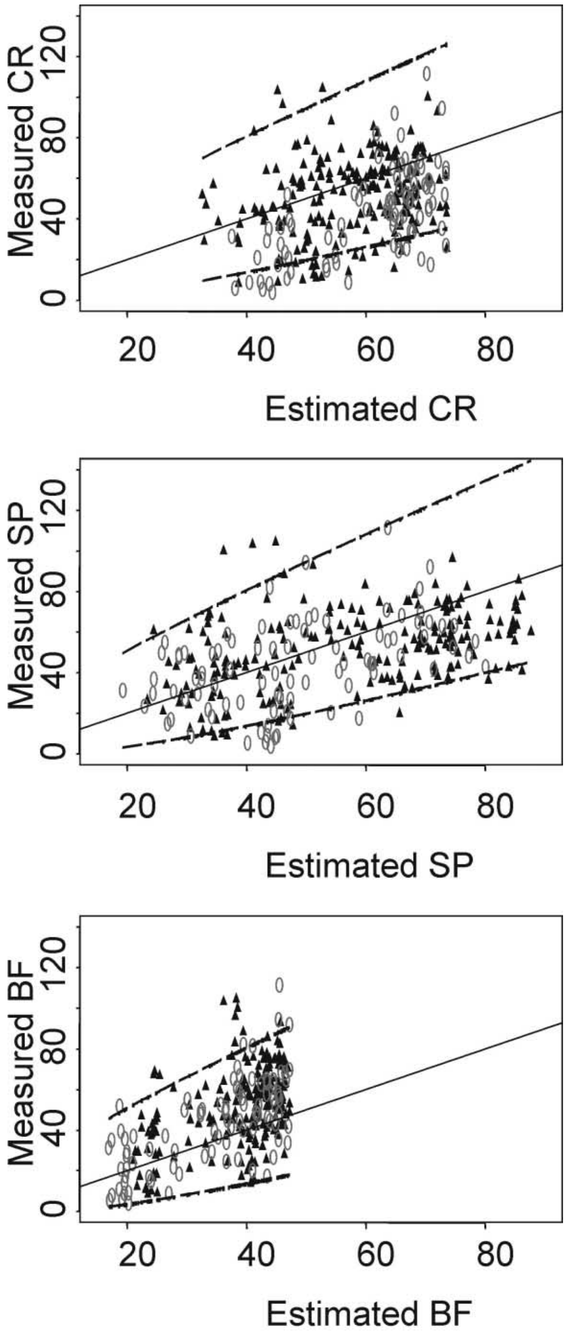

Fig. 4. Measured versus estimated stomatal conductance (for the same hour and day as measured values) in mmol H_2O $m^{-2}s^{-1}$ for all sites; 1993 (▲), 1994 (○). The dotted lines represent the 95th percentile about the regression line.

by TREGRO. In all cases, the adjusted r^2 was >0.92 and $P<0.0001$, determined for each site and year.

4. Discussion

Relative to more continental climates where the variability in occurrence and intensity of summer thunderstorms confound modeling efforts, estimates of gs in a summer-dry, mediterranean climate appear to be straight forward. The west coast (USA) variety of ponderosa pine appears to achieve a maximum gs in early summer, then declines until late September (Helms, 1970, 1971; Barnes, 1972; Coyne and Bingham, 1982; Temple and Miller, 1998; Grulke, 1999; Grulke and Retzlaff, 2001).

In the montane ecosystems of the Transverse Range, 95% of the annual precipitation is received over winter by mid May (San Bernardino County Water District). The winter precipitation recharges ground water. The date when upper soil horizons dry out varies from mid July to early August, depending on total overwinter ground water recharge (Hubbert, 1999). In a summer with above-average regional precipitation (1992) and summer rainstorms large enough to increase gs, the onset of drought stress was delayed by only one week relative to an average precipitation year without summer precipitation (1993; Temple and Miller, 1998). In a drought year, the recharge is less effective and the onset of physiological drought stress is three to four weeks earlier than in years of average precipitation (Grulke and Retzlaff, 2001; Grulke, 1999; Temple and Miller, 1998).

Phenological events in the west coast variety of ponderosa pine differ by only 10–15 days throughout its range, and its control is most likely initiated by photoperiod (Oliver and Ryker, 1990). Elongation growth is altered or truncated by drought stress. Elongation growth continues until approximately 2 weeks after the upper soil horizons dry out (Grulke and Retzlaff, 2001, northern California; Grulke et al., 1998, southern Californian trees). Near-surface soil drying contributes to increased water use efficiency (Grulke and Retzlaff, 2001). Perhaps this shift is mediated hormonally by [some] fine root die-off in the near surface soil horizons (via ABA, Ross et al., 1983; Christmann et al., 1995), because pre-dawn xylem potential in the needles does not reliably drop to a threshold level in every year (Grulke et al., 1998). Lower O_3 uptake was significantly correlated to declining soil moisture even at a pine plantation with relatively high soil water availability (Panek and Goldstein, 2001). Perhaps this is why this simple, statistical model appears to work for ponderosa pine in a mediterranean climate: day length (julian day in this model) and or drying soil signal the change in water use efficiency and decline of maximum daily gs in mid summer.

Site-specific statistical models for gs were based on time of day, day of year, hourly PPFD, and pre-dawn xylem potential in a year of average precipitation. Estimates of gs were compared to field measurements of gs in a year of average or 20% below-average precipitation. The median difference between estimated and measured gs was within 10 mmol H_2O $m^{-2}s^{-1}$ of measured gs in a given hour and julian day in either year.

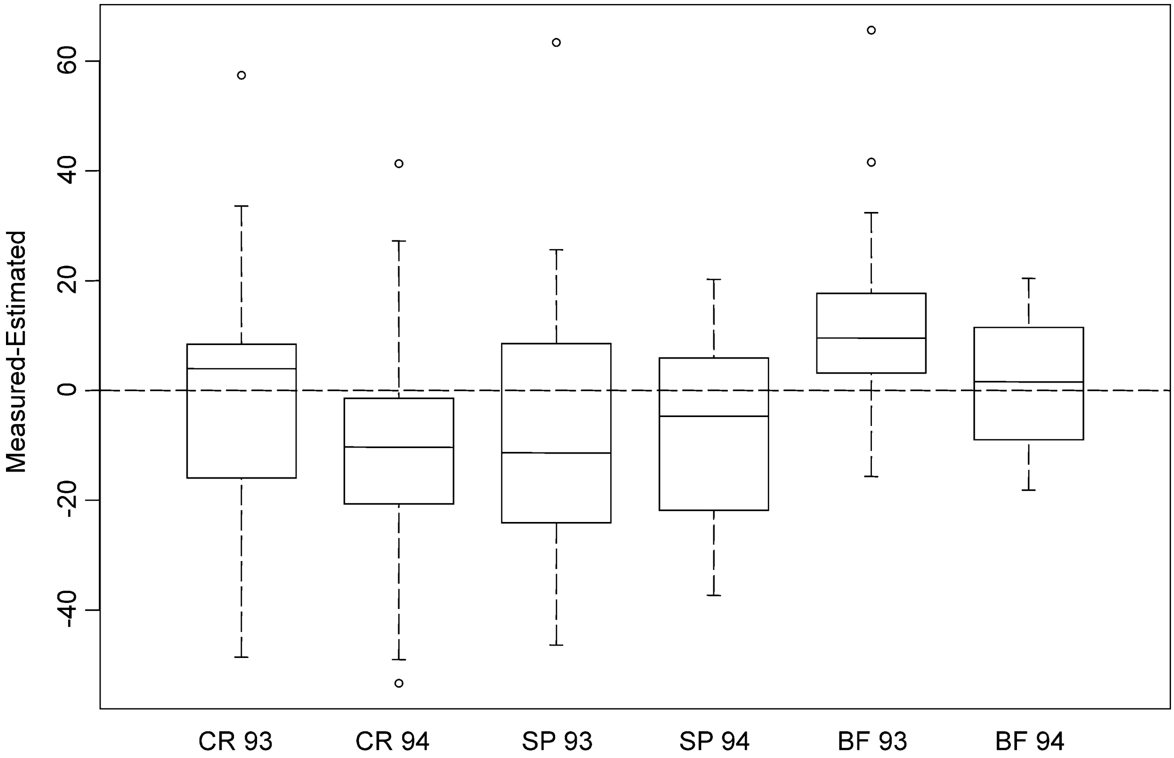

Fig. 5. Statistical summaries for the difference between [observed] and [estimated] stomatal conductance at each site during the 1993 and 1994 growing season. Stomatal conductance is given in mmol H_2O $m^{-2}s^{-1}$.

Table 3
Cumulative O_3 exposure and uptake at the three sites, calculated on a diurnal basis from 15 April through 15 October

Site	O_3 exposure	O_3 uptake (empirical)	O_3 uptake (TREGRO)	
Barton Flats				
1993	1993	270	4463	2698
1994	1994	277	4371	1367
Strawberry Peak				
1993	1993	328	5783	1779
1994	1994	292	5530	3243
Crestline				
1993	1993	340	6653	3101
1994	1994	338	6653	3448

Ozone exposure is reported in ppm h, and uptake is in μmol O_3 m^{-2}.

The statistical model gave lower gs values than measured in the average precipitation year (by 4 mmol H_2O $m^{-2}s^{-1}$), and higher values than measured in the below-average precipitation year (by 5 mmol H_2O $m^{-2}s^{-1}$). Winter precipitation after the drought in 1994 may not have been sufficient to recharge ground water supplies for the summer of 1995 at these sites, and thus contributed to these errors. However, the errors in the estimated gs are well within typical field measurement errors.

The best fit of observed versus fitted gs was at the moderate pollution site in a dry year ($r^2=0.75$). Gs in an average precipitation year at Barton Flats gave a poorer fit ($r^2=0.44$), although the relationship between observed and fitted gs was still highly significant ($P=0.0001$). The poorer fit indicates that additional explanatories were needed. At this site, VPD improved the fit in the afternoon in early summer (Grulke, 2000). In late summer, VPD was positively correlated with gs in mid morning, but was negatively correlated with gs in the afternoon. Both VPD and T_L co-varied with hourly PPFD. Because VPD became a poor explanatory with increasing pollutant exposure (e.g. at Crestline), it was not used in the general model.

Stomatal behavior with respect to light, VPD, and C_i was aberrant at the high pollution exposure site

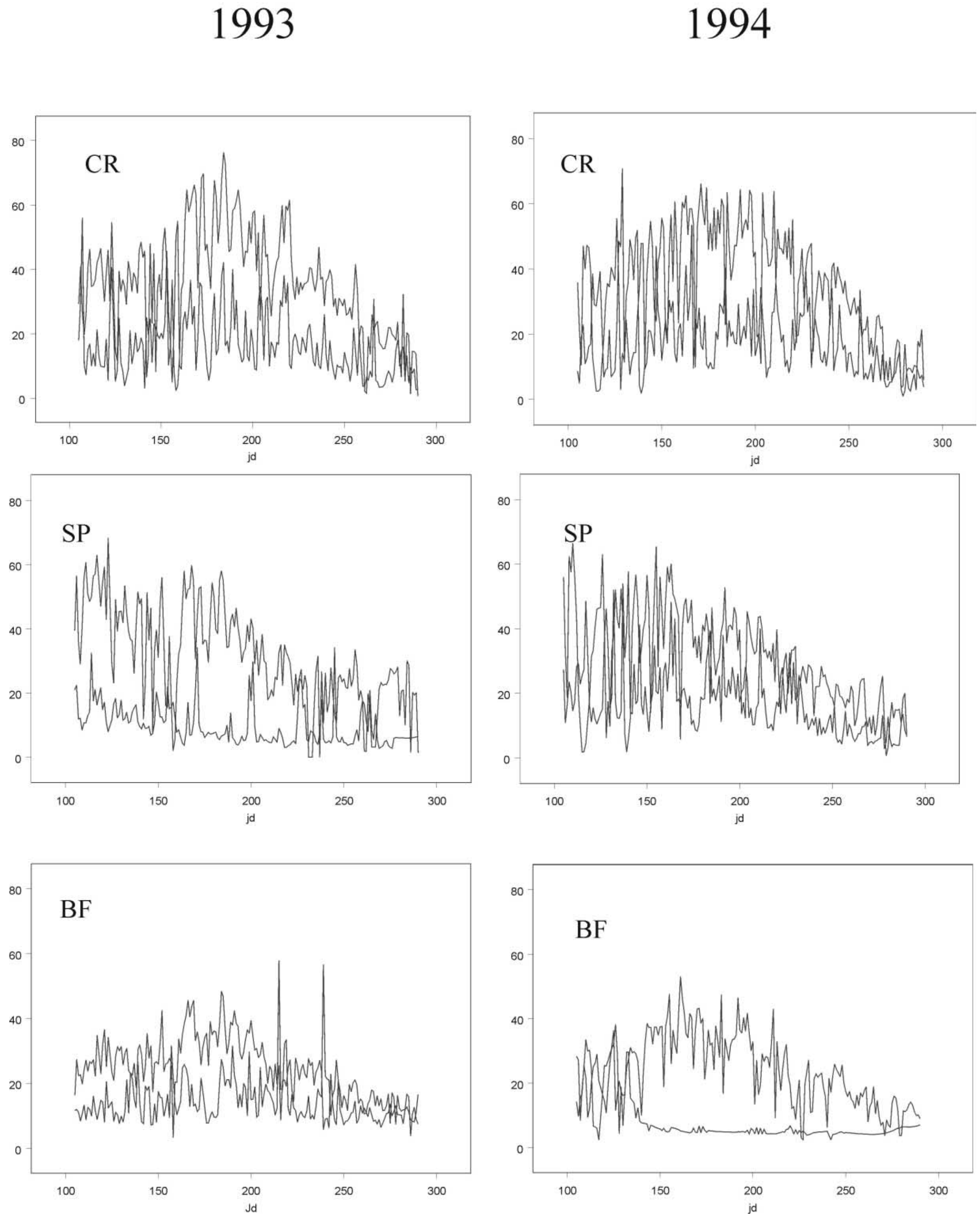

Fig. 6. Seasonal course of statistically estimated and simulated O_3 flux (in µmol O_3 $m^{-2}day^{-1}$) at the three sites for 1993 and 1994.

(Grulke, 1999). Although the median estimated gs was within 3 mmol H_20 $m-^2s^{-1}$ of field measurements at this site, only 14% of the variation in the data were explained by the statistical model in the average- ($P = 0.02$) or below-average precipitation year ($P = 0.03$). There is a clear need to better understand the mechanism of stomatal response to O_3 and other pollutant exposure under field conditions. Models developed in moderately polluted sites (e.g. <260 ppm h cumulative O_3 exposure) may not be applied with confidence to more polluted sites.

Diurnal gs was previously described with a trapezoidal model: gs ramps up quickly with exposure to light, remains high through much of the day, and then ramps down slowly at the end of the day (Temple and Miller, 1998). The data presented in this paper suggest a more parabolic diurnal response of gs. Mature yellow pine[2] had a total O_3 uptake for April 15 through October 15 of 8.5 in 1992, 4.5 in 1993, and 5.5 mmol O_3 m^{-12} in 1994 (Temple and Miller, 1998). The O_3 uptake estimates in 1993 and 1994 at the Barton Flats site in this study (4 km from Temple and Miller's study site) were comparable in 1993 but were 27% lower than their calculations in 1994. In that year, the 40-year-old trees in this study may have been more drought-stressed than mature trees in Temple and Miller (1998). Their estimates also suggest that between-year variability in O_3 uptake is much greater than between-site O_3 uptake presented in this paper. Their estimated O_3 uptake over the growing season was greater in 1992 (8.2 mmol O_3 m^{-2}) than estimates made at more polluted sites in this paper. In 1992, several precipitation events occurred during the summer which were large enough to wet the upper soil horizons to 15 cm at their site. These events influenced gs and thus their estimated uptake. The maximum daily uptake for ponderosa pine in 1992 (approximately 97 μmol O_3 m^{-2} day^{-1}) was similar to that reported for sugar maple in a short-term O_3 exposure experiment under high light (101 μmol O_3 m^{-2} day^{-1}; from Tjoelker et al., 1995, as calculated in Health and Taylor, 1997). Significant interannual differences in O_3 uptake were also reported for a ponderosa pine plantation in the central Sierra Nevada (Bauer et al., 2000).

Overall seasonal patterns of gas exchange, site O_3 concentrations, and weather conditions combined to influence the estimates of O_3 uptake across the pollution gradient. Patterns of O_3 uptake were site-specific and consistent between the two years when estimated by the statistical model. The most polluted site, Crestline, had intermediate O_3 uptake in early summer due to foggy mornings that persisted through mid June in both years. Higher O_3 concentrations during the time of the highest gas exchange rates at Crestline (mid June through early August) significantly contributed to increased O_3 uptake by ponderosa pine at that site. Ozone uptake at the moderately high pollution site, Strawberry Peak, was the highest of the three sites in early summer in both average and below-average precipitation years.

TREGRO calculated a much lower O_3 uptake for all sites relative to estimates generated by the statistical model. TREGRO simulated gs is proportional to A, but this may not be an appropriate assumption at moderately high and high pollution sites. One source of error in the model is how, where, and when the tree accesses soil moisture: the model does not account for deep water sources used by ponderosa pine after mid July (Hubbert, 1999). Insufficient soil moisture availability reduce *gs* and thus calculated O_3 uptake in the model. The most extreme example of this is statistically calculated versus simulated O_3 uptake at Barton Flats in 1994. Simulated O_3 uptake was low from mid July through the end of the growing season in 1994. According to the model, late summer drought stress at Barton Flats in 1994 was sufficient to effectively close stomata and significantly limit O_3 uptake. Field measurements in late summer indicate that gs in the below-average precipitation year was half that of an average precipitation year, but stomata were still open mid morning accompanying pre-dawn xylem potentials of−1.7 MPa. At Strawberry Peak, low temperatures in early summer, unaccounted for in the statistical model, may explain some of the differences in statistically calculated versus simulated O_3 uptake observed. Both the statistical model and TREGRO calculations of O_3 uptake were highly correlated to site O_3 exposure in both years.

Goldstein et al. (2000) estimated ponderosa pine plantation (whole ecosystem) O_3 uptake at approximately 80 mmol m^{-2} over the 6 month growing season in 1998. By harvesting trees from the site and measuring before and after ecosystem O_3 uptake, approximately one third of the ecosystem flux was attributable to tree uptake[3]. Ignoring O_3 uptake on woody tissue surfaces, and using the site leaf area index estimates of 6–9 m^{-2} (Bauer et al. 2000), their direct measures of O_3 uptake per m^{-2} of foliage surface area (3.0 to 4.4 mmol O_3 m^{-2}) at a site with moderate O_3 exposure are in the same range as those estimated by the statistical model (4.4–4.5 mmol O_3 m^{-2}) and calculated by TREGRO (1.4–2.7 mmol O_3 m^{-2}) for our moderate O_3 exposure site. Leaf level gs of plantation ponderosa pine was higher in early summer (Panek and Goldstein, 2001) relative to 40-year-old trees measured in this study, but was comparable in late summer to those in this study for the same tree age class (saplings) at the least polluted site (Grulke, 1999). Another mechanistic model based on the Farquhar model simulated low O_3 uptake relative to measured values in mid afternoon over a deciduous forest canopy (Amthor et al., 1994).

Although simple, the statistical model was effective at estimating seasonal gs at all sites. Refinements for adequately describing stomatal responses at the moderate pollution site are possible (Grulke, 2000), but responses in high O_3 exposure and N deposition sites will require several modifications to existing physiological models including changes in (1) guard cell membrane permeability to K^+ to accommodate temporal delays in stomatal response coupled to O_3 exposure, (2) enzyme kinetics for both rubisco and triphosphate export (see

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[2] Data from two Jeffrey and one ponderosa pine approximately 150 years old were combined for the response.

[3] Dominated by ponderosa pine, but also included Douglas fir, white fir, giant sequoia, incense-cedar, and California black oak.

Heath and Taylor 1997; Grantz and Farrar, 2000), and (3) nitrogen deposition modifications to antioxidant systems (Polle, 1998) which will likely alter a threshold response (Martin et al., 2001).

Ozone exposure is still not considered regularly as a background stressor in ecological field studies (Grulke et al., 2001). Industrial activity resulting in the release of O_3 precursors over the past century has doubled the global background O_3 concentrations from 20–40, to 40–60 ppb, a trend that is expected to continue into the next millennia at approximately 1% year^{-1} (National Academy of Science, 1992). An hourly O_3 exposure of 60 ppb h translates to the moderate O_3 exposure described in this paper of 260 ppm h accumulated over the growing season. Areas influenced by air transport from urban areas would of course be exposed to higher levels, and more complex mixtures of pollutants. Despite the ability of air quality regulators to limit peak hourly O_3 concentrations, cumulative O_3 exposure will continue to increase to state or federal limits as population and fossil fuel consumption continues to increase. Improved models to handle moderately high and high O_3 exposures, and its interaction with other environmental stressors would aid risk assessments to maintain future forest health.

Acknowledgements

This research was funded by US Environmental Protection Agency agreement, DW-12934530, but has not been subjected to the Agency's review and therefore does not necessarily reflect the view of the Agency. No official endorsement should be inferred. Field data for this research was primarily collected by N.G., Lisa Balduman, Janet Kirsch, and Cathy Rose. Special thanks to Bill Hogsett, who supported this research, and to Paul Miller who did much of the initial research in the effects of pollution on ponderosa pine at these sites.

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