Environmental Pollution 207 (2015) 21–30

Contents lists available at ScienceDirect

Environmental Pollution

journal homepage: www.elsevier.com/locate/envpol

Concentration- and flux-based ozone dose–response relationships for five poplar clones grown in North China

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ARTICLE INFO

Article history:
Received 25 June 2015
Received in revised form
14 August 2015
Accepted 20 August 2015
Available online xxx

Keywords:
Stomatal conductance
Ozone flux
Dose–response relationship
Critical level
Poplar

ABSTRACT

Concentration- and flux-based O_3 dose–response relationships were developed for poplars in China. Stomatal conductance (g_s) of five poplar clones was measured to parameterize a Jarvis-type multiplicative g_s model. The maximum g_s and other model parameters varied between clones. The strongest relationship between stomatal O_3 flux and total biomass was obtained when phytotoxic ozone dose (*POD*) was integrated using an uptake rate threshold of 7 nmol m^{-2} s^{-1}. The R^2 value was similar between flux-based and concentration-based dose–response relationships. Ozone concentrations above 28–36 nmol mol^{-1} contributed to reducing the biomass production of poplar. Critical levels of AOT_{40} (accumulated O_3 exposure over 40 nmol mol^{-1}) and POD_7 in relation to 5% reduction in total biomass for poplar were 12 μmol mol^{-1} h and 3.8 mmol m^{-2}, respectively.

1. Introduction

Tropospheric ozone (O_3) is an important phytotoxic air pollutant and a significant greenhouse gas formed by photochemical reactions among nitrogen oxides, volatile hydrocarbons and carbon monoxide in the atmosphere (Dumont et al., 2014; Feng et al., 2015). The background O_3 level over the mid latitudes of the Northern Hemisphere has increased continuously between 0.5% and 2% per year over the last 30 years (Vingarzan, 2004). It is predicted to rise up to 80 nmol mol^{-1} in 2100, accompanied by episodes of O_3 peaks occasionally exceeding 200 nmol mol^{-1} (IPCC, 2013; Richet et al., 2012).

Forest ecosystems, which cover 31% of the Earth's land area and constitute the most important carbon sinks, could be highly vulnerable to O_3 damage (FAO, 2010; Fowler et al., 2009; Sitch et al., 2007). Ozone affects tree health through stomatal uptake, causing visible foliar injury, accelerated leaf senescence, reduced photosynthesis, and impaired stomatal function (Feng et al., 2014; Zhang et al., 2011, 2014b). The increasing ozone concentration contributes to a decline in forestry productivity linked to economic losses (Felzer et al., 2007; Percy and Karnosky, 2007; Pye, 1988). A variety of detrimental changes at the biochemical, physiological and molecular levels have been demonstrated in conifers and deciduous trees (Koch et al., 1998; Kopper and Lindroth, 2003; Richet et al., 2011, 2012; Zhang et al., 2014a). Experimental evidence indicates that not only species but also cultivars, genotypes and clones, show different sensitivity to O_3 (Calatayud et al., 2011; Castagna et al., 2015; Dumont et al., 2014; Häikiö, 2009; Paoletti and Grulke, 2010; Zhang et al., 2012, 2014c).

In the past, the impact of O_3 on vegetation was quantified using the dose–response relationship based only on atmospheric O_3 concentrations, such as AOT_{40} (accumulated hourly O_3 concentration over a threshold of 40 nmol mol^{-1} during daylight hours) (Fuhrer et al., 1997). Nowadays, it has been gradually replaced by the POD_Y (phytotoxic ozone dose over a threshold of *Y*) index defining the amount of O_3 entering the leaves through the stomata (Gerosa et al., 2015; LRTAP Convention, 2010; Mills et al., 2011). This latter index, which has been described using different names in the

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http://dx.doi.org/10.1016/j.envpol.2015.08.034
0269-7491/

literature, such as *CUO* (cumulative uptake of O_3), or *AFstY* (accumulated stomatal flux above a threshold of *Y*) (Azuchi et al., 2014; Uddling et al., 2004), takes into account the O_3 flux by modeling O_3 stomatal conductance (g_{sto}) using multiplicative algorithms, such as Jarvis (1976).

It is generally accepted that the damage of O_3 to plants is mainly caused by the amount of O_3 entering into the leaf interior through the open stomata (Kerstiens and Lendzian, 1989). Stomatal conductance of plants is one of the crucial mechanisms of O_3 sensitivity (Dumont et al., 2013; Guidi et al., 2001). Stomatal O_3 uptake considers both biological traits and climatic factors. Therefore, it is better for assessing the adverse effects of O_3 on the plants than external O_3 concentrations (Emberson et al., 2000b; Uddling et al., 2004).

Among different tree species, poplars have received particular attention because of the fast growth rate and relatively high stomatal conductance (Woo and Hinckley, 2005). There are 53 species of poplar distributed in 22 provinces of China (not including crossbreed and imported species), covering a total area of over 10 million ha with a total standing stock of 426 million m^3 (Liang et al., 2006; Xu et al., 2009). Different species or clones of poplar have shown different sensitivity to O_3 with respect to visible leaf injury (Hoshika et al., 2012; Novak et al., 2005; Ryan et al., 2009; Strohm et al., 1998), damaged photosystems (Bernacchi et al., 2003; Guidi et al., 2001; Ranieri et al., 2001), and reduced growth (Isebrands et al., 2001; Matyssek et al., 1993; Mooi, 1980). However, the knowledge of O_3 dose–response relationships for poplar, which could be an effective tool for O_3 risk assessment, is still limited. It was hypothesized that the difference in O_3 sensitivity could lead to different parameter values of stomatal conductance model for different poplar species or clones.

The overall objectives of the present study were: (1) to parameterize the stomatal conductance model of poplar clones widely used in China; (2) to develop exposure concentration and flux-based O_3 dose–response relationships; and (3) to define the critical levels for protecting poplar against O_3 damage.

2. Materials and methods

2.1. Experimental site and plant materials

The experiment was conducted in Changping (40°19′N, 116°13′E), northwest of Beijing in a warm temperate and semi-humid continental climate. The annual mean temperature in Changping is 11.8 °C and the total annual precipitation is 550 mm.

Rooted cuttings of five hybrid poplar clones were cultivated at the Chinese Academy of Forestry Sciences. The following clones were used: ‘84 K’ (*Populus alba* × *P. glandulosa*), ‘107’ (*P.* × *euramericana* cv. ‘74/76’), ‘90’ (*P. deltoides* × *P. cathayana* cv. Senhai 2), ‘546’ (*P. deltoides* cv. ‘55/56’ × *P. deltoides* cv. ‘Imperial’), and ‘156’ (*P. deltoides* × *P. cathayana*). The cuttings were transplanted into 20 L circular plastic pots on April 20, 2014 when they were about 65 days old. Pots were filled with local light loamy farmland soil. Seedlings with similar height and stem basal diameter (Table S1 and S2) were selected for this study and pre-adapted to open-top chamber conditions for 10 days before O_3 fumigation. All plants were manually irrigated up to soil field capacity with tap water at 1–2 days interval to avoid drought stress.

When most leaves turned yellow and the growth of height and basal stem diameter had stopped, the plants were harvested and separated into leaves, stems, and roots. All plants were harvested between Sep. 13 and 28, 2014 (see details in Table S3). The plant organs were oven-dried at 80 °C until a constant mass was reached.

2.2. Ozone treatments

The experiment was conducted in six open-top chambers (OTC, octagonal base, 12.5 m^2 of growth space and 3.0 m of height, covered with toughened glass) with different treatments: charcoal filtered ambient air (CF), non-filtered ambient air (NF), and NF with targeted O_3 addition of 20 (NF+20), 40 (NF+40), 60 (NF+60), and 80 (NF+80) nmol mol^{-1} for 8-h average O_3 concentration. Ozone was generated from pure oxygen by an O_3 generator (HY003, Chuangcheng Co., Jinan, China) and then mixed with ambient air using a fan (1.1 kW, 1080 Pa, 19 m^3 min^{-1}, CZR, Fengda, China) to achieve the target O_3 concentration at the top of the canopy in the fumigation treatments. There was no chamber replication for each treatment. In order to eliminate the positional and chamber effects (Potvin and Tardif, 1988), the plant positions within each OTC were changed every 3–5 days, and all seedlings were switched between chambers once a month, according to Feng et al. (2011). Each chamber functioned as one of six treatments randomly at each month during the growing season. The concentration in each chamber was changed every month according to the corresponding treatment.

Six to eight potted plants per poplar clone were exposed to each treatment except there was no ‘156’ in the NF+20 chamber. The daily maximum fumigation period was 9 h (from 08:30 to 17:30) when there was no rain, fog, mist, or dew, according to the protocols in Free Air Ozone Concentration Enrichment System (Feng et al., 2010; Morgan et al., 2006).

2.3. Ozone and climate monitoring

The concentrations of O_3 in the OTCs were continuously monitored using an UV absorption O_3 analyzer (Model 49i, Thermo Scientific, USA), via a Teflon solenoid valve switch system, which collected air from sampling points at approximately 10 cm above the plant canopy. The monitors were calibrated by a 49i-PS calibrator (Thermo Scientific, USA) before the experiment and once a month during the experiment. Fumigation targets and average O_3 concentrations (24 h, 12 h, and 8 h) for all treatments are presented in Table 1.

Air temperature (*T*), relative air humidity (*RH*), and photosynthetic photon flux density (*PPFD*) under the ambient condition and inside the OTCs were measured every 5 min using a weather station (Campbell Scientific, North Logan, Utah, USA). Water vapor pressure deficit (*VPD*) was estimated using Eq. (1):

$$VPD = \left(1 - \frac{RH}{100}\right) \times 0.611 \times \exp\left(\frac{17.502 \times T}{T + 240.97}\right). \tag{1}$$

Hourly means of environmental variables were employed in O_3 flux calculations. The range of *T*, *VPD*, and *PPFD* was 6.94–46.48 °C, 0.00–9.15 kPa, and 0–1358 μmol m^{-2} s^{-1}, respectively, from May 7 to Sep. 29, 2014. Compared with ambient air, the average air temperature and relative humidity in the OTCs were increased by 1.52 °C and 1.65%, respectively.

Table 1
O_3 concentrations (nmol mol^{-1}, mean ± SE) during the experiments from May 7 to Sep. 29, 2014.

Target	24-h average	12-h average, 08:00–20:00	8-h average, 09:00–17:00
CF	25.7 ± 0.9	28.1 ± 1.1	28.0 ± 1.3
NF	35.7 ± 1.2	48.4 ± 1.6	53.2 ± 1.8
NF+20	41.4 ± 1.3	59.4 ± 1.7	67.1 ± 1.9
NF+40	46.3 ± 1.4	69.4 ± 2.0	79.5 ± 2.3
NF+60	52.8 ± 1.6	81.9 ± 2.5	95.4 ± 2.9
NF+80	58.1 ± 1.8	92.4 ± 2.8	108.7 ± 3.3