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Dose-response relationships for ozone effect on the growth of deciduous broadleaf oaks in mediterranean environment

Riccardo Marzuoli[a,*], Filippo Bussotti[b], Vicent Calatayud[c], Esperanza Calvo[c], Rocío Alonso[d], Victoria Bermejo[d], Martina Pollastrini[b], Robert Monga[a,e], Giacomo Gerosa[a]

[a] Dept. of Mathematics and Physics, Università Cattolica del Sacro Cuore, via Musei 41, Brescia, Italy
[b] Dept. of Agri-Food Production and Environmental Sciences (DISPAA), Università degli Studi di Firenze, Piazzale delle Cascine 18, Firenze, Italy
[c] Fundación CEAM, c/ Charles R. Darwin 14, Parque Tecnológico, 46980, Paterna, Valencia, Spain
[d] Ecotoxicology of Air Pollution, CIEMAT, Avda. Complutense 40, Madrid, Spain
[e] Département de Gestion des Ressources Naturelles Renouvelables, Université de Lubumbashi, Democratic Republic of the Congo

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ABSTRACT

This study presents a reanalysis of ozone (O_3) exposure experiments performed on deciduous broadleaf oak species in the Mediterranean region and a proposal of critical levels to improve the O_3 risk assessment in this area for these widely distributed forest species. Two experiments performed in Spain and Italy were considered, and the following 3 oak species were studied: *Quercus pyrenaica*, *Q. faginea* and *Q. robur*. All the experiments were performed with irrigated potted seedlings growing in Open-Top Chambers exposed to different O_3 levels (with charcoal-filtered air as the control treatment) for two consecutive growing seasons. The Phytotoxic Ozone Dose above an instantaneous threshold of 1 nmol O_3 m^{-2} s^{-1} (POD_1) was calculated by applying a Jarvis type model for the estimation of the stomatal conductance (g_s), and by adopting a big-leaf resistive scheme to account for the O_3 deposition on the vegetation. Two parameterisations were used for the g_s multiplicative model: one species-specific based on the "local" g_s measurements performed during each experiment, and the other "generic" based on the "Deciduous Mediterranean broadleaf" parameterisation described in the Manual on Methodologies and Criteria for Mapping Critical Loads and Levels and Air Pollution Effects, Risks and Trends of the UN/ECE (CLRTAP, 2015). The two different parameterisations were used to derive dose-response functions and ozone critical levels for the biomass loss of the deciduous oak species. The dose-response functions for roots and total biomass were statistically significant, with both the parameterisations tested ($p < 0.05$). The O_3 critical levels obtained indicate that deciduous broadleaf oaks in Mediterranean environment could be more tolerant to O_3 than other European broadleaf species and that O_3 is more harmful to the below-ground biomass of the plants rather than the above-ground biomass.

1. Introduction

Tropospheric ozone pollution is one of the major threats for forests in the Mediterranean region, because climatic conditions and anthropogenic emissions of O_3 precursors tend to facilitate its photochemical formation and persistence during the spring/summer period. As a consequence, O_3 levels in this area frequently exceed the target values established by the 2008/50/CE directive to protect vegetation (EEA, 2013; Cristofanelli and Bonasoni, 2009; Proietti et al., 2016).

The negative effects of O_3 on the physiology and growth of several forest species have been well documented in the last several decades, and many studies have highlighted detrimental effects such as visible leaf injuries (Gerosa et al., 2008; Marzuoli et al., 2009; Feng et al., 2014), accelerated leaf senescence (Gielen et al., 2007), reductions in stomatal conductance (g_s) and photosynthesis (Wittig et al., 2007), and declines in productivity (Wittig et al., 2009; Li et al., 2017).

Although these effects have been studied and characterized in controlled or semi-controlled experiments mostly performed with potted young trees, there is also evidence that O_3 can affect mature trees in field conditions (Matyssek et al., 2007; Karnosky et al., 2005; Braun et al., 2010, 2014). Moreover, some authors found that the physiological and growth responses to O_3 of potted woody plants do not significantly differ from those of field-grown plants (Oksanen, 2003a; Coleman et al., 1996), although the pot volume under some circumstances can be an important modifying factor for plants response to abiotic stresses (Poorter et al., 2012).

* Corresponding author.
E-mail address: riccardo.marzuoli@unicatt.it (R. Marzuoli).

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The Convention on Long-Range Transboundary Air Pollution (CLRTAP/UNECE) has proposed O_3 critical levels to assess the risk of O_3 effects in European vegetation and to support emission reduction protocols for O_3 precursors (Mills et al., 2011). Critical levels are defined as the cumulative exposure or dose above which direct adverse effects on sensitive receptors (i.e. vegetation) may occur according to present knowledge (CLRTAP, 2015). The most biologically meaningful approach for setting O_3 critical levels to protect forest trees is based on the quantification of the O_3 flux entering the leaves through stomata, i.e. the Phytotoxic Ozone Dose (POD) absorbed by plants per unit leaf area. The response variable typically used is the reduction in the mean annual growth of plants, using changes in the whole tree biomass as an indicator of growth. A large amount of evidence has shown a better correlation between the O_3 stomatal flux, rather than the simple O_3 exposure, and the total biomass loss of forest species (Karlsson et al., 2004; Uddling et al., 2004; Fares et al., 2013; Alonso et al., 2014; Gerosa et al., 2015; Hu et al., 2015; Gao et al., 2017).

The divergence between the relatively moderate O_3 effects observed in Southern Europe forests and the expected effects estimated with the available exposure-based and flux-based dose-response relationships for deciduous trees (Mills et al., 2011) is likely due to the uncertainty of the current dose-response relationships for assessing the O_3 effects on Mediterranean forest trees and/or the higher O_3 tolerance of Mediterranean vegetation (Bussotti and Gerosa, 2002; Alonso et al., 2014). Current critical levels for broadleaf trees (CLRTAP, 2015) are based on experiments conducted mainly in Northern and Central Europe which are characterized by very different climatic conditions and forest species composition (Büker et al., 2015; Karlsson et al., 2004; Oksanen, 2003b).

Therefore, specific dose-response functions and critical levels for deciduous trees growing in Mediterranean environment are needed and are expected to be more consistent with field evidence of O_3 impacts. In any case, it has to be taken into account that under field conditions response indicators of O_3 like growth can be influenced by many factors that render the quantification of the contribution of O_3 to the observed responses very difficult (Ferretti et al., 2007).

The present study is a reanalysis of the available data from experiments on O3 effects on the growth of deciduous broadleaf oaks in Mediterranean environment. The main objective of the work was to develop new dose-response functions and O_3 flux-based critical levels for the O_3 risk assessment within the CLRTAP. A comparison between the performance of two different g_s parameterisations for the POD calculation and the dose-response function definition is also presented and discussed.

This work was conducted as a part of the revision process of the "Chapter 3" of the "Manual on Methodologies and Criteria for Mapping Critical Loads and Levels and Air Pollution Effects, Risks and Trends" (Mapping Manual from now on; CLRTAP, 2015).

2. Material and methods

2.1. Data-set establishment

Data from two experiments involving deciduous *Quercus* spp. performed in Valencia (Spain) and Curno (Italy) in 2006–2007 and 2012–2013, respectively, were used to derive flux-based response functions for biomass losses in deciduous oak species. The experiments were characterized by very similar methodologies and experimental design (Table 1), and had one oak species (*Q. robur*) in common. In both experiments plants originated from seeds of the same local provenience.

Two-year-old seedlings growing in Open-Top Chambers (OTCs) were exposed to 2 and 4 different O_3 treatments in Spain and Italy, respectively. The control treatments in the experiments were charcoal-filtered OTCs, which provided an abatement of between 40 and 50% of the ambient O_3. Ozone fumigation systems provided an increase of O_3 concentrations inside some of the OTCs by approximately 30 ppb in the NF + 30 treatments in Spain, and by 30% and 75% of the ambient O_3 in Italy in the NF + and NF + + treatments, respectively. The average O_3 concentrations and AOT40 exposure index (Accumulated Ozone over a Threshold of 40 ppb) for each O_3 treatments are reported in Table 2. The AOT40 index was calculated by summing up all the exceedances of the hourly O_3 concentrations above 40 ppb during the daylight hours (8 a.m. - 8 p.m., local time) from 1st April to 30th September.

Table 1
Summary of the O_3 exposure experiments on deciduous broadleaf oak species performed in the Mediterranean environment and considered for this study. CF = Charcoal-Filtered air; NF = Non-Filtered air; NF + 30 = Non-Filtered air + 30 ppb of O_3; NF + = Non-Filtered air + 30% of ambient O_3; NF + + = Non-Filtered air + 75% of ambient O_3.

	Valencia (Spain)	Curno (Italy)
Years of experiment	2006–2007	2012–2013
Duration	18 months	18 months
Species	*Quercus robur, Q. faginea; Q. pyrenaica*	*Quercus robur*
Plants age at the beginning of the exp.	2 years	2 years
O_3 treatments	CF, NF + 30	CF, NF, NF +, NF + +
Pots volume	10 L	15 L
Plants number for species/ OTC	9	5
N° of replicates for each O_3 level	3	3
Fumigation period	25 Apr – 19 Sep 2006 2 May – 14 Sep 2007	20 Apr – 26 Sep 2012 6 May – 13 Sep 2013
Reference	Calatayud et al. (2011)	Marzuoli et al. (2016)

Details on the experiments can be found in Calatayud et al. (2011) and Marzuoli et al. (2016), while meteorological and climatic conditions during the growing seasons of the experiments are reported in Table 3.

2.2. Calculation of O_3 stomatal fluxes and Phytotoxic Ozone Dose (POD)

Ozone fluxes at the leaf level were calculated according to the methodology suggested by the Mapping Manual (CLRTAP, 2015).

Hourly g_s to O_3 was calculated using the multiplicative approach proposed by Jarvis (1976) and modified by Emberson et al. (2000) and included in the Mapping Manual with the following formulation:

$$g_s = g_{max} \times f_{phen} \times f_{light} \times \max\{f_{min}; (f_{temp} \times f_{VPD} \times f_{SWP})\}$$

where g_{max} represents the maximum g_s to O_3 (derived from the g_{max} to water) exhibited by plants under optimal conditions, and the f functions (all ranging from 0 to 1) describe the relative effect of leaf phenology (f_{phen}) and environmental conditions such as photosynthetically active radiation (f_{light}), air temperature (f_{temp}), air vapour pressure deficit (f_{VPD}), and soil water potential (f_{SWP}) on g_{max}. All the modifying functions are described in detail in CLRTAP (2015). The values of g_{max} and the modifying functions were defined according to two different parameterisations: a local species-specific parameterisation based on field g_s measurements performed during the experiments, and a generic parameterisation for deciduous broadleaf in Mediterranean Europe provided in Table A2.6 of the Mapping Manual (CLRTAP, 2015) and mainly based on data of beech (Aranda et al., 2000; Mediavilla and Escudero, 2003; Nunn et al., 2005).

The modifying function related to the soil water potential (f_{SWP}) was set to 1 for both parameterisations because in both experiments, plants were irrigated, and the soil water content was maintained close to the field capacity, avoiding any water stress. The local and generic parameterisations for the g_s model are presented in Table 4.

The meteorological data recorded inside the OTCs and collected