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Assessing the negative effects of severe powdery mildew infection on chlorophyll *a* fluorescence and stomatal characteristics of *Quercus robur* L.

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INTRODUCTION

Pedunculate oak (*Quercus robur* L.) is a widespread tree species in European forests with high economic benefits due to the quality of timber. However, in the last few decades, a diminishing in the vitality of *Q. robur* stands related to biotic and abiotic stress factors, has been observed (Thomas et al., 2002). In case of biotic stress factors, oak powdery mildew (*Erysiphe alphitoides* (Griffon and Maubl.) U. Braun and S. Takam.)) has been recognized as one of the most important and widespread foliar pathogenic organisms for oaks (Pap et al., 2014).

The present study was meant to investigate the effectiveness of chlorophyll *a* fluorescence kinetics and chlorophyll *a* fluorescence transient in combination with the stomatal traits for the evaluation and monitoring of severe biotic stress levels caused by powdery mildew fungi on one-year-old *Q. robur*.

MATERIAL AND METHODS

To assess, evaluate and monitor the negative effects of oak powdery mildew, an experiment in semi-controlled conditions was set up and constituted of 10 one-year-old disease-free seedlings of *Quercus robur* L., and the same number of seedlings with 50% and 75% of leaf coverage by epiphytic mycelia of oak powdery mildew. During the experiment, the infection progressed by its natural course, and in the end, the leaves of the first infected treatment group have reached a coverage of 75% with epiphytic mycelia, and the second infected group has progressed to 100% leaf infection coverage.

Fast chlorophyll fluorescence induction curves (OJIP) were recorded with a PAM-2500 portable chlorophyll fluorometer (Walz, Germany). Slow chlorophyll fluorescence kinetics were assessed using the automatic rapid light curve function of the PAM-2500 portable chlorophyll fluorometer (Walz, Germany). The following parameters were deduced by the OJIP test analysis of chlorophyll *a* fluorescence transient: fluorescence value at the J-step (F_J) and I-step of OJIP (F_I), maximum variable fluorescence from dark adapted leaf (F_v), maximum quantum yield of primary PSII photochemistry (Φ_{Po}), efficiency of the water-splitting complex on the donor side of PSII (F_V/F_0) and performance index on absorption basis (Φ_{Po}). In terms of slow chlorophyll fluorescence kinetics the following parameters were derived: quantum yield of non-regulated heat dissipation and fluorescence emission (Y(NO)), Stem-Volmer type non-photochemical fluorescence quenching (NPQ), relative electron transport rate (ETR), coefficient of non-photochemical fluorescence quenching (qN) and coefficient of photochemical fluorescence quenching (qP).

Stomatal imprints were made using the collodion method according to the described protocol by Stojnić et al., (2018). The following stomatal characteristics were assessed: stomatal density per mm^2 (SD), stomata guard cell length (L_A [µm]) and width (W_B [µm]), and stomatal aperture length (L_a [µm]) and width (W_b [µm]).

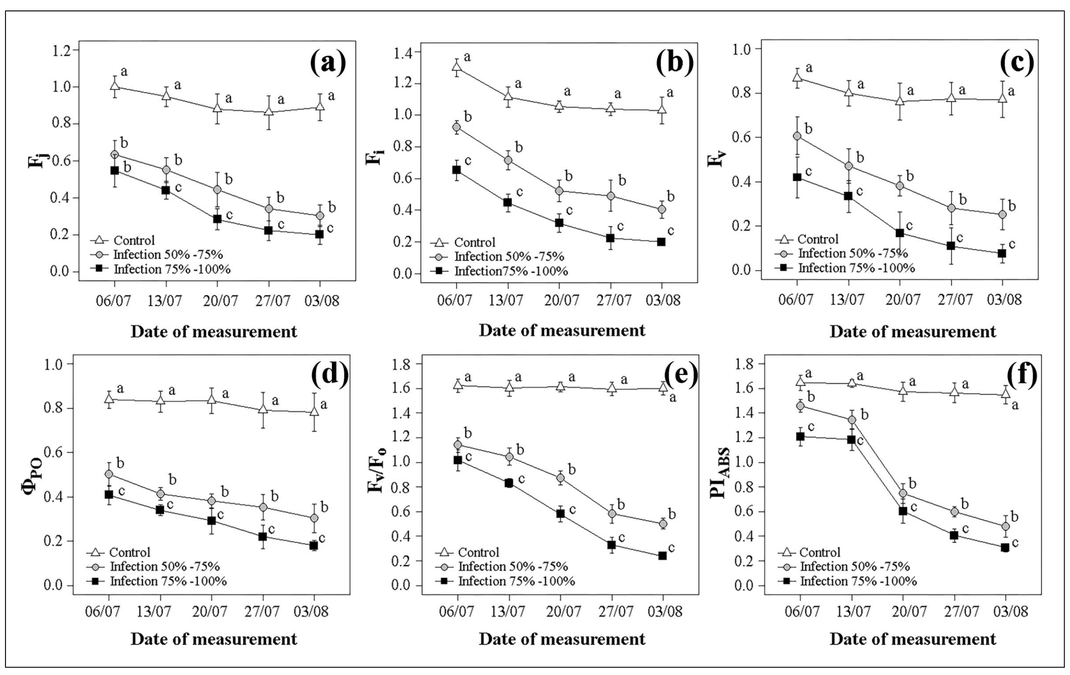

FIGURE 1: Progress curves of chlorophyll *a* fluorescence transient parameters measured on leaves of *Q. robur* seedlings.

RESULTS

All parameters of chlorophyll *a* fluorescence transient were significantly influenced by the studied obligate leaf parasite, in both of the studied treatment groups, detected during the first round of conducted measurements (Figure 1). We observed a significant reduction of the Φ_{Po} parameter, compared to the control plants, in both of the treatment groups. While the values of the control group were between 0.75 and 0.85, a range documented for healthy plants, the first treatment group exhibited a decline of 41.9% and the second a 53.5% decline during the first measurements. At the end of the experiment, the reduction reached 61.2% and 77.1% for *Q. robur* leaves infected 75% and 100% with *E. alphitoides* respectively (Figure 1d).

Concerning the parameters of slow fluorescence kinetics, we observed that Y(NO) showed an increasing trend, meanwhile NPQ, ETR, qN and qP were decreased under severe biotic stress caused by *E. alphitoides* (Figure 2).

Regarding stomatal characteristics, we detected that infection by *E. alphitoides*. caused a significant increase in SD and a simultaneous reduction in Wb. In case of SD, the highest values were observed in leaves covered 75% by mycelia at the start of the measurements, as well as at the end of the experiment, when the mycelia reached 100% of leaf coverage.

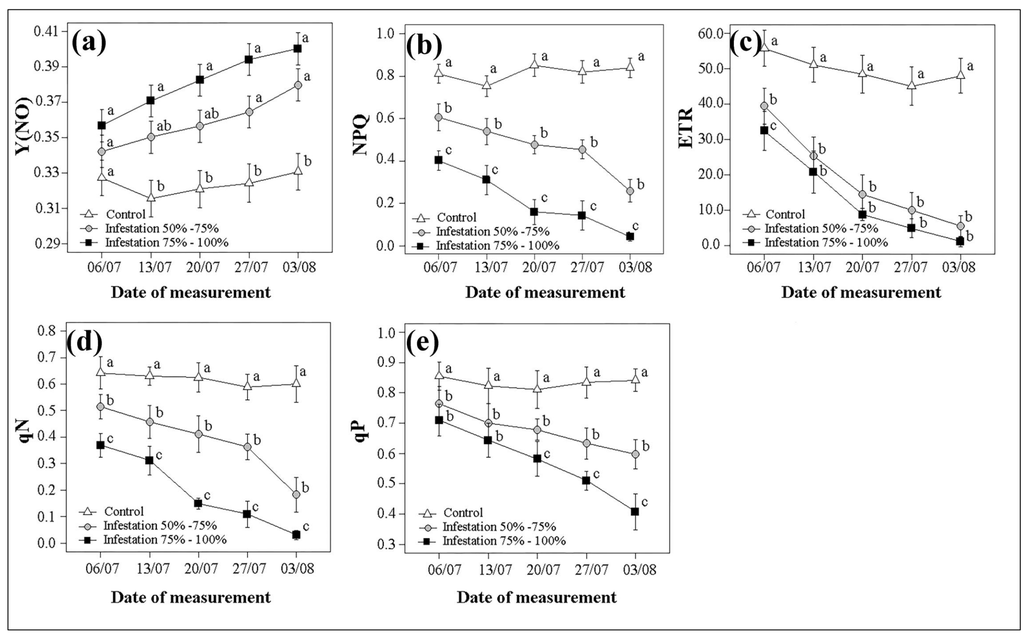

FIGURE 2: Progress curves of slow fluorescence kinetics parameters measured on leaves of *Q. robur* seedlings at high light conditions (1389 µmol (photon) m^{-2} s^{-1}).

CONCLUSIONS

The results of the present study point to a disturbance in the photosynthetic apparatus under biotic stress caused by *E. alphitoides*, which progressed in time and depended on the severity of the infection. Significant negative effects of the studied pathogen on leaf physiology were detected by all of the observed parameters of the chlorophyll *a* fluorescence transient, showing its effectiveness in evaluation and monitoring of severe biotic stress.

Furthermore, parameters of slow fluorescence kinetics were considerably influenced as well, with qN and NPQ parameters showing the fastest responses.

Concerning the harmful effects of the studied pathogen on stomatal traits, we detected a significant increase of SD followed by a simultaneous reduction of W_b, as a protective mechanism of *Q. robur* against the stressor.

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