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VOLATILE ORGANIC COMPOUND EMISSIONS AND PHOTOSYNTHETIC PARAMETERS OF *QUERCUS RUBRA* UNDER TEMPERATURE STRESSES

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Abstract

Red oak (Quercus rubra) is a fast growing tree which could be met in a various forests in Central Europe and North America. In a normal condition, it can live until 300-years and can be 5-6 m tall. Anyway due to the climate change, the oak forest is affected by stress conditions including long period of high temperature and drought.
Regarding to volatile organic compounds (VOC) emission, Quercus rubra is a high isoprene and low monoterpenes emitted. The emission is affected by different stress which can be a sensitive signal of stress responses especially in heat stress which can particularly significantly influence all the metabolic processes of the deciduous trees. Our work has been shown that long term heat stress treatments affect foliage photosynthesis as well as different terpene emissions. Under non-stressed conditions, lipoxygenase pathway products LOX emissions were close to detection limit, while in stress condition those compounds emission enhanced.

Key words*: volatile organic compounds emission, Quercus rubra, .plant stress.*

INTRODUCTION

Almost all plant species are emitters of different volatile organic compounds (VOC). The term *biogenic volatile organic compounds* (biogenic VOCs or BVOC) includes organic atmospheric trace gases other than carbon dioxide and monoxide (Kesselmeier and Staudt, 1999). Volatile isoprenoids – isoprene (5 carbon atoms, C5) and volatile terpenes consisting of isoprene building blocks, monoterpenes, (C10), homoterpenes, sesquiterpenes (C15, SQT), diterpenes (C20), triterpenes (C30), tetraterpenes (C40), polyterpenes (C>40) fatty acid derivatives, benzenoids, alcohols, aldehydes (C5 and C6), ketones, phenylpropanoids, and amino acid derived metabolites – form a major part of plant-generated biogenic volatile organic compounds (BVOC). Plants emissions consist of a complex blend of volatile isoprenoids are usually emitted by plants sometimes a single species emit more than 20 different monoterpenes (Niinemets and Reichstein, 2003; Loreto et al., 2009).
Isoprenoids are synthesized *via* two different pathways from isopentenyl diphosphate. 2-C-methyl-*D*-erythritol 4-phosphate pathway (MEP) is responsible for monoterpene and diterpene production is localized in plastid (Edreva et al., 2007) and the mevalonate pathway (MVA), localized in cytoplasm, and is used to synthesize cytosolic and mitochondrial isoprenoids (Baldwin, 2010).
During different stress conditions plants start to emit green leaves volatiles(GLV) as C6-compound, (Z)-3-hexenal, (Z)-3-hexenal, (Z)-3-hexenol, (E)-2-hexenol, (E)-3-hexenol or (E)-2-hexenal which can be release from plant membranes by phospholipases (Feussner and Wasternack, 2002; Matsui, 2006). Abiotic stress as heat, cold, flooding or drought induces the emission of GLV (see for review (Loreto and Schnitzler, 2010)).
Many studies have been shown a high emission of those C6 compounds as well for biotic stress

(Copolovici et al., 2011; Junker and Tholl, 2013)

The Romanian forests with deciduous trees are dominated by Quercus genus (Popa et al., 2013). Red oak (*Quercus rubra*) is a fast growing tree which could be met in a various forests in Central Europe and North America. In a normal condition, it can live until 300-years and can reach 5-6 m tall.

Our hypothesis has been that VOC emission from plants can be used to quantify the stress impact on the plant.

The long time exposure to middle –high temperature have been used to check the VOC emission in *Quercus rubra.*

MATERIALS AND METHODS

Plant material

Seedlings of *Quercus rubra* are from local origin (Arad National Forest Department ROMSILVA) and were grown in a local greenhouse in 5 L clay pots filled with a 1:1 mixture of commercial potting soil and sand. The plants were watered daily and fertilized once per month with a fertilizer containing microelements.

In all experiments, we used similar-sized 2-yr-old seedlings with 20-25 leaves. The experiment was conducted with plants having fully mature leaves.

Stress application

The heat stress has been applied by keeping one individual leaf in the cuvette at 45 ^{o}C for 6 hours (other parameters have been kept constants: PAR 1000 μmol m^{-2} s^{-1}, CO_2 concentration 385 ppm, relative humidity 65%).

The experiment has been repeated 3 times with individual plants.

Photosynthesis measurements

The monitoring of plants photosynthetic parameters were performed using the GFS 3000 Portable Gas Exchange System (Walz, Effeltrich, Germany) as described before in (Copaciu et al., 2013).

The measurements were performed at a chamber CO_2 concentration of 385 μmol mol^{-1}, photosynthetic quantum flux density was kept at 1000 μmol m^{-2} s^{-1}, and chamber relative humidity at 65%. The air flow rate was 750 μmol s^{-1}.

The leaf were enclose in the cuvette and it was left to stabilize until steady-state values of net assimilation rate (A) and stomatal conductance to water vapor (g_s) (stomata opened) were obtained.

The rates of net assimilation (A) and stomatal conductance to water vapor (g_s) were calculated from these measurements according to (von Caemmerer and Farquhar, 1981).

Isoprene and green leaves volatile measurements

After stabilization following the stress application, the leaves subject to stress were mounted in the clipping cuvette of a commercial gas-exchange system (GFS-3000, Heinz Walz GmbH, Effeltrich, Germany). VOC sampling was performed on adsorbent cartridges via the outlets of each cuvette every day with a flow rate of 200 ml min^{-1} for 15 min by using a constant flow air sample pump (1003-SKC, SKC Inc., Houston, TX, USA). Adsorbent cartridges were analyzed for isoprene and lipoxygenase pathway products concentrations using a Shimadzu TD20 automated cartridge desorber and Shimadzu 2010Plus GC-MS instrument (Shimadzu Corporation, Kyoto, Japan) according to the GC-MS method detailed in our previous studies (Copolovici et al., 2009; Toome et al., 2010). The background (blank) VOC concentrations were subtracted from the emission samples with the seedlings.

RESULTS AND DISCUSSIONS

The emission of isoprene from plants from *Quercus* genus have been shown in many other papers (Monson et al., 2013). *Quercus rubra* have been demonstrated to be isoprene emitters.

The emission of isoprene is constant in the first 30 minutes of temperature stress and decline drastically in the following period of time.

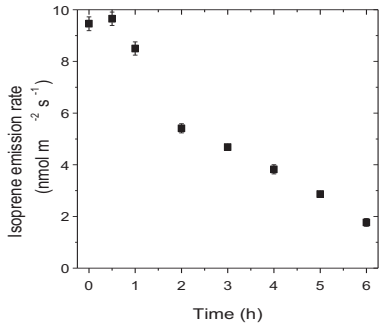

Figure 1. Isoprene emission rate variation function of the time of heat stress for *Quercus rubra* plants

There are a burst of green leaf volatiles (GLV) in the first minutes after the stress application which has been shown as well in many other papers, in case of the temperature stress (Brilli et al., 2011; Copolovici et al., 2012).
The GLV found in the samples were 1-hexanol, (*Z*)-3-hexenol, (*Z*)-2-hexenal, and (*Z*)-3-hexenyl acetate.
Our determinations show that GLV emission increase over first 4 hours and the concentration become constant in the following 2 hours.

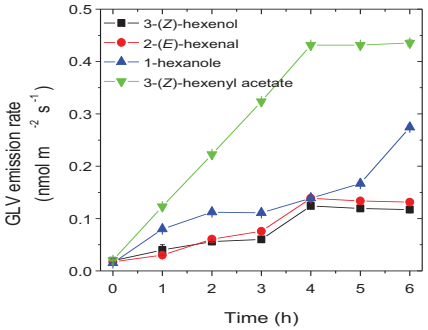

Figure 2. Green leaves volatiles (GLV) emission rates variation function of the time of heat stress for *Quercus rubra* plants

Regarding to monoterpene emission there are an increase after first hour followed by a plateau.
Usually terpene emission cannot be correlated with the time of exposure of stresses (Copolovici et al., 2012).

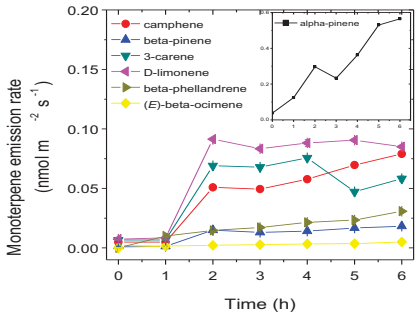

Figure 3. Monoterpenes emission rate variation function of the time of heat stress for *Quercus rubra* plants

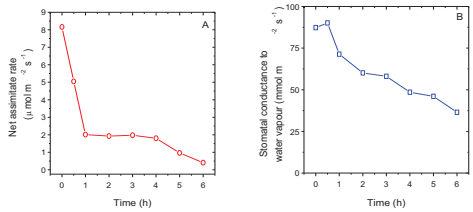

Figure 4. Changing in net assimilation rate (A) and stomatal conductance (B) depending on the time of application of heat stress

The photosynthetic parameters (assimilation rate and stomatal conductance to water vapour) decreased drastically after 30 minutes of stress conditions (Figure 4).

CONCLUSIONS

The results have been shown that photosynthetic parameters decreased drastically after first 30 minutes of exposure while green leaves volatile emissions are increased.
The monoterpene emission rates increased after 1 hours of exposure until a maximum of 0.2 nmol m^{-2} s^{-1} for most of the terpenes. Isoprene emission rate is decreasing, after the first burst, due to isoprene synthetase decreasing activity. The first burst explains the thermotolerance role of the isoprene for *Q. robur* leaves.

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