Advances in Forest Fire Research

Domingos Xavier Viegas

EDITOR

2014

Effect of prescribed burning on chlorophyll fluorescence and sap flow of *Pinus laricio*, a preliminary study

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Abstract

Forest fires constitute important perturbations in Mediterranean ecosystems, and preventing methods are used (e.g. prescribed burning) to avoid large scale fires during dry periods. This preliminary study aims to investigate the effects of prescribed burning on Corsican pine (*Pinus nigra* ssp. *laricio* (Poir.) Maire var. *corsicana* (Loud.) Hyl.) in field conditions. Two complementary approaches were tested: chlorophyll fluorescence and sap flow measurements. Chlorophyll fluorescence parameters allowed showing short term effects of prescribed burning, with a decrease of photochemical process on burned pines. However, at medium term, no significant difference was visible between burned and reference trees. Sap flow monitoring provided information about daily and seasonal cycles, it was greatly correlated with vapour pressure deficit, but no effect of prescribed burning was observed.

Keywords: Prescribed burning, Pinus laricio, chlorophyll fluorescence, sap flow

1. Introduction

Forest fires constitute one of the major perturbations for Mediterranean ecosystems, with real human, economic and ecological hazard. Mediterranean pines are well known for their flammability and their vulnerability to natural fires, and in Corsica (France), in order to prevent large scale fires and protect forests, prescribed burning are conducted under Corsican pine forests (*Pinus nigra* ssp. *laricio* (Poir.) Maire var. *corsicana* (Loud.) Hyl.). Today, a scientific framework is asked by forest managers to help guide this practice.

Previous studies (Ferrat *et al.* 2009; Cannac *et al.* 2009, 2011) brought beginnings of answers with laboratory experiments on needles and experimental fires in nursery, and it is now necessary to go on with tools that can be used on the field. The aim of this preliminary study is to test the potentialities of some vitality/stress indicators in quantifying impacts of a prescribed burning in a natural pine stand: chlorophyll fluorescence and sap flow measurements.

Chlorophyll fluorescence is widely used in plant physiological studies due to the ease of the field measurements and the quality of information provided. Indeed, changes of chlorophyll fluorescence are correlated with changes of CO_2 assimilation and therefore with changes of photosynthetic rate (Baker 2008), it is also a good indicator of stress (Peñuelas *et al.* 1998). Three parameters are usually monitored, the quantum yield photosystem II (ΦPSII), the maximum quantum yield of PSII (F_v/F_m) and the non-photochemical quenching (NPQ). The first are indicators of photochemical process: ΦPSII corresponds to the proportion of absorbed light used in photochemistry, and F_v/F_m is an indicator of photosynthetic performance, with optimal value around 0.83, lower value indicating a photoinhibition. NPQ is correlated to energy dissipated as heat and is studied as a mechanism of protection (Maxwell and Johnson 2000; Calatayud *et al.* 2006; Baker 2008).

These parameters are influenced by soil mineral content (Laing *et al.* 2000; Shangguan *et al.* 2000; Gough *et al.* 2004) and by drought (Shangguan *et al.* 2000; Pukacki and Kamińska-Rożek 2005;

Boureima *et al.* 2012), as demonstrated on coniferous and crop plants. They also undergo important seasonal variations (Vogg *et al.* (1998); Gielen *et al.* (2000) on coniferous; and Damesin (2003) on deciduous trees).

Sap flow is widely used to measure whole tree water use, due to its high degree of reliability and accuracy (Lu *et al.* 2004). Many studies have investigated effects of abiotic parameters on sap flow, mainly drought, with disparate results depending on the species considered (Anfodillo *et al.* 1998 for pinaceae). It is greatly influenced by climatic conditions (radiation, rain, VPD…) on different coniferous (Köstner *et al.* 1996; Simpson 2000; Iijima *et al.* 2004) and deciduous trees (Zalesny Jr *et al.* 2006; Nasr and Mechlia 2007). Ducrey *et al.* (1996), highlighted an alteration of sap flow after an important experimental heating of young *Pinus halepensis* trunk.

To our knowledge, literature regarding the effect of prescribed burning with chlorophyll fluorescence and sap flow on a natural pines stands is quite poor (Ducrey *et al.* 1996; Ferrat *et al.* 2009; Pasqualini *et al.* 2009) and generally realized experimentally and for short periods.

2. Methods

2.1. Study site

The study site was a mountain (900 m a.s.l.) natural pure Corsican pine stand situated in the Venaco Forest, Corsica, France (42°11'51.3"N 9°06'53.5"E). Trees were 20-25 years old, 11-17 m tall, with 13.5-18.5 cm DBH in average. Prescribed burning was performed March 29, 2012 on an usual fuel load surface (pine needles bed) of about 1500 $g.m^{-2}$, leading to a fire intensity between 50 and 100 $kW.m^{-1}$ (Trabaud 1979; Ferrat *et al.* 2011).

Measurements of chlorophyll fluorescence and sap flow were realised daily, then monthly, on burned and reference (unburned) trees.

2.2. Chlorophyll fluorescence

Measurements of chlorophyll fluorescence were realised in situ on attached needles of the previous year with a Portable Chlorophyll Fluorometer (PAM-21000, Walz, Germany), needles were dark-adapted during 30 minutes before measurement to open the reaction centres of the PSII. All of these measurements were realised at the sunrise, before the burning and daily then monthly after the burning, from March 27 to July 13, 2012. When light energy is absorbed by chlorophyll, the light not used in photosynthesis is dissipated as heat or is re-emitted as light fluorescence. Chlorophyll fluorescence is measured by exposing a leaf to light and measuring light re-emitted, different intensity/flash of light is used to measuring different parameters (figure 1). Measurement light (MB) allows measurement of the zero fluorescence level (F_o), the maximum fluorescence level (F^o_m) is measured after a saturating flash (SP), a second saturating flash is applied under an actinic light (AL) to measuring the maximum fluorescence in the light (F'_m) and the fluorescence level under actinic light, before the second flash (F_t), the zero level fluorescence (F'_o) is measured under far-red light (Maxwell and Johnson 2000).
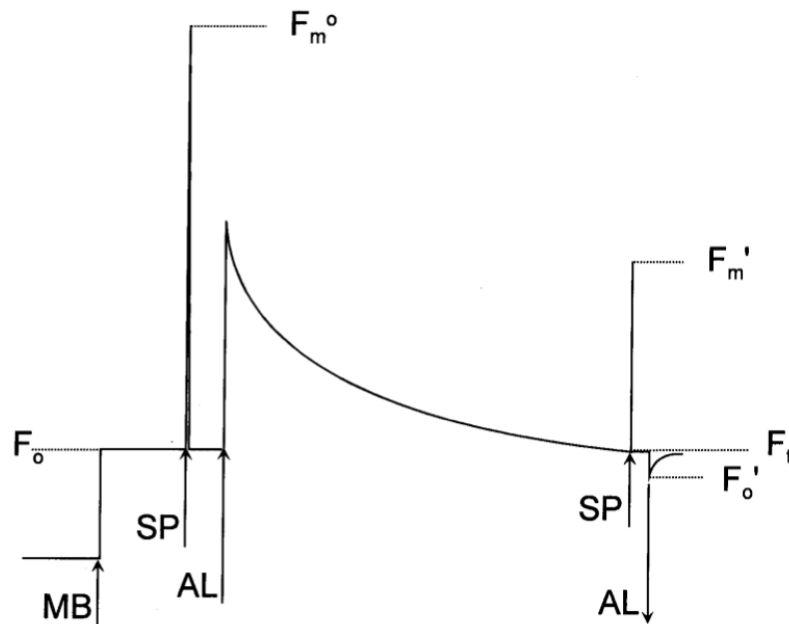
Figure 1. Sequence of a typical fluorescence trace (from Maxwell and Johnson 2000).

Three parameters were monitored: the quantum yield of photosystem II (ΦPSII), the maximum (or intrinsic) quantum yield of photosystem II (F_v/F_m) and the non-photochemical quenching. These parameters were calculated according to Maxwell and Johnson (2000), i.e.:

$$\Phi\text{PSII} = \frac{(F'_m - F_t)}{F'_m}$$

$$F_v/F_m = \frac{(F^o_m - F_o)}{F^o_m}$$

$$NPQ = \frac{(F^o_m - F'_m)}{F'_m}$$

with F'_m is the maximum fluorescence in the light, Ft is the level of fluorescence before the second saturating flash, F^o_m is the maximum fluorescence from dark-adapted leaves.

2.3. Sap flow

Sap flow measurements were realised with thermal dissipation probes (TDP, Dynamax, USA) according to the method developed by Granier (Granier 1985; Granier and Gross 1987). This method is based on temperature differences between two probes inserted radially into the trunk. The upper probe is continuously heated, both probes have a thermocouple. When sap flow is null, the difference between the two probes is maximal, when sap flow is high it dissipates heat produced by the upper probe by convection, and the difference of temperature between the two probes will depend on the speed of sap (Granier 1985; Granier and Gross 1987).

Depending on the thickness of sapwood, two length of probes were used (30 for DBH between 13.5 and 14.5 cm and 50 mm for DBH between 15.5 to 18.5cm) to improve the measurement (Lu *et al.* 2004). All probes were installed at East to avoid the influence of azimuth (Do and Rocheteau 2002). These measurements were recorded continuously before and after the burning from March 28 to October 30, 2012. Data were recorded in a CR3000 Micrologger (Campbell Scientific, USA) supplied by a battery charged by a solar panel. Together relative humidity and temperature were monitored, these allow computation of vapour deficit pressure (VPD, in kPa):

$$VPD = e_s - e_a$$

$$e_s = 0.6108 \times e^{\left(\frac{17.27 \times T}{T+273.3}\right)}$$

$$e_a = e_s \times \frac{RH}{100}$$

with e_s is the saturation vapour pressure (kPa), e_a is the actual vapour pressure (kPa), T is the temperature (°C) and RH is the relative humidity (%).
Sap flow were calculated according to Granier and Gross (1987):

$$k = \frac{\Delta TM - \Delta T}{\Delta T}$$

$$V = 0.0119 \times k^{1.231}$$

with k is a dimensionless number, ΔTM is the maximal temperature difference between the two probes (°C), ΔT is the temperature difference between the two probes (°C) and V is the sap velocity (cm.s^{-1}).

3. Results - Discussion

The prescribed burning influenced all chlorophyll fluorescence measurements at short term, the maximum quantum yield of burned pines decreased strongly immediately after the burning, from 0.84 to 0.76. It increased few days later but remained always under the reference values (resp. 0.82 vs. 0.84 during summer). F_v/F_m of the reference pines remained constant during the entire measurement period (approx. 0.84, figure 2).
Quantum yield increased the day after the burning, from 0.22 to 0.37 (68% up), but there was no difference between burned and reference pines few days later. Furthermore, this parameter underwent seasonal variations with a peak (0.44) at the end of May (figure 3).
Non-photochemical quenching decreased 2 and 3 days after the burning, from 2.3 to 1.2 (48% down) then increased to the initial value the 4th day. Later, it was under the reference (65% of the reference value, May, 5), except for the last record (figure 4).

The decrease of F_v/F_m immediately after the burning could reveal an alteration of photosynthetic performances due to a thermal stress. Previous study (Pasqualini *et al.* 2009) showed a decrease of this parameter after a burning on *Pinus laricio* needles, but this decrease did not have any effect on the survival of populations. It was shown that F_v/F_m was not long term affected if the value was kept upper than 60% of reference value (Ferrat *et al.* 2009), it is in accordance with our results, as Fv/Fm was about 98% of the reference value at the end of the experiment while it was about 90% of the reference value 2 days after the burning. NPQ corresponds to energy dissipated at heat, its increase would be a protection mechanism to dissipate energy excess and photo damage of PSII (Calatayud *et al.* 2006), this mechanism has not been implemented here. In the medium term, none of these three parameters seemed significantly affected by the burning.

Figure 2. F_v/F_m for burned and reference pines. Means ± standard error.

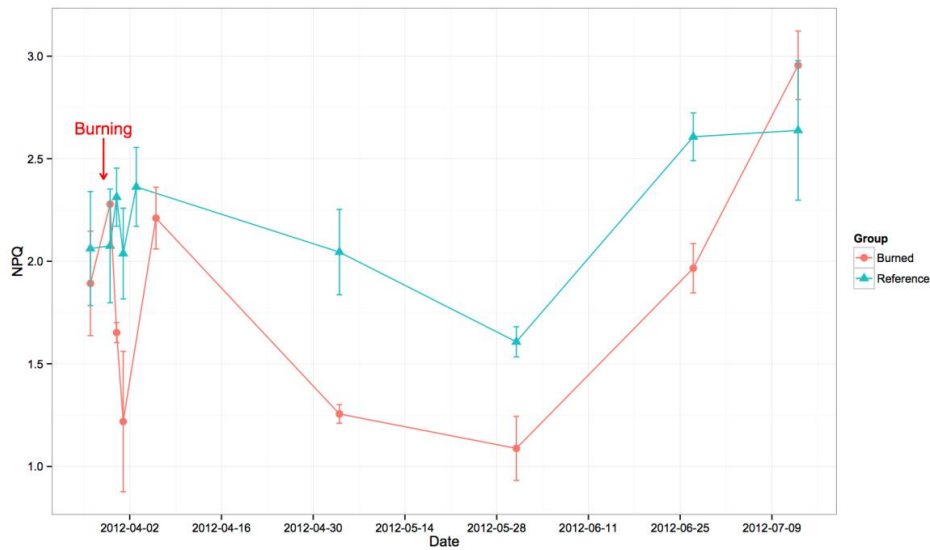*Figure 3. ΦPSII for burned and reference pines. Means ± standard error.*

Figure 4. NPQ for burned and reference pines. Means ± standard error.

Figure 5 shows the sap velocity for two representative trees (a burned and a reference) and the VPD. Sap flow of reference tree was lightly under the sap flow of burned tree during spring (mean: 7.8×10^{-4} cm.s^{-1}, max: 4.39×10^{-3} cm.s^{-1} for reference; mean: 1×10^{-3} cm.s^{-1}, max: 6.3×10^{-3} cm.s^{-1} for burned) and summer (mean: 6.5×10^{-4} cm.s^{-1}, max: 4.5×10^{-3} cm.s^{-1} for reference, mean: 8.5×10^{-4} cm.s^{-1}, max: 4.8×10^{-3} cm.s^{-1} for burned). During autumn they were close (mean: 4.7×10^{-4} cm.s^{-1}, max: 2.2×10^{-3} cm.s^{-1} for reference, mean: 4×10^{-4} cm.s^{-1}, max: 2.9×10^{-3} cm.s^{-1} for burned). The global sap flow difference between reference and burned tree can be explained by difference of DBH (resp. 14 cm vs. 15 cm) and mainly by difference of probe length (resp. 30 mm vs. 50 mm). It will be important in further studies to homogenize probes for a better discrimination of sap flow trends.

Nevertheless, it can be noticed that sap flow of reference and burned trees had similar variation from the start to the end of the monitoring, the prescribed burning did not seem to affect sap flow.
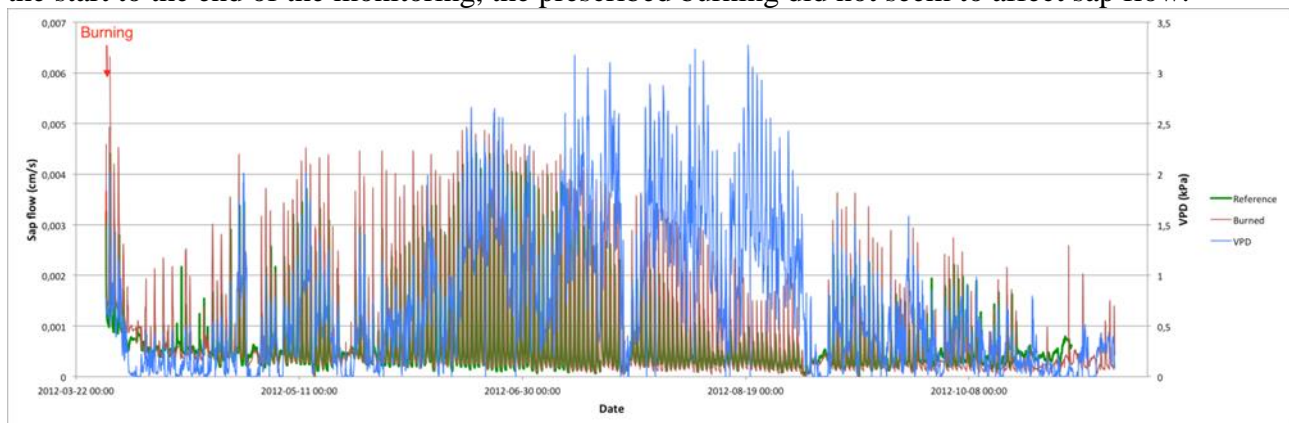

Figure 5. Sap flow velocity for burned and reference pines, and VPD.

The VPD varied daily and seasonally, it was low during spring (mean: 0.47 kPa, max: 2.66 kPa) and greater during summer (mean: 1.19, max: 3.27 kPa) and decreased during autumn (mean: 0.26 kPa, max: 1.58 kPa). Daily and seasonally variation of VPD can be explained by the great influence of relative humidity (RH) on the calculation, indeed during night and rain period RH is very high and thus VPD is very low. Temperature influences also VPD, more the temperature is important, more the saturation pressure is important and more the VPD may be important (Allen *et al.* 1998).

These data show that sap flow was greatly correlated, daily and seasonally, with vapour pressure deficit (VPD). An exception was measured during July and August, with a decrease of sap flow (from 0.004 to 0.002 $cm.s^{-1}$) while VPD was very high (over 3 kPa), this drop could be explained by low water availability during drought period (Anfodillo *et al.* 1998), and would reveal a water saving behaviour (Anfodillo *et al.* 1998; Badalotti *et al.* 2000). It will be interesting to survey this behaviour in case of severe drought.

This preliminary study has provided the beginnings of a reply regarding the effect of prescribed burning on photosynthetic and hydric parameters, and also provided information on daily and/or seasonal variation of these parameters. Chlorophyll fluorescence and sap flow measurements are promising tools to evaluate the impact of thermal stress on field, at short, medium and long term. A longer study with more replicate and complementary tools is in progress to provide more accurate answers.

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