

## Temperature Effect on Plant Leaves Fluorescence and Adaptivity of the Plant on Temperature

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### ABSTRACT

Red fluorescence shift  $\Delta\lambda$  in plant leaves induced by temperature was observed. Investigated temperature range was 263 K-328 K. It was observed that change of  $\Delta\lambda$  for first fluorescence maximum is greater as the temperature change  $\Delta T$  is greater. The red fluorescence shift was correlated with the change in photo system apparatus efficiency  $\Delta\varepsilon$ . It was shown that greater change in  $\Delta\lambda$  was connected with greater changes in  $\Delta\varepsilon$ .  $\Delta\varepsilon/\Delta T$  can be used as an indicator of plant adaptivity on environment temperature changing.

**Keywords:** Fluorescence spectra, Photosynthesis, Red shift

### INTRODUCTION

At a time of global climate change, it is very important, from the human point of view, to protect the green plant because most important reaction for a live on planet earth occur in green plant leaf. It is clear that disappearing the green plants can occur disappearing of whole life on our planet. In general low or high temperature stress results in various observable or measurable symptoms of injury including death of whole plants, visible necrosis of specific tissues and organs or less obvious cellular symptoms that can be detected by vital staining, osmotic responsiveness, chlorophyll fluorescence, or measurement of relative electrolyte leakage (REL) in affected tissues [1]. On the other hand, there is lot of examples that plants injury cause change in plant leaf fluorescence [2-4]. Therefore the effects of temperature on individual life must be known in order to understand the effect of temperature on plants as a whole. In literature date one can find that different plants have different response of photosynthesis on temperature (sunflower, soybean, watermelon, eggplant and jack bean) [5]. Generally different species shown difference in the determined range of environmentally induced photosynthesis adaptation to temperature [6]. This is an important factor to determine the survival of plants in hot/cold climate, as heat/cold damage of photosynthesis apparatus. Also, global warming is predicted to have a general negative effect on plant growth due to the damaging effect of high temperatures on plant photosynthesis.

### MATERIALS AND METHODS

The fluorescence temperature curve is usually measured in temperature range of 298.15 K-348.15 K (25°75°) [7,8]. Taking account mentioned above and our previous work, Jovanic et al. [4] have assumed that chlorophyll fluorescence can be used a useful method for determining plant resistivity upon high environment temperature not only for low temperature as is noted mentioned above.

Laboratory configuration used for the Light Induced Fluorescence (LIF) measurement is shown in **Figure 1**. A 470 nm ( $\pm 20$  nm) high-power light emitted diode (LED) with a power output of 8mW was used as the excitation source. At area of the 10 mm diameter on each leaf was irradiated by LED from the distance of 1 cm. Fluorescence emitted light from irradiated region of the leaf was collected by an optical fiber with a N.A. of 0.22 and a 120  $\mu$ m diameter core that was coupled to the portable high-

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resolution CCD spectrometer (HR-2000, Ocean Optics, Inc., Dunedin, Fla.). Optical resolution was 0.015 nm. The entrance aperture of the fiber was placed 1 cm away from the leaf and directed to the center of the illuminated area at 45° to the light axis. Data collected and spectral processing and analysis were conducted in real time with a microcomputer. We have followed literature data that minimum thirty (30) samples are necessary for the evaluation of a middle value and a standard deviation of some physical phenomena which depend on one stochastic argument, with the precision higher than 1% [9]. In this work over 92 measurements were performed for each plants space at given temperature, hence the sample size satisfies the mentioned criteria. One-way analysis of variance ANOVA was carried out using Statgraphics software (Statistical graphics Corporation,

USA)  $\tau$  test for significant differences. Heat damage is reflected in drastic change in light induced fluorescence and that was the reason why we used this method for researching. Samples, plant leaves, were preheated/cooled for 120 min at the indicated temperatures and after that measured actual fluorescence spectrum. Relative temperature change  $\Delta T$  was defined as difference between preheated temperature  $T_{PH}$  and 263 K:  $\Delta T = T_{PH} - 263$ . Objects of the experiments were four weak old plant leaves from: palm (*Cocos nucifera*), locust (*Robinia pseudoacacia*), linden (*Tilia americana*), birch (*Silver birch*), poplar (*Populus nigra*), walnut (*Juglans nigra*), chestnut (*Castanea sativa*), mulberry black (*Morus nigra*), crabtree (*Malus*), oak (*Quercus robur*), sour cherry (*Prunus cerasus*), willow (*Salix alba*), birch (*Betula pendula*).

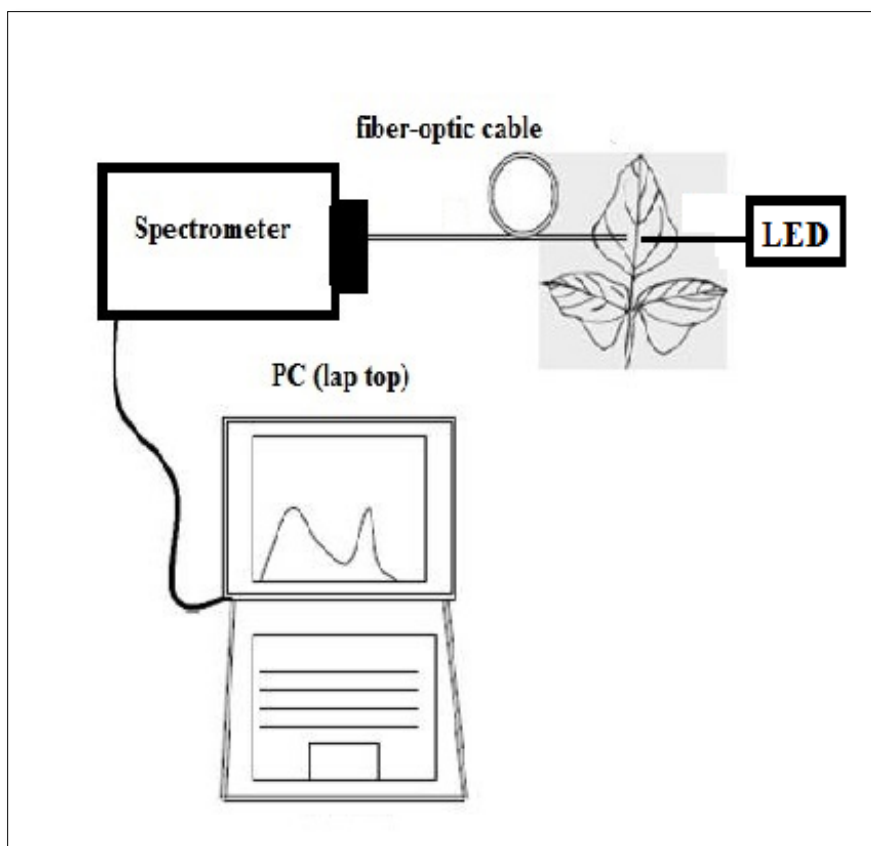


Figure 1. Experimental set up.

Following previous researching the relative change in photosynthetic apparatus efficiency  $\Delta \epsilon$  induced due the damage was calculated using relation between  $\Delta \epsilon$  and relative shift of the position in first fluorescence maximum  $\Delta \lambda$  [4]:

$$\Delta \epsilon [\%] = 5.209 \Delta \lambda^{0.653} \dots \dots \dots Eq.(1)$$

**RESULTS AND DISCUSSION**

Typical fluorescence spectra for plant leaf on three different temperatures were present in **Figure 2**. It is well known that

the temperature at which drastic fluorescence change can be observed depending on plant species and growth temperature [3,10]. There is little difference in the shape of the chlorophyll fluorescence spectra and intensity ratio F690/F735. Significant changes in the ratio F690/F733 could not be detected. This is an agreement with literature date [2]. Therefore the strong decrease of the chlorophyll fluorescence in plants which remained outside in winter may be explained due a partial dehydration of the leaf issue and cytoplasm.

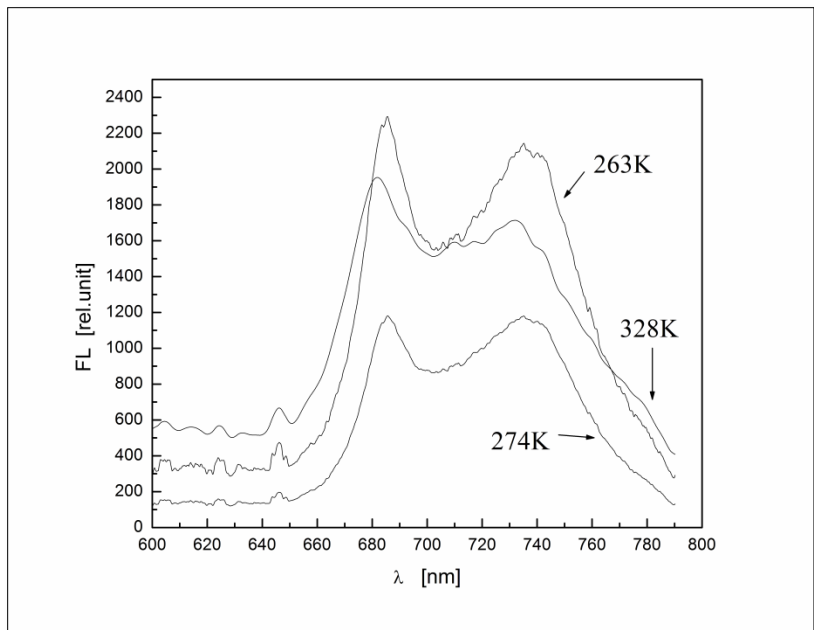


Figure 2. Fluorescence spectra of the plant leaves on three different temperatures.

Possible influences of leaf age and plant age were well explored in literature and shown that it have only a small, neglected, effect on thermal stability [3,11]. Beside this all laves was fore weak old and at this way was eliminate motioned small effect due the leaf age. In order to determinate plant leaf sensitivity upon temperature it was drown relative red shift  $\Delta\lambda$  [%] for the first luminescence maximum FLM (at about 690 nm) upon relative temperature change  $\Delta T$  [K]. In this way we were able to compare the

temperature effect on the difference in position of the FLM for different plant species. Using relative change of  $\Delta T$ , as the difference between control temperature (room temperature) and stress temperature, the effect of the variation in control temperature was eliminated. For all experiments the same temperature intervals were obtained. For all fourteen plant species functional dependence  $\Delta\lambda$  [%] =  $f(\Delta T)$  was linear (Figures 3 and 4).

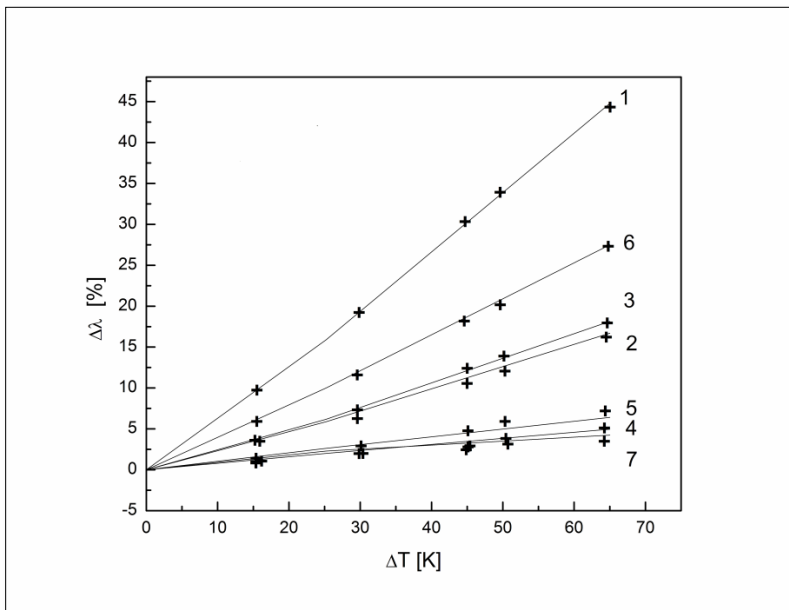
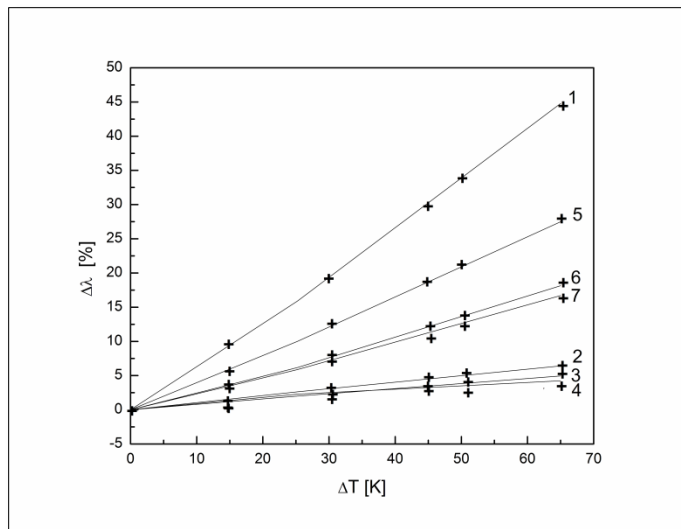


Figure 3. Relative change of the red shift FLM upon temperature. 1- Plum (*Syringa mill*), 2- locust (*Robinia pseudoacacia*), 3- linden (*Tilia americana*), 4- birch (*Silver birch*), 5- poplar (*Populus nigra*), 6- walnut (*Juglans nigra*), 7- chestnut (*Castanea sativa*).



**Figure 4.** Relative change of the red shift FLM upon temperature. 1- palm (*Cocos nucifera*), 2- mulberry black (*Morus nigra*), 3- crabtree (*Malus*), 4- oak (*Quercus robur*), 5- sour cherry (*Prunus cerasus*), 6- willow (*Salix alba*), 7- birch (*Betula pendula*).

It is well known that any change over usual grown condition (298 K ± 5 K) cause dramatic change photosynthetic apparatus efficiency through change in lipid membrane composition, activation oxygen scavenging enzymes, water loss and altered growth morphology [12-14]. As the measure for temperature sensitivity was used the slope of linear regression obtained drawing  $\Delta\lambda [\%] = f(\Delta T)$  (Figures 3 and

4). Obtained slope was presented in Table 1. One can see that different plant species shown different temperature sensitivity. In the other words different plant species will “answer” at different way on environment temperature which depends of its adaptation on climate condition. This is an agreement with literature data [6,12,13]. Some of them are excellent adapted on low temperature [15].

**Table 1.** Relative change of the red shift  $\Delta\lambda$  and in photosynthetic apparatus efficiency  $\Delta\epsilon$  in temperature range (263 K-328 K).

Objects	$\Delta\lambda$ [nm]	$\Delta\lambda/\Delta T$ [nm/K]	$\Delta\epsilon$ [%] Eq.(1)	$\Delta\epsilon/\Delta T$ [%][K <sup>-1</sup> ]
Plum	44.8	0.692	62.38	0.959
Locust tree	15.94	0.245	31.77	0.489
Linden tree	18.26	0.281	34.75	0.535
Birch silver	16.06	0.241	31.92	0.489
Poplar	6.69	0.103	18.02	0.277
Walnut	27.51	0.423	45.37	0.698
Chestnut	3.80	0.058	12.47	0.192
Palm tree	44.48	0.699	62.36	0.959
Mulberry black	6.65	0.102	17.95	0.276
Crabtree	5.21	0.081	15.31	0.236
Oak tree	5.92	0.054	16.64	0.259
Sour cherry	27.53	0.424	45.36	0.698
Willow	18.17	0.279	34.60	0.532
Carb	16.37	0.252	35.32	0.543

Other one being excellent adapted to high temperature [13]. On the other words, it is well known that: a) heat damage is reflected in drastic changes of the light-induced fluorescence spectra (fluorescence intensity) which is correlated with inhibition to Photosystem II [16] and b) chilling also cause dramatic change in light-induced fluorescence spectra which is correlated with in cells morphology [6].

Also, using Eq. (1) relative change in photosynthetic apparatus efficiency  $\Delta\varepsilon$  was calculated. The rate of the relative change of photosynthetic apparatus efficiency  $\Delta\varepsilon/\Delta T$  was obtained as the slope of the line  $\Delta\varepsilon [\%] = f(\Delta T)$  and are given in **Table 1**. One can see that for all species  $\Delta\varepsilon/\Delta T$  is close to each other except for Poplar, Chestnuts and Oak. This mean that photosynthetic apparatus efficiency  $\varepsilon$  for all species, except three mentioned, are very sensitive to temperature change. Also, small temperature sensitivity  $\Delta\varepsilon/\Delta T$  may be result of the excellent acclimation to the climate condition and great resistivity of photosynthetic apparatus on temperature fluctuation. Also, it mean that photosynthetic apparatus and its function as well as the chlorophyll content of the leaf will good and fast adjust to new environment temperature. This can be explained with in different change in plant morphology and physiology caused by temperature. For example, temperatures which induce irreversible damage in lipid membrane of plant leaves are different for different plant species [10]. Plants may be differ in heat tolerance, and studied have shown that some plants are capable of physiological acclimation to increase both their heat tolerance and their temperature optimum for net photosynthesis [10]. Therefore one can say that temperature tolerance is specific and determinate with plants genetically.

Obtained difference in for different plant species in not unexpected. Namely, in literature data one can find that the high temperature effects depend on species and genotype [17-20]. Some experimental data have shown that in poplar photosynthesis significantly decreased after heat stress so after 24 h exposing to the 315 K photochemical quenching is about 45.2% [21].

In literature data one can find lot of examples that change of temperature, outside the optimum condition can cause change in damage of the photosynthetic apparatus [3,22,23].

Taking account mentioned discussions and results from literature date for 73 trees species [1] we assumed that if the change of the photosynthesis apparatus is change less that 40% due the temperature change mentioned plants have great ability to survive in given environmental conditions.

Preliminary discussions indicate the possibility of practical application of the examination of optic properties of the green plant leaf. Namely, mmentioned above discussion lead to say two thinks. First, that the plants with smaller  $\Delta\varepsilon/\Delta T$  have greater ability for adapted (greater possibility) to environment temperature conditions. Second, measuring the  $\Delta\varepsilon/\Delta T$  can it would be an indicator, for young or by crossing

obtained the new plant species that indicates how much the plant is able to overcome the resulting temperamental changes in the environment.

## CONCLUSION

The temperature has great effect on red shift of the first fluorescence maximum in fluorescence spectra of the plant leaves. The shift rate  $\Delta\lambda/\Delta T$  induced with temperature is different for different plant species. Direct correlation between red shift and change in photo system apparatus efficiency  $\Delta\varepsilon$  exist, greater change in  $\Delta\lambda$  was connected with greater change in  $\Delta\varepsilon$ .  $\Delta\varepsilon/\Delta T$  can be used as indicator of plant adaptivity on environment temperature changing.

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