

## THE EFFECT OF THE LIGHT ON THE CONTROL OF ANTHOCYANIN PIGMENTATION OF FRUITS, FLOWERS, AND SHOOTS OF CITRUS AND RELATIVE'S

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In plant anthocyanins are the results of a manifold interaction between biosynthetic pathway, controlled by a Myb-bHLH-WD complex, and other transcription factors (TF) interacting with each other coping with external factors, such as cold and light.

In citrus the anthocyanins pigmentation is a trait quite diffused among primitive, wild, and cultivated species. In particular, the purple coloration is under the control of a positive activator (Ruby1), of a competitor of that (Ruby2), and of a repressor working in an activator-and-repressor loop (CsMYB3). The role of HY5 (bZIP, binding to the G-box motif within the promoter of R2R3 Myb-like) and COP1 (in the dark it degrades, by ubiquitination, all the positive regulators) has been also evaluated similarly to *Arabidopsis* and *Malus*.

In order to investigate the effect of the light and the role of a new TF (Zing Finger-like) in the anthocyanin production, the co-expression analysis (which infers the function of a gene basing on similar expressions) have been carried out on comparing several tissues under dark-light conditions, focusing on already known TFs (Ruby1, Ruby2, CsMYB3, HY5, COP1) and two biosynthetic genes, CHI and DFR. We selected accessions of sweet orange (*Citrus sinensis*) and finger lime (*Microcitrus spp*) not equally anthocyanins-rich in the flesh and in the rind. Sweet orange fruits were covered with black envelope bags before the external turning from green to orange; fruits were collected after three months. Finger lime fruits were covered in the early phase of fruit development, and they were collected after six months. In addition, stigma of pigmented and non-pigmented sweet orange varieties, petals of lemon (*C. limon*), citron (*C. medica*), and *C. latipes*, fruits and shoots of Chinese box orange (*Atalantia buxifolia*, an ancient Citrus species) were covered and sampled after from 3 to 4 weeks. The color was measured using a Konica minolta CR-400 and through image analysis. The genes expression was evaluated through Real time PCR.

Phenotypically the light affected the pigmentation of all tissues, except fruits of Chinese box orange (looking light and cold-independent). “L” showed that no-covered fruits of sweet orange and finger lime were externally much deep-colored for the presence of anthocyanins. Otherwise, any difference was observed in the flesh pigmentation of sweet oranges. Moreover, in the finger lime ‘Sanguinea’ variety (naturally internally and externally pigmented) the enveloping induced the develop of overall non-pigmented fruits. This phenomenon led us to suppose the existence of a genetic source that is different compared to sweet

orange, also totally cold-independent.

(Co)Expression analysis showed the putative involvement of the new Zing Finger-like in the response to light, leading to hypothesize that the atavism of anthocyanins accumulation in response to external factors has been recovered in pigmented oranges, similarly to other *Citrus* species.