Light intensity is positively correlated with the synthesis of condensed tannins in Lotus corniculatus

S. Arcioni, T. Bovone, F. Damiani and F. Paolocci
Plant Genetic Institute – Research Division of Perugia-CNR, via Madonna Alta 130, 06128 Perugia, Italy, Email: sergio.arcioni @igv.cnr.it

Keywords: condensed tannins, DFR, gene expression, light, real time PCR

Introduction The importance of Condensed Tannins (CT) in forage legumes has been well documented in several studies. The role of plant genetics in this field is the acquisition of competences in order to be able to modulate CT synthesis in leaves of these species. The role of light has been investigated in this work on the increase of condensed tannin levels in leaves of two contrasting genotypes of birdsfoot trefoil (*Lotus corniculatus*).

Materials and methods Well developed cuttings of two plants S41 and S50, with high and low CT, were grown under two contrasting light regimes: high light (>1000 μE m^{-2} s^{-1}), and low light (150 μE m^{-2} s^{-1}). After 4 weeks leaf CT, using DMACA protocol (Li *et al*., 1996) and gene expression of DFR, through Real Time RT-PCR, were quantified. A dissociation protocol of amplified products was performed and information about the presence of polymorphic cDNA fragments was obtained.

Results Light affected CT accumulation (Figure 1) with high light intensity increasing plant staining for both S50 and S41 than at low light intensity, S41 produced more CT than S50 at both treatments. Only at a high light intensity did DFR expression differ significantly between plants (Table 1). The qualitative expression of DFR monitored through the dissociation protocol showed that the two plants mainly differed for the allelic expression at both light intensity. In fact from previous experiments the profile of amplification of different DFR alleles was established (Paolocci *et al*., in press) so it was possible to determine which alleles were expressed in plants at different light intensity. Figure 2 show the DFR profiles: at high light S41 expressed mainly allele 3 and trace amounts of allele2 while S50 expressed allele 2 and 3 at similar rate; at low light S41 expressed only allele 3 and 16 mean while in S50 all the three alleles were present. It seems that allele 16 is less responsive to light than the two others and that allele 3 is the most relevant for CT synthesis.

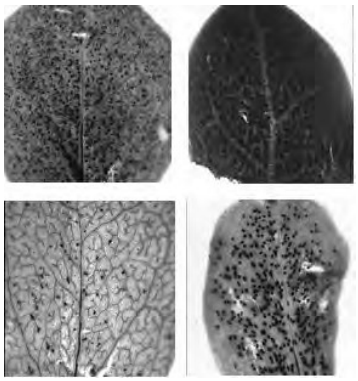

Figure 1 CT stain in S41 and S50 leaves at low (L) and high (H) and low (L) light

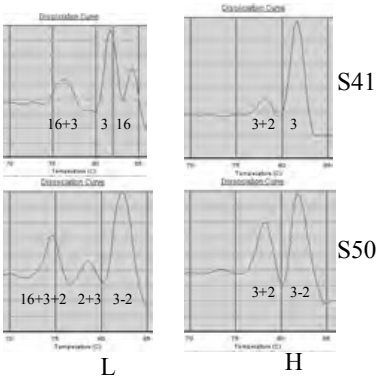

Figure 2 Dissociation profiles of S41 and S50 plants at high (H) and low (L) light

Table 1 DFR level measured through real time RT-PCR and expressed in relative standard unit

	L	H
S41	1.2a	1.6b
S50	0.9a	1.1a

Means with the same letter do not differ significantly

Conclusions Light positively affects CT accumulation. DFR, a key enzyme of the pathway, is quantitatively and qualitatively regulated by light and some alleles play a determinant role on the rate of CT accumulation while some others probably have a competitive negative effect.

References

Li, Y.G., G.J. Tanner & P.J. Larkin (1996) The DMACA-HCl protocol and the threshold proanthocyanidin content for bloat safety in forage legumes. *Journal of the Science of Food and Agriculture,* 70**,** 89-101.

Paolocci, F., T. Bovone, N. Tosti, S. Arcioni & F. Damiani (in press) Light and an exogenous transcription factor qualitatively and quantitatively affect the biosynthetic pathway of condensed tannins in *Lotus corniculatus* leaves. *Journal of Experimental Botany.*