Article

C3H expression is crucial for methyl jasmonate induction of chicoric acid production by *Echinacea purpurea* (L.) Moench cell suspension cultures

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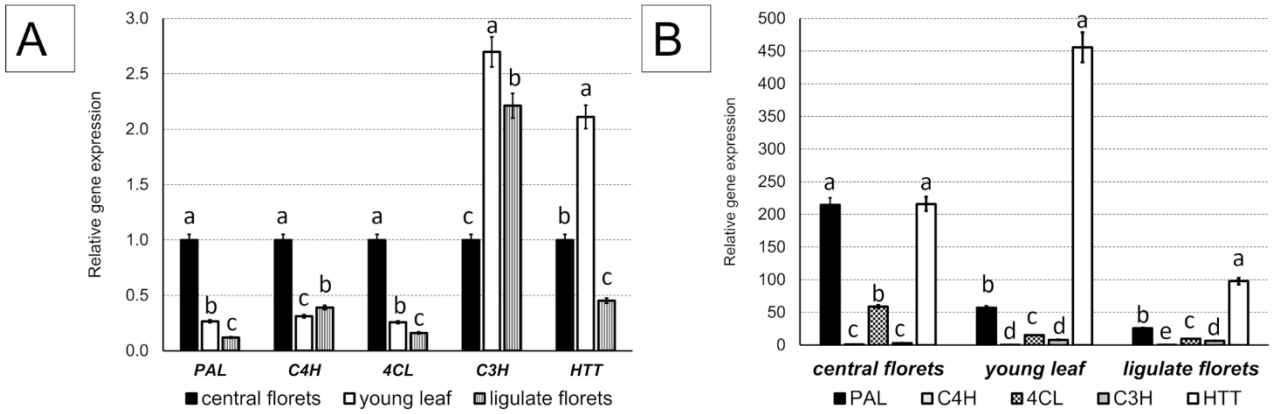

Figure S1. *In vivo* gene expression analysis of *Echinacea purpurea* genes involved in chicoric acid biosynthesis in different tissues. **Panel A** shows the relative gene expression of each gene with respect to the value in the central florets. **Panel B** shows the relative gene expression of every gene compared to the value of *C4H* in the central florets and expressed as 1. Data are mean ± SE for three biological replicates. The expression levels of genes are presented using mRNA levels normalized to *ACT*. Different letters above the bars indicate statistically significant difference at $p<0.05$ for ANOVA.

To set up the methodology, gene expression of genes related to chicoric acid biosynthesis was initially tested on samples obtained from *E. purpurea* tissues, in particular central florets, ligulate florets and young leaf (**Figure S1**). The expression of all the five genes representative of chicoric acid biosynthesis was markedly higher in central florets for *PAL*, *C4H* and *4CL*, while it was more than 2-times higher in the young leaf for *C3H* and *HTT*. The gene expression of all the chicoric acid biosynthesis gene appeared decisively down-regulated in the ligulate florets, with the only exception of *C3H* that showed a 2-times up-regulation in this tissue (**Figure S1A**).
Considering the level of expression in each tissue (**Figure S1B**), *PAL* and *HTT* were the most expressed in the central florets, with *HTT* showing the highest expression also in the young leaf and ligulate florets, followed by *PAL* and *4CL*. *C4H* and *C3H* appeared as the less expressed genes in the three tissues.