

## Anthocyanin biosynthesis mystery in gerbera cultivars Estelle and Ivory

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Flavonoids in our model ornamental plant *Gerbera hybrida*, consist of three subgroups, flavones, flavonols and anthocyanins. Anthocyanins accumulate in the adaxial surface of petals and give the different cultivars their different color. Both pelargonidin and cyanidin derivatives are found in gerbera, but none of the cultivars contain delphinidin.

The acyanic cultivar Ivory is a sport of the pelargonidin containing pink cultivar Estelle, i.e., it originates from an acyanic branch of Estelle. Ivory is apparently a transposon mutant of Estelle, since revertant sectors are regularly observed (Figure 1). In spite of complete loss of anthocyanin pigmentation, all genes encoding enzymes necessary for pelargonidin biosynthesis (*PAL*, *C4H*, *4CL*, *CHS*, *CHI*, *F3H*, *F3'H*, *DFR*, and *ANS*) are expressed in Ivory at similar levels as in Estelle.

We performed a comprehensive flavonoid analysis using UHPLC MS/MS for Estelle and Ivory, collecting samples from whole ray flower petals and from their isolated adaxial epidermi. Except for pelargonidin derivatives, which are present in Estelle but lack nearly completely from Ivory, we found that both cultivars have similar amounts of flavones and flavonoids (mainly apigenin and kaempferol glycosides).

We further analyzed the cultivars using RNA sequencing and produced on average 10 million Illumina reads from two developmental stages of Estelle and Ivory petals. Mapping of the reads to an assembly of gerbera Sanger (*I*), 454 and Illumina reads confirms that all anthocyanidin biosynthesis genes are expressed similarly in the two samples. Surprisingly, none of the assembled contigs show differential expression between these two cultivars.

Although dramatically different to the eye, the difference in Estelle and Ivory at transcript level eludes our attempts of analysis. Pelargonidin biosynthesis is intact at least up to the point of dihydrokaempferol synthesis. In reads mapping to transcripts for *DFR* and *ANS* we have not observed anomalies that would be indicative of a transposon insertion. None of the glucosyl transferase encoding transcripts are down regulated or anomalous either. However, we do not have biochemical evidence which one of the contigs would encode the gerbera anthocyanidin 3-*O*-glucosyltransferase.



**Figure 1.** The gerbera cultivar Ivory (A) is a transposon mutant sport of cultivar Estelle (B).

### References

1. Laitinen RAE, Immanen J, Auvinen P, Rudd S, Alatalo E, Paulin L, Ainasoja M, Kotilainen M, Koskela S, Teeri TH, Elomaa P: Analysis of the floral transcriptome uncovers new regulators or organ determination and gene families related to flower organ differentiation in *Gerbera hybrida* (Asteraceae). *Genome Research* 2005, 15:475-486.