Flavonoid metabolons in *Gerbera hybrida*

Teemu Teeri*[1], Hany Bashandy[1] and Stefan Martens[2]
[1]*Department of Agricultural Sciences, University of Helsinki, Finland*
[2]*Department of Food Quality and Nutrition, Fondazione Edmund Mach, Italy*
**teemu.teeri@helsinki.fi*

The formation of enzyme complexes, metabolons, and the channeling of intermediates of secondary metabolism has been discussed for at least 40 years (*1*). Metabolons and channeling enable plants to perform a highly effective synthesis of specific natural products without or with reduced metabolic interference and to avoid the accumulation of toxic intermediates (*2*). Despite a long tradition of the concept, precise examples of complete metabolons are scarce. For the anthocyanin pathway, soluble enzymes of the pathway have been shown to interact in yeast cells (*3*), and to associate with membranes in plant cells (*4*), the latter presumably through interactions with the membrane anchored P450 hydroxylases of the pathway.

The ornamental plant *Gerbera hybrida* contains flavones, flavonols and anthocyanins in its petals, the exact composition reflecting the color of each gerbera cultivar. Orange, pink and red cultivars contain 4' hydroxylated pelargonidin derivatives, while magenta and purplish cultivars are rich in 3', 4' hydroxylated cyanidin derivatives. Remarkably, 3', 4' hydroxylated flavones and flavonols (luteolin and quercetin) can be found in cultivars which completely lack cyanidin. The gerbera cultivar Terra Regina, with orange pelargonidin containing petals, starts to synthesize cyanidin when overexpressing the transcriptional regulator encoding *GMYB10* gene, but not in cells where pelargonidin accumulates. These observations are indicative of metabolon control of flavonoid biosynthesis in gerbera petals.

We have developed deep transcriptome data using Sanger, 454 and Illumina sequencing of different cultivars of gerbera (*5* and *unpublished*). Assembly of the reads indicates that gerbera expresses small gene families for some of the flavonoid biosynthesis genes (e.g., *PAL*, *4CL*, *CHS* and *CHI*) and single genes for others (e.g., *DFR* and *F3'H*). We are interested if the corresponding isoenzymes have different roles in biosynthesis of the array of flavonoids present in gerbera, particularly if they are able to form metabolons of different composition and biosynthetic capabilities. In order to map their interaction properties, we are running an all-against-all assay in yeast cells using vectors designed for membrane proteins.

References

1. Stafford HA (1974). Possible multi-enzyme complexes regulating the formation of C_6- C_3 phenolic compounds and lignins in higher plants. Rec Adv Photochem 8: 53-79.
2. Winkel-Shirley B (1999) Evidence for enzyme complexes in the phenylpropanoid and flavonoid pathways. Physiology Plant 107: 142-149.
3. Burbulis IE, Winkel-Shirley B(1999) Interactions among enzymes of the Arabidopsis flavonoid biosynthetic pathway. Proc Natl Acad Sci USA 96:12929–12934.
4. Hrazdina G, Wagner GJ(1985) Metabolic pathways as enzyme complexes: evidence for the synthesis of phenylpropanoids and flavonoids on membrane associated enzyme complexes. Arch Biochem Biophys 237:88–100.
5. 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 Res 2005, 15:475-486.