

ABSTRACT

Understanding the Flavonoid Biosynthetic Pathway towards bioengineering novel colours in Anthurium (*Anthurium andraeanum* Hort.)

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The core objective of this dissertation was to understand the flavonoid biosynthetic pathway towards bioengineering novel colours in anthurium (*Anthurium andraeanum* Hort.). This study was complemented with an historical analysis of the regional industry and an exploration of its status, to determine the impact this research could have as a competitive strategy to be introduced in a regional breeding programme. Three studies were completed, 1st was determining spathe colour expression regulatory controls, by analysing the flavonoid levels and expression profiles of CHS, F3'H, ANS and DFR genes from total-RNA of 16 cultivars separated by colour and developmental stages using Northern analysis. The Results showed that F3'H and ANS were controlled by one regulatory system, DFR controlled by another, and CHS by yet another mechanism; supporting in general the Elibox and Umaharan (2008b, 787-791) "three-gene model", with some exceptions. The 2nd and 3rd study established the relationship between vacuolar pH and spathe colour originated from flavonoids pigments with white/green colour group as control, by studying 23 cultivars with a range of spathe colours, at different developmental stages, at various spathe locations, at the abaxial and adaxial spathe surface, and at various time after harvest. Six randomized complete block design experiments were evaluated for pH measurements versus chromospectral analysis of pigmentation using the CIE (L^* , a^* , b^*) colour space method. The results confirmed the relationship between colour and vacuolar pH, where lighter colours had higher pH values and vice versa, with significant differences in all experiments. The results showed the white/green group with the highest average pH (5.65) both, followed by coral (5.38), pink (5.20), red (5.10), and orange with the lowest with (4.51). In conclusion, three clear regulatory controls of major genes were identified for the anthurium anthocyanin biosynthetic pathway, but need to be further confirmed with quantitative real-time PCR and Western analysis. For blue hues in anthuriums, the coral group (5.38) seems to be most suitable to target for transformation, having the highest pH value with lower levels of pH-reducing anthocyanins. Alternately, a larger number of cultivars of the red group should be screened for genotypes with high pH values. Stakeholders according to the Caribbean industry survey still considering the anthurium as a viable business for the region; yet long lasting impact depends on implementing a sustainable regional breeding programme of adapted cultivars using bioengineering and regional genetic studies.

Keywords: Anthurium; regulatory control; flavonoid biosynthetic pathway; vacuolar pH; gene expression; Chalcone synthase (CHS); Flavonoid 3'hydroxylase (F3'H); Dihydroflavonol 4-reductase (DFR); Anthocyanidin synthase (ANS).