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"The colours and antioxidants of fruits and vegetables: what genes and horticultural practices can do"

Myrtle domestication, fruit pigments and antioxidant properties

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Introduction

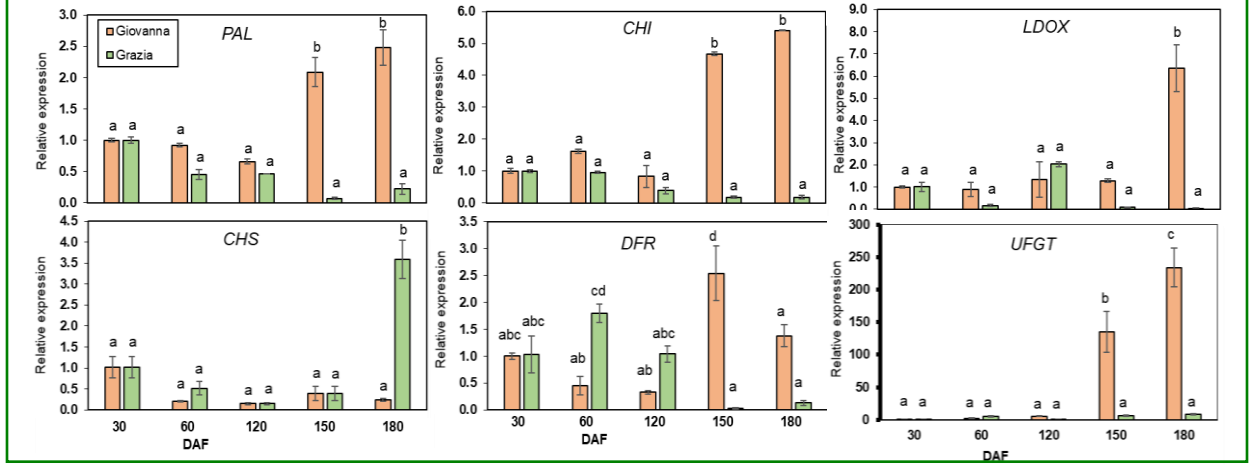
Interest in myrtle (*Myrtus communis* L.) due to biological properties by food, cosmetics, and pharmaceutic industries generated the integration of biomasses harvested from wild populations as raw materials with yields of cultivated orchards. With the aim to preserve wild myrtle populations and to select suitable genotypes for cultivation and industrial uses, the domestication and selection process of myrtle species was promoted. The studies following the domestication process have highlighted the key role of phenolic compounds with respect to aromatic and antioxidant properties of myrtle. Within the species, different genotypes may produce dark-blue berries or white berries depending on the peel color upon ripening. Most of the biological properties of myrtle are linked to the antioxidant activity of the phenolic compounds present in the extracts. In addition, previous studies have shown that the white genotype has a similar or higher total phenol content despite the lack of anthocyanins. This study provides information about the molecular mechanisms leading to myrtle berry pigmentation by studying the expression of common genes of flavonoid biosynthesis during the physiological ripening of the fruit at different stages of development. In addition, evaluation of the antioxidant activity of berries of the two genotypes was performed.

Materials and Methods

Two cultivars of myrtle with dark-blue berries ("Giovanna") and white berries ("Grazia") were analyzed. These cultivar from Berry sampling was carried out over five stages of development (30, 60, 120, 150, and 180 DAF). The relative expression levels of PAL, CHS, CHI, DFR, LDOX and UFGT were evaluated using Quantitative Real-Time RT-PCR. Relative expression levels were estimated with the 2 −ΔΔCt method and were denoted as the fold difference from the expression present at the calibrator sample (30 DAF from dark-blue samples). The specificity of the products was validated with dissociation curve analysis and with agarose gel; the sequences were confirmed via Sanger sequencing at Secugen (Madrid, Spain). For antioxidant evaluation, the samplings of 'Giovanna' and 'Grazia' cultivar were carried out at 60, 150, 210, 225 DAF. Acidified ethanol was used as extraction solvent. The assays used were ABTS, FRAP and DPPH assays and the spin trapping method coupled with Electron Paramagnetic Resonance Spectroscopy.

Results

Cultivar	DAF	DPPH (µmol TE/g DW)	FRAP (mmol TE/g DW)	ABTS (mmol TE/g DW)	EPR (mmol GAE/g DW)
Giovanna	60	20.44 ± 0.65de	110.77 ± 8.46bc	65.54 ± 3.69e	0.12 ± 0.04c
	150	29.66 ± 1.00b	119.28 ± 9.05ab	148.32 ± 8.19b	3.89 ± 0.72b
	210	24.97 ± 0.06c	114.29 ± 1.11abc	121.85 ± 10.5cd	4.67 ± 1.00ab
	225	22.96 ± 0.73cd	118.97 ± 5.92ab	132.40 ± 1.53bc	0.13 ± 0.02c
Grazia	60	19.45 ± 0.73e	100.80 ± 0.01c	109.58 ± 8.79d	0.16 ± 0.01c
	150	30.07 ± 1.80ab	109.17 ± 2.53bc	118.91 ± 11.7cd	4.63 ± 0.77ab
	210	32.74 ± 0.33a	119.79 ± 1.58ab	176.61 ± 7.27a	6.64 ± 1.77a
	225	22.07 ± 1.65cde	127.12 ± 1.70a	71.68 ± 0.87e	0.26 ± 0.11c



Conclusions

The overall results indicated that the differences in coloration between the dark-blue "Giovanna" cultivar and the white "Grazia" fruit might be due to the over-regulation of some structural genes in dark-blue fruit, particularly the LDOX and UFGT genes.

The use of different methodologies in the determination of antioxidant properties of plant extracts reflects a different mode of action of antioxidant molecules and can provide additional information on antioxidant activity and use of the extract. The trend of antioxidant activity was dependent on the cultivar, the stage of development of the fruit and within the same cultivar the methodology of evaluation.