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Effects of the Degree of Ripeness on the Phenolic and Carotenoid Profiles in Different Tissues of *Citrus* Fruits from Southern Italy

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Citrus are fruit crops that are consumed worldwide and have a great importance in the agricultural sector of Southern Italy. The region of Sicily is one of the largest producer of citrus fruits in Europe, and about 60% of the fruits produced in Italy are produced in this region.¹ Citrus fruits are popular amongst consumers due to their pleasant organoleptic characteristics and the high content of bioactive compounds, e.g., carotenoids and phenolic compounds. To date, a large number of phytochemicals have been identified in juices from ripe fruits, however, few investigations have researched the changes in phytochemicals occurring in the different tissues, e.g., albedo, flavedo and juice vesicles, during fruit development. In this study, the effects of the ripening stage on the different tissues of four citrus cultivars grown in Sicily, Southern Italy, were investigated.

Fruits from pummelo (*C. grandis* L. Osbeck) cv 'Chandler', lemon (*C. limon* L. Burm. f) cv. 'Akragas', and sweet orange (*C. sinensis* L. Osbeck) cvs 'Tarocco 57-1E-1' and 'Washington navel', were grown in the experimental fields of Acireale (37°36'31" N, 15°09'56" E) and Palazzelli (37°20'22" N, 14°53'31" E), Sicily, Southern Italy, and harvested every five weeks from September 2018 to January 2019. In total, fruits were collected at four ripening stages, from immature to full maturity. Fruits were washed with distilled water, and manually sectioned in albedo, flavedo and juice vesicles. Then, the citrus tissues were stored at -80°C, freeze-dried, milled, and kept under vacuum at RT.

Phenolic compounds were extracted and analysed by UHPLC-MS/MS as described by Ferguson *et al* (2018).² Carotenoids were extracted and analysed by HPLC-DAD as described by Multari *et al* (2018).³

In all the selected fruits, albedo and flavedo had higher cumulative phenolic contents than the juice vesicles, at all the ripening stages. Hesperidin resulted the main phenolic compound in the tissues of *C. limon* and *C. sinensis* cvs Tarocco and Washington, whereas *C. grandis* was the richest in narirutin, e.g., 4172 ± 240 mg kg⁻¹ dw (unripe fruit). Overall, cumulative phenolic acids reduced during ripening, whereas elevated levels of flavonoids were detected at full maturity. Caffeic, *p*-hydroxybenzoic and ferulic acids were major compounds in the juice vesicles of unripe *C. grandis*, whereas apigenin-7-*O*-glucoside was the predominant compound in the flavedo from full mature *C. limon* (43.1 ± 6.11 mg kg⁻¹ dw).

The carotenoid composition of the selected fruits resulted diverse. Across the samples, the flavedos were the tissues with the highest carotenoid content, and distinguished from albedos and juice vesicles due to great levels of lutein, β -cryptoxanthin, and (*all-E*)-violaxanthin. (*all-E*)-Violaxanthin increased significantly ($p < 0.05$) in the flavedo of *C. limon* during ripening. Juice vesicles showed carotenoid contents that increased constantly during ripening. *C. sinensis* cv. 'Washington navel' had juice vesicles with the highest content of cumulative carotenoids.

Results from the present investigation showed that the levels of phenolic compounds and carotenoids of citrus fruits from Sicily changed significantly over the ripening process. Overall, the tissues flavedo and albedo resulted valuable sources of phytochemicals. This information is useful for a comprehensive utilization of all the citrus tissues by the food industry.

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