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Salvatore Multari[1*], Silvia Carlin[1], Vincenzo Sicari[2], and Stefan Martens[1]

[1] Research and Innovation Centre, Fondazione Edmund Mach, via Edmund Mach 1, 38010, San Michele all' Adige, Trento, Italy.

[2] Università Mediterranea di Reggio Calabria, Dipartimento di Agraria, Contrada Melissari, 89124 Reggio Calabria, Italy

*Corresponding author (S. Multari). Tel.: +39 0461 615756; e-mail: salvatore.multari@fmach.it; s.multari@hotmail.it

ORCID: S Multari (0000-0002-0540-5352), S Carlin (0000-0003-0398-6947), V Sicari (0000-0002-0540-5352), S Martens (0000-0001-9913-4882).

ABSTRACT

Processing citrus fruits into juice generates large amounts of by-products, mainly pomaces. This study aimed to perform a comprehensive analysis of the composition in phenolic compounds, carotenoids, and volatile organic compounds (VOCs) of juices and pomaces of four citrus fruits from Southern Italy, i.e., mandarin, lemon, orange, and bergamot. Juices were produced by squeezing the fruits into an electrical juicer, whereas pomaces were obtained as by-products of the juice extraction. The phytochemical content of the samples was studied by targeted LC and GC approaches. Results indicated that lemon provided the juice with the greatest phenolic content. It was abundant in eriocitrin (90.9 ± 10.8 mg kg^{-1} FW), isorhamnetin 3-*O*-rutinoside (47.3 ± 8.03 mg kg^{-1} FW), and rutin (78.9 ± 14.5 mg kg^{-1} FW). Likewise, lemon pomace was the richest in phenolics, mostly narirutin (130 ± 14.7 mg kg^{-1} FW). As regards carotenoids, mandarin and orange pomaces were equally ($p > 0.05$) prominent sources of the compounds, providing primarily lutein and *β*-cryptoxanthin. The phytochemical profile of lemon and mandarin pomaces was unknown up to date. Bergamot accumulated great amounts of VOCs. In particular, bergamot juice was rich in monoterpenes, e.g., α-pinene (375 ± 62.7 mg kg^{-1} FW) and γ-terpinene (551 ± 67 mg kg^{-1} FW). The study investigated for the first time the carotenoid and VOCs profiles of bergamot products, and of mandarin and lemon pomaces. Since, citrus pomaces contained great amounts of phytochemicals, they should find new applications in the food and cosmetic industries.

Keywords: aroma; mandarin; lemon; sweet orange; bergamot; pigments.

1. INTRODUCTION

Citrus juices are popular products across the world. Italy is a principal producer of citruses in the Mediterranean area, and in 2017 it produced > 2.5 million tons of fruits, originating generally from the Southern regions [1]. About 40% of the harvested citruses are transformed into juices. As a consequence, vast amounts of by-products are generated, predominantly pomaces that consist of endocarp membranes, seeds, residues of the juice sacs, and albedo. The chemical composition of the pomace depends on the type of citrus utilized. In any case, valuable phytochemicals can be obtained from the citrus pomaces, such as phenolic compounds, carotenoids, and VOCs. It is acknowledged that numerous factors, such as the genotype, environmental conditions, and degree of maturity, influence the phytochemical profile of citrus and fruits in general [2]. Although there is no homogeneity in the data, it is inferred that temperate climates might promote the synthesis of phytochemicals, due to the protective effects against the UV radiations [3]. As formerly reported [4], phytochemicals in citrus fruits are modulated by the ripening process, with phenolic compounds decreasing over maturity, whereas, carotenoids are at the greatest levels in fully mature fruits.

Phenolic compounds are widespread in the plant kingdom, with the groups of flavanones and flavones, e.g., naringenin, hesperidin, narirutin, being the most ubiquitous in the citrus fruits [5]. They are bitter and colorless compounds abundant in citron, lemon, bergamot, orange, and kumquats [6]. Carotenoids are a particular group of isoprenoids that are classified in carotenes (compounds having only carbon and hydrogen atoms) and xanthophylls (compounds containing oxygenated functional groups) [7]. Citruses are usually richer in xanthophylls than in carotenes. The color of citruses depends mainly on

62 the presence of carotenoids [8], hence, the pigments influence the marketability of the
63 fruits. VOCs have a great effect on citrus aroma. Hundreds of VOCs have been found in
64 citrus fruits [9], however, only few tens contribute substantially to the perceived aroma,
65 e.g., linalool, β-myrcene, limonene, and valencene. The orange flavor is one of the most
66 sold flavoring agent worldwide [10], as a result, the economic value of the aroma-active
67 compounds found is citruses is huge. Over the last decade, the technique of SPME has
68 given momentum to the study of citrus aroma, however, researchers have investigated
69 largely the essential oils.

70 Despite the huge volume of byproducts generated by the juice industry, the
71 phytochemical composition of the pomaces is largely unknown, and the recovery rate is
72 rather low. Several researchers have investigated the citrus juices [11–13], and to the
73 authors' knowledge, there is only the study performed by Russo et al. [14] that evaluated
74 at the same time the flavonoid composition of bergamot juice and pomace. Even with this
75 study, the research performed on bergamot fruit is limited, as the fruit is almost exclusively
76 produced in a small area of the Southern Italy. Indeed, the carotenoid and VOCs
77 compositions of bergamot remain unknown. In addition, the pomaces derived from lemon
78 and mandarin processing have not been fully investigated hitherto [15]. Exploring the
79 phytochemical composition of different citruses and discriminating between the fractions
80 of juice and pomace, would help understand the distribution of bioactive compounds in
81 the tissues, following juice preparation. The purpose of this investigation was (i) to
82 perform a comprehensive analysis of the phytochemical composition of the selected citrus
83 tissues, (ii) to study how the two types of matrices (juice *versus* pomace) would
84 differentiate in the distribution of individual phenolics, carotenoids, and VOCs, (iii) to

reflect on the utilization of citrus pomaces as new functional ingredients. To this aim, four species of citrus from the Province of Reggio Calabria (Southern Italy), namely mandarin cv. 'Satsuma', lemon cv. 'Femminello Comune', orange cv. 'Valencia', and bergamot cv. 'Fantastico', were investigated. Information from this study will assist the food industry in the search of sources of bioactive compounds, and new substrates to produce food and beverages.

2 MATERIALS AND METHODS

2.1 Plant materials and sample preparation

Fruits from mandarin [*Citrus reticulata* Blanco (L).] cv. 'Satsuma', lemon [*C. limon* (L.) Burm.] cv. 'Femminello Comune', sweet orange [*C. sinensis* (L.) Osbeck] cv. 'Valencia', and bergamot [*Citrus bergamia* Risso] cv. 'Fantastico', were collected in Reggio Calabria (Latitude: 37°56′06″ N, Longitude: 15°55′02″ E, Height: 138.747m, Calabria, Southern Italy). Fruits were picked at full maturity in January 2019. Experiments were performed with four biological replicates (four trees), and each biological replicate consisted of ten healthy fruits. Trees were grown under conventional agricultural practices. Fruits were washed and peeled (removal of the flavedo). Samples were prepared by introducing the fruit pulps into an electric juicer (Bosch – MESM731M). Juices were collected and filtered through paper filters (Melitta® Coffee Filters 1x2), whereas, pomaces were obtained as by-products from the juice extraction. Fresh samples were used to perform the VOCs analysis. For phenolic and carotenoid analyses, samples were freeze dried and milled. The percentage of moisture was recorded (Table S1) and the final data were corrected for the moisture content, to have consistency with data from VOCs. Data are expressed

as mg kg^{-1} of FW. Overall, the following samples were produced: bergamot juice (BeJ), bergamot pomace (BeP), lemon juice (LeJ), lemon pomace (LeP), mandarin juice (MaJ), mandarin pomace (MaP), orange juice (OrJ), and orange pomace (OrP).

2.2 Extraction of free phenolic compounds and analysis by UPLC-MS/MS

The extraction of the free phenolic compounds was performed as described by Multari et al. [4], and rosmarinic acid was used as internal standard to a final concentration of 3 mg L^{-1}.Approx. 100 mg of samples were weighed into 15 mL tubes and 4 mL of 80% methanol were added. Samples were vortexed, and mixed on an orbital shaker for 20 min at room temperature (RT), then transferred into a US bath (Falc Intruments s.r.l., Bergamo, Italy) and extracted for 10 min at RT, 40 KHz. The extraction continued through maceration for 48 at 5 °C in the dark, although every 24 h, the samples were brought to RT and mixed on an orbital shaker for 20 min. Eventually, samples were centrifuged (10 min; 1800×*g*; 4°C), filtered through 0.22 µm PTFE membranes, and stored at -80 °C until MS analysis. The UPLC-MS/MS analysis was conducted on a Waters Acquity UPLC system (Milford, MA, USA), equipped with a binary pump, an online vacuum degasser, an autosampler, and a column compartment. The identification of compounds was performed on a Waters Xevo TQMS instrument, equipped with an electrospray (ESI) source. Compounds were identified by comparing the retention time and the spectral characteristic of the peaks with those of authentic standards. Multiple reaction monitoring (MRM) was used for quantification (Table S2). The cumulative contents of phenolic compounds (CPC) was determined by summing up the individual compounds found in the same tissue.

2.3 Extraction of carotenoids and analysis by HPLC-DAD

The extraction of carotenoids was performed as described by Multari et al. [4]. Carotenoids were extracted with the mixture MeOH/Acetone/Hex, 25/25/50, v/v/v, and saponified overnight at RT with 12% KOH in MeOH (w/v). Carotenoids were analyzed on an Agilent 1200 HPLC-DAD instrument (Agilent Technologies, Wokingham, UK). The chromatographic separation was carried out at 35 °C on an YMC C30 Carotenoid column (250 x 2.1 mm; 3mm i.d.) from CPS (Milan, Italy). The mobile-phase solvents were (A) acetonitrile/MBTE/methanol, 60/20/20, v/v/v, and (B) water. The flow rate was 0.4 mL min^{-1}, the injection volume was 10 μL, and the DAD was set at 400–500 nm. The identification of carotenoids was performed by comparing the UV-Vis spectral data of the samples with those of the external standards, namely, (*all-E*)-violaxanthin, lutein epoxide, antheraxanthin, (*all-E*)-lutein, (*all-E*)-zeaxanthin, β-criptoxanthin, α-carotene, and (*all-E*)-β-carotene (Table S3). The tentative identification of luteoxanthin was carried out by means of published information about the UV-vis spectrum and RT, on analogous YMC C30 Carotenoid columns [16]. Luteoxanthin was quantified against (*all-E*)-violaxanthin. For quantification purposes, calibration curves of the authentic standards were prepared with twelve points and injected in duplicate in the HPLC-DAD. The cumulative contents of carotenoids (CCC) was determined by summing up the individual compounds found in the same tissue.

2.4 SPME extraction and GC-MS analysis of VOCs

SPME was performed adapting the conditions described by Šuklje et al.[17]. In a 20 mL vial were introduced approx. 100 mg of sample, 2.0 g of sodium chloride, and 2.5 mL of

153 purified water. The compound 2-octanol was used as internal standard to a final
154 concentration of 21.3 μg L^{-1}. The GC analysis was performed on a Trace GC Ultra gas
155 chromatograph coupled with a TSQ Quantum Tandem mass spectrometer (Thermo
156 Electron Corporation, Waltham, MA USA). The compounds were separated using a VF-
157 Wax® column (100% polyethylene glycol; 30 m × 0.25 mm × 0.25 μm, from Agilent,
158 Folsom, CA). The GC oven parameters were as follows: initial temperature was 40 °C,
159 maintained for 4 min, followed by an increase to 60 °C at a rate of 2 °C min^{-1}, the oven
160 was then maintained at 60 °C for 1 min, then a rate of 5 °C min^{-1} until 190 °C for 1 min
161 and a rate of 10 °C min^{-1} until 230 °C maintained for 4 min; GC inlet temperature of 250
162 °C. The total cycle time was 50 min. The electron ionization occurred at 70 eV, and
163 spectra were collected in the mass range 40–400 *m/z* with an acquisition rate of 200
164 spectra/s and acquisition delay of 120 s. Data were processed with the software
165 XCALIBUR™ 2.2.

166 VOCs were identified by comparison with reference standards (Table 3). Standards were
167 dissolved in pure ethanol and introduced into a mixture having sugar and acid
168 compositions equivalent to those of the citrus juices. When the reference standards were
169 not available, mass spectral databases were used: NIST 2.0, Wiley 8, and FFNSC 2. The
170 cutoff for the similarity match was > 850. VOCs were semi-quantified using the internal
171 standard and their relative amount was expressed as mg kg^{-1} FW.

172

173 **2.5 Statistical analysis**

174 Data reported are means of four independent observations and values are expressed as
175 mean ± SD. Differences among groups were considered significant at $p < 0.05$. Principal

component analysis (PCA) was applied to identify the components that contributed mostly to the variability amongst the samples. The citrus samples (observations) represented the scores, whereas, the phytochemical variables represented the loadings. The missing values (those below LOD) were imputed using a random value between the percentile 2.5 and 5 of the LOD value for the corresponding compound. The PCA was performed on mean-centered data that were scaled by the standard deviation ("unit variance" scaling). Selected data were analyzed using one-way ANOVA followed by post-hoc Tukey HSD test for multiple comparison. The statistical analysis was performed using PAST 3.24 (Oslo, Norway).

3 RESULTS AND DISCUSSION

3.1 Determination of free phenolic compounds

At both tissue and species levels, the selected citrus fruits presented dissimilarities in the phenolic profiles, indicating different phenolic metabolism and distribution. Figure 1 shows that pomaces were richer in CPC than juices all the time. Significant differences were observed amongst the CPC of pomaces ($p < 0.001$), which were very abundant in phenolics. The CPC ranged from 410 ± 36.8 to 886 ± 43.2 mg kg^{-1} in BeP and LeP, respectively. OrP and LeP ($p > 0.05$) showed the greatest CPC. Similarly, significant differences were found amongst the CPC of the juices ($p < 0.001$). LeJ presented the highest value of CPC (357 ± 51.3 mg kg^{-1}), followed by MaJ (259 ± 21.9 mg kg^{-1}), OrJ (198 ± 30.0 mg kg^{-1}) and BeJ (169 ± 10.1 mg kg^{-1}). Bergamot resulted the matrix with the lowest phenolic content, having the smallest CPC in both juice and pomace. Results indicate that when fruits are juiced, only a small portion of the phenolic compounds migrates into the juice, whereas, the pomace retains most of them. In this regard, pomaces can be considered side-products of great value.

Concerning the individual phenolics, Table 1 shows that citrus juices and pomaces differed highly in the quantity of the single compounds. Across the samples, the most prominent compound was hesperidin, followed by narirutin and eriocitrin, with levels being higher in pomaces than in juices. Hesperidin peaked in OrP (377 ± 17.2 mg kg^{-1}), but amongst the juices, MaJ was the richest in the compounds (157 ± 14.3 mg kg^{-1}). Eriocitrin was abundant in lemon, with levels being almost two times higher in LeP (156 ± 26.2 mg kg^{-1}) than in LeJ (90.9 ± 10.8 mg kg^{-1}). Narirutin concentrations were nearly ten times higher in OrP (141 ± 11.5 mg kg^{-1}) than in OrJ (15.6 ± 2.89 mg kg^{-1}). These results agree

209 with other investigations performed on oranges and lemons, in which hesperidin, narirutin,
210 and eriocitrin resulted the predominant compounds, regardless of the degree of fruit
211 processing and/or origin [18–20]. Flavonoids such as luteolin, isorhamnetin 3-*O*-
212 rutinoside, and rutin were ubiquitous but with concentrations that varied largely, and
213 resulted minor in some samples. Luteolin was a major compound of MaP (18.8 ± 2.03 mg
214 kg^{-1}) and OrP (41.7 ± 3.13 mg kg^{-1}), however, it was found at minor concentrations in LeP,
215 LeJ, and BeJ (approx. 1 mg kg^{-1}; $p = 0.04$). Isorhamnetin 3-*O*-rutinoside was marginal in
216 OrJ and BeJ (< 2.5 mg kg^{-1}; $p = 0.013$), nevertheless, it was detected at great levels in
217 LeP (47.3 ± 8.03 mg kg^{-1}). Rutin fluctuated to a great extent amongst the juices, as peaked
218 in LeJ (7.89 ± 1.45 mg kg^{-1}), remained sizeable in BeJ (4.18 ± 0.70 mg kg^{-1}), and dropped
219 dramatically in MaJ and OrJ (approx. 1.30 mg kg^{-1}; $p > 0.05$). The high levels of rutin in
220 LeP (> 110 mg kg^{-1}) are confirmed by the investigation performed by Papoutsis et al [21].
221 Amongst the eight detected phenolic acids, ferulic, neochlorogenic, and vanillic, resulted
222 the principal. Since phenolic acids are very hydrophilic compounds [22], it was partly
223 unexpected that in some cases they were more abundant in the pomaces than in the
224 juices. For instance, mandarin provided 8.45 ± 1.49 and 1.53 ± 0.23 mg kg^{-1} of ferulic acid
225 in pomace and juice, respectively. A similar trend was observed in orange for vanillic acid,
226 e.g., 20.0 ± 3.85 mg kg^{-1} in pomace, and 9.91 ± 0.89 mg kg^{-1} in juice. This could be
227 ascribed to the processing method, as pomaces retained a high degree of moisture (>
228 70% w/w; Table S1). The levels of two phenolic acids identified in OrP were comparable
229 to those reported by Montero-Calderon et al. [23], namely ferulic acid (> 5 mg kg^{-1}) and
230 *p*-coumaric acid (< 0.5 mg kg^{-1}). It is worth mentioning that the flavonol quercetin and its
231 derivative quercetin 3,4'-*O*-diglucoside were found only in bergamot. Quercetin was

232 detected at elevated concentrations in both pomace (126 ± 18.2 mg kg^{-1}) and juice (56.7
233 ± 9.01 mg kg^{-1}). Additionally, bergamot stood out for the unique presence of luteolin 7-*O*-
234 glucoside (17.5 ± 1.06 and 5.56 ± 1.06 mg kg^{-1} in BeP and BeJ, respectively), and the
235 prominent levels of hesperidin 7-*O*-glucoside (38.5 ± 3.11 and 16.1 ± 2.87 mg kg^{-1} in BeP
236 and BeJ, respectively). These are flavonoids of pharmacological importance exerting anti-
237 inflammatory, anti-proliferative, and anti-hypertensive activities [24]. Despite several
238 authors having investigated the phenolic composition of a number of citrus species, few
239 studies have explored the profile of bergamot (*C. bergamia*) [14, 25, 26]. This is likely due
240 to bergamot being cultivated exclusively in a small geographical area of the Southern Italy
241 [26]. Thus, the indications obtained here on the phenolic profile of bergamot, could
242 contribute to the valorization of this fruit by the local food industry.

243 PCA was performed to group the citrus samples according to their phenolic profile, using
244 the metabolites listed in Table 1. Figure 2 shows that the first two principal components
245 (PC) justified a variation of 62.8%, with PC1 and PC2 accounting for 40.3 and 22.5% of
246 variation, respectively. In the PCA plot, the citruses took up different spatial distributions.
247 PC1 enabled a clear discrimination between the orange and mandarin samples on the
248 right-hand side, and the bergamot and lemon samples on the left-hand side. This might
249 be ascribed to mandarin and orange being the only samples to provide *p*-coumaric acid
250 and 3-hydroxyphloretin, whereas, lemon and bergamot provided hesperidin 7-*O*-
251 glucoside and bergaptol. Conversely, PC2 did not allow a clear-cut distinction, and
252 samples of oranges located on both the positive and negative sides. This was due to OrJ
253 having a phenolic profile close to that of MaJ, e.g., no significant differences ($p > 0.05$) in
254 the levels of eriocitrin, diosmin, and naringenin. In general, PCA discriminated the

255 samples according to the species, but did not separate distinctly between the tissues, i.e.,
256 juice and pomace. Results from the PCA analysis (Figure 2a) suggest that the noticeably
257 high levels of hesperidin 7-*O*-glucoside, eriocitrin, and neochlorogenic acid can be
258 considered characteristics of bergamot, lemon and mandarin products, and could be used
259 as markers of fruit authenticity.

260

261 **3.2 Determination of carotenoids**

262 The levels of carotenoids varied amongst the selected citrus fruits, and significantly
263 different ($p < 0.001$) concentrations were found within the two groups of juice and pomace.
264 The CCC of the samples is shown in Figure 3. Amongst the juices, the highest CCC was
265 determined in orange (12.5 ± 1.07 mg kg^{-1}), followed by lemon (4.12 ± 0.48 mg kg^{-1}) and
266 bergamot (4.06 ± 0.28 mg kg^{-1}). Colorful carotenoids were not found in mandarin juice,
267 which indeed resulted colorless (Figure S1). In the pomaces, carotenoids were found at
268 high levels, with concentrations being greater than those of the juices. The CCC of
269 mandarin (44.8 ± 4.13 mg kg^{-1}) and orange (43.4 ± 4.75 mg kg^{-1}) pomaces were in the
270 same range ($p > 0.05$), on the contrary, the CCC of bergamot (11.0 ± 1.21 mg kg^{-1}) and
271 lemon (6.05 ± 0.45 mg kg^{-1}) pomaces were significantly ($p < 0.05$) lower. In both types of
272 tissues, lemon provided the lowest values of CCC. Previous studies performed on lemons
273 demonstrated that the flesh and pomace of the fruit are modest sources of carotenoids
274 [27, 28]. It is likely that pomaces had greater CCC than juices due to the lipophilic nature
275 of carotenoids, which restrain from leaking into the aqueous environment of the juice [29].
276 Previous studies have demonstrated that the pomaces obtained following juice production
277 are richer in carotenoids than the juice counterparts [30, 31].

The qualitative and quantitate carotenoid composition of the samples is detailed in Table 2. A total of eleven carotenoids were detected by HPLC-DAD. In the juices, lutein was the main pigment, since being the predominant compound in all the samples, namely, OrJ (5.64 ± 0.90 mg kg^{-1}), LeJ (3.89 ± 0.45 mg kg^{-1}), and BeJ (3.67 ± 0.25 mg kg^{-1}). Juices provided exclusively xanthophylls, apart from orange that held appreciable levels of carotenes, i.e., α-carotene (1.29 ± 0.20 mg kg^{-1}) and *β*-carotene (1.87 ± 0.27 mg kg^{-1}). OrJ provided also antheraxanthin (1.58 ± 0.26 mg kg^{-1}). Indeed, it is known that juices from *Citrus sinensis* exhibit a complex carotenoid profile [32, 33]. In LeJ, lutein resulted the principal compound. This observation is corroborated by investigations performed on the different lemon tissues [34, 35]. The fruit of bergamot is cultivated mainly to produce aroma compounds intended for the cosmetic industry, and its juice remains largely unused [36]. To the authors' knowledge, the literature does not provide studies detailing the carotenoid composition of BeJ. In the present study, BeJ showed a similar profile to LeJ, with lutein and *β*-cryptoxanthin being main carotenoids. In general, juices were characterized for the most part by xanthophylls. This is confirmed by other investigations performed on citruses from the Mediterranean basin [33, 37].

Citrus pomaces retained considerable amounts of carotenoids. Contrary to what observed in the juice, mandarin pomace accumulated large amounts of compounds. β–Cryptoxanthin was the main carotenoid (9.14 ± 1.11 mg kg^{-1}), followed by antheraxanthin (7.17 ± 1.33 mg kg^{-1}), and neoxanthin (6.75 ± 0.52 mg kg^{-1}). The substantial level of *β*-cryptoxanthin in mandarin is nutritionally relevant as the compound is a precursor of the vitamin A [38]. In orange pomace, antheraxanthin (15.5 ± 2.88 mg kg^{-1}) and lutein (7.89 ± 1.02 mg kg^{-1}) were the predominant carotenoids, and opposite to

301 the juice, orange pomace retained ample levels of violaxanthin (6.14 ± 1.01 mg kg^{-1}).
302 Investigations performed on orange cv. 'Valencia' showed that the fruit can accumulate
303 the different isomers of violaxanthin, namely 9-*cis*-violaxanthin and all-*E*-violaxanthin [35,
304 39]. It is worth highlighting that OrP provided the highest concentrations of violaxanthin
305 and the lowest levels of *β*-cryptoxanthin, whereas, MaP was low in violaxanthin and rich
306 in *β*-cryptoxanthin. This opposite pattern is not accidental or due to environmental factors,
307 as investigations performed on 'Satsuma' mandarin and 'Valencia' orange indicated that
308 the *β*-cryptoxanthin/violaxanthin ratio is genetically regulated [27, 40]. In LeP and BeP,
309 the levels of carotenoids lessened compared to the other samples, nevertheless, lutein
310 was found at substantial concentrations (4.09 ± 0.39 and 6.43 ± 1.02 mg kg^{-1} in LeP and
311 BeP, respectively). LeP was the only to provide *β*-carotene (0.73 ± 0.04 mg kg^{-1}), yet at
312 low quantities, whereas BeP presented appreciable levels of neoxanthin (2.51 ± 0.29 mg
313 kg^{-1}). Comparable concentrations of *β*-carotene were also found in lemon tissues by
314 Yungyuen et al. [35]. On the whole, citrus pomaces were richer in every single carotenoid
315 than juices. This might be due to pomaces being complex matrices, and carotenoids
316 being conjugated with the components of the matrix [41]. Citrus pomaces have relatively
317 high hydrophobicity (crude fat, 5% w/w), viscosity, and dietary fiber contents (crude fibers
318 7% w/w) [42]. Pomaces, having the carotenoids embedded into the cells, might release
319 bioactive compounds if subjected to effective extraction methods. Nevertheless, juices
320 provide carotenoids with a greater degree of bioaccessibility [43], and for this reason, the
321 bioaccessibility e/o bioavailability of carotenoids must be considered when reflecting on
322 the consumption of citrus products.

323 The PCA (Figure 4) was applied to discriminate the samples according to their carotenoid
324 profile. The two PCs accounted for 72.1% of the total variance (PC1, 50.5%; PC2, 21.6%).
325 PC1 revealed that the pomaces located on the positive side, apart from LeP. The
326 carotenoids accountable for this separation were mostly antheraxanthin, lutein epoxide,
327 and neoxanthin, since LeP was the only to be devoid of them. Key carotenoids for PC2
328 were *α*-carotene and *β*-carotene, which supported the discrimination of OrJ on the
329 positive side of the axis. Indeed, the carotenoid profile of OrJ was qualitatively more
330 complex than the other juices. It is worth mentioning that LeJ and LeP, having limited
331 metabolite variability, assembled on the negative side of PC1. Similarly, bergamot and
332 lemon juices shared the same quadrant, as being characterized by a close metabolite
333 profile, mostly in terms of luteoxanthin, lutein, and *β*-cryptoxanthin.

334

335 **3.3 Determination of VOCs**

336 The list of the VOCs identified in the citrus samples is reported in Table 3. Overall, 79
337 compounds were detected, of which seven were identified only in the juices, i.e.,
338 tridecene, octanal, 3-hexenyl acetate, 2-hexenol, α-selinene, γ-muurolene, and dimethyl
339 anthranilate. VOCs were grouped into nine categorizes (Figure 5B), namely
340 monoterpenes, monoterpene alcohols, sesquiterpenes, alcohols, esters, ketones,
341 aldehydes, acids, and others (including alkenes, lactones, furans, etc.). Monoterpenes
342 stood out as the main class, regardless of the species and/or the tissue examined. Juices
343 resulted richer in VOCs than pomaces, apart from mandarin in which VOCs distributed
344 equally ($p > 0.05$) between the two fractions. This might be due to mandarin having a
345 unique physical composition characterized by the lack of albedo, i.e., the white inner layer

346 of the peel [44]. Mandarin pomace consists of seeds, endocarp membranes, and residues
347 of juice sacs, and is qualitatively and quantitatively different from the other citrus pomaces
348 (Table S1).

349 Limonene was by far the most abundant compound in all the samples, accounting on
350 average 43.8 ± 7.62 and 44.4 ± 6.13% of cumulative VOCs, in juice and pomace
351 respectively. For this reason, it was considered separately from the other VOCs (Figure
352 5A). Lemon and bergamot provided, respectively, the lowest and highest concentrations
353 of limonene in both tissues, e.g., 1138 ± 89.8 and 3553 ± 557 mg kg^{-1} in juices, 998 ±
354 60.5 and 2920 ± 160 mg kg^{-1} in pomaces. For orange juice, the concentrations of
355 limonene reported here (> 12000 mg kg^{-1}) were in line with those reported by Wei et al
356 [45] (> 13000 mg kg^{-1}), who also investigated juices from orange cv. 'Valencia'. Limonene
357 has a dim odor activity due to its high odor threshold (> 200 ppb), however, it is present
358 at such great levels that takes on a key role in the aroma of citrus products. Linalool was
359 another major VOC of the citrus samples, with concentrations ranging in juices from 11.4
360 ± 1.56 (MaJ) to 221 ± 23.2 (BeJ) mg kg^{-1}, and in pomaces from 13.5 ± 1.17 (MaP) to 195
361 ± 29.5 (BeP) mg kg^{-1}. Previous authors already regarded linalool as the most prominent
362 monoterpene alcohol of citruses [46, 47]. It is acknowledged that terpinen-4-ol and α-
363 terpineol could be generated from limonene and linalool following thermal treatments [48],
364 hence, the process of juicing might have augmented their development. This could justify
365 their occurrence at elevated levels across the samples. Although most of the VOCs were
366 ubiquitous, several compounds resulted species specific, e.g., citronellal, linalyl acetate,
367 *o*-cymene, α-bergamotene, and γ-muurulene were found in bergamot, α-salinene, 2-
368 hexenol, and octanal were distinctive of orange, 3-methylbutanol, dimethyl anthranilate,

369 and tridecene were detected in mandarin. From the perspective of the tissue
370 distinctiveness, pomaces provided large amounts of nonpolar compounds, e.g., alkenes
371 and monoterpenes hydrocarbons. The concentration of β-pinene ranged from 30.6 ± 4.84
372 to 77.3 ± 14.5 mg kg^{-1} in OrP and BeP, respectively. B-myrcene peaked in mandarin (14.4
373 ± 2.84 mg kg^{-1}), and distributed entirely in the pomace. About 100% of the detected *o*-
374 cymene was found in pomaces, 48.9 ± 7.51 and 87.7 ± 16.5 mg kg^{-1} for BeP and MaP,
375 respectively. In addition, differences in the concentrations of individual compounds were
376 observed amongst the species, e.g., BeP provided VOCs, such as α-pinene, hexanal,
377 and 3-carene in at least twice the levels found in LeP.

378 The juices were abundant in polar oxygenated VOCs, e.g., linalool, 1,8-cineole,
379 dodecanal, ethyl hexanoate, and citronellal. This was due to the hydrophilic compounds
380 migrating into the aqueous environment following processing. In MaJ, alcohols
381 constituted > 45% of cumulative VOCs (not including limonene), with 1-heptanol (475 ±
382 41.7 mg kg-1) being by far the main compound. OrJ was rich in esters, which accounted
383 for approximately 20% of cumulative VOCs, e.g., ethyl butyrate and neryl acetate were
384 found at levels of 197 ± 37.0 and 18.8 ± 3.80 mg kg^{-1}, respectively. The relative proportion
385 of aldehydes was below 3% in BeJ and LeJ. Both samples showed similar levels (p >
386 0.05) of nonanal (< 0.9 mg kg^{-1}), hexanal and benzaldehyde (approx. 15 mg kg^{-1}),
387 however, two aldehydes were distinctive of BeJ and found in large quantities, e.g.,
388 citronellal and dodecanal, 114 ± 22.1 and 75.1 ± 11.2 mg kg^{-1}, respectively. Data from
389 the juices gave emphasis to the distinctiveness of bergamot in terms of aroma. Besides
390 being rich in hydrophilic compounds, BeJ resulted a remarkable source of hydrocarbon
391 terpenes, e.g., α-pinene (375 ± 62.7 mg kg^{-1}), γ-terpinene (551 ± 67.2 mg kg^{-1}), and

caryophyllene (54.1 ± 10.8 mg kg^{-1}). Results from this study agree with previous investigations in which monoterpene hydrocarbons, esters and aldehydes were major VOCs of citrus juices [49, 50].

The statistical tool of PCA was applied to discriminate the samples and identify the metabolites accountable for their separation. All VOCs listed in Table 3 were included in the analysis. PCA explained 51.5% of the total variability, with PC1 and PC2 accounting for 33.6 and 17.9% of the total variance, respectively. PC1 was negatively correlated (data not shown) with the highly polar esters and alcohols, e.g., neryl acetate, octyl acetate, ethyl butyrate, 1-pentenol, 1-hexanol, and 1-heptanol, and justified the positioning of LeJ, OrJ, and MaJ on the negative side of the axis. PC2 was positively correlated with the low polar terpenes, e.g., 1,8-cineole, β-pinene, γ-terpinene, sabinene, *p*-cymene, and limonene, and explained the presence on the positive side of BeP, MaP, and OrP, which resulted particularly rich in terpenes. In relation to VOCs, the PCA showed that pomaces tended to differentiate from the juices.

4 CONCLUSIONS

The present study investigated the phytochemical profiles, e.g., phenolic compounds, carotenoids, and VOCs of juice and pomace from citrus fruits cultivated in Southern Italy, namely mandarin cv. ‘Satsuma’, lemon cv. ‘Femminello Comune’, orange cv. ‘Valencia’, and bergamot cv. ‘Fantastico’. The selected samples presented complex phytochemical profiles, characterized by the detection of 30 phenolic metabolites, eleven carotenoids, and 79 volatile compounds. The pomaces were more abundant in phenolic compounds and carotenoids than the juices, whereas, juices were richer in VOCs. Remarkably, each species distinguished in a different group of metabolites. In particular, orange and lemon pomaces stood out as the richest in free phenolic compounds. Mandarin pomace was found to be a prominent source of carotenoids, mostly xanthophylls. Bergamot juice held a very diverse volatile profile, characterized by great levels of monoterpenes. Despite bergamot juice being exceptionally abundant in VOCs, bergamot pomace maintained a notable volatile profile. This investigation is the first exploring in depth the carotenoid and volatile profiles of bergamot juice and pomace. Additionally, the phytochemical profile of lemon and mandarin pomace was unknown up to date. Overall, the data provided put emphasis on the potential of the by-products generated from the juice industry. Regardless of the species, the citrus pomaces resulted rich in phytochemicals and could be added to processed products to provide the citrus flavor and/or to supplement the content of bioactive compounds. They are valuable raw materials, and should find applications in the food, cosmetics, and pharmaceutical industries.

ABBREVIATIONS

BeJ, bergamot juice; BeP, bergamot pomace; CCC, cumulative carotenoid content; CPC, cumulative phenolic content; CV, cultivar; LeJ, lemon juice; LeP, lemon pomace; MaJ, mandarin juice; MaP, mandarin pomace; OrJ, orange juice; OrP, orange pomace; PCA, principal component analysis; RI, retention index; RT, retention time; VOCs, volatile organic compounds.

DECLARATIONS

Funding. This study was funded by Symrise AG, Holzminden, Germany.

Conflicts of interest. Authors declare no conflict of interest.

Ethics approval. Not applicable. **Consent to participate**. Not applicable.

Consent for publication. All authors have reviewed and approved the final version of the manuscript for publication.

Availability of data and material. Raw data can be provided following individual requests by the reader(s).

Code availability. Not applicable.

Authors' contributions. S.M. designed the work, performed the analyses, analyzed the data, and wrote the manuscript; S.C. supervised the work on volatiles; S.V. provided the fruit samples and contributed to the design of the work; St.M. supervised the whole work.

Acknowledgments. Authors are thankful to Dr. Franciosi from E. Mach Foundation for sharing her lab facilities**.**

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601

TABLES AND FIGURES

Table 1. Phenolic contents of the juice and pomace samples.

	mandarin	orange	lemon	bergamot	mandarin	orange	lemon	bergamot
	juice				pomace			
	phenolic acids							
***p*-hydroxy-benzoic acid**	0.00 ± 0.00				0.37 ± 0.07			
***p*-coumaric acid**	0.00 ± 0.00	0.00 ± 0.00			0.43 ± 0.05	0.41 ± 0.05		
ferulic acid	1.53 ± 0.23	0.52 ± 0.06	0.00 ± 0.00	0.50 ± 0.09	8.45 ± 1.49	5.36 ± 0.43	0.35 ± 0.06	0.52 ± 0.10
caffeic acid	0.54 ± 0.09	0.00 ± 0.00	0.41 ± 0.08		0.00 ± 0.00	0.78 ± 0.10	0.00 ± 0.00	
crypto-chlorogenic ac.	0.41 ± 0.02			0.26 ± 0.03	0.98 ± 0.10			
chlorogenic acid	0.00 ± 0.00				1.45 ± 0.27			
neochlorogenic acid	1.64 ± 0.39			0.01 ± 0.00	7.90 ± 1.43			0.25 ± 0.02
vanillic acid	9.91 ± 0.89	1.56 ± 0.32	6.00 ± 0.92	5.25 ± 0.90	20.0 ± 3.85	16.0 ± 2.04	0.83 ± 0.15	4.23 ± 0.63
vanillin	1.18 ± 0.15	0.23 ± 0.04	1.25 ± 0.12	0.73 ± 0.14	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00

	mandarin	orange	lemon	bergamot	mandarin	orange	lemon	bergamot
	juice				pomace			
flavanones								
eriocitrin	3.39 ± 0.52	2.68 ± 0.46	90.9 ± 10.8	8.46 ± 0.47	13.8 ± 2.26	6.43 ± 0.61	156 ± 26.2	109 ± 12.9
narirutin	8.81 ± 1.01	15.6 ± 2.89	9.24 ± 0.17	5.74 ± 0.56	42.8 ± 4.44	141 ± 11.5	130 ± 14.7	67.7 ± 10.3
naringenin	0.72 ± 0.11	0.62 ± 0.10	0.47 ± 0.08	2.10 ± 0.36	0.59 ± 0.10	1.40 ± 0.22	4.66 ± 0.92	0.36 ± 0.07
apigenin	0.08 ± 0.01	0.22 ± 0.04	0.16 ± 0.04	0.00 ± 0.00	0.00 ± 0.00	0.25 ± 0.06	0.19 ± 0.04	0.21 ± 0.05
apigenin 7-*O*-glucoside	1.41 ± 0.26	0.00 ± 0.00	4.19 ± 0.61	2.66 ± 0.03	8.60 ± 1.05	1.44 ± 0.11	15.3 ± 1.11	7.64 ± 0.27
flavones								
tangeretin	0.26 ± 0.05	0.03 ± 0.00	0.03 ± 0.00	0.03 ± 0.00	17.1 ± 2.96	0.79 ± 0.15	0.03 ± 0.00	0.10 ± 0.02
diosmin	0.14 ± 0.02	0.16 ± 0.03	2.27 ± 0.40	0.61 ± 0.12	0.17 ± 0.03	0.09 ± 0.02	4.33 ± 0.79	0.99 ± 0.15
luteolin	3.25 ± 0.48	5.99 ± 1.00	1.06 ± 0.20	0.76 ± 0.14	18.8 ± 2.03	41.7 ± 3.13	0.74 ± 0.15	3.66 ± 0.63
luteolin 7-*O*-glucoside				5.56 ± 1.06				17.5 ± 1.06
furanocoumarins								
bergaptol			0.02 ± 0.00	0.11 ± 0.02			0.00 ± 0.00	0.09 ± 0.01

	mandarin	orange	lemon	bergamot	mandarin	orange	lemon	bergamot
	juice				pomace			
psoralen				0.42 ± 0.03				0.53 ± 0.08
	flavonols							
quercetin				56.7 ± 9.01				126 ± 18.2
quercetin 3-*O*-rhamnoside	60.7 ± 5.04	40.1 ± 7.08	5.96 ± 1.15	1.81 ± 0.34	125 ± 11.1	150 ± 11.4	81.5 ± 13.1	3.66 ± 0.65
isorhamnetin 3-*O*- glucoside	0.19 ± 0.03	0.00 ± 0.00	6.46 ± 1.06	0.17 ± 0.03	2.15 ± 0.35	5.51 ± 0.91	9.27 ± 1.25	0.00 ± 0.00
isorhamnetin 3-*O*-rutinoside	5.80 ± 1.08	1.49 ± 0.25	47.3 ± 8.03	2.41 ± 0.46	17.5 ± 3.49	42.3 ± 2.17	80.7 ± 14.0	5.70 ± 0.57
kaempferol 3-*O*-rutinoside	0.24 ± 0.04	0.14 ± 0.03	0.46 ± 0.08	0.04 ± 0.01	3.54 ± 0.66	6.05 ± 0.85	1.58 ± 0.29	0.09 ± 0.01
rutin	1.25 ± 0.23	1.28 ± 0.22	7.89 ± 1.45	4.18 ± 0.70	5.40 ± 0.68	33.0 ± 3.77	141 ± 15.8	8.63 ± 0.73
quercetin 3,4'-diglucoside				1.32 ± 0.24				4.01 ± 0.77
	flavanons							
hesperidin	157 ± 14.3	126 ± 20.1	172 ± 21.6	54.0 ± 3.52	314 ± 21.2	377 ± 17.2	254 ± 30.5	10.1 ± 1.94
hesperidin 7-*O*-glucoside			0.13 ± 0.02	16.1 ± 2.87			5.88 ± 0.98	38.5 ± 3.11

	mandarin	orange	lemon	bergamot	mandarin	orange	lemon	bergamot
	juice				pomace			
	dihydrochalcones							
3-hydroxy-phloretin	0.01 ± 0.00	0.02 ± 0.00			0.05 ± 0.01	0.16 ± 0.03		

Data are expressed as means ± standard deviation (n =4). Values are expressed as mg kg^{-1} FW.

Table 2. Carotenoid contents of the juice and pomace samples.

	mandarin	orange	lemon	bergamot	mandarin	orange	lemon	bergamot
		juice				pomace		
violaxanthin					1.64 ± 0.38	6.14 ± 1.01		
neoxanthin					6.75 ± 0.52			2.51 ± 0.29
lutein epoxide			0.13 ± 0.02		4.81 ± 0.87	4.89 ± 0.38		0.58 ± 0.11
antheraxanthin		1.58 ± 0.26			7.17 ± 1.33	15.5 ± 2.88		
luteoxanthin		1.39 ± 0.19	0.89 ± 0.10	0.87 ± 0.11	3.65 ± 0.48	4.57 ± 0.59		3.95 ± 0.56
lutein		5.64 ± 0.90	3.89 ± 0.45	3.67 ± 0.25	4.31 ± 0.84	7.89 ± 1.02	4.09 ± 0.39	6.43 ± 1.02
zeaxanthin					4.30 ± 1.51	3.06 ± 0.60		
unknown 420/450/480		2.01 ± 0.33	0.65 ± 0.11		5.01 ± 0.59	4.46 ± 0.42	0.87 ± 0.15	

	mandarin	orange	lemon	bergamot	mandarin	orange	lemon	bergamot
		juice				pomace		
β-cryptoxanthin		0.74 ± 0.08	0.56 ± 0.09	0.52 ± 0.07	9.14 ± 1.11	1.21 ± 0.19	1.36 ± 0.17	1.51 ± 0.12
α-carotene		1.29 ± 0.20						
β-carotene		1.87 ± 0.27					0.73 ± 0.04	
CCC		12.5 ± 1.07	4.12 ± 0.48	4.06 ± 0.28	44.8 ± 4.13	43.4 ± 4.75	6.05 ± 0.45	12.0 ± 1.21

Data are expressed as means ± standard deviation (n =4). Values are expressed as mg kg^{-1} FW.

Compounds are listed in order of elution.

CCC means Cumulative Carotenoid Content.

Table 3. Volatile compounds (VOCs) from citrus juices and pomaces

code	compounds	RI	RT	identification	BeJ	LeJ	OrJ	MaJ	BeP	LeP	OrP	MaP
1	α-pinene	1011	5.27	MS, Std	375 ± 62.7	76.9 ± 3.00	39.0 ± 5.26	19.0 ± 3.10	211 ± 34.9	95.7 ± 19.1	11.1 ± 2.12	188 ± 26.1
2	α-thujene	1017	5.41	RI, MS	0.15 ± 0.02			0.10 ± 0.00	0.13 ± 0.01	0.24 ± 0.08		0.47 ± 0.09
3	1-decene	1035	5.86	RI, MS	0.37 ± 0.07	1.48 ± 0.25	1.65 ± 0.36	2.89 ± 0.37	0.66 ± 0.13	1.52 ± 0.29	1.45 ± 0.32	1.02 ± 0.18
4	ethyl butyrate	1035	5.87	MS, Std	271 ± 37.2	41.2 ± 8.06	197 ± 37.0	47.3 ± 2.25	430 ± 68.4	44.1 ± 8.75	83.8 ± 12.6	158 ± 29.2
5	camphene	1051	6.27	RI, MS	0.10 ± 0.01	0.14 ± 0.02			0.10 ± 0.01	0.14 ± 0.01		
6	hexanal	1075	6.86	MS, Std	12.6 ± 2.05	18.9 ± 3.59	69.8 ± 13.3	6.84 ± 1.32	48.7 ± 7.09	17.5 ± 2.35	62.6 ± 10.7	63.6 ± 12.2
7	3-carene	1134	8.41	MS, Std	9.90 ± 1.84	17.4 ± 3.37	0.25 ± 0.04	11.7 ± 1.01	7.86 ± 1.52	0.22 ± 0.01	1.07 ± 0.11	4.09 ± 0.80
8	β-pinene	1135	8.43	MS, Std	231 ± 25.3	48.9 ± 6.45	56.6 ± 10.4	108 ± 13.9	77.3 ± 14.5	49.6 ± 6.49	30.6 ± 4.84	50.3 ± 8.39
9	α-phellandrene	1152	8.89	MS, Std	143 ± 26.1	114 ± 4.19	138 ± 27.4	12.1 ± 1.89	144 ± 25.7	139 ± 15.2	27.2 ± 4.02	388 ± 48.8
10	β-myrcene	1155	8.95	MS, Std		7.03 ± 0.82	9.00 ± 1.12		2.73 ± 0.43	8.10 ± 0.74	6.23 ± 0.67	14.4 ± 2.84
11	α-terpinene	1168	9.31	RI, MS	1.12 ± 0.07	0.14 ± 0.01		0.18 ± 0.02	0.70 ± 0.04	0.55 ± 0.10		1.53 ± 0.29
12	1,8-cineole	1190	9.89	MS, Std	876 ± 101	139 ± 9.14	118 ± 22.2	6.35 ± 0.89	505 ± 92.8	143 ± 15.3	67.9 ± 5.08	178 ± 18.1
13	limonene	1198	10.11	MS, Std	3553 ± 557	1138 ± 89.8	1234 ± 178	1456 ± 259	2920 ± 160	998 ± 60.5	1003 ± 86.9	1031 ± 84.6

14	3-methylbutanol	1205	10.26	MS, Std				0.10 ± 0.00					
15	2-hexenal	1213	10.48	MS, Std	8.95 ± 1.49		2.41 ± 0.17	3.34 ± 0.42	7.67 ± 1.40		4.15 ± 0.67	7.81 ± 1.44	
16	ethyl hexanoate	1229	10.88	MS, Std	0.91 ± 0.14	0.61 ± 0.03	3.77 ± 0.49	1.10 ± 0.21	1.08 ± 0.17	0.55 ± 0.07	0.85 ± 0.10	0.77 ± 0.09	
17	1-pentanol	1234	10.99	MS, Std	6.40 ± 1.00				244 ± 21.5				
18	tridecene	1237	11.06	RI, MS				3.10 ± 0.28					
19	γ-terpinene	1239	11.12	MS, Std	551 ± 67.2	83.3 ± 7.03	1.77 ± 0.26	7.79 ± 1.09	325 ± 62.0	93.5 ± 8.90	1.50 ± 0.28	389 ± 49.0	
20	2-carene	1240	11.14	RI, MS	3.06 ± 0.16							7.75 ± 1.32	
21	*p*-cymenene	1263	11.73	MS, Std	198 ± 33.2	34.2 ± 2.64	23.6 ± 2.05	11.4 ± 2.04	91.1 ± 17.3	26.6 ± 3.63	15.2 ± 2.38	264 ± 36.6	
22	α-terpinolene	1276	12.05	MS, Std		27.3 ± 2.96		2.34 ± 0.40	149 ± 26.7	21.4 ± 1.37		12.2 ± 1.43	
23	*o*-cymene	1284	12.24	MS, Std	0.11 ± 0.02				48.9 ± 7.51			87.7 ± 16.5	
24	octanal	1284	12.25	MS, Std			0.37 ± 0.07						
25	3-hexenyl acetate	1308	12.83	MS, Std	90.8 ± 16.8	18.1 ± 2.33							
26	2-heptenal	1317	13.02	RI, MS					0.09 ± 0.01				
27	methyl heptenone	1319	13.4	RI, MS				0.10 ± 0.00	0.09 ± 0.01				

28	1-hexanol	1350	13.79	MS, Std	0.09 ± 0.01	2.05 ± 0.37	0.12 ± 0.03	4.17 ± 0.84	10.9 ± 1.90		1.17 ± 0.13	0.14 ± 0.02
29	3-hexenol	1370	14.25	RI, MS	0.24 ± 0.03				0.14 ± 0.02		0.71 ± 0.05	0.12 ± 0.09
30	5-hexenol	1388	14.67	MS, Std	20.8 ± 2.18	6.37 ± 0.96	3.08 ± 0.42	4.07 ± 0.64	31.8 ± 4.06	2.83 ± 0.46	5.28 ± 0.78	25.3 ± 1.52
31	nonanal	1390	14.71	MS, Std	0.82 ± 0.15	0.80 ± 0.14	0.66 ± 0.12	1.07 ± 0.19	0.75 ± 0.13	0.54 ± 0.07	0.52 ± 0.06	0.59 ± 27.5
32	2-hexenol	1403	15.0	RI, MS			1.50 ± 0.09					
33	tetradecene	1443	15.86	RI, MS	0.82 ± 0.07	1.42 ± 0.26	1.93 ± 0.24	2.26 ± 0.28	0.96 ± 0.17	1.69 ± 0.08	1.67 ± 0.32	1.11 ± 0.01
34	1-heptanol	1445	15.89	MS, Std	134 ± 9.72	172 ± 29.9	198 ± 35.3	475 ± 41.7	118 ± 22.2	209 ± 36.7	9.29 ± 0.94	57.4 ± 11.2
35	sabinene	1445	15.9	MS, Std	8.61 ± 1.46	2.82 ± 0.52	3.89 ± 0.59	4.65 ± 0.90	6.54 ± 1.11	3.26 ± 0.63	3.71 ± 0.45	3.52 ± 0.32
36	acetic acid	1451	16.02	RI, MS	0.90 ± 0.05	0.11 ± 0.02	0.10 ± 0.01	0.10 ± 0.00	1.06 ± 0.12			
37	citronellal	1469	16.4	MS, Std	114 ± 22.1				20.8 ± 3.69			
38	octyl acetate	1475	16.54	MS, Std	0.77 ± 0.02				0.27 ± 0.04			
39	2-ethyl-1-hexanol	1487	16.79	MS, Std	0.12 ± 0.02	0.52 ± 0.10	0.33 ± 0.04	0.28 ± 0.03	0.13 ± 0.03	0.59 ± 0.06	0.94 ± 0.10	0.20 ± 0.01
40	decanal	1499	17.04	MS, Std	0.09 ± 0.01		0.28 ± 0.05	0.22 ± 0.05	0.11 ± 0.02	0.10 ± 0.01		0.19 ± 0.05
41	camphor	14.99	17.05	MS, Std	13.9 ± 0.96	4.81 ± 0.80	9.73 ± 1.19	7.69 ± 1.43		3.79 ± 0.18	5.04 ± 0.94	8.90 ± 0.04

42	2-nonanol	1511	17.29	MS, Std	6.15 ± 1.18		3.66 ± 0.52		5.72 ± 1.14		2.29 ± 0.44	3.31 ± 0.51
43	benzaldehyde	1527	17.61	MS, Std	15.2 ± 2.65	13.9 ± 1.02	14.8 ± 0.70	26.7 ± 4.11	17.5 ± 3.37	13.9 ± 0.59	12.5 ± 2.42	18.5 ± 2.00
44	linalool	1551	18.09	MS, Std	221 ± 23.2	64.1 ± 2.26	84.4 ± 1.36	11.4 ± 1.56	195 ± 29.5	91.7 ± 11.0	63.9 ± 11.3	13.5 ± 1.17
45	longifolene	1555	18.17	MS, Std	4.05 ± 0.57	0.20 ± 0.04			0.21 ± 0.03	0.30 ± 0.04	0.14 ± 0.01	0.61 ± 0.09
46	1-octanol	1558	18.23	RI, MS			1.03 ± 0.06	0.34 ± 0.04		0.14 ± 0.01	1.61 ± 0.26	0.46 ± 0.09
47	linalyl acetate	15.61	18.32	RI, MS	7.84 ± 0.68				9.89 ± 1.35			
48	5-methylfurfural	1568	18.44	MS, Std	35.5 ± 33.12		32.3 ± 1.61		39.6 ± 5.87	35.0 ± 3.15	26.6 ± 3.68	
49	α-bergamotene	1574	18.55	RI, MS					0.85 ± 0.10			
50	nonyl acetate	1580	18.67	MS, Std	0.14 ± 0.02							
51	caryophyllene	1601	19.09	MS, Std	54.1 ± 10.8	0.15 ± 0.03	1.01 ± 0.16	0.40 ± 0.09	22.3 ± 3.85	1.38 ± 0.19	0.72 ± 0.14	3.98 ± 0.26
52	terpinen-4-ol	1604	19.15	MS, Std	2.80 ± 0.16	1.66 ± 0.40	0.33 ± 0.04	1.67 ± 0.25	1.05 ± 0.13	0.40 ± 0.06	0.35 ± 0.06	0.91 ± 0.04
53	ethyl decanoate	1640	19.86	RI, MS			0.16 ± 0.02	0.51 ± 0.08	0.09 ± 0.01			
54	1-hexadecene	1648	20.02	RI, MS	0.28 ± 0.05		0.10 ± 0.01	0.68 ± 0.15	0.25 ± 0.05		0.62 ± 0.11	0.18 ± 0.05
55	nonanol	1659	20.23	MS, Std	0.07 ± 0.01	0.11 ± 0.01	0.10 ± 0.01	0.12 ± 0.02	0.09 ± 0.01	0.10 ± 0.01		0.12 ± 0.01

56	α-humulene	1670	20.45	MS, Std	83.3 ± 14.8		5.35 ± 0.82	14.9 ± 2.96	0.12 ± 0.01		6.02 ± 0.92	11.1 ± 2.14
57	γ-muurolene	1689	20.81	RI, MS	0.11 ± 0.02							
58	α-terpineol	1696	20.96	MS, Std	355 ± 10.8	275 ± 17.4	35.7 ± 2.45	17.9 ± 2.79	125 ± 20.1	219 ± 15.9	18.1 ± 3.00	26.3 ± 1.36
59	dodecanal	1715	21.34	MS, Std	75.1 ± 11.2	±	3.82 ± 0.41	2.63 ± 0.17	13.0 ± 2.55		3.21 ± 0.28	2.26 ± 0.27
60	valencene	1717	21.39	MS, Std	278 ± 51.7	12.0 ± 1.55	311 ± 44.9	0.86 ± 0.16	132 ± 25.0	37.1 ± 5.04	303 ± 22.0	6.45 ± 0.75
61	α-selinene	1722	21.49	RI, MS			0.21 ± 0.04				0.33 ± 0.06	
62	neryl acetate	1724	21.53	MS, Std	110 ± 19.2	25.1 ± 1.26	18.8 ± 3.80	13.3 ± 1.27	213 ± 41.6	25.3 ± 2.14	17.1 ± 2.88	7.93 ± 0.01
63	citral	1730	21.65	MS, Std	235 ± 42.8	141 ± 7.72	104 ± 3.82	104 ± 6.60	102 ± 16.9	97.5 ± 8.39	124 ± 18.0	44.9 ± 0.56
64	carvone	1731	21.66	RI, MS					0.10 ± 0.02			0.12 ± 0.01
65	geranial	1731	21.67	RI, MS		0.23 ± 0.04	0.10 ± 0.01			0.51 ± 0.10		
66	geranyl acetate	1751	22.09	MS, Std	100 ± 17.8	0.27 ± 0.05	11.3 ± 0.72	9.59 ± 0.88	52.8 ± 8.18	0.36 ± 0.09	13.9 ± 2.76	4.12 ± 0.56
67	β-citronellol	1758	22.23	MS, Std	0.10 ± 0.02	0.12 ± 0.02		0.25 ± 0.02	0.06 ± 0.01			0.19 ± 0.60
68	(-)-myrtenol	17.67	22.42	MS, Std	6.27 ± 0.89	0.54 ± 0.05	1.11 ± 0.31	0.69 ± 0.10	4.99 ± 0.80		1.54 ± 0.28	0.57 ± 0.34
69	perillylaldehyde	1777	22.62	RI, MS		0.10 ± 0.01		0.10 ± 0.00		0.10 ± 0.01		

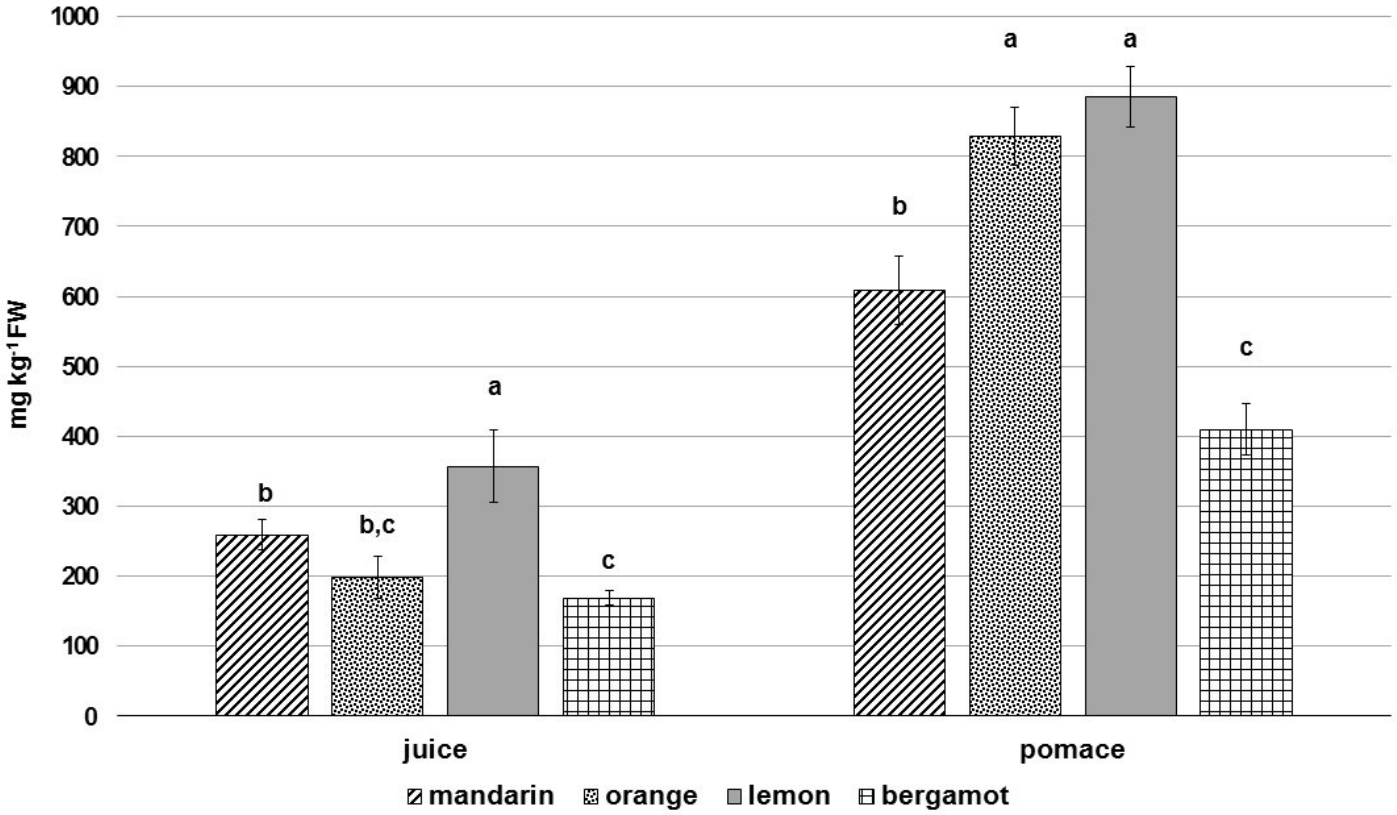
70	nerol	1788	22.85	MS, Std	16.9 ± 3.27	2.54 ± 0.13	0.10 ± 0.01	5.09 ± 0.82	52.7 ± 4.23	1.31 ± 0.14	0.10 ± 0.01	4.09 ± 0.60
71	geraniol	1827	23.68	MS, Std	6.19 ± 1.07	0.37 ± 0.07			3.01 ± 0.40	0.74 ± 0.11		0.15 ± 0.06
72	octadecene	1830	23.75	RI, MS	0.06 ± 0.01	0.10 ± 0.01		0.21 ± 0.04	±			
73	geranyl acetone	1834	23.83	RI, MS					0.10 ± 0.01			
74	limonen-10-yl acetate	1837	23.9	RI, MS	0.28 ± 0.03		1.38 ± 0.17		0.18 ± 0.02			
75	jasmone	1886	24.98	MS, Std					6.57 ± 1.31			
76	β-ionone	1893	25.12	MS, Std	3.99 ± 0.26	1.59 ± 0.30	2.22 ± 0.44	2.58 ± 0.44	65.4 ± 11.3	1.49 ± 0.13	0.62 ± 0.09	2.20 ± 0.24
77	phenethyl acetate		26.12	MS, Std	4.91 ± 0.51	0.69 ± 0.09	0.81 ± 0.11	5.09 ± 0.82	6.33 ± 1.09	3.19 ± 0.38	0.46 ± 0.08	2.08 ± 0.34
78	nerolidol		26.06	MS, Std			0.39 ± 0.03	0.63 ± 0.11		0.54 ± 0.08		1.52 ± 0.11
79	dimethyl anthranilate		27.55	MS, Std				0.14 ± 0.02				

Data are expressed as means ± standard deviation (n =4). Values are expressed as mg kg^{-1} FW.

RI, retention index calculated on the basis of the RT from the alkane mixture C_9-C_{20} on the VF-Wax® column.

RT, retention time; Std, standard; MS, mass spectrum data.

1



2

3 **Figure 1**. Cumulative contents of phenolic compounds in citrus juices and pomaces. Data (mg kg^{-1} FW) are expressed as
4 mean ± SD (n =4). Values with different superscript letters are significantly different ($p < 0.05$).

Fig. 2A

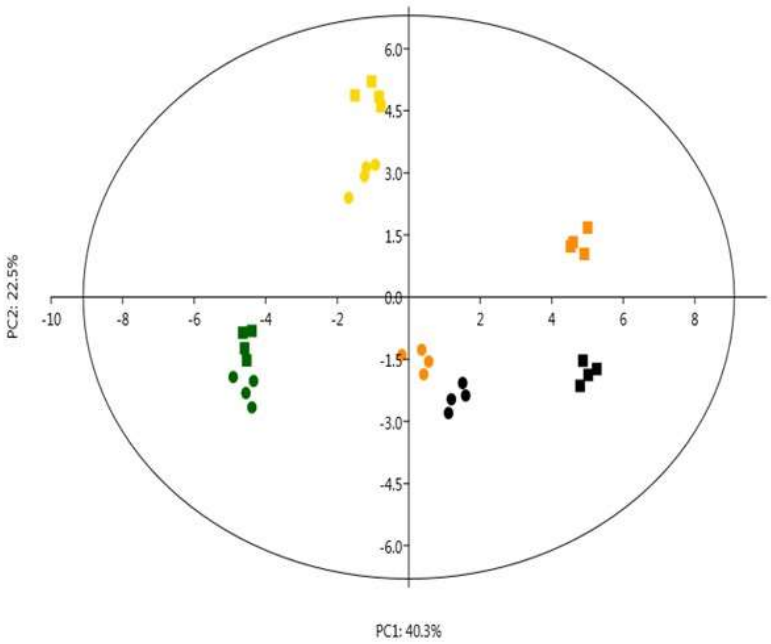


Fig. 2B

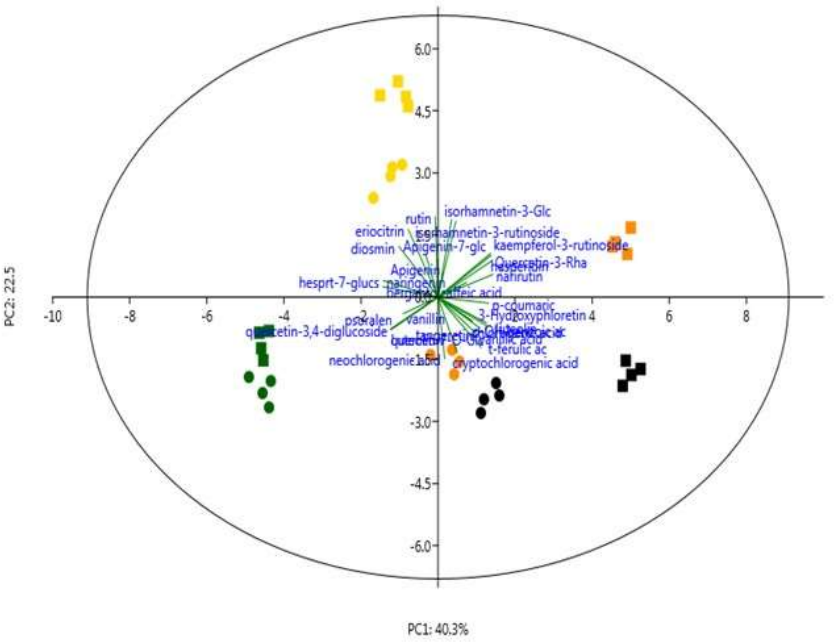


Figure 2. PCA of juices and pomaces in function of PC1 and PC2. (A) Score plot, (B) biplot. The considered phenolic compounds are those listed in Table 1. Symbol legend: (●) = juice and (■) = pomace. Color legend: black (mandarin), yellow (lemon), orange (orange), and green (bergamot).

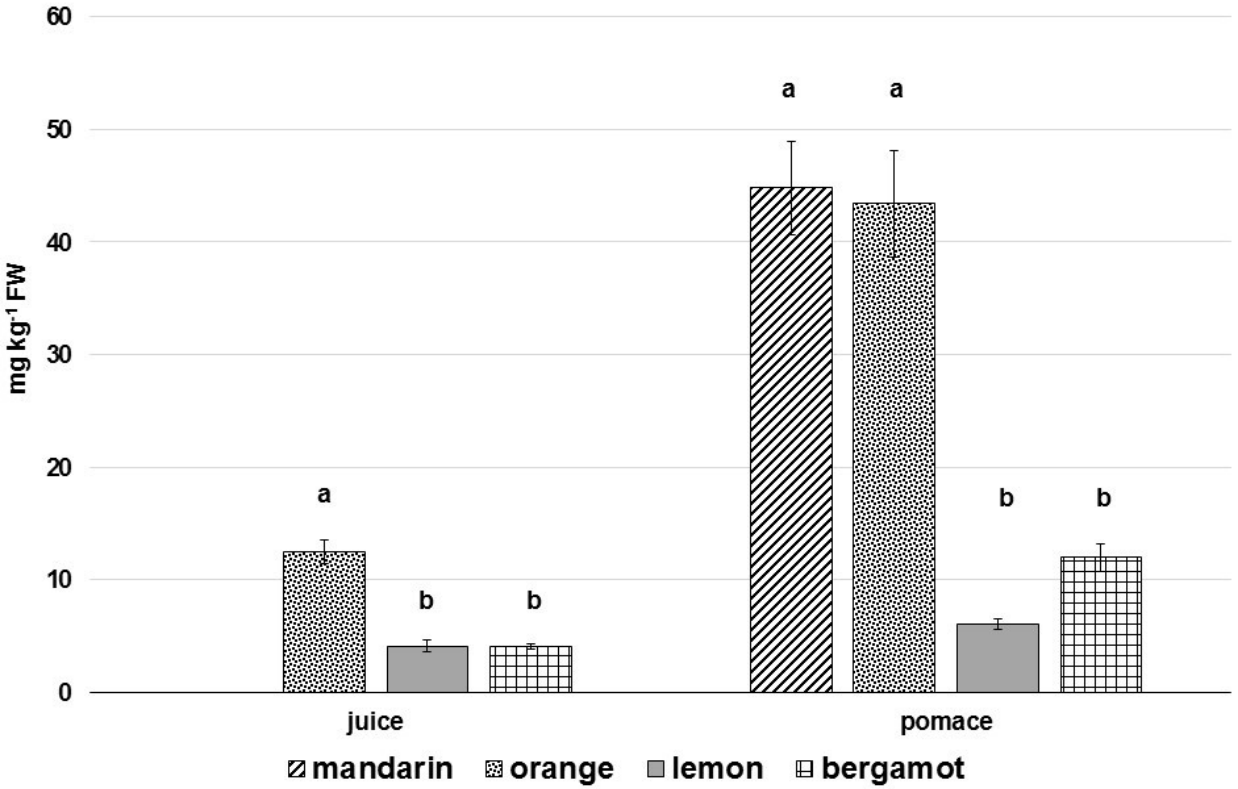


Figure 3. Cumulative contents of carotenoid in citrus juices and pomaces. Data are expressed as mean ± SD (n =4). Values with different superscript letters are significantly different ($p < 0.05$).

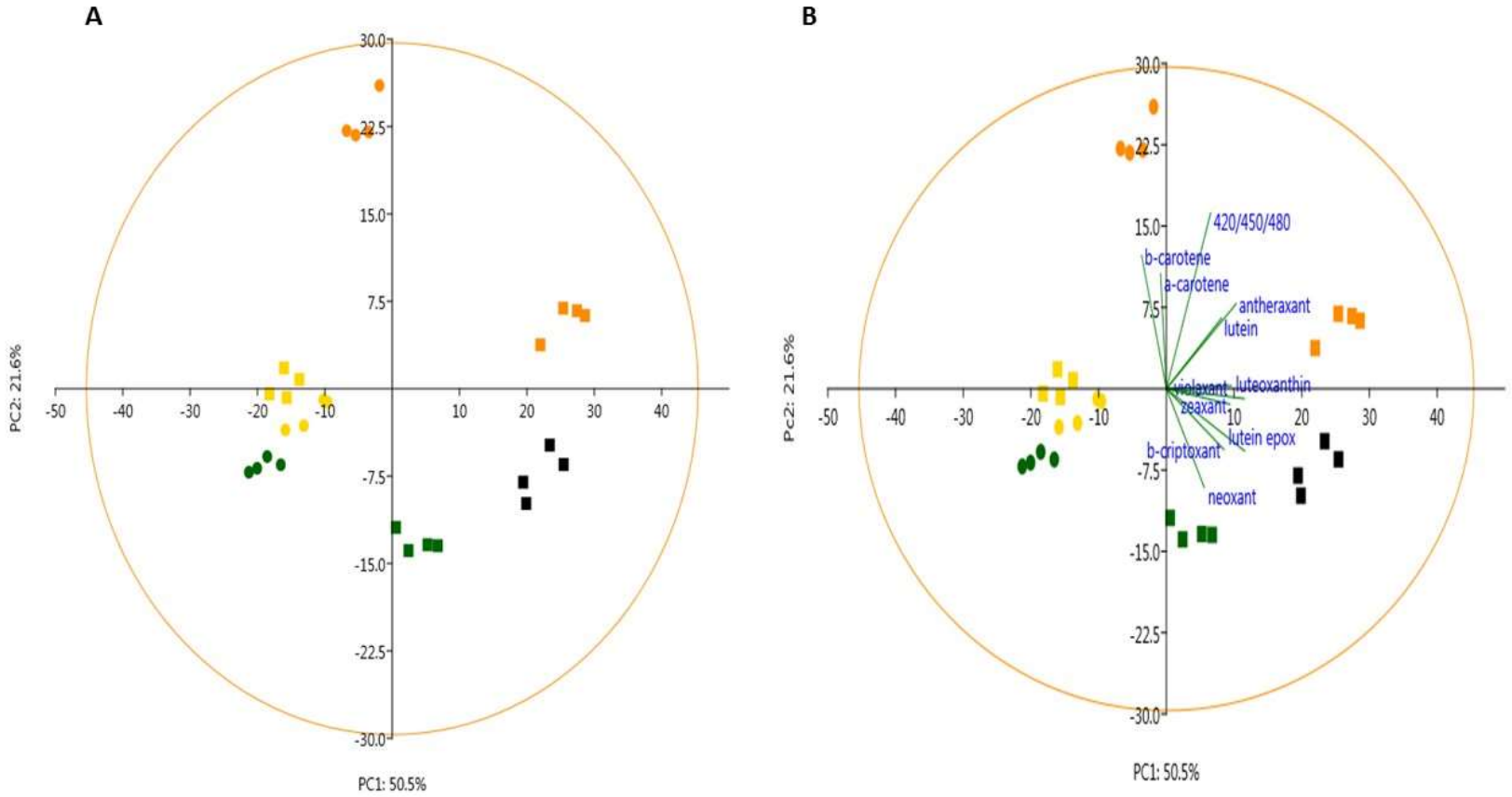


Figure 4. PCA of juices and pomaces in function of PC1 and PC2. (A) Score plot, (B) biplot. The considered carotenoids are those listed in Table 2. Symbol legend: (●) = juice and (■) = pomace. Color legend: black (mandarin), yellow (lemon), orange (orange), and green (bergamot).

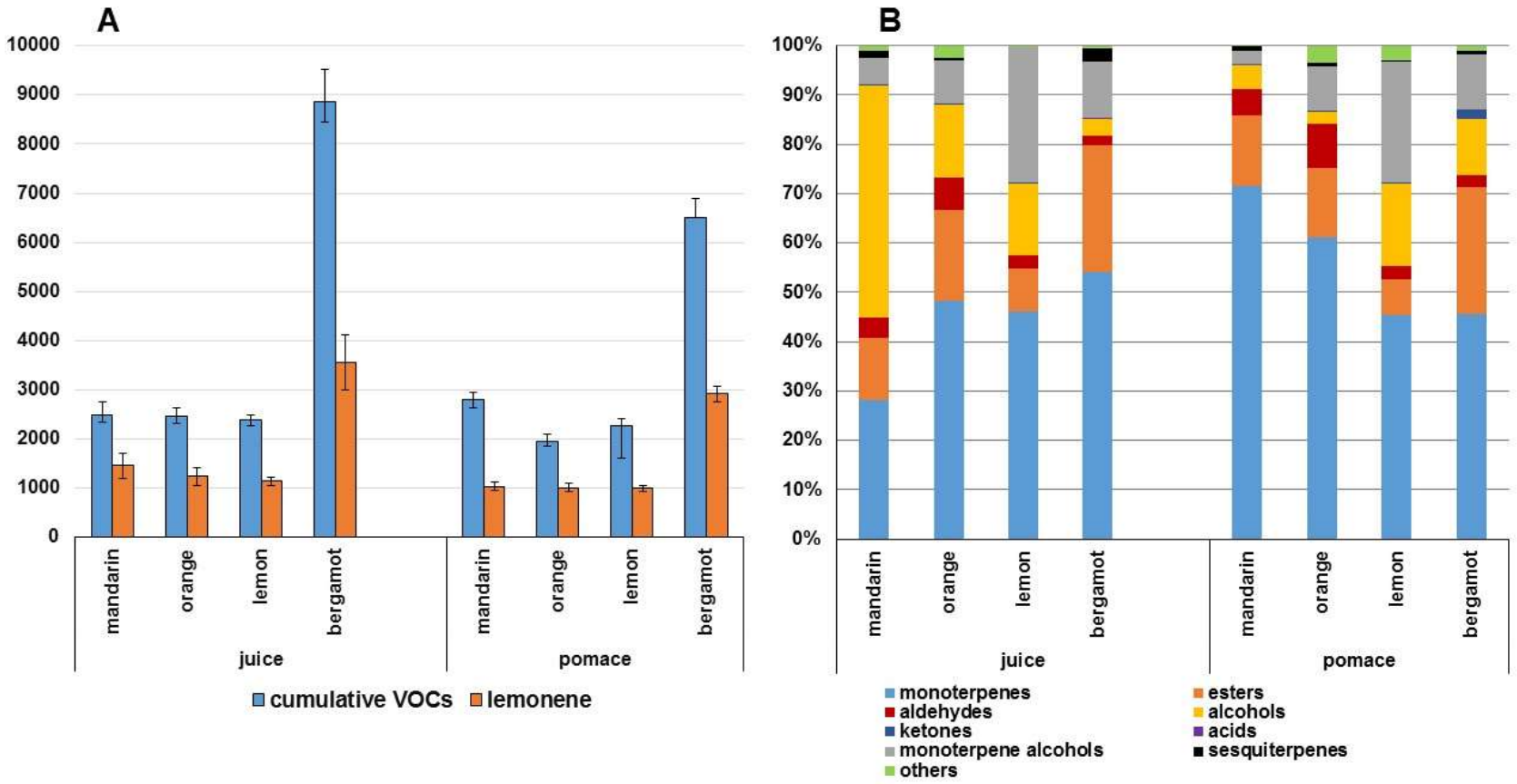


Figure 5A represents the cumulative content of VOCs and limonene of the samples, expressed as mg kg^{-1} FW.

Figure 5B represents the relative proportion (%) of the major chemical classes of VOCs.

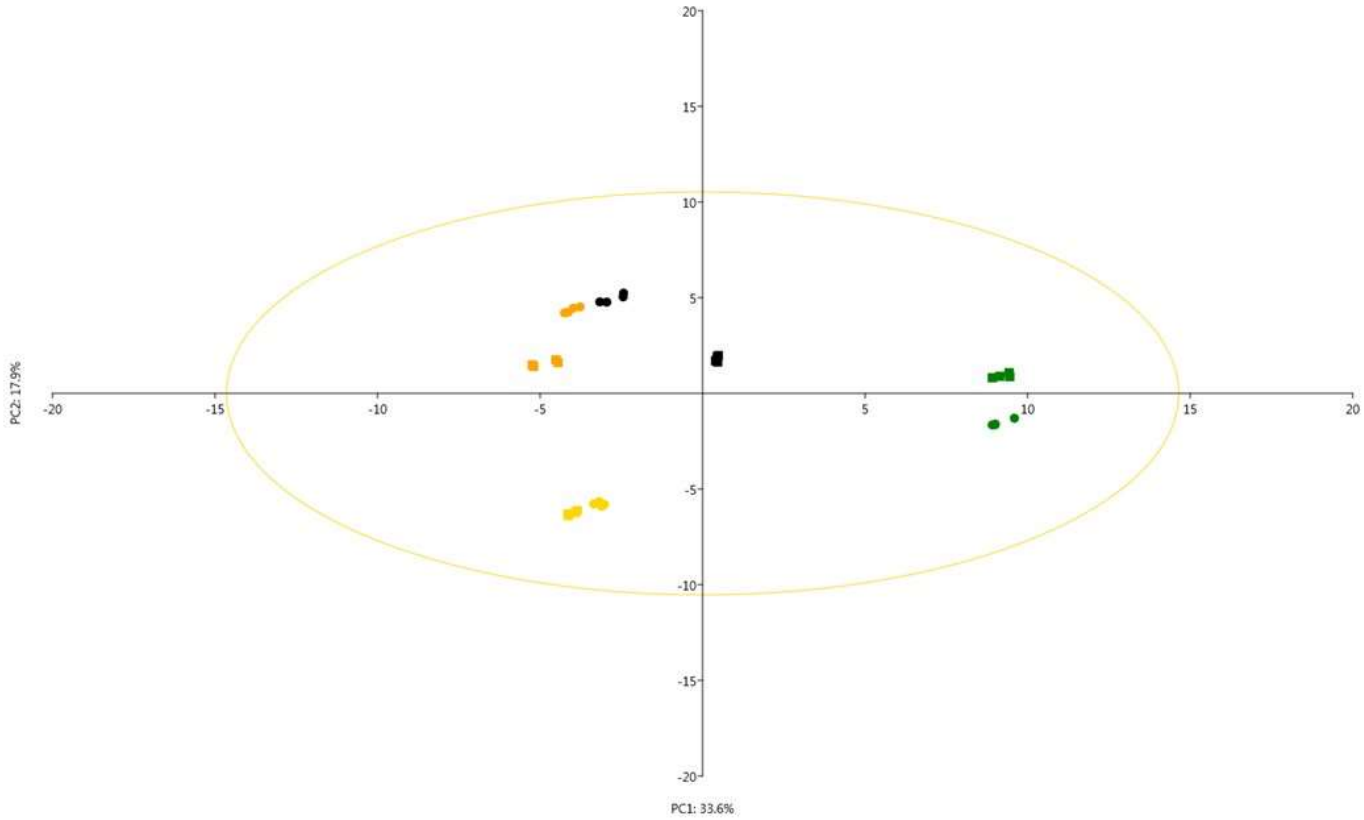


Figure 6. PCA score plot of the juices and pomaces in function of PC1 and PC2. The considered VOCs are those listed in Table 3. Symbol legend: (●) = juice and (■) = pomace. Color legend: black (mandarin), yellow (lemon), orange (orange), and green (bergamot).