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Bergamot (*Citrus bergamia*, Risso): the effects of cultivar and harvest date on functional properties of juice and cloudy.

Angelo Maria Giuffrè

Università degli Studi Mediterranea di Reggio Calabria, AGRARIA - Dipartimento di Agricoltura, Risorse forestali, Ambiente Risorse zootecniche, Ingegneria agraria, Alimenti – Contrada Melissari, 89124 - Reggio Calabria (Italia).

Correspondence: Dr. Angelo M. Giuffrè, Dipartimento di Agraria, Università degli Studi 'Mediterranea' di Reggio Calabria, Contrada Melissari, (89124, Reggio Calabria, Italy). E.mail: amgiuffre@unirc.it.

Phone +39 (0) 965.1694362.

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Abstract

Reggio Calabria province (South Italy) is known for being almost the only area of cultivation of the bergamot fruit, grown principally for its essential oil, but today much studied for the health benefits of its juice. The biometrics and physico-chemical properties of the three (*Citrus bergamia* Risso) existing genotypes namely Castagnaro, Fantastico and Femminello were studied during fruit ripening from October to March. Castagnaro cv had the biggest and heaviest fruit during this harvest period. °Brix (7.9-10.0), pH (2.2-2.8) and Formol number (1.47-2.37 mL NaOH 0.1N/100mL) were shown to be influenced by both the genotype and by the harvest date. Titratable acidity (34.98-59.50 g/L) and Vitamin C (Ascorbic acid) (341-867 g/L) decreased during fruit ripening. The evolution of flavonoids such as neoeriocitrin, naringin, neohesperidin brutieridin and melitidin was studied both in bergamot juice and in the bergamot cloudy which is the aqueous extract of bergamot during fruit processing. Bergamot cloudy contained a higher quantity of flavonoids compared to the juice. This study gives important information regarding the cultivar and the harvest date for producers who want to obtain the highest juice quantity or the highest juice quality from the bergamot fruit.

1. Introduction

Bergamot (*Citrus bergamia*, Risso) is an evergreen tree almost exclusively growing on the Ionian and Tyrrhenian Coast of Reggio Calabria Province (South Italy), in a strip of land between 1 to 12 km wide. It is very important for the region where it is cultivated given its economic benefits [1]. Three varieties (genotypes) are known: Castagnaro, Fantastico and Femminello. In 2017, 1,500 hectares of bergamot trees were cultivated in Reggio Calabria, producing 18,750 tons of fruits [2]. Bergamot is a non-climateric fruit and in the past was picked when the highest essential oil content in the peel was reached. Today the juice has also to be considered. The fruit was commonly cultivated for its essential oil extracted from the peel and which is used in cosmetic, perfumery and in food industry. Very recently the bioconversion of juice and peel into wines and vinegars was positively conducted [3]. More recently there has been an increasing interest in the use of its juice as a beverage also in a blend with other fruit juices. This interest is related to the demand for minimally processed foods and functional foods containing antioxidants and biomolecules whose beneficial effect on human health have been widely studied regarding diabetes, cancers, Alzheimer’s disease, insulin resistance and neuro-disease [4]. There is no universally accepted definition of functional food. The following definition could be applied: 'Natural or processed foods that contain known or unknown biologically-active compounds; which in defined amounts provide a clinically proven and documented health benefit for the prevention, management, or treatment of chronic disease [5]. This merges and updates the definitions stated by the National Academy of Sciences Food and Nutrition Board in the US [6], by the Institute of Food Technologists [7], the American Dietetic Association [8] and what De Felice stated for Nutraceuticals [9]. The aim of this research was to investigate the evolution of biometrics and the anti-oxidative properties during the fruit ripening, the effect of cultivar was also studied.

2. Materials and methods

2.1. Plant Material

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All three cultivars were grown in the same area, in mono-cultivar plots. The bergamot trees were cultivated on level ground and were planted with a distance of 6 m between each row and the trees 5 m apart within each row. All the trees were irrigated and fertilized in the same way. Fruits were collected early in the morning and carefully placed in plastic containers commonly used for citrus fruit picking. Thereafter, fruits were immediately transferred to the laboratory for biometric analyses.

2.2. *Juice extraction*

The bergamot fruit is cultivated for essential oil extraction (from peel) and juice extraction. In the present study both the 'albedo' and the remaining pulp which are commonly known as 'pastazzo' were processed into a hammer mills where they were grinded and homogenised with water to solubilise polyphenols. The obtained mixture undergoes various steps in steel tanks to allow the flavonoids to be extracted as much as possible from the 'pastazzo' and transferred to the liquid phase. A first rough separation is conducted by a press, which divides the pulp destined for the subsequent recovery of the pectins in another production plant, from the liquid phase which is stored in steel tanks for the subsequent processing. The pressed pulp contains a big quantity of pectins, both in the water soluble and non soluble, which makes difficult the separation of pulp from the liquid fraction. For this reason pectinase was used as a pectolytic enzyme at 50-60 °C to facilitate pectins degradation. The juice obtained by this procedure is called 'cloudy'.

2.3. *Chemicals*

Chemicals of both analytical grade and chromatographic grade were purchased from Carlo Erba, Milan, Italy. Pure standards of fatty acid methyl esters and sterols were from Sigma Chemical Co., St Louis, MO, USA.

2.4. *Pulp content*

Pulp content is the solid fraction quantified as a percentage in a bergamot juice after centrifugation for 10 mins at 3500 rpm. Pulp and juice are separated by difference of gravity.

2.5. *Turbidity*

A 12% Bergamot juice in bi-deionised water was prepared and transmittance was read at 578 nm. Turbidity was expressed as percentage ratio between the intensity of the incident light and intensity of light emission from the cuvette.

2.6. *pH*

A Mettler-Toledo instrument was used after calibration pH 7.0 and pH 4.0.

2.7. °Brix

The degree Brix was determined by a Mettler-Toledo refractometer on a drop of bergamot juice sample after zero-set of the instrument by a drop of bi-deionised water.

2.8. Titratable Acidity

A 10 g aliquot of bergamot juice and 150 mL of bi-deionised water were placed in a glass beaker. The mixture was boiled for 10 mins. Thereafter acidity was determined by titration with a 0.1 NaOH aqueous solution up to pH 8.1. Acidity was expressed as g of citric acid monohydrate per litre of juice [10].

2.9. Vitamin C

Vitamin C was quantified by an iodomeric titration. In a glass beaker, 1 mL of bergamot juice and 5 mL of bi-deionised water were mixed and titrated by a 0.01 N iodine solution using a 2% starch solution as an indicator. The result was expressed as mg ascorbic acid / L of juice [10].

2.10. Formol number

In a glass beaker, 10 mL of bergamot juice, 10 mL of 40% by volume formaldehyde solution (pH 2.8) and 7 drops of phenolphthalein (1% in ethanol) were measured. The mixture was stirred and titrated by a 0.1 NaOH solution (IFUMA 30, method EN 1133) [11].

2.11. Flavonoids

The analysis was carried out using the method suggested by Giuffrè et al.[10] and modified using a HPLC-PDA system coupled with a column conditioning system at 25°C. The separation column was a Kinetex 5μ C18 100 Å, 150 mm length, 4.6 mm internal diameter. The system was supported by Chromera software version 3.4.0.5712.

2.12. DPPH and FRAP assays

The analyses were conducted spectrophotometrically as suggested by Panuccio et al and Sicari et al [12-13].

2.13. Statistical analysis

Thirty kilogrammes of bergamot fruits were randomly collected from 20 trees at each harvest date for each cultivar (Castagnaro, Fantastico and Femminello) in the mid of each month from October 2016 to March 2017. For each cultivar, two batches (15 kg each) were prepared at each harvest date and two replicates were obtained from each batch. The same experimental design was replicated in the harvest year 2017-2018. Means and standard deviations were calculated on 8 replicates [4 replicates x two harvest years) by Excel 2010 software. Statistical differences were calculated by one-way ANOVA and Tukey test for post hoc analysis at $p < 0.05$ using the SPSS 17.0 software (SPSS Inc., Chicago, IL, USA); the variables were: the cultivar and the harvest date of bergamots. Principal component analysis (PCA) was carried out using the software XLSTAT version 2009.1.01.

3. Results and discussion

3.1. Biometrics

The vertical diameter length was longest in Castagnaro fruit which showed the most constant increase in length, from 6.63 cm in October to more than 9 cm after December. The vertical diameter length of Fantastico and Femminello fruits showed a slight decrease in March at the end of the ripening period (7.15 and 6.50 cm respectively), (Table 1). The horizontal diameter was greatest in Castagnaro at each monthly sampling and showed a tendency to increase during the ripening of Castagnaro and Fantastico, whereas in Femminello a slightly fluctuating rate was found (Table 1). The pulp in juice content showed very high significant differences ($P<0.001$) for each cultivar during fruit ripening but no significant differences were found between cultivars on October, December and January (Table 1). Vertical diameter increased with horizontal diameter ($r = 0.958$), with fruit weight ($r = 0.87$) and with peel weight ($r = 0.880$), (Table 7). Fruit weight increased very highly significantly ($P<0.001$) during fruit ripening of all cultivars. From October to March, Castagnaro fruit showed both the highest increase in weight during ripening (72%) and the highest weight in each month (245 g on October and 421 g on March). Fantastico fruit increased by 49% during ripening (173 g on October and 258 g on March). Femminello fruits showed both the lowest weight on each harvest date and the lowest increase in weight (23%) from October to February (Table 2). The peel weight was always greatest in Castagnaro, 44.1 g on October, 55.01 g on December and 74.02 g on February but a substantial fall in weight was measured in March in all the three cultivars (Table 2). The percentage of juice was highest in Fantastico at the first stage of ripening (29.33-30.50%) and in Fantastico and Femminello at the end of the ripening period (39.97% and 40.01% respectively), (Table 1). Pulp in juice is a negative parameter because it has to be removed during the industrial process before using or storing the fruit juice. The juice turbidity was very highly significantly different ($P<0.001$) during ripening and the same significance of differences was found between cultivars at each monthly sampling (Table 2). In the correlation matrix, fruit weight had a strong positive correlation with the vertical diameter ($r = 0.875$; $P <$

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0.001; r^2 = 0.766; t = 19.33) and stronger with the horizontal diameter (r = 0.920; P < 0.001; r^2 = 0.846; t = 19.25), (Table 7).The increase in peel weight in the three cultivars was strongly correlated with the vertical diameter (r = 0.880; P < 0.001; r^2 = 0.774; t = 14.76) and with the horizontal diameter (r = 0.830; P < 0.001; r^2 = 0.689; t = 14.62), (Table 7). Fruit weight showed a weak correlation with juice content (r = 0.112; P < 0.001; r^2 = 0.013; t = 17.23) but it was strongly correlated with peel weight (r = 0.815; P < 0.001; r^2 = 0.664; t = 16.56) this means that the increase in weight during fruit ripening is mainly due to the increase in peel and not that of juice.

3.2. pH

Bergamot juice is very acidic and mainly contains ascorbic and citric acid which contribute significantly to the composition of this parameter. Between cultivars, no significant differences were found in November, February and March. A very high significant pH increase (P<0.001) was found in Castagnaro and Femminello fruit juices and high significant differences were found in Fantastico juice (P<0.01) (Table 3). pH of juice was negatively and moderately correlated with Vitamin C (r = 0.643; P < 0.001; r^2 = 0.413; t = 31.32) but strongly and negatively correlated with titratable acidity (r = 0.740; P < 0.001; r^2 = 0.548; t = 50.76), (Table 8). The pH of the bergamot juice was lower than Grapefruit juice (3.05), Orange juice (3.63) and Tangerine juice (3.41) but similar or higher than Lemon juice (2.43) [14].

3.3. °Brix

The degree Brix is the sugar content expressed as g/100 g juice. It is directly proportional to the sweetness of the fruit and therefore to its organoleptic pleasantness. This value did not exceed 10, which was reached by Fantastico cv in November (Table 3). The analysis of variance showed very highly significant differences during ripening (P < 0.001) in all the cultivars. If the cultivar effect is considered, very high significant differences in November, January and March (P<0.001) were found, high significant differences in October and December (P < 0.01) and significant differences (P < 0.05) in February (Table 3). The Brix (%)/Titratable acidity (%) is a maturity index and the highest value for all the three cultivars was found in the last month of sampling, with a tendency to increase during ripening (Table 3). The °Brix (%)/Titratable Acidity (%) ratio had a strong negative correlation with total flavonoids in juice (r = 0.920; P < 0.001; r^2 = 0.846; t = 16.61), (Table 8). Other Authors, in other citrus juices found a different °Brix: 5.10 (grapefruit), 1.16 (lemon), 4.53 (orange), 6.50 Tangerine[14], and 11.0 °Brix in squeezed blood orange juice cultivated in Calabria [15].

3.4. *Titratable Acidity*

The titratable acidity is an important parameter to determine the maturity of the fruit and the acidic taste in citrus fruit. The degree of maturity of a fruit is one of the most important factors to determine conservation methods and control quality parameters such as taste and flavour. An immature fruit usually has a low sugar content in relation to acidity, compared to the ripe fruit that has a high level of sugar in relation to acidity. In bergamot juice a very high significant difference in titratable acidity between cultivars ($P < 0.001$) was observed, from a low 53.86 g/L in Castagnaro to a high 58.67 g/L in Fantastico measured at the earliest sampling event (Table 3). During fruit ripening a decreasing trend in titratable acidity in all cultivars was observed. At the last sampling event in Castagnaro the lowest content (34.98 g/L) was seen, namely a decrease of 35.05% from October to March, whereas the highest value was not found in Fantastico (as at the earliest sampling event) but in Femminello (41.90 g/L) with a decrease rate of 22.81%.

3.5. *Vitamin C*

The human body cannot synthesize vitamins, therefore they have to be part of our diet. Vitamin C (ascorbic acid) is water soluble, has an antioxidant potential [16], prevents scurvy [17] and degenerative diseases, particularly those that are ageing-related [18] and has possible protective effect on the bones of older adults. Vitamin C can be oxidised by storage at room temperature, the addition of baking soda, overcooking and contact with copper contact and the over intakes of zinc (cooking tools), alcohol and pectin [19]. In the studied samples, vitamin C decreased dramatically during fruit ripening: Castagnaro (59%), Fantastico (47%) and Femminello (48%) from October to March. In October a very high significant difference in vitamin C between cultivars ($P<0.001$) was found: 831 mg/L in Castagnaro, 867 mg/L in Fantastico and 669 mg/L in Femminello (Table 3). Vitamin C content was not influenced by cultivars in February but very highly affected by this variable ($P<0.001$) in all other months (Table 3). Findings of other Authors on vitamin C content in citrus fruit juices have revealed 680 mg/L and 455 mg/L respectively in Marsh and Star Ruby i.e. two grapefruit cultivars [20], 680 mg/L in blood orange [10], 220 mg/L in pomelo [21] and 355 mg/L in lemon analysed by HPLC [14].

3.6. *Formol Number*

The Formol number can represent, in a normal chemical industrial control, a useful index for the global quantitative evaluation of amino acids present in fruit juices. The Formol number is not influenced by the presence of many natural constituents of fruit juice (sugars, vitamins, flavourings, colourings) and it is applied in the quality determination of fruit juice and beverages because it expresses the total number of amino acids found. In bergamot juice the cultivar did not influence the Formol number in October but very high significant differences (P<0.001) were found between cultivars from November to March. The harvest date had significant influence (P<0.001) on the Formol Number for the three cultivars (Table 3). The Formol number varied from 23.7 mL NaOH 0.1N/100 mL (Femminello in March) to 14.3 mL NaOH 0.1N/100 mL (Castagnaro in December and Femminello in February) and exceeded 20 mL NaOH 0.1N/100 mL at the same time in all the three cultivars only in October. No correlation was found between Formol number and melitidin in juice (r = 0).

3.7. Flavonoids

Flavonoids are polyphenols with an antioxidant and radical scavenging role, and are described by the scientific literature to have many beneficial effects on human health. They are biomolecules that prevent from the risk of primary open-angle glaucoma [21], have an antiplatelet effect [22], maintain the anti-inflammatory action of cortisol under pro-oxidant conditions [23], protect vascular endothelial function [24], have an anti-obesity activity [25], reduce the risk of cardiovascular disease [26] and have antimicrobial [27] antiviral [28] and anti-inflammatory effects [29]. Neoerocitrin was significantly high in Femminello and low in Castagnaro and Fantastico (P<0.001) at the earliest sampling event. This compound showed a tendency increase in bergamot juice of Castagnaro and Fantastico as fruits ripened from October to February, with a fall in March (Table 4). Naringin, neohesperidin and brutieridin were the major flavonoids in the bergamot juice (Table 4) whereas neoeriocitrin, naringin and neohesperidin prevailed in bergamot cloudy (Table 5) this was probably due to a higher water solubility. Naringin was very highly significantly influenced (P<0.001) by both cultivar and harvest date variables (Tables 4-5). Neoeriocitrin in both bergamot juice and bergamot cloudy was highest in the last fruit sample dates (February and March) for all the three cultivars (Tables 4-5). In the bergamot juice, naringin content was highest on the last sample date (42.61%, 28.63% and 42.30% of the total flavonoids, respectively for Castagnaro, Fantastico and Femminello). Neohesperidin in bergamot juice was significantly different at each sample date (P<0.001) with January being the month in which the highest quantity was measured. Almost always and in both the juice and the cloudy of the three cvs of bergamot, the neohesperidin content was highest in Fantastico (Tables 4-5). Brutieridin and melitidin are two molecules identified and described in the bergamot juice by Di Donna et al. [30-33] and Fiorillo et al [34]. The name brutieridin comes from the ancient name of one of the Calabrian cities (Brutium, today Cosenza) where brutieridin and melitidin were studied, whereas melitidin derives from the name of one of the most important towns (Melito Porto Salvo) where the bergamot tree is cultivated. In the samples studied in our work, brutieridin was always greater than melitidin both in bergamot juice

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and in bergamot cloudy (Tables 4-5). On the first sample dates (October-December), melitidin was higher in Castagnaro and Femminello juice than in Fantastico, the same situation was found on the last sample date (Table 4). Melitidin in cloudy was significantly lower in March when its content was lower than 3% in all the three cultivars, whereas it was double or almost double in the early period of ripening from October to December (Table 5). A significant decreasing tendency in the brutieridin content of bergamot juice was recorded in Castagnaro and Fantastico ($P<0.001$) whereas a fluctuating rate was found, during fruit ripening, in Femminello juice and in the cloudy of the three cultivars. Brutieridin detected in both bergamot juice and bergamot cloudy was almost always greatest in Fantastico cv from October to March, except in February when the highest Brutieridin content was found in Femminello (26.94%) for Bergamot juice and in Castagnaro (14.33%) for bergamot cloudy. Harvest date and cultivar variables showed very high significant differences ($P<0.001$) between means (Tables 4-5). The correlation between each single flavonoid in juice and its homologous in cloudy was between $r = 0.217$ of brutieridin and $r = 0.582$ of melitidin (Table 8). In the bergamot juice the total flavonoid content constantly increased with fruit ripening in Fantastico from 361 mg/L in October to 678 mg/L in March (namely an increase rate of 87.81%) and in Femminello from 287 mg/L in October to 824 mg/L in March (namely an increase of 187.11%). Also in Castagnaro juice the flavonoid content was higher in the last period of fruit ripening compared to October and November (Table 4). Studies on flavonoid content in other citrus juice during storage at 4°C showed a decreasing trend[35], which indicates fruit should be picked later, and juice should be consumed as soon possible after picking.

3.8. DPPH assay and FRAP assay

A citrus juice contains more than one class of antioxidants which have different behaviours. For this reason many authors suggest applying more than one assay to evaluate antioxidant activity. In the present study we applied DPPH assay and FRAP assay which are two of the most common tests used on many matrices such as common orange [13] blood orange juice [10], edible vegetable oils and potential industrial vegetable oils [36-38], apples, bananas, strawberries, kiwifruit, cauliflower [39]. Vitamin C and flavonoids are the most important antioxidants in bergamot juice and show an inverse ratio during fruit ripening: the vitamin C showed a decreasing trend (Table 3) in opposition to total flavonoid content which increased with harvest date (Table 4). In all the three cultivars DPPH value showed a very high significant difference at each month of sampling (Table 6). The correlation between antioxidant activity of the bergamot juice measured with the DPPH assay was high with total flavonoid content ($r = 0.764$; $P < 0.001$; $r^2 = 0.583$; $t = 11.87$). This was in accordance with results of Roussos [40] which found a strong positive correlation between DPPH and flavonoids in blood orange juice. FRAP assay had an almost strong positive correlation with the titratable acidity by ($r = 0.673$; $P < 0.001$; $r^2 = 0.453$; $t = 29.45$) and with Brutieridin in juice by ($r = 0.699$; $P < 0.001$; $r^2 = 0.488$; $t = 9.73$), and a moderate positive correlation with °Brix ($r = 0.441$; $P < 0.001$; $r^2 = 0.194$; $t = 41.07$) and neohesperidin in cloudy ($r = 0.385$; $P < 0.001$; $r^2 = 0.148$; $t = 8.97$). FRAP assay was also moderately correlated with melitidin in both juice ($r = 0.402$; $P < 0.001$; $r^2 = 0.161$; $t = 35.96$) and cloudy ($r = 0.406$; $P < 0.001$; r2 = 0.165; $t = 44.75$) showing a very similar Pearson coefficient (Table 8). Lastly, vitamin C was found to be responsible of the antioxidant activity measured by FRAP assay ($r = 0.962$; $P < 0.001$; $r^2 = 0.926$; $t = 29.45$) (Table 8) similar to findings of other authors on citrus juices [41-43] but in partial disagreement with other results [44].

3.9. Hierarchical Cluster Analysis and Principal Component Analysis

The three cultivars were found to cluster into two clades (Figure 1). Clade 1 contained Fantastico and Femminello which showed a high similarity and were clustered at a distance of 1. The second cluster contained the Castagnaro cv alone, with the highest fruit and peel weights, in particular Castagnaro showed a peel weight double or more than double than Femminello. In Castagnaro the highest vertical and horizontal diameters were also found, the lowest Flavonoids content in juice and cloudy, the lowest titratable acidity. Principal Component Analysis (PCA) was performed on the three cultivars and 30 parameters were included in the test. (Figure 2). Two Eigen values were obtained and together accounted for 100% of the cumulative variability. The Eigen values and the percentage of total variance were 19.7481 (65.8%) and 10.2519 (34.2%). The visualization of the discrimination between the different orange cultivars on the plane of the first two functions led to a distinct separation. The cultivars were split between three sides of the plane which demonstrate the significant difference among the cultivars. The graphic also shows how the parameters are linked or separated from the cultivar factor. The Castagnaro cultivar located in the right corner of the plane, is linked to the neoeriocitrin cloudy, fruit weight/juice content ratio. The Fantastico cultivar located in the lower center of the plane, is more influenced by the brutieridin in juice and in cloudy. Finally, the Femminello cultivar, which is located in the left corner of the plane, is influenced by the °Brix % / titratable acidity ratio, turbidity, juice content/peel weight ratio. Some parameters were showing independency from the cultivar factor because of the their location in the plane, such us the naringin in cloudy, the naringin in juice, °Brix %, FRAP value, DPPH value, titratable acidity, neohesperidin, total flavonoids in juice and in cloudy. Also, some parameters are correlated negatively, such as neohesperidin and naringin, as these parameters are located in opposite directions in the plane.

4. Conclusions

Bergamot is a tree and fruit with a very strong geographical connotation, growing almost exclusively in Reggio Calabria province (South Italy). Three cultivars of this Citrus genus are known (Castagnaro, Fantastico and Femminello) and this study has shown a strong effect of both the cultivar (genotype) and the harvest date. These variables were found to influence the biometrics and the physico-chemical properties of fruits and fruit juice. Castagnaro is the cultivar producing the heaviest fruit, with the highest vertical and horizontal diameter and with the highest peel content during fruit ripening from October to March. Vitamin C content decreases during bergamot fruit ripening and it is close to the mean or in a higher quantity compared to other citrus fruit juice. The findings of this study have shown that bergamot fruit is a very good source of flavonoids which can be directly used in food and beverage preparation when obtained from fruit juice and pulp, or which can be extracted from cloudy for food, beverages and pharmaceutical purposes. Naringin, and neohesperidin were two flavonoids predominating in both the bergamot juice and in the bergamot cloudy with brutieridin as one of the two most represented flavonoid in bergamot juice and naringin as the most represented flavonoid in the bergamot cloudy. Brutieridin and melitidin are two flavonoids characterising the bergamot juice.

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Table 1. Biometrics of bergamot fruit. Results are presented as the mean value ± standard deviation, n=8, (2016-2017 and 2017-2018 harvest years). ***significance at $P < 0.001$; ** significance at $P < 0.01$; * significance at $P < 0.05$. Means in the same line are distinguished by small letters. Means in the same column are distinguished by capital letters .

	Cultivar	October	November	December	January	February	March	*Sign.*
Vertical Diameter (cm)	Castagnaro	8.63±0.25 cA	8.67±0.10 bcA	9.02±0.10 abA	9.07±0.06 aA	9.04±0.09 aA	9.0±0.10 abA	**
	Fantastico	7.33±0.06 cB	7.3±0.10 cB	8.12±0.10 aB	8.11±0.09 aB	7.65±0.09 bB	7.15±0.09 cB	***
	Femminello	6.90±0.1 aC	6.53±0.25 abC	6.47±0.15 bC	6.83±0.06 abC	6.83±0.06 abC	6.5±0.17 abC	**
	Sign.	***	***	***	***	***	***	
Horizontal Diameter (cm)	Castagnaro	8.8±0.10 bA	8.7±0.20 bA	9.77±0.15 aA	9.60±0.10 aA	9.43±0.03 aA	9.77±0.12 aA	***
	Fantastico	7.13±0.06 cB	7.2±0.10 cB	8.40±0.05 aB	8.36±0.06 aB	7.98±0.08 bB	8.04±0.06 bB	*
	Femminello	7.07±0.25 aB	6.53±0.21 bC	6.67±0.12 a bC	7.1±0.10 aC	7.03±0.25 abC	6.77±0.15 abC	*
	Sign.	**	***	***	***	***	***	
Pulp in juice (%)	Castagnaro	10.17±0.29 bA	10.00±0.50 bB	7.00±0.50 cA	9.83±0.29 bA	10.33±0.58 bA	11.67±0.58 aAB	***
	Fantastico	10.33±0.58 aA	10.33±0.58 aA	7.33±0.58 bA	10.33±0.58 aA	10.33±0.58 aA	10.00±1.0 aB	***
	Femminello	10.00±0.0 bA	9.00±0.0 cB	8.00±0 dA	10.00±0.0 bA	7.03±0.06 eB	12.17±0.29 aA	***
	Sign.	n.s.	*	n.s.	n.s.	***	**	

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Table 2. Biometrics of bergamot fruit. Results are presented as the mean value ± standard deviation, n=8, (2016-2017 and 2017-2018 harvest years). ***significance at $P < 0.001$; ** significance at $P < 0.01$; * significance at $P < 0.05$. Means in the same line are distinguished by small letters. Means in the same column are distinguished by capital letters .

	Cultivar	October	November	December	January	February	March	*Sign.*
Fruit weight (g)	Castagnaro	245±6 eA	277±8 dA	354±14 cA	363±5 cA	397±5 bA	421±7 aA	***
	Fantastico	173±3 dB	201±2 cB	241±4 bB	194±5 cB	266±3 aB	258±12 aB	***
	Femminello	150±2 cC	174±1 bC	131±2 dC	170±2 bC	185±4 aC	146±7 cC	***
	Sign.	***	***	***	***	***	***	
Peel weight (g)	Castagnaro	44.01±0.26 cdA	49.77±0.15 bcA	55.01±0.25 bA	55.03±0.67 bA	74.02±5.29 aA	41.97±0.65 dA	***
	Fantastico	31.43±0.21 dB	34.81±0.08 cB	41.13±0.86 bB	42.23±0.67 bB	51.58±0.10 aB	26.53±0.12 eB	***
	Femminello	25.83±0.12 bC	22.8±0.17 cC	20.97±0.06 dC	20.87±0.15 dC	26.90±0.10 aC	19.53±0.50 eC	***
	Sign.	***	***	***	***	***	***	
Juice Content (%)	Castagnaro	19.73±0.68 fC	21.73±0.21 eB	28.77±0.16 dC	40.08±0.16 aA	36.07±0.49 bB	34.73±0.17 cB	***
	Fantastico	29.33±0.25 eA	30.50±0.36 dA	31.93±0.23 cB	33.57±0.21 bB	30.12±0.03 dC	39.97±0.42 aA	***
	Femminello	21.00±0.10 eB	22.13±0.15 dB	39.03±0.42 bA	33.73±0.21 cB	39.07±0.12 bA	40.01±0.46 aA	***
	Sign.	***	***	***	**	***	***	
Fruit weight / Peel weight	Castagnaro	5.58±0.13 cAB	5.57±0.18 cB	6.44±0.22 bA	6.60±0.07 bA	5.38±0.32 cB	10.04±0.26 aB	***
	Fantastico	5.50±0.09 aB	5.78±0.06 aB	5.85±0.13 aB	4.59±0.14 cC	5.16±0.05 bB	9.73±0.51 aA	***
	Femminello	5.81±0.05 dA	7.64±0.07 dA	6.26±0.08 cA	8.16±0.08 bB	6.87±0.17 cdA	7.49±0.32 aA	***
	Sign.	*	***	**	***	***	***	
Fruit weight / Juice content	Castagnaro	12.44±0.17 bcA	12.76±0.29 bA	12.32±0.52 bA	9.07±0.14 dA	11.01±0.25 cdA	12.13±0.15 aA	***
	Fantastico	5.90±0.14 dC	6.60±0.15 cC	7.54±0.15 bB	5.78±0.18 dB	8.83±0.10 aB	6.46±0.31 cB	***
	Femminello	7.14±0.10 bB	7.87±0.04 cB	3.37±0.09 dC	5.05±0.10 dC	4.73±0.12 eC	3.66±0.18 aC	
	Sign.	***	***	***	***	***	***	
Juice content / Peel weight	Castagnaro	0.45±0.02 eC	0.44±0.01 bC	0.52±0.00 dC	0.73±0.01 aC	0.49±0.04 cC	0.83±0.02 bC	***
	Fantastico	0.93±0.01 bA	0.88±0.01 aB	0.78±0.01 eB	0.79±0.01 cB	0.58±0.00 dB	1.51±0.02 eB	***
	Femminello	0.81±0.01 fB	0.97±0.00 eA	1.86±0.02 bA	1.62±0.02 cA	1.45±0.00 dA	2.05±0.03 aA	***
	Sign.	***	***	***	***	***	***	

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Table 3. Physico-chemical properties of bergamot juice. Results are presented as the mean value ± standard deviation, n=8, (2016-2017 and 2017-2018 harvest years). ***significance at $P < 0.001$; ** significance at $P < 0.01$; * significance at $P < 0.05$. Means in the same line are distinguished by small letters. Means in the same column are distinguished by capital letters .

	Cultivar	October	November	December	January	February	March	*Sign.*
Turbidity	Castagnaro	33.67±0.25 eB	36.55±0.13 cA	34.48±0.28 dB	48.78±0.30 aB	36.37±0.15 cB	39.50±0.17 bB	***
	Fantastico	34.20±0.10 eB	32.28±0.16 fC	36.14±0.09 cA	49.12±0.08 aB	35.27±0.06 dC	39.23±0.21 bB	***
(%)	Femminello	35.27±0.31 dA	35.33±0.23 dB	35.7±0.46 dA	54.03±0.45 aA	38.50±0.26 cA	40.03±0.15 bA	***
	Sign.	**	n.s.	*	*	n.s.	n.s.	
pH	Castagnaro	2.4±0.06 cA	2.4±0.06 cA	2.5±0.06 bcA	2.7±0.06 abA	2.7±0.06 abA	2.8±0.06 aA	***
	Fantastico	2.4±0.0 bA	2.4±0.06 bA	2.5±0.10 abAB	2.5±0.10 abAB	2.6±0.12 abA	2.7±0.10 aA	**
	Femminello	2.2±0.12 dB	2.3±0.06 dA	2.3±0.06 dA	2.4±0.06 bcB	2.6±0.0 abA	2.7±0.0 aA	***
	Sign.	**	n.s.	*	*	n.s.	n.s.	
°Brix	Castagnaro	9.4±0.06 aA	9.5±0.06 aB	8.3±0.06 dB	8.7±0.10 bB	8.5±0.06 cA	8.3±0.06 cdA	***
	Fantastico	9.1±0.06 bB	10.0±0.10 aA	8.7±0.10 cA	9.1±0.10 bA	8.2±0.20 dAB	7.9±0.60 eB	***
	Femminello	9.1±0.12 aB	8.6±0.06 bC	8.6±0.12 bA	9.3±0.06 aA	8.0±0.06 cB	8.2±0.06 cA	***
	Sign.	**	***	**	***	*	***	
Titratable Acidity	Castagnaro	53.86±0.29 aB	51.77±0.06 bB	49.74±0.26 cA	42.2±0.26 dC	40.67±1.42 dC	34.98±0.2 eC	***
(g/L)	Fantastico	58.67±0.06 bA	59.50±0.10 aA	47.58±0.06 cB	46.31±0.06 dB	46.23±0.13 dB	39.83±0.15 eB	***
	Femminello	54.28±0.32 aB	49.87±0.85 bC	46.63±0.81 cB	55.37±0.32 aA	49.0±0.36 bA	41.90±0.25 dA	***
	Sign.	***	***	***	***	***	***	
Brix (%)/TA (%)	Castagnaro	1.75±0.02 bcB	1.84±0.01 bcB	1.66±0.01 bAB	2.06±0.02 bB	2.08±0.07 cB	2.38±0.01 aA	***
	Fantastico	1.56±0.01 bcB	1.68±0.02 bB	1.83±0.02 bB	1.97±0.02 dC	1.77±0.05 cdB	1.97±0.02 aA	***
(Maturity Index)	Femminello	1.68±0.03 bA	1.72±0.02 aA	1.84±0.01 bA	1.67±0.00 aA	1.64±0.00 bA	1.97±0.02 bB	***
	Sign.	**	***	*	***	***	***	
Formol Number	Castagnaro	2.07±0.06 aA	1.87±0.06 bB	1.43±0.06 cC	2.1±0.10 aA	2.13±0.06 aA	2.03±0.06 abB	***
	Fantastico	2.07±0.06 aA	2.10±0.10 aA	1.97±0.12 aA	1.90±0.10 aA	1.53±0.06 bB	1.60±0.10 bC	***
(mL NaOH	Femminello	2.03±0.15 bA	1.47±0.06 cdC	1.67±0.06 cB	1.47±0.06 cdB	1.43±0.06 dB	2.37±0.06 aA	***
	Sign.	n.s.	***	***	***	***	***	
Vitamin C	Castagnaro	831±7 aB	593±7 cB	672±13 bA	474±12 dC	498±6 dA	341±4 eB	***
	Fantastico	867±6 aA	582±7 bB	566±3 bB	571±9 bA	504±4 cA	457±5 dA	***
(mg/L)	Femminello	669±4 aC	635±7 bA	556±13 cB	543±6 cB	492±4 dA	349±9 eB	***
	Sign.	***	***	***	***	n.s.	***	

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Table 4. Main Flavonoids in bergamot juice. Results are presented as the mean value ± standard deviation, n=8, (2016-2017 and 2017-2018 harvest years). ***significance at $P < 0.001$; ** significance at $P < 0.01$; * significance at $P < 0.05$. Means in the same line are distinguished by small letters. Means in the same column are distinguished by capital letters.

	Cultivar	October	November	December	January	February	March	*Sign.*
Neoeriocitrin	Castagnaro	10.16±0.08 eC	13.85±0.11 cB	10.14±0.05 eC	12.64±0.17 dA	20.58±0.19 aB	17.95±0.12 bA	***
	Fantastico	12.84±0.07 cB	12.35±0.05 dC	12.82±0.08 cB	11.72±0.09 eB	22.75±0.05 aA	16.67±0.06 bC	***
(%)	Femminello	15.26±0.19 cA	15.98±0.29 bA	15.35±0.11 cA	10.30±0.05 dC	15.23±0.03 cC	17.41±0.15 aB	***
	Sign.	***	***	***	***	***	***	
Naringin	Castagnaro	29.13±0.06 dB	28.62±0.26 dA	29.01±0.15 dB	39.52±0.47 bA	33.36±0.18 cA	42.61±0.13 aA	***
	Fantastico	25.15±0.08 dC	26.35±0.05 cB	26.11±0.06 cC	28.42±0.09 aC	27.21±0.16 bB	28.63±0.12 aB	***
(%)	Femminello	30.69±0.19 cA	23.92±0.20 eC	30.8±0.05 cA	37.18±0.03 bB	26.65±0.03 dC	42.3±0.26 aA	***
	Sign.	***	***	***	***	***	***	
Neohesperidin	Castagnaro	17.87±0.06 bC	16.75±0.20 cC	17.75±0.13 bB	33.11±0.10 aA	13.82±0.12 dC	17.92±0.07 bC	***
	Fantastico	21.05±0.23 bA	20.51±0.12 cA	20.06±0.23 cA	29.16±0.08 aC	18.51±0.11 dB	29.34±0.24 aA	***
(%)	Femminello	18.35±0.18 eB	19.51±0.17 cB	18.01±0.10 eB	32.62±0.15 aB	22.91±0.02 bA	18.78±0.03 dB	***
	Sign.	***	***	***	***	***	***	
Melitidin	Castagnaro	11.65±0.29 bA	13.32±0.21 aA	11.51±0.13 bA	5.21±0.08 dC	13.68±0.11 aA	7.13±0.11 cA	***
	Fantastico	8.13±0.09 cB	8.36±0.08 bC	8.19±0.10 b cC	7.23±0.06 dA	9.23±0.07 aB	3.30±0.10 eB	***
(%)	Femminello	10.56±0.06 aB	10.43±0.15 aB	10.51±0.03 aB	6.91±0.03 dB	8.27±0.03 bC	7.22±0.08 cA	***
	Sign.	***	***	***	***	***	***	
Brutieridin	Castagnaro	31.49±0.05 aB	27.46±0.12 bC	31.47±0.15 aB	9.52±0.08 eC	18.56±0.07 cC	14.39±0.16 dB	***
	Fantastico	32.83±0.07 aA	32.43±0.16 bA	32.82±0.08 aA	23.47±0.16 cA	22.3±0.13 dB	22.06±0.07 dA	***
(%)	Femminello	25.50±0.11 cC	30.16±0.05 aB	25.33±0.05 cC	12.99±0.12 eB	26.94±0.14 bA	14.29±0.01 dB	***
	Sign.	***	***	***	***	***	***	
Total Flavonoids	Castagnaro	276±3 dC	212±7 eC	488±11 aA	425±7 bB	419±8 bC	348±18 cC	***
	Fantastico	361±9 cA	303±6 dA	233±17 eC	509±8 bA	675±10 aB	678±4 aB	***
in juice (g/L)	Femminello	287±1 eB	408±4 cB	364±4 dB	390±12 cC	845±11 aA	824±7 bA	***
	Sign.	***	***	***	***	***	***	

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Table 5. Main Flavonoids in the aqueous extract: *Cloudy*. Results are presented as the mean value ± standard deviation, n=8, (2016-2017 and 2017-2018 harvest years). ***significance at $P < 0.001$; ** significance at $P < 0.01$; * significance at $P < 0.05$. Means in the same line are distinguished by small letters. Means in the same column are distinguished by capital letters .

	Cultivar	October	November	December	January	February	March	*Sign.*
Neoeriocitrin	Castagnaro	17.74±0.09 dB	25.53±0.10 bA	17.86±0.11 dA	18.66±0.09 cC	25.23±0.15 bA	28.89±0.14 aB	***
	Fantastico	18.06±0.10 dA	16.43±0.09 eC	18.09±0.14 dA	25.74±0.07 bA	24.3±0.20 cB	26.13±0.17 aC	***
(%)	Femminello	16.23±0.12 eC	23.36±0.23 bB	16.33±0.07 eB	19.11±0.10 dB	20.16±0.18 cC	33.49 ±0.05 aA	***
	***	***	***	***	***	***	***	
Naringin	Castagnaro	39.52±0.18 bB	35.78±0.18 dB	39.35±0.09 bB	37.64±0.08 cA	31.35±0.10 eC	42.07±0.15 aA	***
	Fantastico	33.44±0.23 cC	38.98±0.11 aA	33.50±0.10 cC	27.18±0.16 eC	34.38±0.08 bA	29.12±0.25 dC	***
(%)	Femminello	42.20±0.11 aA	33.36±0.07 cC	42.33±0.06 aA	31.17±0.06 eB	32.31±0.04 dB	37.96±0.06 bB	***
	***	***	***	***	***	***	***	
Neohesperidin	Castagnaro	25.71±0.27 bC	20.76±0.12 dC	25.64±0.2 bC	27.08±0.06 aC	22.05±0.22 cC	18.91±0.15 eB	***
	Fantastico	32.29±0.25 aA	24.26±0.12 dB	32.15±0.07 aA	31.19±0.16 bB	22.69±0.13 eB	30.43±0.15 cA	***
(%)	Femminello	29.52±0.34 cB	28.25±0.28 dA	29.22±0.03 cB	35.73±0.09 aA	32.19±0.03 bA	16.11±0.09 eC	***
	***	***	***	***	***	***	***	
Melitidin	Castagnaro	6.22±0.09 cA	6.52±0.06 bB	6.24±0.12 b cA	6.24±0.08 b cA	7.04±0.18 aA	2.60±0.05 dA	***
	Fantastico	4.28±0.07 cB	7.81±0.15 aA	4.27±0.10 cC	3.82±0.15 dB	6.69±0.14 bB	2.38±0.08 eB	***
(%)	Femminello	4.45±0.14 aB	4.3±0.24 aC	4.55±0.05 aB	3.37±0.07 bB	4.32±0.07 aC	2.13±0.05 cC	***
	***	***	***	***	***	***	***	
Brutieridin	Castagnaro	10.81±0.07 cB	11.61±0.23 bB	10.91±0.14 cB	10.38±0.14 dB	14.33±0.08 aA	7.53±0.10 eC	***
	Fantastico	11.93±0.15 bA	12.52±0.15 aA	11.99±0.06 bA	12.07±0.16 bA	11.94±0.15 bB	11.94±0.24 bA	**
(%)	Femminello	7.56±0.32 cC	10.73±0.22 a bC	7.56±0.06 cC	10.62±0.08 abB	11.02±0.06 aC	10.31±0.07 bB	***
	***	***	***	***	***	***	***	
Total Flavonoids	Castagnaro	4272±17 cC	4027±12 dC	4778±22 bC	6546±34 aC	4066±14 dC	4273±18 cC	***
	Fantastico	6680±28 cB	5926±12 dB	7816±14 bB	7960±25 aB	5671±30 eB	6671±34 cB	***
in Cloudy (mg/L)	Femminello	8271±82 dA	7753±42 eA	8489±12 cA	8651±9 bA	9254±10 aA	8212±23 dA	***
	***	***	***	***	***	***	***	

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Table 6. Antioxidant activity of bergamot juice. AAE = Ascorbic Acid Equivalent. Results are presented as the mean value ± standard deviation, n=8, (2016-2017 and 2017-2018 harvest years). ***significance at $P < 0.001$; ** significance at $P < 0.01$; * significance at $P < 0.05$. Means in the same line are distinguished by small letters. Means in the same column are distinguished by capital letters .

	Cultivar	**October**	**November**	**December**	**January**	**February**	**March**	*Sign.*
DPPH assay - juice	Castagnaro	380.8±1.16dC	320.3±2.30eC	494.9±2.17aA	431.2±1.11bA	386.6±2.21cC	305.6±1.85fC	***
	Fantastico	423.4±2.17bA	361.7±3.79cB	340.7±1.70dB	416.8±2.41bB	453.3±4.62aB	450.9±4.23aB	***
(mg AAE/100mL)	Femminello	394.3±4.01cB	420.6±5.11bA	330.8±3.53dC	340.0±7.71dC	478.4±6.59aA	473.9±4.10aA	***
	Sign.	***	***	***	***	***	***	
	Castagnaro	45.21±0.96aA	34.44±1.93cdB	40.67±0.56bA	32.66±0.10dB	35.77±0.47cA	28.41±0.08eB	***
	Fantastico	45.84±0.39aA	36.93±0.64bAB	35.91±0.64bcB	35.05±0.64cdA	34.17±0.75dA	33.65±0.57dA	***
FRAP assay - juice	*Femminello*	39.67±0.28aB	38.04±0.58aA	35.24±0.66bB	33.89±0.78AbcB	31.82±0.76cB	26.26±1.35dC	***
	Sign.	***	*	***	**	***	***	

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Table 7. The correlation matrix of biometrics is built on the basis of 48 values for each parameter (4 replicates x 2 harvest years x 3 cultivars). In the South-West section of the matrix there are the r value (above) and the significance level (below) with *P* < 0.05, *. In the North-East section of the matrix there is the *t* value (in italics) with the significance of the *t*-Test calculated at 95% confidence interval and the R^2 value (underlined).

	Vert ical Diameter	Horizontal Diameter	Pulp in juice	Fruit Weight	Peel Weight	Juice conent	Fruit Weight/ Peel weight	Fruit Weight/ Juice content	Juice content/ Peel Weight	Turbidity
Vertical Diameter	**1**	*1.38* 0.918	*7.99* 0.002	*19.33* 0.766	*14.76* 0.774	*26.50* 0.013	*4.59* 0.028	*0.28* 0.702	*45.50* 0.587	*37.96* 0.000
Horizontal Diameter	0.958 n.s.	**1**	*6.50* 0.001	*19.25* 0.846	*14.62* 0.689	*26.10* 0.002	*5.47* 0.000	*0.37* 0.638	*41.89* 0.426	*37.44* 0.009
Pulp in juice	0.048 ***	0.024 ***	**1**	*19.12* 0.001	*13.78* 0.000	*24.05* 0.000	*10.60* 0.031	*3.98* 0.003	*40.54* 0.001	*34.96* 0.034
Fruit Weight	0.875 ***	0.920 ***	0.090 ***	**1**	*16.56* 0.664	*17.23* 0.013	*19.37* 0.034	*19.26* 0.619	*19.84* 0.345	*16.69* 0.000
Peel Weight	0.880 ***	0.830 ***	0.000 ***	0.815 ***	**1**	*2.65* 0.006	*15.27* 0.158	*14.45* 0.552	*18.08* 0.619	*0.35* 0.009
Juice content	-0.114 ***	0.042 ***	0.001 ***	0.112 ***	-0.075 ***	**1**	*27.33* 0.129	*24.29* 0.242	*34.11* 0.360	*5.52* 0.186
Fruit Weight/Peel Weight	-0.166 ***	0.020 ***	0.175 ***	0.185 ***	-0.397 ***	0.359 ***	**1**	*2.70* 0.003	*26.13* 0.220	*38.63* 0.056
Fruit Weight/Juice content	0.838 n.s.	0.799 n.s.	0.057 ***	0.787 ***	0.743 ***	-0.492 ***	-0.054 ***	**1**	*16.53* 0.682	*34.16* 0.071
Juice content / Peel Weight	-0.766 ***	-0.653 ***	0.032 ***	-0.587 ***	-0.787 ***	0.600 ***	0.469 ***	-0.826 ***	**1**	*46.66* 0.081
Turbidity	0.022 ***	0.093 ***	0.185 ***	0.010 ***	-0.093 n.s.	0.431 ***	0.236 ***	-0.266 ***	0.284 ***	**1**

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Table 8. The correlation matrix of the physico-chemical properties of bergamot juice and bergamot *Cloudy* is built on the basis of 48 values for each parameter (4 replicates x 2 harvest years x 3 cultivars). In the South-West section of the matrix there are the r value (above) and the significance level (below) with $P < 0.05$, *. In the North-East section of the matrix there is the *t* value (in italics) with the significance of the *t*-Test calculated at 95% confidence interval and the R^2 value (underlined).

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Table 8

		pH juice	°Brix juice	TA juice	°Brix(%)/ TA/%) juice	Vit. C juice	Formol N juice	Neoerioc juice	Naringin juice	Neohesp juice	Melitidin juice	Brutieridin juice	Tot. Floids juice	Neoerio *Cloudy*	Naringin *Cloudy*	Neohesp *Cloudy*	Melitidin *Cloudy*	Brutieridin *Cloudy*	Tot. Floids *Cloudy*	DPPH assay juice	FRAP assay juice
Juice	pH	**1**	*76.65* 0.326	*50.76* 0.548	*32.83* 0.169	*31.32* 0.413	*14.31* 0.015	*26.24* 0.120	*36.48* 0.251	*24.92* 0.023	*17.65* 0.146	*21.45* 0.285	*17.55* 0.249	*28.87* 0.378	*53.78* 0.048	*34.16* 0.149	*10.13* 0.046	*24.26* 0.047	*28.44* 0.072	*50.78* 0.060	*50.94* 0.330
	°Brix	-0.571 ***	**1**	*43.67* 0.529	*24.10* 0.153	*30.98* 0.413	*78.41* 0.119	*12.73* 0.309	*28.34* 0.036	*16.67* 0.002	*0.40* 0.072	*15.18* 0.094	*17.30* 0.490	*19.42* 0.184	*43.33* 0.002	*25.30* 0.011	*16.34* 0.180	*8.36* 0.008	*28.41* 0.021	*49.98* 0.272	*41.07* 0.194
	TA	-0.740 ***	0.727 ***	**1**	*18.93* 0.432	*28.73* 0.588	*51.46* 0.010	*32.97* 0.317	*14.50* 0.309	*22.54* 0.001	*40.53* 0.078	*17.97* 0.377	*15.75* 0.147	*23.51* 0.446	*11.54* 0.003	*18.49* 0.177	*46.73* 0.135	*40.02* 0.016	*28.24* 0.025	*44.67* 0.006	*11.34* 0.453
	°Brix(%)/ TA(%)	0.411 ***	-0.391 ***	-0.657 ***	**1**	*29.97* 0.008	*33.71* 0.076	*13.39* 0.003	*4.34* 0.046	*4.54* 0.306	*21.51* 0.252	*1.88* 0.003	*16.61* 0.173	*4.55* 0.006	*9.78* 0.278	*0.62* 0.432	*28.29* 0.209	*20.18* 0.004	*25.07* 0.846	*47.54* 0.017	*9.55* 0.109
	Vit. C	-0.643 ***	-0.643 ***	0.767 ***	0.089 ***	**1**	*31.62* 0.002	*30.63* 0.280	*29.71* 0.392	*30.23* 0.028	*30.96* 0.168	*30.08* 0.513	*3.58* 0.236	*30.22* 0.415	*29.46* 0.002	*29.93* 0.166	*31.19* 0.124	*30.85* 0.016	*25.98* 0.013	*8.27* 0.000	*29.45* 0.926
	Formol Number	0.124 ***	0.345 ***	-0.098 ***	0.276 ***	-0.042 ***	**1**	*27.60* 0.001	*37.30* 0.161	*25.77* 0.039	*19.40* 0.000	*22.10* 0.030	*17.57* 0.064	*29.82* 0.040	*54.78* 0.078	*35.06* 0.208	*12.91* 0.001	*36.64* 0.001	*28.44* 0.055	*50.86* 0.048	*51.90* 0.002
	Neo eriocitrin	0.346 ***	-0.556 ***	-0.563 ***	0.054 ***	-0.529 ***	0.032 ***	**1**	*17.80* 0.020	*7.56* 0.150	*9.96* 0.004	*8.35* 0.096	*17.07* 0.166	*8.65* 0.303	*26.99* 0.002	*14.26* 0.232	*19.67* 0.003	*7.38* 0.002	*28.39* 0.016	*49.13* 0.014	*26.06* 0.196
	Naringin	0.501 ***	-0.190 ***	-0.556 ***	0.214 ***	-0.626 ***	0.401 ***	0.141 ***	**1**	*8.62* 0.032	*25.67* 0.080	*5.47* 0.759	*16.43* 0.019	*8.67* 0.202	*4.71* 0.100	*3.77* 0.201	*32.18* 0.121	*24.57* 0.162	*28.32* 0.005	*46.93* 0.023	*4.61* 0.390
	Neo hesperid.	0.153 ***	0.045 ***	-0.024 ***	-0.553 ***	-0.166 ***	-0.197 ***	-0.387 ***	0.179 ***	**1**	*14.97* 0.599	*1.95* 0.191	*16.80* 0.042	*0.29* 0.006	*14.36* 0.218	*5.17* 0.316	*20.99* 0.100	*13.30* 0.002	*28.36* 0.194	*48.13* 0.022	*14.05* 0.039
	Melitidin	-0.382 ***	0.269 n.s.	0.280 ***	0.502 ***	0.410 ***	0.000 ***	0.066 ***	-0.282 ***	-0.774 ***	**1**	*14.19* 0.174	*17.30* 0.163	*17.01* 0.032	*37.46* 0.077	*22.54* 0.085	*9.54* 0.339	*4.57* 0.008	*28.41* 0.182	*49.91* 0.045	*35.96* 0.161
	Brutier.	-0.534 ***	0.306 ***	0.614 ***	0.050 n.s.	0.716 ***	-0.173 ***	-0.310 ***	-0.871 ***	-0.437 n.s.	0.417 ***	**1**	*16.69* 0.097	*1.80* 0.236	*9.88* 0.001	*2.40* 0.063	*18.67* 0.104	*12.72* 0.047	*28.35* 0.004	*47.66* 0.001	*9.73* 0.488
	Total Floids	0.499 ***	-0.700 ***	-0.384 ***	-0.416 ***	-0.486 ***	-0.252 ***	0.407 ***	0.137 ***	0.205 ***	-0.404 ***	-0.311 ***	**1**	*16.79* 0.236	*16.25* 0.111	*16.58* 0.010	*17.46* 0.126	*17.22* 0.016	*26.37* 0.125	*11.85* 0.583	*16.25* 0.242
Cloudy	Neo eriocitrin	0.615 ***	-0.429 ***	-0.668 ***	0.080 ***	-0.644 ***	0.200 ***	0.550 ***	0.449 ***	-0.077 n.s.	-0.178 ***	-0.486 n.s.	0.486 ***	**1**	*15.17* 0.071	*5.23* 0.370	*24.07* 0.195	*15.37* 0.013	*28.36* 0.026	*48.14* 0.021	*14.78* 0.435
	Naringin	-0.219 ***	-0.048 ***	0.054 ***	0.527 ***	0.048 ***	0.280 ***	-0.040 ***	0.316 ***	-0.467 ***	0.278 ***	0.035 ***	-0.333 ***	-0.266 ***	**1**	*9.19* 0.187	*46.96* 0.031	*37.38* 0.497	*28.30* 0.045	*46.42* 0.068	*0.00* 0.003
	Neo hesperid.	-0.386 ***	0.105 ***	0.421 ***	-0.657 n.s.	0.408 ***	-0.456 ***	-0.482 ***	-0.448 ***	0.562 ***	-0.291 ***	0.251 ***	-0.099 ***	-0.608 ***	-0.432 ***	**1**	*29.49* 0.038	*21.27* 0.001	*28.33* 0.358	*47.47* 0.000	*8.97* 0.148
	Melitidin	-0.214 ***	0.424 ***	0.367 ***	0.457 ***	0.352 ***	0.032 ***	-0.052 ***	-0.348 ***	-0.316 ***	0.582 ***	0.323 ***	-0.355 ***	-0.442 ***	0.175 ***	-0.195 ***	**1**	*18.21* 0.171	*28.43* 0.282	*50.46* 0.002	*44.75* 0.165
	Brutier.	0.217 ***	0.091 ***	0.125 ***	-0.062 ***	0.127 ***	0.022 ***	0.044 ***	-0.403 ***	0.047 ***	0.092 ***	0.217 ***	0.128 ***	0.116 ***	-0.705 ***	0.032 ***	0.414 ***	**1**	*28.40* 0.073	*49.69* 0.065	*35.65* 0.029
	Total	-0.268 ***	-0.146 ***	0.158 ***	-0.920 ***	-0.114 ***	-0.235 ***	-0.126 ***	-0.071 ***	0.441 ***	-0.427 ***	-0.066 ***	0.353 ***	-0.161 ***	-0.213 ***	0.598 ***	-0.531 ***	-0.271 ***	**1**	*26.73* 0.040	*28.30* 0.043

	Floids																				
Juice	DPPH assay	0.245 ***	-0.522 ***	-0.079 ***	-0.128 ***	0.000 ***	-0.220 ***	0.116 ***	-0.151 ***	0.149 ***	0.211 ***	0.024 ***	0.764 ***	0.143 ***	-0.260 ***	0.017 ***	-0.048 ***	0.256 ***	0.199 ***	**1**	*46.40* 0.001
	FRAP assay	-0.574 ***	0.441 ***	0.673 ***	-0.330 ***	0.962 ***	0.044 ***	-0.309 ***	-0.624 ***	-0.196 ***	0.402 ***	0.699 ***	-0.492 ***	-0.659 ***	0.054 n.s.	0.385 ***	0.406 ***	0.171 ***	-0.206 ***	0.024 ***	**1**

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* * * * * * H I E R A R C H I C A L   C L U S T E R   A N A L Y S I S * * * * * *

Dendrogram using Complete Linkage

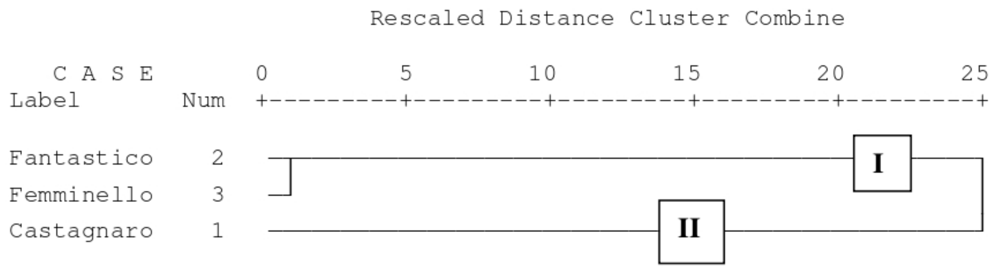

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Figure 1. Two-dimensional dendrogram obtained from the cluster analysis of the fruit biometrics and of the physico-chemical properties of juice and cloudy of bergamot fruit (Citrus bergamia, Risso).

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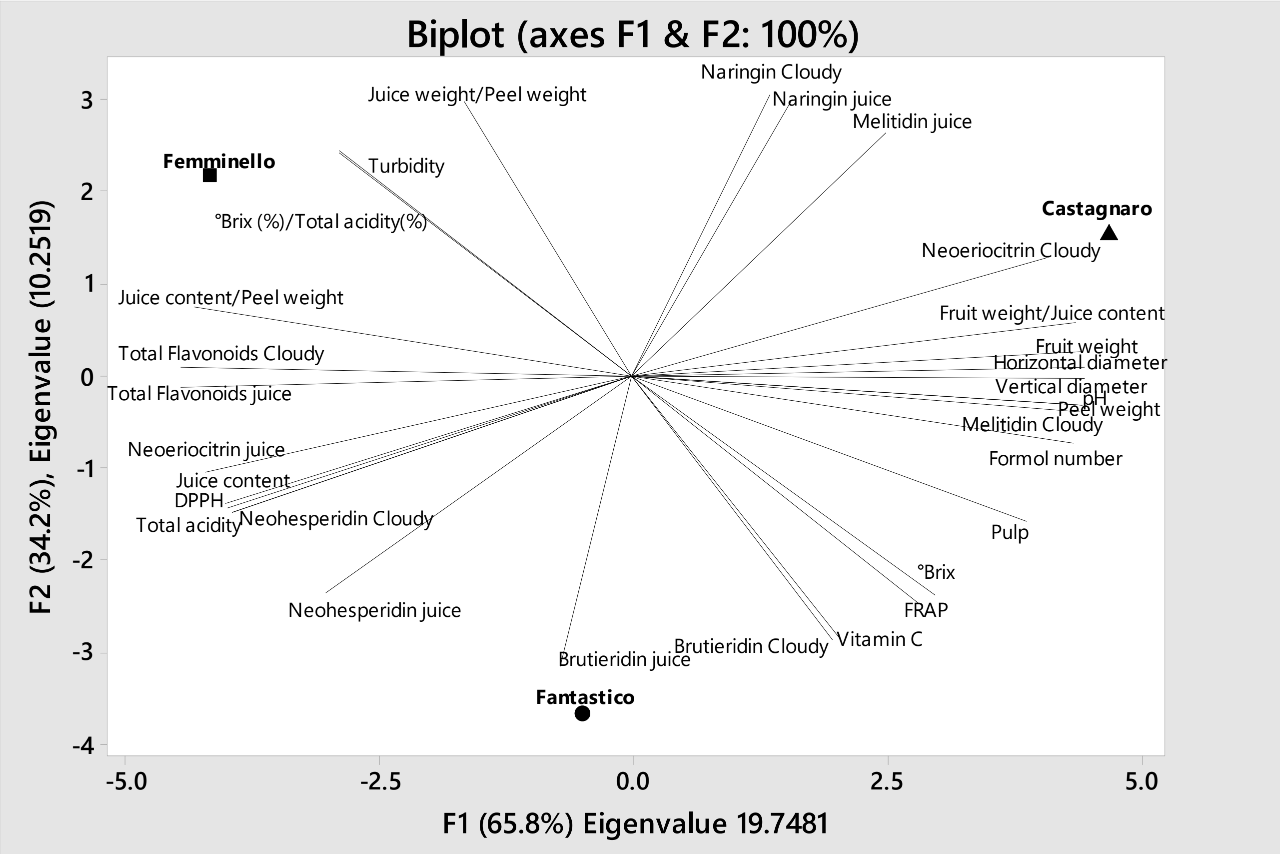

Figure 2. Score plot of the PCA performed on the biometrics and the physico-chemical properties of juice and cloudy of the three cultivars (*Castagnaro*, *Fantastico* and *Femminello*) of bergamot fruit (*Citrus bergamia*, Risso).