Evaluation of granulation and quality parameters of Monte Parnaso late navel orange from South Brazil

Marlise Perini[1], Wendel Paulo Silvestre[2,], Camila Bonatto Vicenço[2], Gabriel Fernandes Pauletti[1,2]*

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ABSTRACT

This work aimed to evaluate granulation incidence and severity and quality parameters of 'Monte Parnaso' navel orange in different harvest and collection dates. The 2018 and 2019 harvests were evaluated, and the collection dates of 0, 15, 30, and 45 days after commercial collection. The evaluated parameters were granulation incidence and severity, fruit weight, longitudinal and transversal diameters, peel thickness, juice content, soluble solids content, titratable acidity, SS/TA ratio, and ascorbic acid content. The results underwent chi-squared test to evaluate granulation incidence and severity, and analysis of variance for the quality parameters; occurring statistical difference, the means were compared by Tukey's multiple range test at 5 % probability. Granulation incidence and severity were not related to collection time. Only severity was related to the harvest. The quality parameters were influenced significantly only by the harvest factor, with exception of the parameters of titratable acidity, influenced only by the factor collection date, and ascorbic acid content, which presented interaction between factors. Granulation incidence and severity were higher in the larger and heavier fruits. The frequency of fruits in the categories of higher severity (> 50 %) was higher in the 2018 harvest than in the one of 2019.

Keywords: *Citrus sinensis* (L.) Osb.; *Poncirus trifoliata* (L.) Raf.; *Citrus* physiology; Fruit quality.

INTRODUCTION

Brazil is the largest world *Citrus* producer; the Rio Grande do Sul state presents itself as the fifth largest *Citrus* producer of the country (IBGE, 2018), and with a large potential in the area of citriculture, once *Citrus* plants may be cultivated in most of its territory. The state also has a large market for *in natura* consumption, especially regarding late seedless oranges and mandarins (Oliveira *et al.*, 2008a). Data from the Agroclimatic Zoning for *Citrus* in the Rio Grande do Sul state reported several thousands of hectares suitable for the cultivation of Citrus, especially the table varieties, and that the region has climatic and soil conditions considered as ideal for the production of high-quality fruits (Wrege *et al.*, 2004; Sulzbach *et al.*, 2016).

The Monte Parnaso navel orange [*Citrus sinensis* (L.) Osb] is widely cultivated in Rio Grande do Sul; its cultivation is generally done in small family properties. The plant is vigorous and the fruits are seedless, with a late ripening (the harvest period lies between August to the end of November), which renders the fruits a high marketing value. This variety has as characteristics large fruits and with an intense yellow color of both the peel and the pulp (Efrom & Souza, 2018).

Considering that the Monte Parnaso navel orange is a table variety (for *in natura* consumption), high-quality fruits are essential; thus, the cultivation demands balanced nutrition management and water supply. In this sense, the knowledge of the physical-chemical and biologic attributes of the soils, the nutritional demand of the plants, and the edaphoclimatic conditions of the cultivation site are fundamental to guarantee and improve the final quality of the fruits (Oliveira & Scivittaro, 2011).

The rootstock also influences the characteristics and quality of the fruits of the scion cultivar; thus, caution is necessary for its choosing. In Rio Grande do Sul, the most

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[1] Universidade de Caxias do Sul, Caxias do Sul, Rio Grande do Sul, Brazil.
[2] Universidade de Caxias do Sul, Laboratório de Estudos do Sistema Solo, Planta e Atmosfera e Metabolismo Vegetal, Caxias do Sul, Rio Grande do Sul, Brazil.
*Corresponding author: wpsilvestre@ucs.br

used rootstock is the trifoliate [*Poncirus trifoliata* (L.) Raf.], especially due to its resistance to cold and for providing high fruit quality. Although being less vigorous than other rootstocks, the trifoliate confers resistance to important diseases, such as gummosis (*Phytophthora* spp.), the nematode *Tylenchulus semipenetrans*, the *Citrus* tristeza virus (CTV), and the *Citrus* sudden death associated virus (CSDaV) (Oliveira *et al.*, 2008b).

Koller *et al.* (2006) cited that the Monte Parnaso navel orange has low productivity (about 20 kg·plant^{-1}); however, depending on the orchard management and the edaphoclimatic conditions of the cultivation site, the average production may reach 25 t·ha^{-1}. According to Reis *et al.* (2008), the rootstock also has a strong influence on the overall productivity of this scion cultivar. The causes for this low productivity may be the high percentage of fruit drop and factors associated with physiological, hormonal, and nutritional aspects.

Granulation is a physiological disorder that affects *Citrus* fruits. The affected fruits develop gelled vesicles that are hard and firm. This disorder initiates in the pedicel end and moves to the center of the fruit; the affected parts of the pulp become whitish. It is very difficult to identify the affected fruits and remove them, once there is no external evidence of the presence of granulation, neither of possible losses to fruit quality (Ritenour *et al.*, 2004).

Little is known about the causes of this disorder, but it is believed that they may be related to plants excessively vigorous, fruits with large size, plant age, pruning severity, rootstock kind and its interaction with the scion cultivar, among other factors. It is considered that granulation is also associated with environmental factors, such as temperature, relative humidity, soil kind and its nutritional status, the position of the fruit in the tree, and fruit mass, among others. This disorder causes the reduction of the juice content and the mass of the fruit, which may render it unfit for marketing, depending on the severity of the disorder (Gravina, 2014; Wang *et al.*, 2014; Jie *et al.*, 2021).

Granulation is a common problem in the Serra Gaúcha region and several other *Citrus* productive regions worldwide. In the Serra Gaúcha, this disorder may be linked mainly to harvest delay, which is carried out aiming to increase the market value of the fruits. According to Oliveira *et al.* (2010), this problem may be mitigated by an early harvest. However, Ortuzar *et al.* (2003) reported that, in Chile, granulation was already present even after the regular harvest period, not being the harvest delay the main factor associated with granulation. In Brazil, granulation is considered to occur in all productive regions, affecting especially late harvest oranges and mandarins (Efrom & Souza, 2018; Brugnara, 2019).

In this sense, the objective of the present work was to evaluate the degree of incidence and severity of granulation and the quality parameters of Monte Parnaso navel oranges in different harvests and harvest dates in the Serra Gaúcha region, South Brazil.

MATERIALS AND METHODS

Plant material and sample preparation

The experiment was carried out in a *Citrus* orchard located in the municipality of Caxias do Sul, RS, under the geographical coordinates of 29º12’ S and 51º13’O, and an altitude of 705 m. According to Alvares *et al.* (2013), the Serra Gaúcha region has a Cfb climate (subtropical highland climate with no dry season), with an average yearly temperature of 17.2 °C. The experiment was conducted for two harvest seasons (2017/2018 and 2018/2019), from September 2017 to November 2019.

The scion cultivar studied was the Monte Parnaso one [*Citrus sinensis* (L.) Osbeck], grafted on the trifoliate rootstock [*Poncirus trifoliata* (L.) Raf.]. The orchard had an age of ten years. The spacing was 4.5 m between lines and 3.0 m between plants. The stage of ripening of the fruits was monitored through the determination of the soluble solids content (SS) of the oranges, being the fruits considered as ripe when the SS reached the range of 9-11 °Brix. When the fruits were considered ripe, the first harvest (day of harvest zero) was carried out; the average SS/TA ratio of the fruits was 17.7 and 20.3 for the 2018/2019 and the 2019/2020 harvests, respectively. The other harvests were carried out after periods of 15 days between them (zero, 15, 30, and 45 days).

Evaluation of fruit granulation

To determine the granulation incidence and severity, it was carried out the procedures proposed by Santos *et al.* (2010). The fruits were cut transversally in the basal position (¼ pedicellar). The granulation was evaluated using a visual scale of incidence (presence/absence) and severity (1-25 %; 25-50 %; 50-75 %; 75-100 %; Figure 1). Relative to the visual scale, it was considered the range of 1-25 % when up to ¼ of the fruit presented granulation, 25-50 % when ¼ to half of the fruit presented granulation, 50-75 % when from half to ¾ of the fruit had granulation, and 75-100 % when more than ¾ of the fruit was granulated (Santos *et al.*, 2010).

Evaluation of the quality parameters

The following quality parameters were evaluated: average fruit mass, longitudinal and transversal diameters, peel thickness, juice content, soluble solids content, titratable acidity, SS/TA ratio, and ascorbic acid (vitamin C) content.

To determine the average fruit mass, the fruits were weighed individually using a semi-analytical balance (AS1000C, Marte, Brazil) with a capacity of 1 kg and a resolution of 0.01 g. The transversal and longitudinal diameters of the fruits and the peel thickness were measured using a digital caliper (PD150, TMX, Brazil), with a measurement range of 0-150 mm and a resolution of 0.01 mm.

The juice was extracted using an electric juice extractor. The extracted juice was measured using a glass graduated cylinder with a capacity of 1 L and a resolution of 10 mL; the content was calculated using equation 1.

$$Jc\ (\%\ v\backslash m) = 100 \times \frac{V}{M} \qquad (1)$$

Being 'Jc' the juice content, 'V' the volume of juice extracted (mL), and 'M' the mass of the fruits used in the extraction (g).

The soluble solids content was determined using an analogic refractometer (RT-30ATC, Incoterm, Brazil), with a measuring range of 0-30 °Brix and a resolution of 0.25 °Brix, using as samples the juice extracted from each replicate. The titratable acidity was determined following the method 310/IV, proposed by Instituto Adolfo Lutz (IAL, 2008). The SS/TA ratio value was calculated as the quotient between the soluble solids content and the titratable acidity (ratio = SS/TA). The ascorbic acid content was determined following the method 364/IV, from Instituto Adolfo Lutz (IAL, 2008).

Experimental design and statistical analysis

The experimental design was bifactorial, completely randomized. The evaluated factors were the harvest (2017/2018 and 2018/2019) and the date of harvest (zero, 15, 30, and 45 days after the date recommended for harvesting). Each treatment was composed of 160 fruits collected from ten random plants in each treatment. 100 fruits were used in the determination of granulation incidence and severity and 60 were used in the determination of the quality parameters. For the quality parameters, the 60 fruits of each treatment were grouped in six replicates of 10 fruits to carry out the statistical analysis.

The granulation data (incidence and severity) underwent the chi-squared test at 5 % probability ($\alpha = 0.05$) to verify the existence of possible correlations between the studied factors and the patterns of granulation appearance. The granulation data and the quality parameters underwent Spearman correlation analysis at 5 % probability ($\alpha = 0.05$); the tests were carried out using the Statistica 12 software (Statsoft, USA). The data on the quality parameters of the fruits underwent analysis of variance (ANOVA) and the means were compared by Tukey's multiple range test at 5 % probability ($\alpha = 0.05$), using the AgroEstat software.

RESULTS AND DISCUSSION

Fruit granulation

The data on granulation incidence and severity underwent the chi-squared test; the results of the chi-squared test are presented in Table 1.

Considering the results of the chi-squared test, it was possible to verify that both granulation incidence and severity were not related to the harvest date. Relative to the harvest, only granulation severity was related to this factor; no significant relationship was observed between granulation incidence and the harvest.

According to Ortuzar *et al.* (2003), who studied the Lane Late and Navelate late navel oranges [both *C. sinensis* (L.) Osb.] in the Chilean regions of Peumo, which has a Csb (Mediterranean) climate, and San Pedro, which has a Bsk (cold desertic) climate (Sarricolea *et al.*, 2017), granulation was presented even before the harvest time, being probably associated to fruit development. However, the same authors commented on the absence of granulation before the harvest period for the Navelate orange.

The results obtained in this work differ from the ones obtained by Ortuzar *et al.* (2000), who studied the Lane Late and Navelate late navel oranges in two harvests and different harvest dates and reported that granulation incidence followed a trend of increase with harvest delay, with a significant increase in overall incidence, independently of the orange variety, the harvest, and the cultivation

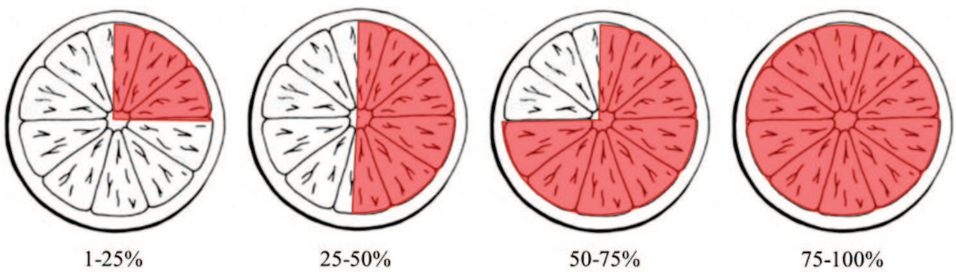

Figure 1: Visual scale of granulation severity considering the percentage of the fruit affected. Source: authors (2021).

site. Nauer *et al.* (1990), evaluating the Lane Late variety, also reported an increase of granulation incidence with the delay in the harvest (longer time the fruit remains on the mother plant).

The data of relative frequency (percentage of fruit sum) of granulation for the harvests are presented in Table 2.

By analyzing Table 1, it was possible to observe that the fruits of the 2019 harvest had lesser granulation incidence and severity relative to the 2018 harvest; it is also noteworthy that most of the fruits that presented granulation in the 2019 harvest were classified in the severity degree of 1-25 % (81.2 %); about half (57.6 %) of the fruits from the 2019 harvest were classified in this category. For granulation severities above 50 %, the fruits become commercially unfeasible, therefore, it is desirable that, if granulation occurs, this should be as mild as possible. In this sense, it can be seen from Table 2, that 22.1 % of the fruits of the 2018 harvest would be lost; in the 2019 harvest, this percentage reduced to 8.6 %, a reduction of about 2.5 times. It is important to highlight that in the 2018 harvest both granulation incidence and severity in all categories with exception of 1-25 % were higher relative to the 2019 harvest, even without statistical difference by the chi-squared test.

Sinclair & Jollife (1961) and Burns & Achor (1989) cited that granulation severity may be associated with the senescence of the fruits. This way, the longer the time the fruit remains on the mother plant, the higher the granulation severity. Ortuzar *et al.* (2003) also reported similar behavior, showing that, in 1997 harvest, when the 'Lane Late' and 'Navelate' navel oranges were collected in November and December, there was severe granulation; on the 1998 harvest, in which the last collection was carried out in November, the fruits presented only mild granulation. It could be inferred that granulation incidence and severity may be highly related to the harvest (productive cycle) and linked to nutritional or edaphoclimatic effects.

It could be considered that the relationship observed between the harvest and granulation severity (Table 1) is a result of an association of climatic and physiological effects that may likely vary with the harvest, such as water balance, temperature, and the presence of several simultaneous physiological stages in the plant (flowering, vegetative growth, fructification), that may cause stress in the developing fruits.

Santos *et al.* (2010) studied the occurrence of granulation in tangerines and navel oranges, both early and late, in the region of Viçosa (Southeast Brazil), which has a Cwa – subtropical humid – climate according to Vianello & Alves (2000). The authors reported that the fruits from the Navelina and Salusiana oranges, which ripe earlier and are harvested between April and June, have not presented granulation. On the other hand, the Navelate orange, which is of late ripening, being harvested between August and November, presented granulation with a small degree of severity. However, this behavior was not observed in the tangerines; they have had a higher incidence and severity of granulation. Sharma *et al.* (2006) observed that granulation tends to be more intense and severe in tangerines than in oranges.

The behavior observed in these studies may explain why there was not a relationship between the harvest date and the incidence and severity of granulation seen in the present work (Table 1). Considering that the Monte Parnaso orange is a late variety, it is likely that granulation was already established before the start of the harvest,

Table 1: Results of the chi-squared test for the parameters of granulation incidence and severity for Monte Parnaso navel oranges relative to the harvest and the date of harvest

Factor	Parameter	Chi-squared test	
		p-value	Significance*
Harvest	Incidence	0.6730	No
	Severity	> 0.001	Yes
Date of harvest	Incidence	0.9981	No
	Severity	0.3495	No

* - p-values higher than 0.05 do not have statistical significance by the chi-squared test, indicating that the tested variables did not correlate themselves.

Table 2: Relative frequencies (percentage) of granulation incidence and severity of Monte Parnaso navel oranges relative to the productive years of 2018 (2017-2018) and 2019 (2018-2019)

Harvest	Incidence (%)	Severity			
		1-25 %	25-50 %	50-75 %	75-100 %
2018	79.2	57.5	20.4	7.4	14.7
2019	38.3	81.2	10.2	4.3	4.3

with little to no influence of the additional time of the fruit on the plant.

According to Wrege *et al.* (2004), the Rio Grande do Sul state is divided into six regions based on their potential for *Citrus* cultivation, based on the variables of thermal summation and risk of frost. The site in which the studied orchard was implanted belonged to region 4, with a frost risk of 35-40 % and a thermal summation of 2000-2500 growing degree days.

Cunha (2003) cited that frost is the weather event that poses the greatest threat to citriculture in South Brazil, where there is large weather variability. Although the damage in *Citrus* is caused by temperatures equal to or lower than -2 °C, temperatures below 15 °C during fruit ripening are considered ideal to produce fruits with high quality (João, 2002; Wrege *et al.*, 2004). The data of average monthly temperature for the harvest periods of 2018 and 2019 are presented in Figure 2(a).

Observing Figure 2(a), it was possible to see that there was no important difference between the average monthly temperatures from the start of the cycle (November) until May (a difference of about 2 °C in this period); the amplitude was of approximately 16 °C (11 to 27 °C), following the normal climatic trend of lower temperatures in winter months (June to August). It is important to observe that the harvesting period for the Monte Parnaso navel orange lies from the end of September up to the start of November.

The average temperature for June presented the highest difference, of about 5 °C (12 °C in the 2017-2018 harvest and 17 °C in the 2018-2019 one); the fall in the average temperatures for the 2018-2019 harvest was also sharper than in the 2017-2018 harvest, whose reduction was approximately linear. From July on, it was possible to observe that the average temperatures increased steadily and more regularly in the 2018-2019 harvest; this behavior was not observed in the 2017-2018 harvest, in which the

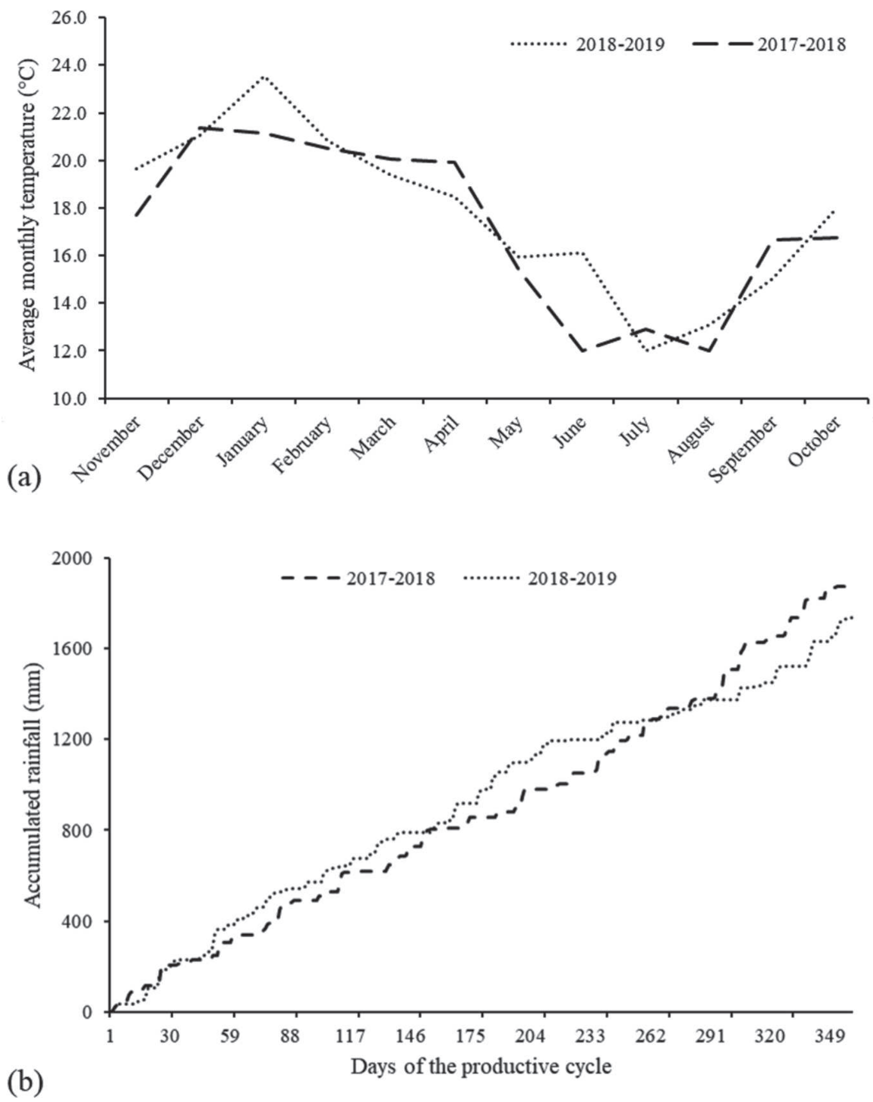

Figure 2: Average monthly temperature (a) and accumulated rainfall (b) relative to the productive cycles of 2018 (2017-2018) and 2019 (2018-2019) for the municipality of Caxias do Sul, Serra Gaúcha region, South Brazil. Source: adapted from INMET (2019).

average temperatures presented an erratic behavior, with a sharp increase from August to September (about 5 °C, from 12 to 17 °C).

Sudden temperature variations and water stress affect negatively and severely *Citrus* production; periods of stress reduce and can even paralyze both plant and fruit growth (Oliveira *et al.*, 2012). However, it was observed that the average temperatures in both harvests have not presented a high variation, with exception of May, in the middle of the productive cycle. Thus, it was not possible to establish a safe association between average temperature and the presence of granulation, at least, not only these two parameters isolated.

The data of accumulated rainfall for the two productive cycles are presented in Figure 2(b).

According to Figure 2(b), the rainfall was evenly distributed throughout the productive cycle in both harvests, indicating the absence of water stress, or the occurrence of little stress in short periods. This may also be drawn by verifying the total accumulated rainfall, once this, in both cycles, was above 1,500 mm (approximately 1,900 mm in the 2017-2018 harvest and 1,800 mm in the 2018-2019 harvest). Relative to the rainfall distribution, the 2018-2019 has had a higher accumulated rainfall in the approximate period between days 60 (January) and 270 (end of July) of the cycle; from day 270 on, the 2017-2018 harvest has had a higher accumulated rainfall. The accumulated rainfall, associated with the average monthly temperature, may have contributed to the appearance/ increase of granulation.

The water necessity of the *Citrus* plants varies with plant age, species, cultivar, planting system and orchard management, developmental stage, and climatic conditions, which affect the characteristics of water absorption by the roots, water transport through the plant, stomata opening, and plant transpiration (Suassuna *et al.*, 2012; Mesejo *et al.*, 2016). Mesejo *et al.* (2016) commented that drought or excess rainfall/soil humidity may have a stressing effect on the plants, rendering them susceptible to the development of physiological disorders, such as granulation and/or splitting.

The amount of rainfall considered as ideal to supply the water demand of *Citrus* plants is 1,500 mm·year^{-1}, with a balanced distribution throughout the entire year. However, rainfall in the range of 600-1,300 mm·year^{-1} is already considered as enough for plant development and cycle completion, if properly distributed (Böettcher *et al.*, 2018; Koller *et al.*, 2013).

Evaluation of fruit quality parameters

The parameters of average fruit mass (AFM), transversal and longitudinal diameters (Ø), peel thickness, juice content, soluble solids content, and SS/TA ratio were only influenced by the factor harvest, not being influenced by the harvest date. The results of these parameters relative to each harvest are presented in Table 3.

The fruits of the 2018 harvest presented higher average fruit mass, longitudinal and transversal diameters, and peel thickness than the ones of the 2019 harvest. However, juice content, soluble solids content, and SS/TA ratio values were higher in the 2019 harvest (Table 3).

Relative to the transversal diameter of the fruits, Santos *et al.* (2010) studying oranges and tangerines, observed that the Navelate and Navelina varieties (both *Citrus sinensis* (L.) Osb.) have had the higher average transversal diameters (91.6 and 96.0 mm, respectively), whereas the Salustiana (*Citrus sinensis* (L.) Osb.) and Marisol (*Citrus clementina* Hort. ex Tan.) have had the smallest average transversal diameters (67.6 and 63.9 mm, respectively). The same authors reported that the fruits with the largest longitudinal diameters were the Ortanique (*Citrus reticulata* × *Citrus sinensis*) and Okitsu (*Citrus unshiu* Marc.) varieties (93.2 and 89.4 mm, respectively); the smallest longitudinal diameters occurred in the Marisol and Salustiana varieties (71.4 and 68.6 mm, respectively). In the present work, relative to the 2018 harvest, the transversal diameter was larger (96.75 mm) than the largest values reported by Santos *et al.* (2010); the longitudinal diameter was between the range cited by the same authors. In the 2019 harvest, both the transversal and longitudinal diameters were within the ranges cited previously.

In this work, the peel thickness was 5.63 and 5.00 mm for the 2017-2018 and 2018-2019 harvest, respectively. Efrom & Souza (2018) cited the peel thickness of the Monte Parnaso navel orange as ‘medium’, but without any specific value; however, these authors reported as average peel thickness of the Lane Late, Salustiana, and Navelate varieties (which are similar to Monte Parnaso) as 3.0, 6.0, and 3.3 mm, respectively. Duarte *et al.* (2011), evaluating the quality of Valência oranges [*Citrus sinensis* (L.) Osb.] with crop loading, reported peel thickness in the range of 4.0-5.2 mm. Wright (2008) reported an average peel thickness of 4.01 mm for the Lane Late and 2.48 mm for the Cara Cara [*Citrus sinensis* (L.) Osb.] navel oranges.

Schäfer *et al.* (2001), studying the effect of phytoregulators on Monte Parnaso navel oranges grafted on the trifoliate rootstock in the Brazilian municipality of Butiá (South Brazil), which has a Cfa (subtropical humid) climate, reported the following results relative to the control treatment (no application of phytoregulators): average fruit mass of 286 g; soluble solids content of 8.07 °Brix, and a SS/TA ratio of 8.86, values lower than those found in this work. Relative to the juice content, the same authors reported a juice content of 46.78 % v/

w, which was higher than the one found in the 2017-2018 harvest (42.81 % v/w), but lower than the one of the 2018-2019 harvest (47.98 % v/w).

Since granulation causes the reduction of the amount of juice present in the fruit; in severe cases this disorder may cause the drying of the fruits, gelling their internal vesicles (Ritenour *et al.*, 2004). Relating the dimensional parameters of the fruits (transversal and longitudinal diameters), the average mass, and the juice content, it was possible to verify that, although in 2018 the fruits have had larger size and mass, the juice content was smaller (42.81 % v/w for the 2018 harvest and 47.98 % v/w for the 2019 harvest, respectively; Table 3). In 2018 the granulation incidence was 79.2 %, in which 57.5 % of the fruits presented a severity classified as 0-25 %; this harvest also presented the highest number of fruits with high severity (> 50 %). In 2019, although the fruits have had a smaller size and average mass, they have presented higher juice content and lower granulation incidence (38.3 %; Table 1) in which the severity was always almost mild (81.2 % in the severity class of 1-25 %). Thus, it could be inferred that larger fruits may be more susceptible to the appearance and development of granulation, either by fruit hypertrophy or due to other factors (Ortuzar *et al.*, 2003).

It is important to highlight that in 2018 the fruits also had a lower soluble solids content and SS/TA ratio; these parameters are linked to fruit quality and stage of ripeness. They may be affected by climatic factors, whose variation may also be associated with the presence of fruit granulation. According to Santos *et al.* (2010), granulation may accelerate the decomposition of sugars and organic acid, causing an unbalance between these classes of compounds, and changing the quality parameters associated with them.

Relative to the titratable acidity, it had a distinct behavior from the other quality parameters; the factor harvest has not influenced the titratable acidity of the fruits, but the harvest date did have influence. Table 4 presents the data of titratable acidity as a function of the harvest date.

There was a progressive reduction of the titratable acidity of the fruits with the delay of the harvest date (Table 3). This behavior was also observed by Ortuzar *et al.* (2003), who evaluated two navel orange varieties (Navelate and Lane Late) in two regions of Chile; in all of the studied orchards, there was a trend of decrease of the titratable acidity as the harvest delay increases and also with storage in a cold chamber. The authors also commented that values of titratable acidity smaller than 0.6 % w/v may indicate that the fruits have had low quality. According to Efrom & Souza (2018), the average titratable acidity for the varieties cited previously for the Rio Grande do Sul state would be about 0.9 % w/v. Schäfer *et al.* (2001) reported titratable acidity values of 0.91 % w/v for Monte Parnaso navel oranges cultivated in the municipality of Butiá, South Brazil, in which the titratable acidity content reported was higher than the highest value observed in the present work (0.61 % w/v).

Efrom & Souza (2018), in a data compilation of several *Citrus* varieties, cited that a titratable acidity of 0.71 % w/v is characteristic of the Monte Parnaso navel orange; value higher than the ones obtained in this work. Ortiz *et al.* (1987) observed that the titratable acidity of the Navelina, Washington, and Navelate oranges reduced gradually, from 0.30 % to 0.07 % w/v as the ripening advanced, which may be related to the degradation of organic acids present in the fruit pulp.

In the state of Rio Grande do Sul, the main productive region of Citrus is the ‘Vale do Caí’ region (Sulzbach *et al.*, 2016), in which the harvest of the Monte Parnaso navel orange occurs earlier than in the Serra Gaúcha region (where this work was conducted). Thus, the lower values of titratable acidity may be a result of a long time of the fruit on the mother plant.

Table 4: Variation of the titratable acidity of the Monte Parnaso navel oranges as a function of the harvest date

Harvest date	Titratable acidity (% w/v)
0	0.61 a
15	0.57 ab
30	0.54 b
45	0.52 b
Coefficient of variation (%)	15.06

Means followed by the same letter have not differed statistically by Tukey’s multiple range test at 5 % probability (α = 0.05).

Table 3: Means relative to the parameters of average fruit mass (AFM), transversal and longitudinal diameters (Ø), peel thickness, juice content, soluble solids content, and SS/TA ratio, which were influenced only by the harvest factor

Harvest	AFM (g)	Transversal Ø (mm)	Longitudinal Ø (mm)	Peel thickness (mm)	Juice content (% v/w)	Soluble solids (°Brix)	SS/TA ratio
2018	417.97 a	96.75 a	92.47 a	5.63 a	42.81 b	9.76 b	17.42 b
2019	339.36 b	87.73 b	87.51 b	5.00 b	47.98 a	10.22 a	19.00 a
CV[1] (%)	12.33	4.40	3.73	10.24	7.35	7.76	14.79

[1] – coefficient of variation.

Teruel *et al.* (2000), studying Baianinha navel oranges [*Citrus sinensis* (L.) Osb.] under room and cold storage conditions for 15 days, reported that the values of titratable acidity of the oranges reduced about 20 % under room storage and about 8 % under cold storage. Theanjumpol *et al.* (2019), reported that, for mandarins, the values of titratable acidity reduced with an increase in the granulation intensity; the authors distributed the mandarins in five classes (A to E), corresponding to the granulation intensities of zero, 1-25 %, 26-50 %, 51-75 %, and 76-100 % granulation, respectively. Class-A fruits (without granulation) had a titratable acidity of 0.45 % w/v, whereas class-E fruits (76-100 % granulation) had 0.22 % w/v. These results diverge from the ones observed in the present work, in which the reduction of the titratable acidity was related to the factor harvest date.

The ascorbic acid content was the only parameter that was influenced by both the harvest and the harvest date, with a significant interaction between the two factors. The results of the factorial analysis of the ascorbic acid content relative to the two factors are presented in Table 5.

It could be seen that, for the 2018 harvest, there was no statistical difference in the ascorbic acid content for the different harvest dates. Comparing the two harvests, it was possible to observe that only occurred statistical difference from 30 days of harvest date, when the ascorbic acid content was smaller in the fruits of the 2019 harvest. Relative to the harvest date for 2019, there was a progressive reduction of the ascorbic acid content, in which the highest value occurred on day zero (457.91 mg·L^{-1}) and the lowest value occurred on day 45 (416.82 mg·L^{-1}), also occurring significant differences on days 15 and 30, as shown in Table 4.

Nascimento & Santos (2013) verified, studying the Tahiti acid lime (*Citrus* × *latifolia* Tan., also known as Persian lime), a reduction in the ascorbic acid content when there was a delay in the harvest. Chitarra & Chitarra (2005) cited that ascorbic acid content tends to decrease as the fruit ripes and with further storage; this may be the result of the direct action of the ascorbinase enzyme, or by the action of other oxidative enzymes, such as peroxidase. However, in the present work, this behavior was only observed in the 2019 harvest since no statistical difference occurred in the ascorbic acid content relative to the harvest date for the 2018 harvest (Table 5).

Ramalho (2005), who studied the Pêra orange of orchards from several locations of São Paulo state (Southeast Brazil), reported average ascorbic acid content ranging from 129.5 to 617.1 mg·L^{-1}. Couto & Canniatti-Brazaca (2010), comparing the ascorbic acid content and the antioxidant activity of several *Citrus* varieties, concluded that there is a large variability in the ascorbic acid content of oranges and mandarins, also occurring important variations in the antioxidant activity. The same authors also commented that the oranges had higher ascorbic acid content and also a higher antioxidant activity than the mandarins; for the mandarins, the ascorbic acid content ranged between 214.7 and 324.7 mg·L^{-1}, the oranges had contents in the range of 625.0-840.3 mg·L^{-1}. It was also observed that considering the same species, the ascorbic acid content varied largely among cultivars; however, the final content of ascorbic acid is also a result of other factors, such as the site of cultivation, edaphoclimatic factors, harvest period and storage, among others (Magwaza *et al.*, 2017). This may explain the observed differences between the data observed in this work and some of the results reported in the literature.

Sharma & Saxena (2004), evaluating Kinnow mandarins grafted on three different rootstocks, observed a reduction of 20.88 % in the ascorbic acid content, comparing normal fruits and ones that presented granulation. In other work, Sharma *et al.* (2006) also compared normal fruits and granulation-affected ones. Among six Citrus groups and twenty different cultivars, the normal fruits presented an ascorbic acid content in the range of 162-462 mg·L^{-1}; the fruits with granulation had contents in the range of 121-429 mg·L^{-1}. These works suggest that an inverse relationship between the presence of granulation and the ascorbic acid content in *Citrus* fruits may exist, however, further research is needed to draw any definitive conclusion.

Relationships between granulation and the quality parameters of the fruits

Granulation and quality parameters data underwent a correlation analysis. The Spearman correlation coefficients (r_s) for granulation relative to the quality parameters and the harvest date are presented in Table 6.

Table 5: Results of ascorbic acid content (mg·L^{-1}) relative to the harvest and the harvest date for Monte Parnaso navel oranges cultivated in Serra Gaúcha region, South Brazil

Harvest	Harvest date			
	0	15	30	45
2018	478.46 Aa	468.92 Aa	471.12 Aa	488.00 Aa
2019	457.91 Aa	451.31 Aab	424.15 Bbc	416.82 Bc

Means followed by the same letter, uppercase in column (factor harvest) and lowercase in line (factor harvest date), have not differed statistically by Tukey's multiple range test at 5 % probability (CV = 6.49 %).

Table 6: Spearman correlation coefficients (r_s) for the granulation parameters of incidence and severity relative to the quality parameters of the fruits and the harvest date

Test/quality parameters	Granulation parameters				
	Incidence	Severity			
		0-25 %	25-50 %	50-75 %	75-100 %
Harvest date	**0.3573**	0.0218	0.0276	0.1083	0.1857
Average fruit mass	**0.5338**	-0.2408	**0.4150**	0.0009	0.3260
Transversal diameter	**0.5702**	-0.3361	**0.4380**	0.0424	**0.4383**
Longitudinal diameter	**0.5262**	-0.2215	**0.4283**	-0.0480	0.3393
Peel thickness	**0.3766**	0.0947	0.0939	-0.1305	**0.3993**
Juice content	**-0.3988**	0.0571	-0.3197	0.0937	-0.2718
Soluble solids content	0.1683	-0.0177	0.0599	0.2420	-0.1892
Ascorbic acid content	**0.6640**	-0.2565	0.2655	**0.3659**	0.1285
Titratable acidity	0.1412	0.0148	0.0028	0.1815	-0.0688
SS/TA ratio	-0.1018	0.0861	-0.1078	-0.0567	-0.0826

Significant correlations ($p < 0.05$) are highlighted in bold.

By analyzing Table 6, some significant correlations were low ($0.30 < r_s < 0.50$) to negligible ($r_s < 0.30$), according to the classification proposed by Agunbiade & Ogunyinka (2013). The correlations between granulation incidence and ascorbic acid content (0.664), with longitudinal (0.526) and transversal (0.570) diameters, and with average fruit mass (0.534) were considered as moderate ($0.50 < r_s < 0.70$). Ortuzar *et al.* (2003) commented that bigger fruits tend to have higher granulation incidence and severity; this may explain the positive correlation between fruit granulation and the biometric parameters (transversal and longitudinal diameters and average fruit mass), although no significant correlation was observed regarding granulation severity in the present work.

Harvest date and peel thickness were also significantly correlated with granulation incidence (0.357 and 0.377, respectively); however, the correlations were low. The juice content was the only parameter negatively correlated with granulation incidence (-0.399) at a low degree; this was expected since granulation causes a reduction of fruit juice due to jellification of the pulp glands (Jie *et al.*, 2021).

Relative to granulation severity, no significant correlation was observed for the severity of 0-25 %. For the range of 25-50 %, the biometric parameters (average fruit mass and transversal and longitudinal diameters) were positively correlated. The correlations were low, with coefficients of 0.415, 0.438, and 0.428 for average fruit mass and transversal and longitudinal diameters, respectively. For the severity range of 50-75 %, only the ascorbic acid content was correlated with granulation incidence at a low level (0.366). For severities in the range of 75-100 %, the parameters of transversal diameter and peel thickness were correlated at a low level (0.438 and 0.399, respectively).

No specific relationship could be established for granulation severity since the correlated parameters for each category varied, and all significant correlations were low ($r_s < 0.5$). However, the parameter of transversal diameter was the only one that was significantly related with two severity categories (25-50 % and 75-100 %), and with granulation incidence.

According to Ortuzar *et al.* (2003) and Mesejo *et al.* (2016), bigger fruits are prone to physiological disorders such as granulation and splitting; in this sense, larger (greater transversal diameter) fruits may tend to develop a more severe granulation than smaller fruits.

CONCLUSION

Granulation incidence and severity were not related to the harvest date; only granulation severity was related to the harvest. The parameters of average fruit mass, longitudinal and transversal diameters, peel thickness, juice content, soluble solids content, and SS/TA ratio were influenced only by the factor harvest not being influenced by the harvest date. The titratable acidity was influenced only by the harvest date, in which there was a decrease in the titratable acidity with a delay in the gravest date. Ascorbic acid content was influenced by both factors, being lower in the 2019 harvest and differing between harvest 30 and 45 days of delay relative to the harvest date. Although some quality parameters were correlated to granulation incidence and severity, most of the correlations were low ($r_s < 0.30$). Correlation incidence was positively correlated with the biometric parameters, peel thickness, and ascorbic acid content; the only negative correlation was between granulation incidence and juice content. No specific trend could be observed for granulation severity since only the transversal diameter had significant correlations with granulation incidence, and two categories of granulation severity. In this sense, larger fruits tend to develop more severe granulation than smaller fruits.

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