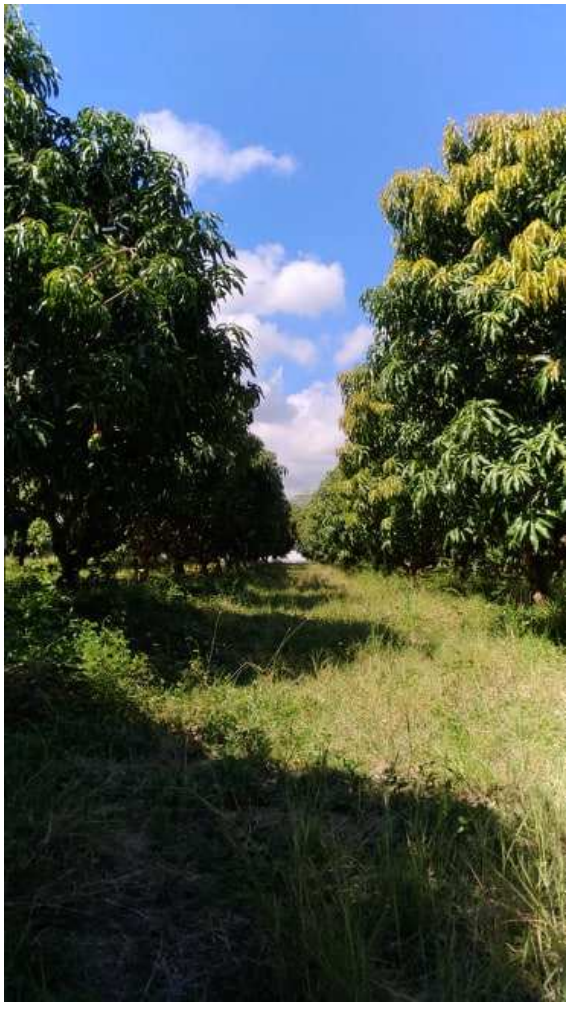
Station Bassin Plat

97455 Saint-Pierre

La Réunion

CHARACTERIZATION OF COGSHALL MANGO QUALITY IN PRE AND POST HARVEST STAGES

Analysis of the effects of growing conditions, maturity stage at harvest and storage conditions on fruits quality



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1. INTRODUCTION

General background

Mango is commonly produced in more than a hundred countries, among which India covers 75% of the total world production. Mango has a great importance for contributing to food security of millions of people of tropical countries, since it is a fruit with high nutritional value (Mitra 2016). Until the 60s, mango was only consumed locally, while in the latest years it's getting more and more importance in US and EU countries. For the moment, 3-4% of the total production is exported, but trends show that the demand of both fresh and processed mango is steeply increasing around the word (Figure 1), having for now US and Netherlands as main importers (Mitra 2016).

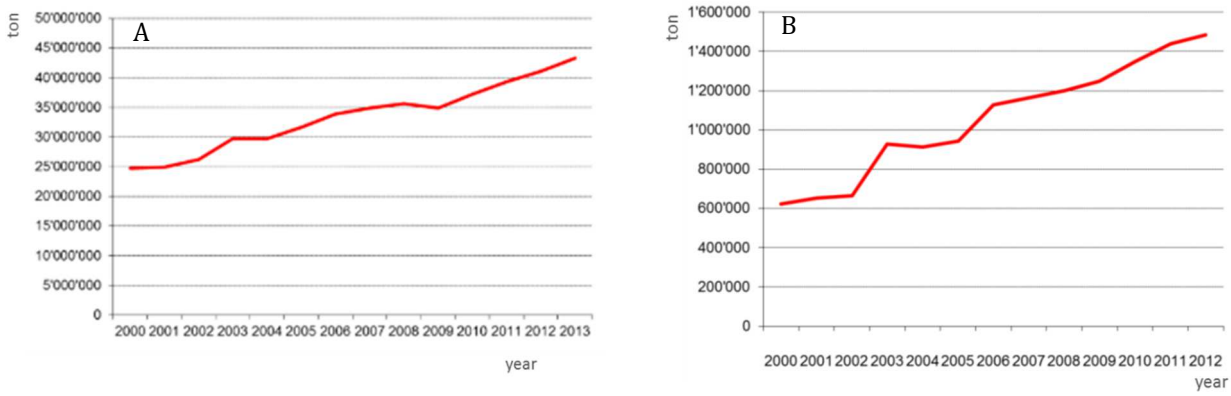


Figure 1 : Trends of (A) mango's world production (tons) from 2000 to 2013 and (B) mango's world exportation (tons) from 2000 to 2012 (Source: United Nations, 2016).

Mango productivity highly varies depending on the area of production. When well managed and irrigated, mango trees can produce around 15-20 t/ha, but eventually in some subtropical cultivars can reach 30 t/ha. However, the greatest part of developing countries does not have irrigations systems, water availability or good management knowledge and their mango productivity varies around 5-8 t/ha (Mitra 2016, Menzel and Le Lagadec 2017). Incomes from mango production for farmers, might be not at the optimal. Furthermore, lot of qualitative requisites obstacle farmers from selling their fruits to supermarkets or for international export.

Lot of different factors, both in pre-harvest and post-harvest, influence final mango quality and must be controlled in order to obtain homogeneous and high quality.

Pre-harvest factors include light availability, temperature, carbon and water availability (Joas et al. 2012). They can be partially controlled by a good orchard management, such as pruning, irrigation or fruit thinning (Léchaudel and Joas 2007). These factors can have a huge influence on both fruit growth and quality, which can be highly heterogeneous within a tree and between trees or orchards. For instance, light and temperature can highly affect fruit transpiration, water and osmotic potentials, which are all highly related to fruit quality (Léchaudel et al. 2013). Carbon availability is a main growing factor that affects fruit growth and quality. Lot of different indicators can be used to define and control carbon availability. In this study we concentrated on Leaf/Fruit ratio at a branch scale, which represents the amount of leaves per fruit in a branch (Joas et al. 2012, Léchaudel et al.

2005). Leaves indeed provides carbohydrate assimilates to fruits through carbon assimilation (photosynthesis). A low Leaf/Fruit ratio results in a lower fruit growth and a lower final quality, including a lower dry matter content (Léchaudel et al. 2005).

Maturity stage at harvest is another key factor that influence the final quality of mango (Joas et al. 2012). Harvesting too green mangos causes a high variability in mangos quality, with some of them that can even not ripen. While, harvesting mangos too late causes a short shelf life and a higher sensibility to diseases (Joas et al. 2012, Léchaudel et al. 2013).

After harvesting, fruits can be either sold directly or stored in cold chambers. Storage conditions, such as temperature and humidity, are post-harvest factors that have a huge impact on the shelf life of a fruit and on its maturation. Fruits are put at different conservation condition depending on if they are intended for export or for local markets.

Fruits are very complex organisms and their growth and quality depends on multiple interconnected factors. Therefore, an integrated approach that considers both pre- and post-harvest, would be suitable to reach a good optimization of mango quality (Léchaudel and Joas 2007, Génard 2007, Lescourret and Génard 2005). As an addition to experiments, models that can represents mango fruit functioning in relation with their surrounding environment, would give a better understanding of mango production and would provide useful information to optimize mango quality while minimizing its variability.

Main goal of the study

This study is part of a PhD project. The main aim of this project is to analyze the dominant factors contributing to mango maturity and quality, and how to control them in order to increase quality. The general objectives of this project are to:

1. Understand how growing conditions (carbohydrate availability) affects final quality of mango and its development on the tree and during storage.
2. Understand how maturity stage at harvest affects final quality of mango and its development during storage.
3. Understand how conservation conditions affect final quality of mango and its development during storage.
4. Develop a process-based model for fruit growth, fruit maturation and sugars evolution, that could give general guidelines on the management of the three previous factors, to help farmers optimizing fruit quality.

In my personal study, I adopted an experimental approach and the obtained experimental data will later on be used to develop the model. The specific objectives of my study were to:

1. Study and compare the effects of two different leaf/fruit ratios (25 and 100) on fruit quality and its development on the tree and during storage. Note that the effect of Leaf/Fruit ratio on fruit quality development on the tree was however not considered in this report.
2. Harvest mango fruits at three different maturity stages (green = G, green-mature = GM and yellow point = YP) to study how it affects mango quality development during storage.
3. Conserve mango fruits in two different temperature conditions (10°C and 20°C), to analyze how it affects mango quality development during storage.

2. MATERIAL AND METHODS

Experiment design and Studied factors

Experimental orchard

Data were collected in an experimental orchard of mango trees (cv. 'Cogshall') grafted onto 'Maison Rouge' rootstock and planted in 2004. The orchard was located in Bassin Plat (CIRAD), St Pierre, Reunion Island (20°52'S, 55°31'E, 125 m a.s.l.). It was managed according to standard commercial practices.

Experimental design

The following factors have been taken into consideration:

1- Fruits growing conditions (leaf/fruit ratio)

Leaf/Fruit ratio (LF) is defined as the number of leaves per fruit applied at a branch scale. Two different Leaf/Fruit ratios (25 and 100) were applied to modify assimilates' supply to the fruits in order to study the effects of these different growing conditions on fruits' size and quality.

Branches were selected depending on the thickness of the diameter (between 1 and 2 cm), on the number of leaves and on the number of the healthy fruits. In order to get the exact leaf/fruit ratio for each branch selected, defoliation and/or fruit thinning have been executed at the beginning of the experiment, but also during the experiment in order to adjust for eventual dropped fruits. Additionally, each selected branch was girdled in order to stop any phloem flow between the branch and the rest of the tree, while maintaining the xylem flow.

Both Leaf/Fruit ratios and branch girdling were applied around 59 days after bloom (DAB).

2- Fruits harvest conditions (maturity stage)

Fruits were harvested at three different maturity stages: green (G), green-mature (GM) and yellow point (YP). These different stages were determined primarily by using a fluorescence measurement, in particular for stages G and GM, where fruits are entirely green (Léchaudel et al. 2010). Additionally, a visual indicator based on the percentage of yellow coloration of the fruit (see Figure 2) helped to determine the maturity stage, mainly starting from the YP stage.

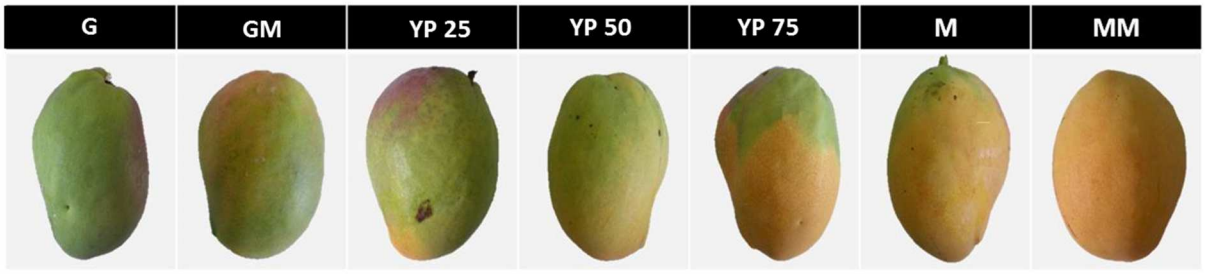


Figure 2: Maturity stages of Cogshall mango fruits and their colors characteristics. G= green, GM= green mature, YP = yellow point (at different percentages), M= mature, MM= over mature.

The fluorescence measurement was executed with a portable fluorimeter PEA (Fluorescence Monitoring System FMS 2, Hansatech) on the fruit surface, near the apex zone where the first changes in peel colors appear. As output, it gives 4 parameters: Fv, Fo, Fm, Fv/Fm. Among them, Fv (variable fluorescence), was used to characterize the maturity stage of fruits (Léchaudel et al. 2010). The maturity stage is established by the amount of chlorophyll present on the skin of the fruit and measured as a response of its fluorescence. Determining a maturity stage with fluorescence is one of the best methods, since it is non-destructive and it can give specific information at a fruit level (Léchaudel et al. 2013, Léchaudel et al. 2010)

3- Fruits' conservation conditions (temperature at storage)

Two conservation conditions were considered for harvested fruits. The first treatment, called T20, consisted in leaving fruits in a chamber at 20°C for 15 days for G and GM and for 5 days for YP. For the second treatment, called T10, fruits were left in a chamber at 10°C for 18 days, then shifted to the chamber at 20°C for 5 days. In both chambers, relative humidity was maintained around 90% during the whole conservation period. The length of conservation has been settled depending on the initial maturity stage, in order to get mature fruits at the end of the treatments. YP fruits, indeed, were conserved for a much shorter time, since they were almost mature. T20 treatment is thought to represent the storage that is usually applied for local markets fruits, while T10 is the storage for exported fruits.

Under the T20 conservation treatment, fruits of all maturity stages (G, GM and YP), have been studied. Whereas, under the T10 conservation treatment, only fruits of maturity stage G and GM have been used. YP fruits have not been put under T10 treatment, since they are already too mature for being exported. In both cases, fruits with 25 and 100 Leaf/Fruit ratios have taken part to the experiment.

Therefore, having 2 treatments for growing conditions (25 and 100), three treatments for maturity stage (G, GM, YP) and 2 treatments for conservation conditions, the combination of all of them gives a total of 10 treatment combinations (Table 1).

Leaf/Fruit ratios	**Maturity stages**	**Conservation Conditions**	
		T20	**T10**
25	**G**	25 – G – T20	25 – G – T10
	GM	25 – GM – T20	25 – GM – T10
	YP	25 – YP – T20	/
100	**G**	100 – G - 20	100 – G - 10
	GM	100 – GM - 20	100 – GM - 10
	YP	100 – YP - 20	/

Table 1: Combination of growth treatments, maturity treatments and conservation treatments.

Sampling and monitoring

As represented in Figure 3, fruits have been harvested in 4 different sets: G, GM, YP for 100 Leaf/Fruit treatment (YP_{100}) and YP for 100 Leaf/Fruit treatment (YP_{25}). YP fruits have been harvested in two different moments, since they reached YP maturity on different dates: 100 Leaf/Fruit on January 9th, and 25 Leaf/Fruit on January 14th, of 2019. The dates of field harvest representing those maturity stages have been approximately estimated by knowing the blooming date and by looking at previous studies that formulated an evolution trend of fruit maturity over the number of days after blooming (DAB). Additionally, these dates were adjusted by measurements of fluorescence and visual estimation of maturity stages. The day of full bloom was on September 1st, 2018.

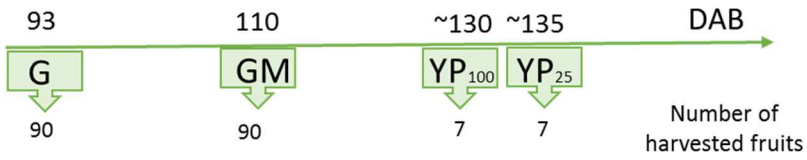


Figure 3: Days after bloom (DAB) when each maturity stage has been harvested, and number of fruits harvested for each date.

Fruits that were harvested at stages G and GM included 45 fruits from 100 Leaf/Fruit and 45 fruits from 25 Leaf/Fruit treatments and subsequently they have been randomly divided in 3 subsamples (Figure 4).

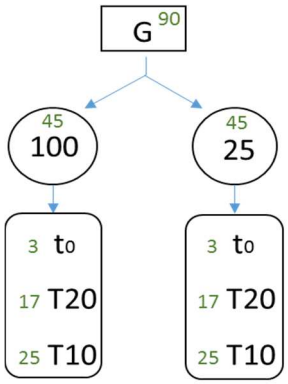


Figure 4 : Representation of fruits division in subsamples. Valid for both G and GM fruits.

As schematized in figure 5, fruits have been sampled and analyzed in different moments during their conservation treatments:

- **t0** harvest: fruits from G, GM and YP stages have been analyzed right after being harvested in order to study the starting point of fruits before putting them under the conservation treatments.
- **ti** harvest: fruit sampling and analysis have been executed at 1 (for YP) and 4 (for G and GM) dates in T20 and 6 (only for G and GM) dates in T10 during the conservation treatments, in order to study the kinetic of fruit's quality and maturity.

For each time point, three fruits have been analyzed. For T10 fruits, t6 is considered the final time point (tf), and for T20 fruits it is t4 for G and GM fruits and t1 for YP fruits.

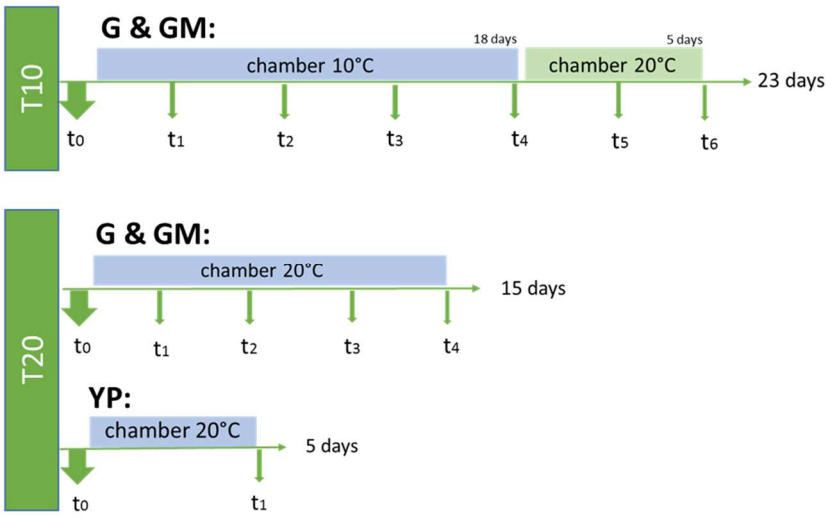


Figure 5: Sampling and analysis plan for T20 and T10 conservation treatments. T20 treatment lasted 15 days for G and GM and 5 days for YP. T10 treatment lasted 23 days in total and has been applied to fruits G and GM only.

Measured variables

Multiple variables were measured during the analysis in order to have a complete characterization of fruits quality and maturity. These variables are fresh mass of the fruit, peel and seed, fresh mass loss of the fruit, fruit transpiration, fruit dimensions, fruit fluorescence, color of the pulp, fruit emission and fruit internal content in gas (CO_2 and C_2H_4), and osmotic and water potentials, dry matter content, titratable acidity and brix degree of the pulp. A complete scheme of destruction and analysis steps of fruits coming from the field or the storage chambers is given in Appendix A. A table resuming the studied variables, their meanings and how they were obtained is given in Appendix B.

Evaluation of sampled fruits

Before to start the analysis of the results, three different hypotheses where tested on the variables FM (fruit fresh mass) and Fv (variable fluorescence) for:

- Homogeneity of fruits between conservation treatments T10 and T20
- Heterogeneity of fruits between Leaf/Fruit ratios 100 and 25
- Heterogeneity of fruits between Maturity Stages G, GM and YP

All these tests were executed with ANOVA, and additionally, for the heterogeneity tests, also interactions between Leaf/Fruit ratios and Maturity stages were checked.

The hypothesis of homogeneity was tested on sampled fruits that were harvested but not put yet under their assigned conservation treatments, in order to check if fruits were homogeneously divided between the two treatments. This is important to verify, in order to be sure that any significant difference in the final results (after the conservation treatments) is representing of the effect of the temperature, and not of an eventual initial experimental effect.

The hypothesis of heterogeneity was tested on the same fruits, to check whether there is an initial effect of Leaf/Fruit ratio and of Maturity stage on fruits, without taking in to account the temperature effect. Furthermore, it was investigated if these two factors are interconnected, since from a biological point of view, this can be possible.

Homogeneity between T10 and T20

In the following graphs, samples of both 25 (Figure 6) and 100 (Figure 7) Leaf/Fruit ratios are represented and analyzed to prove for homogeneity. ANOVA test was executed on G and GM fruits only. YP only has T20, therefore, no statistical analysis has been executed on YP fruits. Results indicate that fruits of both maturity stage G and GM, both for FM and Fv, are not statistically different, therefore, homogeneity between samples of the two conservation treatment sets has been proven.

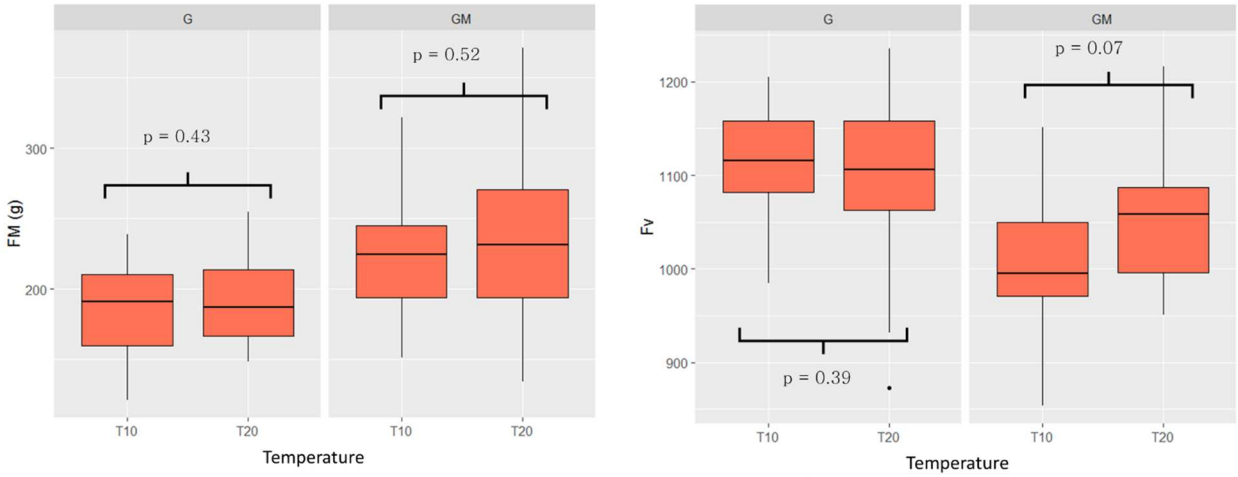


Figure 6: Boxplot graphs representing Fresh Mass (FM) and variable fluorescence (Fv) values of 25 Leaf/Fruit samples of fruits on the day of their harvest, distinguished by maturity stage and conservation treatment. In these two graphs, only fruits that have been lately analyzed, were taken into account. P-values of the ANOVA tests comparing T10 and T20 sets of fruits are indicated.

Also for samples of 100 Leaf/Fruit, the ANOVA test proved for homogeneity for FM variable, while for Fv, only GM fruits were homogeneous. In G fruits, a slightly significant difference is present.

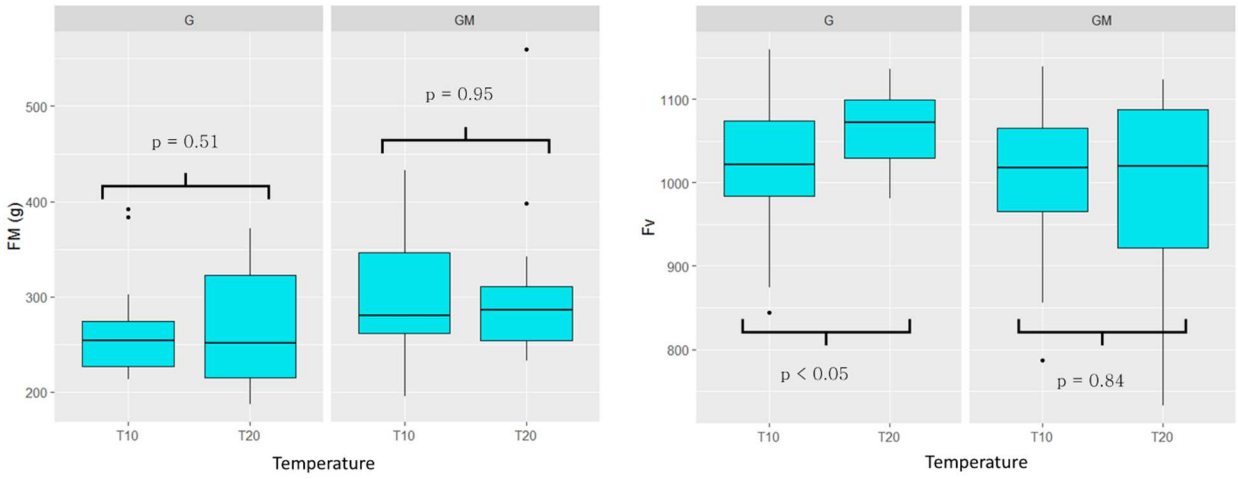


Figure 7 Boxplot graphs representing Fresh Mass (FM) and variable fluorescence (Fv) values of 100 Leaf/Fruit samples of fruits on the day of their harvest, distinguished by maturity stage and conservation treatment. In these two graphs, only fruits that have been lately analyzed, were taken into account. P-values of the ANOVA tests comparing T10 and T20 sets of fruits are indicated.

Heterogeneity

By doing an ANOVA test, it resulted that samples are heterogeneous for both Leaf/Fruit ratio and Maturity Stage treatments, both in fresh mass (FM) and in fluorescence (Fv) (Figure 8).

Looking at the trends in FM, 100 Leaf/Fruit fruits have a higher fresh mass than 25 Leaf/Fruit fruits. On the contrary, for Fv, 100 Leaf/Fruit fruits have a lower value than the 25 Leaf/Fruit (lower amount of chlorophyll), which indicates that they are more mature. This difference between 100 and 25 Leaf/Fruit was mostly observed for G and YP fruits. In conclusion, 100 Leaf/Fruit fruits have a higher FM and are more mature then 25 Leaf/Fruit.

Looking at Maturity stages, it is possible to detect a difference among them. For FM, YP fruits have a higher value than GM and G fruits and the difference is more accentuated for 100 Leaf/Fruit. For Fv, G fruits have a higher value than G fruits and YP fruits. In conclusion, YP fruits are bigger and more mature than G and GM fruits, reflecting for expected differences in the Maturity stage.

Tests for interactions, both in FM and Fv, resulted significantly different. This means that the two factors are dependent one from the other and that work together, having a combined effect on the results. Indeed, for FM, its relationship with Maturity stage varies between LF25 and LF100 (the increase of FM with maturity stage is higher for LF100 than LF25) and this is the interaction between Maturity and LF. In addition, the interaction between Maturity and LF for Fv could be explained by the fact that it seems that Fv is lower for LF100 than LF25 only for G and YP, whereas Fv seems to be not different between LF25 and LF100 for GM. This has to be taken in to consideration over the whole statistical analysis, because it will not allow a clear explanation of each individual factor, since they are interconnected.

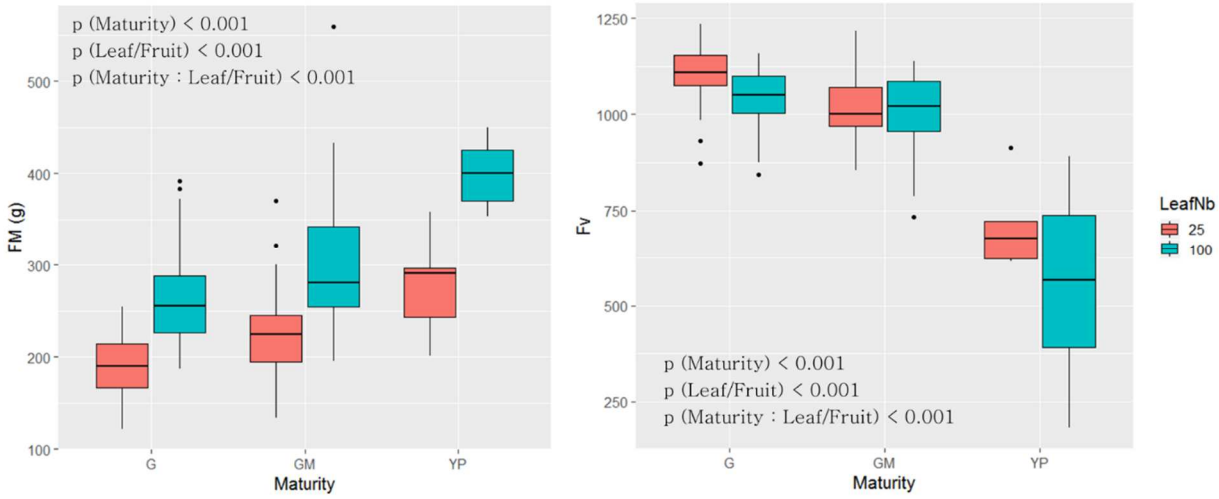


Figure 8: Boxplot graphs representing Fresh Mass (FM) and variable fluorescence (Fv) values of fruits on the day of their harvest, distinguished by maturity stage and Leaf/Fruit ratio treatment. In these two graphs, all harvested fruits have been taken in to account. P-values of the ANOVA tests comparing Maturity stages, Leaf/Fruit ratios and testing the two factors interaction are indicated.

Statistical analysis

Individual effect of pre- and post-harvest factors was evaluated on all the analyzed variables using ANOVA tests. In particular, ANOVA tests were executed to:

a) Test for each time point (t0, ti, tf), within a specific maturity stage and a specific conservation treatment, an effect of Leaf/Fruit ratio on the studied variable.

b) Compare each final time point (tf), within a specific maturity stage and a specific Leaf/Fruit ratio, and check for an effect of conservation treatment on the studied variable. This analysis was executed only for maturity stages G and GM because only T20 treatment was applied on stage YP.

c) Compare each initial (t0) and final (tf) time point, within a specific conservation treatment and a specific Leaf/Fruit ratio, and check for an effect of maturity stage on the studied variable. Furthermore, when an effect was observed, a test of multi-comparison (Tukey HSD) was executed among maturity stages to check which ones were different. This analysis was executed by comparing G and GM fruits at T10 (for tf points only), and G, GM and YP fruits at T20 (both for t0 and tf points).

All statistical analysis was executed with R software version 3.4.2 (2017-09-28) (R Core Team 2017). Multi-comparison tests were performed using function from the multcomp R package (Hothorn et al. 2008). Data have been represented in graphs using functions from the lattice (Sarkar and Deepayan 2008) and ggplot2 (H. Wickham 2016) R packages.

In the report, results were presented only for some measured variables, considered as important indicator fruit quality. These variables are fresh mass loss of the fruit, internal content of the fruit in ethylene (C_2H_4) and dry matter content, titratable acidity, brix degree and color (Hue angle) of the pulp. Full results of the ANOVA tests described in part b) and c) are reported in Appendix C.

2. RESULTS

Fresh Mass Loss

a) Analysis of Leaf/Fruit ratios effect, for each time point

From the ANOVA results, it is shown that Leaf/Fruits treatments might have an effect on G fruits, for both T10 and T20 sets, reporting a very higher Fresh Mass Loss (FML, %) for 100 Leaf/Fruit (Figure 9). On fruits GM and YP, Leaf/Fruit treatments did not show a significant effect at T20. Only a weak effect was observed on GM fruits at T10, with a slightly higher FML for 25 Leaf/Fruit.

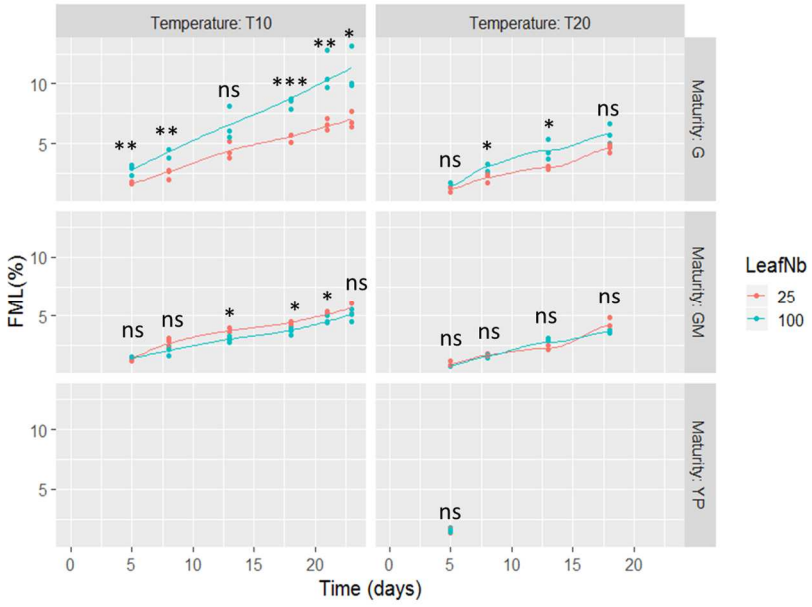


Figure 9: dynamic of Fresh Mass loss (%) over time during storage, and differentiated by Maturity stage, Temperature of Conservation Conditions and Leaf/Fruit Ratio. P-values of the ANOVA tests comparing Leaf/Fruit ratios at each time point are indicated (ns: P≥0.05, *:P<0.05, **: P<0.01, ***: P<0.001).

b) Analysis of Conservation conditions effect, for final time points

Conservation treatments resulted to effect both G and GM fruits, at both Leaf/Fruit ratios (Table 1 in Appendix C). As an overall, FML is higher in fruits storage at T10. Furthermore, looking at the dynamic of FML over time, it is possible to see that the more the fruits stay in storage, the more their loss is higher.

c) Analysis of Maturity stages effect, for final time points

Note that t0 time points haven't been analyzed, since applying the formula for FML, the result would have been zero.

tf: At the final time points, Maturity stages showed an effect on fruits of all Maturity stage and Leaf/Fruit ratios combinations beside the ones at T20 with 25 Leaf/Fruit (Table 2 in Appendix C). As an overall, G fruits have a higher FML than GM and YP fruits.

Dry Matter Content

a) Analysis of Leaf/Fruit ratios effect, for each time point

Results of the ANOVA with its significant time points are represented in Figure 10. From the results, it is shown that Leaf/Fruit treatments might have an effect on G fruits, reporting a significantly higher dry matter content for 100 Leaf/Fruits for the mayor part of time points. The same result is clearly reported for fruits YP. On GM fruits, even if the same trend is also observed, Leaf/Fruit treatments had almost no significant effect on dry matter content.

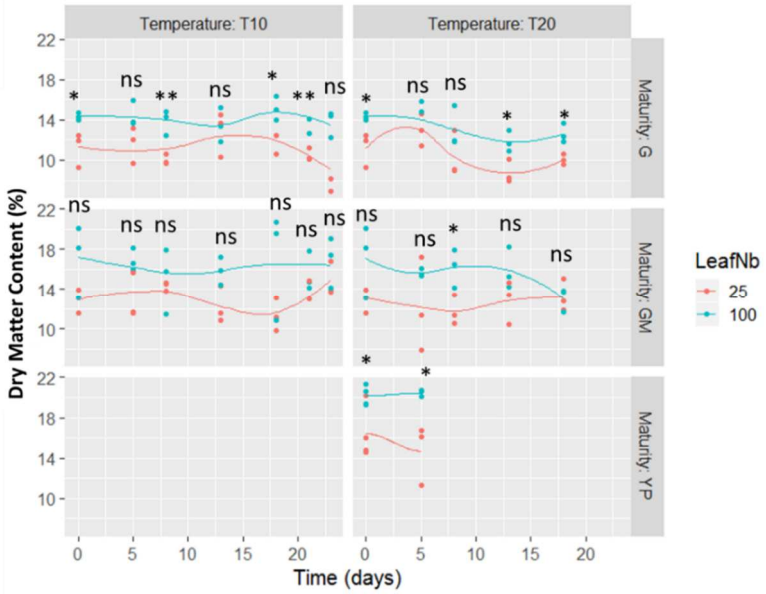


Figure 10: Dynamic of Dry Matter Content over time during storage, and differentiated by Maturity stage, Temperature of Conservation Conditions and Leaf/Fruit Ratio. P-values of the ANOVA tests comparing Leaf/Fruit ratios at each time point are indicated (ns: P≥0.05, *:P<0.05, **: P<0.01, ***: P<0.001).

b) Analysis of Conservation conditions effect, for final time points

Conservation condition treatments did not show significant effects on DMC (Table 3 in Appendix C).

c) Analysis of Maturity stages effect of initial (t0) and final(t_f) time points

t0: Maturity stage treatments resulted to have an effect on all fruits at T20. At 25 Leaf/Fruit ratio, YP fruits resulted to have 20% of DMC, while G fruits only 14%. At 100 Leaf/Fruit, YP fruits are composed of approximatively 16%, while G fruits of 11%. In conclusion, YP fruits have a higher DMC percentage then G fruits (Table 4 in Appendix C).

tf: At the final time point, both for T10 and T20, maturity stages resulted to have a significant effect only on fruits at 25 Leaf/Fruit: GM fruits resulted in having a higher final dry matter content than G fruits (Table 4 in Appendix C).

Hue Angle

a) Analysis of Leaf/Fruit ratios effect, for each time point

Results of the ANOVA with its significant time points are represented in Figure 11. From the results, it is shown that Leaf/Fruit treatments have not any significant effects on fruit color.

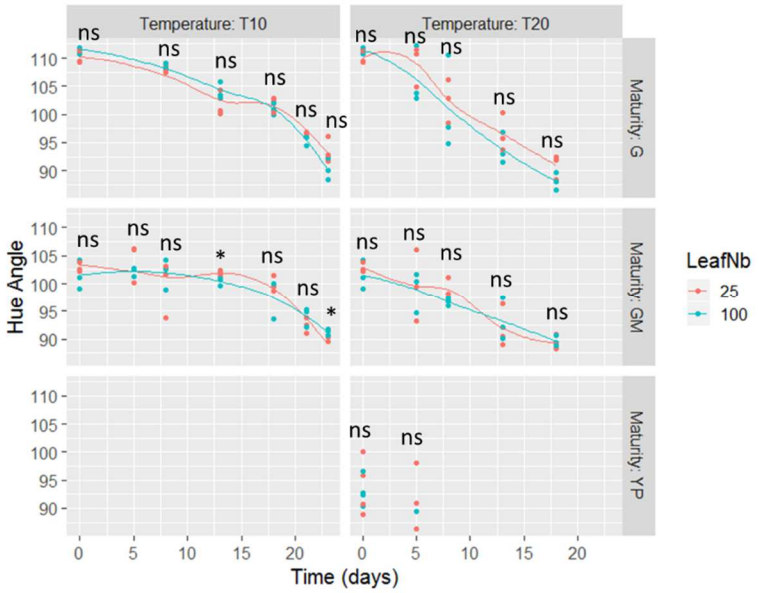


Figure 11: Dynamic of Hue Angle over time during storage, and differentiated by Maturity stage, Temperature of Conservation Conditions and Leaf/Fruit Ratio. P-values of the ANOVA tests comparing Leaf/Fruit ratios at each time point are indicated (ns: P≥0.05, *:P<0.05, **: P<0.01, ***: P<0.001).

b) Analysis of Conservation conditions effect, for final time points

Conservation condition treatments resulted to have no significant effects on fruit color (Table 5 in Appendix C).

c) Analysis of Maturity stages effect of initial (t0) and final(t_f) time points

t0: Maturity stage have a significant effect on Hue Angle both at 25 and 100 LF. At LF25, the Hue Angle value was significantly lower for YP fruits than for G and GM fruits. At LF100, the Hue Angle was significantly different between YP, GM and G fruits, with YP fruits having the lower values, followed by GM fruits and G fruits having the highest values (Table 6 in Appendix C).

tf: Hue Angle reported a significant effect on fruits at 25 Leaf/Fruit and at T10: G fruits reported a higher Hue Angle then GM fruits, reflecting for a whiter color for G fruits while a more yellow color in GM fruits (Table 6 in Appendix C).

Titratable acidity

a) Analysis of Leaf/Fruit ratios effect, for each time point

Results of the ANOVA with its significant time points are represented in Figure 12. Leaf/Fruit treatments showed to not have any significant effect on Titratable acidity.

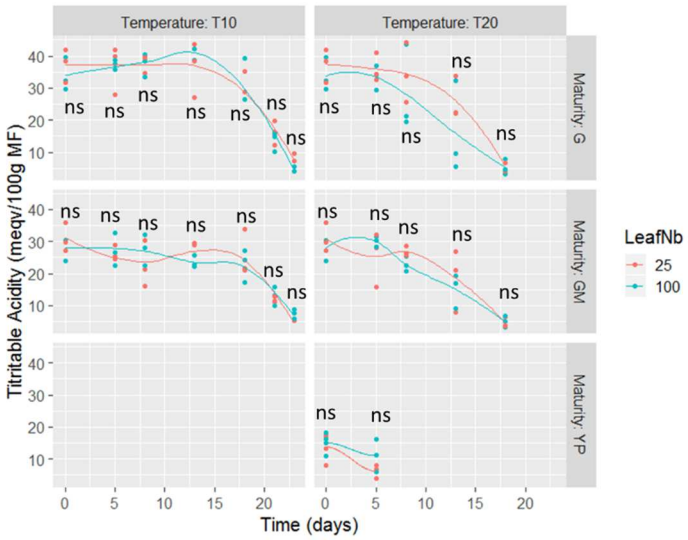


Figure 12: Dynamic of TA over time during storage, and differentiated by Maturity stage, Temperature of Conservation Conditions and Leaf/Fruit Ratio. P-values of the ANOVA tests comparing Leaf/Fruit ratios at each time point are indicated (ns: P≥0.05, *:P<0.05, **: P<0.01, ***: P<0.001).

b) Analysis of Conservation conditions effect, for final time points

Conservation condition treatments reported no significant effects on Titratable Acidity (Table 7 in Appendix C).

c) Analysis of Maturity stages effect of initial (t0) and final(t_f) time points

t0: Maturity stage treatments resulted to have significant effects on all fruits. Results showed a much higher TA in G and GM fruits than in YP fruits, reflecting therefore a higher acidity in less mature fruits (Table 8 in Appendix C).
tf: Only for fruits at 25 Leaf/Fruit and at T10, Maturity stages treatments reported significant effects: TA in G fruits resulted the double of TA in GM fruits (Table 8 in Appendix C).

Brix degree

a) Analysis of Leaf/Fruit ratios effect, for each time point

Results of the ANOVA with its significant time points are represented in Figure 13. Leaf/Fruit treatments resulted to have a significant effect on G fruits, showing a higher °Brix value, therefore a higher sugar content, at 100 Leaf/Fruit ratio.

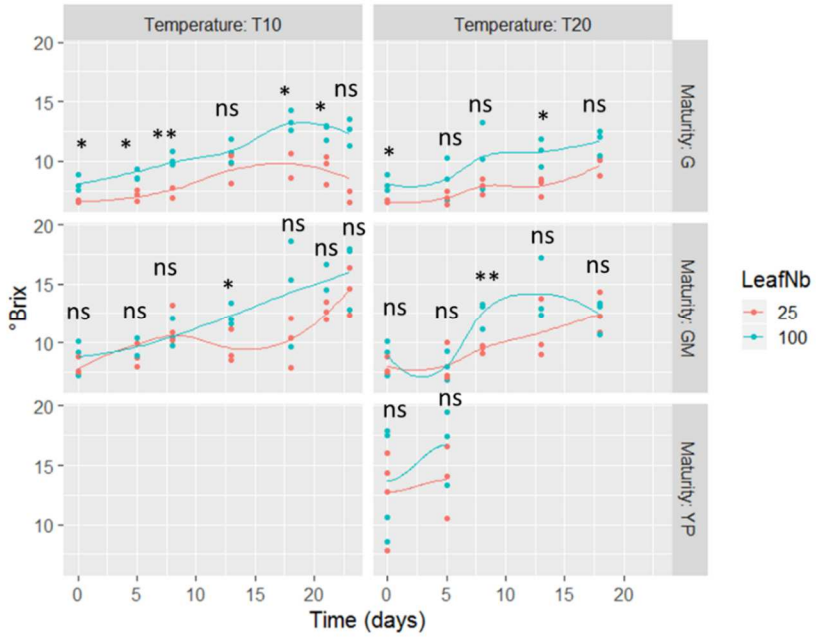


Figure 13: Dynamic of °Brix over time during storage, and differentiated by Maturity stage, Temperature of Conservation Conditions and Leaf/Fruit Ratio. P-values of the ANOVA tests comparing Leaf/Fruit ratios at each time point are indicated (ns: P≥0.05, *:P<0.05, **: P<0.01, ***: P<0.001).

b) Analysis of Conservation conditions effect, for final time points

Conservation condition treatments reported no significant effects on sugar content of fruits (Table 9 in Appendix C).

c) Analysis of Maturity stages effect of initial (t0) and final(t_f) time points

t0: Only at 25 Leaf/Fruit, Maturity stage treatments resulted to have a significant effect on °Brix. The results showed that YP fruits had a higher °Brix than G fruits, reflecting for a higher content in sugars in more mature fruits (Table 10 in Appendix C).

tf: Only fruits at 25 Leaf/Fruit fruits and at T10, showed a significant difference among Maturity Stages: GM fruits reported a °Brix mean value that doubles the one of G fruits, reflecting a higher sugar content in GM fruits (Table 10 in Appendix C).

C_2H_4

a) Analysis of Leaf/Fruit ratios effect, for each time point

Results of the ANOVA with its significant time points are represented in Figure 14. Leaf/Fruit treatments showed to have not much significant effect on ethylene content.

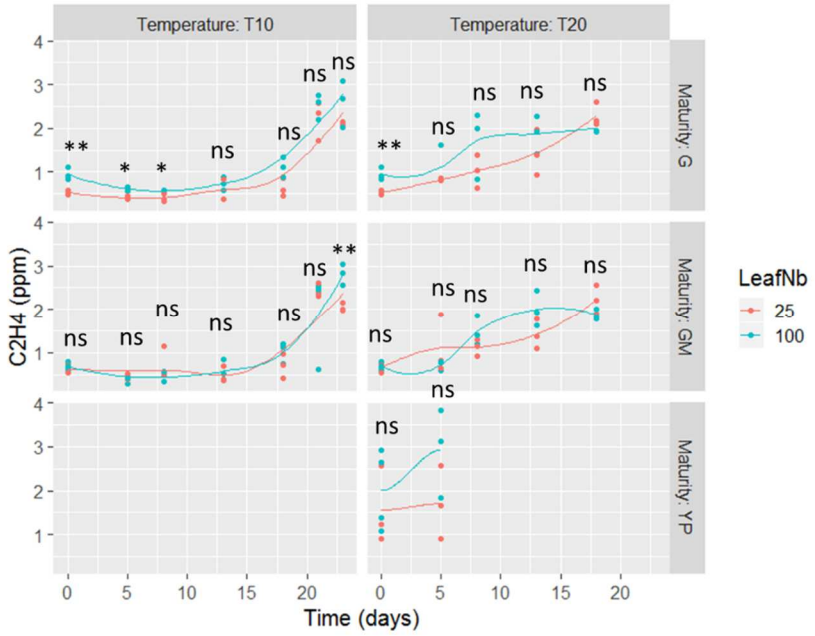


Figure 14: Dynamic of C2H4(ppm) over time during storage, and differentiated by Maturity stage, Temperature of Conservation Conditions and Leaf/Fruit Ratio. P-values of the ANOVA tests comparing Leaf/Fruit ratios at each time point are indicated (ns: P≥0.05, *:P<0.05, **: P<0.01, ***: P<0.001).

b) Analysis of Conservation conditions effect, for final time points

Conservation condition treatments resulted to have a significant effect on GM fruits only, at 100 Leaf/Fruit ratio, showing a higher concentration of ethylene for fruits storage at T10 (Table 11 in Appendix C).

c) Analysis of Maturity stages effect of initial (t0) and final(t_f) time points

t0: On fruits at both 25 and 100 Leaf/Fruit ratios, Maturity stage treatments resulted to have a significant effect. At 25 Leaf/Fruit, YP fruits showed a higher ethylene concentration than G fruits. At 100 Leaf/Fruit, YP fruits showed a higher ethylene concentration than GM fruits (Table 12 in Appendix C).

tf: Maturity stage treatments did not show any significant effects on ethylene concentration (Table 12 in Appendix C).

3. DISCUSSION

Limits of the project

Decisions, materials availability and methods used could have had an impact on the reported results. Harvesting homogeneous fruits at the same maturity stage was difficult. For G fruits there was a higher range among which they could be chosen, but at the moment of harvesting YP fruits, there were very few fruits left on trees, due to environmental causes, previous harvests and limited amount of initial fruits. The harvested YP fruits resulted indeed quite variable for size and maturity, resulting in a high fruit variability within treatments. In the statistical analysis, significance was quite difficult to obtain since for each time point only three fruits per sampling date were analyzed. Harvesting more fruits would have been an excessive work load and available fruits were limited. Since the main project was focused on developing a model to study the dynamics of the variables, it was decided to harvest less fruits on several sampling dates, rather than more fruits on fewer sampling dates.

Fruit quality and factors effects

Fruit quality is a combination of several chemical and physical characteristics that are controlled by very complex processes that depends both on pre- and post-harvest factors (Génard et al. 2007, Joas et al. 2009). In this study only a few variables that compose or contribute to fruit quality have been studied. FML and DMC are two important factors that reflects characteristics for its quality. A high DMC is better since sugars are less diluted and the flavor results better. In this study, results showed that both FML and DMC were higher for 100 Leaf/Fruit (only for G fruits in FML and for G and YP fruits in case of DMC). This result reflects indeed that at a higher Leaf/Fruit, there is a higher Carbon availability due to a higher foliar surface for photosynthesis. This result is in accordance with our expectations and is similarly reported by Léchaudel et al. (2005). Our results also reported that Maturity stage at harvest has an effect on both FML and DMC. For FML, G fruits reported a higher % than GM fruits. DMC was higher for late harvested fruits (YP>GM>G), both before and after storage, even though after storage only for 25 Leaf/Fruit. In general, these results respect our expectations, since later harvested fruits had more time to accumulate dry matter than early harvested (Nordey et al. 2016, Lechaudel and Joas 2006). However, late harvest is better for local markets only, since a higher DMC reflects in a shorter shelf life (Lechaudel and Joas 2006).

Hue Angle represents the color of the pulp, which changes from green/yellow to yellow/orange during maturation. Color resulted to be not very much affected by Leaf/Fruit ratios, however Maturity stages at harvest had an effect on it. Usually the color starts changing after GM stage, and usually no particular differences are encountered between G and GM fruits (Nordey et al. 2014), as we observed in the results. At t0, indeed, there was a significant difference only between YP and GM, for 25 Leaf/Fruit. However, at 100 Leaf/Fruit, G and GM resulted to have a slight difference, but mainly YP was differentiating from them, having an intense yellow color.

TA is an indicator of fruit acidity and together with sugars, highly influence fruit flavor (Lechaudel and Joas 2006). It resulted to be not much affected by Lead/Fruit ratios, while much more from the maturity stage. Our results showed that at t0 there was not a great difference between G and GM

fruits, while at YP acidity was much lower, in accordance with previous studies (Nordey et al. 2014, Lechaudel and Joas 2006). Decrease in TA seems to depend on decrease in citric acid, which is the predominant acid in Cogshall mango (Lechaudel and Joas 2006).

Brix degree represents the amount of soluble compounds present in a fruit, but it is mainly representing for sugars. Sugars are the key factor for fruit quality and flavor (Léchaudel and Joas 2006). From our results it seems that higher Leaf/Fruit ratios, only for G fruits, contribute to higher sugar content. Indeed, at 100 Leaf/Fruit, there is more carbon assimilates available for sugar synthesis, and at 25 Leaf/Fruit, fruits have a delay in sucrose formation (Léchaudel and Joas 2006). Furthermore, sugars are mainly synthetized in later stages and therefore present in higher concentrations in late harvested fruits (Nordey et al. 2016, Lechaudel and Joas 2006). As reported, the results showed that YP fruits had a higher sugar content than, GM, and GM fruits a higher sugar content than G fruits, both at t0 and at tf, but only at 25 Leaf/Fruit ratios.

As last, ethylene production is a fundamental indicator for fruit ripening and it indeed evidently increase during the climacteric phase (Nordey et al. 2016). It is also brought evident that changes in fruit gas content associated with ripening are modulated by pre and post-harvest factors, influencing respiration rate and ethylene production by modifying its metabolism or/and changing fruit resistance to gas diffusion (Nordey et al. 2016). Among these factors, maturity stage at harvest can influence ethylene concentration: Late harvested fruits have a higher ethylene concentration in comparison to early harvested fruits. Also Leaf/Fruit ratios seem to influence ethylene, in particular, fruits with a low Leaf/Fruit ratios report a delayed production of ethylene respect to fruits with high Leaf/Fruit ratios. (Léchaudel and Joas 2006). However, in our results, only maturity stage reported a significant effect on ethylene concentration, being higher in YP fruits. Also storage conditions are included among the influencing factors and lower conservation temperatures seems to delay ethylene production (Nordey et al. 2016), but it did not result in our study.

4. CONCLUSIONS

This study tries to represent some of the effects that pre and post-harvest factors can have on fruit quality. Results shows that maturity stage at harvest is the main factor influencing for fruit quality, reporting an effect on all studied variables that were in accordance with previous researches. Also Leaf/Fruit treatments reported an effect on fruit quality, even though not on all studied variables. In contrary, conservation treatments did not show clear effects on fruit quality. The obtained results might be influenced by the previously mentioned experimental limits, however these results can be considered a good starting point for future researches and a good database for model developing. Since this particular study focused only on the effect of each individual factor, it would be interesting as a next step, to analyze eventual interactions among factors and to interpret them in order to have a deeper understanding and a better explanation of their effects on fruit quality. In addition, deeper analysis should be executed on Leaf/Fruit treatments and Conservation conditions to get clearer explanations of their effects on fruit quality. Lastly, including more variables characterizing for fruit quality, could give a completer idea of fruit functioning and the complex relations that occurs inside it.

In conclusions, all the reported data will be a useful material for developing a model that will allow an integrated approach for fruit quality analysis, combining all pre and post-harvest factors that influence fruit quality and development. Knowing and understanding better their functioning is essential to lately adopt cultural practices that could provide high quality fruits.

5. BIBLIOGRAPHY

Literature references

Génard et al. "Towards a virtual fruit focusing on quality: modelling features and potential uses." Journal of Experimental Botany 58.5 (2007): 917-928.

H. Wickham. (2016) ggplot2: Elegant Graphics for Data Analysis. Springer-Verlag New York.

Joas et al. "Physiological age at harvest regulates the variability in postharvest ripening, sensory and nutritional characteristics of mango (*Mangifera indica* L.) cv. Coghshall due to growing conditions." *Journal of the Science of Food and Agriculture* 92.6 (2012): 1282-1290.

Joas Jacques, Yanis Caro, and Mathieu Lechaudel. "Comparison of postharvest changes in mango (cv Cogshall) using a Ripening class index (Rci) for different carbon supplies and harvest dates." Postharvest biology and technology 54.1 (2009): 25-31.

Léchaudel and Joas. "An overview of pre-harvest factors influencing mango fruit growth, quality and postharvest behavior." Brazilian Journal of Plant Physiology 19.4 (2007): 287-298.

Léchaudel and Joas. "Quality and maturation of mango fruits of cv. Cogshall in relation to harvest date and carbon supply." Australian Journal of Agricultural Research 57.4 (2006): 419-426.

Lechaudel et al. "An analysis of elastic and plastic fruit growth of mango in response to various assimilate supplies." Tree Physiology 27.2 (2007): 219-230.

Léchaudel et al. "Leaf: fruit ratio and irrigation supply affect seasonal changes in minerals, organic acids and sugars of mango fruit." Journal of the Science of Food and Agriculture 85.2 (2005): 251-260.

Léchaudel et al. "Modeling effects of weather and source–sink relationships on mango fruit growth." Tree Physiology 25.5 (2005): 583-597.

Léchaudel et al. "Response of the physiological parameters of mango fruit (transpiration, water relations and antioxidant system) to its light and temperature environment." Journal of plant physiology 170.6 (2013): 567-576.

Lechaudel, Laurent, and Joas. "Chlorophyll fluorescence, a nondestructive method to assess maturity of mango fruits (Cv. 'Cogshall') without growth conditions bias." Journal of agricultural and food chemistry 58.13 (2010): 7532-7538.

Lescourret, and Génard. "A virtual peach fruit model simulating changes in fruit quality during the final stage of fruit growth." Tree physiology 25.10 (2005): 1303-1315.

Menzel, Christopher M., and M. D. Le Lagadec. "Can the productivity of mango orchards be increased by using high-density plantings?" Scientia horticulturae 219 (2017): 222-263.

Mitra, S. K. "Mango production in the world–present situation and future prospect." XXIX International Horticultural Congress on Horticulture: Sustaining Lives, Livelihoods and Landscapes (IHC2014): IV 1111. 2014.

Nordey et al. "Factors affecting ethylene and carbon dioxide concentrations during ripening: Incidence on final dry matter, total soluble solids content and acidity of mango fruit." Journal of plant physiology 196 (2016): 70-78.

Nordey et al. "Spatial and temporal variations in mango color, acidity, and sweetness in relation to temperature and ethylene gradients within the fruit." Journal of plant physiology 171.17 (2014): 1555-1563.

R Core Team (2017). R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. URL https://www.R-project.org/.

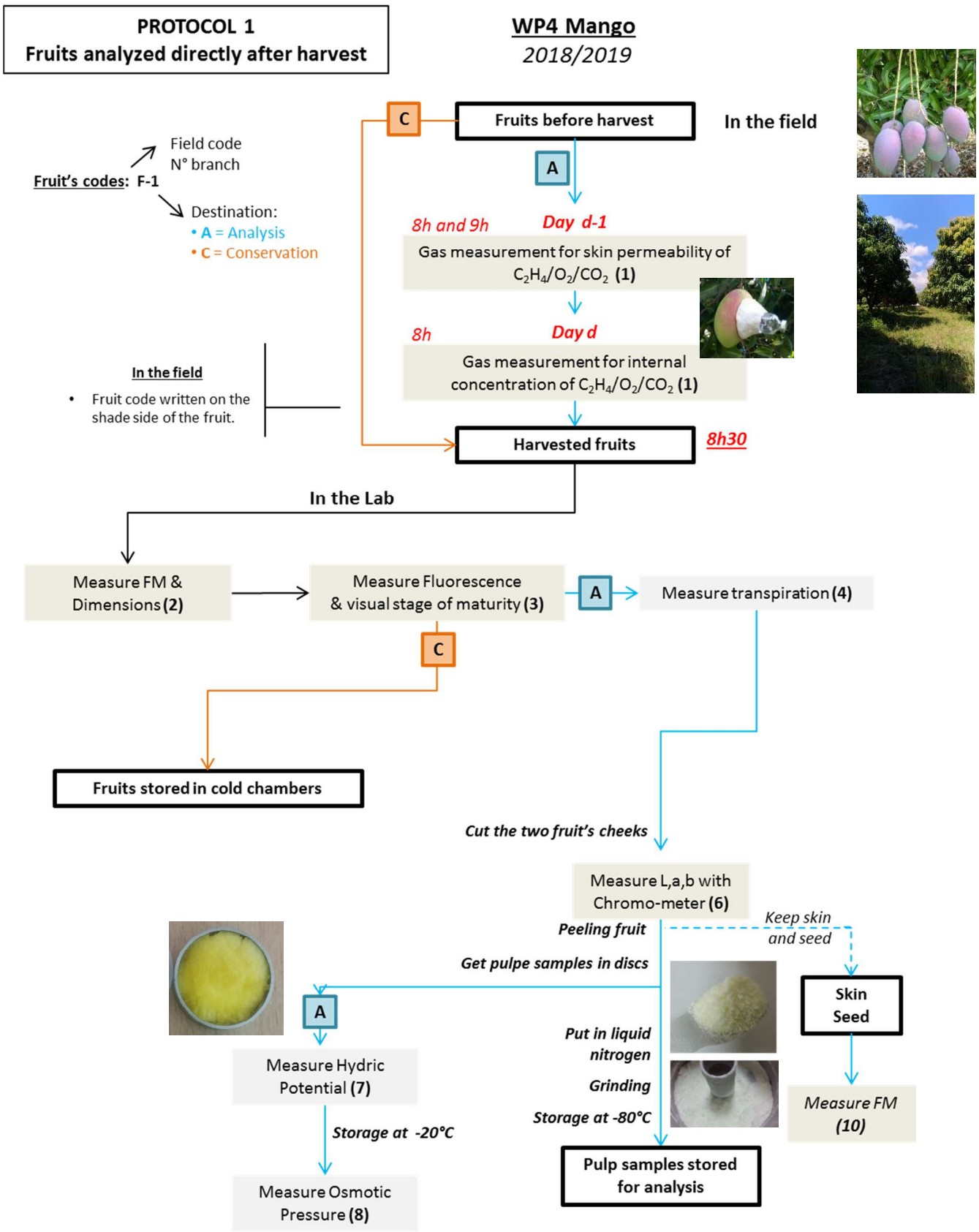
Sarkar, Deepayan (2008) Lattice: Multivariate Data Visualization with R. Springer, New York. ISBN 9 78-0-387-75968-5.

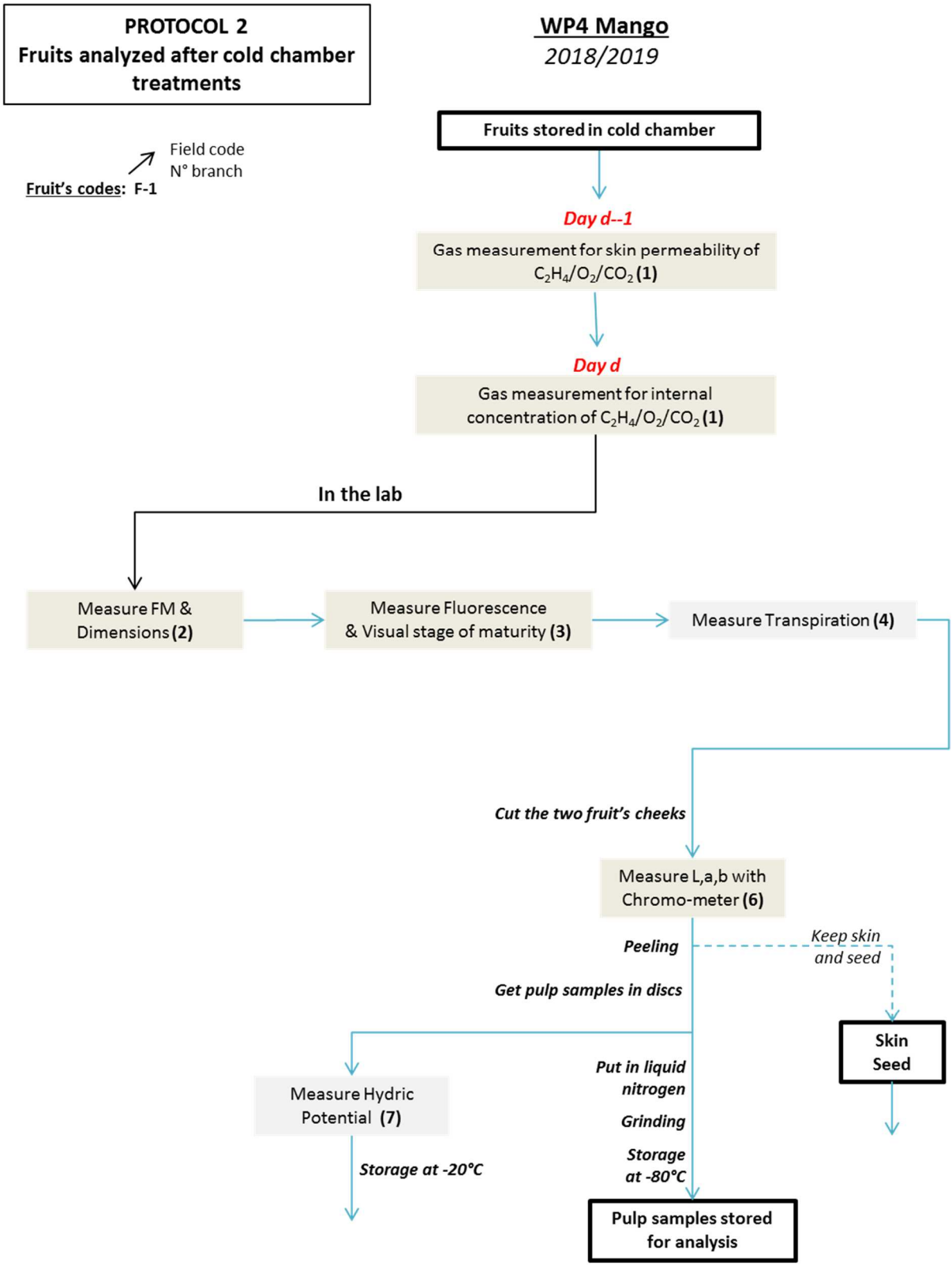
Torsten Hothorn, Frank Bretz and Peter Westfall (2008). Simultaneous Inference in General Parametric Models. Biometrical Journal 50(3), 346--363.

United Nations. “MANGO” An INFOCOMM Commodity Profile, UNCTAD Trust Fund on Market Information on Agricultural Commodities (2016).

APPENDICES

Appendix A. Complete scheme of destruction and analysis steps of fruits coming directly from the field (protocol 1) or from cold chambers (protocol 2).





(Adapted from Isabelle Grechi and Antoine Drouillard, 2017).

Appendix B. Table representing the studied variables, their meanings and how they were obtained.

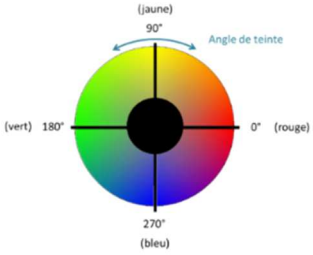
MEASURED VARIABLES	INDICATORS OF	PART OF FRUIT ANALYSED	MEASURMENT METHOD AND/OR MEANING	MEASUREMENT MATERIAL
Fresh mass loss (%)	Growth	Whole fruit	Fresh mass loss during storage, from t_0 to t_i, is: $FML(\%) = 100 * \frac{FM(to) - FM(ti)}{FM(to)}$ with FM the fresh mass of the fruit.	 Precision balance
Transpiration (g/h)	Transpiration	Whole fruit	Fruit transpiration is equal to the weight loss per unit time, calculated from fruit fresh mass measured 4 times over 1 hour. Together with temperature, relative humidity and fruit surface, fruit transpiration is used to calculate "fruit surface conductance" for water, a parameter of the model predicting fruit growth in fresh mass.	 Precision balance
Dimensions: length, width, depth(cm)	Growth	Whole fruit	Fruit dimensions are used to estimate surface of fruits	 Digital caliper
Dry matter content (%)	Organoleptic quality	Pulp	Dry matter content is: $DMC\ (\%) = 100 * \frac{DM}{FM}$ A sample of about 2 g of fresh pulp was weighted with a precision balance before and after 72 hours of drying at 72°C	 Precision balance
Titratable acidy (meqv/100g FM) and pH	Acidity	Pulp	They give information on fruit acidity. Measurements were made on 2 g of grinded pulp (under liquid nitrogen), diluted in 2 ml of distilled water. Titratable acidity was measured by diluting the sample with a NaOH solution at 0.05 mol L^{-1} until the whole sample solution reached pH 8.1	Automated tetrameter and pH-meter
Osmotic and water potentials	Hydric status	Pulp	They are used to calculate turgor pressure and to model water-based processes in the fruit	Osmometer and  Potential-Meter

Brix degree (° Brix)	Soluble sugars content	Pulp	It gives information on all soluble compounds present in water, which will be rounded and used as a rough estimation of soluble sugars, since they constitute the biggest % of the total soluble components	 Digital refractometer
(L*,a*,b*) colorimetric coordinates	Color	Pulp	They are used to calculate Hue angle and represents pulp coloration (*), and can give information on secondary metabolism of fruits	 Chroma-meter
Variable fluorescence	Maturity	Skin	Variable fluorescence Fv is an indicator of the amount of chlorophyll present on fruit skin, giving information on the maturity stage	 Fluorimeter
CO_2 and C_2H_4 (ppm)	Maturity	Skin/ Whole fruit	They give information on respiration and climacteric crisis. After applying a hermetic vial on skin fruits, gas was taken in the vial with a syringe after 1, 2 and 24h. Gas concentrations after 1, 2 and 24h were used to calculate skin resistance to gas diffusion, and internal gas content of the fruit was set as gas concentration in the vial after 24h (Nordey et al. 2016). Gas concentration was measured via chromatographer	Gas chromatographer

Table 1: Resume of studied variables and their explanation.

*Color information are given by the Hue Angle (°), which is an indicator calculated from a and b coordinates measured with the Chroma-meter: $Hue\ Angle\ (°) = \arctan(\frac{a}{b})$. CIELAB (L*a*b) is a color space.

A mango with a Hue Angle 90° has a yellow color. Values higher than 90° represents a color approximately green, while values lower than 90° represents a color that tends to orange. Here in the figure, it is represented the Hue Angle on a chromatic diagram.



Appendix C: Tables containing ANOVA results of Conservation conditions and Maturity stages effects on studied variables.

Fresh mass loss (FML)

Maturity stage	Leaf/Fruit	Statistical results	
		F	p
G	25	33.619	< 0.01
	100	26.767	< 0.01
GM	25	24.523	< 0.001
	100	28.079	< 0.01

Table 1: ANOVA results (F statistics and p-values) for the Conservation conditions effects on fresh mass loss at tf within a specific Maturity stage and a specific Leaf/Fruit ratio treatment.

Time point	Conservation	Leaf/Fruit	Statistical results	
			F	p
tf	T10	25	8.6668	< 0.05
		100	47.478	< 0.01
	T20	25	0.9913	0.37
		100	31.272	< 0.01

Table 2 : ANOVA results (F statistics and p-values) for Maturity stage effects on fresh mass loss at tf within a specific Conservation treatment and a specific Leaf/Fruit ratio treatment.

Dry matter content (DMC)

Maturity stage	Leaf/Fruit	Statistical results	
		F	p
G	25	0.3524	0.58
	100	1.5704	0.28
GM	25	1.3615	0.31
	100	5.6284	0.07

Table 3: ANOVA results (F statistics and p-values) for the Conservation conditions effects on dry matter content at tf within a specific Maturity stage and a specific Leaf/Fruit ratio treatment.

Time point	Conservation	Leaf/Fruit	Statistical results	
			F	p
t0	T10	25		
		100		
	T20	25	5.7797	<0.05
		100	7.257	<0.05
tf	T10	25	9.1096	<0.05
		100	3.6097	0.13
	T20	25	10.904	<0.05
		100	0.2747	0.63

Table 4: ANOVA results (F statistics and p-values) for Maturity stage effects on dry matter content at t0 and tf within a specific Conservation treatment and a specific Leaf/Fruit ratio treatment.

Hue Angle

Maturity stage	Leaf/Fruit	Statistical results	
		F	p
G	25	1.9505	0.23
	100	2.2197	0.21
GM	25	3.279	0.10
	100	7.1331	0.06

Table 5: Table representing the ANOVA results (F statistics and p-values) for Conservation conditions effect.

Time point	Conservation	Leaf/Fruit	Statistical results	
			F	p
t0	T10	25		
		100		
	T20	25	19.872	0.0013
		100	59.303	<0.001
tf	T10	25	8.0059	0.047
		100	0.9238	0.39
	T20	25	1.1578	0.34
		100	2.1874	0.21

Table 6: ANOVA results (F statistics and p-values) for Maturity stage effects on Hue Angle at t0 and tf within a specific Conservation treatment and a specific Leaf/Fruit ratio treatment.

TA

Maturity stage	Leaf/Fruit	Statistical results	
		F	p
G	25	3.7461	0.13
	100	0.0691	0.81
GM	25	0.93	0.39
	100	3.5944	0.13

Table 7: Table representing the ANOVA results (F statistics and p values) for Conservation conditions effect.

Time point	Conservation	Leaf/Fruit	Statistical results	
			F	p
t0	T10	25		
		100		
	T20	25	22.153	<0.001
		100	21.755	<0.001
tf	T10	25	12.534	0.024
		100	2.6089	0.18
	T20	25	0.7494	0.44
		100	0.0054	0.95

Table 8: ANOVA results (F statistics and p-values) for Maturity stage effects on TA at t0 and tf within a specific Conservation treatment and a specific Leaf/Fruit ratio treatment.

°BRIX

Maturity stage	Leaf/Fruit	Statistical results	
		F	p
G	25	1.047	0.36
	100	0.8012	0.42
GM	25	1.649	0.27
	100	3.8141	0.12

Table 9: Table representing the ANOVA results (F statistics and p values) for Conservation conditions effect.

Time point	Conservation	Leaf/Fruit	Statistical results	
			F	p
t0	T10	25		
		100		
	T20	25	9.1852	0.01103
		100	3.5044	0.08819
tf	T10	25	7.8908	0.04836
		100	4.2538	0.11
	T20	25	7.5999	0.05102
		100	0.3855	0.57

Table 10: ANOVA results (F statistics and p-values) for Maturity stage effects on °BRIX at t0 and tf within a specific Conservation treatment and a specific Leaf/Fruit ratio treatment.

C2H4

Maturity stage	Leaf/Fruit	Statistical results	
		F	p
G	25	1.4222	0.29
	100	3.6261	0.13
GM	25	0.5938	0.48
	100	42.533	0.0028

Table 17: Table representing the ANOVA results (F and p values) for Conservation conditions effect.

Time point	Conservation	Leaf/Fruit	Statistical results	
			F	p
t0	T10	25		
		100		
	T20	25	7.3888	0.0241
		100	7.9013	0.016
tf	T10	25	1.0461	0.36
		100	0.4723	0.53
	T20	25	0.1116	0.76
		100	1.6113	0.27

Table 8: ANOVA results (F statistics and p-values) for Maturity stage effects on TA at t0 and tf within a specific Conservation treatment and a specific Leaf/Fruit ratio treatment.