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A multivariate approach to determining and predicting the internal postharvest quality of Hass avocado

Pablo Rodríguez [1,*], Iris Soto [1], Jairo Villamizar [1,2] and Alexander Rebolledo [2]

[1] Corporación Colombiana de Investigación Agropecuaria-Agrosavia. Centro de Investigación La Selva. Km. 7, vía Rionegro - Las Palmas, Sector Llanogrande, Rionegro-Antioquia, 054048, Colombia (Research unit ITAV: Innovaciones Tecnológicas para Agregar Valor a Recursos Agrícolas). prodriguezf@agrosavia.co (P. R.); isoto@agrosavia.co (I. S.).

[2] Corporación Colombiana de Investigación Agropecuaria-Agrosavia. Centro de Investigación Palmira. Diagonal a la intersección de la Carrera 36A con Calle 23, Palmira-Valle del Cauca, 763531, Colombia. arebolledor@agrosavia.co (A. R.); javillamizar@agrosavia.co (J. V.)

* Correspondence: prodriguezf@agrosavia.co; Tel. : +57-317-6582802

Abstract: There is immense variability in the postharvest quality of Hass avocados. However, there is a lack of knowledge to guaranteeing the robustness of fruit with consistent quality. The aims of this work were to develop a multivariate methodology for evaluating the postharvest quality of avo-cado and to determine predictive quality markers to manage fruit quality. Fruit samples produced under different nutrient management, elevation, date-to-harvest, and growing-cycle conditions were analyzed. The results highlighted soil and weather differences among orchards. Nutrient management practices based on index balancing in some samples increased both productivity and the fruit size. High variability was observed in the dry matter related to the age of the fruit at harvest. Ripening heterogeneity was very large in low-elevation orchards where the fruit was picked relatively early. High flesh mineral contents delayed fruit ripening. At low growing temperatures, more oleic and linoleic acids were present in fruits. The sensory texture and taste descriptors were affected by the fruit age and related to the flesh composition. Logistic, PLS-DA, and biplot models effectively represented the variabilities in the ripening pattern, composition, and sensory profile of avocado fruits and allowed the samples to be grouped according to the internal fruit quality.

Keywords: ripening heterogeneity; Hass avocado; quality predictive markers, internal disorders; fruit composition

1. Introduction

The growth dynamics of Hass avocado consumption around the world have been maintained since 2016. The USA is the main avocado-importing market, with a per-capita avocado consumption between 4 and 7 kg, followed by Canada, EU27 West, and EU27+UK [1]. The Asian market is emerging but is currently in the last place, and consumption in Asian countries is expected to increase with campaigns promoting its use in Mexican food, new preparation methods, and the dissemination of its health effects. In the harvest season of 2020-21, Latin America was by far the largest exporter of avocado (82%), with México, Perú, Chile, and Colombia being the countries with the highest export volumes globally. This tendency is due to the desire of consumers to eat healthfully while experiencing new and exotic flavors.

The nutrient management strategies and agroecological characterization of orchards are essential for understanding the behavior of nutrients in plants and for establishing whether each structure has the optimal levels to form quality fruits and increase productivity. It is also necessary to study the elimination and accumulation patterns of nutrients in different agroecological zones if management programs are to be established in avocado orchards to optimize the yield, quality, and efficiency of nutrients in avocado crops [2].

Previously published works have found that the yield and fruit size of avocado could improve with appropriate nutrient management [3,4]. However, there is a lack of available knowledge to balance increasing production without affecting the internal quality and ripening pattern of Hass avocado in the target markets. Production is crucial for crop profitability, but providing markets with high-quality fruit is essential for ensuring the competitiveness of the value chain.

Despite its commercial popularity, the marketing of 'Hass' avocado is constrained by several postharvest disorders. Vast postharvest losses are incurred when fruit are rejected for not meeting consumers' expectations regarding fruit-quality standards. The main challenges faced in this production chain are ripening heterogeneity, internal disorders (both physiological and pathological), and guaranteed sensory quality (nutty, creamy, buttery, flavor, and texture factors). Preharvest factors such as the climate, soil, and agronomic management strategy, among other factors, have also been associated with these problems.

Hofman was among the pioneer researchers who studied the relationship between the mineral flesh composition and internal avocado disorders, focusing on Ca in reducing internal damage due to the role of this element in the cell wall and membrane function [5]. Subsequently, works were performed to optimize the fruit quality [6] and the robustness of the avocado supply chain through mineral nutrition mechanisms [7]. These works took a univariate approach to the quality problem: each study considered one mineral vs. an internal disorder.

Heterogeneity in ripening has been focused on as a main logistical factor [8] but has not been associated with an increase in internal problems when avocado fruits require longer durations to ripen. Although fruit can ripen without internal disorders, such fruit does not always have a high sensory quality [9].

Studies performed in the sensory area have implemented acceptance tests mainly with a focus on postharvest or breeding [10]. Other studies have proposed the content of fatty acids as a marker of origin [11]. However, metabolism during fruit growth limits the application of such methods to countries with highly contrasting production conditions, such as certain regions in Chile, Mexico and Colombia. In addition, few works have sought to determine the relationships between fatty-acid profiles and the sensory quality of avocado fruit.

One aspect of great importance is the lack of multivariate studies addressing avocado quality not only from the aspect of heterogeneity but also from the perspective of the internal and sensory quality to guarantee consistency among the different destination markets of the fruit. Producing countries must include such variables in their production plans to carry out adequate agronomic management programs that guarantee the productivity (ton/Ha) of the crop to achieve economic feasibility without negatively affecting the integral quality of the fruit.

Currently, the harvest indices used, including the dry matter (DM) and oil content (OC) indices, do not allow fruits of high or low integral postharvest quality to be distinguished. Despite past efforts to develop nondestructive techniques, existing methods follow the univariate approach and are often restricted by the high variabilities in the measurement indicators. In a univariate approach, it is not possible to identify orchards, areas and countries with differentiated fruit qualities and thus determine how to manage such regions according to market demands. Therefore, the aim of this work was to develop a methodology for evaluating the integral quality of Hass avocados and to determine predictive quality markers to manage avocado fruit productions.

2. Materials and Methods

2.1 Field conditions

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In the municipalities of Morales (orchard Recuerdo (R), 2°45′37″N, 76°38′02″W) and El Tambo (orchard Nápoles (N), 02°27′05″N, 76°48′39″W) in Cauca, Colombia, fifteen commercial fruit-bearing 15-year-old 'Hass' avocado trees grafted on seed rootstock (possibly of the Antillean ecological group) were selected. Another previously analyzed, orchard, El Banco (EB) (located at 6°29'39.6"N, 75°31'42.0"W, Antioquia Colombia), was used as a quality reference orchard because it produces fruit with a high internal quality consistently during the different studied harvest seasons [9,12].

In the same year in each orchard, trees with different flowering intensities are often present. In this work, trees were selected due to their high flowering level, vigor, and good phytosanitary status. For the R and N orchards, the study was conducted over two harvest cycles, the first corresponding to the intermediate flowering period with anthesis in August 2020 and the second corresponding to main flowering period with anthesis in March 2021 (Figure 1).

Climatic conditions were monitored during two consecutive years with a weather station (Watch Dog 2900ET; Spectrum Technologies, Plainfield, IL, US); the following weather variables were recorded: the daily wind speed, rainfall, relative humidity, sunshine, and maximum, minimum, and average air temperatures. The recorded climatic variables were used to determine the water balance and the high and low rain seasons according to the daily reference evapotranspiration (ET0) based on the FAO (Food and Agriculture Organization)-Penman Monteith method [12].

The nutrient status and management were diagnosed and oriented by the Balanced Indices of Kenworthy (BIK) [13] for the orchards located in the department of Cauca in August 2020 (Figure 1), and a fertilizer plan was applied monthly. Nutrient removal with a 20-ton target yield was used to calculate the fertilization plan from the data obtained by Salazar-García and Lazcano-Ferrat [3].

Soil chemical analyses of the top 45-cm layer were performed at the beginning of the experiment, and the results were used to calculate the nutrient supply capacity of the soil. To determine the plant nutrient status, fifty leaves from the vegetative growth were collected from trees at a height of 1.5 m every two months during the two consecutive years of study and washed, oven-dried to a constant mass and sent to the laboratory at the Corporación Colombiana de Investigación Agropecuaria - AGROSAVIA for mineral nutrient analysis.

The yield (kilogram/tree) and the number of fruits per tree were determined at the time of harvest during the two flowering cycles. The mass of a hundred randomly selected fruits from trees with high crop loads were collected and used to calculate the total number of fruits per tree. The harvested fruit were then classified according to the Codex Alimentarius standard (Codex Stand 197, 1995) with the following packing carton sizes (range in grams per fruit): size 10 (364–462), size 12 (300–371), size 14 (258–313), size 16 (227–274), size 18 (203–243), size 20 (184–217), size 22 (165–196), size 24 (151–175), size 26 (144–157), size 28 (134–147), size 30 (123–137), and size 32 (80–123).

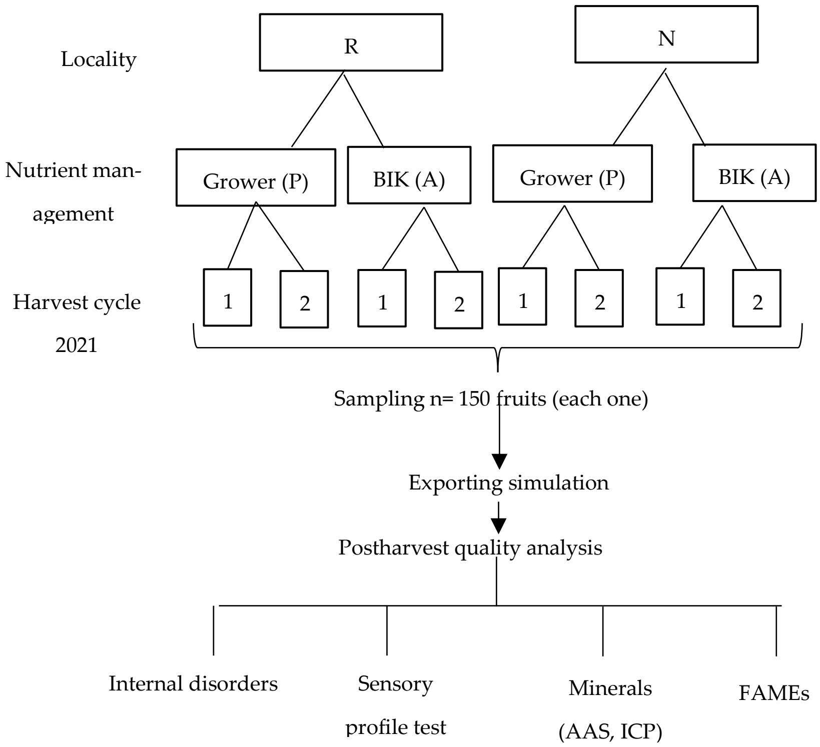

Figure 1. Schematic representation of the experimental design and postharvest quality analysis. Locality (orchard): **R:** Recuerdo, **N:** Nápoles. **BIK:** Balanced Indices of Kenworthy. **P:** nutrient management of the farmer. **A:** Nutrient management based on the BIK. Harvest cycle: **1.** Harvest from the intermediate flowering period. **2.** Harvest from the main flowering period. **FAMEs:** fatty acid methyl esters. **AAS:** Atomic absorption spectroscopy. **ICP:** Inductively Coupled Plasma.

2.2 Postharvest quality

2.2.1 Sampling

In each orchard, a sample of 150 fruits per nutrient management strategy was taken for each harvest cycle (Figure 1). As described before, one sample was taken from the EB orchard for comparison with the R and N orchard results. The fruit was harvested when it reached the dry matter (DM) value at commercial maturity employed by the grower according to the practices of the packing houses. The DM and OC contents were analyzed according to the AOAC methods (American Organization of Analytical Chemists, No. 934.01 and 963.15, respectively) [14].

2.2.2 Storage and ripening

The samples were disinfected following the packing house practices and the methods reported by Hofman et al. (2003) [5]. The fruits were immersed in a prochloraz solution of 0.05% w/v (Mirage 45 ADAMA ANDINA) for 30 seconds. Then, the samples were dried at room temperature and packed into corrugated cardboard boxes. The fruits were stored for 2 weeks, simulating European market export conditions (at 5 °C/90% relative humidity (RH), Memmert HPP 110).

After that, the fruit was ripened (20 °C/90% RH, Memmert HPP 110). Ripening ended when fruits reached a rating of 5 according to White et al. (2009), corresponding to the firmness range from 8 to 12 N, as measured in the fruit's equatorial zone using a TA-XT Plus texture analyzer (Texture Technologies Corp., New York City, NY, US) equipped with a 30-kg load cell [15]. Each day, two measurements were performed on unpeeled fruit with a stainless-steel compression plate (P/75.75-mm diameter).

2.2.3 Fruit postharvest quality analyses

- **Internal defects or disorders**

Internal defects were evaluated by cutting fruits longitudinally in half, and the damage was identified as described in the international quality manual [15]. The internal disorder incidence (damage frequency into the sample) and severity (percentage of the flesh surface area affected) were evaluated.

- **Sensory analysis**

The method used involved a sensory profile developed through a multidimensional approach [16]. Fruits without internal damage were cut longitudinally in two halves, and the seeds were removed. At the equatorial fruit zone, a cube (2 cm^3 each) was taken and placed into a 30-cm^3 plastic dish labeled with a random three-digit number.

The test was then performed by a trained panel. The descriptors were selected based on previous works [9] and preliminary tests to verify the consensus of the panel about the Hass avocado sensory descriptors corresponding to the quality profile. Appearance, smell, taste, texture, and general quality descriptors were evaluated on a scale from 0 to 5. Panelists were also instructed to take a bite of cracker and drink some water to rinse their palate between each sample [17].

The sensory analysis was performed in the laboratory at the postharvest and agrofood processing pilot plant facilities (Agrosavia La Selva Research Center, Rionegro, Antioquia Colombia).

- **Flesh mineral content**

The analysis was performed following the procedure published by Escobar et al. [18]. Briefly, the flesh was dried at 70 °C in a forced convection oven (Memmert UF 110) until a constant weight was attained. Then, the samples were hydrolyzed, and the mineral content (K, Ca, Mg, S, and Na) was analyzed with an inductively coupled plasma (ICP) emission spectrometer (Thermo Scientific iCAP 6500 duo). Atomic absorption spectroscopy (AAS, Agilent Technologies FS 240) was used to analyze the P, Fe, Cu, Mn, and Zn contents. The B content was determined in a spectrophotometer UV–VIS (Thermo Scientific Spectronic Genesys 10 S) at a wavelength of 430 nm following the methods of McKean [19].

- **Fatty acid methyl esters (FAMEs)**

The FAME profile was identified and quantified according to AOAC 996.06 [14]. The extracted total fat (TF) was methylated to fatty acid methyl esters using BF_3 in methanol. FAMEs were quantitatively measured by gas chromatography and a flame ionization detector (GC-FID) (Thermo Trace 1300) against the C11:0 internal standard.

2.3 Statistical Analysis

The homoscedasticity and normality of the obtained data were analyzed to determine whether parametric or nonparametric tests should be applied. The fruit-ripening heterogeneity was analyzed using the nonparametric Kruskal–Wallis test. After that, Dunn's multiple-comparisons test was applied with a Bonferroni correction ($p < 0.05$). For the incidence of internal disorders, descriptive analyses of the qualitative data were used. Then, the relative frequency was analyzed by a k proportion test using a Chi-square test (α=0.05).

The disorder severity was analyzed by a Kruskal–Wallis test. The incidence of internal disorders was modelled through nonlinear regression using a logistic model (confidence interval 95%). Multivariate data of minerals and FAME flesh composition were analyzed by unsupervised (PCA) and supervised (PLS-DA) analysis methods. Sensory product characterization was performed to identify which descriptors best discriminated a set of fruits according to the orchard-specific nutrient management program and the growing cycle. For each descriptor, an ANOVA model was applied to check whether the scores given by the assessors differed significantly (XLSTAT 2022.2.1 Addinsoft).

3. Results

3.1 Field conditions

The experimental avocado zones of R and N in the department of Cauca are located at elevations of 1600 and 1708 m.a.s.l, respectively. The reference orchard EB is located in the department of Antioquia at an elevation of 2464 m.a.s.l. The variations in climatic conditions among the EB, R and N orchards were registered (Figure 2). Significant differences of approximately 5 °C were observed in the maximum and minimum temperatures among the R, N and EB orchards (Figure 2a,b).

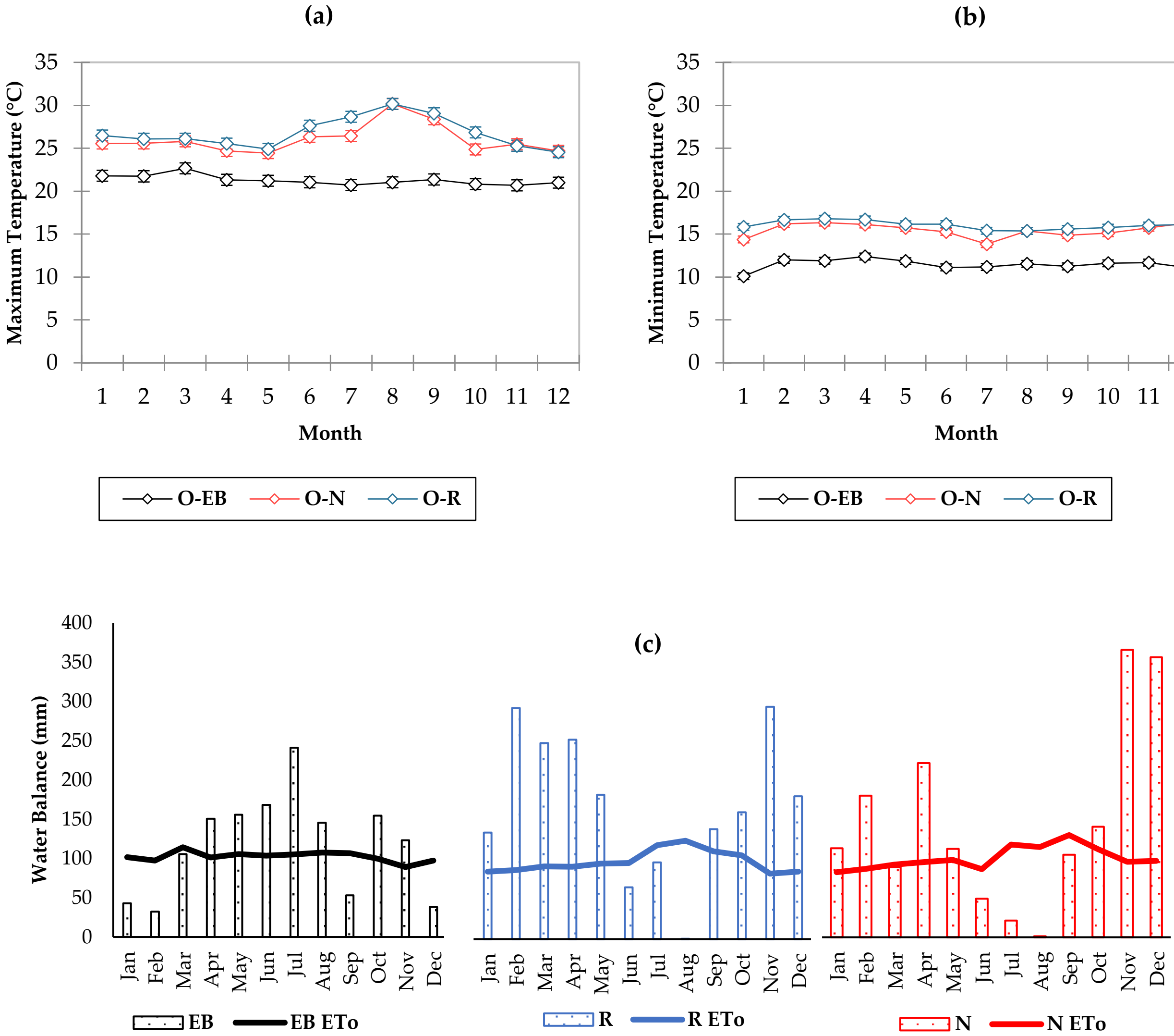

Figure 2. Climate conditions of the Hass avocado experimental zones corresponding to the following variables: (a) maximum temperature, (b) minimum temperature and (c) evapotranspiration. **O-EB:** Orchard El Banco. **O-N:** Orchard Nápoles. **O-R:** Orchard Recuerdo. **ET0:** evapotranspiration.

The cumulative annual precipitation totals were 2060 mm, 1795 mm and 1411 mm in the R, N and EB study areas, respectively, with a differential monthly distribution pattern throughout the year. An average of 211 mm of precipitation was recorded in the high-rain seasons from January to May and from October to December, while 61 mm of precipitation was recorded in the low-rain season from June to September in both R and N. A variation was reported in the precipitation distribution pattern in the EB location. A low-rain season occurred in January, February, September, and December, with 42 mm of precipitation falling (Figure 2c).

The soil characteristics in the experimental zones located in the Cauca region showed a sandy loam texture and pH conditions between moderate to hard acidic (Table 1). The electrical conductivity values indicated that these are salt-free soils, in addition to having high organic matter (OM) contents, indicating a high nitrogen availability.

The cation exchange capacity (CEC) showed values less than 10 meq/100 g, indicating the very poor ability of the soil to exchange nutrients such as calcium, magnesium, sodium, and potassium. These characteristics should be explained by the loamy-sandy texture of this kind of soil (Table 1). Some of these characteristics are common for the EB orchard, but the contents of mineral nutrients such as Ca, Mg, P, and minor elements were within the normal concentration ranges.

Table 1. Ranges of chemical contents of the soils in the experimental avocado orchards.

Mineral	R	N	EB
	Min - Max	Min - Max	Min - Max
Depth (cm)	0. 45	0.45	0.45
pH	5.42 ± 0.10	5.50 ± 0.06	5.12 ± 0.05
EC (ds/m)	0.33 ± 0.10	0.35 ± 0.06	1.05 ± 0.11
OM (%)	12.36 ± 0.47	14.85 ± 0.88	12.53 ± 0.69
P*	4.69 ± 0.72	5.45 ± 0.70	37.79 ± 5.08
S*	67.03 ± 7.4	75.13 ± 7.9	30.52 ± 1.83
Mg*	0.97 ± 0.15	0.99 ± 0.04	1.64 ± 0.27
Ca**	2.87± 0.40	3.90 ± 0.5	4.04 ± 0.61
K**	0.62 ± 0.09	0.44 ± 0.12	0.62 ± 0.05
Na**	0.09 ± 0.003	0.08 ± 0.008	0.10 ± 0.001
CEC**	4.64 ± 1.00	5.29 ± 0.6	6.98 ± 0.82
B*	0.22 ± 0.04	0.26 ± 0.03	0.33 ± 0.06
Mn*	2.39 ± 0.30	2.19 ± 0.20	13.05 ± 0.88
Cu*	2.49 ± 0.90	0.69 ± 0.20	5.65 ± 0.41
Fe*	101.95 ± 2.72	80.20 ± 5.60	309.32 ± 22.23
Zn*	1.54 ± 0.20	3.76 ± 1.20	8.56 ± 1.35
Soil texture	SL	SL	SL

R: Orchard Recuerdo. **N:** Orchard Nápoles. **EB:** Orchard El Banco. **EC:** electric conductivity. **OM:** organic matter**.** **CEC:** cation exchange capacity. **SL:** sandy loam. * (%), ** (mg/kg).

Two flowering periods were recorded in two consecutive years, characterized as the intermediate flowering period or cycle 1 (July September) and the main flowering period or cycle 2 (February-April); these flowering period peaked with the largest proportion of flowers in anthesis in August and March, respectively.

The harvest from the intermediate flowering period was recorded in April, and that from main flowering period was recorded in November 2021. An increase in the per-tree yield was registered in the harvest from main flowering period with a nutrient management program based on the BIK standards. The wide standard error recorded in all treatments reflects the variation in the per-tree crop yield due to variations in the flowering intensity (Figure 3).

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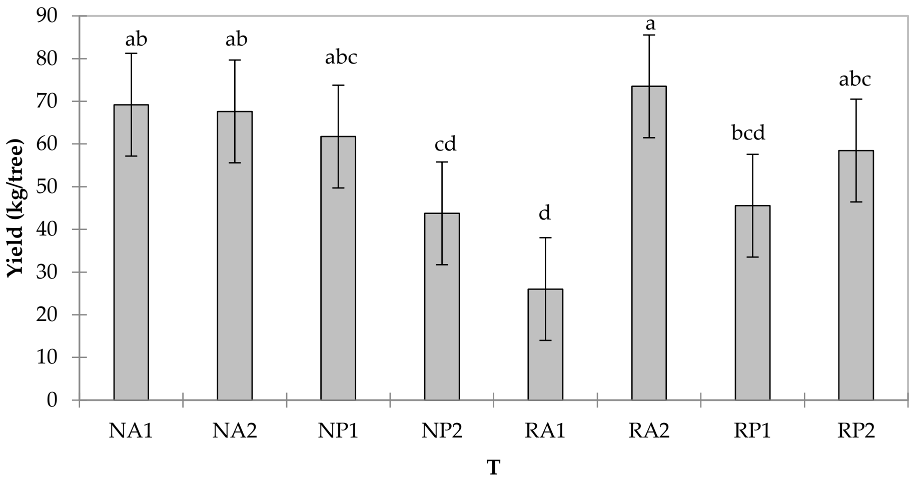


Figure 3. Yield (production average, kg/per tree) of Hass avocados in each orchard using 2 nutrient management practices during 2 production cycles. Different letters represent significant differences (P < 0.05) as determined using ANOVA and Tukey's post hoc test. **T:** treatments. **N:** Nápoles. **R:** Recuerdo. **P:** nutrient management of the farmer. **A:** Nutrient management based on the BIK standards. Harvest cycles: **1.** Harvest from the intermediate flowering period. **2.** Harvest from the main flowering period.

In accordance with the Codex Alimentarius standard [20], the fresh fruit weight at harvest showed an increase in the percentage of fruits from size 22 (165-196 g) in both the R and N orchards (Figure 4a) with the nutrient management program based on the BIK standards [13]. In the N orchard, no effect of nutrient management on improving the fruit weight from the intermediate flowering period was observed (Figure 4b).

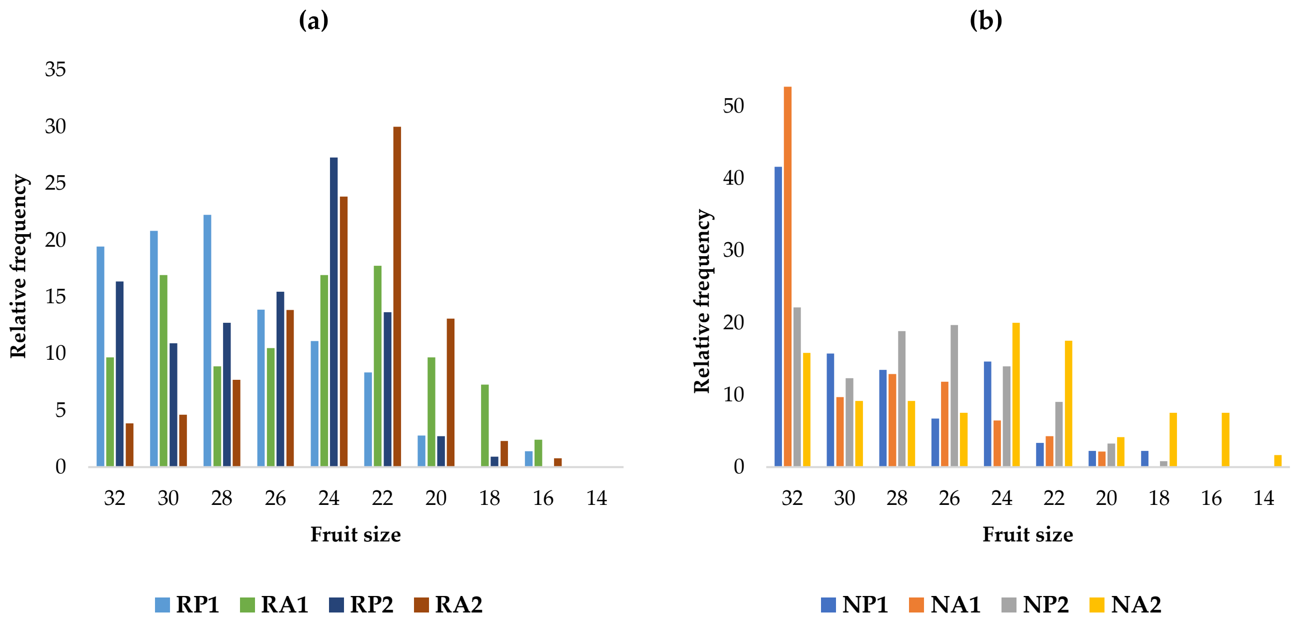


Figure 4. Fruit harvest size classification results according to the Codex Stand 197, 1995. **R:** Recuerdo orchard. **N:** Nápoles orchard. **P:** Nutrient management of the farmer. **A:** Nutrient management program based on the BIK standards. **1.** Harvest from the intermediate flowering period. **2.** Harvest from the main flowering period.

3.2 Postharvest quality

In Figure 5, the Hass avocado ripening heterogeneity is shown. The harvest index (DM or fruit development time) has a greater impact on the ripening pattern of Hass avocados than either the nutrient management program or growing season cycle does. The orchard located above 2400 m.a.s.l had a shorter ripening ranges than the orchards situated at elevations below 1710 m.a.s.l. At high elevations, the development of fruit is slower than at low elevations because of the temperature differences (approximately 5 °C) among the N, R and EB orchards (Figure 2a and 2 b).

In terms of the fruit development time, differences of at least 50 days were observed (Figure S1). In other words, the growers at low elevations harvested their fruit earlier than those at high elevations. Moreover, the mineral accumulations in the R and N orchards were larger than those in EB, as will be discussed later. Even though the EB orchard had a high range of variability at harvest (Figure 1S), its softening pattern occurred faster. Therefore, other preharvest factors must influence the ripening heterogeneity pattern, and the need thus exists to include not only classical harvest indices but also other quality markers when predicting the robustness of fruit to exportation.

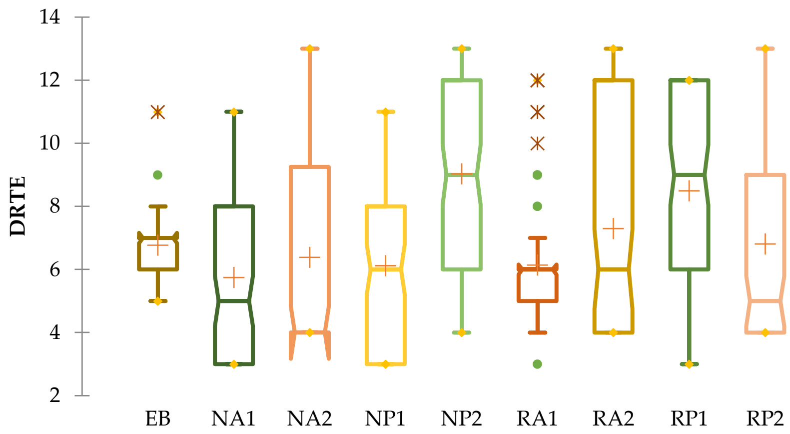


Figure 5. Ripening heterogeneity results of Hass avocados. Orchards: **EB:** El Banco, **N:** Nápoles, and **R:** Recuerdo. **A:** Nutrient management based program on the Balanced Indices of Kenworthy (BIK) standards. **P:** Nutrient management of the farmer. **1.** Harvest from the intermediate flowering period. **2.** Harvest from the main flowering period. **DRTE:** Days to ready-to-eat. The crosses represents the mean values, the yellow dots indicate the minimum and maximum values, and the green dots and brown asterisks denote outliers.

The incidence rates of internal disorders and the corresponding severity were higher in the N and R orchards than in the EB orchard (Figure 2Sa). Nutrient management A (BIK) reduced the incidence of disorders, but there was no significant difference from grower management. Moreover, the disorder severity was lowest at the EB orchard. This is contrary to the literature that reports that at high DM contents, the highest severity of flesh damage occurs. Therefore, these results confirm that internal damage appears when fruits take a long time to reach the RTE stage. For the N and R orchards, the mineral contents impeded the Hass avocado ripening pattern, as discussed above, and promoted the increased incidence and severity of internal disorders as the DTRE was lengthened (Figure 3S).

Vascular browning was the main internal disorder found (37-76%), and the N and R orchards had the highest occurrence rates of this disorder (Figure 4Sa). Stem end rot was the second most common internal disorder in the samples (6-46%), for which EB orchard had the lowest incidence rate. Flesh discoloration (FD), flesh bruising (FB), flesh rots and body rots were found in the samples with a low relative incidence frequencies.

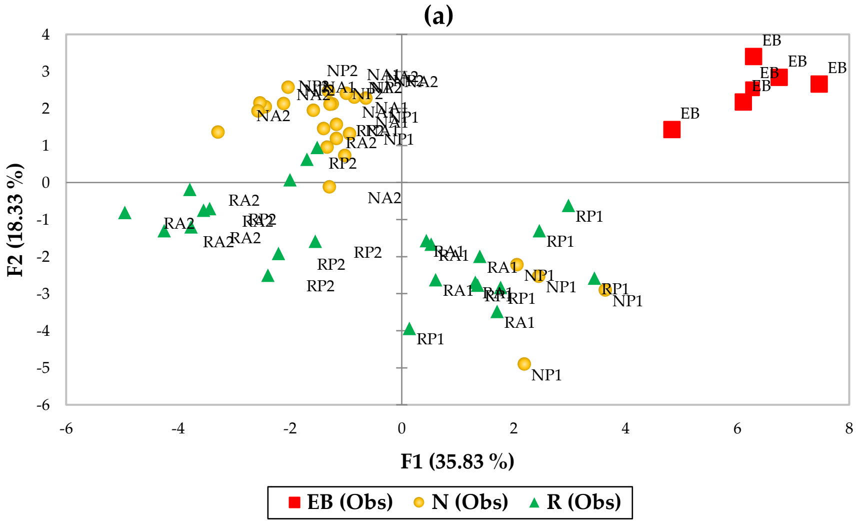
For both growth cycles, the percentages of internal damage severity at the R and N orchards were higher than 20% (Figure 5S, 6S). The severity increased after the 8 DRTE (days to ready-to-eat) and, in many cases, exceeded the control limit and could affect the consumer acceptance of fruit at the international market [21] because of the disorder severity [22].

The principal component analysis (PCA) results of the flesh mineral and fatty acid contents are shown in Figure 6. The score plot shows that the orchards were effectively grouped according to their composition. In Figure 6b, the loading plot shows that the N orchard had the highest B, N, Zn, and S contents, except for 4 samples in growth cycle 1 with nutrient management of the grower, which showed low values of these minerals. The R orchard had more Mg, Ca, Cu and Mg, especially during growth cycle 2. This could be the cause of a reduction in the internal quality (increased internal disorders and a reduced sensory quality), as will be discussed. The EB orchard had intermediate quantities of B, Fe, and Mn even though it had the highest fruit development time and OC.

The samples far from the EB grouping had a low internal quality, high ripening heterogeneity and even lower sensory quality, as will be shown later. Descriptive statistics of the flesh mineral composition for each orchard are presented in Table 4S. These EB mineral contents could be used as a reference to determine Hass avocado heterogeneity and internal quality. Moreover, the results show that despite growing season 2 having low harvest indices, the mineral contents were high at this time. The PLS-DA analysis shows that the principal discriminant variables between orchards indicate that K, N, B, P and S correspond to the first factor and B, Cu, S, N and K correspond to the second factor (Figure 7).

Factors 1 and 2 explain 80% of the variance observed in the Y variables (orchards), and with 3 factors, the model has good robustness with regards to both prediction and stability (R^2= 0.889, SCE= 0.106, SECV= 0.122). Therefore, the model could be useful for identifying whether new samples have a composition similar or not to the EB samples (colored in red and defined as an ellipse quality in Figure 7a). These models could allow us to predict the internal fruit quality and guarantee the mineral composition necessary to produce a robust fruit to the international supply chain of fresh fruit.

(a)



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(b)

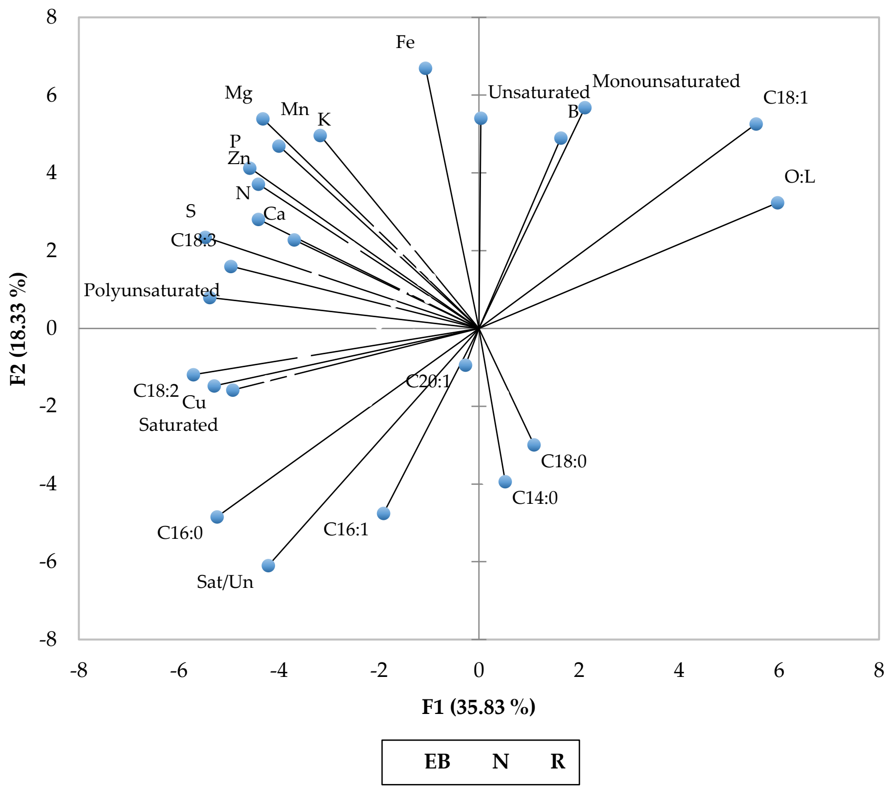
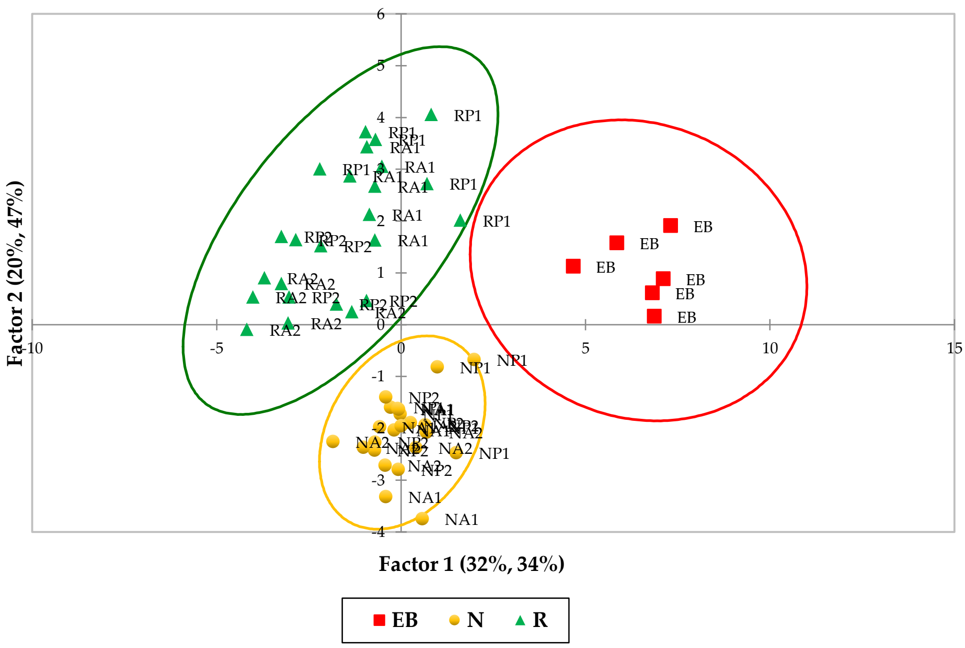


Figure 6. Principal component analysis (PCA) results of the mineral and FAME contents grouped by the samples. **a)** Score plot. **b)** Loading plots. Blue dots: active variables. Orchards: **EB:** El Banco, **N:** Nápoles, and **R:** Recuerdo. **A:** Nutrient management based on the Balanced Indices of Kenworthy (BIK). **P:** Nutrient management of the farmer**. 1.** Harvest from the intermediate flowering period. **2.** Harvest from the main flowering period.

(a)



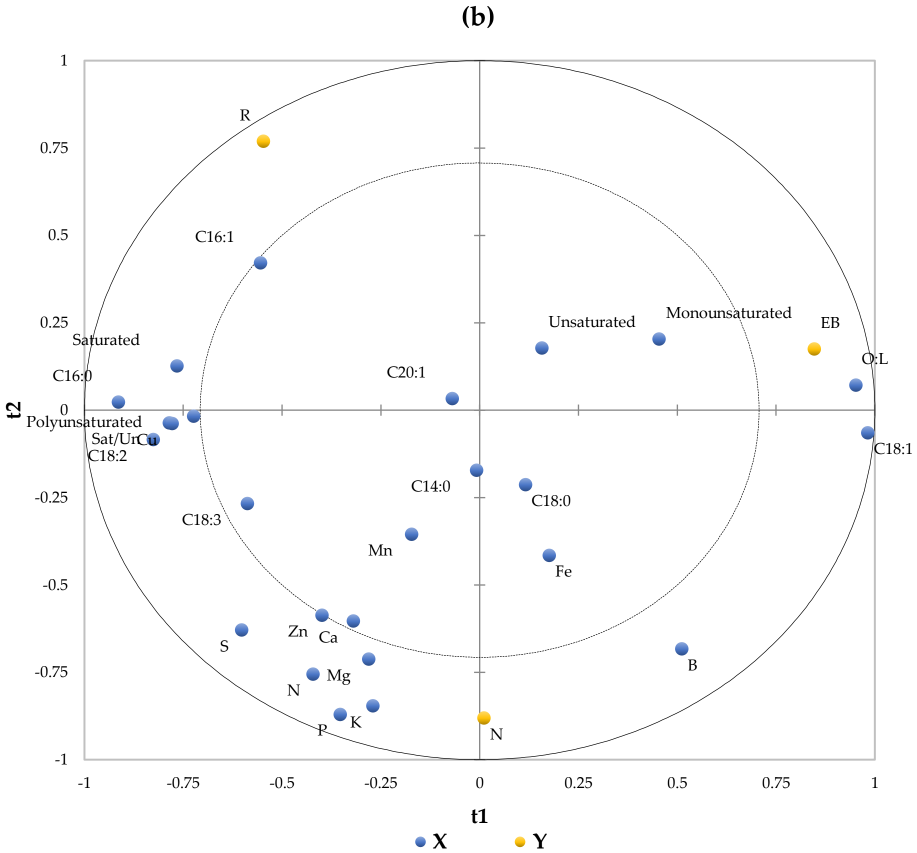


Figure 7. Partial least squares discriminant analysis (PLS-DA) results of the fatty acid and mineral contents grouped by orchard. Score plot (a) and loading plots (b). R^2= 88.9 (3 factors). Orchards: **EB:** El Banco, **N:** Nápoles, and **R:** Recuerdo. **A:** Nutrient management based on the Balanced Indices of Kenworthy (BIK). **P:** Nutrient management of the farmer**. 1.** Harvest from the intermediate flowering period. **2.** Harvest from the main flowering period.

The quality of Hass avocado fruits must be qualified not only as the absence of internal disorders but also as an optimal sensory profile related to the origin (country, region, etc.), rootstock, management, and harvest practices, among other factors. A multivariate profile indicating the orchards, growing season and nutrient management factors is shown in Figure 8.

The results show that the sensory descriptors related to a high general quality are coconut sensation, hazelnut sensation, flux texture and creamy sensation (oily sensation), and the means adjusted for these descriptors were high in EB, RA1 and RP1 tests (Figure 8).

Therefore, as discussed before, the composition has an impact on the overall Hass avocado fruit quality, and the "quality ellipse" established (Figure 7 a) in the flesh mineral composition indicates that the oleic content and oleic: linoleic relationship are in accordance with the sensory quality profile. A high mineral content is related to hard or firm, lumpy and fibrous textures, astringent sensations, and sweet, acid, and bitter tastes, effectively explaining how the sensory profile is related to the RP1, NA1, NP1 samples. Growing cycle 2 had a low intensity in almost all of the descriptors, and the factor-2 fruits of the biplot showed a hard and fibrous texture with a high green color (Factor 2) and poor development of the discriminant descriptors related to a high sensory quality (Figure 8).

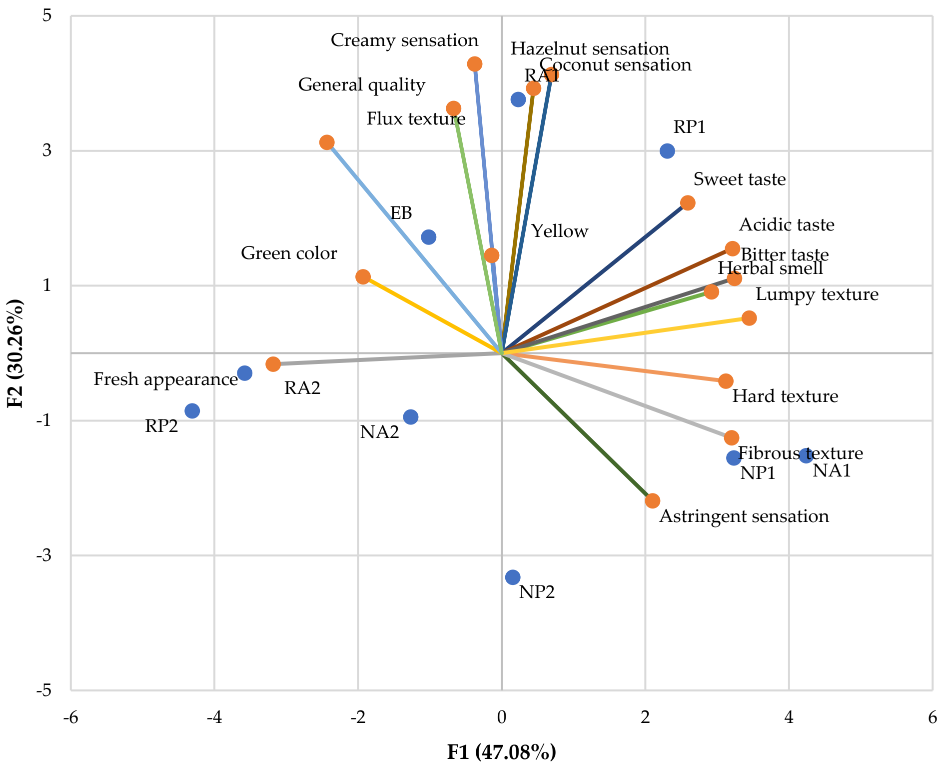


Figure 8. Sensory profile of fruits from different orchards under different nutrient management (P and A) and growth cycle (1 and 2) conditions. Orchards: **EB:** El Banco, **N:** Nápoles, and **R:** Recuerdo. **A:** Nutrient management based on the Balanced Indices of Kenworthy (BIK). **P:** Nutrient management of the farmer**. 1.** Harvest from the intermediate flowering period. **2.** Harvest from the main flowering period.

Moreover, the mineral composition and the internal and sensory quality of fruits are affected by the fatty acid composition. The EB orchard had a high oleic acid content and a strong O:L (oleic: linoleic) relationship (Figures 6 and 7). The R and N orchards had greater saturated fatty acid contents (myristic, palmitic, and stearic acids).

One difference between the R and N orchards was that unsaturated fatty acids were more abundant in fruits grown in the R orchard. The variability found in the fatty acid composition between orchards was well-represented by the PLS-DA analysis (Figure 7). These results explain the differences found in the sensory analysis. The samples with more unsaturated (EB and R) fatty acids (oleic, linoleic, linolenic and palmitoleic acids) had the highest sensory quality (creamy sensation, flux texture, and hazelnut and coconut sensations).

4. Discussion

As a common characteristic in avocado-producing areas in Colombia, a bimodal rainfall distribution pattern is presented [23–27], as was registered in the studied avocado-producing area of Cauca. In this region, the high rainy season registered from January to May supplies the necessary water for nutrient mobility during the main flowering period. However, the intermediate flowering matches the low rainy season recorded from June to September. The flowering phase and initial fruit growth are highly demanding in terms of water and nutrients, and stress factors resulting from these two resources can increase the abscission of flowers and set fruits and can affect fruit growth [28–33]. Herein, in the experimental orchards in Cauca, the nutrient management program based on BIK supplied nutrients monthly, but the low water availability in the low rainy seasons potentially limited the transport of these nutrients to the plants during critical periods of high demand within the intermediate flowering period.

The soils in Cauca orchards showed a low capacity to exchange nutrients such as calcium, magnesium, sodium, and potassium. Previous reports showed a relationship between the nutritional condition of avocado trees and the availability of elements in the soil [34], and some characteristics, such as acidic pH values, low levels of organic matter and high to very high levels of Cu, Fe, K, Ca, B and Zn, lead to deficit concentrations of nutrients in the leaves [35]. BIK is a widely used tool for diagnosing the nutrimental status of Hass avocado trees [35,36].

In the methodology adopted herein, we considered an optimum standard value corresponding to each mineral nutrient in trees with yields exceeding 85 kg per tree and obtain the standard deviation for each nutrient in this population of trees. Using this tool to orientate nutrient management in the orchards of the Cauca region, yield increases of 55% and 26% in N and R orchards, respectively, were recorded in the harvest from the main flowering period. Additionally, improvements in fruit weight were recorded. The results shown herein are in accordance with the reports of Salazar-García and Lazcano-Ferrat, who oriented nutrient management based on the BIK methodology and increased fruit calibers with sizes greater than 170 g by 72% of the orchard yield, using specific fertilization with doses calculated for each nutrient throughout the growth cycle of the Hass avocado [4].

Ripening heterogeneity has been reported as a severe logistical problem for marketers and results in fruits of inconsistent quality being delivery to consumers. This phenomenon is the result of the complex avocado fruit physiology and preharvest and postharvest factors [8]. Rapid softening during ripening has been associated with C7 sugars, organic acids, and the lauric acid, stearic acid, and oleic acid contents.

Therefore, avocado-producing countries with high variability face challenges due to the primary and secondary metabolites of Hass avocados produced during fruit development being affected by the climate, harvest practices, rootstock, etc. [8,37]. Moreover, the preharvest factors and harvest indices are variable in Colombian conditions [38]. In practice, the harvest begins in low-elevation regions at a DM value close to 23%. Our results agree with those of Fuentealba et al.: at low DM values, more ripening heterogeneity occurs [39].

This is because the biological age of the fruit does not guarantee the quantity of fatty acids or other metabolites that soften flesh. Therefore, in Cauca orchards, growers must increase their development time to ensure adequate Hass avocado ripening.

The flesh mineral content has an effect on delaying the firmness of mesocarp softness because some elements are present and associated with the cell wall structure. Previously, nutrition has been reported to play an important role in postharvest Hass avocado quality. The effect on internal quality of some minerals has been discussed separately, as briefly described below. High levels of calcium in avocado fruits have been associated with a delay in ripening due to the effects on respiration, the impedance of the ethylene peak, greater firmness after storage and reduction in internal defects (stem end rot, body rot, vascular browning flesh discoloration, etc.) [40]. An excess of nitrogen fertilization induces the activation of cytokinins that inhibit the production of ethylene and ABA [41], making fruit more vulnerable to fungal pathogen attack [42] and more susceptible to postharvest rot and vascular browning.

Similar to cell wall-associated calcium, boron differences might be another factor contributing to the observed ripening heterogeneity because 95% of the flesh B content is present in the cell wall [8] and acts as a bridge between the cell wall and membrane [8]. In contrast, B-deficient tissues have greater PPO activity and accumulate more phenolic compounds, the substrates for PPO-mediated browning reactions. Increasing the K supply beyond the minimum required value may lead to K-induced deficiencies of Ca and Mg, which could cause increased diffuse discoloration and body rot severity. Attention has been given to the balance between Mg, Ca and K cations in the fruit, as this balance seems to be more strongly associated with avocado fruit quality than the Mg content alone [7].

According to Rooyen and Bower, copper is known to activate a group of oxidizing enzymes, such as polyphenol oxidase, monophenol oxidase, laccase and systems of oxidizing ascorbic acid [38,43]. Copper acts as a catalyst for the enzymatic systems that lead to enzymatic browning and is thought to be interrelated with zinc in various oxidation–reduction reactions. Zinc is often coordinated with copper in the various oxidation–reduction reactions that lead to browning. For the other minerals, there is a lack of knowledge of their roles in affecting the internal quality of Hass avocado fruits.

The approaches of past studies aimed to find a simple relationship (linear or nonlinear) between mineral contents and internal disorders. However, our results show that no one mineral plays the main role in affecting the internal quality of Hass avocado fruits, and the associated variabilities could be well-represented in a multivariate approach. PCA and PLS-DA were very useful because the confidence of the ellipse of the high-quality group (EB) allowed the identification of multiple vectors (variables) that cause heterogeneity, internal quality and sensory profile differences related to the flesh composition.

After the internal disorders, the other quality challenge of Hass avocado origins is obtaining a sensory quality that is satisfactory to consumer expectations. Therefore, a high sensory quality could serve as a competitiveness strategy to improve the success of certain avocado origins in the international market, especially in emerging avocado markets. Studies have found that the harvest season has an effect on the sensory quality of avocado fruits.

Arpaia et al. reported that likeability increased with an advancing harvest date (DM content), and their research was performed with preference sensory tests [44]. Rodríguez et al. employed a sensory profile test and showed that the discriminant descriptors related to unmature fruit were green, bitter, acid, spicy taste, and sensations, whereas flux texture, oily taste, oily texture, and sweet taste were descriptors found in fruits at high DM contents [9].

The results of this work confirmed that harvest practices had an important impact on the Hass avocado fruit quality profile. The main issue under field conditions is that farmers do not have nondestructive equipment or lack knowledge regarding what markers to measure to sort fruit and guide the harvest decisions to guarantee the robustness and sensory quality of the fruit at the destination market [45,46]. Our results show that the sensory descriptors change among localities and growth cycles and could be explained as discussed above with regards to the flesh mineral and FAME composition.

There is no information that reports the role of the mineral contents on the sensory quality of Hass avocado fruits. However, the role of the fruit mineral composition on fruit softness and mesocarp discoloration in 'Pinkerton' avocado fruits was studied by Rooyen and Bower, who published mineral concentrations related to low, medium and high risks of developing internal flesh disorders [38]. The results of this work contribute not only to reporting optimal mineral ranges for reducing internal damage but also to improving the sensory profile of Hass avocados.

The flesh fatty acid relationship is influenced by the temperature of the localities or regions where the Hass avocado is produced. This phenomenon has been found in many Hass avocado-producing countries around the world. Some studies have reported that Hass avocados produced in regions with low temperatures had greater oil contents and higher proportions of oleic acid than those grown in high-temperature areas. In contrast, fruits grown in high-temperature regions had more palmitic acid [47,48]. Ferreyra et al. found that the fatty acid proportion changed with elevation in Chile [40].

The authors reported that high oleic acid contents were related to the maximum mean temperature. Therefore, different studies have postulated the fatty acid profile as a biomarker of fruit origin. However, high variabilities exist in avocado-producing countries such as México, Colombia, Chile, and Peru, thus making such classifications difficult. The EB locality has an oleic acid content similar to the cool regions or farms from South Africa [48], New Zealand [47], and Chile [40,49], whereas the low-elevation orchard has a FAME profile similar to producing localities in Peru, Spain [49] and California, USA [50]. Fruits grown at the R orchard at both nutrient management levels had more palmitoleic acid than fruits of other international origins (low and high elevation or temperature) and were similar to the avocados produced in Florida, USA [51]. There is a lack of information relating the fatty acid profile to the sensory quality. In this work, it is clear that the sensory analysis was influenced by the proportion of fatty acids in the samples, especially with regards to unsaturated fats.

The mouth sensory texture perception could be associated with a change in the mesocarp tissue due to enzymatic (polygalacturonase and cellulase) action during fruit ripening and the mineral and fatty acid compositions. The parenchyma tissue contains oil bodies as a finely dispersed emulsion surrounded by a thin cellulose wall [52], whereas idioblasts are specialized cells containing large oil sacs covered by a thick cellular wall.

During Hass avocado ripening, the degradation of parenchyma cell walls and the release of some oil bodies in emulsion form occur [53]. Despite the importance of the role of fatty acids in Hass avocado composition and quality, few studies have reported the role of nutrient management in modulating fatty acids. This work contributes to identifying the quality markers related to nutrient management under field conditions. Our results show the importance of having a robust fruit flesh composition, and it is necessary to consider the rootstock selection, nutrition management and orchard location to guarantee a consistent quality to consumers.

It is necessary to increase the knowledge on the development of nondestructive devices, NIR applications, or other technologies focused on analyzing the discriminant biomarkers found in this work to predict the integral quality of Hass avocados and their shelf life before export. These strategies could reduce postharvest losses and satisfy consumer expectations worldwide.

5. Conclusions

Nutrient BIK management increased the fruit sizes and yields without affecting the postharvest quality. Differences in growing temperatures among localities influenced the fruit compositions, especially the fatty acid composition and postharvest internal quality. The fruit produced at low temperatures had relatively high oleic acid contents, while fruits from orchards at high growing temperatures had more palmitic, palmitoleic and stearic fatty acids and more internal disorders. Fruits harvested at more development days ripened faster and thus had reduced internal disorder incidence rates and severities.

The multivariate approach allowed us to find a robust model for representing and following the variability in the postharvest internal avocado fruit quality independently of the harvest season, locality, and nutrient management. The PLS-DA model could be a powerful tool for predicting the internal quality of fruits in avocado-producing countries. Therefore, in countries with highly variable growing conditions and production practices, the use of destructive analyses and multivariate data analyses could reduce the heterogeneity and allow the shipment of fruits with better internal quality to the international market.

Supplementary Materials: The following supporting information can be downloaded at www.mdpi.com/xxx/s1. Figure S1: Variability in the dry matter (DM) content of Hass avocado fruits during harvest cycles. Figure S2: Percentage of incidence (a) and severity (b) of internal disorders. Figure S3: Logistic regression analysis of internal disorder incidence. Figure S4: Incidence of internal disorders in avocado: relative frequency in vascular browning (a), stem end rot (b), flesh discoloration (c), flesh bruising (d), flesh rots (e), and body rots (f). Figure S5: Severity of internal disorders through Hass avocado ripening for growing cycle 1 of 2021. Figure S6: Severity of internal disorders through Hass avocado ripening for growing cycle 2 of 2021. Table S1: K proportion test results regarding the incidence of internal disorders (Marascuilo procedure). Table S2: Kruskal–Wallis two-tailed test results regarding disorder severity (multiple pairwise comparisons using Dunn's procedure with Bonferroni correction). Table S3: K proportion test results regarding the incidence of internal disorders (Marascuilo procedure comparison). Table S4: Descriptive statistics of the flesh mineral composition of Hass avocados at the RTE (ready to eat) stage.

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Formal analysis, Writing—original draft. J. V.: Methodology, Writing—original draft. A. R.: Conceptualization, Methodology, Funding acquisition, Writing—original draft. All authors have read and agreed to the published version of the manuscript.

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Conflicts of Interest:

The authors declare no conflict of interest. The funders had no role in the design of the study; in the collection, analyses, or interpretation of data; in the writing of the manuscript; or in the decision to publish the results.

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