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# Altitude Modulates the Effect of Agricultural Management on Phenolic Compounds, Caffeine, and Cup Quality of Roasted Coffee (*Coffea arabica* L.) from Veracruz, Mexico

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## Abstract

Coffee quality results from the interplay between genotype, environment, and cultivation practices, but whether the effect of agricultural management depends on altitude remains poorly understood. We hypothesized that the response of roasted coffee chemistry and cup quality to management would differ among altitudinal zones. Composite samples representative of sustainable and conventional management in three altitudinal zones (low, <900; mid, 900–1100; high, >1100 m.a.s.l.) were obtained from eighteen commercial *Coffea arabica* L. farms in Veracruz, Mexico (management monitored in 2023–2024 and 2024–2025; cherries sampled in the 2024–2025 harvest), and their physicochemical properties, phenolic compounds, caffeine, in vitro antioxidant capacity, and Specialty Coffee Association (SCA) cup quality were evaluated. Altitude was the main source of variation in pH, total phenolic and flavonoid contents, and total cup score. In contrast, management mainly affected chlorogenic acids (CGA), caffeine, and hydrolyzable tannins, which were higher under sustainable management (CGA: 22.52 vs. 16.82 mg $g^{-1}$). The management × altitude interaction was significant ($p \leq 0.017$) for all phenolic compounds, caffeine, and antioxidant assays, with the largest differences between systems in the mid zone, where sustainable samples showed the highest CGA (30.27 mg $g^{-1}$) and FRAP values (675.30 µmol Trolox equivalents $g^{-1}$). All samples reached specialty grade (82.08–84.75 points); total cup score increased with altitude and did not differ between management systems. These results indicate that management effects on coffee chemistry depend on altitude.

**Keywords:** antioxidant capacity; chlorogenic acids; SCA cupping; specialty coffee; sustainable management

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## 1. Introduction

Coffee is one of the most consumed beverages worldwide and an agricultural product with significant economic and social relevance. More than 25 million families depend on its

production, with over 160 million bags commercialized annually. Brazil, Vietnam, Indonesia, and Colombia lead global production, while Mexico is the world's ninth-largest coffee producer [1]. Its popularity stems from a combination of sensory qualities, stimulating effects, and antioxidant properties, driven by a complex chemical profile that includes chlorogenic acids, sugars, trigonelline, lipids, caffeine, and other metabolites that shape flavor, aroma, mouthfeel, and balance [2]. Caffeine, the main coffee alkaloid, exerts stimulating effects by blocking adenosine receptors in the central nervous system [3]. At the same time, phenolic compounds such as chlorogenic acids, caffeic acid, ferulic acid, p-coumaric acid, quercetin, and tannins contribute to the prevention of chronic diseases through their antioxidant, anti-inflammatory, antipyretic, and antineoplastic properties [4,5].

These metabolites activate antioxidant pathways such as Nrf2 and HO1, reinforcing their potential for health benefits [6–8]. The chemical composition of coffee, and thus its quality, depends on factors such as variety, geographical origin, agricultural management, and environmental conditions [9]. *Coffea arabica* L. is the most appreciated species for its sensory complexity, attributed to its allotetraploid genome, which allows broad regulation of metabolic pathways that favor the accumulation of caffeine, lipids, terpenes, and chlorogenic acids, key compounds in the sensory and functional profile of coffee [10,11]. Among the environmental factors, altitude is one of the most decisive for coffee quality [12]. At higher altitudes, the decrease in temperature modifies photosynthetic and transpiration rates [13], which induces slower fruit ripening and favors the accumulation of sucrose, fatty acids, and volatile compounds that intensify attributes such as aroma, taste, mouthfeel, acidity, and sweetness [14,15].

In contrast, low altitude zones with elevated temperatures accelerate ripening, reduce desirable flavor precursors, increase compounds associated with green or green earthy notes, and increase the incidence of sensory defects [16]. Agricultural management also influences coffee quality. Conventional management, based on synthetic fertilizers and pesticides, increases the availability of nitrogen and phosphorus, stimulating the expression of methyltransferase genes and favoring the biosynthesis of caffeine and flavonoids such as quercetin [7,8].

As a result, medium-light roasted coffees under conventional management have shown elevated levels of caffeine, phenols, flavonoids, and chlorogenic acids [17]. However, evidence also supports a superior bioactive profile in coffees produced under sustainable management. Studies report significantly higher concentrations of phenolic acids and flavonoids in sustainable systems [18], attributable to conditions such as increased organic matter, active soil microbiota, UV-shading, optimized water balance, and careful post-harvest handling factors that favor the synthesis and conservation of bioactive metabolites [19–21]. In addition, sustainable management brings environmental and social benefits such as conservation of biodiversity, use of organic inputs, soil and water protection, fair trade, and access to differentiated markets [22]. In contrast, conventional management can lead to erosion, soil acidification, changes in the soil microbial community, and water pollution.

Despite these advances, the interaction between altitude and management systems remains poorly resolved. Previous work has typically examined altitude and management as independent factors or assessed them within a single production system, so few studies have tested, as an explicit interaction term, the extent to which altitude modulates bean chemistry responses to management practice. Moreover, studies that integrate physicochemical composition, bioactive profile, antioxidant capacity, and certified sensory grading on the same set of commercial farms are scarce, and Mesoamerican mid-altitude coffee (below 1400 m.a.s.l.) is markedly under-represented in this literature relative to Andean, Brazilian, and East African origins. This absence is particularly relevant for the state of Veracruz, one

of the main coffee-growing regions, recognized for its high yields, agroecological diversity, and cup quality, which makes it a national benchmark for *Coffea arabica* L. production.

Accordingly, this study aimed to evaluate the effect of agricultural management (sustainable and conventional) in three altitudinal zones (low, medium, and high) of the state of Veracruz, Mexico, on the physicochemical properties, phenolic compounds, caffeine, in vitro antioxidant capacity, and sensory quality of roasted coffee (*Coffea arabica* L.). We hypothesized that bean chemistry and cup quality would respond differently to agricultural management across altitudinal zones, meaning we expected a significant management × altitude interaction.

## 2. Materials and Methods

### *2.1. Plant Material, Management System and Production Area*

We conducted the study at eighteen commercial *Coffea arabica* L. farms in the coffee-producing region of Veracruz, Mexico, distributed across three altitudinal zones: low zone (<900 m.a.s.l.), mid zone (900–1100 m.a.s.l.), and high zone (>1100 m.a.s.l.). Within each altitudinal zone, six farms were included, three managed under a sustainable system and three under a conventional system, yielding three farms per management × altitude combination for a total of eighteen farms across the study area (Table 1). Plant selection criteria were standardized across all sites: only five-year-old plants of cultivar Colombia, planted at a density of 2 × 1.5 m and exhibiting similar canopy diameter, were eligible for inclusion. Farms under the sustainable system had been managed under certified organic practices for an average of 12 years, whereas the conventional farms had been under uninterrupted conventional management over a comparable period; both systems therefore represented long-established production conditions rather than recent conversions. Farms were selected according to the following criteria: (i) certified organic or uninterrupted conventional management for at least ten years; (ii) plantations of cultivar Colombia of the same age and planting density; and (iii) location within the elevation range of the corresponding altitudinal zone. To verify that each farm consistently followed its declared management system, we monitored agronomic practices over two consecutive production cycles (2023–2024 and 2024–2025) through farm visits and producer records. Coffee cherries were sampled only during the second cycle (2024–2025 harvest; November 2024–February 2025).

#### 2.1.1. Management System

Sustainable management: Sustainable management was characterized by the absence of synthetic fertilizers or chemical inputs. Nutrition was provided exclusively through compost prepared on-farm by the producers with coffee pulp, goat manure, zeolite, dolomitic lime, and phosphoric rock, materials locally available in the region and permitted under organic certification standards, with a nutritional composition of 35% organic matter, 1.1% nitrogen (N), 1% phosphorus (P), 2% potassium (K), and 1.8% magnesium (Mg). Compost was applied at 2 kg per plant per season, with timing adjusted by altitudinal zone: February for low-zone farms and March for mid- and high-zone farms, in each monitored cycle (2023 and 2024), to align with the crop's phenological stage at each elevation. Weed control was performed manually four times per year (January, March, July, and October). We pruned coffee plants and shade trees in February of each monitored cycle (2023 and 2024).

Conventional management: Nutrition was provided through a commercial NPK fertilizer (20-10-20; Yara, Isla Veracruz, Mexico), applied in two doses of 120 g per plant in June and October of each monitored cycle (2023 and 2024). Weed control was performed manually twice per year (February and August). Pruning was carried out in February of each monitored cycle (2023 and 2024). In both systems, workers selectively hand-harvested fully ripe coffee cherries.

**Table 1.** Characterization of the eighteen *Coffea arabica* L. farms included in the study, distributed across altitudinal zones and management systems in Veracruz, Mexico.

| Farm | Management | Zone | Area (ha) | Altitude (m.a.s.l.) | Latitude (°N) | Longitude (°W) |
|---|---|---|---|---|---|---|
| 1 | Sustainable | High | 2.50 | 1378 | 19.164444 | −96.985833 |
| 2 | Sustainable | High | 2.00 | 1343 | 19.158056 | −96.960278 |
| 3 | Sustainable | High | 2.5 | 1335 | 19.1591800 | −96.9602900 |
| 4 | Sustainable | Mid | 0.50 | 1014 | 19.307500 | −96.920556 |
| 5 | Sustainable | Mid | 2.00 | 1008 | 19.310278 | −96.922778 |
| 6 | Sustainable | Mid | 3.00 | 954 | 19.318056 | −96.911667 |
| 7 | Sustainable | Low | 3.00 | 885 | 19.146111 | −96.847778 |
| 8 | Sustainable | Low | 2.00 | 882 | 19.101353 | −96.855844 |
| 9 | Sustainable | Low | 1.50 | 876 | 19.099872 | −96.857458 |
| 10 | Conventional | High | 4.00 | 1362 | 19.159392 | −96.985880 |
| 11 | Conventional | High | 1.00 | 1334 | 19.144444 | −96.975556 |
| 12 | Conventional | High | 0.25 | 1300 | 19.168333 | −96.950000 |
| 13 | Conventional | Mid | 1.00 | 1084 | 19.124167 | −96.910833 |
| 14 | Conventional | Mid | 1.00 | 1083 | 19.124444 | −96.910833 |
| 15 | Conventional | Mid | 1.09 | 973 | 19.102500 | −96.915000 |
| 16 | Conventional | Low | 1.00 | 799 | 19.328431 | −96.859686 |
| 17 | Conventional | Low | 0.50 | 799 | 19.333636 | −96.858530 |
| 18 | Conventional | Low | 1.00 | 767 | 19.067314 | −96.815186 |

#### 2.1.2. Coffee-Growing Region and Altitudinal Conditions

We defined zone boundaries based on elevation alone, following the altitudinal strata conventionally used in Mexican coffee production agroecology; we used the Köppen–Geiger climate classification [23] only to describe each zone's climate, not to delimit the altitudinal strata. We unambiguously assigned all eighteen farms to their respective zones based on GPS-recorded altitude measurements.

Low zone (<900 m.a.s.l.): Six farms were located in this zone, three under sustainable management (farms 7–9; 876–885 m.a.s.l.; 19.100–19.146° N, 96.848–96.857° W) and three under conventional management (farms 16–18; 767–799 m.a.s.l.; 19.067–19.334° N, 96.815–96.860 °W). The area is characterized by Aw, Cwa, and Cfa climates, with mean annual temperatures of 18–26 °C and annual precipitation of 900–2100 mm. The predominant soils are Leptosol (70.44%), Andosol (13.52%), Luvisol (10.67%), Vertisol (2.79%), and Phaeozem (2.08%).

Middle zone (900–1100 m.a.s.l.): Six farms were located in this zone, three under sustainable management (farms 4–6; 954–1014 m.a.s.l.; 19.308–19.318° N, 96.912–96.923° W) and three under conventional management (farms 13–15; 973–1084 m.a.s.l.; 19.103–19.124° N, 96.910–96.915° W). The climate is Aw and Cwa, with mean annual temperatures of 20–26 °C and annual precipitation of 1100–1600 mm. The predominant soils are Leptosol (65.28%), Luvisol (28.06%), Phaeozem (6.02%), and Vertisol (0.33%).

High zone (>1100 m.a.s.l.): Six farms were located in this zone, three under sustainable management (farms 1–3; 1335–1378 m.a.s.l.; 19.158–19.164° N, 96.960–96.986° W) and three under conventional management (farms 10–12; 1300–1362 m.a.s.l.; 19.144–19.168° N, 96.950–96.986° W). The climate is Cfa and Aw, with mean annual temperatures of 16–26 °C and annual precipitation of 1100–1600 mm. The predominant soils are Andosol (44.28%), Luvisol (42.88%), and Leptosol (9.87%).

### 2.2. Experimental Design

A 2 × 3 factorial arrangement was adopted, with management system (sustainable, S; and conventional, C) and altitudinal gradient (low, L; mid, M; and high, H) as fixed factors, yielding six factor-level combinations (SL, SM, SH, CL, CM, CH).

Within each farm, five representative plants were randomly selected and spatially distributed across the entire farm surface. Each selected plant met the same morphological criteria (canopy diameter, stem diameter, plant age, and cultivar). We harvested all ripe cherries from each of the five selected plants separately at commercial maturity (full red color) during the 2024–2025 harvest (November 2024–February 2025). For each plant, three independent subsamples were obtained from different canopy positions (upper, middle, and lower thirds), yielding 15 subsamples per farm (5 plants × 3 positions). The subsamples from the three farms of each management × altitude combination (45 subsamples) were pooled to obtain a composite sample representative of each production condition, analogous to a commercial lot. All analytical determinations were performed in triplicate on each composite sample.

### 2.3. Sample Processing

Postharvest processing followed a standard two-stage wet-dry method. In the wet processing stage, cherries were pulped on the day of harvest using a disc pulper (10 kg capacity). Natural fermentation removed mucilage for 24 h at ambient temperature, followed by thorough washing with pressurized potable water. Beans were then dried in the shade to a moisture content of 12% (w.b.), verified with a grain moisture meter. In the dry processing stage, we weighed parchment coffee (250 g per replicate) and hulled it using a mechanical coffee huller. The resulting green coffee beans were visually selected following [23] quality standards, packed in sealed airtight bags, and stored at room temperature in a dry environment until roasting.

We roasted the beans in a drum roaster (BYZ-4-L550, Nanyang Dongyi Machinery Equip. Co., Ltd., Nanyang, China) at 180 °C for 6 min, reaching a medium roast level confirmed by visual color assessment against the Agtron roast color reference scale (Agtron Inc., Reno, NV, USA). The beans were immediately air-cooled to room temperature and degassed for 24 h before grinding and laboratory analysis.

### 2.4. Determination of Physicochemical Properties

The physicochemical properties of roasted coffee were determined from 10 g of ground sample. The pH was measured following the methodology described by Huang et al. [24], using a pH meter (OAKTON, pH550, Cole-Parmer, Vernon Hills, IL, USA). The titratable acidity was quantified according to AOAC International [25] method 942.15, by titration with NaOH (0.1 N) and using bromothymol blue (1%) as an indicator; results were expressed as chlorogenic acid equivalents (% CGA eq), since chlorogenic acids are among the main acid compounds in roasted coffee. Color was evaluated with a colorimeter (CM508d, Minolta Camera Co., Ltd., Osaka, Japan), and the L*, a*, and b*values were used to calculate chroma (C*) and hue angle (h°).

### 2.5. Determination of Phenolic Compounds, Tannins and Caffeine

All reagents were of analytical grade (≥98% purity) and were purchased from Sigma-Aldrich (St. Louis, MO, USA). Extracts were prepared from 10 g of ground roasted coffee with methanol 80% ($v\ v^{-1}$) by ultrasound-assisted extraction (Ultrasonic Cleaner 32V118A, Ultrasonic Power Corporation, Freeport, IL, USA) for 30 min, followed by centrifugation at 10,000× $g$ for 10 min. Total phenolic content was determined by the Folin–Ciocalteu method described by Singleton et al. [26]. Absorbance was measured at 725 nm on a UV/Visible

spectrophotometer (GENESYS 150, Thermo Fisher Scientific, Waltham, MA, USA). The results were expressed as milligrams of gallic acid equivalents per gram of dry weight (mg GAE $g^{-1}$ DW). The flavonoid content was quantified based on the formation of the $AlCl_3 \cdot 6H_2O$ complex [27]. Results were expressed as milligrams of quercetin equivalents per gram of dry weight (mg QE $g^{-1}$ DW). Hydrolyzable tannins were quantified according to Willis [28] using 0.5 mL of sample and 2.5 mL of 2.5% potassium iodate; absorbance was recorded at 550 nm; results were expressed as milligrams of tannic acid equivalent per gram of dry weight (mg TAE $g^{-1}$ DW). Condensed tannins were determined using the vanillin assay described by Wu et al. [29] at 4% by measuring absorbance at 500 nm, and the results were expressed in milligrams of catechin equivalents per gram of dry weight (mg CE $g^{-1}$ DW). Caffeine and chlorogenic acids (CGA) were determined using the methodology of Atlabachew et al. [30]. After acetonitrile extraction, the upper phase was diluted 1:20 ($v/v$) and used for subsequent spectrophotometric readings at 274 nm (caffeine) and 325 nm (CGA), with results expressed in mg $g^{-1}$. Quantification was performed using external calibration curves prepared with caffeine and chlorogenic acid standards ($R^2 = 0.9973$ and $R^2 = 0.9957$, respectively).

### *2.6. Determination of Antioxidant Activity*

Antioxidant activity was evaluated using three complementary methods: $DPPH^{\bullet}$ radical scavenging activity [31], reduction of the $ABTS^{\bullet+}$ radical cation according to [32] and the ferric reducing antioxidant power (FRAP) assay according to Benzie & Strain [33]. Absorbance readings were recorded on the same UV/Visible spectrophotometer described in Section 2.5 (GENESYS 150, Thermo Fisher Scientific, Waltham, MA, USA) at 515 nm (DPPH), 734 nm (ABTS) and 593 nm (FRAP). The results were expressed as micromoles of Trolox equivalents per gram of dry weight (µmol TE $g^{-1}$ DW). All assays were performed in triplicate.

### *2.7. Sensory Evaluation Procedure*

The sensory quality of roasted coffee was evaluated by a panel of three Q-Graders certified by the Coffee Quality Institute (CQI) under the standards of the Specialty Coffee Association (SCA) [23]. Samples were presented in randomized and blinded order using three-digit codes to prevent bias. Each evaluator independently assessed five cups per sample following the SCA Cupping Protocol [23] fragrance/aroma, flavor, aftertaste, acidity, body, balance, and overall impression were scored from 6 to 10 points in 0.25-point increments, whereas uniformity, clean cup, and sweetness were scored by assigning 2 points per acceptable cup (maximum 10 points). For each cup, we extracted medium-roasted ground coffee (8.25 g) with 150 mL of water at 93 °C. The cupping session was conducted in a standardized environment free of interfering odors, at a controlled room temperature of 20–25 °C.

The sensory evaluation was conducted in accordance with the ethical principles of the Declaration of Helsinki [34]. Participation was voluntary, and all Q-Graders provided informed verbal consent before the sessions and could withdraw at any time. Verbal rather than written consent was obtained because the evaluation was a no-risk professional grading of a food product and no personal data were collected. In coffee cupping, the evaluators act as trained judges of product quality rather than as research subjects; no personal, physiological, or health-related information about them was collected, and the samples were commercially acceptable food products [35]. Under Mexican regulations, this activity corresponds to research without risk (Article 17, Section I) [36].

### *2.8. Statistical Analysis*

Data were analyzed by two-way ANOVA with management systems, altitudinal zone, and their interaction as fixed factors, using the analytical replicates (n = 3) of each composite sample; sensory scores were analyzed using the Q-Grader as a blocking factor (n = 3 evaluators). F statistics and $p$ values are reported in Table S1. Normality and homoscedasticity of residuals were verified with the Shapiro–Wilk and Levene tests; slight departures from normality were found only for total flavonoid content and overall impression ($p = 0.039$ and 0.006), to which ANOVA is robust given the balanced design. Means were compared by Tukey's HSD test ($p \leq 0.05$). We applied PCA to the standardized sensory attributes and RDA to the standardized bioactive compounds, constrained by management systems and mean elevation, with significance tested by 999 permutations. All analyses were performed in Python 3.12 (statsmodels 0.15, SciPy 1.17, and NumPy 2.4), and figures were generated with Matplotlib 3.10.

## 3. Results and Discussion

### *3.1. Physicochemical Properties*

Table 2 shows the effects of management system and altitudinal zone on pH, titratable acidity (TA), and color parameters. Management affected only TA, which was higher in the conventional samples (8.94 vs. 8.12% CGA eq.; $p = 0.022$). A possible explanation is the higher nitrogen supply under NPK fertilization, although this was not assessed in the present study; TA is a key chemical descriptor associated with perceived sourness in coffee [37]. Altitude had a stronger effect (Table S1): the mid-altitude samples showed the highest TA (9.95% CGA eq.) and the lowest pH (4.77), whereas the low-altitude samples showed the lowest TA (7.24% CGA eq.). Higher temperatures at low altitude accelerate fruit respiration and ripening, which has been associated with lower organic acid contents in the bean [38,39].

**Table 2.** Effect of agricultural management system, altitudinal zone and their interaction on the physicochemical properties in coffee beans (*Coffea arabica* L.).

| Factor | Level | pH | TA (%CGA eq.) | Color | | |
|---|---|---|---|---|---|---|
| | | | | L* | C* | h° (°) |
| Management system (M) | S | 5.05 ± 0.25 $^{a}$ | 8.12 ± 0.99 $^{b}$ | 41.51 ± 0.79 $^{a}$ | 5.56 ± 0.81 $^{a}$ | 43.21 ± 1.88 $^{a}$ |
| | C | 5.00 ± 0.18 $^{a}$ | 8.94 ± 1.63 $^{a}$ | 41.77 ± 0.97 $^{a}$ | 6.06 ± 1.47 $^{a}$ | 44.36 ± 3.24 $^{a}$ |
| Altitudinal zone (A) | L | 5.21 ± 0.14 $^{a}$ | 7.24 ± 0.29 $^{c}$ | 41.04 ± 0.80 $^{a}$ | 4.85 ± 0.87 $^{b}$ | 41.49 ± 2.14 $^{b}$ |
| | M | 4.77 ± 0.05 $^{b}$ | 9.95 ± 1.06 $^{a}$ | 41.95 ± 0.97 $^{a}$ | 6.70 ± 1.21 $^{a}$ | 45.41 ± 1.98 $^{a}$ |
| | H | 5.10 ± 0.05 $^{a}$ | 8.41 ± 0.92 $^{b}$ | 41.92 ± 0.59 $^{a}$ | 5.88 ± 0.66 $^{ab}$ | 44.44 ± 2.26 $^{ab}$ |
| M × A | SL | 5.30 ± 0.17 $^{a}$ | 7.09 ± 0.31 $^{c}$ | 41.04 ± 1.17 $^{a}$ | 5.12 ± 1.13 $^{ab}$ | 42.11 ± 2.68 $^{a}$ |
| | SM | 4.76 ± 0.01 $^{b}$ | 9.08 ± 0.26 $^{ab}$ | 42.02 ± 0.37 $^{a}$ | 6.12 ± 0.42 $^{ab}$ | 44.79 ± 0.20 $^{a}$ |
| | SH | 5.09 ± 0.04 $^{a}$ | 8.18 ± 0.91 $^{bc}$ | 41.47 ± 0.49 $^{a}$ | 5.43 ± 0.64 $^{ab}$ | 42.71 ± 0.97 $^{a}$ |
| | CL | 5.13 ± 0.01 $^{a}$ | 7.39 ± 0.23 $^{bc}$ | 41.05 ± 0.47 $^{a}$ | 4.57 ± 0.63 $^{b}$ | 40.87 ± 1.76 $^{a}$ |
| | CM | 4.78 ± 0.07 $^{b}$ | 10.81 ± 0.69 $^{a}$ | 41.88 ± 1.48 $^{a}$ | 7.28 ± 1.57 $^{a}$ | 46.03 ± 2.94 $^{a}$ |
| | CH | 5.11 ± 0.06 $^{a}$ | 8.63 ± 1.07 $^{bc}$ | 42.38 ± 0.05 $^{a}$ | 6.33 ± 0.31 $^{ab}$ | 46.17 ± 1.67 $^{a}$ |
| P (M) | | 0.248 | 0.022 | 0.523 | 0.256 | 0.234 |
| P (A) | | <0.001 | <0.001 | 0.142 | 0.012 | 0.012 |
| P (M × A) | | 0.084 | 0.163 | 0.516 | 0.284 | 0.155 |

Values are mean ± SD (n = 9 for management system, n = 6 for altitudinal zone, and n = 3 analytical replicates of each composite sample for management × altitude combinations). Within each column and factor, different superscript letters indicate significant differences (Tukey's HSD, $p \leq 0.05$). $p$ values from two-way ANOVA. TA: titratable acidity expressed as chlorogenic acid equivalents; L*: lightness; C*: chroma; h°: hue angle. S: Sustainable; C: Conventional; L: Low; M: Mid; H: High; SL: Sustainable × Low; SM: Sustainable × Mid; SH: Sustainable × High; CL: Conventional × Low; CM: Conventional × Mid; CH: Conventional × High.

Altitude also affected color: the mid-altitude samples showed the highest chroma (C* = 6.70) and angle (h° = 45.41°), whereas the low-altitude samples showed the lowest values (4.85 and 41.49°, respectively); lightness (L*) did not differ among treatments. Since all samples were roasted under the same conditions, these differences likely reflect variations in the precursors of Maillard reactions and caramelization, such as sugars, free amino acids, and CGA, which determine melanoidin formation and the color of roasted coffee [40,41]. The management × altitude interaction was not significant for pH, TA, or color parameters ($p$ = 0.084–0.516; Table S1), indicating that these properties were mainly determined by altitude.

### 3.2. Total Phenolic Content, Flavonoids and Chlorogenic Acids

Total phenols, flavonoids, and chlorogenic acids (CGAs) differed significantly between treatments (Table 3). The conventional samples showed slightly higher total phenolic (38.59 vs. 36.99 mg GAE $g^{-1}$) and flavonoid contents (27.25 vs. 24.41 mg QE $g^{-1}$) than the sustainable samples. A plausible but untested explanation is the higher nitrogen supply under NPK fertilization (20-10-20), since nitrogen availability has been associated with stimulation of phenylalanine ammonia-lyase (PAL) activity, a key enzyme of the phenylpropanoid pathway, in other crops [16,42].

**Table 3.** Effect of agricultural management system, altitudinal zone, and their interaction on total phenolic content, total flavonoid content, chlorogenic acids, tannins, and caffeine in roasted coffee (*Coffea arabica* L.).

| Factor | Level | TPC (mg GAE $g^{-1}$ DW) | TFC (mg QE $g^{-1}$ DW) | CGA (CGA mg $g^{-1}$ DW) | CT (mg CE $g^{-1}$ DW) | HT (mg TAE $g^{-1}$ DW) | Caffeine (mg $g^{-1}$ DW) |
|---|---|---|---|---|---|---|---|
| Management system (M) | S | 36.99 ± 5.43 $^{b}$ | 24.41 ± 9.65 $^{b}$ | 22.52 ± 8.30 $^{a}$ | 21.01 ± 4.53 $^{a}$ | 26.22 ± 2.94 $^{a}$ | 28.92 ± 6.45 $^{a}$ |
| | C | 38.59 ± 2.22 $^{a}$ | 27.25 ± 6.82 $^{a}$ | 16.82 ± 3.45 $^{b}$ | 20.41 ± 2.15 $^{a}$ | 18.15 ± 2.29 $^{b}$ | 19.52 ± 6.55 $^{b}$ |
| Altitudinal zone (A) | L | 37.76 ± 1.68 $^{b}$ | 27.65 ± 5.13 $^{b}$ | 12.31 ± 1.39 $^{b}$ | 22.93 ± 3.90 $^{a}$ | 24.68 ± 4.73 $^{a}$ | 18.26 ± 2.96 $^{c}$ |
| | M | 42.14 ± 1.59 $^{a}$ | 33.66 ± 3.01 $^{a}$ | 24.54 ± 6.42 $^{a}$ | 17.31 ± 1.37 $^{b}$ | 21.47 ± 6.06 $^{b}$ | 24.93 ± 11.13 $^{b}$ |
| | H | 33.47 ± 2.65 $^{c}$ | 16.18 ± 2.47 $^{c}$ | 22.17 ± 3.82 $^{a}$ | 21.88 ± 1.42 $^{a}$ | 20.40 ± 3.14 $^{b}$ | 29.47 ± 2.37 $^{a}$ |
| M × A | SL | 36.31 ± 0.46 $^{d}$ | 23.12 ± 1.78 $^{c}$ | 11.93 ± 1.92 $^{d}$ | 26.09 ± 2.66 $^{a}$ | 28.81 ± 1.22 $^{a}$ | 20.72 ± 1.29 $^{c}$ |
| | SM | 43.56 ± 0.27 $^{a}$ | 36.07 ± 0.96 $^{a}$ | 30.27 ± 1.13 $^{a}$ | 16.18 ± 0.83 $^{d}$ | 26.91 ± 1.28 $^{ab}$ | 35.04 ± 0.80 $^{a}$ |
| | SH | 31.09 ± 0.52 $^{e}$ | 14.05 ± 1.16 $^{e}$ | 25.38 ± 0.53 $^{b}$ | 20.75 ± 0.71 $^{bc}$ | 22.94 ± 2.13 $^{bc}$ | 31.00 ± 0.72 $^{ab}$ |
| | CL | 39.21 ± 0.72 $^{c}$ | 32.18 ± 0.96 $^{b}$ | 12.70 ± 0.83 $^{d}$ | 19.77 ± 1.00 $^{bc}$ | 20.55 ± 1.84 $^{cd}$ | 15.81 ± 1.49 $^{d}$ |
| | CM | 40.72 ± 0.44 $^{b}$ | 31.26 ± 2.10 $^{b}$ | 18.81 ± 1.84 $^{c}$ | 18.45 ± 0.41 $^{cd}$ | 16.03 ± 1.12 $^{e}$ | 14.81 ± 1.52 $^{d}$ |
| | CH | 35.85 ± 0.53 $^{d}$ | 18.31 ± 0.56 $^{d}$ | 18.96 ± 2.32 $^{c}$ | 23.02 ± 0.84 $^{ab}$ | 17.86 ± 0.90 $^{de}$ | 27.95 ± 2.57 $^{b}$ |
| P (M) | | <0.001 | <0.001 | <0.001 | 0.349 | <0.001 | <0.001 |
| P (A) | | <0.001 | <0.001 | <0.001 | <0.001 | <0.001 | <0.001 |
| P (M × A) | | <0.001 | <0.001 | <0.001 | <0.001 | 0.017 | <0.001 |

Values are mean ± SD (n = 9 for management system, n = 6 for altitudinal zone, and n = 3 analytical replicates of each composite sample for management × altitude combinations). Within each column and factor, different superscript letters indicate significant differences (Tukey's HSD, $p \leq 0.05$). $p$ values from two-way ANOVA. TPC: total phenolic content; TFC: total flavonoid content; CGA: chlorogenic acids; CT: condensed tannins; HT: hydrolyzable tannins; GAE: gallic acid equivalents; QE: quercetin equivalents; CE: catechin equivalents; TAE: tannic acid equivalents; DW: dry weight. S: Sustainable; C: Conventional; L: Low; M: Mid; H: High; SL: Sustainable × Low; SM: Sustainable × Mid; SH: Sustainable × High; CL: Conventional × Low; CM: Conventional × Mid; CH: Conventional × High.

In contrast, CGAs were 33.9% higher in the sustainable samples (22.52 vs. 16.82 mg $g^{-1}$). Although CGAs, flavonoids, and other phenolics share the phenylpropanoid pathway, they do not necessarily respond uniformly to nitrogen availability; in other plant systems, a higher nitrogen supply has been associated with a preferential allocation toward the flavonoid branch [43], and in coffee, PAL isoforms with different roles in CGA and flavonoid accumulation have been described [44]. Whether these mechanisms operate under the

field conditions of this study remains to be tested. Total phenolic and CGA contents were comparable to those reported by Liao et al. [45] for medium-light roasted Arabica coffee from Brazil, Ethiopia, and Mexico.

Altitude strongly affected these compounds. The mid-altitude samples showed the highest total phenolic (42.14 mg GAE $g^{-1}$) and flavonoid contents (33.66 mg QE $g^{-1}$), followed by the low-altitude samples, whereas the high-altitude samples showed the lowest values (33.47 mg GAE $g^{-1}$ and 16.18 mg QE $g^{-1}$), 20.57% and 51.93% lower than in the mid zone. Similar peaks of phenolic compounds at intermediate elevations have been reported by Zaman et al. [46] and attributed to moderate temperatures and radiation levels.

CGAs showed a different pattern: it was similarly high in the mid (24.54 mg $g^{-1}$) and high zones (22.17 mg $g^{-1}$) and lowest in the low zone (12.31 mg $g^{-1}$). Lower temperatures and higher UV radiation at higher elevations have been associated with the accumulation of defense-related phenolics, including CGA [47]. These CGA contents were similar to those reported by Rusinek et al. [48] for Arabica coffees from Peru, Costa Rica, Guatemala, and Ethiopia.

The management × altitude interaction was significant for total phenolics, flavonoids, and CGAs ($p < 0.001$; Table S1), indicating that the effect of management differed among altitudinal zones. The sustainable × mid-altitude samples had the highest total phenolic and flavonoid contents (SM: 43.56 mg GAE $g^{-1}$ and 36.07 mg QE $g^{-1}$). It is possible that the intermediate conditions of this zone favored the response to organic matter inputs, which release nutrients gradually and stimulate soil microbial activity [20,21] however, this hypothesis requires confirmation in studies including farm-level replication.

In contrast, in the low and high zones, conventional samples showed higher total phenolic and flavonoid contents than sustainable samples (Table 3); hence, the management effect changed with altitude. For CGA, sustainable samples showed higher values in the mid (SM: 30.27 mg $g^{-1}$) and high zones (SH: 25.38 mg $g^{-1}$), whereas the low zone showed no differences between management systems (11.93–12.70 mg $g^{-1}$). Thus, the largest management-related difference in CGAs occurred at mid altitude, a pattern that may be related to the moderate temperatures and UV radiation of this zone [46]. In the low zone, conventional farms were located at slightly lower elevations than sustainable farms (767–799 vs. 876–885 m.a.s.l.; Table 1), which may have contributed to the differences observed between management systems in this zone.

### *3.3. Content of Hydrolyzable and Condensed Tannins*

Tannins are water-soluble polyphenols able to form complexes with proteins and polysaccharides, and they act as defense compounds against herbivores and pathogens [49]. In coffee, condensed (CT) and hydrolyzable (HT) tannins have been associated with antimicrobial and antioxidant activities [50]. CT did not differ between management systems (21.01 vs. 20.40 mg CE $g^{-1}$; $p = 0.349$), suggesting that their accumulation is less sensitive to nitrogen availability [42]. In contrast, HT were higher in the sustainable samples (26.22 vs. 18.15 mg TAE $g^{-1}$). A plausible but untested explanation is the high C/N ratio of organically managed systems, which in other species has been associated with greater allocation of carbon to gallic acid-derived compounds [13].

Altitude also affected tannin contents. CT were highest in the low (22.93 mg CE $g^{-1}$) and high zones (21.88 mg CE $g^{-1}$) and lowest in the mid zone (17.31 mg CE $g^{-1}$), whereas HT were highest in the low zone (24.68 mg TAE $g^{-1}$). Higher temperatures and herbivore pressure at low elevations, and lower temperatures and higher UV radiation at high elevations, have been associated with the accumulation of defense-related polyphenols, including tannins [42,49]. In the mid zone, where CGAs and flavonoids reached their highest values, CT showed its lowest values, suggesting a different allocation among phenolic classes under

these conditions. The interaction was significant for both tannin types (Table S1): the highest CT and HT contents were found in the sustainable × low-altitude samples (SL: 26.09 mg CE $g^{-1}$ and 28.81 mg TAE $g^{-1}$), while the lowest values corresponded to SM for CT (16.18 mg CE $g^{-1}$) and to CM for HT (16.03 mg TAE $g^{-1}$).

### *3.4. Caffeine Content*

Caffeine contents ranged from 14.81 to 35.04 mg $g^{-1}$ (Table 3). A value of 17.19 mg $g^{-1}$ has been reported for medium-roasted coffee by Atlabachew et al. [30], close to the lowest values found in this study (CM and CL), Duangjai et al. [51] reported 27.65 mg $g^{-1}$ in medium-roasted Arabica coffee. The sustainable samples showed 48.15% more caffeine than the conventional samples (28.92 vs. 19.52 mg $g^{-1}$). Viana et al. [20] reported that shaded, organically managed systems generate microenvironments with moderate physiological stress and greater microbial activity, conditions that have been associated with caffeine accumulation. Because we quantified caffeine by UV spectrophotometry, we cannot completely rule out a partial contribution of other UV-absorbing compounds, such as CGA, to the measured values.

Caffeine increased with altitude (low: 18.26; mid: 24.93; high: 29.47 mg $g^{-1}$), a pattern that has been attributed to lower temperatures and slower fruit maturation at high elevations [52]. The high-altitude farms (1300–1378 m.a.s.l.) were located approximately 500 m above the low-altitude farms (767–885 m.a.s.l.), which, considering a typical decrease in air temperature of about 0.6 °C per 100 m of elevation [53] corresponds to mean temperatures roughly 3 °C lower. The interaction followed a pattern similar to CGA: sustainable × mid-altitude samples showed the highest caffeine content (SM: 35.04 mg $g^{-1}$), and the largest difference between management systems occurred in the mid zone. Caffeine and CGAs were strongly correlated ($r = 0.82$; Figure 1C), which agrees with reports of co-regulation of both compounds in coffee [54,55].

### *3.5. Antioxidant Activity*

Management significantly affected the three assays (Figure 1A). Conventional samples showed higher DPPH (526.88 ± 20.32 µmol TE $g^{-1}$ DW) and ABTS values (668.78 ± 20.39 µmol TE $g^{-1}$ DW), consistent with their higher total phenolic and flavonoid contents, which correlated most strongly with these assays (Figure 1C). In contrast, the sustainable samples showed the highest FRAP value (566.29 ± 83.30 µmol TE $g^{-1}$ DW), and FRAP was positively correlated with caffeine and CGA, which were higher in these samples. Caffeine and CGAs can form π-π stacking complexes that are stable in aqueous solutions at pH 3–5 [56]; since the FRAP assay is performed at pH 3.6, the formation of these complexes may have contributed to the reducing activity measured [57], although this was not directly evaluated.

Regarding the altitudinal gradient (Figure 1A), all three assays differed significantly among zones ($p < 0.001$). Mid-altitude samples showed the highest antioxidant capacity (DPPH: 545.17 ± 6.89; ABTS: 681.29 ± 18.80; FRAP: 594.47 ± 89.55 µmol TE $g^{-1}$ DW), whereas high-altitude samples showed the lowest DPPH and ABTS values. This agrees with Hernández-Ayala et al. [4], who reported that the antioxidant activity of coffee is mainly due to phenolic acids rather than caffeine. The management × altitude interaction was also significant for the three assays ($p \leq 0.004$; Figure 1B); the sustainable × mid-altitude samples (SM) showed the highest FRAP value (675.30 µmol TE $g^{-1}$ DW) and, together with CL and CM, the highest DPPH values, whereas the sustainable × high-altitude samples (SH) showed the lowest DPPH and ABTS values (445.75 and 593.46 µmol TE $g^{-1}$ DW, respectively).

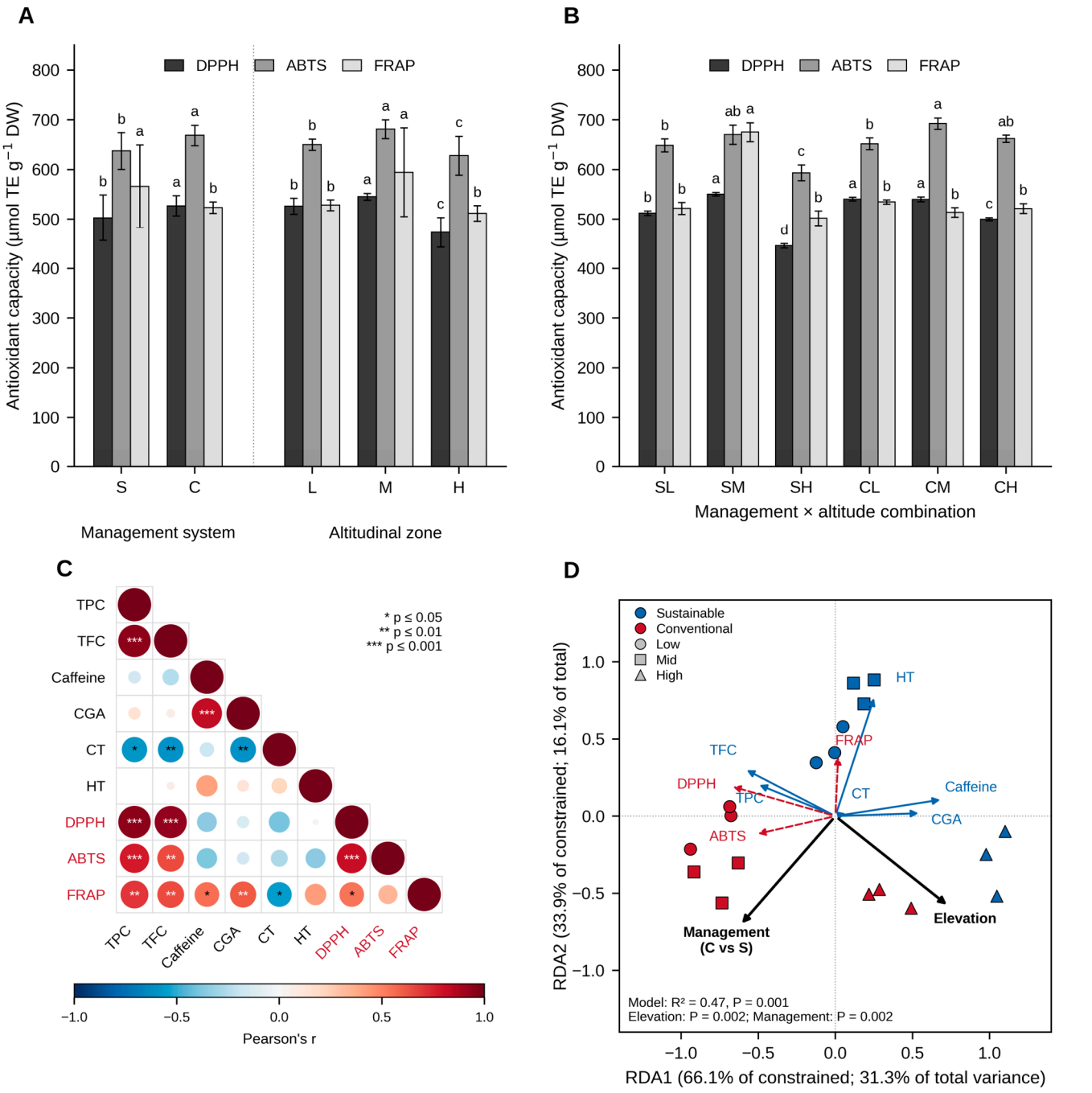



**Figure 1.** In vitro antioxidant capacity of roasted coffee (*Coffea arabica* L.): (**A**) Main effects of management system (S, sustainable; C, conventional) and altitudinal zone (L, low; M, mid; H, high); (**B**) management × altitude combinations (SL, SM, SH: sustainable management in the low, mid, and high zones; CL, CM, CH: conventional management in the low, mid, and high zones). (**C**) Pearson correlation matrix between bioactive compounds and antioxidant assays (n = 18), antioxidant assays are labeled in red and bioactive compounds in black; (**D**) redundancy analysis (RDA) of bioactive compounds constrained by management system and mean elevation of each management × altitude combination, with antioxidant assays projected as supplementary variables (dashed red arrows); solid blue arrows represent bioactive compounds and solid black arrows represent the constraining variables; model significance was assessed with 999 permutations. Bars represent mean ± SD (n = 9 for management, n = 6 for altitude, and n = 3 analytical replicates for combinations). Within each assay and factor, different lowercase letters above the bars indicate significant differences (Tukey's HSD, $p \leq 0.05$). TPC: total phenolic content; TFC: total flavonoid content; CGA: chlorogenic acids; CT: condensed tannins; HT: hydrolyzable tannins; TE: Trolox equivalents; DW: dry weight.

In addition, this study evaluated medium-roasted coffee, a roast degree that retains a high proportion of thermally stable polyphenols and shows higher antioxidant activity than darker roasts. The Redundancy Analysis (Figure 1D) showed that management systems and elevation jointly explained 47.4% of the variance in bioactive composition ($p = 0.001$), with RDA1 accounting for 66.1% of the constrained variance; both elevation and management contributed significantly to the model ($p = 0.002$). Caffeine and CGAs were associated with higher elevations, HT with sustainable samples, and total phenolics, flavonoids, DPPH, and ABTS with conventional low- and mid-altitude samples. This distribution is consistent with the patterns described above for each compound: greater UV radiation and lower temperatures at high elevations have been associated with the accumulation of caffeine and CGAs as part of plant defense responses [52].

Taken together, the sustainable × mid-altitude samples (SM) had the highest total phenolic, flavonoid, CGA, and caffeine contents (Table 3), along with the highest FRAP and DPPH values, although not the highest ABTS value (Figure 1B). In the RDA, these samples were positioned in the direction of HT and FRAP (Figure 1D). This concurrence suggests that the moderate temperature and radiation conditions of the mid zone, together with sustainable management, were associated with the simultaneous accumulation of several compounds with antioxidant activity [58], among which CGAs are considered major contributors to the antioxidant capacity of coffee [4]. However, since these results correspond to composite samples from a single harvest, they should be interpreted as associations between production conditions and bean composition rather than as causal relationships, an approach that requires linking treatment effects through a coherent physiological framework and should be confirmed in farm-level, multi-season studies [59,60].

### *3.6. Sensory Evaluation*

The first principal components explained 78.7% of the variability in the sensory attributes (Figure 2; Table 4). We excluded uniformity, clean cup, sweetness, and balance from the analysis because they did not vary among samples (Table 5). PC1 (eigenvalue = 3.82; 63.7% of the variance) showed high positive loadings for all attributes (0.727–0.869; $p \leq 0.001$) and was therefore interpreted as overall sensory quality. PC2 (eigenvalue = 0.90; 15.0%) contrasted aftertaste (0.559) with overall impression (−0.524) but explained a small proportion of the variance. Body provides viscosity and mouthfeel; acidity confers brightness associated with organic acids such as citric, malic, and phosphoric acids; flavor integrates basic tastes and retronasal aroma; and aftertaste reflects the persistence of the finish [2].

**Table 4.** Loadings of SCA sensory attributes on the first two principal components.

| Attribute | PC1 (63.7%; Eigenvalue = 3.82) | PC2 (15.0%; Eigenvalue = 0.90) |
|---|---|---|
| Fragrance/aroma | 0.760 *** | 0.130 (NS) |
| Flavor | 0.869 *** | 0.374 (NS) |
| Aftertaste | 0.778 *** | 0.559 * |
| Acidity | 0.843 *** | −0.274 (NS) |
| Body | 0.727 *** | −0.284 (NS) |
| Overall | 0.803 *** | −0.524 * |

Loadings are Pearson correlations between each attribute and the principal component score (n = 18). *, *** indicate significance at $p \leq 0.05$, 0.01 and 0.001, respectively; NS: not significant.

**Table 5.** SCA sensory scores of roasted coffee (*Coffea arabica* L.) by management system, altitudinal zone, and their interaction.

| Factor | Level | Fragrance/ Aroma | Flavor | Aftertaste | Acidity | Body | Overall | Total Score |
|---|---|---|---|---|---|---|---|---|
| Management system (M) | S | 7.72 ± 0.20 $^{a}$ | 7.42 ± 0.18 $^{a}$ | 7.42 ± 0.18 $^{a}$ | 7.64 ± 0.22 $^{a}$ | 7.69 ± 0.17 $^{a}$ | 7.78 ± 0.15 $^{a}$ | 83.17 ± 0.76 $^{a}$ |
| | C | 7.61 ± 0.31 $^{a}$ | 7.44 ± 0.27 $^{a}$ | 7.39 ± 0.22 $^{a}$ | 7.69 ± 0.27 $^{a}$ | 7.75 ± 0.22 $^{a}$ | 7.83 ± 0.13 $^{a}$ | 83.22 ± 1.23 $^{a}$ |
| Altitudinal zone (A) | L | 7.46 ± 0.25 $^{b}$ | 7.25 ± 0.22 $^{b}$ | 7.21 ± 0.19 $^{b}$ | 7.42 ± 0.13 $^{c}$ | 7.62 ± 0.14 $^{b}$ | 7.71 ± 0.10 $^{b}$ | 82.17 ± 0.38 $^{c}$ |
| | M | 7.62 ± 0.14 $^{ab}$ | 7.42 ± 0.13 $^{b}$ | 7.46 ± 0.10 $^{a}$ | 7.67 ± 0.13 $^{b}$ | 7.67 ± 0.20 $^{ab}$ | 7.75 ± 0.00 $^{b}$ | 83.08 ± 0.41 $^{b}$ |
| | H | 7.92 ± 0.13 $^{a}$ | 7.62 ± 0.14 $^{a}$ | 7.54 ± 0.10 $^{a}$ | 7.92 ± 0.13 $^{a}$ | 7.88 ± 0.14 $^{a}$ | 7.96 ± 0.10 $^{a}$ | 84.33 ± 0.47 $^{a}$ |
| M × A | SL | 7.58 ± 0.14 $^{ab}$ | 7.25 ± 0.25 $^{b}$ | 7.25 ± 0.25 $^{ab}$ | 7.42 ± 0.14 $^{b}$ | 7.58 ± 0.14 $^{b}$ | 7.67 ± 0.14 $^{c}$ | 82.25 ± 0.25 $^{de}$ |
| | SM | 7.67 ± 0.14 $^{ab}$ | 7.50 ± 0.00 $^{ab}$ | 7.50 ± 0.00 $^{ab}$ | 7.67 ± 0.14 $^{ab}$ | 7.75 ± 0.25 $^{ab}$ | 7.75 ± 0.00 $^{bc}$ | 83.33 ± 0.29 $^{bc}$ |
| | SH | 7.92 ± 0.14 $^{a}$ | 7.50 ± 0.00 $^{ab}$ | 7.50 ± 0.00 $^{ab}$ | 7.83 ± 0.14 $^{a}$ | 7.75 ± 0.00 $^{ab}$ | 7.92 ± 0.14 $^{ab}$ | 83.92 ± 0.14 $^{b}$ |
| | CL | 7.33 ± 0.29 $^{b}$ | 7.25 ± 0.25 $^{b}$ | 7.17 ± 0.14 $^{b}$ | 7.42 ± 0.14 $^{b}$ | 7.67 ± 0.14 $^{ab}$ | 7.75 ± 0.00 $^{bc}$ | 82.08 ± 0.52 $^{e}$ |
| | CM | 7.58 ± 0.14 $^{ab}$ | 7.33 ± 0.14 $^{b}$ | 7.42 ± 0.14 $^{ab}$ | 7.67 ± 0.14 $^{ab}$ | 7.58 ± 0.14 $^{b}$ | 7.75 ± 0.00 $^{bc}$ | 82.83 ± 0.38 $^{cd}$ |
| | CH | 7.92 ± 0.14 $^{a}$ | 7.75 ± 0.00 $^{a}$ | 7.58 ± 0.14 $^{a}$ | 8.00 ± 0.00 $^{a}$ | 8.00 ± 0.00 $^{a}$ | 8.00 ± 0.00 $^{a}$ | 84.75 ± 0.00 $^{a}$ |
| P (M) | | 0.239 | 0.651 | 0.628 | 0.352 | 0.426 | 0.207 | 0.591 |
| P (A) | | 0.006 | 0.002 | 0.002 | <0.001 | 0.026 | 0.001 | <0.001 |
| P (M × A) | | 0.526 | 0.049 | 0.402 | 0.418 | 0.080 | 0.647 | <0.001 |
| P (Q-Grader) | | 0.751 | 0.049 | 0.053 | 0.168 | 0.339 | 0.647 | 0.009 |

Values are the mean ± SD of three certified Q-Graders, each evaluating five cups per sample (n = 9 for management system, n = 6 for altitudinal zone, and n = 3 combinations). Within each column and factor, different superscript letters indicate significant differences (Tukey's HSD, $p \leq 0.05$). $p$ values are from a two-way ANOVA with Q-Grader as a blocking factor. Uniformity, clean cup, and sweetness scored 10.00, and balance scored 7.50 in all samples. All samples scored ≥ 80 points and were classified as specialty coffee. S: Sustainable; C: Conventional; L: Low; M: Mid; H: High; SL: Sustainable × Low; SM: Sustainable × Mid; SH: Sustainable × High; CL: Conventional × Low; CM: Conventional × Mid; CH: Conventional × High.

Along PC1, samples were ordered mainly by altitude: high-altitude samples (CH and SH) were on the positive side, mid-altitude samples near the origin, and low-altitude samples (SL and CL) on the negative side (Figure 2). Accordingly, altitude was the main source of variation in total SCA score ($p < 0.001$; Table 5), increasing from the low (82.17 points) to the mid (83.08) and high (84.33) zones. All samples reached specialty grade (≥80 points), with total scores ranging from 82.08 (CL) to 84.75 (CH). Fragrance/aroma, flavor, aftertaste, acidity, body and overall impression were also highest, or among the highest, in the high-altitude samples (Table 5). Lower temperatures and slower fruit ripening at high elevations favor the accumulation of sugars, acids, and aroma precursors, which has been associated with more complex profiles with higher acidity and body [15,47]. Correlations between chemical variables and sensory attributes, calculated on the six treatment means, showed positive trends of caffeine and CGAs with fragrance/aroma, flavor, and aftertaste (r = 0.53–0.74) and negative trends of flavonoids and DPPH with fragrance/aroma (r = −0.74 and −0.72); however, none were significant ($p > 0.05$; Table S2), so these associations should be confirmed with a larger number of samples.

Management did not affect total SCA score (sustainable: 83.17; conventional: 83.22 points; $p = 0.591$) or any individual attribute ($p \geq 0.207$), indicating that both systems produced coffees of equivalent overall cup quality. However, the management × altitude interaction was significant for total score ($p < 0.001$): in the high zone, the conventional samples scored higher than the sustainable ones (CH: 84.75 vs. SH: 83.92), whereas in the low and mid zones the differences between management systems were not significant. The expression of sensory attributes also depends on roasting conditions and the associated non-enzymatic reactions (Maillard reaction, Strecker degradation and caramelization), which determine aroma formation and flavor modulation [61,62].

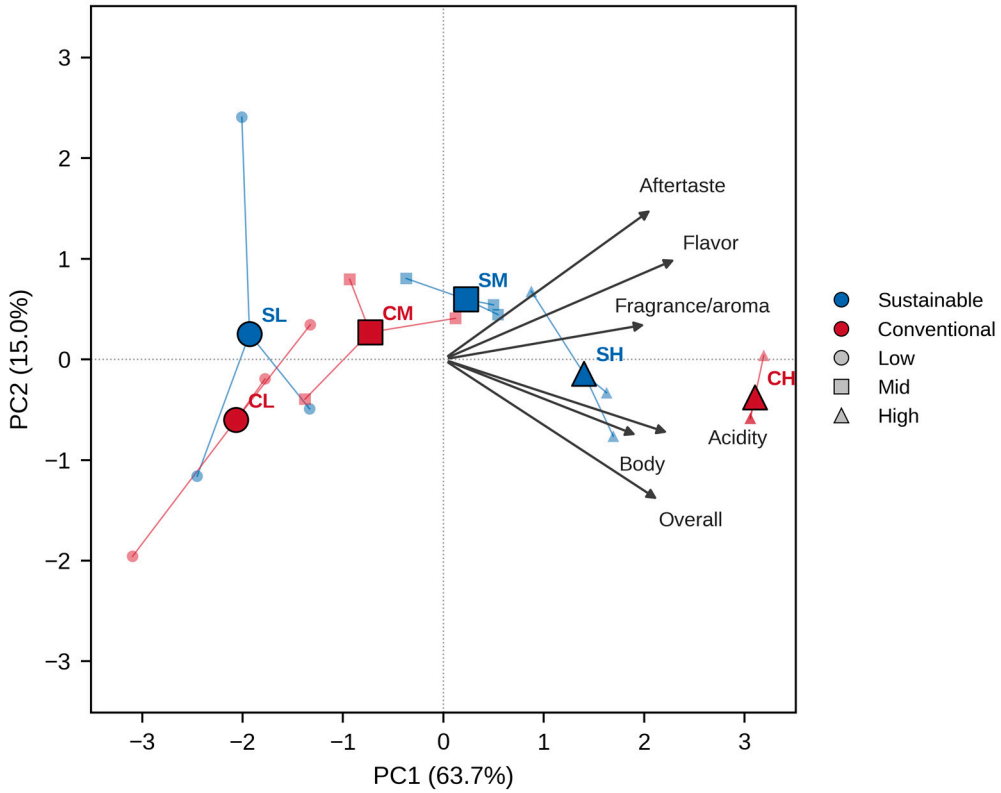



**Figure 2.** Principal component analysis (PCA) biplot of SCA sensory attributes of roasted coffee (*Coffea arabica* L.) by management systems and altitudinal zone. Small symbols represent individual Q-Grader scores, and large symbols represent the mean of each management × altitude combination. We excluded uniformity, clean cup, sweetness, and balance because they did not vary among samples. SL: Sustainable × Low; SM: Sustainable × Mid; SH: Sustainable × High; CL: Conventional × Low; CM: Conventional × Mid; CH: Conventional × High.

## 4. Conclusions

Under the conditions evaluated, altitude was the main factor associated with the physicochemical properties, phenolic and flavonoid contents, and sensory quality of roasted *Coffea arabica* L. from Veracruz, Mexico. In contrast, agricultural management mainly affected chlorogenic acid, caffeine, and hydrolyzable tannins, which were higher in the sustainable samples. Management effects depended on altitude for all phenolic compounds, caffeine, and antioxidant assays, with the largest differences between systems in the mid-altitude zone, where sustainable samples showed the highest total phenolic, flavonoid, CGA, and caffeine contents and FRAP values. Total cup score increased with altitude and did not differ between management systems; all samples reached specialty grade, with the highest score in the conventional × high-altitude samples. These results indicate that management strategies aimed at improving coffee's bioactive profile should consider altitudinal context, and mid- and high-altitude zones are promising for producing specialty coffee with high CGA and caffeine contents under sustainable practices. Further studies, including farm-level replication and multiple harvest seasons, are recommended to confirm these patterns.

**Supplementary Materials:** The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/appliedchem6040082/s1, Table S1. F statistics and *p* values (in parentheses) from two-way ANOVA for management system (M), altitudinal zone (A) and their interaction (M × A). Sensory attributes were analyzed with the Q-Grader as a blocking factor; Table S2. Pearson correlation coefficients (r) between chemical variables and SCA sensory attributes, calculated on the means of the six management × altitude combinations (n = 6).

**Author Contributions:** Conceptualization, A.G.-Z., A.D.H.-F., J.M.P.-E., Z.H.P.-G. and C.U.L.-P.; methodology, A.G.-Z., A.D.H.-F., J.G.-T., Z.H.P.-G. and C.U.L.-P.; validation, A.G.-Z., J.M.P.-E., Z.H.P.-G., J.P.-M. and C.U.L.-P.; formal analysis, A.G.-Z., A.D.H.-F., J.G.-T. and C.U.L.-P.; investigation, A.G.-Z., A.D.H.-F., A.M.-N., J.G.-T. and C.U.L.-P.; resources, A.G.-Z., A.D.H.-F. and C.U.L.-P.; data curation, A.G.-Z., A.D.H.-F. and C.U.L.-P.; writing—original draft preparation, A.G.-Z., J.G.-T. and C.U.L.-P.; writing—review and editing, A.G.-Z. and C.U.L.-P.; visualization, A.G.-Z., A.D.H.-F., J.P.-M. and C.U.L.-P. All authors have read and agreed to the published version of the manuscript.

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**Institutional Review Board Statement:** Under Mexican legislation, ethics committee approval is required for research involving human beings within the scope of health research, as established in the General Health Law (Ley General de Salud (Articles 96 and 98; https://www.diputados.gob.mx/LeyesBiblio/pdf/LGS.pdf, accessed on 29 October 2026) and in the Regulation of the General Health Law on Health Research (Reglamento de la ley General de Salud en Materia de Investigacion para la salud; https://www.diputados.gob.mx/leyesBiblio/regley/Reg_LGS_MIS.pdf, accessed on 29 October 2026). According to this legislation, healt research comprises actions that contribute to the knowledge of biological and psychological processes in human beings, the relationships between the causes of disease, medical practice and social structure, the prevention and control of health problems, and the provision of health services. Since the present study consisted of the sensory quality grading of a food product (roasted coffee) by adult certified professional evaluators (Q-Graders), whose scores describe the coffee samples rather than the evaluators, and did not involve clinical, biomedical or health-related procedures, it does not fall within the scope of this legislation; therefore, ethics committee approval was not required. In any case, this activity corresponds to research without risk according to Article 17, Section I, of the aforementioned Regulation. The study was conducted in accordance with the principles of the Declaration of Helsinki.

**Informed Consent Statement:** Verbal informed consent was obtained from the participants. Verbal consent was obtained rather than written because the study consisted of a non-invasive, no-risk sensory quality grading of a commercial food product, performed by adult certified professional evaluators (Q-Graders) as part of their routine professional activity, and no personal, physiological or health-related data about them were collected o recorded.

**Data Availability Statement:** The original contributions presented in this study are included in the article/Supplementary Material. Further inquiries can be directed to the corresponding authors.

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**Conflicts of Interest:** The authors declare no conflicts of interest.

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