



Research article

Profile of chemical compounds, aroma, and cup quality of arabica coffee from ohmic-heated carbonic maceration

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Abstract: In this study, we investigated the effects of ohmic-heated carbonic maceration on the chemical composition (including bioactive compounds), aroma, and cup quality of Arabica coffee. Coffee cherries were fermented at two temperature levels (31 °C and 36 °C) and three durations (48, 144, and 240 hours) in an ohmic-heated fermenter under carbonic (CO₂-rich, anaerobic) conditions. The coffee samples were analyzed using HPLC and GC-MS to identify bioactive and volatile compounds, and cupping tests were performed using the Specialty Coffee Association of America (SCAA) protocol. The results showed that increasing fermentation duration decreased 3-caffeoylquinic acid (3-CQA) and 4-CQA, while 5-CQA remained stable or increased at 36 °C for 240 hours. Trigonelline exhibited relative stability at low temperatures but declined at prolonged durations, particularly at 36 °C. Caffeine concentrations peaked at intermediate durations before stabilizing. Sugar content generally decreased with increasing fermentation duration, with significantly lower levels observed after 10 days of fermentation. Principal component analysis of volatile compounds revealed distinct clustering patterns among treatments, with long fermentation durations positively correlated with the production of phenols, alcohols, pyrroles, benzenes, pyridines, and alkanes. Sensory evaluation using the SCAA protocol showed that four fermentation treatments provided slightly higher cupping scores (85.0–85.75) than the control (84.5), with flavor notes including nutty,

flowery, lemony, brown sugar, spicy, fruity, and winey aromas. These findings indicated that Arabica coffee processed under ohmic-heated carbonic fermentation conditions exhibits measurable modulation of key chemical constituents, sugar content, and volatile composition, leading to improved cup quality and distinct sensory attributes within the studied processing conditions.

Keywords: Arabica coffee; chemical composition; carbonic maceration; ohmic heating; sensory quality

Abbreviations

Control = Fermentation Temperature 0 °C, Fermentation Duration 0 hours (not fermented, denoted as C); T1F2 = Fermentation Temperature 31 °C, Fermentation Duration 48 hours; T1F6 = Fermentation Temperature 31 °C, Fermentation Duration 144 hours; T1F10 = Fermentation Temperature 31 °C, Fermentation Duration 240 hours; T2F2 = Fermentation Temperature 36 °C, Fermentation Duration 48 hours; T2F6 = Fermentation Temperature 36 °C, Fermentation Duration 144 hours; T2F10 = Fermentation Temperature 36 °C, Fermentation Duration 240 hours

1. Introduction

Different post-harvest processing methods have been shown to affect the chemical composition and sensory characteristics of roasted coffee [1–3]. The introduction of controlled fermentation has become one of the major drivers in specialty coffee amid the dynamics of consumer preferences. Today's coffee consumers demand more defined flavors, consistent quality, and process transparency. One notable approach is carbonic maceration, defined as an anaerobic, CO₂-rich fermentation process in which whole coffee cherries are fermented under limited oxygen availability. This approach is adapted from winemaking practices. Moreover, this method modulates microbial metabolism and the biotransformation pathways of aroma precursors in coffee cherries, resulting in changes to chemical composition and sensory attributes [4–7]. To precisely control the fermentation process, ohmic-assisted fermentation has been developed. Ohmic heating has emerged as a volumetric heating technology that enables rapid and uniform temperature distribution throughout the fermentation matrix, offering precise temperature control and improved energy efficiency compared to conventional conductive heating methods [8–10]

The combination of the carbonic maceration process with ohmic heating offers a fermentation environment with enhanced control over temperature and processing time, which has implications for the stability and interconversion of key compounds, including chlorogenic acids (particularly 5-caffeoylquinic acid, 5-CQA) and alkaloids (trigonelline and caffeine). It also affects the dynamics of sugars (sucrose and reducing sugars), which act as important precursors for Maillard reactions during roasting [5,11,12].

Integrated microbial–metabolomic studies in coffee fermentation have reported that post-harvest fermentation can drive rapid sucrose hydrolysis, lactic acid accumulation, and the formation of fermentation-derived volatiles that contribute to distinctive floral and fruity sensory notes [13]. Given that more than 800 volatile compounds have been identified in roasted coffee, but only a limited subset contributes decisively to aroma perception, there is a need for fermentation strategies capable of directing volatile formation toward targeted sensory outcomes. Based on this context, we examined

the chemical composition and key quality-related constituents, aroma profile, and cup quality of Arabica coffee processed through ohmic-heated carbonic maceration, focusing on changes in chlorogenic acid isomers (3-, 4-, and 5-CQA), alkaloids, sugars, volatile compound clusters, and cupping scores.

The application of carbonic maceration under controlled conditions presents methodological challenges and strategic opportunities for improving specialty coffee quality. Variability in indigenous microbiota, mucilage composition, seasonal conditions, and agroecological factors can influence biotransformation pathways during fermentation, making reproducibility difficult and highlighting the need for precise and scalable process control [14]. Comparable insights from other fermented commodities (e.g., cocoa) show that spontaneous fermentation induces substantial metabolite variation, affecting taste, aroma, and quality, and that metabolomics combined with chemometrics can identify key metabolites/biomarkers and support standardization of fermentation time [15]. Studies have shown that carbonic maceration can alter bacterial community structure, including shifts in lactic acid bacteria dominance, which are associated with changes in sensory attributes; however, these effects are highly sensitive to temperature, fermentation duration, aeration, and pH, necessitating well-defined and auditable processing protocols [6].

Literature on controlled coffee fermentation highlights two converging trends: (i) The use of carbonic maceration to modulate chemical and sensory profiles through regulated microbe–substrate interactions, and (ii) the adoption of ohmic heating and moderate-intensity electric fields to enhance heating uniformity, energy efficiency, and process parameter control [4,9,16].

Several researchers have reported that carbonic maceration influences volatile composition, lactic acid bacteria dynamics, and sensory attributes in coffee; however, these investigations are often conducted under broad and variable processing conditions, including wide temperature and time ranges, different coffee species, and diverse agroecological contexts, which limits reproducibility and generalization [6,17].

Guided by ohmic-assisted coffee fermentation work [18] that evaluated 30, 35, and 40 °C and reported the highest cupping score at 35 °C after 12 h, we investigate the effects of fermentation temperature (31 °C and 36 °C) and duration (48, 144, and 240 h) during ohmic-heated carbonic maceration on (i) temporal changes in chlorogenic acid isomers (3-, 4-, and 5-CQA), trigonelline, and caffeine; (ii) sugar content and volatile profiles; and (iii) their associations with sensory attributes and SCAA cupping scores, compared with a non-fermented control.

Sensomic and chemometric approaches, such as GC-MS combined with principal component analysis (PCA), have been widely used to associate classes of volatile compounds (e.g., phenols, alcohols, pyrroles, benzenes, pyridines, and alkanes) with aroma perception. In parallel, Omics-based fermentation research emphasizes the need to link microorganisms and their corresponding metabolite signatures, typically supported by multivariate tools (e.g., PCA) to interpret complex chromatographic and GC-MS datasets [15]. Nevertheless, an integrated assessment combining key chemical constituents (3-, 4-, and 5-CQA; trigonelline; caffeine), sugars, volatile profiles, and SCAA cupping scores under precisely controlled ohmic-assisted carbonic fermentation conditions remains lacking.

In this study, we provide an integrated evaluation of chemical composition, volatile profiles, and sensory quality under well-defined ohmic-assisted carbonic fermentation conditions. By systematically examining fermentation temperatures of 31–36 °C and durations of 48–240 h, the study contributes evidence on how controlled fermentation parameters influence key chemical compounds, sugars, and aroma-active volatiles. In this sense, the work complements emerging microbe–metabolite

(Omics-informed) perspectives on coffee fermentation by providing a controlled-process dataset that links targeted chemical markers, volatile clusters, and SCAA cup quality [13]. Rather than comparing ohmic heating with conventional heating, the findings are intended to inform process optimization within ohmic-assisted systems, offering practical guidance for temperature- and time-based fermentation control in specialty coffee production. This framework supports the development of replicable and auditable fermentation protocols and provides a basis for future studies aiming to compare heating technologies or scale up controlled fermentation strategies for the coffee industry.

2. Materials and methods

2.1. Coffee cherries

The material used in this study was fully mature Arabica coffee cherries harvested in the highlands of Tanah Toraja, Kaduaja Village, Sangbua, South Sulawesi, Indonesia (approximately 1,400 m above sea level). The coffee cherries were then transported to Hasanuddin University in Makassar, South Sulawesi, Indonesia, for fermentation. The time between harvest and the start of the fermentation process was approximately 36 hours. The fermenter consisted of polyvinyl chloride (PVC) tubes with an internal diameter of 12.7 cm and a length of 130 cm. The system was supplied with 220 V alternating current (AC), generating an electric field strength of approximately 1.7 V cm^{-1} . Approximately 10 kg of whole coffee cherries were loaded into each fermenter (Figure 1), and potable water was added to serve as an electrical conductor.

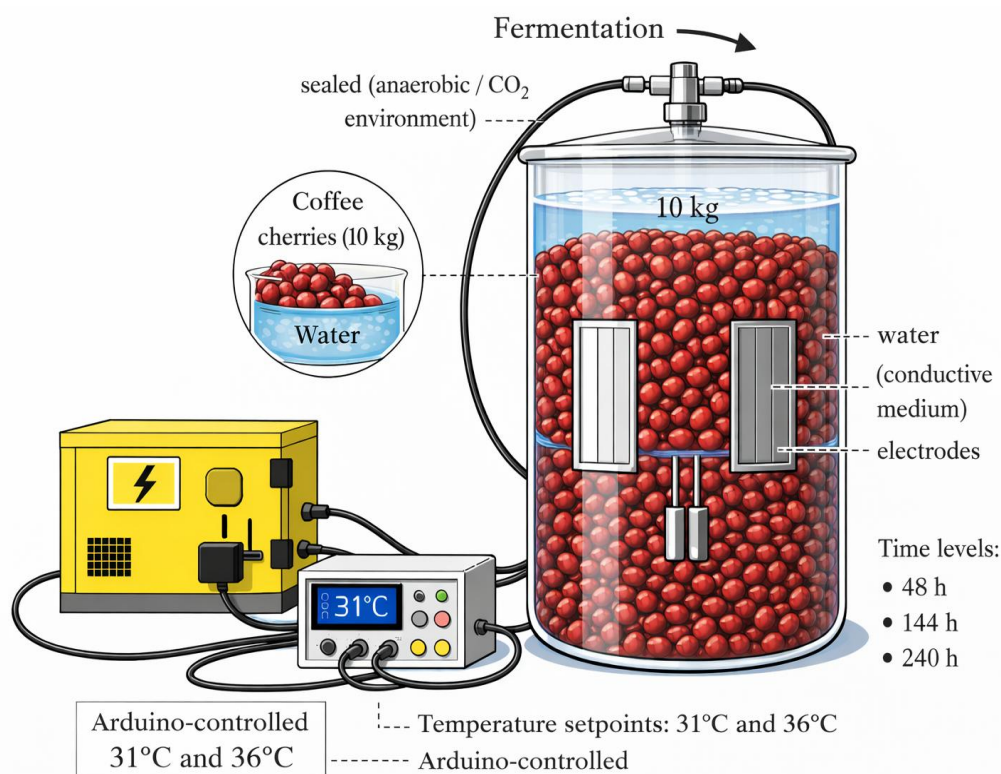


Figure 1. Illustration of ohmic-assisted carbonic maceration (anaerobic fermentation) of coffee cherries in an ohmically heated fermenter.

Two fermentation temperatures (31 °C and 36 °C), denoted as T1 and T2, and three fermentation durations (48, 144, and 240 h), denoted as F2, F6, and F10, were selected based on preliminary trials and reported ranges commonly applied in controlled coffee fermentation studies. Following fermentation, cherries were removed from the fermenter, depulped, washed, and sun-dried to approximately 8% moisture content. The dried parchment was then dehulled to obtain green coffee beans (GCBs). The GCBs were subsequently stored in airtight containers at room temperature. All GCBs were roasted prior to analysis using a Cafemasy coffee roaster (Cafemasy, Guangdong, China). For comparison, control (untreated or honey processed) sample was prepared by depulping the coffee cherries directly after harvest and sun-dried. This sample is denoted as Control (C) throughout this article.

2.2. Chemical compound analysis

Coffee samples roasted to a medium-dark roast level were finely ground and extracted using the method described in [19]. The coffee extract (20 µL) was injected into an HPLC system equipped with a UV-Vis detector (LC-20AD; Shimadzu, Kyoto, Japan). Six concentrations of 3-, 4-, and 5-caffeoylquinic acid (CQA) standards were used to generate calibration curves. Concentrations ranged from 16 to 500 mg L⁻¹ (triplicate); the limit of detection (LoD) for 3-CQA was 13.27 mg L⁻¹ (R² = 0.997) and for 4-CQA was 15.37 mg L⁻¹ (R² = 0.996). Alkaloid quantification was performed according to the method described by the researchers in [20] using a Zorbax C18 column (5 µm particle size). Theobromine, caffeine, and trigonelline were quantified using six-point calibration curves (6–200 mg L⁻¹, triplicate). The LoD values were 1.08 mg L⁻¹ for trigonelline (R² = 0.999), 2.40 mg L⁻¹ for theobromine (R² = 0.997), and 0.93 mg L⁻¹ for caffeine (R² = 0.999).

2.3. Reducing sugar analysis [20]

A Cafemasy coffee roaster (Cafemasy, Guangdong, China) was used to roast up to 80 g of coffee at a moderately dark roasting level for 7 minutes. The reducing sugar content was determined following the method described by the researchers in [20]. Standard glucose solutions with concentrations of 2, 4, 6, 8, and 10 mg per 100 mL were prepared by diluting glucose anhydrous (10 mg per 100 mL), followed by serial dilution. Next, six clean test tubes were prepared, each containing 1 mL of standard glucose solution, with distilled water used as a blank. One milliliter (1 mL) of Nelson's reagent was added to each tube, followed by heating for 20 min in a boiling water bath and cooling to 25 °C. One milliliter (1 mL) of arsenomolybdate reagent was then added and mixed to dissolve all Cu₂O precipitates. Once the precipitate had completely dissolved, 7 mL of purified water was added to obtain a homogeneous solution. Absorbance was measured at 540 nm to generate a standard calibration curve. To determine reducing sugar content in samples, 1 mL of lead-free sample solution was diluted to 100 mL, and 1 mL of the diluted sample was measured at 540 nm.

2.4. Total sugar determination [20]

Approximately 25 mL of filtrate-free sample was placed in an Erlenmeyer flask, followed by 15 mL of distilled water and 5 mL of 30% HCl (specific gravity 1.15). The solution was heated in a water bath at 67–70 °C for 10 min and then rapidly cooled to 20 °C. The solution was neutralized with 40%

NaOH and diluted to a final volume of 100 mL. Sucrose content was determined using the Nelson–Somogyi method.

2.5. *Volatile compound analysis*

Volatile compounds were analyzed using gas chromatography–mass spectrometry (GC-MS; Agilent 7890A GC and Agilent 5975C XL EI/CI MS; Santa Clara, CA, USA) with the injector temperature set at 250 °C in splitless mode. Helium was used as the carrier gas at a flow rate of 0.8 mL min⁻¹. Coffee samples (80 g) were roasted using a Cafemasy roaster (Cafemasy, Guangdong, China) for 7 min at a moderately dark roasting level. Extraction was performed using 3.5 g of roasted coffee in a 22 mL SPME vial, with 0.2 µL of 2,4,6-trimethylpyridine (0.01%) as an internal standard. Samples were extracted at 80 °C for 45 min using a 2 cm DVB/CAR/PDMS SPME fiber.

2.6. *Sensory analysis of coffee samples*

Cup tests were conducted in accordance with the Specialty Coffee Association of America (SCAA) standard protocol at the Indonesian Coffee and Cocoa Research Center, and were performed by two certified cuppers. All coffee samples were roasted, ground, and brewed according to SCAA preparation guidelines prior to sensory evaluation.

2.7. *Data analysis*

Data for chlorogenic acids, alkaloids, and sugars were analyzed using two-way analysis of variance (ANOVA) to assess the effects of fermentation temperature and duration, followed by Tukey's post-hoc test when significant differences were observed ($p < 0.05$). Quadratic polynomial regression was applied to model non-linear trends observed across temperature and fermentation duration, following Steel and Torrie (1997). All statistical analyses were performed using SPSS (version 25; IBM Corp., Armonk, NY, USA), and data visualization was conducted using Orange Data Mining (version 3.40.0).

3. Results and discussion

3.1. *Chlorogenic acid*

Chlorogenic acid is an important antioxidant that contributes to coffee flavor. Furthermore, 5-caffeoylquinic acid (5-CQA) is the predominant chlorogenic acid (CQA) isomer in green coffee, accounting for up to 76–84% of total chlorogenic acids, although its stability is highly sensitive to temperature, pH, and light exposure [11].

The levels of chlorogenic acids (3-CQA, 4-CQA, and 5-CQA) presented in Table 1 show a pattern in relation to temperature and fermentation duration. Relative to the control, 3-CQA and 4-CQA generally decreased with increasing fermentation duration at 31 °C and 36 °C. In contrast, 5-CQA showed a different trend, remaining comparatively stable across most conditions and increasing at 36 °C for 240 h. In summary, longer durations reduced 3-/4-CQA, while 5-CQA remained relatively stable or increased at 36 °C for 240 h.

Mechanistically, carbonic fermentation in coffee is strongly influenced by temperature and fermentation duration (Figure 2), which alters the chemical profile and may be associated with cup quality. In this study, the observed shifts in CQA isomers are discussed in relation to processing parameters; however, causal links to sensory outcomes should be interpreted cautiously because sensory responses are multifactorial. Studies indicate that controlled fermentation (including variations in temperature and time) can modulate the biotransformation pathways of aroma precursors and phenolic acids, with implications for cup quality [7].

Table 1. The effect of fermentation temperature and duration on 3-caffeoylquinic, 4-caffeoylquinic, and 5-caffeoylquinic acids.

Treatment		Variable \pm std		
Temperature ($^{\circ}$ C)	Duration (hours)	3 CQA	4 CQA	5 CQA
Control		127.1 \pm 1.41 ^a	165.80 \pm 1.27 ^a	449.6 \pm 2.83 ^a
31	48	127.2 \pm 1.48 ^b	169.30 \pm 2.97 ^a	440.90 \pm 2.83 ^b
	144	113.0 \pm 0.71 ^c	152.10 \pm 1.27 ^b	407.90 \pm 1.41 ^c
	240	96.20 \pm 1.41 ^d	132.90 \pm 0.98 ^c	414.40 \pm 1.41 ^d
36	48	117.9 \pm 1.41 ^b	157.10 \pm 1.56 ^a	406.40 \pm 1.41 ^b
	144	97.0 \pm 1.41 ^c	133.30 \pm 1.56 ^b	413.10 \pm 4.24 ^c
	240	88.33 \pm 0.33 ^d	125.60 \pm 3.11 ^c	515.60 \pm 2.12 ^d

Description: Superscript letters within the same column indicate significant differences between treatments ($p < 0.05$) at a 95% confidence level.

Regarding the role of ohmic heating, this process provides rapid and relatively uniform volumetric heating, enabling stable temperature control during fermentation. Because we did not include a conventional-heating control, statements about superiority over conventional heating are not concluded here; instead, ohmic heating is discussed as a process-control approach that is energy efficient and offers precise temperature regulation, which may help preserve heat-sensitive components such as phenolics [21]

Interconversion of CQA isomers has been reported in studies, where 5-CQA can isomerize to 3-/4-CQA and form other transformation products depending on processing conditions; the extent is influenced by pH, matrix composition, and heating conditions [22]. In this dataset, the decline in 3-/4-CQA with increasing duration, together with the higher 5-CQA at 36 $^{\circ}$ C–240 h, may reflect shifts in the isomer distribution and/or release of bound chlorogenic acids during fermentation; however, the mechanisms were not directly measured in this study. This interpretation is consistent with work highlighting the roles of temperature and time during carbonic or semi-carbonic fermentation [4].

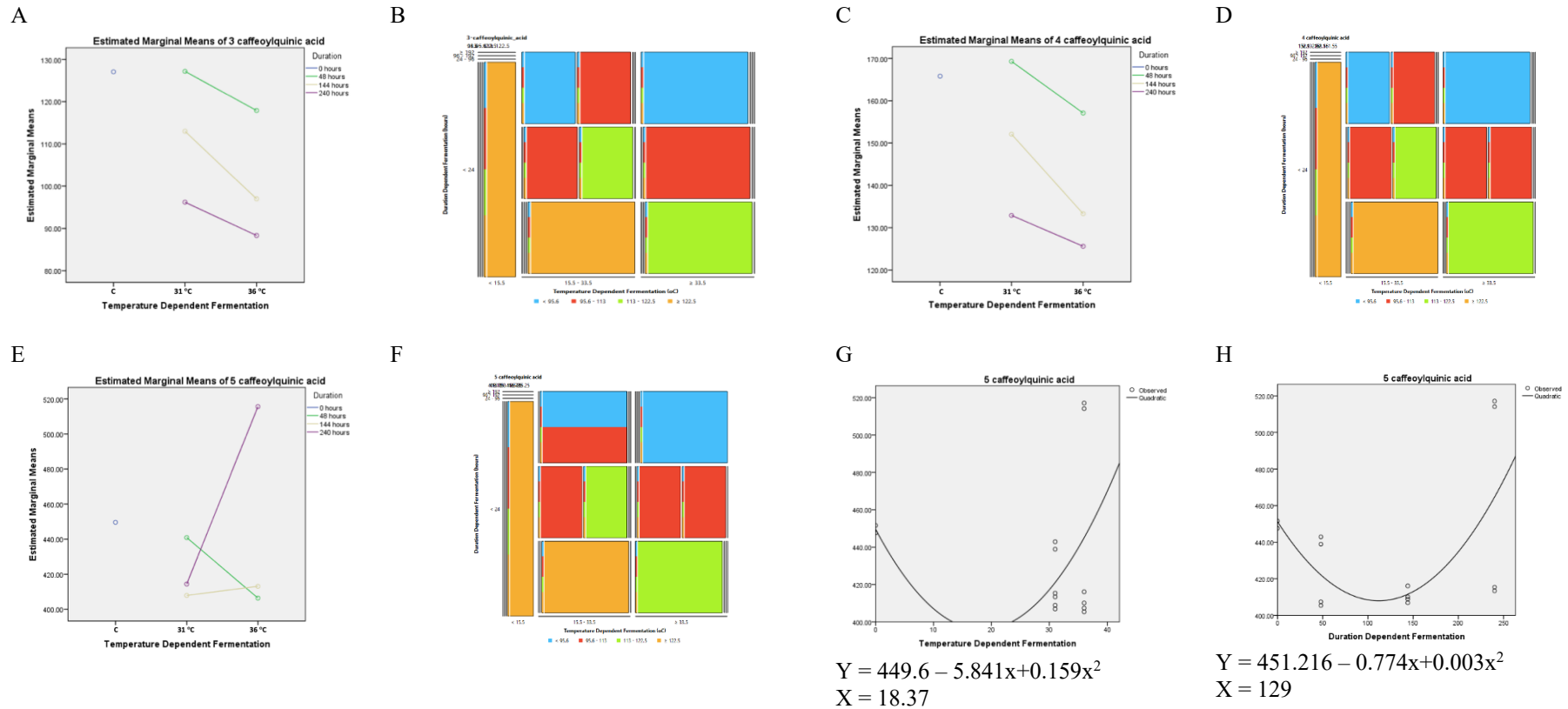


Figure 2. (a) Estimated marginal means (EMMs) of 3-caffeoylquinic acid (3-CQA) as a function of fermentation temperature (Control (C), 31, and 36 °C) across four durations (Control (C), 48, 144, and 240 h). (b) Heatmap of mean 3-CQA across temperature–duration combinations (Control, 31, and 36 °C; Control, 48, 144, and 240 h). (c) EMMs of 4-caffeoylquinic acid (4-CQA) as a function of fermentation temperature (Control, 31, and 36 °C) across four durations (Control, 48, 144, and 240 h). (d) Heatmap of mean 4-CQA across temperature–duration combinations (Control, 31, 36 °C; Control, 48, 144, 240 h). (e) EMMs of 5-caffeoylquinic acid (5-CQA) as a function of fermentation temperature (Control, 31, and 36 °C) across four durations (Control, 48, 144, and 240 h). (f) Heatmap of mean 5-CQA across temperature–duration combinations (Control, 31, 36 °C; Control, 48, 144, and 240 h). (g) Quadratic model of 5-CQA versus temperature (open circles = observed data; line = fitted quadratic), showing a minimum near 18.37 °C and an upward trend at higher temperatures within the study range. (h) Quadratic model of 5-CQA versus duration (open circles = observed data; line = fitted quadratic), showing a minimum near 129 h and an increase at longer durations (≥200–240 h).

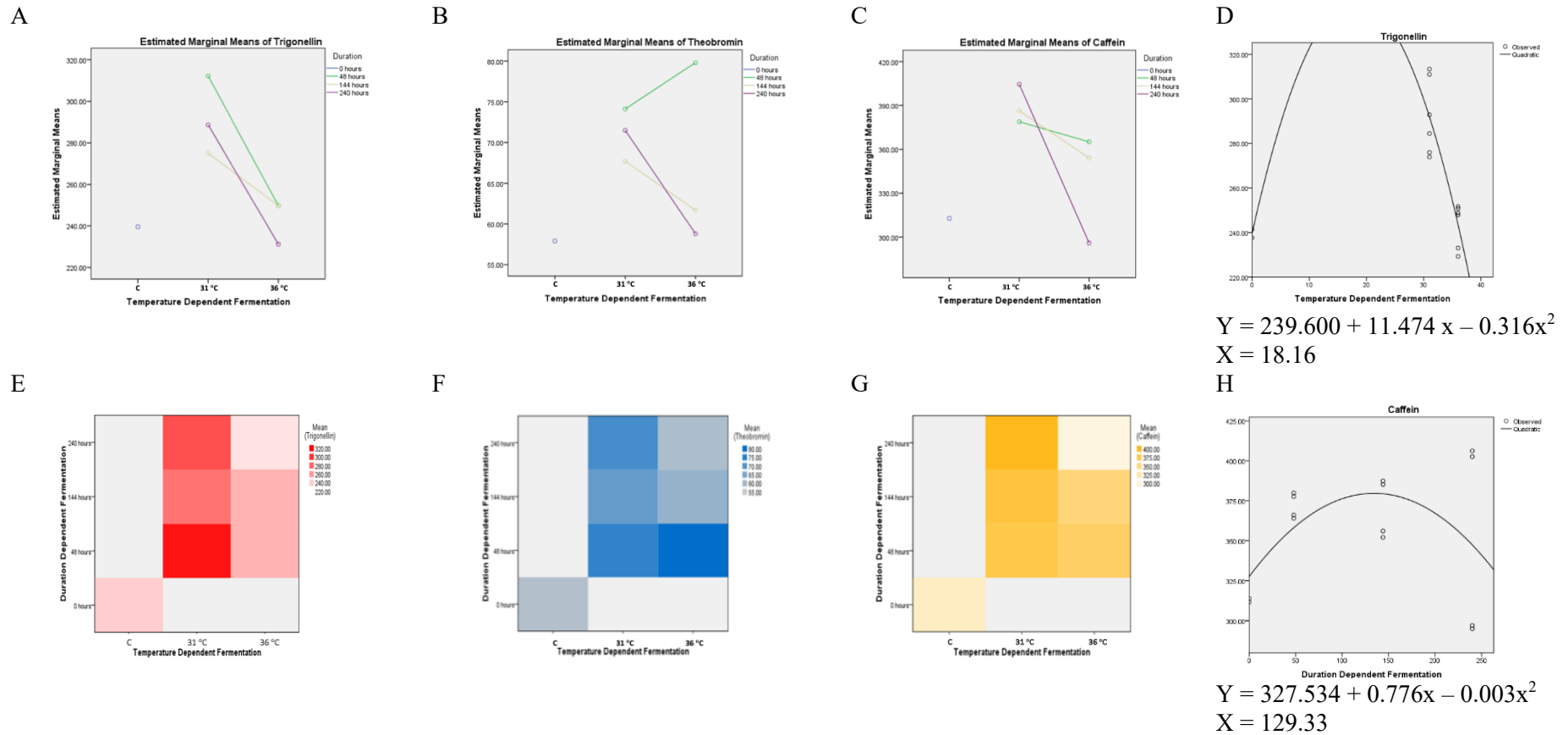


Figure 3. (a) Estimated marginal means (EMMs) of trigonelline as a function of fermentation temperature (Control (C), 31, and 36 °C) across four durations (Control (C), 48, 144, and 240 h). (b) EMMs of theobromine versus fermentation temperature (Control, 31, and 36 °C) across the exact four durations. (c) EMMs of caffeine versus fermentation temperature (Control, 31, and 36 °C) across the same four durations. (d) Quadratic model of trigonelline versus temperature (open circles = observed; line = fitted quadratic), showing a peak at an intermediate temperature within the study range. (e) Heatmap of mean trigonelline across temperature–duration combinations (Control, 31, 36 °C; Control, 48, 144, 240 h). (f) Heatmap of mean theobromine across the same combinations. (g) Heatmap of mean caffeine levels across the same combinations. (h) Quadratic model of caffeine versus fermentation duration (open circles = observed; line = fitted quadratic), showing a maximum near the mid-range (~129 h) and a decline at longer durations.

3.2. Alkaloid compounds

Trigonelline, theobromine, and caffeine are key alkaloids in coffee brews, and recent profiling of espresso-based Arabica beverages has shown that their concentrations vary with bean origin and brewing method [22]. Here, we investigated the impact of fermentation temperature and duration on trigonelline, theobromine, and caffeine levels, as shown in Table

Table 2. The effect of fermentation temperature and duration on trigonelline, theobromine, and caffeine levels.

Treatment		Variable \pm std		
Temperature ($^{\circ}$ C)	Duration (hours)	Trigonelline	Theobromine	Caffeine
Control		239.60 \pm 2.69 ^c	57.90 \pm 1.41 ^c	312.80 \pm 1.56 ^c
31	48	312.20 \pm 1.70 ^a	74.13 \pm 1.39 ^a	378.30 \pm 1.56 ^a
	144	275.00 \pm 1.41 ^b	67.70 \pm 3.11 ^b	386.30 \pm 1.56 ^a
	240	288.70 \pm 5.94 ^b	71.50 \pm 1.56 ^b	404.40 \pm 2.55 ^b
36	48	249.80 \pm 1.41 ^a	79.80 \pm 1.34 ^a	365.10 \pm 1.41 ^a
	144	249.80 \pm 2.69 ^b	61.70 \pm 1.69 ^b	354.10 \pm 2.83 ^a
	240	231.20 \pm 2.69 ^b	58.80 \pm 1.56 ^b	296.00 \pm 1.41 ^b

Description: Superscript letters within the same column indicate significant differences between treatments ($p < 0.05$) at a 95% confidence level.

The levels of trigonelline, theobromine, and caffeine are listed in Table 2. Across treatments, trigonelline increased at 31 $^{\circ}$ C after 48 h relative to the control and then varied with longer durations, with the lowest value observed at 36 $^{\circ}$ C at 240 h. This pattern is consistent with reports indicating that trigonelline is water soluble and relatively stable during early post-harvest stages but is thermolabile and undergoes substantial degradation during roasting [23,24].

Fermentation temperature and duration under ohmic-assisted carbonic maceration influenced alkaloid levels (Figure 3). These changes may be associated with time-dependent biochemical changes during fermentation and temperature stability achieved by ohmic volumetric heating. Studies under carbonic maceration conditions (18–38 $^{\circ}$ C; 24–120 h) report increases in microbial diversity and shifts in lactic acid bacteria dominance (e.g., *Leuconostoc* \rightarrow *Lactobacillus*) with higher temperature and longer duration, which have been associated with sensory differences [6]. In this study, microbial community composition was not directly measured; therefore, the discussion of microbial contributions is based on published literature rather than direct evidence from our dataset.

Ohmic heating provides uniform and rapid heating to reach the target temperature, resulting in a flatter temperature-time curve and supporting stable thermal conditions during fermentation [25]. In the context of this study, ohmic heating is discussed as a means of improving temperature control rather than as a comparative advantage over conventional heating (which was not tested). Ohmic heating can accelerate attainment of the set temperature using a moderate-intensity electric field and may influence fermentation kinetics, which in turn may affect non-volatile compounds such as alkaloids over the fermentation duration [8].

In anaerobically fermented coffee, trigonelline has been reported to increase during early stages and decrease with extended fermentation, while caffeine may peak at intermediate durations before stabilizing; these patterns are consistent with changes in the fermentation environment and compound

transformation pathways. In our data, caffeine increased under several fermentation conditions (e.g., 31 °C after 240 h) but decreased at 36 °C for 240 h relative to the control, indicating that the direction of change depended on the temperature–duration combination. Overall, measurable shifts in trigonelline, caffeine, and theobromine were observed across treatments, consistent with the literature indicating that controlled fermentation can modulate key coffee chemical constituents through processing conditions and reported microbially mediated transformations [26].

3.3. Sugar content

The sugar content in coffee samples (Table 3) produced from carbonic maceration ranged from 1.35% to 2.79% for maceration at 31 °C and from 1.73% to 2.34% for maceration at 36 °C. In general, sugar content tended to decrease as maceration duration increased, and significantly lower sugar contents were obtained from samples that underwent 10 days (240 h) of carbonic maceration. The effect of maceration temperature on sugar content was less consistent across durations and was therefore interpreted cautiously. Researchers have reported that sugar content in coffee can be affected by pre- and post-harvest processes [12].

Table 3. The effect of fermentation temperature and duration on reducing sugar, total sugar, and sucrose.

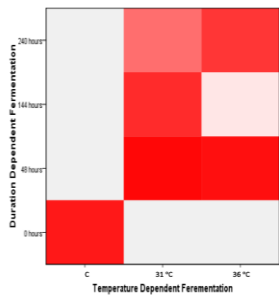
Treatment		Variable ± std		
Temperature (°C)	Duration (hours)	Reducing Sugar	Total sugar	Sucrose
Control		1.36 ± 0.03 ^b	2.48 ± 0.00 ^b	1.06 ± 0.03 ^a
31	48	1.46 ± 0.03 ^a	2.79 ± 0.03 ^a	1.26 ± 0.00 ^a
	144	1.25 ± 0.03 ^d	2.42 ± 0.03 ^c	0.11 ± 0.00 ^c
	240	0.86 ± 0.01 ^c	1.35 ± 0.01 ^d	0.47 ± 0.14 ^b
36	48	1.41 ± 0.03 ^a	2.34 ± 0.04 ^a	0.88 ± 0.01 ^a
	144	0.16 ± 0.01 ^d	1.93 ± 0.02 ^c	0.75 ± 0.01 ^c
	240	1.19 ± 0.02 ^c	1.73 ± 0.01 ^d	0.52 ± 0.00 ^b

Description: Superscript letters within the same column indicate significant differences between treatments ($p < 0.05$) at a 95% confidence level.

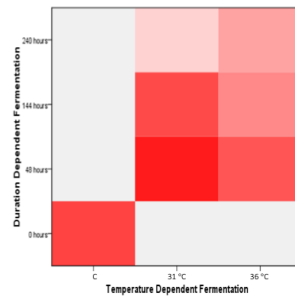
The sucrose content tended to decrease as maceration duration increased, but an anomaly was observed for maceration at 31 °C for 144 h, which indicated an extremely low sucrose content (Figure 4). This value was reported as measured and may have reflected treatment-specific variability and/or analytical variation; additional confirmation (e.g., repeat extraction/quantification) would be required to determine whether it represents a systematic effect. Sucrose is an important flavor precursor in coffee beans because it can degrade or react with other constituents in the beans to form aroma compounds. Studies have indicated that sucrose can produce aromatic compounds, such as furans, aldehydes, and carboxylic acids, which contribute to coffee taste and aroma [27]. Comparable Omics-based insights from other fermented commodities (e.g., cocoa) highlight coordinated shifts in sugars and other metabolites that can influence sensory quality [28].

Reducing sugar content also tended to decrease as maceration duration increased, except for maceration at 36 °C for 144 h, which showed significantly less sugar than at 240 h at the same temperature. Overall, temperature effects on reducing sugar and sucrose depended on duration rather

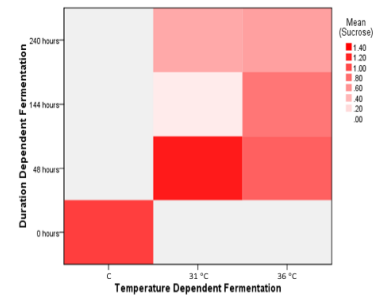
than showing a uniform trend. It is important to note that the reducing sugar content in the control sample (honey process) was 1.36%.



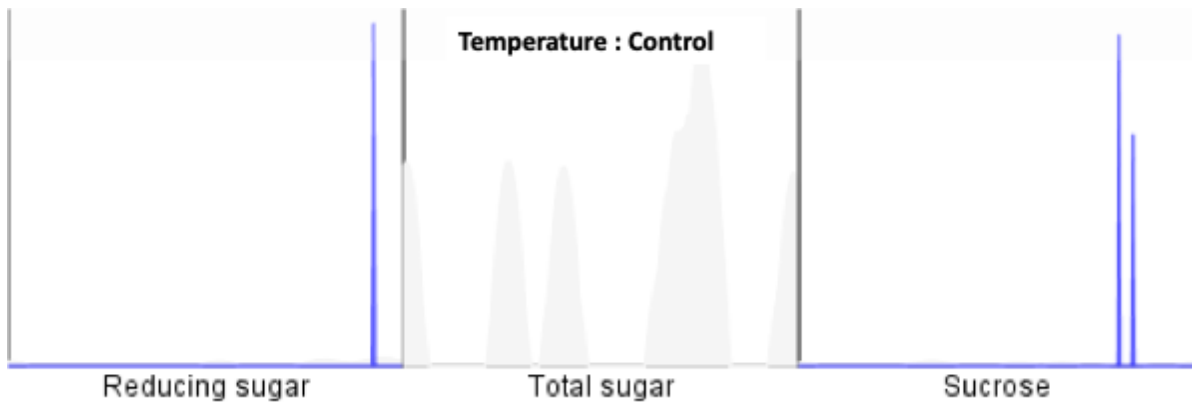
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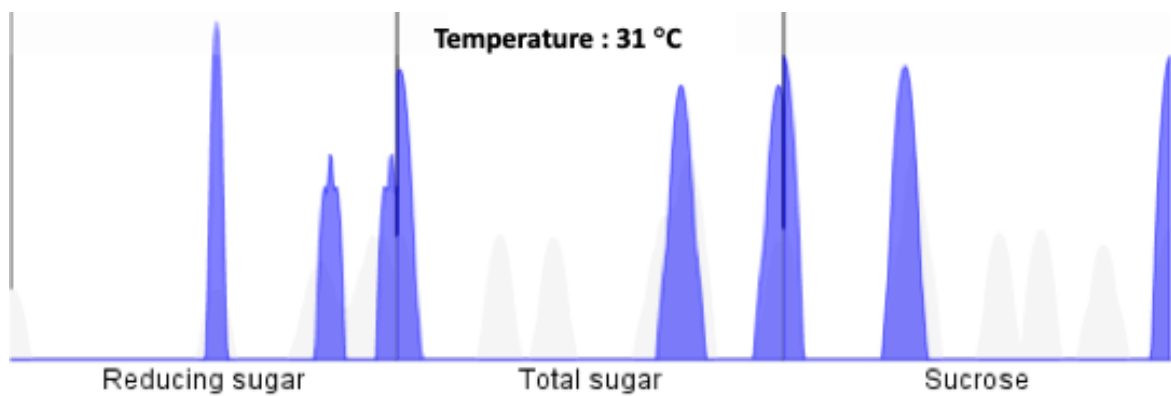
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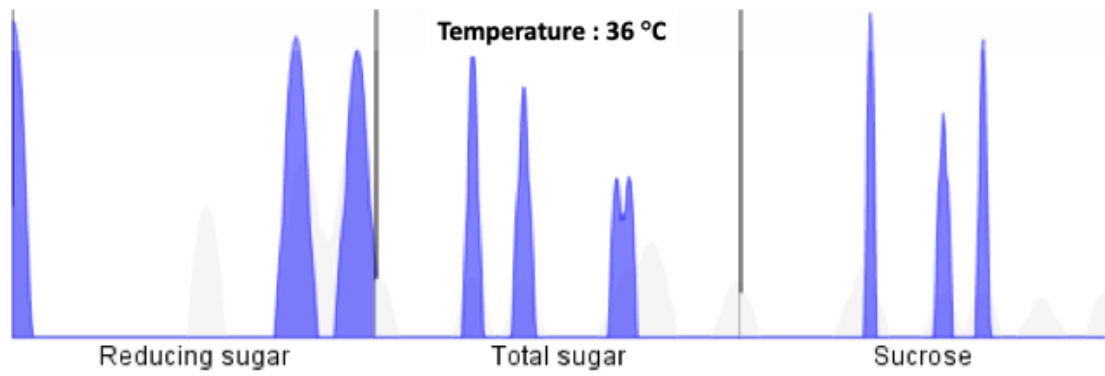
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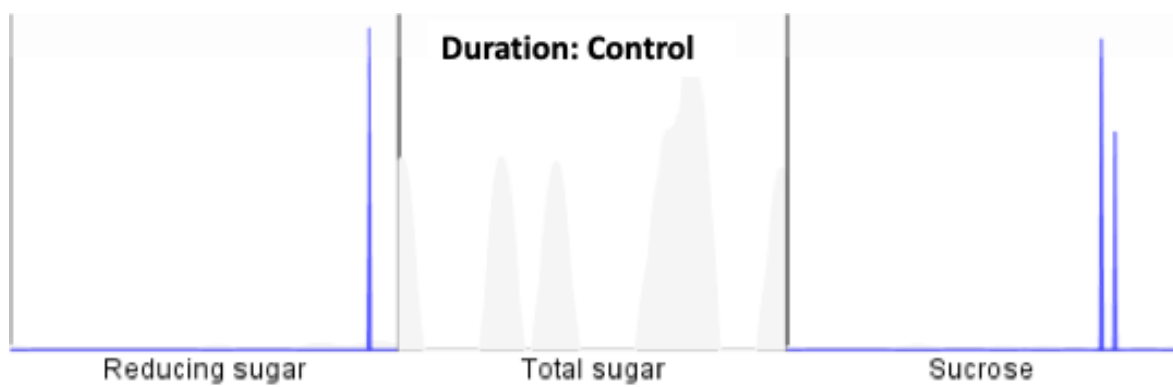
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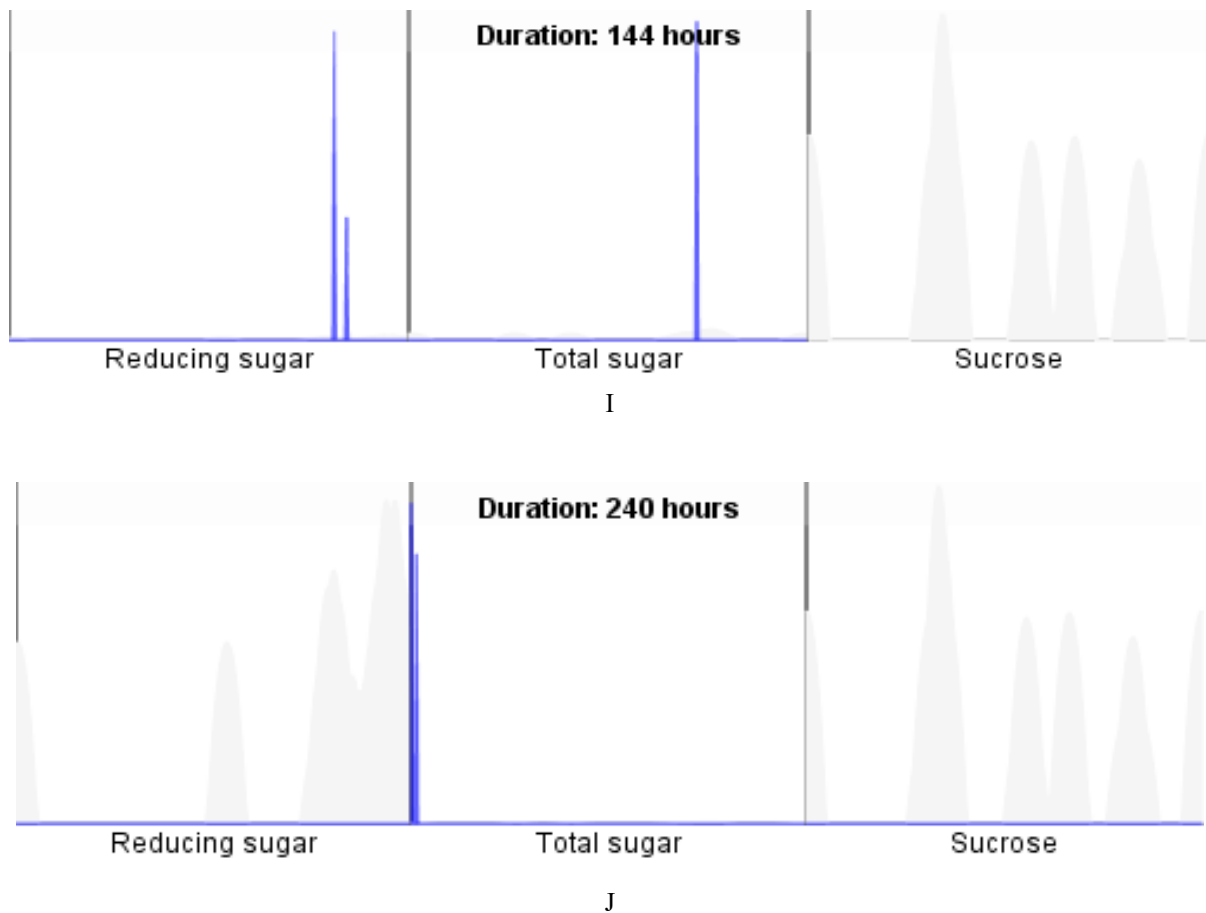


Figure 4. (a) Estimated marginal means (EMMs) of reducing sugar as a function of fermentation temperature (Control (C), 31 °C, and 36 °C) at four durations (Control (C), 48, 144, and 240 hours). (b) EMMs of total sugar versus temperature at the same durations. (c) EMMs of sucrose versus temperature at the same durations. (d) Quadratic model of sucrose versus temperature (open circles = observed data; line = quadratic fit) showing a peak at intermediate temperatures within the study range. (e) Heatmap of mean reducing sugar at temperature–duration combinations (Control (C), 31 °C, and 36 °C; Control (C), 48, 144, and 240 h). (f) Heatmap of mean total sugar at the same combinations. (g) Heatmap of mean sucrose at the same combinations. (h–j) Quadratic model of reducing sugar versus duration (open circles = observed data; line = quadratic fit).

The sugar content presented in Figure 4 shows that the combination of temperature and fermentation duration in ohmic-heated carbonic maceration significantly altered the sugar profile. At higher temperatures (31–36 °C), reducing sugars and sucrose appeared to increase in the initial phase (consistent with sucrose hydrolysis and mucilage removal) and then decrease as fermentation progressed, which is consistent with reported sugar utilization during coffee fermentation (microbial dynamics were not measured here). This agrees with the researchers in [29] who found that during wet fermentation of coffee, sucrose, glucose, and fructose in the mucilage decrease sharply over time; with yeast inoculation, the decline can occur more quickly and extensively. The reduction of sugars in the endosperm can also be accelerated through mucilage removal and microbially mediated consumption, which is important because reducing sugars trigger the Maillard reaction during roasting. This is consistent with the researchers in [15,30], who reported that fermentation time and temperature shift chemical precursors, particularly through the conversion of sugars into metabolites/organic acids, which affects sensory quality.

Epiphytic microorganisms have been reported to metabolize mucilage sugars and produce organic acids, alcohols, and esters that affect sensory quality [14]. Because microbial populations were not quantified in this study, this interpretation is presented as literature-consistent rather than a direct mechanistic conclusion. This is consistent with the trend shown in Figure 4, where, at 31–36 °C, sucrose hydrolysis is accelerated (creating a temporary spike in reducing sugars), but over longer durations, total sugar decreases as substrates are depleted during fermentation.

The role of ohmic heating is to quickly and uniformly control the temperature, making the kinetics of hydrolysis-fermentation more predictable. Studies have shown that ohmic heating provides uniform heating and high energy efficiency; its performance is strongly influenced by electrical conductivity and increases linearly with temperature [9]. Because conventional heating was not evaluated as a comparator in this study, we focus on temperature-control advantages within the ohmic-assisted system rather than comparative performance. With this type of thermal control, increasing fermentation temperature to 31–36 °C is associated with faster sucrose conversion early in the process, followed by decreasing total sugars at later stages [9].

Fermentation of Arabica coffee with variations in fermentation time and temperature has also been reported to decrease fructose (approximately 12 to 5 g L⁻¹), while sucrose and glucose decrease as temperature and time increase [18]. Overall, this evidence supports the interpretation that under ohmic-heated carbonic maceration, raising the temperature (31–36 °C) can accelerate sucrose hydrolysis and is associated with reducing-sugar depletion. Furthermore, extended duration leads to lower total sugars, which may influence the availability of flavor precursors during roasting [17].

3.4. Volatile compounds

The effect of carbonic maceration on volatile compounds was evaluated using solid-phase microextraction coupled with gas chromatography-mass spectrometry (SPME-GC-MS), following established protocols [5,31]. Volatile compounds are critical determinants of coffee flavor, as they originate from diverse chemical precursors in green beans that undergo thermal degradation and transformation reactions during roasting [32]. Although more than 800 volatile compounds have been identified in roasted coffee, only a limited subset, primarily sulfur-containing compounds, aldehydes, ketones, furans, pyrroles, and pyrazines, plays a dominant role in defining coffee aroma perception.

Principal component analysis (PCA) of volatile compound classes (Figure 5) explained 76.1% of the total variance, with Dim1 and Dim2 accounting for 44.3% and 31.8%, respectively. Fermented samples were separated from the control along Dim1, indicating a strong effect of fermentation on volatile composition. In contrast, the temperature difference between 31 °C and 36 °C contributed primarily to dispersion along Dim2. The variable biplot revealed that alcohols and their derivatives aldehydes, and ester-related variables were associated with longer fermentation durations, whereas treatments conducted at the higher temperature (36 °C) were more strongly associated with pyrazine- and benzenoid-related compounds.

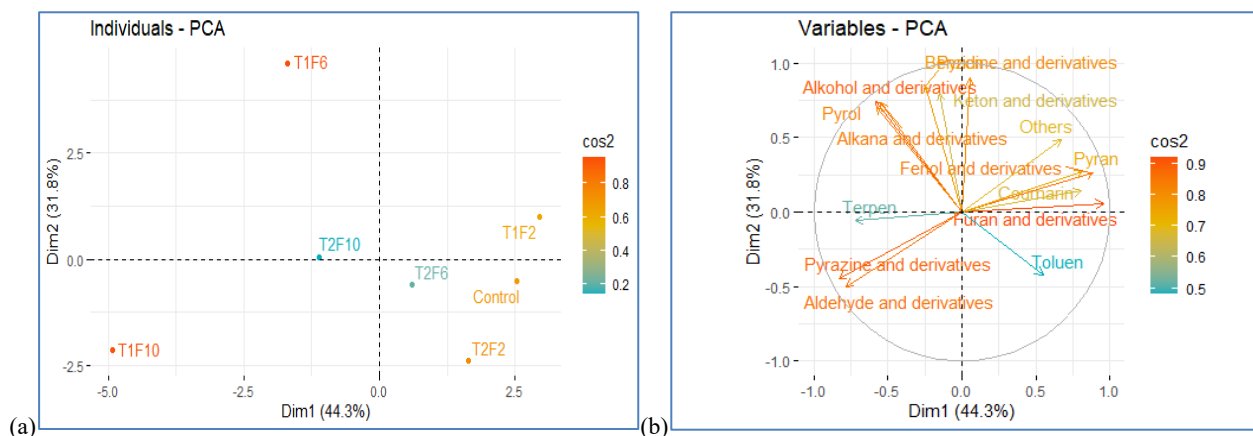


Figure 5. Principal component analysis (PCA) of volatile components (% peak area) in Arabica coffee produced by ohmic-assisted carbonic maceration fermentation: (a) score plot; and (b) loading plot.

These patterns are consistent with studies demonstrating that fermentation temperature and duration jointly modulate volatile synthesis and retention. Higher temperatures may favor the formation of higher alcohols and promote volatilization losses, shifting the aroma profile away from ester dominance, whereas moderate temperatures may favor ester accumulation [33]. Prolonged fermentation can enhance the release of certain alcohols and monoterpenes, while some ester classes decline, which is consistent with the increased contribution of alcohol-related variables observed at extended durations [34]. Under very long fermentation conditions, total volatile abundance may decrease despite increases in specific compounds, explaining why samples fermented for 240 h were not necessarily ester-rich but instead exhibited alcohol- and aldehyde-driven profiles [35].

Last, the finding from individual PCA that separates samples mainly by temperature on Dim2 is consistent with fermentation aroma literature, in which temperature influences the balance between ester formation and higher alcohol production (these mechanisms are used here only as context and not as direct evidence for coffee-specific pathways). Overall, the combination of 31 °C for 144 h showed a relatively stronger ester-related contribution in the PCA, whereas 36 °C for 240 h were associated with a more alcohol/aldehyde-dominant profile, consistent with temperature–time effects on volatile dynamics. To avoid overextension from non-coffee substrates, detailed wine-specific examples (e.g., individual grape-wine marker compounds) were removed, and the interpretation was on the PCA patterns observed in this dataset.

Reviews on fermented beverages further support the role of temperature regulation in shaping aroma matrices dominated by alcohols, aldehydes, ketones, esters, and terpenoids, with higher fermentation temperatures generally favoring higher alcohol formation [36]. Because these mechanisms are largely derived from non-coffee systems, we use them only to contextualize the temperature-driven shifts observed in our coffee volatile profiles, without implying identical pathways.

Studies on atmosphere-based fermentation report that carbonic maceration alters polyphenolic content and selected volatile fractions, and that temperature–time interactions are major drivers of volatile differentiation and cluster separation in multivariate analyses [37]. In this study, the GC-MS results are therefore interpreted primarily in relation to the experimentally controlled factors

(temperature and duration) under ohmic-assisted carbonic maceration. The concentrations of volatile compound classes quantified by GC-MS are summarized in Table 4.

Table 4. GC-MS concentrations (ppb) of volatile compound classes in coffee produced by ohmic-assisted carbonic maceration fermentation.

Classes	Control	Fermentation temperature 31 °C			Fermentation temperature 36 °C		
		48 h	144 h	240 h	48 h	144 h	240 h
Aldehyde and derivatives	213.71	177.22	109.68	333.92	247.77	250.82	194.59
Pyrazine and derivatives	596.02	534.96	397.55	872.09	766.04	606.48	376.96
Alcohol and derivatives	45.14	47.11	91.98	57.00	60.91	62.80	47.16
Phenol and derivatives	220.26	179.94	122.99	121.82	221.98	160.05	101.79
Alkane and derivatives	13.11	19.37	116.30	42.61	48.43	31.49	26.26
Furan and derivatives	3020.20	2144.54	1422.72	1282.88	2880.50	2014.57	1095.59
Pyridine and derivatives	35.27	49.30	37.88	19.11	21.09	23.53	16.59
Ketone and derivatives	199.07	208.90	156.23	139.13	180.06	151.01	108.86
Coumarin and derivatives	22.89	17.19	10.96	11.08	19.51	17.20	9.05
Benzene and derivatives	39.21	24.45	30.25	21.03	26.09	24.24	18.40
Terpene and derivatives	33.98	26.14	20.59	24.83	30.75	25.68	20.42
Pyrrole and derivatives	168.29	138.43	133.97	121.48	150.94	157.00	97.73
Toluene and derivatives	11.96	10.84	2.72	5.43	14.49	11.05	7.80
Pyran and derivatives	23.68	16.92	12.17	10.34	22.54	16.20	10.62
Others and derivatives	1097.58	874.61	698.34	646.69	1127.44	966.22	515.11

Table 4 demonstrates that aldehydes, pyrazines, and furans exhibited pronounced time- and temperature-dependent fluctuations during fermentation. For instance, at 36 °C, pyrazine concentrations increased at 48 h before declining at extended durations, whereas at 31 °C, pyrazine levels were comparatively more stable across durations. These trends suggest that temperature and duration jointly shape the abundance of key volatile classes within the ohmic-assisted system. Consistent with other reports, fermentation under ohmic heating can influence the volatile profile in coffee as a function of fermentation time and temperature [25]. Moreover, since we did not include a non-ohmic or conventional-heating comparator, we describe these changes within the experimental design rather than attributing them to superiority of ohmic heating. Links to sensory outcomes are evaluated directly in Section 3.5 using cupping results, rather than being inferred solely from volatile changes.

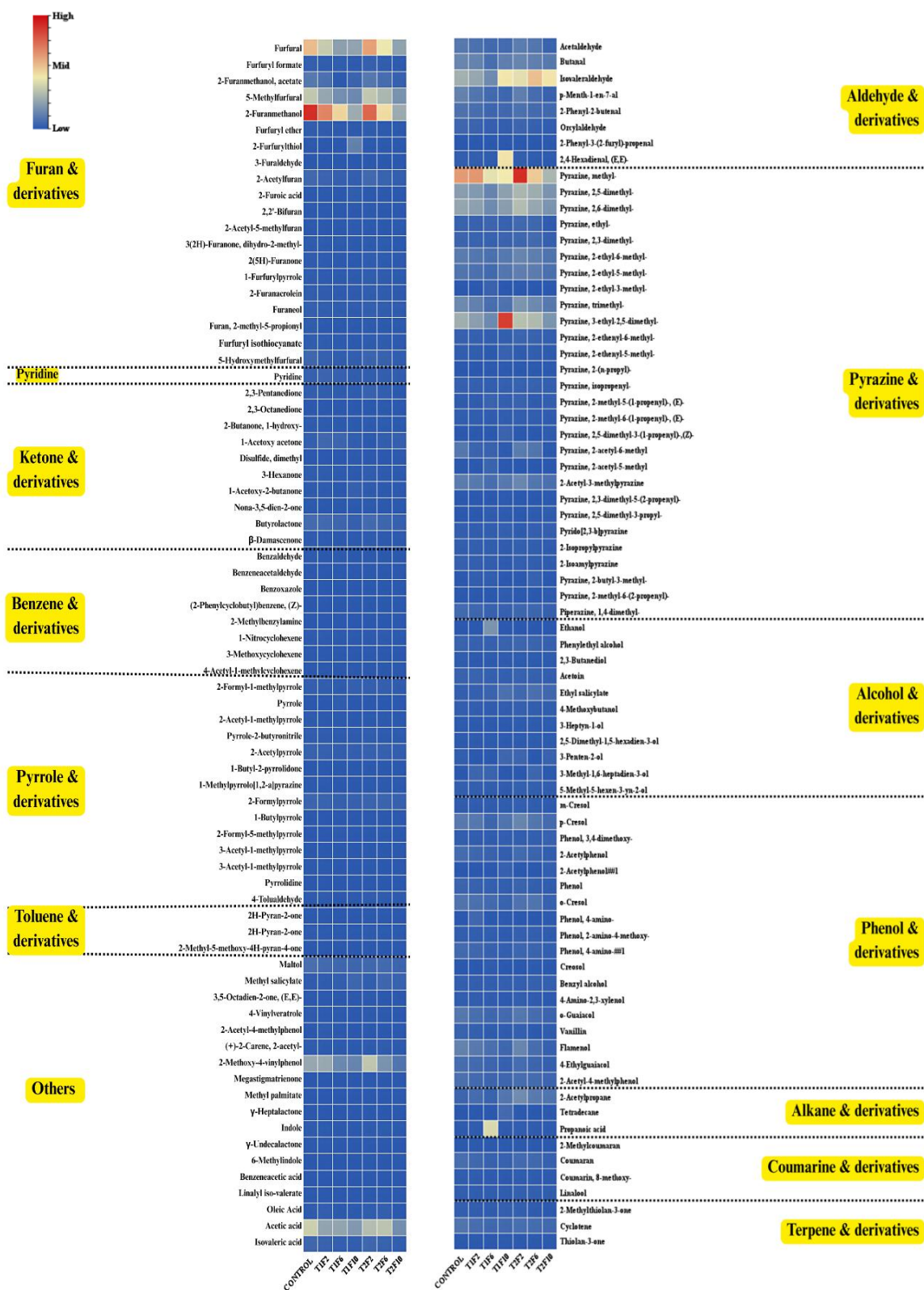


Figure 6. Heatmap visualization of 149 volatile compounds identified by GC-MS across treatments. Color intensity reflects relative fold change, where red indicates higher abundance and blue indicates lower abundance.

Heatmap visualization of GC-MS data (Figure 6) revealed that increasing fermentation temperature and duration shifted the volatile profile toward higher relative contributions of alcohol-related compounds and non-acetate esters, accompanied by a relative reduction in ethyl/acetate ester-associated signals compared with the non-fermented control. This pattern aligns with observations in fermentation-driven aroma systems, where prolonged maceration favors alcohol accumulation over ester retention [34].

Several key aroma-active compounds were identified, including 2-furfurylthiol, which possesses an extremely low sensory threshold and contributes strongly to roasted coffee aroma. Additional markers such as pyrazines, furans, aldehydes, and phenolic derivatives were detected and are known to differentiate coffee samples based on processing conditions and roasting level [5,38].

Taken together, the observed shifts in volatile compound classes, particularly the balance between esters, alcohols, pyrazines, and aldehydes, may contribute to measurable differences in sensory perception. To examine how these chemically driven changes are reflected at the sensory level, we evaluate the cupping profiles of the fermented coffee samples using the SCAA protocol in the following section.

3.5. *Sensory properties of coffee*

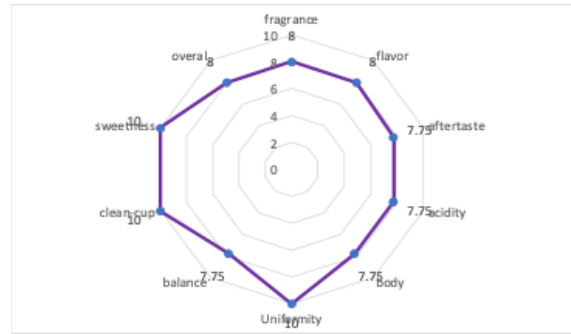
Sensory evaluation was conducted following the Specialty Coffee Association of America (SCAA) standard protocol at the Indonesian Coffee and Cocoa Research Center. The results of the sensory test (cupping) indicate that the cup test score of the coffee samples produced from carbonic maceration ranged from 83.0 to 85.75. From the six carbonic maceration treatments, four treatments (31 °C for 48 h; 31 °C for 240 h; 36 °C for 144 h; and 36 °C for 240 h) provided slightly higher (85.0–85.75) cupping scores than the control, one treatment (31 °C for 144 h) produced a cupping score comparable to the control (84.5), and one treatment (36 °C for 48 h) produced a lower score (83.0) than the control.

Based on Figure 7 (Aroma/Fragrance, Flavor, Aftertaste, Acidity, Body, Balance, Uniformity, Clean Cup, Overall, and Final SCAA score), the sensory profiles of treatments varied across temperature–duration combinations. In particular, 36 °C for 144–240 h tended to increase Body/Aftertaste and Balance scores, whereas 31 °C with 144 h tended to show a more balanced acidity–flavor profile with higher Clean Cup/Uniformity scores, contributing to higher overall scores. The role of fermentation time in shifting sensory attributes has been reported in prior work; for example, the SIAF study indicates that fermentation duration (0–96 h) can be associated with shifts in dominant notes (e.g., “woody,” “fruity,” and “herbaceous”), suggesting that the choice of duration should be aligned with the target profile [39].

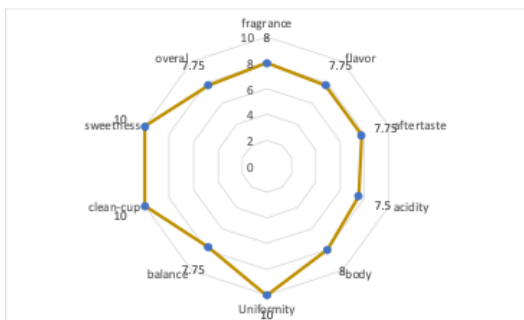
A = nutty, flowery-coffee blossom, and grassy



B = nutty, flowery-coffee blossom, lemony



C = brown sugar, spicy, chill-like, fruity, coffee pulp aroma, grassy



D = brown sugar, spicy chili-like, nutty



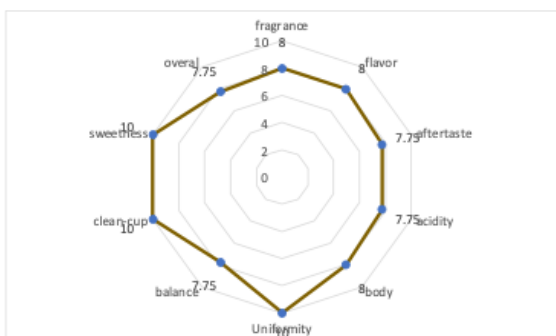
E = brown sugar, spicy-chili like, nutty



F = brown sugar, spicy-chili like, nutty, winery



G = brown sugar, spicy-like, fruity, winery



H = final SCAA score

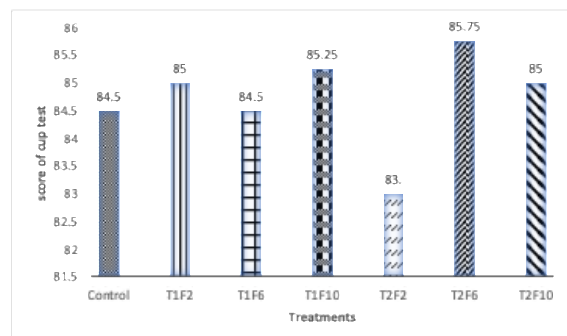


Figure 7. Effect of fermentation temperature and duration on SCAA sensory attributes of Arabica coffee: (A) Control, (B) T1F2, (C) T1F6, (D) T1F10, (E) T2F2, (F) T2F6, (G) T2F10, and (H) Final SCAA score. Brief cupping descriptors (aroma/flavor notes) for each treatment are provided to support interpretation of the sensory profiles. Decimal notation uses a period (e.g., 7.75).

In addition to duration, controlled fermentation techniques have been reported to influence sensory quality under SCA protocols, including at a bioreactor scale across 24–96 h [39]. Changes in sugars and volatile compounds can also arise under controlled fermentation conditions [13]. Regarding the temperature and anoxic environment typical of carbonic maceration, research indicates that natural/washed methods versus induced methods (including carbonic maceration) can be distinguished sensorially, and specialty samples may achieve maximum scores for Uniformity and Clean Cup [40]. In this study, ohmic heating is discussed as a means of maintaining stable fermentation temperature and reaching the set point more quickly; however, comparisons with conventional heating are not made because such a control was not included.

Carbonic fermentation in coffee has been reported to be influenced by temperature and time control [41]. Thus, the scores in Figure 7 are consistent with other findings: 31 °C at 144 h is associated with higher Clean Cup/Balance and a favorable acidity–flavor balance, while 36 °C after 144–240 h is associated with higher Body/Aftertaste, representing two processing pathways that can be tailored to the desired sensory profile.

4. Conclusions

The results showed that increasing fermentation duration decreased 3-caffeoylquinic acid and 4-caffeoylquinic acid, while 5-caffeoylquinic acid remained stable or increased at 36 °C for 240 h. Trigonelline exhibited relative stability at low temperatures but declined at prolonged durations, particularly at 36 °C. Caffeine concentrations peaked at intermediate durations before stabilizing. Sugar content generally decreased with increasing maceration duration, with significantly lower levels after 10 days of fermentation. Principal component analysis of volatile compounds revealed distinct clustering patterns among treatments, with long fermentation durations positively correlated with the production of phenols, alcohols, pyrroles, benzenes, pyridines, and alkanes. Sensory evaluation showed that four maceration treatments provided slightly higher cupping scores (85.0–85.75) than the control (84.5), with flavor notes including nutty, flowery, lemony, brown sugar, spicy, fruity, and winery aromas. To avoid ambiguity, “winery” is used here as a sensory descriptor (wine-like/fermentative note) reported during cupping, not as an indication of wine addition or wine-based processing. These findings demonstrate that Arabica coffee processed under ohmic-assisted carbonic maceration conditions exhibited measurable modulation of key chemical constituents, sugars, and volatile composition, leading to distinct sensory attributes under the studied processing conditions.

Use of AI tools declaration

AI tools were not used for data generation or analysis; minor language editing was assisted using digital tools.

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Conflict of interest

All authors mentioned above declare no conflict of interest.

Author contributions

A.M: investigation, formal analysis, writing-original draft. S.S: validation, resources, supervision. A.L: validation, visualization, supervision. F.B: validation, visualization, supervision. A.H: validation, visualization, supervision. M.M: validation, visualization, supervision. A.D: validation, visualization, supervision. A.I: validation, visualization, supervision. R.R: validation, visualization, supervision.

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