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Sensory profiles of Robusta coffee (*Coffea canephora*) genetic resources from the Democratic Republic of the Congo

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Introduction: The genetic diversity of Robusta coffee (*Coffea canephora*), a cornerstone in the global coffee industry, remains not fully explored, leading to a significant gap in our understanding of its sensory intricacies. Our study evaluated the sensory quality potential of the Robusta cultivars from the INERA Coffee Collection in Yangambi (the Democratic Republic of the Congo), the local wild diversity, and their hybrids.

Methods: We evaluated the sensory attributes of 70 genotypes representing the genetic structure of the coffee collection. Of those 70, 22 genotypes were evaluated for two consecutive years to assess the consistency of the sensory quality. Standard coffee cupping with the Fine Robusta Standards and Protocols was enhanced through sensory descriptors from the Coffee Taster's Flavor Wheel. Each genotype's sensory profile was constructed based on the Total cupping score and the frequency of reported sensory descriptors. The Total cupping score ranged from 75.75 to 84.75, with a substantial variation in sensory profiles, even within a genetic cluster.

Results and discussion: *Nutty/Cocoa* was the most frequently reported descriptor class. The sensory profile ideotype exhibits a high frequency of *Fruity*, *Sweet*, and *Sour/Fermented* descriptors and a low frequency of *Green/Vegetative*, *Other*, and *Roasted* descriptors. Evidence suggests that the sensory profile of a genotype is consistent over two harvest years. Genotypes with promising and unique sensory profiles were discovered within the cultivars and the wild – cultivar hybrids. The genetic diversity of wild and cultivated Robusta in the Democratic Republic of the Congo could play an essential role in understanding and improving its sensory quality.

KEYWORDS

Robusta, *Coffea canephora*, coffee quality, sensory descriptor, genotype, cultivar, hybrid, wild

1 Introduction

Robusta coffee (*Coffea canephora*) is a cornerstone in the global coffee industry, and its global market share is steadily increasing (ICO, 2023). However, in terms of quality, the spotlight often falls on Arabica (*Coffea arabica*) due to its more complex flavor profiles. Chemical compounds with a positive impact on the sensory characteristics of coffee have been found in higher concentrations in Arabica coffees. In contrast, compounds that impart negative sensory descriptors have been reported in higher concentrations in Robusta coffee (Toledo et al., 2016). Furthermore, Robusta breeding programs often did not consider the sensory quality of the coffee. When quality traits were considered, they focused on bean size, extractable soluble solids, and compounds such as caffeine (Leroy et al., 2011). Breeding for coffee quality in Arabica has seen consistent investments and development programs, resulting in improved varieties related to the sensory profile of the coffee (Montagnon et al., 2019). The sensory profile of Arabica coffee is associated with a milder taste, fruity flavor, and more acidic notes, whereas that of Robusta coffee is characterized by a bitter profile and earthy notes (Sunarharum et al., 2014; Seninde and Chambers, 2020). Although there is evidence that Robusta coffees can exhibit good quality and promising flavor profiles (Ngugi and Aluka, 2016; Augusto De Souza et al., 2018; Dalazen et al., 2020; Lemos et al., 2020; Morais et al., 2021) research on the distinct sensory profiles of Robusta has been minimal so far (Lingle and Menon, 2017).

Sensory science is commonly applied in the beverage industry. In coffee science, this includes the development of coffee cupping protocols and lexicons for sensory descriptors. The first cupping protocol for Arabica coffee was proposed in 2003 by the Specialty Coffee Association of America and has since become the industry standard (Lingle and Menon, 2017). The often inferior quality of Robusta reported in standard coffee cupping is attributed to its intrinsic genetic background, suboptimal cultivation, and post-harvest processing, resulting in different green coffee chemical compositions (Toledo et al., 2016). Robusta is therefore used to produce espresso, instant coffee, and coffee blends. However, a recent improvement in sensory quality with nuances has surprised coffee specialists and consumers (Baqueta et al., 2020). In 2019, the Fine Robusta Standards and Protocols were published to strengthen the market evaluation of Robusta coffee quality (Coffee Quality Institute, 2019). Both protocols focus on the quantitative evaluation of coffee quality through a scoring system on a 100-point total scale, with coffees of 80 points or more classified as Specialty (for Arabica) or Fine (for Robusta). However, the total score of a coffee reveals limited information about its intrinsic sensory profile. Two coffee samples with the same score could differ substantially in their aroma and flavor descriptors, so without sensory descriptors, the differentiation between coffees remains a matter of points on a scale. For this reason, lexicons and the so-called Coffee Taster's Flavor Wheel have been developed to enhance standard coffee cupping with sensory descriptors (Chambers et al., 2016; Spencer et al., 2016). However, these descriptors were skewed towards Arabica coffee, and Robusta coffee properties are underrepresented. Sensory descriptors have been used to predict Arabica coffee scores and market prices in which floral, fruity, sweet, and sour descriptors were most valued (Traore et al., 2018). Sensory descriptors were also used to predict the coffee roasting and brewing method (Bhumiratana et al., 2011) and describe the different terroirs' main characteristics (Scholz

et al., 2018). Overall, sensory descriptors are an excellent tool for better understanding sensory quality aspects and helping differentiate coffee samples. However, they are best used in parallel with the standard coffee cupping method (Di Donfrancesco et al., 2014).

Despite Robusta's market pervasiveness, the coffee species' genetic diversity remains not fully explored, leading to a significant gap in our understanding of its sensory intricacies. Robusta coffees show a greater difference in their sensory quality than Arabica, and these nuances are essential to understanding the difference between commercial and "Fine" Robusta (Lingle and Menon, 2017). An integrated development of new varieties should aim to characterize and exploit the rich genetic resources of this species (Prakash, 2018). In this context, characterizing genetic collections and identifying accessions with desirable sensory profiles can provide access to more genetic variation for breeding programs (Anthony et al., 2011). Therefore, the genetic diversity of wild and cultivated *C. canephora* in the Democratic Republic of the Congo (DR Congo) could play an essential role in valorizing wild and cultivated genetic resources (Bramel et al., 2017; Stoffelen et al., 2019).

The introduction of Robusta coffee from the Lomami River region in DR Congo (formerly Zaire) around 1900 was the starting point of the commercial cultivation of Robusta coffee (Leplae, 1936). This genetic material spread to Java for breeding and later returned to Central Africa (Van Der Vossen, 1985). From 1930 until 1960, l'Institut Nationale pour l'Étude Agronomique du Congo Belge (INEAC) was the leading breeding institute for Robusta coffee, with the principal research stations in Lula, Luki and Yangambi. By the 1950s, Yangambi had become the leading research station for Robusta coffee. The "Java" lines at the principal research stations were complemented with material from Gabon, Congo Brazzaville, and western DR Congo, as well as Ugandan material (Coste, 1955; Capot, 1962; Berthaud and Charrier, 1988). The developed elite lines were distributed throughout the tropics. This "INEAC" material is still the basis of Robusta production in several regions, e.g., West Africa (Montagnon et al., 1998), Vietnam (Vi et al., 2023), and DR Congo (Vanden Abeele et al., 2021). Other wild Congolese genetic resources have rarely been exploited since.

The National Agricultural Study and Research Institute (INERA), formerly INEAC, manages the *ex-situ* coffee collection in Yangambi, DR Congo. Due to instability in the region, weak governance, and lack of resources, the INERA Coffee Collection in Yangambi was decimated, and much documentation was lost. In collaboration with Meise Botanic Garden (Belgium), this collection has been screened, rehabilitated, and enriched with new genetic material from the wild and local home gardens (Stoffelen et al., 2019; Vanden Abeele et al., 2021). Previous research differentiated eight genetic groups of *C. canephora* corresponding to different geographic origins across West and Central Africa (Merot-Lanthoene et al., 2019). The geographical regions of Congolese subgroups A, B, E, and R overlap with the west, north, central, and southeast of DR Congo, respectively. A recent, comprehensive genetic analysis of the INERA Coffee Collection revealed a relatively broad genetic diversity. Materials from the Congolese subgroup A (likely corresponding to materials initially derived from the INERA Research Station in Luki) and subgroup BE (hybrid between subgroups B and E; likely corresponding to "Wild" genotypes from the rainforest in the Yangambi region) were discovered. However, the most abundant materials in the INERA

Coffee Collection in Yangambi are known and distributed as “Lula” cultivars, and are currently of unknown origin with respect to the natural distribution range (see detailed explanation in Verleysen et al., 2023). In addition, the INERA Coffee Collection also contains individuals with an admixed genotypic background and are likely derived from hybridization between “Lula” and subgroup BE (local “Wild”) or “Lula” and subgroup A. The coffee collection has not been phenotyped for coffee bean quality and sensory profiles since 1960 (Coste, 1955). The new hybrids and introduced wild genetic resources from the Yangambi bioserve have never been evaluated.

This study aims to unravel the sensory quality potential of the Robusta genetic resources from the present INERA Coffee Collection in Yangambi, the successor of the former INEAC coffee collection. Therefore, we capitalized on the available genotyping data of the collection from Verleysen et al. (2023), selected representative genotypes (including clonal replicates, i.e., sets of genetically identical trees), and harvested beans from coffee plants grown under the same environmental conditions. We combined standard coffee cupping following the Fine Robusta Standards and Protocols with sensory descriptors defined by the Coffee Taster’s Flavor Wheel to discover genotypes with promising sensory profiles. We were specifically interested in the differences in sensory profiles among local forest and cultivar accessions.

2 Materials and methods

2.1 Site description and genetic material

The INERA Coffee Collection in Yangambi is located in the Tshopo province of DR Congo (Lat: 0°50′59.60″N Long: 24°27′50.85″E, 485 m altitude). The region is classified as Köppen Af (rainforest) with a dry season, average annual precipitation of 1837 mm, and a mean annual temperature of 25.1°C (Kasongo Yakusu et al., 2023). The soil of the Yangambi region is classified as Ferralsol, a strongly weathered soil with low nutrient-holding capacity (Jones et al., 2013). The coffee collection is maintained as an unshaded monoculture system with coffee trees spaced 2.5×2.5 meters apart (Supplementary Figure S1A). Agricultural management practices are limited to mulching, weeding, and pruning without irrigation and no use of fertilizer and pesticides.

A recent genetic fingerprinting analysis of the INERA Coffee Collection by Verleysen et al. (2023) revealed the genetic structure of the collection and delineated three main groups of origin. First, the “Lula” cultivars originated in the Lula breeding station of DR Congo and are currently of unknown geographic origin. Second, the INERA accession belonging to the previously identified Congolese subgroup A (Merot-Lanthoene et al., 2019), comprise “Luki” cultivar material originating from another INERA breeding station in Luki, DR Congo (see Supplementary Figure S1B) and “Petit-Kwilu” material. Third, local Wild accessions that were introduced in the collection between 2015 and 2019 from the nearby rainforest in the Yangambi region belonging to the Congolese subgroup BE (Verleysen et al., 2023). Finally, admixed genotypes that are likely Lula – Wild hybrids and Lula – Congolese subgroup A hybrids were identified. An overview of the genetic structure of the INERA Coffee Collection in Yangambi in relation to the three origin groups as described in Verleysen et al. (2023) is given in Supplementary Figure S1C.

We selected seventy genotypes and subdivided these into five classes: Wild ($n = 3$), Lula – Wild hybrids ($n = 14$), Lula ($n = 39$), Lula – Congolese subgroup A hybrids ($n = 13$), and Congolese subgroup A ($n = 1$). Within this selection, coffee cherries of 22 genotypes (1, 2, 13, and 6, respectively, from each class) were harvested for two consecutive years (2021 and 2022). If different clones were available for a genotype, based on the genetic fingerprinting of Verleysen et al. (2023) up to three clones were selected, and their harvested coffee cherries were pooled. When applicable, the same chosen clones were studied during both harvest years. The age of the trees was based on the planting dates from recent INERA documentation (Supplementary Table S1). By pooling and processing coffee cherries of three different clones, biological replicates were taken for G0002 and G0003 for harvest year 2021 and G0008 for harvest year 2022. This resulted in 95 coffee cherry samples for this study.

2.2 Coffee cherry harvest and processing

Cherries of 55 genotypes were harvested from November to December 2021, and 37 genotypes were harvested from November to December 2022, with 22 genotypes harvested in both years. Between 250 and 500 grams of ripe red cherries were hand-picked per tree in the morning and pooled when clonal material was available. Harvested samples were floated in water to remove unripe and infected cherries. The cherries were sun-dried on a concrete patio during the day and covered during the nighttime and periods of rain. Moisture content was monitored daily with a TG pro coffee moisture meter (Draminski S.A., Poland). The dried samples with a moisture content between 10 and 13% (wet basis) were hulled with a manual huller. The green coffee samples were transported to Meise Botanic Garden (Belgium) in Ziplock bags. Green coffee samples were stored in the dark at room temperature (20°C) for four months.

2.3 Sensory analysis

2.3.1 The Fine Robusta Standards and Protocols

The quality of the genotypes was assessed through a standard coffee cupping by the Fine Robusta Standards and Protocols (Coffee Quality Institute, 2019). The green coffee samples were cleaned of any green bean defects (black, sour, immature, broken, malformed, fungus, and insect-damaged beans). Before roasting, moisture content was measured with a Sinar BeanPro Meter (Graintec, Australia). Fifty grams of green beans were roasted to a medium degree with the same roast profile. A medium roast profile was developed with an IKAWA® Sample Roaster V2 Pro (London, UK) and evaluated with a colorimeter COLORTTEST II (Neuhaus Neotec, Germany). Variations in the roast degree for each sample were monitored as the ratio of roasted coffee weight (g) to green coffee weight (g), i.e., weight loss ratio. Roasted samples were ground with a Mahlkönig Guatemala grinder (Zürich, Switzerland). A ratio of 8.75 ± 0.25 g coffee to 150 mL water at $93.5 \pm 1.5^\circ\text{C}$ was used for infusion of the coffee. The Total Dissolved Solids of the water was 130 mg/L (Volvic, France). Three licensed quality graders (Q-graders) performed a blind standard coffee cupping evaluation of the coffee samples. The attributes of Fragrance/Aroma, Flavor, Aftertaste, Salt/Acid ratio, Bitter/Sweet ratio, Mouthfeel, Balance, and Overall score of the coffee were

evaluated. Each attribute was scored from 0 to 10 in steps of 0.25. This score was added to the Uniformity and Clean Cup scores, penalizing any sensory defect. The sum resulted in a total score of a maximum of 100, with scores equal to or above 80 being considered high-quality Robusta (Fine). The mean total score of the three Q-graders was used for this study as the Total score per sample.

2.3.2 Sensory descriptors

The Coffee Taster's Flavor Wheel was used as a standard language tool for notating sensory descriptors, hereafter referred to as "descriptors" (Spencer et al., 2016). The three Q-graders were trained in reporting these descriptors during three standard coffee cupping sessions. Descriptors were reported as detailed as possible for three attributes: dry ground coffee fragrance, water-infused coffee aroma, and flavor during gustation. Each reported descriptor was then categorized into one of the nine main descriptor classes of the Coffee Taster's Flavor Wheel, i.e., *Green/Vegetative*, *Other*, *Roasted*, *Spices*, *Nutty/Cocoa*, *Sweet*, *Floral*, *Fruity*, and *Sour/Fermented*. If at least one descriptor was reported for a main class, the value of "one" was assigned. Otherwise, the attribute received a value of zero. The sum of the reported descriptors for dry fragrance, wet aroma, and flavor was made per coffee sample, in which each class can take a value between zero and three. The sum of the reported frequencies was made over the three Q-graders, resulting in a frequency table for each main descriptor class with values between zero and nine.

2.4 Data analysis

A multiple linear regression model was built with the 95 coffee samples to quantify the relationship between the Total score and the descriptor classes. The descriptor classes were the independent variables, and the Total score was the dependent variable while controlling for tree age and the roasted weight loss ratio. The full regression model was reduced through backward elimination by iteratively removing the non-significant independent variables. The *F*-statistic, AIC, and BIC values were used to select the best-fit reduced model. Pearson correlations were calculated for the tree age and roasted weight loss ratio with the Total score and nine descriptor classes. The probability levels of the correlation coefficients were Bonferroni corrected. A Paired Samples *t*-test was used to test for significant differences between the mean Total score for the selected 22 genotypes harvested in two years and between the biological replicates. Multiple logistic regression of the nine descriptor classes (independent variables) on the harvest year (dependent dummy variable) was performed with the 22 selected genotypes. The mean of the Total score and the mean frequency of the descriptor classes over the two harvest years were used for the 22 genotypes. The biological replicates were not used in creating the sensory profile of the genotypes. Descriptive statistics, correlations, and regressions were performed in R (version 4.2.3) with RStudio (2023.09.1). A sensory profile was established for each evaluated genotype based on the Total score and the frequency table of the main descriptor classes. Sensory profile figures were created with Tableau Desktop (version 2023 2.0), in which the reported mean Total scores were rounded to the 0.25 decimal.

3 Results

3.1 Standard coffee cupping results

The Total score of the 95 evaluated coffee samples ranged from 75.75 to 84.75 points (Supplementary Table S1). An outlier in the total score (87.75) for one Q-grader was found for genotype G0131 and was omitted. The mean Total score of this single genotype was calculated from the results of the remaining two Q-graders (80.00 and 79.00). The frequency of descriptors was the sum of the three Q-graders. The standard deviation (SD) of the mean Total score of the three Q-graders ranged from 0.14 to 3.50, and the absolute spread in Total points ranged from 0.25 to 6.50. The average SD was 1.05, and the average spread was 2.00 over the 95 coffee samples, which was low compared to previous research on the reproducibility of standard coffee cupping (Worku et al., 2016).

3.2 Relationship between the Total score and the nine descriptor classes

The estimated regression coefficients and goodness of fit of the full regression model (Total score regressed on the frequency of the descriptor classes, the tree age, and the weight loss ratio) are reported in Supplementary Table S2. After backward elimination, the independent variables *Floral*, *Nutty/Cocoa*, and *Spices* were omitted from the model. The tree age and the roasted weight loss ratio variable were insignificant and eliminated. The regression model was significant ($p < 0.001$) and explained 64.37% (R^2 -adj) of the variation in the Total score (Table 1). *Green/Vegetative*, *Other*, and *Roasted* variables had significant negative coefficients, whereas *Sweet*, *Fruity*, and *Sour/Fermented* were significantly positive. For example, every increase in the frequency of the *Fruity* descriptor reported during sensory analysis increased the Total score of the coffee by 0.27 points.

TABLE 1 Regression coefficients from the reduced multiple linear regression model of the descriptor classes on the Total score for the 95 coffee samples.

| Variable | Estimate | Std. error |
|---------------------|----------|------------|
| (Intercept) | 81.31*** | (0.52) |
| Green/Vegetative | -0.30*** | (0.06) |
| Other | -0.25** | (0.09) |
| Roasted | -0.20** | (0.07) |
| Sweet | 0.22* | (0.10) |
| Fruity | 0.27*** | (0.06) |
| Sour/Fermented | 0.39** | (0.15) |
| <i>F</i> -statistic | 29.31 | |
| <i>p</i> -value | <0.001 | |
| <i>df</i> | 88 | |
| AIC | 307.15 | |
| BIC | 327.58 | |
| R^2 | 0.67 | |
| R^2 -adj | 0.64 | |

* $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$.

The tree age ranged from 3 to 18 years and did not significantly correlate with the Total score or any of the nine descriptor classes. The roasted weight loss ratio ranged from 0.82 to 0.90 and correlated significantly negatively with the *Green/Vegetative* class ($r = -0.34$, $p = 0.009$) and positively with the *Fruity* class ($r = 0.30$, $p = 0.029$) (Supplementary Table S3). More roasted weight loss correlated weakly with more reported *Green/Vegetative* descriptors and less reported *Fruity* descriptors in the coffee.

3.3 The consistency of the sensory analysis

The Pairwise *t*-test reported a significant difference ($p = 0.048$) in the mean Total score of the three Q-graders between the two harvest years for genotype G0120 (Figure 1). The mode of the absolute difference in frequency over the two harvest years was one for the *Other*, *Roasted*, *Spices*, *Nutty/Cocoa*, *Sweet* and *Fruity* class and zero for the *Green/Vegetative*, *Floral* and *Sour/Fermented* class. The absolute difference in frequency ranged from zero to seven over the nine classes (Figure 1). Additionally, there was no significant difference in the mean total score of the three Q-graders between the biological replicates of G0002, G0003, and G0008. The absolute difference in frequency of descriptors was the highest for the *Other*, *Fruity*, and *Green/Vegetative* classes for biological replicates of G0002, G0003, and G0008, respectively (Figure 1). The logistic model evaluating the environmental effect of the harvest year on the descriptor class frequency was significant (LR $\chi^2 = 23.25$, $p = 0.006$). The *Green/Vegetative* and *Nutty/Cocoa* descriptor classes were significantly associated with the harvest year. Both descriptor classes significantly predict a coffee sample to belong to the 2022 harvest year (Table 2). The Pairwise *t*-test showed one significant difference between the mean Total score of the 22 genotypes at the 5% level. The mode of absolute difference in frequency of the nine descriptor classes was no higher than one. Two descriptor classes were associated with the harvest year. The mean Total score and frequency of the descriptor

classes over the two harvest years were used to create the sensory profiles of these 22 genotypes.

3.4 Sensory profiles of the genotypes

The total reported descriptors of the nine descriptor classes for the 70 genotypes are illustrated in Figure 2. Additionally, we report descriptors for the genetic classes Lula – Wild hybrid, Lula, and Lula – subgroup A hybrid. Overall, the *Nutty/Cocoa* descriptor class was most common (333), followed by the *Roasted* (227), *Spices* (199), and *Fruity* (196) classes. The *Floral* (7) class was the least reported descriptor. The Lula – Wild hybrid class and Lula – subgroup A hybrid class exhibited relatively more *Fruity* descriptors than the Lula class. The Lula – Wild hybrid class exhibited a relatively higher fraction of reported *Green/Vegetative* and *Other* descriptors. The sensory profiles of the 70 genotypes are reported based on the Total score and reported frequency of the nine descriptor classes (Figure 3). The sensory profiles were sorted into the assigned genetic classes, i.e., Wild, Lula – Wild hybrid, Lula, Lula – subgroup A hybrid, and Congolese subgroup A, representing the genetic structure of the INERA Coffee Collection. Genotypes with a remarkable sensory quality were highlighted and further described for each class based on the plant label information from available INERA documentation.

The three Wild genotypes had a relatively low Total score and did not exhibit promising sensory profiles.

Within the Lula – Wild hybrid class, G0028 scored a high Total score (83.25). G0042 had a low Total score (76.75) and was predominantly described by *Green/Vegetative* descriptors. Genotype G0058 (Total score 82.50) had a unique sensory profile and was the only sample from this study with reported descriptors in each of the nine classes. This accession originated from the local forest. Remarkable in this genetic class were G0064 (83.75) and G0067 (83.00), which both exhibited a high frequency of *Fruity* descriptors, with G0064 reporting the most *Sour/Fermented* descriptors. These

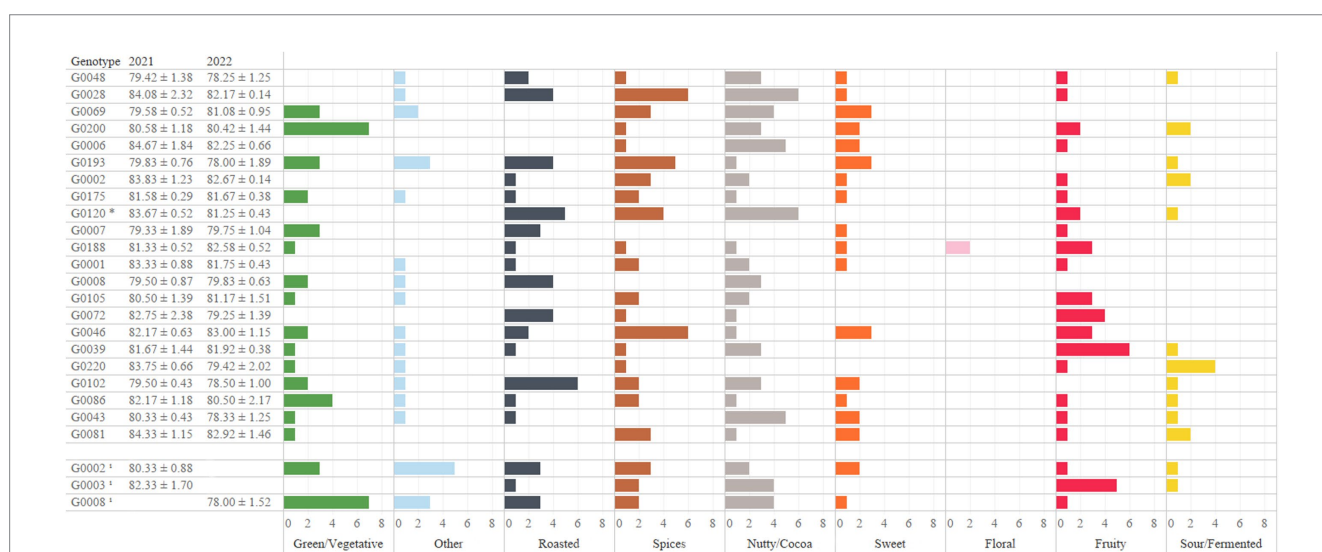


FIGURE 1 Mean and standard deviation of the Total score alongside the absolute difference in descriptor class frequency between harvest year 2021 and 2022 for the 22 genotypes and three biological replicates. *Significant difference between the mean total score of harvest year 2021 and 2022 ($p < 0.05$). [†]Biological replicates of the genotype for the same harvest year. Parallel measurement of G0003 is reported in Figure 3.

accessions originated from the local forest. G0023 (83.75) also reported a high frequency of *Fruity* descriptors. G0035 (83.25) reported a high frequency of *Spices* and *Nutty/Cocoa* descriptors. It was labeled as a Beni/Uganda accession, as were G0175 and G0192 from this genetic class.

For the Lula class, several sensory profiles stood out. G0262 (84.00) was exceptional due to the high frequency of *Fruity* and *Spices* descriptors combined with the presence of *Sweet* and *Floral* descriptors. *Green/Vegetative*, *Other*, and *Roasted* descriptors were absent. G0262 was labeled as Beni/Uganda origin, but its sensory profile stood out from genotypes with the same accession origin. Another remarkable genotype was G0103 (84.25), with a high frequency of *Fruity* and *Sour/Fermented* descriptors. Other noteworthy *Fruity* profiles were found in G0026 (83.25) and G0095 (81.75). The latter was labeled with the same Beni/Uganda accession as genotype G0262. Genotypes G0002, G0094, G0119, G0131, and G0134 were all labeled by INERA as clonal material from the same elite Lula line and harvested from the same plot. These unique genotypes scored between 79.50 and 83.75 points. The lowest Total score in this study was for genotype G0071 (75.75), with the maximum reported frequency of

Green/Vegetative descriptors. This accession originated from the local forest.

In the Lula – subgroup A hybrid class, G0079 scored high (83.50) and was free of *Green/Vegetative*, *Other* and *Roasted* descriptors. It was labeled as an old remnant of breeding (“époque belge”). G0222 scored 84.00 and stood out from the other genotypes in this class due to its tall tree size. G0073 scored the highest Total score in this study (84.75) with a high frequency of *Fruity* and *Sweet* descriptors and an absence of *Green/Vegetative*, *Other* and *Roasted* descriptors. It was labeled as a “Petit-Kwilu” accession. G0081 was notable due to its high Total score (83.75) and *Fruity* profile. It was labeled as Indonesian origin. Only one genotype represented the Congolese subgroup A class. G0087 had a Total score of 78.25 and was described mainly by *Other* descriptors.

4 Discussion

A detailed sensory analysis of the numerous genotypes from the INERA Coffee Collection in Yangambi was worthwhile, as we discovered high-quality genotypes that would otherwise remain undervalued. This aligns with previous research on germplasm banks and accessions in Arabica and shows the importance of screening coffee genotypes for cupping quality potential and further breeding (Sobreira et al., 2016). In our study, natural processed (sundried coffee cherries) and clean coffee samples (no green bean defects) were used and revealed sensory quality scores between 75.75 and 84.75 points. A comparison of the quality of the studied genetic resources with Robusta material worldwide must be made with caution as sensory quality scores depend on growing conditions (shade, altitude, management practices) and post-harvest processing methods like drying and sorting of the beans. For example, a previous sensory evaluation of Robusta coffee samples from 18 Robusta-producing countries reported a considerable variation in sensory quality (67.75 to 82.75 points). However, if we only consider the natural processed coffees free from defects, the range in sensory quality was 78.00 to 80.25 points (ICO, 2010). For references to the sensory quality of Robusta, only sensory quality scores from natural processed coffee samples with proper processing are discussed here. For instance, the sensory quality screening of Robusta germplasm material and cultivars from Uganda revealed a considerable variation in sensory quality, in which six out of the 206 genotypes were categorized as Fine Robusta

TABLE 2 Regression coefficients from the multiple logistic regression of the descriptor classes on the harvest year.

| Variable | Estimate | Std. error |
|------------------|----------|------------|
| (Intercept) | -8.50* | (3.65) |
| Green/Vegetative | 0.81* | (0.38) |
| Other | -0.14 | (0.4) |
| Roasted | 0.29 | (0.27) |
| Spices | 0.21 | (0.27) |
| Nutty/Cocoa | 1.02** | (0.34) |
| Sweet | 0.20 | (0.46) |
| Floral | -8.32 | (1199.77) |
| Fruity | 0.32 | (0.28) |
| Sour/Fermented | -0.98 | (0.78) |
| N | 44 | |
| LR χ^2 | 23.25 | |
| p-value | 0.006 | |

*p < 0.05, **p < 0.01, ***p < 0.001. A dummy variable was used for the harvest year (2021 value zero, 2022 value one).

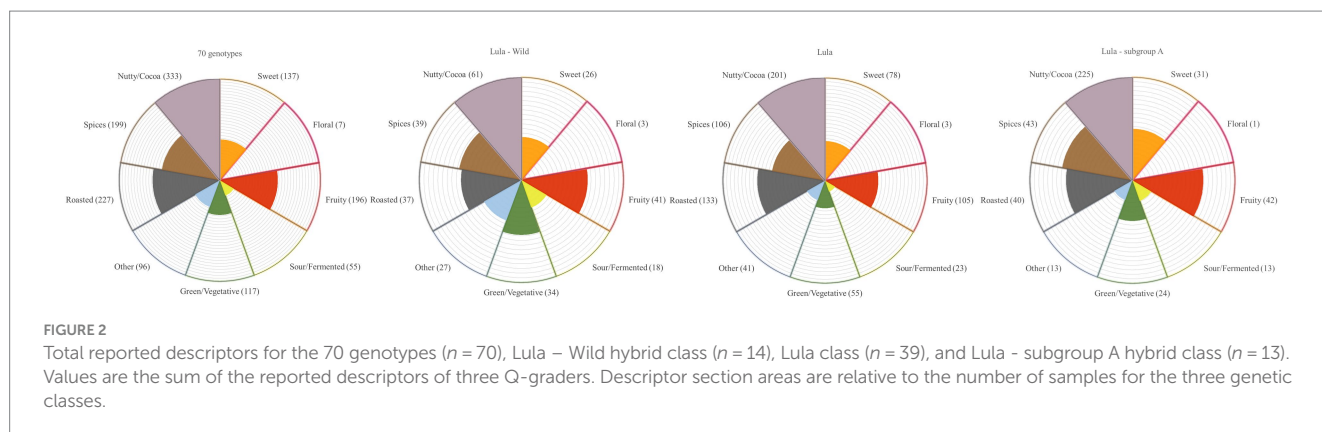




FIGURE 3 Sensory profiles of the 70 genotypes representing the INERA Coffee Collection in Yangambi. Genetic structure based on the fastSTRUCTURE bar plot representing three genetic clusters ($K = 3$). Colors define subpopulations: orange (Wild), blue (Congolese subgroup A), and red ("Lula" cultivars) (Verleysen et al., 2023). Wild, Lula - Wild, Lula, Lula - subgroup A hybrid, and Congolese subgroup A classes. ¹ Sensory profile created from the mean Total score and mean frequency of descriptors over the two harvest years.

(quality score above 80 points) (Ngugi and Aluka, 2016). A study on Brazilian Robusta cultivars categorized two out of the six genotypes as Fine Robusta (dos Santos Gomes et al., 2023) while another study categorized one out of four genotypes as Fine Robusta (Lemos et al., 2020). Robusta cultivars from the Western Amazon ranged between 76.50 and 81.50 points (Dalazen et al., 2020). A large-scale comparison of inter-varietal hybrids between genetic material from Congolese subgroups A and E did not report genotypes with a quality score above 80 points (Augusto De Souza et al., 2018) New Robusta varieties from Vietnam scored between 72.50 and 81.75 points (ICO, 2019). Previous research focused on the scores of standard coffee cupping, while our study enhanced the

screening and differentiation of coffee sensory profiles through sensory descriptors.

The relationship between the Total score and the frequency of the nine descriptor classes indicated a desired sensory profile for Robusta coffee. A sensory profile ideotype would exhibit a high frequency of Fruity, Sweet, and Sour/Fermented descriptors and a low frequency of Green/Vegetative, Other, and Roasted descriptors. This is similar to the sensory profile ideotype of Arabica coffee. In order of importance, the preferred quality attributes for cuppers and buyers of high-quality Arabica coffee were: Fruity, Floral, Sweet, and Sour descriptors. Other and Roasted were least valued (Traore et al., 2018). Interestingly, the Sour/Fermented class had the highest impact on the Total score out of

the nine descriptor classes. This was attributed to the reported frequency of the “winey” descriptor from this descriptor class. The *winey* descriptor is defined as the “sharp, pungent, somewhat fruity, alcohol-like aromatic associated with wine” (Chambers et al., 2016). Even though all coffee samples in our study were subjected to blind evaluation and descriptors were reported as objectively as possible during sensory analysis, a bias in the relationship between descriptors and the Total score should be considered. Q-graders have been trained to associate certain descriptor classes with quality, as these classes are desired commercially (Traore et al., 2018). This bias can also be found in the Fine Robusta Standards and Protocols, which states that flavor notes in high-quality Robusta include fruity, nutty, spices, and sweet descriptors. Lower-grade Robustas commonly include vegetative, cereal, burnt, chemical, and papery sensory notes, which coincide with the *Green/Vegetative*, *Roasted*, and *Other* classes (Coffee Quality Institute, 2019). The *Other* class sensory descriptors were uncommon in the evaluated coffee samples of our study. This was not unexpected, as the coffee samples were cleaned of any defects associated with this descriptor class. A previous study that used properly processed cleaned coffee samples rarely observed “earthy” descriptors (*Other* class) during the sensory evaluation of the genotypes (Moschetto et al., 1996).

The variation in the roast degree, expressed as the weight loss ratio, correlated weakly with the *Green/Vegetative* and *Fruity* class. Coffee aroma evolves with the roast degree, in which light roasts are described by more fruity and sweet aromas, whereas dark roasts are linked with cocoa, spices, and ashy aromas (Poisson et al., 2017). The fruitiness of coffee was negatively correlated with roast degree and roasted descriptors in Münchow et al. (2020). Our study used the same medium roast profile for all coffee samples, yet a range in roasted weight loss ratio was observed. Two of the nine descriptor classes significantly correlated (weakly) with roasted weight loss ratio, so the control for the roast degree in the analysis of descriptors was acceptable. The consistency of the Total score for genotypes evaluated over the two harvest years was good, as only one significant difference in Total score was reported. The same can be concluded for the three biological replicates from the same harvest year, where no significant differences in Total scores were reported. Regarding the frequency of reported descriptors, the consistency was less clear. The *Green/Vegetative* and *Nutty/Cocoa* classes were more frequent, depending on the harvest year. The exact genotypes were known, so environmental factors from the coffee samples’ harvesting, processing, and preparations are more likely to explain the variations in descriptors between the harvest years. A correlation was found between the *Green/Vegetative* class and roasted weight loss ratio. However, the difference in frequency of the *Green/Vegetative* class between the harvest years was not significantly correlated ($r = -0.17$) with the difference in weight loss ratio between the harvest years for the 22 genotypes. Climatic factors could explain this harvest year effect, as a study on Arabica coffee reported a positive correlation of green (*Green/Vegetative*) flavors with temperature and solar radiation during seed development (Bertrand et al., 2012).

We presented a range in Total scores and frequency in descriptor classes for the 70 genotypes of the coffee collection. While the predominant descriptors were *Nutty/Cocoa* and *Roasted* (Figure 2), the substantial presence of the *Fruity*, *Sweet*, and *Sour/Fermented* classes was promising. *Nutty/Cocoa* was the most

reported descriptor in all but two coffee samples and can be considered a typical descriptor class of Robusta coffee. The *Floral* descriptor class was only reported seven times. This descriptor class is the most valued in high-quality Arabica coffee (Traore et al., 2018), so its rare occurrence in the studied Robusta genotypes must not be undervalued. Our findings are in line with previous research. Brazilian Robusta cultivars were predominantly described by *Nutty/Cocoa* and *Roasted* descriptors and scored between 70 and 82 points in different environments. *Fruity* descriptors were only found in the highest-quality sample (Teixeira et al., 2020). High-quality Amazonian Robustas were characterized by a higher intensity of *Fruity*, *Sweet*, and *Sour/Fermented* descriptors (Manfrin Artêncio et al., 2023). Dalazen et al. (2020) and Lemos et al. (2020) found *Fruity* and *Sweet* descriptors in their highest quality Robusta clones. Promising Robusta genotypes from Sumatra were described by *Sweet* and *Floral* descriptors (Wicaksono et al., 2022).

The Wild genotypes did not exhibit desired sensory profiles and scored a low Total score. The Lula—Wild hybrid class contained possible ideotypes, i.e., a high Total score and frequency of *Fruity* descriptors combined with *Sweet* and *Sour/Fermented* descriptors. This Wild – cultivated hybrid class had not previously been evaluated, so the results of our first screening are promising. In the Lula class, possible ideotypes were also discovered. The genetic base from the (presumed) seven mother plants was small for the subpopulation of “Lula” cultivars (Capot, 1962). The multiplication of accessions through seedlings and open pollination resulted in the hybridization of the initial accessions into many unique genetic fingerprints (Verleysen et al., 2023). The range in Total score and variation of sensory profiles within this group was surprising. The Lula – subgroup A hybrid class contained multiple ideotypes and the overall highest Total scoring genotype (G0073) with a *Fruity* sensory profile. INERA researchers already knew that the G0073 hybrid (labeled “Petit-Kwilu”) exhibited good agronomic characteristics, but its remarkable sensory profile was unknown. Commercial, wet-fermented Petit-Kwilu from DR Congo was described as a “neutral cup with balanced acidity” (Wilkins, 2019). Previous studies in Robusta field trials reported good sensory quality of hybrids between Conilon (Congolese subgroup A) and Robusta (Congolese subgroup E) botanical varieties, exhibiting fruit-like (*Fruity*) characteristics (Augusto De Souza et al., 2018). Moschetto et al. (1996) demonstrated the effect of genotypes on the sensory quality of Robusta coffee and reported considerable variation in the sensory quality of genotypes from the same genetic origin. The continued screening of the INERA Coffee Collection is advised, as only 70 of the 263 unique genetic identities reported in Verleysen et al. (2023) have been evaluated. Furthermore, the continued introductions of local forest accessions from conservation projects increase the genetic resources at disposal for further evaluation.

The ideal sensory profile ideotype would exhibit a high Total score and high frequency of *Fruity*, *Floral*, *Sweet*, and *Sour/Fermented* descriptors and a complete absence of *Green/Vegetative*, *Other*, and *Roasted* descriptors. This was the case for genotypes G0073, G0079 and G0262. The next step would be to evaluate if the sensory profiles of these ideotypes can be generalized for clonally propagated trees (Montagnon et al., 2012). The heritability of sensory profiles could

then be studied to evaluate if breeding for these ideotypes is possible. It would be interesting to assess if the presence of *Green/Vegetative*, *Other*, and *Roasted* descriptors in genotypes could be suppressed through negative selection for these descriptor classes. The parentage analysis of Verleysen et al. (2023) revealed that genotypes G0002 (83.25), G0003 (82.00), G0006 (83.25) and G0103 (84.25) are commonly used parents in the INERA Coffee Collection, which exhibit a *Fruity* profile. As a starting point for breeding, the direct progeny material of these parent genotypes could be screened for sensory profile ideotypes.

The evaluated Robusta genotypes were cultivated in an unshaded, monoculture system at a low altitude (485 m). Environmental conditions such as growing altitude and shade impact the sensory quality of coffee. Previous research reported a positive impact of higher altitude on the sensory quality of Robusta (Velásquez et al., 2022) and Arabica cultivars (Avelino et al., 2005; Tolessa et al., 2017; Worku et al., 2018; Koutouleas et al., 2023). Previous studies also reported both a positive effect (Vaast et al., 2006; Worku et al., 2018) and a negative effect (Bosselmann et al., 2009; Tolessa et al., 2017) of shade on the sensory quality of Arabica coffee. A study on Robusta cultivars indicated a negative effect of shade on the sensory quality (Vaast et al., 2010). The effect of shade on the sensory quality of coffee, especially Robusta, remains unclear, and more research is needed to understand these environmental effects (Koutouleas et al., 2022). Future research could evaluate whether the sensory profiles of the genotypes, especially the discovered ideotypes, change at higher altitudes or in a shaded system, e.g., an agroforestry system. Furthermore, the Wild and some Lula – Wild genotypes were introduced in the coffee collection from the Yangambi rainforest, a shaded environment. These genotypes, however, were cultivated in an unshaded monoculture system, so an evaluation of their sensory profiles in a shaded environment is advised. Lastly, Ferralsol soils, like those in Yangambi, lack plant nutrients and bind phosphorus fertilizers, which is problematic for crop cultivation (Jones et al., 2013). A shaded coffee system could improve the nutrient availability for plants as previous research reported a higher carbon stock and nutrient concentration in a shaded Robusta coffee system than in an unshaded system (Dossa et al., 2008) and warrants further research.

5 Conclusion

Our study highlights the sensory quality potential of Robusta cultivars from the INERA Coffee Collection in Yangambi, the local wild *C. canephora* diversity, and their hybrids. High sensory quality scores were reported in the cultivated and hybrid materials. Enhancing standard coffee cupping by Q-graders with sensory descriptors revealed substantial variations in the sensory profiles of closely related genotypes. Genotypes with promising and unique sensory profiles were discovered. The evidence from this study also showed consistency in coffee quality and sensory profile over two harvest years for Robusta genotypes. The sensory profile ideotype exhibits *Fruity*, *Sweet*, and *Sour/Fermented* descriptors and is free from *Green/Vegetative*, *Roasted*, and *Other* descriptors. The current study only evaluated a fraction of the diversity of cultivated and wild *C. canephora* genetic resources available in DR Congo, yet already revealed the potential to improve the sensory quality of cultivated Robusta coffee.

Data availability statement

The datasets presented in this study can be found in online repositories. The names of the repository/repositories and accession number(s) can be found in the article/Supplementary material.

Author contributions

RB: Conceptualization, Data curation, Formal analysis, Investigation, Methodology, Project administration, Supervision, Validation, Visualization, Writing – original draft, Writing – review & editing. LV: Conceptualization, Data curation, Formal analysis, Investigation, Methodology, Writing – review & editing. BK: Data curation, Investigation, Supervision, Writing – review & editing. J-LK: Data curation, Investigation, Supervision, Writing – review & editing. TE: Data curation, Investigation, Supervision, Writing – review & editing. TR: Conceptualization, Data curation, Formal analysis, Investigation, Methodology, Writing – review & editing. FV: Conceptualization, Funding acquisition, Project administration, Resources, Supervision, Writing – review & editing. OH: Funding acquisition, Project administration, Supervision, Writing – review & editing. PS: Conceptualization, Data curation, Funding acquisition, Project administration, Resources, Supervision, Writing – review & editing.

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Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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Supplementary material

The Supplementary material for this article can be found online at: <https://www.frontiersin.org/articles/10.3389/fsufs.2024.1382976/full#supplementary-material>

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