CZECH UNIVERSITY OF LIFE SCIENCES PRAGUE

Faculty of Tropical AgriSciences

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Caffeine content of Honduran coffee on the Czech market

BACHELOR'S THESIS

Prague 2023

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Declaration

I hereby declare that I have done this thesis entitled Caffeine content of Honduran coffee on the Czech market independently, all texts in this thesis are original, and all the sources have been quoted and acknowledged by means of complete references and according to Citation rules of the FTA.

In Prague, April 14, 2023

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Acknowledgements

First and foremost, I would love to thank my supervisor Ing. Olga Leuner, Ph.D. for her invaluable guidance, expert leadership, and support throughout this entire journey. I am also deeply grateful to Ing. Petra Škvorová from the Department of Microbiology, Nutrition, and Dietetics at the Faculty of Agrobiology, Food and Natural Resources at CZU, who helped with conducting the HPLC analysis, for her expertise and guidance.

Furthermore, I would like to thank my dear friends and classmates Evgeniia Shershukova and Matyáš Němeček for their invaluable assistance with my sensory analysis. Their hard work and enthusiasm to help were important to the success of this thesis.

Last but not least, I would like to thank myself for the persistence, dedication, and hard work that I put into this thesis. This research has taught me a great deal about the scientific process and has been an incredibly rewarding and fulfilling experience.

Abstract

Many elements, including internal factors such as coffee terroir and post-harvest techniques like roasting and external ones like caffeine content, have a great impact on coffee’s complex flavour. However, the relationship between caffeine content and sensory attributes in coffee is a subject of ongoing research. The main aim of the thesis, which focuses on both specialty and commercial coffee, was to determine the strength of a correlation between caffeine content and sensory characteristics in Honduran coffee, if any, using HPLC, sensory analysis, and Pearson’s correlation coefficient. Results of correlation coefficient analysis revealed that there was only a very weak and negligible indication of a relationship between caffeine content and various coffee attributes. Overall, the findings showed that there was no significant relationship between caffeine content and sensory properties including flavour, aroma, or colour. The study provided evidence that caffeine content does not necessarily correlate with certain sensory attributes, highlighting the complexity of factors that contribute to coffee taste and quality. The thesis should help further research to explore potential mechanisms underlying the relationship between caffeine and coffee sensory properties which could eventually help to acquire knowledge that would help growers and roasters in optimizing their processes and producing coffee with the desired profile.

Key words: *Coffea arabica*, sensory analysis, HPLC, specialty coffee, commercial coffee beans

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List of the abbreviations used in the thesis

C. – Coffea

CO – Carbon dioxide

CQA – Chlorogenic acid

DAD – diode-array detection

DNA – Deoxyribonucleic acid

FTA – Faculty of Tropical AgriSciences

HPLC – High-performance liquid chromatography

LMW – Low molecular weight

masl- meters above sea level

PTFE – Polytetrafluorethen

SHG – Strictly High Grown

spp. – several species

1. Introduction

Every year, people around the world drink an increasing number of cups of coffee. The demand for this product has been rising for ages. With a mean annual growth rate of 1.9 % over the previous 50 years, global coffee consumption reached 9.98 million tons in 2020/21 season (International Coffee Organization 2021). Most importantly, coffee is now related to an entire culture that spans many other spheres rather than just being a plant, a commodity, or a beverage. Coffee drink consumption can be analysed from a social perspective, seeking for symbolism and pragmatic justifications. The coffee culture affects most of the people, regardless of the how the coffee is made, served, where and when it is consumed, or the occasion.

With the growth of cafés and roasters, not just in traditional coffee nations like Italy but also in nations with less evident historical contexts like the Czech Republic, knowledge of various methods of coffee preparation—which go far beyond instant—is also becoming more widespread. The method of coffee preparation depends on the desired taste and strength of the drink. Depending on the difficulty or speed of preparation, it is then divided into different preparation options, including boiling (Turkish coffee), steeping (French Press), dripping (Chemex, Clever dripper), or applying pressure (moka pot, Aeropress, espresso machine). The name is also often prescribed in the name of the coffee drink itself. Generally, they can be split into two fundamental types: dripped through a filter and leached in water (or a combination thereof).

Espresso-based drinks are a unique subset of barista and coffee culture. In cafés, these are more often the drinks of choice (Morris 2013). Frequently, milk or sugar are just the main alternatives people put in coffee. Ristretto, espresso, cappuccino, latte macchiato, and flat white are a few examples of these beverages. The preparation of so-called latte art, or the method of embellishing milky coffee beverages like cappuccinos or flat whites, has been a recent barista trend. This method of adding milk froth to espresso (or double espresso) aims to imprint a pattern on the coffee cream.

Aside from its cultural significance, coffee is frequently sought after for its positive health effects, particularly because it contains substances like caffeine and other compounds that are widely known for their energizing properties. As coffee consumption continues to grow, so does the demand for unique and distinctive flavours. Coffee enthusiasts often choose their coffee based not only on its origin but also on the specific type of beans used. This is because different varieties of coffee beans can contain diverse amounts of chemical compounds and in general produce distinct sensory characteristics, including taste, and aroma. Nevertheless, there is a scanty number of studies on presumable importance of particular components linkage to coffee's flavour profile (Sunarharum et al. 2014). Caffeine content is one of the factors that can influence these sensory qualities, and its impact on the final product is a subject of ongoing research. Therefore, this thesis aims to investigate the correlation between caffeine content and sensory characteristics in various types of coffee samples from Honduras, one of the largest coffee producers in the world, which can contribute to a deeper understanding of coffee chemistry and its resulting profile.

2. Literature Review

2.1. Genus Coffea

A large plant family called Rubiaceae contains more than 6,000 species and 500 genera (Ukers 1922). The *Coffea* genus is the most notable plant in this family of higher dicotyledonous plants. Rubiaceae also includes other genera like *Cinchona* and *Carapichea* which are suppliers of different alkaloids highly valued in medicine and pharmacology (Ferreira Júnior et al. 2012). According to Davis et al. (2006), the *Coffea* genus include more than 100 species, many of which have been grown for generations in the tropical belt. The cotyledons of seeds from plants of the genus *Coffea* are used to make the actual coffee beverage. *Coffea canephora* Pierre ex A. Froehner, or robusta coffees, have a more bitter flavour than *Coffea arabica* and are mostly employed with instant coffees and in commercial espresso blends to encourage the creation of "crema" whereas specialty coffee production primarily uses the seeds of *C. arabica*, or so-called arabica coffees, which account for 60 % of the global market (Crozier et al. 2012; International Coffee Organization 2021).

2.1.1. Physiology

Although they are frequently referred to as fruit bushes, coffee trees can be considered as trees because of their height, which can reach up to 10 meters. In reality, they are frequently trimmed to make harvesting them simpler. Although the plant can survive up to 50 years, its peak fruitfulness only lasts for 30 years. Coffee is a perennial plant, and it normally takes 3–5 years for it to start bearing fruit until it reaches its peak output of 6–8 years. Afterward, depending on the farmer's management methods, plants may live for 20–30 years or longer (Noponen et al. 2017). The first crop of a coffee tree is usually harvested three to four years later, depending on the species.

Root system

A shallow root system or insufficient soil are detrimental to a plant's ability to grow because they make it difficult to retain the water and nutrients (auxins and other phytohormones) that are necessary for growth. According to a study on the root system of coffee plants, deeper rooting and more root dry mass are both related to drought resistance (Pinheiro et al. 2005). However, as the number of roots increases, so does the amount of competition for water and minerals between the roots of nearby plants as well as between the roots of the same plant, which lowers the total intake of these nutrients and stunts plant nutrition and growth (Kramer & Boyer 1995).

Trunk and branches

The coffee bush's trunk, or main plant stem, is orthotropic. It develops buds on its surface that form plagiotropic shoots and eventually branches that are nearly horizontally to the ground as it grows vertically to the soil. Only one branch can be produced by each of these buds. Coffee plants become stumped in the event of unfavourable weather (frost, hail, drought), which encourages the establishment of new orthotropic stems with fresh buds capable of generating additional primary plagiotropic branches.

The development and growth of the entire plant are affected by the general environmental factors, species, and variation of the coffee plant. Within the first year, arabica varieties normally grows 6–10 layers of plagiotropic branches. Two years after planting, the 1.5–2-meter-tall orthotropic stem blooms for the first time. The next year the coffee plant matures and starts to produce fruit (Wintgens 2004).

Figure 1. Coffea arabica

Source: Köhler (1887)

Leaves

As shown in **Figure 1**, the elliptical leaves of the coffee plant are lustrous, thin, clearly veined, and waxy on the surface. They typically sprout on branches facing each other. The largest distinction between *C. arabica* and *C. canephora* is seen in the size and colour of the leaves, with *C. arabica* leaves often being smaller, glossier, and with a dark surface (Ferreira et al. 2019).

The cuticle gives the impression of the leaves waxy surface. It acts as a barrier against water evaporation and shields the leaf from abiotic harm. In reality, just 5 % of the water lost by leaves is thought to escape through the cuticle. Stomatal water diffusion accounts for the majority of water loss from leaves (Yeats & Rose 2013).

Flowering

Coffee beans, seeds from ripe fruits, are how the coffee tree reproduces. It typically takes approximately a month for it to germinate after being placed in light, nutrient-rich, acidic soil; during this time, it must have a layer of parchment covering it. The seeds themselves germinate in three to four weeks, and the plant can reach a height of 15 cm with broad, gleaming green leaves. In a few days all the coffee plants' small white blooms are in the bloom. The green coffee cherry starts to grow after flowering, and it does not turn its ultimate dark red hue until a few weeks before harvest.

The two separate stages of flowering in coffee plants are flower bud initiation and flower opening, otherwise known as anthesis. When serial buds of plagiotropic branches are induced to develop into buds, flower bud initiation takes place (Wintgens 2004). After flowering, pollination happens within 6 hours, followed by fermentation in the next 48 hours (Murthy & Madhava Naidu 2012).

Buds expand by 4–6 mm before going dormant, which in most growing regions occurs during the dry season (Wintgens 2004). 7–10 mm of moisture will then start to appear after the dry time (Eira et al. 2006). Coffee flowering and fertility might be restricted, for instance, by an excessive amount of shadow (Lambot et al. 2017).

Coffee sherry

Different aspects of the hydric, energetic, and geographical components are associated with coffee maturation (De Oliveira Aparecido et al. 2018). Coffee tree fruits mature over a period of 7–12 months, and depending on the plantation's location, they are harvested once a year or perhaps more frequently (Davies Veselá 2018). Coffee cherries are a type of drupe, which is a fruit that can be eaten and has a sweet pulp inside along with a carpel and seeds. Each cherry typically contains two coffee beans, which serve as stores for minerals, lipids, carbs, and proteins for germination.

The exocarp, which is typically the green skin of the coffee cherry that starts to become red after the chlorophyll pigment vanishes, is the first component of the pericarp, or the outer layer of the coffee bean (De Melo Pereira et al. 2019).

The mesocarp, the fleshy layer of the coffee cherry, is known as the middle section, or mucin. The mesocarp's firmness or softness to the touch is a sign of how ripe the fruit is. When pectin chains are broken down by pectinolytic enzymes into a hydrogel that is full of sugars and pectin, softness results. Two green coffee beans are flat sided together and found inside the mesocarp (Avallone et al. 2000).

The endocarp, often known as parchment, is the covering of coffee beans. It shields the seed physically, stops germination, and guards against chemical reactions between the layers. During germination, it stays on the beans but is taken off before roasting (Nabais et al. 2008).

The actual coffee cherry seeds are three-layered and elliptical in shape. Their exterior is covered in silver skin. Although it is unclear which biological components are exactly moved and how this happens, it is believed that they serve to accumulate and transport them from the pericarp to the endosperm. The endosperm is the live tissue that aids in the first growth of plants following germination. The embryo, the third component of the seed, also depends on it because of its nutrients (Heeger et al. 2017).

The coffee plant yields products apart from the beans found inside the coffee cherries. The dried husk of the coffee cherry, known as cascara, is popular for beverage preparation in addition to its traditional function as fertilizer. It is well-liked mostly for its high caffeine level and flavour like rosehip or hibiscus tea (Heeger et al. 2017).

2.1.2. Coffea arabica

Arabica (*Coffea arabica*) is generally considered to be the better and higher quality type of coffee, making up roughly 60 % of the world's production (International Coffee Organization 2021). It is mostly grown in Central and South America, partly also in Africa, its producer is also in Australia. This coffee plant is more demanding to grow, it can have its first harvest after about 5 years. It benefits from a higher altitude between 600–2,500 meters above sea level. As for diseases, Arabica is less resistant to diseases and pests, on the other hand, it is not susceptible to weather changes. The ideal growing temperature is between 15–24 °C. The most common types of Arabica are Typica, Bourbon, or Caturra (Anthony et al. 2001; Davies Veselá 2018; Melese & Kolech 2021).

The most basic type of Arabica developed from different lines of varieties is Typica. It comes from Ethiopia, as substantiated by using DNA-based genetic markers, and also grew on the first coffee plantations in Yemen (Anthony et al. 2001). The tree grows to a height of approximately 4 meters and has a conical shape, straight trunk, and relatively open crown (Tausend et al. 2000). The tree has a relatively low production compared to other Arabica species, but its red fruits are of excellent quality.

La Réunion obtained coffee plants from Yemen. One tree, which significantly expanded during the 17th century, served as the principal source of growth for the plantations. The proliferation of the different kinds that eventually gave rise to the Bourbon variety was substantially aided by this time of seed propagation (Lécolier et al. 2009). Nevertheless, its mutations are grown mainly in Brazil and South America. Although its harvest is still smaller than other species, it produces 30 % more coffee than the aforementioned Typica (Davies Veselá 2018). However, they are similar in quality. The Bourbon variety is susceptible to weather changes, its mostly yellow coffee cherries fall off easily in wind and rain.

Many other types of coffee are mutations of some of the types already mentioned. Caturra is a species bred in Brazil near the city of the same name. It is a mutation of the Bourbon species with high production and quality. As for the taste, it has a strong acidity and a soft body. Other such variations are Mundo Nuovo, Catuai, Maragogype, or Geisha.

2.1.3. Coffea canephora

A cheaper and much lower quality type of coffee is Robusta (*Coffea canephora*). This coffee plant grows at a lower altitude than Arabica, at 200–600 meters above sea level. In contrast, it requires a warmer and more stable climate with a temperature of 24–29 °C. It is distinguished for its disease resistance and easier cultivation. After two to three years, the first coffee cherries are plucked. Because of its shallow root system, it needs more rainfall that is distributed more frequently. Although full sunlight produces the maximum yields, shade has a beneficial effect in highlands because it reduces the temperature swings between day and night. *Albizia* spp., *Gliricidia purpureum*, and other legumes with broad crowns and light foliage are favoured to intercrop for shade (Rehm & Espig 1991).

2.1.4. Differences between the main coffee varieties

The distiction between the two types can be recognized both by the taste and by the look of the beans themselves. The Arabica coffee cherry is elongated and flat, and the groove in the middle is visibly curved. The colour of the fruit is light green, exceptionally with a bluish tone. In contrast, Robusta has a pale green, sometimes greyish bean. It has a straight groove and is more rounded overall. After roasting, you can definitely tell the difference mainly due to their shape (Davies Veselá 2018).

In terms of taste, Robusta is easily recognizable due to its earthier taste and strong body, leaving a bitter taste in the mouth. The chemical structure compared to Arabica is completely different, by the way, it contains up to twice as much caffeine which is reflected in its resulting taste. Vietnam is among the largest producers of this variety (Vo 2022). Robusta is also a coffee that the average consumer buys in a supermarket, in both ground and instant powder form.

2.2. Coffee terroir

The conditions under which agricultural crops are grown as well as their harvesting and subsequent processing are undeniably important factors that influence the overall taste of the final product. For centuries, the importance of the so-called terroir has been demonstrated, for example, by viticulture. Less well-known, but no less important, is the coffee terroir, which, just as with wines or teas, significantly affects the resulting taste of the coffee.

A terroir is a system of complex interactions between a set of operations and techniques practiced by man, agricultural production, and a physical environment to be exploited by a product on which it confers specific original features (Salette 1998). According to Cadot et al. (2012) within the terroir notion, a special significance is placed on the fact where some products are given a distinctive identity that will influence production and have varying effects on their final features. The terroir approach is comprehensive and based on a synthesis of social, technological, and natural aspects that gives rise to the uniqueness of products (Soto Vázquez et al. 2010). In the already mentioned viticulture, the representation of a certain number of factors is essential for the resulting sensory properties of the final drink (Vaudour et al. 2015). The expression "coffee terroir" refers to a broad range of factors that govern the development of the coffee cherry and its transformation into a green coffee bean (Lambot et al. 2017). Coffee's potential for differentiation and segmentation is examined through an examination of the beverage's final quality, particularly in the case of specialty coffees. Physical features as origin, variety, colour, and size, sensory characteristics such as flavour, colour and aroma, and even environmental and social concerns with production systems and agricultural practices are some of the differentiating aspects of specialty coffee (Davies Veselá 2018). In relation to coffee terroir, we are discussing the variables that affect the development of the coffee tree plant, the ripening of the coffee cherries, their harvest, and their post-harvest treatment, including roasting and further preparation. Climate, geography, soil, and tradition are the four primary determinants of coffee terroir.

2.2.1. Climate

More than 50 countries in the intertropical belt grow coffee. The two most crucial water-related variables for the two farmed species are the annual rainfall and the length of the dry season, both of which have a big impact on the yield. The most crucial climate conditions for good coffee cultivation are precipitation and temperature. High and consistent atmospheric humidity is ideal for both species (Smith 1985).

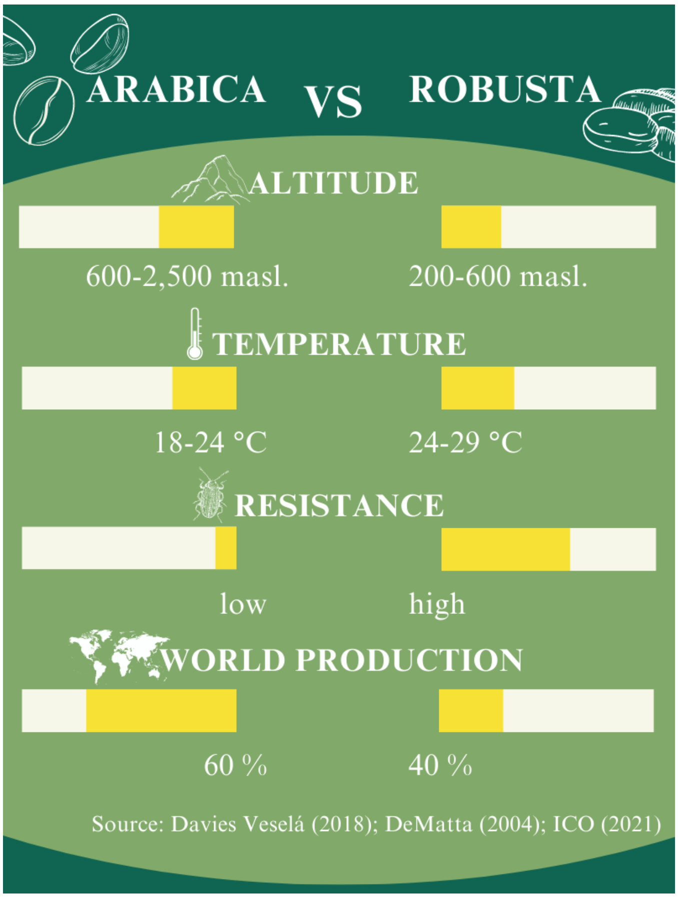


Figure 2. Coffee subspecies comparison regarding coffee terroir

Figure 2 above highlights important variations in temperature, altitude, resistance, and global output between Arabica and Robusta coffee beans. Overall, it is obvious that these two coffee varieties are very different from one another. Robusta coffee beans favour warmer regions, while Arabica coffee beans typically flourish in cooler ones. As a result, these two varieties of coffee are often grown in very distinct regions. While Robusta thrives in lower-lying areas where it is warmer and more humid, Arabica is frequently found at higher altitudes where it is colder. Moreover, Arabica coffee is better able to withstand intraseasonal variation and lower temperatures than Robusta coffee, despite Robusta having a reputation for being more resilient due to its ability to withstand higher temperatures. On the other hand, it is not as resistant to pests. Robusta has a low susceptibility to frost. Therefore, it grows best at lower elevations; its leaves and fruits cannot endure temperatures below 5–6 °C or even continuous exposure to temperatures below 15 °C (DaMatta & Cochicho Ramalho 2006). Natural climatic oscillations have a significant impact on Arabica productivity because it is closely correlated with climatic variability (De Camargo 1985).

Weather patterns around the world are significantly changing as a result of climate change. The provision of natural resources and ecosystem services, on which we all rely, will be impacted by changes in how ecosystems function and the loss of biodiversity. Many locations in the equatorial belt, where coffee is grown, are already enduring greater temperatures, protracted droughts, or bursts of strong rains. These factors can have a variety of effects on coffee production, including increasing the spread and prevalence of pests and diseases that harm coffee plants, interfering with pollination, or reducing the uptake of essential nutrients, which will reduce crop yields and quality. Plants can sustain direct harm from heat stress in the form of denaturation and aggregation of proteins as well as an increase in the fluidity of membrane lipids. Indirect damage brought on by heat stress includes the inactivation of mitochondrial and chloroplast enzymes, the suppression of biosynthesis, an increase in protein breakdown, a loss of membrane integrity, and alterations to the plant cell wall (DaMatta & Cochicho Ramalho 2006).

Recent climate change research claim that extended exposure to temperatures of up to 30 °C stresses plants, which manifests as slowed growth and abnormalities such as the formation of stem tumours and yellowing leaves. The disruption of cellular homeostasis caused by these kinds of damage eventually results in hunger, decreased ion flow, the creation of hazardous chemicals and reactive oxygen species, and the limitation of plant growth (Wahid et al. 2007). Growth is also slowed down in areas where the annual average temperature is below 17– 18 °C. Even intermittent frosts can drastically reduce the crop’s ability to provide income (DaMatta & Cochicho Ramalho 2006).

2.2.2. Terrain (Altitude)

One element that influences a coffee’s overall flavour character is altitude. The best Arabica coffees in the world are grown in mountainous areas of the Coffee Belt (Goodinid et al. 2019). The principal coffee-growing regions in the globe include Central and South America, southern Asia, and several Pacific islands, and mid to southern Africa.

All coffee plants are produced in tropical climates, but the altitude at which they are grown has a big impact on the flavour of the final beans. The coffee tree thrives best at elevations, where there is little risk of frost, moderate annual rainfall of around 2,032 mm, and enough of sunlight. Coffee trees grow more slowly in cooler highland climates, which delays bean development. Due to the prolonged maturing period, the coffee bean is infused with more complex sugars, resulting in richer and more enticing aromas. Higher elevations with better drainage also have less water in the fruit, which further concentrates the flavours (Bardin et al. 2010).

Although coffee species can be found from sea level to 2,500 m above, the majority of species (67 %) are only able to survive in an altitude range of less than 1,000 m. Some species, such as *C. canephora*, *C. liberica*, *C. salvatrix*, or *C. brevipes*, have a broad distribution in elevated areas, ranging from lowlands up to even 800 masl, others are mostly restricted to narrow ranges of variation (e.g., *C. heterocalix, C. kapakata*, or *C. stenophylla*). Additionally, *C. arabica* thrives even in altitudes exceeding 2,000 masl. On the other hand, the majority of the species with a widespread distribution on the continent of Africa (such as *Canephora canephora*, *Eugenioides canephora*, and *Liberica canephora*) are typically found in wet habitats typified by evergreen or gallery forest. Other species, however, such as *Coffea congensis*, *Coffea racemosa*, or *Coffea pseudozanguebariae*, show specific adaptations to environments with particular soil and climate conditions (Davis et al. 2006).

Coffees produced at high altitudes tend to have good bean quality (Decazy et al. 2003) and are related to superior sensory experience scores for the coffee beverage (Smith 2018) because they mature during longer maturation periods and are shaded by cooler air temperatures during the ripening phase. The sensory score is significantly affected by even slight temperature variations of 2.5 °C, affecting the coffee’s quality grade and indicating the potential impact of microclimates on terroir (Da Silva et al. 2005). It is widely acknowledged that greater altitudes are related to better-quality beans and cups.

Although the best coffees in the world are grown at elevations of at least 1,200 meters, there are a few exceptional cases. For instance, the renowned Hawaiian Kona is so far north of the equator that coffee cannot be cultivated beyond 600 meters in that area (Powers 1932).

Contrarily, high-altitude specialty coffees typically fetch a much higher market price because of their great flavour and vibrancy, lesser yield per coffee tree, and difficulty in production and marketing for coffee farmers in isolated mountainous regions. High-altitude coffee growing provides truly exceptional coffees that are among the most reasonably priced luxury goods available (Avelino et al. 2005).

According to reports, terroir connected to high altitudes (>1,000 m) and little precipitation (1,600 mm/year) produced coffee that was more favoured since it was aromatic, slightly bitter, acidic, and had a body. Contrarily, coffee cultivated at lower elevations (about 850 m) and with high levels of precipitation (>2,110 mm/year) was less liked because of its enhanced astringency, grassy flavour, more pronounced bitterness, and low aroma. The higher altitude and a lower average temperature are thought to lengthen the maturity period of the coffee cherry, resulting in a nutrient-dense coffee bean that contributes more compounds and flavour to the finished beverage (Decazy et al. 2003).

According to DaMatta et al. (2007), coffee berries ripes more slowly at higher elevations and cooler air temperatures or when shaded, which results in a superior bean-filling process (Vaast et al. 2006). Compared to coffee produced at lower altitudes, the beans grown under these conditions are denser and have a far more potent flavour. One of the main factors contributing to good cup quality appears to be a longer maturation phase, possibly by a more comprehensive biochemical mechanism needed for the development of beverage quality (Da Silva et al. 2005).

The future of coffee plants will once again depend on expanding the site of coffee plantations because climate change will still have an impact on this situation. By 2050, changes in temperature and precipitation may cause Central America's area under coffee cultivation to increase the required elevation for coffee production from around 800 m–1,400 meters above sea level upwards to 1,200–1,600 meters above sea level (Baca et al. 2014).

Kenyan coffee crops may experience significant delays due to the increase in minimum elevation from 1,000–1,400 meters above sea level across the Atlantic. Kenya's eastern and (particularly) western highlands could lose adequate arable land (Davis et al. 2012).

2.2.3. Soil

Environmental protection is significantly aided by soil, and healthy plant growth is largely influenced by its condition. Not just with coffee, it is a valuable asset to the preservation of natural ecosystems.

Coffee thrives in soils with specific physical and chemical characteristics. The most crucial of the physical qualities is a soil structure that allows for adequate drainage because water logging drastically affects yield and, if it persists for a long enough period of time, can even destroy coffee trees.

As coffee plants were historically moved around the tropics, they landed on a variety of soil types. Coffee was frequently cultivated in agroforest systems, which retain shade trees, or, increasingly, in open, deforested areas, where the soil and plants are exposed to equatorial sun, monsoon rains, trade winds, and other natural occurrences. Because of this, the risk of erosion is considerably increased when coffee is cultivated on steep hillsides.

Higher yields are attained without shade when coffee is grown under ideal soil and climate conditions, with high standards of cultural practices and enough inputs (DaMatta et al. 2007). It is advised to utilize shade to maintain regular yields and prevent overbearing if soil conditions are less than ideal, rainfall is heavy, temperatures are excessively high or low, and maybe where there is a lengthy season of many hours of bright sunlight.

As per Clemente et al. (2015), coffee quality is a product of the chemical components of coffee and the effects of enzymes on some of these components, which result in compounds that have an impact on coffee quality. These soil-related factors affect coffee quality. Since it influences the quality of the coffee and the crop's overall output, proper coffee nutrition is necessary (Melke & Ittana 2014).

Different plants have different nutrient needs and different capacities for absorbing nutrients from the soil (Martins et al. 2015). Typically, 17 essential nutrients are needed by plants for optimum growth and development (Fageria 2009). Due to their particular metabolic roles in plants, these nutrients are crucial (Hopkins & Hüner 2009).

One of the pillars of the 2030 Agenda for Sustainable Development endorsed by United Nations Member States is the sustainable use of terrestrial ecosystems. That makes soil degradation one of society's biggest problems, if not threats, in the twenty-first century (IAASTD 2009).

There is abundant knowledge about what soil types are most suitable for coffee and how to amend and maintain soil for crop productivity and quality. In general, heavy clay soils are not suitable for coffee cultivation as they have poor soil drainage. In such soil, root penetration and growth, in general, are very difficult.

Other two characteristics necessary for coffee production include water capacity and depth. Since it makes enough water available throughout the dry season, water capacity helps to maintain evapotranspiration. By providing a bigger volume of soil that holds more water and nutrients surrounding the coffee trees, deep soil promotes the growth of roots. Deep soils are essential, particularly, in regions with extended dry seasons and little rainfall.

Deep soil allows root proliferation by offering a larger volume of soil which contains more water and nutrients around the coffee trees. Especially in places where there is a long dry season coupled with lower rainfall, deep soils are necessary. It has been suggested that 3 meters depth of soil is ideal. Bleeker (2017) cites coffees successfully produced in Papua New Guinea with high rainfall, a short dry season, regular cloud cover on clay soils just 15–20 cm deep over hard clay that is not pierced by coffee roots, and on steeper slopes, confirms that this is not the general rule. Crop yields can be significantly decreased under these circumstances in years with considerable rainfall or an extremely protracted dry season.

The pH and the number of nutrients available to the plants are the two most crucial soil characteristics for the growth and output of coffee. According to several publications, coffee is produced on soils that range in pH from very acidic ($pH < 4.0$) to somewhat alkaline (pH up to 8.0). But neither of these extremes is suitable for high-output economic production. A slightly acid soil is preferred. Arabica coffee is known to grow best on deep, slightly acidic, fertile, well-drained loams that are of lateritic or volcanic origin and have a reasonable amount of humus content (Sadeghian et al. 2008).

Given the vast occurrence of wild plants in diverse altitudes and habitats, it may be expected that Robusta coffee has a wider range of adaptability and less specialized requirements than Arabica coffee. Robusta coffee is grown on red sandy, clay, or gravely loams (oxisols, ultisols) (Lambot et al. 2017). The primary habitats for robusta variety growth are flat to gently sloping, well-drained, acidic soils with minimal natural fertility. Additionally, a variety of soil types, including peat, clay, and less fertile soils, can be used to grow *C. liberica* coffee. Compared to the other two species, it can withstand more abuse (Teketay 2003).

Although it might not seem like it from a nutritional standpoint, the tropical regions where the majority of coffee trees are cultivated have very loose soil. Because nutrients are necessary for the vegetative growth of coffee trees as well as the production of high-quality beans, nutrient imbalances can have an impact on the quality of the coffee (Lambot et al. 2017). The two main nutrients needed for coffee are potassium and nitrogen, with nitrogen being more crucial for vegetative growth and potassium for fruit development. Phosphorus is necessary for the growth of crop plants' roots, flower buds, and fruits, as well as for the storage and transfer of energy (Fageria 2009). Although the amounts needed are typically tiny to negligible in coffee, calcium, magnesium, and other major micronutrients are crucial for the coffee plant's balanced nutrition (Yadessa et al. 2019). When critical nutrients are not provided, plants have recognizable deficiency symptoms (Nagao et al. 1986).

While insects and bacteria quickly decompose materials in the tropics, preventing the development of rich topsoil, temperate soils build up nitrogen and carbon that persist in the soil as organic matter. Unfortunately, economic growth is hampered, and poverty is made worse in the tropics by poorer agricultural output and greater disease rates (Masters & Mcmillan 2001).

To keep soil's nutrients intact, either fertilizers or agroforestry systems can be used. Trees offer a variety of environmental services. For instance, some trees fix nitrogen in the soil, so lowering the demand for fertilizer. Since nitrogen is the most limited element in coffee production, trees that can produce more than 100 kg of nitrogen annually play a crucial ecological and biochemical role in coffee cultivation (Binkley et al. 1994).

2.2.4. Tradition

Tradition, the final component of coffee terroir, is equally significant to the other elements. Each plantation uses different cultivation, gathering, and processing techniques for coffee cherries. Because of the many flavour profiles this results in, each coffee manufacturer still has its individuality. The quality of the coffee is influenced by the attention given by the growers, the roasters, and the baristas who make it. According to published data, preharvesting factors define 40 % of the physical, chemical, and sensory characteristics, while postharvest processing methods determine 60 % of the final taste (Musebe et al. 2007). Perhaps as a result of this, it is possible to assert that the traditional component of coffee terroir—important for both flavour and price—is very likely the most crucial.

In the lengthy process of growing and processing coffee, which takes more than a year before they produce a cup of coffee, the aforementioned elements—altitude, soil, and climate—that affect the growth of the coffee plant, and the sensory taste that results are only the first stage (Davies Veselá 2018). Individual methods vary, as it is grown as a commodity or specialty coffee on small or large plantations, by businesses or individual farmers.

Harvesting

The geographic location of the plantation affects coffee harvesting. In regions near the equator, where the climate is more consistent, coffee can be collected all year long; however, in other locations, the best time to harvest the ripe fruits of the coffee tree vary considerably. At lower altitudes, where the harvest starts, fruits ripen more quickly. However, their quality, and eventually their price, which tends to be lower than that of the rest of the harvest, reflects their quicker ripening time. The fact that the entire process takes 6–8 weeks is another justification for harvesting in lower areas.

Traditions, the type of coffee, commodity or specialty, the size of the plantations, and their accessibility at higher altitudes are only a few of the variables that influence current harvesting practices. Methods are generally categorized as “stripping” and “selected picking”.

Stripping, or simply strip picking, is a harsh method for gathering coffee cherries. When stripping, the coffee tree branch's fruits are all plucked simultaneously into baskets, starting from the bush's trunk and working outward to the edge. The harm done to the coffee plant itself counterbalances the method's advantage in terms of speed. Damage to the bush's limbs and the assortment of green, immature fruits result from the method's lack of prudence. The blending of green and overripe cherries with ripe ones can lower the quality of the coffee (Poltronieri & Rossi 2016). The ability to use this strategy manually or automatically is an undeniable advantage. An appropriate environment is needed for harvesting with machines. Here, it is influenced by elements like topography, inclination, spacing, alignment, or the height of the plant (Haile & Hee Kang 2020). The device, which has a harvester-like appearance, drives over coffee plants or through rows of planted bushes, removing all the cherries and leaves. Mechanical collection, despite being reckless, is becoming more and more common in nations like Brazil and Australia, mostly because of how quickly and cost-effectively it is handled.

Unquestionably, high-quality coffee beans matter. The coffee cherry reaches its greatest quality when certain characteristics and chemical processes are present. Only a few days a year, when they are mature and prepared for harvest, they reach these attributes. The best way to identify such beans is by hand-picking (Sanz-Uribe et al. 2017). As opposed to strip picking, this approach involves choosing the coffee cherries by hand, one at a time. This method is gentler on the coffee plant and only harvests fruits that are fully ripe. Manual collecting requires a lot of time and resources in terms of logistics. For plantations situated on steep terrain or high in the mountains, it is frequently the only option available. Given this, coffee harvested in this manner is likewise more expensive, but of course also of a higher quality. Specialty coffee is typically hand-picked, yet, plucking only ripe cherries and receiving an incentive for quantity rather than quality can result in such coffee not necessarily being of the highest quality (Haile & Hee Kang 2020).

Processing

Farmers have a few hours to process the cherries they have collected. Processing has a significant impact on both the flavour and the cost of coffee. Although the result of the three fundamental processing techniques—dry, wet, and honey—variates in difficulty, the removal of the skin from the coffee cherry is always the outcome (Hameed et al. 2018). The chosen drying method matters from the perspective of the sensory qualities that are produced since it impacts the chemical processes in the coffee cherry. According to Nayak et al. (2017), the amount of LMW present in green coffee beans was greatly impacted by the drying process.

Dry processing method

Dried, freshly picked cherries are called natural, dry-processed, or unwashed. The climate is the primary factor in this method's popularity; historically, it has been and continues to be frequently applied in regions with inadequate water supplies and in regions with little annual precipitation. Because of this, it is seen as a more widely used, easier, and more economical method. The cherries are dried using the drying method, which involves spreading them out in a thin layer on concrete patios or African beds after they have been picked. African beds are popular because they allow for ventilation from all sides. They are composed of bamboo and stretched canvas. The major objective is to provide equitable access to sunlight and air for all cherries (Davies Veselá 2018).

These factors cause the coffee cherries to gradually dry out; the entire process can take up to a month. Mechanical dryers are another option for additional drying; however, they risk damaging the bean during rapid drying. Controlling the drying temperature is essential when using mechanical drying; it should not be higher than 45 °C (Davies Veselá 2018). In a study by Silva et al. (2008) bacterial populations were predominant in the coffee cherries as they represented 96.3 % of the total isolated microorganisms (bacteria, filamentous fungi, and yeasts). To avoid the growth of mould and undesired fermentation, the coffee must be tossed frequently. Because the mucilage, which is extremely hygroscopic and remains with the coffee sherry, there is a considerable possibility of subsequent fermentation with natural (Gratuito et al. 2008; Lee et al. 2015). The fruits are covered at night to keep moisture away from them.

Regardless of variety of terroir, dry processing alone imparts some flavour characteristics to the coffee. Frequently favourable, but if not dried properly, they can be damaging, completely destroying the lot (part of the harvest). Because chemicals from the skin and pulp penetrate the bean, beans typically have a stronger and more prominent body. Because it may have fermented or even alcoholic notes, kvass, or even manure, if the coffee is not properly dried, it is not very well-liked on the global market. On the other hand, the dry process always imparts fruity aromas and a creamy texture to the cup when drying is done properly (Poltronieri & Rossi 2016).

Wet processing method

A fully washed or so-called wet-processed processing method frequently yields coffee of higher quality. In special tanks, coffee cherries are first washed with water while unripe or otherwise substandard fruits that float to the surface are separated. The upper skin and a portion of the pulp are removed during this processing step after washing in a water bath and selection. This step must be completed within 24 hours of harvesting; if not, it would be more challenging to remove because the skins have a tendency to dry up along with the bean's pulp. When cherries are processed using the washed method, the skin, and pulp are removed by striking the cherry quickly against metal discs with projections or spikes. This process leaves a green bean with pulp.

The beans' fermentation occurs in the next phase. According to Stanbury et al. (1995), fermentation is any process that involves mass-culturing microorganisms to produce a product. Tanks are used for the fermentation of hulled beans. The entire process can take up to 36 hours and is influenced by the surrounding temperature, the number of microorganisms in the tank, and the pulp and parchment residue. The beans may rot if the fermentation process lasts too long. Enzymes break down the remaining sticky layer that covers both the parchment seed skin and the parchment itself during fermentation. Hereafter, the sticky characteristics are eliminated, and the beans develop a rough surface. Another alternative is to ferment the fruit pulp directly in its own juice, i.e., without the addition of water. Panama, among other places, uses this method. The final freshness, fruitiness, and acidity of the coffee are impacted by fermentation and washing.

Like the dry processing approach, the green beans handled in this way are next spread out on concrete floors or African beds because they still contain an unfavourable amount of moisture—roughly 50 %. They are also turned over at this stage and covered as necessary during the night or in direct sunshine, just like naturals. The use of manual machinery is widespread, but there is once again a risk that the quality will decrease.

However, not all plantations can afford to use the wet technique of processing due to ecological and financial limitations. During the washing, subsequent cleaning of the tanks, and water exchange, about 75 litres of water are used for every kilogram of green coffee (Davies Veselá 2018).

Honey method

Each plantation uses the honey method in a unique way, and the variations can be seen in the amount of water utilized during processing. A technique called "pulped-natural" is similar to the "wet method," but it uses less water overall, making it more popular with farmers in places with limited water supplies. The sorted fruits from the water bath are peeled in machines to remove the skin and the majority of the pulp. They are then cleaned with water and dried in the sun on parchment as in the dry technique (Sanz-Uribe et al. 2017).

In the context of honey-processed coffee, the coffee can be classified according to the total amount of surface pulp that is still present on the bean, which can be identified by its colour. The cherry is immediately peeled, very little water is used, and the beans are dried with parchment, primarily with the pulp on top. The final beverage has a different flavour depending on whether it contains a large, medium, or little amount of pulp known as black honey, red honey, or yellow honey respectivelly. Overall, it can be argued that coffee that has undergone this processing is sweeter tasting, more prominent, and free of sour overtones.

The so-called reposo phase, which is a 1–2 month interval after the beans have been processed, is when the green coffee beans rest. Before being exported, the dried peel or parchment is removed in so-called mills, the beans are separated by size and quality using special sieves, any flaws are eliminated, and then the various batches that will be transported to the roasters or the stock market are tasted (Davies Veselá 2018).

Roasting

The roastery is where green beans of different coffee varieties from around the world, which are kept in jute bags, go through the greatest change, and develop all of their tastes. The cup score of a good coffee should be high, and it should deliver the outstanding flavour that coffee enthusiasts expect. Coffee taste (cup quality) is significantly correlated with the quality of the green beans, which includes a number of elements collectively referred to as coffee terroir, including the roasting method (Wei & Tanokura 2015). Flament (2001) notes that the application of heat during bean roasting sparked the Maillard reactions, which result in the production of a special chemical composition that gives coffee its distinct flavour characteristics. Varied combinations of volatile and non-volatile compounds produced by the same type of coffee can generate distinctly different coffee flavours, tastes, and aromas (Ferreira et al. 2019).

The temperature at which coffee beans are roasted is high and gradually rises, usually between 200–250 °C (Rehm & Espig 1991). We divide samples into three categories based on the degree of roasting: light city roast, city plus roast (also known as medium roast), and full city roast (Davies Veselá 2018). 70 % of coffee beans are made of cellulose, which also includes water, proteins, carbohydrates, sugars, oils, acids, and aromatic compounds, including caffeine and trigonelline. Although coffee beans of various species, origins, roasting levels, or analytical procedures have varying compositions, the degradation of polysaccharides, oligosaccharides (particularly sucrose), chlorogenic acids, and trigonelline is frequently seen while roasting (Vignoli et al. 2011; Kreuml et al. 2013). Flament's study is also supported by other research from 2015 proving that considerably fewer green coffee beans components such as sucrose, free amino acids, chlorogenic acids, and trigonelline, as well as the breakdown of polysaccharides and proteins, are thought to be the primary cause of the roasted coffee bean's distinctive scent, flavour, taste, and colour (Wei & Tanokura 2015).

The most prevalent simple carbohydrate in green coffee beans, sucrose, functions as an aroma precursor during roasting to produce a variety of classes of chemicals that have an impact on the flavour of the coffee. These compounds include carboxylic acids, furans, and aldehydes. It has been observed that sucrose degrades quickly during the initial stages of roasting (Wei & Tanokura 2015).

At least 30 chlorogenic acids, including caffeoylquinic acids, dicaffeoylquinic acids, feruloylquinic acids, and p-coumaroylquinic acids, have been found to be present in coffee beans (Clifford et al. 2003). Wei & Tanokura (2015) reveals that the amount of CQAs significantly drops after roasting, whilst the levels of quinic acid, and syllo-quinic acid increased. From this we can deduce that one of the measures of the degree of roasting can be the decomposition of chlorogenic acids.

A pyridine derivative known as trigonelline is known to indirectly help produce products with appealing flavours. Trigonelline's significance as a precursor of flavour and aroma molecules as well as a favourable nutritional factor has been extensively studied in the past. Trigonelline increases continually during roasting. There is no question that trigonelline have both direct and indirect effects on other physicochemical characteristics of a cup of coffee, including flavour and aroma (Wei & Tanokura 2015). Green coffee beans have significant quantities of trigonelline, which decrease as the beans are roasted.

The process of roasting results in significant physical modifications as well as the creation of compounds that give the beverage its unique sensory characteristics. Green coffee beans naturally lack the roasted coffee's distinctive colour and aroma, which are both created during the roasting process. Most of the coffee aroma is carried by coffee oil, which makes up roughly 10 % of the roasted beans (Buffo & Cardelli-Freire 2004). The processes by which coffee aroma is created are incredibly complex. A diverse blend of volatile chemicals makes up the aroma. All the pathways involved in the Maillard reaction, caramelization, Strecker degradation, and the breakdown of sulfur amino acids, hydroxy-amino acids, proline and hydroxyproline, trigonelline, quinic acid moiety, carotenoids, and minor lipids clearly have a wide range of interactions. Less than 20 of the approximately 900 volatile chemicals have been shown to be relevant to the aroma of coffee (Buffo & Cardelli-Freire 2004).

The moisture content of each batch of green beans varies, affecting how they should be roasted. The major shift in roasting occurs when moisture evaporates, going from an initial 11–12 % to 2 % at the end of the procedure (Davies Veselá 2018). The majority of the water evaporates during the initial roasting process (which then turns into drying), and then the so-called yellowing, or gradual conversion of green beans to yellowish ones, happens. Browning, which causes beans to turn light brown, occurs at the end.

The Maillard reaction is a chemical reaction that occurs when proteins, peptides, and other amino acids with free amino groups are combined with reducing carbohydrates (Wei & Tanokura 2015). During the thermal processing of foods, it plays a key role in the formation of distinctive scents and tastes. Amino acids and sugars react with heat and so the Maillard reaction, the process of simultaneously browning beans and caramelizing sugars, begins. The remaining water is taken out, which causes the bean to create carbon dioxide, which builds up inside of it. The so-called first crack is caused by joint pressurization with steam, which causes the bean's cell structure to break.

The most significant non-aroma volatile ingredient in roasted coffee, in terms of quantity, is carbon dioxide produced by pyrolysis and the Strecker degradation reaction. For whole-bean coffee, the degree of roast has no impact on emissions (Lebouf & Aldridge 2018). The type of coffee and the roasting methods affect how much carbon dioxide is created during roasting (Anderson et al. 2003). Although some degrees can go up to 10 ml/g of coffee, Anderson et al. (2003) summarize that most roasting degrees are evaluated between 2 and 5 ml/g of coffee.

A profile, or the best curve, of the coffee, is created during the roaster's initial coffee tasting. Finding the appropriate sequence of temperature, time, and other criteria is required for this (drum rotation, air volume). When the green beans are put in a roaster that is hotter than 200 °C, the complete roasting process takes around 10–15 minutes. First-crack, which happens in the ninth minute of roasting, is a sign that chemical reactions have begun in the coffee. The remaining roasting time is determined by the desired sensory outcome. Coffees with a fine, light roast that is 11–12 minutes long are more acidic and fruitier.

The coffee begins to take on sweeter tones during the 13^{th} minute of roasting, and the acidity on the tongue begins to decrease. These are the characteristics of medium-roasted coffee. The flavour turns increasingly bitter as the roasting time increases. This type of roasting coffee can take up to 15 minutes and is known as dark or intensely roasted coffee. When coffee beans are over roasted, a second crack occurs. Such roasting produces a highly bitter, almost burnt flavour that is particularly popular, for instance, in Italy. Nevertheless, roasting times can be as short as 90 seconds to make high-yield coffees or as long as 40 minutes, as it is frequently the case in Brazil. Roasting time affects the reactions occurring inside the bean: longer roasting times result in a bitter coffee missing a pleasing aroma, but very short roasting times may not be long enough to complete all pyrolytic reactions, producing coffee with underdeveloped organoleptic characteristics (Buffo & Cardelli-Freire 2004; Davies Veselá 2018).

The coffee beans are stored in barrels, plastic boxes, or, in the case of larger roasters, silos after being chilled in special coolers to stop any chemical reactions. During this process, a number of gases are released from the beans.

The key determinants of hydrolysis processes in roasted coffee are moisture and temperature because hydrolytic enzymes are thermally inactivated during coffee roasting. High moisture content in food storage systems minimizes food's interaction with oxygen, which tends to inhibit oxidation reactions while promoting hydrolysis ones (Toci et al. 2013). When these changes were correlated with storage time, the environment and temperature both had an impact. The free fatty acid loss was slowed down by using an inert atmosphere and low temperature (Toci et al. 2013).

LeBouf's recent study found that the carbon monoxide emissions rates from roasted coffee showed that unventilated or insufficiently ventilated storage rooms should be watched and vented as necessary to maintain CO concentrations at safe levels (Lebouf & Aldridge 2018). After storing roasted coffee beans for nine months, the conducted sensory evaluation found that the quality of coffee beverages was negatively impacted. The intensity of characteristics that point to oxidation processes increased after 18 months. As a result, we can draw the conclusion that sensory alterations to roasted and vacuum-packed coffee beans are probably going to occur during storage (Kreuml et al. 2013).

The coffee industry uses and recommends inert atmosphere, vacuum packaging, and low storage temperatures to limit degradation processes in coffees and increase shelf life (Makri et al. 2011). Nevertheless, these measures do not stop chemical reactions during storage; they simply postpone the effects (Toci et al. 2013).

2.2.5. Honduran coffee terroir

Honduras is the top producer of coffee in Central America and ranks 6th globally in terms of production (International Coffee Organization 2021). Though it has not always been the case, its terroir is recognised for creating unusual and exceptional coffees. Before speciality coffee became increasingly popular, Honduras farmed coffee primarily for the commercial market, where quantity is more important than quality.

The majority of Honduran coffee plantations are situated at high altitudes (up to 1,800 metres above sea level), which supports the production of the Arabica variety. Due mostly to its tropical environment, Honduras is also unique in terms of the composition of its soil and its rugged relief. With access to both the Pacific and Atlantic oceans as well as having volcanoes on its ground, Honduras has an extremely diversified terrain. Overall, the distinctive flavour and quality of the coffee from this Central American state are impacted by the high altitude, mineral-rich volcanic soil, fluctuations in rainfall patterns influenced by the oceans, and other factors (Ohashi 1989; Guerra 2017).

2.3. Chemical components related to coffee sensory characteristics

The main chemical components in coffee beans surely contribute to both the beverage's sensory qualities and its impact on the human body. From the most eluted ones like chlorogenic acid or caffeine to polyphenols, minerals, or vitamins. Their volume generally causes a favourable or negative final taste of the coffee drink.

2.3.1. Caffeine

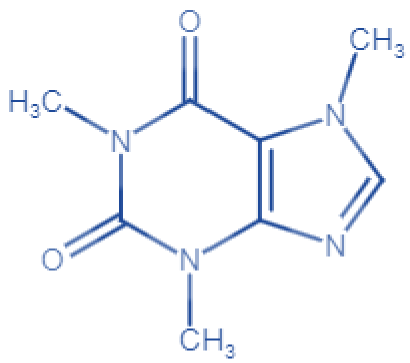


Figure 3. Genetic molecular structure of caffeine

Caffeine is currently consumed by 80 % of the world's population on a daily basis (Ogawa & Ueki 2007), primarily for its stimulating effects, making it the most extensively used psychoactive stimulant globally (Smit & Rogers 2002). Coffee beans naturally contain caffeine, also known as 1,3,7-trimethyl xanthine, a purine alkaloid (see **Figure 3** above) (Aguilar-Navarro et al. 2019). Caffeine appears to exert the majority of its biological effects at ingestion levels associated with coffee consumption via adenosine receptor subtypes. Its administration has a number of physiological consequences, including stimulation of the central nervous system, an abrupt increase in blood pressure, an increase in metabolic rate, and diuresis (Chou & Benowitz 1994). Coffee is not the only naturally occurring caffeine source, despite being the most well-known. This alkaloid is also found in more than 60 other plants, such as yerba mate, guarana, and the Chinese tea tree (Guest et al. 2021).

At room temperature, caffeine is colourless, odourless, and bitter. It dissolves easily in boiling water, and the addition of acids and the creation of complexes like benzoate, citrate, and salicylate at high temperatures both boost its solubility (Arnaud 2005). Caffeine is a non-ionized chemical in an aqueous solution at physiological pH. The melting point is between 234 and 239 °C (Rameshaiah & Ashoka 2015).

The primary function of caffeine in coffee seeds and plants appears to be defence. The so-called chemical defense theory, which contends that caffeine in young leaves, fruits, and flower buds' functions to shield soft tissues against plant-pathogenic bacteria (such as *Pectobacterium atrosepticum*), is supported by several studies, including one from Sledz et al. (2015).

There has not been any discernible variation in caffeine values between seeds produced using various post-harvest techniques (wet, dry, and honey). Regarding the level of maturity, a study by Koshiro found that during the growth of *C. arabica* fruits, there were minor fluctuations in the amount of caffeine in the seeds (between 2 and 10 %). Significant differences between seeds from different maturation stages have been seen after fruit growth. Mature and ripening pericarp and seeds showed little to no biosynthetic activity (Koshiro et al. 2006).

Although entering the body as a xenobiotic chemical, caffeine is swiftly transported throughout most tissues and bodily fluids (such as saliva) after absorption (Velička et al. 2021). The distribution pattern can be substantially higher in women than in males, and it typically does not vary over the course of a person's lifetime. Yet, although this amount was reduced with weight loss, severely obese people showed a greater volume of dispersion (Abernethy et al. 1985). The thermogenic impact and resting metabolic output are both enhanced by caffeine. It is an ergogenic resource because the brain mechanisms that cause a series of physiological reactions cause it to have a favourable impact on exercise capacity and endurance (Guest et al. 2021). It was a 1 % rise in resting metabolic production in Davoodi et al. (2014) study. Following caffeine consumption, Belza et al. (2006) note a 90 Kj increase in resting metabolism.

Caffeine is the most extensively ingested psychoactive substance in the world. The physiological effects of caffeine consumption include diuresis, an abrupt rise in metabolic rate, and acute blood pressure elevation. Current research demonstrates that, at least in healthy adults, caffeine consumption (2–3 cups or 300 mg daily) is not linked to negative consequences such as cardiovascular stimulatory effects and behavioural abnormalities. Although generally safe, caffeine has some risks. In fact, caffeine quickly reaches a similar dosage in the mother and foetus after crossing the human placenta. Excessive caffeine use has been linked to spontaneous abortion and stunted foetal growth. It is recommended that pregnant women and those who are nursing consume no more than 300 mg of caffeine each day. Children can be considered to be another risk category since they exhibit altered behaviour, such as restlessness or anxiety (Tuomilehto 2013). Caffeine intake for children and adolescents is deemed to have a maximum limit of 3 mg/kg body weight per day by European Food Safety Authority (EFSA 2015).

According to a Lang et al. (2013) study, excessive caffeine use may cause the metabolism to become saturated. Caffeine is one of the substances responsible for many of these advantages, and several epidemiological studies have linked moderate coffee drinking with a lower relative risk of developing chronic degenerative diseases and death. These include hepatoprotective properties and a decreased risk of Parkinson’s and Alzheimer’s diseases. Antioxidant and anti-inflammatory processes are among the mechanisms (Kolahdouzan & Hamadeh 2017).

The amount of caffeine in coffee is directly proportional to both its sensory and stimulating effects. Coffee drinker’s final caffeine intake is influenced by all of the beans’ constituents, such as the blend’s composition, which is largely genetic in nature, or the level of maturation (Koshiro et al. 2006). By using the most popular hot brewing techniques, caffeine is effectively extracted. The dosage ratio, water temperature, particle size, and brewing time are all controlling variables (Davies Veselá 2018). The method used to prepare the coffee, the grind size, and the amount of steeping time will all have an impact on the caffeine content. The amount of caffeine in coffee increases with the length of the steeping process. Although it is common knowledge that espresso-based coffee is richer in caffeine, some studies, despite this belief, claim otherwise stating that filtered coffee is higher in caffeine content (Caporaso et al. 2014; Davies Veselá 2018; Bobková et al. 2021).

2.3.2. Other components affecting the sensory characteristics

Acidity, bitterness, sweetness, and sourness are the four prominent taste descriptors that can be used to categorize substances that have an impact on the sensory qualities of coffee drinks (Sunarharum et al. 2014). Many volatile and non-volatile chemicals can be discussed from this perspective. In addition to chlorogenic acid lactones, bitterness also refers to phenolic acid breakdown, which has an impact on astringency (Variyar et al. 2003). Polysaccharides, particularly glucose and sucrose, affect the overall sweetness, and furanone gives coffee a caramel flavour (Oestreich-Janzen 2010; Bressanello et al. 2018). The molecules of guaiacol, phenolic substances that are particularly appealing in malt whiskey, which already imparts a smoky touch, are responsible for the flavours that determine unpleasant tastes, such as the burnt taste of coffee in the mouth (Seninde & Chambers 2020).

2.4. Specialty and commercial coffee

Coffee is divided into two types - specialty and commercial. That is, the one that can only be purchased in businesses focusing on high-quality coffee and the one that can be purchased in supermarkets.

Specialty coffee must meet a number of criteria in order to be referred as such. Selected coffee must pass evaluations of quality during coffee tastings, as specific requirements are set by which the beans are assessed, with the SCA cupping form being the most well-known. Coffee that passes this test is always given at least 80 out of 100 possible scores. In general, we can say that specialty coffee comes from higher altitudes where the climate has a good impact on the coffee’s flavour profile. Of course, specialty coffee also depends on post-harvest processing, such as roasting and coffee preparation (Specialty Coffee Association 2021).

Contrarily, it is frequently impossible to identify the country of origin of commercial coffee. Whatever its flavour profile, so-called commodity coffee is farmed only for financial gain, as the name suggests. With this coffee, quantity always wins out over quality. The robusta variety is most frequently cultivated on large plantations, and the simplest and least expensive post-harvest processing techniques are most frequently used. Given that it is impossible to predict how long after harvest a coffee crop will be sold, this typically has a negative impact on the flavour and quality of commercial coffee. The most popular form of this coffee that is sold in supermarkets is instant coffee. It is also available in the form of beans or ground coffee. However, the consumption of commodity coffee worldwide exceeds specialty one (Davies Veselá 2018).

2.5. An overview on caffeine and coffee profile correlation studies

Researchers are still being conducted to determine the relationships between the chemical components of coffee and its flavour or other sensory attributes. Although there has been a lot of research on this subject, the conclusions are unclear, making it impossible to state with certainty whether or not and to what extend there is a connection between caffeine and the flavour profile of coffee at this time.

The coffee taste profile and chemical components, including bioactive chemicals like caffeine, were analysed by De Oliviera Fassio et al. (2016). The study used trained panellists to assess the basic sensory characteristics of selected coffee samples, the caffeine concentration was evaluated using HPLC and the correlation matrix was used to express the relationship between the variables. The majority of the study's correlations showed a poor or non-existent relationship with every sensory attribute, hence the hypothesis linking coffee profile and caffeine level was disproved. In Bertrand et al. (2006) study on coffee from Central America, drink quality and chemical composition were also assessed using sensory analysis and HPLC analysis. The study's findings demonstrated some effects of coffee terroir but did not demonstrate a direct link between the two. Avelino et al. (2005) also claim that there is no likelihood for a positive relationship between sensory analysis evaluations and chemical composition in their investigation of specialty coffees from Costa Rica, another Central American nation. The correlation matrix for the study on Coffee quality and its interactions with environmental factors shows a negative correlation as well (Barbosa et al. 2012).

Contrarily, there are studies demonstrating the opposite; a study by Gimase et al. (2014) confirmed a positive link between all sensory qualities and chemical components. In studies on Arabica coffees, caffeine has been linked to agreeable cup quality (Farah & Donangelo 2006). Non-volatile substances, including caffeine, have a significant impact on how consumers perceive flavours and how much they like and appreciate coffee (Sunarharum et al. 2014).

Most of the studies do not focus on detailed caffeine analysis, trying to cover as many variables possible, and thus do not provide a deeper insight and greater contribution to this issue. The reason for continuing the research is the significant variation in the data that are currently available. Additional study can help to better comprehend the subject, clarify the existing material, and provide fresh perspectives.

3. Aims of the Thesis

The aim of this thesis was to determine the effect of caffeine content on the sensory characteristics of coffee (*Coffea* sp.) from Honduras.

The specific objectives of the thesis were:

- Investigate the caffeine content of five different coffee bean samples from Honduras.
- Conduct a sensory analysis to evaluate among others the flavour, aroma, and colour of the coffee samples.
- Determine if there is a correlation between the sensory properties of the coffee and its caffeine content.
- Compare and contrast the caffeine content and sensory attributes of specialty coffee and commercial coffee in order to identify any significant differences between them.

Research question: To what extent, if ever, is there a correlation between caffeine content and sensory characteristics in different types of coffee samples from Honduras?

4. Methods

4.1. Sampling

The coffee samples used in this study were carefully chosen to represent various processing techniques and geographical locations. Five samples total were chosen, comprising one commercial coffee and four specialty coffees. The specialty coffees were sourced from three different regions in Honduras, namely Santa Barbara, Copán, and La Paz, while the commercial coffee's origin was unknown. The samples were obtained from roasters and coffee shops in the Czech Republic, ensuring their freshness and quality.

Before preparation, the coffee samples were stored in their original packaging at room temperature (up to 20 °C) to prevent exposure to light and air, which could potentially impact aroma and rancidity. Samples were handled with care to avoid contamination or degradation prior to sensory and HPLC analysis. Prior to the analysis, the samples were equilibrated at room temperature for at least 24 hours.

Each sample was given a special code to guarantee an impartial examination. These codes were used to blind the panellists and the analysts from knowing the specific brand or origin of the coffee samples. Lidl Deluxe, First Crack Coffee, Dos Mundos, Kafé Křižka and Amáres Coffee all had codes of 147, 264, 389, 514, and 903, respectively. This coding system was established prior to the analysis and was used for recording in the sensory analysis questionnaire. Such classification technique ensured a fair and impartial study of the samples by removing any biases that might have resulted from prior assumptions or knowledge of the coffee brands.

Samples used in this thesis:

Honduras Soleado- Deluxe, Lidl

The only coffee that is commercial rather than specialty. The retail organization Lidl is the seller. Although Honduras is mentioned, no specific region is named on the packaging. The beans were roasted in Germany using the traditional drum roasting method, although the level of roasting is not described in more detail. Yet, the beans appear to be roasted on a dark level at first glance, see **Figure 4**. The coffee is 100 % Arabica and has overtones of chocolate.

Commercial sample 147 has an unknown origin, which means that it can be traced back to a specific farm or region. Without knowing the specific origin of the sample, it is difficult to provide detailed information about the unique combination of factors that contribute to its flavour profile.

Figure 4. Coffee beans sample 147

Honduras Caballero – JR #1, First Crack Coffee

An anaerobically washed coffee beans of the Catuai variety from the Caballero family farm. The farm is in the Chinacla region, at an elevation of 1,600 masl. The coffee carries aroma of peaches and dried orange, the roaster observes medium acidity and a cupping score of 87.5. The coffee was roasted to a light degree of roasting, as the roastery's name implies (see **Figure 5**).

The Chinacla region's Sample 264 was cultivated in the perfect altitude for the cultivation of coffee. The coffee plants flourish in this area's special climate, which has considerable humidity and rainfall. Because of the strong nutrient content of the local soil, the coffee has a unique flavour profile. The area is renowned for its conventional coffee-growing techniques as well, which have been handed down through generations of coffee farmers. The distinctive flavour and quality of the coffee are enhanced by these techniques.

Figure 5. Coffee beans sample 264

Honduras San Rafael, Dos Mundos

Coffee from the Prague roaster Dos Mundos of the Parainema and Lempira variety (100 % Arabica). The achieved cupping score is 86.5 points. It originates from San Rafael farm owned by Norma Iris Fiallos Calidonio based in the area of Los Limos, Corquin, Copán. The plantation is situated at an altitude of 1,300 meters above sea level. It has notes of star anise, chocolate mousse, pear jam. It undergoes natural, i.e., dry, processing before being light to medium light roasted (see **Figure 6**). A filter is the suggested preparatory technique.

In comparison to the other samples, sample 389 from the Copán region was grown at a lower elevation. Nevertheless, the special microclimate and rich soil of the area make it the perfect place for coffee to flourish. Moreover, the area is renowned for its rich volcanic soil, which adds to the distinctive flavour character of the coffee. Most of the coffee in this area is grown in shadow, which shields the plants from the harsh sun and gives the coffee a softer flavour.

Figure 6. Coffee beans sample 389

Honduras Capucas, Kafé Křižka

Coffee beans produced and processed in the Honduran San Pedro, Copán region. It comes from the Cooperativa Cafetalera farm, which is situated between 1,350 and 1,600 meters above sea level in the northwester Honduras. The beans (shown in **Figure 7**) underwent a wet processing method and are therefore so-called fully washed. It has flavours reminiscent of ripe fruit, caramel, and chocolate. The obtained SHG badge indicates that the taste should be denser, fuller, and sweeter. These coffee beans are roasted to a level of espresso city to city roast, which corresponds to a medium roast. The roaster states that the beans may be used to make filtered coffee as well as espresso.

Sample 514 was grown under optimal conditions in the area renowned for its microclimates, which give the coffee a variety of flavours. The distinctive flavour profile of the coffee is influenced by the region's varied wet and dry seasons. Also rich in nutrients is the local soil, which gives the coffee a flavourful complexity.

Figure 7. Coffee beans sample 514

Honduras Capucas, Amáres Coffee

Coffee of the Catuaí variety, it is 100 % Arabica (*Coffea arabica*). It originates in the Santa Barbara region, one of Honduran main coffee-producing areas. It is grown at an altitude between 1,400 and 1,800 meters above sea level. One can distinguish caramel and nuts, with hints of tobacco and almonds, and it has a mild acidity. Wet processing and medium roasting were applied to the coffee beans (see **Figure 8**). Due to its medium body, it can be used for both filter and espresso-based coffee.

Sample 903 from the Santa Barbara region, region with a unique climate, moderate rainfall, and dry winters, which provides ideal growing conditions for coffee. The soil in the region is also rich in nutrients, which contributes to the unique flavour profile of the coffee. Additionally, the region is known for its traditional coffee-growing methods, which include the use of organic fertilizers and hand-picking of the coffee cherries. These methods contribute to the distinct taste and quality of the coffee.

Figure 8. Coffee beans sample 903

The geographic distribution of the coffee bean samples utilised in the study is depicted on a map bellow (see **Figure 9**). The locations of the plantations where the coffee beans were cultivated are indicated by the spots on the map, showcasing the variety in the samples' places of origin. Two of the samples are from the Santa Barbara region, and the remaining samples are from the La Paz and Copán regions. The map gives a visual depiction of the vast geographic area the samples came from.

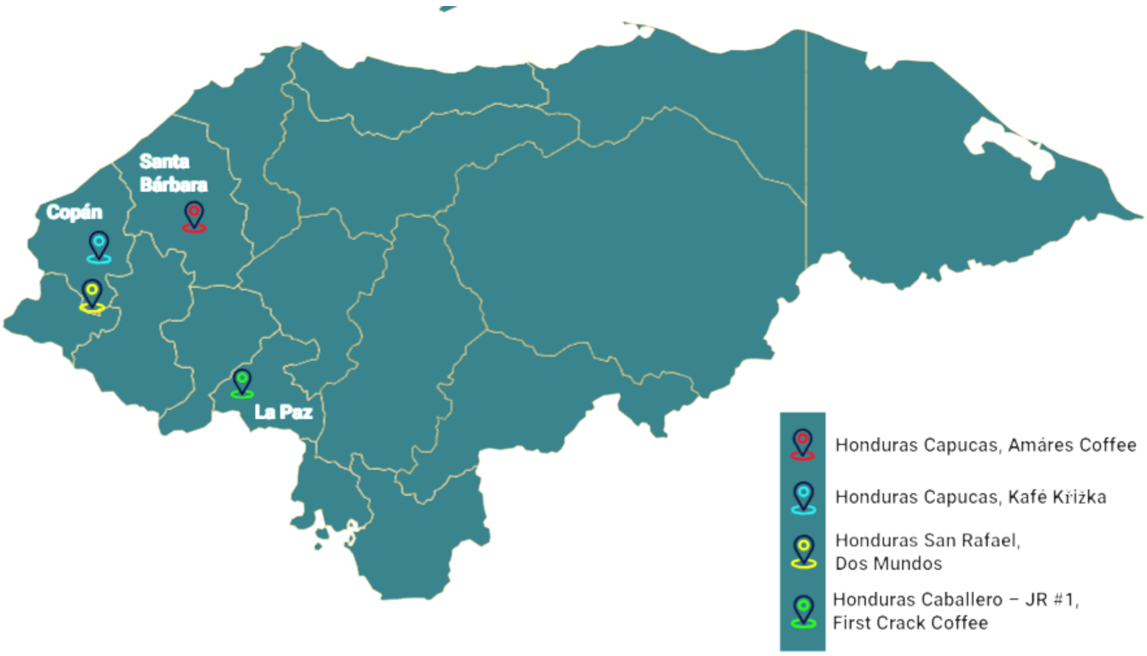


Figure 9. Map of samples bean origin Honduras

All the materials for this study, which investigates coffee beans available on the Czech market, were obtained from regional roasters in Prague and Poděbrady, except for the Deluxe brand coffee, which was bought in Chlumec nad Cidlinou and is distributed by the Lidl company.

4.2. HPLC Analysis

In the fields of chemistry and biochemistry, high-performance liquid chromatography (HPLC) is a commonly used analytical technique that allows for the separation, identification, and quantification of individual components in mixtures. In HPLC, the sample is divided into discrete components by their differential partitioning between a stationary phase and a mobile phase, which is based on the chromatographic separation principle. By engaging in specific interactions with the constituents of the sample, the stationary phase—often a solid or liquid packed in a column—provides the separation mechanism. The sample components are transported through the stationary phase by the mobile phase, which is typically a liquid that flows through the column. Individual components elute at various intervals as a result of the stationary phase's variable retention of the sample's constituents, which produces the separation (Lough & Wainer 1996).

As comparison to other analytical methods, HPLC provides several benefits, including high sensitivity, selectivity, and reproducibility. It enables the investigation of a variety of substances, including proteins, peptides, tiny molecules, and nucleic acids. HPLC also offers the capacity to quantify and distinguish several versions of a chemical, such as isomers and chiral compounds. The ability to separate and quantify distinct components is crucial for reliable analysis in complex mixtures such as those present in biological samples, environmental samples, and food products, which are frequently the subject of HPLC studies (Riley 1996).

4.2.1. Determination of caffeine content using HPLC-DAD

The technical background of the HPLC-DAD caffeine measuring is described subsequently in **Table 1** below:

Table 1. HPLC measuring background

HPLC-DAD measuring background	**Chemicals used for measuring:** Demineralized water – purifying using Milli-Q Plus (Millipore, Germany) Methanol for HPLC (Lach-Ner, Czech Republic) Caffeine (Sigma-Aldrich, Germany)
	Apparatus: Analytical scales (0.1 mg accuracy) Mettler AE 200 (Mettler Toledo, Switzerland) Ultrasonic bath (Elma, Germany) Infinity 1260 II. HPLC system (Agilent, USA): Wide-range DAD detector 1260 Infinity II. (Agilent, USA) Automatic Vialsampler 1290 Infinity II. Vortex SA 7 (Stuart, United Kingdom) Syringe with PTFE membrane filter (0.45 µm) DELL computer with an OpenLab software
	Analysis conditions: Column: Infinity Lab Poroshell 120, 2.7 µm C 18, size 150 x 3 mm (Agilent, USA) Mobile phase: methanol:demineralized water (ratio 40:60) – isocratic elution Detection: DAD at 264 nm Mobile phase flow rate: 0.25 ml/min Column temperature: 35 °C Length of analysis: 7 minutes Analyte retention time: 4.5 minutes

Five extracts were created from five samples of Honduran coffee beans. These extracts were then each introduced one at a time into a high-performance liquid chromatography (HPLC) machine. As shown in **Figure 10**, for the HPLC analysis, 5 grams of each coffee sample were used, which were diluted 10 times by adding 1,350 microliters of tap water and 150 microliters of prepared filtered coffee to a vial with a volume of 1.5 millilitres, using an adjustable laboratory pipette and special reloads.

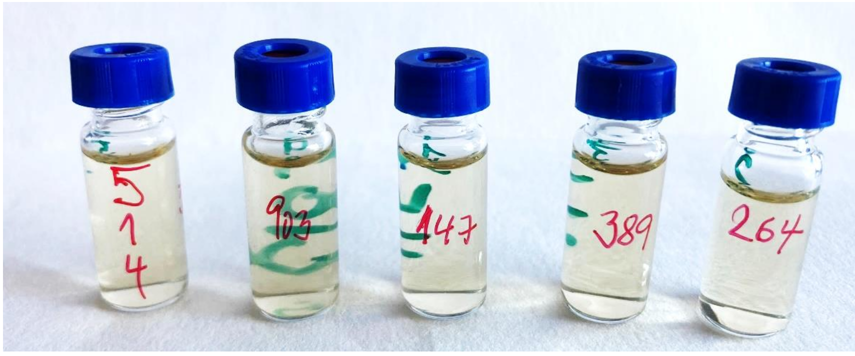
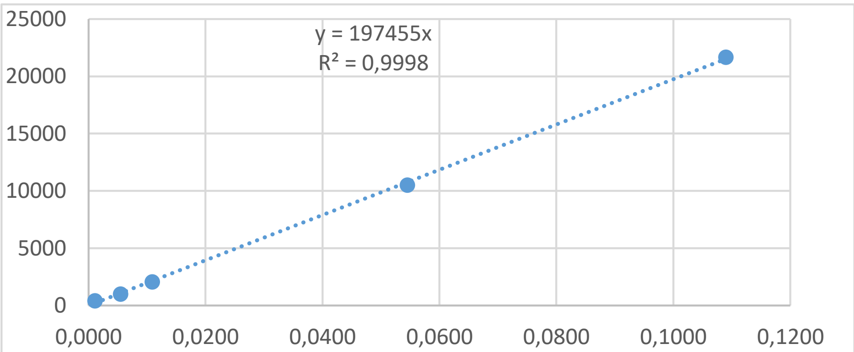


Figure 10. Vials prepared for HPLC analysis

Base solution was prepared dissolving 10 mg of caffeine in the mobile phase (100 ml) creating a concentration of 100 µg/ml. A calibration set was prepared from the base solution with caffeine concentrations of 1; 5; 10; 50; 100 µg/ml. Volumes of base solution (0.25; 1.25; 2.5;12.5 ml) were pipetted into 25 ml volumetric flasks and filled until the graduation marking with the mobile phase. The linear calibration curve was created using Microsoft Excel programme, refer to **Table 2**.

Table 2. Calibration curve for the samples analysed



4.3. Sensory analysis

A scientific method called sensory analysis is used to quantify and examine how people react to stimuli including taste, aroma, texture, and appearance. It is an essential tool for food research and development since it enables scientists to assess and quantify the sensory qualities of food products in an unbiased manner (Civille et al. 2016).

Many techniques, such as descriptive analysis, hedonic testing, and discrimination testing, can be used to undertake sensory analysis (Heymann & Lawless 2010; Rogers 2010). The sensory characteristics of a product are described by trained panellists using a common vocabulary in descriptive analysis. Consumers' subjective preferences for a product are gauged by hedonic testing, whilst the ability to distinguish between two or more products is assessed by discrimination testing (Heymann & Lawless 2010).

Semi-trained panellists are people who have received some training to recognize and explain particular sensory characteristics in a product, but they might not be as knowledgeable as fully trained panellists. To aid in their training, taste buds can be calibrated to assist them better discern particular flavours and scents. This approach is frequently utilized in circumstances when it may not be practical to assemble and completely train a panel of experts, such as in consumer testing or research with a larger participant pool (Heymann & Lawless 2010).

The inclusion of semi-trained panellists can help to enhance its diversity, which can result in a more accurate assessment of a products' sensory attributes (Ares & Varela 2017). Nonetheless, it is crucial to make sure that the panellists' assessments are consistent and that the outcomes are accurate and repeatable. Although semi-trained panellists' consistency and accuracy in rating sensory qualities can be improved with training, it is crucial to remember that their ratings may still be subject to some degree of variability as panellists may not be able to detect sensory defects (Mörlein 2012).

Sensory analysis can be used to assess the body, flavour, aroma, and overall quality of coffee samples in the context of coffee research. This knowledge can be used to distinguish the distinctive sensory qualities of coffee from various growing locations, roasting levels, and processing techniques. In order to better understand the connection between the chemical makeup and the sensory qualities of coffee, sensory analysis is frequently completed with chemical analysis, such as HPLC.

4.3.1. Sensory analysis of coffee samples from Honduras

Sensory analysis is a crucial aspect of evaluating food and beverages, including coffee. This study used a sensory analysis to investigate the relationship between the caffeine and sensory properties of coffee beans. The sensory analysis was conducted at the sensory laboratory of the Faculty of Tropical Agriculture, Czech University of Life Sciences in Prague, from March 14th to March 16th, 2023. A total of 31 panellists took part in the analysis, all of whom filled out the questionnaire correctly and therefore all 31 questionnaires were processed for this thesis (refer to **Table 3** below). The age group categories were selected based on differences in taste perception with age, as demonstrated by several studies (Weiffenbach et al. 1986; Kennedy et al. 2010; Krishnaa & Jayaraj 2017). The number of around 30 participants was a desired sufficient number for several surrounding factors such as the time-consuming sensory analysis or the number of samples to be evaluated. The age groups were chosen based on commonly used categories in researchers on sensory evaluation.

Table 3. Sensory analysis participants data

Number of participants: 31

Age

- 0-19 years: 0
- 20-39 years: 25
- 40-59 years: 6
- 60 and more years: 0

Sex

- female: 20
- male: 11

Prior to conducting the sensory analysis, the panellists, who were faculty members and students, undertook a sensory calibration procedure to guarantee that their assessments of the coffee samples were reliable and consistent. The calibration process involved the use of several reference solutions to train the panellists' sensory buds to identify and distinguish between different tastes and aromas. For sweetness, panellists were exposed to two reference solutions of varying sugar concentrations (a concentrate of 30 g of sugar in 1.5 l of water, and a concentrate of 150 g of sugar in 1.5 l of water) while two citric acid solutions of varying concentrations were used to calibrate for sourness (0.75 g in 1.5 l and 3 g in 1.5 l). A reference solution containing 1 g of caffeine in 1 l was used to calibrate for bitterness. Additionally, panellists were exposed to reference aromas of caramel, burnt peanuts, and a 1:1 concentrate of strawberry and kiwi syrup to calibrate their sense of smell for sweet, burnt, and fruity aromas, respectively. The panellists were also given a detailed explanation of the various sensory perceptions in words. Fresh tap water was also provided to neutralize between calibrations.

Based on prior research and industry norms, the amount of each sample utilized for analysis was calculated to be 46.5 grammes to make 31 cups of coffee, with 25 ml of coffee in each cup, for the sensory analysis. Using the French press method, 300 grammes of 92–96 °C water was combined with 18 grammes of coarsely ground coffee. The coffee samples were prepared using tap water available at the university campus. During the sensory evaluation, each participant got one cup of coffee per sample, a glass of water, and rohlík, type of bread roll, whose crispy texture and mild taste helps to neutralize the palate (see **Figure 11**). Prior to the examination, the roasted beans were kept in a cool, dry environment to preserve their quality and freshness.

To minimize potential bias, the samples were randomized and presented in a randomized order to the panellists during the sensory analysis. The respondents were instructed to cleanse their palate between samples with water and rohlík. The panellists were also provided with a scoring sheet to rate the coffee samples on various sensory attributes, including aroma, flavour, colour, overall palatability, and aftertaste.

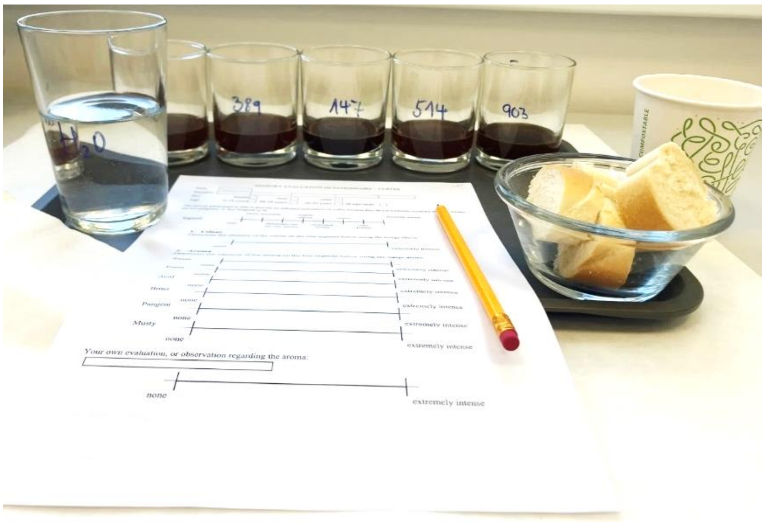


Figure 11. Sensory analysis plate

Questionnaire

The sensory analysis questionnaire that was utilised in this study had sections for colour, aroma, flavour, overall palatability, and aftertaste (see **Appendix 2**). The questionnaire started off with basic information on the participant's sex and age group, followed by a statement stating that the participant was able to provide an unbiased assessment of coffee due to specific limits, including not regularly taking alcohol, not smoking, being pregnant, or feeling fatigued or ill.

To help the participant assess each sensory attribute, the questionnaire had a line segment and a legend. The remaining categories, including aroma, flavour, overall palatability, and aftertaste, were organised in the same way, with subsections of the following: sweet, fruity, acid, bitter, pungent, and musty (with three-line segments for aftertaste evaluation at different time intervals and just a single line segment for overall palatability).

As for the evaluation, participants were instructed to mark the line segment at the point that best represented the intensity of the attribute for each sample, and to write the sample number on the line segment. The sensory analysis questionnaire used in this study included a section on overall palatability, which was evaluated on an 8-point scale ranging from 1 (non) to 8 (extremely intense), see **Figure 12**.

It is important to note that in this case, the scale was used primarily as a measure of hedonic evaluation, rather than intensity-related factors such as bitterness or acidity. Following the data collection, the ratings were assessed based on the numbers that corresponded to each description on the scale. As a result, means and standard deviations for each attribute across all samples could be calculated. The scale was a visual guide for participants and a helpful tool for quantifying the results that was included in the questionnaire.

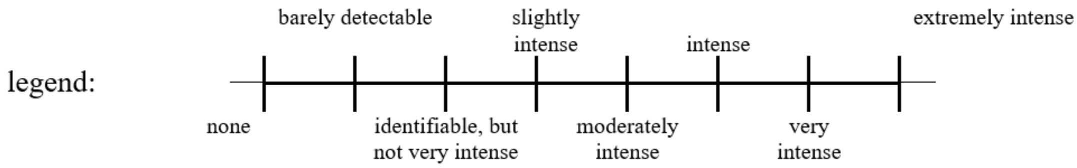


Figure 12. Sensory analysis questionnaire legend

An optional comment section on the questionnaire allowed participants to offer extra feedback on the flavour, aroma, and general acceptability of the coffee samples.

4.4. Data analysis

The initial stage after gathering the data from the sensory study was to calculate the arithmetic mean scores and standard deviations for each characteristic (colour, aroma, flavour, overall palatability, and aftertaste) for each sample. The average of the score obtained by each participant for each attribute was used to determine the final scores of each sample in the sensory analysis.

Pearson's correlation coefficient was utilised to determine the correlation between caffeine content and sensory characteristics calculating the correlation for the entire data set. Pearson's correlation coefficient is a statistical measure that determines the strength and direction of the relationship between two variables. The coefficient is a value between -1 and 1, where -1 represents a perfect negative correlation, 0 represents no correlation, and 1 represents a perfect positive correlation (Trahorsch et al. 2019). For its calculation, the sensory analysis scores, i.e., the average of all scores from 31 questionnaires, for each sensory attribute and the result of high-performance liquid chromatography was used. Pearson's correlation coefficient was calculated by the JASP software (0.17.1. version).

4.5. Ethics

Ethical considerations were considered throughout the entire study. Prior to the start of the study, all participants were informed of the purpose of the research, the nature of the sensory analysis, and what is expected of them as participants. They were also informed that their participation was voluntary and that they could withdraw from the study at any time. All participants began participating in the study only after being informed.

To protect the privacy and confidentiality of the participants, all data collected during the study were collected anonymously. Only members of the research team had access to the data (sex, age group), and there was not any identifying information. Overall, the study was conducted with the utmost respect for the rights and welfare of the participants, and all ethical considerations were considered.

It was brought to attention during the study that it might not have been ethical to include people under the age of 18 in the questionnaire. The questionnaire was corrected right away to make sure that future participants would be aware that only adults are permitted to participate and to remedy the problem. The previously completed questionnaires was also checked, and none had been completed by anyone in the first age group 0–19 years. Although the author regrets this lapse, an action was taken quickly and appropriately to make sure the study was carried out morally and in conformity with all relevant regulations.

5. Results

For each coffee sample that was evaluated, an adequate number of samples were gathered through the French press brewing method. Duplicate samples were obtained from the same batch of coffee as the originals, thanks to the selected preparation method that ensures a large volume of coffee in one batch, which avoids the samples showing differences. Each obtained sample underwent two analyses in order to eradicate mistakes from the HPLC analysis. Using linear trend estimation discovered by caffeine standard preparation, the peak area data was converted into milligrams concentrations per 100 ml. Calculated arithmetic mean values are shown in **Table 4** below. Sample 264 had the greatest caffeine concentration (42.34±0.0060 mg/100 ml), while sample 514 had the lowest caffeine concentration (36.65±0.3462 mg/100 ml).

Table 4. Caffeine content in examined samples

Sample code	Caffeine concentration: mg/100 ml (Mean values ± SD)
147	41.49 ± 0.0795
264	42.34 ± 0.0060
389	40.75 ± 1.0219
514	36.65 ± 0.3462
903	36.78 ± 0.1452

In addition to the HPLC analysis, sensory evaluation was conducted on each of the coffee samples using a panel of semi- trained tasters. The samples were evaluated based on aroma, flavour, overall palatability, colour, and aftertaste using an 8-point scoring system.

Table 5. Sensory analysis evaluation- the colour of examined samples

COLOUR EVALUATION:

Colour	**Mean score**
147	6.45±0.88
264	3.16±1.16
389	4.65±1.14
514	4.74±1.44
903	4.29±1.24

Based on the findings of the colour evaluation (see **Table 5**), the sample with the highest caffeine concentration (42.34 mg/100 ml) received the lowest colour score. In contrast, the sample with the highest colour score was produced using the same preparation technique with the second-highest caffeine content (41.49 mg/100 ml). The standard deviation numbers for each sample are very high, indicating that there is some variance in how the panellists perceive colour.

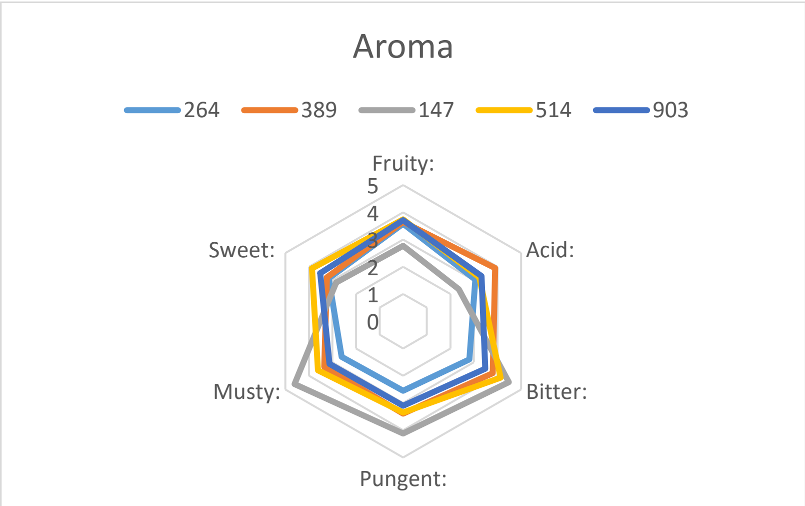


Figure 13. Sensory analysis evaluation- the aroma of examined samples

As **Figure 13** indicates, sample 147 had the highest mean score for the bitter, pungent and musty subcategories but the lowest mean score for the sweet, fruity, and acid subcategories. Sample 389 had the highest mean score for the acid subcategory, while sample 514 had the highest mean score for the sweet and fruity subcategories. It is noteworthy that sample 514 obtained high scores for the sweet and fruity aroma subcategories while having a lower caffeine amount, even though there may not be a direct association between caffeine content and aroma evaluation. Similarly, sample 903 scored high in most of the aroma subcategories despite having less caffeine. Given the information, there is a chance that sample 147's high caffeine content may have contributed to the evaluation of its strong bitter aroma (for the complete data set, see **Appendix 1**).

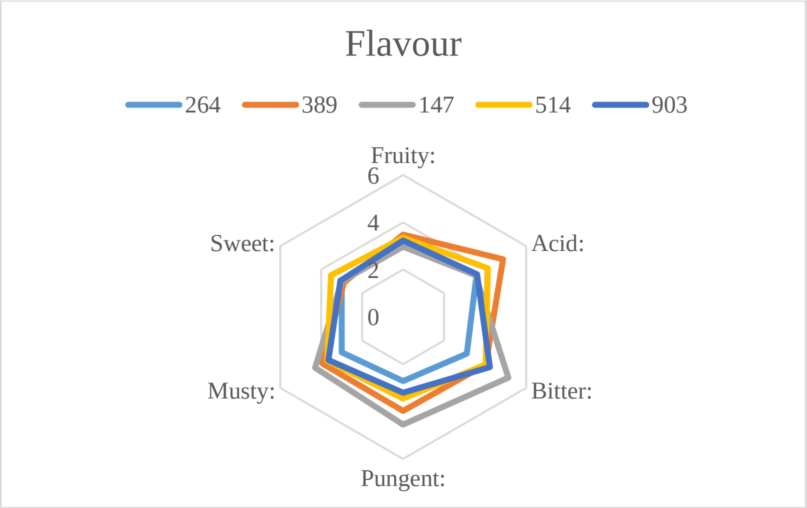


Figure 14. Sensory analysis evaluation- the flavour of examined samples

For sample 147, there may be a correlation between caffeine quantity and flavour ratings in the bitter and pungent categories, but more statistical research is needed to prove a meaningful connection (see **Figure 14**). High acidity was observed with sample 389 (for the complete data set, see **Appendix 1**).

Table 6. Sensory analysis evaluation- the overall palatability of examined samples

OVERALL PALATABILITY EVALUATION:

Overall palatability	**Mean score**
147	4.81±1.82
264	3.71±1.66
389	4.61±1.43
514	4.42±1.67
903	4.65±1.31

The **Table 6** shows the scores of overall palatability of the coffee samples, with sample 147 having the highest score of 4.81±1.82, followed by samples 903, 389, 514 and 264. This suggests that commodity coffee sample 147 was the one that the tasters preferred the most. Nevertheless, scores between the samples varied just slightly and might not be statistically significant. It is questionable if sample 147, which had the second-highest caffeine concentration, earned a good overall palatability score purely because of the caffeine content which could contribute to its complex profile or other factors like other element of coffee terroir.

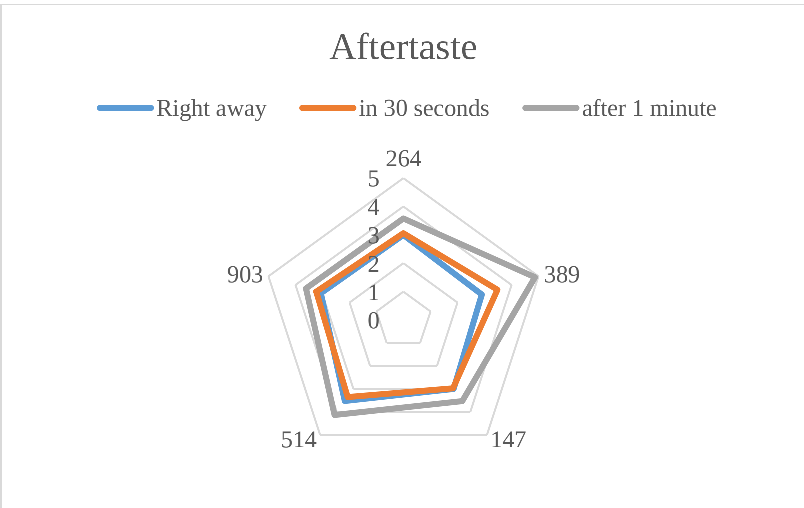


Figure 15. Sensory analysis evaluation- the aftertaste of examined samples

As shown in **Figure 15**, at each of the three intervals, sample 389 has the biggest drop in intensity of aftertaste going down from 5.06 to 2.74. Closely followed by sample

514, samples 389, 903 and 147 received the highest score for intensity of aftertaste. Through all, sample 264 consistently received the lowest overall aftertaste rating. Despite having highest caffeine content such evaluation could indication weak or no correlation between these variables. It is interesting to note that, there was no obvious trend in the aftertaste ratings across the time periods, suggesting that the aftertaste's quality did not necessarily get better or worse with time (for the complete data set, see **Appendix 1**).

Table 7. Pearson's correlation coefficient

Variable		**Caffeine content**
Colour	Pearson's r	0.001
	p-value	0.999
Aroma- sweet	Pearson's r	-0.783
	p-value	0.117
Aroma- fruity	Pearson's r	-0.360
	p-value	0.552
Aroma- acid	Pearson's r	0.043
	p-value	0.945
Aroma- bitter	Pearson's r	-0.167
	p-value	0.788
Aroma- pungent	Pearson's r	0.032
	p-value	0.960
Aroma- musty	Pearson's r	-0.011
	p-value	0.985

Continued Table 7. Pearson's correlation coefficient		
Flavour-sweet	Pearson's r	-0.752
	p-value	0.142
Flavour- fruity	Pearson's r	-0.129
	p-value	0.836
Flavour- acid	Pearson's r	0.284
	p-value	0.644
Flavour- bitter	Pearson's r	-0.135
	p-value	0.829
Flavour- pungent	Pearson's r	0.262
	p-value	0.671
Flavour- musty	Pearson's r	0.027
	p-value	0.965
Overall palatability	Pearson's r	-0.219
	p-value	0.723
Aftertaste- right away	Pearson's r	0.166
	p-value	0.789
Aftertaste- in 30 seconds	Pearson's r	0.342
	p-value	0.574
Aftertaste- in 1 minute	Pearson's r	-0.117
	p-value	0.852

The Pearson's correlation coefficient and p-values for several sensory attributes and chemical components of coffee are shown in **Table 7**. HPLC was used to determine the chemical components, and panellists evaluated the sensory properties giving each a score. The correlation coefficient's level of significance is indicated by the p-values; a p-value of 0.05 or less indicates a significant correlation.

With a Pearson's r of 0.001 and a p-value of 0.999, the results show a weakly positive correlation between the colour and chemical components of coffee. The aromas of musty and pungent, with Pearson's r values of -0.011 and 0.032, respectively, and p-values over 0.9, may demonstrate weak to no association with the caffeine content.

The flavour correlation coefficients result in negative or no linear relationship in specific variables. Bitter flavour, which could be expected as one of the positive correlations, showed negative values with a relatively high p-value. On the other hand, the strong negative correlation of sweet flavour with caffeine content is less surprising.

Pearson's correlation coefficient for overall palatability shows large deviations and also no positive correlation with r of -0.219.

The aftertaste also shows some interesting results. The aftertaste in 30 seconds has a weak positive correlation with a Pearson's r of 0.342 and a p-value of 0.574, while the aftertaste in 1 minute has a weak negative correlation with a Pearson's r of -0.117 and a p-value of 0.852.

Interesting correlations will now be highlighted in following scatter plots:

Correlation plot n.1

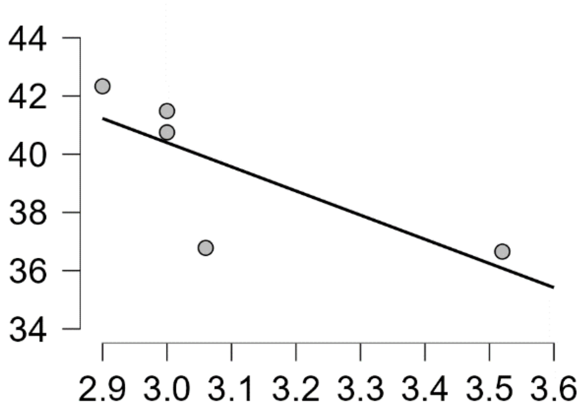


Figure 16. Scatter plot: caffeine and sweet flavour correlation

As shown in **Figure 16**, the first scatter plot shows the relationship between sweet flavour and caffeine concentration, which is negative (r -0.752). This indicates that the sweetness of coffee reduces as the amount of caffeine increases. Although it may seem that sweet flavour clearly demonstrates a negative correlation with caffeine due to variables (147, 264, and 389), the deviation of sample 903 from the regression line weakens the strength of the correlation. The p-value is 0.142 meaning that there is a 14.2 % chance of observing this correlation by chance alone.

Correlation plot n.2

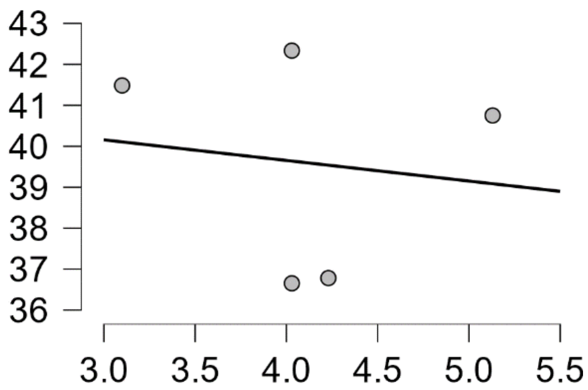


Figure 17. Scatter plot: caffeine and bitter flavour correlation

Another scatter plot (see **Figure 17**) demonstrates the connection between bitter flavour and caffeine content, which has a weak negative association (r -0.135). This suggests that the bitter flavour of the coffee may be caused by more than just the caffeine concentration. This is another unexpected result, considering it is common knowledge that caffeine quantity alone determines how bitter a flavour is.

Correlation plot n.3

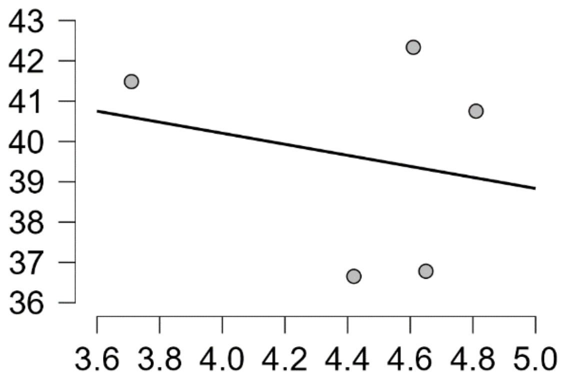


Figure 18. Scatter plot: caffeine and overall palatability correlation

The final scatter plot (see **Figure 18**) is made for overall palatability versus caffeine content because there is a slightly negative relationship between the two (r -0.219). This indicates that the overall palatability of coffee declines as the amount of caffeine in it increases. Some consumers could believe that a higher caffeine content would result in a more intense and complex flavour profile, which might enhance overall palatability.

6. Discussion

The theses original objectives were to determine whether there is a correlation between caffeine content and sensory attributes of coffee, and if even, to what extent. To achieve these goals, a sensory analysis of selected samples was conducted, HPLC analysis was performed, and the correlation between caffeine content and individual sensory analysis scores was determined using statistical measurements.

The use of sensory analysis in coffee research has been demonstrated to provide valuable insights into various aspects of coffee flavour and aroma, including the first indications of both positive and negative correlations between caffeine content and sensory attributes. The findings suggest a possible relationship between caffeine level and specific sensory characteristics of coffee, such as bitterness and pungency. Pearson's correlation coefficient pointed to certain correlations, however, none of them proved to be strong enough to answer the research question in the affirmative, although there were hints. Different results that either positively or negatively correspond to the research question of this thesis only confirm this.

De Oliviera Fassio et al. (2016) study contradicts the assumption that caffeine and the sensory attributes of coffee are actually correlated by concluding that "caffeine did not have a good relationship to the sensory attributes". Another recent study makes a same conclusion, claiming that "caffeine concentration in beans had a negative correlation with all cup quality attributes". Furthermore, the author emphasises that it is in opposition to the chlorogenic acid chemical compound. Correlations with bean hedonic characteristics were non-significant according to Dessalegn et al. (2008).

In contrast to the belief that there is a clear general link when evaluating the results of aroma and flavour correlation, research on the physicochemical and sensory properties of coffee points to a weak to zero association of aftertaste with caffeine level (Patria et al. 2018). The result of the Gimase et al. (2014) study focused on the correlation of not only caffeine and sensory characteristics in coffee completely supports the idea of correlation stating, "positive significant correlations between all sensory characteristics and chemical composition". Another study from the same year that asserting that "non-volatile compounds have a great influence on flavour perception and customer acceptance" provides indirect evidence for this (Sunarharum et al. 2014).

The variation in the coffee samples utilised in various experiments must be taken into account. The origin, method of processing, degree of roasting, and storage conditions of coffee beans can all significantly impact the chemical composition and sensory qualities of the beverage. As a result, the findings of a study on one type of coffee bean or one kind of processing technique might not necessarily apply to other types of coffee.

6.1. Caffeine and colour correlation

As for the colour perception and amount of caffeine, the fact that the sample with the highest caffeine concentration had the lowest colour score and the sample with the second-highest caffeine content had the highest colour score indicates that there might not be a straight relationship between caffeine concentration and colour. Several elements that influence colour perception, including processing techniques, roasting temperatures and times, and storage conditions, can be attributed to this. After all, statistical data also supports this assertion. A Pearson's correlation coefficient of 0.001 and a p-value of 0.999 indicate that there is almost no relationship between caffeine content and colour. As a result, it appears unlikely that the variations in colour ratings between samples are caused by their different caffeine contents.

The colour of the coffee is more likely to be influenced by other elements like the type of coffee bean, the roasting procedure, and the brewing technique. However, it should be noted that each sample's high standard deviation in colour ratings points to some variation in the panellists' perceptions of the coffee's colour. Individual perception differences frequently cause this. The FTA's sensory laboratory is sufficiently prepared to prevent the possibility that the perception of colour itself would be influenced by lighting conditions. Whatever the situation, it is likely that the total standard deviation accurately identifies potential factors that could have impacted the correlation results.

6.2. Caffeine and aroma correlation

Regarding aroma, based on the sensory analysis data, sample 147 had the highest mean scores for the bitter, pungent and musty aroma subcategories but the lowest mean scores for the sweet, fruity, and acid subcategories. Although this might suggest that sample 147's high caffeine level might have influenced how strongly bitter it smelled, correlation coefficient unequivocally disproves it. The high concentration of caffeine in sample 147 may have affected how the panellists perceived the aroma of the coffee because caffeine is known to have a bitter taste. Yet it is important to remember that other factors, such as processing methods, and coffee terroir, may have also contributed to the development of the coffee's aroma profile.

For the acid subcategory, sample 389 received the greatest mean score, while sample 514 had the highest mean score for the sweet and fruity subcategories. There might still not be a direct correlation between caffeine level and aroma evaluation, sample 514 scored highly for the sweet and fruity aroma evaluation despite having less of it which again statistics only supports, showing no significant correlation. These results imply that the aroma profile of the coffee may not be solely determined by the amount of caffeine present.

The high p-value, which exceeded the accepted limit of 0.05, contributes to these not conclusive findings. For instance, the correlation coefficients for sweet and fruity aroma elements are negative, indicating that increased caffeine content is linked to lower ratings for these properties, but the correlations are not strong enough to suggest a substantial linkage.

It appears that there is not a significant association between caffeine content and aroma attributes based on the data from Pearson's correlation coefficient calculation. The caffeine concentration and each aroma element's Pearson's correlation coefficients point to either a weak or non-existent linear relationship between them.

Overall, the given data does not provide strong evidence in favour of the theory that coffee's caffeine concentration and aroma sensory properties are related which is in accordance with already mentioned research of Patria et al. (2018). Nevertheless, although the results do not support the idea of correlation between caffeine content and sensory attributes, it implies possible use in developing coffee products with specific caffeine content while maintaining desirable both colour and aroma.

6.3. Caffeine and flavour correlation

According to the Pearson's correlation coefficients for the caffeine concentration and flavour components, there also does not appear to be a significant connection between them. Samples with higher caffeine levels may have lower sweetness ratings, as indicated by the correlation coefficient between caffeine content and sweet flavour, however this association is not statistically significant. It may seem that sweet flavor clearly shows a negative correlation with caffeine due to the variable (samples 147, 264, 389), however, the deviation of sample 903 weakens the strength of the correlation. Thus, no significant correlation can be demonstrated.

A weak positive correlation exists between caffeine content and pungent flavour, while there is no apparent relationship between caffeine content and fruity flavours. The p-values for each of these correlations are over the conventional threshold of 0.05, indicating that the observed correlations could be due to chance.

There was no significant link between the bitter flavour and caffeine levels in the samples studied, with Pearson's r value of -0.135 and p-value of 0.829. This supports research of De Oliviera Fassio et al. (2016) that there may be various causes for the bitter flavour in coffee and they may not all be related to the amount of caffeine present, CQA content for instance.

The large standard deviation values for each sample in the sensory evaluation also indicate some diversity in the panellists' perceptions of flavour, which could have an impact on the strength of any found relationships. If there is a connection between caffeine level and flavour in this context, more study with a larger sample size and presumably also fully trained panellists may be required to come to a more definitive conclusion. The conclusion that there is a relationship between flavour sensory characteristics and caffeine level in terms of flavour is therefore not strongly supported by these results.

6.4. Caffeine and overall palatability correlation

Caffeine concentration and overall palatability do not appear to be significantly correlated, according to the insignificantly negative Pearson's correlation coefficient of -0.219. Suggesting that other elements, including roast degree, brewing method, and coffee origin, have an impact on the overall flavour of coffee and are not always influenced by its caffeine content. Due to the fact that certain tasters might prefer a milder flavour while others would prefer a stronger and more robust flavour, there may be a slightly negative relationship between caffeine level and overall palatability.

Also, the samples' overall palatability scores were quite similar, suggesting that there may not be much of a flavour variation between them, perhaps since they are all from the same country of origin. Hence, determining total palatability may depend more on criteria besides caffeine concentration. Individual preferences for specific aroma and flavour components could have also affected the overall palatability score. On the other hand, the scatter plot's barely detectable negative association between overall palatability and caffeine content may suggest that there is some relationship between sensory qualities and caffeine content. Conclusions are difficult to draw because of the correlation coefficient's small effect size.

6.5. Caffeine and aftertaste correlation

The association between caffeine concentration and aftertaste does not appear to be particularly strong when the Pearson's correlation coefficients are examined. Since the coefficients for each time period range from -0.117 to 0.342, there is weak (both positive and negative) to no correlation. Patria et al. (2018) study support this finding in their own research on Physicochemical and sensory characteristics of coffee emphasizing week linear negative relationship of caffeine with aftertaste. Hence, it seems unlikely that the samples' caffeine content had a considerable impact on mouthfeel right away, or with time gap. The aftertaste ratings may also be influenced by other variables, such roasting methods, bean varieties, or even taster preferences.

6.6. Correlation with commercial and specialty coffee

Sample 147 consistently outperformed the other samples in terms of overall palatability and aftertaste, according to the sensory analysis results, indicating that it possesses distinctive qualities that appeal to panellists more. This shows that commercial coffee may have a more uniform flavour profile that appeals to a wider spectrum of customers since it is frequently manufactured in bigger numbers and goes through more standardised processing. It is also possible that commercial coffee's higher caffeine content relates to its more appealing flavour profile. Furthermore, sample 147 had the second-highest caffeine concentration of all the samples, which raises the question of whether the sample's high palatability scores are due to the caffeine content of the sample, or if they instead reflect characteristics derived from its place of origin, processing, or other factors.

There could be a number of explanations why sample 147 had higher scores in terms of overall palatability and aftertaste compared to the other samples, while having a higher caffeine content. It is likely that despite being a commercial sample, this one's flavour was not negatively affected by the higher caffeine content because it was made from high-quality coffee beans with a good flavour profile. Another possibility is that this sample's roasting was optimised to maximise flavour while reducing any adverse effects from the caffeine concentration. In addition, it is possible that tasters in the sensory analysis panel favoured sample 147's flavour profile over that of the other samples regardless sample 147's caffeine content.

According to the results of the sensory analysis and the data on the caffeine content, there was no obvious relationship between the caffeine content and the sensory properties of the specialty coffee samples. Although while some samples had more caffeine than others, this did not always reflect into a better sensory profile.

In fact, some of the specialty coffee samples with less caffeine, such sample 264, obtained high sensory scores. This suggests that other elements, such as coffee variety, processing technique, and origin, can determine specialty coffees' sensory profile more significantly.

It is worth mentioning that specialty coffee samples often received higher sensory analysis scores than commercial coffee samples. This indicates that specialty coffee, which is often cultivated and processed with greater attention and quality control, may provide a more consistent and higher sensory experience than commercial coffee. After all, that is the reason speciality coffee has become so popular lately.

6.7. Limitations

Despite the valuable insights gained from this study, there are some limitations that need to be acknowledged. One of the main limitations is the small sample size of only five samples. While this was a deliberate decision to allow for a more in-depth analysis of each individual sample and conduct high-quality sensory analysis, it also limits the generalizability of the findings to a larger population of coffee samples. Additionally, the samples used in this study were selected based on geographic variability, which may have limited the range of sensory attributes and caffeine content observed.

Another drawback is that all samples were evaluated by a single sensory panel, which may have caused some results to vary. Moreover, the panel was consisted of only semi-trained respondents whose experience and training levels may not be standardized or consistent, leading to variations in their evaluations and potentially affecting the reliability of the results. Given that the majority of respondents were from age groups 20-39, it cannot be assumed that the difference in results is a consequence of the different age of the respondents, as is the case in many cases. Finally, the analysis omitted other potential variables that can affect the sensory profile of coffee, such as roast degree, processing method, or brewing technique, focusing solely on the correlation between caffeine content and sensorial properties. Future research should aim to address these limitations in order to provide a more comprehensive understanding of the relationship between caffeine content and sensory characteristics of coffee.

7. Conclusions

According to the analysis of the sensory attributes and caffeine content data, there was no clear relationship between sensory attributes and caffeine content. A few sensory attributes showed some weak associations, but they were not statistically significant enough to make definite conclusions. Moreover, additional factors including processing methods, roast level, and coffee terroir may have a certain impact on the sensory characteristics of coffee. Hence, claiming that there is a direct correlation between sensory attributes and caffeine content would be premature. This study showed that caffeine content does not necessarily correlate with certain sensory attributes, highlighting the complexity of factors that contribute to the coffee characteristics.

The results suggested that there may not be a strong relationship between caffeine content and certain sensory attributes in coffee, such as colour and aroma, but there could be a correlation with attributes such as taste and aftertaste. This is demonstrated by the Pearson's correlation coefficient values, which, in contrast to the other attributes examined, did not indicate an entirely negative correlation but instead suggested a correlation in positive numbers rather than negative. Under certain conditions, such as the use of trained panellists for sensory analysis or a different technique for preparing coffee, this may be proven right. The results can be attributed to the complexity of the chemical components of roasted beans, and it can therefore be said that it is difficult for the panellists to accurately identify it. Hence, it is important to remember that the sensory analysis of coffee is a complex method, and the results should only be interpreted with caution.

Based on the findings of this study, further research might investigate the relationship between terroir, sensory qualities, and caffeine content in coffee. It is likely that other factors, such as growth conditions and soil composition, could influence the connection between caffeine and sensory characteristics, even if the study did not find a significant correlation. Also, researching the effects of various roasting techniques on caffeine levels and sensory qualities may reveal important information for the coffee industry.

Further research could also look into the impact of antioxidants and polyphenols such as chlorogenic acid on sensory properties as well as their possible connections to caffeine content. Ultimately, researchers may contribute to advancing our understanding of coffee and supporting the development of better standard coffee products by continuing to investigate the complex relationships between coffee chemistry and sensory experience. It is crucial to carry out further research on a subject in order to clarify and confirm earlier conclusions or to spot any knowledge gaps that might exist.

The findings of my study might be valuable to a variety of people, including coffee roasters, coffee shops, and coffee enthusiasts. The data might be used by coffee roasters to adjust the amount of caffeine in their product to meet the preferences of their target market. The association between caffeine level and general palatability is one such outcome that a coffee roaster might employ. If a roaster wishes to make a coffee with a lot of caffeine, they could believe that adding more caffeine will immediately make the coffee taste better. The roaster would want to re-evaluate this assumption and instead concentrate on other characteristics that affect palatability, such as coffee terroir or roast degree, in light of the study's negative correlation finding. The development of coffee products with improved flavours and more commercial viability may result from this information.

Coffee shops could use the information to make informed decisions about which coffee beans to use in their blends or to provide more information to their customers about the caffeine content of their products. Based on their preferences for caffeine, coffee drinkers might use the knowledge to make more informed judgements about which coffee beans to buy and drink. The results of the study may also be helpful to researchers and academics engaged in the sensory analysis and coffee science fields.

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Appendices

List of the Appendices:

Appendix 1 Sensory analysis evaluation

1. **AROMA EVALUATION:**

Sweet	Mean score
147	2.84±1.49
264	3.16±1.63
389	3.23±1.48
514	3.87±1.8
903	3.52±1.57

Fruity	Mean score
147	2.77±1.36
264	3.58±1.76
389	3.65±1.36
514	3.74±1.61
903	3.71±2.08

Acid	Mean score
147	2.36±1.50
264	3.06±1.59
389	3.90±1.92
514	3.23±1.73
903	3.32±1.4

Bitter	Mean score
147	4.48±2.05
264	2.81±1.58
389	3.80±1.76
514	4.13±1.76
903	3.48±1.79

Pungent	Mean score
147	4.12±1.93
264	2.55±1.52
389	3.39±1.61
514	3.35±1.91
903	3.1 ±1.68

Musty	Mean score
147	2.77±1.36
264	3.58±1.76
389	3.65±1.36
514	3.74±1.61
903	3.71±2.08

2. **FLAVOUR EVALUATION:**

Sweet	Mean score
147	3±1.79
264	3±1.86
389	2.9±1.94
514	3.52±2.11
903	3.06±1.93

Fruity	Mean score
147	2.97±1.91
264	3.06±1.91
389	3.48±1.95
514	3.35±1.58
903	3.23±1.93

Acid	Mean score
147	3.52±2.01
264	3.58±1.88
389	4.87±1.91
514	4.13±2
903	3.61±1.82

Bitter	Mean score
147	5.13±2.01
264	3.1±1.89
389	4.03±1.78
514	4.03±1.87
903	4.23±1.78

Pungent	Mean score
147	4.55±2
264	2.71±1.66
389	3.97±2.21
514	3.45±1.67
903	3.2±2.04

Musty	Mean score
147	4.3±2.34
264	3±1.73
389	3.94±2.16
514	3.71±1.81
903	3.65±2.26

3. AFTERTASTE EVALUATION:

Right away	Mean score
147	3±1.79
264	3±1.86
389	2.9±1.94
514	3.52±2.11
903	3.06±1.93

In 30 sec	Mean score
147	2.97±1.91
264	3.06±1.91
389	3.48±1.95
514	3.35±1.58
903	3.23±1.93

In 1 min	Mean score
147	3.52±2.01
264	3.58±1.88
389	4.87±1.91
514	4.13±2
903	3.61±1.82

Appendix 2 Sensory analysis questionnaire

SENSORY EVALUATION QUESTIONNAIRE – COFFEE

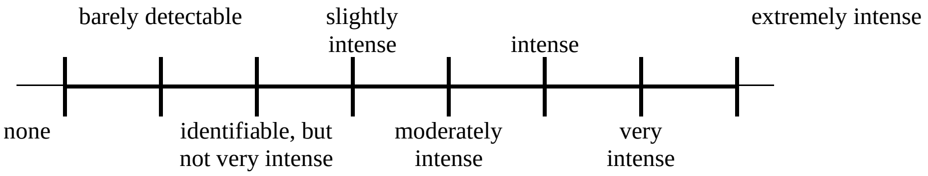
Date					
Samples					

Sex: female ☐ male ☐ other ☐

Age: 0-19 years ☐ 20-39 years ☐ 40-59 years ☐ 60 and more ☐

The survey participant is able to provide an unbiased assessment of coffee because they do not routinely consume alcohol, smoke, are not pregnant, or feel fatigued or ill.

legend:



1. Colour

Determine the intensity of the colour on the line segment below using the image above.

none — extremely intense

2. Aroma

Determine the intensity of the aroma on the line segment below using the image above.

Sweet: none — extremely intense

Fruity: none — extremely intense

Acid: none — extremely intense

Bitter: none — extremely intense

Pungent: none — extremely intense

Musty: none — extremely intense

Your own evaluation, or observation regarding the aroma:

none extremely intense

3. Flavour

Determine the intensity of the flavour on the line segment below using the image above.

Sweet

none extremely intense

Fruity

none extremely intense

Acid

none extremely intense

Bitter

none extremely intense

Pungent

none extremely intense

Musty

none extremely intense

Your own evaluation, or observation regarding the flavour:

none extremely intense

4. Overall palatability

Determine the overall palatability (taste, smell) on the line segment below using the image above.

none extremely intense

5. Aftertaste

Determine the aftertaste (mouthfeel) with the time gap on the line segment below using the image above.

right away

none — extremely intense

in 30 seconds

none — extremely intense

in 1 minute

none — extremely intense

6. Overall impression (optional comment - add a note on the taste, aroma and overall acceptability of the coffee)