

# Altering the physical-chemical properties of coffee using digestive enzymes

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**Abstract.** The article explored an innovative bioreactor-based coffee fermentation process, closely mirroring the natural digestive system of civets. This intricate process unfolded in three distinct stages: Stomach Replication: In the first stage, the bioreactor replicates the solution present in the stomach of civets. Intestine Emulation: Moving to the second stage, the bioreactor emulates the conditions found within the intestines of these animals. Colon Fermentation: The third and final stage involves the introduction of lactic acid bacteria, mirroring the fermentation process that occurs in the colon. Lactic acid bacteria are added at concentrations aligning with those observed in the civet's small intestine. The research presented the proximate analysis of coffee, encompassing parameters such as water content, ash content, lipid, protein, and carbohydrate. Notably, the findings reveal that while there are no significant differences in water content, ash content, and lipid content between bioreactor/artificial luwak coffee and the original luwak coffee, the protein content in artificial luwak coffee exceeds that of the original luwak coffee. This suggests that the bioreactor process effectively mitigates protein degradation that typically occurs during natural fermentation. This degradation is attributed to the acidic environment within the civet's digestive system, with the article emphasizing that the addition of an acid solution during research does not harm the coffee beans. In fact, it enhances the quality of coffee beans, as supported by previous studies.

**Keywords.** Coffee, Enzyme, Fermentation, Exotic, Quality.

## 1. Introduction

Coffee stands as one of the most commonly consumed beverages, with its popularity extending across the globe. It ranks as the second most traded commodity in international markets, closely following petroleum. The primary producer and exporter of *Coffea arabica* are Brazil, trailed by Indonesia, Ethiopia, the Philippines, Mexico, Vietnam, along with 40 other nations. People indulge in coffee for relaxation, savoring its diverse range of flavors and captivating aromas. Beyond its nutritional advantages, coffee exerts physiological and psychological effects. In recent times, an increased appreciation for premium coffee has elevated the significance of specialty blends, each boasting unique aromas and exceptional flavors. These distinctive characteristics are intrinsically tied to the specific regions where the coffee is cultivated and the genetic variations of the coffee

plants. Some scholars have even noted significant distinctions among beverages crafted through different coffee processing techniques. The prevailing belief is that distinct processing methods trigger varying metabolic reactions in coffee fruits, ultimately influencing the chemical composition of coffee beans and, consequently, their cupping quality [3].

The global significance of coffee's impact on health is underscored by its widespread consumption, with FAO data indicating an annual global intake of approximately 7 million tons. Coffee, in its essence, is a brewed beverage crafted from the roasted seeds of the *Coffea* genus. Enclosed within berries, these coffee seeds, or beans, are meticulously processed and dried upon reaching maturity. The coffee's historical journey originates in Ethiopia, where it was believed to possess invigorating properties prior to the 14th century. By the middle of the 15th century, evidence points to the consumption of

coffee within Yemeni Sufi monastic communities, serving as a gateway for its propagation across the wider Middle East and northern Africa. The robust trade conducted by Venetian vessels with the Middle East played a pivotal role in introducing coffee to Europe, subsequently paving its way to the shores of the Americas [1].

The priciest coffee globally goes by the name Kopi Luwak, commanding a staggering \$90 per serving in the United States. Regrettably, not widely known is the dark truth behind its production, entailing exploitation and infliction of suffering upon animals, particularly the civet mammal native to Indonesia, as reported by ABC News network [7].

The coffee fruit consumed by the civet encompasses the entire package, including the fruit skin (pulp), mucilage layer (mucilage), and coffee beans. However, only the beans remain intact after processing. The primary constituents present in the fruit skin and mucilage layer undergo degradation facilitated by microorganisms during the fermentation process of civet coffee, along with the enzymes it generates. The fruit skin and mucilage layer of coffee boast substantial pectin and protein content, reaching as high as 42.3% and 18.9%, respectively. It is believed that the bacteria responsible for fermenting civet coffee possess the capability to break down pectin and protein. These pectinolytic and proteolytic bacteria are recognized for their ability to decompose sugars, with each capable of utilizing either pectin or protein as a substrate. Pectin degradation results in various simple sugar compounds, while protein decomposition yields a plethora of peptides that are considered precursors in the creation of aroma and flavor compounds. Consequently, this study's objective is to identify the bacterial population within the mongoose's digestive tract and assess its potential for protein and pectin breakdown [6].

The present work aims to demonstrate that it is possible to alter the physicochemical properties of coffee with the use of some digestive enzymes in order to mimic the action of other enzymes of the digestive system of omnivorous animals, previously mentioned, in the production of exotic coffee allowing the coffees submitted to enzymatic treatment and fermentation to present different characteristics (more desirable) from those of commercial coffees.

## 2. Methodology

Articles published between 2004 and 2023 were selected after crossing coffee and exotic with a list of relevant keywords.

## 3. Results and Discussions

### 3.1 Use of digestive enzymes to mimic natural digestive processes for exotic coffee production

The bioreactor-based fermentation process of coffee mirrors the natural fermentation that occurs within the digestive system of civets, encompassing the stomach, intestines, and colon. This intricate process unfolds in three distinct stages.

Firstly, the initial phase involves replicating the solution present in the stomach of civets.

Moving on to the second stage, the bioreactor emulates the conditions encountered within the intestines of these animals.

The third and final stage entails the introduction of lactic acid bacteria, mirroring the fermentation process within the colon. The quantity of lactic acid bacteria added to the bioreactor typically ranges from  $10^8$  cfu/ml to  $10^9$  cfu/ml, aligning with the concentrations observed in the civet's small intestine, which surpasses  $10^7$ - $10^8$  cfu/ml [2].

Figure 1 visually illustrates the various stages of producing artificial or bioreactor-derived luwak coffee, starting from coffee berries (a), followed by coffee beans with endocarp skin (b), green coffee beans (c), and finally roasted coffee (d). The coffee beans with endocarp skin are obtained after undergoing the fermentation process in the bioreactor, followed by meticulous washing and drying using either an oven or natural sunlight [2].



**Fig. 1** - Coffee berries (a), coffee bean with endocarp skin (b), green bean coffee (c), and roasted coffee (d) [2].

The research findings are presented in Table 1, illustrating the proximate analysis of coffee, encompassing parameters such as water content, ash content, lipid, protein, and carbohydrate.

Notably, there are no significant differences observed in the water content, ash content, and lipid content between the bioreactor/artificial luwak coffee and the original luwak coffee. However, it's worth highlighting that the protein content in artificial luwak coffee surpasses that of the original luwak coffee. This suggests that the bioreactor process effectively mitigates the protein

degradation that typically occurs during the natural fermentation process.

The degradation of protein can be attributed to the acidic environment within the civet's digestive system, as the pH of gastric fluid in civets is lower than the pH of the simulated gastric fluid in the bioreactor. It's important to note that the addition of acid solution in this research does not inflict damage upon the coffee beans. In fact, there is an enhancement in the quality of coffee beans following acid and enzyme treatment [2].

**Tab. 1** -Proximate analysis of roasted coffee.

Treatment	Water (%)	Ash (%)	Lipid (%)	Protein (%)	Carbohydrate (%)
Artificial luwak coffee	2.05 ± 0.11	5.38 ± 0.22	12.21 ± 0.19	13.11 ± 0.23	67.25 ± 0.33
Original luwak coffee	2.00 ± 0.15	5.34 ± 0.17	12.13 ± 0.22	11.08 ± 0.31	69.44 ± 0.28

Data are presented as mean ± SD (n = 3) [2].

### 3.2 Structural analysis of exotic coffee beans

The research conducted by Marcone in 2004 represents the inaugural scientific inquiry into the diverse physicochemical attributes of palm civet coffee beans (Kopi Luwak) from Indonesia, contrasting them with the coffee beans obtained from African civets in Ethiopia, Eastern Africa. Through scanning electron microscopy, it was unveiled that all civet coffee beans exhibited surface micro-pitting when examined at a magnification of 10,000×. These minute irregularities were the result of the interaction between gastric juices and digestive enzymes during the digestion process.

Furthermore, comprehensive mechanical rheology testing divulged that civet coffee beans possessed a greater degree of hardness and brittleness in comparison to their control counterparts. This finding hinted at the infiltration of digestive juices into the beans, thereby modifying the micro-structural properties of the beans themselves. The SDS-PAGE analysis lent further credence to this observation by illustrating the infiltration of proteolytic enzymes into all civet beans, leading to a significant breakdown of storage proteins.

It's important to note that variations were observed in the susceptibility of different subunits to proteolysis between civet coffee types, ultimately resulting in disparities in Maillard browning products, and consequently, distinctive flavor and aroma profiles.

Lastly, the data generated by the electronic nose discerned clear distinctions between civet coffees

and their control counterparts, underlining the substantial transformation these coffees undergo as they traverse the gastrointestinal tract of civets [5].

## 4. Conclusion

This comprehensive exploration of coffee covers its global importance, ethical concerns, physiological changes during production, and innovative methods for altering coffee properties, offering valuable insights for coffee enthusiasts and industry professionals alike.

The authors demonstrate, through well-founded physical-chemical analyses, that it is possible to alter the physical-chemical properties of coffee with the use of pre-selected enzymes that simulate the action of enzymes that act on the digestive system of some omnivorous animals in the coffee fruit to the production of exotic coffee, considered superior in quality. The use of such a technique can be explored at an industrial level, given the high added value of the product and the idea of no longer using the animal as a biological reactor, contributing positively to sustainability.

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