

Composition and properties of Indonesian palm civet coffee (Kopi Luwak) and Ethiopian civet coffee

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Abstract

This research paper reports on the findings of the first scientific investigation into the various physicochemical properties of the palm civet (Kopi Luwak coffee bean) from Indonesia and their comparison to the first African civet coffee beans collected in Ethiopia in eastern Africa. Examination of the palm civet (Kopi Luwak) and African civet coffee beans indicate that major physical differences exist between them especially with regards to their overall color. All civet coffee beans appear to possess a higher level of red color hue and being overall darker in color than their control counterparts. Scanning electron microscopy revealed that all civet coffee beans possessed surface micro-pitting (as viewed at 10,000× magnification) caused by the action of gastric juices and digestive enzymes during digestion. Large deformation mechanical rheology testing revealed that civet coffee beans were in fact harder and more brittle in nature than their control counterparts indicating that gestive juices were entering into the beans and modifying the micro-structural properties of these beans. SDS-PAGE also supported this observation by revealing that proteolytic enzymes were penetrating into all the civet beans and causing substantial breakdown of storage proteins. Differences were noted in the types of subunits which were most susceptible to proteolysis between civet types and therefore lead to differences in maillard browning products and therefore flavor and aroma profiles. This was confirmed by electronic nose analysis which revealed differences between the palm civet coffee (Kopi Luwak) and African civet coffee aroma profiles. Analytical techniques for the authentication of palm civet (Kopi Luwak) and African civet coffee are also explored. It would appear that SDS-PAGE may serve as the most reasonable and reliable test to help confirm the authenticity of civet coffee. Electronic nose data was able to distinguish both civet coffees from their control counterparts and further indicated that processing through the civets gastro-intestinal track substantially modified these coffees.

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1. Introduction

Coffee is grown in over 80 countries around the world which lie within 1000 miles north and south of the equator. Of the many varieties grown world-wide a few varieties have achieved a special reputation and notoriety based upon their rarity and overall flavor. Of these Jamaican Blue Mountain and Tanzanian Peaberry are the most notable and as such command a premium price. Although these coffees are in short supply, no coffee is perhaps in shorter supply and has a more distinct flavor and history than a coffee called Kopi Luwak from In-

donesia. With an annual production of under 500 pounds and a price tag of 600 dollars (Canadian) per pound, it commands the undisputed reputation of being the rarest and most expensive coffee or beverage in the world.

Although Kopi Luwak (the Indonesian words for coffee and civet) comes from the Indonesian islands of Java, Sumatra and Sulawesi, it is not its exotic location of origins but rather its unusual and quite unexpected method of production which contribute to its mystique and price. The desire to consume unique food products is a characteristic of passionate coffee drinkers. To this end, an unique coffee emerged from the jungles of Indonesia and became known as Kopi Luwak in the West. This is indeed a rare and unique coffee as it is processed in the digestive system of the indigenous palm civet

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(*Paradoxurus hermaphroditus*). This 3–10 pound animal is an expert tree climber and lives in the trees. During the night it uses its eyesight and smell to seek out and eat only the ripest, reddish coffee cherries. The coffee cherry fruit is sweet and is completely digested by the Luwak but the beans are excreted in their feces. This internal fermentation and action by different digestive enzymes add a unique flavor to the beans which has been described as earthy, musty, syrupy, smooth, and rich with both jungle and chocolate undertones. It is believed that the Kopi Luwak coffee has its origins in the Dutch coffee plantation estates (approx. 200 years ago) before Indonesian achieved its independence in 1945. It is believed that the local people could bring in these unusual beans and, in return, receive a small payment from the estate managers (Schoenholf, 1999).

In the West, the knowledge about the existence of this coffee was not well known until the March 1981 issue of National Geographic which mentioned this coffee by name in one of their feature articles entitled “The Bonanza Bean – Coffee” (Starbird, 1981).

Curiously, Kopi Luwak is not the first nor only human food produced by its passage through or part of the digestive tract of an animal and used for human consumption.

The world’s first sweetener, honey, is a product produced from a combination of flower nectar and pollen and bee ‘excretions’. The bee transports its nectar to its hive to make it into honey by first swallowing it. They then regurgitate it and mix it with other substances, removing a majority of the liquid from it and it becomes honey which they use to feed their young and each other.

Another is Bird’s nest soup often referred to as the “Caviar from the east”, is produced from the saliva of a swiftlet (a small bird native to Southeast Asia). It is consumed in many parts of the world with the United States being one of its biggest importers. This tiny bird makes its nest from strands of its own gummy saliva which hardens when exposed to air.

Argan oil is produced by goats in Morocco who are encouraged by their herders to climb acacia-like trees and eat the fruit which is similar to olives. The locals remove the pits from the feces and grind them to extract their oil. This unusual and expensive oil is used in massage and cooking, and is repudiated to be an aphrodisiac.

Presently, the main island producer of Kopi Luwak is Sulawesi (Indonesia) but due to a local war, few people venture into the forest to collect this rare, unique coffee for fear of being killed by the warring parties. As a result, the coffee supply has virtually dried up and it is extremely difficult to locate authentic sources of Kopi Luwak coffee.

The desire to possess and drink unique coffee has not ceased so other sources of animal ‘processed’ coffee were explored. The search began by determining the coffee growing areas of the world. Researching the cultivation areas world of coffee, it was determined that coffee only

grows within 1000 miles north and south of the equator. The next criteria was to find possible animals within this region who could possibly eat coffee cherries and, again through some research, the African civet (*Civettictis civetta*) was the best possible candidate. This 15–45 pound animal lives in sub-Saharan Africa, i.e., from Senegal to Somalia and south to Namibia and eastern South African. Unlike the Indonesian civet, this animal lives mainly on the ground in forests, savannah, and areas with long grasses and thickets. It usually lives near permanent water systems and is repudiated to be a good swimmer. It is nocturnal and is an omnivore – it eats fruit, carrion, rodents, insects, eggs, reptiles, birds and vegetation. It is known that this civet deposits its feces in special piles called civetries which are located near the edge of its territory. The Ethiopian people also capture this animal and harvest the musk from the perineal glands which are located just under the tail. This musk has traditionally been used in the perfume industry but its use has been greatly reduced due to the influence of people who are concerned about the animals’ welfare. The third hurdle was to determine which of the countries within the geographical area of the civet was politically stable. The choice was Ethiopia, the birthplace of coffee which is known world-wide for its long history of growing quality Arabica coffee. With this determination, a research expedition into Ethiopia in December 2003 to find and study this civet, was undertaken to hopefully determine scientifically if this cat-like animal eats coffee cherries. This was indeed established, without a doubt, that this animal does indeed eat the coffee cherries and excretes the ‘processed’ beans in their feces. Extensive documentation of the findings and samples from various locations in Ethiopia were taken back to Canada for authentication and analysis.

The objectives of this study were first to determine the physico-chemical properties of Kopi Luwak and to determine and compare them to those of the African civet to determine if the former may one day serve as a substitute for Kopi Luwak.

2. Materials and methods

2.1. Materials

Kopi Luwak and control beans (not having gone through the palm civet) were obtained from Holland Coffee California Inc. (Novato, CA).

Both the Kopi Luwak and control coffee beans were of the *Coffea robusta* (canephora variety) and originated in Aceh Province in Sumatra, Indonesia during the 2002 harvest season.

African civet coffee and its respective control beans (not having gone through the African civet) were collected from both the regions around Abdelah and

Nekemte in western Ethiopia during the period of December 3–21, 2003.

All the beans were dried to 11% moisture before experiments were begun.

2.2. Methods

2.2.1. Colorimetric determination

The color (L^* , a^* , and b^* values) of the raw coffee beans was determined using a Minolta Chroma Meter CR-200b (Minolta Camera Co, Ltd., Osaka, Japan).

2.2.2. Scanning electron microscopy

Beans were mounted on aluminum stubs and coated with gold/palladium (60/40) to a thickness of 25–30 nm using an Anatech Hummer VII sputter coater (Alexandria, VA). Following coating, beans were viewed under a Hitachi S-570 scanning electron microscope with a high voltage setting of 10–20 kV.

2.2.3. Proximate analysis of beans and elemental (P, K, Mg, and Ca) analysis

Proximate analysis was performed as prescribed in the official standard methods of the American Association of Cereal Chemists, Inc. (American Association of Cereal Chemists, 1983) after beans were ground using a coffee grinder (Black & Decker, Toronto, Ont.). For elemental analysis, 0.250 g samples of oven-dried bean material were wet digested and subjected to atomic absorption analysis as described by Marcone and Yada (1997) and Marcone (2000).

2.2.4. SDS–PAGE

SDS–PAGE electrophoresis was performed on raw coffee beans after normalization for differences in protein content. Approximately 50 mg of ground coffee bean material was dissolved in 1 ml of SDS–PAGE Standard derivitization and procedure run according to the method outlined by Marcone and Yada (1997).

2.2.5. Microbial tests

0.5 g of ground coffee bean (unroasted and roasted) were diluted into 4.5 ml of 0.1% sterile peptone water. 10-fold dilutions were made in peptone water and 0.1 ml spread plated onto Tryptic Soy Agar (TSA) and Violet Red Bile Agar (VRB). 1.0 ml of each dilution was also plated on 3 M *Escherichia coli* coliform petrifilms. Plates and films were incubated at 37 °C for 24 h prior to enumeration.

2.2.6. Roasting of coffee beans

All green coffee beans (control and civet) were roasted in a Probat Twin Roaster (Probat Burns Inc., Memphis, TN) under identical roasting conditions to achieve a medium colored roast. Roasting times to the onset of the first and second cracks for each roast were

kept constant at 8 and 11 min, respectively followed by forced air cooling for 1 min. Each type of roasted bean resulted in beans that were uniform in color as measured by a Minolta colorimeter.

2.2.7. Cupping of coffee

All roasted beans (controls and civet coffee) were ground to a standard cupping grind of 800 μm essential as described by Lyman, Benck, Dell, Merle, and Murray-Wijelath (2003). Coffee was brewed by adding six ounces of hot distilled water (98 °C) to exactly 8 g of ground coffee into a standard tasting cup/bowl and allowed to steep for 4 min. The crust floating on top of each cup was then broken and the coffee aroma noted as per official steep cup methodology. The various roasted coffee beans were then tasted by a (blinded) certified cupper and evaluated as per official coffee cupping procedures.

2.2.8. Electronic nose analysis

Tests were conducted using an Alpha MOS Fox 3000 Electronic nose equipped with 12 metal oxide sensors. Prepared samples are placed into a sample tray for the auto-sampler and held at room temperature during sampling process. Samples were transferred sequentially to the incubator/heating block and gently agitated at constant rpm/directional cycle to facilitate headspace sample production. The headspace sample (1000 or 500 μL) was drawn into the syringe and transferred to the injection port of the electronic nose. Sensor response data was collected for 120 s followed by a 1080 s delay before injection of the next sample. The carrier gas (flow rate maintained at 150 ml/min) is oxygen/nitrogen at 20% (i.e., 19.8–20.2% O_2) and with impurities specified as $\text{H}_2\text{O} < 5$ ppm, $\text{C}_n\text{H}_m < 5$ ppm, $\text{O}_2 + \text{N}_2 > 99.95\%$, $\text{O}_2 = 20 \pm 1\%$. For data analysis the numerical values of the changes in the sensor resistance are recorded as individual response patterns for each of the twelve metal oxide sensors by the computer system operated through the Alpha MOS AlphaSoft V8.0 software. This data is used for the multivariate statistical analyses contained in the program. This allows the evaluation of the presence/detection of differences within and among sample groups. The principal component analysis (PCA) subroutine in the program was used to generate the maps to compare the data within and between samples.

2.2.9. Large deformation mechanical rheology

Yield force (F_y) and compression modules (k) measurements were conducted using a Stable Mirco System Materials Tester model MT-LQ (Surrey, England) fitted with a 50 kg load cell. The geometry attached to the load cell was a 38 mm diameter stainless steel compression platen and positioned exactly at the top of the bean and then lowered at a constant rate of deformation (10 mm/s). Large deformation test parameters, F_y and k ,

were obtained using the software provided by the manufacturer.

3. Statistical analysis

Statistical analysis was performed using a SAS Statistical Analysis System package. Significant differences among treatments (samples) were determined by Duncan's multiple range test ($p \leq 0.05$) (SAS, 1990).

4. Results and discussion

Physical characterization of the palm civet coffee beans (otherwise known as Kopi Luwak) collected in the province of Apec in Indonesia and the two African civet coffee bean types collected in Abdela and Nekemte in Western Ethiopia indicate the existence of major distinguishable differences between them with regards to their overall size, weight and color.

Preliminary examination of the unroasted beans by analytical color measurement equipment (Minolta colourimeter) indicated substantial differences in overall color between the three types of civet beans and their corresponding control beans (control bean being defined as those not having gone through the civet's digestive tract (GI)). (Table 1, Fig. 1). In general, control beans were more uniformly green in color with little discernable (quantifiable) color difference between them (Table 1, Fig. 1). The largest of the civet coffee beans were from the palm civet (Kopi Luwak) followed closely by those from Nekemte with the smallest beans collected being those from Abdela (Table 1). Although palm civet (Kopi Luwak) coffee beans were the largest, they were actually the lowest in weight compared to Nekemte and Abdela civet beans (with Nekemte and Abdela civet bean weights agreeing with their overall size). (Fig. 1). Instron (compression measurements) on the civet beans as compared to their control beans indicated that they were substantial harder and more brittle in nature than their corresponding control beans which were softer and more elastic in nature (with bean fracturing occurring at applied perpendicular forces of 200–275 and 80–100 N, respectively). The observed differences would indicate that same penetration of gastric juices and or digestive enzymes were occurring entering into the beans and a change in the micro structural properties of the civet beans was occurring during transit through the civet's GI track.

The palm civet (Kopi Luwak) coffee beans were significantly higher in their level of red and yellow hues rendering them with an overall beige color compared to the African civet beans, which were greener in color.

Both types of African civet beans were darker in color (i.e., lower L^* value) than their control beans and both higher in the reddish color than their respective controls.

These differences in color would indicate that in the case of all three civet bean types some form and level of chemical reaction was occurring at the very surface of the bean which was rendering them discernibly darker in color than their corresponding control counterparts. Although this was true the actual color hue was different between them with the commonality being that all three civet bean types were slightly but significantly higher in the reddish hue tone as well as being significantly darker in overall color. At this point it can be indicated that the various digestive biochemicals (gastric juices and proteases) are actually penetrating the outer coffee cherry (pericarp) after ingestion by the civets and reaching the actual bean surface where the chemical color change was occurring. It would be difficult to use color analysis as the sole means to determine the authenticity of civet beans as it appears that the magnitude of these differences differ between palm and African civet beans as well as within the sub-grouping of the two African civets studied.

Fecal examination by scanning electron microscopy of both African civet dung collected from their respective civetries revealed that similar to its close relative the palm civet, these animals have similar dietary patterns, including the consumption of insects, meat (animal) and vegetable matter thus making them both omnivores (Fig. 2).

In order to study further the observed changes occurring at the surface of civet beans (indicated earlier) microscopic examination by scanning electron microscopy was conducted. Scanning electron microscopy did in fact reveal substantial differences between all control beans and those having passed through the two types of civets. In the case of all controls, examination of the beans at 1000 \times (Fig. 3) and 5000 \times magnification (Fig. 4) revealed that their surfaces were substantially rougher in nature than their civet counterpart. The relative smoothness of those beans passing through either the palm civet or African civet could be due to the fact of exfoliation of the outer surface of the bean during passage and parastasis action through the digestive system of the civet. In a similar way all coffee beans passing through the civet's digestive system were observed having surface micro-pitting as observed at 10,000 \times magnification, respectively but not observed in their control counterparts (Fig. 5). This would indicate that the acidic substances and proteolytic enzymes found in the gastric juices of each of the civets were permeating through the endocarp of the coffee cherry and reaching and reacting with the actual bean surface in a similar fashion. This observation together with the observed changes in color may potentially serve as a

Table 1

Physical, chemical and microbiological characterization of Kopi Luwak (Indonesian Palm Civet) coffee beans and Nekemte Civet and Abdela Civet (Ethiopian Civet) coffee beans and respective coffee bean controls

	Kopi Luwak (Indonesian Palm Civet)	Kopi Luwak (control)	Nekemte Civet (Ethiopian Civet)	Nekemte (control)	Abdela Civet (Ethiopian Civet)	Abdela (control)
<i>Proximate analysis (%)</i>						
Moisture	9.2 ^a	11.7 ^c	10.9 ^b	12.0 ^c	11.2 ^b	13.0 ^c
Protein	13.5 ^d	14.5 ^c	12.1 ^b	13.2 ^c	11.4 ^a	12.7 ^c
Fat	13.0 ^c	12.0 ^a	12.5 ^b	12.1 ^a	13.1 ^c	12.5 ^b
Ash	3.6 ^c	3.4 ^b	3.2 ^a	3.4 ^b	3.4 ^b	3.8 ^d
Carbohydrate (by difference)	60.7	58.4	61.3	59.3	63.9	61.0
<i>Seed characteristics</i>						
Color						
<i>L</i> *	37.62 ^a	40.00 ^b	44.66 ^c	60.12 ^c	44.94 ^c	54.3 ^d
<i>a</i> *	6.23 ^f	2.78 ^c	4.10 ^c	2.21 ^a	3.0 ^d	2.57 ^a
<i>b</i> *	20.41 ^f	11.93 ^c	12.92 ^d	6.31 ^a	14.00 ^c	9.00 ^b
Av. bean wt (g)	1.4 ^a	1.6 ^b	1.8 ^c	2.0 ^d	1.6 ^b	1.9 ^{cd}
Av. bean (mm)	12 ^c	12 ^c	9 ^b	8 ^b	7 ^a	7 ^a
<i>Large deformation mechanical rheological properties (N)</i>						
	275 ^d	100 ^a	200 ^b	80 ^a	240 ^c	92 ^a
<i>Minerals (ppm)</i>						
Potassium (K)	15,000 ^a	18,200 ^d	16,500 ^b	19,000 ^c	17,000 ^c	18,100 ^d
Phosphorus (P)	20,00 ^a	24,50 ^d	22,00 ^c	25,00 ^d	21,00 ^b	24,60 ^d
Calcium (Ca)	14,50 ^a	18,00 ^c	15,00 ^b	18,00 ^c	15,50 ^b	18,00 ^c
Magnesium (Mg)	14,00 ^a	17,00 ^b	14,00 ^a	17,50 ^b	14,40 ^a	17,60 ^b
Iron (Fe)	12,00 ^a	15,00 ^c	12,50 ^a	14,50 ^b	13,00 ^a	15,00 ^c
<i>Microbial counts (green)^A</i>						
APC	8.5 × 10 ^{5b}	17 × 10 ^{5a}	8.0 × 10 ^{5b}	1.4 × 10 ^{5a}	7.2 × 10 ^{5b}	1.1 × 10 ^{5a}
Enterics	3.2 × 10 ^{2c}	2.2 × 10 ^{4a}	4.0 × 10 ^{2d}	2.5 × 10 ^{4b}	4.4 × 10 ^{2d}	2.9 × 10 ^{4b}
Coliforms	100 ^a	1.4 × 10 ^{4c}	110 ^a	1.8 × 10 ^{4c}	1.0 × 10 ^{3b}	1.4 × 10 ^{4c}
Mold	5.0 × 10 ^{3c}	1.2 × 10 ^{3a}	4.1 × 10 ^{3d}	1.9 × 10 ^{3d}	3.2 × 10 ^{3c}	2.3 × 10 ^{3b}
<i>Microbial counts (roasted)^A</i>						
APC	5.5 × 10 ^{2a}	2.5 × 10 ^{2b}	1.1 × 10 ^{2a}	2.0 × 10 ^{2a}	1.8 × 10 ^{2a}	2.4 × 10 ^{2b}
Enterics	50 ^a	<100 ^b	<100 ^b	<100 ^b	<100 ^b	<100
Coliforms	ND	ND	ND	ND	ND	ND
Mold	ND	ND	ND	ND	ND	ND

APC, aerobic plate count; Enterics, Enterobacteriaceae count.

Values in each category row with the same letter are not significantly different ($p \geq 0.05$).

ND, not detected.

^A Colony forming units per gram.

Fig. 1. Photograph of (a) Kopi Luwak coffee beans (b) Nekemte-African Civet coffee beans (c) Abdela-African Civet coffee beans.

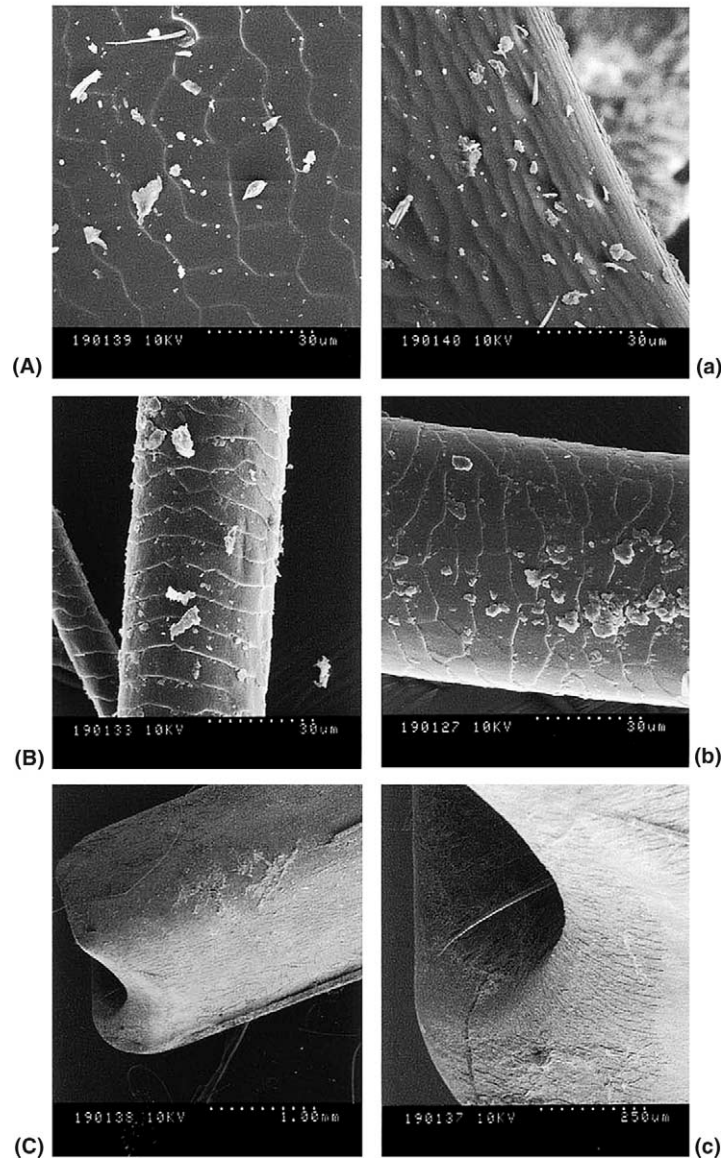


Fig. 2. Scanning electron micrographs of (A and a) insect parts found in African Civet dung from Nekemte (B and b) (B) Rodent hair (b) Civet hair and (C and c) bone material found in African Civet dung from Abdela.

preliminary way of determining the authenticity of civet coffee beans.

Although it would appear that enzymatic action of proteolytic enzymes (i.e., pepsin and trypsin, etc.) was reaching and affecting the surface of the bean more information was sought in order to determine if these compounds were actually penetrating and entering into and modifying the actual bean. If in fact these enzymes were making their way into these beans, breakdown of susceptible storage proteins would be expected and observable. SDS-PAGE in fact confirmed this hypothesis as civet beans finger prints revealed a loss in intensity of “protein bands” which would be indicative of proteolysis (Fig. 6). Although this was found to occur, it appeared that both the acidic (30,000 Da) and basic (20,000 Da)

subunits described by (Marcone, Kakuda, & Yada, 1998), in seeds and beans respectively of the palm civet (Kopi Luwak) beans were equally susceptible to proteolysis whereas only the basic subunit of the African civet showed substantial breakdown. Since the breakdown of the proteins would lead to the exposure of more amino acids (and free amino acids) their reaction through maillard browning on roasting would lead to the production of measurable affects on the aromatic and flavor characteristics of civet coffee compared to their control beans. Since the degree and specificity of proteolysis was observed to be different between the palm civet vis-a-vis the African civets (as indicated above) measurable differences in flavor and aromatic characteristics of the palm vis-a-vis African civet could be expected.

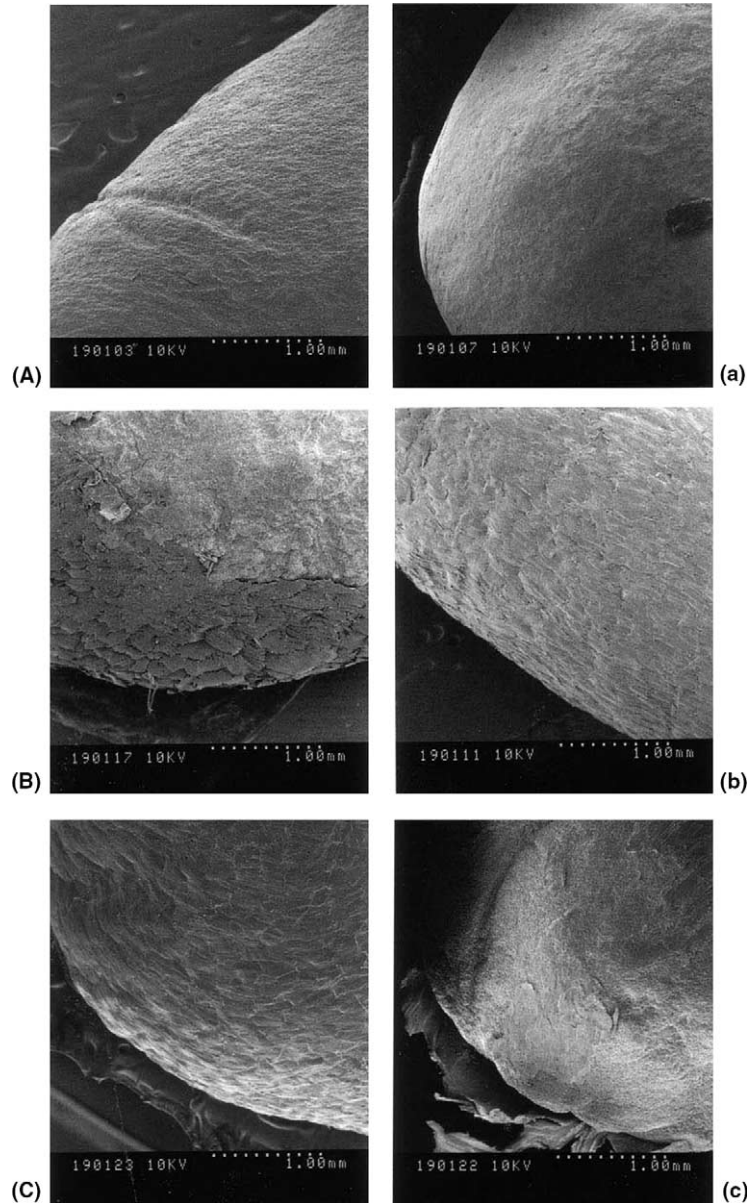


Fig. 3. Scanning electron micrographs of (A) Kopi Luwak control coffee bean (not gone through animal) (a) Kopi Luwak coffee bean (B) Nekemte control coffee bean (b) Nekemte African Civet coffee bean (C) Abdela control coffee bean (c) Abdela African Civet coffee bean.

It is interesting to note that all civet beans were lower in total protein indicating that during digestion not only were proteins being partially broken down but also leached out of the bean. It is interesting to note that the lower levels of proteins would lead to a decrease in the levels of coffee bitterness since proteins serve as the precursor of certain bitter compounds on roasting (Mc Camey, Thorpe, & McCarthy, 1990). This may help to explain some of the perceived flavor differences noted by the consumers between Kopi Luwak and its controls. Examination of the other components in the various beans (including minerals) did not reveal any other substantial difference between

them. As such it would appear that little other modification of the coffee beans occurs as they pass through the civets' GI track.

Microbiological testing (aerobic plate counts – APC) showed that all types of green civet coffee beans were in fact significantly more contaminated than their respective control beans (Table 1) something that would be expected. In the civet bean coffee fewer colony forming types were observed than their respective control beans. Quite surprisingly the Enteric Organism counts were on average two log orders lower for the green civet coffee beans than their controls. In fact, coliform counts were also shown to be significantly

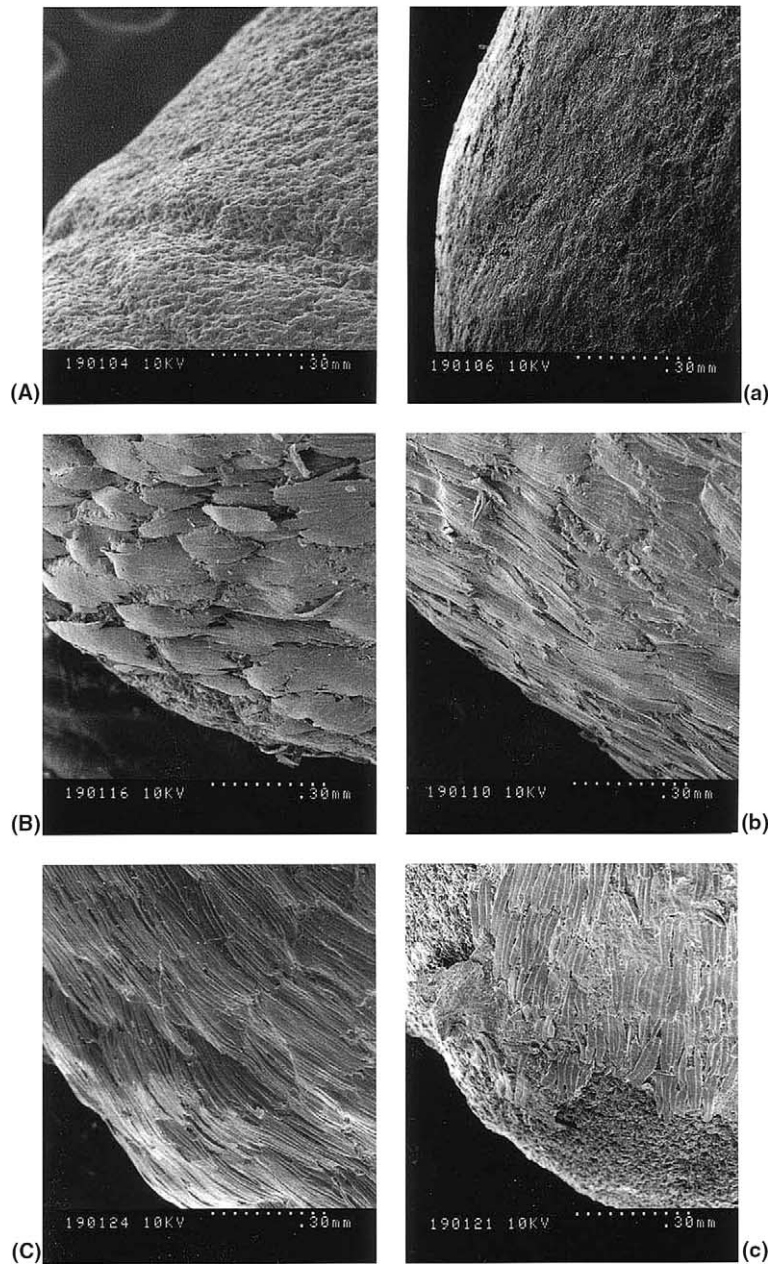


Fig. 4. Scanning electron micrographs of (A) Kopi Luwak control coffee bean (not gone through animal) (a) Kopi Luwak coffee bean (B) Nekemte control coffee bean (b) Nekemte African Civet coffee bean (C) Abdela control coffee bean (c) Abdela African Civet coffee bean.

lower in the civet beans as compare to their respective control. A possible explanation of this observation could be that civet coffee beans are typically extensively washed under running water after collection which would dislodge bacteria and lower their overall counts. These results would agree with the above observation of fewer colony types for civet coffee beans since different bacteria would have different levels of physical attachment to the beans. Gram stain results indicated predominately gram positive rods on the surface of the beans. Other types of microorganism such as molds were also found on both civet and control beans with

significantly more molds being found on the civet beans (this could be due to the slow rate of drying after washing). Upon roasting of all coffee beans reduction of colony counts occurred to near undetectable levels. Use of the electronic nose to measure the aroma profile of roasted ground coffee beans of the three civet coffees and three control coffees indicate good differentiation between samples at a discrimination index of 95% (Table 2). Examination of the distance between clusters on the PCA map indicated that the control sample could be differentiated from one another but do not appear to be very different as a group. On the other

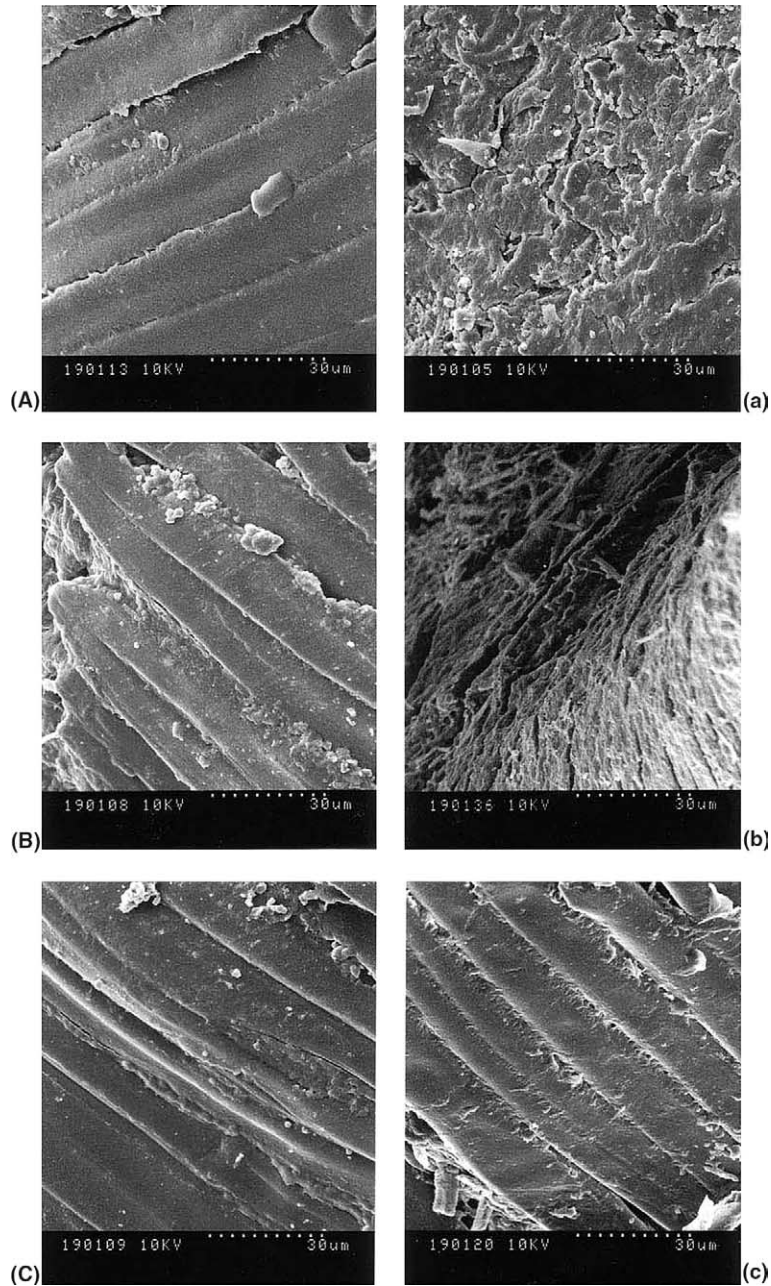


Fig. 5. Scanning electron micrographs of (A) Kopi Luwak control coffee bean (not gone through animal) (b) Kopi Luwak coffee bean (B) Nekemte control coffee bean (b) Nekemte African Civet coffee bean (C) Abdela control coffee bean (c) Abdela African Civet coffee bean.

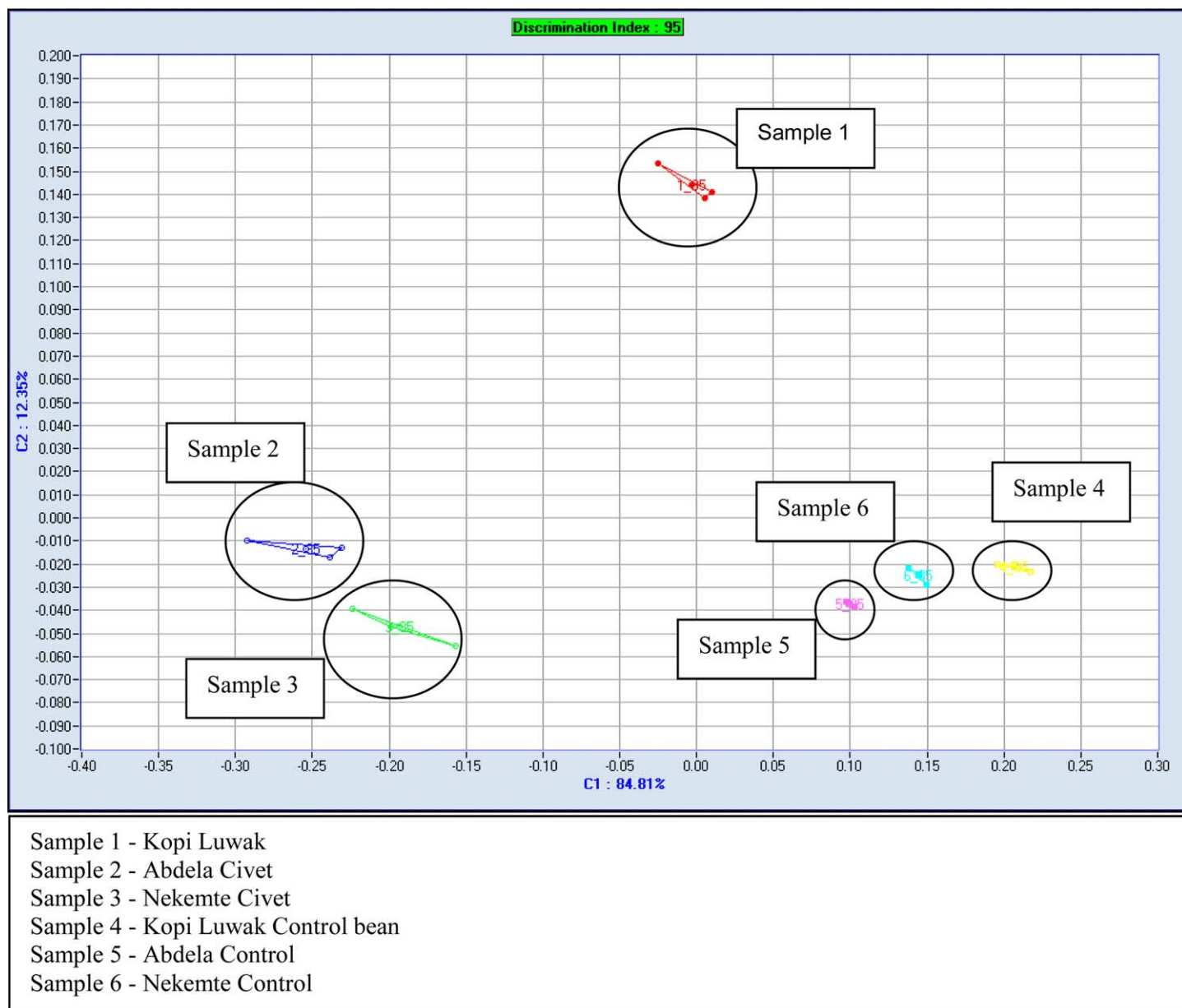
hand Kopi Luwak (Indonesian civet coffee) was the most different from all coffees with the African civets (although being differentiable from one another) appear to be very similar in their overall profiles. It can be concluded at this point that the electronic nose does indicate that aroma/flavor profiles of the palm and African civet coffee beans are affected as they are processed through the civet's GI tract but are modified differently. These results would be in direct agreement with the differences observed in proteolytic protein

finger prints also mentioned earlier which would lead to the production of different maillard browning by products partly responsible for the aroma/flavor profiles of the palm and African and civet coffees.

Coffee cupping results by an experience, certified cupper revealed very little difference in the overall flavor and aromatic attributes of all three control coffee beans except that the palm civet control was slightly lower in body and slightly higher in acidity which correlated well with the electronic nose data obtained. Cupping results

Table 2

PCA map of electronic nose data of ground roasted coffee tested at 35 °C



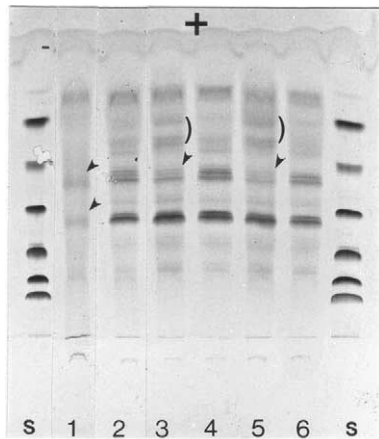


Fig. 6. SDS-PAGE (reducing) (1 μ l of 1 mg⁻¹ protein solutions) applied to Homogenous 20 Phast gels (Pharmacia LKB). (Lane S) standards [α]-lactalbumin, 14,400 Da; Soybean trypsin inhibitor, 20,100 Da; Carbonic anhydrase, 30,000 Da; ovalbumin, 43,000 Da; bovine serum albumin, 67,000 Da; phosphorylase b, 94,000 Da; (Lane 1) Kopi Luwak (Lane 2) Kopi Luwak Control (Lane 3) Nekempt African Civet Coffee bean (Lane 4) Nekempt Control (Lane 5) Abdela African Civet Coffee bean (Lane 6) Abdela Control bean.

also showed that all three civet coffee beans to be differentiable from the actual controls which again agreed well with the electronic nose data. Major differences were noted in the level of acidity and body (being lower) for all civet coffee as compared to their controls. Although the palm civet (Kopi Luwak) coffee was found to be differentiable by the electronic nose procedure from the two African civet coffees, little difference in the overall aroma and flavor profile was noted between them by the certified blinded cupper.

According to several researchers, fermented coffees (i.e., wet processed) have a better overall quality than those prepared by dry-processing (Avallone, Brillouet, Guyot, Olguin, & Guiraud, 2002; Puerta-Quintero, 1999).

In the coffee industry wet processed or fermented coffees are known to be of superior flavor and as such command a much higher price than their dry-processed coffee counterparts. It is interesting to note that when the coffee cherries are processed through the digestive tract of both the palm (Indonesian) and African civet they do indeed undergo a type of wet processing due to acidification in the stomach and then fermentation due to the natural intestinal microflora. In wet processing of coffee, it is important to remove all mucilage which could otherwise lead to secondary fermentation during drying and storage and ultimately lead to the development of flavor defects (Avallone et al., 2002; Woelore, 1993). It is interesting to note that several researchers have found that mucilage degradation seems to be correlated to acidification (similar to what the coffee cherries would experience due to gastric juices in the civets' stomach).

Although researchers have found that microbial growth is necessary, it does not directly participate in mucilage degradation by enzyme production (pectate-lyase – enzymatic pectolysis) but limits off-flavor development due to the production of various organic acids. It is also interesting to note that research shows that lactic acid bacteria are preferred in wet processing systems in order to stay as close as possible to a natural neutral fermentation. It is interesting to note that lactic acid bacteria are a major colonizing bacteria in the digestive tract of civets. It would be interesting to submit that a possible reason for the unique and characteristic flavor of civet coffee could be due to the type of wet process it undergoes in the GI tract of civets.

In closing, it would appear that color analyses coupled with microscopic surface analysis for the presence of micro-pitting on civet coffee beans could be potentially used as a method to authenticate the origins (or process) of civet coffee beans. SDS-PAGE electrophoresis was also shown to be an excellent tool to determine the authenticity of civet coffee beans but also having the added advantage of being able to distinguish between those having gone through the digestive tract of the palm civet *vis-a-vis* those having gone through the African civet.

Electronic nose authentication analyses was also found to be an alternate technique and also being able to distinguish between those from the palm civet *vis-a-vis* those from the African civet.

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