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Phenotypic Identification of the Fungal Flora Associated with Peanut Paste Sold Commercially in Korhogo (Côte d'Ivoire)

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ABSTRACT

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Peanut paste is widely used in the preparation of many traditional dishes consumed by local populations. However, its sanitary quality may be compromised by fungal contamination occurring during processing or storage. This study aimed to determine the fungal flora of peanut paste marketed in the city of Korhogo, Côte d'Ivoire. A total of thirty (30) peanut paste samples, representing two varieties, with fifteen (15) samples collected from each variety, were obtained from two markets in Korhogo. The analyses included the determination of pH and moisture content of the collected samples. In addition, mold counts were determined by culture on agar medium. Finally, the isolated molds were identified phenotypically based on the observation of their macroscopic and microscopic characteristics. The pH values ranged from 6.47 to 6.50, whereas the moisture content varied from 2.63% to 8.50%. Mold counts ranged from 2.01 to 2.60 log₁₀ CFU/g in Virginia peanut paste samples and from 2.43 to 2.68 log₁₀ CFU/g in Valencia peanut paste samples. Three fungal species, namely *Aspergillus flavus*, *Aspergillus niger*, and *Rhizopus* sp., were identified in the analyzed samples. The presence of these mold species may constitute a potential health risk to consumers. These findings highlight the need to strengthen good hygiene, processing, and storage practices in order to improve the sanitary quality of peanut paste marketed in Korhogo.

Introduction

Peanut (*Arachis hypogaea*) is a nutrient-rich legume

containing vitamins A and E, folate, magnesium, zinc, iron, calcium, phosphorus, and dietary fiber. It provides approximately 593 kcal per 100 g, making it an important

source of energy and nutrients that can contribute significantly to meeting human nutritional requirements (Reddy *et al.*, 2010). In addition, several studies have demonstrated that the consumption of peanuts and peanut-based products exerts beneficial effects on brain function and cardiovascular health (Basse, 2020, Çiftçi and Suna, 2022). Global peanut production exceeded 17 million tonnes in 2024. The four leading producers—China, India, Nigeria, and the United States—account for more than two-thirds of the world's total production (FAOSTAT, 2024). In Côte d'Ivoire, peanut production increased by more than 10% annually between 2015 and 2024 as a result of government investments through several agricultural development projects. Consequently, national production rose from 178,800 tonnes in 2015 to more than 363,000 tonnes in 2025, meeting approximately 64.2% of the country's estimated demand. Côte d'Ivoire therefore imports peanuts from neighboring countries, particularly Mali and Burkina Faso, to compensate for the production deficit (Gouv.ci, 2026).

Peanuts are consumed in a wide variety of forms. They are mainly processed into oil, flour, and several value-added products, including peanut butter and peanut paste. Peanut-derived ingredients are widely incorporated into confectionery products such as chocolate, biscuits, and cakes, as well as infant foods and coating or breading materials used in fried products (Arya *et al.*, 2016; Moharana *et al.*, 2020). In peanut-producing regions of Africa, peanuts are primarily consumed either roasted, boiled in the shell, or processed into peanut paste (Compaore *et al.*, 2020).

In Côte d'Ivoire, peanut paste is one of the major processed peanut products. It is widely used as a basic ingredient in the preparation of traditional sauces and is also incorporated into locally produced complementary infant flours to improve their sensory and nutritional quality. Peanut paste production is predominantly carried out by women and remains largely artisanal, with limited mechanization (Boli *et al.*, 2018; Compaore *et al.*, 2020). The production of peanut paste involves several successive processing steps. It begins with the selection of peanut kernels, which are winnowed and sorted to remove foreign materials such as stones, shell fragments, and damaged or spoiled seeds. The kernels are then manually roasted in large pans or in roasting equipment using wood or gas as a heat source. This step is critical because it greatly influences the final quality and color of the peanut paste. Roasting time depends on both the quantity of kernels and the intensity of the heat source.

After roasting, the kernels are spread out and cooled to ambient temperature. Once cooled, they are manually skinned and winnowed again to remove the seed coats, followed by a final sorting step. The roasted and dehulled kernels are then ground into peanut paste using a milling machine (Diakite *et al.*, 2017; Compaore *et al.*, 2020).

Several studies have reported the occurrence of molds belonging to the genera *Aspergillus*, *Penicillium*, *Fusarium*, *Claviceps*, and *Alternaria* in peanut kernels and peanut-derived products (Ahébé *et al.*, 2020; Oswald and Parent-Massin, 2020). Some of these fungi are capable of producing mycotoxins that pose serious health risks (Manizan *et al.*, 2018; Sogbossi *et al.*, 2025). Mycotoxins may persist in food products long after the disappearance of the producing molds and can withstand the high temperatures reached during cooking. Their adverse effects may occur immediately as acute intoxication or develop over the long term following chronic dietary exposure (Boli *et al.*, 2020; Alfriana and Anis, 2023). The artisanal production process and inadequate storage conditions of peanut paste favor microbial contamination, particularly by molds. The presence of mycotoxigenic fungi therefore represents a significant public health concern. However, little information is currently available regarding the microbiological quality of peanut paste marketed in the Korhogo region. This study was therefore undertaken to determine the fungal flora associated with peanut paste sold in the markets of Korhogo, Côte d'Ivoire.

Materials and Methods

Study Area

This study was conducted in the city of Korhogo, Côte d'Ivoire. Located at 9.4580° N and 5.6295° W, Korhogo is the largest city in northern Côte d'Ivoire and one of the country's major peanut-producing areas (Figure 1).

Biological Material

The biological material consisted of peanut paste prepared from the Virginia and Valencia peanut varieties and marketed in the markets of Korhogo (Figure 2).

Methods

Sampling

Peanut paste samples were collected from the main

market located in the « Soba » district and from the « Belleville » market in Korhogo, Côte d'Ivoire. The « Soba » market is the city's largest commercial market, whereas the « Belleville » market hosts a peanut paste production unit. These characteristics motivated the selection of the two sampling sites. A preliminary survey was conducted to identify peanut paste sellers in both markets.

Five sellers were then randomly selected from each market for sample collection. From each seller, three samples of each peanut paste variety (Virginia and Valencia) were collected. In total, thirty (30) samples were obtained, comprising fifteen (15) samples from each peanut variety. The peanut paste was packaged in plastic containers of approximately 250 g. Each sample was appropriately labeled and transported to the laboratory in an insulated cooler containing ice packs to maintain sample integrity until analysis.

Determination of pH and Moisture Content

The pH of the peanut paste samples was determined according to the method described by Compaore (2020). Briefly, 10 g of peanut paste was homogenized with 90 mL of distilled water. The resulting mixture was stirred for 30 min and allowed to stand for a few minutes.

The pH was then measured using a calibrated pH meter (Hanna Instruments), previously standardized with pH 4.0 and pH 7.0 buffer solutions. The electrode was immersed in the supernatant, and the pH value was recorded directly from the instrument display.

Moisture content was determined by the oven-drying method according to AOAC Official Method No. 925.40. Briefly, 5 g of each sample were weighed (M_1) into a crucible. The mass of the crucible containing the sample was recorded as M_2 .

The crucible was then dried in a hot-air oven at 105°C and weighed after cooling in a moisture-free environment at 1-h intervals until a constant weight (M_3) was obtained. The moisture content (%) was calculated by the gravimetric method using the equation described by Otmane *et al.*, (2022).

$$TH = \frac{M_2(g) - M_3(g)}{M_1(g)} \times 100$$

M_1 : mass of the fresh sample; M_2 : mass of the sample plus crucible; M_3 : mass of the dried sample.

Microbiological Analyses

Preparation of Culture Media

The culture media were reconstituted according to the manufacturer's instructions provided on the media containers.

Preparation of the Initial Suspension and Decimal Dilutions

The preparation of the initial suspension and the serial decimal dilutions was carried out according to ISO 6887-1 standard. Briefly, five grams (5 g) of each peanut paste sample were aseptically collected and homogenized in a Stomacher bag containing 45 mL of buffered peptone water. The suspension obtained after manual homogenization represented the initial suspension corresponding to the 10^{-1} dilution. A tenfold decimal dilution (1:10) was then prepared from the initial suspension to obtain the 10^{-2} dilution.

Inoculation and Enumeration of Molds

Oxytetracycline-Glucose-Yeast Extract (OGYE) agar was used for the enumeration of fungal flora. Inoculation was performed by the surface-spreading method according to the NF V08-059 standard. For this purpose, 0.1 mL of each decimal dilution from each sample was spread onto the surface of previously prepared OGYE agar plates. The inoculated plates were then incubated at 30°C for 3 to 5 days. Colony counting was performed on plates containing fewer than 150 colonies. The results were expressed according to Formula 2.

$$N = \sum C/V.d (n_1+0,1n_2)$$

ΣC : sum of the colonies counted on all plates from the two successive dilutions; V : volume of inoculum applied to each plate, expressed in milliliters (mL); n_1 and n_2 : number of plates retained for the first and second selected dilutions, respectively; d : dilution factor corresponding to the first dilution showing countable colonies (lowest dilution).

Purification and Identification of Fungi

The purification of fungal strains was performed to

obtain colonies composed of genetically and morphologically homogeneous cells. A well-isolated colony with characteristic morphology was aseptically collected using sterile forceps previously sterilized in a Bunsen burner flame and then transferred onto a new Petri dish containing OGYE agar. After incubation for 48 to 72 h, macroscopic and microscopic examinations were carried out. The macroscopic examination was performed by observing both the upper and reverse surfaces of Petri dishes containing purified mold strains.

The observations focused on pigmentation, growth rate, colony appearance, and texture. For microscopic examination, a fragment of the fungal strain was collected and placed on a microscope slide. After adding a drop of methylene blue stain, the preparation was covered with a coverslip and observed under a microscope at $\times 10$ and $\times 40$ magnifications. Microscopic observations included the morphology of the mycelium, the shape of conidia, conidial heads, and other characteristic structures. Fungal species identification was carried out using the identification keys proposed by [Talaiekhosani and Mohanadoss \(2015\)](#), combined with the observed morphological characteristics.

Determination of Isolation Frequency and Fungal Contamination Rate

According to [Alfrian and Anis \(2023\)](#), the isolation frequency (IF) was calculated using formula 3. The fungal contamination rate (FCR) was determined using formula 4, as described by [Compaore et al., \(2020\)](#).

$$FI = (NF/NT) \times 100$$

NF: total number of samples from which a given fungus was isolated; NT: total number of samples from which fungal isolations were performed.

$$TC = (NI/NT) \times 100$$

NI: number of fungal genera isolated from a sample; NT: total number of fungal genera isolated from all samples.

Statistical Analysis

IBM SPSS Statistics software version 20 was used for the statistical analysis of microbiological and physicochemical data. Differences in the distribution of physicochemical and microbiological parameters

between groups were assessed using the one-way Kruskal–Wallis ANOVA test at a significance level of 5% ($p < 0.05$). Principal component analysis (PCA) was performed using the ade4TkGUI software.

Results and Discussion

pH and Moisture Content of Samples

The pH values of the samples were slightly acidic, ranging from 6.47 to 6.51. Only two samples (Evi5 and Eva2) exhibited pH values slightly above the optimal range (6.0–6.5). No statistically significant difference was observed among the pH values of the different samples at the 5% significance level (Table I).

The moisture content of the samples ranged from 2.63% to 8.50%, with an average value of 5.5% for EVI samples, and from 2.3% to 7.5%, with an average value of 4.5% for EVA samples. The moisture content of the samples was higher than the optimal recommended range (0.5–2%). A statistically significant difference at the 5% significance level was observed among the moisture contents of samples belonging to the same type of peanut paste (Table I).

Results of Microbiological Analyses

The results of fungal flora enumeration showed that all analyzed peanut paste samples were contaminated with molds. Mold counts ranged from 2.01 to 2.60 $\log_{10}$ CFU/g for Virginia variety peanut paste and from 2.43 to 2.68 $\log_{10}$ CFU/g for Valencia variety peanut paste. Sample EVA5 exhibited the highest mold load (2.68 $\log_{10}$ CFU/g), whereas the lowest count was recorded in sample EVI4 (2.01 $\log_{10}$ CFU/g). Overall, peanut pastes prepared from the Valencia variety showed slightly higher mold loads than those produced from the Virginia variety. These findings indicate widespread fungal contamination of marketed peanut pastes, with varying contamination levels among samples (Table II).

Macroscopic and microscopic observations of the fungal isolates allowed the identification of three mold species, namely *Aspergillus flavus*, *Aspergillus niger*, and *Rhizopus* sp., according to the identification keys described by [Talaiekhosani and Mohanadoss \(2015\)](#). The isolates of *A. flavus* exhibited yellow-green to olive-green colonies with a powdery or granular texture. Microscopically, they were characterized by hyaline

conidiophores terminating in globose vesicles bearing phialides and spherical to subglobose conidia with rough walls. The colonies of *A. niger* were dark brown to black in color, with a velvety to powdery texture. Microscopic examination revealed long black conidial heads, globose vesicles completely covered with metulae and biseriate phialides, as well as highly pigmented spherical conidia. The isolates of *Rhizopus* sp. formed rapidly growing, cottony colonies. Microscopically, they displayed long sporangiophores bearing globose sporangia, spherical sporangiospores, and well-developed rhizoids at the base of the sporangiophores.

All these morphological characteristics were consistent with the descriptions reported in the literature and confirmed the identification of the isolated fungal species (Talaiekhosani and Mohanadoss, 2015) (Table III).

Isolation Frequencies

Aspergillus flavus and *Aspergillus niger* were isolated from both peanut paste varieties. *Rhizopus* sp. was detected only in peanut paste samples prepared from the Valencia variety. The isolation frequencies of *A. flavus* were 60% and 20% in Virginia and Valencia peanut paste samples, respectively. The isolation frequency of *A. niger* was 40% in both types of peanut paste. Regarding *Rhizopus* sp., its isolation frequency was 40% (Table IV).

Contamination Rate

The contamination rate was 100% for all Virginia peanut paste samples. Valencia peanut paste samples EVA1 to EVA3 showed a contamination rate of 100%, whereas samples EVA4 and EVA5 exhibited a contamination rate of 50% (Table V).

The pH values of the different peanut pastes (6.47–6.51) indicate that this product is slightly acidic. These results are consistent with those reported by Boli *et al.*, (2018) and Compaoré *et al.*, (2020), who recorded pH values ranging from 6.2 to 6.7 in traditionally produced peanut pastes. These pH values are favorable for the growth of several microorganisms, particularly yeasts and molds, unlike highly acidic foods in which low pH naturally limits microbial development (Roberts *et al.*, 2005). The absence of significant differences among samples suggests that the processing methods and raw materials used were relatively similar across the different supply sites.

In contrast, the moisture contents recorded (2.30–8.50%) were considerably higher than the values generally recommended for peanut paste, which should remain below 2% to ensure good microbiological stability (Meneely *et al.*, 2022). This relatively high moisture content may result from insufficient drying of peanut kernels, incomplete roasting, moisture absorption during cooling, or poor storage and marketing conditions. Several authors have demonstrated that increased water content promotes mold growth and enhances the risk of mycotoxin production in peanut-derived products (Manizan *et al.*, 2018).

All analyzed peanut paste samples were contaminated with molds, indicating suboptimal microbiological quality. Similar findings were reported by Boli *et al.*, (2020) in peanut pastes marketed in Côte d'Ivoire.

The presence of molds in these samples is concerning because some species are capable of producing mycotoxins. These toxic metabolites may persist in food matrices even after the disappearance of the producing molds (Schrenk *et al.*, 2020, Lare *et al.*, 2021).

The identified species mainly belonged to the genera *Aspergillus* and *Rhizopus*. The predominance of *Aspergillus flavus* and *Aspergillus niger* has frequently been reported in peanut kernels, peanut pastes, and other oilseed products marketed in tropical regions (Manizan *et al.*, 2018). High temperatures and humidity levels characteristic of tropical environments favor the development of these molds.

The high isolation frequency of *A. flavus* is of particular concern. This species is recognized as the main producer of aflatoxins B1, B2, G1, and G2, with aflatoxin B1 classified as carcinogenic to humans by the International Agency for Research on Cancer (Syraji *et al.*, 2025). The presence of *A. niger* also represents a potential health concern, as some strains are able to produce ochratoxins, although this ability is less common (Laré *et al.*, 2021).

The isolation of *Rhizopus* sp. exclusively from peanut pastes prepared from the Valencia variety may reflect differences in processing conditions, storage practices, or raw material sources rather than a true varietal effect. Species belonging to the genus *Rhizopus* are generally considered post-harvest contaminants responsible for organoleptic deterioration, but they are not regarded as major mycotoxin producers (Samson *et al.*, 2019).

Table.1 pH and moisture content of peanut paste samples

Samples	pH	Optimal value	TH (%)	Optimal rate
Evi ₁	6,48±0,05 ^a	6 – 6,5	8,5±0,52 ^c	0,5 % et 2 %
Evi ₂	6,5±0,03 ^a		6,7±0,52 ^{de}	
Evi ₃	6,47±0,01 ^a		2,63±0,11 ^a	
Evi ₄	6,5±0,01 ^a		4,57±0,56 ^{bc}	
Evi ₅	6,51±0,02 ^a		5,33±0,57 ^{cd}	
Mean	6,5		5,55	
Eva ₁	6,5±0,02 ^a	6 – 6,5	2,3±0,2 ^a	0,5 % et 2 %
Eva ₂	6,51±0,01 ^a		5,87±0,47 ^{cde}	
Eva ₃	6,47±0,05 ^a		4,23±0,41 ^{abc}	
Eva ₄	6,49±0,03 ^a		2,77±0,61 ^{ab}	
Eva ₅	6,48±0,01 ^a		7,5±1,2 ^{de}	
Mean	6,49		4,5	

EVI: Virginia variety sample; **EVA:** Valencia variety sample.

Within the same column, mean values followed by the same letter are not significantly different at the 5% significance level.

Table.2 Mold counts in peanut paste samples

	Samples	Mold Contamination (log ₁₀ ufc/g)
Paste obtained from Virginia variety peanuts	Evi ₁	2,6±0,001 ^c
	Evi ₂	2,55±0,03 ^{de}
	Evi ₃	2,30±0,001 ^b
	Evi ₄	2,01±0,02 ^a
	Evi ₅	2,57±0,006 ^{de}
Paste obtained from Valencia variety peanuts	Eva ₁	2,43±0,04 ^{bc}
	Eva ₂	2,5±0,01 ^{cd}
	Eva ₃	2,43±0,03 ^{bc}
	Eva ₄	2,44±0,02 ^{bc}
	Eva ₅	2,68±0,02 ^f

Within the same column, mean values followed by the same letter are not significantly different at the 5% significance level.

Figure.1 Location of the Study Area (Korhogo Department) in Côte d'Ivoire

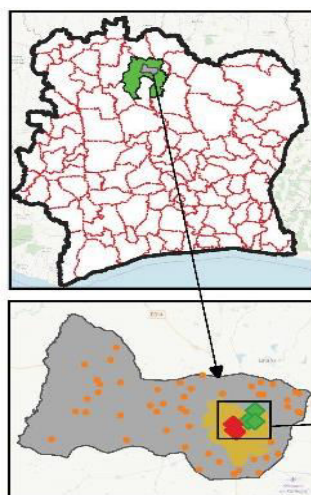


Table.3 Identification of molds

Macroscopic observation		Microscopic observation	Species
Front	Back		
			<i>Aspergillus flavus</i>
			<i>Aspergillus niger</i>
			<i>Rhizopus</i> sp.

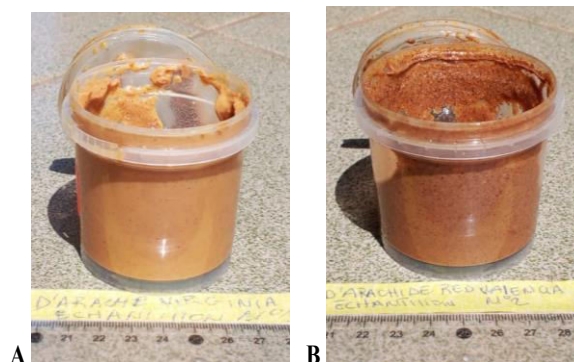
Table.4 Isolation frequencies of the isolated molds

Species	Frequency of isolation (%)		
	Evi (N=15)	FI Eva B (N=15)	Mean
<i>Aspergillus flavus</i>	60 (9)	20 (3)	40
<i>Aspergillus niger</i>	40 (6)	40 (6)	40
<i>Rhizopus</i> sp.	ND	40 (6)	20

Table.5 Mold Contamination Rates in Samples by Month

Samples	Fungal contamination rate (%)	
	Virginia variety	Valencia variety
E ₁	100	50
E ₂	100	50
E ₃	100	50
E ₄	100	100
E ₅	100	100

Figure.2 Photographs of Peanut Paste Samples: (A) Virginia Variety; (B) Valencia Variety



The high isolation frequencies of *Aspergillus flavus* (40%) and *A. niger* (40%) confirm that these two species constitute the dominant fungal flora of peanut pastes marketed in Korhogo. Similar observations have been reported in Benin, Burkina Faso, and Côte d'Ivoire, where *Aspergillus* species largely dominate the fungal flora of peanut-derived products (Manizan *et al.*, 2018; Sogbossi *et al.*, 2025). This predominance is generally attributed to traditional drying practices, storage conditions, and prolonged exposure of products in open markets (N'pagyendou *et al.*, 2022).

Overall, this study highlights widespread fungal contamination of peanut pastes marketed in Korhogo. Although the observed microbial loads remain relatively moderate, the recurrent presence of potentially toxigenic species, particularly *Aspergillus flavus*, represents a significant health concern. These findings emphasize the need to improve drying, roasting, processing, packaging, and storage practices in order to limit mold development and potential mycotoxin contamination of peanut pastes.

The limitations of this study include the molecular identification of molds and the detection of mycotoxins in the analyzed samples. However, we plan to expand the study to a larger area and conduct more in-depth analyses.

This study enabled the identification of molds contaminating peanut pastes marketed in Korhogo. The analyses revealed contamination of all samples, with the identification of *Aspergillus flavus*, *Aspergillus niger*, and *Rhizopus* sp. The pH values, which were close to neutrality, combined with moisture contents generally exceeding recommended levels, provided favorable conditions for mold development. The predominance of species belonging to the genus *Aspergillus*, which are

recognized for their toxigenic potential, highlights the potential risk of mycotoxin contamination in peanut pastes and, consequently, a possible health risk for consumers. These findings emphasize the need to strengthen good manufacturing practices, particularly during drying, roasting, processing, packaging, and storage of peanut pastes, in order to reduce fungal contamination.

Further studies focusing on the determination of major mycotoxins, especially aflatoxins, as well as the molecular characterization of the isolated strains, are required to better assess the health risk associated with the consumption of this product.

Author Contributions

TRAORE Moumouny: Conceptualization, Formal Analysis, Methodology, Validation, Visualization, Writing – original draft; KOUAME N'zebo Desire: Analysis and interpretation of data, - writing the article; BOSSON Ya Stephanie Roxane: Investigation, Formal analysis. YORO Thierry Dezay: Investigation. - proofreading and editing the manuscript; KAMBIRE Olo: final approval of the version to be published.

Data Availability

The datasets generated during and/or analyzed during the current study are available from the corresponding author on reasonable request.

Declarations

Ethical Approval Not applicable.

Consent to Participate Not applicable.

Consent to Publish Not applicable.

Conflict of Interest The authors declare no competing interests.

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