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Isolation and identification of *Aspergillus* species isolated from different soil environments in Jos Metropolis, Plateau State, Nigeria

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ABSTRACT

Aspergillus species are among the most ubiquitous filamentous soil fungi and play significant ecological, industrial, and medical roles. However, some species are opportunistic pathogens and producers of harmful mycotoxins, making their occurrence in environmental soils a public health concern. This study aimed to isolate and identify *Aspergillus* species from different soil environments within Jos Metropolis, Plateau State, Nigeria, and to determine their distribution across selected anthropogenic habitats. A total of 30 soil samples were collected in triplicate from ten sampling locations representing gardens, cultivated lands, mechanic workshops, industrial waste sites, industrial effluent areas, abattoirs, and vegetable oil-contaminated soils. Fungal isolation was carried out using the serial dilution pour-plate technique on Potato Dextrose Agar supplemented with chloramphenicol. Pure cultures were obtained through repeated subculturing, while identification was based on cultural and microscopic characteristics using standard taxonomic keys. Thirty-six *Aspergillus* isolates representing four species were recovered. *Aspergillus niger* was the predominant species, accounting for 15 (41.7%) isolates, followed by *A. fumigatus* with 13 (36.1%), *A. flavus* with 7 (19.4%), and *A. nidulans* with 1 (2.8%). Cultivated soils and industrial effluent sites recorded the highest proportion of isolates (19.4% each), whereas mechanic workshop and oil-contaminated soils yielded comparatively fewer isolates. The predominance of *A. niger* suggests its superior ecological adaptability and ability to thrive in soils enriched with organic matter, while the occurrence of *A. fumigatus* and *A. flavus* highlights the potential public health risks associated with exposure to opportunistic and mycotoxin-producing fungi. The study concludes that soils from different environments within Jos Metropolis constitute important reservoirs of diverse Aspergillus species, with their distribution largely influenced by environmental conditions and anthropogenic activities. Continuous environmental surveillance and the incorporation of molecular identification techniques are recommended to improve species characterization and support environmental, agricultural, and public health management.

Keywords: *Aspergillus* species, soil fungi, *Aspergillus niger*, fungal isolation, Jos Metropolis, environmental microbiology.

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INTRODUCTION

Fungi constitute one of the most diverse groups of microorganisms in nature and play indispensable roles in nutrient recycling, decomposition of organic matter, and maintenance of ecosystem stability (Tortora et al., 2021). Among filamentous fungi, the genus *Aspergillus* is one of the most widespread and economically important, owing to its occurrence in soil, air, water, decaying vegetation, stored agricultural products, and indoor environments. Soil serves as a principal ecological reservoir for *Aspergillus* species because it provides abundant organic substrates, suitable moisture conditions, and favorable temperatures for fungal growth and sporulation. The remarkable adaptability of *Aspergillus* species enables them to survive under diverse environmental conditions,

including variations in pH, temperature, oxygen availability, and water activity, making them dominant members of many soil fungal communities. Beyond their ecological significance, these fungi are recognized for producing numerous extracellular enzymes and secondary metabolites that contribute to the decomposition of plant residues and nutrient cycling within terrestrial ecosystems (Deacon, 2006; Dighton, 2016). Consequently, understanding the diversity and distribution of *Aspergillus* species in soil has become an important aspect of environmental microbiology and fungal ecology.

Several species of *Aspergillus*, including *A. niger*, *A. flavus*, *A. fumigatus*, *A. terreus*, and *A. nidulans*, possess considerable agricultural, industrial, medical, and environmental importance. *Aspergillus niger* is widely utilized in the industrial production of citric acid and numerous commercial enzymes, whereas *A. oryzae* is employed in the fermentation of traditional foods and beverages (Bennett, 2010). Conversely, *A. flavus* is a well-known producer of aflatoxins, potent carcinogenic mycotoxins capable of contaminating cereals, legumes, and nuts, thereby posing serious food safety and public health concerns. Likewise, *A. fumigatus* is one of the leading opportunistic fungal pathogens responsible for aspergillosis, particularly among immunocompromised individuals, while increasing reports of antifungal resistance have heightened global concern regarding this species. The World Health Organization (WHO) has recognized several *Aspergillus* species as priority fungal pathogens because of their increasing clinical significance and impact on human health (WHO, 2022). Therefore, accurate isolation and identification of *Aspergillus* species from environmental sources remain essential for disease surveillance, food safety, environmental monitoring, and industrial applications.

Soil fungal diversity is greatly influenced by environmental variables such as vegetation type, organic matter content, soil texture, moisture, altitude, land-use practices, climatic conditions, and anthropogenic activities (Paul, 2015). These factors determine not only the abundance of fungal populations but also the composition of different *Aspergillus* species inhabiting a particular environment. Jos Metropolis, located on the Jos Plateau of north-central Nigeria, possesses a unique highland climate characterized by relatively low temperatures, moderate rainfall, and diverse vegetation compared with many other regions of the country. These ecological conditions, coupled with rapid urbanization, agricultural activities, waste disposal practices, and increasing industrial development, create favorable ecological niches for the proliferation of various soil fungi, including species of *Aspergillus*. Despite the ecological and public health importance of these fungi, information regarding their occurrence, diversity, and distribution in soils from different locations within Jos Metropolis remains relatively limited. This knowledge gap underscores the need for systematic studies aimed at documenting the *Aspergillus* species present in different soil environments and evaluating their distribution patterns.

The isolation and identification of *Aspergillus* species traditionally involve culture-based techniques using selective fungal media, followed by characterization based on colonial morphology, microscopic structures such as conidiophores, vesicles, phialides, and conidia, and, where available, molecular identification methods for improved taxonomic accuracy (Samson et al., 2014). Accurate species identification provides valuable information for ecological studies, environmental monitoring, industrial biotechnology, agricultural disease management, and public health surveillance. Furthermore, identifying soil-borne *Aspergillus* species contributes to understanding potential reservoirs of opportunistic pathogens and mycotoxin-producing fungi, thereby facilitating appropriate preventive and control measures. Therefore, this study aimed to isolate and identify *Aspergillus* species from different soil samples collected within Jos Metropolis, Plateau State, Nigeria, and to evaluate their distribution among selected anthropogenic environments.

MATERIALS AND METHODS

Study area and soil sample collection

Soil samples were collected from 10 different sampling locations within Jos Metropolis, Plateau State, Nigeria. The selected sites represented different anthropogenic environments, including gardens, automobile mechanic workshops, industrial areas, abattoirs, and palm oil-selling areas, to ensure broad representation of soil ecosystems and the diversity of *Aspergillus* species associated with different soil environments. At each sampling location, three independent soil samples (triplicates) were collected, giving a total of 30 soil samples.

Prior to sampling, surface debris, stones, leaves, and other extraneous materials were carefully removed from the soil surface. Using sterile disposable gloves and sterile sampling tools, soil was aseptically collected from a depth of approximately 3–5 cm, where fungal propagules are more abundant and less susceptible to transient environmental contamination. Approximately 100 g of soil was transferred into sterile polyethylene bags, appropriately labeled according to the sampling site, and transported immediately to the Microbiology Laboratory for mycological analysis. All samples were processed within 24 h of collection to minimize changes in the indigenous fungal population.

Isolation of *Aspergillus* species

Isolation of *Aspergillus* species was performed using the

serial dilution plate technique as described by Raja et al. (2017), with slight modifications. Briefly, 1.0 g of each soil sample was aseptically transferred into a sterile test tube containing 9.0 mL of sterile distilled water to obtain a 10^{-1} dilution. The suspension was vigorously vortexed for 2–3 min to ensure uniform dispersion of fungal spores and mycelial fragments. Ten-fold serial dilutions were subsequently prepared up to 10^{-3}.

Thereafter, 0.5 mL aliquots from the appropriate dilution were aseptically inoculated onto sterile Potato Dextrose Agar (PDA) plates using the pour-plate technique. The inoculum was evenly distributed by gently swirling the Petri dishes before the medium solidified. To minimize bacterial contamination, the PDA medium was supplemented with chloramphenicol (50 mg/L) prior to inoculation. The inoculated plates were incubated aerobically at 30 ± 2°C for 5 days, after which emerging fungal colonies exhibiting morphological characteristics typical of *Aspergillus* species were selected for further purification and identification.

Purification of fungal isolates

Distinct fungal colonies presumptively identified as *Aspergillus* spp. were purified by repeated subculturing onto freshly prepared Potato Dextrose Agar (PDA) plates using the hyphal-tip technique. Individual colonies were aseptically transferred onto fresh media and incubated at 30 ± 2°C for 3-5 days. Successive subculturing was carried out until morphologically uniform colonies free from contamination were obtained. Pure cultures were subsequently maintained on PDA slants at 4°C for further morphological characterization and identification.

Identification of *Aspergillus* species

Identification of the purified fungal isolates was based on their macroscopic and microscopic characteristics following the taxonomic keys of Raja et al. (2017), Samson et al. (2014), and Pitt and Hocking (2009). Macroscopic examination included assessment of colony morphology, including colony diameter, texture, pigmentation, topography, margin characteristics, and pigmentation of the reverse surface after incubation on Potato Dextrose Agar.

Microscopic characterization was performed using the lactophenol cotton blue staining technique. A small portion of the fungal mycelium was mounted on a clean glass slide containing a drop of lactophenol cotton blue stain, covered with a coverslip, and examined under a compound light microscope using the ×100 objective lens. Identification was based on characteristic microscopic structures, including the conidiophores, vesicles, phialides, and conidia, and the observed features were compared with those described in the relevant taxonomic keys (Samson et al., 2014; Pitt and Hocking, 2009).

RESULTS AND DISCUSSION

The distribution and frequency of *Aspergillus* species isolated from different soil environments are presented in Tables 1 and 2 and Figure 1. The findings of this study revealed that soils collected from different environmental settings within Jos Metropolis harboured diverse *Aspergillus* species, with *Aspergillus niger* being the predominant isolate, accounting for 15 (41.7%) of the total isolates, followed by *Aspergillus fumigatus* (13; 36.1%), *Aspergillus flavus* (7; 19.4%), and *Aspergillus nidulans* (1; 2.8%).

The predominance of *A. niger* corroborates previous studies that identified this species as one of the most ubiquitous saprophytic fungi in soil because of its exceptional ecological adaptability, efficient spore production, and ability to utilize a wide range of organic substrates (Samson et al., 2014; Klich, 2002). The high frequency of *A. niger* in cultivated soils, abattoir soils, and industrial effluent sites suggests that environments enriched with decomposing organic matter and anthropogenic wastes provide favourable conditions for its proliferation.

Table 1. Distribution and percentage occurrence of *Aspergillus* species isolated from different soil environments in Jos Metropolis.

Sampling Site	No. of Samples	*A. niger* n (%)	*A. fumigatus* n (%)	*A. flavus* n (%)	*A.nidulans* n (%)	Total Isolates n (%)
Abattoir soil (AE)	3	3 (100)	2 (66.7)	1 (33.3)	0	6 (16.7)
Cultivated soil (HG)	3	3 (100)	2 (66.7)	2 (66.7)	0	7 (19.4)
Garden soil (MG)	3	1 (33.3)	1 (33.3)	0	1 (33.3)	3 (8.3)
Industrial waste (IA)	3	2 (66.7)	2 (66.7)	1 (33.3)	0	5 (13.9)
Industrial effluent (ND)	3	3 (100)	3 (100)	1 (33.3)	0	7 (19.4)
Mechanic workshop 1 (KM1)	3	0	1 (33.3)	1 (33.3)	0	2 (5.6)
Mechanic workshop 2 (KM2)	3	2 (66.7)	0	1 (33.3)	0	3 (8.3)
Mechanic workshop 3 (KM3)	3	0	1 (33.3)	0	0	1 (2.8)
Vegetable oil soil (KO)	3	1 (33.3)	0	0	0	1 (2.8)
Terminus oil soil (TO)	3	0	1 (33.3)	0	0	1 (2.8)
Total	30	15 (41.7)	13 (36.1)	7 (19.4)	1 (2.8)	36 (100)

Table 2. Overall frequency of *Aspergillus* species isolated from soil samples.

Species	Number of Isolates	Percentage (%)
Aspergillus niger	15	41.7
Aspergillus fumigatus	13	36.1
Aspergillus flavus	7	19.4
Aspergillus nidulans	1	2.8
Total	36	100

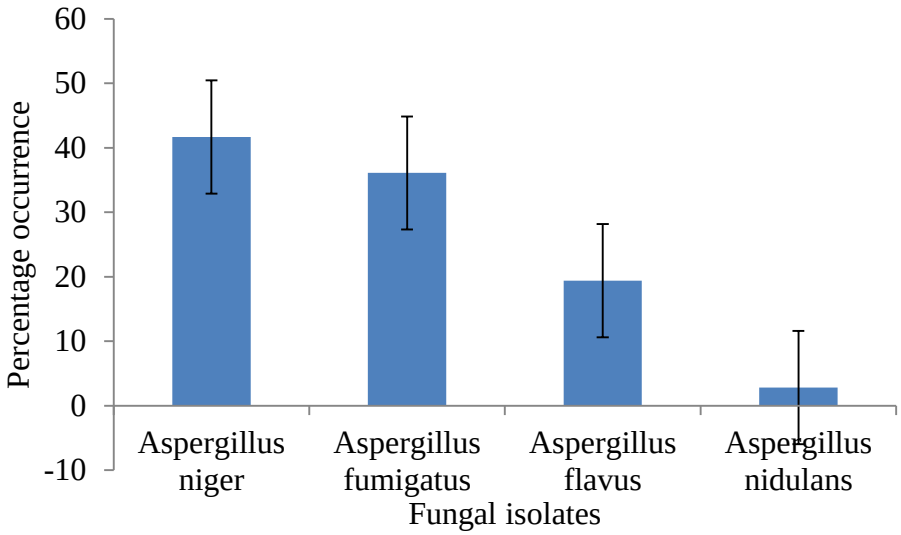


Figure 1. Overall frequency of *Aspergillus* species isolated from soil samples.

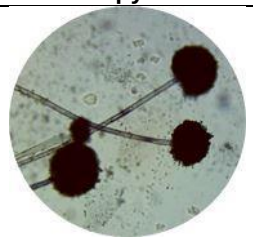
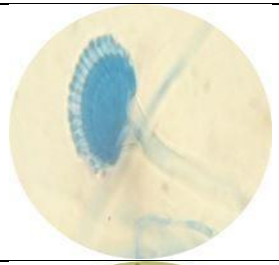
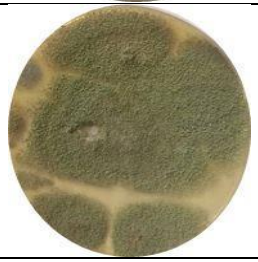
Similarly, the widespread occurrence of *A. fumigatus*, particularly in industrial effluent and organically contaminated soils, may be associated with its thermotolerant nature and ability to colonize composting organic materials, animal wastes, and decaying vegetation (Latgé and Chamilos, 2019).

The relatively lower occurrence of *A. flavus* agrees with reports by Pitt and Hocking (2009), who noted that although *A. flavus* is a common soil inhabitant, its distribution is often influenced by climatic conditions, soil moisture, nutrient availability, and competition with other fungal species. The isolation of *A. nidulans* from only one garden soil sample indicates its comparatively low prevalence in the study area, possibly reflecting its narrower ecological niche compared with the more dominant *Aspergillus* species.

Furthermore, the highest proportions of fungal isolates were recovered from cultivated soils and industrial effluent sites (19.4% each), emphasizing the influence of organic enrichment and anthropogenic activities on soil fungal diversity. In contrast, the comparatively lower recovery of isolates from mechanic workshop and oil-contaminated soils may be associated with the inhibitory effects of petroleum hydrocarbons and heavy metals, which can suppress the growth of susceptible fungal species while selecting for more tolerant microorganisms (Atlas and Bartha, 1998).

The cultural and microscopic characteristics observed in Table 3 confirmed the identification of the isolated *Aspergillus* species based on their distinct morphological features. *Aspergillus niger* was characterized by rapid growth, white colonies that developed into black, dense powdery conidial heads, with septate hyphae and globose vesicles bearing radiating chains of conidia. *A. fumigatus* exhibited blue-green, suede-like colonies with short conidiophores, conical terminal vesicles and globose to subglobose conidia, while *A. flavus* produced yellow-green to olive colonies with granular texture and rough conidiophores terminating in globose vesicles. These cultural and microscopic features are consistent with the characteristic morphological descriptions commonly used for the identification of *Aspergillus* species, demonstrating that colony colour, texture, conidiophore structure, vesicle shape and arrangement of conidia are useful diagnostic characteristics for differentiating the isolates. Overall, these findings demonstrate that soil characteristics, the degree of organic matter accumulation, and anthropogenic activities influence the occurrence and distribution of *Aspergillus* species within Jos Metropolis. The findings further

Table 3. Cultural and microscopic features of *Aspergillus* species isolated from different soil environments in Jos Metropolis.

Isolates	Cultural characteristics	Morphology	Macroscopic	Microscopy ×100	Organism
1	Colonies showed rapid growth, initially white becoming black with dense powdery conidial heads; texture granular; reverse pale yellow	Septate hyphae; erect conidiophores with globose vesicles bearing radiating chains of conidia			*Aspergillus niger*
2	On PDA, colonies showed typical bluegreen surface pigmentation with a suede-like surface consisting of a dense felt of conidiophores. Texture was powdery and the colour on the reverse side was yellow	Septate hyphae with thin-walled Conidiophore stipes are short, smoothwalled and had conical-shaped terminal vesicles. Conidia were produced in basipetal succession forming long chains and are globose to subglobose			*Aspergillus Fumigatus*
3	Colonies showed rapid growth, yellow-green to olive with granular texture; reverse gold to reddish	Septate hyphae; rough conidiophores terminating in globose vesicles with chains of conidia			*Aspergillus flavus*

highlight the importance of environmental monitoring of these fungi because of their ecological roles, industrial relevance, and potential public health implications associated with opportunistic infections and mycotoxin production.

CONCLUSION

The present study demonstrated that soils from different environmental settings within Jos Metropolis serve as important reservoirs of *Aspergillus* species. Among the identified isolates, *A. niger* was the predominant species, followed by *A. fumigatus*, *A. flavus*, and *A. nidulans*. The distribution of these fungi varied among the sampling locations, with cultivated soils and industrial effluent sites yielding the highest proportions of isolates. These findings indicate that ecological conditions and anthropogenic activities influence the occurrence and distribution of *Aspergillus* species in the study area.

The presence of medically important and potentially mycotoxin-producing species, particularly *A. fumigatus* and *A. flavus*, highlights the environmental, agricultural, and public health significance of these fungi. The findings underscore the need for continuous environmental surveillance and provide baseline information for future ecological and molecular investigations in the study area.

RECOMMENDATIONS

Based on the findings of this study, regular

monitoring of soil fungal communities in different environments within Jos Metropolis should be encouraged to detect changes in the distribution of potentially pathogenic *Aspergillus* species. Future studies should employ molecular identification techniques, such as ITS rDNA and β-tubulin gene sequencing, to complement morphological identification and improve species-level accuracy.

In addition, investigations into the antifungal susceptibility profiles and mycotoxin-producing potential of the isolated *Aspergillus* species are recommended to better assess their environmental, agricultural, and public health risks. Finally, proper management of industrial wastes, abattoir effluents, and other organic wastes should be strengthened to reduce environmental conditions that favour the proliferation of opportunistic fungal pathogens.

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