

Antifungal Susceptibility Pattern of Fungi Associated with Selected Tomato (*Solanum lycopersicum* L.) Cultivars in Makurdi Metropolis, Benue State, Nigeria

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Abstract

Tomato (*Solanum lycopersicum* L.) is an economically important vegetable crop widely consumed in Nigeria; however, its production and postharvest quality are frequently compromised by fungal contamination. This study investigated the antifungal susceptibility pattern of fungi associated with selected tomato cultivars sold in Makurdi metropolis, Benue State, Nigeria. A total of 200 rotten tomato fruits were randomly collected from five major markets (North Bank, Wurukum, High Level, Wadata, and Modern markets) and analysed using standard mycological techniques. Fungal isolation was performed using serial dilution and plating on Potato Dextrose Agar (PDA), followed by purification and identification based on macroscopic and microscopic characteristics. Four fungal species were identified: *Candida albicans*, *Rhizopus arrhizus*, *Aspergillus fumigatus*, and *Fusarium oxysporum*. The highest occurrence was recorded for *Candida albicans* (30.83%), followed by *Rhizopus arrhizus* (26.66%), *Aspergillus fumigatus* (21.66%), and *Fusarium oxysporum* (20.83%). Antifungal susceptibility testing using the agar diffusion method showed that all isolates were highly susceptible to fluconazole, with mean inhibition zones ranging from 24.20 ± 0.10 mm to 27.47 ± 0.55 mm. Moderate susceptibility was observed with ketoconazole, while clotrimazole and griseofulvin showed little or no inhibitory activity. These findings highlight the prevalence of pathogenic fungi in market-sourced tomatoes and emphasize the need for improved postharvest handling practices and continuous monitoring of antifungal resistance to ensure food safety and sustainable tomato production.

Keywords: Tomato, Fungal Isolates, Antifungal Susceptibility, Fluconazole, Postharvest Spoilage, Makurdi Metropolis, Food Safety.

I. INTRODUCTION

Tomato (*Solanum lycopersicum* L.) is one of the most widely cultivated and consumed vegetable crops worldwide, valued for its nutritional contribution as a rich source of vitamins, minerals, antioxidants, and bioactive compounds such as lycopene (FAO, 2020).

Postharvest losses of fruits and vegetables remain a major challenge in many developing countries, largely due to inadequate storage facilities and poor handling practices (Kassa and Senay, 2019). Because of their highly perishable nature, fruits and vegetables contribute

substantially to overall postharvest losses. It has been estimated that nearly half of the fruits and vegetables produced globally are lost or wasted at different stages along the agri-food supply chain, from harvesting to consumption (Tiong *et al.*, 2018).

Tomato (*Lycopersicon esculentum*), a member of the *Solanaceae* family, is among the most widely cultivated and consumed vegetables worldwide (Lamidi *et al.*, 2020). Also known scientifically as *Solanum lycopersicum*, tomato is one of the most versatile and commonly consumed fruits globally. It is highly valued for its bright red colour, unique flavour, and numerous health benefits.

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Tomatoes are widely used in different cuisines, eaten fresh in salads, and incorporated into a variety of dishes such as soups and stews. Their nutritional richness, historical importance, and culinary flexibility make them an essential component of many diets (Rao et al., 2022). Regular consumption of tomatoes as part of a balanced diet has also been associated with improved health and overall wellbeing (Rao et al., 2022). Tomatoes are nutritionally rich, containing carbohydrates, organic acids, water, minerals, vitamins, and natural pigments. On average, ripe tomato fruits consist of approximately 94% water, 4.3% carbohydrates, 1% protein, 0.1% fat, and about 0.6% dietary fibre, along with various vitamins and Kim, 2020). They are also known to contain natural antioxidants such as carotenoids, phenolic compounds, flavonoids, dietary glutathione, and several endogenous metabolites that help neutralize harmful free radicals in the body (Ali et al., 2020).

Due to their importance as a food crop, significant efforts have been made to improve tomato varieties in terms of productivity, fruit quality, and resistance to both biotic and abiotic stresses (Yang and Kim, 2020). Tomatoes have also served as an important model crop for scientific research. The crop is cultivated under different production systems, including open fields, greenhouses, and net houses, and its popularity has increased significantly over the past century (Bihn and Gravani, 2016). Globally, tomatoes are cultivated as a vegetable crop, and Nigeria ranks as the second-largest producer in Africa after Egypt (FAO, 2020).

In Benue State, particularly in Makurdi, tomato cultivation typically occurs between April and June. However, significant yield losses often occur as a result of disease outbreaks affecting the crop (Aernan et al., 2014). To obtain optimal yield and quality fruits, tomato farmers must therefore plant at the appropriate time and adopt effective crop management practices. Unfortunately, inadequate storage facilities and the absence of sufficient processing industries contribute greatly to postharvest losses caused by microbial activities. Aernan et al. (2014) reported several major pathogens associated with tomato plants in Makurdi, Gboko, Buruku, and Tarka. These include *Alternaria solani*, *Septoria lycopersici* isolated from fruits and stems, and *Fusarium oxysporum*, which was recovered from different parts of the plant.

Among vegetable crops, tomatoes are particularly delicate and highly susceptible to damage during handling, transportation, and storage. Environmental conditions, pests, and plant diseases can significantly influence tomato yield and overall production. This vulnerability is largely attributed to the high-water content of the fruit, which makes it prone to spoilage by bacteria, fungi, and other microorganisms (Abubakar et al., 2023). Fruits and vegetables naturally harbour epiphytic microflora on their surfaces. During cultivation, harvesting, transportation, processing, and handling, these products may become contaminated with both pathogenic and non-pathogenic

microorganisms originating from soil, humans, and animals (Abubakar et al., 2023).

This study therefore aims to investigate the susceptibility pattern of fungi associated with selected tomato cultivars in Makurdi metropolis. By identifying the predominant fungal species and assessing cultivar responses, the findings are expected to contribute to improved disease management, enhance sustainable tomato production, and support evidence-based recommendations for farmers and agricultural stakeholders in the region.

II. MATERIAL AND METHODS

➤ *The Study Area*

The study was carried out in Makurdi, the capital of Benue State in Nigeria. Makurdi town serves as both the capital of Benue State and the seat of the Makurdi Local Government Area. The city, which is located between latitudes 7° 44' north of the equator and longitudes 8° 31' east of the Greenwich meridian, is home to 300,000 people (National Population Commission, 2007). It encompasses 804 km² of land and has a radius of 16 km. With heights ranging from 73 to 167 meters above sea level, the Local Government Area (L.G.A.) has a generally low relief due to its location in the Lower Benue Valley. According to Onyia et al. (2019), Makurdi's soils are typically extremely ferruginous tropical soils. Makurdi is situated in the Benue Valley in the North Central region of Nigeria. It is traversed by the second-largest river in the country, the river Benue. The settlement is dominated by guinea savanna vegetation types. Large grasses, oil palm, and locust bean trees comprise much of the vegetative cover. Food production is supported by the average annual rainfall. It is between 150 and 180 millimeters, and the average annual temperature is between 27 -28⁰C.

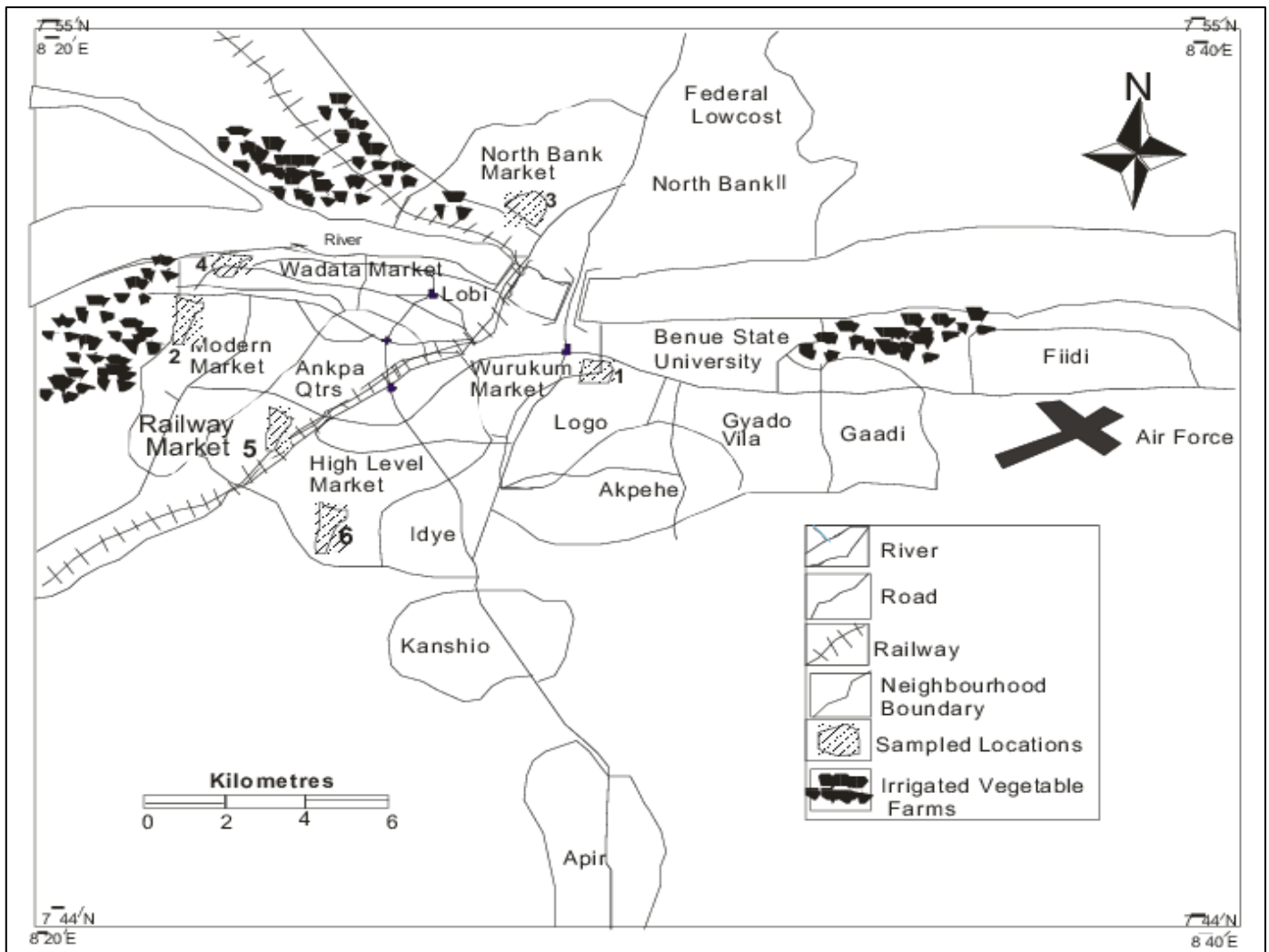


Fig 1 Map of Makurdi Showing Study Area.
(Source: Ministry of Land and Survey Makurdi, 2023).

➤ Sample Collection

Random sampling was employed to collect tomato samples from five major markets in Makurdi metropolis: North Bank, Wurukum, High Level, Wadata, and Modern markets. Four commonly consumed cultivars *Solanum lycopersicum* ‘Roma VF’, ‘UC82B’, ‘Beske’, and ‘Nagina’ were selected. A total of 200 rotten tomato fruits were obtained (10 samples per cultivar from each market). Samples were rinsed with sterile distilled water, aseptically packaged in sterilized containers following Gwa and Sani (2018), properly labeled, and transported to the Microbiology Laboratory of Joseph Sarwuan Tarka University Makurdi (JOSTUM) for fungal isolation.

➤ Isolation of Fungi

Serial dilution (10^1 – 10^5) was carried out following Elijah *et al.* (2023). Aliquots from the 10^3 , 10^4 , and 10^5 dilutions were inoculated onto Potato Dextrose Agar (PDA) plates and incubated at room temperature for 2–5 days to allow fungal growth.

➤ Fungal Count

Using the method of Olanbiwoninu *et al.* (2025), inoculated plates were incubated at $25 \pm 2^\circ\text{C}$ for five days. Total viable fungal counts were determined after incubation and expressed as colony-forming units per gram (CFU/g).

➤ Purification of Fungal Isolates

Fungal colonies were purified by subculturing distinct growths after 72 hours of incubation, following Aernan *et al.* (2014). Pure cultures obtained were preserved in sterile McCartney bottles for subsequent analysis.

➤ Identification of Fungi

Fungal isolates were identified based on macroscopic and microscopic characteristics using standard identification keys. Lactophenol Cotton Blue staining was used to prepare slides, which were examined under $\times 4$, $\times 10$, and $\times 40$ objective lenses to observe vegetative and reproductive structures such as hyphae and spores (Olanbiwoninu *et al.*, 2025).

➤ Antifungal Susceptibility Test:

Antifungal susceptibility was evaluated using the agar diffusion method (Macha *et al.*, 2021). Four antimicrobial agents—Ketoconazole, Fluconazole, Clotrimazole, and Griseofulvin—were tested against fungal isolates incorporated into agar plates. After seven days of incubation at room temperature, zones of inhibition were measured to determine isolate sensitivity.

Table 1 Standard of Susceptibility and Resistance of Antifungal Disks

S/N	Antifungal drugs	Potency	Sensitive (mm)	Intermediate(mm)	Resistance(mm)
1	Fluconazole	25 µg	≥19	15–18	≤14
2	Ketoconazole	15 µg	≥30	23–29	≤22
3	Clotrimazole	10 µg	≥20	12–19	≤11
4	Griseofulvin	25 µg	≥10	–	No zone

(Macha *et al.* (2021).

➤ *Data Analysis*

Data on fungal isolation frequency and antifungal susceptibility test were analysed using descriptive statistics of mean and standard deviation (Takam *et al.*, 2019).

III. RESULTS AND DISCUSSION

➤ *Distribution and Frequency of Fungal Species Across Sampling Locations*

The present study identified four major fungal species associated with spoiled tomato fruits in Makurdi metropolis: *Candida albicans*, *Rhizopus arrhizus*, *Aspergillus fumigatus*, and *Fusarium oxysporum*. Among these, *Candida albicans* showed the highest frequency of occurrence (30.83%), while *F. oxysporum* recorded the lowest (20.83%). The predominance of *Candida* and *Rhizopus* species in market-sourced tomatoes may be attributed to poor postharvest handling, high moisture content, and warm environmental conditions that favour rapid fungal proliferation. Similar results were obtained by Aernan *et al.* (2020), Gwa and Lum (2023), Ghosh (2009) and Sam *et al.* (2024) who indicated that fungi are major spoilage causing pathogens in tomato fruits. In another study, Onuorah and Orji (2015) isolated *Aspergillus niger*, *Fusarium oxysporum*, *Rhizopus stolonifera*, *Alternaria*

alternate, *Saacharomyces cerevisiae*, *Penicillium spp* and *Geotrichum candidum* from spoiled tomato fruits from Awka, Nigeria. However, they did not isolate *Candida spp*. This disparity in the result seems to suggest that although a lot of tomato rot causing organisms start from the farm, and *Candida spp* could be post-harvest contaminants acquired probably during transportation from the farm to the market. Also, some fungi species were not detected in this present study, these might be due to difference in the study area and samples used. The higher number of these fungi in this present studied could probably be due to environmental conditions such as temperature and relative humidity that favour the growth and activity of these fungi species in the area.

The distribution and frequency of occurrence of the isolated fungal species across the sampling locations are presented in Table 1. Among the markets surveyed, Wadata Market recorded the highest diversity and frequency of fungal pathogens, whereas North Bank Market exhibited the lowest occurrence. *Candida albicans* showed the highest frequency of occurrence (30.83%), followed by *Rhizopus arrhizus* (26.66%), *Aspergillus fumigatus* (21.66%), and *Fusarium oxysporum* (20.83%), which recorded the lowest percentage occurrence.

Table 2 Fungal Species Isolated and Identified According to Their Location

Isolates	North bank market	Wurukum market	High-level market	Wadata market	Modern market	Total	% Occurrence
<i>Aspergillus fumigatus</i>	5	4	3	10	4	26	21.66
<i>Fusarium oxysporum</i>	3	4	5	8	5	25	20.83
<i>Rhizopus arrhizus</i>	4	7	5	9	7	32	26.66
<i>Candida albicans</i>	10	9	3	6	9	37	30.83
Total	22	24	16	31	25	120	100

➤ *Antifungal Susceptibility of Fungal Isolates*

The antifungal susceptibility pattern of the isolated fungi was determined using four antifungal agents, namely fluconazole, ketoconazole, clotrimazole and griseofulvin. The zones of inhibition obtained from three replicate plates were analysed and expressed as mean ± standard deviation (mm). The Clinical and Laboratory Standard Institute (CLSI) first authorized the antifungal susceptibility testing procedure for dermatophytes in 2008, and it underwent additional modifications in 2010 Macha *et al.* (2021). The most popular antifungal for the treatment of dermatophyte fungal infections are azoles, allylamines, and polyenes. The azoles are divided into two groups: triazoles (such as fluconazole and itraconazole) and imidazoles (such as clotrimazole and ketoconazole). For the majority of severe fungal infections, triazoles are employed as systemic first-line treatments Macha *et al.*

(2021). The effectiveness of different antifungal medications against different fungal genera and species can be assessed using a variety of techniques. The standard disk diffusion assay was recommended by Macha *et al.* (2021) as a suitable paradigm for antifungal testing in routine laboratory diagnosis and evaluation of dermatophyte resistance to antifungal drugs. In this present study, the disk diffusion method was adopted to ascertain the activities of four antifungal viz; Fluconazole, ketoconazole, clotrimazole and griseofulvin against four fungi isolates (*Aspergillus fumigatus*, *Fusarium oxysporum*, *Rhizopus arrhizus* and *Candida albicans*) associated with tomato fruits sold within Makurdi metropolis. The zones of inhibition obtained from three replicate plates were analysed and expressed as mean ± standard deviation (mm).

Antifungal susceptibility results (Table 2) showed clear differences among the tested drugs. Fluconazole demonstrated 100% susceptibility against all fungal isolates. Ketoconazole showed intermediate activity against *Aspergillus fumigatus* and *Rhizopus arrhizus*, but resistance was observed in *Candida albicans* and *Fusarium oxysporum*. Griseofulvin exhibited complete resistance against all isolates tested.

➤ *Comparative Antifungal Activity of Tested Drugs*

The fungal isolates exhibited varying antifungal susceptibility patterns to the three antifungal agents used. Fluconazole exhibited the highest antifungal activity against all fungal isolates with mean standard deviation of 26.43 ± 0.49 against *Aspergillus fumigatus*, 26.43 ± 0.49 against *Fusarium oxysporum*, 24.20 ± 0.10 against *Rhizopus arrhizus* and 27.47 ± 0.55 against *Candida albicans* indicating 100% susceptibility rate. This report agrees with the findings of Olanbiwoninu *et al.* (2025) and disagrees with findings of Macha *et al.* (2021) and El-Hamd *et al.* (2019).

Ketoconazole demonstrated moderate antifungal activity against the isolates, with most values falling within the intermediate susceptibility range. Clotrimazole showed minimal inhibitory activity with mean standard deviation of 23.23 ± 0.06 to *Rhizopus arrhizus* and 23.13 ± 0.21 to *Aspergillus fumigatus* (intermediate) indicating 50% intermediate and resistant to *Candida albicans* and *Fusarium oxysporum* with mean standard deviation of

(0.00 ± 0.00) indicating 100% resistant. This report agrees with findings of Macha *et al.* (2021).

Clotrimazole and griseofulvin showed total inhibition rate to the four isolates with mean standard deviation of (0.00 ± 0.00) indicating 100% resistant to all the isolates. This is consistent with the findings of Macha *et al.* (2021) that griseofulvin has been losing popularity as a result of considerable treatment failure, expensive cost, and lengthy treatment duration. However, species-specific variations in treatment responsiveness and mutations like missense substitution could be the cause of this of this ineffectiveness. It is concerning for public health when these antifungal-resistant fungus are found in ready-to-eat fruits and vegetables. The findings of this study suggest that fluconazole could be utilized as therapeutic agent for managing fungal infections. Comparatively, fluconazole was the most effective antifungal agent, while ketoconazole showed moderate (50%) activity. Clotrimazole and griseofulvin showed little to no inhibitory effect on most isolates, indicating high resistance.

➤ *Mean Zones of Inhibition of Antifungal Agents*

Mean zones of inhibition (from triplicate tests in Table 5) further confirmed fluconazole’s superior activity, with inhibition zones of 26.43 ± 0.49 mm (*A. fumigatus*), 25.43 ± 0.45 mm (*F. oxysporum*), 24.20 ± 0.10 mm (*R. arrhizus*), and 27.47 ± 0.55 mm (*C. albicans*). All isolates were classified as susceptible to fluconazole based on standard breakpoints.

Table 3 Antifungal Susceptibility Pattern of the Isolated Fungi

S/N	Fungal isolates	FLU	KET	CLT	GRV
1	<i>Aspergillus fumigates</i>	26	23	10	0
2	<i>Fusarium oxysporum</i>	25	22	0	5
3	<i>Rhizopus arrhizus</i>	24	23	9	4
4	<i>Candida albicans</i>	27	22	0	0

- Key: FLU- Fluconazole, KET- Ketoconazole, CLT- Clotrimazole, GRV- Griseofulvin

Table 4 Test Results of the Susceptibility to Fungi Isolates Against Selected Antifungal Drugs.

Antifungal	Susceptible (S) n (%)	Intermediate (S) n (%)	Resistance (S) n (%)
Fluconazole	4 (100%)	0 (0%)	0 (0%)
Ketoconazole	0 (0%)	2 (50%)	2 (50%)
Clotrimazole	0 (0%)	0 (0%)	4 (100%)
Griseofulvin	0 (0%)	0 (0%)	4 (100%)

Table 5 Mean Zone of Inhibition (mm ± SD) and Susceptibility Pattern of Fungal Isolates to Selected Antifungal Agents

Fungal Isolate	Antifungal Drug	Mean ± SD (mm)	Interpretation
<i>Aspergillus fumigatus</i>	Fluconazole	26.43 ± 0.49	Susceptible
<i>Aspergillus fumigatus</i>	Ketoconazole	23.13 ± 0.21	Intermediate
<i>Aspergillus fumigatus</i>	Clotrimazole	10.17 ± 0.21	Resistant
<i>Aspergillus fumigatus</i>	Griseofulvin	0.00 ± 0.00	Resistant
<i>Fusarium oxysporum</i>	Fluconazole	25.43 ± 0.45	Susceptible
<i>Fusarium oxysporum</i>	Ketoconazole	22.20 ± 0.10	Resistant
<i>Fusarium oxysporum</i>	Clotrimazole	0.00 ± 0.00	Resistant
<i>Fusarium oxysporum</i>	Griseofulvin	5.23 ± 0.15	Resistant
<i>Rhizopus arrhizus</i>	Fluconazole	24.20 ± 0.10	Susceptible
<i>Rhizopus arrhizus</i>	Ketoconazole	23.23 ± 0.06	Intermediate
<i>Rhizopus arrhizus</i>	Clotrimazole	9.10 ± 0.20	Resistant
<i>Rhizopus arrhizus</i>	Griseofulvin	4.20 ± 0.26	Resistant
<i>Candida albicans</i>	Fluconazole	27.47 ± 0.55	Susceptible
<i>Candida albicans</i>	Ketoconazole	22.43 ± 0.15	Resistant
<i>Candida albicans</i>	Clotrimazole	0.00 ± 0.00	Resistant
<i>Candida albicans</i>	Griseofulvin	0.00 ± 0.00	Resistant

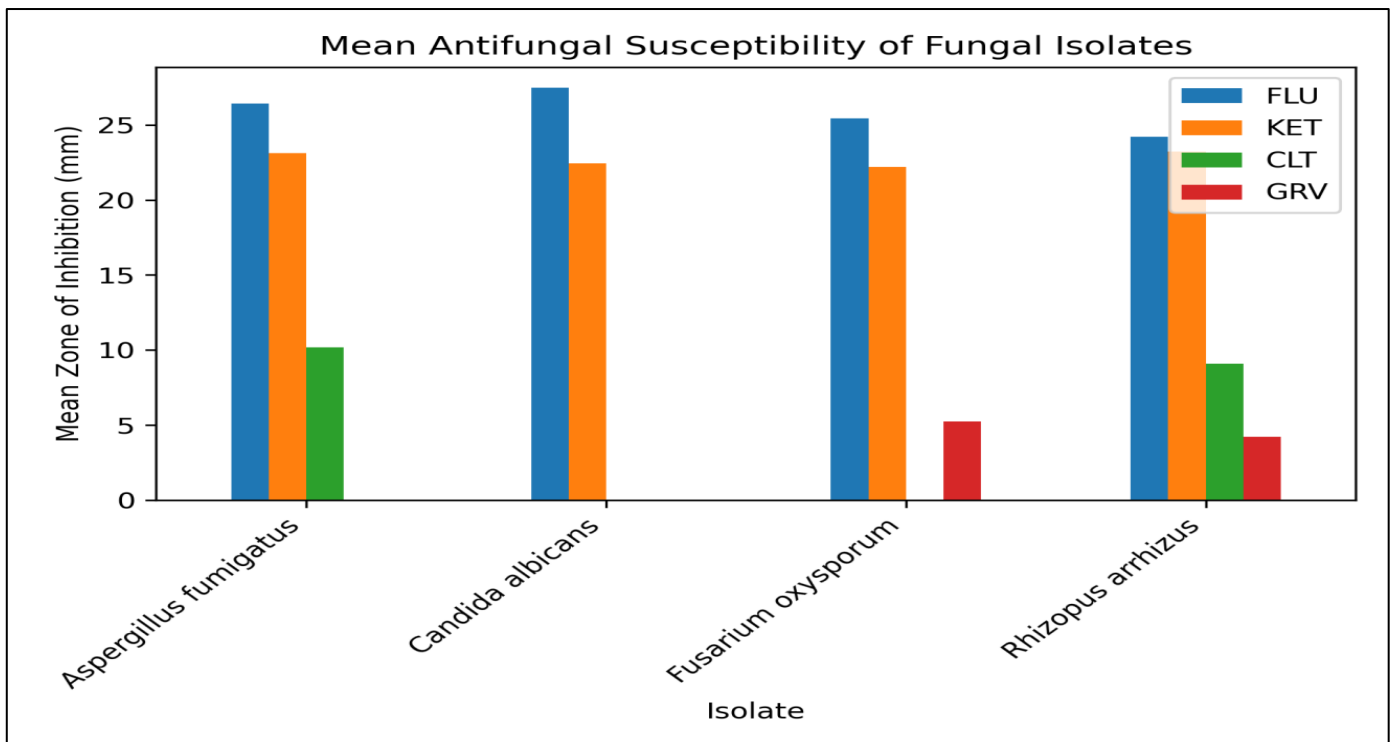


Fig 2 Mean Antifungal Susceptibility of Isolated Fungal

IV. CONCLUSION AND RECOMMENDATIONS

➤ Conclusion

The findings of the present study indicate that fluconazole remains highly effective against the fungal isolates associated with spoilt tomatoes in Makurdi, while resistance to other antifungal agents is evident. These results underscore the importance of continuous monitoring of antifungal susceptibility patterns, particularly in regions where agricultural produce is exposed to diverse environmental contaminants.

This study demonstrated that *Candida albicans*, *Rhizopus arrhizus*, *Aspergillus fumigatus*, and *Fusarium oxysporum* are the major fungal pathogens associated with spoilt tomato fruits in Makurdi metropolis. Among the

antifungal agents tested, fluconazole exhibited the highest inhibitory activity against all isolates, while significant resistance was observed against clotrimazole and griseofulvin. These findings highlight the need for improved postharvest handling practices, regular monitoring of fungal contamination, and effective management strategies to reduce postharvest losses and potential health risks associated with contaminated tomatoes.

➤ Recommendations

The following is recommended:

- Farmers and vendors should adopt better harvesting, transportation, and storage practices to minimize mechanical damage and fungal contamination.

- Regular cleaning and proper ventilation in markets should be encouraged to reduce fungal proliferation.
- Periodic monitoring of fungal species and their antifungal susceptibility patterns should be conducted to track emerging resistance trends.
- The use of resistant tomato cultivars, biological control methods, and good agricultural practices should be promoted to reduce reliance on chemical antifungals.
- Awareness programs should be organized to educate farmers, vendors, and consumers on the risks associated with fungal contamination and improper handling of tomatoes.

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