# Phytochemical potentiality and antifungal activity of extracts from *Cymbopogon citratus* and *Xylopia aethiopica* : a biopreservation strategy for charmout against fungal strains in N'Djamena, Chad

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## Abstract

The use of medicinal plants is crucial in traditional and modern medicine. In Africa, fungal infections, particularly those caused by foodstuffs such as *charmout* (dried meat) in Chad, are a significant public health problem. The present study evaluated the phytochemical and antifungal potential of extracts from *Cymbopogon citratus* leaves, *Xylopia aethiopica* fruit, and essential oil *from Cymbopogon citratus* against the microflora that causes *charmout* spoilage (*Aspergillus niger, Mucor sp*., *Fusarium sp*.). Phytochemical analysis revealed that both plants are rich in secondary metabolites. The levels of phenol in X. aethiopica and C. citratus were found to be 72.37 ± 0.59 mgEqAG/gMS and 74.96 ± 0.85 mgEqAG/gMS, respectively. The presence of saponins was also detected, with X. aethiopica being particularly rich in these compounds. X. *aethiopica* was distinguished by a greater abundance of tannins and alkaloids in comparison the C. *citratus.* The ethanolic extracts of both plants exhibited strong inhibitory activity at concentrations of 1 and 2 mg/mL. The ethanolic extract of *Cymbopogon citratus* demonstrated a high level of inhibition against *Mucor sp.* and *Fusarium sp.* over a period of seven days at elevated concentrations. In contrast, the aqueous extracts showed negligible activity after three days, highlighting the ineffectiveness of water in extracting the major active compounds. The study corroborates the substantial potential of these plants as effective biopreservatives for the fungicidal protection of *charmout*, emphasising the necessity for optimisation of concentrations to ensure sustainable protection.

## INTRODUCTION

The utilisation of medicinal plants is an age-old practice that persists in the global healthcare systems. According to the World Health Organisation (WHO), herbal preparations constitute a fundamental component of primary healthcare for up to 80% of the global population (WHO, 2007). The significance of natural resources is increasing not only in traditional medicine, but also in modern medicine, where almost 74% of plant-based medicines have uses that overlap with their traditional applications (Saggar et al. 2022; Al-worafi et al.2022). In the face of emergence of antimicrobial resistance and persistent challenges in the treatment of various diseases, the search for new bioactive molecules particularly antiviral, antibacterial and antifungal compounds has become a priority (Kieltyka-Dadasiewicz et al. 2024). These phytochemical compounds, obtained through traditional or modern processes (e.g. maceration, decoction, extraction, etc.), offer significant therapeutic potential against a wide range of human and animal diseases (Agaie et al. 2007; Da et al. 2015; Ouédraogo et al. 2019; Ouédraogo et al. 2021). In numerous developing countries, opportunistic fungal diseases and fungal contamination of foodstuffs constitute a substantial public health problem (Okigbo et al. 2008). It is evident that the preservation of foodstuffs represents a significant challenge. *Charmout* (smoked and dried meat), a widely consumed foodstuff in Chad, is particularly vulnerable to spoilage by fungal flora. This contamination not only compromises the quality and shelf life of the product, but also exposes consumers to the risk of mycotoxins. (Maloba et al. 2025). In this context, the use of natural substances is particularly appropriate.

The present study evaluated the phytochemical potential and activity of extracts of *Xylopia aethiopica* and *Cymbopogon citratus* for the conservation and preservation of *charmout* quality.

## MATERIAL AND METHODS

# Plant material

The plant material utilised in this study encompassed fresh leaves of *Cymbopogon citratus* (Fig. 2a) harvested on 22 October 2024 in the Sabangali district and fruits of *Xylopia aethiopica* (Fig. 2b) procured at the central market in N'Djamena (Fig. 1) on 25 October 2024.

The samples were dispatched to the Food Science and Nutrition Research Laboratory (LaRSAN). Subsequent to the process of transportation, the material underwent a drying procedure at ambient temperature, in conditions that were devoid of both light and dust. The drying time for *Cymbopogon citratus* leaves was three to four days.

The samples were then subjected to grinding using a mechanical grinder, with approximately 1 kg of powder being obtained from each sample. This powder was subsequently packaged in plastic bags and stored in a manner that ensured it was protected from light and moisture pending extraction.

## Fungal material

The fungal material utilised in this study comprises moulds previously isolated from samples of *charmout* collected from diverse markets in N'Djamena. The aforementioned samples were identified as Aspergillus niger, Mucor sp. and Fusarium sp., which are considered to be the primary agents of contamination and biodeterioration of the foodstuff in question. These organisms are frequently implicated in the production of mycotoxins (Pandey et al. 2023).

The fungal strains were maintained and cultured on Sabouraud medium, with the addition of chloramphenicol to inhibit bacterial growth and ensure the purity of the cultures.

## Phytochemical study

A preliminary phytochemical study of the extract powder was conducted using conventional analytical chemistry methods with the objective of determining the presence of major classes of phenolic and nitrogenous compounds, including alkaloids, saponins, tannins, glycosides and flavonoids.

**Alkaloid test (Wagner's reagent)**:
The add a few drops of Dragendorff's reagent to a 2-mL extract is required to facilitate the desired reaction. The presence of an orange colour or precipitate is indicative of the presence of alkaloids. In the event of a positive test result, the presence of polyphenols must be confirmed using Nessler's reagent.

Subsequently, two to three drops of Nessler's reagent should be addited to 2 mL of extract. The presence of a yellowish precipitate is indicative to the presence of alkaloids (Harbone, 1973; Wagner, 1983).

### Saponin test (foaming test)

The quantity of plant powder required for this experiment is 1 g. This should be placed in a 250 mL Erlenmeyer flask. The next step is to added 100 mL of distilled water. The mixture should be heated gently. The substance should then be filtered, cooled and made up to 100 mL with distilled water. The filtrate (10 mL) should then be transferred into a test tube and vigorously shaken for 15 seconds. The tube should be placed in an upright position for a period of 15 minutes. Should the foam persist beyond this designated period, it can be deduced that the plant drug contains saponins (Wagner, 1983).

## Tannin test:

A quantity of 5 g of the powder is to be placed in 100 mL of boiling water in an Erlenmeyer flask. Following a 15-minute infusion, the solution should be filtered and made up to 100 mL with distilled water. Five (5) mL of the 5% infusion is placed into a test tube, followed by the addition of 1 millilitre of a 1% aqueous solution of $FeCl_3$. In the presence of gallic tannins, a greenish or blue-black colouration is observed to develop. The presence of catechin tannins is revealed through the use of Stiasny's reagent, as evidenced by research conducted by Edeoga et al.(2005) and Koffi et al.(2015).

**Glycoside test (Benedict's reagent)**:

A quantity of 0.5 g of the powder was mixed with 10 mL of boiling distilled water for the purpose of extraction. Glycoside hydrolysis was performed on 2 mL of the filtrate by the addition of concentrated hydrochloric acid, followed by the subsequent alkalisation with ammonia. Five drops of the hydrolysed solution were then boiled with 2 mL of Benedict's reagent. The formation of a reddish-brown precipitate was indicative of the presence of glycosides.

### Flavonoid test

In a test tube, 2 mL of hydrochloric alcohol (a mixture of 95% ethanol, distilled water and concentrated hydrochloric acid, equivalent to the volume of the mixture) was added to 1 mL of n-amyl alcohol, along with a few magnesium shavings, to which 2 mL of infusate was added. The presence of a pink-orange (flavones) or pink-purple (flavanones) or red (flavonones, flavanonols) hue in the isoamyl alcohol superiores layer is indicative of the presence of a free flavonoid (genin). The presence of flavone glycosides results in a reduction of colour intensity (Wagner, 1983).

## Quantitative study

The quantitative analysis of secondary metabolites in *Xylopia aethiopica* and *Cymbopogon citratus* is based on the use of spectrophotometric methods, which require the establishment of calibration curves (Fig. 3) to determine the concentration of the compounds of interest.

## Evaluation of the antifungal bioactivity of extracts

## Preparation of total extracts

The pulverised plant samples were then subjected to extraction. For each plant organ (*Cymbopogon citratus* and *Xylopia aethiopica*), two types of crude extracts were prepared: an aqueous extract and an ethanolic extract.

## Preparation of aqueous extracts

A 50 g sample of plant powder was macerated in 500 mL of distilled water for 48 hours at room temperature, with constant agitation using a magnetic stirrer. Following the maceration process, the extract solution was subjected to a dual filtration procedure on cotton (utilising a funnel) with the objective of removing plant debris. The filtrate obtained was then collected in a ceramic dish and placed in an oven at 50°C for a period of 72 hours, in order to facilitate complete drying. Subsequent to the desiccation of the extract, the residual aqueous solution was meticulously removed, weighed and transferred to an airtight container. This procedure enabled the calculation of the extraction yield (Compaore et al. 2021).

## Preparation of ethanol extracts

The extraction of Ethanolic extraction was conducted through the process of maceration, involving the immersion of 50 g of plant materiel in 500 mL of ethanol solution at ambient temperature for a duration of 24 hours, with continuous mechanical agitation. Following filtration, the filtrate was subjected to complete evaporation at 50°C using an oven in order to obtain the crude extract (Compaoré et al. 2021).

## Determination of extraction yield

The extraction yield (R) was determined for each type of extract (aqueous and ethanolic) using the following formula :

$$\varvec{R}\ (\varvec{\%}) = \frac{\varvec{M}\varvec{a}\varvec{s}\varvec{s}\ \varvec{o}\varvec{f}\ \varvec{e}\varvec{x}}{\varvec{M}\varvec{a}\varvec{s}\varvec{s}\ \varvec{o}\varvec{f}\ \varvec{v}\varvec{e}\varvec{g}\varvec{e}\varvec{t}\varvec{a}\varvec{}}$$

## In vitro evaluation of antifungal activity

The antifungal activity of the crude extracts and essential oil was determined *in vitro* by the agar incorporation method according to Senhaji *et al.* (2005), by measuring the inhibition of mycelial growth of fungal strains isolated from *charmout*. The experimental tests were conducted using crude extracts (aqueous

and ethanolic) from *Cymbopogon citratus* leaves and *Xylopia aethiopica* seeds, in addition to essential oil extracted from *Cymbopogon citratus* leaves.

This evaluation enabled the determination of the primary parameters of antifungal activity, including the diameters of the inhibition zones, the Minimum Inhibitory Concentration (MIC) and the Minimum Fungicidal Concentration (MFC) (Skandamis and Nychas, 2001). The A method was utilised to determine this. The immersion of discs in oil B was conducted in a Petri dish containing agar, with an essential oil disc positioned centrally. The experimental part comprised the following materials materials and methods : micro-dilution in liquid medium. The essential oil is initially prepared by means of emulsion in an ethanol solvent at a concentration of 1 mg/µl, with a view to dispersing the compounds and improve enhancing their contact with the germs to be tested. Subsequent dilutions of the solution obtained are then carried out by geometric progression to obtain the following dilutions. According to the Clinical and Laboratory Standards Institute (CLSI, 2008), the following values should be used : 1/2, 1/4, 1/8, 1/16, 1/32, 1/64 and 1/128.

## Preparation of extract solutions

The essential oil (EO) of *Cymbopogon citratus* was solubilised in ethanol at a ratio of 1:9 (v/v).

Stock solutions of aqueous and ethanolic extracts of C. *citratus* and *X. aethiopica* at a concentration of 20 mg/mL were prepared under aseptic conditions in a laminar flow hood.

A series of dilutions of each stock solution was carried out in Sabouraud Dextrose Agar (SDA) culture medium, with the addition of chloramphenicol as a preservative. The following final concentrations were obtained in the agar for testing : 0.15, 0.25, 0.5, 0.75, 1, and 2 mg/mL (Bakarnga-VIA et al. 2022).

## Preparation of Petri dishes

The prepared media, containing the different concentrations of essential oils, were poured into sterile Petri dishes (90 mm in diameter) at a rate of 20 mL/dish and left under the fume hood until solidification. Dishes devoid of any extract were utilised as growth controls.

## Inoculation and monitoring of fungal growth

Each Petri dish was inoculated in the centre with a 6 mm diameter mycelial disc (explant) taken from a young culture (3 to 4 days old) using a sterile cookie cutter. The boxes were hermetically sealed and then placed into an inverted position within an incubator set to a temperature of 37°C. Mycelial growth was monitored on a daily basis over a period of seven days. Growth was quantified by measuring two perpendicular diameters (D1 and D2) marked on the reverse of each Petri dish (Akakpo et al. 2023).

The diameter of daily mycelial growth (D) was calculated by subtracting the diameter of the explant (DE) from the average of the two measured diameters :

$$\mathbf{D} = \frac{\varvec{D}1 + \varvec{D}2}{2} - \varvec{D}\varvec{E}$$

The antifungal activity of the essential oil was evaluated by the percentage of inhibition (% I), calculated in relation to the average diameter of fungal growth in the control box (Dt), according to the following formula :

$$\% \mathbf{I} = \frac{\varvec{D}\varvec{t} - \varvec{D}\varvec{e}}{2} 100$$

Where "**De**" represents the average diameter of fungal growth in the box containing the extract.

## Determination of Minimum Inhibitory and Fungicidal Concentrations

Following a period incubation spanning seven days, the Minimum Inhibitory Concentration (MIC) was determined as the lowest extract concentration that resulted in no observable macroscopic mycelial growth in the Petri dishes.

The minimum fungicidal concentration (MFC) and the nature of the inhibition were determined by subculture. Explantations that exhibited no visible growth after seven days of incubation at the MIC and higher concentrations were transferred to new Petri dishes containing sterile culture medium without extract supplementation.

Following a subsequent incubation period :

- Should mycelial growth be observed once more, the extract will be designated as fungistatic, a classification that denotes its capacity to impede fungal growth without exerting an lethal effect.
- In the absence of observed growth, the extract is designated as fungicidal, that is to say, it is lethal to the fungus. The lowest of these concentrations is thus defined as the FIC.

The fungicidal nature of the extract was quantified by the MIC/CMF ratio. A ratio equal to 1 or 2 indicates a fungicidal effect, while a ratio greater than 2 indicates a fungistatic effect (CLSI, 2002).

# RESULTS

## Extraction yields

The extraction yields for both plants (*Xylopia aethiopica* and *Cymbopogon citratus*) were calculated using two solvents (water and ethanol), and the results are presented in Table 1.

Table 1
Extraction yield

| Plants | Part used | Aqueous extract | Ethanolic extract |
|---|---|---|---|
| **Xylopia aethiopica** | Fruit | 8.2 | 30 |
| **Cymbopogon citratus** | Leaf | 16 | 19 |

The extraction yield ranged from 8.2 to 30%. The lowest yields were obtained with the aqueous extract for both species. In contrast, the ethanolic extract yielded the highest yields for *Xylopia aethiopica* (30%) and *Cymbopogon citratus* (19%).

# Phytochemical screening results

## Qualitative phytochemistry

The results of the comparative phytochemical analysis of Guinea fruit (*Xylopia aethiopica*) and lemongrass leaves (*Cymbopogon citratus*) are presented in Table 2, with the emphasis on the richness and relative abundance of four major classes of chemical compounds in each plant sample.

Table 2
Chemical characterisation results *for Cymbopogon citratus and Xylopia aethiopica*

| Chemical groups | *Xylopia aethiopica* | *Cymbopogon citratus* |
|---|---|---|
| **Alkaloids** | ++ | + |
| **Tannins** | ++ | + |
| **Saponins** | +++ | ++ |
| **Phenols** | +++ | +++ |

**Key**

**(-)** *= Totally absent ;* **(+)** *= Slightly present ;* **(++)** *= Moderately abundant ;* **(+++)** *= Very abundant.*

## Quantitative phytochemistry

As illustrated in Fig. 3, the secondary metabolism curves for *Xylopia aethiopica* and *Cymbopogon citratus are presented.*

Table 3 below shows the quantified contents of the main chemical groups in the crude extract of the two plants, expressed in milligrams of standard equivalent per gram of dry matter (mg/gDM), with their margins of error.

Table 3
Results of quantitative analysis of secondary metabolites of *Cymbopogon citratus and Xylopia aethiopica*

| Chemical groups | *Cymbopogon citratus* | *Xylopia aethiopica* | Calibration unit |
|---|---|---|---|
| **Phenols** | 72.37 ± 0.59 | 74.96 ± 0.85 | mgEqAG/gMS |
| **Tannins** | 26.41 ± 0.13 | 44.35 ± 0.62 | mgEqCat/gDM |
| **Saponins** | 63.29 ± 0.78 | 61.47 ± 0.73 | mgEqGal/gDM |
| **Alkaloids** | 41.52 ± 0.44 | 46.17 ± 0.67 | mgEqQui/gDM |
| **The following abbreviations are employed in this text**: **mgEqAG/gDM**: *milligram of gallic acid equivalent per gram of dry matter;* **mgEqCat/gDM**: *milligram of catechin equivalent per gram of dry matter;* **mgEqGal/gDM**: *milligram of galactose equivalent per gram of dry matter;* **mgEqQui/gDM**: *milligrams of quinine equivalent per gram of dry matter* | | | |

# Evaluation of the antifungal activity of crude extracts

## Summary of inhibitory activity at 72 hours

Macroscopic observations made after 72 hours of incubation revealed strong inhibition of fungal growth in the boxes containing the extracts in comparison to the control (Sabouraud medium alone). The results of this study are presented in Table 4.

Table 4
Results of exposure to the crude extract after 72 hours of incubation

| Plant species | PU | 24H | 48H | 72 hours | Macroscopic characteristics of colonies (size, appearance, colour, etc.) |
|---|---|---|---|---|---|
| **Xylopia aethiopica** | EE | - | - | + | Small, round colonies |
| | EA | + | + | + | Small, granular, beige colonies |
| **Cymbopogon citratus** | EE | - | + | + | Small, flat, yellow colonies |
| | EA | + | + | + | Medium, granular, yellowish colonies |
| Key: **PU**: *Part used;* **EE**: *Ethanolic extract;* **EA**: *Aqueous extract* | | | | | |

The antifungal activity of the aqueous and ethanolic extracts is demonstrated in Figs. 4 and 5, respectively, at varying concentrations and after an incubation period of 7 days

## Qualitative evaluation of the efficacy of the extracts on fungal strains

As illustrated in Table 5, the evaluation of antifungal activity on various fungal genera was conducted after 72 hours peroid. The findings of this evaluation demonstrate that the ethanol extracts exhibited superior inhibitory activity.

Table 5
Results of the evaluation of the antifungal activity of extracts on fungal genera

| Strains | Plants | Aqueous extract | Ethanol extract |
|---|---|---|---|
| *Aspergillus niger* | *Xylopia aethiopica* | + | - |
| | *Lemongrass* | - | - |
| *Mucor* | *Xylopia aethiopica* | - | - |
| | *Lemongrass* | + | - |
| *Fusarium* | *Xylopia aethiopica* | + | + |
| | *Lemongrass* | + | - |
| key: **-** *= No colony;* **+** *= Colony present* | | | |

This observation indicates that the ethanol extracts of *Xylopia aethiopica* and *Cymbopogon citratus* demonstrate a degree of activity, in contrast to the aqueous extracts, which exhibit reduced activity at the concentrations examined.

## Evaluation of the inhibitory effects of the extracts

Daily monitoring of mycelial growth over on seven days at a temperature of 27°C permitted the inhibitory effect of the various concentrations of extracts to be evaluated (Table 6).

Table 6
Results of the inhibitory effects of aqueous and ethanolic extracts of *C. citratus* and *X. aethiopica* on the three fungal strains

| Extracts | Plants | Microorganisms | Day | Concentrations | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | | | | 0.15 | 0.25 | 0.5 | 0.75 | 1 | 2 |
| **Aqueous** | *C. citratus* | *A. Niger* | 1 | 1.01 | 1.10 | 1.09 | 0.55 | 0.02 | 0.03 |
| | | | 2 | 1.02 | 1.91 | 1.01 | 0.98 | 0.53 | 0.08 |
| | | | 3 to 7 | + | + | + | + | + | + |
| | *X. aethiopica* | *A. Niger* | 1 | 1.01 | 1.18 | 1.06 | 0.93 | 0.06 | 0.06 |
| | | | 2 | 1.23 | 1.71 | 1.13 | 0.96 | 0.13 | 0.09 |
| | | | 3 to 7 | + | + | + | + | + | + |
| | *C. citratus* | *Mucor sp.* | 1 | 1.01 | 0.98 | 1.06 | 0.93 | 0.04 | 0.06 |
| | | | 2 | 1.02 | 1.02 | 1.01 | 0.96 | 0.13 | 0.09 |
| | | | 3 | 0.99 | 1.00 | 0.06 | 1.06 | 0.03 | 0.79 |
| | | | 4 | 0.41 | 0.28 | 1.01 | 0.75 | 0.08 | 0.07 |
| | | | 5 to 7 | + | + | + | + | + | + |
| | *X. aethiopica* | *Mucor sp.* | 1 | 1.01 | 1.96 | 1.06 | 0.93 | 0.04 | 0.03 |
| | | | 2 | 1.62 | 1.01 | 1.01 | 0.73 | 0.13 | 0.08 |
| | | | 3 to 7 | + | + | + | + | + | + |
| | *C. citratus* | *Fusarium sp.* | 1 to 5 | - | - | - | - | - | - |
| | | | 6 | 0.07 | - | - | - | - | - |
| | | | 7 | 0.81 | 0.48 | - | - | - | - |
| | *X. aethiopica* | *Fusarium sp.* | 1 | 1.62 | 1.07 | 0.24 | 0.91 | 0.07 | 0.06 |
| | | | 2 | 1.76 | 1.22 | 0.27 | 1.06 | 0.13 | 0.09 |
| | | | 3 to 7 | + | + | + | + | + | + |
| **Ethanolic** | *C. citratus* | *A. Niger* | 1 to 4 | - | - | - | - | - | - |
| | | | 5 | 0.04 | 0.01 | 0.03 | 0.07 | 0.84 | 0.02 |
| | | | 6 | 0.07 | 0.20 | 0.05 | 1.05 | 0.69 | 0.12 |
| | | | 7 | 0.81 | 0.46 | 0.08 | 1.99 | 0.31 | 0.22 |
| | *C. citratus* | *Mucor sp.* | 1 to 4 | - | - | - | - | - | - |
| | | | 5 | 0.38 | 1.08 | 0.015 | - | - | - |
| | | | 6 | 1.03 | 0.63 | 0.035 | - | - | - |
| | | | 7 | 0.96 | 0.01 | 0.024 | - | - | - |
| | *C. citratus* | *Fusarium sp.* | 1 to 5 | - | - | - | - | - | - |
| | | | 6 | 0.07 | - | - | - | - | - |
| | | | 7 | 0.81 | 0.48 | - | - | - | - |
| | *X. aethiopica* | *A. Niger* | 1 | 0.15 | 0.25 | 0.5 | 0.75 | 1 | 2 |
| | | | 2 to 5 | - | - | - | - | - | - |
| | | | 6 | 1.65 | 0.14 | 0.33 | 0.31 | - | - |
| | | | 7 | 1.71 | 1.22 | 0.88 | 1.03 | - | - |
| | *X. aethiopica* | *Mucor sp.* | 1 and 2 | - | - | - | - | - | - |
| | | | 3 | 0.03 | 1.12 | 0.06 | - | - | - |
| | | | 4 | 0.53 | 0.18 | 1.01 | - | - | - |
| | | | 5 | 0.43 | 0.24 | 0.06 | 0.33 | 1.09 | 0.01 |
| | | | 6 | 1.21 | 0.60 | 0.08 | 0.54 | 1.63 | 0.05 |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | | 7 | 1.81 | 0.78 | 0.09 | 0.97 | 1.71 | 0.03 |
| *X. aethiopica* | *Fusarium sp.* | 1 to 4 | - | - | - | - | - | - |
| | | 5 | 0.04 | 0.01 | 0.03 | 0.38 | - | - |
| | | 6 | 1.72 | 0.30 | 0.02 | 0.01 | 0.63 | 1.02 |
| | | 7 | 1.81 | 0.44 | 0.07 | 0.03 | 0.51 | 1.02 |

**Legend**

*[C] = concentration in mg/mL. Values are colony diameter in mm. (+) = Full colony; (−) = No colony.*

**NB**

below (last page) find Table 6

## Inhibitory effects of aqueous extracts from both plants

The aqueous extracts exhibited an insignificant inhibitory effect on all strains examined, particularly during the initial 3 to 4 days of incubation (Table 6).

In the case of *Aspergillus niger*, *Cymbopogon citratus exhibited minimal* mycelial growth during the first two days, but underwent accelerated development from the third to the seventh day across all concentrations (0.15 to 2 mg/mL).

The aqueous extract of *Xylopia aethiopica* demonstrated significant growth from the first three days, followed by uninhibited growth from the fourth to the seventh day.

In the case of *Mucor sp.*, the aqueous extract of *Cymbopon citratus* and the aqueous extract of *Xylopia aethiopica* exhibited no significant inhibitory effect on this genus, with growth observed from the first to the last day.

In the case of *Fusarium sp.*, the aqueous extract of *Xylopia aethiopica* and the aqueous extract of *Cymbopogon citratus* demonstrate no inhibitory effects, with high mycelial development observed from the first day.

## Inhibitory effects of ethanolic extracts from both plants

Ethanolic extracts exhibit considerable inhibitory potential, particularly at elevated concentrations (Table 6).

In the case of *A. niger*, ethanol extracts of *Cymbopogon citratus* have been observed to result in a complete cessation of growth inhibition from the first to the fourth day at all concentrations. Although there is a resumption of partial growth resumes from the fifth to the seventh day, inhibition is maintained at low concentrations.

In the case of *Mucor sp.*, the ethanol extract of *Cymbopogon citratus* demonstrated complete inhibition on the first four days. Growth resumed from the fifth to the seventh day at low concentrations (0.15, 0.25 and 0.5 mg/mL), but inhibition was maintained at high concentrations (0.75, 1 and 2 mg/mL).

In the case of *Fusarium sp.*, for the ethanolic extract of *Cymbopogon citratus* exhibited significant inhibition from the first to the fifth day. Mycelial growth was found to be weak on the sixth and seventh days at low concentrations (0.15 and 0.25 mg/mL), but inhibition was complete at higher concentrations.

The effect of the ethanolic extract of *Xylopia aethiopica* on *A. niger* shows total inhibition of mycelial development from the first to the fourth day. Growth is found to be completely inhibited at high concentrations (1 and 2 mg/mL) from the fifth to the seventh day.

In the case of *Mucor sp.*, *Xylopia aethiopica*, the ethanolic extract demonstrated complete inhibition on the first two days. Inhibition is sustained at elevated concentrations (0.75, 1 and 2 mg/mL) on the ensuing days.

In the case of *Fusarium sp.*, the inhibition of the ethanolic extract of *Xylopia aethiopica exhibits complete inhibition at all concentrations and over the entire duration of the experiment*, from the first to the fourth day. It is evident that, with the exception of the highest concentrations, growth resumes partially from the fifth to the seventh day.

The findings indicate that the ethanol extracts of both plants demonstrate robust antifungal properties, frequently resulting in lethal or sustainable inhibition, particularly at the highest concentrations. In contrast, the aqueous extracts exhibit only marginal effectiveness.

## DISCUSSION

Phytochemical screening revealed that both plants are abundant in secondary metabolites including phenols, tannins, saponins and alkaloids. Quantitatively, *Xylopia aethiopica* has higher levels of tannins (44.35 ± 0.62 mgEqCat/gMS) and alkaloids (46.17 ± 0.67 mgEqQui/gMS) than *Cymbopogon citratus.* These classes of compounds, most notably polyphenols (phenols and tannins), are widely recognised for their antifungal properties, which arise from the disruption of the cell membrane of fungi.

A thorough analysis of the results unequivocally demonstrates the antifungal potential of the plant species studied, thereby highlighting a general inhibitory effect on the growth of the isolated fungal genera. However, it should be noted that the efficacy of this method is contingent upon the concentration of the

extract employed, as well as the type of extraction solvent utilised.

The effectiveness of the ethanolic extract on fungal growth varies depending on the concentrations considered. Consequently, optimal inhibition is frequently only attained at elevated concentrations (Syed *et al.* 2018).

Concomitantly, other researchers, including Kaboré *et al.* (2007), have documented substantial inhibitory effects, including complete mycelial inhibition (100%) with *Cymbopogon citratus* extract, thereby validating the pronounced potential of this species as a natural antifungal agent.

The solvent employed for extraction exerts a substantial influence on the type and quantity of phytochemicals extracted and, consequently on the resulting biological activity.

The results obtained in this study are similar to those reported by Tiendrebeogo *et al.* (2017) on *C. citratus*, as well as the observations of Chidozie *et al.* (2024), which tend to indicate that the ethanolic extract is often the most fungitoxic.

*Xylopia aethiopica*, for instance, exhibited the most pronounced fungitoxic activity with the ethanolic extract, exhibiting the capacity to inhibit the majority of the fungal flora examined (Chidozie *et al.*2024).

This phenomenon is attributed to the capacity of ethanol to extract a broader spectrum of secondary metabolites, including polyphenols, flavonoids, terpenes, and alkaloids. These secondary metabolites are frequently the active ingredients responsible for antifungal activity and are less soluble in water.

The low activity observed with aqueous extracts can be attributed to the fact that water is predominantly responsible for the extraction of polar compounds. In the event that the key antifungal compounds are not very polar, it can be hypothesised that the aqueous extract may prove to be less active than hydroalcoholic extracts. A substantial body of research has demonstrated that ethanol extracts exhibit superior antifungal properties in comparison to aqueous extracts (Petrasch *et al.* 2019).

In contrast in to the results reported by Chidozie *et al.* (2024), the inactivity observed in the present study may also be attributable to experimental factors, such as the extraction method, the specific plant part utilised, and the fungal strain employed. Alternatively, these outcomes could be attributed to geographical and seasonal variations in the plant's metabolites.

The antifungal effect of *C. citratus* on pathogenic species such as *Aspergillus niger* is well established (Syed *et al.* 2018), thus supporting the hypothesis that this plant can be used as a natural antifungal agent. C. *citratus* essential oil has also been shown to be very effective against A. *niger*, A. *flavus* and P. *digitatum* (Bakarnga-Via *et al.* 2022).

The phenomenon of C. citratus's occasional ineffectiveness, particularly in the context of aqueous extracts, can be attributed to the inherent resilience of moulds. It has been established that microorganisms possess the capacity to develop defence mechanisms against natural substances (efflux of compounds, modification of the cell wall). Consequently higher minimum inhibitory concentrations (MICs) are required to ensure effective control.

## CONCLUSION

The study demonstrated that both plants are rich in phenols, tannins, saponins and alkaloids. Extracts of *Xylopia aethiopica* and *Cymbopogon citratus* have been identified as promising natural sources for controlling fungal biodeterioration of *charmout*. The antifungal activity exhibited by these samples was found to be strongly correlated with the extraction solvent employed. The ethanolic extract demonstrated greater efficacy in solubilising secondary metabolites, resulting in complete and prolonged inhibition of moulds (A. *niger, Mucor sp., Fusarium sp.)*. The essential oil of *Cymbopogon citratus*, whose activity is corroborated by its lipophilic nature, appears to be the most relevant agent for biopreservation. The results of the study indicate the necessity of employing concentrated, lipophilic matrices to achieve a sustained fungicidal action capable of controlling mycelial growth on *charmout.*

## Declarations

# Author Contribution

A.B et C ont écrit le texte, D.E et FGHI ont préparé les tableaux et les figures. Tous les auteurs ont écrit le texte du manuscrit

## References


1. **A. Tiendrebeogo, I. Ouedraogo, S. Bonzi and A.I. Kassankogno** Study of the antifungal activity of extracts of *Cymbopogon citratus* (DC.) Stap, Eclipta alba L., Lippia multiflora M. and Agave sisalana P. *Int. J. Biol. Chem. Sci. 11(3): 1202-1211, June 2017.*
2. **Abney K. Pendey, Mahesh K. Samota, Qbhishek Kumar, Ana Sanches Silva, Nawal K. Dubey. (**2023) Fungal mycotoxins in food: Current status and future prospects. *Front. Sustain. Food Syst., Sec. Food Safety volume 76 2023 https:// doi.org/10.3389/fsufs.2023.1162595*
3. **Agaie; Onyeyili; Muhammad; Landan. 2007.** Some toxic effects of aqueous leaf extract of *Anogeissus leiocarpus* in rats. Journal of Pharmacology and Toxicology, 2: 396–401.
4. **Akakpo, A. Y., Kabore, D., Somda, M. K., Sawadogo, A., Mihin, H. B., Semde, Z., & Ouattara, A. S. (2023)**. Potential antifungal activity of essential oil from *Cymbopogon citratus* (DC) Stapf and its performances as biostabilisation of tiger nut milk (Cyperus exculentus). *Scientific African, 19.* https://doi.org/10.1016/j.sciaf.2022.e01497
5. **Al-Worafi, Y.M.** Herbal medicines safety issues. In Drug Safety in Developing Countries; Academic Press: Cambridge, MA, USA, 2020; pp. 163–178.

6. **Bakarnga-Via Issakou, Espérance, L., Walbang Ossoga, G., Hissein Ousman, A., Béral Valéry, M., Tidjani, A., & Chantal, M. (2022)**. Antifungal activity of *Cymbopogon citratus* (DC) Stapf (Poaceae) essential oil: Case of Moulds Isolated from Smoked and Dried Fish in the Markets of N'Djamena. *HEALTH SCIENCES AND DISEASE*. 23, 12 (Dec. 2022). DOI:https://doi.org/10.5281/hsd.v23i12.4080.
7. **Chidozie Azubuike A.**, **Chibuzo-Ogelle C.P., Achukbu Adaeze N., Ogbuozobe Okwudili Gabriel and Nwakuche Adaugo, Ozioma**. Control of Yam Spoilage Fungi Using *Xylopia aethiopica* and *Monodora myristica* Seed Extracts. *Asian J. Plant Pathol., 18 (1): 59-66, 2024.*
8. **CLSI** (Clinical and Laboratory Standards Institute), 2002, Reference method for broth dilution antifungal susceptibility testing of conidium-forming filamentous fungi. Wayne (Approved Standard M38-A)
9. **COMPAORE H, GUENNE S, DIANDE T, PARE A, SAMA H, BAMBARA A, RATONGUE H, TRAORE L, SAWADOGO-LINGANI H, HILOU A, KIENDREBEOGO M.** (**2021)** Evaluation of the antibacterial activity of the aqueous extract of Vachellia nilotica (L) PJH Hurter et Mabb pod powder on multidrug-resistant bacteria, phytochemical screening and quality control. *SNA [Internet]. 27 Nov. 2023 [cited 14 Nov. 2025];40(2):114-26. Available from: https://revuesciences-techniquesburkina.org/index.php/sciences_naturelles_et_appliquee/article/view/1050 vol-40 No. 2 (2021): July-December*
10. **Da O, Coulibaly MT, Ouédraogo J, Yaro B, Yerbanga R, Kini F, Koama B, Dakuyo Z, Nikiema J, Ouédraogo G. 2015.** Phytochemical screening of Saye, a traditional herbal remedy for malaria. *International Journal of Biological and Chemical Sciences, 9*(6): 2940-2946. DOI: 10.4314/ijbcs. v9i6.33.
11. **Edeoga, H. O., Okwu, D. E., & Mbaebie, B. O. (2005).** Phytochemical constituent of some Nigerian medicinal plants. African Journal of Biotechnology, 4(7), 685-68. DOI: 10.5897/AJB2005.000-3127.
12. **Harborne, J. B. (1973).** Phytochemical Methods: A Guide to Modern Technique of Plant Analysis. Ed. Chapman and Hall, London, 107 150.
13. **Kaboré B, Koita E, Ouedraogo I, NEBIE R. 2007**. "Efficacy of local plant extracts in seed treatment against mycology." Technique, 1(1): 49-57.
14. **Kiełtyka-Dadasiewicz, A.; Esteban, J.; Jabłonska-Trypuc**, A. Antiviral, Antibacterial, Antifungal, and Anticancer Activity of Plant Materials Derived from *Cymbopogon citratus* (DC.) Stapf Species. *Pharmaceuticals* **2024**, *17*, 705. https://doi.org/10.3390/ph17060705.
15. **Koffi, A. J, Bla, K. B., Yapi, H. F., Bidie, A. P., & Djaman, A. J. (2015). Phytochemical** Screening of Some Medicinal Plants in Côte D'ivoire and Evaluation of their Extraction Efficiency. International Journal of Pharmacognosy and Phytochemical Research, 7(3), 563–569.
16. **Maloba MJ, Mbayo KM, Monga MM, Dikala OF, Kanda KJ, Muamba ML, Ngoy KE, Lumbu SJ, 2025.** Ethnobotanical survey of medicinal plants used in the treatment of human fungal infections in Lubumbashi and its surroundings (DR Congo). *J. Appl. Biosci. Vol: 206: 21838–21851.*
17. **Okigbo RN and Ramesh P, 2008.** Effects of plants and medicinal plant combinations as anti-infectives. *Afr. J. Pharma.* 2(7): 130.
18. **Ouédraogo B, Yoda J, Kini BF, Koala M, Yaro A, Bonzi-Coulibaly Y. 2019.** Phytochemical screening and in vitro antioxidant study of six plants used for the treatment of hypertension in traditional medicine. *World Journal of Pharmacy and Pharmaceutical Sciences, 8*(**4**): 1665-1678. DOI: 10.20959/wjpps20194-13559.
19. **Petrasch S., Knapp S.J., Kan J., Blanco-Ulate B. Grey (2019)** Mould strawberry, a devastating disease caused by the ubiquitous necrotrophic fungal pathogen Botrytis cinera. *Molecular Plant Pathology. 20:877-892. Ahon, G. M., Ackah, J. A. A. B., Golly, K. J., Kra, A.*
20. **Saggar, S.; Mir, P.A.; Kumar, N.; Chawla, A.; Uppal, J.; Kaur, A.** Traditional and Herbal Medicines: Opportunities and Challenges. Pharmacogn. Res. 2022, 14, 107–114.
21. **Senhaji, O., Faid, M., Elyachioui, M., & Dehhaoui, M. (2005)**. Study of the antifungal activity of various cinnamon extracts. *Journal of Medical Mycology*, *15*(4), 220–229. https://doi.org/10.1016/j.mycmed.2005.07.002.
22. **Skandamis P.N., Nychas G.J.E, (2001)**, Effect of oregano essential oil on microbiological and physico-chemical attributes of minced meat stored in air and modified atmospheres. Journal of Applied Microbiology, 91(6), 1011-1022p
23. **Syed Nyamath and Karthikeyan B. (2018)**. *In vitro* antifungal activity of lemongrass (*Cymbopogon citratus*) leaf extracts. *Journal of Pharmacognosy and Phytochemistry*, **7**(3), 1163-1166.
24. **Wagner, H. (1983).** Drug analysis, thin-layer chromatographic analysis of medicinal drugs. Springer Verlag Berlin Heidelberg New York, 522 p.
25. **World Health Organisation**. WHO Guidelines for Assessing Quality of Herbal Medicines with Reference to Contaminants and Residues. 5 June 2007. Available online: https://www.who.int/publications/i/item/9789241594448 (accessed on 15 November 2023).

## Figures

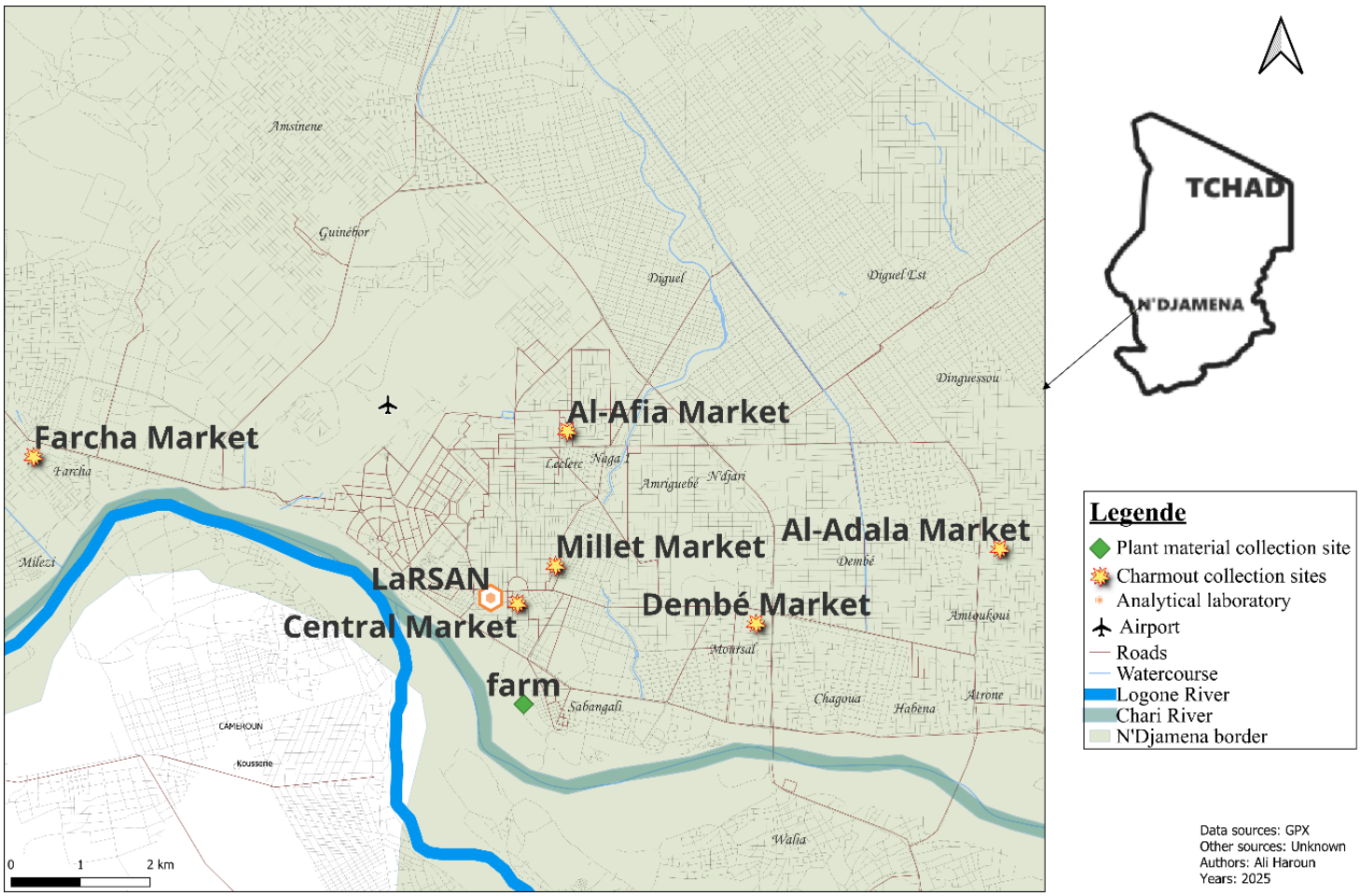


**Figure 1**

Study area

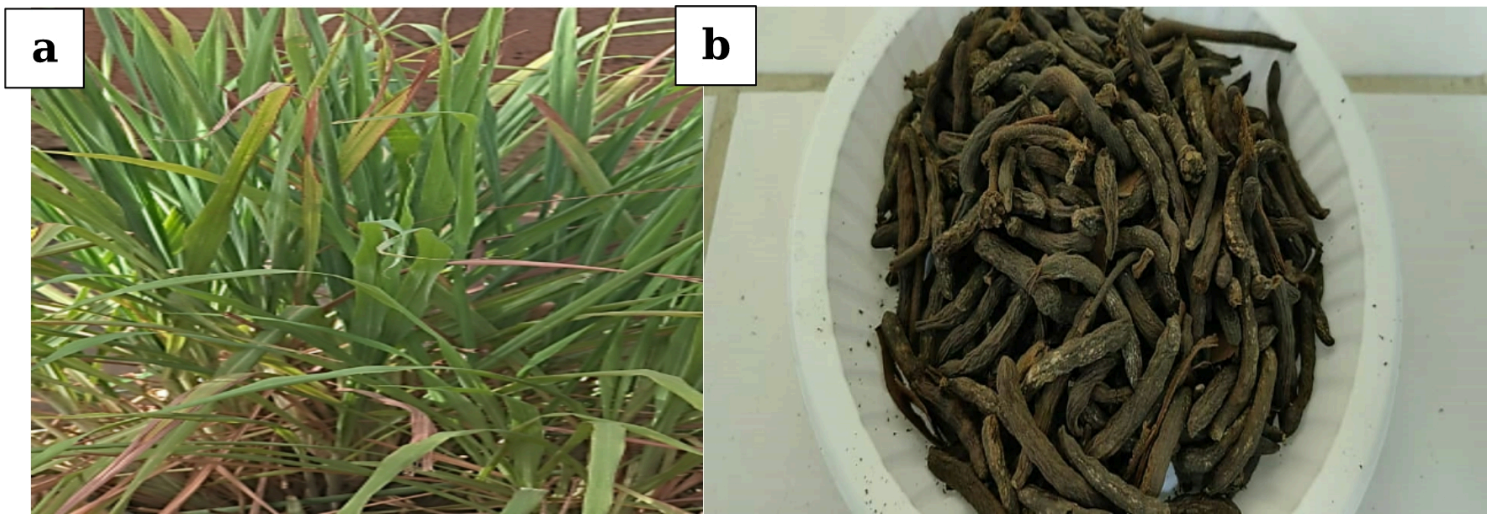


**Figure 2**

(a) Leaves of *Cymbopogon citratus* and (b) Fruits of *Xylopia aethiopica*

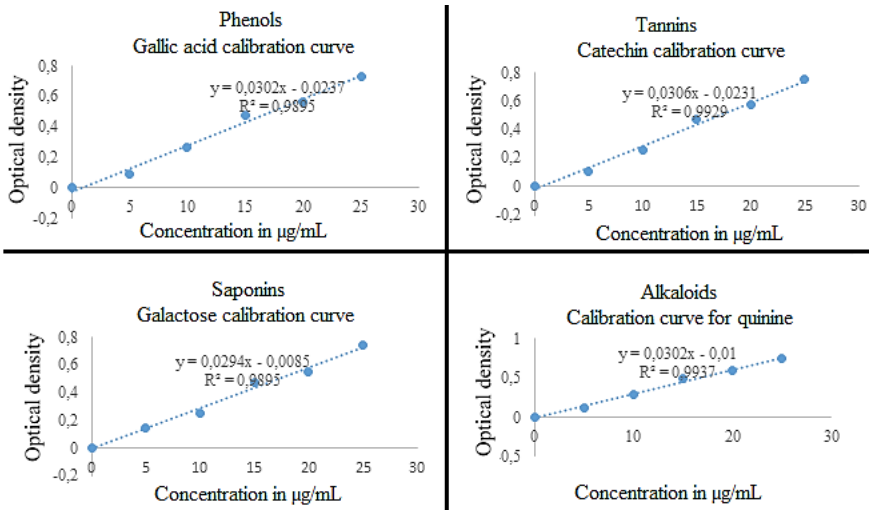


Figure 3

Calibration curves for secondary metabolites of *Xylopia aethiopica* and *Cymbopogon*

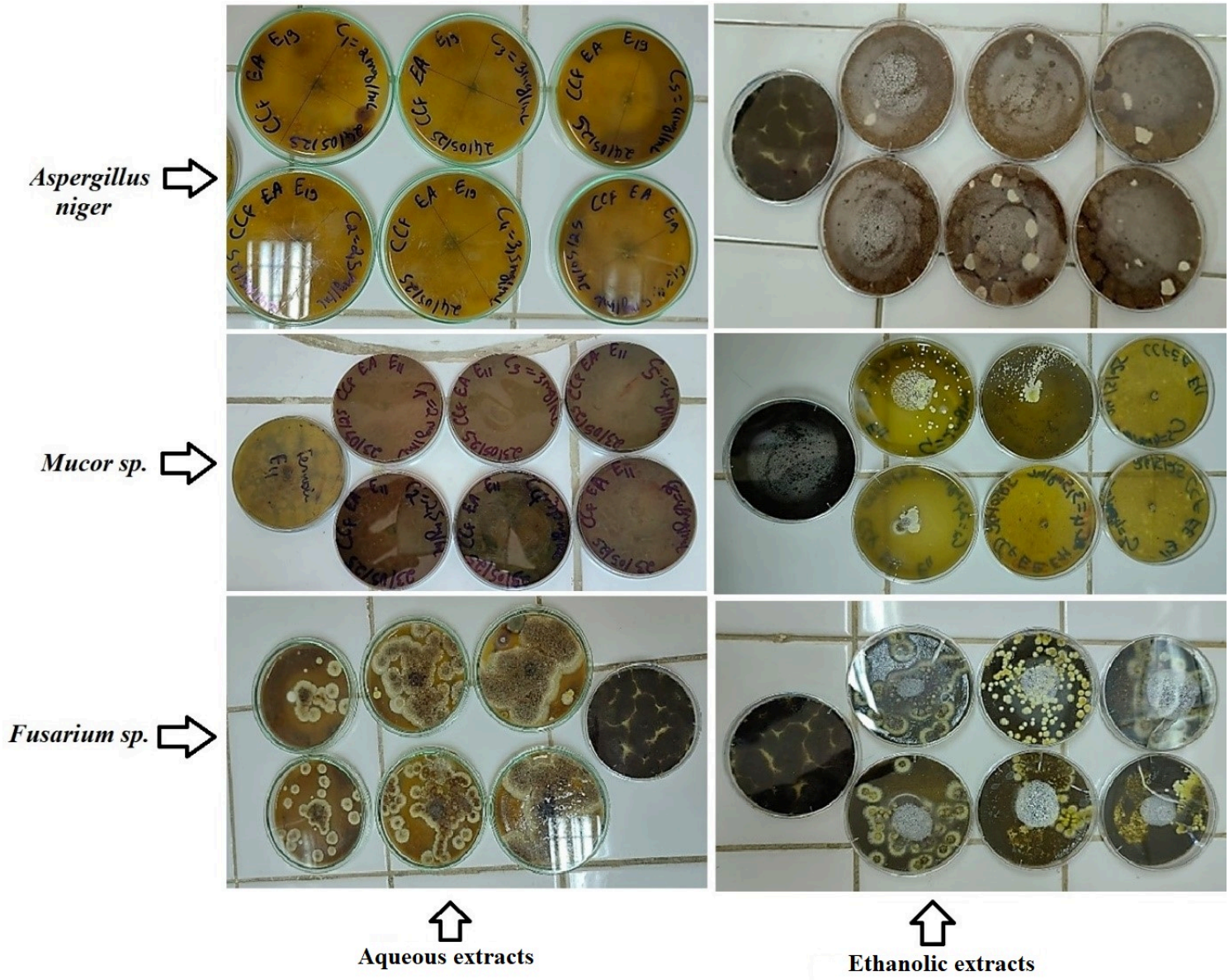


Figure 4

Antifungal activity of crude extracts of *Cymbopogon citratus*

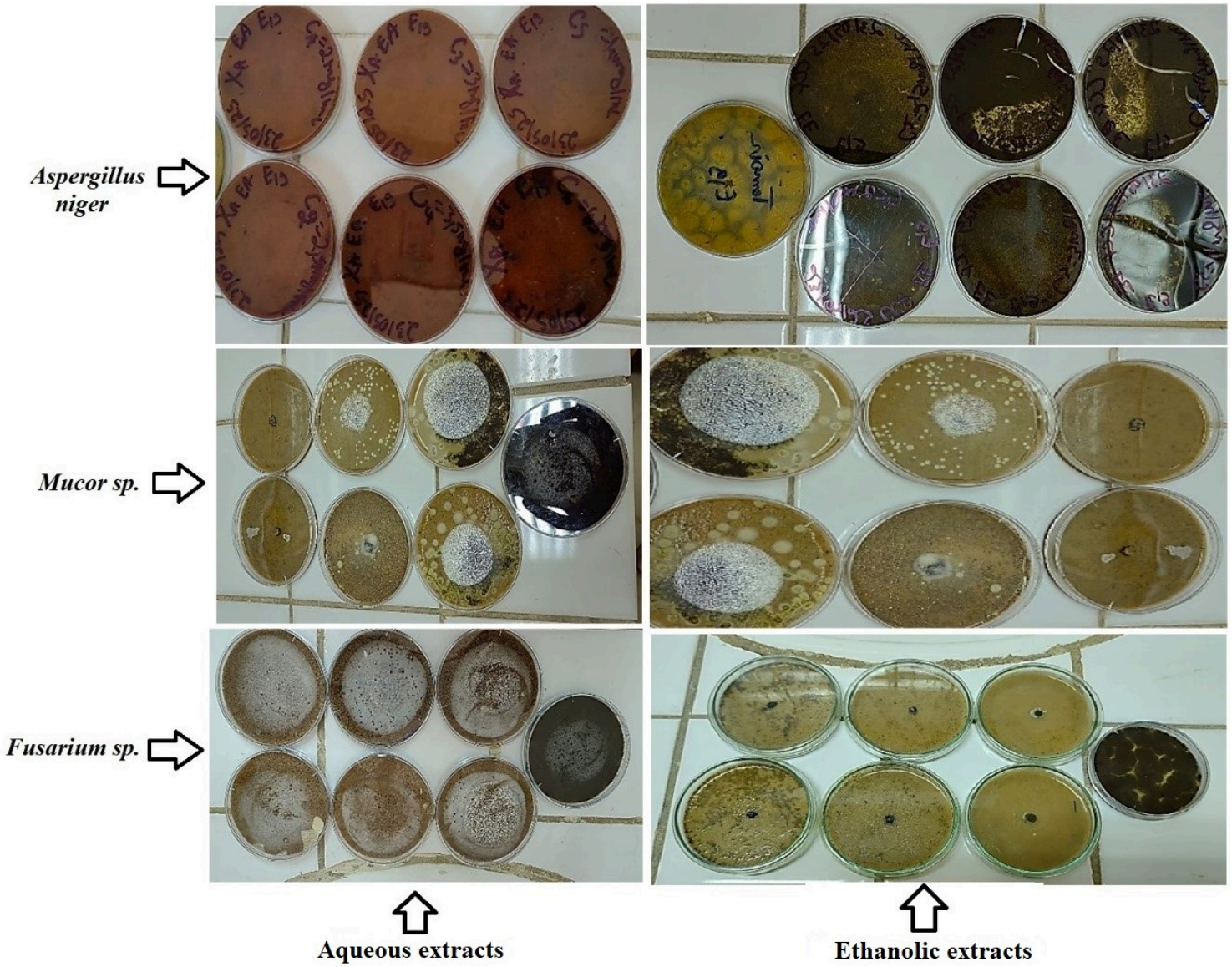


Figure 5

Antifungal activity of crude extracts of *Xylopia aethiopica*