ISOLATION AND CHARACTERIZATION OF FUNGI FROM FRIED BOTTLED GROUNDNUTS SOLD IN BENIN CITY

BY

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DEPARTMENT OF PLANT BIOLOGY AND BIOTECHNOLOGY
FACULTY OF LIFE
UNIVERSITY OF BENIN, BENIN CITY SCIENCES
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OCTOBER, 2025.

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A PROJECT REPORT SUBMITTED TO THE DEPARTMENT OF PLANT BIOLOGY AND BIOTECHNOLOGY, FACULTY OF LIFE, UNIVERSITY OF BENIN, BENIN CITY SCIENCES IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR THE AWARD OF BACHELOR OF SCIENCE (HONOURS) DEGREE (BSC.) IN PLANT BIOLOGY AND BIOTECHNOLOGY

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OCTOBER, 2025.

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Head of Department	Signature /Date	Email

CERTIFICATION

This is to certify that this project work was carried out by Tolulope Samuel AROWOSAFE in the Department of Plant Biology and Biotechnology, Faculty of Life Sciences, University of Benin, Benin City, Nigeria.

Signed: ---------- ----------

Prof. (Mrs) F.I. OKUNGBOWA Date

(Project Supervisor)

Signed: ---------- ----------

Prof. B. IKHAJIAGBE Date

(Head of Department)

DEDICATION

This work is dedicated to Almighty God for his undying love and care for me and my parents for their care, love and support towards me.

ABSTRACT

Chemical preservatives are commonly used in food preservation, but rising safety concerns have shifted attention toward natural alternatives such as ginger (*Zingiber officinale*), a spice known for its antimicrobial and antioxidant properties. This study aimed to isolate and characterize fungi associated with fried bottled groundnuts sold in Benin City, and to evaluate the antimicrobial activity of aqueous ginger extract at varying concentrations (20 g/mL, 40 g/mL, and 60 g/mL) against the isolates. Groundnut samples were collected from three markets (New Benin and Ring Road) and June 12 (a commercial hub) inoculated on Potato Dextrose Agar, and subjected to microscopic and cultural identification. Pure cultures of *A. niger* and mould (*A.clavatus*) were obtained and treated with ginger extract using agar well diffusion, and zones of inhibition were measured. Results showed that ginger extract had significant antifungal activity, with inhibition zones generally increasing with higher concentrations. *A. niger* isolates from New Benin and Ring Road samples exhibited a clear dose-dependent response, while the June 12 isolate showed optimal inhibition at 40 g/mL. Mould isolates from June 12 and Ring Road responded consistently, with the highest inhibition recorded at 60 g/mL (26.0 mm and 24.3 mm, respectively). The overall trend confirmed that 60 g/mL ginger extract had the strongest inhibitory effect (mean inhibition: 37%), supporting its concentration-dependent efficacy. The findings confirm that ginger extract possesses promising antifungal activity against common contaminants of fried bottled groundnuts and can serve as a safe, affordable, and locally available natural preservative. This is important for enhancing food safety, lowering spoilage, and reducing aflatoxin health risks in areas lacking advanced storage options. Further studies should improve extraction methods, use larger samples, and investigate combining this with other natural agents for sustainable food preservation.

ACKNOWLEDGEMENTS

Firstly I want to appreciate God Almighty for his unlimited grace upon my life throughout the duration of my project. It wouldn't have been possible without Him. In addition, I want to thank my Supervisor Prof. (Mrs) F.I Okungbowa for her wisdom, knowledge and guidance. May God really bless you, ma. Special thanks go to the Head of Department, Prof. B. Ikhajiagbe. I am also grateful to all the lecturers and the entire staff of the Department of Plant Biology and Biotechnology at the University of Benin for providing me with the knowledge and resources I needed throughout my academic development.

My deepest gratitude goes to my parents, Mr. and Mrs. Sesan Arowosafe, my siblings, Jummy and Tobi for their love, care, advice and support throughout this period. I could not have done this without you both.

To my friends amongst whom are Praise, Jane, Jonathan and Dpaulz, I love you all.

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INTRODUCTION

1.1 GROUNDNUT

Groundnut (*Arachis hypogaea*), commonly known as peanut, is a major source of protein and oil globally and forms a staple snack in many parts of Nigeria (Adebiyi *et al*., 2016). Fried bottled groundnuts are widely consumed for their taste, convenience, and shelf-life; however, poor processing and storage can lead to contamination by fungi such as *Aspergillus* species, *Penicillium*, *Rhizopus*, and *Fusarium*, which are well-documented contaminants of groundnuts and can produce mycotoxins like aflatoxins which are potent carcinogens (Ezekiel *et al*., 2012). Fungal contamination frequently occurs during post-harvest handling, storage, and marketing, especially under the high humidity and temperatures typical of tropical regions (Klich, 2007).

1.1.1 Uses of Groundnut

Groundnuts are remarkably versatile. They serve as a plant-based protein source and are used in peanut butter, roasting, and confectionery (Withinnigeria.com, 2024). They are also pressed for oil utilized in cooking, cosmetics, biodiesel, soaps, lubricants, and even nitroglycerin production. The residual cake is processed into animal feed. Additionally, shells are repurposed into mulch, fuel, building materials, and artisanal crafts (FeasibilityReportsInNigeria, 2024; Withinnigeria.com, 2024).

Culinary uses in Nigeria include peanut soup, *kuli-kuli*, peanut butter, and snacks especially in street food culture as well as sweet treats and local sauces (Dutable.com, 2024; AgricultureNigeria.com, 2024).

1.1.2 Groundnut- Producing Areas and Economic Contribution in Nigeria

Groundnut production is concentrated in the northern and central States like Kano, Katsina, Kaduna, Jigawa, Sokoto, Zamfara, Kebbi, Bauchi, and parts of Benue, Plateau, Nasarawa, and Kwara where favorable agro-ecological conditions prevail (Ajeigbe *et al*., 2014). Historically, groundnuts accounted for about 70% of Nigeria's non-oil export earnings (1956–1967), symbolized by the iconic groundnut pyramids of Kano (Ajeigbe *et al*., 2014; Wikipedia-Groundnut pyramids, 2023).

Currently, Nigeria remains a key global producer, contributing around 5% of world production (2 million metric tons), and is Africa's top producer. Groundnut output has risen to over 5 million metric tons as of 2024, spurred by industrial demand for products like oil, butter, snacks, and confectionery (BusinessDay NG, 2024; MSME Africa, 2024). For many small-scale farmers in States like Benue and Niger, groundnut production contributes 23% of household cash revenue and is a profitable enterprise (MyProject.ng, 2024; Baba *et al*., 2022).

1.1.3 Geographical Distribution of Groundnut (Africa and Global)

Globally, groundnuts are cultivated across nearly 100 countries in both tropical and subtropical regions between approximately 40° N and 40° S latitude (FAO manual, 2020). Africa has become a secondary center of diversity despite the crop's South American origin (Infonet Biovision, 2019). In Nigeria, production encompasses both Sahelian and savanna zones ranging from semi-arid to more humid environments suitable for groundnut cultivation.

1.1.4 Cultivation and Growth Requirements of Groundnut

Groundnuts thrive best in light, sandy loam or sandy clay loam soils with good drainage, moderate acidity (pH ~5.9–7), and optimal germination temperatures around 30 °C (AgricultureNigeria.com, 2024; TheJunction.ng, 2024). They require a warm growing season of 75–150 days with rainfall or irrigation of approximately 500–1,200 mm (FAO manual, 2020; TheJunction.ng, 2024). These conditions allow for yields between 300–1,000 kg/ha typically, with well-managed farms yielding up to 5 t/ha (TheJunction.ng, 2024).

1.1.5 Use of Natural Antimicrobial Agents for Food Preservation

Due to rising concerns over the health implications of chemical preservatives, there is growing interest in natural antimicrobial agents in food safety (Prakash *et al*., 2013). Ginger (*Zingiber officinale*) is widely recognized for its antimicrobial, antioxidant, and medicinal properties (Singh *et al*., 2008). Its extracts contain bioactive compounds such as gingerol, shogaol, paradol, and

zingerone, which have demonstrated significant antifungal activity (Ali *et al*., 2008). Recent in vitro studies have further confirmed ginger's antifungal and antibiofilm effects against *Candida albicans* and *C. krusei* at concentrations ranging from 0.625–5 mg/mL (PubMed study, 2016). Therefore, investigating ginger extract's antimicrobial activity against fungal isolates from fried bottled groundnuts could offer a promising natural alternative for enhancing food safety.

1.2 LITERATURE REVIEW

Groundnuts are highly susceptible to fungal invasion, particularly by *Aspergillus* species, due to their high oil and protein content that foster fungal growth (Bankole and Adebanjo, 2003). In Nigeria, *Aspergillus niger*, *A. flavus*, *Penicillium*, and other moulds are commonly identified in stored groundnuts (Ezekiel *et al*., 2012). Contamination is often linked to poor drying, humid storage, and inadequate packaging (Ogar *et al*., 2015). Mycotoxins especially aflatoxin B_1 pose serious health risks, including liver cancer, immune suppression, and stunted growth in children (Wild and Gong, 2010).

1.2.1 Antimicrobial Property of Ginger

Ginger (*Zingiber officinale*) is a tropical plant whose rhizome is extensively utilized as a spice and traditional remedy. Its antimicrobial effects are attributed to phenolic compounds including gingerols, shogaols, paradols, and zingerone (Ali *et al*., 2008). Research has repeatedly demonstrated ginger's inhibitory effects on various bacteria and fungi, including foodborne pathogens (Prakash *et al*., 2013; Singh *et al*., 2008). Ginger extract has also shown promise as a natural preservative in food processing (Omoya and Akharaiyi, 2012). More recent studies have confirmed its antifungal and antibiofilm efficacy against *Candida* species at low concentrations (PubMed study, 2016).

1.2.2 Mechanisms of Antimicrobial Action

The antimicrobial action of ginger is linked to its ability to disrupt microbial cell membranes, interfere with enzyme systems, and inhibit toxin production (Nanasombat and Lohasupthawee, 2005). For fungi, ginger extracts have been shown to inhibit spore germination and hyphal growth (Adegoke and Gopalakrishnan, 1998).

1.2.3 Antifungal Efficacy of Ginger Extract

Previous studies confirm ginger's antifungal activity against various species, including *Aspergillus niger* and other moulds commonly found in food (Ekwenye and Elegalam, 2005; Singh *et al.,*

2008). Ekwenye and Elegalam (2005) demonstrated that ethanolic ginger extract inhibited the growth of *A. niger* and *Penicillium* species isolated from food samples.

Given its bioactive compounds, ginger extract has demonstrated strong antifungal effects against foodborne fungi. Investigating its effect on fungal isolates from fried bottled groundnuts could offer a practical, safe, and locally accessible means of reducing fungal contamination and extending shelf life (Prakash *et al.,* 2013).

Several studies have isolated and characterised fungi from groundnuts sold in open markets, revealing the prevalence of *Aspergillus niger*, *A. flavus*, *Penicillium* species, and other storage fungi (Ezekiel *et al.,* 2012; Ogar *et al.,* 2015). These fungi not only reduce the quality of groundnuts but also pose significant health risks due to mycotoxin production.

1.2.4 Public Health Implications of Consuming Fungi-contaminated Groundnut

The health implications of consuming fungi-contaminated groundnuts are significant, particularly due to aflatoxins. Therefore, using natural antifungal agents such as ginger extract can contribute to safer food preservation, especially in regions with limited access to modern storage facilities (Bankole and Adebanjo, 2003).

1.3 JUSTIFICATION OF STUDY

Despite the known risks of fungal contamination in groundnuts and the proven antimicrobial potential of ginger extract, there is limited research specifically targeting fried bottled groundnuts sold in Benin City. Therefore, research is needed to identify and characterize the dominant fungal contaminants and assess the effectiveness of ginger extract.

1.4 AIM/OBJECTIVES

Aim

To isolate and characterise fungi from fried bottled groundnuts sold in Benin City and to investigate the antimicrobial activity of ginger extract against the isolates.

Objectives

- To isolate fungal contaminants from fried bottled groundnuts from different markets in Benin City.
- To identify and characterise the isolated fungi using cultural characteristics (colony colour, texture, growth pattern) and microscopic features (spore shape, hyphae type, reproductive structures).
- To prepare ginger extract and assess its antimicrobial activity against the isolated fungi.
- To determine the optimal concentration of ginger extract for effective inhibition.

CHAPTER TWO

MATERIALS AND METHODS

2.1 Sample Collection

Groundnut samples were collected from three locations in Benin City: New Benin, June 12, and Ring Road.

2.1.2 Preparation of Potato Dextrose Agar (PDA)

Forty grams (40 g) of Irish potatoes were peeled, weighed, and placed in 200 ml of distilled water in a conical flask. The mixture was boiled for 5 minutes to extract the potato nutrients, then filtered through a fine sieve to remove solids and allowed to cool. Next, 4 g of agar powder and 4 g of dextrose were weighed and added to the potato extract. The solution was gently swirled to ensure thorough mixing and covered with aluminum foil plugged with cotton wool. It was then sterilized for 15 minutes in a manual autoclave. After sterilization, the Potato Dextrose Agar was allowed to cool to approximately 45–50°C before being poured aseptically into six sterile Petri dishes labeled with the collection sites of the groundnuts: New Benin, June 12, and Ring Road.

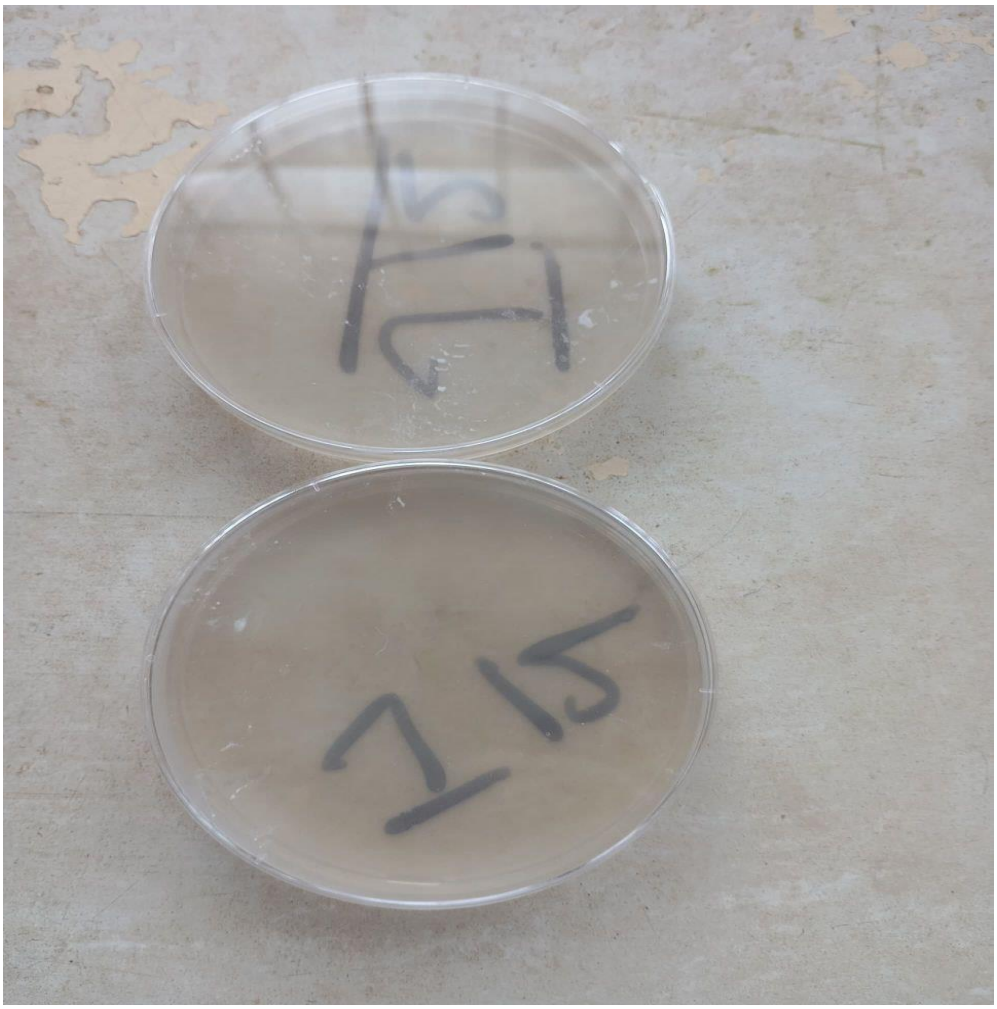

Plate 2.1: Potato Dextrose Agar (PDA) in Petri dishes

2.1.3 Preparation of Mixed Culture

Six groundnut grains were aseptically taken from each bottle and inoculated onto freshly prepared Potato Dextrose Agar (PDA) plates. These plates were incubated in a sterile environment at room temperature for three days to promote maximum fungal growth. By the third day, fungal colonies rapidly developed. The colonies exhibiting dark brown to black coloration indicated the presence of *Aspergillus niger*, while colonies with a greenish-whitish appearance suggested mould contamination.

Plate 2.2: Groundnut seeds inoculated on PDA to get a mixed culture

2.1.4 Preparation of Pure Cultures

Freshly prepared Potato Dextrose Agar (PDA) was used as the growth medium for fungal isolation. A sterile swab stick was used to carefully collect samples of each organism (*Aspergillus niger* and mould) from the mixed cultures on the different plates. The swab was then used to inoculate separate replicates of freshly prepared PDA plates. These inoculated plates were incubated for three days to allow for maximum fungal growth.

2.1.5 Microscopic Examination of Fungal Isolates

A clean drop of water was placed on a glass slide using a needle. A thin colony from the plate was then picked and transferred onto the slide. The slide was mounted on the microscope stage and initially examined under low power magnification (×40). After that, a drop of lactophenol cotton blue stain was added to the prepared slide, which was then covered with a coverslip. Upon closer observation at the same low power magnification, structures such as hyphae, mycelium, and a mass of hyphae were revealed.

2.1.6 Preparation of Ginger Extract

Twenty grams, forty grams, and sixty grams of ginger were weighed using a digital weighing balance. Each quantity was then chopped into smaller pieces and blended manually. After blending, each respective amount of ginger was transferred into sterile bottles labeled 20g, 40g,

and 60g. One hundred milliliters of distilled water was measured with a plastic measuring cylinder and added to each labeled bottle containing the ginger. The bottles were left undisturbed for 24 hours to allow fermentation of the ginger. On the following day, the ginger mixtures were sieved to separate the fermented liquid, which was then transferred into MacConkey bottles for further use.

Plate 2.3: Prepared Ginger Extract

2.1.7 Treatment of Isolated Organisms

Three replicates of pure cultures were prepared for each isolated organism (*Aspergillus niger* and mould). Each replicate was labeled with a permanent marker to indicate the locations and the concentration of ginger extract used: 20 mg/mL, 40 mg/mL, and 60 mg/mL. Using an 8 mm cork borer, wells were aseptically created in the agar of each replicate plate. Then, 1 mL of the respective ginger extract concentration was carefully introduced into each well using a sterile plastic pipette. The treated plates were then covered with a wooden net sprayed with sodium hypochlorite and incubated for two days to allow the ginger extract to exert its effect on the isolates.

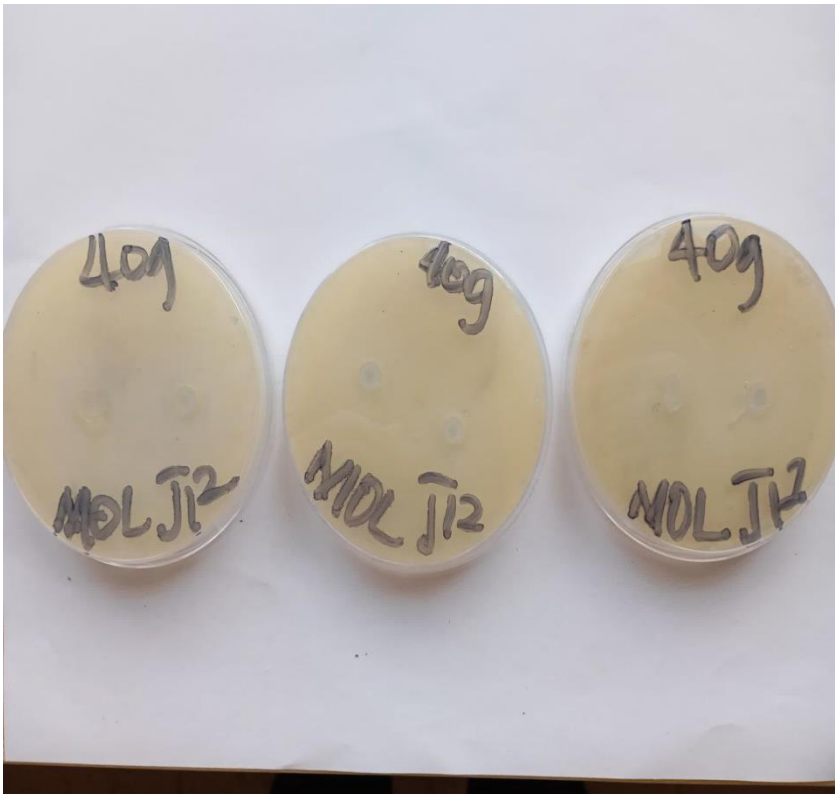

Plate 2.4: Replicate of Pure Culture of Mould

Measurement of zone of inhibition (mm)

Zone of inhibition produced by three concentrations of ginger extract: 20 mg/mL, 40 mg/mL, and 60 mg/mL was measured. Each treatment was replicated on pure cultures of *Aspergillus niger* and mould isolates from New Benin, June 12, and Ring Road. The mean inhibition zones were then used to compare the antimicrobial effect of the different concentrations.

2.2 Data Collection

Data were recorded during the incubation period, capturing the following variables:

- Source of groundnut samples.
- Types of fungal isolate (*Aspergillus niger* or mould).
- Concentration of ginger extract applied (20 mg/mL, 40 mg/mL and 60 mg/mL),
- Zone of Inhibition (mm) for each replicate.

2.3 Data Analyses

Mean zones of inhibition were calculated for each isolate, location and ginger extract concentration to summarise the antimicrobial effect.

Percentages were derived to illustrate the proportion of mean inhibition for each concentration.

CHAPTER THREE

RESULTS

This research explored the antimicrobial activity of ginger extract against isolates from fried bottled groundnuts in Benin City, focusing on two specific microbial types: *Aspergillus niger* and other moulds (filamentous fungi), across different sampling locations (June 12, New Benin, and Ring Road). The activity was measured as the zone of inhibition (mm) at three concentrations of ginger extract: 20 g/mL, 40 g/mL, and 60 g/mL. A pie chart also presented the mean zone of inhibition (%) by ginger extract concentration.

Plate 3.1 shows the growth of *Aspergillus niger* on Potato Dextrose Agar (PDA). The colonies appeared dark with dense mycelial growth and characteristic black conidial structures, confirming the presence of the fungus used for antimicrobial testing.

Plate 3.2 shows the pure cultures of the isolates. Plate A represents *Aspergillus niger*, characterized by dense dark mycelial growth, while Plate B represents mould (filamentous fungi) with lighter growth and distinct colony morphology.

Plate 3.3 presents the microscopic view of *Aspergillus niger* stained with lactophenol cotton blue, showing the fungal structures more distinctly. The microscopic examination revealed dichotomous branching of hyphae. The conidiophores, which are structures that bear asexual spores, were observed to be smooth-walled and darker towards the apex. The mycelium appeared black in colour and consisted of a filamentous, cottony mass of hyphae, while the mass of hyphae formed the mycelial colony with characteristic black conidial heads.

Plate 3.4 shows the microscopic view of branched *Aspergillus niger* hyphae. Hyphae showed dichotomous branching. The conidiophores, which are structures that bear asexual spores, were observed to be smooth-walled and darker towards the apex.

Plate 3.5 presents the prepared replicate of the pure culture of *Aspergillus niger*. The replicates were inoculated and incubated to confirm purity and reproducibility of growth, showing consistent morphological characteristics across the culture plates.

Plate 3.6 also presents the microscopic view of a long strand of mould hyphae. The general mould isolate was characterized by the following features: hyphae were observed as long, filamentous structures; the mycelium was a network of hyphae that formed a cottony mass; and the mass of hyphae appeared as a dense network.

The macroscopic features of the fungal isolates were also observed. For *Aspergillus niger*, the colony appeared black in colour, with a rough texture, flat elevation, and rapid growth rate. For the mould isolate, the colony exhibited a whitish colour, with a fluffy texture, raised elevation, and rapid growth rate.

The pie chart titled "Mean Zone of Inhibition (%) by Ginger Extract Concentration" provides an overview of the ginger extract's overall effectiveness across all isolates and locations. The 60 g/mL concentration showed the highest mean inhibition (36.7% or 37%), followed closely by 40 g/mL (33.5% or 33%) and 20 g/mL (29.8% or 30%). This general trend indicates that the antimicrobial activity of ginger extract increases with higher concentrations.

Figure 1 shows the bar chart for the inhibitory effect of ginger extract against *Aspergillus niger* from the June 12 location. The ginger extract exhibited inhibition, with the zone of inhibition being 18.2 mm at 20 g/mL, sharply increasing to 19.2 mm at 40 g/mL but then decreasing to 18 mm at 60 g/mL, suggesting an optimal concentration around 40 g/mL for inhibiting *A. niger* from this location (Table 3.1). Figure 2 presents the bar chart showing the antimicrobial activity against *A. niger* from New Benin. The ginger extract demonstrated a clear dose-response relationship, with the zone of inhibition starting at 17 mm at 20 g/mL, increasing to 18.5 mm at 40 g/mL, and further rising to 19.5 mm at 60 g/mL (Table 3.1), indicating that higher concentrations were more effective. Mould did not grow in New Benin because it was overshadowed by the *Aspergillus* isolate.

Figure 3 displays the bar chart for the inhibitory effect on *A. niger* isolated from Ring Road. The inhibition zones were 16.7 mm at 20 g/mL, 18 mm at 40 g/mL, and 18.5 mm at 60 g/mL (Table 3.1), similar to the New Benin isolate, showing a direct relationship between ginger extract concentration and inhibitory effect. Figure 4 shows the bar chart illustrating the effect on mould (filamentous fungi) isolated from June 12. For this mould, the ginger extract showed a consistent increase in inhibitory activity with rising concentration. The zone of inhibition was 16.8 mm at 20 g/mL, increasing to 23.2 mm at 40 g/mL, and reaching its highest at 26 mm at 60 g/mL (Table 3.1), indicating a dose-dependent inhibitory effect against this isolate. Figure 5 depicts the bar chart for the activity against mould from Ring Road. For this isolate, the ginger extract's activity also increased with concentration. The zone of inhibition was 16.8 mm at 20 g/mL, 18.2 mm at 40 g/mL, and peaked at 24.3 mm at 60 g/mL (Table 3.1), highlighting enhanced efficacy at higher concentrations against this mould.

The results, as illustrated in the bar charts, Table 3.1, and the accompanying pie chart, indicate that ginger extract possesses antimicrobial properties against both *Aspergillus niger* and moulds isolated from fried bottled groundnuts in Benin City. While a general trend of increasing inhibition with higher ginger extract concentrations was observed across most isolates, specific variations were noted. Notably, for *A. niger* from the June 12 location, the optimal inhibitory effect was observed at 40 g/mL, with a slight decrease at 60 g/mL. In contrast, for mould isolates from both June 12 and Ring Road, and *A. niger* from New Benin and Ring Road, the highest concentration (60 g/mL) consistently yielded the largest zones of inhibition. These findings suggest that ginger extract could be a potential natural antimicrobial agent for controlling microbial contamination in fried bottled groundnuts.

Table 3.1 Mean zone of Inhibition (mm) of Ginger Extract Against Isolated Fungi (*Aspergillus niger* and Mould) at Different Concentration and Location

Organism Isolated	Location	Mean of Zone of Inhibition (mm)		
		20g/ml	40g/ml	60g/ml
Aspergillus niger	New Benin	17.00	18.50	19.50
	June 12	18.20	19.20	18.00
	Ringroad	16.70	18.00	18.50
Mould	June 12	16.80	23.20	26.00
	Ring Road	16.80	18.20	24.30
	Mould	0.00	0.00	0.00
Average		86.20	97.10	106.30

Key to Parameters

g/mL – grams per millilitre (concentration of ginger extract)

mm – millimetres (diameter of zone of inhibition)

Plate 3.1: Growth of *Aspergillus niger* on PDA

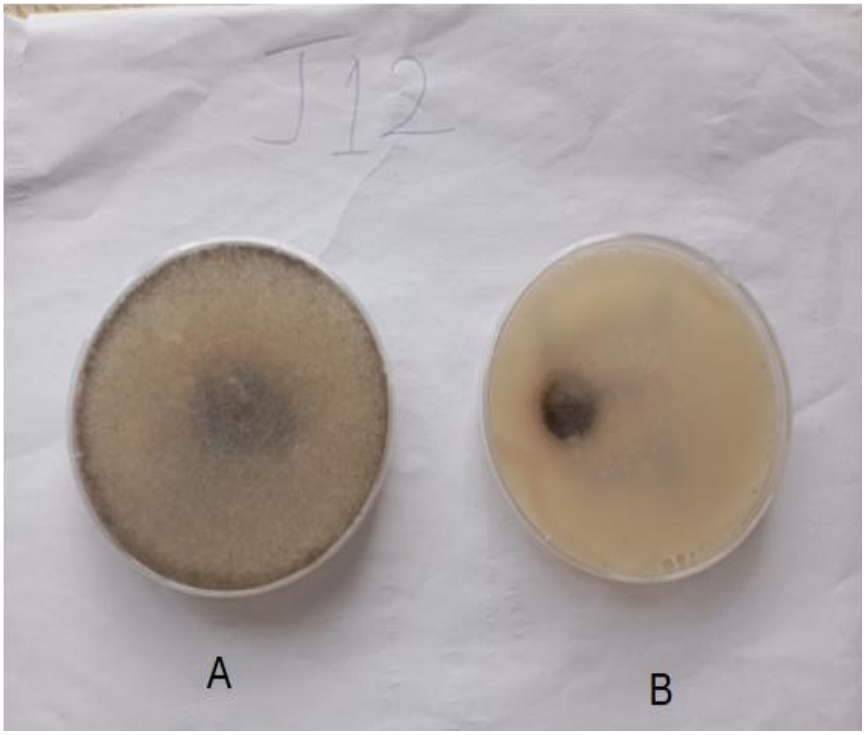

Plate 3.2: Pure Cultures of A**:** *Aspergillus niger* and B: Mould

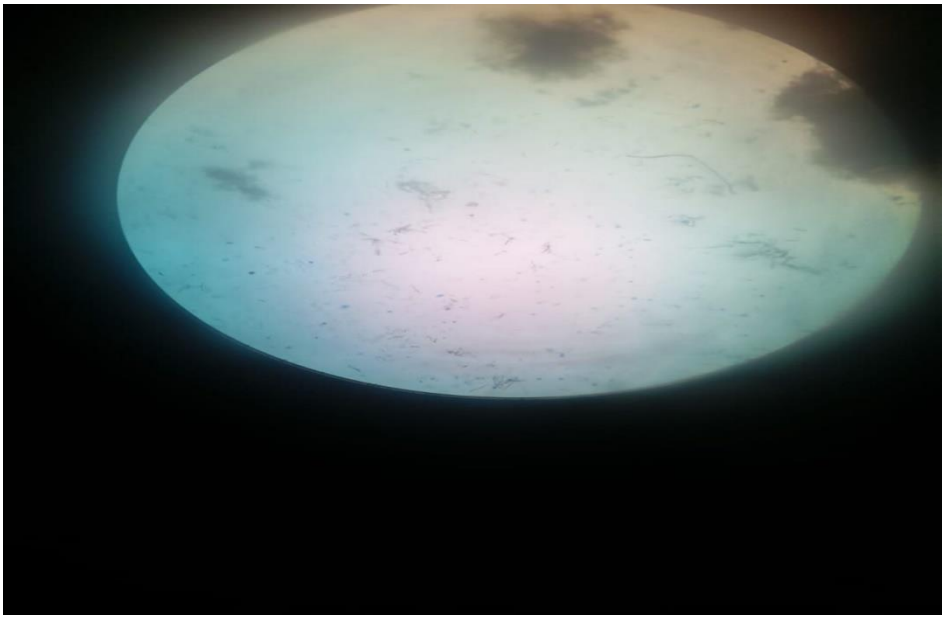

Plate 3.3:

Microscopic View of *Aspergillus niger* (Magnification = ×40)

Plate 3.4: Microscopic View of Branched *Aspergillus niger* Hyphae

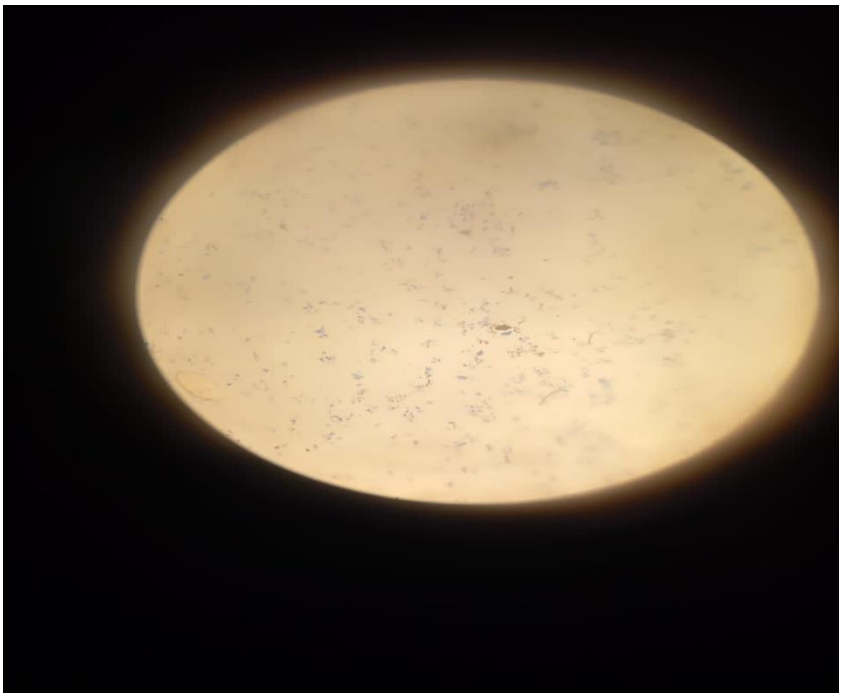

Plate 3.5: Microscopic View of Mould

Plate 3.6: Prepared Replicate of Pure Culture of *Aspergillus niger*

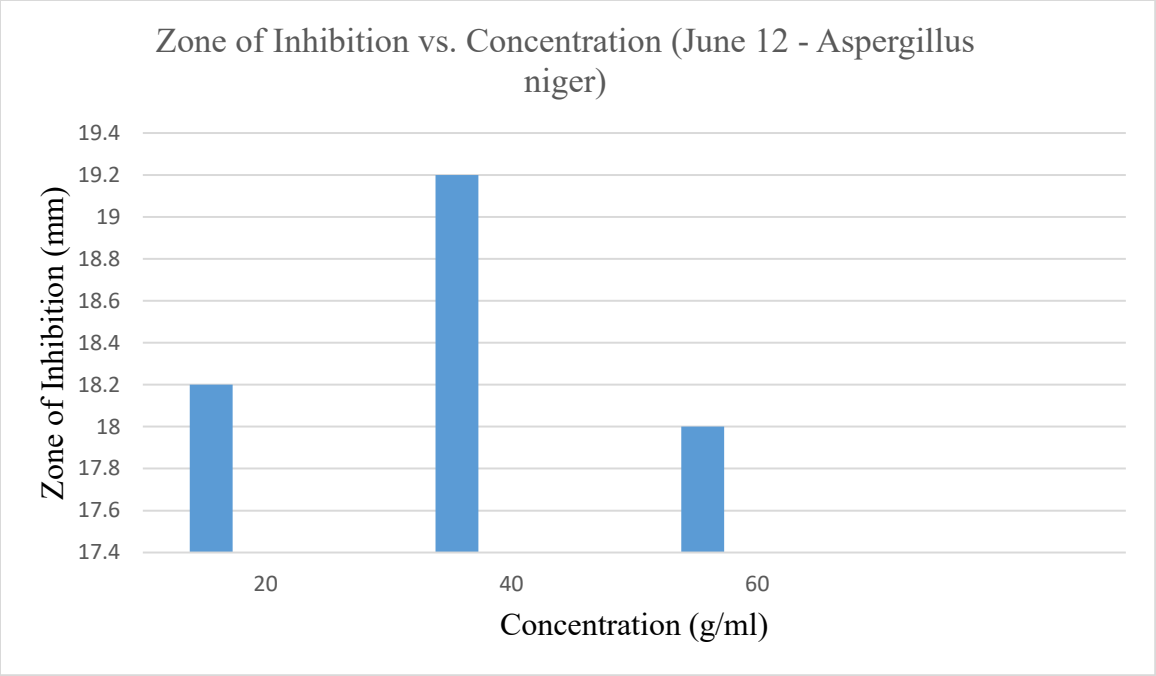

Figure 1: Bar graph representation of the Zone of Inhibition vs. Concentration (June 12 - *Aspergillus niger*)

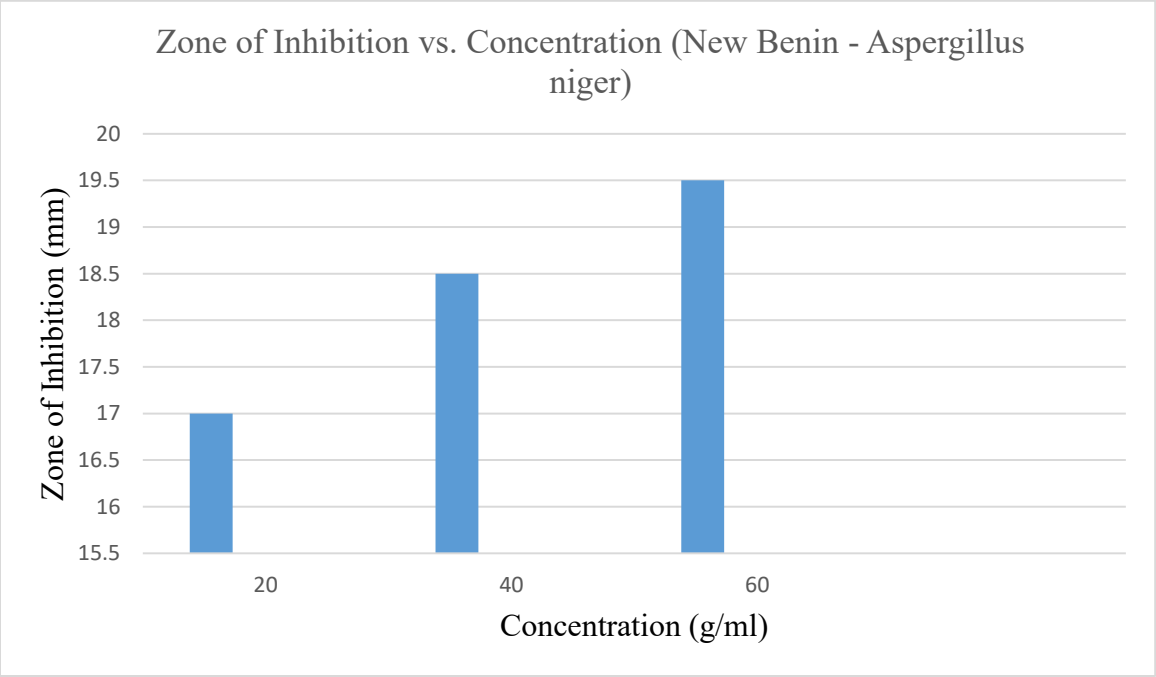

Figure 2: Bar graph representation of the Zone of Inhibition vs. Concentration (New Benin - *Aspergillus niger*)

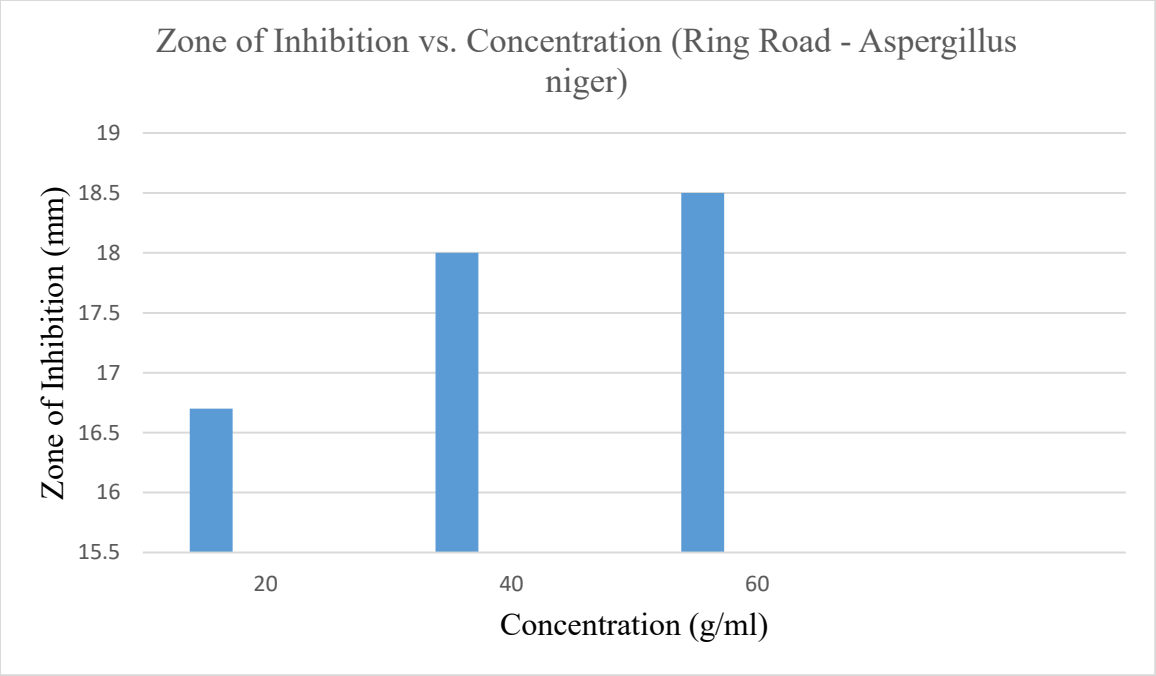

Figure 3: Bar graph representation of the Zone of Inhibition vs. Concentration (Ring Road - *Aspergillus niger*)

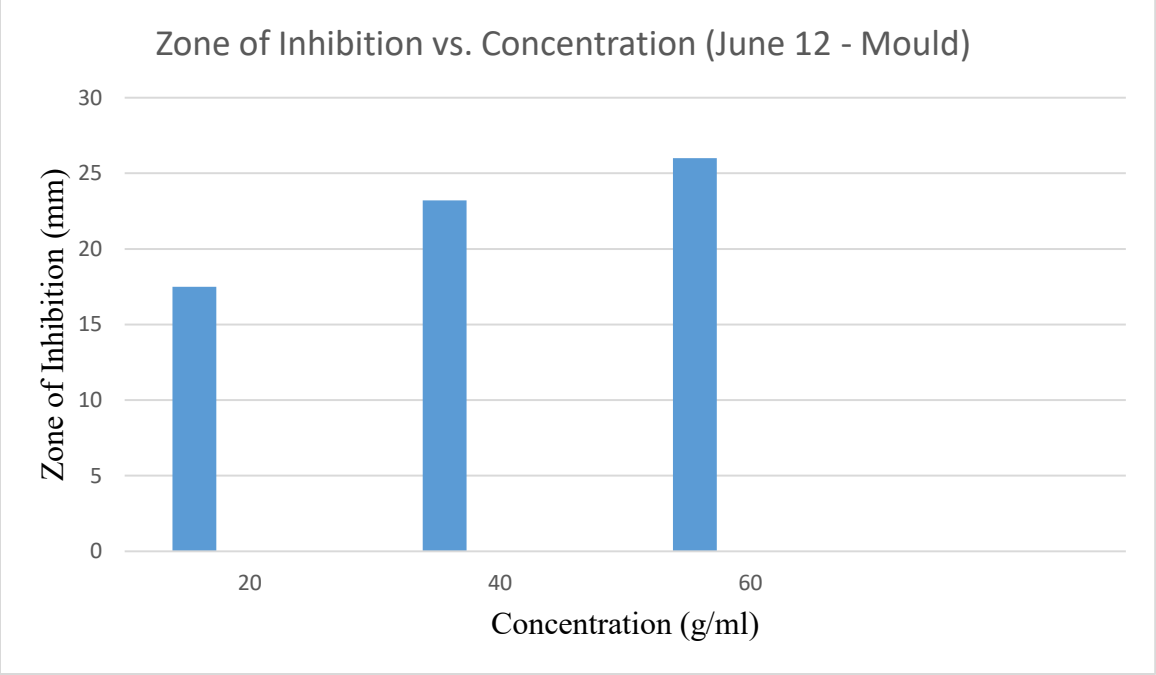

Figure 4: Zone of Inhibition vs. Concentration (June 12 – Mould)

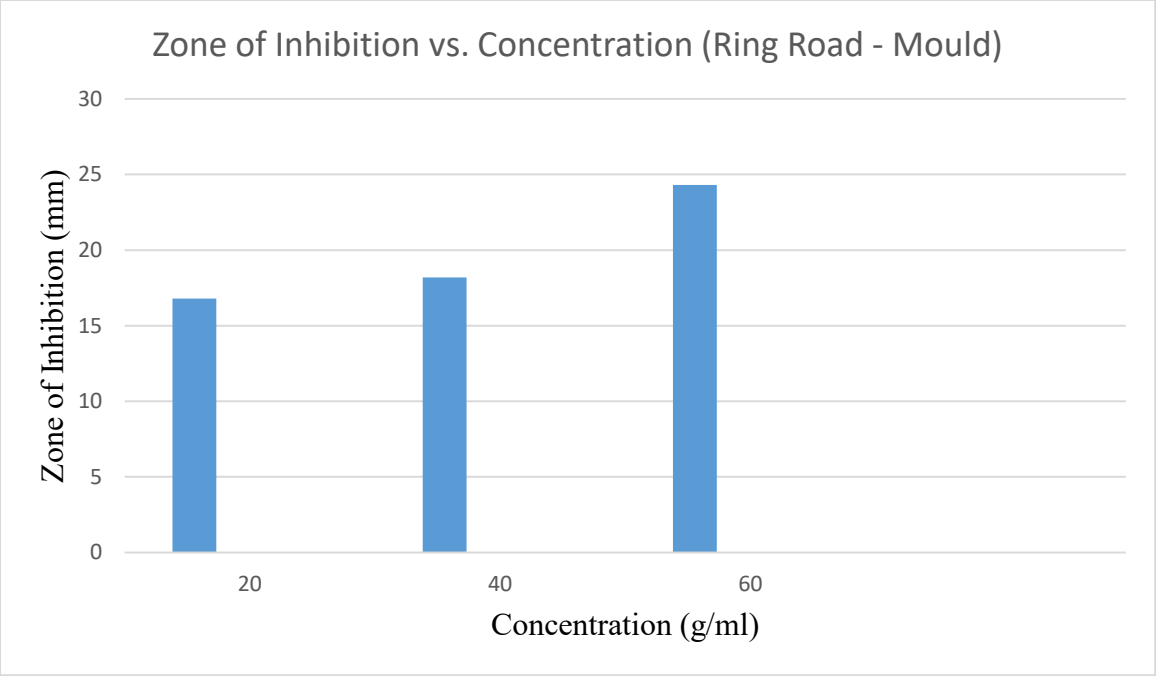

Figure 5: Zone of Inhibition vs. Concentration (Ring Road - Mould)

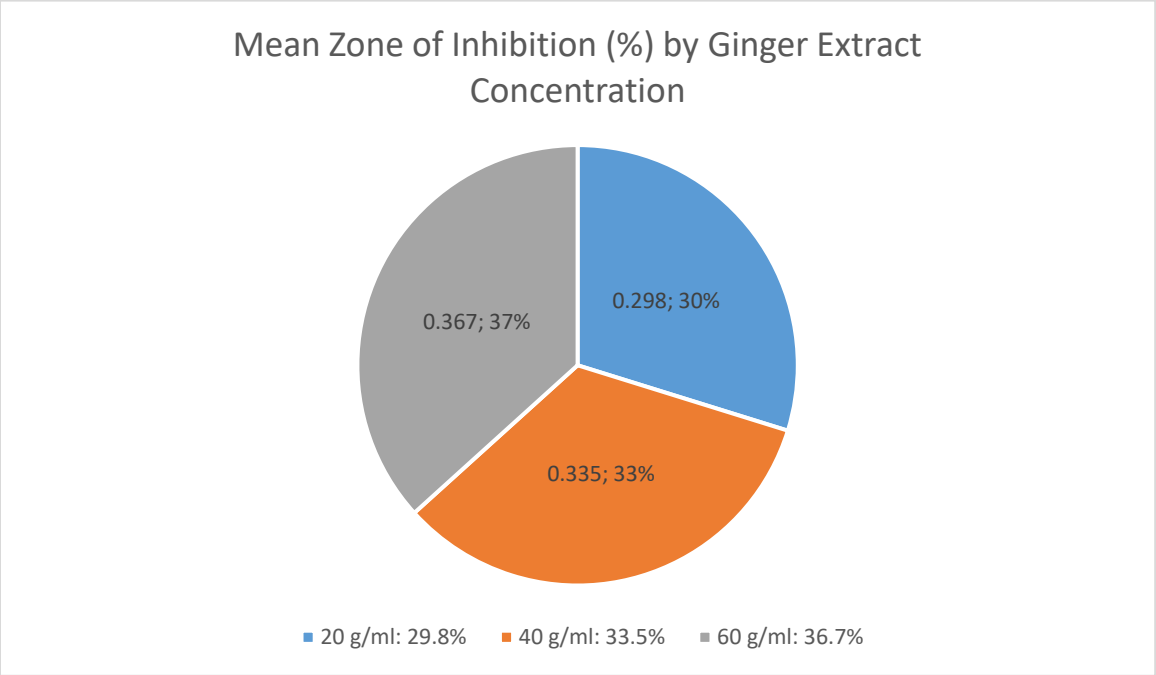

Figure 6: Mean Zone of Inhibition (%) by Ginger Extract Concentration

CHAPTER FOUR

DISCUSSION

The overall trend, as presented in the pie chart titled "Mean Zone of Inhibition (%) by Ginger Extract Concentration," showed that antimicrobial activity increased with concentration. The highest mean inhibition (37%) was observed at 60 g/mL, followed by 33% at 40 g/mL, and 30% at 20 g/mL. This dose-dependent antimicrobial effect agrees with reports by Prakash *et al.,* (2013) and Singh *et al.,* (2008), who demonstrated that higher concentrations of ginger extract exhibited stronger antibacterial and antifungal activities due to elevated levels of bioactive compounds such as gingerol, shogaol, and zingerone.

Isolate-specific responses were also evident. For *A. niger* from the June 12 location, the zone of inhibition increased from 18.2 mm at 20 g/mL to 19.2 mm at 40 g/mL, but slightly decreased to 18 mm at 60 g/mL, suggesting a possible concentration threshold beyond which no further inhibition occurs or antagonistic effects set in. Such non-linear responses have been observed in other studies on plant extracts, where excessive concentrations can sometimes inhibit the diffusion or activity of phytochemicals (Saheed *et al.,* 2015). In contrast, *A. niger* from New Benin and Ring Road demonstrated clear dose-response relationships, with inhibition zones increasing progressively up to 19.5 mm and 18.5 mm at 60 g/mL, respectively. This supports the idea that ginger extract retains potent antifungal properties when concentration is optimized for the target strain, as noted by Ali *et al.,* (2008).

The mould isolates, however, showed a more consistent pattern of increasing inhibition with concentration. For the June 12 mould, inhibition increased markedly from 17.5 mm at 20 g/mL to 26 mm at 60 g/mL, while the Ring Road mould showed similar results, increasing from 16.8 mm to 24.3 mm. These results align with the work of Ekwenye and Elegalam (2005), who demonstrated that ginger extract effectively inhibits a broad range of moulds, highlighting its potential as a natural antifungal agent for food preservation. The differences in inhibition patterns between *A. niger* and general moulds could be attributed to structural differences in their cell walls, enzymatic systems, or spore resilience, which affect susceptibility to ginger's bioactive compounds (Sulong *et al.,* 2019). Additionally, environmental factors at the sampling sites, such as moisture content and storage conditions, may have contributed to isolate-specific resistance levels. Overall, this study confirms that ginger extract has promising antimicrobial properties that can be harnessed to control fungal contamination in fried bottled groundnuts. However, the

findings also indicate that optimal concentration must be carefully considered for different fungal strains and storage conditions. Further research should explore synergistic effects between ginger extract and other natural preservatives, as combined treatments have been shown to enhance antifungal efficacy (Rahman *et al.,* 2011).

CONCLUSION

This study suggests that ginger extract possesses measurable antimicrobial activity against *Aspergillus niger* and general moulds isolated from fried bottled groundnuts in Benin City. The results demonstrated that the effectiveness of the ginger extract generally increased with higher concentrations, with the 60 g/mL concentration yielding the highest zones of inhibition for most isolates tested. However, slight variations were observed, as seen with *A. niger* from the June 12 location, where an optimal effect was noted at 40 g/mL.

Overall, the findings suggest that ginger extract can serve as a potential natural antimicrobial agent for controlling fungal contamination in fried bottled groundnuts. By selecting an appropriate concentration based on the specific fungal strain and storage conditions, the extract could help reduce spoilage and enhance food safety. Future studies should focus on refining extraction methods, testing on larger sample sizes, and exploring the use of ginger extract in combination with other preservation techniques to improve its practical application in food storage and processing.

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