Scholars Academic Journal of Biosciences
Abbreviated Key Title: Sch Acad J Biosci
ISSN 2347-9515 (Print) | ISSN 2321-6883 (Online)
Journal homepage: https://saspublishers.com
NLM ID:101629416
OPEN ACCESS
Biology

Uses of *Eucalyptus camaldulensis* Dehnh (Myrtaceae) in the City of Korhogo and in Vitro Evaluation of the Antifungal Activity of its Essential Oil by Microatmosphere on Two Fungi Found in Stored Foodstuffs

Kanga Yao[1*], Ambe Alain Serge Augustin[2], Yapo Yomeh Cynthia Viviane[3], Zirihi Guédé Noel[4]

[1]UFR Biological Sciences, Department of Plant Biology, Peleforo Gon Coulibaly University (Korhogo, Côte d'Ivoire)
[2]Plant Biology Laboratory, Department of Science and Technology, École Normale Supérieure d'Abidjan, Côte d'Ivoire
[3]Faculty of Medical Sciences, Department of Science and Technology, Alassane Ouattara University, Bouaké, BP V 18 Bouaké 01, Côte d'Ivoire
[4]Laboratory of Natural Environments and Biodiversity Conservation, Félix HOUPHOUET-BOIGNY University

DOI: https://doi.org/10.36347/sajb.2025.v13i08.023 | **Received:** 02.06.2025 | **Accepted:** 11.08.2025 | **Published:** 29.08.2025

***Corresponding author:** Kanga Yao
UFR Biological Sciences, Department of Plant Biology, Peleforo Gon Coulibaly University (Korhogo, Côte d'Ivoire)

Abstract | **Original Research Article**

The objective of this study was to explore the antifungal activity of *Eucalyptus camaldulensis* essential oil against phytopathogenic fungi in stored foodstuffs. To this end, an ethnobotanical survey was conducted in the city of Korhogo on the uses of *Eucalyptus camaldulensis*. Next, the essential oil was extracted from *Eucalyptus camaldulensis* leaves by hydrodistillation, followed by an exploration of the in vitro antifungal activity of its essential oil. The ethnobotanical survey was conducted on 24 households in the city of Korhogo. A total of 72 people were interviewed. The survey revealed that the leaves of *Eucalyptus camaldulensis* are the most commonly used parts of the plant (50%), followed by the bark (29.16%). The antifungal power of *E. camaldulensis*, tested using the microatmosphere method on agar medium against fungal strains of *Aspergillus niger* and *Botryodiplodia* sp, revealed fungistatic activity at respective quantities of 25, 40, 60, and 80 µL of *E. camaldulensis essential* oil. The inhibitory effect of *E. camaldulensis* oil suggests potential applications in the preservation of certain foodstuffs.

Keywords: *Eucalyptus Camaldulensis*, Essential Oil, Antifungal Activity.

INTRODUCTION

Cereal consumption is deeply rooted in the culinary traditions and heritage of many civilizations. This agricultural sector is one of the pillars of the global agricultural economy. Unfortunately, the cereal sector faces many challenges, such as global warming (Liu *et al*., 2021), declining genetic variability (Jia *et al*., 2020), global population growth (United Nations, 2019) and contamination by biological agents (Jedidi *et al.*, 2018). One of the main contaminants affecting cereal production is certain filamentous fungi. Under favorable conditions, they attack crops in the field or in storage. They develop in the grains, affecting yields and reducing their organoleptic and technological qualities (Gautier *et al*., 2020). Furthermore, these fungi can produce secondary metabolites that are toxic to humans, livestock, and pets. These metabolites, called mycotoxins, are generally heat-stable substances that are not easily broken down and are resistant to treatments and processing (Alshannaq and Yu, 2017). To combat these harmful molds, chemical molecules are currently commonly used (Magan and Olsen, 2004). However, their intensive and indiscriminate use is a source of contamination of the biosphere and the food chain. In this context, the search for other solutions is essential. Several studies have highlighted the various biological activities of aromatic and medicinal plants, in particular their antifungal (Jazet *et al*., 2009) and insecticidal (Cheng *et al*., 2009) properties. Essential oils, which represent a very interesting group of metabolites, have been explored. They have antimicrobial and antifungal properties, which make them attractive due to their effectiveness and ease of use. Given these properties, essential oils could therefore be used as food preservatives. In this context, numerous studies have shown that extracts from certain aromatic plants have an inhibitory effect on the growth and toxin production of several bacteria and fungi responsible for foodborne infections (Tzortzakis, 2007). The objective of this study was to highlight the antifungal power of Eucalyptus camaldulensis essential oil using micro-atmosphere

technology, after conducting an ethnobotanical survey on its uses in the city of Korhogo.

MATERIALS AND METHODS

Materials

The plant material used consisted of dried leaves of *Eucalyptus camaldulensis*. The fungal material consisted of two fungal strains, namely *Aspergillus Niger* and *Botryodiplodia* sp.

Methods

Ethnobotanical Survey

Preliminary Ethnobotanical Survey

An initial visit was made to a few households in the city of Korhogo with the aim of establishing a climate of trust and defining the interview channels in order to facilitate the survey.

Sampling Plan

The ethnobotanical survey was conducted in the city of Korhogo (Figure 1). Five neighborhoods were selected for this purpose: Sinistré, Nouveau Quartier, Cocody, Petit Paris, and Nagnénéfou. Twenty-four households were visited for this study.

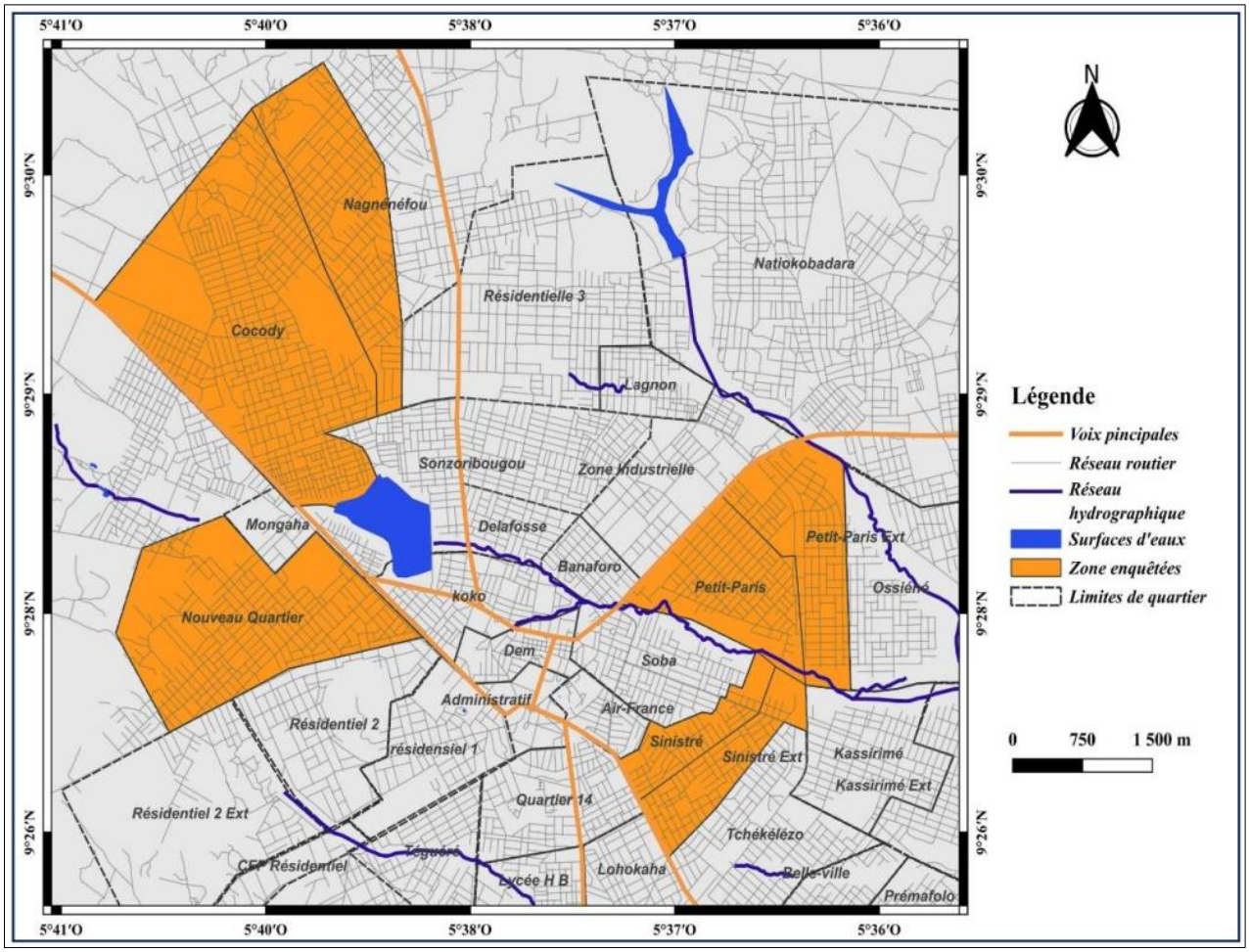

Figure 1: Map of the city of KORHOGO with the areas surveyed

The distribution was as follows:

- Petit Paris 12 surveyed
- Sinistré 20 surveyed
- Nouveau Quartier 17 surveyed
- Cocody 13 surveyed
- Nagnénéfou 10 surveyed

Conduct of the Ethnobotanical Survey

Information on the uses of *Eucalyptus Camaldulensis* was collected using a pre-established questionnaire form. This was divided into two parts, allowing information to be gathered on the respondents and on the plant used by the population. During the survey, the respondents agreed to provide all the information on *Eucalyptus camaldulensis*.

Processing of Ethnobotanical Data

The data collected was processed according to the characteristic parameters of the population surveyed, information on the organs used, and the method of preparation.

Evaluation of Antifungal Activity

Extraction of Essential Oil from Eucalyptus Camaldulensis Leaves by Hydrodistillation

The essential oil was extracted in the laboratory by hydrodistillation using a stainless steel essential oil distiller. This technique is based on the ability of water vapor to transport essential oils. The process involves placing the dry plant material in a steam generator containing water, and boiling the mixture for 1 hour. The essential oil obtained is collected and stored in opaque glass bottles.

Essential Oil Yield from Eucalyptus Leaves

Essential oil yield is the ratio between the mass of essential oil (EO) extracted and the mass of the plant organ to be processed (AFNOR, 1986). Yield is expressed as a percentage using the following formula:

RHE (%) = Mass of EO/Mass (MVS) ×100
RHE (%): Essential oil yield in %
Mass (EO): Mass of essential oil in grams
Mass (MVS): Mass of dry plant material in grams

Evaluation of Antifungal Activity Using Micro-Atmosphere

The antifungal activity of the volatile compounds in the essential oil was tested using the micro-atmosphere method described by Neri *et al*., (2006), derived from the original method developed by Kellner and Kober (1955). Eighteen Petri dishes per fungus containing 20 ml of PDA medium were prepared, including four replicates per concentration and one control per strain. Inoculation was performed by placing a mycelium disc approximately 6 mm in diameter, taken from three-day preculture dishes (106 spores/ml), in the center of each dish. A sterile filter paper disc (FIORONI S.A. Italy) (Ø = 6 mm) is placed in the center of the lid of each Petri dish, and then, using a micropipette, 25 µl, 40 µl, 60 µl, or 80 µl of the essential oil is deposited on the filter paper. The untreated control was prepared under the same conditions with a piece of paper impregnated with distilled water. The dishes are sealed with parafilm (placed upside down on the lid of the dish). These Petri dishes are incubated at 30°C.

Incubation

The Petri dishes were incubated (on the lid so that the volatile compounds could reach the fungal strains) at 30°C for 2 to 14 days, until the growth in the control dishes reached the edges of the Petri dishes. Each test was repeated four times. Every day, the growth of filaments on each dish was recorded and the diameters of different filamentous fungal colonies were measured to calculate the inhibition rate (I%) or percentage of mycelial growth inhibition.

Expression of Results

The percentage of mycelial growth inhibition, compared to the control, was calculated using the following formula:

IAvap (%) = (DC – DT) / DC.100
IAvap (%): antifungal index of volatile compounds in essential oil;
DC: diameter of control mycelium discs;
DT: diameter of mycelium discs with essential oil.

Antifungal Parameters (CMFs and CMFc)

After obtaining the MIC for the oil, a fragment was taken from the box where there was total inhibition of the fungus and transferred to another box containing only PDA. The purpose of this method is to determine whether the oil is fungicidal or fungistatic against the fungus used. When mycelial growth resumes, the concentration is said to be fungistatic (CMFS); however, if there is no further growth, it is called fungicidal (CMFC).

RESULTS

Ethnobotanical Survey

Characteristics of Respondents

The ethnobotanical survey was conducted on 24 households in the city of Korhogo. A total of 72 people were interviewed. The profile was established based on age, gender, and level of education.

❖ **Age**

The age of the respondents ranged from 20 to 80 years old. The majority of them, 50%, belonged to the 30-60 age group. The group of respondents aged between 20 and 30 was less represented, at 16.66%. (Figure 2)

❖ **Gender**

The sample was composed of equal numbers of men and women (Figure 3).

❖ **Level of Education**

In terms of educational attainment, 75% of respondents had no schooling. The remaining 25% were divided between primary schooling (1%) and secondary schooling (24%), as shown in Figure 4.

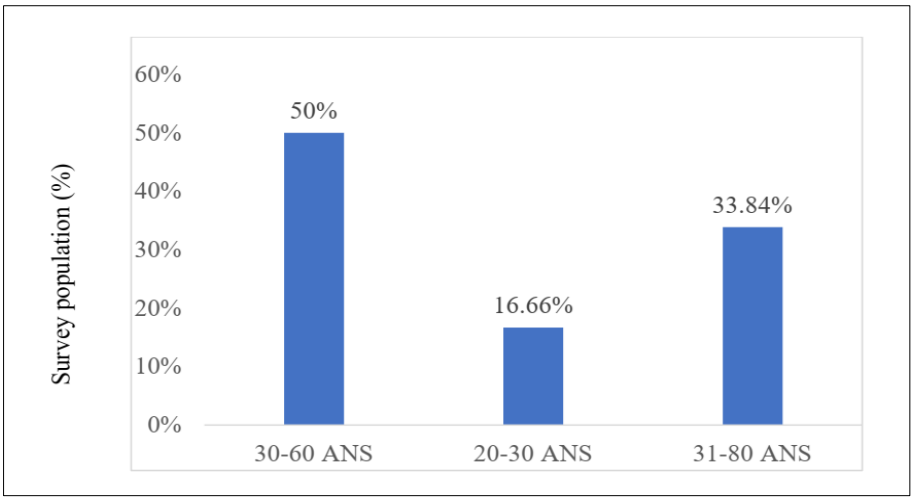

Figure 2: Age range of respondents

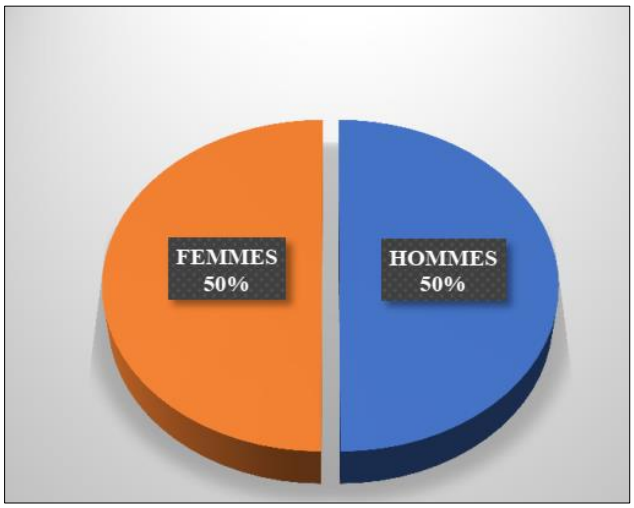

Figure 3: Distribution of respondents by gender

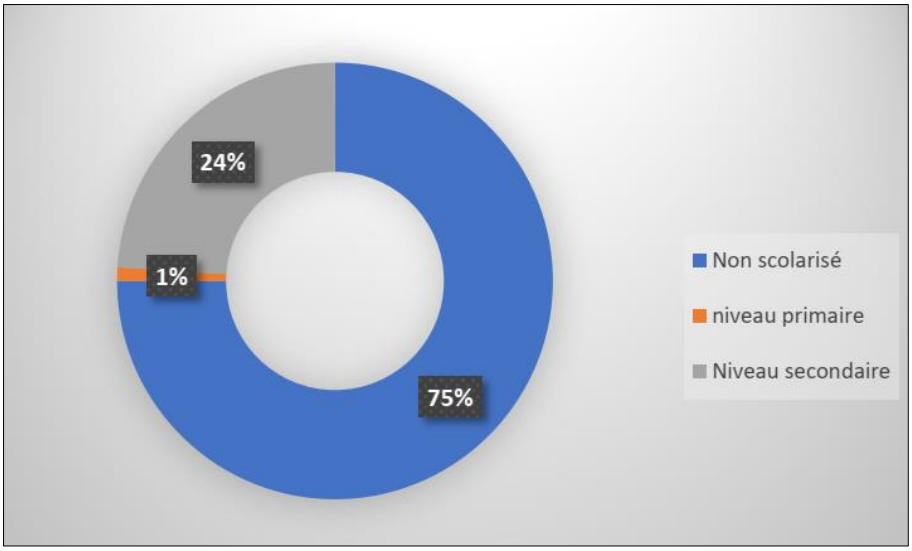

Figure 4: Distribution of respondents by level of education

Parts of plant organs used

Various parts of the organs are used. Leaves are the most commonly used organs (50%), followed by bark (29.16%), then fruits and the whole plant (both 8.33%), and finally roots and flowers (4.16%) (Figure 5).

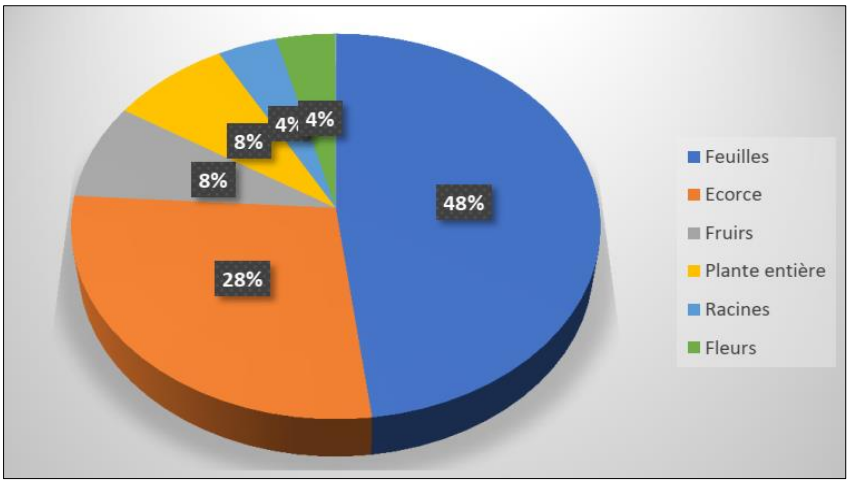

Figure 5: Distribution of organs used from *Eucalyptus camaldulensis*

Method of Preparation of the Organs Used

Following the ethnobotanical survey, only one method of preparation was identified: decoction.

Yield of Eucalyptus Camaldulensis Essential Oil

Analysis of the results in Table I shows that the yield of essential oil obtained from *Eucalyptus camaldulensis* leaves is around 0.9%. The essential oil obtained is yellow in color, liquid in appearance, and has good fluidity, with an aromatic odor.

Table I: Essential oil yield of *Eucalyptus camaldulensis*

Mass of plant material used in (g)	Mass of essential oils extracted in (g)	Essential oil yield (%)
4000	36,39	0,9

Antifungal Activity by Microatmosphere

Antifungal activity is revealed by the absence or presence of mycelial growth for different concentrations of *Eucalyptus camaldulensis* essential oil on two fungal strains: *Aspergillus niger* and *Botryodiplodia* sp. The results indicated that E. Camaldulensis EO had a significant inhibitory effect on the mycelial growth of *Aspergillus niger* at concentrations C1 to C4, i.e., from 25 µl to 80 µl, with respective inhibition rates of 87, 94, 87.5, 88.52, and 90.44% after 10 days (Figure 6). For *Botryodiplodia* sp, the results showed that the oil had a moderate effect compared to *Aspergillus niger*, with inhibition rates of 48.52% for C3, 93.82% for C4, and 0% for C1 and C2 after 10 days (Figure 7). Mycelial growth as a function of fungal strains observed using the microatmosphere method on the 10th day showed that *Eucalyptus camaldulensis* essential oil had a strong inhibitory effect on *Aspergillus niger* with an inhibition rate of 87.87%, unlike *Botryodiplodia* sp, which had an inhibition rate of 55.15%.

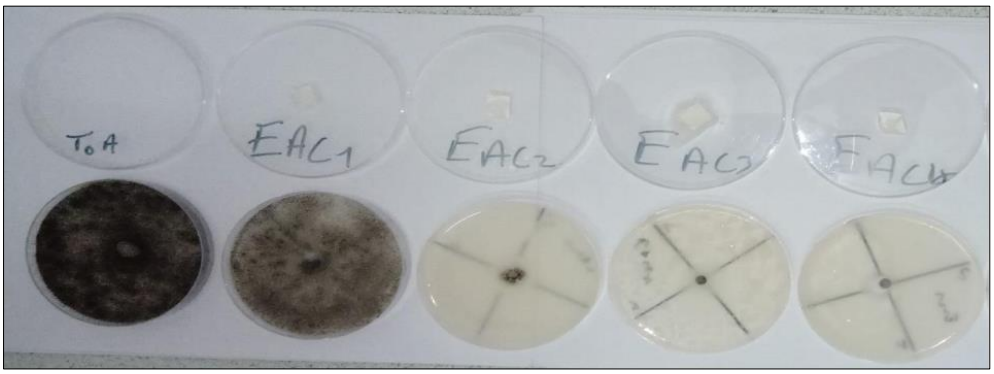

Figure 6: Antifungal activity of *Eucalyptus camaldulensis* essential oil on *Aspergillus niger* after 10 days of incubation in a microatmosphere

T0 A: Control without essential oil on *Aspergillus niger*
EAC1: 25 µl of essential oil on *Aspergillus niger*
EAC2: 40 µl of essential oil on *Aspergillus niger*
EAC3: 60 µl of essential oil on *Aspergillus niger*
EAC4: 80 µl of essential oil on *Aspergillus niger*

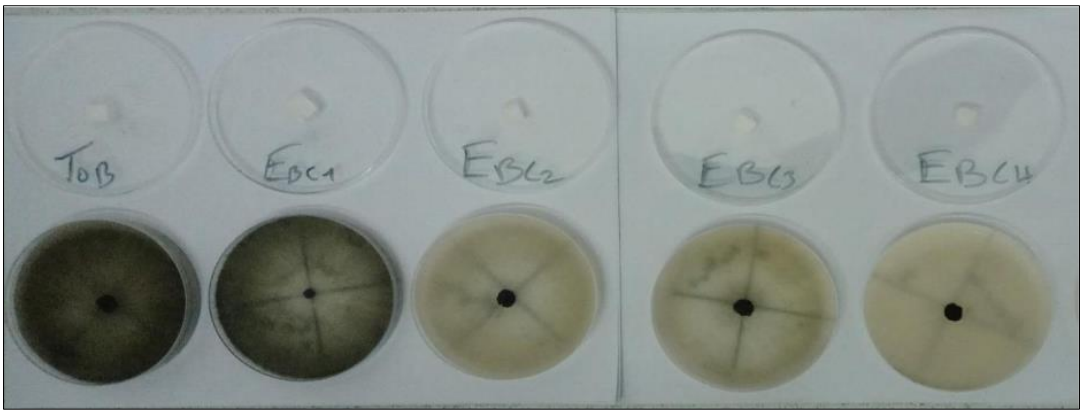

Figure 7: Antifungal activity of *Eucalyptus camaldulensis* essential oil on *Botryodiplodia* sp using micro-atmosphere after 10 days of incubation.

T0 B: Control without essential oil on *Botryodiplodia* sp.
EBC1: 25 µl of essential oil on *Botryodiplodia* sp.
EBC2: 40 µl of essential oil on *Botryodiplodia* sp.
EBC3: 60 µl of essential oil on *Botryodiplodia* sp.
EBC4: 80 µl of essential oil on *Botryodiplodia* sp.

DISCUSSION

This study was conducted in the Poro region, involving 72 people, through an ethnobotanical survey in the department of Korhogo. The majority of respondents were aged between 50 and 80. This can be explained by the fact that knowledge of the uses and properties of medicinal plants is generally acquired through long experience and passed down from one generation to the next, according to Klotoé *et al*., (2013). The results of this study showed that leaves were the most commonly used parts (48%), followed by bark (28%). The interest in leaves and bark can be explained by the fact that they are the main site of biosynthesis and even storage of the secondary metabolites responsible for the plant's biological properties (Nacoulma Ouedraogo, 1996). The high frequency of use of leaves is also due to the ease and speed of harvesting, according to Bitsindou. (1986). Decoction is the most commonly used method of preparation (100%). This method of preparation allows for the collection of the most active ingredients and reduces or eliminates the toxic effect of certain recipes (Salhi *et al*., 2010). Decoction as the most commonly used method of preparation is also confirmed by N'Guessan *et al*., (2009).

The microatmosphere technique revealed the antifungal activity of E. camaldulensis essential oil on both fungal strains. The microatmosphere method for *E. camaldulensis* essential oil showed significant inhibitory activity against *Aspergillus niger* and lesser activity against *Botryodiplodia* sp. The inhibition diameter and mycelium growth rate decreased each time the concentration of essential oil was increased. This could be explained by the presence of certain molecules in the essential oil, namely pinene, cineol, and sabinene. According to Giordani *et al*., (2008), the presence of these molecules in the essential oil gives it antifungal properties.

CONCLUSION

The results of this study revealed that the essential oil extracted from *E. camaldulensis* leaves has antifungal activity and can be used to inhibit the growth of *Aspergillus niger and Botryodiplodia* sp during food storage. The essential oil of *E. camaldulensis*, which has a fairly high antifungal potential per microatmosphere on *Aspergillus niger*, could be used as a natural alternative means of protecting food against mycotoxins.

REFERENCES

- AFNOR (1986). Recueil des normes française ″huiles Essentielles″. AFNOR, Paris,57p.
- Alshannaq, A., & Yu, J.-H. (2017). Occurrence, Toxicity, and Analysis of Major Mycotoxins in Food, IJERPH 14, 632 p.
- Bitsindou M. (1986). Enquête sur la phytothérapie traditionnelle à Kindamba et Odzala (Congo) et analyse de convergence d'usage des plantes médicinales en Afrique Centrale. Mémoire Doc (ined.), Université libre de Bruxelles, Belgique, 482 p.
- Cheng, S., Huang, C., Chen Y., Yu, J., Chen, W., & Chang, S. (2009), Chemical compositions and larvicidal activities of leaf essential oils from two eucalyptus species, Bioresour. Technol., 100, 452-456.
- Gautier, C., Pinson-Gadais, L. & Richard-Forget, F. (2020). Fusarium Mycotoxins Enniatins : An Updated Review of Their Occurrence, the Producing Fusarium Species, and the Abiotic Determinants of Their Accumulation in Crop Harvests. J. Agric. Food Chem, 68: 4788–4798.
- Giordani, R., Hadef, Y., & Kaloustian J. (2008). Compositions and antifungal activities of essential oils of some Algerian aromatic plants. Fitoterapia, 79(3) :199-203.
- Jazet, D P.M., Tatsadjieu, L.N., Tchinda Sonwa, E., Kuate, J., Amvam, Z.P.H., & Menut C. (2009).

Essential oils of Citrus aurantifolia from Cameroon and their antifungal activity against Phaeoramularia angolensis, African Journal of Agricultural Research, 4(4): 354-358.

- Jedidi, I., Soldevilla, C., Lahouar, A., Marín, P., González-Jaén, M.T., Said, S. (2018). Mycoflora isolation and molecular characterization of *Aspergillus* and *Fusarium* species in Tunisian cereals. Saudi Journal of Biological Sciences, 25: 868–874.
- Jia, J., Claude-Taupin, A., Gu Y., Choi, S.W., Peters, R., Bissa B., Mudd, M.H., Allers, L., Pallikkuth, S., Lidke, K.A., Salemi, M., Phinney, B., Mari, M., Reggiorie, F., &Deretic, V. (2020). MERIT, a cellular system coordinating lysosomal repair, removal and replacement. Autophagy, 16 (8) :1539-1541
- Kellner, W. & Kober, W. (1955). Moeglichkeiten derVerwendung aetherischer Oele zurRaumdesinfektion. Arzneimittel-Forsch., 5, 224.
- Klotoé, J R., Dougnon, T V., Koudouvo, K., Atègbo, JM., Loko, F., Akoègninou, A., Aklikokou ,K., Dramane, K., & Gbeassor, M. (2013). Ethnopharmacological survey on antihemorrhagic medicinal plants in South of Benin. European Journal of Medicinal Plants, 3(1): 40-51.
- Liu, N., Liu, C., Dudaš, T., Loc, M. C., Bagi, F. F., & Van der Fels-Klerx, H.J. (2021) Improved aflatoxin and fumonisin forecasting models for maize (PREMA and PREFUM), using combined mechanistic and Bayesian network modelling–Serbia as a case study. Front. Microbiol., 12: 630
- Magan, N., & Olsen, M. (2004). Mycotoxins in food: Detection and control. Woodhead Ltd and CRC Press LLC, 460p
- N'Guessan, K., Kadja, B., Zirihi, G.N., Traoré, D. & Aké-assi, L. (2009). Screening phytochimique de quelques plantes médicinales ivoiriennes, Siences et Nature, 6 : 1-15.
- Nacoulma-Ouedraogo, O. (1996). Plantes médicinales et pratiques médicales traditionnelles au Burkina Faso : cas du plateau central. Thèse de Doctorat ès Sciences Naturelles, option Biochimie, Université de Ouagadougou, (Burkina-Faso), 320 p.
- Neri, F., Mari, M., & Brigati, S. (2006). Control of *Penicillium expansum* by volatile compounds. Plant Pathol., 55 : 100-105.
- Salhi, S., Fadli M., Zidane, L., & Douira, A. (2010). Etude floristique et ethnobotanique des plantes médicinales de la ville de Kénitra. Maroc. 31: 133-146.
- Tzortzakis, N. G. (2007). Maintaining postharvest quality of fresh produce with volatile compounds, Innovative Food Sci. Emerg. Technol., 8 (1) :111-116.
- United Nations (2019) Environment Assembly at its Fourth Session. Innovative Solutions for Environmental Challenges and Sustainable Consumption and Production UNEP/EA.4/HLS. Consultée le 24/08/2024.