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STUDY OF METAL LEVELS IN LEMONGRASS ESSENTIAL OIL

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ABSTRACT
Lemongrass has been noted for its use in various parts of Nigeria to treat conditions such as fever, convulsions in children, throat inflammation, digestive problems, skin disorders, and infections affecting the ears and eyes. It is also a common ingredient in dishes like pepper soup, curries, and local drinks. Given this context, it is important to assess the metal content, as high consumption of lemongrass oil or extended exposure may pose health risks. In this research, lemongrass oil was extracted using a Soxhlet apparatus, and its metal content was evaluated using an Atomic Absorption Spectrophotometer (AAS). The results were compared with similar studies in the literature and the acceptable limits for medicinal plants as established by the WHO. The findings revealed the presence of several metallic elements along with their respective concentrations: magnesium (14.159 mg/kg), lead (1.379 mg/kg), zinc (0.844 mg/kg), mercury (0.728 mg/kg), chromium (0.530 mg/kg), copper (0.267 mg/kg), iron (0.167 mg/kg), cadmium (0.080 mg/kg), manganese (0.009 mg/kg), and arsenic (not detected). The metal composition is deemed acceptable and tolerable according to WHO standards, with the exception of mercury. Therefore, it is vital to continuously monitor metal concentrations to ensure the safety of those who consume or utilize lemongrass, and to prevent excessive intake to avoid associated health issues such as neuropathy, dementia, Parkinson's disease, and cancer.

KEYWORDS: *Materials and methods, Arsenic, Extraction, Heavy metals, Lemongrass, Medicinal plants, Soxhlet extractor,*

1. INTRODUCTION

The lemongrass plant is classified under the Gramineae (Poaceae) family and is part of the genus Cymbopogon. As noted in [1], lemongrass is native to Africa, Australia, Oceania, and Asia, where it remains a staple in traditional medicine and various cosmetic applications. This plant is recognized for its fragrant aroma and is endowed with both anti-fungal and anti-bacterial properties [2]. According to [3], extracts from lemongrass are utilized across several industries, including pharmaceuticals, cosmetics, food processing, and perfumery. For instance, in nations such as India, China, and Thailand, lemongrass is predominantly employed as a flavoring agent in culinary dishes and beverages, attributed to its benefits in digestion, boosting immunity, and enhancing blood circulation. In some areas of Nigeria, lemongrass is reportedly used to alleviate fever, treat convulsions in children, reduce throat inflammation, address stomach issues, and combat skin diseases as well as ear and eye infections. Specifically, in Nigeria's Isoko region, lemongrass features in pepper soup, curries, and local beverages. Furthermore, lemongrass extract has been applied to manage diarrhea, skin infections, and painful irregular menstruation in women. Globally, lemongrass is also believed to assist in lowering blood pressure [4]. The efficacy of lemongrass extracts for local treatment of gastrointestinal discomfort has been validated by [5]. The chemical composition of lemongrass can differ based on soil quality, geographical factors, and fertilizer usage during its cultivation [6] Lemongrass is rich in essential minerals like magnesium, potassium, and calcium, which are required in varying amounts for optimal growth, health maintenance, and overall well-being. For example, magnesium aids the enzymatic functions of all cellular life; potassium is vital for normal brain function and helps in stroke prevention; and calcium is essential for body metabolism as well as the development of bones and teeth. Lemongrass has been recognized for its natural ability to absorb metallic elements from the soil, effectively transferring these elements between the roots and shoots within a specific concentration range, which the plant uses for its biological processes and growth [10]. For instance, the species Cymbopogoncitratus has been utilized to absorb Cu2+, Ni2+, Pb2+, Cd2+, and Zn2+, while Cymbopogonflexuosus can remove As3+ and Cr4+ from water solutions [11-15]. Moreover, lemongrass may contain certain toxic metallic elements, including heavy metals such as lead, mercury, chromium, cadmium, and arsenic [16]; [17]; [18]. These metals are important for human health as they contribute to the maintenance of normal bodily functions. However, excessive levels or prolonged exposure and accumulation, particularly of heavy metals, can pose serious risks to human health, potentially leading to multi-organ dysfunction, acute or chronic poisoning [19-20]. For example, this

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could result in permanent disorders or dysfunctions of the Central Nervous System [21-24]. Additionally, excessive intake or exposure to lead (Pb) can result in high blood pressure, cadmium (Cd) can lead to respiratory problems, renal failure, and cardiovascular issues, an overdose of zinc (Zn) may cause fever, nausea, and general weakness, while excessive iron intake, especially in children, can result in gastrointestinal and skin problems [25].

Recently, there has been an increasing utilization of plant extracts, accompanied by reported health issues associated with their use. The essential analysis of toxic heavy metals, including mercury, lead, arsenic, and cadmium, in plant extracts is vital for ensuring human safety and maintaining quality control. Current literature suggests that lemongrass oil/extract possesses notable pharmacological properties; however, there is a lack of information regarding its metal content, which could affect health. Therefore, this study intends to evaluate the concentration levels of magnesium (Mg), lead (Pb), zinc (Zn), mercury (Hg), chromium (Cr), copper (Cu), iron (Fe), cadmium (Cd), manganese (Mn), and arsenic (As) in lemongrass oil extracted with ethanol from Nigeria.

2. MATERIALS AND METHODS

2.1. Materials

A new sample of lemongrass was gathered from a botanical garden located in Ozoro at the coordinates 5° 32′ 18″ N, 6° 12′ 58″ E, in Delta State, Nigeria. The sample underwent a thorough cleaning process to eliminate any dirt, was washed with water, and then dried for a period of 4-5 hours. The dried leaves were subsequently chopped into 0.5 cm pieces and stored in an airtight bag until required. Ethanol of analytical grade was used as the solvent.

2.2. Experimental method

2.2.1 Oil Extraction

The experiment was carried out according to the methodology described in [28], using a 500mL Shuniu GG-17 Soxhlet extractor. Approximately 150g of lemongrass sample was precisely measured with an electronic digital balance. The weighed sample was placed into the thimble of the Soxhlet extractor, and 250mL of ethanol was added to the round bottom flask of the extractor. The heating mantle was set to maintain a temperature of 78°C, which is the boiling point of ethanol. Throughout the process, the applied heat caused the solvent to vaporize into the extractor thimble containing the sample oil, as the solvent boiled in the round bottom flask. When the liquid reached the overflow level, siphons extracted the mixture from the thimble-holder and transferred it into the distillation flask. The experiment was repeated five times, with each session lasting one hour. Afterward, the experimental apparatus was dismantled, and the extracted oil was separated from the ethanol through an evaporation process under reduced pressure using a rotary evaporator, yielding solvent-free oil. The separated oil was then stored in an airtight container in a dark cupboard for subsequent analysis.

2.3. Atomic Absorption Spectroscopy (AAS) Analysis procedure

The quantitative analysis was performed using the Agilent FS240 AAS machine model, adhering to the approved standard procedure. A 10g oil sample was placed in a dish and heated in a fume cupboard until no smoke was visible. The ash from the heated oil sample was then transferred to desiccators to maintain its moisture content. Following this, 0.1 M HCl acid was added to the oil ash. The resulting mixture was filtered and appropriately diluted for quantitative analysis. Suitable metal salts were used to prepare standard solutions for analysis. An acetylene and air mixture was utilized as the fuel gas. Subsequently, the appropriate metallic hollow lamp and their corresponding standard solutions were used to establish a calibration curve for the respective metals. All samples and standards were analyzed in triplicate, and the average concentrations of the suspected metals were recorded accordingly.

2.4. Validation of the metal contents of the lemongrass

The metal composition of the lemongrass oil was validated by comparing it with the results from previous studies and the specifications detailed in [29], as shown in Table 1.

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Table 1. Metal concentrations of lemongrass oil from literature and acceptable level in medicinal plants by WHO

Heavy Metals	Conc. of metals Ethopia	Conc. of metals Indian	Conc. of metals in medicinal plant (mg/kg) WHO
Magnesium(Mg)	23.9-36.3	0.76-0.79	2000
Lead (Pb)	0.13-0.20	0.14	10
Zinc (Zn)	0.59-1.07	2.23	50
Mercury (Hg)			
Chromium (Cr)			
Copper (Cu)	1.48-2.5	0.05-0.07	20-150
Iron (Fe)	10.35-22.3	1.5-1.98	261-1239
Cadmium (Cd)			
Manganese (Mn)	10.0-12.7	0.19-0.27	20-150
Arsenic (As)			
	[6]	[30]	[29]

3. RESULTS AND DISCUSSION

3.1. Determination of the metal concentration of lemongrass oil

The extraction of lemongrass oil was carried out using the Soxhlet method, followed by an analysis through atomic absorption spectroscopy (AAS) as described in Section 2 to assess the metal content in the oil. The results are depicted in Fig. 1, which shows that the oil contains a range of metals including magnesium (Mg), lead (Pb), zinc (Zn), mercury (Hg), chromium (Cr), copper (Cu), iron (Fe), cadmium (Cd), manganese (Mn), and arsenic (As) at different concentrations. The detection of these metals in the oil can be attributed to the soil and the specific area where the plant was grown [31]. Since the sample was obtained from a garden and cultivated without fertilizers, there was no impact on the metallic composition from fertilizers.

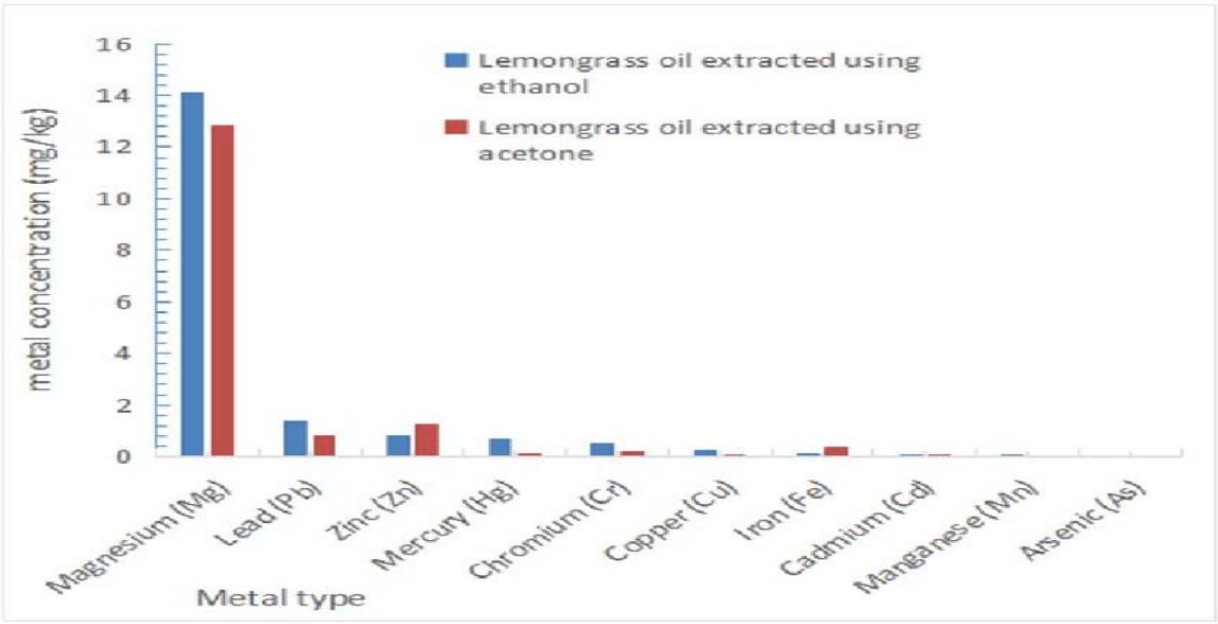


Figure 1. Metal concentrations of lemongrass oil extracted by Soxhlet extraction with ethanol and acetone solvents obtained using AAS.

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Magnesium (Mg)
The magnesium levels obtained from the two solvents, ethanol and acetone, are 14.159 mg/kg and 12.819 mg/kg, respectively. These figures are lower than those indicated by [6] and higher than those from [30]. The differences may be due to factors like soil type, geographical location, or the use of fertilizers to enhance growth. However, the magnesium concentrations are within the standard levels recommended by WHO (Table 1). When lemongrass is used in soups or for medicinal applications, it can support enzymatic functions.

Zinc (Zn)
The concentration of zinc extracted using ethanol was measured at 0.844 mg/kg, while the concentration from acetone was found to be 1.259 mg/kg. The zinc concentration obtained from ethanol extraction falls within the range indicated by [6], whereas the concentration from acetone is marginally higher. Both concentrations exceed those reported by [30], yet they remain compliant with the specifications outlined by [29] (refer to Table 1 and Fig. 1). This discrepancy may be attributed to similar factors affecting magnesium.

Copper (Cu)
The concentration of copper obtained using ethanol as a solvent was 0.267 mg/kg, whereas the concentration achieved with acetone was 0.094 mg/kg. When these results are compared to existing literature, it is evident that both concentrations derived from solvents are lower than those documented by [6] and higher than those recorded by [30]. Nevertheless, they do not meet the range of 20-150 mg/kg specified by [29] (see Table 1 and Fig. 1). The variations in results could be attributed to factors such as climatic conditions, fertilizer usage, and soil properties [6]. These findings imply that lemongrass oil does not provide adequate copper, and its incorporation into food may not sufficiently fulfill the body's copper needs.

Iron (Fe)
The measured iron concentrations were 0.167 and 0.353 mg/kg, obtained from extraction processes utilizing ethanol and acetone, respectively (Fig. 1). When compared to the data presented in Table 1, these findings are lower, and even when taking into account literature values, they do not meet the specifications detailed by [29]. This suggests that lemongrass oil does not contain adequate iron and is improbable to supply a meaningful quantity of this element for its intended application. Iron is essential for blood formation, and a deficiency can result in anemia.

Lead (Pb)
The levels of lead (Pb) in the solvents ethanol and acetone are recorded at 1.379 and 0.840, respectively (see Fig. 1). Although these levels exceed those documented in references [6] and [30], they still fall within the acceptable limits established by [29] (see Table 1). Lead is categorized as a heavy metal, and prolonged exposure can result in chronic poisoning symptoms in humans and other living beings, although certain species can absorb and retain lead in their bodies. Regular intake of products with minor lead contamination can lead to chronic cumulative poisoning. Exposure to lead and its compounds may cause diabetic nerve pain and polyneuropathy in adults, as well as gout and cognitive deficits in children [26]. A major contributor to lead (Pb) pollution is the use of petrol in vehicles, which accounts for roughly 80% of the lead (Pb) found in the air. Approximately 50% of this lead settles within 100 meters of roadways, while the remainder is spread throughout the environment [32]. The lemongrass oil produced in this research contains a safe level of lead, which is not expected to result in immediate harmful effects as previously noted.

Mercury (Hg)
The measured mercury levels were 0.728 and 0.145 mg/kg from the ethanol and acetone extracts, respectively. These levels surpass those recorded by [6, 30], as well as the 5.0 μg/g threshold established by [29]. As a result, there is a potential risk that lemongrass oil may be harmful if consumed, considering that mercury is a toxic element. A report by [33] suggests that exposure to mercury above acceptable levels can result in severe health problems, and [34] highlighted that mercury accumulates in vital organs such as the brain, nervous system, heart, liver, kidneys, and bone marrow, among others. Moreover, mercury significantly contributes to conditions like neuropathy, dementia, Parkinson’s disease, and various cancers in humans, among other issues.

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Chromium (Cr)
The concentration of chromium varied between 0.201 and 0.530 mg/kg. Chromium is essential for humans in trace amounts. Nevertheless, it can be detrimental if its levels exceed the established maximum permissible limits. Chromium is involved in the production of the glucose-tolerance factor [35]; [36]. An excessive intake of chromium may also contribute to cancer.

Cadmium (Cd)
The concentration of chromium varied between 0.201 and 0.530 mg/kg. Chromium is essential for humans in trace amounts. Nevertheless, it can be detrimental if its levels exceed the established safety thresholds. Chromium is involved in the production of the glucose-tolerance factor [35]; [36]. An overconsumption of chromium may also result in cancer

Cadmium (Cd)
The cadmium concentration was found to be between 0.08 and 0.09 mg/kg. In comparison to the permissible limit of 0.3 µg/g as indicated by [29], this suggests that lemongrass oil contains a low level of cadmium. This low concentration may be due to the lack of fertilizer used during the cultivation of lemongrass, since phosphate fertilizers are known to influence cadmium levels, along with the application of sewage sludge and the burning of fossil fuels [37]. These results are encouraging, indicating that lemongrass oil is devoid of the detrimental effects of cadmium, which can accumulate in the kidneys and cause damage to essential cells and bodily functions [38]

Manganese (Mn)
The highest recorded concentration of manganese was 0.009 mg/kg, which is below the levels indicated by [6] and [30], and also falls outside the specified range of 20-150 mg/kg established by [29]. This suggests that the use or intake of lemongrass oil is unlikely to provide a significant amount of manganese to the body. Manganese plays a crucial role in most biological systems. [38] It has been noted that a lack of manganese can lead to serious skeletal and reproductive issues in mammals, along with potential weakening of the brain and lungs due to excessive exposure, which may result in neurological disorders.

Arsenic (As)
No traces of arsenic were detected in the sample based on the conditions tested and the method used. Nevertheless, the detection of arsenic (As) above the permissible limit of 0.2µg/g presents considerable health hazards, as it could result in cancers affecting the lungs, skin, liver, and bladder.

4. CONCLUSIONS
The study revealed that lemongrass is composed of several metallic elements, such as magnesium, zinc, copper, iron, lead, mercury, manganese, chromium, and cadmium, each found in varying concentrations. Most of these metals fall within the acceptable limits established by WHO, with mercury being the notable exception. While metals like magnesium, zinc, copper, and iron are advantageous, the presence of heavy metals including lead, mercury, chromium, cadmium, and manganese poses considerable health risks if ingested or with prolonged exposure. Therefore, it is crucial to limit the consumption or use of lemongrass, and it is advisable to conduct regular analyses of its metal content to ensure health and safety.

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