



## EVALUATION OF THE ANTHELMINTIC ACTIVITY OF CRUDE LEAF EXTRACTS OF SELECTED PLANTS FROM JALINGO, NIGERIA AGAINST *FASCIOLA SP*

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### ABSTRACT

This study evaluates the anthelmintic activity of crude leaf extracts of selected plants from Jalingo Local Government Area of Taraba State, Nigeria. Fresh leaves of *Cymbopogon citratus*, *Ocimum gratissimum* and *Musa paradisiaca*, were collected from local farms in Jalingo metropolis. The collected plant materials were washed thoroughly to remove debris and dried under shade at room temperature for 7–10 days. The anthelmintic activity of the plant extracts was evaluated using an in vitro model involving intestinal helminths that were collected at the abattoir in Jalingo metropolis. The parasites of uniform size (approximately 5–7 cm) were placed in Petri dishes containing various concentrations (10 mg/mL, 20 mg/mL, 40 mg/mL, and 80 mg/mL) of each plant extract dissolved in distilled water. Albendazole was used as a positive control, while distilled water served as the negative control. The paralyzing and death times of the helminths was recorded. The analysis revealed the presence of flavonoids, saponins, tannins, terpenoids, phenols, and glycosides for all the plants, while alkaloids and steroids were absent in *C. citratus* and tannins absent for *Ocimum gratissimum*. As the concentration of *C. citratus* extract increased from 10 mg/ml to 80 mg/ml, the immobilization time decreased from  $69.33 \pm 23.86$  to  $49.50 \pm 16.26$  minutes, indicating a dose-dependent anthelmintic activity. At 10 mg/ml, the immobilization time was  $133.67 \pm 4.73$  minutes, while at 80 mg/ml, it was to  $69.67 \pm 17.39$  minutes for *M. paradisiaca*. The immobilization time drops sharply from  $176.00 \pm 41.61$  minutes at 10 mg/ml to just  $30.67 \pm 2.08$  minutes at 80 mg/ml for *O. gratissimum*. The synergistic formulation shows enhanced potency compared to individual extracts. At 10 mg/ml, the immobilization time was  $55.81 \pm 2.25$  minutes, and at 80 mg/ml, it drops dramatically to  $26.65 \pm 1.57$  minutes. In conclusion, the results of this study strongly support the ethnomedicinal use of *Cymbopogon citratus*, *Musa paradisiaca*, and *Ocimum gratissimum* in managing parasitic infections. The presence of multiple phytochemicals in these plants contributes to their biological activity, particularly in inhibiting the viability of *Fasciola hepatica*.

### 1. Introduction

Helminth infections, caused by parasitic worms such as trematodes, nematodes and cestodes, represent a widespread global health issue, particularly in developing countries with inadequate sanitation and water supply systems (Hotez *et al.*, 2014). According to the World Health Organization, over two billion people are affected by soil-transmitted helminths (STHs) and schistosomes, leading to significant morbidity and, in severe cases, mortality (WHO, 2020). The burden of helminthiasis disproportionately impacts children and the impoverished, causing malnutrition, cognitive impairment, stunted growth, and weakened immune systems (Bethony *et al.*, 2006). Moreover, helminth infections exacerbate other diseases by increasing susceptibility to secondary infections and reducing the effectiveness of vaccines (Cooper *et al.*, 2010).

Despite the availability of synthetic anthelmintics such as albendazole, mebendazole, and ivermectin, challenges such as drug resistance, side effects, and the high cost of treatment have led to the exploration of alternative therapies

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(Geary, 2012). Drug resistance, in particular, has emerged as a serious concern, especially in veterinary medicine, where the overuse of anthelmintics has led to resistant strains of parasites (Prichard *et al.*, 2012). In humans, the repetitive use of the same anthelmintic drugs in mass drug administration (MDA) programs for controlling helminth infections has raised concerns about the potential for resistance (Vercruysse *et al.*, 2011). Consequently, there is an urgent need to identify new, effective, and sustainable treatments that can be used either alone or in combination with existing drugs to manage helminth infections.

Plants have been used for centuries in traditional medicine for the treatment of parasitic infections, including helminthiasis. Their widespread use in various cultures is often attributed to their accessibility, affordability, and perceived safety compared to synthetic drugs (Wink, 2015). Medicinal plants contain a rich diversity of bioactive compounds such as alkaloids, flavonoids, tannins, and terpenoids, many of which have demonstrated anthelmintic activities (Adamu *et al.*, 2013). These compounds may act through various mechanisms, including the inhibition of parasite motility, disruption of the parasite's metabolism, and impairment of its ability to reproduce (Egualé *et al.*, 2011).

One of the most promising plant species with documented anthelmintic properties is *Cymbopogon citratus*, commonly known as lemongrass. This plant is widely used in traditional medicine for treating digestive issues, fever, and infections. Studies have shown that the essential oils derived from *Cymbopogon citratus* possess antimicrobial, antifungal, and antiparasitic activities (Shah *et al.*, 2011). The key bioactive compounds in lemongrass include citral, geraniol, and myrcene, which have demonstrated efficacy against a variety of parasitic organisms, including helminths (De Moraes *et al.*, 2019).

Similarly, *Ocimum gratissimum*, commonly referred to as African basil or scent leaf, is another medicinal plant with potent anthelmintic properties. It has been used traditionally in African and Asian communities for the treatment of various ailments, including gastrointestinal disorders, respiratory infections, and parasitic diseases (Akinmoladun *et al.*, 2014). The leaves of *Ocimum gratissimum* contain essential oils, flavonoids, and phenolic compounds, which are responsible for its anthelmintic, antimicrobial, and anti-inflammatory activities (Ezekwesili *et al.*, 2004). Previous studies have demonstrated the plant's ability to inhibit the survival and reproduction of nematodes, providing a scientific basis for its traditional use as an anthelmintic agent (Adjanohoun *et al.*, 1996).

*Musa paradisiaca*, or plantain, is another plant with potential anthelmintic properties. While it is primarily known for its nutritional value, various parts of the plant, including the peel, root, and leaves, have been used in traditional medicine for treating wounds, infections, and parasitic diseases (Aiyeloja and Bello, 2006). The phytochemicals present in *Musa paradisiaca*, such as alkaloids, saponins, and tannins, have been shown to exhibit anthelmintic activities against a range of helminths (Bakr *et al.*, 2021). Although research on the anthelmintic properties of plantain is limited compared to *Cymbopogon citratus* and *Ocimum gratissimum*, the available evidence suggests that it could be a valuable component of plant-based anthelmintic formulations (Fasoyiro *et al.*, 2005).

The increasing interest in plant-based anthelmintics is driven by the limitations of conventional drugs and the growing awareness of the potential health and environmental benefits of natural remedies (Hoste *et al.*, 2015). Unlike synthetic drugs, plant-based anthelmintics are often biodegradable and less likely to contribute to the development of drug resistance due to their complex mixtures of bioactive compounds (Githiori *et al.*, 2006). Furthermore, the use of plants with multiple bioactive components may reduce the likelihood of helminths developing resistance, as the parasites would need to simultaneously adapt to several different mechanisms of action (Waller, 2006).

Recent studies have focused on evaluating the efficacy of crude plant extracts *in vitro* and *in vivo*, with promising results. For instance, *in vitro* studies using *Cymbopogon citratus* extracts have shown significant inhibitory effects on the motility and survival of gastrointestinal nematodes (Iqbal *et al.*, 2004). Similarly, the essential oils of *Ocimum gratissimum* have been reported to cause paralysis and death in adult helminths in both laboratory and field settings (Ola-Fadunsin & Ademola, 2013). These findings support the hypothesis that plant-based treatments could serve as an effective alternative to synthetic anthelmintics, particularly in areas where access to conventional drugs is limited.

## 2. Materials and Methods

### 2.1 Study Area

This study was conducted in Jalingo, the capital city of Taraba State, located in the northeastern region of Nigeria. Jalingo lies between latitudes 8°54'N and 9°09'N, and longitudes 11°09'E and 11°30'E, with an average elevation of approximately 349 meters above sea level. It covers an area of about 195 square kilometers and serves as the administrative and commercial hub of Taraba State.

The climate in Jalingo is typically tropical with distinct wet and dry seasons. The rainy season usually begins in April and extends through October, while the dry season spans from November to March. The area receives an annual rainfall of approximately 1,200 to 1,800 mm, with average daily temperatures ranging from 25°C to 35°C (Nigerian Meteorological Agency, 2022). These climatic conditions support diverse vegetation and make the region suitable for

the cultivation of medicinal plants, fruits, and vegetables.

Jalingo's population is predominantly agrarian, with many residents engaging in subsistence farming and the cultivation of medicinal plants used in traditional health care systems. The local flora includes a wide variety of plants with ethnomedicinal importance, which are commonly used in the treatment of parasitic infections and other ailments. This makes Jalingo an ideal location for evaluating the anthelmintic potential of crude leaf extracts against *Fasciola* species, a liver fluke parasite responsible for fascioliasis in both livestock and humans.

## 2.2 Research Design

The study adopted an experimental design to evaluate the anthelmintic activities of crude extracts from *Cymbopogon citratus*, *Ocimum gratissimum*, and *Musa paradisiaca*. The research followed a structured process involving phytochemical screening, in vitro sensitivity testing, and comparison of individual and synergistic activities of the plant extracts against selected helminths.

## 2.3 Plant Materials Collection

Fresh leaves of *Cymbopogon citratus*, *Ocimum gratissimum*, and *Musa paradisiaca* were collected from local farms in the Jalingo metropolis. Identification and authentication of the plant species were carried out by a botanist at Taraba State University, Jalingo. The collected plant materials were washed thoroughly to remove debris and dried at room temperature for 7–10 days. Once dried, the samples were pulverized into fine powder using a mechanical grinder and stored in airtight containers until extraction.

## 2.4 Collection of Parasites

The liver of the slaughtered animals was carefully removed and transported to a clean, designated examination area within the abattoir premises. Using sterile scalpels and forceps, the bile ducts and gallbladders were dissected open, and the *Fasciola* spp. were gently extracted.

The collected parasites were initially identified based on morphological features such as leaf-shaped, dorsoventrally flattened bodies, oral and ventral suckers, and overall length and width. These features were compared to standard descriptions in parasitology manuals for confirmation of *Fasciola* spp (Soulsby, 1982).

## 2.5 Preparation of Plant Extracts

The extraction of the plant materials was performed using the cold maceration method. Briefly, 500 grams of each powdered plant material were soaked in 1,500 mL of distilled water for 72 hours, with occasional stirring. The mixture was filtered through muslin cloth followed by Whatman No. 1 filter paper to obtain the crude ethanol extract. The filtrates were concentrated under reduced pressure using a rotary evaporator and then further dried in a desiccator. The dried crude extracts were stored in sealed containers and kept at 4°C until use.

## 2.6 Phytochemical Screening of Plant Extracts

Preliminary phytochemical screening of the crude extracts was conducted using standard methods described by Harborne (1998) to identify bioactive compounds such as alkaloids, flavonoids, saponins, tannins, glycosides, and terpenoids. Each phytochemical test was conducted in triplicate to ensure accuracy.

## 2.7 Evaluation of Anthelmintic Activity

### In Vitro Sensitivity Model

The anthelmintic activity of the plant extracts was evaluated using an in vitro model involving intestinal helminths that were collected at the abattoir in the Jalingo metropolis. The *Fasciola* spp of uniform size (approximately 30–32 cm) were placed in Petri dishes containing various concentrations (10 mg/mL, 20 mg/mL, 40 mg/mL, and 80 mg/mL) of each plant extract dissolved in distilled water. Albendazole (20 mg/mL) was used as a positive control, while distilled water served as the negative control. The immobilization times of *Fasciola hepatica* were recorded. Immobilization was determined when the worms did not move after gentle stimulation, and death was confirmed when no movement was observed even after exposure to hot water (50°C). The experiment was carried out in triplicate, and the mean paralysis and death times for each concentration were calculated.

### Comparison of Anthelmintic Activity

The anthelmintic activities of the different plant extracts were compared based on their mean immobilization times at various concentrations. A dose-response curve was plotted for each extract to assess its efficacy.

### Evaluation of Synergistic Activity

To evaluate the potential synergistic anthelmintic effects of the plant extracts, combinations of *Cymbopogon citratus*, *Ocimum gratissimum*, and *Musa paradisiaca* extracts were prepared in a 1:1 ratio (by weight). The anthelmintic assay was repeated using the combined extracts at the same concentrations as in the individual assays (10 mg/mL, 20 mg/mL, 40 mg/mL, and 80 mg/mL). The mean immobilization times were recorded for each combination.

## 2.8 Statistical Analysis

The data obtained from the phytochemical screening and anthelmintic assays were expressed as mean  $\pm$  standard

deviation (SD). Chi-square was used to determine the statistical significance of differences between the groups, with p-values less than 0.05 considered statistically significant. All statistical analyses were performed using the Statistical Package for Social Sciences (SPSS).

### 3. Results and Discussion

Table 1: Phytochemical Screening

Phytochemical Constituents	Plants		
	<i>Cymbopogon citratus</i>	<i>Musa paradisiaca</i>	<i>Ocimum gratissimum</i>
Alkaloids	–	+	+
Flavonoids	+	+	+
Saponins	+	+	+
Tannins	+	+	+
Terpenoids	+	–	+
Phenols	+	+	+
Glycosides	+	+	+
Steroids	–	–	+

Key: += positive; -=Negative

Table 2: Effect of *C. citratus* on *Fasciola spp*

Concentration	Immobilization time (Minutes)
10 mg/ml	69.33 ± 23.86
20 mg/ml	61.67 ± 23.03
40 mg/ml	55.50 ± 24.75
80 mg/ml	49.50 ± 16.26

( $\chi^2=11.89$ ; p-value=0.0644)

Table 3: Effect of *M. paradisiaca* on *Fasciola spp*

Concentration	Immobilization time (Minutes)
10 mg/ml	133.67 ± 4.73
20 mg/ml	116.00 ± 7.81
40 mg/ml	77.33 ± 22.50
80 mg/ml	69.67 ± 17.39

( $\chi^2=18.86$ ; p-value=0.0044)

Table 4: Effect of *O. gratissimum* on *Fasciola* spp

Concentration	Immobilization time (Minutes)
10 mg/ml	176.00 ± 41.61
20 mg/ml	108.33 ± 11.93
40 mg/ml	32.67 ± 3.06
80 mg/ml	30.67 ± 2.08

( $\chi^2=14.21$ ; p-value=0.0273)

Table 5: Synergistic effect of *C. citratus*, *M. paradisiaca* and *O. gratissimum* on *Fasciola* spp

Concentration	Immobilization time (Minutes)
10 mg/ml	55.81 ± 2.25
20 mg/ml	47.62 ± 4.67
40 mg/ml	37.19 ± 3.61
80 mg/ml	26.65 ± 1.57

( $\chi^2=36.52$ ; p-value=0.014)

## Discussion

The evaluation of the anthelmintic activity of crude leaf extracts of *Cymbopogon citratus*, *Musa paradisiaca*, and *Ocimum gratissimum* against *Fasciola hepatica* offers valuable insights into their pharmacological potentials. These plants are widely used in African ethnomedicine for treating microbial and parasitic infections, and the findings of this study align with that traditional usage. The differential composition of secondary metabolites among the plants suggests that variations in phytochemistry directly influence their biological activity.

*Cymbopogon citratus* contained flavonoids, saponins, tannins, terpenoids, phenols, and glycosides, while alkaloids and steroids were absent. These compounds are known for antimicrobial, antioxidant, and antiparasitic properties. The moderate anthelmintic activity observed can be attributed to these constituents, especially tannins and flavonoids, which have been reported to impair parasite metabolism by binding to enzymes and structural proteins. Adeniyi *et al.* (2019) similarly reported the presence of these compounds in *C. citratus* and confirmed its moderate antiparasitic efficacy, aligning with the present study. Ogunleye and Osho (2015) also observed dose-dependent anthelmintic effects of *C. citratus* extracts, though with limited potency at lower concentrations, supporting our results. However, contrary findings were reported by Ibrahim *et al.* (2021), who found that *C. citratus* exhibited strong anthelmintic activity even at low concentrations, suggesting that geographical and extraction variations may influence efficacy.

*Musa paradisiaca* showed a slightly different phytochemical profile, containing alkaloids, flavonoids, saponins, tannins, phenols, and glycosides, but lacking terpenoids and steroids. The presence of alkaloids may explain its higher potency, as these compounds interfere with parasite neuromuscular functions. The current study revealed significant anthelmintic activity ( $p = 0.0044$ ), consistent with Irokanulo *et al.* (2017), who reported that plantain extracts possess potent antiparasitic and immunomodulatory properties due to their alkaloid and saponin content. Similarly, Okafor *et al.* (2018) found that *M. paradisiaca* extracts were effective against gastrointestinal parasites in small ruminants. Conversely, Adebayo *et al.* (2020) observed limited anthelmintic effects of *M. paradisiaca* in vitro, suggesting that efficacy may depend on the type of parasite or solvent used in extraction.

*Ocimum gratissimum* demonstrated the most comprehensive phytochemical profile, containing all eight tested constituent's alkaloids, flavonoids, saponins, tannins, terpenoids, phenols, glycosides, and steroids. This extensive profile correlates with its strong efficacy, as seen in the significant reduction of immobilization time ( $p = 0.0273$ ). These findings align with Ugboogu *et al.* (2020), who reported that *O. gratissimum* extracts showed broad-spectrum antiparasitic and antimicrobial activity due to synergistic effects of multiple bioactive compounds. Akinyemi *et al.* (2020) similarly found that *O. gratissimum* exhibited superior efficacy against intestinal worms compared to single-compound extracts. However, contrasting observations were made by Eze *et al.* (2021), who found *O. gratissimum* less effective than *Azadirachta indica* extracts, highlighting that activity may vary across parasite species and concentration ranges.

The pattern of activity across the three plants revealed a dose-dependent trend, with *O. gratissimum* showing the highest potency, followed by *M. paradisiaca*, and then *C. citratus*. The immobilization time decreased significantly as concentration increased, which reflects the concentration-dependent action typical of phytochemical-rich plant extracts. This observation agrees with the findings of Ojo *et al.* (2018), who noted that herbal extracts generally show enhanced antiparasitic efficacy at higher doses. On the contrary, Ekanem *et al.* (2021) argued that increasing concentration may lead to cytotoxic effects, reducing the therapeutic index of certain plant extracts. Hence, while the dose–response relationship is beneficial, the safety threshold must be considered.

Interestingly, the combination of the three plant extracts exhibited superior anthelmintic activity compared to individual extracts, with immobilization time dropping significantly ( $p = 0.00014$ ). This finding supports the principle of phytochemical synergy, where multiple bioactive agents interact to enhance efficacy. Onwuka *et al.* (2021) reported similar synergistic effects in combined herbal formulations used against intestinal helminths. Likewise, Ezeonu *et al.* (2020) emphasized that polyherbal preparations enhance pharmacological outcomes by targeting multiple biological pathways simultaneously. However, in contrast, Ajayi *et al.* (2022) cautioned that combining extracts could also result in antagonistic interactions or toxicity if not well standardized, which underscores the need for further toxicological evaluation of combined formulations.

#### 4. Conclusion

The results of this study strongly support the ethnomedicinal use of *Cymbopogon citratus*, *Musa paradisiaca*, and *Ocimum gratissimum* in managing parasitic infections. The presence of multiple phytochemicals in these plants contributes to their biological activity, particularly in inhibiting the viability of *Fasciola hepatica*. The observed synergistic effect of the combined extracts offers promising potential for developing more effective, plant-based anthelmintic formulations.

Extensive *in vivo* studies and toxicity assessments should be carried out by researchers to validate the safety and efficacy of these extracts in live animal models before progressing to human

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