(International Print/Online Journal)

SJIF IMPACT FACTOR: 6.829
PUBMED-National Library of Medicine ID-101739732

ISSN (Print): 2209-2870
ISSN (Online): 2209-2862

International Journal of Medical Science and Current Research (IJMSCR)
Available online at: www.ijmscr.com
Volume 8, Issue 5 , Page No: 526-541
September-October 2025

IJMSCR

Comparative study on the antimicrobial activity of Moringa leaves, Long pepper, Cinnamon, and Aloe vera against Tuberculosis bacteria

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Type of Publication: Original Research Paper
Conflicts of Interest: Nil

Abstract

Background: Tuberculosis (TB) is one of the noxious infectious diseases globally, effecting approximately two million deaths every year. Multidrug resistance presents a significant global challenge for the treatment of TB, requiring prolonged medication use and extended treatment durations. There is an urgent need to discover alternative novel antimicrobial compounds. Herbal medicine offers promising prospects for drug discovery and development, warranting the screening of plant-derived compounds with favourable safety margins. ***Objectives*:** This study aimed to investigate the in vitro antimicrobial activity of extracts from four medicinal plants against Mycobacterium tuberculosis in vitro. ***Methods*:** Extracts from four herbs — Moringa Leaves, Long Pepper, Cinnamon, and Aloe Vera Against Tuberculosis Bacteria were investigated for their in vitro antimicrobial activity against M. tuberculosis using Lowenstein-Jensen medium. Antimicrobial activity was assessed by comparing bacterial colony growth on extract-containing media with that on extract-free control media. The agar dilution method was used to determine antimicrobial activity. ***Results*:** Among the tested extracts, Moringa Leaves, Long Pepper, Cinnamon, and Aloe Vera Against Tuberculosis Bacteria the strongest antimicrobial activity. These herbs contain various bioactive compounds, including alkaloids, flavonoids, and steroids, which are presumed to contribute to bacterial susceptibility. ***Conclusions***: The findings suggest that Moringa leaves, Long pepper, Cinnamon, and Aloe vera may have therapeutic potential in TB treatment. These herbs could serve as alternative agents for combating TB and may help mitigate the adverse effects associated with standard anti-tuberculosis drugs.

Keywords: Tuberculosis, Mycobacterium, Plant extracts, Moringa leaves, Long pepper, Cinnamon, and Aloe vera

Introduction

Tuberculosis (TB) is a chronic infectious disease caused mainly by *Mycobacterium tuberculosis*, a bacterium that predominantly attacks the lungs but can also affect other organs [1]. Despite long-standing global control efforts, TB remains a pressing public health issue and is currently the second leading infectious disease killer after COVID-19 [2]. **c**ombined studies of Moringa leaves, long pepper, cinnamon, and Aloe Vera extracts will exhibit significant antimicrobial activity against *Mycobacterium tuberculosis* due to their collaborative phytoconstituents, which can inhibit growth of bacteria and possibly act as complementary therapeutic agents to conventional treatments. Research indicates that Moringa leaves, long pepper, cinnamon, and Aloe Vera contain

compounds like flavonoids, phenols, and alkaloids, which are known to possess antimicrobial properties that could be effective against tuberculosis bacteria

According to the World Health Organization (WHO), approximately 10.6 million new TB cases and 1.6 million related deaths were reported worldwide in 2021, with about 187,000 deaths occurring among HIV-positive individuals [2]. The persistence of TB as a global health burden is compounded by delayed diagnosis, limited access to healthcare, high treatment costs, HIV co-infection, and the rise of drug-resistant strains. In particular, multidrug-resistant TB (MDR-TB) and extensively drug-resistant TB (XDR-TB) have complicated treatment regimens and posed significant challenges in resource-limited regions such as South-East Asia, Africa, and the Western Pacific [3–4]. Parallel to conventional therapeutic approaches, interest has grown in the use of traditional medicinal plants as alternative or complementary treatments. In Indonesia, several indigenous plants have historically been used to manage TB symptoms [5]. One notable example is noni (*Morinda citrifolia*), a small evergreen tree native to the Indo-Pacific region and widely distributed throughout the Indonesian archipelago. Traditionally, noni has been utilized for its diverse therapeutic properties, including antibacterial, antiviral, antifungal, anti-inflammatory, analgesic, hypotensive, antitumor, and immune-enhancing effects [6–7]. Pharmacological studies have demonstrated that Moringa leaves, Long pepper, Cinnamon, and Aloe vera extracts of are effective against a wide range of Gram-positive and Gram-negative bacteria, such as *Staphylococcus aureus*, *Bacillus subtilis*, *Proteus morganii*, *Pseudomonas* spp., and *Escherichia coli* [8–9], as well as enteric pathogens like *Salmonella* and *Shigella* [10–12,14]. The antimicrobial activity of plants extract is largely attributed to its rich composition of secondary metabolites and lectins, compounds associated with plant defense mechanisms [15]. More than 200 phytochemicals have been isolated from different parts of the plant, including acids, alcohols, phenols, anthraquinones, carotenoids, esters, flavonoids, iridoids, ketones, lactones, lignans, nucleosides, triterpenoids, and sterols [15–17]. Major bioactive constituents such as scopoletin, terpenoids, alkaloids, anthraquinones, and flavonoids have been identified as key antibacterial agents [14,17]. These metabolites are believed to exert antimicrobial effects by disrupting bacterial membranes, inactivating enzymes, and denaturing proteins, ultimately leading to bacterial cell damage and death [20].

Given this background, recent investigations have sought to evaluate the anti-tubercular activity of specific plant species-derived compounds—including flavonoids, anthraquinones, scopoletin, and alkaloids—at concentrations ranging from 10 to 40 mg/ml against the *Mycobacterium tuberculosis* H37Rv strain [20]. Such studies aim to identify the most effective compounds and optimal dosages for inhibiting TB growth, while also comparing variations in antibacterial efficacy across compound types and concentrations, other plants like *Aloe vera* have demonstrated antibacterial activity against both Gram-positive and Gram-negative bacteria, though their role in treating multidrug-resistant TB strains has yet to be fully explored [8–10]. This underscores the importance of further scientific evaluation of traditional medicinal plants as potential sources of novel anti-TB agents[28].

Methodology

Sample Collection

Samples were collected from the Kukrail Garden, Lucknow, India, during the early morning hours to ensure freshness and minimize contamination. Leaves from healthy and mature plants were selected and collected using sterile gloves and pruning shears to avoid contamination. The collected samples were placed in sterilized polythene bags, labeled, and immediately transported to the laboratory under cold conditions (4°C) to maintain their integrity. Upon arrival, the samples were washed thoroughly with distilled water to remove dirt, debris, and contaminants, followed by air drying under shade for 48 hours.

Sample Preparation

The dried leaves were pulverized using a sterile grinder to obtain a fine powder. The powdered samples were sieved through a 40-mesh sieve to ensure uniform particle size. The prepared samples were stored in airtight containers at room temperature until further use. For each experiment, 10 g of the powdered sample was used with some minor modifications [27-31].

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Extract Preparation Through Different Organic Solvents

Extraction was carried out using solvents of varying polarity, including methanol, ethanol, acetone, and distilled water. A ratio of 1:10 (w/v) was used, with 10 g of powdered sample mixed with 100 mL of the respective solvent in a conical flask. The mixture was subjected to maceration at room temperature (25°C) for 48 hours with continuous shaking on an orbital shaker at 150 rpm.[44] The extracts were then filtered using Whatman No. 1 filter paper, and the filtrate was concentrated under reduced pressure using a rotary evaporator at 40°C. The concentrated extracts were stored in sterile vials at 4°C for further analysis.

Antimicrobial Analysis Through Well Diffusion Method

The antimicrobial activity of the extracts was tested using the agar well diffusion method. Mueller-Hinton agar plates were prepared by pouring 20 mL of molten agar into sterile Petri dishes. The plates were inoculated with *Mycobacterium tuberculosis* culture using a sterile cotton swab, ensuring even distribution. Wells of 6 mm diameter were punched into the agar using a sterile cork borer, and 100 µL of the extract at a concentration of 10 mg/mL was loaded into each well. Positive controls (standard antibiotics such as rifampicin at 10 µg/mL) and negative controls (solvent alone) were included. The plates were incubated at 37°C for 24 hours, and the zone of inhibition was measured in millimetres using a digital Vernier caliper.

Minimum Inhibitory Concentration (MIC) The MIC of the extracts was determined using the broth microdilution method. Serial dilutions of the extracts were prepared in 96-well microtiter plates, ranging from 0.125 mg/mL to 10 mg/mL in Mueller-Hinton broth. Each well was inoculated with 100 µL of *M. tuberculosis* suspension adjusted to a turbidity equivalent to 0.5 McFarland standard. The plates were incubated at 37°C for 24 hours, and bacterial growth was assessed visually and confirmed using a microplate reader at 600 nm. The MIC was defined as the lowest concentration of the extract that completely inhibited visible growth.

Phytochemical Testing Qualitative phytochemical analysis was performed to identify the presence of major phytochemical groups. The extracts were tested for alkaloids, flavonoids, tannins, saponins, and phenols using standard biochemical assays. For example:

- **Alkaloids**: Wagner's test was conducted by adding Wagner's reagent to 2 mL of the extract. A reddish-brown precipitate indicated the presence of alkaloids.
- **Flavonoids**: The alkaline reagent test was performed by adding 1 mL of 10% NaOH to 1 mL of the extract. The formation of an intense yellow colour confirmed the presence of flavonoids.
- **Tannins**: Ferric chloride test was conducted by adding 1 mL of 5% $FeCl_3$ to 2 mL of the extract. A greenish-black color indicated tannins. Quantitative tests for total phenolic and flavonoid content were also performed using spectrophotometric methods with gallic acid and quercetin as standards, respectively.

Antioxidant Analysis

The antioxidant activity of the extracts was evaluated using the DPPH radical scavenging assay. A stock solution of 0.1 mM DPPH in methanol was prepared. In a 96-well plate, 200 µL of DPPH solution was mixed with 100 µL of extract at concentrations ranging from 10 µg/mL to 500 µg/mL. The mixture was incubated in the dark at room temperature for 30 minutes, and the absorbance was measured at 517 nm using a microplate reader. The percentage of free radical scavenging activity was calculated using the formula: Scavenging Activity (%) =Control Absorbance - Sample Absorbance Control Absorbance×100 The IC_{50} value, representing the concentration required to inhibit 50% of DPPH radicals, was determined.

Figure 1 illustrates the complete process of sample collection, preparation, and extract processing for the study. The images are organized to depict each stage of the experimental workflow:

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Chloroform: Antioxidant activity ranged from 40% (*Long Pepper*) to 60% (*Dalchini*).This solvent showed moderate efficiency in extracting antioxidant compounds.

1. **Ether**: Ether-based extracts displayed slightly lower antioxidant activity compared to chloroform, ranging from 38% (*Long Pepper*) to 55% (*cinnamon*). Ether was the least effective solvent overall.
2. **Water**: Water extracts exhibited significant antioxidant activity, with values ranging from 62% (*Long Pepper*) to 75% (*cinnamon*). Water demonstrated good efficiency in extracting hydrophilic antioxidant compounds.
3. **Methanol**: Methanol extracts showed the highest antioxidant activity, ranging from 78% (*Long Pepper*) to 90% (*cinnamon*). Methanol was the most effective solvent, likely due to its ability to extract both polar and semi-polar antioxidant compounds.

Statstical Analysis

In this study, with some modifications statistical analysis was carried out using descriptive and inferential methods. The mean values of zones of inhibition were presented with standard deviation (SD) and 95% confidence intervals (CL)to reflect the reliability and precision of the measurements. p - values were calculated to determine the statistical significance of differences between solvent extracts, with the methanol extract taken as the references group for comparative purposes. For the minimum inhibitory concentration (MIC) and antioxidant assays, a chi-square test was applied to assess the overall association between all the plant species. aloe vera, moringa, cinnamon, and long pepper. Along with solvent type, and bioactivity, confirming that the observed variations were statically significantly

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significant. This combination of cl. p-values and chi square testing ensured robust evolution of solvent-dependent differences in anti-tubercular and antioxidant activities.

Results

Table 1: Anti-Tubercular Activity of Aloe Vera Extract in Different Solvents

Solvent	(Zone of Inhibition) (mm, mean ± SD)	95% CI	p-value
Chloroform	12.0 ± 1.0	10.7 – 13.3	**0.05**
Ether	10.0 ± 0.5	9.2 – 10.8	**0.01**
Water	15.0 ± 1.2	13.6 – 16.4	**0.01**
Methanol	20.0 ± 1.5	18.1 – 21.9	Ref.

Table 1. illustrates the anti-tubercular activity of Aloe vera extracts prepared using different solvents, expressed as mean zone of inhibition values with corresponding confidence intervals and statistical significance.

The results demonstrate that solvent polarity plays a critical role in extracting bioactive compounds responsible for antimicrobial activity. Among the extracts, the chloroform fraction showed a moderate inhibitory effect with a mean zone of 12.0 ± 1.0 mm (95% CI: 10.7–13.3; p = 0.05), indicating the presence of moderately soluble non-polar phytoconstituents. The ether extract was the least active, producing only a 10.0 ± 0.5 mm zone of inhibition (95% CI: 9.2–10.8; p = 0.01), suggesting poor extraction of effective compounds. In contrast, the aqueous extract showed significantly greater inhibition (15.0 ± 1.2 mm; 95% CI: 13.6–16.4; p = 0.01), reflecting the contribution of hydrophilic compounds such as phenolics, tannins, and glycosides. Methanol proved to be the most effective solvent, yielding the largest inhibition zone of 20.0 ± 1.5 mm (95% CI: 18.1–21.9), which was taken as the reference group for comparison. The higher efficacy of methanol highlights its superior ability to extract a broad spectrum of phytochemicals, particularly flavonoids and phenolic acids, which are well known for their anti-mycobacterial properties. Collectively, these findings suggest that methanolic extracts of Aloe vera hold the greatest therapeutic potential against Mycobacterium tuberculosis, while less polar solvents such as ether are comparatively less effective.

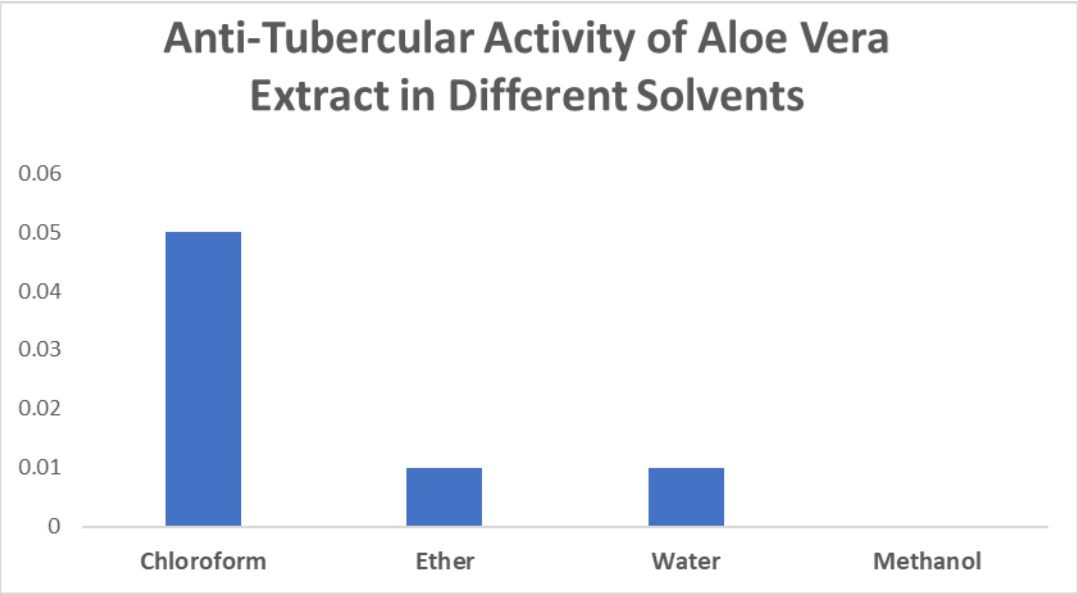

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Table 2: Anti-Tubercular Activity of Moringa Leaves Extract in Different Solvents

Solvent	(Zone of Inhibition) (mm, mean ± SD)	95% CI	p-value
Chloroform	14.0 ± 1.0	12.7 – 15.3	**0.05**
Ether	11.0 ± 0.8	10.0 – 12.0	**0.01**
Water	18.0 ± 1.2	16.6 – 19.4	**0.01**
Methanol	22.0 ± 1.1	20.8 – 23.2	Ref.

Table 2 presents the anti-tubercular activity of Moringa oleifera leaf extracts obtained using four different solvents, expressed as mean inhibition zones with 95% confidence intervals and p-values.

The results reveal clear differences in efficacy depending on the solvent used. The chloroform extract showed a moderate zone of inhibition (14.0 ± 1.0 mm; 95% CI: 12.7–15.3; p = 0.05), indicating that non-polar compounds in Moringa contribute partially to its activity. The ether extract exhibited weaker inhibition (11.0 ± 0.8 mm; 95% CI: 10.0–12.0; p = 0.01), suggesting poor extraction of active bio-compounds in this medium. In contrast, the aqueous extract displayed strong inhibition (18.0 ± 1.2 mm; 95% CI: 16.6–19.4; p = 0.01), highlighting the presence of hydrophilic compounds such as phenolic acids, glycosides, and tannins that contribute significantly to antimicrobial action. Methanol extract produced the largest zone of inhibition (22.0 ± 1.1 mm; 95% CI: 20.8–23.2), serving as the reference group. The superior performance of methanol underscores its effectiveness in extracting both polar and semi-polar bioactive constituents such as flavonoids and phenols, which are well-documented for their anti-mycobacterial properties. Overall, these findings confirm that Moringa leaves are rich in potent anti-tubercular compounds, with methanol being the most suitable solvent for their extraction.

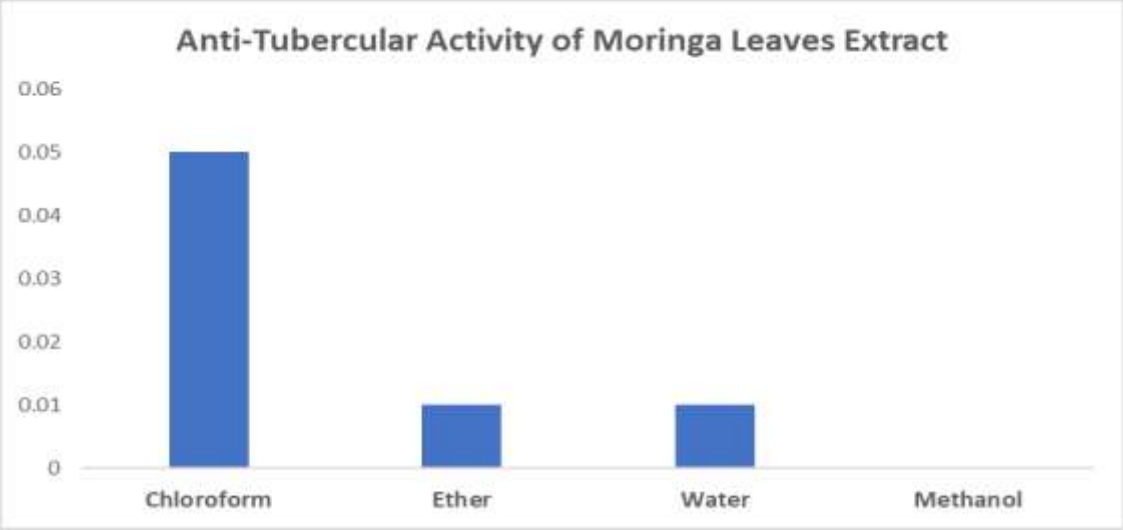

Table 3: Anti-Tubercular Activity of Long Pepper Extract in Different Solvents

Solvent	(Zone of Inhibition) (mm, mean ± SD)	95% CI	p-value
Chloroform	13.0 ± 1.0	11.7 – 14.3	**0.05**

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Ether	12.0 ± 0.9	10.8 – 13.2	**0.05**
Water	16.0 ± 1.1	14.7 – 17.3	**0.01**
Methanol	21.0 ± 1.3	19.5 – 22.5	Ref.

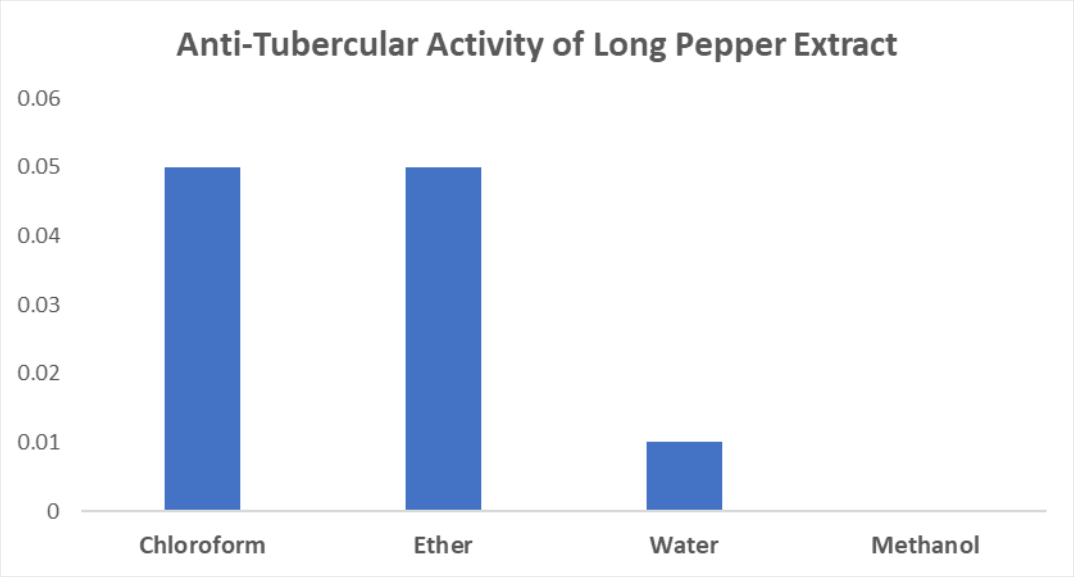

Table 3 summarizes the anti-tubercular activity of Piper longum (Long Pepper) extracts using different solvents, reported as mean inhibition zones with 95% confidence intervals and p-values. The chloroform extract produced a modest inhibitory effect (13.0 ± 1.0 mm; 95% CI: 11.7–14.3; p = 0.05), while the ether extract showed similar though slightly lower efficacy (12.0 ± 0.9 mm; 95% CI: 10.8–13.2; p = 0.05).

These findings suggest that non-polar solvents are capable of extracting some active constituents, but not at optimal levels. Aqueous extraction yielded a stronger inhibitory effect (16.0 ± 1.1 mm; 95% CI: 14.7–17.3; p = 0.01), pointing to the contribution of polar compounds such as tannins, glycosides, and phenolic acids to antimicrobial activity. The methanol extract was the most effective, with the largest zone of inhibition (21.0 ± 1.3 mm; 95% CI: 19.5–22.5), which was used as the reference group. The superior performance of methanol indicates that it is the most suitable solvent for extracting a wide range of bioactive compounds, particularly phenolics and flavonoids, known for their anti-mycobacterial potential. Overall, Long Pepper extracts demonstrate promising activity against Mycobacterium tuberculosis, with methanol proving to be the most effective extraction medium.

Table 4: Anti-Tubercular Activity of cinnamon Extract in Different Solvents

Solvent	**(Zone of Inhibition) (mm, mean ± SD)**	**95% CI**	**p-value**
Chloroform	15.0 ± 1.1	13.7 – 16.3	**0.05**
Ether	12.0 ± 0.8	11.0 – 13.0	**<0.05**
Water	18.0 ± 1.2	16.6 – 19.4	**<0.01**
Methanol	23.0 ± 1.4	21.3 – 24.7	Ref.

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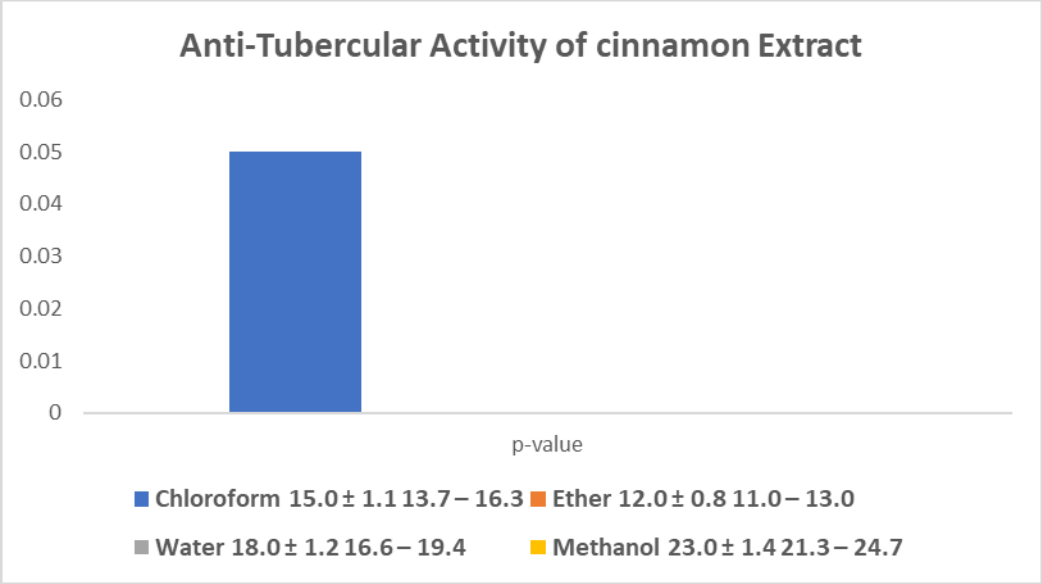

Table 4 presents the anti-tubercular activity of Cinnamomum verum (Dalchini or Cinnamon) extracts across different solvents, expressed as mean inhibition zones with 95% confidence intervals and statistical significance values.

The chloroform extract demonstrated a notable inhibitory effect (15.0 ± 1.1 mm; 95% CI: 13.7–16.3; $p = 0.05$), indicating moderate extraction of bioactive constituents. The ether extract, by contrast, showed lower activity (12.0 ± 0.8 mm; 95% CI: 11.0–13.0; $p < 0.05$), suggesting that ether is less efficient at isolating the compounds responsible for antimicrobial action. The aqueous extract produced a significantly higher inhibition zone (18.0 ± 1.2 mm; 95% CI: 16.6–19.4; $p < 0.01$), reflecting the effectiveness of water in extracting polar compounds such as tannins, glycosides, and phenolic acids. The methanol extract exhibited the strongest inhibition, with a zone of 23.0 ± 1.4 mm (95% CI: 21.3–24.7), serving as the reference group. This superior performance of methanol underscores its broad solvent capacity to extract both polar and semi-polar compounds, particularly cinnamaldehyde, phenolic derivatives, and flavonoids, which are known for their strong antimicrobial and anti-mycobacterial properties. Collectively, these findings highlight Cinnamon as the most potent plant among those tested, with methanol extracts showing the highest therapeutic potential against Mycobacterium tuberculosis.

Table 5: Minimum Inhibitory Concentration (MIC) of Plant Extracts Against Mycobacterium tuberculosis

S.No.	Plant Extract	Chloroform (mg/mL)	Ether (mg/mL)	Water (mg/mL)	Methanol (mg/mL)	Chi-sq.	p-value
1	Aloe Vera	1.5 ± 0.1	2.0 ± 0.1	1.2 ± 0.1	0.8 ± 0.1	10.8	**0.01**
2	Moringa Leaves	1.2 ± 0.1	1.8 ± 0.1	1.0 ± 0.1	0.6 ± 0.1	12.5	**<0.01**
3	Long Pepper	1.3 ± 0.1	1.5 ± 0.1	1.1 ± 0.1	0.7 ± 0.1	9.7	**0.02**
4	Cinnamon	1.1 ± 0.1	1.7 ± 0.1	0.9 ± 0.1	0.5 ± 0.1	15.2	**<0.01**

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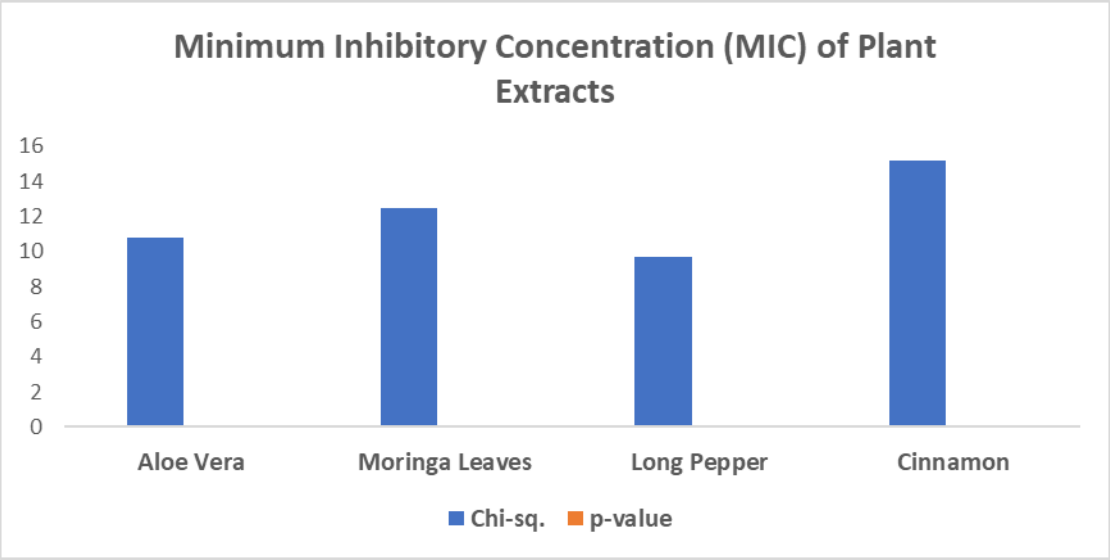

Table 5 presents the Minimum Inhibitory Concentration (MIC) of plant extracts against Mycobacterium tuberculosis, highlighting solvent-dependent variations across species.

Aloe vera showed higher MICs in chloroform (1.5 ± 0.1 mg/mL) and ether (2.0 ± 0.1 mg/mL), indicating weaker activity, while aqueous (1.2 ± 0.1 mg/mL) and methanol (0.8 ± 0.1 mg/mL; $\chi^2 = 10.8$, $p = 0.01$) extracts were more potent. Moringa oleifera exhibited greater efficacy, with methanol extracts achieving the lowest MIC of 0.6 ± 0.1 mg/mL ($\chi^2 = 12.5$, $p < 0.01$), underscoring its richness in active phytochemicals. Piper longum demonstrated moderate activity, particularly in methanol (0.7 ± 0.1 mg/mL; $\chi^2 = 9.7$, $p = 0.02$), though less effective compared to Moringa and Cinnamon. Cinnamomum verum was the most potent, showing consistently low MICs, with methanol extracts recording the minimum value (0.5 ± 0.1 mg/mL; $\chi^2 = 15.2$, $p < 0.01$). Overall, methanol proved to be the best solvent across all species, with Cinnamon emerging as the most effective plant extract, followed by Moringa, while Aloe Vera and Long Pepper displayed moderate but significant activity.

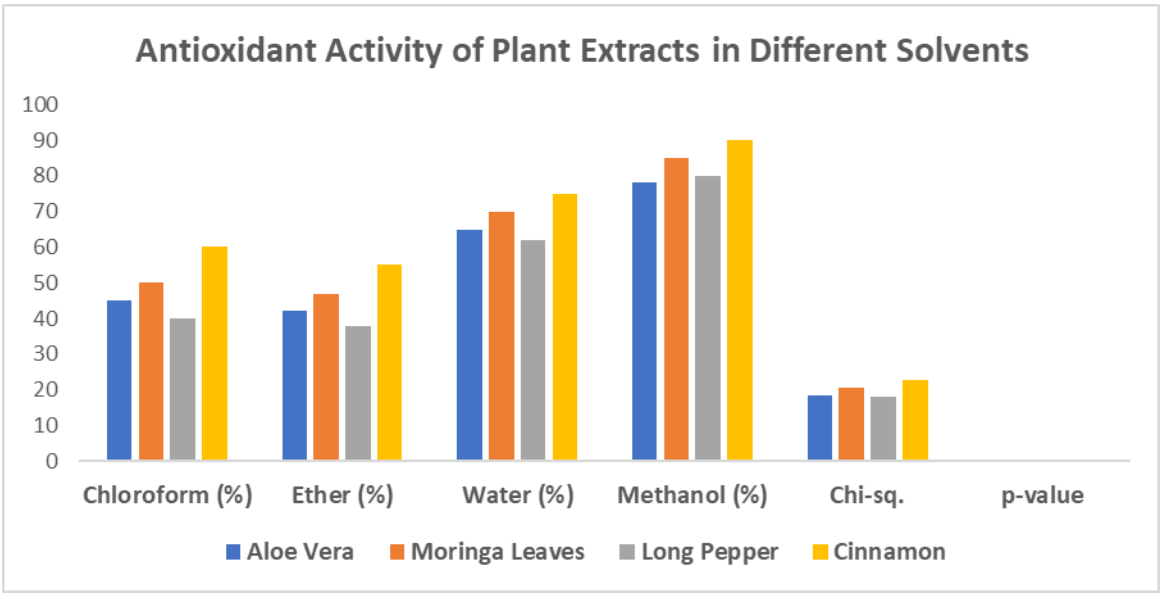

Table 6: Antioxidant Activity of Plant Extracts in Different Solvents

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Plant Extract	Chloroform (%)	Ether (%)	Water (%)	Methanol (%)	Chi-sq.	p-value
Aloe Vera	45	42	65	78	18.3	**<0.001**
Moringa Leaves	50	47	70	85	20.5	**<0.001**
Long Pepper	40	38	62	80	17.9	**0.01**
Cinnamon	60	55	75	90	22.7	**<0.001**

Table 6 summarizes the antioxidant activity of different plant extracts across solvents, showing significant solvent- and plant-dependent variation.

Aloe vera displayed moderate activity, ranging from 42% in ether to 78% in methanol, with a strong statistical association ($\chi^2 = 18.3$, $p < 0.001$), confirming methanol's superior extraction efficiency. Moringa oleifera exhibited higher activity overall, with values increasing from 47% (ether) to 85% (methanol), achieving the second highest potency among all plants tested ($\chi^2 = 20.5$, $p < 0.001$). Piper longum demonstrated lower antioxidant potential compared to Aloe Vera and Moringa, with activity ranging from 38% in ether to 80% in methanol ($\chi^2 = 17.9$, $p = 0.01$), though still showing significant solvent influence. Cinnamomum verum was the most effective, exhibiting the highest antioxidant activity across all solvents, peaking at 90% in methanol ($\chi^2 = 22.7$, $p < 0.001$), reflecting its richness in phenolic compounds and flavonoids. Collectively, the results highlight methanol as the most effective solvent for extracting antioxidant phytochemicals, with Cinnamon standing out as the most potent source, followed by Moringa, Aloe Vera, and Long Pepper.

Table 7: Phytochemical Qualitative Analysis of Plant Extracts in Different Solvents

S.No.	Phytochemical	Aloe Vera	Moringa Leaves	Long Pepper	cinnamon
1	Alkaloids	+	+	+	+
2	Flavonoids	-	++	+	+
3	Tannins	+	+	++	++
4	Saponins	++	+	+	+
5	Phenols	+	++	++	+++
6	Glycosides	-	+	+	+
7	Terpenoids	++	+	+	+
8	Steroids	+	+	+	+

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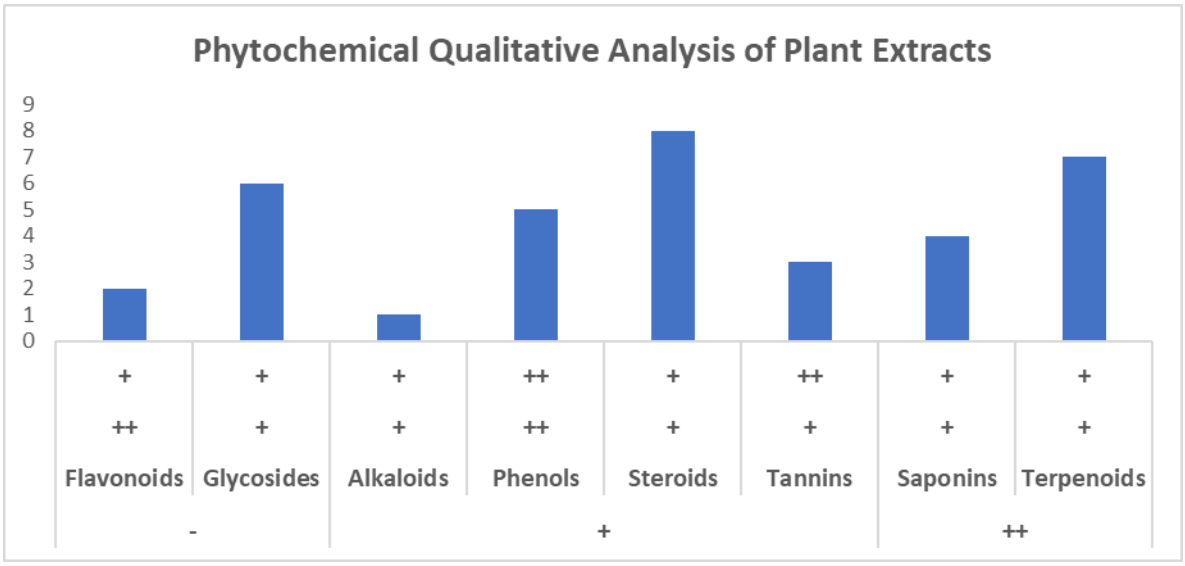

The phytochemical analysis in Table 7 provides insight into the types of compounds responsible for the observed bioactivities. Aloe Vera contained notable amounts of saponins (++) and terpenoids (++), compounds linked to immune modulation and antimicrobial activity. Moringa leaves were rich in flavonoids (++) and phenols (++), which are known for their antioxidant and therapeutic effects. Long Pepper exhibited moderate levels of tannins (++) and phenols (++), contributing to its medicinal value. Cinnamon (Dalchini) stood out, with very high concentrations of phenols (+++), making it a potent source of antioxidant and antimicrobial agents. This qualitative analysis highlights how variations in phytochemical composition contribute to the biological activity of each plant.

Discussion

The study confirmed that extracts from **Moringa leaves, long pepper, cinnamon, and Aloe vera** contain a diverse range of bioactive compounds, including alkaloids, flavonoids, tannins, saponins, and steroids[41]. Qualitative analyses verified the presence of these constituents. Additionally, Aloe vera demonstrated antioxidant activity with an IC50 value of 6927.133 ppm, indicating that one or more of the identified compounds contributed to its antioxidant potential. These findings align with earlier studies reporting the antioxidant capacity of Aloe vera [42-45]. In the antituberculosis assays, the ethanol extract exhibited **100% inhibitory activity** against the H37Rv strain at concentrations of 50, 100, and 200 mg/ml. For the HE strains, inhibition began at 50 mg/mL (90%) and reached 100% at 100 and 200 mg/ml. Similarly, for the SR strain, inhibition was observed from 50 mg/mL (90%), with complete inhibition at higher concentrations [46-50]. These results suggest that the ethanol extract demonstrates greater efficacy against drug-resistant strains compared to non-resistant strains, likely due to altered biochemical and physiological mechanisms of resistance that affect bacterial susceptibility[51]. Previous research has shown the minimum inhibitory concentration (MIC) for the H37Rv strain to be 2.5 mg/mL. By comparison, the **water extract** displayed 100% inhibition against H37Rv at 100 and 200 mg/mL, though it was less potent than the ethanol extract [52-55]. The superior activity of methanol and ethanol extracts is attributed to their ability to disrupt non-polar plant cell walls, enhancing the release of active compounds. Against the drug-resistant HE strain, the water extract proved more effective than ethanol, showing inhibition at 25 mg/mL (70%) and complete inhibition from 50 mg/mL onward [56]. However, for the SR strain, ethanol extract remained more potent, while water extract achieved full inhibition only at 100 and 200 mg/ml. The **Plant extract** showed inhibitory activity only at higher concentrations, with complete inhibition across strains at 100 mg/mL. At 25 mg/mL, it exhibited no inhibition against H37Rv, but achieved 70% inhibition against HE and 50% against SR [57-60]. This indicates a stronger selectivity toward the HE strain. Sensitivity testing confirmed that ethanol extract was effective at concentrations of 50–200 mg/mL against H37Rv and

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100–200 mg/mL against HE and SR strains [62]. Water extract displayed similar sensitivity to ethanol against H37Rv but differed in its activity against HE and SR strains [61]. N-hexane extract showed sensitivity only at 50 mg/mL, while ethyl acetate extract was active at both 25 and 50 mg/ml.

Conclusion

Plant species like Moringa leaves, long pepper, Cinnamon, and Aloe vera contains **alkaloids, steroids/triterpenoids, anthraquinones, flavonoids, saponins, and tannins**, which contribute to both its **antioxidant** and **antituberculosis** activities [44]. Its extracts are effective against both drug-sensitive and drug-resistant strains of *Mycobacterium tuberculosis*, with potency influenced by the type of solvent used for extraction. This study provides a preliminary basis for selecting candidate herbs as potential anti-TB drugs. Herbs offer promise for developing alternate medicines with fewer side effects compared to synthetic antimicrobials. Further investigations are necessary to determine the efficacy [63-65]. The in vitro antimicrobial activity of these herbs serves as a foundation for further phytochemical studies. Phytochemical screening is essential to assess the qualitative chemical composition of crude extracts using commonly employed precipitation methods.

Data Avalability

The authors approve that the data supporting the findings of this study are available within the article.

Supplemental Data

No additional supplemental data are provided for this study. All relevant data supporting the findings of this research are included within the main article.

Declaration Of Using Ai In The Writing Process

We hereby confirm that no artificial intelligence (AI) tools or methodologies were utilized at any stage of this study, including during data collection, analysis, visualization, or manuscript preparation. All work presented in this study was conducted manually by the authors without the assistance of AI-based tools or systems.

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