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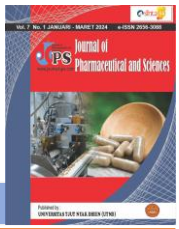

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Effectiveness of Mimosa pudica Extract on Reducing the Number of *Blastocystis sp*. in Wistar Rats with Acute Diarrhea Model

Efektivitas Ekstrak Mimosa Pudica Terhadap Penurunan Jumlah *Blastocystis sp*. Pada Tikus Wistar Model Diare Akut

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Abstract

Introduction: *Blastocystis* sp. infection is a prevalent cause of gastrointestinal disorders, contributing to acute diarrhea through inflammatory mechanisms and intestinal mucosal damage. Although metronidazole remains the standard antiparasitic therapy, its use is constrained by side effects and variable efficacy, underscoring the need for safer and more effective alternative treatments derived from natural sources. *Mimosa pudica* is known to contain bioactive compounds, including flavonoids, tannins, saponins, and alkaloids, which exhibit antiparasitic and anti-inflammatory activities. **Objective:** This study aims to evaluate the effect of *Mimosa pudica* leaf extract on reducing parasite numbers in a Wistar rat model of acute diarrhea induced by *Blastocystis* sp. **Methods:** Leaf extracts were prepared using ethanol as a solvent and were characterized using Fourier Transform Infrared (FTIR) spectroscopy and phytochemical screening. An *in vivo* test was conducted in 40 male Wistar rats, divided into six groups: a negative control, a positive control, two metronidazole-treated groups (9 and 13.9 mg), and two extract-treated groups (40 and 80 mg). The primary parameters observed included parasite count in fecal samples, stool consistency, and motor activity. **Results:** Phytochemical analysis confirmed the presence of flavonoids, tannins, saponins, and alkaloids in the extract. FTIR spectra revealed functional groups such as hydroxyl (–OH), carbonyl (C=O), and aliphatic groups (C–H), which are characteristic of bioactive polyphenolic compounds. *In vivo* results demonstrated that *Mimosa pudica* extract significantly reduced Blastocystis sp. burden and improved clinical conditions, including stool consistency, in a dose-dependent manner. The 80 mg dose exhibited the highest efficacy, with 80% of samples becoming parasite-free by day 7 of treatment. **Conclusion:** The findings indicate that *Mimosa pudica* extract exhibits significant antiparasitic activity against Blastocystis sp. and shows potential as a therapeutic agent for treating protozoan infections, particularly in the context of acute diarrhea. Further research is warranted to elucidate the precise molecular mechanisms and to evaluate its safety and clinical applicability.

Keywords: Mimosa pudica, Extract, Blastocystis sp, Diarrhea

Abstrak

Pendahuluan: Infeksi *Blastocystis* sp. merupakan penyebab umum gangguan saluran pencernaan yang berkontribusi terhadap diare akut melalui mekanisme inflamasi dan kerusakan mukosa usus. Meskipun metronidazol masih menjadi terapi antiparasit standar, penggunaannya dibatasi oleh efek samping dan variasi efektivitas, sehingga mendorong perlunya terapi alternatif yang lebih aman dan efektif yang berasal dari sumber daya alam. *Mimosa pudica* diketahui mengandung senyawa bioaktif seperti flavonoid, tanin, saponin, dan alkaloid yang memiliki aktivitas antiparasit dan antiinflamasi. **Tujuan:** Penelitian ini bertujuan untuk mengevaluasi efek ekstrak daun *Mimosa pudica* terhadap penurunan jumlah parasit pada model tikus Wistar yang mengalami diare akut yang diinduksi oleh *Blastocystis* sp. **Metode:** Ekstrak daun disiapkan menggunakan pelarut etanol dan dikarakterisasi menggunakan spektroskopi Fourier Transform Infrared (FTIR) serta dilakukan skrining fitokimia. Uji *in vivo* dilakukan pada 40 ekor tikus Wistar jantan yang dibagi menjadi enam kelompok: kontrol negatif, kontrol positif, dua kelompok metronidazol (9 dan 13,9 mg), serta dua kelompok perlakuan ekstrak (40 dan 80 mg). Parameter yang diamati meliputi jumlah parasit pada sampel feses, konsistensi tinja, dan aktivitas motorik. **Hasil:** Analisis fitokimia mengonfirmasi adanya flavonoid, tanin, saponin, dan alkaloid dalam ekstrak. Spektrum FTIR menunjukkan gugus fungsi seperti hidroksil (–OH), karbonil (C=O), dan gugus alifatik (C–H) yang merupakan karakteristik senyawa polifenol bioaktif. Hasil *in vivo* menunjukkan bahwa ekstrak *Mimosa pudica* secara signifikan menurunkan jumlah *Blastocystis* sp. dan memperbaiki kondisi klinis, termasuk konsistensi tinja, pada berbagai dosis. Dosis 80 mg menunjukkan efektivitas tertinggi, dengan 80% sampel bebas parasit pada hari ke-7 pengobatan. **Kesimpulan:** Temuan ini menunjukkan bahwa ekstrak *Mimosa pudica* memiliki aktivitas antiparasit yang signifikan terhadap *Blastocystis* sp. dan berpotensi menjadi agen terapeutik alternatif yang menjanjikan untuk infeksi protozoa, terutama pada kasus diare akut. Penelitian lebih lanjut diperlukan untuk menguraikan mekanisme molekuler yang spesifik serta mengevaluasi keamanan dan potensi aplikasinya secara klinis.

Kata kunci: Mimosa pudica, ekstrak, Blastocystis sp, Diare.

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Introduction

The prevalence of intestinal protozoan infections in humans and animals has led to increased global attention to the pathogen *Blastocystis* sp. This protozoan is not only widely known for its cosmopolitan distribution but also for the complexity of its clinical manifestations, which range from asymptomatic to causing chronic gastrointestinal disorders such as diarrhea, abdominal pain, and irritable bowel syndrome [1]. *Blastocystis* sp. is a common intestinal protozoan found in humans and animals, and its presence has attracted growing clinical attention due to its association with gastrointestinal disorders, including diarrhea, abdominal pain, and irritable bowel syndrome. Although metronidazole has long been the conventional therapy for *Blastocystis* sp., resistance and long-term side effects have prompted the search for safer, more effective, and sustainable alternative therapies [2].

This situation highlights the need for the development of safer, more effective, and less resistant alternative antiparasitic therapies. In recent years, phytotherapy has emerged as a promising approach to enhance the efficacy of natural therapies by utilizing plant bioactive compounds, showing significant potential in addressing the challenges of parasitic infection therapy. This approach emphasizes the use of plant extracts rich in bioactive compounds as natural therapeutic agents. The combination of various secondary metabolites in plant extracts is known to enhance bioactivity, stability, and target specificity [3]. This method is not only environmentally friendly but also enables the incorporation of various plant-derived active compounds, thereby enhancing the biological activity and stability of formulations [4,5]. One promising approach is the use of plant extracts containing bioactive compounds with synergistic effects on antimicrobial and antiparasitic activities [6].

Mimosa pudica, or the sensitive plant, is a medicinal plant widespread in tropical regions and is known to contain various bioactive compounds such as flavonoids, tannins, alkaloids, and saponins. These compounds have been reported to exhibit antioxidant, antimicrobial, and antiparasitic activities [7]. However, the potential of Mimosa pudica extract in treating *Blastocystis* sp. infections has not been comprehensively studied, especially in animal models. Previous studies have primarily focused on in vitro testing or conventional formulations, without directly assessing the biological efficacy of the extract against parasitic infections [8,9].

This study aims to characterize Mimosa pudica extract and test its biological activity against *Blastocystis* sp. infection in experimental animals. Comprehensive analysis was conducted, including phytochemical testing, physicochemical characterization (FTIR), and in vivo validation. By combining pharmacognostic approaches and the exploration of plant bioactive compounds, this study is expected to make significant scientific contributions to the development of more effective and safe plant-based antiparasitic agents for therapeutic applications.

Experimental Section

Materials and Equipment

The materials used in this study include Mimosa pudica leaves, $FeCl_3$ solution, concentrated HCl, magnesium powder, Mayer and Dragendorff reagents, and distilled water. Feces, horse serum. The main equipment used includes Fourier Transform Infrared Spectroscopy (FTIR), a Scanning Electron Microscope with an EDX detector, and SEM processing software (such as SmartPLS or AMOS).

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Extraction of Active Compounds from Mimosa Leaves

Fresh Mimosa pudica leaves were washed, dried, and ground into a fine powder. A total of 100 grams of leaf powder was extracted with 96% ethanol by maceration for 72 hours, with frequent stirring. The filtrate was filtered and evaporated to obtain a concentrated extract.

Qualitative Phytochemical Test

Phytochemical tests are conducted to detect the presence of tannins, saponins, flavonoids, and alkaloids in the extract. The method used includes Tannin: the extract is added to a 1% $FeCl_3$ solution; a greenish-black color change indicates a positive result. Saponin: The extract is shaken with distilled water, and the formation of stable foam indicates the presence of saponins. Flavonoid: The extract is mixed with magnesium powder and concentrated hydrochloric acid; a brownish-yellow color change indicates a positive reaction. Alkaloid: The extract is added to Mayer's or Dragendorff's reagent; the formation of a precipitate indicates the presence of alkaloids.

FTIR Analysis

Functional group analysis was performed using FTIR in the wavenumber range of 4000–500 cm^{-1}. The detected peaks were compared with reference spectra to identify the main functional groups.

Test Animals and Acclimatization

A total of 40 male Wistar rats (*Rattus norvegicus*), aged 8–10 weeks and weighing between 160 and 190 grams, were utilized in this study. The animals were sourced from a certified laboratory animal facility and underwent a seven-day acclimatization period prior to the commencement of the experiment. During this period, the rats were housed individually in standard polypropylene cages under controlled environmental conditions, maintained at a room temperature of 22–25°C with a consistent 12-hour light/dark cycle. All animals were provided with standard commercial rodent chow and had *ad libitum* access to drinking water.

Dosage Determination

The doses for both the *Mimosa pudica* extract and the positive control drug, metronidazole, were calculated based on the average body weight of the experimental animals. For the *Mimosa pudica* extract, two treatment doses were prepared. The low dose was calculated as 200 mg/kg body weight, which, for a 0.2 kg rat, corresponds to 40 mg. The high dose was calculated to be 400 mg/kg body weight, equivalent to 80 mg per rat. For the standard therapy, metronidazole was administered at two different dose levels. The first dose was prepared at 750 mg/kg of body weight, resulting in a 13.5 mg dose per 0.2 kg rat. The second dose was prepared at 500 mg/kg body weight, corresponding to 9 mg per animal.

Treatment Group Design

Table 1. Mice were randomly divided into four main groups, each consisting of 5 mice:

Group	Treatment
K- (Negative Control)	not infected with *Blastocystis* sp., not treated, and only given distilled water
K+ (Positive Control)	Inoculated with *Blastocystis* sp. without treatment and only given distilled water
K+O (Positive Control with Treatment)	Inoculated with *Blastocystis sp.* and given Metronidazole 9 mg and 13.9 mg
P (extractTreatment)	Inoculated with *Blastocystis sp.* and extracted from *Mimosa pudica* at doses of 40 and 80 mg

Preparation and Inoculation of *Blastocystis* sp.

Blastocystis sp. was obtained from human fecal isolates of patients who had previously been cultured on Jones medium for 72 hours. The suspension was concentrated by centrifugation (1500 rpm, 15 minutes), washed twice with PBS pH 7.4, and diluted to a final concentration of 10^5 cells/mL. Each mouse in the K+, K+O, and P groups was inoculated orally with 0.5 mL (10^5 cells/mouse) using a probe on day 7 after acclimatization. Parameters Observed Observations were made before inoculation (day 0), 14 days after inoculation, and after treatment, including: Wistar rats' body weight (grams), Motor activity (active, less active) based on direct observation.

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Research Ethics

This research is an experimental study with ethics approval from the Health Research Ethics Committee of the Faculty of Medicine, Andalas University, under ethics review number 22/UN/16.3/KEP-FK/2025.

Results and Discussion

Phytochemical Analysis

Phytochemical analysis was conducted to identify bioactive compounds in the nanoparticle system. The results of the phytochemical analysis of Mimosa pudica extract in this study are presented in Table 3. Qualitative examination results indicated that it contained bioactive compounds, including flavonoids, tannins, saponins, and alkaloids. Detection of each compound was performed using the test tube method, which uses specific reagents and observes color changes or foam formation as positive indicators. The presence of secondary metabolites in the nanoparticle system indicates that the active compounds from the plant extract remain stable after synthesis and likely bind to or adsorb onto the surfaces of the metal particles [10].

Table 2. Phytochemical Test Results of Mimosa Extract

Type of Compound	Test Method	Positive Indicator	Test Results
Tannin	Addition of 1% $FeCl_3$ solution	Blackish green color	Positive (+)
Saponin	Shake vigorously in distilled water, observe foam formation	Stable foam with a bright green color	Positive (+)
Flavonoids	Addition of magnesium powder and concentrated hydrochloric acid	Yellow brown/turbid color	Positive (+)
Alkaloid	Addition of Mayer's reagent or Dragendorff's reagent	Blackish-green precipitate	Positive (+)

The detected flavonoids play a crucial role as reducing agents in synthesis, thus functioning in particle nucleation and growth. Furthermore, flavonoids act as stabilizing (capping) agents that inhibit particle agglomeration, maintain colloidal stability, and preserve particle size [5]. The presence of tannins in the extract also contributes significantly, as these compounds have a polyphenolic structure that enables them to form strong bonds with metal surfaces via phenolic and carbonyl groups. A study by Sreenivasulu et al. [11] showed that Mimosa pudica extract, which is rich in tannins, can produce irregular yet stable and bioactive morphologies.

Saponins detected in phytochemical tests also increase compound solubility in aqueous solutions and provide additional stability to the system. Saponins are amphiphilic and can form a natural surfactant layer that protects the extract from coagulation. Furthermore, saponins exhibit immunomodulatory and cytotoxic effects against pathogenic microorganisms, which may contribute to antiparasitic activity [12]. The alkaloids identified also have potential as bioactive agents that disrupt the structure and function of microbial cell membranes and increase the permeability of parasite cell walls, thereby enhancing their antimicrobial efficacy [13]. Compared with previous research, these results align with the findings of Rizwan et al. [12], who reported that Mimosa pudica extract contains active compounds that contribute to its high biological activity. Research by Sreenivasulu et al. [11] also stated that the combination of tannins and flavonoids in the extract can accelerate wound healing and reduce inflammation in animal models.

The presence of various secondary metabolites supports the hypothesis that the Mimosa pudica-derived system relies not only on the antimicrobial effect of silver ions but also on a synergistic mechanism between the particle core and the surrounding phytochemicals, thereby enhancing biological activity. A study by Singh et al. [14] showed that extracts coated with plant-derived active compounds exhibited higher bioactivity than synthetic extracts lacking natural protective agents. In the context of this study, the successful preservation of active compounds after further synthesis is supported by FTIR results, which indicate the presence of hydroxyl (–OH), carbonyl (C=O), and ether (C–O–C) groups characteristic of polyphenols and glycosides. Overall, the presence of complex secondary metabolites in Mimosa pudica extract supports its use as an active ingredient in extraction.

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FTIR spectroscopic characterization

FTIR spectroscopic characterization was used in this study to verify the functional groups involved in particle synthesis and stabilization. The FTIR results from this study are presented in Table 3.

Table 3. Identification of Functional Groups Based on FTIR Spectra of *Mimosa pudica* Extract

Wavenumber (cm^{-1})	Type of Vibration	Indicated Functional Groups	Interpretation
3313.7	O–H Stretching (broad)	Hydroxyl (–OH) from phenol or alcohol	Polyphenolic compounds or flavonoids
2945.35	C–H stretch (asymmetric)	Methyl/methylene groups ($–CH_3$, $–CH_2$)	Aliphatic organic compounds
2833.48	C–H stretching (symmetric)	Alkyl group (alkanes)	Lipid components or hydrocarbon chains
1659.77	C=O or C=C stretching	Carbonyl (amide, ester) or double bond	Indicates the possible presence of phenolic compounds/amides
1450.49	C–H bending	Alkane group	Simple aliphatic compounds
1112.95	C–O–C or C–N stretching	Ether or amine	Flavonoid or glycoside structure
1021.33	C–O stretching	Alcohol/ester	Polar compounds, phenolic or sugar

Fourier Transform Infrared Spectroscopy (FTIR) analysis was performed to identify the main functional groups in the extract sample using infrared absorption patterns at specific wavenumbers. Interpretation of the FTIR spectra of complex extracts is preliminary because a single absorption peak can arise from multiple overlapping functional groups. Therefore, the FTIR results in this study are used as an initial indication of the presence of a particular functional group and not to confirm the specific identity of the compound [15]. The broad absorption peak at 3313.7 cm^{-1} indicates O–H stretching vibrations, typically associated with compounds containing hydroxyl groups, such as phenols or alcohols. This group is often found in secondary plant metabolites, particularly phenolic compounds.

The absorption at 2945.35 cm^{-1} and 2833.48 cm^{-1} indicate aliphatic C–H stretching vibrations of methyl and methylene groups, respectively. These peaks indicate the possible presence of aliphatic organic compounds in the extract. The peak at 1659.77 cm^{-1} indicates the possible presence of vibrations of the C=O group or a C=C double bond. Absorption in this region could be attributed to carbonyl-containing or conjugated organic compounds, but specific identification cannot be determined from FTIR analysis alone [17]. Furthermore, the absorption at 1450.49 cm^{-1} indicates C–H bending vibrations, supporting the presence of aliphatic compounds in the sample. The peaks at 1112.95 cm^{-1} and 1021.33 cm^{-1} indicate vibrations of oxygen-containing groups, such as C–O, which are commonly found in alcohols, phenolics, or other polar organic compounds. However, determining the specific type of functional group requires further analysis. In general, FTIR results indicate the presence of several major functional groups, including hydroxyl (O–H), aliphatic (C–H), carbonyl or double bonds (C=O/C=C), and oxygen-containing groups (C–O). However, because the sample is a complex extract, FTIR results cannot be used to identify the specific compound type. Therefore, supporting analyses such as phytochemical screening, thin-layer chromatography (TLC), or other characterization methods are needed to confirm the compound content in the

This finding is consistent with previous research by Singh et al. [14], who reported that the FTIR spectra of biologically synthesized extracts consistently exhibit peaks below 700 cm^{-1}, indicating the formation of metal-biomolecular bonds. The presence of these functional groups indicates that the bioactive compounds in Mimosa pudica extract are not degraded during synthesis and remain active in the system. Thus, the extract not only carries an antimicrobial metal load but is also coated with phytochemical compounds that exert a synergistic effect on its biological activity. Madaniyah et al. [15] stated that one advantage of green synthesis methods is plants' ability to produce multifunctional nanoparticles through the action of naturally occurring bioactive compounds. These results are consistent with previous research by Sreenivasulu et al. [11], who found that Mimosa pudica extract is rich in flavonoids, tannins, and hydroxyl-containing compounds that can reduce metal ions to high-stability states. Another study by Singh et al. [14] showed that phenolic compounds from this plant play a crucial role in the formation of extracts with high antibacterial activity.

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In-Vivo Analysis

Table 4. Fecal Consistency and Motor Activity in Mimosa Pudica Extract-treated and Control Wistar rats Model Acute Diarrhea

Group	n	Stool Consistency		
		Beginning	14 days of diarrhea	End
Negative Control	5	0	0	0
Positive Control	5	1	1	1
Metronidazole 9 mg	5	0	1	0
Metronidazol 13,9 mg	5	0	1	0
Mimosa pudica extract 40 mg	5	0	1	0
Mimosa pudica extract 80 mg	5	0	1	0

*Use Fecal Score: Score 0 (solid), 1 (soft/formed), 2 (paste/unformed), 3 (liquid).

The results showed that administration of Mimosa pudica extract at doses of 40 mg and 80 mg improved stool consistency from semi-solid to solid in Wistar rats after 14 days of diarrhea induction. These findings indicate that Mimosa pudica extract has a therapeutic effect in improving the clinical condition of acute diarrhea caused by *Blastocystis* sp. In contrast, the positive control group showed stool that remained semi-solid, indicating an untreated parasitic infection. The improvement in the condition in the treatment group is thought to be related to the content of secondary metabolites in Mimosa pudica, such as flavonoids, tannins, saponins, and alkaloids, which have antiparasitic and anti-inflammatory activities [18]. Flavonoid compounds are known to damage parasite cell membranes and inhibit energy metabolism, thereby reducing parasite numbers [13]. In addition, tannins help precipitate proteins and disrupt the integrity of the microorganism's cell wall [12].

The results of this study also indicate that the effectiveness of Mimosa pudica extract in improving clinical conditions is comparable to, if not superior to, standard therapies such as metronidazole, which in this study still showed a decrease in activity at the end of treatment. This is in line with reports that the use of natural ingredients is associated with fewer side effects and broader pharmacological activity [8]. In addition, the anti-inflammatory activity of Mimosa pudica plays an important role in reducing intestinal inflammation caused by Blastocystis sp. infection, thereby helping to restore stool consistency and gastrointestinal function. Phenolic compounds have been reported to suppress the production of inflammatory mediators, such as pro-inflammatory cytokines, and to improve tissue regeneration [18]. Another study reported that Mimosa pudica extract exhibits antioxidant activity that can protect tissues from oxidative damage during infection [20]. Overall, the results of this study indicate that Mimosa pudica extract has significant potential as an alternative therapy in treating *Blastocystis* sp. infection in acute diarrhea. However, further research is still needed to evaluate the specific molecular mechanisms, conduct long-term toxicity testing, and validate in humans to ensure its safety and clinical effectiveness.

Table 5. Differences In the Number of *Blastocystis* Sp. in Feces Samples After Mimosa Pudica Extract Treatment

Kelompok	n	f (%)
Negative Control	5	
There is no *Blastocystis* sp.		5 (100,0)
Positive Control	5	
There is *Blastocystis* Sp		5 (100,0)
Metronidazole 9 mg	5	
No *Blastocystis* Sp on the 3rd day of treatment		1 (20,0)
No *Blastocystis* Sp on the 4th day of treatment		2 (40,0)
No *Blastocystis* Sp on the 5th day of treatment		2 (40,0)
Metronidazol 13,9 mg	5	
No *Blastocystis* Sp on the 4th day of treatment		1 (20,0)
No *Blastocystis* Sp on the 5th day of treatment		4 (80,0)
Ekstrak *Mimosa pudica* 40 mg	5	
No *Blastocystis* Sp on the 8th day of treatment		2 (40,0)
No *Blastocystis* Sp on the 10th day of treatment		3 (60,0)
Ekstrak *Mimosa pudica* 80 mg	5	
No *Blastocystis* Sp on the 7th day of treatment		4 (80,0)
No *Blastocystis* Sp on the 8th day of treatment		1 (20,0)

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The results of the study showed a clear difference in the elimination time of *Blastocystis* sp. between treatment groups. The negative control group did not detect Blastocystis sp. in any sample (100%). In contrast, the positive control group detected parasites in all samples (100%), confirming successful induction of infection in the mouse model. In the standard therapy group, administration of Metronidazole at a dose of 9 mg showed gradual parasite elimination from day 3 to day 5. In contrast, the 13.9 mg dose showed faster elimination, with 80% of samples being parasite-free on day 5. This is in accordance with reports that metronidazole works by damaging protozoan DNA through the formation of free radicals, thereby inhibiting replication and causing parasite cell death [21]. However, the variation in responses found in this study is also consistent with reports of resistance and inconsistent effectiveness of metronidazole against *Blastocystis* sp. [3]. In the Mimosa pudica extract treatment group, the 40 mg dose showed slower parasite elimination, occurring on days 8 to 10. Conversely, the 80 mg dose was more effective, with 80% of samples parasite-free on day 7 and 100% on day 8. This suggests a dose-response relationship, in which increasing the extract dose accelerates parasite elimination.

The effectiveness of Mimosa pudica extract is thought to be related to its content of bioactive compounds, including flavonoids, tannins, saponins, and alkaloids, which exhibit antiparasitic activity. Flavonoids are known to disrupt cell membranes and impair parasite energy metabolism, while saponins can increase cell membrane permeability, leading to cell lysis [13]. Tannins also play a role in protein precipitation and in inhibiting important enzymes in microorganisms. Furthermore, the phenolic compounds in this plant have anti-inflammatory effects that help repair intestinal mucosal damage caused by infection [21]. Although the parasite elimination time with Mimosa pudica extract is relatively longer than with metronidazole, the results of this study indicate that this extract remains significantly effective, especially at high doses. The main advantages of nature-based therapies are the potential for fewer side effects and broader pharmacological activity, including antioxidant and immunomodulatory effects [8]. Overall, the results of this study indicate that Mimosa pudica extract has potential as an alternative therapy in the treatment of *Blastocystis* sp. infections, especially in conditions where standard therapies are limited. However, further research is needed to evaluate the molecular mechanism of action, conduct toxicity tests, and conduct clinical trials in humans to ensure its safety and effectiveness more broadly.

Form Of *Blastocystis* Sp. In Fecal Samples After Mimosa Pudica Extract Treatment

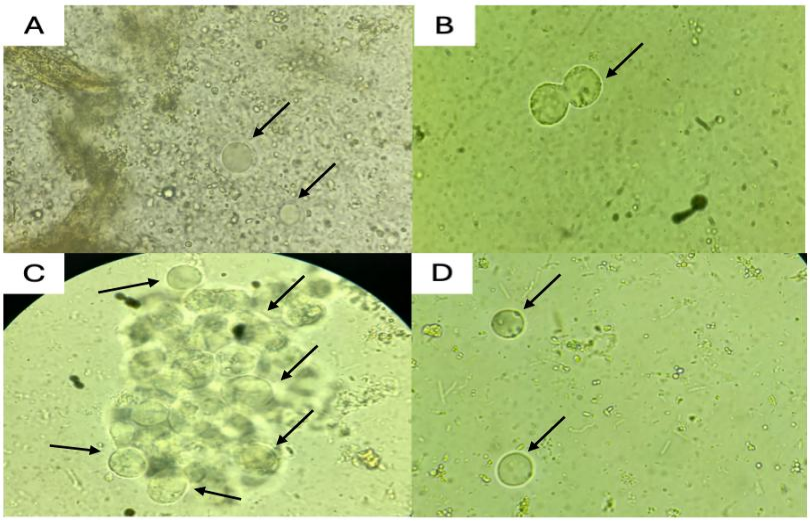

Figure 1. Various types of stool samples were used in the study. A: Child stool sample; B and C: stool samples cultured in horse serum; D: Wistar rats' stool sample. Each image shows a different stool condition, both in its natural state and after culture.

Figure 1 shows the variation in the morphology and composition of microorganisms in stool samples from different sources, both in their natural state and after culture. In panel A (child stool sample), the diversity of microscopic structures is visible, reflecting the complex composition of the human gut microbiota. This diversity is a normal physiological condition that helps maintain the balance of the gut ecosystem and digestive function [15]. In panels B and C (fecal samples cultured in horse serum), an increase in the number and regularity of microorganisms is visible.

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The culture process allows the growth and proliferation of protozoa, including *Blastocystis* sp., thus facilitating microscopic identification. Culture media, such as serum, are known to provide optimal nutrition for the growth of intestinal protozoa and to increase detection sensitivity compared with direct examination [4]. This explains why the structure of microorganisms in panels B and C appears clearer and more organized than in panel A. Meanwhile, panel D (mouse fecal sample) shows differences in microflora composition compared to humans. Species factors, diet, and the physiological condition of the test animal influence this difference. Mice, as an animal model, have a simpler gut microbiota but remain representative for the study of gastrointestinal infections, including *Blastocystis* sp. infections [16]. Overall, the morphological differences observed in each panel indicate that environmental conditions, culture methods, and host types greatly influence the characteristics of microorganisms in feces. This finding is important for supporting the validity of the Blastocystis sp. identification method and the interpretation of research results, especially for distinguishing infection status before and after treatment. In addition, the use of culture methods has been shown to increase the accuracy of detecting intestinal protozoa, which are often difficult to identify in direct preparations [3].

Conclusion

This study demonstrates that Mimosa pudica extract is effective at reducing Blastocystis sp. burden in a model of acute diarrhea. This reduction in parasite numbers indicates the extract's ability to reduce the burden of infection significantly. The results also demonstrate the potential antiparasitic and anti-inflammatory activity of the bioactive compounds contained in Mimosa pudica. Overall, these findings support the potential of Mimosa pudica extract as an alternative therapeutic candidate for treating protozoan infections, particularly *Blastocystis* sp., with promising efficacy. However, further research is needed to evaluate its safety, mechanism of action, and effectiveness on a broader scale before it can be applied clinically.

Conflict of interest

The author conducted this research independently, without involving any other parties that could influence the final results. Every stage of this research was conducted properly and with high integrity. There are no conflicts of interest that could affect the objectivity of this article.

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