# INHIBITORY EFFECT OF THE ETHANOLIC EXTRACT OF *Bidens pilosa* L. LEAVES ON THE GROWTH OF *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Escherichia coli* AND *Saccharomyces cerevisiae*

# EFEITO INIBITÓRIO DO EXTRATO ETANÓLICO DE FOLHAS DE *Bidens pilosa L.* SOB O CRESCIMENTO DE *Staphylococcus aureus, Pseudomonas aeruginosa, Escherichia coli* E *Saccharomyces cerevisiae*

# EFECTO INHIBIDOR DEL EXTRACTO ETANÓLICO DE LAS HOJAS DE *Bidens pilosa* L. SOBRE EL CRECIMIENTO DE *Staphylococcus aureus, Pseudomonas aeruginosa*, *Escherichia coli Y Saccharomyces cerevisiae*

https://doi.org/10.56238/rcsv16n5-003

**Enviado el**: 04/09/2026 **Aprobado el**: 05/09/2026

**Bruna Oliveira de Souza[1], Anna Carolina de Oliveira Souza[2], Nayara Lança de Andrade[3], Vitor Teixeira[4], Tadeu Aparecido Martins[5], Edvânia Ferreira do Nascimento Tiezi[6], Luiz Flávio José dos Santos[7]**

## ABSTRACT

The increase in bacterial resistance to conventional antimicrobials has driven the search for new therapeutic alternatives, with natural products standing out as promising sources of bioactive compounds. This study evaluated the antimicrobial potential of the ethanolic extract of Bidens pilosa L. (picão-preto) leaves. The extract was obtained by macerating 10g of leaves in 150mL of 80% ethanol for seven days, filtered, and concentrated in a water bath at 60°C to 20% of the original volume. Antimicrobial activity was evaluated in liquid medium against Saccharomyces cerevisiae and in solid medium by the disk diffusion method against Staphylococcus aureus, Pseudomonas aeruginosa, and Escherichia coli. Results showed a 19.04% reduction in S. cerevisiae cell viability after exposure to the extract. In solid medium assays, inhibition zones were observed for all microorganisms: S. aureus (21-24 mm), P. aeruginosa (30-33 mm), and E. coli (21-23 mm). Activity against E. coli particularly relevant, considering the characteristic resistance of Gram-negative bacteria. The results confirm the broad-spectrum antimicrobial potential of Bidens pilosa extract, attributed to the presence of secondary metabolites such as flavonoids and phenolic compounds.

**Keywords:** Plant Extract Production. Antimicrobial Activity. *Bidens pilosa L.*

[1] High School and Technical Education. Centro Paula Souza. Etec Bento Carlos Botelho do Amaral. E-mail: bruna.souza531@etec.sp.gov.br Orcid: https://orcid.org/0000-0002-1669-8453 Lattes: 0461306323954134
[2] Dr. Centro Paula Souza. Etec Bento Carlos Botelho do Amaral. E-mail: anna.souza87@etec.sp.gov.br Orcid: https://orcid.org/0000-0002-8028-5418 Lattes: 3489602585225522
[3] Master's degree. Centro Paula Souza. Fatec Nilo de Stéfani. E-mail: nayara.andrade@cps.sp.gov.br Orcid: https://orcid.org/0000-0003-0343-7358 Lattes: 0742084904407782
[4] Dr. Centro Paula Souza. Etec Bento Carlos Botelho do Amaral. E-mail: vitorteixeira@ites.com.br Orcid: https://orcid.org/0000-0002-2179-2946 Lattes: 5727302355912744
[5] Specialist. Centro Paula Souza. Dr. Adail Nunes da Silva. E-mail: tadeu.martins5@etec.sp.gov.br Orcid: https://orcid.org/0009-0000-1152-0659 Lattes: 1324668420470908
[6] Master's degree. Centro Paula Souza. Etec Bento Carlos Botelho do Amaral. E-mail: edvania.nascimento2@etec.sp.gov.br Orcid: https://orcid.org/0000-0002-2182-4485 Lattes: 7137375311039875
[7] Dr. Centro Paula Souza.Fatec. E-mail: luiz.santos02@cps.sp.gov.br Orcid: https://orcid.org/0000-0002-2868-5430 Lattes: 5888302973425312

**RESUMO**
O aumento da resistência bacteriana aos antimicrobianos convencionais tem impulsionado a busca por novas alternativas terapêuticas, destacando-se os produtos naturais como fontes promissoras de compostos bioativos. Este estudo avaliou o potencial antimicrobiano do extrato etanólico das folhas de *Bidens pilosa* L. (picão-preto). O extrato foi obtido por maceração de 10g de folhas em 150mL de etanol 80% por sete dias, filtrado e concentrado em banho-maria a 60°C até 20% do volume original. A atividade antimicrobiana foi avaliada em meio líquido contra *Saccharomyces cerevisiae* e em meio sólido pelo método de difusão em disco contra *Staphylococcus aureus*, *Pseudomonas aeruginosa* e *Escherichia coli*. Os resultados demonstraram redução de 19,04% na viabilidade celular de *S. cerevisiae* após exposição ao extrato. Nos ensaios em meio sólido, observaram-se halos de inibição para todos os microrganismos: *S. aureus* (21-24 mm), *P. aeruginosa* (30-33 mm) e *E. coli* (21-23 mm). A atividade contra *E. coli* oi particularmente relevante, considerando a resistência característica de bactérias Gram-negativas. Os resultados confirmam o potencial antimicrobiano de amplo espectro do extrato de *Bidens pilosa*, atribuído à presença de metabólitos secundários como flavonoides e compostos fenólicos.

**Palavras-chave:** Produção de Extratos Vegetais. Atividade Antimicrobiana. *Bidens pilosa* L.

**RESUMEN**
El aumento de la resistencia bacteriana a los antimicrobianos convencionales ha impulsado la búsqueda de nuevas alternativas terapéuticas, destacándose los productos naturales como fuentes prometedoras de compuestos bioactivos. Este estudio evaluó el potencial antimicrobiano del extracto etanólico de las hojas de *Bidens pilosa* L. (amor seco). El extracto fue obtenido mediante maceración de 10 g de hojas en 150 mL de etanol al 80% durante siete días, filtrado y concentrado en baño maría a 60°C hasta alcanzar el 20% del volumen original. La actividad antimicrobiana fue evaluada en medio líquido contra *Saccharomyces* cerevisiae y en medio sólido mediante el método de difusión en disco contra *Staphylococcus aureus*, *Pseudomonas aeruginosa* y *Escherichia coli*. Los resultados demostraron una reducción del 19,04% en la viabilidad celular de S. cerevisiae tras la exposición al extracto. En los ensayos en medio sólido, se observaron halos de inhibición para todos los microorganismos: *S. aureus* (21–24 mm), P. aeruginosa (30–33 mm) y E. coli (21–23 mm). La actividad contra *E. coli* fue particularmente relevante, considerando la resistencia característica de las bacterias Gram negativas. Los resultados confirman el potencial antimicrobiano de amplio espectro del extracto de *Bidens pilosa*, atribuido a la presencia de metabolitos secundarios como flavonoides y compuestos fenólicos.

**Palabras clave:** Producción de Extractos Vegetales. Actividad Antimicrobiana. *Bidens pilosa L.*

## 1 INTRODUCTION

The discovery of antibiotics revolutionized modern medicine, providing effective treatment for infectious diseases that were previously considered fatal. Since the identification of penicillin by Alexander Fleming in 1928, these compounds have become fundamental not only for treating infections but also for the success of complex surgical procedures, organ transplants, and immunosuppressive therapies (Davies; Davies, 2010). However, parallel to the emergence of antibiotics, the development of bacterial resistance mechanisms was observed, a phenomenon identified as early as the 1940s, even before the therapeutic introduction of penicillin (Davies; Davies, 2010). This scenario highlighted the adaptive capacity of microorganisms and inaugurated a constant race between the development of new drugs and the evolution of microbial resistance.

The indiscriminate and inadequate use of antibiotics over the last decades has significantly accelerated the process of antimicrobial resistance. The industrial production of millions of tons of these compounds, associated with their wide application in human medicine, veterinary medicine, agriculture, and even personal hygiene products, has created unprecedented selective pressure on bacterial populations (Angelini, 2024). This scenario resulted in the selection and dissemination of multidrug-resistant microorganisms, popularly known as antimicrobial-resistant bacteria which currently represent one of the greatest threats to global public health (Davies; Davies, 2010). It is estimated that infections by resistant bacteria were directly responsible for approximately 1.27 million deaths in 2019, with alarming projections indicating the possibility of 10 million annual deaths by 2050 if effective measures are not implemented (Angelini, 2024).

Bacterial resistance to antimicrobials manifests through various biochemical and genetic mechanisms, including the production of inactivating enzymes, such as β-lactamases, alteration of antibiotic target sites, reduction of outer membrane permeability, and the expression of efflux pumps that expel drugs from the cellular interior (Davies; Davies, 2010; Angelini, 2024). Particularly concerning is the capacity for horizontal gene transfer of resistance genes between different bacterial species, mediated by plasmids, transposons, and integrons, which enables the rapid dissemination of these genetic determinants in clinically relevant microbial populations (Davies; Davies, 2010). This phenomenon has seriously compromised the therapeutic efficacy of broad-spectrum antibiotics, including β-lactams, aminoglycosides, tetracyclines, and fluoroquinolones, limiting the options available for treating common infections.

Given this concerning outlook, the search for new therapeutic alternatives has intensified, directing the scientific community's attention to natural products with antimicrobial potential (Angelini, 2024). Studies demonstrate that compounds derived from plants, fungi, lichens, and marine microorganisms constitute promising sources for the development of new antimicrobial agents (Angelini, 2024).

Thus, the goals of the project was to produce Picão-Preto (*Bidens pilosa*) extract and test its potential antimicrobial effect in different culture media and microorganisms.

## 2 THEORETICAL FOUNDATION

The study of ethnobotany can provide information regarding active substances for the development of new drugs (Souza *et al*., 2020). According to the World Health Organization (WHO), approximately 80% of the African population utilizes traditional medicine to meet their health needs, highlighting the relevance of natural products as a therapeutic alternative (Conde *et al*., 2014).

The antimicrobial activity of plants is directly related to the presence of secondary metabolites in their composition. These metabolites are natural chemical compounds produced by plants, playing an essential role in defense against various environmental and biological factors (Echeverria *et al*., 2025). They act as barriers against herbivores, protect against oxidative damage caused by free radicals and ultraviolet radiation, and combat pathogens such as bacteria, fungi, and viruses (Kaur *et al*., 2022). Among the primary classes of secondary metabolites with antimicrobial activity, phenolic compounds, alkaloids, terpenes, steroids, flavonoids, tannins, and coumarins stand out (Angelini, 2024; Echeverria *et al*., 2025).

Phenolic compounds, including flavonoids and tannins, constitute one of the most important groups of secondary metabolites with antimicrobial properties. These compounds act through multiple mechanisms, including the inhibition of efflux pumps, damage to the bacterial cell membrane, inhibition of essential enzymes, and interference with nucleic acid synthesis (Angelini, 2024). Studies demonstrate that flavonoids such as quercetin, kaempferol, and baicalein show significant inhibitory activity against Gram-positive bacteria, including methicillin-resistant *S. aureus* strains (Farhadi *et al*., 2019). Tannins, in turn, act by precipitating bacterial proteins and forming complexes with cell wall polysaccharides, compromising the integrity and functionality of the microbial cell (Simões, 2003).

Alkaloids represent another relevant class of metabolites with antimicrobial action. These compounds, which include more than 12,000 substances isolated from plants, act

primarily as efflux pump inhibitors (EPIs), representing a fundamental antibacterial mechanism (Angelini, 2024). Piperine, an alkaloid isolated from *Piper nigrum*, demonstrated the capacity to inhibit the growth of mutant *S. aureus* when combined with ciprofloxacin, significantly reducing the minimum inhibitory concentration (MIC) values. Berberine, an isoquinoline alkaloid found in Berberis species, intercalates into bacterial DNA, inhibits RNA polymerase, gyrase, and topoisomerase IV, and interferes with cell division through the inhibition of the FtsZ protein (Domadia *et al*., 2008; Boberek; Stach; Good, 2010).

*Bidens pilosa* L., popularly known as "picão-preto" or "carrapicho", is a plant belonging to the Asteraceae family, considered an invader of commercial crops, yet possessing important medicinal potential, particularly in its leaves (Lima *et al.,* 2019; Santos; Cury, 2011). Despite being frequently treated as a weed, *B. pilosa* possesses various substances that exert significant pharmacological activities, including salicylic acids, tannins, and limonenes (Haida et al., 2007).

Studies demonstrate that extracts produced from *Bidens pilosa* show sensitivity against Gram-positive bacteria, notably *Staphylococcus aureus*. However, a difference in antimicrobial action is observed for each plant organ. The active principles contained in the leaf epidermis proved to be more efficient for inhibiting the growth of Gram-positive bacteria than those present in the stem (Kwieciński *et al*., 2008; Santos; Cury, 2011). This variation in antimicrobial activity between different parts of the same plant species reflects the differentiated distribution of secondary metabolites in various tissues, which can be influenced by factors such as phenological stage, environmental conditions, and extraction methods (Echeverria *et al*., 2025).

Research conducted by Lima *et al*. (2019) evaluated the antimicrobial activity of extracts from different organs of *Bidens pilosa* (root, stem, and leaf) against *Escherichia coli* and *Staphylococcus aureus*. The results demonstrated that the stem and leaf extracts were effective against *S. aureus*, while the bacterium *E. coli* proved resistant to all tested extracts. This pattern of differentiated susceptibility between Gram-positive and Gram-negative bacteria is consistent with the literature, being attributed to the greater complexity of the Gram-negative bacterial cell wall, which features an outer membrane rich in lipopolysaccharides (LPS), providing greater protection against antimicrobial agents (Gladwin; Trattler, 2004; Nogueira; Miguel, 2009).

The efficacy of the antimicrobial activity of plant extracts is influenced by multiple factors, including the part of the plant used, the developmental stage, environmental cultivation conditions, and the extraction methods employed (Echeverria *et al*., 2025).

Comparative studies demonstrate that all these factors directly influence the chemical composition of the extracts and, consequently, their biological and pharmacological properties.

The choice of extraction solvent is particularly critical, as it determines which classes of compounds will be extracted from the plant matrix. Solvents of different polarities show differentiated affinity for various secondary metabolites. Ethanol, for example, is effective in extracting phenolic compounds and flavonoids, while non-polar solvents like hexane and chloroform are more suitable for extracting terpenes and essential oils (Abubakar; Haque, 2020; Echeverria *et al*., 2025).

The phenological stage of the plant at the time of collection also significantly influences the composition and concentration of secondary metabolites. Ramana and Bhaskar (2015) observed that flower extracts of *Couroupita guianensis* collected during the flowering period were more effective compared to extracts from flowers collected after full flowering, confirming that variations in compounds based on the plant's developmental stage can lead to different microbial inhibition results (Abreu *et al*., 2025).

Environmental conditions, including water availability, soil nutrients, temperature, and luminosity, also modulate the production of secondary metabolites in plants. These compounds are synthesized in response to environmental stresses, acting as defense and adaptation mechanisms (Taiz; Zeiger, 2004). Consequently, plants of the same species cultivated in different regions may present distinct phytochemical profiles and, by extension, variable biological activities (Abreu *et al*., 2025).

The evaluation of the antimicrobial activity of plant extracts can be performed through different methodologies, the most common being the disk (or well) diffusion method and broth microdilution for determining the minimum inhibitory concentration (MIC) (Angelini, 2024; Echeverria *et al*., 2025).

The disk diffusion method, also known as the antimicrobial susceptibility test (AST), consists of applying filter paper disks impregnated with the extract onto the surface of agar previously inoculated with the test microorganism. After incubation, the formation of inhibition zones around the disks indicates the sensitivity of the microorganism to the evaluated extract (Lima *et al*., 2019; Maciel *et al*., 2024). This method is widely used due to its simplicity and low cost, allowing for the initial screening of multiple extracts simultaneously.

Broth microdilution is considered the reference method for quantitative determination of antimicrobial activity, allowing for the establishment of the minimum inhibitory concentration

(MIC) and the minimum bactericidal or fungicidal concentration (MBC/MFC) (Echeverria *et al*., 2025).

## 3 MATERIALS AND METHODS

### 3.1 PREPARATION OF PLANT EXTRACTS

To produce the *B. pilosa* extract in alcoholic solution, leaves were used in a proportion of 10g of plant material to 150mL of 80% ethanol; the leaves were macerated and then stored in sealed glass flasks at room temperature for seven days (adapted from Lima *et al*., 2019). The resulting material was filtered using qualitative filter paper and heated in a water bath at 60°C until reduced to 20% of its volume, then stored in a sterile container at 8°C for up to one week.

### 3.2 ANTIMICROBIAL ACTIVITY TEST IN LIQUID MEDIUM

To evaluate the effect of the *Bidens pilosa* extract on eukaryotic microorganisms, a liquid medium assay was conducted using *Saccharomyces cerevisiae* as the test organism (Teixeira *et al*., 2019). Initially, 1 mL of a previously activated liquid yeast culture was inoculated into tubes containing 4 mL of sterilized nutrient broth. In three of these tubes, 0.1 mL of the *B. pilosa* ethanolic extract was added, while a fourth tube, without the extract, was kept as the negative control for the experiment. All tubes were properly identified and incubated at 32°C for 24 hours.

After the incubation period, 0.1 mL aliquots from each tube were transferred to 9.9 mL of sterilized saline solution. For the determination of cell viability, 0.5 mL of each dilution was mixed with 4.5 mL of Ringer's Blue vital dye. The samples were homogenized and allowed to rest for 10 minutes to permit the uptake of the dye by non-viable cells.

Cell counting was performed in a Neubauer chamber with the aid of an optical microscope, differentiating between live and dead cells. Cell viability was calculated by the ratio between the number of live cells and the total number of cells, expressed as a percentage.

### 3.3 ANTIMICROBIAL ACTIVITY TEST IN SOLID MEDIUM WITH DIFFUSION DISKS

The evaluation of the antimicrobial activity of the *Bidens pilosa* extract on bacteria was performed by the disk diffusion method, following an adaptation of the standardized technique for antibiograms. Three bacterial species of clinical interest were used: *Staphylococcus aureus*, *Pseudomonas aeruginosa*, and *Escherichia coli*, as well as a mixed sample of

microorganisms collected from the school environment. Initially, Petri dishes containing nutrient agar were properly identified for each experimental condition.

For the preparation of the inoculum, 0.1 to 0.2 mL aliquots of each previously activated liquid bacterial culture were transferred to the surface of the nutrient agar and spread uniformly. The plates were kept at rest until the inoculum was completely absorbed by the culture medium. Subsequently, sterile filter paper disks (6 mm in diameter) were impregnated with 20 µL of the *B. pilosa* ethanolic extract and applied onto the agar surface using sterilized forceps, applying light pressure to ensure proper contact.

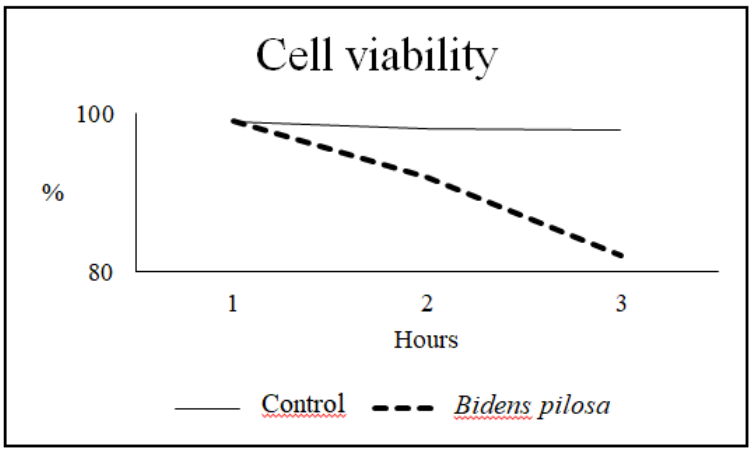
The plates were incubated at 32°C for 24 hours. After the incubation period, antimicrobial activity was evaluated by measuring the inhibition zones of bacterial growth around each disk using a caliper. The diameters of the zones were recorded in millimeters, with the presence of any clear and visible zone of microbial growth inhibition considered a positive result. The assays were performed in triplicate for each microorganism tested.

## 4 RESULTS AND DISCUSSION

The results obtained in this study demonstrate that the *Bidens pilosa* extract exhibited antimicrobial activity in both liquid and solid media. In the liquid medium assay, a progressive reduction in the cell viability (CV) of *S. cerevisiae* was observed (Figure 1), reaching a decrease of 19.04% after 3 hours of exposure. This result suggests that the bioactive compounds present in "picão-preto" exert a direct effect on yeasts, corroborating previous findings that the species possesses a broad antimicrobial spectrum (Rojas et al., 2006; Souza et al., 2010). According to Singh, Saini, and Prakash (2025), the diversity of secondary metabolites such as flavonoids, tannins, and polyacetylenes provides various plant extracts with antimicrobial properties, possibly related to the destabilization of the cell membrane and inhibition of essential metabolic processes.

**Figure 1**

*Cell viability of S. cerevisiae under the effect of B. pilosa*

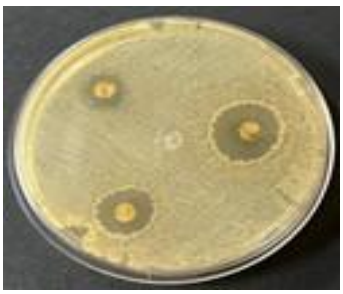



Source: The author (2025).

In the solid medium assays, the *B. pilosa* extract showed significant inhibition against the cultivation of microorganisms collected from the school environment (Figure 2, inhibition zones measuring 13, 18, and 23 mm), as well as *Staphylococcus aureus* (Figure 3, inhibition zones measuring 21, 22, and 24 mm), *Pseudomonas aeruginosa* (Figure 4, inhibition zones measuring 30, 31, and 33 mm), and *Escherichia coli* (Figure 5, inhibition zones measuring 21, 23, and 23 mm), confirming the efficacy of the "picão-preto" extract against both Gram-positive and Gram-negative bacteria.

**Figure 2**

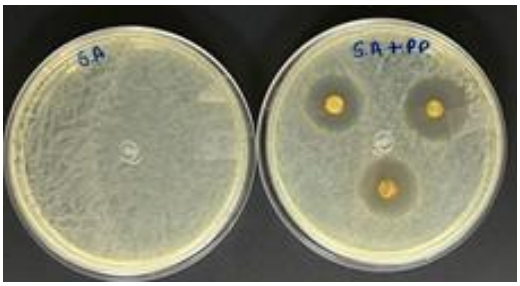
*Inhibitory effect of B. pilosa extract on microorganisms collected in a school environment.*



Source: The author (2025).

**Figure 3**

*Inhibitory effect of B. pilosa extract on S. aureus culture – control culture on the right.*




Source: The author (2025).

**Figure 4**

*Inhibitory effect of B. pilosa extract on P. aeruginosa culture – control culture on the right.*


Source: The author (2025).

**Figure 5**

*Inhibitory effect of B. pilosa extract on E. coli culture – control culture on the right.*


Source: The author (2025).

The inhibition of *S. aureus* observed in our study is particularly relevant, given that this pathogen is associated with hospital-acquired infections and shows increasing resistance to conventional antibiotics (Elfadadny *et al*., 2024). In this sense, the use of plant extracts such as "picão-preto" may represent a promising alternative for controlling resistant strains.

The results obtained in the assays with *Pseudomonas aeruginosa* revealed particularly expressive inhibition zones, ranging from 30 to 33 mm, indicating a notable sensitivity of this

Gram-negative bacterium to the *Bidens pilosa* extract. This finding is especially significant since *P. aeruginosa* is recognized as a difficult-to-control opportunistic pathogen, frequently exhibiting resistance to multiple classes of antibiotics, including β-lactams and aminoglycosides (Davies; Davies, 2010). Recent literature confirms the relentless search for new alternatives for treating infections caused by this bacterium, highlighting the potential of plant-derived compounds (Elfadadny et al., 2024). The efficacy of the "picão-preto" extract against *P. aeruginosa* can be attributed to the presence of metabolites such as flavonoids and terpenoids, which have the capacity to damage the bacterial cell membrane, inhibit efflux pumps, and interfere with biofilm formation, mechanisms essential for the pathogenicity and resistance of this species (Angelini, 2024).

The consistent inhibition of *Escherichia coli*, with zones of 21 to 23 mm, reinforces the broad-spectrum antimicrobial potential of the *B. pilosa* extract, especially against Gram-negative bacteria. As discussed by Lima *et al*. (2019), *E. coli* resistance to plant extracts is frequently observed due to the complexity of its cell wall, which includes an outer membrane rich in lipopolysaccharides (LPS) that acts as an effective barrier to various antimicrobial agents (Gladwin; Trattler, 2004; Nogueira; Miguel, 2009). Overcoming this barrier in our study suggests that the extraction methodology employed and the subsequent concentration of bioactive compounds were effective in reaching the cellular target. Abreu *et al*. (2025), in their review on *Couroupita guianensis*, highlight that antimicrobial activity against *E. coli* can vary significantly according to the plant organ and the solvent used; the ethanolic extract of the leaves often presents promising results, which corroborates the efficacy observed with the *B. pilosa* extract.

The divergence between our results for *E. coli* and those reported by Lima *et al*. (2019), who did not observe inhibition for this bacterium with "picão-preto" extracts, reinforces the importance of methodological factors in evaluating antimicrobial potential. Comparative studies demonstrate that the part of the plant used, the developmental stage, environmental conditions, and, crucially, the extraction method directly influence the chemical composition of the extract and, consequently, its biological properties (Echeverria *et al*., 2025; Abubakar; Haque, 2020). The concentration of the extract in a water bath, reducing it to 20% of the original volume as described in the methodology, may have acted as a concentration factor for the active principles, explaining the activity observed against *E. coli* in our study, in contrast to the work of Lima *et al*. (2019), which used extracts without this concentration step.

In addition to the solvent, cultivation conditions and the phenological state of the plant at the time of collection are determining factors for phytochemical variability. Ramana and

Bhaskar (2015) observed that flower extracts of *Couroupita guianensis* collected during flowering were more effective than those from post-flowering flowers, evidencing that the developmental stage can alter the concentration of active metabolites (Abreu *et al*., 2025). The moderate but consistent antimicrobial activity observed against *S. aureus*, *P. aeruginosa*, and *E. coli* described here, compared to variable results in the literature (Mistura *et al*., 2019; Lima *et al*., 2019), suggests that the plant material used possessed a robust phytochemical profile. The presence of compounds such as tannins and flavonoids, known for their action on multiple bacterial targets, such as the inhibition of efflux pumps (Angelini, 2024) and complexation with cell wall proteins (Simões, 2003); probably contributed to the observed efficacy, demonstrating the potential of "picão-preto" as a valuable source of natural antimicrobial agents.

In a similar study, Mistura *et al*. (2020) found that different parts of the plant (roots, stems, and leaves) extracted in varying solvents show distinct inhibition patterns against *S. aureus* and *E. coli*, emphasizing the influence of the solvent type and the plant organ used. This variation in results between studies reinforces the need for methodological standardization for comparability between different plant extracts, as already highlighted by Alara *et al*. (2019).

The results obtained in this study also align with observations by Abalaka *et al*. (2012), who demonstrated the antimicrobial potential of *Moringa oleifera* against enteric bacteria and its use in the decontamination of food and beverages. These findings suggest that plant extracts are not limited to pharmaceutical applications but can also be employed in sanitization and preservation processes, strengthening their applicability across different sectors.

## 5 FINAL CONSIDERATIONS

Thus, the results presented herein confirm the potential of the *Bidens pilosa* L. extract as an antimicrobial agent in both culture media and for the tested microorganisms; however, additional studies are necessary for the detailed chemical characterization of the active compounds, as well as for the development of safe and effective large-scale formulations.

## ACKNOWLEDGEMENTS

To the National Council for Scientific and Technological Development – CNPq – Brazil, for granting the Scientific Initiation scholarship (PIBIC).

## REFERENCES

Abreu, A. D. S., et al. (2025). Atividade antimicrobiana de extratos de Couroupita guianensis Aubl.: Uma revisão bibliográfica. Paubrasilia, 8, e0171. https://doi.org/10.33447/paubrasilia.2025.e0171

Abalaka, M. E., et al. (2012). The antibacterial evaluation of Moringa oleifera leaf extracts on selected bacterial pathogens. Journal of Microbiology Research, 2(2), 1–4.

Abubakar, A. R., et al. (2020). Preparation of medicinal plants: Basic extraction and fractionation procedures for experimental purposes. Journal of Pharmacy and Bioallied Sciences, 12(1), 1.

Alara, O. R., et al. (2019). Extraction and characterization of bioactive compounds in Vernonia amygdalina leaf ethanolic extract comparing Soxhlet and microwave assisted extraction techniques. Journal of Taibah University for Science, 13(1), 414–422.

Alcarde, A. R. (2014). Cachaça: Ciência, tecnologia e arte. Editora Blucher.

Amorim, H. V. (2005). Fermentação alcoólica: Ciência e tecnologia. Fermentec.

Angelini, P. (2024). Plant-derived antimicrobials and their crucial role in combating antimicrobial resistance. Antibiotics, 13(8), 746. https://doi.org/10.3390/antibiotics13080746

Boberek, J. M., et al. (2010). Genetic evidence for inhibition of bacterial division protein FtsZ by berberine. PLoS ONE, 5(11), e13745.

Brito, F. E. S., et al. (2021). Análise microbiológica da qualidade da água do povoado Barra Nova, Cocal de Telha – Piauí, Brasil. RAPA, 6(3), 174–182.

Conde, P., et al. (2014). A Missão Botânica de Moçambique (1942–1948): Contribuições para o conhecimento da flora medicinal de Moçambique. História, Ciências, Saúde-Manguinhos.

Cordeiro, M. M. C. A. (2011). Caracterização molecular de cepas de Staphylococcus aureus isolados no Hospital Municipal de Ipatinga/MG [Dissertação/Tese, Universidade Federal de Ouro Preto].

Davies, J., et al. (2010). Origins and evolution of antibiotic resistance. Microbiology and Molecular Biology Reviews, 74(3), 417–433. https://doi.org/10.1128/MMBR.00016-10

Domadia, P. N., et al. (2008). Berberine targets assembly of Escherichia coli cell division protein FtsZ. Biochemistry, 47(10), 3225–3234.

Echeverria, J. S., et al. (2025). Atividade antimicrobiana dos extratos etanólicos de especiarias brasileiras. Revista Eletrônica Acervo Saúde, 25, e18574. https://doi.org/10.25248/REAS.e18574.2025

Elfadadny, A., et al. (2024). Antimicrobial resistance of Pseudomonas aeruginosa: Navigating clinical impacts, current resistance trends, and innovations in breaking therapies. Frontiers in Microbiology, 15, 1374466.

Farhadi, F., et al. (2019). Antibacterial activity of flavonoids and their structure–activity relationship: An update review. Phytotherapy Research, 33(1), 13–40.

Gladwin, M., et al. (2004). Clinical microbiology (4th ed., pp. 12–18).

Haida, K. S., et al. (2007). Avaliação in vitro da atividade antimicrobiana de oito espécies de plantas medicinais. Arquivos de Ciências da Saúde da UNIPAR, 11(3), 185–192.

Kaur, S., et al. (2022). How do plants defend themselves against pathogens—Biochemical mechanisms and genetic interventions. Physiology and Molecular Biology of Plants, 28(2), 485–504.

Kwieciński, M. R., et al. (2008). Study of the antitumor potential of Bidens pilosa (Asteraceae) used in Brazilian folk medicine. Journal of Ethnopharmacology, 117(1), 69–75.

Lima, B. O., et al. (2019). Atividade antimicrobiana de Bidens pilosa (picão-preto) e do própolis da região de Divinolândia. In 20 Congresso Nacional de Iniciação Científica.

Lopes, A. C., et al. (2006). Diagnóstico e tratamento. Manole.

Macedo, E. R., et al. (2018). Epidemiological profile of acute diarrhea diseases notified in the municipal hospital of Una-BA in the period from 2013 to 2014. REES, 7(2), 25–30.

Maciel, C. K., et al. (2024). Antimicrobianos naturais: Potencial bacteriostático do extrato de orégano. Brazilian Journal of Implantology and Health Sciences, 6(10), 3014–3027. https://doi.org/10.36557/2674-8169.2024v6n10p3014-3027

Martínez-Orgániz, A., et al. (2020). Patotipos y resistencia a antibióticos de Escherichia coli en agua residual. Revista Internacional de Contaminación Ambiental, 36(4), 957–966.

Mickymaray, S., et al. (2018). Antibacterial efficacy of bacteriocin produced by marine Bacillus subtilis against clinically important extended-spectrum betalactamase strains and methicillin-resistant Staphylococcus aureus. International Journal of Medical Research & Health Sciences, 7(2), 75–83.

Mistura, I. D., et al. (2020). Atividade antimicrobiana de Bidens pilosa (picão-preto) e do própolis da região de Divinolândia. In Anais do 20º Congresso Nacional de Iniciação Científica (CONIC-SEMESP). SEMESP.

Nandhini, P., et al. (2022). Recent developments in methicillin-resistant Staphylococcus aureus (MRSA) treatment: A review. Antibiotics, 11(5), 606–626.

Nogueira, J. M. R., et al. (2009). Bacteriologia. In E. M. Molinaro, L. F. G. Caputo, & M. R. R. Amendoeira (Orgs.), Conceitos e métodos para a formação de profissionais em laboratórios de saúde (4th ed., pp. 221–397). EPSJV/IOC.

Ramana, C. V., et al. (2015). Screening of antimicrobial activity of flower extracts on human bacterial pathogens. Journal of Pharmacognosy and Phytochemistry, 3(6), 153–156.

Rodrigues, J. A., et al. (2024). Potencial antimicrobiano de extratos vegetais com foco na odontologia. RECIMA21 - Revista Científica Multidisciplinar, 5(11), e5115843. https://doi.org/10.47820/recima21.v5i11.5843

Rojas, J. J., et al. (2006). Actividad antibacteriana de extractos de Bidens pilosa L. frente a bacterias Gram-positivas y Gram-negativas. Revista de Biología Tropical, 54(1), 1–10.

Santos, J. B., et al. (2011). Picão-preto: Uma planta daninha especial em solos tropicais. Planta Daninha, 29(1).

Silva, J. F. M., et al. (2017). Staphylococcus aureus em alimentos. Desafios, 4(4), 15–31.

Simões, C. M. O. (2003). Farmacognosia: Da planta ao medicamento. Editora da UFSC; Editora da UFRGS.

Singh, K. K., et al. (2025). Screening of natural plant extracts for antimicrobial activity against Streptobacillus moniliformis. bioRxiv. https://doi.org/10.1101/2025.04.05.647341

Soni, J., et al. (2024). Understanding bacterial pathogenicity: A closer look at the journey of harmful microbes. Frontiers in Microbiology, 15, 1370818.

Souza, S., et al. (2010). Atividade antimicrobiana de extratos de Bidens pilosa L. sobre microrganismos multirresistentes. Revista Brasileira de Farmacognosia, 20(1), 32–37.

Souza, G. A., et al. (2020). Novas isoflavonas isoladas das folhas de Vatairea guianensis Aubl. e atividade antiproliferativa. Scientia Plena, 16(11).

Taiz, L., et al. (2004). Fisiologia vegetal. Artmed.

Teixeira, V., et al. (2019). Using Moringa oleifera Lamarck seed extract for controlling microbial contamination when producing organic cachaça. International Journal of Food Microbiology, 308, 108287. https://doi.org/10.1016/j.ijfoodmicro.2019.108287

Torres, A. R., et al. (2018). Atividade antimicrobiana de extratos de própolis e plantas medicinais. Brazilian Journal of Development, 4(4), 1234–1250.