

Proceeding Paper

Evaluation of the Antimicrobial Potential of Extracts from Plants of the Family Asteraceae [†]

Marta Barral-Martinez ¹, Lucia Cassani ^{1,2}, Maria Carpena ¹, Antia G. Pereira ^{1,2},
Paula Garcia-Oliveira ^{1,2}, Franklin Chamorro ¹, Sepidar Seyyedi Mansour ¹, Aurora Silva ^{1,3},
Fatima Barroso ³, Hui Cao ¹, Miguel A. Prieto ^{1,2,*} and Jesus Simal-Gandara ^{1,*}

- ¹ Nutrition and Bromatology Group, Department of Analytical and Food Chemistry, Faculty of Food Science and Technology, Ourense Campus, University of Vigo, E32004 Ourense, Spain; marta.barral@uvigo.es (M.B.-M.); lucivictoria.cassani@uvigo.es (L.C.); mcarpena@uvigo.es (M.C.); antia.gonzalez.pereira@uvigo.es (A.G.P.); paula.garcia.oliveira@uvigo.es (P.G.-O.); franklin.noel.chamorro@uvigo.es (F.C.); sepidar.seyyedi@uvigo.es (S.S.M.); mass@isep.ipp.pt (A.S.); hui.cao@uvigo.es (H.C.)
- ² Centro de Investigação de Montanha (CIMO), Instituto Politécnico de Bragança, Campus de Santa Apolonia, 5300-253 Bragança, Portugal
- ³ Associated Laboratory for Green Chemistry (LAQV) of the Network of Chemistry and Technology (REQUIMTE), Department of Chemistry and Biochemistry, Faculty of Sciences, University of Porto, Rua do Campo Alegre s/n, 4169-007 Porto, Portugal; mfb@isep.ipp.pt
- * Correspondence: mprieto@uvigo.es (M.A.P.); jsimal@uvigo.es (J.S.-G.)
- [†] Presented at the 2nd International Electronic Conference on Antibiotics—Drugs for Superbugs: Antibiotic Discovery, Modes of Action and Mechanisms of Resistance, 15–30 June 2022; Available online: <https://eca2022.sciforum.net/>.



Citation: Barral-Martinez, M.; Cassani, L.; Carpena, M.; G. Pereira, A.; Garcia-Oliveira, P.; Chamorro, F.; Seyyedi Mansour, S.; Silva, A.; Barroso, F.; Cao, H.; et al. Evaluation of the Antimicrobial Potential of Extracts from Plants of the Family Asteraceae. *Med. Sci. Forum* **2022**, *12*, 17. <https://doi.org/10.3390/eca2022-12726>

Academic Editor: Marc Maresca

Published: 15 June 2022

Publisher's Note: MDPI stays neutral with regard to jurisdictional claims in published maps and institutional affiliations.



Copyright: © 2022 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (<https://creativecommons.org/licenses/by/4.0/>).

Abstract: Today, medicinal plants have multiple different uses since they can play an important role in the design of modern medicines and new healthy foods. Although their level of consumption is not high, these plants are attributed pharmacological properties with beneficial properties, such as antioxidant, anti-inflammatory, or antimicrobial, among others. This study aimed to evaluate extracts derived from *Chamaemelum nobile* and *Arnica montana*, two plants from the Asteraceae family that could be of interest for the industry, in terms of their antimicrobial capacity. After a previous bibliographic study, heat-assisted extraction (HAE) was selected and used to obtain extracts rich in bioactive compounds. Then, the antimicrobial activity was determined using three Gram-negative species, *Pseudomonas aeruginosa* (ATCC 10145), *Escherichia coli* (NCTC 9001), and *Salmonella enteritidis* (ATCC 13676), and two Gram-positive species, *Bacillus cereus* (ATCC 25923) and *Staphylococcus aureus* (ATCC 25923). The results obtained showed that *A. montana* showed a moderate antimicrobial activity with inhibition zones ranging from 6.82 to 8.36 mm while the *C. nobile* showed inhibition zones of 7.55 and 7.91 mm against *S. enteritidis* and *P. aeruginosa*, respectively. None of the plants showed activity against *S. aureus*. These results provide scientific evidence for the use of medicinal plant extracts for the development of new products with antimicrobial properties.

Keywords: medicinal plants; Asteraceae; antimicrobial activity

1. Introduction

Since ancient times, medicinal plants have been used as a traditional treatment for various diseases or ailments. Several scientific studies have shown that these plants have different beneficial properties, such as antioxidant, anti-inflammatory, or antimicrobial among others [1]. In addition, they have been recognized as interesting matrices for the recovery of bioactive compounds, specific molecules that promote beneficial properties for health. Therefore, the content of these compounds in medicinal plants has been related to their possible health effects [2]. Northwest Spain is particularly rich in botanical and folk medicine traditions. Among the known groups, Asteraceae species (plants belonging

to the family Asteraceae) are promising candidates for their bioactive compounds and associated beneficial properties. This family of cosmopolitan distribution and easy adaptability includes 8–10% of all known angiosperms and represents 24,000–25,000 species and 1600–1700 genera [3]. In particular, two plants belonging to this family called *Chamaemelum nobile* and *Arnica montana* are the subject of this study. *Chamaemelum nobile*, commonly called Roman chamomile, is a medicinal plant that has been traditionally used to relieve fever and sunstroke, insomnia, back pain, neuralgia, rheumatism, skin conditions, or headache [4]. In addition, beneficial properties has been ascribed to these plants, such as digestive health [5], antioxidant, antibacterial, and antifungal activities [4]. Parts of the *A. montana* plant, such as flowers and root extracts, have been used in ointments for the relief of various injuries, including bruises, sprains, rheumatic pains, or inflammation. Their anticancer, antimicrobial, hepatoprotective, or antitumor bioactivities have not been still studied in great depth [6]. For this reason, the main objective of this study was to evaluate the antimicrobial capacity of *C. nobile* and *A. montana* extracts.

2. Materials and Methods

2.1. Preparation of Plants

Dry *C. nobile* and *A. montana*, were crushed until pore size was below 2 mm to obtain a fine and homogeneous powder. The parts used from each plant were the leaves, stems, and flowers.

2.2. Heat-Assisted Extraction (HAE)

HAE was applied to obtain extracts rich in bioactive compounds. To this aim, 5 g of sample was placed in a glass bottle with 100 mL of methanol–water mixture (60:40 *v/v*) and a magnetic stirrer. The extraction was carried out in a thermostatic bath at 45 °C for 1 h. Extracts were then freeze-dried using Telstar LyoAlfa 15, sourced in Terrassa, Spain, equipment to obtain dry extracts that were used in subsequent analyses.

2.3. Determination of Antimicrobial Activity

The antimicrobial activity was determined through plate diffusion test by measuring the inhibition zones produced by the addition of extracts.

Three Gram-negative (Gram-) species were used for this assay, *Pseudomonas aeruginosa* (ATCC 10145), *Escherichia coli* (NCTC 9001), and *Salmonella enteritidis* (ATCC 13676), and two Gram-positive species, *Bacillus cereus* (ATCC 25923) and *Staphylococcus aureus* (ATCC 25923). These bacteria were selected since they are some of the most common microorganisms causing food poisoning [7].

First, the stock cultures were inoculated into 10 mL of Mueller–Hinton Broth (MHB) and grown from 12 to 24 h at 37 °C. The evaluation of the number of colonies was performed by UV spectrometry at 600 nm and was set between 1 and 2×10^8 colony forming units (CFUs). Of the previously cultured inoculum, 100 μ L were seeded and spread on a Petri dish containing Muller–Hinton agar, which was divided into four quadrants. Then, 15 μ L of dimethyl sulfoxide (DMSO) was added as negative control, 15 μ L of 40% lactic acid as positive control, and 15 μ L of extracts (20 mg/mL in DMSO) in each of the remaining quadrants, as can be observed in Figure 1 Petri dishes were incubated for 24 h at 37 °C. After that, inhibition zones were determined [8]. Determinations were performed in triplicate and the inhibition halos were measured with a pachymeter in millimeters.

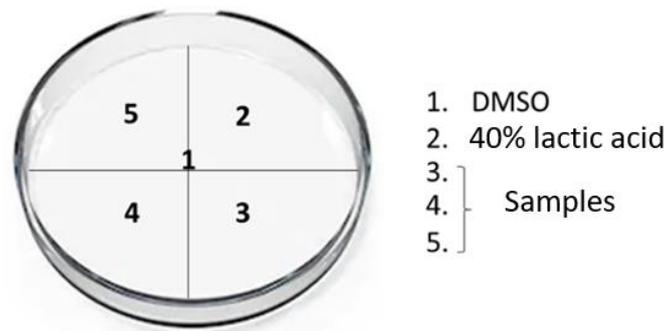


Figure 1. Distribution of different solutions and extracts in a Petri dish containing Muller–Hinton agar.

3. Results and Discussion

Table 1 shows results regarding the antimicrobial activity of the two studied plants. For this trial, the most common food-related microorganisms were selected to consider the possible use of these two plant extracts as food additives. Especially the microorganisms *S. aureus* and *S. epidermidis* are considered to cause opportunistic infections [7].

Table 1. Antimicrobial activity of *A. montana* and *C. nobile*. Bacterial growth inhibition zones diameter expressed in mm.

Plants	Gram-Negative Species			Gram-Positive Species	
	<i>P. aeruginosa</i>	<i>E. coli</i>	<i>S. enteritidis</i>	<i>S. aureus</i>	<i>B. cereus</i>
<i>A. montana</i>	6.82 ± 0.66	7.5 ± 1.47	8.36 ± 1.03	Ni*	7.73 ± 0.470
<i>C. nobile</i>	7.55 ± 0.54	Ni*	7.91 ± 0.83	Ni*	7.73 ± 0.46

P. aeruginosa; *Pseudomonas aeruginosa*; *E. coli*: *Escherichia coli*; *S. enteritidis*: *Salmonella enteritidis*; *S. aureus*: *Salmonella enteritidis* and *B. cereus*: *Bacillus cereus*. Ni*: no inhibition.

From Table 1, it can be observed that in both plant extracts, inhibitory effects were produced against at least one of the bacterial strains. *A. montana* showed a moderate range of antimicrobial activity (except for *S. aureus*), with inhibition halos ranging from 6.82 to 8.36 mm. Previous studies have evaluated the antimicrobial activity of selected plants. In the case of *C. nobile*, extracts rich in phenolic compounds showed inhibitory activity against *S. aureus*, *Bacillus* sp., *P. aeruginosa*, and *E. coli*, with inhibition zones ranging from 10 to 12.66 mm [9,10]. In the present study, no inhibition of *S. aureus* and *E. coli* species by *C. nobile* extracts was observed. In the case of *A. montana*, to our knowledge, no studies evaluating its antimicrobial properties have been carried out. Of the selected bacteria, *S. enteritidis* was the most affected by both plants, showing the highest ranges of inhibition (8.36 mm with *A. montana* and 7.91 mm with *C. nobile*). Figure 2 shows, as an example, the antimicrobial activity of *A. montana* and *C. nobile* against *E. coli* and *B. cereus*, respectively.

In general, differences observed in antimicrobial activity between studies may be attributed to the phytochemical composition of the studied plants, which differ according to the part used, geographic location, season, climatic conditions, etc.

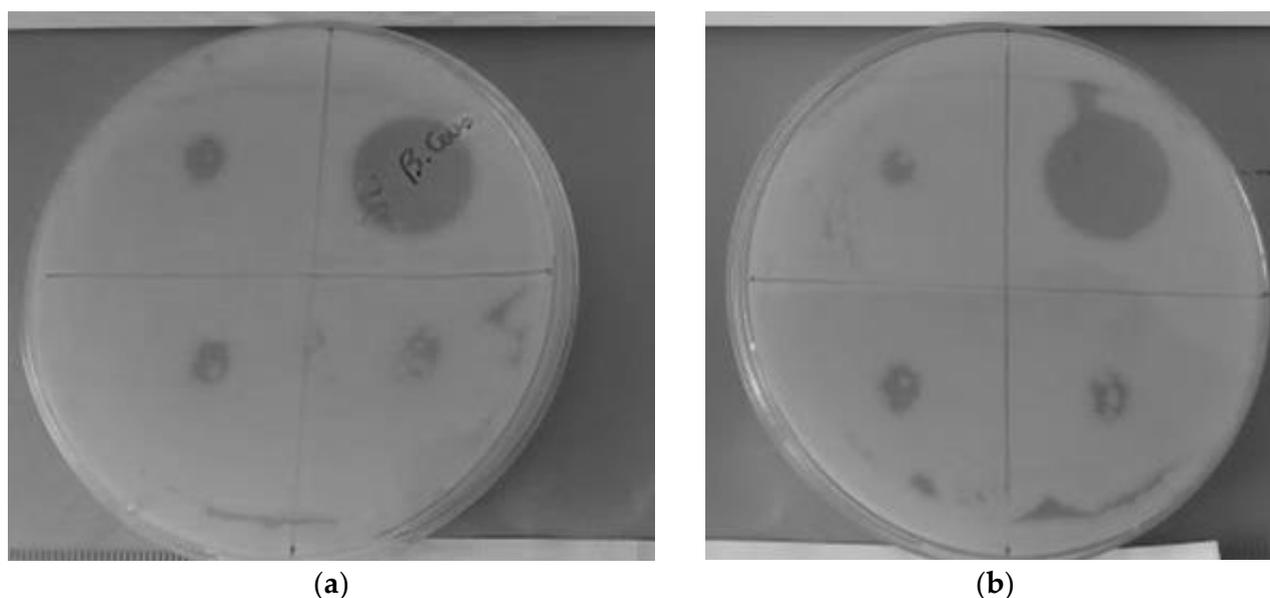


Figure 2. Antimicrobial activity by *C. nobile* against *B. cereus* (a) and by *A. montana* against *E. coli* (b) using plate diffusion method.

4. Conclusions

In recent years, there has been a growing interest in discovering new bioactivities from the phytochemical compounds of plants used in traditional medicine with possible industrial applications. At present, *C. nobile* and *A. montana* are used in herbal products. Both plants showed moderate antimicrobial activity against selected bacteria strains, providing scientific evidence for the potential of medicinal plant extracts in the development of new products with antimicrobial properties.

Author Contributions: Conceptualization, M.B.-M., L.C., P.G.-O., M.C., A.G.P., A.S., H.C., J.S.-G. and M.A.P.; methodology, M.B.-M., P.G.-O., A.G.P., A.S., F.C. and F.B.; software, S.S.M., M.C., A.S. and F.B.; validation, F.B., L.C., J.S.-G. and M.A.P.; formal analysis, A.S., P.G.-O., M.C. and L.C.; investigation, M.B.-M., P.G.-O., A.S. and F.B.; writing—original draft preparation, M.B.-M. and L.C.; writing—review and editing, M.B.-M., M.C. and L.C.; visualization, L.C., P.G.-O., M.C., A.G.P. and F.B.; supervision, L.C., A.S., F.B., J.S.-G. and M.A.P. All authors have read and agreed to the published version of the manuscript.

Funding: Authors are grateful to Ibero-American Program on Science and Technology (CYTED—AQUA-CIBUS, P317RT0003), to the Bio Based Industries Joint Undertaking (JU) under grant agreement No 888003 UP4HEALTH Project (H2020-BBI-JTI-2019). The JU receives support from the European Union’s Horizon 2020 research and innovation program and the Bio Based Industries Consortium. The project SYSTEMIC Knowledge hub on Nutrition and Food Security, has received funding from national research funding parties in Belgium (FWO), France (INRA), Germany (BLE), Italy (MIPAAF), Latvia (IZM), Norway (RCN), Portugal (FCT), and Spain (AEI) in a joint action of JPI HDHL, JPI-OCEANS and FACCE-JPI launched in 2019 under the ERA-NET ERA-HDHL (n° 696295).

Institutional Review Board Statement: Not applicable.

Informed Consent Statement: Not applicable.

Data Availability Statement: All data included in the Manuscript are attached here.

Acknowledgments: The research leading to these results was supported by MICINN supporting the Ramón y Cajal grant for M.A. Prieto (RYC-2017-22891), the Juan de la Cierva Incorporación for Hui Cao (IJC2020-046055-I), by Xunta de Galicia for supporting the program EXCELENCIA-ED431F 2020/12, the post-doctoral grant of L. Cassani (ED481B-2021/152), and the pre-doctoral grants of P. Garcia-Oliveira (ED481A-2019/295), A. G. Pereira (ED481A-2019/0228), and M. Carpena (ED481A 2021/313). The authors thank the program BENEFICIOS DO CONSUMO DAS ESPECIES

TINTORERA-(CO-0019-2021) that supports the work of F. Chamorro. Authors are grateful to Ibero-American Program on Science and Technology (CYTED—AQUA-CIBUS, P317RT0003), to the Bio Based Industries Joint Undertaking (JU) under grant agreement No 888003 UP4HEALTH Project (H2020-BBI-JTI-2019) that supports the work of M. Barral-Martínez.

Conflicts of Interest: The authors declare no conflict of interest.

References

1. Garcia-Oliveira, P.; Fraga-Corral, M.; Pereira, A.G.; Lourenço-Lopes, C.; Jimenez-Lopez, C.; Prieto, M.A.; Simal-Gandara, J. Scientific basis for the industrialization of traditionally used plants of the Rosaceae family. *Food Chem.* **2020**, *330*, 127197. [[CrossRef](#)] [[PubMed](#)]
2. Wang, W.; Xu, J.; Fang, H.; Li, Z.; Li, M. Advances and challenges in medicinal plant breeding. *Plant Sci.* **2020**, *298*, 110573. [[CrossRef](#)] [[PubMed](#)]
3. Nikolić, M.; Stevović, S. Family *Asteraceae* as a sustainable planning tool in phytoremediation and its relevance in urban areas. *Urban For. Urban Green.* **2015**, *14*, 782–789. [[CrossRef](#)]
4. Sharifzadeh, A.; Jebeli Javan, A.; Shokri, H.; Abbaszadeh, S.; Keykhosravy, K. Evaluation of antioxidant and antifungal properties of the traditional plants against foodborne fungal pathogens. *J. Mycol. Med.* **2016**, *26*, e11–e17. [[CrossRef](#)] [[PubMed](#)]
5. Ghaedi, M.; Naghiha, R.; Jannesar, R.; Dehghanian, N.; Mirtamizdoust, B.; Pezeshkpour, V. Antibacterial and antifungal activity of flower extracts of *Urtica dioica*, *Chamaemelum nobile* and *Salvia officinalis*: Effects of Zn[OH]₂ nanoparticles and Hp-2-minh on their property. *J. Ind. Eng. Chem.* **2015**, *32*, 353–359. [[CrossRef](#)]
6. Sugier, D.; Sugier, P.; Jakubowicz-Gil, J.; Winiarczyk, K.; Kowalski, R. Essential oil from *Arnica Montana* L. Achenes: Chemical characteristics and anticancer activity. *Molecules* **2019**, *24*, 4158. [[CrossRef](#)] [[PubMed](#)]
7. Miliuskas, G.; Venskutonis, P.R.; Van Beek, T.A. Screening of radical scavenging activity of some medicinal and aromatic plant extracts. *Food Chem.* **2004**, *85*, 231–237. [[CrossRef](#)]
8. Silva, A.; Rodrigues, C.; Garcia-Oliveira, P.; Lourenço-Lopes, C.; Silva, S.A.; Garcia-Perez, P.; Carvalho, A.P.; Domingues, V.F.; Barroso, M.F.; Delerue-Matos, C.; et al. Screening of bioactive properties in brown algae from the northwest iberian peninsula. *Foods* **2021**, *10*, 1915. [[CrossRef](#)] [[PubMed](#)]
9. Boudieb, K.; Kaki, S.A.; Oulebsir-Mohandkaci, H.; Bennacer, A. Phytochemical Characterization and Antimicrobial Potentialities of Two Medicinal plants, *Chamaemelum nobile* (L.) All and *Matricaria chamomilla* (L.). *Int. J. Innov. Approaches Sci. Res.* **2018**, *2*, 126–139. [[CrossRef](#)]
10. Cockerill, F.R.; Wikler, M.A.; Alder, J.; Dudley, M.N.; Eliopoulos, G.M.; Ferraro, M.J.; Hardy, D.J.; Hecht, D.W.; Hindle, J.A.; Patel, J.B.; et al. *Performance Standards for Antimicrobial Disk Susceptibility Tests: Approved Standard*, 11th ed.; Clinical and Laboratory Standards Institute: Berwyn, PA, USA, 2012; Volume 32, ISBN 1562384856.