Since 1987
REVIEW OF CLINICAL PHARMACOLOGY AND PHARMACOKINETICS, INTERNATIONAL EDITION | 38 (Sup2):165-168 (2024) Rcpp

Received: 13 February 2024 | **Accepted**: 27 March 2024 | **Published**: 5 May 2024

Open Access | Research

Chemical analysis and antibacterial activity of the *Achillea millefolium* extract

Walaa Jubair Sabbar[1], Raghad S. Mouhamad[2,*], Ibrahim A. Murad[3], Entisar E. Al-Abodi[4]

[1]Directorate General of Education in Baghdad (Karkh1), Baghdad, Iraq
[2]Ministry of Science and Technology, Baghdad, Iraq
[3]University of Garmian, Kurdistan, Iraq
[4]Chemistry Department, College of Education for Pure Science / Ibn Al-Haitham, University of Baghdad, Baghdad, Iraq

***Corresponding author:** Raghad S. Mouhamad, Ministry of Science and Technology, Baghdad, Iraq
Tel.: +964-(0)7702719152
E-mail: raghad1974@yahoo.com

Abstract

Achillea millefolium L., a traditional medicinal plant from the Asteraceae family, is known for its anti-inflammatory, anti-ulcer, and anti-cancer properties, attributed to its rich content of phytochemicals (such as flavonoids, alkaloids, terpenes, tannins, and phenolic acids). This study has focused on the chemical analysis of the oil extracted from the aerial parts of *Achillea millefolium* subsp. *millefolium*, cultivated and harvested in Kurdistan, Iraq. The oil was extracted by using a Clevenger apparatus for hydro-distillation and was analysed by using gas chromatography with a flame ionization detector and an HP-5 MS capillary column. The analysis revealed the presence of 1,8-cineole (eucalyptus oil), Artemisia ketone, camphor, linalyl acetate, and D-limonene, with Artemisia ketone having the highest concentration at 15.04% and D-limonene the lowest at 7.51%. The extract of *Achillea millefolium* has been shown to aid in the treatment of oral mucositis (a common side effect of anticancer chemotherapy), likely due to the flavonoids and tannins present in the plant.

KEYWORDS
yarrow, gas chromatography - mass analysis, camphor, Artemisia ketone, 1,8-cineole

How to cite: Sabbar W. J., Mouhamad R. S., Murad I. A., Al-Abodi E. E. Chemical analysis and antibacterial activity of the *Achillea millefolium* extract. *Rev. Clin. Pharmacol. Pharmacokinet. Int. Ed.* 38 (Sup2): 165-168 (2024).
https://doi.org/10.61873/CGWA5841

Publisher note: PHARMAKON-Press stays neutral with regard to jurisdictional claims in published maps and institutional affiliations.

1. INTRODUCTION

Many people use yarrow (*Achillea millefolium* L.), a Asteraceae family plant, as an ancient medicinal herbaceous plant [1]. Yarrow is native to Europe and Asia, but it has spread to North America. It is also known as “gordaldo”, “nosebleed plant”, “old man’s pepper”, “devil’s nettle”, “sanguinary”, “milfoil”, “soldier’s woundwort”, “thousand-leaf”, and “thousand-seal” [2]. Yarrow grows naturally in pastures, meadows, roadside ditches, and open forests. The herb has anti-bleeding qualities and promotes wound healing. The yarrow root, on the

other hand, has anaesthetic properties; it has been used in order to relieve toothaches, by applying fresh roots or leaves to the gums [3]. *Achillea millefolium* has been used in traditional medicine for centuries [4,5]. It possesses anti-inflammatory, anti-ulcer, and anti-cancer properties. Flavonoids, alkaloids, terpenes, tannins, phenolic acids, and other phytochemicals are abundant in *Achillea millefolium*, with flavonoids and phenolic acids being the most important components. The main component of the *Achillea millefolium* essential oil is chamazulene [6]. In terms of its beneficial effects in dental pathology, it has been demonstrated that the plant's extract assists in the treatment of oral mucositis; a typical side-effect of standard anticancer therapies [7].

2. MATERIALS AND METHODS

Achillea millefolium plants were cultivated and harvested from their natural habitats in Kurdistan, Iraq. The aerial parts of the plants were air-dried and stored in paper bags for 30 days. The dried plant materials (100 g) were then powdered and subjected to hydro-distillation by using Clevenger-type equipment for a duration of 3 h [8]. The extracted essential oil from *Achillea millefolium* was analysed using an HP 6890 gas chromatograph equipped with a flame ionization detector (FID) and an HP-5 MS capillary column (30 m; 0.25 mm i.d.; film thickness 0.25 µm). The same column and analytical conditions were used as in gas chromatography (GC) - mass spectrometry. The percentage composition of the essential oil was estimated by using GC-FID peak areas, without the use of adjustment factors. Each bioactive compound that was isolated and evaluated in the essential oil of *Achillea millefolium* has been studied in the literature. The potential bioactivity of these compounds against the common cavity-causing bacterium, *Streptococcus mutans*, was highlighted, particularly in the context of mouthwash production. The antibacterial activity of the yarrow oil extract was assessed against several bacterial species (*Streptococcus salivarius*, *Pseudomonas aeruginosa*, *Staphylococcus aureus*, and *Staphylococcus epidermidis*) by using the agar diffusion technique. A bacterial suspension (100 µL) containing approximately 1×10^8 cells/mL (McFarland solution) was uniformly spread on the surface of a Mueller-Hinton agar in a petri dish (90-mm diameter). Then, 0.1 mL of each concentration (0%, 0.1%, 0.3%, and 0.5%) of the yarrow extract was aseptically introduced into the wells (5-mm diameter). The plates were left for 30 min before their incubation at 37°C for 24 h. The inhibitory zones (in mm) were measured, indicating the antibacterial activity [9].

3. RESULTS AND DISCUSSION

Our GC-mass spectrometry analysis has shown that the yarrow oil contains numerous anti-inflammatory chemicals. The chemical analysis spectrum of the yarrow essential oil is influenced by intrinsic factors such as the number of chromosomes and whether the plant is diploid or tetraploid. Extrinsic factors, including the stage of harvest, the part of the plant, the harvesting season, the geographical origins, and the method used for the oil extraction and analysis, can also affect the chemical composition and yield of the oil obtained from the yarrow. Artemisia ketone, a member of the enone functional group with the structure RC(=O)CR', has been found to exist at 3.1% to 12.9%. Artemisia ketone is almost insoluble in water at 0.29 g/L and is a very weak basic (essentially neutral) molecule. This green, herbal chemical with a berry flavour is also found in the sunflower and in the tarragon. Linalyl acetate, which makes up 12.08% of the *Achillea millefolium* essential oil, is a phytochemical molecule found in many flowers, and has a lavender scent. Linalyl acetate is chemically composed of linalool acetate ester; it has a pleasant odour and a taste that is similar to its smell, and it can be used as a safe fragrance substance. 1,8-Cineole was found to be the third component at 11.08%, which varied from the oil recovered from commercial yarrow plants. Eucalyptol is another name for 1,8-cineole, which is naturally formed as a cyclic ether and monoterpenoid by several plants, most notably eucalyptus. Camphor and D-limonene, both terpenes, were found at 10.75% and 7.51%, respectively, in the extracted oil of the yarrow. Camphor was found to be a prominent ingredient of the yarrow oil, with a range of 0.6% to 17.6%. Camphor oil is primarily derived by steam distillation from the wood of camphor trees. D-limonene, a key ingredient in various citrus oils, is generally regarded as safe and may be found in popular food items. Camphor is often used in creams, ointments, and lotions so as to reduce skin irritation and itching, and may help enhance the skin's overall appearance.

Yarrow oil has been found to have antibacterial and antifungal properties, making it effective in treating infections. It is suggested that the yarrow extract could be used as a mouthwash to alleviate mucositis and ulcers following chemotherapy, and to reduce harmful bacterial dysbiosis. It could also be used in creams or lotions to prevent acne in young people, based on the bioactive ingredients identified in the oil. Oil extracted from *Achillea* spp. was tested against *Propionibacterium acnes* and *Staphylococcus epidermidis*, showing varying degrees of antibacterial activity. Yarrow oil, which is

rich in eucalyptol, could potentially be used in the manufacturing of mouthwash and cough suppressants. Eucalyptol has been shown to regulate airway mucus hypersecretion and asthma through the suppression of inflammatory cytokines. It can effectively treat nonpurulent rhinosinusitis and, when applied topically, can relieve inflammation and discomfort. The antibacterial efficacy of the oily yarrow extract against common cavity bacterial species has been discussed. The *Achillea millefolium* extract has demonstrated a dose-response antibacterial activity. All tested bacteria were susceptible to yarrow oil extract at its higher concentration of 0.5%. *Streptococcus salivarius* was less susceptible, with an inhibition zone reaching 11 mm, followed by *Staphylococcus epidermidis* with an inhibition zone of 13 mm. The yarrow oil extract showed higher antibacterial activity against *Staphylococcus aureus*, with an inhibition zone reaching 21 mm. An ethanol extract of the *Achillea millefolium* aerial parts was previously tested for antimicrobial activity against *Escherichia coli*, *Bacillus cereus*, *Pseudomonas aeruginosa*, *Salmonella enteritidis*, and *Candida albicans*. The highest MIC value of 62.50 mg/mL was observed against *Bacillus cereus* and *Salmonella enteritidis*, with no activity observed in the other three tested strains.

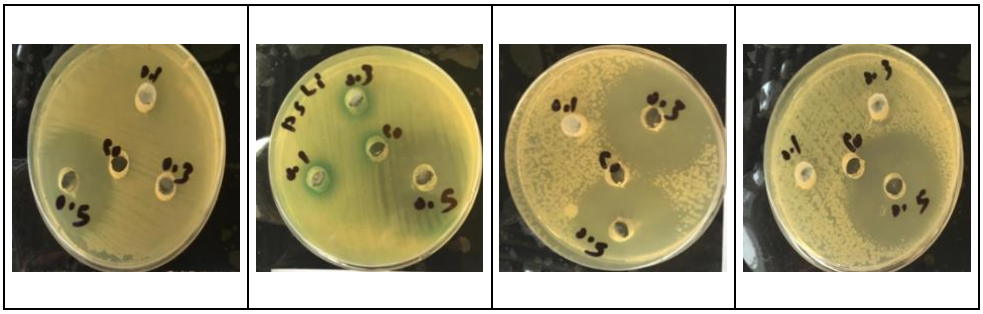

Figure 1. Antibacterial activity of the yarrow extract against various bacterial species at different concentrations.

4. CONCLUSION

A total of 19 compounds were discovered in our GC-mass spectrometry analysis of *Achillea millefolium*. The most common components were Artemisia ketone (15.04%), camphor (10.27%), linalyl acetate (12.08%), 1,8-cineole (11.08%), and D-limonene (7.51%).

ACKNOWLEDGEMENTS

We would like to extend our thanks to the institutions and funding bodies that supported our work. Their backing was crucial in carrying out our research and achieving our objectives. Finally, we wish to acknowledge the tireless efforts of the technical staff who contributed significantly to the successful completion of our research. Their dedication and hard work have not gone unnoticed and we are immensely grateful for their support.

CONFLICT OF INTEREST STATEMENT

The authors declare no conflicts of interest.

REFERENCES

1. Ghasemzadeh A., Jaafar H.Z., Rahmat A.: Antioxidant activities, total phenolics and flavonoids content in two varieties of Malaysia young ginger (*Zingiber officinale* Roscoe). *Molecules* 15(6): 4324-4333 (2010).
DOI: **10.3390/molecules15064324**
PMID: **20657444**

2. Saeidnia S., Gohari A., Mokhber-Dezfuli N., Kiuchi F.: A review on phytochemistry and medicinal properties of the genus *Achillea*. *Daru* 19(3): 173-186 (2011).
PMID: **22615655**

3. Khorasani G., Hosseinimehr S.J., Zamani P., Ghasemi M., Ahmadi A.: The effect of saffron (*Crocus sativus*) extract for healing of second-degree burn wounds in rats. *Keio J. Med*. 57(4):190-195 (2008).
DOI: **10.2302/kjm.57.190**
PMID: **19110531**

4. Candan F., Unlu M., Tepe B., Daferera D., Polissiou M., Sökmen A., *et al.*: Antioxidant and antimicrobial activity of the essential oil and methanol extracts of *Achillea millefolium* subsp. *millefolium* Afan. (Asteraceae). *J. Ethnopharmacol*. 87(2-3): 215-220 (2003).
DOI: **10.1016/s0378-8741(03)00149-1**
PMID: **12860311**

5. Stojanović G., Radulović N., Hashimoto T., Palić R.: *In vitro* antimicrobial activity of extracts of four *Achillea* species: the composition of *Achillea clavennae* L.

(Asteraceae) extract. *J. Ethnopharmacol*. 101(1-3): 185-190 (2005).
DOI: **10.1016/j.jep.2005.04.026**
PMID: **15978758**

6. Başer K.H.C.: Essential oils of *Achillea* species of Turkey. *Nat. Volatiles Essent. Oils* 3(1): 1-14 (2016).

7. Mahdi M.S., Mouhamad R.S., Al-Khafaji K.A., Al-Abodi E.E.: The production of nano based medicinal herbal mixture and its application in an inhibition and elimination of burn-contaminating germs. *Int. J. Agricult. Stat. Sci*. 18(s1): 1491-1497 (2022).

8. Emad A., Al-Abodi E.E. Anti-inflammation effects of silver nanoparticles - zinc polycarboxylate cement (AGNPS-ZPCCEM). *Pak. J. Med. Health Sci*. 16(4): 943-946 (2022).
DOI: **10.53350/pjmhs22164943**

9. Mouhamad R.S., Al Khafaji K.A., Al-Dharob M.H., Al-Abodi E.E. Antifungal, antibacterial and anti-yeast activities evaluation of oxides of silver, zinc and titanium nanoparticles. *Chem. Int*. 8(4): 159-166 (2022).
DOI: **10.5281/zenodo.7317423**