Vacuum oven in determination of phenolic compound characteristics

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> **The Memmert vacuum oven VO as part of a university study that looks at phenolic compounds of artemis rutifolia.**

The Memmert vacuum oven VO was used in a 2017 study conducted jointly by the Government College Women University and University of Agriculture, in Pakistan. It was published in the International Journal of Food Properties, Volume 20, Issue 11 (2017).

Detailed information is now available in the vacuum oven whitepaper. Register here to access it in our downloads section.

The study is titled "*Phenolic compounds' characterization of Artemisia rutifolia spreng from Pakistani flora and their relationships with antioxidant and antimicrobial attributes*". The full study can be read here. This article discusses the highlights of the report.

Artemisia rutifolia. (c) B.Byambajav – some rights reserved (CC BY-NC).

Artemisia rutifolia. Courtesy of Ji-Elle.

For more applications of the laboratory vacuum drying oven, read our blog. Or contact us for a personal consultation. For further information on drying ovens, its technology, uses, versatility and more, read this explainer blog.

Background

Phenolics, a class of bioactive compound, have antioxidative and antimicrobial properties. The science community has long been engaged in studying organic sources of phenolic compounds for wellness purposes.

This brings forth the objective of this study where a Memmert heating and drying oven – vacuum oven VO was used. The researchers looked into valorizing the medical plant known as *Artemisia rutifolia*, from Pakistan, and determine its *novel natural therapeutic agents*.

...there was complete gap of knowledge about phytochemical and pharmacological aspects of A. rutifolia. Therefore, in the current study, for the first time, an effort was made to explore phenolic profile as well as antioxidant and antimicrobial activities of different extracts from A. rutifolia leaves.

Materials and methods

Chemicals and reagents

- Gallic acid (GA),2,2′-diphenyl-1-picrylhydrazyl (DPPH)
- Ascorbic acid
- Quercetin
- Other reference compounds
- Cell culture reagents
- Sodium carbonate
- Folin-Ciocalteu's phenol reagent
- Ferric chloride
- Aluminum chloride
- Potato dextrose agar
- Nutrient agar
- Potassium ferricyanide
- Agar powder
- Methanol
- Chloroform
- Hexane

Preparation of extract with Memmert VO

In the beginning, the sample of A. rutifolia was shade-dried and later powdered by a food processor. The result was filtered through a strainer (0.50 mm). Next follows extraction and filtration. Later, the filtrates undergo evaporation in a Memmert vacuum oven to constant weight.

After this stage, the dried extracts are scratched with a spatula that has been sterilized, stored in extract vials in the refrigerator at −4°C.

Continue reading for more information on how a vacuum oven works, its features and applications, or reach out for expert consultation. Latest information on vacuum ovens and other heating and drying ovens can be found here.

Determination of phenolic content

Total phenolic content was determined using Folin-Ciocalteu's reagent method. 1 mL of individual extract solution, at an appropriate dilution level, was mixed with 500 µL of Folin-Ciocalteu's reagent and 7.5 mL of double-deionized water. To this, mL of 5% Na2CO3 (W/V) solution is further added whose resulting mixture was set to incubation at room temperature for a 90-minute period.

The absorbance is measured, and the total phenolic contents was given as µg gallic acid equivalents (GAEs) per mg of extract.

Total flavonoid content

Aluminum chloride method is used to determine total flavonoid content. Extract solutions, at 0.5 mL, is mixed with 1.5 mL of 95% ethanol (V/V), 0.1 mL of 10% aluminum chloride (m/V), 0.1 mL of 1 mol L–1 potassium acetate and 2.8 mL of water. The resulting mixture, like before, is set to incubation at room temperature for a 30-minute period. Absorbance is measured using a spectrophotometer and the total flavonoid content is given as µg quercetin equivalents (QEs) per mg of plant extract.

HPLC analysis of phenolic compounds

The hydrolysis technique for *A. rutifolia* leaf extracts follows the methods of this study with Mengkudu leaf extracts.

50g of the test sample extracts are dissolved in 24 mL of methanol to allow for homogenization. 16 mL of distilled water and 10 mL of 6M HCl are added. This followed thermostatic process for 2 hours at 95°C. Later, the solution is filtered and subsequently goes through high-performance liquid chromatography (HPLC) analysis.

The separation of plant samples on the gradient HPLC is done. Phenolic acid and flavonoid identities are established, as well as the limit of detection (LOD) and limit of quantification (LOQ) are calculated.

Free radical scavenging activity (DPPH assay)

This is carried about as per the methods applied in the study exploring the antioxidant properties of flowers and roots of Pyrostegia venusta (Ker Gawl) Miers.

Extract solution is mixed with equal volumes of 100 µM DPPH solution in methanol whereby its resulting mixture is incubated for 15 minutes at room temperature. The absorbance is recording by using a UV–visible spectrophotometer at 517nm.

Ferric reducing–antioxidant power (FRAP) assay

Reducing power was determined following ferric reducing–antioxidant power (FRAP) assay. Detailed information on it by reading the antioxidant and antibacterial activity of leaves of *Etlingera* species (Zingiberaceae).

The absorbance of reaction mixtures, whose details and steps are better explained in the report, is measured via UV–vis spectrophotometer set to 700nm. Total oxidant activity is recorded as well as documenting total oxidant content as mg gallic acid equivalent/g of plant extract.

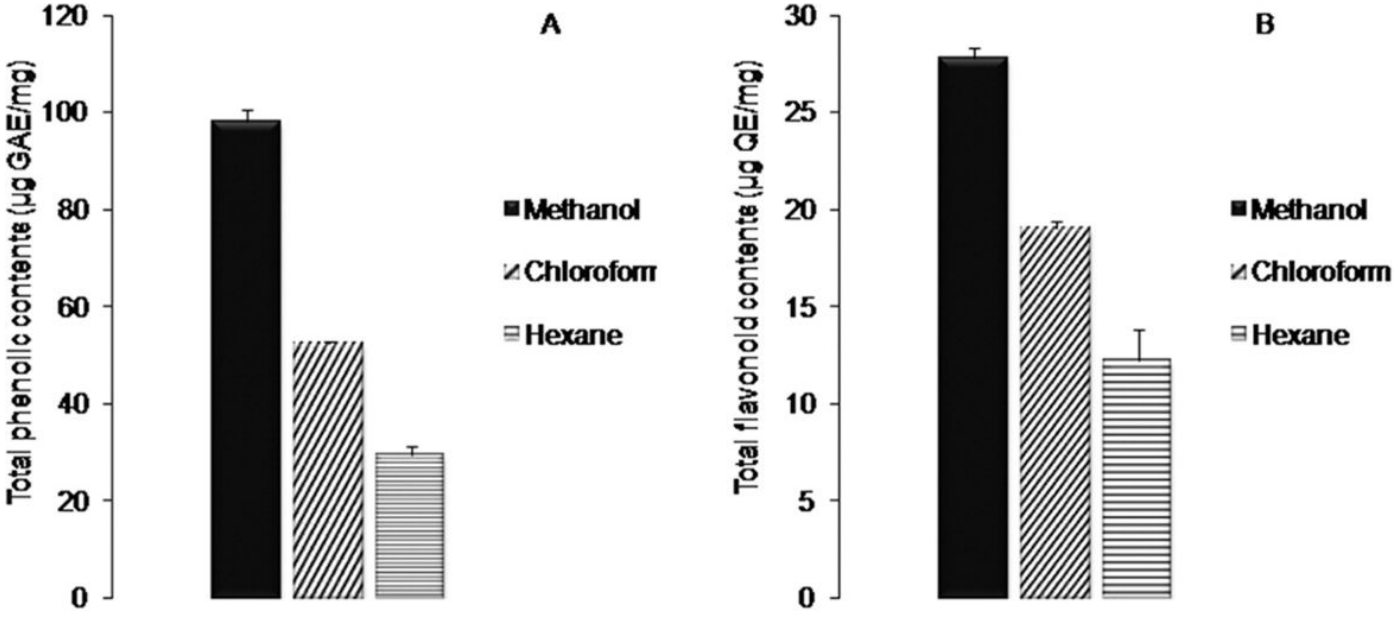
Antimicrobial activity

In this step, in order to be able to calculate the antimicrobial activity of *A. rutifolia* leaf extracts, the researchers used agar disc diffusion assay Inoculation of nutrient agar and potato dextrose took place. Afterwards, these were placed into sterilized petri plates. Dilutions of the extracts were made with biological-grade dimethyl sulfoxide (DMSO). Sterile filter discs impregnated with 50 µL of diluted plant extract solution are then placed into the aforementioned petri plates. These places undergo incubation at 37°C for 24-hour and at 27°C for 48-hour time period. Later, Antibacterial and antifungal activities are determined by using a zone reader.

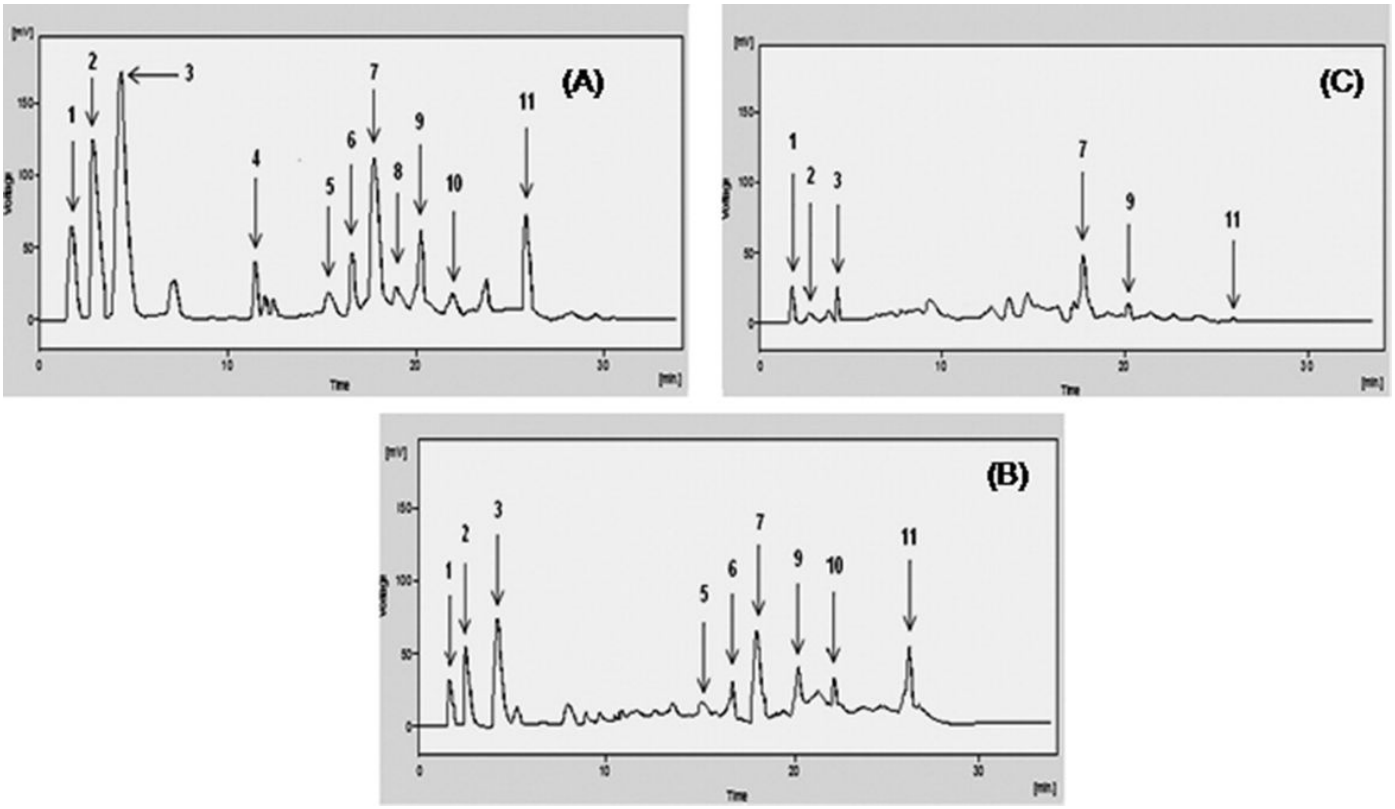
Estimation of minimum inhibitory concentration (MIC) values

The calculation of the minimum inhibitory concentration (MIC) was carried out by following the methods laid out in the broth microdilution method. After experimentation, the plates are incubated at 37°C for 24 hours for bacteria and 27°C for 48 hour for fungi. It is noted that the reason for the color change of indicator, resazurin, varies as per microbial growth.

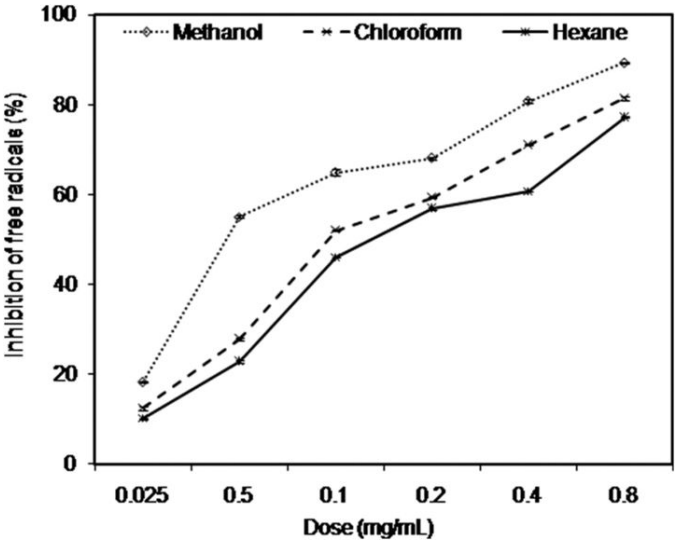
Results



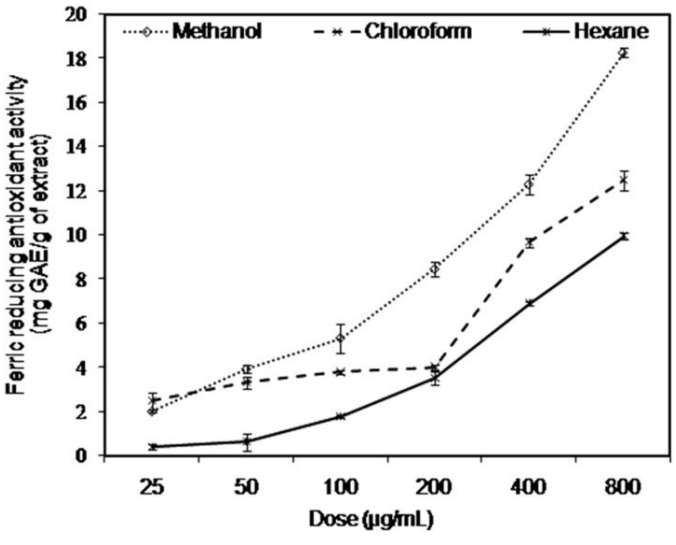
Polyphenolic compounds in different solvent extracts of A. rutifolia leaves. (A) Total phenolic compounds expressed as microgram gallic acid equivalent per milligram of plant extract. (B) Total flavonoid compounds expressed as microgram quercetin equivalent per milligram of plant extract. Courtesy of Taylor & Francis Online.



HPLC phenolic profile of (A) methanol, (B) chloroform, and (C) hexane extracts of A. rutifolia leaves. Peaks: (1) myricetin, (2) quercetin, (3) gallic acid, (4) caffeic acid, (5) chlorogenic acid, (6) syringic acid, (7) p-coumaric acid, (8) vanillic acid, (9) m-coumaric acid, (10) ferulic acid, and (11) sinapic acid. Courtesy of Taylor & Francis Online.



Inhibition (%) of free radicals (DPPH) by A. rutifolia leaf extracts. Courtesy of Taylor & Francis Online.



Ferric reducing–antioxidant power (FRAP) of A. rutifolia leaf extracts. Courtesy of Taylor & Francis Online.

Conclusion

With regards to the objective set forth by the researchers for this study, upon completing the experiments, key findings were recorded.

...extracts presented relatively potent antioxidant and antimicrobial potential that could explain their promise for prevention or treatment of diseases.

- Profiling of polyphenols led to identifying medicinally valuable phenolic acids and flavonoids.
- The experiment steps in the right direction towards *A. rutifolia* leaves' potential as a key source of natural antioxidants for the food and the pharmaceutical industries.
- Comparisons made of phenolic composition as opposed to solvents of variable polarity can assist in solvent optimization of phenolic compounds.

About Memmert vacuum oven

Memmert GmbH + Co.KG, operating out of Schwabach and manufacturing from Büchenbach, produces the vacuum oven VO as part of its diverse heating and drying oven range.

It offers features such as:

- Temperature range up to +200 °C
- Vacuum control range: 5 to 1100 mbar
- 3 model sizes (29 to 101 litres volume)
- 1 model variant: TwinDISPLAY
- Anti-splinter; VDE-tested door construction for all models
- Pump control: Optimised rinsing of the pump membrane as well as signal output for switching the pump ON/OFF according to requirements.
- Optional: Pump base cabinet and energy-efficient vacuum pump
- Nearly exclusive use of high-quality, rust-resistant and easy to clean stainless steel for interior and exterior housing
- Precise and homogenous temperature control thanks to a product-specific heating concept
- A wide range of options for programming and documentation using interfaces, integrated data loggers and the AtmoCONTROL software
- 3 years guarantee worldwide

Memmert vacuum oven VO.

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