

A Study on Phytochemical, Elemental Analysis and Antimicrobial Activity of Leaves of *Mentha arvensis* Linn.

Thida Kyaw*, Pa Pa San**, Khin Htwe Kyaing***

Abstract

Mentha arvensis Linn. leaves have been given to treat indigestion, skin diseases, coughs, rheumatic pains, arthritis and as remedy for inflamed joints. Above the pros, this research definitely was done. It deals with the study of phytochemical constituents, elemental analysis and antimicrobial activity of leaves of *Mentha arvensis* Linn. (Pudina). It is usually consumed as a vegetable. The phytochemical investigation of pudina indicated the presence of alkaloids, α -amino acid, flavonoids, glycosides, phenolic compounds, reducing sugar, saponins tannins and steroids whereas starch, organic acids and terpenoids were not detected. The qualitative elemental analysis showed that K, Ca, S, P, Fe, Mn, Ti, Zn, Cu, Rb and Br were present in sample by using EDXRF spectrophotometer. Antimicrobial activity was studied by agar well diffusion method against *Bacillus subtilis*, *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Candida albicans* and *Escherichia coli*. Water extract did not show activity on all species of microorganisms. Ethanol extract of pudina leaves showed higher antimicrobial activity against than other extracts by tested organisms.

Keywords: Pudina, Phytochemistry, EDXRF, Antimicrobial activity

Introduction

During the past decades, traditional system of medicine has become a topic of global interest. Current estimates suggest that in many developing countries a large proportion of the population relies heavily on traditional practitioners and medicinal plants to meet primary health care needs. The WHO recognizes that nearly 80% of world's population is depending on traditional medicinal for the treatment of disease using roots, fruits, stems, leaves, barks, seeds, flowers or any other parts of plant (Phillipson and Wright, 1991). Herbs have provided humankind with medicine from the earliest beginning of civilization. It is estimated that there are 250,000 – 500,000 plants on earth (Borris RP., 1996) and relatively small percentage (1 – 10%) of these are used as a food for human and other animal species. It is likely that even more of these are used for medicinal purpose (Moerman DE. 1996).

Mentha arvensis Linn. is used as a food seasoner, household remedy, and industrial purposes. Juice of leaves is given in diarrhea and dysentery. These leaves are mostly used as salad and medicinally used for stomach problems and allergy (Khan and Khatoun, 2008). It is also used for treatment of liver and spleen disease, asthma and jaundice. It is traditionally used in hypertension and for patients with ischemic heart disease.

Pudina is an edible green plant. Its English name is field mint and its Botanical name is *Mentha arvensis* Linn. belonging to the family of Lamiaceae. Pudina is Bengali name. The Myanmar name is Pusinan. Pudina is distributed in central and southwestern Asia and Europe. They grow well at the riverbanks, edges of marches and moist prairies. These plants need regular watering in order to grow properly. Plenty of sunlight is ideal for their proper growth.

Mentha arvensis Linn. is packed with vitamins such as vitamin C, vitamin A and vitamin B and minerals like magnesium, iron, phosphorus, zinc, potassium, calcium and folic acid. These leaves are used as herb in various cuisines and as Herbal tea, for treatments of fever, headache, cold, asthma, cough syrups, and to reduce toothaches, chest pains and other heart ailments. The leaves as well as the oil of these plants are used in toothpastes and mouth

* Associate Professor, Dr, Department of Chemistry, Yadanabon University

** Lecturers, Dr, Department of Chemistry, Yadanabon University

*** Lecturers, Dr, Department of Chemistry, Yadanabon University

washes. It is a good source of the bioflavonoid quercetin with many other flavonoids which exhibits antioxidant, antispasmodic, antiphlogistic and antidepressant.

Botanical Description

Scientific name	: <i>Mentha arvensis</i> Linn.
English name	: Field mint
Family	: Lamiaceae
Myanmar name	: Pusinan
Bengali name	: Pudina
Part used	: Leaves



Fig 1 Leaves of *Mentha arvensis* Linn. (Pudina)

Aim and Objectives

The aim of the present study is to determine the elements from the sample and to investigate the antimicrobial activity of the leaves of *Mentha arvensis* Linn. (Pudina). To fulfill the aim, the following objectives were carried out.

- To collect the sample
- To investigate the preliminary phytochemical constituent in the sample
- To detect the elemental contents of the sample
- To determine the antimicrobial activity of the crude extract

Materials and Methods

General Experimental Techniques

Mentha arvensis Linn. was collected from Mahaangmyae Township, Mandalay Region. These samples were cut into small pieces and air-dried at room temperature. This sample was ground into powder in an electric blender and stored in airtight container. Preliminary phytochemical constituent was investigated by test tube method. Qualitative analysis of some elements in leaf samples was measured by Energy Dispersive X-Ray Fluorescence (EDXRF) Spectrometer. Antimicrobial activity was studied by agar well diffusion method.

Preliminary Phytochemical Test

A few grams of dried sample powder was subjected to the tests of alkaloids, α - amino acids, carbohydrates, flavonoids, glycosides, organic acids, phenolic compounds, reducing sugars, saponins, starch, steroids, Terpenoids and tannins according to the standard procedures, (Trease and Evans, 1980; Harborne, 1984; Marini-Bettolo, 1981).

Elemental Analysis of *Mentha arvensis* Linn.

In order to know the concentrations of some elements in leaf samples, it was investigated by using EDXRF spectrometer at department of Physics, University of Mandalay. The resultant EDXRF spectral data were shown in Table 1.

Preparation of Crude Extracts

About 10 g of dried powder sample was percolated with 100 cm³ of pet-ether for 24 hours and 2 days. It was filtered and filtrate was placed in a weighted porcelain basin and evaporated to dry on a water-bath until it was completely dried. The residue with the basin was weighted. The difference in weights of basin before and after the experiment was taken to be the pet-ether soluble matter content. Ethyl acetate and 95 % ethanol extracts of dried powder sample were also prepared by percolation method. Water extract of this sample was prepared by boiling 10 g of sample with 200 cm³ distilled water for 3 hours and filtered. According to similar manner mentioned in above procedure to yield the respective water extract. Each extract was stored in refrigerator for screening of antimicrobial activities.

Screening of Antimicrobial Activity

Medicinal plants are known to produce certain bioactive molecules which react with other organisms in the environment, inhibiting bacterial and fungal growth (Chopra *et al.*, 1992). Bacteria and fungi are various types of organisms. They can also cause tissue damage and temper with our body functions to the point of causing disease. The antimicrobial activity was screened by employing agar well diffusion method at Biotechnological department, Mandalay Technological University. The antimicrobial activity of four crude extracts such as petroleum ether, ethylacetate, ethanol and water from the leaves of *Mentha arvensis* Linn. was determined against six strains of microorganism. The results are described in Table 2 and Figure 2 and 3.

Results and Discussion

The phytochemical constituents of *Mentha arvensis* Linn. was investigated by test tube method. Alkaloids, α -amino acid, flavonoids, glycosides, phenolic compounds, reducing sugar, saponin, tannins and steroids whereas starch, organic acids and terpenoid were not detected. The qualitative elemental analysis showed that K, Ca, S, P, Fe, Mn, Ti, Zn, Cu, Rb and Br were present in sample by using EDXRF spectrophotometer. Alkaloids are reported to have anti-inflammatory, antimicrobial and antifungal effect and also act as an antihypertensive agent. Tannins from plants are reported to have healing properties. Flavonoids and phenolic are most important group of secondary metabolites and bioactive compounds in plants. Flavonoids are protective against cardiovascular diseases, cancers and other related diseases. Phenolic is major group of compound having capability of scavenging reactive oxygen species. Hence it may act as antioxidant.

According to the results of mineral compositions, potassium and calcium were the highest. K, Ca, S, P, Fe, Mn, Ti and Zn are essential elements for living organism, being required for growth and developments. In human body, potassium is essential for building muscles, preventing strokes, maintaining body growth and controlling acid-base balance. A diet rich in potassium helps to offset some of sodium's harmful effects on blood pressure. The main function of the most of calcium is utilized structuring the human skeleton along with collagenous fibers. Sulfur plays a critical role in detoxification, as it is part of one of the most important antioxidants that our body produces glutathione. Phosphorus is essential for the normal function of every cell, performing a variety of functions. The toxic metal of copper was observed in this sample but the amounts of these metals were little trace. Therefore, the sample can be used for medicinal purpose.

Antimicrobial activity of crude extracts such as pet-ether, ethylacetate, 95% ethanol and water extracts from *Mentha arvensis* Linn. was investigated by agar well diffusion method against six microorganisms. The measurable zone diameter, including the agar well diameter showed the degree of antimicrobial activity. The results are described in Table 2 and the inhibition zones of extracts against microorganisms tested are shown in figure 2 and 3.

According to the data, it was found that water extract cannot show any antimicrobial activity. Pet-ether extract showed low activities on all selected microorganisms. Ethylacetate extract showed medium activities on *Bacillus subtilis*, *Pseudomonas aeruginosa* and *Candida albicans*.

Table 1. Relative Abundance of Element Contents in Leaves of *Mentha arvensis* Linn. by EDXRF Method

No.	Elements	Relative abundance (%)
1	Potassium (K)	2.986
2	Calcium (Ca)	0.935
3	Sulphur (S)	0.416
4	Phosphorus (P)	0.310
5	Iron (Fe)	0.042
6	Manganese (Mn)	0.016
7	Titanium (Ti)	0.005
8	Zinc (Zn)	0.004
9	Copper (Cu)	0.002
10	Rubidium (Rb)	0.001
11	Bromine (Br)	0.001

Table.2 Results of Antimicrobial Activities of Crude Extracts from the Leaves of *Mentha arvensis* Linn.

Samples	Solvents	Organisms					
		1	2	3	4	5	6
<i>Mentha arvensis</i>	H ₂ O	-	-	-	-	-	-
	pet-ether	+	+	+	+	+	+
	EtOAc	++	++	++	++	++	++
	EtOH	+++	++	+++	++	+++	++

Organisms

Diameter of Agar well ~ 10 mm

- (1) *Bacillus subtilis* 10 mm ~ 14 mm = (+)
- (2) *Staphylococcus aureus* 15 mm ~ 19 mm = (++)
- (3) *Pseudomonas aeruginosa* 20 mm above = (+++)
- (4) *Bacillus pumilus* no activity = (-)
- (5) *Candida albicans*
- (6) *Escherichia coli*



Bacillus subtilis



Staphylococcus aureus



Pseudomonas aeruginosa



Bacillus pumilus



Candida albicans



Escherichia coli

Fig. 2. Effect of Different Extracts from Pudina Leaves on Six Microorganisms

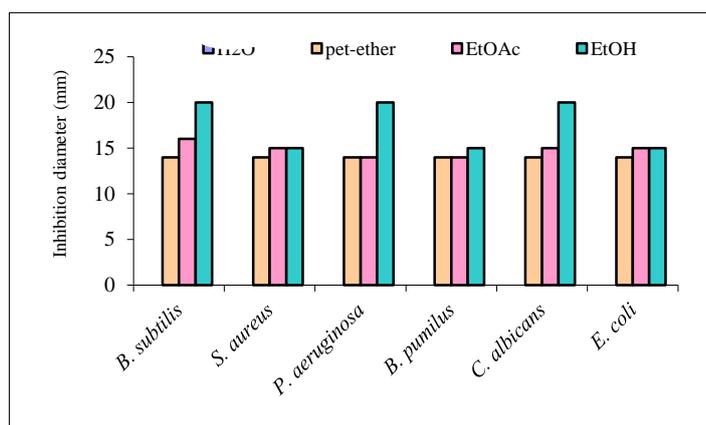


Fig.3 Bargraph of bacterial inhibition zone diameter (mm) of different crude extracts of Pudina on six microorganisms

Conclusion

The following inferences could be deduced from the chemical and biological activity investigations on the leaves of *Mentha arvensis* Linn. The preliminary phytochemical tests revealed the presence of alkaloid, flavonoids, α -amino acid, carbohydrates, glycosides, phenolic compounds, reducing sugar, saponins, tannins and steroids. Therefore, this sample contains valuable phytochemical constituents for human's health. But terpenoids, starch and organic acids were absent in the sample. According to EDXRF elemental analysis, it can be observed that K, Ca, S, P, Fe, Mn, Ti, Zn, Cu, Rb and Br were found in this sample. Among them, potassium was not abundant content in sample. From the experimental results, potassium was found to be highest amount in this sample. These minerals are essential dietary constituents for human life. However toxic metals were not detected. From antimicrobial investigation tests, almost all of the extracts (pet-ether, ethyl acetate and ethanol) exhibited antimicrobial and antifungal activities except watery extract. Among them, ethanol extract showed the higher potency with inhibition zone diameter up to 20 mm on *Bacillus subtilis*, *Pseudomonas aeruginosa*, *Candida albicans*. So, these results indicated that locally cultivated leaves of *Mentha arvensis* Linn. extract possess antimicrobial activity and they can be used for infections caused by bacteria and fungi.

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