ACHILLEA MILLEFOLIUM: DETERMINATION OF METALS AND MICROSCOPIC ANALYSIS OF TWO GROUPS OF DIFFERENT ORIGINS FROM BIHOR

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Abstract

At present in our countryand elsewhere in the world, a special interest is on the use of medicinal and aromatic plants. Medicinal flora of our country is represented by eight hundred species, of which two hundred and eighty-three have proven therapeutic properties.

The present paper is analyzing Achillea millefolium from two areas of Bihor county, one exposed to pollution, other pollution away on urbanization, using optical microscopy; nine metals were determined beneficial or not for human body by X-ray fluorescence.

Microscopic analysis showed that plant product harvested plants are different in the form of pollen and the section stalk plants from the two areas, even if metal content does not differ very much.

Keywords: *Achillea millefolium,* microscopic analysis, metals content, X ray fluorescence

INTRODUCTION

X-Ray fluorescence analysis is a versatile technique used for easy determination of different metallic elements into the liquid and also solid samples. Main problem with natural products is represented by the concentration of different oligoelements, because the higher and also lower concentrations of these elements can represent a danger for human health. Tea preparation involves the dissolution of metals from herbals used during preparation, and because of that is important that the used plants are not containing higher quantities of metals. Because, a large number of plants used in alternative medicine are coming from spontaneous flora, is important to know the elemental composition of used plants (Chuparina et al, 2011).

A recent study performed in Romania revealed that most used natural treatments are represented by: brotherwort (*Thymus serpyllum*),

milfoil (*Achillea millefolium*), chamomile (*Matricaria recutita*), rattle *(Hypericum perforatum*), marigold (*Calendula officinalis*), linden (*Tilia platyphyllos*) and peppermint (*Mentha piperita*).

In numerous cases the human activities are leading at environmental pollution is really important to evaluate the elemental composition of natural plants used into the alternative medicine for tea preparation, because the heavy metals can be transferred from soil and accumulate in plants during growth.

Milfoil (*Achillea millefolium*) was known since 2000 years ago, and was recorded firstly into the medical textbook of Dioscoride. Achillea millefolium is a perennial plant which belongs to Asteraceae family, having wedge shaped leaves, with fine hair and also white till pink flowers. This plant was cultivated in Europe and also into the west part of Asia. Originally milfoil plant was used as treatment for wounds, gangrene and also for infectious diseases. Later, into the Romanian alternative medicine have an important place being used as treatment for at least 100 diseases – digestive, dermatological immune and endocrine system diseases. Milfoil is a widespread plant which like direct sunlight, and have a highly resistance at lack of moisture. In order to better preserve the active principles is recommended to collect the plant in dry weather, preferable at noon, followed by dried in layers with thickness of maximum 8 cm in places free of moisture, shaded and well ventilated.

Although the milfoil plant has such a large application a detailed study was not performed until several years ago.

Fig.1. *Achillea millefolium*

In Romania the milfoil plant is used as: powder (as treatment for hormonal activities and also for bleeding), combined infusion (treatment for biliary, digestive and intestinal diseases), juice (as treatment against hepatitis and cirrhosis), tincture (hypertension and cardiovascular diseases), cataplasm (treatment of allergies), cream (treatment for sores and hemorrhoids), oils (burns treatment), and also for vaginal douches.

Into the latest years were carried out several tests in order to demonstrate the antimicrobial, anti-inflammatory and also antioxidant

capacity of milfoil (Benedek, B et al, 2007, Candan F. Et al, 2003, Benedek, B.et al, 2007, Innocenti, G. et al, 2007, Benedek, B. et al, 2006, Cavalcanti, A.M., 2006, Potrich, F.B. et al, 2010). Also was studied the plant composition (Chandler, R.F. et al, 1982, Csupor‐Löffler, B. et al, 2009, Pallag A., et al, 2011), and the properties of different extracts (Thorsell, W. et al, 2006, Yaeesh, S., et al, 2006, Lemmens-Gruber, R., et al, 2005, de Souza, P., et al, 2011, Dalsenter, P.R., et al, 2004).

Present paper is dealing with microscopic analysis of Achillea millefolium stem and pollen correlated with determination of plant metallic content, for plants collected from two different areas – one area near a pollution source (Oradea) and a different area faraway from the pollution source (Alesd). All metals concentrations were determinated by using X – Ray Fluorescence Analysis.

MATERIAL AND METHOD

It was analyzed plants harvested from Oradea city and also for an unpolluted area near Alesd (located at 40 km faraway from Oradea). The metal content were determined using three different samples harvested from each location.

Microscopic analysis was realized by using and optical microscope with 100 and 200 X magnification. *Achillea millefolium* metal content was determinate with Niton X-Ray fluorescence analyzer.

RESULTS AND DISSCUSIONS

Microscopic analysis

In order to perform the microscopic analyze, after the harvest of fresh plants, stains were cut crosswise. The obtained samples were analyzed by optical microscopy using 100 magnifications.

Achillea millefolium L. pollen derived from unpolluted area (Alesd) presents a tipical morfological structure (fig. 2.a).

Achillea millefolium L. pollen derived from near industrial area (Oradea) presents minor deviations, morphological and structural changes, changes in the number of pores and colpi, substance sedimentation, that appear as unspecific excrescences visible with an optical microscope (fig.2.b).

Fig.2.a. *Achillea millefolium L.* pollen (from unpolluted area – Alesd)

Fig.2.b. *Achillea millefolium L.* pollen (from near industrial area - Oradea)

At level of crosssections through main stem were not observed the significant differences, however at samples harvested from unpolluted areas was observed the presence of central lacunae (fig.3.a), and at samples harvested from polluted area was observed the presence of medullar parenchyma is present also into the spinal center (fig.3.b)

Fig.3.a. Cross section through the *Achillea millefolium L.* main stem from unpolluted area (100X): 1- colenchima; 2- assimilated parenchyma; 3- leading bundles; 4 - medular parenchyma;5- main gap marrow; 6- epidermis

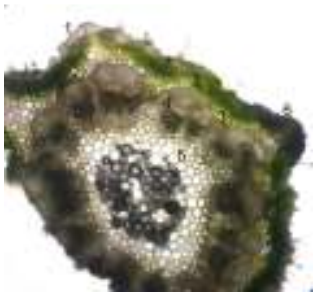

Fig.3.b. Cross section through the *Achillea millefolium L.* main stem from Oradea (100X): 1- epidermis; 2- assimilated parenchyma; 3- pericycle; 4- leading bundles; 5 - medular parenchyma; 6 – colenchima

Detection of metals

Metal detection was realized using Niton XRF techniques on vegetable *Achillea millefolium* when it was determinate the concentration for nine metals, representing the main components and also the essential traces and possible toxic products. The obtained results are depicted in table 1 and table 2.

Table 1

Elemental content of three samples containing vegetal product of *Achillea millefolium* of Oradea

Sample/Me (µg/g)	Mo	Zr	Sr	Ca	Fe	K	Zn	Ti	Sc
Sample 1	7.2	6.0	15.2	23900.0	75.4	49100.4	18.5	81.4	86.2
Sample 2	7.0	6.0	15.3	23875.1	75.3	49098.1	17.9	80.9	85.7
Sample 3	7.3	6.1	14.9	23903.1	75.6	49095.3	18.3	80.5	84.9

Table 2

Elemental content of three samples containing vegetal product of *Achillea millefolium* of Alesd

Sample/Me (µg/g)	Mo	Zr	Sr	Ca	Fe	K	Zn	Ti	Sc
Sample 1	6.5	7.2	13.4	33700.2	52.2	48200.6	20.4	186.7	178.2
Sample 2	6.2	7.6	13.1	33695.8	52.6	48199.2	19.9	185.9	177.9
Sample 3	6.6	7.1	13.7	33701.1	51.9	48195.9	20.5	186.3	178.1

Analyzing the data presented in table 1 and also table 2 can conclude that the most abundant metal into the studied plant is potassium. This metal is an important oligoelement for human body where present an important role for neural impulse transmission. Calcium represent an important element for plant development, his absorption is passive without any energy consumption. Calcium is assuring the mechanical resistance of tissues, and also is involved into the physiological processes (water and salt circuit through cell walls) (http://www.gazetadeagricultura.info/plante/protectia-plantelor). In human body, calcium plays an important role in blood coagulation and also into the B12 vitamin adsorption. Because of his importance into the human body development was analyzed the calcium content in all harvested samples.

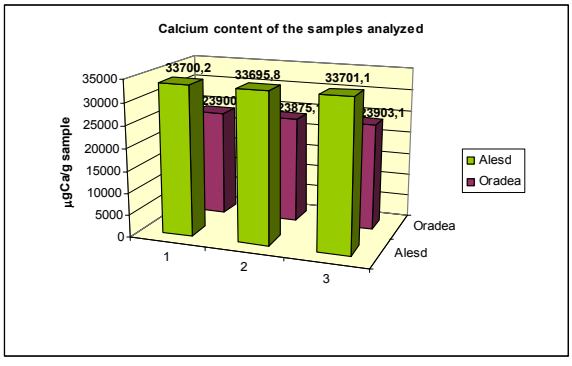

Fig. 4. The calcium content of the three samples analyzed

Another important oligoelement is zinc, which play an essential role into the human body due to his regulatory activity for the vitamin A concentration, adjusting the amount in which these vitamins are released in the liver, influencing in this way the body growth and development.The zinc concentration isn't situated within the range of non-polluting plants (15-100 mg/kg) (Senila M et al, 2006, Antal DS., 2005, Antal D.S et al 2005, Humadi S.S. et al, 2009). The zinc content in medicinal plants is very low, as compared to normative requirements for humans (4,6 mg/day) (Saper R.B. et al, 2004).

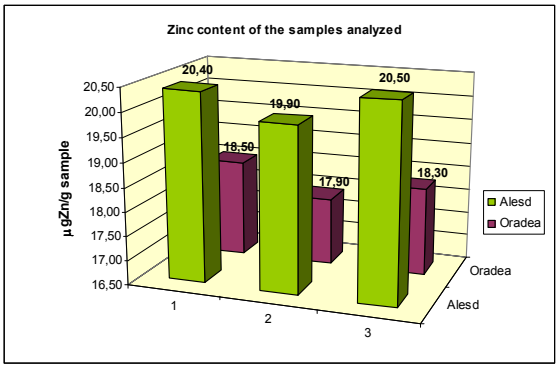

Fig. 5. The zinc content of the three samples analyzed

Another important oligoelement is iorn, which is present in all human body cells where is influencing many metabolic processes. Iron from plants has a low adsorption rate (2-3 time lower rate in comparison with iron from meat, because of that is possible that the vegetarians are not able to cover their iron need from diet. Data presented revel that the iron content is different for samples harvested from two different areas (fig.6).

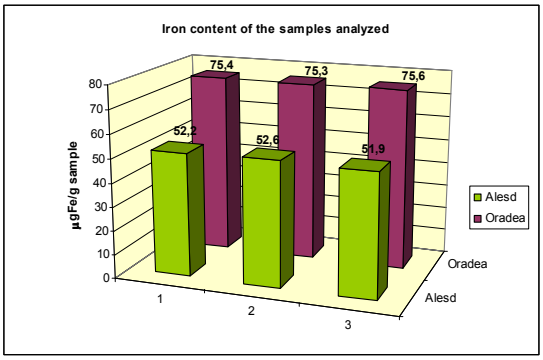

Fig. 6. The iron content of the three samples analyzed

Higher differences were observed in case of titanium and scandium, and that can be associated with the chemical composition of soils in that two different areas.

CONCLUSIONS

Today the medications and nutritional supplements factories pay a special attention to natural resources, and tend to use a large amount of local flora into the production.

In present study was analyzed the *Achillea millefolium* plant harvested from two different areas of Bihor county: three samples were harvested from a relative polluted area – Oradea town, and other three samples were harvested from some unpolluted areas – Alesd town.

From microscopic analysis can observe some differences into the plant structure. After that, was determined the metal content into the different samples.

High metallic content recommend *Achillea millefolium* as medical plant which can be used for improvement of human health.

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