

TRACE ELEMENTAL ANALYSIS OF SOME MEDICINAL HERBS TRADITIONALLY USED FOR ANTI-CANCER DISEASE BY EDXRF TECHNIQUE

S. Jyothsna*, G. Manjula and AS. Nageswara Rao

Department of Physics, Kakatiya University, Warangal-506 009, Telangana State, India.

ABSTRACT

The elemental analysis was carried out by using EX-3600 spectrometer at Kolkata Centre. A quantitative Energy Dispersive X-ray Fluorescence technique (EDXRF) is used to analyze 13 elements (P, S, Cl, K, Ca, Mn, Fe, Cu, Zn, Se, Br, Rb and Sr) in different anti-cancer medicinal plants. This present study gives the information of major and minor concentrations of trace elements in these samples located in Telangana region. These results can be compared to standard reference values of NIST 1515 apple leaf, which gives a good agreement. The benefits of plants are safe, no toxic or no minimal side effects and cheaper drugs from the plant resources those are available on earth.

Keywords: Trace elements, Medicinal plants, Cancer disease and EDXRF- Spectrometer.

1. INTRODUCTION

Cancer is a major public health burden in both developed and developing countries¹. Cancer is a disease of cells in the body, there are different types of cells in the body and many different types of cancer which arise from different types of cells. These cells are born due to imbalance in the body and by correcting this imbalance the cancer may be treated². The major causes of cancer are smoking, dietary and imbalance of hormones³. Today's global scenario indicates that Breast cancer and colorectal cancer is the most prominent cancer in the case of women and men. So cancer is not just one condition, in case it is important to know exactly what type of cancer has developed, how large it has become, whether it has spread and how well it usually respond to treatment.

Cancers affecting the digestive track are among the most common of all the cancers associated with aging². The most important preventive method in cancer treatment includes dietary changes. Several chemopreventative agents are used to treat cancer but they cause toxicity that prevents their usages⁴. Because of high death rate associated with cancer and side effects of chemotherapy and radiation therapy many

cancer patients search alternative methods for cancer treatment.

Micronutrients and trace elements play a significant role in maintaining health and preventing disease including cancer through a wide range of mechanisms like anti-oxidation, anti-proliferation and repair of DNA damage. Medicinal plants contain high amount of micro nutrients and trace elements, so they have long been used in the treatment of cancer⁵. Medicinal plants are basic raw material for many of the transfer of trace elements from soil to man. According to World Health Organization (WHO) 80% of people living in rural areas depending on medicinal plants offer a safe, cheap and reliable alternative to chemical drugs⁶. Traditionally in recent times medicinal plants occupy an important position for drug discovery⁷.

Medicinal plants contain both organic and inorganic constituents and the human body requires a number of constituents in order to maintain good health⁸⁻⁹. Most of the studies have been done important constituents of diet like essential oils, vitamins, glycosides and other organic components and little has been reported about the elemental composition of the plants¹⁰. Every constituent play an important role in the formation of these compounds¹¹ and deficiency of any constituent

may lead to abnormal development in the human body. Because of their potential on human health, medicinal plants can play an important role in healing and soothing of various diseases². So it is important to measure the trace elements and one of the prime importances to understand the effect of trace elements in medicinal plants on human health⁸. Another advantage of medicinal plants using in drugs, they can control disease without side effects¹². We have focused on some of the medicinal plants containing anti cancer properties. This could be evaluated for effective anti cancer compounds in future days. Direct and indirect relationship between micronutrients and health has been discussed in the present study.

The aim of this work principally to utilize EDXRF technique for the determination of elements in anti-cancer medicinal plants and to find a presumed correlation between their curative properties. Five Anti-cancer leaf samples of medicinal plants are chosen for this present study i.e, Catharanthous roseus, Annona squamosa, Datura metal, Vitex negundo, Calotropis procera. These medicinal plants are used locally to maintain the body immunity and herbal to heal anti-cancer disease.

2. MATERIALS AND METHODS

2.1 Sample preparation

Fresh leaf samples of 5 different medicinal plants were collected from Ramagiri khilla forest is located in 40 km away from Karimnagar district, Telangana state, India. The list of medicinal plants selected for present study, their sample code, botanical name, local name, used for representation part was shown in Table1.

Table 1: List of anti-cancer medicinal plants selected for this present study

Sample code	Botanical Name	Vernacular Name	Parts used for anti- cancer disease
CTR	<i>Catharanthous roseus</i>	Billa ganneru	Leaves
ANO	<i>Annona squamosa</i>	Seethapalam	Leaves
DT	<i>Datura metal</i>	Ummetta	Leaves
VAV	<i>Vitex negundo</i>	Tella vavili	Leaves
CAL	<i>Calotropis procera</i>	Jilledu	Leaves

CTR-Catharanthous roseus, ANO- Annona sqamosa, DT- Datura metal, VAV-Vitex negundo and CAL- Calotropis procera.

These leaves were washed in tap water and rinsed thoroughly with double distilled water in order to remove surface contamination, dried in an oven at about 60⁰C overnight (24 hours) and subsequently powdered by using agate mortar. A quantity of pure 150 grams of each powder sample was weighted and compressed using a 150 ton hydraulic press and made in to pellets of 13mm diameter and about 1mm thickness. Triplicates of each sample were done. These pellets were then used as targets for the EDXRF experiment. Biological reference material NIST 1515 (Apple leaf) used as a reference multi elemental standard.

2.2 Experimental (EDXRF) analysis

Present study reports the elemental analysis of medicinal plant samples was carried out at trace elemental laboratory, UGC-DAE CSR Kolkata center, Kolkata, India. The setup consist of Xenometric (previously Jordan valley) EX-3600, energy dispersive X-ray fluorescence (EDXRF) spectrometer, which consist of an oil cooled Rh anode X-ray tube (maximum voltage 50kV, current 1mA).The measurements were carried out in vacuum chamber using different filters (between the source and sample) for optimum detection of elements. Si (Li) detector with a resolution of 143eV at 5.9KeV and 10 samples turret enables mounting and analyzing 10 samples at a time.

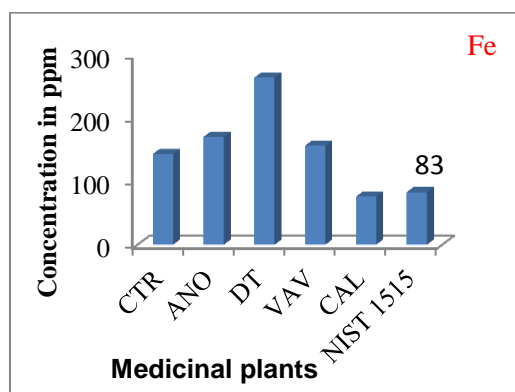
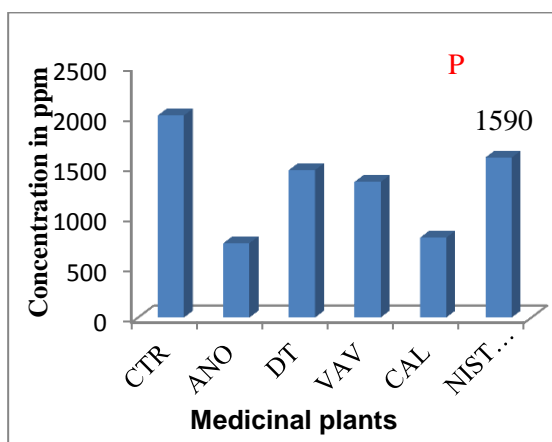
The targets were positioned at an angle of 45⁰ to the beam direction. The X-ray beam was collimated to a diameter of 4mm and was made to fall on the targets. The detector was kept at an angle of 45⁰ to the target position and at an angle of 90⁰ to the X-ray beam direction. The characteristic X-rays emitted from each sample were recorded with a high resolution Si (Li) detector which has sensitive area of 30sq mm and provided with a thin beryllium window of 8mm thickness. The spectra were collected for a sufficiently long time so that good statistical accuracies can be achieved. The quantitative analysis is carried out by using the nEXT software.

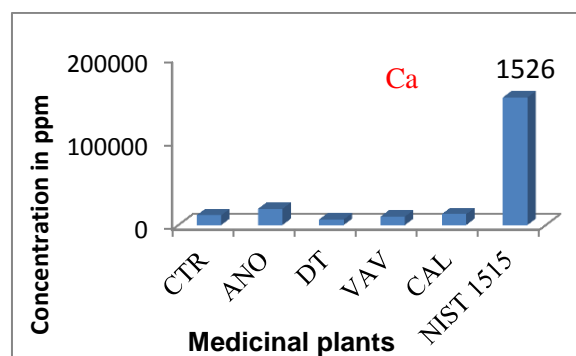
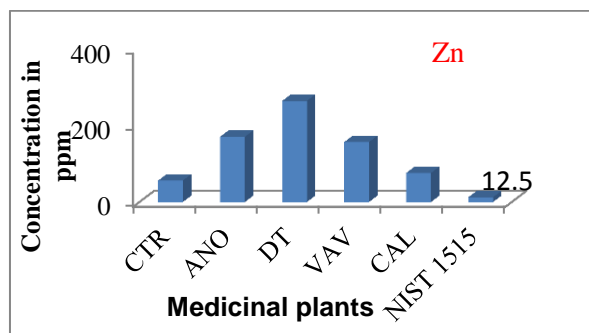
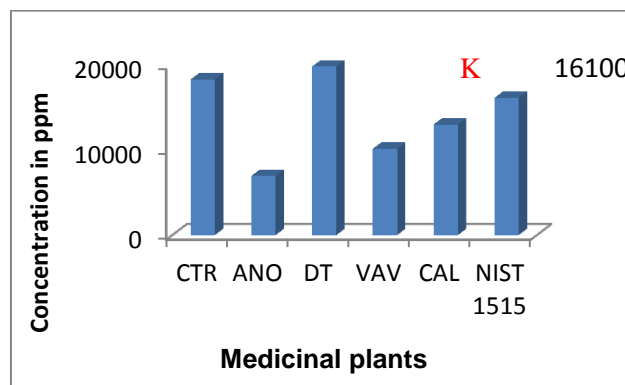
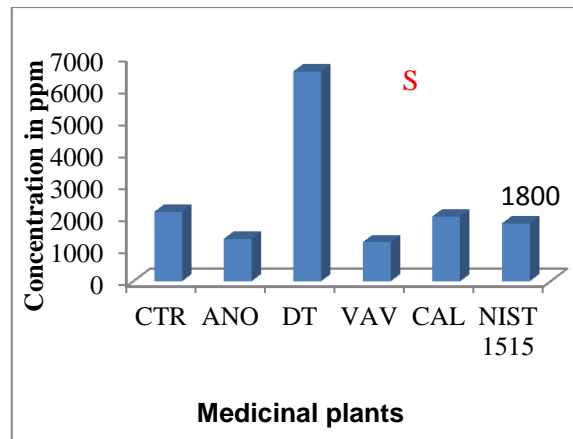
3. RESULTS AND DISCUSSION

Elemental concentrations were calculated by using EX-3600 spectrometer. Maximum, minimum and trace elemental composition were measured in these selected medicinal plants and these are compared with standard reference material of NIST1515 apple leaf.

Table 2: Average elemental concentration values of the present studied plant samples

Elements	NIST1515 Apple leaves	Elemental concentrations in ppm level otherwise mentioned				
		CTR	ANO	DT	VAV	CAL
K (%)	1.61	1.827±12.4	0.6949±5.81	1.9786±5.10	1.0133±52.32	1.298±3.4
Ca (%)	1.52	1.1743±7.69	1.936±93.3	0.6772±0.94	0.7721±6.37	0.963±2.44
P	1590	2007.17±538	737.99±161	1464.34±131	1346.07±279	792.43±77
S	1800	2161.72±263	1320.97±90	6537.72±383	1218.45±121	2017.32±124
Cl	579	4416.43±520	1694.26±168	4916.51±220	579.00	8998.65±345
Mn	54.00	47.00±1.88	14.33±1.13	24.33±0.57	17.19±0.81	45.07±2.1
Fe	83.00	143.32±143	170.47±6.44	264.03±13.2	156.58±2.04	75.91±3.0
Cu	5.64	9.47±2.41	7.44±0.48	10.60±0.42	8.67±0.32	5.36±0.6
Zn	12.50	55.95±0.47	10.91±0.96	41.16±0.86	25.27±2.13	17.8±0.7
Se	0.05	0.28±0.89	0.55±0.61	0.39±0.32	0.62±0.37	0.24±0.1
Br	1.80	4.95±0.23	17.36±0.25	14.25±0.19	0.22±0.31	180.47±15
Rb	10.20	9.48±0.08	4.61±1.25	20.23±1.7	9.44±0.54	52.74±4.3
Sr	25.00	53.16±1.40	128.04±2.29	27.76±2.2	25.43±1.38	98.59±8.3





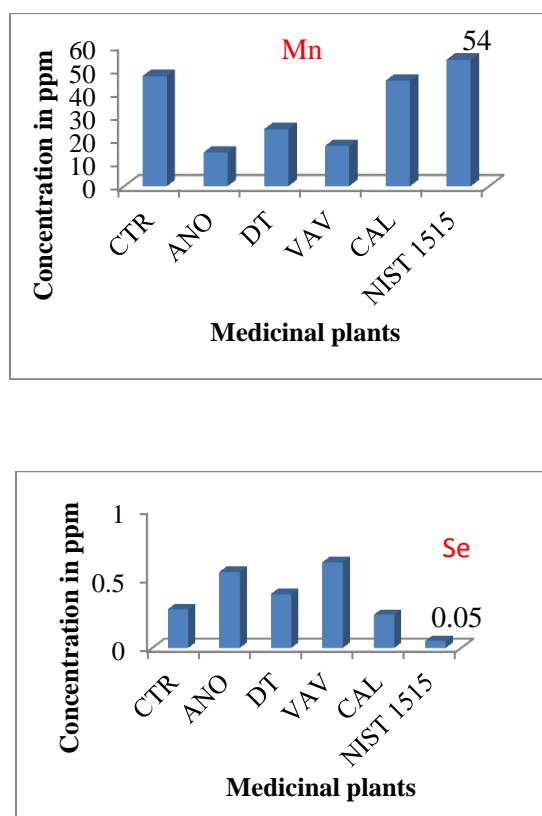


Fig. 1: Graphs of P, S, K, Ca, Fe, Zn, Mn and Se concentrations of the present studied plant samples with NIST 1515 standard values

The thirteen elements namely P, S, Cl, K, Ca, Mn, Fe, Cu, Zn, Se, Br, Rb and Sr were done in the present plant samples by using EDXRF technique. Table 2 shows the measured concentration values of the present study. The results of EDXRF measurements are among all the trace elements it is clear that potassium (K) and calcium (Ca) are the most abundant detected elements (major elements) which are quantified in percentage level whereas other elements are found to be in parts per million (ppm) levels. The essential elemental concentrations of P, S, K, Ca, Mn, Fe, Zn and Se are present in these plant samples which are compared to certified values of NIST 1515 apple leaf. This gives a wide variation in their elemental concentration values of essential trace elements shown in fig.1.

The maximum concentration of K is observed in *Datura metal* (1.978%) and minimum concentration is observed in case of *Annona squamosa* (0.6949%), which is compared certified value of NIST 1515 apple leaf (1.61%). K is an important part in regulation of water balance of the body, it is extremely important to cell and without it we could not survive¹². The concentration of Ca is high in

Annona squamosa (1.9366%) and low in *Datura metal* plant sample (0.6947%) (NIST1515 value 1.52%). Ca is essential element for maintaining healthy bones and teeth¹³. The maximum concentration of P is found in *Catharanthou roseus* (2007.17ppm) and minimum concentration is found in *Annona squamosa* (737.99ppm) (NIST value of P is 1590ppm). P works with Calcium to help build bones and is an essential mineral primary used for growth of body cells and tissues. High level of S is found in *Datura metal* (6573.72ppm) and low S is found in *Vitex negundo* (1218.45ppm) plant (NIST1515 value of S 1800ppm). S helps in protecting the cells in the body from environmental hazards like air pollution and radiation. The concentration of Cl is varies from 8998.65ppm to 579.00ppm. It is found to be maximum in the case of *Calotropis procera* (8998.65ppm) and equal amount of Cl is found in *Vitex negundo* (579ppm) plant sample (compared to NIST 1515 certified value 579ppm). Cl helps man to digest his food properly and to absorb other important elements that e need to survive¹⁴.

Mn present at minor levels in all the medicinal plants (compared to certified values of NIST

1515 apple leaf 54ppm) with their concentrations vary from 47ppm to 14.33ppm. The maximum concentration (in this minor level of concentration) of Mn is observed in *Catharanthus roseus* and minimum concentration is observed in *Annona squamosa*. Mn is an important mineral in maintaining the body immune system since it is responsible for several metabolic and enzymatic processes¹⁵. The excess of Mn can cause toxic, in brain it can cause a Parkinson type syndrome¹⁶. All medicinal plants are having high amount of Fe (Iron) except *Calotropis procera* plant sample, which are compared to certified value of NIST 1515 apple leaf 83ppm. Fe is found at minor level in the case of *Calotropis procera* (75.91ppm). Iron is found to be very high concentration in *Datura metal* (264.06ppm) and *Annona squamosa* (170.47ppm). Fe plays an important role in the formation of hemoglobin and certain enzymes. Iron deficiency can lead to anemia and one of the most serious forms of iron overload is acute Fe poisoning¹⁷.

The maximum concentration of Cu (Copper) is observed in *Datura metal* (10.60ppm) and minimum in *calotropis procera* (5.36ppm) (NIST1515 value is 5.64ppm). Cu is an important enzymes involved in a number of vital processes¹⁸. Excessive dietary of Zn can cause Cu deficiency, chronic Cu toxicity is rare in humans and mostly associated with liver damage. The concentration of Zn is found in the range of 55.95ppm to 10.91 ppm. Zn is found to be very high in *Catharanthus roseus* (55.95ppm), *Datura metal* (41.16ppm) and minimum Zn is found in *Annona squamosa* (10.91ppm) (which are compared to NIST 1515 standard values 12.50ppm). Zn is involved in the activity of about 100 enzymes example RNA polymerase and Carbonic anhydrate. It is also essential for the creation, release and use of hormones in the body. Zn deficiency is common in under developed countries and is mainly associated with malnutrition, affecting the immune system¹⁷. High concentration of Selenium (Se) is observed in *Vitex negundo* (0.62ppm), *Annona squamosa* (0.55ppm) and *Datura metal* (0.39ppm) (NIST1515 value of Se 0.05ppm). Se is used in the synthesis of ascorbic acid and helps to revive patients¹⁴. Se deficiency contributes to a form of heart disease, hypothyroidism and a weakened immune system¹⁹. It is most important element used in the treatment of cancer. Bromine (Br) concentrations vary from 180.47ppm to 0.22ppm, is present at major level in the case of *Calotropis procera* (180.47ppm) and minor level of concentration is found in *Vitex negundo* (0.22ppm) NIST1515 standard value

of Br is 1.80ppm. Br is obviously harmful to human health. The highest concentration of Rb is found in *Calotropis procera* (52.74ppm) and lowest concentration is found in *Annona squamosa* (4.61ppm) and NIST1515 apple leaf of Rb value is 10.20ppm. Rb reacts with skin moisture to form rubidium hydroxide which causes chemical burns of eye and skin. In the present study we have discussed with their maximum and minimum concentrations of selected anti cancer medicinal plant samples and also discussed their significances of the trace elements.

4. CONCLUSION

EDXRF technique has been used to analyze 13 elements namely P, S, Cl, K, Ca, Mn, Fe, Cu, Zn, Se, Br, Rb and Sr in five anti-cancer medicinal plants. The results obtained from the present study shows that there is a substantial difference of trace elemental concentration of Mn, Cu, Fe, Zn and Se of these anti-cancer medicinal plant samples. The variation in elemental concentrations is mainly attributed to the differences in botanical structure, mineral composition of the soil in which the plants are cultivated and environment effect. These trace elements contain a vital importance in mans metabolism, development, preventative and healing of anti-cancer disease. The data obtained in the present work Se, Zn and Mn is needed by the cancer patients in order to prevent and to increase the change for natural immunological remission of cancer. Our measured concentrations are almost near to the concentrations of certified NIST 1515 apple leaves except Br and Sr. The selected use of this plants may definitely in anti-antigenic therapy and thus in cancer management.

5. ACKNOWLEDGEMENT

The authors are thankful to Dr. M. Sudarshan, UGC- DAE CSR, Kolkata centre, for providing the beam line of EDXRF facility to carry out the present work.

6. REFERENCES

1. Mohammad shoeb. Bangladesh. J Pharmacol. 2006;1:35-41.
2. Madhuri S and Govind Pandey. Curr Scien. 2009;96:779-783.
3. Parkin DM, Bray F, Ferlay J and Pisani P. C A Cancer. J Clin. 2002;55(2):74-108.
4. Kathiresan K, Boopathy NS and Kavitha S. Nat Prod Rad. 2006;5:115-119.
5. Hartwell JL. Plants used against cancer: a survey. Lawrence, MA. Quarterman Publications. 1982;438-439.

6. Rajbir Kapur, Ashok Kumar, Navneet Kaur, Mohanty BP, Mumtaz Oswal, Singh KP and Gulzar Singh. *Inter. Jour of PIXE*. 2012;22:113-116.
7. Merina N, Jogen Chandra K and Jibon K. *Int Research Jour of Pharmacy*. 2012;3(6): 26-30.
8. Seetharami Reddy B, Fikre Dessalegn, Abdul Sattar Sheik. *Int Jour of Engineering Scie & Tech*. 2013;3:633-635.
9. Naga Raju GJ, Saritha P, Ramana Murthy GAV, Ravikumar M, Seetharami Reddy B, John Charles M, Lakshminarayana S, Seshi Reddy T, Bhuloka Reddy S and Vijayan V. *App Radia and Iso*. 2006;64:893-900.
10. Singh V and Garg AN. *Jour of Appl Rad Iso*. 1997;48:97-101.
11. Jawad Alzeer, Balayeshwanth R, Vummind, Rami Arafah, Waleed Rimawi, Hatem Saleem and Luedtke NW. *Jour of Medicinal Plant Research*. 2014;8(9):408-415.
12. Singh AK, Rahubanshi AS and Singh J S. *Jour Ethnopham*. 2002;81(1):31-41.
13. Hanauske U, Hanauske MH, Marshall, Muggia VA and Von Hoff DD. *Int J Cell Cloning*. 1987; 5(2):170- 178.
14. Chaturvedi UC, Shrivastava R and Upreti RK. *Curr Science*. 2004;87:1536-1554.
15. Aschner M. *Environ Health Perspect*. 2000;108:429-432.
16. Fraga CG. *Mol Aspects of Med*. 2005;26:235-244.
17. Carri MT, Ferri A, Cozzolino M, Calabrese L and Rotilio G. *Brain Res Bull*. 2000;61:365-374.
18. Lisa M Gaetke and Ching Kuang Chow. *Toxicology*. 2003;189:147-163.
19. Combs GF. *Biofactors*. 2000;12:39-43.