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Research Article

ANALYSIS OF THE ARSENIC IN COMMONLY USED MEDICINAL PLANTS

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ABSTRACT

The demand for herbal products as food supplements, food additive and traditional medicine for health care is increasing globally. There are several reports of adverse effects of these herbal preparations due to the presence of high level of heavy metals such as Lead, Cadmium, Arsenic, Mercury, Chromium, Nickel, Copper etc and this problem has become a matter of concern. The present study was done to determine the presence of Arsenic in some of the selected medicinal plants namely *Hemidesmus indicus* (L.) R.Br. *(Sariba), Cyperus rotundus* L. *(Musta), Glycyrrhiza glabra* L. *(Yashtimadhu), Rubia cordifolia* L. *(Manjishta), Eclipta alba* Hassk *(Bhringaraj), Hedychium spicatum* Ham.ex Smith *(Karchura), Emblica officinalis* Gaertn. *(Amalaki) and Acacia concinna* (Willd.) DC. *(Shikakai),* which were procured from local market of Chennai, Tirupati and Hyderabad. The samples were digested by wet digestion method and analysed by UV-Vis Spectrophotometer. The results were compared with permissible limits recommended by WHO. Mean levels were evaluated with respect to their procurement. It was observed that the analyzed plant species contained safe levels of the heavy metals concentration excepting a very few samples. There was a considerable variation of heavy metal concentration for the examined medicinal plant species. This is due to the difference in physiological properties of plant uptake.

KEY WORDS: Arsenic, Herbal drugs, UV Spectrophotometer, heavy metal concentration.

INTRODUCTION

Human endeavour over the ages has been to enhance its scientific inquisitiveness and through it present standard of life. Today's jet age is marked with much responsibility to save the planet for future generations. This has led to the formation of a binding environmental legislation that is very effective in many nations.

According to the world health organisation (WHO), traditional medicine refers to health practices, approaches, knowledge and beliefs incorporating plant, animal, and mineral-based medicines, spiritual therapies, manual techniques, and exercises, applied singularly or in combination to treat, diagnose, and prevent illnesses or to maintain well-being. If the material being used is of plant origin, then it is called traditional herbal medicine. Plant derived drugs were classified for the treatment and evaluation based on their therapeutic action from the ancient time itself. These Medicinal plants have different chemical compositions due to influence of climatic conditions, nature and properties of soil, fertilizer, pesticide, geographical distribution, age of the plant, source of collection, altitude, period of harvesting, manufacturing practices etc.[1]

Medicinal plants may be easily contaminated by absorbing heavy metals from soil, water and air. Usually soil is subjected to contamination through atmospheric deposition of heavy metals from point sources including metalliferous mining, smelting and different industrial activities. Some other sources of soil contamination involve use of fertilizers, pesticides, sewage sludge and organic manures[2]. Additional sources of these elements

for plants are rainfall, atmospheric dusts and plant protection agents, which could be absorbed through leaf blades[3].

The term heavy metal refers to any metallic chemical element that has a relatively high density and is toxic or poisonous at low concentrations. Some of the heavy metals are essential in very low concentrations for the survival of all forms of life. Heavy metals such as iron, chromium, copper, zinc, cobalt, manganese and nickel are essential metals, since they play an important role in biological systems; whereas mercury, lead, arsenic and cadmium are nonessential metals have toxic and mutagenic effects even at very low concentration. Several cases of human disease, disorders, malfunction and malformation of organs due to metal toxicity have been reported. Along with human beings, animals and plants are also affected by toxic levels of heavy metals. [4-8]

Arsenic is noticed in more than 200 different inorganic minerals, occurs frequently as trivalent (arsenite) or pentavalent (arsenate) ions, and can bind to organic material commonly present in the environment. Arsenic is highly toxic in inorganic form. It is peregrine and found in the Earth's sediment, soil, and water. In addition to being present at naturally occurring levels, arsenic is frequently found at higher concentrations due to anthropogenic contributions, including pesticides, herbicides, industrial waste, and the burning of fossil fuels[2,7]. Chronic exposure to inorganic arsenic through drinking contaminated water, using contaminated water in food preparations and irrigation of food crops, has serious health effects including vomiting, abdominal pain and diarrhoea, gastrointestinal damage, and skin and internal cancers[9].

In this present work, UV-VIS spectrophotometer is used because most of the phenolic compounds, such as flavonoids, anthroquinones, coumarins, anthocyanins, and other compounds containing conjugated double bond (s) with chromophore (s) in herbs have strong UV-Vis absorption. The use of UV-VIS spectrophotometer in determination of heavy metals in medicinal samples is becoming popular in many laboratories because it provides for easy, economical, efficient, robust simple and rapid determination in low and high concentration at cheap cost[16].

AIMS AND OBJECTIVES

The present study is concerned with the assessment of Arsenic [As] content in some of the selected medicinal plants namely *Hemidesmus indicus (Sariba), Cyperus rotundus (Musta), Glycyrrhiza, glabra (Yashtimadhu), Rubia cordifolia (Manjishta), Eclipta alba Hassk (Bhringaraj), Hedychium spicatum Ham.ex Smith (Karchura), Emblica officinalis (Amalaki) and Acacia concinna (Shikakai)* were procured from local market of Chennai, Tirupati and Hyderabad respectively.

MATERIALS AND METHODS[3, 9-15]

Chemicals

Sulphuric acid, hydrogen peroxide, nitric acid, deionised water, Arsenic powder.

Apparatus

1000 ml standard flask, 100 ml standard flask, 50 ml standard flask, Tissue papers, Whatman filter papers, beakers, hot plate, electronic weighing machine, Pipette, measuring jar.

PREPARATION OF STOCK SOLUTION

Arsenic stock solution

Dissolve 1.0g of arsenic powder in 50ml conc. nitric acid by constantly stirring the volumetric flask. Dilute to 1 litre with deionised water.

Arsenic makes arsenic acid with concentrated nitric acid, arsenious acid with dilute nitric acid.

$5\ HNO_3$ (Conc.) + As --> $H_3AsO_4 + 5\ NO_2 + H_2O$

$3\ HNO_3$ (Dilute) + As --> $H_3AsO_3 + 3\ NO_2$

This reaction is very interesting as it is a rare example of the formation of nitrogendioxide from only dilute nitric acids.

SAMPLE PREPARATION

Sample preparation for analysis of Heavy metals in medicinal plants was done according Wet digestion method (AOAC 1995) for non volatile heavy metals. Wet digestion involves the destruction of organic matter through the use of both heat and acid[3].

PROCEDURE

- Weigh accurately 1.0 g of dried sample and place in a beaker or digestion tube.
- Add 16 ml concentrated H_2SO_4 and place the beaker on hot plate and then temperature was gradually increased to 125^0C at which the sample was boiled for 1hour.

- Remove beaker and allow cooling.
- Add 4 ml H_2O_2 (30%) and digest at the same temperature. As the reaction finished another 4 ml H_2O_2 (30%) was added. The mixture was heated till the digestion is complete.
- After cooling, the content was filtered into 100 ml volumetric flask using Whatman filter paper No.41 and the solution was completed to the mark using deionized water.

Concentrated Sulphuric Acid is been used in this procedure. Hydrogen peroxide is also used to enhance reaction speed and complete digestion. Hot plates or digestion blocks are utilized to maintain temperatures of 80 to 125^{0}C. After digestion is complete and the sample is cooled and filtered into standard flask which is filled to volume and dilutions are made to meet analytical requirements.

Critical factors in wet digestion procedures include selection of the digestion vessel, temperature and its control, time, the digestion mixture, and final volume. Selection of a digestion vessel is dependent on the elements of interest and the heat source. Time and temperature are interrelated and are dependent on the digestion mixture.

Wet digestion procedures generally require greater analyst supervision and intervention than dry procedures. Wet digestion is recommended for plant materials.

Instrumentation: Agilent Cary 60 UV-Vis

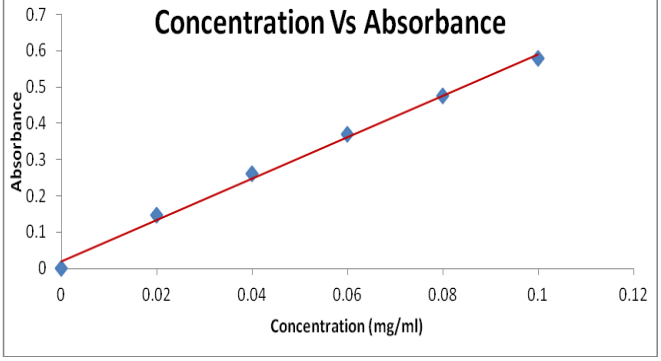
5.1. ARSENIC		
Maximum Wavelength = 300 nm		
S.No	Concentration	Absorbance
1	0	0
2	0.02	0.1472
3	0.04	0.261
4	0.06	0.3691
5	0.08	0.4755
6	0.1	0.578

Spectrophotometer

The Agilent Cary 60 UV-Vis spectrophotometer is efficient, accurate and flexible, and is designed to meet both current and future measurement needs. The proven, robust design of the Cary 60 comprises a double beam, Czerny-Turner monochromator, 190–1100 nm wavelength range, 1.5 nm fixed spectral bandwidth, full spectrum Xenon pulse lamp single source with exceptionally long life, dual silicon diode detectors, quartz overcoated optics, scan rates up to 24, 000 nm/min, 80 data points/sec maximum measurement rate, non-measurement phase stepping wavelength drive, room light immunity, central control by PC with Microsoft® Windows® operating system. Supported by GLP software, optional 21 CFR Part 11 capable software, and dedicated instrument validation software which includes pharmacopeia test suites.

OBSERVATIONS AND RESULTS

Sample is prepared using Wet Digestion method.

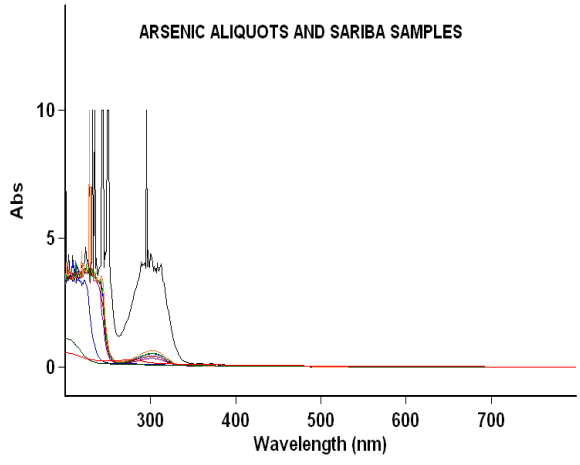


Calibration eqn	Abs = 5.68957 * Conc. +0.02067
Correlation Coefficient	0.99588

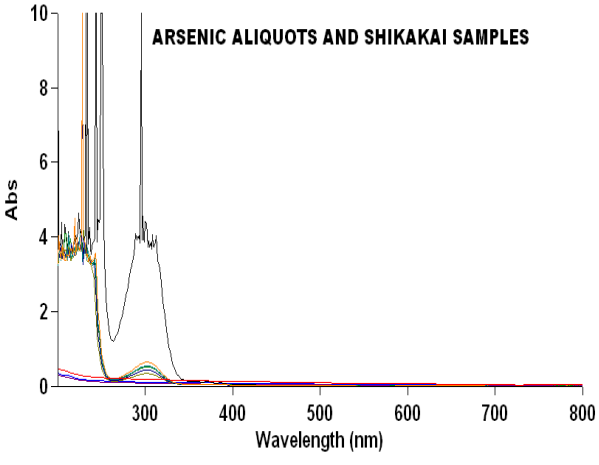
Arsenic was analysed at a maximum wavelength of 300 nm and at different conc. (0, 0.02, 0.04, 0.06, 0.08 and 0.1) and the corresponding absorbance was obtained. A graph is plotted between concentration and absorbance is called Calibration Curve. Based on this graph, the concentration of Arsenic in various samples was identified.

Table 2: Concentration of Arsenic in Various Samples

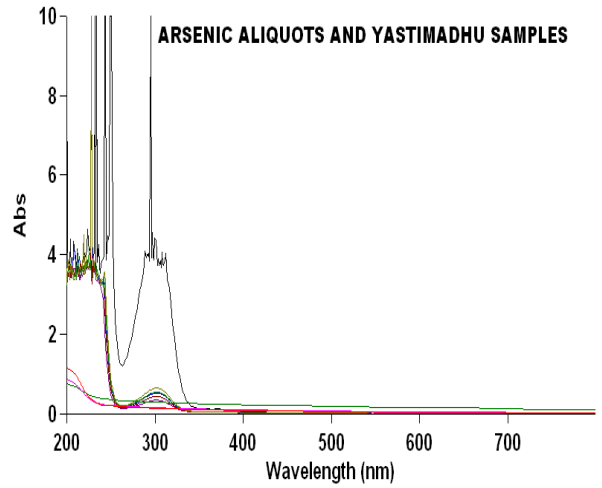
ARSENIC					
Name of the Sample		**Absorbance**	**Concentration (mg/ml)**	**Concentration (ppm)**	**Concentration (mg/kg)**
SARIBA	Chennai sample	0.0323	0.002	2	1.3783
	Tirupati sample	0.0222	0	0	0.0000
	Hyderabad sample	0.0205	0	0	0.0000
MUSTA	Chennai sample	0.0454	0.004	4	2.7108
	Tirupati sample	0.0203	0	0	0.0000
	Hyderabad sample	0.0328	0.002	2	1.3356
YASTIMADHU	Chennai sample	0.0611	0.007	7	4.6941
	Tirupati sample	0.0282	0.001	1	0.7371
	Hyderabad sample	0.0229	0	0	0.0000
KARCHURA	Chennai sample	0.0281	0.001	1	0.5714
	Tirupati sample	0.0433	0.004	4	2.5279
	Hyderabad sample	0.0415	0.003	3	1.7676
MANJISHTA	Chennai sample	0.0445	0.004	4	2.3392
	Tirupati sample	0.0654	0.008	8	5.7971
	Hyderabad sample	0.0515	0.005	5	3.0581
BHRINGARAJ	Chennai sample	0.0145	0	0	0.0000
	Tirupati sample	0.0156	0	0	0.0000
	Hyderabad sample	0.0146	0	0	0.0000
AMLA	Chennai sample	0.017	0	0	0.0000
	Tirupati sample	0.0232	0	0	0.0000
	Hyderabad sample	0.022	0	0	0.0000
SHIKAKAI	Chennai sample	0.0295	0.001	1	0.6173
	Tirupati sample	0.0222	0	0	0.0000
	Hyderabad sample	0.0205	0	0	0.0000



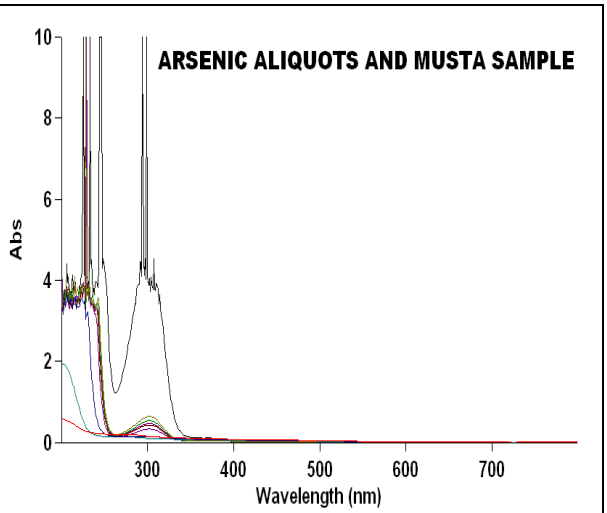
Sariba



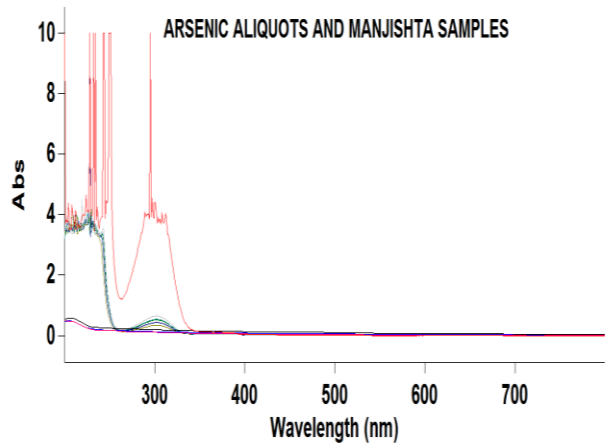
Shikakai



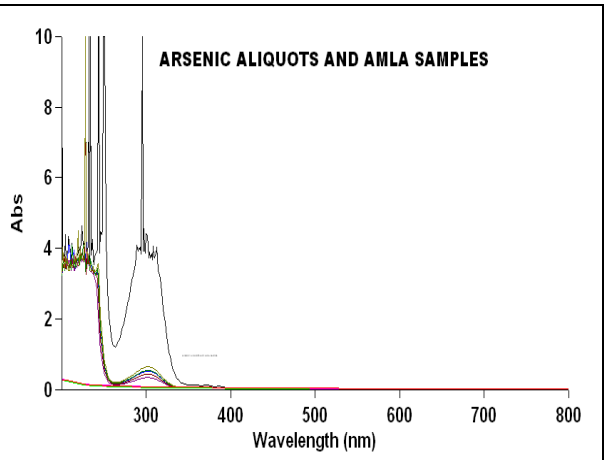
Yashtimadhu



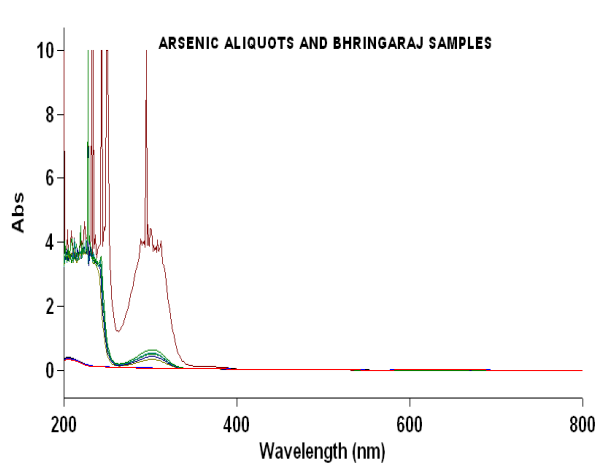
Musta



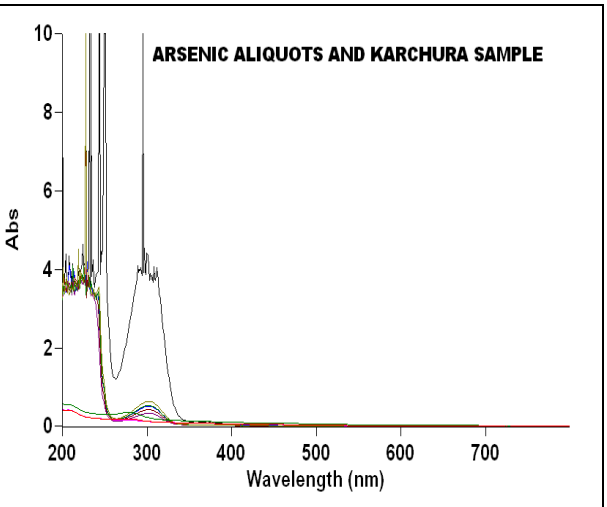
Manjishta



Amalakai



Bhringaraj



Karchura

RESULTS AND DISCUSSION

From the Observation it was found that, the conc. of Arsenic in the Sariba Chennai sample is 2 ppm (1.3783 mg/Kg) whereas Sariba Tirupati sample and Hyderabad sample did not show the trace of Arsenic.

In the Musta samples, Chennai sample has 4 ppm (2.7108 mg/Kg) conc. of Arsenic and Hyderabad sample has 2 ppm (1.3356 mg/Kg) conc. of Arsenic whereas Hyderabad sample has no trace of Arsenic.

In the Yastimadhu samples, Chennai sample has 7 ppm (4.6941 mg/Kg) conc. of Arsenic, Tirupati sample has 1 ppm (0.7371 mg/Kg) conc. of Arsenic and Hyderabad sample has no trace of Arsenic.

Out of the three samples of Karchura, the conc. of Arsenic in the Chennai sample is 1 ppm (0.5714 mg/Kg), Tirupati sample is 4 ppm (2.5279 mg/kg) and Hyderabad sample has 3 ppm (1.7676 mg/kg).

In the Manjishta samples, the conc. of Arsenic in the Chennai sample is 4 ppm (2.3392 mg/Kg), Tirupati sample has 8 ppm (5.7971 mg/kg) and Hyderabad sample has 5 ppm (3.0581 mg/kg).

Bhringaraj and Amalaki samples did not show any traces of Arsenic. In the Shikakai samples, Tirupati sample and Hyderabad sample did not show the traces of Arsenic whereas Chennai sample contained 1 ppm (0.617 mg/kg) of Arsenic.

The results of the present analysis showed that the levels of Arsenic in all samples were 0-8 ppm (0-5.7971 mg/Kg) with a mean of 1.75 ppm, which is much lower than the acceptable limit (5 ppm) recommended by World Health Organization (WHO). The Highest concentration occurred in Manjishta Tirupati sample has 8 ppm (5.7971 mg/kg). It was observed that Amalaki and Bhringaraj samples have not shown any traces of the Arsenic. Results reveal that the contents of Arsenic in some samples like Yastimadhu Chennai sample and Manjishta Tirupati sample are slightly higher than the acceptable safe limit for the body. The elevated level of Arsenic may lead to the Arsenic toxicity and potential health hazards for the consumers. No samples of Sariba, Musta, Karchura, Bhringraj, Amalaki and Shikakai contain Arsenic above allowable limit recommended by WHO.

CONCLUSION

From the above study it can be concluded that the analyzed plant species contained safe levels of the heavy metals concentration excepting a very few samples. There was a considerable variation of heavy metal concentration for the examined medicinal plant species collected from three local markets of Chennai, Tirupati and Hyderabad. This is due to the difference in physiological properties of plant uptake.

It is therefore suggested that awareness of this phenomenon should be disseminated to prevent collecting medicinal herbs from non-cultivated, polluted areas and other sources, which are prone to heavy metal pollution. The analysis of heavy metals is highly essential for raw drugs used for the preparation of compound formulations. The periodic assessment is essential for quality assurance and safer use of herbal drugs.

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