Journal of Applied Pharmaceutical Science Vol. 3 (04), pp. 054-058, April, 2013
Available online at http://www.japsonline.com
DOI: 10.7324/JAPS.2013.3409
ISSN 2231-3354

Pollution Based Study of Heavy Metals in Medicinal plants *Aloe vera* and *Tamarix aphylla*

Hamid Iqbal[1], Baharullah Khattak[1*], Sultan Ayaz[2], Ali Rehman[4], Muhammad Ishfaq[1], Muhammad Naseer Abbas[1], Muhammad Saqib Malik[1], Abdul Wahab[3], Imran[4], Saifullah Mehsud[5]
[1]Department of Microbiology, Kohat University of Science and Technology, Pakistan.
[2]Department of Zoology, Kohat University of Science and Technology, Pakistan.
[3]Department of Pharmacy, Kohat University of Science and Technology, Pakistan.
[4]Department of Chemistry, Kohat University of Science and Technology, Pakistan.
[5]Department of Pharmacy, Hazara University, Mansehra, Pakistan.

ARTICLE INFO

Article history:
Received on: 07/01/2013
Revised on: 25/01/2013
Accepted on: 05/02/2013
Available online: 27/04/2013

Key words:
Aloe vera, *Tamarix aphylla*, Heavy Metals, Atomic Absorption Spectrophotometer.

ABSTRACT

The main objective of this study was to generate perception in community regarding pollution of heavy metals contained in *Aloe vera* and *Tamarix aphylla* plants which have wide range of medicinal use but having toxic concentration of heavy metals. The plant samples were collected at three different areas referred as spots i.e. polluted (spot 1), less polluted (spot 2) and non- polluted (spot 3) from District Kohat, Khyber Pakhtunkhwa, Pakistan. In the present study, essential heavy metals such as Cobalt (Co), Nickel (Ni), Chromium (Cr), Copper (Cu), Iron (Fe) and Zinc (Zn) and non-essential heavy metals Cadmium (Cd), Lead (Pb) were analyzed in *Aloe vera* leaves and *Tamarix aphylla* by using Flame atomic absorption spectrophotometer. Heavy metals concentration in different spots were found to be in order as Spot 1> Spot 2>Spot 3 with the concentration range of found heavy metals., Cd 0.25-0.51 mg/kg, Pb BDL-15.23, Co 1.90-3.31mg/kg, Ni 2.98-4.01 mg/kg, Cr 4.86-6.01 mg/kg, Cu 2.32-3.01 mg/kg, Fe 12.42-22.47 mg/kg, Zn 34.53-53.08 mg/kg for *Aloe vera* leaves and concentrations of the same heavy metals like Cd 0.13-0.31 mg/kg, Pb 1.00-18.01 mg/kg, Co 0.25-2.90 mg/kg , Ni 3.75-5.93 mg/kg , Cr 3.83-5.32 mg/kg , Cu 1.01-1.90 mg/kg, Fe 23.65-30.10 mg/kg , Zn 13.70-25.63 mg/kg were observed in the bark of *Tamarix aphylla*. Thus it was concluded from the present study that the samples collected from polluted area were found more contaminated with heavy metals as compared to less polluted and non-polluted area.

INTRODUCTION

Growing tendencies and increase in demand for food safety has drawn the attention of researchers to the risk factors associated with consumption of contaminated foodstuffs i.e. pesticides, heavy metals and or toxins etc. All plants contain heavy metals in trace or negligible amount as micronutrients and when the uptake of heavy metals become greater, they contaminate the environment thus leads to adulterate food supply (Abdollatif *et al*., 2009; Khair., 2009). These metals have the potential to accumulate in different organs of the body for a long period of time as some of them possess longer biological half-lives and cause toxicities and unwanted effects. (Jarup., 2003; Ata *et al*., 2009). The selection of medicinal plants like *Aloe vera* and *Tamarix aphylla* is based on the fact that these plants were not previously screened for such objectives.

Many preliminary studies on the use of *Aloe vera leaf* and *Tamarix aphylla* have been reported and proved to have important medicinal role for centuries. *Aloe vera* (family: *Liliaceae*) is a xerophytic medicinal plant and also grows even in rain fed condition. Weiner and Weiner (1994) reported a number of biological activities such as antiseptic (saponins and anthraquinones), antitumoral (mucopolysaccharides), anti-inflammatory (steroids and salicylic acid), anti-oxidant (Vitamins) and immune regulator (glucomannans). It is used in various pharmaceutical product, food and cosmetic industries due to its metabolites. Scientific studies provide the evidence that *Aloe vera* extracts have wide spread uses in health products as a skin salve including topical healing of burns, epithelial injuries and treatment of wounds, skin abrasions, minor infections, an ulcer remedy, and an adjuvant cancer treatment due to its immune modulating effects and canker sores, sebaceous cyst, diabetes, and elevated blood lipids in humans Boudreau and Beland, (2006) and antileishmanial activity (Hamid *et al*., 2012).

* *Corresponding Author*
Email: dr.baharullah@kust.edu.pk,

These positive effects are thought to be due to the presence of compounds such as polysaccharides, anthraquinones and lectins *Aloe vera* is now widely used on face tissues, where it is promoted as a moisturized and/or anti-irritant to reduce chafing of the nose of users suffering hay-fever or cold.

Tamarix aphylla (family Tamaracaceae) is an ethno medicinal plant that is widely used by the local practitioners in Pakistan. This plant is widely available in different parts of the country with having antifungal activity and no side effects (Marwat *et al*., 2009). The bark of *Tamarix aphylla* is an astringent and tonic and is commonly used for the treatment of hepatitis, eczema and skin diseases like capititis, syphilis and scaly skin conditions (Panhwar & Abro, 2007). Its bark powder is also used by the local people as a poultice on minor wounds.

MATERIALS AND METHODS

Collection of plant material

Fresh plants samples of *Aloe vera* leaf and *Tamarix aphylla* bark were collected randomly from District Kohat, (Khyber Pakhtunkhwa) Pakistan and authenticated by the herbarium staff of botany department, Kohat University of Science and Technology and were kept in the sterile environment of the laboratory for further process. After collection, the desired plants were first cleaned from extra weeds and seeds and washed with distilled water; air-dried and were chopped and ground into fine powder and passed through sieved of 0.5mm mesh screen and kept separately in clean polythene bags for further processing.

Analysis of Plant Samples

Crucibles rinsed with acid and distilled water were taken and kept in furnace at 600^{o}C for 1hr and weight was noted. Weight of empty crucibles was noted. Weighed quantity of grinded powdered plant samples (2gm) was taken from each sample and was kept in oven at 105^{0}C for 1 hour and then in desiccator to remove moisture. The sample was ashed for about 5hrs until a grey or white ash residue was obtained. Then the moisture free sample was placed in furnace. The combined weight (crucible and crude powder) was noted and these crucibles were kept in furnace at 550 at 550^{0}C for 4 hrs and were kept in desiccators for 40min. The weight was noted again. The contents of china dish were cooled to room temperature in desiccators and 2.5 mL of 6M HNO_3 solution was added into crucibles and when necessary, the mixture was heated to dissolve its content. The solution was filtered through whatman filter paper#42 and diluted to the mark of 25mL flask.

The solutions were stored in clean and dry plastic bottles (Ikhtiar *et al*., 2008) and analyzed for the elements of interest like Cd, Pb, Co, Fe, Ni, Zn, Cu and Cr utilizing Flame Atomic Absorption Spectrophotometer (Parkin elmer 400) with suitable hollow cathode lamps.

RESULTS AND DISCUSSION

In the present study, elemental analysis of heavy metals such as Cadmium (Cd), Lead (Pb), Cobalt (Co), Nickel (Ni), Chromium(Cr), Copper (Cu), Iron (Fe) and Zinc (Zn) were determined in *Aloe vera* leaves and *Tamarix aphylla* bark. In order to ascertain accumulation of the above mentioned heavy metals, investigations were performed by using atomic absorption spectrometry. The permissible concentration of heavy metals makes raw plants admissible to the production of medicines (Gorbanova and Gorbanov, 2004).

Cadmium is highly toxic for the human bio-system, taken up from the soil, water, fertilizers, pesticides treatment and anthropogenic operations even at very low levels of intake (Hunt, 2003). Analysis of heavy metals in the plant samples *Aloe vera* and *Tamarix aphylla* shows that cadmium was found in the least concentration among all the detected elements as shown in table 1. In all the three selected spots, the cadmium was found to be in the range of 0.13-0.51 mg/kg. In general high concentration of cadmium was noted in plant samples collected from Spot 1 as compared to Spot 2 and Spot 3. High concentration of Cadmium 0.51 mg/kg was found in *Aloe vera* collected from spot 1 followed by *Aloe vera* of spot 2, 0.46 mg/kg and 0.31 mg/kg in *Tamarix aphylla* of spot 1 while lower concentration 0.13 mg/kg was found in *Tamarix aphylla* of spot 3. Maximum permissible limit (MPL) for Cadmium (Cd) recommended by World Health Organisation (WHO) is 0.3 mg/kg (WHO, 1998) while the daily dietary intake (DDI) is 0.057 mg. The results show that *Tamarix aphylla is* resistant to heavy metals uptake as compared to *Aloe vera.*

It is revealed from the figure 2 that Lead (Pb) concentration was below the detection limit (BDL) in *Aloe vera* of spot 3. As the table shows that almost negligible amount of lead was found in *Aloe vera* and *Tamarix aphylla* collected from spot 3 while the concentrations reached a toxic level in spot 1.

High concentrations of lead was found in spot 1, *Aloe vera* 15.23mg/kg and *Tamarix aphylla* 18.01mg/kg followed *Aloe vera* 9.09mg/kg and *Tamarix aphylla* 8.43 mg/kg of spot 2. Maximum permissible limit (MPL) for Lead (Pb) recommended by World Health Organization (WHO) is 10 mg/kg (WHO, 1998)

Table. 1: Summarizes the concentration levels of heavy metals in *Aloe vera* leaves and bark of *Tamarix aphylla.*

Spot	Plant Name	Cd	Pb	Co	Ni	Cr	Cu	Fe	Zn
Spot 1	***Aloe vera***	0.51±0.02	15.23±1.01	3.31±0.09	4.01±0.02	6.01±0.28	2.32±0.11	22.47±1.98	34.53±1.32
	Tamarix aphylla	0.31±0.00	18.01±0.41	2.90±0.11	5.93±0.09	5.32±0.43	1.01±0.18	30.10±1.43	13.70±1.11
Spot 2	***Aloe vera***	0.46±0.01	9.09±0.02	2.74±0.32	3.89±0.02	4.00±0.46	4.67±0.10	35.97±1.31	33.41±1.21
	Tamarix aphylla	0.23±0.01	8.43±0.09	3.89±0.21	4.65±0.01	4.20±0.65	1.76±0.21	25.62±0.11	40.34±1.24
Spot 3	***Aloe vera***	0.25±0.00	BDL	1.90±0.08	2.98±0.21	4.86±0.31	3.01±0.22	12.42±0.10	53.08±1.53
	Tamarix aphylla	0.13±0.01	1.00±0.00	0.25±0.01	3.75±0.12	3.83±0.23	1.90±0.31	23.65±1.89	25.63±1.24

Spot 1-polluted area, Spot 2 - less polluted area, Spot 3 - non-polluted area.
BDL indicates below detection limit, ± shows Standard deviation.

while the daily dietary intake (DDI) is 0.415 mg (Gorbanova and Gorbanov, 2004) while concentration exceeding this, limit leads to paralysis and causes anemia, abdominal pain (Goyer 1988, Davies, 1990).

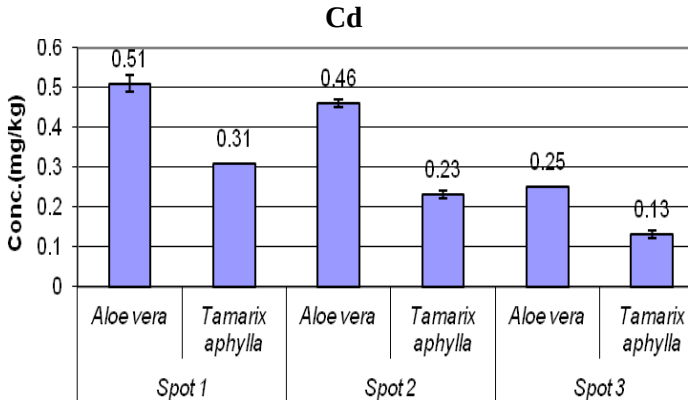

Fig. 1: Concentration of Cadmium in *Aloe vera* and *Tamarix aphylla*.

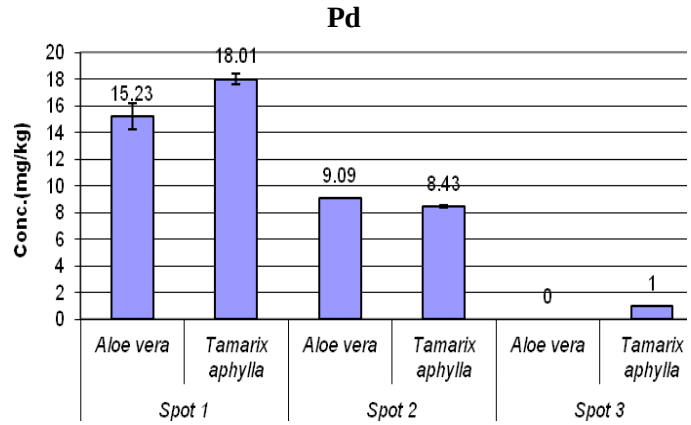

Fig. 2: Concentration of Lead in *Aloe vera* and *Tamarix aphylla.*

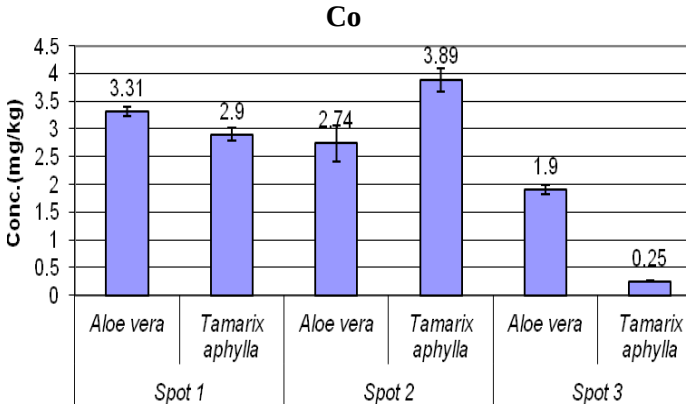

Fig. 3: Concentration of Cobalt in *Aloe vera* and *Tamarix aphylla.*

It is evident from the data (Table 1) that cobalt concentration was found in the range of 0.25-3.89 mg/kg. It is shown by the figure 3 that high concentration of cobalt 3.89mg/kg was found in *Tamarix aphylla* spot 2 followed by Aloe vera 3.31 mg/kg of spot 1 while lowest concentration 0.25 mg/kg was found in *Tamarix aphylla* of spot 3. Maximum permissible limit (MPL) for Cobalt (Co) recommended by World Health Organization (WHO) is 0.2-0.3 mg/kg (WHO, 1998) while the daily dietary intake (DDI) is 0.04 mg. Although cobalt is toxic at elevated concentration, however the body needs only in trace amount. High intake of cobalt causes vomiting, nausea, vision and heart problems and also damage of thyroid (Bethesda 1993, Smith 1990).

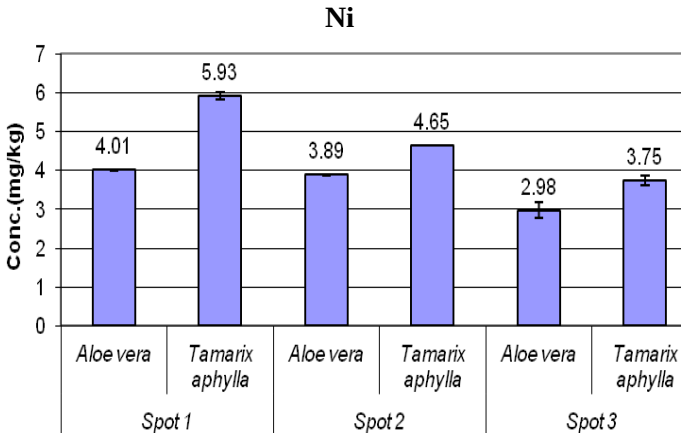

Fig. 4: Concentration of Nickel in *Aloe vera* and *Tamarix aphylla.*

The present study indicates that Nickel concentration in *Aloe vera* and *Tamrix aphylla* was found beyond the acceptable limits i.e 2.98-5.93 mg/kg as shown in figure 4. In general, high concentration of Nickel than the permissible limit was observed in all spots. Aloe vera shows nearly equal concentration in spot 1 (4.01 mg/kg) and spot 2 (3.89 mg/kg) while low concentration was observed in spot 3 having a concentration of 2.98 mg/kg while a considerable maximum and varied concentrations were found in *Tamarix aphylla* in spot 1 (5.93 mg/kg) followed by spot 2 (4.65 mg/kg) and 3.75 mg/kg in spot 3. Environmental Protection Agency (EPA) has recommended less than 1 mg/day beyond which is toxic (WHO, 1998) while the daily dietary intake (DDI) is less than 1 mg (Shad *et al*., 2008). Increased concentration of nickel leads to kidney damage and disorders of liver (McGrath and Smith, 1990).

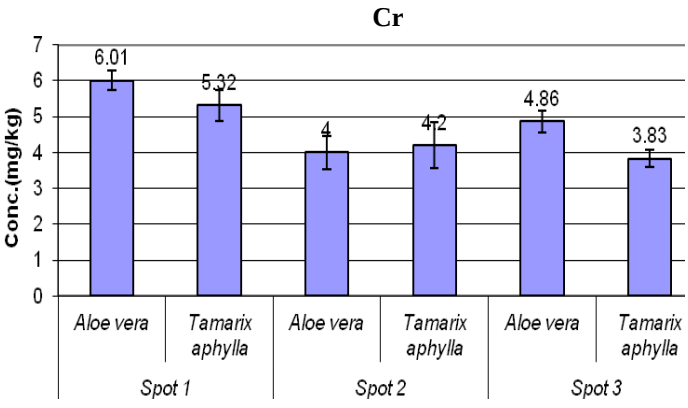

Fig: 5 Concentration of Chromium in *Aloe vera* and *Tamarix aphylla.*

It is revealed from the table that chromium concentration was found in the range of 3.83-6.01 mg/kg in *Aloe vera* and *Tamarax aphylla*. *Aloe vera* contains maximum chromium concentration in spot 1 as it is 6.01 mg/kg followed by spot 3 (4.86 mg/kg) and minimum in spot 2 (4 mg/kg) as shown in figure 5.

Similar observations with less quantity in spot 1 (5.32 mg/kg) and spot 3 (3.83 mg/kg) were observed in *Tamarix aphylla* as compared to *Aloe vera* however *Tamarix aphylla* showed a reasonable greater quantity in spot 2 (4.2 mg/kg) as compared to *Aloe vera*. Maximum permissible limit (MPL) for Chromium (Cr) recommended by World Health Organization (WHO) is 1.5 mg/kg while the dietary intake is 0.2 mg/day (WHO, 1998). An elevated concentration between 5-30 mg/kg can lead to yield reduction in plants (Kabata-Pendias and Pendias, 1992) and bleeding tendencies, ulcer, GI upset, lack of immune system (US Health Services, 2000).

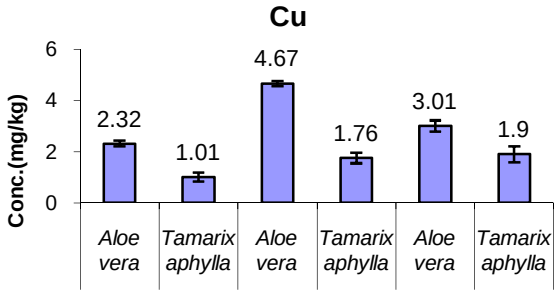

Fig. 6: Concentration of Copper in *Aloe vera* and *Tamarix aphylla*.

The copper concentration was found as 2.32-4.67 mg/kg in *Aleo vera* and concentration of 1.01-1.9 mg/kg was recorded in *Tamarix aphylla* in all the three spots. Figure 6 reveals that copper concentration was found less in spot 1 (2.32 mg/kg), eventually high in spot 3 (3.01 mg/kg) and most in spot 2 (4.67 mg/kg) whereas *Tamarix aphylla* shows variable concentration 1.9 mg/kg in spot 3 followed by spot 2 (1.76 mg/kg) and found less in spot 1 (1.01 mg/kg). Critical concentration for copper in plants is in between 20-100 mg/kg. Maximum permissible limit (MPL) for Copper (Cu) recommended by World Health Organization (WHO) is 10mg/kg (WHO, 1998) while the daily dietary intake (DDI) is 2-3 mg (Gupta, 1975). High levels of copper may cause metal fumes fever with flue like symptoms, hair and skin discoloration, dermatitis, irritation of the nasal mucosa and nausea (Shad *et al*., 2008).

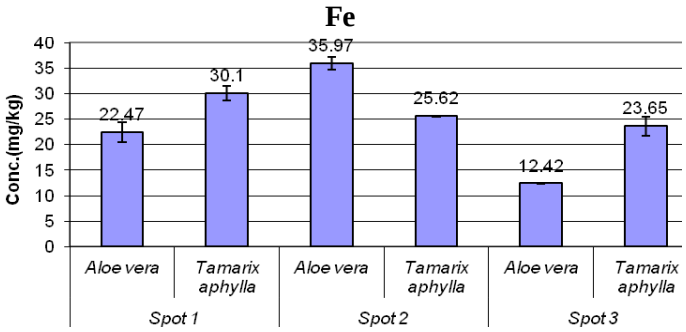

Fig. 7: Concentration of Iron in *Aloe vera* and *Tamarix aphylla*.

After Zinc, Iron was the most detected element with a concentration range of 12.42-35.97 mg/kg in *Aloe vera* while 23.65-30.1 mg/kg concentration of iron was noted in *Tamarix aphylla*. Data shows that Iron in *Aloe vera* was considerable in the normal range in all spots. i.e 35.97 mg/kg in spot 2, followed by spot 1 i.e 22.47 mg/kg and low concentration (12.42 mg/kg) was observed in spot 3 whereas *Tamarix aphylla* shows a maximum concentration (30.1 mg/kg) in spot 1, then in spot 2 (25.62 mg/kg) and low in spot 3 (23.56 mg/kg). Maximum permissible limit (MPL) for Iron (Fe) recommended by World Health Organization (WHO) is 20mg/kg (WHO, 1998). The dietary limit of Fe in the food is 10-60 mg/day (Ali *et al.*, 2011)**.** Iron deficiency in plants produces chlorosis, however its high concentration also affects plant growth (Shad *et al*., 2008).

It is shown by experiment (figure 8) that among all the heavy metals, high concentration of zinc was found in the plant samples of *Aloe vera* 33.41-53.08 mg/kg and 13.7-40.34 mg/kg in *Tamarix aphylla*. *Aloe vera* shows a maximum concentration in spot 3 (53.08 mg/kg) followed by spot 1 i.e. 34.53 mg/kg and low in spot 2 (33.41 mg/kg) whereas *Tamarix aphylla* was found low in spot 1 (13.7 mg/kg), considerably high in spot 3 (25.63 mg/kg) and maximum in spot 2 (40.34 mg/kg). Maximum permissible limit (MPL) for Zinc (Zn) recommended by World Health Organization (WHO) is 50 mg/kg while the dietary intake (DDI) is 11mg/day. Exceed limits of Zn causes many complications and has adverse effects on brain development and dopaminergic system of the body.

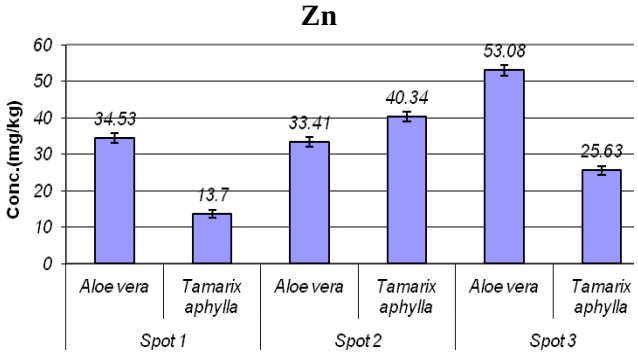

Fig. 8: Concentration of Zinc in *Aloe vera* and *Tamarix aphylla*.

CONCLUSION

It is clear from our investigations that the plant samples collected from polluted area are more contaminated with heavy metals as compared to less polluted and non polluted areas. So each medicinal plant should be thoroughly analyzed for heavy metals used in the preparation of herbal products and standardized extracts before processing for pharmaceutical purposes.

ACKNOWLEDGEMENTS

The authors are grateful and wish to express thank to the Chairman Department of Microbiology, Kohat University of Science and Technology and all the faculty members for their gratitude and making the facilities available for the research.

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How to cite this article:

Hamid Iqbal, Baharullah Khattak, Sultan Ayaz, Ali Rehman, Muhammad Ishfaq, Muhammad Naseer Abbas, Muhammad Saqib Malik, Abdul Wahab, Imran, Saifullah Mehsud., Pollution Based Study of Heavy Metals in Medicinal plants *Aloe vera* and *Tamarix aphylla*. J App Pharm Sci, 2013; 3 (04): 054-058.