Analytical Investigation of Selected Inorganic Nutrients of Desert Growing *Aloes*

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Summary: The use of *Aloe vera* as a family medicine is being promoted for a large variety of conditions. Often general practitioners seem to know little about its inorganic constituents for the treatment of certain diseases. Keeping this in view, the levels of pH, moisture contents, organic contents, soluble salts, Na^+, K^+, Ca^{2+}, Mg^{2+}, Cl^-, CO_3^{2-}, SO_4^{2-} and PO_4^{3-} in both soil and plant samples were studied and a strong positive correlation was observed for certain pairs of nutrients. The results were expressed on X ± SD basis and showed varied levels of selected nutrients in plant and soil samples. In case of soils the observed order in mg kg^{-1} was $> SO_4^{2-} > CO_3^{2-} > Na^+ > K^+ > Ca^{2+} > Cl^- > Mg^{2+} > PO_4^{3-}$ and in case of plant the order was $> CO_3^{2-} > SO_4^{2-} > Na^+ > K^+ > Ca^{2+} > PO_4^{3-} > Cl^- > Mg^{2+}$. This base line data generated will be helpful for the medicinal uses of this plant.

Introduction

Aloes are one of the oldest medicinal plants known to mankind. Ancient Egyptians used it in their beauty regime and for embalming. Ancient Greeks used it to heal numerous internal and external ailments. Ancient Chinese doctors considered *Aloes* to be one of the plants with major therapeutic properties and called it the 'harmonic remedy'. American Indians ate the *Aloes* as a remedy for a number of internal disorders. The plant is known as 'the silent healer'. The Russians call *Aloes* the 'elixir of longevity'. *Aloe* has been marketed as a remedy for coughs, wounds, ulcers, gastritis, diabetes, cancer, headaches, arthritis, immune-system deficiencies and many other conditions [1-3].

Aloe vera is a cactus-like plant (a member of the lily-onion family), which grows in hot fertile regions and has been used for centuries for its healing and health giving properties. Knowledge of the extraordinary healing power of the *Aloe vera* plant has been passed down through the centuries and there is an increasing amount of evidence that *Aloe vera* can help with numerous ailments, both internal and external. Human uses of Aloes are primarily as a herbal remedy in alternative medicines and 'home first aid'. Both the translucent inner pulp as well as the resinous yellow exudate from wounding the Aloe plant are used externally to relieve skin discomforts [4] and internally as purgatives [5]. *Aloe*'s benefits include ingesting aloe juice to lower blood sugar levels in diabetes patients [6, 7]. Aloe as a strong laxative, may have some anti-cancer effects on humans [8] and is even being tested to treat asthma [9].

Aloe vera contains over seventy-five nutrients and twenty minerals, nineteen amino acids including all eight essential amino acids and eleven secondary amino acids as well and twelve vitamins. These vitamins include: A, B1, B6, B12, C and E. It has even been referred to as 'a pharmacy in a plant'. Aloes also contain anthraquinone gycosides, resins, polysaccharides, sterols, gelonins and chromones. It is also a source of a class of chemicals called Aloins [1,2].

In recent years, *Aloe vera* and related products have drawn great attention from cosmetic [10], nutroceutical and pharmaceutical industries [11]. Aloes produce positive medicinal benefits for healing damaged skin [12]. Drinks made from or containing chunks of aloe pulp are popular in Asia as commercial beverages and as a tea additive. *Aloe vera* gel, which is generated in the mucilaginous cells

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of the inter central zone of the leaf, has been very well documented for its wound-healing [13, 14], anti-inflammatory [15, 16] and immune-stimulatory [17] activities. The pH consistently falls in the range 4.4 to 4.7. Total solids content of Aloe gel is 0.66 % and soluble solids are 0.56 % with some seasonal fluctuation [18]. Aloe's clear gel contains amino acids, minerals, vitamins, enzymes, proteins, polysaccharides and biological stimulators [18]. Decades of scientific studies on Aloe chromones reveal hundreds of structures with anti-inflammation [19], anti-ulcer [20], tyrosinase inhibition [21], skin protection [22], laxative effect [23] and other biological activities. Analyses of aloe chromones in different aloe species also have been reported [24-32]. No research has been conducted for inorganic nutrients of *Aloe vera* plant growing in desert soil environment. The present research will discuss and document some medicinally important inorganic cations and anions especially in desert growing environment of the plant. The information generated in this research will aid the Aloe industry in strengthening their products.

Results and Discussion

Medicinal herbs used in indigenous medicines in crude forms for the control of various diseases contain both the organic and inorganic constituents. Plants take inorganic nutrients from the soil for their metabolic activities. Some of the inorganic constituents are essential in trace amounts and play important role in plant and animal nutrition, enzymatic reactions and metabolic processes. The availability of heavy metals depends upon soil properties such as pH, electrical conductivity, lime content, organic matter and texture. Therefore, their correlation studies are of utmost importance.

Average level of Na^+ (Table-1) in soil of Cholistan desert is 8.34 ± 2.79 mg kg^{-1} with the range 5.65-11.22 mg kg^{-1} and is lower than found in plant *i.e.* 31.26 ± 15.02 mg kg^{-1} with the range 13.98-41.23 mg kg^{-1}. Sodium is the major component of the cation of extra cellular fluid. 139 meq/ litre of sodium is present in blood plasma of human beings and controls the osmotic pressure including water retention. High concentration of sodium leads to hypertension [33-36].

Levels of K^+ in *Aloe vera* are 16.30 ± 4.41 mg kg^{-1} with the range of 12.87-21.28 mg kg^{-1}, which are quite higher than the levels found in periphery soil *i.e.* 5.61 ± 1.80 mg kg^{-1} with the range 4.23-7.65 mg kg^{-1}. This means that nature has designed plant cells in such a fashion that they have a tendency to pick up less K^+ than Na^+. K^+ is the major component of the cation of intracellular fluid. 5 meq/ litre of K^+ are present in blood plasma of human beings and control the osmotic pressure including water retention. High concentration of K^+ leads to the dilation of arteries and normalizes the blood pressure. Excessive amount of K^+ leads to the failure of heart [33-36].

Concentration of Ca^{2+} in soil samples (Table-1) has average of 1.38 ± 0.65 mg kg^{-1} with the range 0.96-2.12 mg kg^{-1} and is lower than that found in *Aloe vera* plant *i.e.* 2.88 ± 0.41 mg kg^{-1} with the range 2.45-3.27 mg kg^{-1}. The Ca^{2+} is used in the synthesis of new plant cell walls. A protein found in Cytosol, to form calcium-calmodulin complex might be involved in regulation of many metabolic processes. 5 meq/ liter of calcium are present in blood plasma of humans [33-36].

Table-1: Comparative levels of nutritive inorganic elements studied in soil and *Aloe vera* plant.

Sample Code	Soil/ Plant Samples	pH	Water contents %	Organic contents %	Soluble salts %	Na^+ (mg kg^{-1})	K^+ (mg kg^{-1})	Ca^{2+} (mg kg^{-1})	Mg^{2+} (mg kg^{-1})	Cl^- (mg kg^{-1})	CO_3^{2-} (mg kg^{-1})	SO_4^{2-} (mg kg^{-1})	PO_4^{3-} (mg kg^{-1})
A	Soil	8.21	1.56	4.56	21.28	8.14	4.96	1.05	0.46	0.63	129.15	96.53	0.13
	Plant	–	–	–	–	41.23	21.28	2.45	1.32	1.24	81.96	79.26	0.29
B	Soil	8.25	2.18	4.59	33.37	5.65	7.65	0.96	0.34	0.75	93.87	148.82	0.15
	Plant	–	–	–	–	38.56	14.76	3.27	1.61	1.49	90.18	15.84	0.28
C	Soil	8.87	1.57	3.96	22.69	11.22	4.23	2.12	0.29	1.16	87.94	187.25	0.19
	Plant	–	–	–	–	13.98	12.87	2.92	0.63	1.53	45.65	57.67	4.91
*Soil	Range	8.21-8.87	1.56-2.18	3.96-4.59	21.28-33.37	5.65-11.22	4.23-7.65	0.96-2.12	0.29-0.46	0.63-1.16	87.94-129.15	96.53-187.25	0.13-0.19
	Average	8.44	1.77	4.37	25.78	8.34	5.61	1.38	0.36	0.85	103.65	144.20	0.16
	SD	± 0.37	± 0.36	± 0.36	± 6.61	± 2.79	± 1.80	± 0.65	± 0.09	± 0.28	± 22.28	± 45.54	± 0.03
*Plant	Range	–	–	–	–	13.98-41.23	12.87-21.28	2.45-3.27	0.63-1.61	1.24-1.53	45.65-90.18	15.84-79.26	0.28-4.91
	Average	–	–	–	–	31.26	16.30	2.88	1.19	1.42	72.60	50.92	1.83
	SD	–	–	–	–	± 15.02	± 4.41	± 0.41	± 0.50	± 0.16	± 23.70	± 32.24	± 2.67

*Range, average and standard deviation of samples collected from three different areas of Cholistan desert. Sample of each site is the average of three samples.

Mean level of Mg^{2+} in soil is 0.36 ± 0.09 mg kg^{-1} with the range 0.29-0.46 mg kg^{-1} and is lower in concentration than the plant *i.e.* 1.19 ± 0.50 mg kg^{-1} with the range of 0.63-1.61 mg kg^{-1}. Mg^{2+} is almost never limiting to the plant growth in soils. In plant cells, Mg^{2+} has a specific role in the activation of enzymes involved in respiration, photosynthesis and synthesis of DNA and RNA. Mg^{2+} is also a part of the porphyrin component of the chlorophyll. It combines with ATP and allows it to function [33-36].

Higher chloride levels are found in plant (1.42 ± 0.16mg kg^{-1} with the range 1.24-1.53 mg kg^{-1}) than the adjoining soil *i.e.* 0.85 ± 0.28 mg kg^{-1} with the range 0.63-1.16 mg kg^{-1}. Abnormalities of Na^+ metabolism are generally accompanied by abnormallities in Cl^- metabolism because concentration of Na^+ and Cl^- is equal in blood plasma of humans. Chlorides are also essential in water balance, osmotic pressure regulation as well as acid base equilibrium. Mostly it may be required for cell division in leaves and roots of the plants [33-36].

Levels of CO_3^{2-} observed in soil are 103.65 ± 22.28 mg kg^{-1} with the range of 87.94-129.15 mg kg^{-1} and are found higher than their levels in *Aloe vera* plant *i.e.* 72.60 ± 23.70 mg kg^{-1} with the range 45.65-90.18 mg kg^{-1}. Carbonic anhydrase, an enzyme found in mammalian tissues, produces and uses protons and bicarbonate ions and plays a key role in the regulation of pH and fluid balance in different parts of human body. The transport of the protons and bicarbonate ions produced in kidney and eyes influence the water content of the cells at these locations. Absence or malfunction of this enzyme can lead to diseased states, ranging from the loss of acid production in the stomach to kidney failure [33-36].

Concentration of SO_4^{2-} is higher in soil *i.e.* 144.20 ± 45.54 mg kg^{-1} with the range 96.53-187.25 mg kg^{-1} than the observed levels in *Aloe vera* plant *i.e.* 50.92 ± 32.24 mg kg^{-1} with the range 15.84-79.26 mg kg^{-1}. Sulphur in plants, absorbed as divalent sulphate ions (SO_4^{2-}), is found in proteins especially in amino acids cystein and methionine that are building blocks of proteins. It is an essential component of vitamins, biotin and co-enzyme [33-36].

PO_4^{3-} levels in *Aloe vera* plant *i.e.* 1.83 ± 2.67 mg kg^{-1} with the range 0.28-4.91 mg kg^{-1}, are found higher than observed in adjoining soil *i.e.* 0.16 ± 0.03 mg kg^{-1} with the range 0.13-0.19 mg kg^{-1}. Phosphorus, as phosphate, is an integral component of a number of important compounds present in plant cells. It acts as limiting element in soils and is absorbed primarily as monovalent phosphate anion ($H_2PO_4^-$). The ability of these two forms of ions is controlled by soil pH *i.e.* monovalent is favoured by pH below 7 and divalent form by pH above 7. Phosphorus remains as free phosphate or bound to organic compounds as esters in plants [33-36].

The linear correlation between pairs of selected nutritive parameters studied in soils adjacent to the roots of *Aloe vera* plant is shown in Table-2. Soil pH has strong positive correlations for Na^+, Ca^{2+}, Cl^-, SO_4^{2-} and PO_4^{3-}. Water contents are positively correlated with organic contents, soluble salts and K^+ levels. Organic contents are also positively correlated with K^+, Mg^{2+} and CO_3^{2-}. Soluble salts show positive correlation with K^+ rather than Na^+. Sodium has positive correlations with Ca^{2+}, Cl^-, SO_4^{2-} and PO_4^{3-}. Calcium is positively correlated with Cl^-, SO_4^{2-} and PO_4^{3-} and magnesium with CO_3^{2-}. Chloride has strong positive correlation with both SO_4^{2-} and PO_4^{3-} and

Table-2: Linear correlation coefficient matrix for selected nutritive inorganic parameters studied in soil adjacent to *Aloe vera* plant roots (n = 3).

	pH	Water contents	Organic contents	Soluble salts	Sodium	Potassium	Calcium	Magnesium	Chloride	Carbonate	Sulphate
Water contents	-0.440										
Organic contents	-0.995	0.524									
Soluble salts	-0.355	0.996	0.443								
Sodium	0.870	-0.826	-0.913	-0.770							
Potassium	-0.624	0.976	0.696	0.952	-0.928						
Calcium	0.992	-0.547	-1.000	-0.468	0.924	-0.716					
Magnesium	-0.763	-0.245	0.697	-0.334	-0.344	-0.029	-0.677				
Chloride	0.987	-0.288	-0.966	-0.198	0.777	-0.488	0.959	-0.858			
Carbonate	-0.653	-0.393	0.577	-0.477	-0.193	-0.185	-0.554	0.988	-0.767		
Sulphate	0.849	0.102	-0.794	0.194	0.477	-0.116	0.777	-0.989	0.923	-0.955	
Phosphate	0.961	-0.175	-0.930	-0.083	0.700	-0.384	0.920	-0.912	0.993	-0.836	0.962

Values>0.3 or < -0.3 are significant at P< 0.01.

Table-3: Linear correlation coefficient matrix for selected nutritive inorganic parameters studied in *Aloe vera* plant (n = 3).

	Sodium	Potassium	Calcium	Magnesium	Chloride	Carbonate	Sulphate
Potassium	0.737						
Calcium	-0.172	-0.793					
Magnesium	0.928	0.433	0.206				
Chloride	-0.674	-0.996	0.844	-0.351			
Carbonate	0.966	0.536	0.090	0.993	-0.459		
Sulphate	-0.093	0.605	-0.965	-0.457	-0.672	-0.349	
Phosphate	-0.996	-0.672	0.082	-0.958	0.605	-0.985	0.183

Values>0.3 or < -0.3 are significant at P< 0.01.

SO_4^{2-} also has strong positive correlation with PO_4^{3-} [37].

Linear correlation coefficient matrix for selected nutritive parameters studied in *Aloe vera* plant is given in Table-3. Sodium has strong positive correlations with K^+ (r = 0.737), Mg^{2+} (r = 0.928) and CO_3^{2-} (r = 0.966). Potassium is strongly correlated with Mg^{2+} (r = 0.433), CO_3^{2-} (r = 0.536) and SO_4^{2-} (r = 0.605). Other strong positive correlated pairs in *Aloe vera* plant are Ca^{2+}-Cl^- (r = 0.844), Mg^{2+}-CO_3^{2-} (r = 0.993) and Cl^--PO_4^{3-} (r = 0.605).

Tables-2 and 3 show differences in correlation pairs for *Aloe vera* plant and adjoining soils indicating the selectivity of soil nutrient absorption by the plant cell. This correlation data is very useful to find out the medicinal uses of the plant. However, this needs proper source identification in order to know which sources contribute to a significant correlation of these parameters in plants and adjacent soils [38].

Experimental

During this study, 6 samples of *Aloe vera* plant and adjoining soil near the plant roots were collected during September 1999 to January 2000 from three different areas of Cholistan desert as given in Table-4. The samples were then analysed for different cations and anions like Na^+, K^+, Ca^{2+}, Mg^{2+}, Cl^-, CO_3^{2-}, SO_4^{2-} and PO_4^{3-} using different laboratory methods [33].

Table-4: Plant sampling location of Cholistan desert.

Sample code	Sampling Areas
A	Plant and soil samples collected near Cholistan institute of desert studies, Baghdad-ul-Jadeed Campus, Islamia University, Bahawalpur (Rural).
B	Plant and soil samples collected from Abbasia Campus, Islamia University, Bahawalpur (Urban).
C	Plant and soil samples collected from Ahmadpur East (Urban).

Chemical Reagents

All the reagents and chemicals used were of AR grade procured from E. Merck and BDH. All the solutions of standards and samples were prepared in freshly prepared deionised water.

Plant Sample Preparation

The plants were cleaned visually to remove the dust particles and dried at 150°C to a constant weight. The dried plants were ground to fine powder and then used for dry ashing. The pre-cleaned silica crucible was heated at 600°C to a constant weight. The powdered plant material in the crucible was heated in a muffle furnace at 600°C until there was no evolution of smoke. The crucible containing plant ash was cooled at room temperature and moistened with deionised water to keep it overnight. The undissolved particles were filtered and volume made up to 1000 ml. This solution was used as sample solution [36].

Soil Sample Preparation

20 grams of dried soil was taken in a 500 ml beaker, stirred it with 100 ml deionised water for 30 minutes and filtered through 'Whatman 42' filter paper. The volume of the filtrate was made up to 1000 ml and was used for quantitative estimation of Na^+, K^+, Ca^{2+}, Mg^{2+}, Cl^-, CO_3^{2-}, SO_4^{2-} and PO_4^{3-} [36].

Methods

Sodium and potassium were determined by flame photometer model 'Corning – 40'. Calcium and magnesium were determined by complexometric titration with EDTA. Carbonates were determined by titrimetric method and chlorides were by the standard argentometric method using potassium chromate as an indicator. Sulphates were estimated gravimetrically. Phosphates were determined by calorimetric method using 'ammonium dihydrogen phosphate' as standard solution and 'molybdate' as complexing agent. [38, 39]

Statistical Analysis

The data was statistically analysed by using *SPSS-12* and *STATSICA* (StatSoft 1999) softwares on P-IV system. The concentrations of elements in soil and plant samples were correlated by linear correlation coefficient matrix (Pearson).

Conclusions

Sophisticated analytical techniques of the present day have given a new impetus to the use of plants in medical research. Active ingredients from plants, individually or in combination with different medicines, are generating new interest in modem drug therapies and their combination with synthetics has increased. Plant derived organic, ionic minerals naturally chelate or remove the excess inorganic minerals from the body and don't build up to toxic levels in the soft tissue. They accelerate the healing process of chronic illness as well. This data will help in establishing baseline levels for the use of the plant as medicine.

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