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Mineral Composition of Medicinal Plants Traditionally Used in the Management of Sickle Cell Disease in the Kwilu Province (Congo-Kinshasa)

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Abstract

The aim of this study was to determine the mineral composition of 10 plants especially *Alchornea cordifolia, Alternanthera bettzickiana, Annona senegalensis, Cyttaranthus congolensis, Dissotis brazzae, Justicia secunda, Harungana madagascariensis, Hura crepitans, Hypoxis angustifolia and Vigna unguiculata* used in the management of sickle cell disease by traditional practitioners in Kwilu Province. The Plants collection was done at Kwilu province, sample were evaluated through fluorescence spectrometric analysis. These plants were harvested in Kwilu province in the Democratic Republic of Congo The mineral composition analysis was carried out using the fluorescence spectrometric method. This study revealed presence of Twenty-three (23) mineral elements, in each of these plants among others: Potassium (K), Phosphorus (P), Calcium (Ca), Sodium (Na), Magnesium (Mg), Sulphur (S), Chlorine (Cl) and trace elements such as: Aluminum (Al), Silicon (Si), Vanadium (V), Chromium (Cr), Manganese (Mn), Iron (Fe), Nickel (Ni), Copper (Cu), Zinc (Zn), Selenium (Se), Bromine (Br), Molybdenum (Mo), Tin (Sn), Iodine (I), Barium (Ba) and Lead (Pb). Iron, Zinc (Zn), Selenium (Se), Copper (Cu), Calcium (Ca), Magnesium (Mg) and Manganese (Mn) are mineral elements that are related to sickle cell disease. Of all these elements, Potassium and Calcium were in a higher

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content while Lead, Tin, Bromine, Copper and Nickel are in trace amounts. The correlation coefficients obtained between the mineral elements are significant. The presence of some mineral elements like Fe, Zn, mg and Se can be useful for sickle cell disease patients.

Keywords: Folk medicine; Botanicals; Nutrients; Anemia; Democratic Republic of the Congo.

1. Introduction

Worldwide, medicinal plants represent an important source of health products [1]. Traditional medicines are widely used due to population growth and the inaccessibility to modern medicines [2]. According to the World Health Organization, 80% of populations depend on traditional medicine for their primary health care [3]. In the Democratic Republic of the Congo, medicinal plants represent the key product for both urban and rural populations are increasingly turning to the use of medicinal plants to solve their major health problems [4]. Sickle cell disease (SCD) is an inherited disease that affects nearly 2% of the world's population. It is due to a mutation of the beta globin gene of hemoglobin and is characterized by presence of Hemoglobin S that can polymerize and to severe anemia [2-5], a loss of mineral elements that are important for the functioning of the body [6]. Minerals are natural chemical elements that the body uses to activate certain biochemical reactions. They are part of functionally important inorganic compounds such as iron (Fe) in hemoglobin and Cytochrome or zinc (Zn) in insulin [7, 8]. There is also evidence that leaves are potential sources of minerals and vitamins and are apparently inexpensive [9, 10]. In order to scientifically validate the phyto-therapeutic richness of the DRC, ten plants used in Congolese pharmacopoeia for the management of sickle cell disease in the province of Kwilu were selected.

The general objective of this study was to determine the mineral composition of these plants used in traditional Congolese medicine for the management of sickle cell disease.

2. Material and Methods

2.1. Medicinal Plants

The leaves bark and seeds of these plants used in this study were collected September 2019 and April 2020 in the five Territory of the Province of Kwilu, Democratic Republic of Congo. These samples were dried in the dark at the Basic Science Laboratory of the University of Kikwit. They were then crushed to obtain a fine powder.

2.2. Methods

The detection and quantification of the mineral elements was done by the fluorescence spectrometric method. This method of analysis allows the determination of several elements in the same sample. A quantity of the powder was pressed into pellets through the hydraulic press for each plant and the resulting pellets were fed into the fluorescence spectrometer for reading. The analysis of the results was carried out through Excel. R Studio and SPSS software packages were used for the statistical analyses using Principal Component Analysis (PCA) and Pearson Correlation [11].

3. Results and Discussion

As can be seen in Table 1, the plants studied contain seven macro-elements, in particular K, P, Ca, Na, Mg, S and Cl. The Ca, S and Cl contents are higher in *Hura crepitans* bark than in other plants. The Ca and Cl content are low in *Harungana madagascariensis* leaves that of S are low in the leaves of *Dissotis brazzae*. It was also found that the Mg content is higher in the *Justicia secunda* leaves; it is low in *Harungana madagascariensis* leaves. The Na content is high in the leaves of *Dissotis brazzae*, it is low in *Alchornea cordifolia* leaves, *Alternanthera bettzickiana*, *Justicia secunda*, *Cyttaranthus congolensis*, and in *Hypoxis angustifolia* bulbs.

The mineral composition of *Hura crepitans* bark was previously determined by Oyeleke, *et al.* [12]. Comparing the results of this research with those obtained by these authors, it can be seen that the calcium, potassium and magnesium content obtained by Oyeleke, *et al.* [12] was much lower than that obtained in this work. But the values of calcium and potassium concentrations obtained in this work are of the same order of magnitude as those of *O. basilicum* [10].

Potassium and sodium are electrolytes necessary for the normal functioning of the body and help maintain the volume of fluid and blood in the body. Ca makes up a large part of bone, human blood and extracellular fluid and is necessary for normal heart muscle function, blood clotting and regulation of cell permeability. In humans, Mg is needed in plasma and extracellular fluid, where it helps to maintain osmotic balance. Magnesium may also play an important role as an enzyme cofactor, reducing the number of abnormal erythrocytes in sickle cell disease and improving red blood cell hydration [13].

In addition to macroelements, trace elements have also been identified and quantified in the plants studied. Of all these trace elements, Al has a high content in the leaves of *A. bettzickiana*, while its low content was found in the leaves of *A. senegalensis*. The iron (Fe) content is higher in the bark of *H. crepitans* while it is low in the leaves of *H. madagascariensis*. The content of Selenium (Se) is the lowest in almost all plants studied. The iron content being the highest in the bark of *H. crepitans*, this could prove the use of the macerated extract of the bark of this plant by the traditional practitioners of Kwilu to increase the level of hemoglobin in most cases of sickle cell disease. Indeed, iron is important in the production of hemoglobin. A lack of iron causes the body to produce fewer, smaller red blood cells, resulting in anemia[14].

Comparing the results of this research with those obtained by other authors [15] in the bark of *H. crepitans* bark, it appears that the content of the same elements identified by this team was lower than that obtained in this study. But the numerical values of the contents are of the same order of magnitude as those of the other plants [16, 17]. This difference may be due to the composition of the mineral elements in the soil where the different samples were taken.

The presence of other important trace elements such as Zn, Cu, I... can be noticed. In fact, zinc is a catalyst for many enzymes necessary for the production of red blood cells; therefore, zinc deficiency can be associated with anemia. Deficient iron absorption can be caused by a decrease in trace elements such as zinc, which is found in the structure of enzymes that coordinate or catalyze iron metabolism. Copper is also a component of many enzyme systems such as cytochrome oxidase, lysyl oxidase and ceruloplasmin, an enzyme that oxidizes iron in the blood. The observation of anemia in Cu deficiency can probably be related to its role in facilitating iron absorption and the incorporation of iron into hemoglobin [16, 17].

3.1. Principal Component Analysis (PCA)

The results obtained for this analysis are as follows:

Description of Plan 1:2

Figure-1a. Plot of Individuals (plants) (PCA)

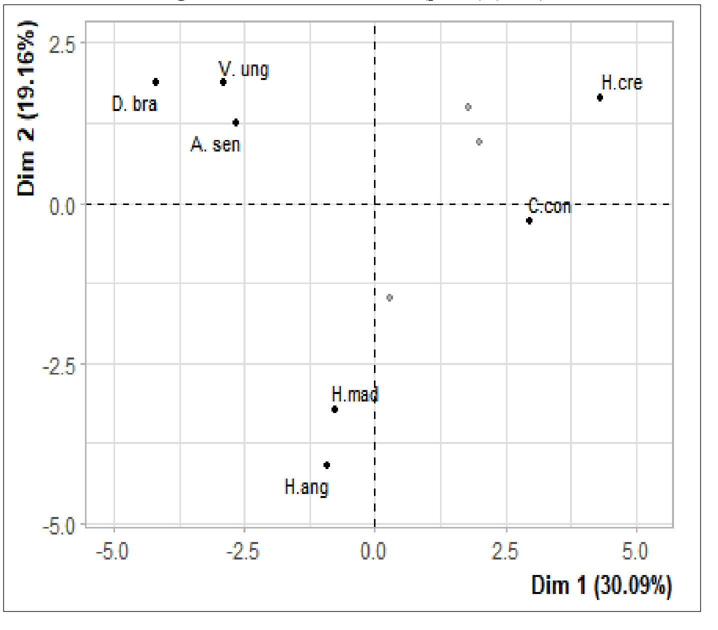
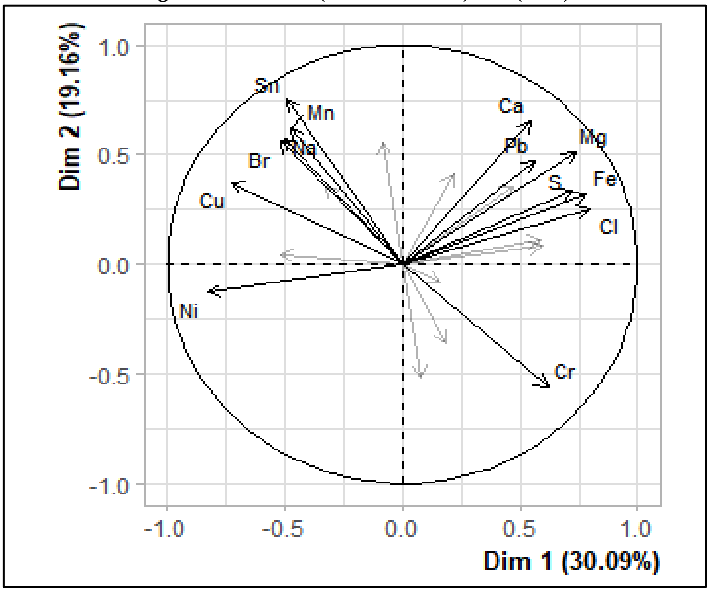


Figure-1b. Variable (mineral element) Plot (PCA)



The variables labelled are those best represented in the plan. Dimension 1 pits individuals such as H.cre and C.con (to the right of the graph, characterized by a strongly positive coordinate on the axis) against individuals such as D. bra, A. sen and V. ung (to the left of the graph, characterized by a strongly negative coordinate on the axis). The group to which individuals H.cre and C.con belong (characterized by a positive coordinate on the axis) shares:

- High values for some variables (mineral elements) such as: Mg, Fe, Cl, Pb and K (from the most extreme to the least extreme).

- Low values for other variables such as: Ni.

The group to which the individuals (plants) *D. bra*, *A. sen* and *V. ung* belong (characterized by a negative coordinate on the axis) shares:

- High values for some variables (mineral elements) like: Sn, Na, Cu and Br (from most extreme to least extreme).

- Low values for the variable Cr. Dimension 2 opposes individuals such as *D. bra*, *A. sen* and V. ung (top of the graph, characterized by a strongly positive coordinate on the axis) to individuals such as H.ang and H.mad (bottom of the graph, characterized by a strongly negative coordinate on the axis). The group to which individuals *D. bra*, *A. sen* and *V. ung* belongs (characterized by a positive coordinate on the axis) shares:

- High values for some variables (mineral elements) such as: Sn, Na, Cu and Br (from most extreme to least extreme).

- Low values for variable Cr.

The group to which the individuals *H.ang* and *H.mad* belong (characterized by a negative coordinate on the axis) shares:

- Low values for variable Sn.

From the graph of the variables, important information is given on the angle formed between two mineral elements. If the angle is less than 90°, the correlation between these two elements is positive whereas if this angle is greater than 90°, the correlation between these two elements is negative.

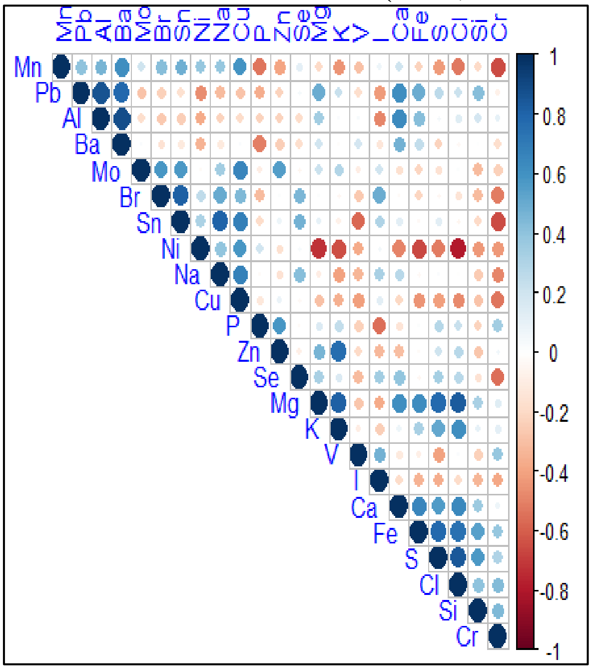
Ultimately, the higher the element content, the more the element is placed to the right of the axis (dimension 2). Fe, Mg, Cu are mineral elements related to sickle cell disease.

1) *Correlation*

A. *Correlation between mineral elements*

The figure 2 shows the correlation obtained between the mineral elements from the R software.

Figure-2. Correlation between mineral elements (Pearson, R studio software)



The analysis in this table presents two classes of correlations taking into account the p-value obtained.

The first class regroups together the elements that are correlated with a p-value between 0.01 and 0.05. The second class is that of highly correlated elements with a p-value less than 0.01. Apart from the negative correlations obtained between Manganese (Mn) and Chromium (Cr), Nickel (Ni) and Magnesium (Mg), Nickel (Ni) and Iron (Fe), Nickel (Ni) and Potassium (K), Nickel and Chlorine (Cl), Tin (Sn) and Cr; the other correlations are positive. This means that all mineral elements (variables) with positive correlations vary on average in the same direction and those with negative correlations vary in the opposite direction.

Starting from the first class, it is noticed that Mn is negatively correlated with Cr, Ni is negatively correlated with Mg, K and Fe. Fe is positively correlated with Calcium (Ca) and with Cl.

Ca is positively correlated with Cl. Mn is negatively correlated with Cr. Copper (Cu) is positively correlated with Sodium (Na) and Molybdenum (Mo). Tin (Sn) is positively correlated with Cu while it is negatively correlated with Cr. The second class of correlation gives the following information: Fe is strongly correlated with Sulphur (S); Mg is strongly correlated with S, Cl and K. S is strongly correlated with Cl; Zinc (Zn) is strongly correlated with K. A strong positive correlation exists between Sn and Na, Sn and Bromine (Br), Barium (Ba) and Aluminum (Al), Lead (Pb) and Ba, Pb and Al. The correlations that are related this study are those of: Fe-S, Mg-S, Mg-Cl, Mg-K, Zn-K, Cu-Na, Cu-Mo, Ni-Fe, Ca- Cl, because Fe, Mg, Zn and Cu are closely related to sickle cell disease. Positive correlations of these elements are beneficial for sickle cell disease, in case of a deficiency of one of these elements. In case of an excess of one element it is the negative correlation of this one which is beneficial for sickle cell disease.

This correlation study was carried out using the Pearson method with two software programs, SPSS and R studio, which produced the same results [18, 19].

The Table 1 gives the content of mineral elements identified in each plant.

Table-1. Mineral element content of the plants studied

	Na	Mg	Al	Si	P	S	Cl	K	Ca	V	Cr	Mn	Fe	Ni	Cu	Zn	Se	Br	Mo	Sn	I	Ba	Pb
A. bet	0.0	3117.1	902.8	239.8	1147.6	1503.2	384.6	12860.5	25708.7	2.5	2.0	681.8	408.6	3.1	9.4	22.8	0.0	4.8	0.0	0.7	1.7	15.6	3.0
D. br	2169.3	1905.4	58.3	37.7	1386.9	1357.3	126.9	8294.2	9433.4	1.8	1.8	524.9	164.5	4.1	23.7	51.6	0.0	47.9	1.7	1.2	2.5	7.0	0.0
H.cre	1582.0	4895.1	315.0	447.4	2472.6	3726.2	5936.6	19431.4	37605.0	2.2	2.5	34.9	795.5	2.3	6.2	32.2	0.8	9.1	0.0	0.9	2.0	6.2	1.2
J.sec	0.0	5538.5	104.7	150.1	2129.3	2561.3	3534.5	51887.5	13136.0	1.8	1.9	85.4	198.5	2.3	8.8	87.4	0.7	24.7	1.1	0.9	2.1	6.4	1.2
H.angu	0.0	550.1	18.5	9.0	1452.6	837.2	586.8	8685.6	4799.1	7.4	2.3	10.2	56.9	3.1	4.9	25.9	0.0	3.5	0.0	0.6	2.8	6.5	0.0
C.cong	0.0	3477.5	148.7	431.7	1561.3	3057.5	2642.2	29856.3	11026.3	2.0	2.1	46.9	790.1	2.5	4.5	47.9	0.0	11.8	0.0	0.9	2.1	6.5	1.5
H.mad	398.7	1646.6	15.1	11.2	3931.2	1920.2	64.0	15373.6	1243.5	0.9	2.3	40.7	101.2	4.8	10.7	67.8	0.0	1.7	0,0	0.7	1.5	2.2	0.0
V. ung	1300.9	1937.5	35.7	57.4	1333.6	2095.3	129.0	7291.5	15865.5	1.1	1.5	475.4	128.2	4.4	9.4	15.3	1.0	54.0	0.0	1.1	2.7	4.5	0.9
A. sen	1815.3	1693.1	27.9	153.9	1566.6	1659.0	118.0	7695.1	13747.3	1.0	1.3	428.5	123.7	4.3	16.6	28.6	0.7	7.3	0.0	1.0	2.4	7.2	0.9
A cord	0.0	1852.1	48.1	696.5	1258.4	2124.7	533.0	9262.1	9441.3	1.3	2.3	197.9	164.2	3.6	11.1	20.4	0.0	9.1	0.0	0.8	2.0	4.1	1.0

(Errors on all measurements did not exceed 5% of the mean value, so they are eliminated to avoid cluttering up the table)

4. Conclusion

The objective of this study was to determine mineral composition of medicinal plants used in traditional medicine in the province of Kwilu in the management of sickle cell disease. The results obtained from the fluorescence spectrophotometric method show that these plants (*Alchornea cordifolia, Alternanthera bettzickiana, Annona senegalensis, Cyttaranthus congolensis, Dissotis brazzae, Justicia secunda, Harungana madagascariensis, Hura crepitans, Hypoxis angustifolia, Vigna unguiculata*) contain each 23 distinct mineral elements. These results also show that calcium and potassium are the most abundant in all the plants studied, some elements are dominant in content and others are low in content. Some elements such as magnesium, zinc, selenium, manganese, copper, cobalt play important roles in sickle cell disease. Magnesium, in addition to being a cofactor of enzymes, reduces the number of abnormal erythrocytes in sickle cell patients and improves the hydration of red blood cells. Comparing the mineral element contents found in this work with those of the other research teams, a clear difference emerges, which is at least due to the method used by each team and also to the mineral element composition of the soil where these different plants are found.

For each of the mineral elements measured, there is either a positive or negative correlation that shows the behaviour of each mineral element with respect to its neighbour. A positive correlation means that these elements vary in content in the same direction while a negative correlation between two elements shows that the content of these elements varies in the opposite direction.

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