

Comparison of Major and Trace Element Concentrations in 16 Varieties of Cuban Mango Stem Bark (*Mangifera indica* L.)

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An aqueous decoction of mango (*Mangifera indica* L.) stem bark (MSB) has been developed in Cuba on an industrial scale to be used as a nutritional supplement, cosmetic, and phytomedicine, with antioxidant, anti-inflammatory, analgesic, and immunomodulatory properties. The concentration of major and trace elements was determined for 16 varieties of MSB belonging to two cultivars and grown in Cuba in the same soil (red ferralytic). Plants were classified into two groups, according to the tree age (12 and 26 year olds) and were analyzed for As, Ca, Cd, Cu, Fe, Hg, K, Mg, Pb, Se, and Zn content by means of ICP-AES technique. Experimental data were processed by ANOVA and principal component analysis in terms of elements, variety, and plant age, to choose the most adequate varieties for industrial purposes.

KEYWORDS: *Mangifera indica* L.; mango stem bark; nutritional supplement; major and trace elements; ICP-AES; PCA

INTRODUCTION

Mango extracts from leaves, fruit seed kernel, fruit pulp, roots, bark, and stem bark for medicinal purposes in many countries have been extensively reported in the Napralert Data Base. Specifically, the ethnomedical use of the mango stem bark aqueous extract (MSB) in Cuba has been documented widely (1) and it has been extensively used in cancer, diabetes, asthma, infertility, lupus, prostatitis, prostatic hyperplasia, gastric disorders, arthralgias, mouth sores, and tooth pain, as the more frequent diseases. Nondocumented verbal references from old Cuban people told about uses of the MSB by African slaves in Cuba since the XIX century. Documented data from more than 7000 patients with 84 diseases have been compiled in Cuba during the past 10 years. Focal studies have been done on specific diseases of relevant importance like cancer (2) to evaluate disease progress and the improvement of the patient's quality of life in a 6-month field trial: 84.8% of patients experienced decreasing level of depression, 82.2% of patients had a better integral evaluation, and 89.7% of them were able to have a normal life with an average dose of 20 mg/kg body

weight (b.w.) after oral administration three times a day of an aqueous decoction of MSB (30 mL).

The above-described results were considered as a ground basis for the development of a new natural product from the MSB with the hypothesis that so many successful applications would be sustained from its antioxidant effect better than a specific medical application. Therefore, the MSB was developed up to the industrial scale, standardized, and formulated to be used as an antioxidant nutritional supplement, a cosmeceutical product, and phytomedicine strongly related to oxidative stress, pain, and inflammation (3) with adequate protection of the intellectual property by a patent and registered brand name (*Vimang*).

The Ministry of Agriculture in Cuba has an inventory of 273 mango varieties which has been introduced into the country from more than 1200 reported worldwide. Sixty-eight mango varieties are distributed along the island for extensive cultivation and fruit collection but only 16 of them have been proven to be effective for the production of *Vimang* according to the polyphenols content by phytochemical screening and in vitro preliminary toxicological tests (data not published). Previous work reported the isolation and the quantitative analysis of phenolic constituents, free sugars, and polyols from an MSB aqueous decoction (4), where mangiferin (1,3,6,7-tetrahydroxyxanthone-C2- β -D-glucoside) was found to be the major component. Terpenoids (β -elemene, β -selinene, hinesol, and β -eudesmol) have also been found to be major components, together

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with sterols (β -sitosterol and β -campesterol), both chemical groups of biological relevance within MSB (5).

The concentrations at which MSB has exhibited its antioxidant effects in terms of lipid peroxidation inhibition, protection from oxidative damage, and scavenger capacity of oxidative chemical species *in vitro* were extremely low (6) and it seems to act by a mechanism different from the classical hydroxyl radical scavengers (7). Highly significant was the liver- and brain-protecting effect from *in vivo* experiments on rats and gerbils, respectively, in ischemia-reperfusion models at doses of 50 mg/kg b.w. (8–10). The analgesic and anti-inflammatory effects of MSB have been reported extensively (11–16). Recent results from a controlled clinical study in Primary Health Care with the use of MSB (formulated as *Vimang* coated tablets) in elderly humans demonstrated that oxidative stress markers were improved and they reached the same values as a young male control group after 2 months of treatment (3 tablets per day), with a high significant improvement of the subjects overall in terms of body pains (17).

MSB has also modulated several immune responses. It has a mytogenic effect on lymphoid populations with a high stimuli on the proliferation of T lymphocytes (18). Other experiments demonstrated that MSB inhibited immunoglobulines G (specifically IgG2a and IgG2b), typical of a T helper 1 (Th1) cells macrophage activation, without affecting the production of IgM (19). We have hypothesized that the strong antioxidant, analgesic, anti-inflammatory, and immunomodulatory effects of the MSB might be attributed to a mixture of components, organic and minerals, and not a single-component effect. Therefore, it was necessary to complete the analytical characterization of MSB by the identification and the quantitative analysis of major and trace elements such as Ca, K, Mg, and Fe (as dietary supplements) and Se, Cu, and Zn (as relevant microelements for the antioxidant defense body system, and the anti-inflammatory and immunomodulatory effects). Also, for toxicological purposes, the presence of heavy metals was investigated (As, Cd, Hg, and Pb).

In the present work, the content of major and trace elements in MSB of 16 Cuban varieties of mango (Group A: H01, C20, M13, CH4, CA5, B69, F77, SH8, BL9, and A10; Group B: M11, R12, N13, S14, D15, and O16) was evaluated by inductively coupled plasma absorption-emission spectrometry (ICP-AES), to identify and quantify the elements of dietary and biological relevance, which might contribute to the assessment of its nutritional and pharmacological effects, respectively. Group A was from a 12 year old cultivar and Group B from a 26 year old cultivar grown on the same type of soil to compare the mineral ions composition and to choose the most adequate varieties for the industrial production of MSB. Finally, principal component analysis (PCA) was applied to the experimental data with the purpose of assessing the possible statistical significance of mango variety, tree age, and mineral content to the observed biological effects in MSB.

MATERIALS AND METHODS

Chemicals. All of the pure standards were purchased from BDH (United Kingdom). All acids (Suprapur Grade) and solvents (Pure for Analysis) were purchased from E. Merck (Darmstadt, Germany). Water was purified by a Milli-Q_{plus} system from Millipore (Milford, MA).

Plant Material. The stem bark of *Mangifera indica* L. (Anacardiaceae) was collected free of biological contamination (fungi) from plants grown in an Experimental Fruit Farm (Instituto de Investigaciones de Cítricos y Frutales, Quivicán, Havana, Cuba) without affecting the ecosystem, according to a standardized collection procedure. Thus, 10 kg of bark was carefully cut along the mango tree stem, without

Table 1. Variety Codes of the Mango Varieties Used for the Elemental Quantitative Analysis of MSB

group A: 12 years old	group B: 26 years old
H01	M11
C20	R12
M13	N13
CH4	S14
CA5	D15
B69	O16
F77	
SH8	
BL9	
A10	

affecting the inner part of it, from the top (25 cm below the lowest branch) to the bottom (25 cm above the highest root). Cut width was not larger than 20 cm. Sixteen varieties, which were grown in two cultivars, with the same type of soil (red ferralytic), were selected for this study and encoded as reported in **Table 1**. Plants were classified into two groups according to the tree age, as described previously, and stored at room temperature (25 °C) in vacuum-sealed PVC bags until sample preparation. Voucher specimens were deposited at the Natural Products Archive, Center of Pharmaceutical Chemistry, Havana, Cuba (Code: 41722).

Sample Preparation. MSB samples were dried at 105 ± 2 °C for 6 h, subsequently milled (Hammer mill 705, UEMI, Havana) and sized to obtain pieces around 3 cm large, and then stored in vacuum-sealed PVC bags at room temperature (25 °C) until processing. Elements from the plant material were extracted by humid digestion according to standard procedures (20). The glassware was cleaned prior to use by soaking overnight with 10% v/v nitric acid. Dried plant material (1 kg) was poured into an Erlenmeyer flask (2.5 L) and heated up to 70 °C; 4 mL of nitric acid (65%) was added twice until full organic combustion. Thereafter, 4 mL of acids mixture (65% nitric acid, 98% sulfuric acid, and 70% perchloric acid; 10:1:3) were added, and the heating temperature was increased up to 130 °C until a transparent and colorless solution was obtained. The same procedure with the acids mixture was repeated and the heating was continued until the solution was reduced to 1 mL. The sample was poured into a 50 mL volumetric flask and filled with distilled water. Samples were stored at 8 °C until spectrometric analysis. A blank sample was prepared in parallel for subtraction in the spectrometric determinations. Each sample had three replicates.

Standard Preparation. Five solutions of pure standards for quantitative analyses were prepared by successive dilutions in distilled water with concentrations of 25, 5, 2.5, 0.25, and 0.025 $\mu\text{g/mL}$ for As, Ca, Fe, K, and Pb; 10, 2, 1, 0.1, and 0.01 $\mu\text{g/mL}$ for Cd, Cu, Mg, and Zn; and seven solutions of 50, 10, 5, 1, 0.5, 0.1, and 0.01 $\mu\text{g/mL}$ for Se. The technique of cold vapor was used for the determination of Hg.

Spectrometric Determination. Samples, both blank and standards, were analyzed by ICP-AES (Model Spectroflame, Spectro, Germany) with the following conditions: Paschen-Runge mode; 128 analytical channels; 5 optic systems; Rowland circle diameter, 750 mm; Zerodur holographic lattice; plasma flux, 13 L/min; auxiliary flux, 4 L/min; nebulizer flux, 1 L/min; sample aspiration velocity, 3 mL/min; radio frequency, 27.12 MHz; power, 2.5 kW (maximum). The absorbance data for each element were recorded at the following wavelengths: Pb ($\lambda = 168.2$ nm), As ($\lambda = 193.1$ nm), Se ($\lambda = 196.0$ nm), Zn ($\lambda = 213.9$ nm), Fe ($\lambda = 259.9$ nm), Mg ($\lambda = 285.2$ nm), Cu ($\lambda = 324.7$ nm), Cd ($\lambda = 327.4$ nm), Ca ($\lambda = 422.7$ nm), and K ($\lambda = 766.5$ nm). The monochromator was direct-access type with 4 inlets and 6 outlets and a photomultiplier. Each sample was analyzed twice.

Quantitation. Calibration curves and equations were obtained for each standard in the concentration ranges as described above. Absorbance values of each element were expressed as the mean value of six determinations \pm relative standard deviation. Concentration of each element was determined by interpolation and calculation from calibration curves and equations, respectively.

Linearity. Linearity was determined for all reference standards. Linearity of response was determined on five levels of concentration

Table 2. Mineral Content of Mango Varieties in Cultivar of Group A (12 Years Old)^a

element	varieties (concentrations in $\mu\text{g/g}$ MSB)										mean value
	H01	C20	M13	CH4	CA5	B69	F77	SH8	BL9	A10	
Ca	309.3 ± 10.9	387.5 ± 6.5	372.5 ± 13.6	358.8 ± 13.0	436.3 ± 11.6	448.8 ± 28.8	651.5 ± 11.1	446.8 ± 13.8	377.3 ± 19.3	432.2 ± 27.3	422.1
K	183.5 ± 8.8	220.1 ± 10.4	328.8 ± 7.9	378.3 ± 11.4	319.7 ± 6.3	215.3 ± 12.7	256.1 ± 1.5	434.7 ± 8.8	379.4 ± 11.3	215.4 ± 8.1	293.1
Mg	32.4 ± 1.6	44.5 ± 1.9	27.8 ± 6.8	32.1 ± 2.1	35.3 ± 0.3	42.5 ± 3.3	13.0 ± 1.3	21.4 ± 0.4	36.4 ± 0.1	16.2 ± 0.2	30.2
Fe	7.0 ± 0.2	19.9 ± 7.0	9.3 ± 0.6	9.5 ± 1.0	9.5 ± 2.6	6.1 ± 0.4	2.9 ± 0.5	5.8 ± 0.5	5.9 ± 0.4	5.8 ± 0.1	8.2
Cu	0.8 ± 0.1	2.6 ± 0.2	1.6 ± 0.1	2.8 ± 0.5	1.7 ± 0.1	1.0 ± 0.1	0.2 ± 0.1	1.2 ± 0.1	0.3 ± 0.01	0.6 ± 0.01	1.3
Zn	0.6 ± 0.2	0.9 ± 0.1	0.4 ± 0.1	0.5 ± 0.1	0.5 ± 0.1	0.5 ± 0.1	0.4 ± 0.1	0.5 ± 0.1	0.4 ± 0.1	0.5 ± 0.01	0.5
Se	2.0 ± 0.1	2.0 ± 0.1	1.2 ± 0.2	1.2 ± 0.1	1.2 ± 0.1	1.2 ± 0.1	1.2 ± 0.1	1.2 ± 0.1	1.2 ± 0.1	1.1 ± 0.1	1.3
Pb	0.12 ± 0.01	0.20 ± 0.01	0.28 ± 0.03	0.08 ± 0.04	0.09 ± 0.03	0.45 ± 0.49	0.08 ± 0.10	0.09 ± 0.01	0.06 ± 0.05	0.08 ± 0.01	0.2
total	535.8	677.7	741.9	783.1	804.3	715.8	925.3	911.8	797.8	671.7	756.5

^aData are the means of three experiments. S.E.M.s were below 10%.

Table 3. Mineral Content of Mango Varieties in Cultivar of Group B (26 Years Old)^a

element	varieties (concentrations in $\mu\text{g/g}$ MSB)						mean value
	M11	R12	N13	S14	D15	O16	
Ca	523.8 ± 14.4	487.5 ± 23.6	554.8 ± 18.2	358.8 ± 0.8	576.2 ± 13.5	639.2 ± 14.2	523.4
K	198.9 ± 4.3	231.5 ± 1.7	154.4 ± 8.2	207.7 ± 15.7	217.5 ± 1.9	126.1 ± 1.5	189.3
Mg	14.6 ± 0.9	21.8 ± 0.6	24.8 ± 0.9	9.9 ± 1.6	37.1 ± 1.3	31.5 ± 0.9	23.3
Fe	2.9 ± 0.4	4.4 ± 0.6	3.5 ± 0.2	3.4 ± 0.4	4.8 ± 0.9	2.1 ± 0.2	3.5
Cu	0.1 ± 0.01	0.1 ± 0.01	0.1 ± 0.01	0.1 ± 0.01	0.2 ± 0.01	0.1 ± 0.01	0.1
Zn	0.2 ± 0.1	0.4 ± 0.1	0.4 ± 0.1	0.5 ± 0.1	0.4 ± 0.03	0.3 ± 0.1	0.4
Se	1.1 ± 0.1	1.1 ± 0.1	1.1 ± 0.1	1.0 ± 0.1	1.1 ± 0.1	1.0 ± 0.1	1.0
Pb	0.01 ± 0.00	0.05 ± 0.03	0.04 ± 0.01	0.02 ± 0.02	0.05 ± 0.03	0.04 ± 0.02	0.04
total	741.5	746.9	739.2	581.4	837.3	800.3	741.1

^aData are the means of three experiments. S.E.M.s were below 10%.

with three determinations for each level. A linear relationship between absorbance and concentration, at the above-described ranges, was observed for each standard with a correlation coefficient $r = 0.99$. Detection limits ranged between 0.005 and 0.02 $\mu\text{g/g}$ with a signal-to-noise ratio of 3:1.

Statistical Analysis. Three independent analyses were carried out for each variety for major and trace elements. Statistical analyses were performed by Mann-Whitney U Test. Minimal acceptance of statistical significance was $p < 0.05$. All data in the tables are expressed as the mean \pm standard error (S.E.M.).

Data Processing. Duncan multivariate test and ANOVA with a factorial design (tree age, variety, and mineral ion) were performed on the experimental data. PCA was applied to the experimental data by considering the correlation between elements and varieties. Elements were taken as mathematical-statistical cases (rows of the matrix) and varieties as variables (columns of the input matrix). PCA was applied to decompose the original matrix into loading (varieties) and score (elements) matrices. The internal correlation circles were constructed to determine vector significance (element and variety).

RESULTS AND DISCUSSION

MSB organic components, such as polyphenols and terpenoids, which account for 45% and 25%, respectively (4), with the glycosylated xanthone, mangiferin, as the main MSB component (ca. 10%) would probably have a higher effect on health and nutrition than the inorganic components. However, the formation of coordination complexes between organic and inorganic components within MSB should be considered to assess the biological effects of MSB. For example, we reported recently from *in vitro* experiments that a complex mangiferin–Fe showed a significantly higher influence on mitochondrial permeability and antioxidant and cytoprotector effects (21) than mangiferin alone.

All mango varieties had a mineral ion content below 1% (between 0.5 and 0.9%) of the total dry weight of MSB, where

organic components are predominant, and therefore insignificant statistical differences were found regarding total mineral ion content. ICP-AES was a useful technique for analyzing the mineral ion content of the stem bark in the 16 mango varieties and results are reported in **Table 2** for Group A (12 years old) and **Table 3** for Group B (26 years old) cultivars, respectively. The total mineral ion content in the 16 varieties was found in the sequence $F77 > SH8 > D15 > CA5 > O16 > BL9 > CH4 > R12 > M13 > M11 > N13 > B69 > C20 > A10 > S14 > H01$. The highest mineral ion content was found in F77, 925.34 $\mu\text{g/g}$ (Group A), whereas H01, within the same Group A, had the lowest, 535.79 $\mu\text{g/g}$.

The mineral ion contents of MSB in both, Groups A and B, had similar sequences of concentrations for major elements, in the order $Ca > K > Mg > Fe$. However, trace elements sequences were different between Group A ($Cu > Se > Zn > Pb$) and Group B ($Se > Zn > Cu > Pb$). The concentrations of K, Ca, Mg, and Fe and the concentration of Cu, Zn, and Se in MSB (w/w) for the varieties of both, Groups A and B, are shown in **Figures 1** and **2**, respectively.

Ca was found to have the higher values within all the analyzed mineral ions (mean value = 460.1 $\mu\text{g/g}$) followed by K (mean value = 254.21 $\mu\text{g/g}$). Ca concentration ranged from a low content in Group A-H01 and Group B-S14 varieties (309.33 and 325.50 $\mu\text{g/g}$, respectively) to a high one (651.50 $\mu\text{g/g}$) in Group A-F77 variety, approximately a 2-fold increase. The lowest content of K was found in Group B-O16 variety (126.10 $\mu\text{g/g}$), while the highest content was found in Group A-SH8 variety (434.72 $\mu\text{g/g}$), which is almost 4 times higher. Mg concentration had a mean value of 27.57 $\mu\text{g/g}$ and ranged from 8.27 $\mu\text{g/g}$ (Group B-S14 variety) to 44.48 and 42.47 $\mu\text{g/g}$ (Group A-C20 and -B69 varieties, respectively), more than a 5-fold increase. Fe had the lowest concentrations in Group A-F77,

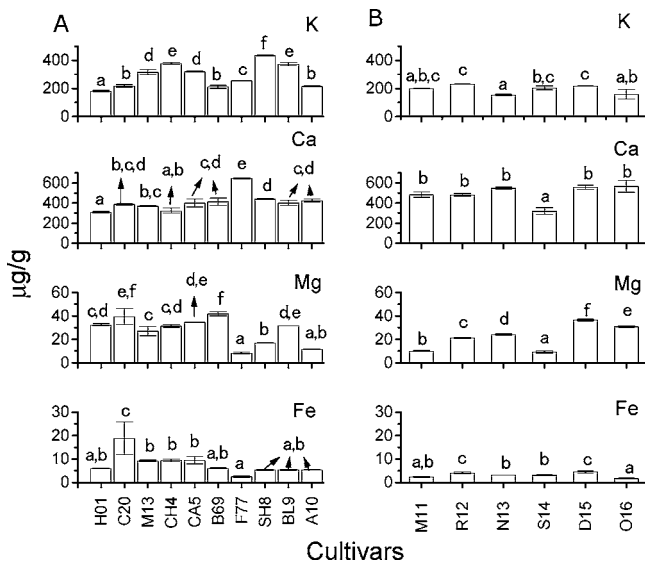


Figure 1. Concentrations of K, Ca, Mg, and Fe in MSB (w/w) of 16 varieties of mango (*Mangifera indica* L.) cultivated in an experimental farm (Havana, Cuba) on a red-ferralytic soil type. Mean value \pm S.E.M. of three replicates are shown in Table 2. Group A was a 12 year old cultivar and Group B a 26 year old cultivar. Different letters above the bars indicate statistical significant difference ($p < 0.05$).

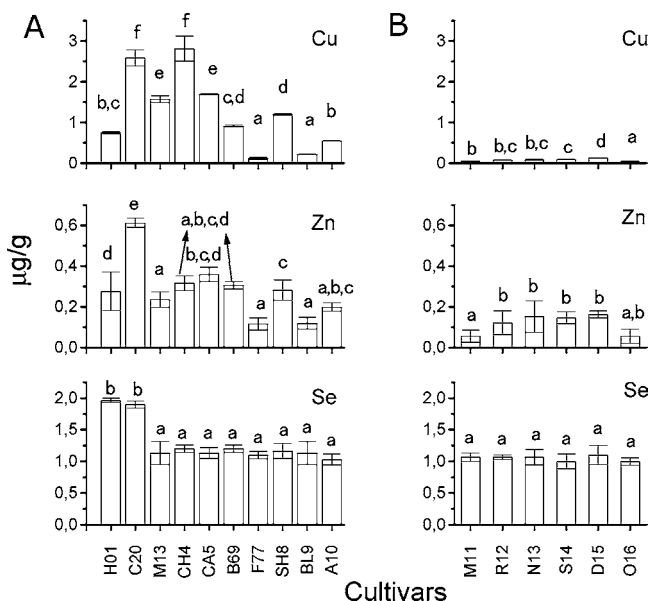


Figure 2. Concentration of Cu, Zn, and Se in MSB (w/w) of 16 varieties of mango (*Mangifera indica* L.) cultivated in an experimental farm (Havana, Cuba) on a red-ferralytic soil type. Mean value \pm S.E.M. of three replicates are shown in Table 3. Group A was a 12 year old cultivar and Group B a 26 year old cultivar. Different letters above the bars indicate statistical significant difference ($p < 0.05$).

Group B-M11, and Group B-O16 varieties (average: 2.62 $\mu\text{g/g}$). On the other hand, Group A-C20 variety had the highest Fe content (19.86 $\mu\text{g/g}$). The mean value of Fe was 6.42 $\mu\text{g/g}$. All these values of major elements were considered insignificant in terms of their biological relevance as food supplements and therefore the influence of the variety was discarded when considering stem bark collection for the production of MSB. However, the interaction of Fe with MSB components in terms of antioxidant and cytoprotecting activities must be taken into account in future in vivo evaluations, according to recent experimental results in vitro (22).

Significant differences were observed when considering the tree age: K and Fe ions were in higher average concentrations in varieties of the Group A than those of the Group B, whereas Ca and Mg were in higher amounts in the Group B. Considering the osmotic importance of K for the human organism and the relevance of Ca supplementation for the human body, it was interesting to note that plants of Group B had higher concentrations of Ca associated with a minor content of K, when compared to Group A, which would probably be related to plant age. The observed differences in Mg and Fe concentrations were not relevant since in both groups and all varieties their concentrations were below the Daily Recommended Allowance (DRA) (23) when considering the preparation of *Vimang* oral pharmaceutical formulations from MSB. The above-described results lead us to recommend the collection of mango stem bark on 26 year old trees for the production of MSB in terms of Ca content as food supplement. Ca daily intake has been considered relevant for human health; however, it varies widely between different regions of the world (Nordic countries, 1,200; European Union, 900; Middle East, 800; Latin America, 500; Africa and Far East, below 400 mg/day) (24). Ca intake has been reported to be considerably low in communities where animal milks are scarce or not habitually consumed.

The most relevant finding was to identify Se in all varieties, and its concentration was not dependent on plant age or the variety. The mean concentration of Se for all varieties (1.49 $\mu\text{g/g}$) was within the DRA as dietary supplement for both Groups A and B. Only two varieties (H01 and C20), both from Group A, had a significantly higher content of Se than the other 14 varieties (see **Figure 2**), but the statistical difference was negligible when compared to the DRA range in terms of MSB dose in *Vimang* formulations. This result would allow us to correlate the presence of Se to the antioxidant effect of MSB for all mango varieties included in this study, where the action of Se as cofactor of glutathion peroxidase has been extensively reported (25). Other relevant information is the increasing importance of Se as a chemoprevention factor for several types of cancer (26, 27). Therefore, the relative constant concentration of Se will not influence variety or plant age when considering stem bark collection for the production of antioxidant *Vimang* formulations.

Cu and Zn concentrations were significantly higher in Group A, although these values in both groups were below the DRA as dietary supplement for MSB and were, therefore, insignificant. There was not a statistically significant difference in Cu content within Group A, with an average Cu concentration of 0.9 $\mu\text{g/g}$. The mean values of Cu and Zn, when considering the 16 varieties, were 0.84 and 0.48 $\mu\text{g/g}$, respectively. In M11 (Group B) the lowest contents of Zn, 0.24 $\mu\text{g/g}$, were found, while C20 (Group A) had the highest content (0.94 $\mu\text{g/g}$) and these values were not significantly different. Cu and Zn play significant roles in the activation of superoxide dismutase (SOD) (28, 29), a key enzyme within the inflammatory response. Recent works about Zn relevance in several physiological systems have been published elsewhere (30, 31).

Pb was detected in all varieties and concentrations ranged from 0.01 (M11) to 0.45 mg/g (B69), with a mean value of 0.11 $\mu\text{g/g}$, which was considerably below the toxic dose for human consumption (< 2 mg/kg body weight) (32). Those results were in correspondence with acute and chronic toxic evaluations of MSB for its use in *Vimang* formulations (33), leading to classify *Vimang* formulations as nontoxic for oral and topical administration before health authorities. Cd, As, and Hg were

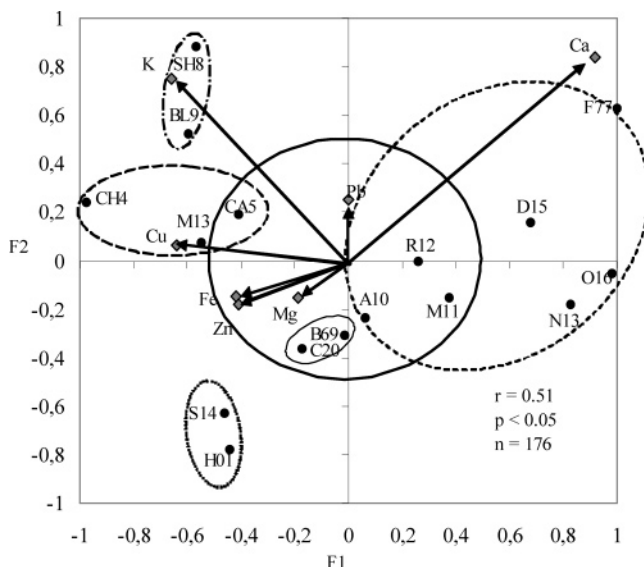


Figure 3. PCA graphics of the concentration of Ca, K, Mg, Fe, Cu, Zn, and Pb in 16 varieties of mango (*Mangifera indica* L.) cultivated in an experimental farm (Havana, Cuba) on a red-ferralytic soil type. Group 1 (thick dark circle) includes 6 varieties (C20, CA5, B69, A10, M11, and R12) which had a significant correlation according to (12) for all elements; Group 2 (short-dot circle) includes Ca-predominant varieties (D15, N13, O16, and F77); Group 3 (dot-point circle) includes K-predominant varieties (SH8 and BL9); Group 4 (long-dot circle) includes Cu-significant varieties (CH4 and M13); Group 5 (point circle) includes varieties of low mineral content (H01 and S14). Se content was excluded from PCA analysis.

not detected in all MSB samples analyzed by ICP-AES at detection limits of 0.005, 0.02, and 0.01 $\mu\text{g/g}$, respectively.

Principal component analysis (PCA) is a powerful tool for pattern recognition, classification, modeling, and other aspects of data evaluation. For this reason, the PCA technique was performed to determine the most attractive varieties for MSB collection in industrial processing. The analysis of the Se concentration was not included in PCA analysis since its concentration did not change significantly within all studied cultivars, being almost centered at the plot.

The PCA plot is shown in **Figure 3**. The first two principal components (PCs) were chosen (60.5% of the total variance) because their eigenvalues were higher than 1. The first PC that explains the higher percentage of variance (38.6%) was associated with Ca and the second with K (23.2%). A third PC, but with an eigenvalue lower than 1, was Cu (11.8%). Four of 16 varieties (25%) were Ca-predominant (F77, D15, O16, and N13), three from Group B and one from Group A, which confirmed the results of the previous discussed statistical analysis. This means that 26 year old trees are better than 12 year old trees as the source of stem bark for the production of MSB as Ca-food supplement. Two varieties (SH8 and BL9) were related to a high content of K (12.5%) both from Group A, with insignificance in their use for the production of MSB. Varieties CH4 and M13 (third vector), both from Group A, were correlated to the Cu content, but without significance.

We concluded that stem bark collection for the production of MSB, in terms of mineral ion content for major and trace elements, could be done for all varieties included in the present study. The presence and concentration of Se in MSB for its use as raw material for antioxidant formulations and also of Ca as food supplementation were noticeable. It would be preferable to perform the collection of mango stem bark for the production of MSB on trees aging around 26 years because of its high Ca

content correlated to a low content of K. Research work will in due course evaluate ionic complexes of Fe, Se, Cu, and Zn with MSB polyphenols, which seems to be of biological relevance to increase the antioxidant and cytoprotecting effects of *Vimang* formulations.

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