

# Characterizing the relationship between aluminum content and pH in coffee (*Coffea arabica* L.) crop soils in Mexico

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**ABSTRACT:** Mexico ranks fifth among coffee producing countries worldwide. Coffee production has economic implications at both a national and an international level. Therefore, the investigation of factors negatively impacting the cultivation of different crops on the American continent is an important area of study. In Mexico, the production of coffee may be affected by the presence of aluminum and other soluble metals in crop soils. Furthermore, coffee grows in acidic soils that have increased availability of  $Al^{3+}$ , which is more accessible to plants than other elements present in the soil. These high levels of  $Al^{3+}$  produce toxic effects in coffee plants. To study the relationship between the pH conditions and aluminum contents of the soils in which coffee crops are produced, we determined the pH,  $Al^{3+}$  and total aluminum levels in the soil from three different regions of Mexico. We found different pH values in the three regions of Mexico: the pH was lower in soils from Veracruz than in the Chiapas and Yucatan soils, coinciding with the higher amount of  $Al^{3+}$  detected in the soil from Huatusco, Veracruz. Additionally,  $Al^{3+}$  determinations were performed on several coffee products used for human consumption. We detected the presence of aluminum in coffee beverages and ground coffee products originating from Chiapas and Veracruz. Our results suggest a direct relationship between acidic pH and  $Al^{3+}$  content. We also detected the metal in products derived from coffee.

**Keywords:** acidic soils, aluminum, *Coffea arabica*, Coffee, pH

## INTRODUCTION

Coffee crops can be cultivated in tropical regions of the world that have suitable climates, such as in Eastern Africa, South-West Arabia, Central and South America and Asia (ICO, 2008). Coffee is an important exported agricultural commodity for Mexico. Moreover, Mexico produces more certified organic coffee than any other country, contributing significantly to conservation of coffee biodiversity. The major coffee-growing regions of Mexico include Chiapas, Oaxaca and Veracruz, and cooperatives in these regions have been innovators in the organic production of and fair-trade practices surrounding coffee (Celis et al., 1991; Pérezgrovas et al., 1997). However, coffee growers are constantly confronted with conditions that do not support optimal agronomic methods, such as wet seasons, parcels of land that are more vulnerable due to deforestation, susceptibility of coffee bushes to disease and increasingly acidic soils. All of these conditions are consequences of anthropogenic climate change (Smith et al., 2008).

Soil properties play a crucial role in plant development, and the reactivity of soil depends on its degree of acidity or alkalinity (Berkelaar, 2001), which also influences the mobility of different soil components (Nye, 1981; Violante, 2010). It has been shown that more than 50% of worldwide arable land contains acidic soil (Aguilera and Martínez, 1990; Vitorello et al., 2005) and that the presence of metals including Cd, Pb and Al in soil can limit plant growth and be toxic to animals and humans (Adriano, 2001; Matsumoto, 2000; Ciamporova, 2002). The toxicity produced from high concentrations of metals can be the result of both the intrinsic toxicity of elements and their accumulation (García and Dorronsoro, 2005), and the effects of metal ions in plants are numerous.

Reduced root growth decreases the absorption of nutrients and water and, consequently, the crop yield. Coffee is often grown in acidic soils, a condition that increases the levels of  $Al^{3+}$ , an exchangeable form of aluminum that can be released to the soil, making it very accessible to plants (Xie et al., 2001). However,  $Al^{3+}$  also produces toxic effects (Kochian et al., 2005; Marschner, 1995; Qiang et al., 2009). Because aluminum can be transferred to plants from the soil, it is necessary to understand the variation in soil composition with respect

to metal content. Investigation of soil compositions includes the evaluation of soil metal concentrations and the determination of the properties and circumstances necessary for optimal agricultural working conditions (Andrade et al., 2000; Arshad and Martin, 2002). However, considering the particular chemical characteristics of aluminum and its capacity to react with a wide spectrum of compounds in soil, studying the toxic effects of aluminum in plants is challenging. Additionally, it is necessary to consider the natural variability of soils (Kinraide, 1991) in order to obtain a more comprehensive picture of the effect of aluminum on plants.

Metal transference can also occur between plants and humans. This information is particularly relevant to this study because the consumption of coffee-containing beverages is common in modern society. Food is considered to be the primary source by which humans ingest aluminum (Yokel, 2004). Throughout history, coffee cultivation and its commercialization has had worldwide relevance; indeed, the coffee market currently exchanges more than 6000 billion kg of coffee per year and employs more than 25 million people globally. In Mexico, coffee is the fourth most popular beverage consumed after water, milk and soda. The average Mexican citizen consumes approximately 1.3 kg of coffee per year, and 53% of the population consumes 2-3 cups of coffee-containing beverages daily. Also, 85% of the population consumes soluble coffee products, and 17% consumes ground coffee.

In Mexico, coffee growers recognize the close relationship between crop yields and climate change; however, as relatively little information regarding aluminum toxicity in coffee crop soils is available, uncertainty exists about the possible dependence of crop yields on the presence of metals in the soil. In one existing report, Pavan and Bingham studied the effect of aluminum on *Coffea arabica* L. plants in four different acidic soils from Brazil and identified a relationship between the activity of the  $Al^{3+}$  cation and reduced growth in *C. arabica* seedlings. Yet, an understanding of the effects of aluminum on coffee production is lacking, and data relating to the aluminum content in Mexican coffee-growing soils are scarce. Through collaboration with coffee growers, we aim to increase our understanding of how soil components such as metals (e.g., aluminum) affect crop yield in order to optimize production. Because there is not enough evidence to suggest that coffee products made for human consumption contain aluminum, another goal of this work is to study the relationship between pH and aluminum content in Mexican soils in which coffee crops are produced.

## MATERIALS AND METHODS

### MATERIALS

Acetylene (research purity, 99.999%) and nitrous oxide (research purity, 99.999%) were obtained from Praxair (Mexico). For aluminum determination, a standard curve was generated using  $Al(NO_3)_3 \cdot 9H_2O$  as the standard; the standard reagent was obtained from Perkin Elmer (USA).  $LaCl_3$  was purchased from J.T. Baker (USA). All other chemicals were obtained from Sigma (USA).

#### **Sample collection**

Soil samples were collected from the three principal coffee-producing states in Mexico (Figure 1). Within the state of Chiapas, zone IV (Soconusco) was selected as the collection site, and a region called Huatusco was selected within the state of Veracruz. The central region of Yucatan was selected as the third site for sample collection because its soil has an alkaline pH.

Following the specified procedures established in the Mexican Official Norm (NOM-021-RECNAT-2000) (Norma Oficial Mexicana), the collected soil samples were assessed by the number of hectares and the number of samples from each region. Three samples were collected in Yucatan, eight samples were collected in Chiapas and eleven samples were collected in Veracruz. Every sample comprised a number of sub-samples combined to form a single compound soil sample (0.5 kg). The samples were collected from the soil at a depth of 20 cm (8 inches). For further analysis, leaf samples from coffee plants were also collected from the same regions as the soil samples. The beans from Chiapas and Veracruz were obtained from a commercial product market in each location.

#### **Treatments**

All soil samples were dried at 80°C for 48 h. Soil samples were cleaned to remove excess rocks and organic material prior to being crushed in a porcelain mortar and sieved using a steel sieve (2 mm pore size) to obtain a fine powder that was used for the analysis of aluminum content. For the analysis of total aluminum present in the coffee leaves, the leaves were washed to remove residual soil and were then dried in an oven at 80°C for 48 h. Prior to analysis, coffee beans were ground into a fine powder, which was then dried at 80°C for 42 h.

### ***pH determination***

The pH of each sample was measured following the procedures from NOM-021-RECNAT-2000 (Norma Oficial Mexicana, 2000). The soil samples were first weighed in triplicate (10 g). Next, 20 ml of degassed water was added to each sample. The samples were mixed with a glass bar for 30 min and then were left to settle. After 15 min, each mixture was agitated and the pH was measured using a Beckman pH meter (USA).

### ***Aluminum determination***

According to our experimental strategy, we used different methods to measure the level of Al, depending on whether the free form or the conjugated form was present in the samples. The quantification of total Al was performed using atomic absorption spectroscopy (AAS) and scanning electron microscopy (SEM). Inductively coupled plasma atomic emission spectroscopy (ICP-AES) was used to measure free Al.

### ***Atomic absorption spectroscopy (AAS)***

Each sample (soil, leaves and beans, previously dried) was analyzed in triplicate. The samples were weighed, placed in a porcelain crucible and oven dried (Novatech, USA) at 80°C for 24 h. The samples were then furnace ashed (Thermolyne, USA) at 400°C (temperature range was 100°C to 400°C, increasing by 100°C every 15 min) for 1 h and then at 550 °C for 2 h. The ashes were next dissolved in 70% nitric acid (Sigma Aldrich, USA) and collected by vacuum filtration on filter paper (Whatman, USA, medium pore size). The ashes were recovered by adding LaCl<sub>3</sub> at a concentration of 50,000 ppm, and the material was transferred to acid-washed test tubes. The total concentration of aluminum was measured using a Perkin-Elmer 3100 atomic absorption spectrometer ( $\lambda = 309.3 \text{ nm}$ ) (the aluminum detection limit was  $450 \mu\text{g L}^{-1}$ ), with a flame generated by mixing air, acetylene and nitrous oxide. A standard curve was generated using a mixture containing Al(NO<sub>3</sub>)<sub>3</sub>·9H<sub>2</sub>O and 50,000 ppm LaCl<sub>3</sub>.

### ***Scanning electron microscopy (SEM)***

The Al analysis was performed using a Jeol Scanning Electron Microscope (SEM), model JSM-6360 LV, with soil samples that had been dried at 80°C for 48 h. Following the drying period, the samples were placed on a glass slide with copper tape for analysis.

### ***Inductively coupled plasma atomic emission spectroscopy (ICP-AES)***

Measurements of aluminum in coffee grounds and beverages were made using a Varian VistaPro (Melbourne, Australia) axially viewed inductively coupled plasma atomic emission spectrometer (ICP-AES) using an autosampler. The detection limit for aluminum was  $1.1 \mu\text{g L}^{-1}$ .

### ***Determination of exchangeable aluminum (Al<sup>3+</sup>) in soil samples***

To determine the Al<sup>3+</sup> levels in soil samples, the approved potassium chloride (KCl) method (NOM-021-RECNAT-2000) cited in the regulations for soil classification, sampling or analysis of soils were employed, in accordance with Mexican regulations. The samples were weighed (5 g) and 1 M KCl was added to each sample. The samples were then agitated using an orbital shaker (Nutator) for 30 min and centrifuged at 1309 x g for 10 min (Centrifuge Legend MACH 1.6R). The supernatant was filtered using Whatman paper No. 40, after which 40 ml aliquots were combined with 5 drops of 0.5% phenolphthalein (Sigma Aldrich). The titration was performed with 0.1 M NaOH, and observation of a pink colored solution indicated the endpoint. After the volume of NaOH used for titration was recorded, 2 ml of 1 M potassium fluoride was added to each sample. Then, 0.1 M HCl was added until the solution became colorless. For a control assay, the same procedure was performed using a 1 M KCl starting solution instead of a soil sample.

### ***Statistical analysis***

The data were analyzed using a one-way analysis of variance (ANOVA), and comparisons of the means were performed using Duncan's method with Statgraphics 5.1. The samples were analyzed in triplicate with a confidence level of 95%.

## **RESULTS**

### ***Qualitative and quantitative analyses of aluminum in different soils***

Collected soil samples were analyzed using SEM as a new tool to determine the presence of metals in the soil. As shown in Figure 2A, the Yucatan soil samples contained of 6.98% calcium, 3.76% iron, 8.3% silicon and 8.54% aluminum. In the soils from Chiapas, silicon and aluminum were present at 19% and 8%, respectively, and represented the two most abundant elements in these samples (Figure 2B). In the soils from Veracruz, a greater relative abundance of aluminum was detected (9.8%), and iron was present at an abundance of 7% (Figure 2C).

### **Aluminum-pH relationship in coffee soils**

The acidity or alkalinity of the soil is crucial for plant development, and the interactions among the soil, elements, minerals, water, animals and plants are highly dependent on pH. pH values were measured as described in the Materials and Methods section and the resulting values are presented in Table I. pH values for the Yucatan soils were approximately  $8.15 \pm 0.08$ , reflecting slightly alkaline conditions. Conversely, the pH of Chiapas soils showed values of 5.45-6.0, reflecting acidic conditions.

The levels of  $Al^{3+}$  were also measured. The amount of  $Al^{3+}$  found in the Yucatan soils ranged from 0.1 to  $0.21 \text{ cmol kg}^{-1}$  (Table 1). These results represent the first report of the presence of exchangeable aluminum in Yucatan soil samples. These values are lower than those observed in other soils because the alkaline pH of the Yucatan soil does not stimulate dissolution of aluminum species. The  $Al^{3+}$  contained in the Chiapas soil samples was  $0.13 \pm 0.04 \text{ cmol kg}^{-1}$ , which was similar to the  $Al^{3+}$  content observed in the Yucatan soils. However, the  $Al^{3+}$  content of Chiapas soils determined in this study differs from the values reported by Ramos and Flores for Chiapas soils in a previous study (Ramos and Flores, 2008). These differences could be due to the specific locations chosen for sample collection within each state. For example, the samples in our study were collected from a locations used for organically grown coffee (Ortiz et al., 2004), which may not have been the case in previous studies. In contrast to Yucatan and Chiapas soil samples, Veracruz soil samples exhibited  $Al^{3+}$  values as high as  $2.76 \text{ cmol kg}^{-1}$  in samples with a pH of 3.6. In the same region, we detected lower concentrations (i.e.,  $0.1 \text{ cmol kg}^{-1}$ ) of  $Al^{3+}$  where the soil pH was higher (i.e., 6.44), thus reflecting an inverse relationship between the  $Al^{3+}$  content and the soil pH. The  $Al^{3+}$  concentration reported in this study ( $2.76 \text{ cmol kg}^{-1}$ ) is higher than previously reported values for  $Al^{3+}$  content ( $0.8 \text{ cmol kg}^{-1}$ ) for this region of Veracruz (Geissert et al., 2000).

### **Total aluminum in soil, coffee leaves and coffee beans**

$Al(OH)_2 \cdot 5H_2O$  is a compound that has negative effects on the development of *Triticum aestivum*, while the compounds  $Al^{3+} \cdot 6H_2O$  and  $Al(OH)^{2+} \cdot 4H_2O$  have been shown to damage *Coffea arabica* plants (Müller et al., 1997). However, it is challenging to determine the specific forms of aluminum that produce toxic effects in plants because these aluminum compounds are highly reactive. Therefore, taking the sum of all forms of monomeric aluminum present in soil provides a good alternative method for investigating the possible effects of aluminum on plants (Casierra-Posada and Aguilar-Avenidaño, 2007). Quantification of total aluminum present in each zone was performed using AAS analysis, as described in the Materials and Methods section. The total aluminum concentrations measured in the Yucatan soils ranged from 19 to  $119 \text{ mg g}^{-1}$ . The relative abundance of aluminum was 8.2%, which is similar to the 8.54% aluminum abundance determined by SEM. In Chiapas soils, the values for total aluminum content ranged from 19 to  $48 \text{ mg g}^{-1}$ , which were lower than the values observed in the Yucatan soils.

Because we were studying aluminum toxicity in plants that are used to make coffee-containing beverages, we also wished to ascertain whether aluminum from the soil is absorbed by plants and mobilized to different plant organs. To investigate this question, we determined the amounts of aluminum contained in the leaves and beans of coffee plants. It should be noted that the  $Al^{3+}$  concentration in crop plants is usually higher in the roots than in aerial tissues and that the first symptom of aluminum toxicity is reduced root growth (Ryan et al., 1992). The total aluminum content of coffee leaves harvested from plants growing in Yucatan ranged from 0.02 to  $0.17 \text{ mg g}^{-1}$ . While these relatively high values for aluminum content are similar to those that have been reported for acidic soils from Brazil (Pavan and Bingham, 1982), it is interesting to note that the soil pH in Yucatan is alkaline (pH 8.07), whereas the soil pH reported in the Brazilian study is 4.3. Nevertheless, alkaline soil conditions in Yucatan permitted aluminum absorption by the coffee plants (Table 2). We detected aluminum at concentrations between 0.02 and  $0.16 \text{ mg g}^{-1}$  in coffee leaves collected in Chiapas, in agreement with the data reported in the Brazilian study ( $0.06\text{--}0.10 \text{ mg g}^{-1}$ ). Additionally, we observed size reductions in roots and shoots (Pavan and Bingham, 1982). The average aluminum content in leaves from Veracruz was  $0.17 \text{ mg g}^{-1}$ , which is greater than the values determined in leaves from Yucatan and Chiapas (Table 2).

According to a report on the daily exposure of humans to aluminum in food and beverage products, food is considered the major source of aluminum ingested by humans (Yokel et al., 2004). Because we detected aluminum in aerial tissues, such as leaves, and not only in roots, aluminum must be mobilized in coffee plants. This observation suggests that aluminum can also be mobilized to fruits. It is important to determine the amount of aluminum contained in coffee beans produced by plants grown in acidic soil conditions because high aluminum contents could allow increased aluminum absorption in humans and represent a significant health risk. After investigating the aluminum levels using ICP-AES, a technique with a higher sensitivity than AAS (Table 3), we detected substantial levels of aluminum in almost all samples of coffee grounds currently available for human consumption.

## DISCUSSION

The high proportion of aluminum and silicon in soil samples can be explained by the fact that aluminum forms oxides with multiple different minerals. These oxides include  $Al_4(SiO_{10})(OH)_8 \cdot 4H_2O$  (halloysite),  $AlO(OH)$  (boehmite) and  $SiO_2$  (quartz) (Bautista and Palacio, 2005) (Figure 2A).

The reduced presence of elements such as calcium, magnesium and potassium has been observed in acidic soils and is consistent with our analyses of Chiapas soils: calcium (0.91%), magnesium (0.56%) and potassium (1.23%) (Adams, 1984).

These high values for aluminum and iron are characteristic of volcanic soils (i.e., andosols) in which Fe and Al commonly form complexes with organic material to increase their stability (Meza and Geissert, 2003) (Figure 2C). Some metals, such as calcium and magnesium, support alkaline conditions in soil. In agreement with this observation, soil samples from Veracruz contained very low concentrations of both elements (i.e., less than 0.1%). A low abundance of base metals results in a strongly acidic soil (Manson et al., 2008).

Soil from Yucatan usually contains high concentrations of calcium carbonate, which is highly insoluble, thus influencing the base saturation. This alkaline condition is also supported by the increased generation of  $OH^-$  ions from the dissociation of calcium hydroxide, compared to the decreased formation of  $H^+$  ions from carbonic acid (Millar et al., 1979). These pH values are similar to the values (pH 5.5) reported from other zones of Chiapas (Triomphe, 2009; Mendoza et al., 2007). The acidic character of Chiapas soil samples is influenced by several agricultural practices conducted by coffee growers. Veracruz soil samples had pH values ranging from 3.6 to 6.4, and these results are similar to previously reported values (Manson et al., 2008) and to values reported for other regions of Veracruz (Zetina and Uriza, 1999; Geissert et al., 2000).

Other coffee-producing countries, such as Costa Rica and Colombia, have reported  $Al^{3+}$  concentrations of  $0.90 \text{ cmol kg}^{-1}$  and  $0.60\text{-}1.61 \text{ cmol kg}^{-1}$ , respectively (Ortiz et al, 2004; Corella and López, 1984).

These results are consistent with the reported abundance of aluminum (8%) in Earth's crust (Peng et al., 2006). Chiapas soil samples have high concentrations of iron and aluminum oxides, and complexes of silicon-aluminum with potassium, sodium and calcium, which are commonly found in igneous rocks (Ortiz and Ortiz, 1990). We report total aluminum contents of 48 to  $147 \text{ mg g}^{-1}$  in Veracruz, which are higher than those reported for Chiapas and Yucatan (Table 2).

In this study, we investigated the link between the presence of aluminum in soil, coffee plants and coffee beans and the possible negative effects of aluminum content on plant development and human consumers.

Previously, a relationship between low pH and increased  $Al^{3+}$  content has been reported, with levels of  $Al^{3+}$  increasing for pH values lower than 5.5 (Marschner, 1995). Soil pH values below 5.5 are accompanied by numerous phenomena, such as increased abundance of aluminum and manganese, reduced amounts of cation macronutrients, reduced solubility of phosphorous and molybdenum and the inhibited root growth that results from a decrease in the absorption of water and other nutrients (Casierra-Posada and Aguiar-Avenidaño, 2007; Zapata, 2004). The variation in aluminum concentration between the two coffee-producing soils selected for this study is remarkable (Table 1). Veracruz soil samples had a pH of 4.77 and Chiapas soil samples had a pH of 5.92. At pH values below 5.5, toxic  $Al^{3+}$  is present at levels high enough to be accessible and absorbed by plant cells. It has been reported that the reduced root growth of soy plantlets was related to the  $Al^{3+}$  content of the soil, which was  $1.25 \text{ cmol kg}^{-1}$  (Hashimoto et al., 2010).

As soil pH decreases, the hydrolysis of clay minerals and other silicate complexes begins, as does the solubilization of aluminum oxides to form  $Al^{3+}$ . This process potentially produces irreversible soil degradation. Therefore, the acidic growth conditions confirmed by analyzing coffee-growing soils from Veracruz and Chiapas most likely promote negative effects on the development and growth of coffee plants due to the presence of toxic aluminum species. The acidic conditions observed in the Veracruz soil samples most likely influence the levels of total aluminum found in coffee leaves collected in the region of Huatusco. Therefore, we suggest that low soil pH is a key factor responsible for the accumulation of exchangeable aluminum in coffee plants grown in soil from Veracruz ( $1.06 \text{ cmol kg}^{-1}$ ), which has also been suggested by previous reports (Bernier, 1999).

Following aluminum absorption, aluminum participates in chemical reactions including hydrolysis, polymerization and ion exchange. The hydrolysis reaction can be compared to the reaction of a strong acid that produces protons (Wallnöfer and Engelhardt, 1995). Such metals as cadmium and aluminum are present in very low levels in the stem, leaves and fruits, whereas higher levels are reported in the roots (Berkelaar, 2001). Based on our results, after aluminum is absorbed by the roots, it is transported by some mechanism to aerial parts of the plant, such as the leaves.

The aluminum content of ground coffee beans has previously been reported to range from 0.01 to  $0.03 \text{ mg g}^{-1}$  (Müller et al., 1997; Vega et al., 2002). We examined the aluminum levels in coffee beans using three different methods (AAS, SEM and ICP-AES); however, we only detected aluminum in the coffee beans using ICP-AES (Table 2). These values are consistent with previously reported data for aluminum, which ranges from 0.01 to  $0.03 \text{ mg g}^{-1}$  in ground coffee beans (Müller et al., 1997; Vega et al., 2002).

Aluminum is present in soil in which coffee crops are grown in Mexico. We also detected aluminum in the leaves and beans produced by these plants. Therefore, this metal could be transferred from the soil to plants and may promote aluminum toxicity and influence coffee production. Moreover, the levels of aluminum in the beans produced by plants grown in these soils can, in some cases, approaches values reported for tea infusions (Müller et al, 1997). These findings support the argument to continue research in other regions of Mexico where coffee crops are cultivated in order to develop a comprehensive picture of soil suitability and propose strategies for preventing potential problematic situations. The toxic effects of Al in humans have been previously described (Müller et al., 1997; Milacic, 2005). Our observations of aluminum levels in coffee beans produced by plants grown in acidic soils suggest that coffee beverages (Table 3) made from these beans are a potential source of aluminum in humans.

Our results suggest a direct relationship between acidic pH and Al<sup>3+</sup> content. We also detected aluminum in products derived from coffee. We conclude that it is necessary to continue accumulating more data to encompass additional regions in Mexico in which coffee plants are cultivated in order to gain information about the aluminum contents of coffee crop soils around the world.

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Table 1. Determination of pH ( $\pm$  standard deviation) and abundance of exchangeable species of aluminum ( $Al^{3+}$ ) ( $\pm$  standard deviation) in soils obtained from Chiapas, Veracruz and Yucatan.

| State    | pH              | $Al^{3+}$ (cmol $kg^{-1}$ ) |
|----------|-----------------|-----------------------------|
| Yucatan  | 8.15 $\pm$ 0.08 | 0.14 $\pm$ 0.07             |
| Chiapas  | 5.92 $\pm$ 0.26 | 0.13 $\pm$ 0.04             |
| Veracruz | 4.77 $\pm$ 0.75 | 1.06 $\pm$ 0.91             |

Table 2. Determination of total aluminum ( $Al_t$ ) content ( $\pm$  standard deviation) by ICP-OES in soil and leaves.

| State    | pH              | $Al_t$ in soils (mg/g) | $Al_t$ in leaves (mg/g) |
|----------|-----------------|------------------------|-------------------------|
| Yucatan  | 8.15 $\pm$ 0.08 | 86.67 $\pm$ 30.20      | 0.08 $\pm$ 0.07         |
| Chiapas  | 5.92 $\pm$ 0.26 | 35.50 $\pm$ 10.15      | 0.08 $\pm$ 0.05         |
| Veracruz | 4.77 $\pm$ 0.75 | 110.8 $\pm$ 26.14      | 0.13 $\pm$ 0.10         |

Table 3. Quantification of the total aluminum content ( $\pm$  standard deviation) of roasted and ground coffee beans and coffee beverages, as measured by ICP-OES.

| Sample    | Coffee ground ( $\square$ g/g) | Beverage ( $\square$ g/ml) |
|-----------|--------------------------------|----------------------------|
| Comercial | 2.93 $\pm$ 0.44                | 0.12 $\pm$ 0.01            |
| Chiapas1  | 3.30 $\pm$ 0.87                | 0.24 $\pm$ 0.03            |
| Chiapas2  | 7.27 $\pm$ 0.26                | 0.33 $\pm$ 0.03            |
| Chiapas3  | 3.99 $\pm$ 0.05                | 0.32 $\pm$ 0.16            |
| Veracruz1 | 10.09 $\pm$ 0.61               | 0.28 $\pm$ 0.06            |
| Veracruz2 | 2.27 $\pm$ 0.61                | 0.27 $\pm$ 0.02            |
| Instant   | 3.48 $\pm$ 0.13                | 0.54 $\pm$ 0.01            |

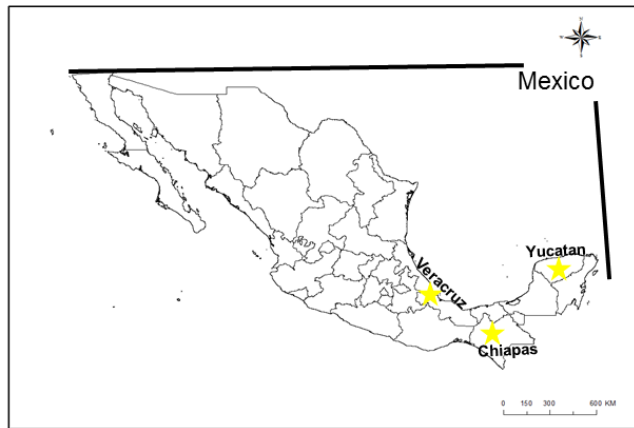


Figure 1. The locations of three regions (Chiapas, Veracruz and Yucatan) in Mexico in which coffee crops are cultivated are indicated with yellow stars.

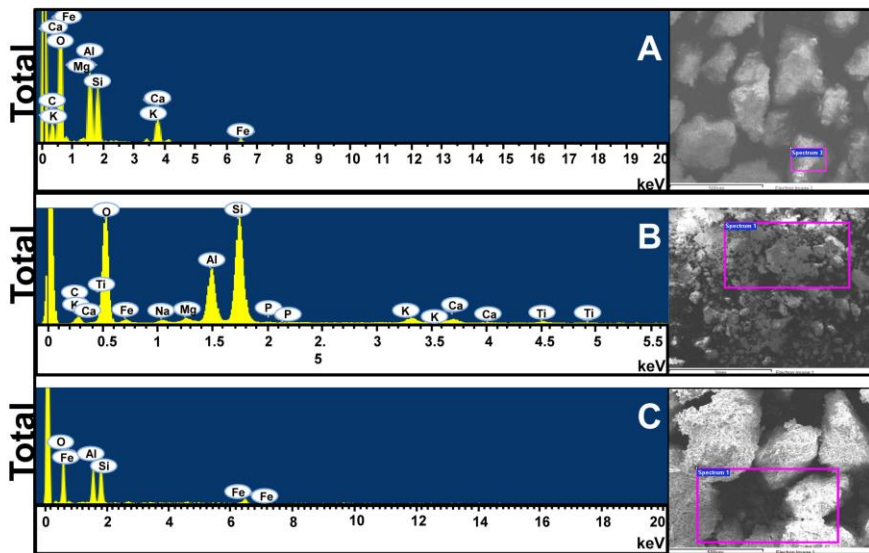


Figure 2. Qualitative analysis of aluminum in different soil samples by SEM: A) Yucatan, B) Chiapas and C) Veracruz. The circle indicates the peak corresponding to aluminum, and the image to the right shows the scanned area.