

## REVISIÓN / REVIEW

## Evaluation of the concentration of heavy metals in vegetables from Ecuador

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**Abstract:** Heavy metals are toxic elements that have contaminated agricultural waters and soils due to their use in anthropic activities, generating bioaccumulation in food and biomagnification in the food chain. In this research, information was collected on the presence of heavy metals in different foods of plant origin in Ecuador, the first link in the trophic chain, to establish a baseline in the field of food toxicology. Information was extracted from the central databases, scientific journals and libraries' digital repositories, analyzing data on Cd, Hg, Pb and the metalloid As in 15 plants. According to the Mercosur standard, 14 exceeded the maximum permissible limit (LMP) of As, distinguishing the avocado ( $19.76 \pm 1.36$  mg/kg As). According to the Codex Alimentarius, 31% of the vegetables exceeded the LMP in terms of Cd; carrots ( $9.71 \pm 8.66$  mg/kg Cd) were the most contaminated. Mercury (Hg) was studied only in potatoes (0.04 mg/kg Hg), which was found to exceed the standard four times, while Pb surpassed the limits in 33% of the samples analyzed, sugarcane the vegetable with the highest concentrations of 4.32 mg/kg Pb. In conclusion, multi-metal contamination was evidenced in some vegetables, representing a risk to consumers' health and food safety.

**Keywords:** bioremediation, contamination, food safety, heavy metals, toxicity

### Introducción

Heavy metals are hazardous pollutants due to their toxicity, environmental persistence, ability to accumulate in organisms, and biomagnification in the food chain<sup>1</sup>. Industrialization and agrochemical use have generated contamination by heavy metals in agricultural soils. These metals are easily absorbed by the roots and transported to the edible parts of the plant, ultimately entering the food chain<sup>2</sup>. Consequently, plants accumulate these elements, generating several diseases in consumers<sup>3</sup>.

Contamination in food can originate naturally, due to environmental contamination, as a consequence of bad agricultural practices or the mishandling of food on the way from field to table. Therefore, food safety assessment is challenging given the diversity of foods and agricultural products<sup>3</sup>.

Among the most toxic heavy metals are Cadmium (Cd), lead (Pb) and mercury (Hg), in addition to the metalloid arsenic (As). The latter has generated public health problems in the United States, Mexico, Chile, Argentina, Bangladesh, India, China, Taiwan and Thailand, where the cases of lung, bladder, and skin cancer associated with the consumption of water contaminated with As have increased<sup>4,5</sup>. Contamination by this metal has been reported in rice in Australia<sup>6</sup>, Bangladesh<sup>7</sup> and Brazil<sup>8</sup> as has also been detected in wheat, corn and potatoes<sup>9,10</sup>.

As causes some serious health effects such as anemia, melanosis (hyperpigmentation or dark spots and hypopigmentation or white spots), hyperkeratosis (hardening of the skin), restrictive lung disease, peripheral vascular disease (black foot disease), gangrene, diabetes mellitus, hypertension, ischemic heart disease, and cancer<sup>11,12</sup>.

On the other hand, chronic exposure to Cd mainly affects the kidneys, causing cardiac nephropathy<sup>13,14</sup>, high blood pressure, diabetes, cardiovascular effects<sup>15,16</sup>, lung and prostate cancer<sup>13,17</sup>, endocrine disruptions related to breast and endometrial cancer<sup>17</sup>, teratogenesis and mutagenesis<sup>18-20</sup>. Additionally, Cd can damage calcium (Ca) metabolism, resulting in a Ca deficiency that can cause bone fractures and cartilage damage<sup>21</sup>.

A classic example of excessive consumption of Cd through contaminated food is the "Itai-Itai" disease (osteomalacia) described in Japan because of the consumption of contaminated rice. Other foods where contamination with Cd has been described are lettuce, peanuts, spinach and soybeans because these plants can accumulate heavy metal in high concentrations, and their hyper-accumulative capacity has been demonstrated<sup>22</sup>. Likewise, high levels of Cd in potatoes have been found in Brazil<sup>10</sup>.

Another essential metal is Hg, which can come from natural sources, but is mainly associated with anthropogenic activities, particularly the extraction and use of fossil fuels and gold<sup>23</sup>. Mercury has several presentations; inorganic Hg is the form in which it is discharged into aquatic systems, and in the upper layers of aquatic sediments and water, bacteria transform inorganic Hg into methylmercury<sup>24</sup>.

Methylmercury is the most toxic form, highly bio-accumulative in organisms and biomagnifies in the food chain<sup>25</sup>. Hg, in its form of methylmercury, produces acrodynia or pink disease. The Environmental Protection Agency (EPA) has declared that methylmercury is highly carcinogenic, and its exposure can alter brain functions

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and cause shyness, memory problems, tremors, irritability and changes in vision or hearing. Excess Hg consumption leads to Minamata disease, as reported in Japan, which is characterized by symptoms of memory loss, fatigue, cortical blindness, and poor motor control<sup>26</sup>. Numerous authors have also described the contamination of foods of plant origin, where spinach, mint, onion and carrot are mentioned<sup>27-29</sup>.

Regarding Pb, since ancient times, poisoning by this metal has been known to generate diseases such as lead poisoning (or plumbism) caused by storing food in Pb containers<sup>30,31</sup>. The main signs of Pb poisoning are predominantly related to the gastrointestinal tract and the central nervous system<sup>32-33</sup>. Its chronic exposure can cause congenital disabilities, muscle weakness, weight loss, kidney damage, allergies, and brain damage, including mental retardation, autism, hyperactivity, psychosis, dyslexia, and paralysis, and can even result in death<sup>34</sup>. In vegetables, Pb contamination has been described in corn, potatoes, paprika and spinach<sup>35,36</sup>.

In Ecuador, 8% of the gross domestic product depends on agriculture<sup>37</sup>. There are 8.81 million hectares with agricultural uses<sup>38</sup> exposed to fertilizers and pesticides that may contain heavy metals that contaminate the soil and enter the food and, consequently, generate diseases in the population.

In Ecuador, there is evidence of contamination by heavy metals in children and pregnant women. In this sense, high lead levels were found in breast milk (3.73 µg / L, max. 28 µg / dL) and blood (7.8 µg / dL, max. 21 µg / dL) of lactating women from the Ecuadorians Andes<sup>39</sup>, and in the city of Guayaquil the presence of Pb in the blood (0.7 µg / L, max. 2.2 µg / dl) was evidenced in people not exposed to this heavy metal at work<sup>40</sup>. On the other hand, there is occupational exposure to lead in battery factory workers in the city of Guayaquil (3.80- 38.30 µg/dL)<sup>40</sup> and in sites of widespread production of Pb glazed tiles and other ceramics in the south of Quito<sup>41</sup>. In children from the population of La Victoria and El Tejar, values of 6.6-119.1 µg / dL were found to be associated with the consumption of water, food, and dust contaminated with lead as a result of glazing of ceramics and were reflected in the health of children as impaired visual pursuit, a deficit in cognitive/perceptual abilities in math calculation skills, neurological deficits in fine motor, perceptual, and non-verbal reasoning skills<sup>41</sup>, and in adults, sensory-neural hearing loss in men, which may be attributable to occupational noise exposure in combination with Pb intoxication<sup>42</sup>. However, these investigations have not considered the intake of food contaminated with heavy metals.

In another study, high concentrations of Pb, Cd, and As<sup>43</sup> were found in the blood of mothers and children from the Ecuadorian Andes and associated with factors such as residence in a dirt-floor home, living near a dirt-paved or cobblestone street, or in local neighborhoods where scrap metal smelters and gas depositories; as well as other anthropogenic and geogenic sources<sup>43</sup>. These authors also mentioned the possible intake of food contaminated with heavy metals as a potential source of contamination<sup>43</sup>.

According to Globoscan<sup>44</sup>, the most frequent types of cancer in Ecuador are breast, prostate, colorectal, and stomach cancer. In 2020, there were 29,273 new cancer cases with a mortality of 15,123 people<sup>44</sup>. The incidence rates of gastric cancer are expected to increase in Ecuador through 2030<sup>45</sup>. Another study showed that there had been an increase in the incidence and mortality rates of breast cancer in Ecuador in the last five years<sup>46</sup>. We wonder if this increase in cancer cases is due to the intake of heavy metals, the lack of food monitoring, and policies that require compliance with standards in food for national consumption described in the Codex Alimentarius. As a food exporting country, Ecuador has strict controls on export products, regulated with the standards of

the European Union and the Codex Alimentarius; however, those rules are not applied to products for the national consumption market.

In light of this evidence, in this research, we decided to study the sources of contamination, targeting the food supplies through the compilation of information on the presence and the amounts of heavy metals in different foods of plant origin, the first link in the trophic chain, comparing to the maximum permissible levels established by the national and international standards to determine a baseline in the field of food toxicology in Ecuador

## Methods

The Pubmed, Scopus and Web of Science databases were extensively searched for previous reports. Finding little information, the keywords (heavy metals, Cadmium, lead, mercury, arsenic, vegetables, crop plants) were searched in Google Scholar and in the Digital Repositories of Libraries, focusing on Ecuador. Subsequently, the investigations carried out between the years 2010 to 2021 that met the inclusion criteria of being either scientific articles, theses or reports were selected, taking into account those whose laboratory analyses had reference material or had been carried out by entities with the parameters of heavy metals accredited under the ISO 17025 standard, to ensure the validity of the information (Table 1).

## DATA ANALYSIS

A descriptive analysis was carried out for each variable. The means, standard deviations, maximums, and minimums were calculated by location and food for each heavy metal. Dendrograms were elaborated to analyze the grouping of vegetables concerning the metal concentrations. All analysis and graphs were performed using R Studio version 4.0.2 programs. To prepare maps, the location coordinates were homogenized to the projected coordinate system UTM WGS 84 using the Google Earth program and plotted on maps for identifying critical points of contamination by heavy metals using the ArcGIS Program. The results were compared with international standards of maximum permissible limits (MPL) in food, including among them, the Codex Alimentarius of the World Health Organization (WHO and FAO), the standard of the European Union (2019) and the MERCOSUR regulation (2011).

### Arsenic (As) detected in products of plant origin

The collected studies showed As a presence in 15 plants, of which 14 exceeded the maximum permissible limits (Table 2). Table 2 shows that the highest concentration occurred in the Persea americana fruit (avocado) with an average of 19.76 mg/kg As recorded in the town of Izamba, Ambato canton<sup>47</sup>. Avocado exceeded 66.9 times the maximum permissible limit (MPL) established by MERCOSUR (0.3 mg/kg As)<sup>48</sup>. This could be related to its long-life cycle; since being a perennial plant, it can accumulate large amounts of pollutants from the cultivation soil, especially in soils of volcanic origin, irrigation water and the type of agrochemicals used for its production.

Plant species beetroot (*Beta vulgaris*) and cabbage (*Brassica oleracea*) also presented high concentrations of As (5.75 ± 3.53 mg/kg; 4.89 ± 1.62 mg/kg, respectively). The food with the lowest As concentration was potato (*Solanum tuberosum*) with 0.80 mg/kg As determined in two farms in the Mejía canton<sup>49</sup>, Pichincha province.

Figure 1A shows an association dendrogram where several groups are distinguished: the first, where the *Persea americana* species

is found, showing the highest accumulation of As; another group formed by *Lactuca sativa*, *Oryza sativa* and *Solanum tuberosum*, which represents the species with the lowest accumulation of As and the third group made up of the species with intermediate concentrations: *Beta vulgaris*, *Brassica oleracea* var. *botrytis*, *Apium graveolens*, *Brassica rapa pekinensis*, *Allium fistulosum*, *Rubus ulmifolius*, *Brassica oleracea* var. *capitata*, *Fragaria x ananassa*, *Brassica oleracea* var. *capitata*, *Solanum lycopersicum* and *Spinacia oleracea*.

Specie	Common name	Element	Author
<i>Allium cepa</i> L.	Red onion	Cd, Pb	Mantilla & Mastrocola, <sup>44</sup>
<i>Allium fistulosum</i> L.	Green onion	As	Fiallos & Arancibia, <sup>47</sup>
		Cd, Pb	Tonato & Briceño, <sup>34</sup>
<i>Apium graveolens</i> L.	Celery	As	Fiallos & Arancibia, <sup>47</sup>
<i>Beta vulgaris</i> L.	Beet root	As	Fiallos & Arancibia, <sup>47</sup>
<i>Brassica oleracea</i> var. <i>botrytis</i>	Cauliflower	As	Fiallos & Arancibia, <sup>47</sup>
<i>Brassica oleracea</i> var. <i>capitata</i>	White cabbage	As	Fiallos & Arancibia, <sup>47</sup>
<i>Brassica oleracea</i> var. <i>capitata</i> f. <i>rubra</i>	Red cabbage	As	Fiallos & Arancibia, <sup>47</sup>
<i>Brassica oleracea</i> var. <i>italica</i> L.	Broccoli	Cd, Pb	Coronel & Mastrocola, <sup>34</sup>
<i>Brassica rapa pekinensis</i> L.	Napa cabbage	As	Fiallos & Arancibia, <sup>47</sup>
<i>Capsicum annuum</i> L.	Pepper	Cd, Pb	Coronel & Mastrocola, <sup>34</sup>
		Cd, Pb	Mantilla & Mastrocola, <sup>44</sup>
<i>Coffea arabica</i> L.	Coffee	Cd, Pb	Viñals & Pernia, <sup>41</sup>
<i>Daucus carota</i> L.	Carrot	Cd, Pb	Coronel & Mastrocola, <sup>34</sup>
<i>Fragaria x ananassa</i> Duch.	Strawberry	Cd, Pb	Escobar, <sup>45</sup>
		As	Fiallos & Arancibia, <sup>47</sup>
<i>Glycine max</i> L.	Soybean	Cd, Pb	Lopez & Pernia, <sup>41</sup>
<i>Lactuca sativa</i> L.	Lettuce	Cd, Pb	Carrillo & Valdés, <sup>36</sup>
		Cd, Pb	Coronel & Mastrocola, <sup>34</sup>
		As	Fiallos & Arancibia, <sup>47</sup>
		Cd, Pb	Velasquez & Mastrocola, <sup>37</sup>
		Pb	Villegas et al., <sup>38</sup>
		Cd, Pb	Torres & Pozo, <sup>39</sup>
<i>Musa x paradisiaca</i> L.	Banana	Cd, Pb	Romero-Estévez et al., <sup>38</sup>
<i>Nasturtium officinale</i> R. Br.	Watercress	Cd, Pb	Cabascango & Valdés, <sup>36</sup>
<i>Oryza sativa</i> L.	Rice	As Total	Estrélla et al., <sup>41</sup>
		Cd, Pb	Huiraococha, <sup>40</sup>
		As Total	Moreno, <sup>36</sup>
		Cd	Muñoz et al., <sup>42</sup>
		Pb	Sinchi & Pernia, <sup>42</sup>
<i>Persea americana</i> L.	Avocado	As	Fiallos & Arancibia, <sup>47</sup>
<i>Rubus ulmifolius</i> L.	Blackberry	As	Fiallos & Arancibia, <sup>47</sup>
<i>Saccharum officinarum</i> L.	Sugarcane	Cd, Pb	Aleivar & Pernia, <sup>41</sup>
<i>Solanum lycopersicum</i> L.	Tomato	As	Fiallos & Arancibia, <sup>47</sup>
		Cd, Pb	Coronel & Mastrocola, <sup>34</sup>
		Cd, Pb	Escobar, <sup>45</sup>
		Cd, Pb	Velasquez & Mastrocola, <sup>37</sup>
		Pb	Villegas et al., <sup>38</sup>
<i>Solanum tuberosum</i> L.	Potato	As, Cd, Hg, Pb	Castillo, <sup>43</sup>
<i>Spinacia oleracea</i> L.	Spinach	As	Fiallos & Arancibia, <sup>47</sup>
<i>Theobroma cacao</i> L.	Cocoa	Cd, Pb	Díaz et al., <sup>42</sup>
		Cd, Pb	Intriago et al., <sup>44</sup>
		Cd	Mite et al., <sup>43</sup>
			Chávez et al., <sup>42</sup>
			Barraza et al., <sup>42</sup>
			Díaz et al., <sup>42</sup>
			Arguello et al., <sup>41</sup>
			Vega, <sup>41</sup>
	Chocolate	As, Cd	Sánchez-Solehdspa et al., <sup>42</sup>
<i>Zea mays</i> L.	Corn	Cd	Condo & Pernia, <sup>46</sup>

**Table 1.** Summary of the food of plant origin analyzed in Ecuador

All the As in plant foods averages exceeded the MPL according to the MERCOSUR standard<sup>48</sup>, except for rice (Table 1). However, *Oryza sativa* presented maximum values above the LMP in the provinces of El Oro, Guayas and Los Ríos, with the highest concentration of 0.95 mg/kg As being observed in the Vinces canton<sup>50</sup>.

These high concentrations of As in food could be due to the influence of volcanoes, one of the leading natural sources of As in Latin America<sup>51</sup>. In addition, it must be taken into account that the foods analyzed were found in the area of influence of the Tungurahua volcano (except for rice), which presented volcanic activity on the sampling dates of study<sup>47</sup>; therefore, the accumulation of As could be due to deposition of volcanic ash.

Likewise, volcanic rocks are a natural contribution of As in aquifers, presenting values between 100 to 794 µg / L As in geo-

Scientific name	Common name	Mean	Standard deviation	Minimum	Maximum	MPL <sup>2</sup>
<i>Persea americana</i>	Avocado	19.76	1.36	18.93	21.33	0.30
<i>Beta vulgaris</i>	Beet root	5.75	3.53	2.55	9.54	0.30
<i>Brassica oleracea</i> var. <i>botrytis</i>	Cauliflower	4.89	1.62	3.11	6.28	0.30
<i>Apium graveolens</i>	Celery	4.58	0.60	3.88	4.95	0.20
<i>Brassica rapa pekinensis</i>	Napa cabbage	4.08	0.55	3.47	4.54	0.20
<i>Allium fistulosum</i>	Green onion	3.93	2.24	1.38	5.56	0.30
<i>Rubus ulmifolius</i>	Blackberry	3.85	0.43	3.58	4.35	0.30
<i>Brassica oleracea</i> var. <i>capitata</i>	White cabbage	3.39	0.95	2.40	4.29	0.30
<i>Fragaria x ananassa</i>	Strawberry	3.05	1.84	1.94	5.17	0.30
<i>Brassica oleracea</i> var. <i>capitata</i> f. <i>rubra</i>	Red cabbage	3.01	1.20	1.74	4.13	0.30
<i>Solanum lycopersicum</i>	Tomato	2.74	1.16	1.42	3.58	0.10
<i>Spinacia oleracea</i>	Spinach	2.16	1.27	1.12	3.57	0.30
<i>Lactuca sativa</i> L.	Lettuce	0.85	0.20	0.67	1.07	0.30
<i>Solanum tuberosum</i> L.	Potato	0.80	0.18	0.50	1.20	0.20
<i>Oryza sativa</i> L.	Rice	0.23	0.16	0.00	0.95	0.30

MPL<sup>2</sup> Maximum permissible limits established by MERCOSUR. Blue color: the concentration does not exceed the maximum permissible limits. Pink color: concentration exceeds the maximum permissible limits

**Table 2.** Summary of arsenic (As) concentration in Ecuadorian vegetables and its comparison with the maximum permissible limits (MPL).

thermal water bodies. These water bodies can be contributors to surface waters; however, anthropogenic sources play a significant role in the distribution of As in these tributaries<sup>52</sup>, hence the importance of carrying out As analysis in irrigation waters. It is essential to mention that the national regulations of Ecuador do not contemplate the arsenic content for agricultural products; therefore, the reported as content was compared with the international regulations. This is an essential factor to consider due to the strong influence of this metalloid in volcanic areas of Ecuador and the high levels of As found in the present work. In addition, the speciation of As should be considered since inorganic arsenic is more toxic than organic, and most of the analyzed studies measured total As. Likewise, digestibility tests should be carried out to evaluate the risk for the Ecuadorian population due to the consumption of these vegetables.

**Cadmium (Cd) presence in products of plant origin**

Regarding Cd, Table 3 shows that of the 17 plant species studied, 5 exceeded the MPL according to the Codex Alimentarius<sup>53</sup>. Carrot (*Daucus carota*) presented the highest concentration with 9.71 mg/kg, and it is important to pinpoint that it was obtained in organic fairs in Quito<sup>54</sup>. This represented 97.06 times more Cd than the MPL for roots, according to Codex Alimentarius (0.1 mg/kg). This value is extremely high compared to that obtained in China and Bangladesh (0.023 mg/kg Cd)<sup>55-56</sup>. The high content of Cd in carrots maybe because it is a storage root, and it has been described that these accumulate many heavy metals<sup>65-67</sup>.

High concentrations were also observed in tomato (*Solanum lycopersicum*) (2.21 mg/kg Cd) and lettuce (*Lactuca sativa*) (0.76 mg/kg Cd) analyzed in Quito. However, these same vegetables obtained in an organic fair presented very high values (atypical) of 7.31 mg/kg and 14.61 mg/kg Cd for tomato and 9.39 mg/kg and 18.77 mg/kg Cd for lettuce<sup>54</sup>, a worrying fact because organic food is assumed to be healthier. Another food of great importance for Ecuador that exceeded the limits was cocoa (*Theobroma cacao*), which presented high Cd contents in the almond (0.695 ± 0.408 mg/kg); however, the Codex Alimentarius has not determined the MPL for cocoa beans; nonetheless, it slightly exceeds the MPL determined by the European Union<sup>52</sup> of 0.6 mg/kg Cd



Scientific name	Common name	Mean	Standard deviation	Minimum	Maximum	MPL <sup>2</sup>
<i>Daucus carota</i> L.	Carrot	9.710	8.660	0.020	19.900	0.10
<i>Solanum lycopersicum</i> L.	Tomato	2.210	4.920	0.000	14.610	0.05
<i>Lactuca sativa</i> L.	Lettuce	0.756	3.364	0.004	18.770	0.20
<i>Theobroma cacao</i> L.	Cocoa	0.695	0.408	0.099	1.860	0.60
<i>Solanum tuberosum</i> L.	Potato	0.323	0.274	0.049	0.898	0.10
<i>Saccharum officinarum</i> L.	Sugarcane	0.150	0.000	0.150	0.150	0.05
<i>Glycine max</i> L.	Soybean	0.137	0.038	0.094	0.183	0.10
<i>Brassica oleracea</i> var. <i>italica</i> L.	Broccoli	0.042	0.008	0.031	0.050	0.05
<i>Fragaria x ananassa</i> (Weston) Duchesne	Strawberry	0.036	0.035	0.011	0.060	0.05
<i>Allium cepa</i> L.	Onion	0.026	0.014	0.016	0.048	0.05
<i>Allium fistulosum</i> L.	Green onion	0.025	0.004	0.018	0.030	0.05
<i>Musa x paradisiaca</i> L.	Banana	0.024	0.009	0.009	0.047	0.05
<i>Oryza sativa</i> L.	Rice	0.018	0.050	0.000	0.190	0.40
<i>Capsicum annum</i> L.	Pepper	0.014	0.006	0.000	0.023	0.05
<i>Zea mays</i> L.	Corn	0.012	0.012	0.000	0.050	0.05
<i>Coffea arabica</i> L.	Coffee	0.000	0.000	0.000	0.000	0.20
<i>Nasturtium officinale</i> R. Br.	Watercress	0.000	0.000	0.000	0.000	0.00

MPL\* Maximum permissible limits established by MERCOSUR and the Codex Alimentarius. Blue color: the concentration does not exceed the maximum permissible limits. Pink color: concentration exceeds the maximum permissible limits

**Table 3.** Cadmium concentration in vegetables from Ecuador and its comparison with the maximum permissible limits (MPL).

for cocoa powder with an average of 0.9 mg/kg of Cd<sup>61</sup> in Ecuadorian cocoa and attributed this value to the capacity of cocoa plants to accumulate high concentrations of Cadmium and to its cultivation in young soils. On the other hand, processed cocoa powder showed higher values than cocoa beans in a cocoa commercial brand<sup>54</sup>. Values below the standard in the cocoa powder were found<sup>62</sup>, with 0.236±0.082 mg/kg Cd for Brand 1 and 0.169±0.066 mg/kg for Brand 2; however, Brand 3 presented 1.44±0.212 mg/kg, a much higher value than that found in the almond, which shows that there may be postharvest contamination. It is important to note that cocoa drinks are consumed mainly by children, increasing the risk of suffering from any diseases generated by excess Cd. These results indicate that the postharvest handling machinery, inputs, and additives could generate heavy metal contamination in the final product for processed or highly processed products.

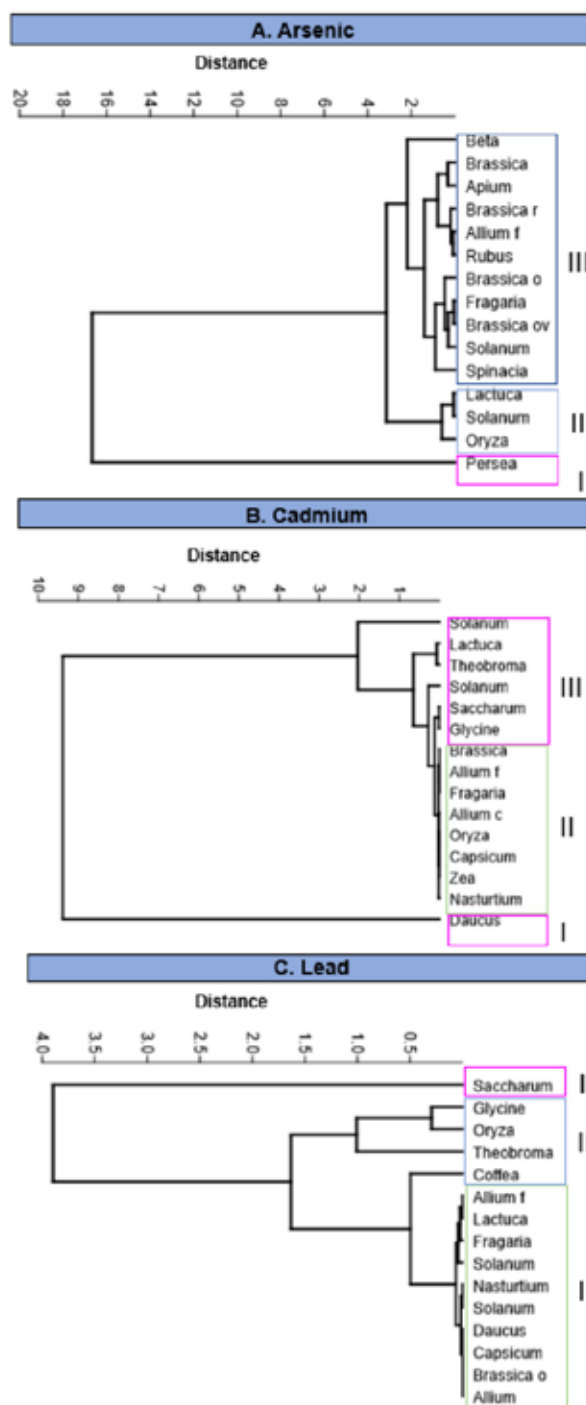
Figure 1B shows a dendrogram composed of three large groups: the one with a maximum accumulation of Cd represented by *Daucus carota*; a second group that exceeds the MPL, made up of *Glycine max*, *Lactuca sativa*, *Saccharum officinarum*, *Solanum lycopersicum*, *Solanum tuberosum* and *Theobroma cacao*; and the third group of foods suitable for consumption since they present MPL values below the Codex Alimentarius: *Brassica oleracea* var. *italica*, *Allium fistulosum*, *Fragaria x ananassa*, *Allium cepa*, *Oryza sativa*, *Capsicum annum*, *Zea mays* and *Nasturtium officinale*.

Among the possible causes of high Cd values in food is natural contamination: contamination of soils by geographical origin<sup>55</sup>, or because of the deposition of ash and volcanic material<sup>64</sup>.

However, there is a greater probability that the contamination of agricultural soils presents anthropogenic contamination due to the use of fertilizers, especially phosphates that, in Ecuador, contain up to 41.30 ± 1.65 mg/kg of Cd, exceeding 41 times the limits recommended by the Washington State Ecology Department<sup>65</sup>. It has also been shown that there is contamination by heavy metals in river water used for irrigation of crops<sup>66-68</sup>, contaminated healthy water and wastewater. In addition, another critical factor is the proximity to roads and populated areas. In this sense, a correlation has been found between the Cd content in the rice cultivation soil and the distance to the roads<sup>65</sup>.

### Mercury (Hg) presence in food products

Mercury was the metal least studied for its occurrence in vegeta-



bles in Ecuador; there is only one study in *Solanum tuberosum*<sup>49</sup> collected in farms in the Aloasí-Pichincha canton. According to the European Union, an average of 0.04 mg/kg Hg was determined, which exceeds the MPL of 0.01 mg/kg Hg. The Hg content evaluated in *Solanum tuberosum* exceeded 4 times the MPL; this value is similar to the maximum values evidenced in foods produced in areas contaminated by coal mines for leaf vegetables (0.06 mg/kg), fruit vegetables (0.06 mg/kg) and grains (0.04 mg/kg)<sup>61</sup>. In contaminated soils, up to 20 mg/kg of Hg has been quantified in this vegetable<sup>70</sup>. One possible explanation is the contamination of agricultural soils. There is evidence that the Hg content in cultivation soils is mainly due to atmospheric depositions and the use of fertilizers<sup>63</sup>; these depositions can come from anthropogenic and natural sources.

Within the natural origins, it has been determined that volcanoes constitute a significant source of Hg to the surrounding environ-

ment<sup>64</sup>. Atmospheric depositions can also contribute to the Hg content in the soil.

This confirms the need to know the characteristics of the crop soils to determine their contribution to pollution, in addition to considering the influence of volcanoes and the industrial activities of the nearest cities. It is recommended that the authorities and universities carry out more studies to determine mercury in foods of plant origin and consider carrying out studies in mining areas where mercury levels are high in the rivers near the plantations.

#### Lead (Pb) detected in products of plant origin

Regarding the analysis of the presence of Pb in foods of plant origin, 6 of the 16 foods studied exceeded the maximum permissible limits according to Codex Alimentarius and MERCOSUR (Table 4). The highest concentration was evidenced in *Saccharum officinarum* from La Troncal-Guayas<sup>65</sup> with 4.32 mg/kg Pb, a quantity that exceeds the MPL of 0.10 mg/kg Pb for sugars<sup>56</sup>. Sugarcane showed the highest Pb values, with 43.2 times the MPL, a value higher than that reported before<sup>66</sup> (0.01 - 1.11 mg/kg) in the sugarcane stem in India. It is important to note that *Saccharum officinarum* can accumulate high concentrations of heavy metals, and therefore it can be considered a potential phytoremediator of Cd and Pb<sup>67-78</sup>.

Likewise, *Theobroma cacao*, studied in the provinces of Los Ríos and Manabí<sup>79,80</sup> obtained an average of 2.38 mg/kg Pb, which exceeds the MPL of 0,50 mg/kg Pb<sup>49</sup>.

The Pb content present in *Oryza sativa*, studied in the provinces of Azuay<sup>73</sup> and Guayas<sup>82</sup> was 1.52 mg/kg, which exceeded the MPL of 0.2mg/kg<sup>49,61</sup>. Notably, atypical values were evidenced in the cantons of Nobol (10.69 and 5.65 mg/kg) and Daule (10.10 mg/kg) belonging to the province of Guayas<sup>82</sup>. This research demonstrated that lead contamination was due to the high bioavailable levels of this metal in cultivated soils and the proximity to the road network<sup>82</sup>. Likewise, critical points for lead contamination have been established due to vehicle emissions<sup>75</sup>. Rice contamination is considered very serious because it is part of many typical dishes of the region, and there is a high consumption nationwide, increasing the intake of this metal and the possibility of suffering from diseases generated by Pb.

The results obtained for *Glycine max* L. were 1.22 mg/kg in different markets of Guayaquil<sup>84</sup>, with the LMP being 0.1 mg/kg Pb<sup>53</sup>. Likewise, coffee (*Coffea* sp.) obtained an average value of 0.52 mg/kg Pb<sup>77</sup>, which is above the MPL (0.50 mg/kg Pb) established by MERCOSUR (2011). In another investigation, the same authors established a relationship between the concentration found in the soil and in the coffee<sup>77</sup>, the source of the contamination being the naturally contaminated soil in the province of Loja. Fortunately, they indicated that Pb was not detectable in coffee infusion, so it does not represent a danger to consumers.

On the other hand, in bananas (*Musa x paradisiaca*), concentration ranges between 0.0369-0.538 mg/kg of Pb<sup>78</sup>. Likewise, these authors found differences between provinces; the highest results were from the samples collected in the provinces of Bolívar and Santa Elena, which had 0.420 mg/kg and 0.538 mg/kg of Pb, respectively; on the contrary, the lowest concentrations in banana were found in the province of Azuay with 0.0369 mg/kg Pb<sup>78</sup>.

Figure 1C shows a dendrogram where 3 groups of plants with different degrees of contamination can be appreciated: the first one is represented by the species *Saccharum officinarum* with the highest accumulation of Pb; the second consists of species that exceed the permissible limits: *Glycine max*, *Oryza sativa*, *Theobroma cacao* and *Coffea arabica*; and a third group made up of species suitable for consumption, which do not exceed the permissible

Scientific name	Common name	Mean	Standard deviation	Minimum	Maximum	MPL <sup>3</sup>
<i>Allium cepa</i> L.	Onion leaves	0.0005	0.0002	0.0003	0.0008	0.10
<i>Capsicum annum</i> L.	Pepper	0.0006	0.0003	0.0003	0.0013	0.05
<i>Brassica oleraceae</i> var. <i>italica</i> L.	Broccoli	0.0006	0.0003	0.0003	0.0009	0.10
<i>Daucus carota</i> L.	Carrot	0.0008	0.0003	0.0002	0.0011	0.10
<i>Nasturtium officinale</i> R. Br.	Watercress	0.0147	0.0063	0.0000	0.0200	0.30
<i>Solanum lycopersicum</i> L.	Tomato	0.0159	0.0349	0.0000	0.1190	0.05
<i>Allium fistulosum</i> L.	Green onion	0.0478	0.0068	0.0400	0.0580	0.10
<i>Lactuca sativa</i> L.	Lettuce	0.0498	0.0590	0.0000	0.2730	0.30
<i>Fragaria x ananassa</i> Duch.	Strawberry	0.0710	0.0056	0.0670	0.0749	0.10
<i>Solanum tuberosum</i> L.	Potato	0.0970	0.1020	0.0000	0.2996	0.10
<i>Musa x paradisiaca</i> L.	Banana	0.1918	0.1362	0.0369	0.538	0.10
<i>Coffea arabica</i>	Coffee	0.5233	0.6557	0.0000	1.4900	0.50
<i>Glycine max</i> L.	Soybean	1.2248	1.5011	0.0000	3.0660	0.10
<i>Oryza sativa</i> L.	Rice	1.5180	2.8777	0.0000	10.6900	0.20
<i>Theobroma cacao</i> L.	Cocoa almond	2.1290	1.8960	0.0000	5.4470	0.50
<i>Saccharum officinarum</i> L.	Sugarcane	4.3200	0.0000	4.3200	4.3200	0.10

MPL: Maximum permissible limits established by MERCOSUR and the Codex Alimentarius. Blue color: the concentration does not exceed the maximum permissible limits. Pink color: concentration exceeds the maximum permissible limits

**Table 4.** Lead concentration in vegetables from Ecuador

limits: *Allium cepa*, *Capsicum annum*, *Brassica oleraceae* var. *italica*, *Daucus carota*, *Nasturtium officinale*, *Solanum lycopersicum*, *Allium fistulosum*, *Lactuca sativa*, *Fragaria x ananassa* and *Solanum tuberosum*.

One of the sources of crop contamination by Pb and Cd are the agrochemicals used. Several studies have reported that the excessive use of fertilizers (phosphate and nitrogenous) and manure of animal origin for an extended period can cause the accumulation of Cd and Pb in the soil and the plant<sup>79,80</sup>.

Another important aspect is the location of the crop since it has been shown that urban soils contain a more significant amount of Pb<sup>65</sup>. It is essential to consider that the vegetables grown in urban gardens present high concentrations of Pb, increasing the risk of suffering diseases in consumers, and therefore, it is recommended to carry out monitoring of urban gardens because the levels of Pb in the soil are very high.<sup>89</sup>

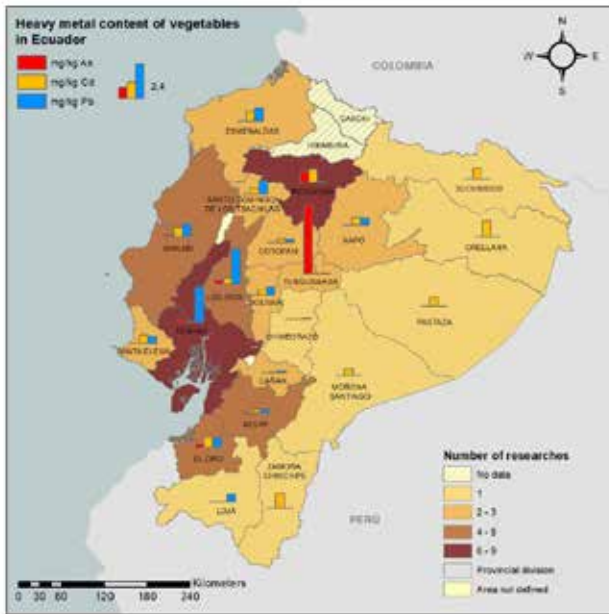
In sum, the factors that influence the content of heavy metals in vegetables are irrigation with contaminated water, the addition of fertilizers and pesticides, industrial emissions, transportation, the harvesting process, storage and the method of sale<sup>90</sup>.

#### Regarding the distribution of metals in vegetables in Ecuador,

Figure 2 shows that the foods with the highest average content of As, Cd and Pb, are located in the province of Tungurahua, Orellana and Guayas, respectively. The highest average concentrations of As in vegetables that occurred in the province of Tungurahua (4.772 mg/kg) could be influenced by volcanic activity; while Cd presented higher concentrations in Zamora Chinchipe (1.050 mg/kg), where mining and agricultural activity takes place. In the province of Guayas and Los Ríos, high concentrations of vegetables were evidenced with 2.520 and 2.170 mg/kg Pb, respectively, where contamination could be due to industrial activity.

Likewise, it was observed that the most significant number of research works had been carried out in the provinces of Guayas and Pichincha, as they are the country's main provinces; meantime, the heavy metal content in the Carchi and Imbabura provinces is yet unknown.

As seen in Figure 3, there are species capable of accumulating several metals/metalloids simultaneously, which could generate synergy and a greater toxic effect than that recorded individually. The species that accumulated the three elements (As, Cd and Pb)



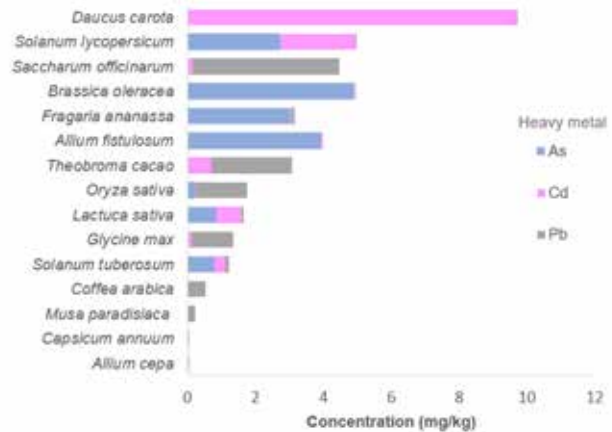
**Figure 2.** Distribution of foods of plant origin contaminated with heavy metals in Ecuador. The colors of the provinces correspond to the number of studies carried out and the bars to the concentrations of As (red), Cd (yellow) and Pb (blue).

were *B. oleracea* (with the prevalence of As), *Fragaria x ananassa*, *L. sativa*, and *S. tuberosum* accumulated four metals since Hg was also found. It is essential to mention that the works analyzed in this study determined the total concentrations of heavy metals; although, to calculate the risk associated with exposure, it is necessary to know what percentage of these elements are bio-accessible<sup>91</sup>. Only one report<sup>92</sup> determined gastric bio-accessibility in chocolate raw material (cocoa beans and liqueurs) showing that almost 100% of the total Cd content is bio-accessible after ingestion, which represents a risk for consumers. Therefore, it is proposed to monitor foods in which heavy metals were measured and determine the real risk to the population's health; additionally, the total amount and the bio-accessibility level in foods of plant origin in Ecuador must be established.

Ecuador is a country with a high incidence of cancer, and it has been estimated that the cases would increase according to a study<sup>45</sup>; one of the sources of carcinogenic agents is vegetables, which is why it was proposed to the authorities:

1. Monitor all foods of plant origin from the field to the consumer's plate, considering the province of production and possible sources of heavy metals.
2. Ensure compliance with INEN and Codex Alimentarius standards is mandatory for foods for internal consumption in Ecuador to ensure food safety for Ecuadorians.
3. Farmers are advised to use heavy metal-free fertilizers, measure metal levels in their soils before sowing seeds, and choose plant varieties with a lower bioaccumulation rate of these items.

This research aimed to construct a baseline of contamination by heavy metals in foods of plant origin in Ecuador. Our research



**Figure 3.** Evidence of multi-metal contamination in vegetables from Ecuador.

## Conclusions

found that there is contamination in avocado, cocoa, potatoes, carrots, sugar cane, rice and soybeans, which represents a risk to the health of Ecuadorians. The genera with the highest concentration of arsenic were *Persea*, in the case of *Cadmium Daucus* and lead *Saccharum*.

In addition, it was shown that there are several metals with simultaneous risen levels in the same food, representing a risk due to multi-metal contamination and the synergy of metals, and their possible impacts on the population's health should be studied. On the other hand, the lack of studies about mercury contamination in food in Ecuador was evidenced. In addition, there are very few studies in the Ecuadorian Amazon and none in Imbabura and Carchi, so we recommended analyzing plant products from these areas of Ecuador.

The authorities are called upon to carry out a nationwide monitoring study about heavy metal contents in food to protect the population's health. Unfortunately, the contamination of agricultural soils is increasing, together with the risk of increasing the levels of heavy metals in agricultural products.

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