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Arsenic accumulation in lettuce (*Lactuca sativa* L.) and broad bean (*Vicia faba* L.) crops and its potential risk for human consumption

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Abstract

Exposure to arsenic (As) is considered one of the primary health risks humans face worldwide. This study was conducted to determine As absorption by broad beans and lettuce crops grown in soil with As contents and irrigated with water contaminated with this toxic element, in Pastos Chicos, Jujuy (Argentina). Total dry biomass (TDB) and total As were determined in soils, roots, leaves, pods and seeds. These data were used to determine several parameters, such as translocation (TF) and bioconcentration (BCF) factors, target hazard quotient (THQ), and carcinogenic risk (CR). Broad bean plants had the lowest biomass production when exposed to As in irrigation water and soil. Lettuce plants presented TDB reductions of 33.3 and 42.8% when grown in soil polluted with As, and in control soil under irrigation with contaminated water, respectively.

The presence of this toxicant in broad bean seeds and lettuce leaves (edible parts) exceeded the limits established by Código Alimentario Argentino, i.e. 0.10 and 0.30 mg/kg, respectively. THQ values for lettuce leaves were higher than 1, the same as those for broad bean seeds when grown in soil with As contents and irrigated with arsenic-contaminated water, thus suggesting that consumers would run significant risks when consuming these vegetables. Furthermore, this type of exposure to As implied a CR that exceeded the acceptable 1×10^{-4} risk level. Hence, we may conclude that consuming lettuce and broad beans grown at the evaluated site brings about considerable health risks for local residents.

Keyword: Environmental science

1. Introduction

Arsenic (As) contamination levels have increased drastically in the last three decades, posing a potential risk to humans and the environment (Rafiq et al., 2017). Is considered a toxic element to humans, other animals and plants. As a consequence of contamination, human health problems have occurred in various countries, often caused by high As concentrations in the diet, originating from both drinking and irrigation waters, from crops, vegetables and meat products (Sadee et al., 2016). Chronic exposure to As has been associated with a variety of health problems, including several types of cancer (skin, lung, bladder, kidney), certain irregular perinatal conditions, and neurological and cardiovascular diseases, among others (Panda et al., 2010; Bardach et al., 2015).

This contamination is caused by natural or anthropogenic activity (Shakoor et al., 2015). Among the most natural sources are volcanic activity, rock erosion and minerals (Martínez-Toledo et al., 2017). Anthropogenic activities, such as coal combustion, mining, smelting, the textile industry, and the use of arsenical pesticides and arsenic-contaminated groundwater for irrigation in agriculture, are contributing to the more pervasive presence of this toxic chemical in the environment (Zhao et al., 2009; Niazi et al., 2016).

In Argentina, there are large areas with groundwaters contaminated with As, which restrains their use for human consumption. According to Bundschuh et al. (2012), this toxicant can be found polluting the Chaco-Pampeana Plain, transitional areas of the Andes mountain range and the mountain range itself. In particular, the northwest province of Jujuy (Puna region) presents high As levels in superficial water (between 5 and 10,000 µg/L) which are easily contaminated by contact with As rich soil and sediments (Farías et al., 2009; Ruggeri et al., 2009). Also, Pastos Chicos at Jujuy province has a history of natural water pollution by As, according to reported by Farías et al. (2009). The main source of drinking water for the population contained

820 μg/L of As which was used for the food preparation and crops irrigation (Choque et al., 2014).

Arsenic concentrations in edible vegetable components depend on As availability in the soil and the ability of the plant to take up As and translocate it to target organs. Arsenic mobility in agricultural soils can vary greatly from one location to another, depending on soil conditions. Physicochemical properties, such as pH, redox potential, organic matter content, texture, and the concentrations of some elements (aluminum, iron, manganese and phosphorus) can dramatically affect As solubility (Zheng et al., 2011; Caporale et al., 2013).

In the regions called "Quebrada" and "Puna", in the province of Jujuy, lettuce (*Lactuca sativa* L.) and legumes such as broad beans (*Vicia faba* L.) are widely grown. Lettuce is an important leafy vegetable that is consumed mainly fresh in salads, and is one of the most commonly consumed vegetables. Its benefits for human health are linked to its contents of vitamin C, phenolic compounds and fibers (Mulabagal et al., 2010). On the other hand, broad beans are characteristic of the Andean regions of South America and are widely consumed for their energetic value and contents of proteins, folic acid, niacin, vitamin C, magnesium, potassium, iron and dietary fiber (Gimenez et al., 2013).

Several methods have been proposed for the assessment of potential human health risks posed by exposure to As. Current non-cancer risk assessment methods are typically based on calculating the target hazard quotient (THQ), a ratio between the estimated dose of a contaminant and the reference dose below which there will be no appreciable risk (USEPA, 2000). The aims of this study were determining total As accumulation by lettuce and broad bean crops and evaluate the potential health risks by the consumption from these crops by local inhabitants of Pastos Chicos.

2. Materials and methods

2.1. Soil collection and preparation

The superficial soil samples (at 0−20 cm depth) were randomly collected from town of Pastos Chicos (23°45′58.8″S- 66°26′14.0″W) and samples were air-dried, passed through a 2 mm sieve and mixed to ensure homogeneity. The soils were stored in polyethylene bags at room temperature for chemical analysis and to study As uptake by lettuce and broad bean plants in the glasshouse.

2.2. Experimental design of broad beens and lettuce exposed to arsenic

A pot experiment was carried out in a greenhouse from the School of Agricultural Sciences, University of Jujuy (Argentina), in a completely randomized design

(CRD) whit a total of 60 pots under ambient temperature and irrigation without leachate, with a light and darkness regime.

Broad bean seeds used in the trials were provided by the "Pro Huerta" program of Ministerio de Desarrollo Social de la Nación Argentina, whereas lettuce seeds were supplied by Takii Seed-USA, and corresponded to the New Red Fire variety.

The crops were grown in soil typical of Pastos Chicos, which contained a 49 mg/kg concentration of As and irrigation water came from the local river (23°42′31.3″S - 66°26′42.3″W), which presents a 1.44 mg/L As concentration (Yañez et al., 2018). As control treatment, a soil with similar texture to the Pastos Chicos soil, which contained 8 mg/kg of As, and distilled water for irrigation was used (Bustingorri and Lavado, 2014).

The lettuce seeds were germinated in multipot trays and transferred to black polyethylene bags, each containing 1 kg of soil. The broad bean seeds, one per bag, were sown directly into 3 kg of soil. The lettuce test lasted 60 days, and that of broad beans 180 days. Once plants had grown, the plants were uprooted, washed with deionized water, and then oven-dried at 70 °C for 72 h. The dry weights of plants were recorded. The root, leaves, pods and seeds samples were ground and powdered using an electric grinder for later total As determination.

2.3. Determination of bioconcentration and translocation factors in crops

To fully understand As mobilization in the evaluated crops, bioconcentration (BCF) and translocation (TF) factors were estimated based on the toxic concentrations obtained. BCF is the ratio of total As concentration in root of vegetable dry weight (d.w) basis with the total As concentration in the soil (Eq. (1)). On the other hand, the translocation of As in crops was evaluated according to Eq. (2)

$$\text{FBC} = \frac{AsT \ root}{AsT \ soil} \tag{1}$$

$$\text{TF} = \frac{AsT \ aerial}{AsT \ root} \tag{2}$$

here AsT root, AsT soil and Ast aerial represent the As concentration in the extracted vegetable and soils, respectively.

2.4. Exposure and risk assessment

Human health risk assessment is the process used to estimate the nature and probability of adverse health effects in humans who may be exposed to hazards in contaminated environmental media now or in the future (USEPA, 2015).

2.4.1. Non-carcinogenic and carcinogenic risk

The non-carcinogenic health risks associated with the consumption of lettuce and broad beans by people from Pastos Chicos were assessed using the target hazard quotient (THQ) which is a unitless value that is calculated using the following Eq. (3):

$$THQ = \frac{EFr \times EDtot \times FIR \times C}{RfDo \times BWa \times ATn} \times 10^{-3} \tag{3}$$

here *EFr, EDtot, FIR, C, RfDo, BWa and ATn* represent exposure frequency, exposure time, daily intake rate, concentration in sample, oral reference dose, body weight, average time for non-carcinogens, respectively. The values of these parameterswere those previously used by Yañez et al. (2018). The daily intake of broad beans and lettuce by local residents (*FIR*) was estimated at 150 and 200 g, respectively, and *EFr* for broad bean seeds and lettuce was estimated to reach rates of 72 and 24 days/year, respectively (Bassett et al., 2013).

Carcinogenic risk (CR) reflects the possibility of carcinogenic risk and is based on the product of the exposure dose of pollutant and carcinogenic potency slope. The equation used to estimate cancer risk is as follows (4):

$$CR = \frac{EFr \times EDtot \times FIR \times CSFo}{BWa \times ATn} \times 10^{-3} \tag{4}$$

CSFo is the oral carcinogenic slope factor as registered in the Integrated Risk Information System (USEPA, 2010) database, which is 1.5 (mg/kg/day) for As. A CR lower than 10^{-6} is considered to be negligible; one between 10^{-6} and 10^{-4} is generally considered acceptable (average risk of developing cancer over a human lifetime is 1 in 100,000), whereas one above 10^{-4} is held as dangerous, is an indication of high-potential cancer risk and As has the high potentiality to develop cancer among the exposed population during lifetime (USEPA, 2010; Monferran et al., 2016).

2.5. Total arsenic determination

The total arsenic quantification in the irrigation water, agricultural, soil and the plant parts (root, leaves, pods and seeds) was performed with hydride generation atomic absorption spectrometer (HG-AAS) with continuous flow system FIAS 400 device attached to an AAnalyst 100 spectrometer (PerkinElmer). The mineralization processes of the vegetable matrix and water used in this work are shown in Fig. 1.

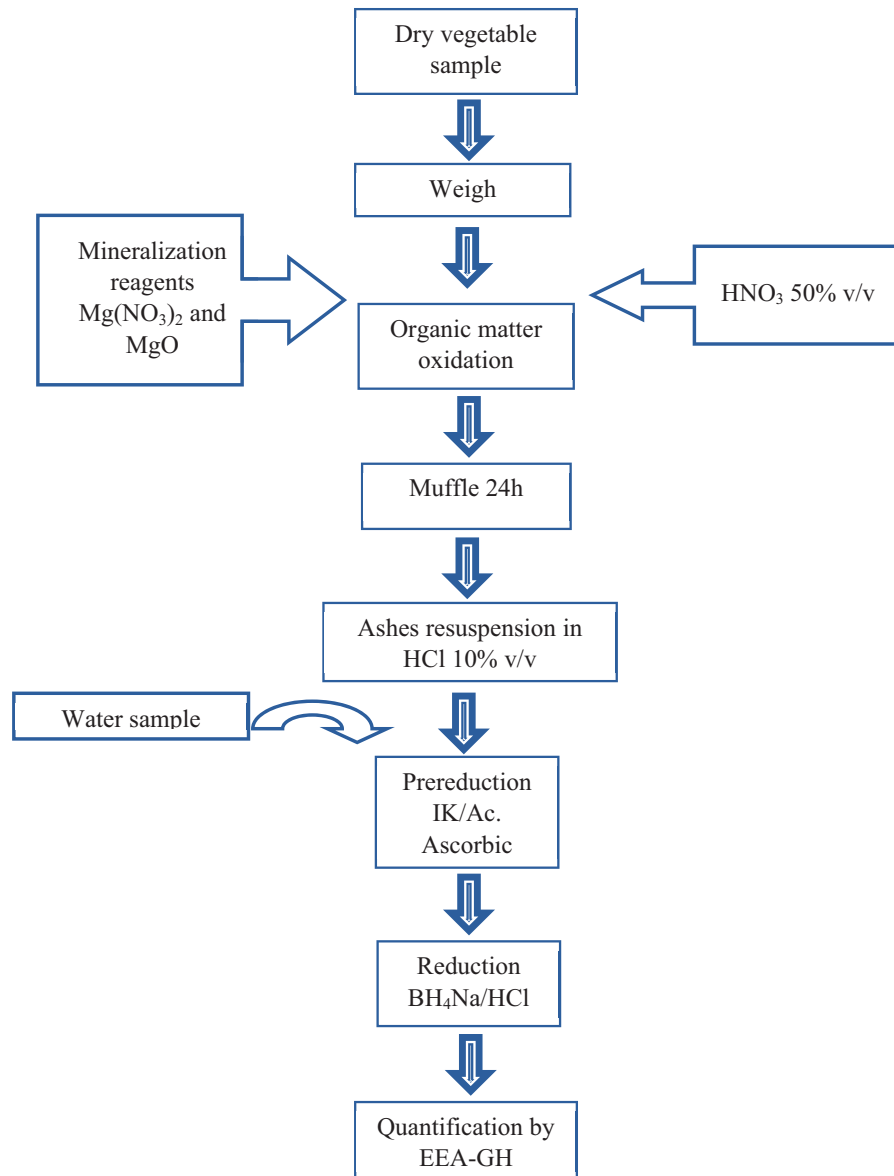Soil samples were processed for total As according to EPA method 3050 B (1996).

Fig. 1. Mineralization processes.

The calibration curve was used in the range of 0,3–5 μg As/L and showed good linearity response of up to 5 μg/L (r = 0.998). The detection and quantification limits was 0.1 and 0.3 μg/L respectively.

2.6. Statistical analysis

The results are expressed as mean values with standard deviations (SDs) as a measure of dispersión (means ± SD). Significant differences were determined using an analysis of variance (ANOVA) by Duncan comparisons test, at $p < 0.05$ significance level using a Version 2008 of InfoStat software.

3. Results and discussion

3.1. Soil characterization

The physicochemical properties the soil used in this study are presented in Table 1. The Pastos Chcicos soil presented a sandy-loan texture. The dominance of sand and small amount of clay contribute to low arsenic retention (Mehmood et al., 2017). For instance, studies conducted by Mehmood et al. (2017) showed at higher As concentration in shoot of maize plants grown in the soil of Narwala (sandy loam) in Punjab, Pakistan.

According to electrical conductivity value (1.98 dS/m), the soil sample in this study was classified as alkaline and slightly saline (USDA, 1954). A high concentration of salt can cause the decrement of crop biomass and would cause negative effects on the growth and physiology. As the salinity increases, growth decreases until the plant becomes chlorotic and dies. High salt levels can affect the stress conditions and alter the nutrient absorption by crops (Shahbaz et al., 2012; Ouzounidou et al., 2014). Although the soil showed at highest salinity level, these agricultural sites present high content of macronutrients as organic matter, nitrogen and phosphorus which should increase salt tolerance.

3.2. The influence of arsenic on the growth of broad bean and lettuce plants

Arsenic is a toxic element and is considered to be non-essential for plant growth (Niazi et al., 2016). Total dry biomass (TDB) is a critical parameter for assessing the impact of As stress on plant growth (Niazi et al., 2017).

Growth of broad bean plants exposed to As in this study is illustrated in Fig. 2. All treatments presented statistically significant differences ($p \leq 0.05$) in TDB with respect to the control. The lowest biomass was obtained in broad bean plants that had been grown in arsenic contaminated soil and irrigated with water containing As, which showed the negative effect the toxicant has on plant development. Similar results were reported by Prieto García et al. (2007), who observed that the presence

Table 1. Physicochemical parameters of the Pastos Chicos soils.

Granulometric analysis		Nutrients		Parameter	
Clay (%)	15.2	Organic Matter (%)	5.12	EC (dS/m)	1.98
Silt (%)	7.5	Organic carbon (%)	2.285	pH water (1:2.5)	8.26
Sand (%)	77.3	Total nitrogen (%)	0.25	Ca meq/L	8.88
USDA textural class	FA	Relación C/N	9.14	Mg meq/L	3.4
		P (mg/kg)	72.8	Carbonate	+

FA: soil presented a sandy-loam texture, P: extractable phosphorus, EC: electrical conductivity; Ca: calcium, Mg: magnesium, Carbonate +: reaction to HCl.
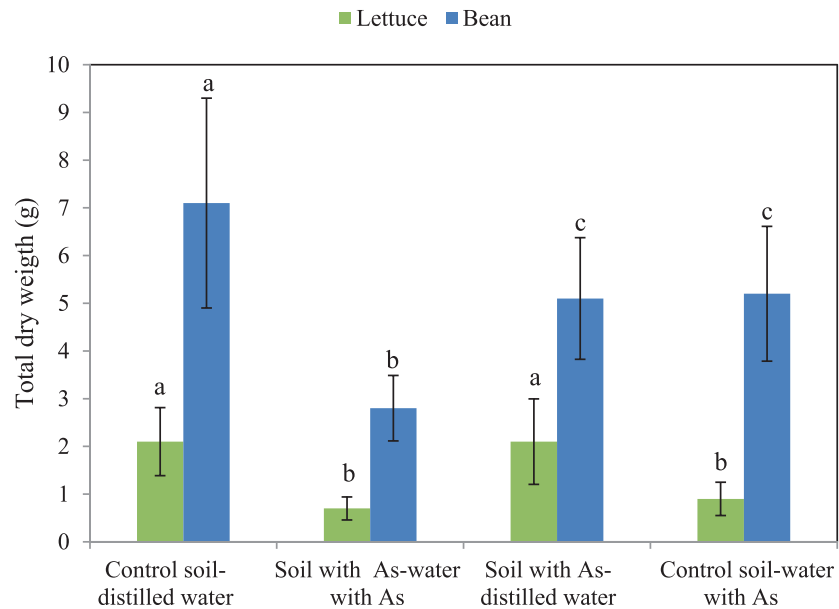
Fig. 2. Total plant dry weight exposed to arsenic. Each value represents the mean of fifteen replicates ± standard deviation. The different letters within a column indicate a significant difference at $p \leq 0.05$ according to Duncan's multiple range tests.

of As led to growth deficits or damage. They also reported problems in root branching and decreases in stem size or height of broad bean plants. Also, Austruy et al. (2013) published that As accumulation in *V. faba* was accompanied by an inhibition of the development of root biomass, which presents a high susceptibility to As toxicity.

Lettuce plant growth was significantly affected by irrigation with arsenic-contaminated water. The results of the present study showed statistically significant differences in TDB in relation to the control when the crop grew in soil polluted with As and in control soil, and also when the plants were irrigated with water containing the toxicant (Fig. 2). In these treatments, there was a biomass reduction of 33.3 and 42.8%, respectively, as compared with the control. Similar results were published by Gusman et al. (2013), when they grew lettuce under hydroponic conditions, using water that presented a 52.8 μmol/L As concentration. They determined a biomass reduction of 39 and 29% for AsV and AsIII, respectively. Also, Colding (2014) evaluated Pb and As accumulation in lettuce grown in soil contaminated with lead arsenate, determining a yield reduction of 31 and 38%. This yield decrease could be due to the fact that the abovementioned toxicants interfere with the photochemical and biochemical stages of photosynthesis (Gusman et al., 2013).

Arsenic interferes with many plant metabolic processes, as it is assumed to impact directly on stomatal apparatus, plant transpiration rate, and root and shoot development (Armendariz et al., 2016). It has to be considered that metabolic energy can be

used for the generation of compounds related to As stress, such as antioxidants and phytochelatins (Srivastava et al., 2016). Arsenic also causes oxidative stress and lipid peroxidation due to the overproduction of reactive oxygen species (ROS), such as hydrogen peroxide (H_2O_2), singlet oxygen, superoxide, and hydroxyl radicals (Flora, 2011). Furthermore, it can change the balance of nutrients and their assimilation, protein metabolism and oxidative phosphorylation in plant tissues (Mirza et al., 2016).

3.3. Total arsenic absorption by broad beans and lettuce

In broad bean crops, accumulated As rose as arsenic concentrations in the medium increased. This coincides with what was reported by Castillo et al. (2013), who showed that As concentration in vegetables increased with higher arsenic contents in irrigation water and soil. Similar results were also published by Rafiq et al. (2017), who exposed *Vicia faba* L. plants to different As concentrations in presence of EDTA and calcium.

In this work, the highest As accumulation in roots, pods and seeds was obtained when broad bean plants were exposed to As by both irrigation and contact with a contaminated soil. These results showed statistically significant differences with respect to the other treatments (Table 2). Roots presented the maximum As concentration, followed by leaves, pods, and finally seeds. Similar results were accomplished by Sadee et al. (2016), who quantified As concentrations in bean crops. They concluded that roots presented the highest concentration, followed by leaves, stems and seeds. Also, studies conducted by Rafiq et al. (2017) focusing on the absorption of As by *Vicia faba* L. crops showed results that coincide with the ones reported in this work.

As for lettuce crops, the maximum concentration of the toxicant was found when they were grown in soil contaminated with As and irrigated with distilled water. This treatment showed statistically significant differences with respect to the control, when comparing As contents in roots (Table 3). The As concentration levels found in lettuce plants in this work coincide with those reported in previous literature. For example, Bunzl et al. (2001) reported values of 11 mg/kg of As in lettuce grown

Table 2. Total arsenic concentration (mg/kg dry weight) in broad beans plants.

Treatments	Root	Leaves	Pods	Seeds
Soil control-distilled water	7.78 $\pm$ 1.81 (a)	1.32 $\pm$ 0.67 (a)	0.27 $\pm$ 0.04 (c)	0.18 $\pm$ 0.04 (c)
Soil with As-water with As	271.3 $\pm$ 38.01 (b)	5.41 $\pm$ 0.11 (b)	3.55 $\pm$ 0.37 (a)	1.28 $\pm$ 0.06 (a)
Soil with As-distilled water	151.32 $\pm$ 10.53 (c)	7.42 $\pm$ 0.19 (b)	0.76 $\pm$ 0.08 (c)	0.42 $\pm$ 0.06 (b)
Soil control-water with As	29.01 $\pm$ 3.56 (a)	7.61 $\pm$ 1.48 (b)	1.61 $\pm$ 0.18 (b)	0.26 $\pm$ 0.0 (bc)

Each value represents the mean of three replicates $\pm$ standard deviation. The different letters within a column indicate a significant difference at $p \leq 0.05$ according to Duncan's multiple range tests.

Table 3. Total arsenic concentration (mg/kg dry weight) in lettuce plants.

Treatments	Root	Leaves
Soil control-distilled water	7.12 ± 1.92 (a)	<DL
Soil with As-water with As	701.94 ± 59.71 (b)	<DL
Soil with As-distilled water	727.59 ± 53.82 (b)	10.1 ± 0.33 (a)
Soil control-water with As	40.51 ± 4.02 (a)	8.76 ± 1.33 (a)

Each value represents the mean of three replicates ± standard deviation. The different letters within a column indicate a significant difference at $p \leq 0.05$ according to Duncan's multiple range tests. DL: below detection limit.

in soils contaminated with slag containing As. Cao and Ma (2004) evaluated As accumulation in lettuce and carrot crops grown in soils with contents of 27 and 43 mg/kg of As. These authors reported toxic concentrations in lettuce and carrots of 4−32 and 9−44 mg/kg, respectively.

The greatest As accumulation value was found in the roots, which could indicate a limited translocation rate of the toxic systemic level (Smith et al., 2009). Also, Smith et al. (2009) reported an As accumulation between 77% and 92% in *Beta vulgaris*, *Raphanus sativus* and *Lactuca sativa* roots. Studies conducted by Kumwimba et al. (2013) regarding As absorption by hydroponic lettuce crops showed that the average As concentration in roots was 19−26 times higher than in shoots. These authors reported values of 534.06 mg/kg of accumulated As in roots. In addition, de Oliveira et al. (2017) evaluated As accumulation in a lettuce crop grown in soil that had been planted with a hyperaccumulating species (*Pteris vittata*) and treated with organic matter addition. These authors determined that the roots accumulated 96% of the total As absorbed. Arsenic concentrations in leaves that were below the detection limit could be due to the inability of some plants to translocate As beyond the roots (Smith et al., 2009). According to reported by Liu et al. (2010), this high As accumulation in the root could be attributed to As complexation with phytochelatins (PCs), which would decrease efflux to the external medium and As translocation from roots to shoots.

An explanation for this limited translocation is that arsenate As(V) is rapidly reduced to arsenite As(III) in roots, followed by complexation of As(III) with PCs and subsequent sequestration in root vacuoles (Zhao et al., 2009). It is believed that the complexation between PCs and As species would be essential for plant tolerance to inorganic As species.

Under the conditions of the present study, As concentration in broad bean seeds and two of the treatments lettuce leaves (edible parts) exceeded the allowable limits established by Código Alimentario Argentino, i.e. 0.10 and 0.30 mg/kg respectively (CAA, Res. Nº 116 and 356/2012). Thus, this represents may pose a significant risk for those people who consume this crops.

3.4. Contribution of crops to total arsenic contents in the soil

Total arsenic contents in the soil after harvest is shown in Table 4. Statistically significant differences were observed when both crops studied in this work were cultivated in soils contaminated with As, as compared with the other treatments. Considering that the contaminated soil had an initial concentration of 49 mg/kg of As, continuous irrigation with contaminated water (without leaching) increased the toxic concentration in the soil where both crops were grown. In control soil treatments, with the soil initially containing 8 mg/kg of As, the concentration of this toxic chemical possibly decreased, since it was absorbed by the plants. Similar results were published by Yañez et al. (2018), when they exposed Swiss chard crops (*Beta vulgaris* var *cicla* and *d'ampuis*) to As both in soil and irrigation water.

In fact, the exchangeable and weakly adsorbed As fraction can be considered as the bioavailable content of As in soil, which can be easily absorbed by plants (Fayiga et al., 2007). Arsenic bioavailability and mobility in soil depends on factors such as pH, organic matter content, speciation, and the concentration of clay oxides and minerals (Castillo-Michel et al., 2012). Plants modify As bioavailability in the rhizosphere, so biogeochemical processes affecting As in soils planted with vegetables and those not planted with them are different. The As present in the soil solution is absorbed by plants after the release of root compounds such as organic acids, which are responsible for changing its speciation in the rhizosphere, and which hence represent the bioavailable fraction of this toxicant (Gonzaga et al., 2009).

The alkaline pH of the soil and water used in this study could contribute to As co-precipitation with sulfate or calcium, reducing its availability (Moreno-Jiménez et al., 2012). High calcium concentrations in the soil (8.88 meq/L) and in irrigation water (3.99 meq/L) could form stable precipitates such as calcium arsenate, which would decrease As absorption by crops (Hassan et al., 2014). Also, Chou et al. (2016) reported an As concentration reduction in *Raphanus sativus*, *Brassica rapa* and *Lactuca sativa* crops when they added CaO_2. According to Rafiq et al. (2017), calcium significantly decreased As accumulation in the organs of hydroponic *Vicia faba* L. In addition, considering that phosphates and arsenates are

Table 4. Total arsenic concentration (mg/kg) in soils where lettuce and broad bean plantations were grown.

Treatments	Broad beans	Lettuce
Control soil-distilled water	4.77 ± 0.03 (a)	4.80 ± 0.36 (a)
Soil with As-water with As	54.8 ± 3.45 (b)	56.43 ± 5.27 (b)
Soil with As-distilled water	48.43 ± 1.96 (b)	54.00 ± 7.60 (b)
Control soil-water with As	6.42 ± 0.13 (a)	5.53 ± 0.23 (a)

Each value represents the mean of three replicates ± standard deviation. The different letters within a column indicate a significant difference at $p \leq 0.05$ according to Duncan's multiple range tests.

structural analogues, there could have been competition between them for sorption sites in the soil.

It is worth noting that due to As cumulative effect, the concentrations of this toxic chemical in the soils where both lettuce and broad beans were grown exceeded the regulatory levels of 20 mg/kg established by National Law N° 24585 for agricultural soils. This happened when the soils contained As and when irrigation was carried out with either contaminated or distilled water.

3.5. Arsenic translocation and bioconcentration factors in lettuce and broad bean plants

Translocation factor (TF) was determined to evaluate the capacity of plants to mobilize the toxicant from roots to aerial parts.

The control treatment in the broad bean crop trial showed a higher TF than the treatments with soils contaminated with As, but this factor was lower when the crop was grown in control soil and irrigated with water containing As (Table 5). According to Huang et al. (2005), the higher As concentration levels turn out to be in the soil, the lower TF becomes, since most plants avoid intoxication by expelling the toxicant. Arsenic efflux by roots is one of the important strategies for plants to reduce As concentrations in plant cells (Chao et al., 2014). Also, As efflux was reported in other plant species, such as rice, wheat and *Holcus lanatus* (Logoteta et al., 2009; Shi et al., 2015).

Similar results were obtained by Austruy et al. (2013) found TF values that agree with those reported in the present study, when exposing a bean plantation to concentrations of 234 and 3,164 mg/kg of As. Yañez et al. (2018) also published similar TF values (0.02−0.16) in chard crops (*Beta vulgaris* var. *cicla*) exposed to As.

TF indexes lower than 1 would indicate that As was transported to a limited extent to the aerial part, and that the plants managed to expel the toxicant (Olowoyo et al.,

Table 5. Translocation and bioconcentration factor values in lettuce and broad bean crops.

Treatments	TF		BCF	
	Broad beans	**Lettuce**	**Broad beans**	**Lettuce**
Control soil-distilled water	0.17	------	1.63	1.49
Soil with As-water with As	0.02	------	4.95	12.44
Soil with As-distilled water	0.05	0.01	3.12	13.47
Control soil-water with As	0.26	0.22	4.52	7.33

(------): TF could not be determined since As concentrations in leaves were lower than detection limits.

2010). In studies conducted by Serbula et al. (2013), TF values higher than 1 indicated that plants effectively translocated As from the root to the aerial part. According to the results in the present study, the crops would not mobilize As to the aerial part effectively, even when concentrations in edible parts exceeded the regulated levels.

As reported by Sinha et al. (2007), plants can act as "accumulators" and/or "excluders". Based on this, the crops evaluated in this work could be considered excluders, since they avoided incorporating As, restricting its transport to the aerial part. Verbruggen et al. (2009), reported that a considerable fraction of absorbed As can be eliminated through the roots. This exclusion could explain why high As concentrations are found in broad bean and lettuce roots. The results of this work would support what was stated by Sun et al. (2008), who published that the roots of some higher plants can play the role of a trap organ in As transfer to aerial parts.

Bioconcentration factor (BCF) is an vital quantifiable indicator of crop/vegetables contamination which has commonly been used for estimating toxic transfer from soil into plants (Chang et al., 2014).

In this study, BCF values varied from 1.1 to 4.3 when treatments were compared between crops. This was also observed by Bui et al. (2016) when comparing lettuce BCF with that of other vegetables. In the present study, in contaminated soil treatments BCF values in lettuce plantations were 2.5−4.3 times higher than in broad bean crops.

Bioaccumulation factor values increased significantly in contaminated soils or when the toxicant was contributed by water (Table 5). This is consistent with the results published by Austruy et al. (2013), when they grew *S. nigrum*, *A. capillaris* and *V. faba* in soils contaminated with high As concentrations.

The high As accumulation levels in the roots of the evaluated plants could influence the bioconcentration of this toxic element. The lettuce crop showed the highest BCF (13.47) when grown in soil containing As and irrigated with distilled water (Table 5). The BCF values determined for this crop were higher than those reported by Kumwimba et al. (2013), when they exposed five lettuce plantations to 6 mg/L of As. These authors found BCF values within a range of 2.34−3.43.

As for broad bean plants, when they were grown in soils contaminated with As and irrigated with water that contained the toxicant, a maximum BCF of 4.95 was found (Table 5). BCF values determined for this crop were much higher than those published by Austruy et al. (2013), when they grew beans in soils with a 3164 mg/kg of As concentration.

According to Serbula et al. (2013), plants with BCF values ≥ 1 are often classified as hyperaccumulators BCF. In fact, this feature could be attributed to the crops evaluated in this study, due to their effective As accumulation in roots. Similar results were published by Yañez et al. (2018), when growing two Swiss chard crops in the same type of contaminated soil used in this work.

3.6. Health risks associated with the consumption of lettuce and broad beans

To assess the human health risk index of As, it is necessary to estimate the level of exposure by quantifying the paths of As exposure to the target organisms. The exposure of toxic elements to humans occurs through several pathways, including inhalation, food intake and dermal contact. The the target hazard quotient (THQ) and carcinogenic risk (CR) were calculated to evaluate the potential human health risk associated with vegetable consumption.

THQ values found for lettuce leaves in this work were higher than 1 (Table 6). Since As quantification in two of the lettuce crop treatments gave values that were below the detection limit, THQ could not be determined. Broad bean seeds had a THQ value higher than 1 when grown in soil containing As and when irrigated with arsenic-contaminated water, showing statistical differences with respect to the other treatments (Table 6). Similar results were published by Yañez et al. (2018), who obtained mean THQ values higher than 1 when exposing two Swiss chard varieties to As. High THQ values (higher than 1) suggest that people who consume these vegetables could experience adverse health effects. In addition, the Total Target Hazard Quotient (TTHQ) represented by the sum of each individual THQ (Chang et al., 2014) would be higher than the individual THQ values. TTHQ has been used in recent public studies as a reliable parameter to assess the combined toxicity risks that result from consuming certain foods and the types of contaminating chemicals involved.

The results obtained in this work showed that consuming lettuce leaves posed a greater risk to health than eating broad beans. Sharifi et al. (2017) actually established a decreasing risk order among certain crops: root and tuber vegetables > leafy vegetables > bulbous vegetables > cereal grains.

Table 6. Estimated target hazard quotient in the crops evaluated in this study.

Treatments	Lettuce leaf	Bean seed
Control soil-distilled water	------	0.26 ± 0.06 (c)
Soil with As-water with As	------	1.81 ± 0.08 (a)
Soil with As-distilled water	6.31 ± 0.21 (a)	0.60 ± 0.12 (b)
Control soil-water with As	4.48 ± 0.83 (a)	0.37 ± 0.0 (c)

Each value represents the mean of three replicates $\pm$ standard deviation. The different letters within a column indicate a significant difference at $p \leq 0.05$ according to Duncan's multiple range tests.

The carcinogenic risk (CR) values for adults who consume lettuce and broad beans exposed to As were 2.82×10^{-4} and 6.34×10^{-4}, respectively. These values exceeded the threshold value (1×10^{-4}) recommended by the US Environmental Protection Agency (USEPA, 2010), hence posing a considerable risk for residents who eat these vegetables. The CR indices determined in this work are consistent with those reported by Ma et al. (2017), who evaluated As absorption by different crops, including lettuce. These authors determined CR values between 1.28×10^{-4} and 4.57×10^{-4}.

The broad bean plantations studied in this work showed higher CR values than those reported by Rehman et al. (2016). These researchers reported that the intake of leafy vegetables harvested in the province of Khyber Pakhtunkhwa in Pakistan presented a CR of 2.3 per 100,000 adults. In addition, food consumption, inhalation of soil particles, drinking water and dermal contact are the important pathways for human exposure to toxic. Therefore, potential health risks for residents are actually higher than those reported in this study.

4. Conclusions

This study provides useful information for a better understanding of the uptake and distribution of As in different parts of the lettuce and broad bean plants (root, leaf, pod and bean). In addition, this study includes the measurement impact that available As from water and the soil it is grown in has on the plants. The results indicated that physicochemical soil and water properties and particularly high available As content play an important role in determining total As content in plants. The root system of lettuce and broad bean plants accumulates the highest level of total As compared with the other plant organs. The two crops evaluated showed effective As mobilization when this toxicant was present both in the soil and in irrigation water, exceeded the limit established by WHO/FAO safe.

The higher As bioaccumulation factors recorded for the roots of the two assessed plant species would indicate that they possess the characteristics of "hyperaccumulating plants".

The results of this study revealed that the values calculated risk index for Pastos Chicos population evaluated by target hazard quotient and arcinogenicity index indicated that ingestion of the As through consuming of vegetables tested is hazardous to the area residents health. These results suggest that, to protect against arsenic toxicity, local residents need vegetable-specific and site-specific information and should pay attention to the kinds and amounts of vegetables consumed. Further research work is need in order to investigate to which As species the evaluated crops are exposed to, and also to assess the bioavailability of this toxicant. Future studies will focus on the potential health risks of As through other exposure pathways.

Declarations

Author contribution statement

Luciano Matias Yañez: Conceived and designed the experiments; Performed the experiments; Analyzed and interpreted the data; Contributed reagents, materials, analysis tools or data; Wrote the paper.

Jimena A. Alfaro: Conceived and designed the experiments; Performed the experiments; Analyzed and interpreted the data.

Natalia Maria Elisa Avila Carreras: Performed the experiments; Wrote the paper.

Graciela Bovi Mitre: Conceived and designed the experiments; Contributed reagents, materials, analysis tools or data; Wrote the paper.

Competing interest statement

The authors declare no interest conflict.

Additional information

No additional information is available for this manuscript.

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