Sewage sludge biofertilizer impacts the accumulation of heavy metals in the leaves and fruits of sweet passion fruit (Passiflora alata Curtis)

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Abstract

Treatment of industrial sewage results in the production of a solid residue, sewage sludge, whose rich organic matter and chemical content makes it useful in agriculture. However, some soil components can be potentially toxic to humans if they enter the food chain. The aim of this study was to analyze the accumulation of heavy metals in the leaves and fruits of *Passiflora alata* Curtis grown on sewage sludge. Sweet passion fruit plants were cultivated in plain Oxisol soil and in lime-adjusted Oxisol enriched with sludge. After fruit maturation, the leaves and fruits were collected, growth variables were measured, and the concentration of arsenic, barium, cadmium, chromium, and lead was determined. The use of sewage sludge increased the concentration of essential elements and organic matter in the soil, thereby favoring plant growth and development. However, the sludge caused also the differential accumulation of heavy metals in the leaves and fruits of sweet passion fruit.

1. Introduction

Intensive agriculture relies heavily on chemical fertilizers derived from non-renewable resources, whose production is highly energy-consuming (Izydorczyk et al. 2022; Samoraj et al. 2022). In addition, as has been made clear by the current war in Ukraine, reliance on a single global source of chemical fertilizers (Russia), may lead to serious dependence and disruptions in their supply. Therefore, it is important to find alternative means of soil fertilization.

A shift to a more sustainable economy is based on the idea of circularity, which prioritizes material recycling over the use of non-renewable resources (Razza et al. 2018). In a circular economy, waste from one process becomes raw material for another. This approach not only prevents the loss of nutrients, but avoids also their transformation into environmental pollutants (Chojnacka et al. 2020; Izydorczyk et al. 2022; Samoraj et al. 2022).

A circular approach within the fertilizer industry aims for the recovery of biogenic elements, such as nitrogen, phosphorus, and potassium. Nitrogen recovery is achieved through reuse of biomass, such as sewage sludge and animal waste (Samoraj et al. 2022). Sewage sludge is also rich in phosphorus (Hamilton et al. 2017; Grigatti et al. 2019; Izydorczyk et al. 2022), as well as organic carbon and other elements essential for soil fertility or plant nutrition, such as sulfur, magnesium, calcium, and micronutrients (copper, manganese, zinc, and iron) (Samoraj et al. 2022).

Given the forecast increase in human population, the treatment and safe exploitation of growing volumes of solid waste from sewage represents a major environmental concern and global challenge (Li et al. 2012; Magna et al. 2014; Berilli et al. 2015; Hossain et al. 2015). With an estimated 212.912.911 inhabitants (IBGE 2021), Brazil sends 97% of untreated waste to landfills (Alfaia et al. 2017). Industrial waste is turned into sewage sludge, which has seen ample application in agriculture (Lobo et al. 2012; Magna et al. 2014; Freitas et al. 2015; Ali et al. 2019). However, this form of sewage disposal has a negative impact on the environment owing to its large content of mineral matter and heavy metals. After being spread on agricultural land, these potentially toxic compounds enter the food chain and, through bioaccumulation, may cause metabolic and chronic diseases in humans. Persistence, bioaccumulation, and toxicity make these chemicals particularly dangerous (Rai et al. 2018; Ali et al. 2019; Palansooriya et al. 2019).

Therefore, the use of sewage sludge must take into account the concentration of metals, their maximum levels in agricultural soils, the presence of pathogens, and the target species (Brasil 2006). Heavy metals are naturally occurring elements, with atomic number above 20 and elemental density above 5 g cm^{-3} (Malavolta 1994; Ali and Khan 2018; Zhang et al. 2021). Some of these elements, such as arsenic, barium, cadmium, lead, and chromium, can be extremely toxic to plants and animals (Qayoom et al. 2022).

Food is one of the main routes through which heavy metals enter the body (Pandey and Sharma 2002; Chapin and Creasy 2012; González-Weller et al. 2013), reach the nervous and endocrine systems, and accumulate in organs, such as the liver and kidneys. They may inhibit essential enzymatic activities, induce the generation of reactive oxygen species, and alter the structure of microbial communities (Taiz and Zeiger 2015).

Passiflora alata Curtis is an economically and ecologically important species, as it can help diversify agricultural crops. Its sweet taste and attractive appearance favor acceptance by consumers. The species presents satisfactory growth when cultivated in soil enriched with sewage sludge as a source of nutrients (Freitas et al. 2015). The aim of this study was to analyze the accumulation of heavy metals in the leaves and fruits of *P. alata* cultivated on sewage sludge, thus assessing the suitability of this residue as an alternative fertilizer. The study highlights the differential accumulation of heavy metals in the leaves and fruits of *P. alata* grown in soil enriched with sludge.

2. Materials And Methods

The experiment was conducted at ‘Ponte da Braúna’ farm, Rive district, Alegre, ES, located at 20° 45' S and 41° 29' W, at an altitude of 138 m. According to the Köppen classification, the region has a warm temperate climate (“Cfa”), with no dry season in winter, and hot and humid summers. The average temperature in the hottest month is above 22°C and that of the coldest month is below 18°C. The average precipitation during the driest month is above 60 mm.

Sewage sludge was collected according to the Brazilian Association of Technical Standards (ABNT) NBR 10007 (ABNT 2004), at the Sewage Treatment Station of Espírito Santense Company of Sanitation in Joana Dark, Vitória, ES. Its chemical and organic composition is presented in Table 1. The sludge was sieved through a 4-mm mesh sieve and then added to the soil together with PRNT 93% limestone (V% = 80) 30 days before planting.

Seeds of *P. alata* were harvested from ripe fruits collected in an open-pollinated orchard in the city of Alegre, ES. Seeds were sown in tubes (90 mL) filled with pure soil (Oxisol - Yellow Red Latosol) or soil +

sludge (Oxisol - Yellow Red Latosol corrected with lime and enriched with sludge at 80 t ha^{-1}). The seedlings were kept under full sun (average 850 µmol m^{-2} s^{-1}) for 50 days after emergence.

After a period in the nursery, the seedlings were transferred to holes of 40 × 40 × 40 cm spaced 4 m between plants and 3 m between rows. The plants were conducted in an espalier on a single stem for up to 11 months after planting, leaving only one sprout opposite the main stem on the wire. After this period, the plant was trimmed, so that the length of the main stem was set to 1.80 m, from which lateral and vertical branches were conducted to form a curtain. The complete espalier structure was evaluated 20 months after planting.

After fruit maturation, the leaves (n = 20) and fruits (n = 16) were collected, and the following variables were evaluated: fruit diameter (mm), mesocarp thickness (mm), fruit weight (g), number of fruits, and soluble solids (°Brix). The concentrations of As, Ba, Cd, Cr, and Pb in the leaves (n = 20) and fruits (n = 16) were determined by air/acetylene flame atomic absorption spectrophotometry at the Quimiplan Laboratory, Vila Velha, ES. An Optima 2100 DV inductively coupled plasma/optical emission spectrometer (Perkin Elmer) was used at the following parameters: power, 1300 W; gas flow, 2.5 L min^{-1}; plasma gas flow, 15 L min^{-1}; auxiliary gas flow, 0.2 L min^{-1}; nebulizer gas flow, 0.5 L min^{-1}; nebulizer type, cyclonic; and plasma vision, axial. The following wavelengths were used: 193.696 nm (As), 455,403 nm (Ba), 228.802 nm (Cd), 267.716 nm (Cr), and 220.353 nm (Pb).

After 21 months of cultivation, samples (n = 4) of pure soil and soil + sludge were collected for chemical analysis, which was performed at the Soil Routine Laboratory - CCAE-UFES, Alegre, ES. Given the normality of samples and homogeneity of variance, the means were compared by *t*-test with 5% significance.

3. Results And Discussion

All heavy metals in sewage sludge were present below the maximum legal limit determined by CONAMA Resolution 375 (Brasil 2006) (Table 1). A 21-month treatment with lime and sewage sludge improved soil chemical composition, as indicated by an increase in P, K, Ca, organic matter, cation exchange capacity, and base saturation (Table 2).

Table 1
Chemical and organic composition of the studied sewage sludge.

Chemical elements	Concentration	CONAMA limit (Brasil, 2006)
As (mg kg^{-1})	11.2	41
Ba (mg kg^{-1})	518.0	1300
Cd (mg kg^{-1})	1.4	39
Ca (mg kg^{-1})	8221.0	Not specified
Pb (mg kg^{-1})	45.4	300
Cu (mg kg^{-1})	315.0	1500
Cr (mg kg^{-1})	41.1	1000
S (mg kg^{-1})	18306.0	Not specified
Mg (mg kg^{-1})	3761.0	Not specified
Mn (mg kg^{-1})	220.0	Not specified
Hg (mg kg^{-1})	2.3	17
Mo (mg kg^{-1})	1.4	50
N (mg kg^{-1})	5505.0	Not specified
Ni (mg kg^{-1})	20.3	420
K (mg kg^{-1})	6.5	Not specified
Se (mg kg^{-1})	25.3	100
Na (mg kg^{-1})	2764.0	Not specified
Zn (mg kg^{-1})	738.0	2800
P (%)	0.1	Not specified
C organic (%)	6.5	Not specified
Organic matter (%)	11.3	Not specified

Table 2
Mean composition of pure soil and soil treated with lime and sludge for 21 months.

TREAT	pH (H_2O)	P	K	Ca	Mg	Al
		mg dm^{-3}		———cmol$_c$ dm^{-3}——		
PS	5.0^{ns}	4.0^{*}	13.9^{*}	1.9^{*}	1.1^{ns}	0.1^{ns}
S + S	5.9^{ns}	11.0^{*}	53.0^{*}	3.9^{*}	1.3^{ns}	0.0^{ns}
TREAT	H + Al	SB	CEC	T	OM	V
	————cmol$_c$ dm^{-3}————				g kg^{-1}	%
PS	3.5^{ns}	3.4^{ns}	6.90^{*}	3.50^{ns}	6.9^{*}	49.4^{*}
S + S	3.0^{ns}	5.39^{ns}	8.39^{*}	5.39^{ns}	20.3^{*}	64.2^{*}

* Significant and ^{ns} non-significant at 5% probability (*t*-test). H + Al, potential acidity; SB, sum of bases; CEC, cation exchange capacity; T, effective CTC; OM, organic matter; V%, base saturation; TREAT, treatment; PS, pure soil; S + S, soil + sludge.

Sewage sludge can improve the chemical, physical and biological characteristics of soil, having a positive impact on the microbial community, organic matter content, cation exchange capacity, and nutrient content (Mattana et al. 2014; Alvarenga et al. 2015; Qayoom et al. 2022). Successful growth of sweet passion fruit in soil enriched with sewage sludge may be associated with greater availability of essential elements required for plant growth (Freitas et al. 2015). However, the addition of sewage sludge increases a form of diethylenetriamine pentaacetic acid that extracts potentially mobile and phytotoxic fractions of heavy metals (Dhanker et al. 2021).

Here, the use of sewage sludge favored the growth of fruits, as indicated by their larger diameter, weight, and number, compared to fruits from plants grown in pure soil (Fig. 1). No differences were observed with respect to soluble solids and fruit mesocarp thickness. The higher concentration of P, K, Ca, and Mg in sludge-treated soil likely favored growth, as these elements are essential for plants (Taiz and Zeiger 2015). At the same time, effective cation exchange capacity and higher organic matter favor nutrient availability (Trannin et al. 2008).

The availability of essential elements ensures stronger plant growth. More abundant and larger fruits were observed in tomato plants cultivated in sewage sludge-supplemented soils (Hossain et al. 2015). Similarly, root, stem, and leaf dry mass, as well as fruit size were larger in cucumbers grown in sludge-treated soil than in control plants (Xie et al. 2020).

The content of As in the leaves and fruits was below 0.025 mg L^{-1}, the limit of detection by atomic absorption spectrophotometry (Fig. 2). Based on DRC N^{o} 42 (Brasil 2013), the acceptable concentration of As in the leaves and fruits is 0.1 and 0.3 mg L^{-1}, respectively. Thus, there was no evidence of As accumulation at potentially toxic levels.

The average Ba concentration in the leaves (5.42 mg L^{-1}) and ripe fruits (1.22 mg L^{-1}) exceeded the established limit (Brasil 2006) of 0.7 mg L^{-1} (Fig. 2) by 7.74 and 1.74 times, respectively. Ba present in contaminated soils can be easily translocated by the plants and accumulate in the leaves. In humans, it may affect the endocrine system, as well as cause abdominal pain, diarrhea, vomiting, anxiety, asthenia, nausea, agitation, sweating, tremor, respiratory distress, cardiac arrhythmia, and seizures (Chang et al. 2009; Li et al. 2011). Maize grown in soils treated with sludge for nine years exhibited an increased concentration of Ba in the ears and grains (Nogueira et al. 2010). Here, the various heavy metals accumulated differently in the leaves and fruits (Fig. 3), with some of them surpassing the recommended concentrations (Brasil 2013).

The concentrations of Cd (0.076 mg L^{-1}) and Pb (1.7 mg L^{-1}) in the leaves surpassed by 1.52 and 5.66 times the legal limit established by DRC Nº 42 (Brasil 2013) (0.05 mg L^{-1} and 0.3 mg L^{-1}, respectively) (Fig. 4). While there is no legal limit for Cr in the leaves, the value in treated soil (1.09 mg L^{-1}) was about twice as much as that recorded in pure soil (0.55 mg L^{-1}) (Fig. 4).

In the fruits of plants grown in sludge-treated soil, Cd was present below (0.025 mg L^{-1}) the limit (0.05 mg L^{-1}) established by RDC Nº 42 (Brasil, 2013). Instead, in the fruits, the values for Cr (0.25 mg L^{-1}) and Pb (0.26 mg L^{-1}) were above the limit established by Brazilian legislation, which is 0.1 and 0.2 mg L^{-1}, respectively. According to Chen et al. (2021), heavy metals transfer to fruits and vegetables according to the following order Cu > Zn > Cd > Cr > As > Pb, although the exact sequence depends on species and soil type, suggesting the selective absorption of elements from the soil (Elmi et al. 2020).

The chemical and biological composition of sewage sludge varies according to region, time, and stabilization procedure. In addition, the bioaccumulation of metals in plants varies according to their chemical requirements, solubility, physical condition, and soil chemistry (Gu et al. 2012). According to Song et al. (2014), plants grown in soils treated with sewage sludge may alter or prevent the accumulation of heavy metals in their green tissues in a temperature-dependent manner. Hence, the use of sewage sludge as a fertilizer should be carefully controlled to avoid any toxic effect. The accumulation of heavy metals was higher when tomato plants were exposed to 10 t ha^{-1} of sewage sludge than other organic fertilizers, with the concentration of Cd surpassing the legal limit (Hossain et al. 2015). According to the authors, the likely reason for such high levels might be the industrial type of sewage sludge used in the experiments, as Cd is commonly used for metal casting and battery manufacturing.

Concentrations of Cr above the limit were observed in the leaves and fruits of *Phaseolus vulgaris*, along with the accumulation of other non-essential elements, including Cd, Cr, Cu, and Pb (Kumar and Chopra 2013). The mobility and accumulation of Cd and Cu was found to be higher than that of Pb in grass leaves grown on treated sewage sludge (Wolejko et al. 2013). Soil treatment with sewage sludge resulted in higher accumulation of Pb, Cr, Cd, Cu, and Ni in *Beta vulgaris*, reaching potentially toxic levels for human consumption (Singh and Agrawal 2007). Analyzing the accumulation of As, Cd, Pb, Cr, and Hg in six species of leafy vegetables grown in soil treated with sewage sludge, Chang et al. (2014) reported that Cd had the greatest capacity to transfer to plants, with values 30 times higher than for Hg and 50 times higher compared to Cr and Pb. However, the authors pointed out that further studies were necessary, due to the strong sensitivity of these plants to metal pollutants, even if the concentrations found in the study were below the limit established by Chinese legislation. In another study, even though wheat grew well in sludge-treated soil, the content of Cu, Zn, and Pb exceeded the food safety limit in China, suggesting that such plants might pose a health risk (Dai et al. 2014).

The results obtained in this work differ from those reported by Oliveira et al. (2009), who also evaluated the effect of adding sewage sludge on sweet passion fruit. Their results pointed to higher concentrations of Zn, Cr, and Cu, but without phytotoxicity or plant contamination. The absence of toxicity in that study can be attributed to a different timing and methodology, as evaluations were carried out 60 days after planting in containers instead of in the field. This discrepancy implies that results obtained under controlled conditions are important but need to be validated by field experiments and at production scale.

The present and other studies highlight the accumulation of heavy metals in plant species cultivated in soil treated with sewage sludge, even if some metals remain below the legal limit. Hence, the use of sewage sludge as an organic fertilizer for the cultivation of edible species such as sweet passion fruit is not recommended. However, it may be recommended for nonedible species, such as wood and cellulose producers.

Practices involving the use of sewage-treated soil for agricultural purposes (Kelessidis and Stasinakis 2012; Grotto et al. 2013; Freitas et al. 2015) should take into account also field conditions, chemical characteristics, translocation, as well as long-term accumulation of metals in plants and soil to determine the toxic potential of such strategies on foods intended for human consumption.

4. Conclusions

Sewage sludge from the Sewage Treatment Plant of Espírito Santense Company of Sanitation in Joana Dark presents levels of heavy metals below the legal limits and, therefore, may be considered adequate for use in agriculture.

The use of sewage sludge augments the concentration of essential elements and organic matter in soil, but it also promotes the accumulation of heavy metals in the leaves and fruits of *P. alata*.

In particular, the concentrations of Ba, Cd, and Pb in the leaves, as well as those of Ba, Cr, and Pb in the fruits exceeded the limit established by Brazilian legislation.

Sweet passion fruit cultivated in soils treated with sewage should not be consumed by humans.

Declarations

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Author Contributions Statement

A.R. Freitas: conceptualization, preparation of experiments, investigation, writing – original draft, writing – review & editing. **N.A.B. Fávaris:** preparation of experiments, investigation. **T. Mello:** writing – review editing. **F.R. Pires**, **W.C. Otoni**, and **R.S. Alexandre:** writing – review and editing, supervision. **J.C. Lopes:** writing: – review and editing, supervision, and supply of resources.

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Data availability

The datasets used and/or analyzed during the current study are available from the corresponding author on reasonable request

Ethics Approval

The authors all agree to ethical approval and understand its related rules and content.

Consent to participate

The authors of this manuscript are all aware of the journal to which the manuscript was submitted, and all agree to continue to support the follow-up work.

Consent to Publication

This manuscript has not been submitted or published in other journals, and the authors agree to consent to publish.

Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Figures

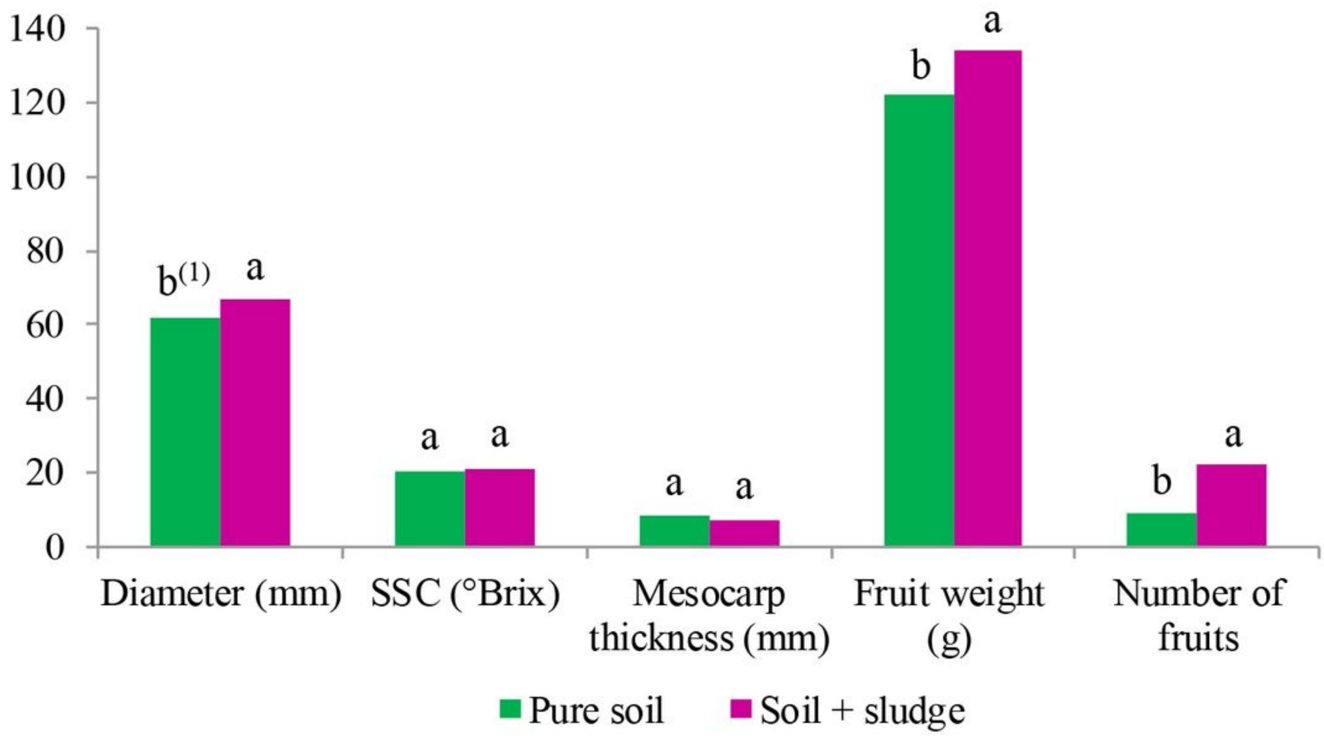

Figure 1

Diameter, soluble solids content (SSC), mesocarp thickness, weight, and number of fruits per *P. alata* plant cultivated in pure soil or soil treated with sewage sludge. [1] Means followed by the same letter across treatments are not statistically different at 5% probability (*t*-test).

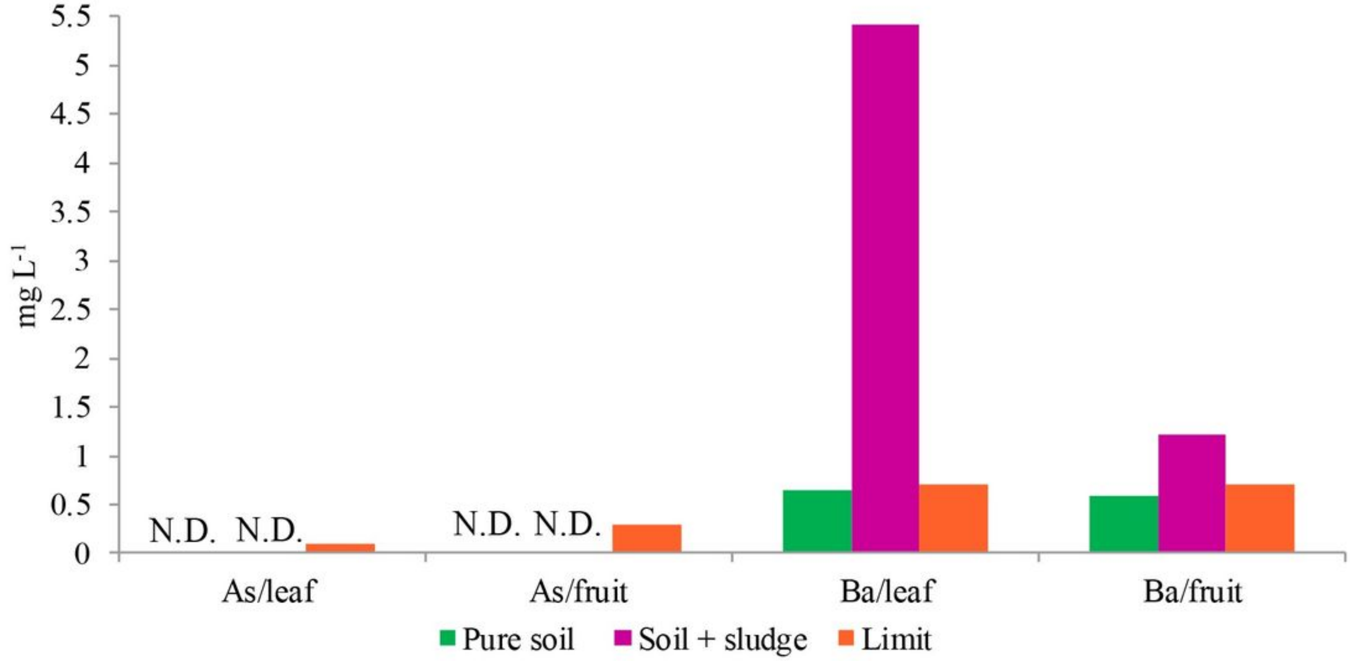

Figure 2

Concentration of As and Ba in the leaves and fruits of *P. alata* plants cultivated in pure soil or soil treated with sewage sludge. The averages were compared by *t*-test at 5% probability. The As limit is based on DRC Nº 42 (Brasil 2013) and the Ba limit is based on CONAMA Nº 375 (Brasil 2006); N.D., not detected.

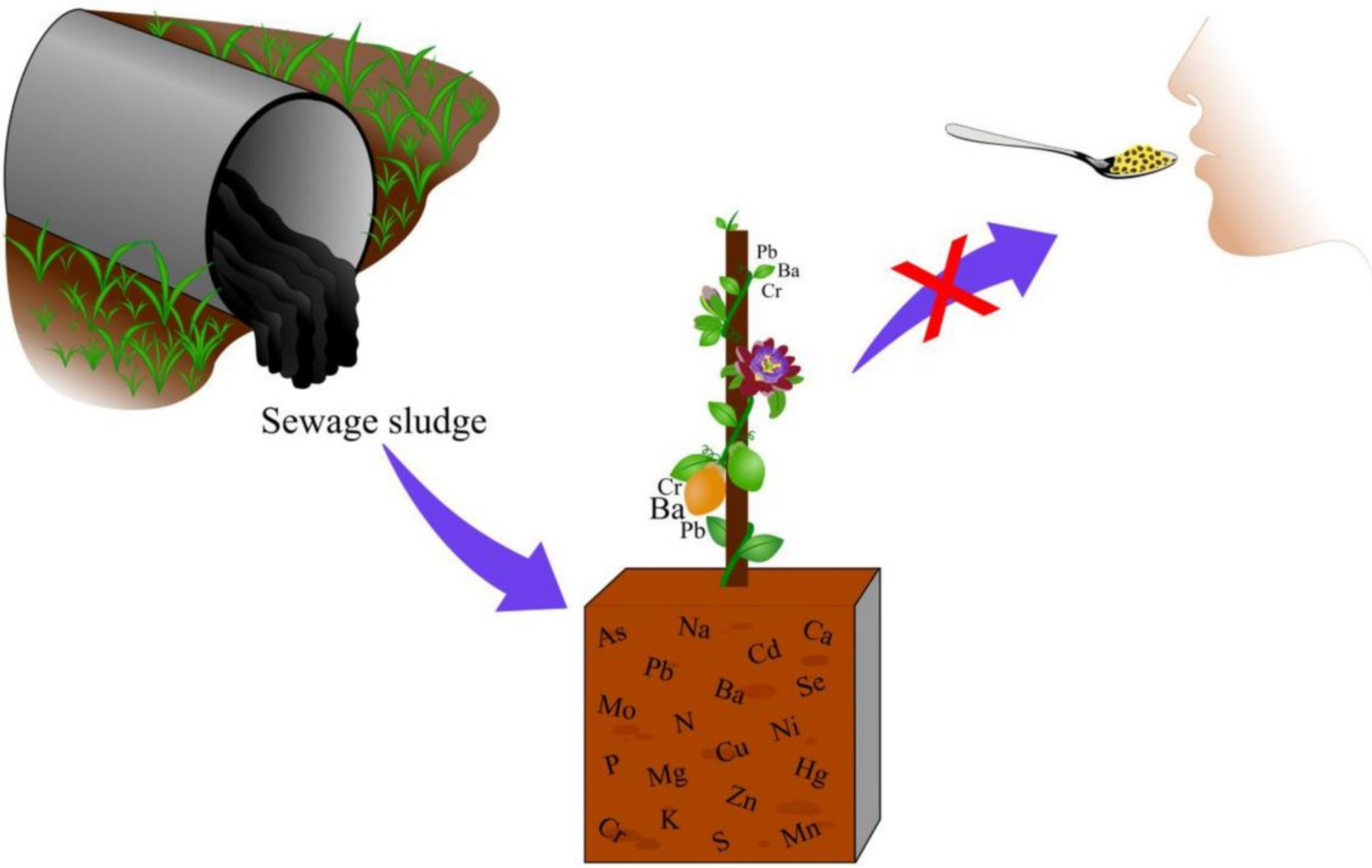

Figure 3

Schematic use of sewage sludge as soil fertilizer for the cultivation of *P. alata*, with consequent accumulation of heavy metals in the plant and production of fruits unsuitable for human consumption.

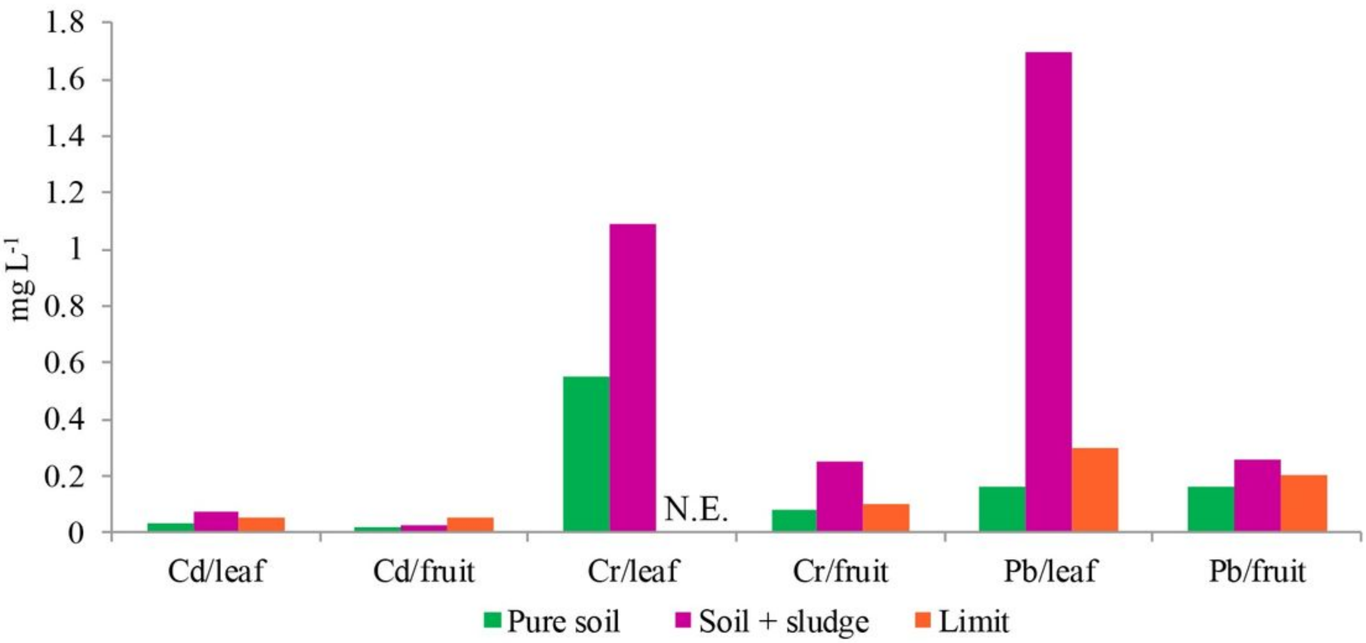

Figure 4

Concentration of Cd, Cr, and Pb in the leaves and fruits of *P. alata* cultivated in pure soil or soil treated with sewage sludge. The averages were compared by *t*-test at 5% probability. The Cd limit is based on DRC N^{o} 42 (Brasil 2013), the Cr limit is based on DRC N^{o} 55871 (Brasil 1965), and the Pb limit is based on DRC N^{o} 42 (Brasil 2013); N.E., not specified.