Science of the Total Environment 612 (2018) 370–378

Contents lists available at ScienceDirect

Science of the Total Environment

journal homepage: www.elsevier.com/locate/scitotenv

Soil cadmium uptake by cocoa in Honduras

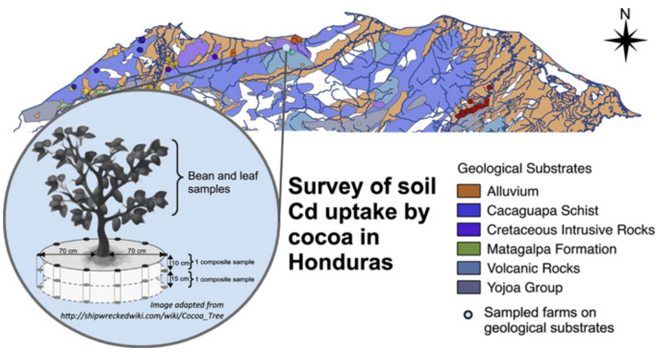
A. Gramlich [a],*, S. Tandy [a], C. Gauggel [b], M. López [b], D. Perla [b], V. Gonzalez [b], R. Schulin [a]

[a] *ETH Zurich, Institute of Terrestrial Ecosystems, Zurich, Switzerland*
[b] *Fundación Hondureña de Investigación Agrícola (FHIA), La Lima, Honduras*

HIGHLIGHTS

- Bean Cd exceeded European standards in some areas, although soils were uncontaminated.
- DGT-available soil Cd (Cd_{DGT}) best predicted bean and leaf Cd.
- Cadmium concentrations in cocoa beans were highest on alluvial substrates.

GRAPHICAL ABSTRACT



ARTICLE INFO

Article history:
Received 31 May 2017
Received in revised form 11 August 2017
Accepted 14 August 2017
Available online xxxx

Editor: F.M. Tack

Keywords:
Cadmium
Theobroma cacao L.
Bioavailability
Diffusive gradients in thin films (DGT)
Soil

ABSTRACT

Cadmium (Cd) is a trace metal without essential biological functions that is toxic to plants, animals and humans at low concentrations. It occurs naturally in soils, but inputs from anthropogenic sources have increased soil Cd contents worldwide. Cadmium uptake by cocoa (*Theobroma cacao* L.) has recently attracted attention, after the European Union (EU) decided to bring into force values for maximum Cd concentrations in cocoa products that would be exceeded by current products of various provenances from Latin America. In order to identify factors governing Cd uptake by cocoa, we carried out a survey on 55 cocoa farms in Honduras in which we determined Cd concentrations in cocoa leaves, pod husks and beans and analysed their relationships to a variety of surrounding soil and site factors. Averaging 2.6 ± 0.4 mg kg^{-1}, the concentrations of Cd were higher in the leaves than in the beans. With an average of 1.1 ± 0.2 mg kg^{-1}, the bean Cd concentrations still exceeded the proposed EU limit, however. The bean Cd showed large differences between geological substrates, even though regional variations in 'total' soil Cd were comparably small and the average concentration was in the range of uncontaminated soils (0.25 ± 0.02 mg kg^{-1}). As we found no influence of fertilizer application or vicinity to industrial sites, we conclude that the differences in soil Cd between sites were due to natural variation. Of all factors included here, DGT-available soil Cd was the best predictor of bean Cd ($R^2 = 0.5$). When DGT was not considered, bean Cd was best predicted by 'total' soil Cd, pH and geology. The highest bean Cd concentrations were found on alluvial substrates.

1. Introduction

Cadmium (Cd) is a trace metal without essential biological functions that is toxic to plants, animals and humans at low concentrations. The primary natural source of Cd in soils is the weathering of parent material

---

* Corresponding author.
E-mail address: anja.gramlich@env.ethz.ch (A. Gramlich).

http://dx.doi.org/10.1016/j.scitotenv.2017.08.145

(Adriano, 2001). The mobility of Cd in soils is high compared to other heavy metals, and it is taken up readily by plants, even though it has no essential biological functions (McLaughlin and Singh, 1999). While soil Cd can be naturally enriched to concentrations of 2 mg kg^{-1} and more, especially in soils developed on alluvial sediments or on andesite and other volcanic rocks (Fauziah et al., 2001; Manton, 2013), anthropogenic inputs originating from sources such as mining and smelting, the plastic and microelectronics industry, and rock-based fertilizers are the primary cause of elevated Cd contents in many soils (He and Singh, 1994; Ross, 1994).

Cadmium uptake by cocoa plants (*Theobroma cacao* L.) has recently attracted increasing public attention, after rather high concentrations of Cd and lead (Pb) were found in cocoa products (Manton, 2013). Many chocolate products contained >100 μg kg^{-1} Cd and thus Cd levels that were higher than those of other food products (European Food Safety Authority, 2012). According to a report of the European Food Safety Authority (2012), chocolate confectionary products contributed on average 4.3% to the weekly Cd intake of the European population. Responding to public health concerns caused by these figures, the European Union (EU) defined maximum values for Cd in cocoa products imported to the EU to be enforced in 2019 (The European Commission, 2014). While protecting the health of consumers, these regulations are a potential threat to the livelihood of many cocoa-growing smallholder farmers, in particular for producers of high-value cocoa used in products with high cocoa contents, such as dark chocolate. Thus, there is a need to identify the factors that govern Cd accumulation in cocoa beans and to find measures to reduce its concentration.

While cocoa products from Latin America generally contain higher Cd concentrations than those from West Africa (Mounicou et al., 2003), considerable variation in bean Cd contents has also been found within Latin America, between as well as within countries (Chavez et al., 2015; Gramlich et al., 2017; Mite et al., 2010; Mounicou et al., 2003). The variability in Cd concentrations in cocoa beans from different sites has been attributed to different soil parent materials (Manton, 2013), but also other factors must be considered, such as Cd inputs with phosphorus (P) fertilizers (McLaughlin and Singh, 1999; Zarcinas et al., 2004) or soil properties influencing the availability of soil Cd for uptake by plants (Chavez et al., 2015).

Honduras is one of the countries where elevated Cd contents in cocoa beans were observed recently by the chocolate industry. Even though most cocoa farmers do not add organic or mineral fertilizers to their plantations and many farms are certified for organic production in Honduras (Fromm, 2013), applications of fertilizers and pesticides in the past are still a possible source of soil contamination Cd. Intensive banana production was widespread in the second half of the last century and many cocoa farms have been established on former banana plantations. Further inputs to be considered as potential sources, at least locally, are atmospheric deposition of emissions from industries in the vicinity of cocoa fields or diffuse pollution due to flooding or irrigation with polluted river water.

Apart from the 'total' soil Cd content, soil pH, texture and organic matter are important soil factors influencing Cd availability to plants (Adams et al., 2004; Fauziah et al., 2001; Kirkham, 2006). Increased Cd uptake has also been found in plants deficient in zinc (Zn) compared to plants with adequate Zn nutrition (Choudhary et al., 1995; Dar et al., 2012; Oliver et al., 1994). One explanation for this finding is competition of Zn and Cd for the same carriers or transporters in the root cell membranes (Hart et al., 2002). However, there are also studies that did not find an antagonistic effect of Zn on Cd uptake and the interaction between the two metals may even vary among different cultivars of the same plant species (Köleli et al., 2004; Nan et al., 2002; Sanaeiostovar et al., 2012). Cadmium accumulation was also found to vary with age and season in trees (Lettens et al., 2011).

Only a few studies have investigated the effects of soil and other environmental factors on heavy metal uptake by cocoa, especially under field conditions (Chavez et al., 2015; Fauziah et al., 2001; Gramlich et al., 2017; Ramtahal et al., 2016; Ramtahal et al., 2015). In two of the studies good correlations between plant Cd and 'total' soil Cd were found (Fauziah et al., 2001; Ramtahal et al., 2016). In a further study EDTA, DTPA and Ammonium-Bicarbonate-DTPA extractions were found to be suitable methods to predict the available Cd to cocoa plants (Ramtahal et al., 2015). In the two other studies from Ecuador (Chavez et al., 2015) and Bolivia (Gramlich et al., 2017), the 'available' soil Cd (Mehlich 3 or diffusive gradients in thin films (DGT) method) in combination with the soil organic matter were good predictors of the plant Cd content. An additional effect of the clay content was observed in one of them. While in the study from Bolivia only the leaf concentration could be modeled, in the one from Ecuador a good prediction of the bean Cd was achieved (Chavez et al., 2015; Gramlich et al., 2017). The reason for this difference may be that a much broader concentration range was covered in the study from Ecuador.

The objective of this study was to identify sources of Cd in the soils of cocoa plantations in Honduras and the factors governing its accumulation in cocoa. For this purpose, we carried out a survey on 55 cocoa farms in northern and eastern Honduras in which we determined Cd concentrations in cocoa leaves, pod husks and beans and analysed their relationships to a variety of soil and site factors at the scale of individual trees.

2. Materials and methods

2.1. Survey area

Cocoa is grown in the northern and eastern parts of Honduras (Fromm, 2013), where the climate is mostly an equatorial monsoon climate (Am) according to the Köppen classification (Kottek et al., 2006). The annual temperatures vary between 20 and 35 °C in the low lands, with maxima in May and June, and the average monthly rainfall varies between about 100 mm in April and May and up to 500 mm in October, November and December.

Cocoa production is mostly in the hands of smallholder farmers in Honduras, with farm sizes ranging from less than one up to six hectares. Most farmers grow cocoa in agroforestry systems in addition to fruit and timber trees (Fromm, 2013). On the surveyed farms, we frequently found the legume tree Madreado (*Gliricidia sepium*), Caoba (*Swietenia macrophylla*), various species of the genus *Inga*, and fruit trees such as orange, mango or lemon. Most smallholder cocoa farms are managed organically. Mineral fertilizers were applied only on a few farms surveyed and not on a regular basis. The age of the sampled cocoa plantations varied greatly, from 3 to >30 years.

The survey was carried out in December 2014 and January 2015 on 55 cocoa farms in the political departments Santa Bárbara, Cortés, Atlántida, Yoro and Gracias a Dios (Figs. 1, S.1). The farms were selected to represent a wide range of soil types, site conditions (geology, topography, climate) and management schemes. The geological substrates included alluvial sediments, cretaceous intrusive and undifferentiated volcanic rocks, Cacaguapa schist, and rocks of the Matagalpa formation and Yojoa group according to USGS classification (US Geological Survey, 2016). We divided the alluvial sites into four geographic groups for the analysis of the results (wherever enough samples were available the division was made by catchment, Fig. 1). According to the FAO Classification Map of Honduras (1998), the soil types included Fluvisols, Nitosols, Cambisols and Regosols.

2.2. Soil and plant sampling

On each farm, two cocoa trees with mature fruits were selected at random and their location recorded using GPS. A total of 10 medium aged leaves were collected from each tree by taking the 9th and 10th leaf of 5 different branches, counting from their tips. In addition, 1–4 mature pods, depending on availability, were collected from each tree, separated into beans and pod husks and combined to one composite

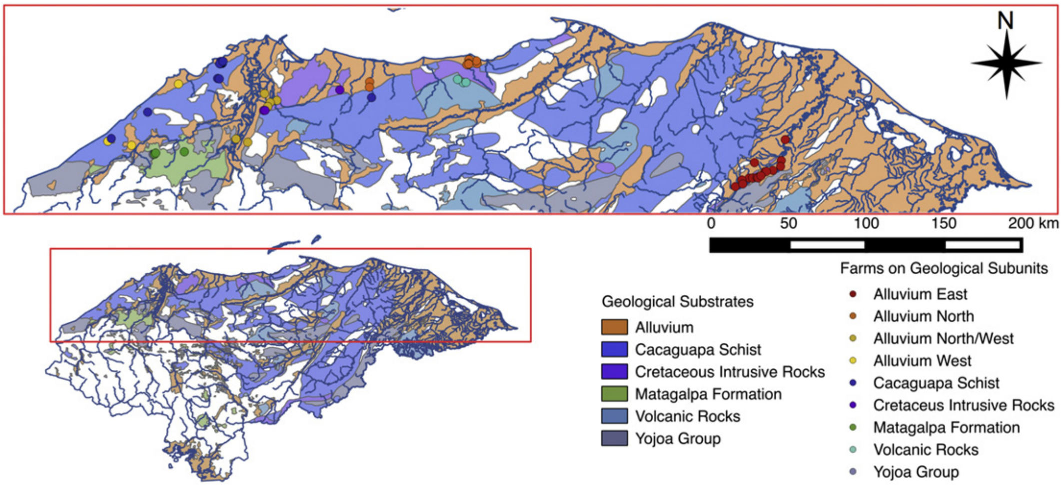


Fig. 1. Sampling area in northern and eastern Honduras with geological substrates. Points indicate farms where samples were taken. Colours of different points indicate the geological substrates, including the subunits of alluvial sediments used for analysis. Blue lines indicate rivers and borders of the country. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

sample for each of the two parts. The trunk circumference was measured at 30 cm above ground. If multiple stems occurred at that height, they were measured individually ($d_{individual}$), and the overall diameter ($d_{overall}$) was calculated as the equivalent diameter of a single stem with the same total cross sectional area (assuming geometrical similarity in shape), as proposed by the (USA Department of Agriculture, 2007):

$$d_{overall} = \sqrt{\sum d^2_{individual}} \tag{1}$$

Below each tree, 8 soil cores were taken at equal distances from each other on a circle with 70 cm radius around the trunk, using an Eijkelkamp soil corer. The cores were separated into two increments each from 0 to 10 cm ("topsoil") and 10–25 cm depth ("subsoil") and combined to give one composite sample for each depth increment. Using a prepared questionnaire, the farmers were asked about whether they applied fertilizers or pesticides, the age of the plantation and former use of the sampled land.

2.3. Analysis of samples

The leaf and pod husk samples were cleaned carefully with nano-pure water and oven-dried at 70 °C for at least 72 h. The bean samples were directly dried in the same way and peeled. All plant samples were then milled, digested following the guidelines of EPA Method 3050 (United States Environmental Protection Agency, 1996), and analysed by means of atomic absorption spectrometry (AAS). Graphite-furnace AAS (Varian SpectraAA-200) was used for the analysis of Cd and flame AAS (Elmer Analyst 200) for the analysis iron (Fe) and Zn.

The soil samples were sieved to 2 mm, oven-dried at 70 °C for at least 72 h, and then subsampled for the individual analyses described in the following. Soil pH was analysed in suspension with deionised H_2O at a 1:2 solid:liquid ratio. The organic matter content was determined using the Walkley Black method (ISRIC, 2002). Soil texture was analysed using the Bouyoucos method (Bouyoucos, 1962). 'Available' P was determined as Olsen-extractable P (ISRIC, 2002) using a Hach Dr/3000 spectrophotometer. 'Available' calcium (Ca), magnesium (Mg), and potassium (K) were determined in ammonium hydroxide extracts, buffered at pH 4.8 with acetic acid, using atomic absorption spectroscopy (AAS, Perkin Elmer Analyst 200). The EPA-3050B extraction method was used to determine 'total' soil Cd and Zn concentrations in the soil (United States Environmental Protection Agency, 1996). To determine 'available' soil Fe DTPA-$CaCl_2$-triethanolamine extraction (Lindsay and Norvell, 1978) and ammonium-acetate-EDTA (AAAcEDTA) buffered at pH 4.65 (Lakanen and Ervio, 1971) were used. For 'available' Zn DTPA, AAAcEDTA and the DGT method, as described in the next section, were used and for Cd AAAcEDTA and DGT were used as indicators of soil Cd availability. Except for DGT-available Cd, all metal analyses were performed by means of AAS in the same way as the plant samples. Two replicates of each soil and plant sample were digested and each digestion was measured by AAS twice. For quality assessment, we included the following WEPAL (Wageningen Evaluating Programs for Analytical Laboratories) soil and plant reference materials: ISE 952 (aqua regia extractable Cd: 0.120 mg kg^{-1}), ISE 992 (aqua regia extractable Cd: 0.539 mg kg^{-1}), and IPE 212 (total Cd concentration: 0.040 mg kg^{-1}). Respective recovery rates for Cd were 102 ± 3%, 94 ± 4%, and 123 ± 17%.

For DGT analysis (Zhang and Davison, 1995; Zhang et al., 1998), we used 45 g aliquots of dried and sieved soil. After adjusting the moisture content to 100% WHC (between 20 and 23 ml nano-pure H_2O), the samples were covered with parafilm and left to equilibrate for 24 h in an incubator at 25 °C and then brought in contact with rinsed DGT samplers for 72 h at 25 °C. The DGT samplers (0.4 mm Chelex-100 gel, 0.8 mm diffusive gel, 2.54 cm^2 window size) were obtained from DGT Research Ltd. at Lancaster LA2 0QJ, UK. After deployment, the samplers were cleaned, and then the resin gel was immersed for 24 h in 1 ml of 1 M ultrapure HNO_3 to extract the accumulated metals. In each batch of 40 samples, we included 3 blank DGT samplers and 3 acid blank samples as controls. Each sample was analysed in 3 analytical replicates for Cd and Zn by means of ICP-MS (ICP-MS 920, Varian, CH). The masses of Cd and Zn accumulated in the resin gel were calculated as

$$M = \frac{C(V_e + V_g)}{f_e} \tag{2}$$

where C is the concentration of the respective metal in the HNO_3 extract, V_e the volume of the extract, V_g the volume of the resin gel and f_e an elution factor accounting for incomplete elution. The respective DGT-available soil metal concentration (C_{DGT}) was then calculated, following Zhang et al. (1998), as

$$C_{DGT} = \frac{M\Delta g}{DtA} \tag{3}$$

where Δg is the thickness of the diffusive gel, D the diffusion coefficient of the gel for the metal, t the exposure time, and A the area of the diffusive gel exposed to the soil slurry.

Transfer factors relating dry weight leaf, pod and bean Cd concentrations to 'total' and DGT-available soil Cd were calculated as

$$\mathrm{TF} = \frac{\text{Plant Cd}}{\text{Soil Cd}} \tag{4}$$

where "Plant Cd" and "Soil Cd" denote the respective concentration values.

2.4. Statistical analysis

All statistical analyses were carried out using the program R (version 3.2.3, R-Development Core Team, 2015). Differences in soil and plant characteristics between geological and geographic units were determined using one-way ANOVA in combination with the Tukey HSD test for post-hoc analysis. Multiple regression analyses relating DGT-available topsoil Cd to other variables were carried out using best-fit weighted least-square estimation (MM estimator) with forward variable selection (modified rlm of the MASS package), starting out from the following selection of 11 explanatory variables with a potential influence on Cd availability: geological substrate, topsoil pH, topsoil organic matter and clay content, and the concentrations of 'total' Cd, DTPA-extractable Zn, DTPA-extractable Fe, 'available' Ca, 'available' K, 'available' Mg, and 'available' P in the topsoil. 'Available' Cd was excluded, as it was very strongly correlated to the 'total' soil Cd concentration. AAAcEDTA-extractable Zn and Fe were excluded, as DTPA-extractable Zn and Fe proved to be better predictors of plant Cd in a preliminary exploratory analysis (pairs plot). The criteria for model improvement were: significance of the variable and decrease in BIC (Bayesian information criterion). The residuals of each model were checked for systematic deviations using Tukey-Anscombe plots, normal QQ plots and residuals vs. leverage plots. Multiple regression analysis was also applied to analyse the dependencies of 'total' soil Cd from potential explanatory variables, including geological substrate, fertilizer application and altitude categories. The same procedure was applied to plant Cd, using trunk diameter and DGT-available Cd in addition to the same 11 variables used to describe the DGT-available Cd in the soil as initial set of variables. Only topsoil variables were included in the analysis, since much better correlations between plant and $\mathrm{Cd_{DGT}}$ from the topsoil were obtained in a previous study on cocoa (Gramlich et al., 2017).

For most numerical variables, $\log_{10}$ transformations were used in the ANOVA and regression analyses to meet the criterion of normal distribution. For variables relating to fractions (texture and organic matter content) we used the asin sqrt transform. The number of considered observations (n) is given for each analysis. The small variation in observation numbers is caused by a few missing soil and plant sample analyses. If not indicated otherwise, the significance level for differences was set at $p \le 0.05$ and error bars indicate standard errors of the means (SE).

3. Results

3.1. Soil characteristics

The soils showed quite large variability within as well as between different geological subunits (Table 1). The topsoil pH ranged from 4.6 to 7.8. The highest values were found in the western part of the country on the Matagalpa Formation, which is composed of calc-alkaline lava of basaltic and andesitic composition and minor pyroclastic layers (Barberi et al., 2013). This was the only region in which the median pH exceeded a value of 6. Also, the highest values in 'available' Ca, K and Mg concentrations and the lowest DTPA-extractable Fe concentrations were found on this substrate. The lowest soil pH's were found on cretaceous intrusive rocks and on undifferentiated volcanic rocks. The soil organic matter was again highest on the Matagalpa Formation and it was lowest on the different alluvial sediments. The soil texture was predominantly loamy, with maximal clay contents of about 50% on some volcanic rocks. AAAcEDTA-extractable Zn and Fe concentrations were generally higher than the respective DTPA-available concentrations, especially in soils with high pH on the Matagalpa Formation.

3.2. Soil Cd and relations to other soil parameters and geological substrates

'Total' soil Cd concentrations were within the range of Cd concentrations normally found in uncontaminated soils all over the world (Kabata-Pendias, 2001). They averaged 0.25 ± 0.02 mg kg^{-1} in the topsoil (0–10 cm) and 0.16 ± 0.01 mg kg^{-1} in the sub soil (10–25 cm). Like 'total' soil Cd, also the concentrations of AAAcEDTA-extractable Cd were higher in the topsoil, with an average of 0.14 ± 0.01 mg kg^{-1}, than in the subsoil concentrations, where they averaged 0.08 ± 0.01 mg kg^{-1}.

We found no indication that 'total' soil Cd concentrations were influenced by any fertilizer application (current application or applications in the past) reported by the farmers in the survey and also not by the altitude of the location, as no dependencies of 'total' soil Cd from these variables were observed. The concentrations were also not enhanced on soils in the vicinity of industrial sites. The values of 'total' soil Cd only depended significantly on the different geological substrates, with the highest median concentration on alluvial sediments (especially in the ones in the north of the country) and the lowest values on Cacaguapa Schist and Cretaceus Intrusive Rocks in the Western parts of Honduras. Similar results were obtained with the AAAcEDTA-extractable and DGT-available soil Cd, but the relative differences between substrates increased in the order 'total' < AAAcEDTA extractable < DGT-available soil Cd (Fig. 2). The biggest relative differences between 'total' and DGT-available soil Cd were found on the Matagalpa Formation with comparably high 'total' soil Cd concentrations, but very low DGT-available Cd (Table 1, Fig. 2).

Using 'total' soil Cd (positive relationship), DTPA-extractable Fe (positive relationship), clay content (positive relationship), organic matter content (negative relationship) and geological substrate as explanatory variables, 88% percent of the total variance of the DGT-available Cd in the topsoil could be predicted by our best regression model (Table 2). Of these explanatory variables, soil organic matter improved the model only marginally and thus could be omitted without relevant loss of predictive power. Being closely correlated with topsoil pH, DTPA-extractable Fe accounted for most of the influence of pH on DGT-available Cd. When DTPA-extractable Fe was replaced by pH, the model still explained 85% of the total variance.

3.3. Plant Cd, Zn and Fe concentrations

Leaf Cd concentrations averaged 2.6 ± 0.4 mg kg^{-1} dry weight (DW). They were thus more than twice as high as the Cd concentrations of the pod husks (1.1 ± 0.2 mg kg^{-1} DW) and beans (1.1 ± 0.1 mg kg^{-1} DW). Likewise, Zn and Fe concentrations were higher in the leaves than in the pod husks and beans. The Zn concentrations in leaves, pod husks and beans averaged 89.7 ± 6.1 mg kg^{-1} DW, 45.8 ± 1.4 mg kg^{-1} DW and 41.8 ± 0.7 mg kg^{-1} DW, respectively, and the average Fe concentrations were 93.4 ± 9.5 mg kg^{-1} DW in the leaves, 67.5 ± 3.7 mg kg^{-1} DW in the pod husks and 30.4 ± 0.8 mg kg^{-1} DW in the beans. These concentrations were for both elements in the respective ranges considered sufficient for plant nutrition (Fageria et al., 2002). The plant Cd concentrations (leaves, pod husks and beans) varied considerably between geological substrates. The lowest values were found on Cacaguapa Schist, Cretaceous Intrusive Rocks and on Matagalpa Formation, the highest values on alluvial sediments, especially in the East and North of the country, and intermediate values on alluvial sediments in the West and North West, as well as on undifferentiated volcanic rocks (Fig. 2, Table 3). The regional variation of plant Zn concentrations was quite different to that of Cd. The median concentration of bean Zn was very similar on all substrates, even though median leaf Zn concentrations were much higher on the alluvial sediments in

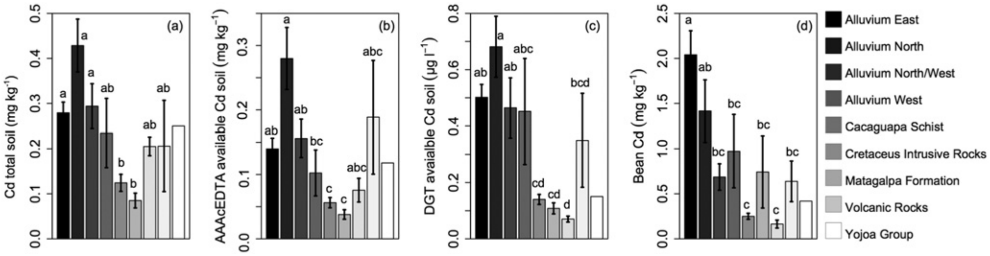


Fig. 2. Distribution of 'total' (a), AAAcEDTA-available (b), DGT-available (c) soil Cd and bean Cd (d) on different geological substrates. Error bars indicate SE. $n = 30$ (Alluvium East), $n = 18$ (Alluvium North), $n = 12$ (Alluvium North/West), $n = 5$ (Alluvium West) $n = 22$ (Cacaguapa Schist), $n = 6$ (Cretaceus Intrusive Rocks), $n = 5$ (Matagalpa Formation), $n = 6$ (undifferenciated Volcanic Rocks), $n = 1$ (Yojoa Group).

the east and north west than on the other substrates. Also, the bean Fe concentrations showed no clear regional pattern and the median concentrations were between 26 and 33 mg kg^{-1} DW in all regions (Table 3).

The log-transformed concentrations of Cd in the leaves, pod husks and beans were highly correlated among each other, with an R^2 value of 0.69 for the correlation between bean and pod husk Cd and an R^2 value of 0.47 for the correlation between bean and leaf Cd. In contrast, there was only a weak correlation between the log-transformed concentrations of Zn in the leaves and pod husks (R^2: 0.32), and no significant correlation of these with bean Zn concentrations. There were also no significant correlations between the Fe concentrations of the different plant parts. Correlations between the concentrations of the three elements in the same plant part were generally very weak ($R^2 < 0.2$). The strongest correlation between these elements in plant tissue was the one between leaf Zn and Cd with an R^2 value of 0.31 (Fig. S.2).

3.4. Correlations of plant Cd and Zn with site factors

Overall, the variation of DGT-available soil Cd among different geological substrate showed a pattern that was comparable to that of bean Cd (Figs. 2, 3). The high Cd concentrations found in beans grown on alluvial sediments in the east of the country (Fig. 2) are reflected in the high median values of the Cd soil-plant transfer factor for this region (Table 4).

The (not weighted) linear correlation between log-transformed bean Cd and log-transformed DGT-available soil Cd was quite high ($R^2 = 0.4$), considering the broad range of soils and plantations included in the survey (Fig. 3). The DGT measurements did not only provide good predictions of bean Cd concentrations, but also of the concentrations of Cd in the leaves and pod husks (Fig. 3). These relationships were closer than the relationships between bean Cd and AAAcEDTA-extractable or 'total' soil Cd (Fig. S.3).

Table 1
Top soil properties (min.–max. (median)) found on different geological substrates. The abbreviations of the geological substrates are: Alluvial (Al), Cacaguapa Schist (CS), Cretaceous Intrusive Rocks (CI), Matagalpa Formation (MF), Undifferentiated Volcanic Rocks (V), Yojoa Group (Y). For each substrate, the number of analysed samples (n) is given.

	Al East $n = 30$	Al North $n = 18$	Al No/We $n = 12$	Al West $n = 5$	CS $n = 21$	CI $n = 4$	MF $n = 5$	V $n = 6$	Y $n = 1$
Soil pH (H_2O)	5.0–6.4 (5.9)	5.2–6.2 (5.9)	5.0–7.8 (5.7)	5.2–6 (5.8)	4.7–6.2 (5.8)	4.6–5.6 (4.9)	6.2–7.0 (6.5)	5.0–5.8 (5.3)	6.8
Soil organic matter (%)	2.1–5.6 (3.6)	2.1–5.8 (3.6)	2.4–4.5 (3.2)	2.3–3.7 (3.5)	1.7–6.9 (4.4)	3.1–6.3 (4.7)	4.3–5.5 (5.1)	3.4–7.8 (4.4)	7.2
Clay content (%)	7–43 (23)	11–42 (25)	8–42 (22)	10–25 (17)	9–29 (18)	7–26 (17)	28–41 (30)	18–49 (25)	25
Silt content (%)	16–52 (38)	16–56 (42)	18–49 (30)	26–32 (30)	15–35 (22)	18–22 (19)	26–36 (35)	18–24 (22)	26
Texture class (median)	Loam	Loam	Loam	Sandy loam	Sandy loam	Sandy loam	Clay loam	Sandy clay loam	Sandy clay loam
Available P [mg kg^{-1}]	2–19 (6)	6–39 (21)	4–16 (11)	3–14 (5)	1–13 (4)	3–7 (7)	2–26 (18)	0.3–30 (18)	14
Available Ca [g kg^{-1}]	0.7–2.8 (1.4)	0.8–2.5 (1.8)	0.5–5.7 (1.6)	0.7–1.1 (0.9)	0.2–2.3 (1.0)	0.3–0.8 (0.5)	1.5–7.9 (4.4)	0.7–1.8 (0.9)	1.3
Available K [mg kg^{-1}]	33–453 (63)	51–569 (82)	42–1540 (63)	23–113 (69)	20–480 (80)	37–103 (55)	104–850 (253)	49–97 (73)	102
Available Mg [mg kg^{-1}]	115–496 (220)	117–496 (326)	93–246 (186)	63–193 (149)	48–600 (163)	50–127 (111)	155–818 (695)	153–566 (192)	216
AAAcEDTA Zn [mg kg^{-1}]	0.8–10.0 (2.3)	1.0–21.9 (5.4)	0.6–10.4 (4.2)	1.4–6.6 (2.2)	0.3–48.9 (1.3)	0.5–1.6 (0.7)	2.0–58.9 (20.3)	1.9–16.5 (4.0)	15.2
DTPA Zn [mg kg^{-1}]	1.3–2.0 (2.7)	0.8–9.6 (3.1)	0.5–6.2 (2.5)	0.2–0.5 (0.2)	0.1–5.3 (1.1)	0.4–0.8 (0.5)	0.1–3.1 (0.6)	1.0–7.0 (2.6)	0.45
DGT available Zn [µg l^{-1}]	1.7–35.2 (5.4)	1.0–25.2 (6.8)	1.8–17.9 (6.0)	1.7–15.4 (8.6)	0.9–30.7 (3.4)	0.6–11.6 (3.5)	1.0–31.2 (3.6)	3.2–50.9 (12.4)	10.6
AAAcEDTA Fe [mg kg^{-1}]	154–740 (265)	146–359 (267)	130–355 (244)	148–226 (187)	41–488 (196)	97–132 (108)	184–363 (286)	198–313 (274)	187
DTPA Fe [mg kg^{-1}]	34–177 (98)	20–125 (55)	7–129 (85)	21–74 (38)	29–155 (66)	35–72 (46)	9–23 (16)	69–154 (125)	7
Total Cd [mg kg^{-1}]	0.1–0.6 (0.2)	0.1–1.1 (0.4)	0.1–0.6 (0.3)	0.02–0.5 (0.2)	0.1–0.5 (0.1)	0.1–0.2 (0.1)	0.1–0.2 (0.2)	0.02–0.6 (0.1)	0.25
AAAcEDTA Cd [mg kg^{-1}]	0.01–0.39 (0.11)	0.06–0.87 (0.24)	0.06–0.43 (0.13)	0.01–0.19 (0.07)	0.02–0.17 (0.04)	0.01–0.05 (0.03)	0.02–0.12 (0.09)	0.01–0.5 (0.08)	0.12
DGT available Cd [µg l^{-1}]	0.20–1.16 (0.42)	0.17–1.69 (0.63)	0.14–1.23 (0.36)	0.10–0.93 (0.21)	0.04–0.34 (0.11)	0.06–0.12 (0.09)	0.05–0.11 (0.07)	0.05–1.04 (0.12)	0.15

Table 2
Multiple linear regression analysis for the description of Cd_{DGT} in top soil (TS) using best fit weighted (MM estimator) least square estimation. Cd_{tot} = 'total' soil Cd, Geology = geological substrate, and $Fe_{available}$ for the DTPA Fe. All soil variables are from the top soil. To assess the quality of each model, the Bayesian Information Criterion (BIC) and the multiple adjusted R^2 are given. All mentioned variables were significant. 104 observations could be used.

Model parameters	BIC	R^2
$Cd_{DGT} \sim 1$	−203.28	–
$Cd_{DGT} \sim Cd_{tot}$	−318.34	0.65
$Cd_{DGT} \sim Cd_{tot} + Fe_{available}$	−349.60	0.77
$Cd_{DGT} \sim Cd_{tot} + Fe_{available}$ + clay content	−371.00	0.81
$Cd_{DGT} \sim Cd_{tot} + Fe_{available}$ + clay content + geology	−372.50	0.87
$Cd_{DGT} \sim Cd_{tot} + Fe_{available}$ + clay content + geology + organic matter	−372.49	0.88

The prediction of plant Cd concentrations further improved when we used a weighted more robust model and also included the 'available' soil Mg (negative relationship) in addition to DGT-available soil Cd in the linear regression model for the leaves (Table 5). The best model for Cd in the pod husks based on DGT-available soil Cd was obtained by including topsoil clay content (negative relationship), while none of the other variables considered in our survey improved the prediction of bean Cd in models that included DGT-available soil Cd (Table 5). When Cd_{DGT} was not taken into account, the best predictions of plant Cd concentrations were obtained with models including 'total' soil Cd (positive relationship) and 'available' Fe (leaves) or soil pH (pod husks and beans) (Table 5). Accounting in addition to 'total' soil Cd for the factor 'geology' led to further improvement of the models for Cd in beans and pod husks, primarily due to the strong effect of the alluvial soils mentioned above, while accounting for 'available' soil K (negative relationship) improved leaf Cd prediction, and accounting for the factor organic matter (negative relationship) significantly improved the predictions for leaves and pod husks (Table 5). In line with the modelling of DGT-available soil Cd, also the regression analysis of plant Cd concentrations in the leaves did not reveal a direct influence of soil pH, as the influence of soil pH was apparently covered by that of DTPA-extractable soil Fe, which was closely correlated with soil pH (R^2 = 0.37).

In contrast to the suitability of the DGT method for predicting plant Cd concentrations, none of the measurements used to characterize plant 'available' soil Zn (Zn_{DGT}, DTPA Zn, AAAcEDTA Zn) was a good predictor of plant Zn concentrations. A considerable correlation was only found between leaf Zn and DTPA-extractable as well as DGT-available Zn (R^2 = 0.30, R^2 = 0.22), while the other correlations between plant and soil Zn parameters were not significant (Fig. S.4).

Table 3
Plant Cd, Fe and Zn concentrations in mg kg^{-1} dry weight (min.–max. (median)) found on different geological substrates. The abbreviations of the geological substrates are: Alluvial (Al), Cacaguapa Schist (CS), Cretaceous Intrusive Rocks (CI), Matagalpa Formation (MF), undifferentiated Volcanic Rocks (V), Yojoa Group (Y). For each substrate, the number of analysed samples (n) is given.

	Al East $n = 26$	Al North $n = 15$	Al No/We $n = 10$	Al West $n = 6$	CS $n = 22$	CI $n = 6$	MF $n = 5$	V $n = 6$	Y $n = 1$
Bean Cd [mg kg^{-1}]	0.1–7.1 (1.8)	0.2–6.0 (1.2)	0.2–1.2 (0.4)	0.1–2.2 (0.5)	0.03–0.6 (0.2)	0.04–2.3 (0.2)	0.05–0.3 (0.1)	0.04–1.5 (0.6)	0.4
Pod husk Cd [mg kg^{-1}]	0.2–6.7 (2.1)	0.1–10.2 (0.8)	0.1–1.2 (0.5)	0.2–2.5 (0.4)	0.04–0.8 (0.2)	0.1–0.2 (0.1)	0.1–0.2 (0.1)	0.1–1.7 (0.2)	0.7
Leaf Cd [mg kg^{-1}]	0.8–28.0 (4.4)	0.2–19.1 (1.7)	0.3–6.5 (1.3)	0.1–3.3 (1.9)	0.1–2.4 (0.5)	0.2–0.8 (0.5)	0.2–1.0 (0.4)	0.1–6.3 (0.3)	0.8
Bean Zn [mg kg^{-1}]	31–52 (40)	38–52 (42)	32–62 (49)	34–53 (43)	19–57 (41)	33–53 (40)	26–46 (35)	38–59 (41)	44
Pod husk Zn [mg kg^{-1}]	39–69 (56)	26–73 (40)	25–57 (39)	26–48 (40)	25–136 (38)	24–55 (37)	21–52 (41)	36–81 (62)	43
Leaf Zn [mg kg^{-1}]	64–215 (131)	29–149 (65)	17–199 (80)	18–96 (40)	17–133 (50)	32–96 (54)	22–100 (54)	31–381 (51)	43
Bean Fe [mg kg^{-1}]	26–46 (33)	12–41 (33)	10–37 (26)	24–40 (34)	5–51 (30)	12–47 (27)	18–35 (26)	23–40 (30)	26
Pod husk Fe [mg kg^{-1}]	44–164 (72)	20–269 (75)	22–72 (42)	35–120 (54)	29–170 (53)	22–138 (41)	255–57 (40)	24–83 (48)	29
Leaf Fe [mg kg^{-1}]	36–111 (51)	47–278 (80)	24–143 (76)	58–149 (128)	42–113 (72)	44–114 (68)	84–444 (114)	40–76 (54)	66

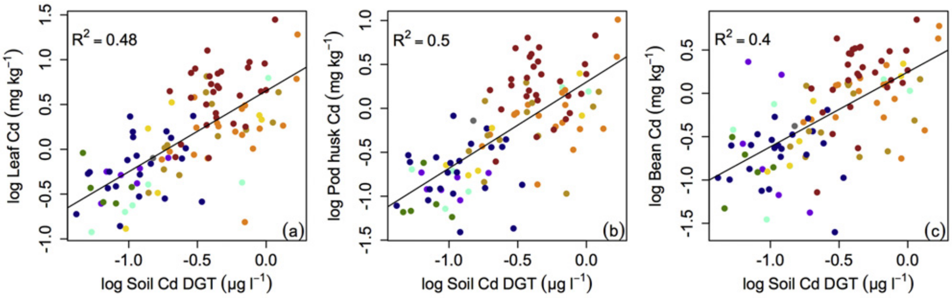


Fig. 3. Linear correlations of logarithmic plant Cd (leaves (a), pod husks (b), beans (c)) and logarithmic Cd_{DGT}. The colours of the points represent geological subunits as defined in Fig. 1. n = 103.

Table 4
Transfer factors (Cd concentrations in plants (DW) divided by soil Cd concentrations) for leaves and beans (min.-max. (median)) on different geological substrates. The abbreviations of the geological substrates are: Alluvial (Al), Cacaguapa Schist (CS), Cretaceous Intrusive Rocks (CI), Matagalpa Formation (MF), Undifferentiated Volcanic Rocks (V), Yojoa Group (Y). For each substrate, the number of analysed samples (n) is given.

Transfer factors	Al East $n = 30$	Al North $n = 18$	Al No/We $n = 12$	Al West $n = 5$	CS $n = 22$	CI $n = 6$	MF $n = 5$	V $n = 6$	Y $n = 1$
Leaf Cd/total soil Cd	4–46 (15)	0.3–26 (5)	1–26 (6)	5–20 (6)	1–25 (7)	2–11 (9)	1–5 (2)	1–10 (7)	3
Bean Cd/total soil Cd	0.4–18 (6)	0.3–11 (3)	0.4–19 (2)	0.9–6 (5)	0.1–7 (3)	0.6–46 (2)	0.3–1 (0.6)	1–22 (3)	2
Leaf Cd/Cd_{DGT}	2–34 (11)	0.2–13 (4)	2–18 (3)	1–16 (3)	0.8–23 (6)	4–8 (5)	3–20 (4)	0.6–6 (2)	5
Bean Cd/Cd_{DGT}	0.3–14 (4)	0.2–4 (2)	0.3–7 (2)	1–4 (2)	0.1–5 (2)	0.2–33 (3)	1–6 (2)	0.4–7 (2)	3

4. Discussion

4.1. Cadmium concentrations in soil and plant samples

The Cd bean concentrations found in this survey were too high in some regions to meet the guidance value for acceptable Cd concentrations in cocoa beans of 0.5 mg kg^{-1} and most likely also to meet future critical values of 0.1–0.8 mg kg^{-1} (0.1 mg kg^{-1} for products with <30% cocoa solids and 0.8 mg kg^{-1} for products with >50% cocoa solids) in final chocolate products set by the European Union (The European Commission, 2014; Netherlands Ministry of Foreign Affairs, 2016). It is interesting to note in this context that the 'total' soil Cd concentrations were within the range of what is normal for uncontaminated soils worldwide (Kabata-Pendias, 2001). This finding however is in line with similarly high Cd accumulation by cocoa reported recently also in other studies (Chavez et al., 2015; Mite et al., 2010; Ramtahal et al., 2016; Zarcinas et al., 2004) and indicates that cocoa trees extract more Cd from soil than plants in general. In fact, the values of the transfer factor relating 'total' soil Cd to the Cd concentrations in leaves and beans found in this study (Table 4) are much higher than those found for wheat and many other crop plants (Smolders, 2001). They are comparable to those found in willow and poplar trees, which are known for their accumulation of Cd (Evangelou et al., 2012; Puschenreiter et al., 2013). This observation remains valid also when comparing plant Cd uptake of cocoa (this study) at comparable DGT-available Cd concentrations with other crops such as wheat and potato (Pérez and Anderson, 2009).

The ability of cocoa trees to extract higher amounts of Cd than other plants may be promoted by enhanced Cd availability in soil resulting from the recycling of Cd with leaf litter and pod husks falling onto the soil below the trees. In contrast to practices in countries such as Ghana (Kwawukume et al., 2014), pod husks are commonly left on the ground after harvest in Honduras to reduce nutrient losses from the cocoa farms. Decomposing quickly in the tropical climate, litter fall from cocoa trees could be an important source of easily available Cd in the uppermost soil layers, where the fine roots of cocoa plants are mainly found (Kummerow et al., 1982). Such a recycling effect would also explain why Cd concentrations were generally higher in the top 10 cm of the sampled soils than in the zone below. A similar effect on the vertical distribution of Cd in soil has been has been observed below 33-years old poplar trees growing on an initially homogeneously contaminated soil, whereas the opposite effect was found below oak, ash and maple trees growing on the same site (Mertens et al., 2007). Elevated concentrations of Cd in the top soils of cocoa plantations could also result from the application of phosphate fertilizers or pesticides, as discussed (Chavez et al., 2015). Fertilizer application could be a relevant factor especially in the low land area around San Pedro Sula (Department Cortés), where also potential soil contamination due to intensive use of fertilizers and pesticides during former land use for banana production needs to be taken into account. However, in our study Cd enrichment in the topsoil was found even in very remote locations where no fertilizers had ever been applied according to the best knowledge of the interviewed farmers, notably in cocoa farms located on hillsides in the departments Cortes and Santa Barbara, in the north-western part of Honduras and in Gracias a Dios in the east of the country (Fig. S.1). Given that we found no relationship between 'total' soil Cd and vicinity to industrial sites or other potential Cd emitters, we conclude that parent material was the main source of soil Cd in the survey area. This does not rule out that in some regions, especially on the northern alluvial sediments, also diffuse industrial or agricultural pollution contributed to the observed Cd enrichment in the top soils. Also, Cd inputs through deposition of sediments transported with inundating river water may have been a relevant source of topsoil Cd on some alluvial sites.

Table 5
Multiple linear regression analysis for Cd in leaves (Cd_{Leaves}), pod husks ($Cd_{Pod\ husks}$) and beans (Cd_{Beans}) using best fit weighted (MM estimator) least square estimation. All soil variables are from the top soil. Cd_{DGT} = DGT-available Cd, Cd_{tot} = 'total' soil Cd content, $Mg_{available}$, $Fe_{available}$ and $K_{available}$ = the 'available' Mg, K and DTPA Fe, Geology = the geological substrate and pH the top soil pH. To assess the quality of each model, the Bayesian Information Criterion (BIC) and the multiple adjusted R^2 were used. All mentioned variables were significant. For leaves and beans 101 observations (n) could be used, for pod husks n equalled 99.

Model parameters	R^2
$Cd_{Leaves} \sim Cd_{DGT} + Mg_{available}$	0.59
$Cd_{Leaves} \sim Cd_{tot} + Fe_{available} + \text{organic matter} + K_{available}$	0.57
$Cd_{Pod\ husks} \sim Cd_{DGT} + \text{clay content}$	0.62
$Cd_{Pod\ husks} \sim Cd_{tot} + \text{pH} + \text{Geology} + \text{organic matter}$	0.73
$Cd_{Beans} \sim Cd_{DGT}$	0.53
$Cd_{Beans} \sim Cd_{tot} + \text{pH} + \text{Geology}$	0.57

4.2. Allocation of Cd, Fe and Zn in different plant parts

Similar correlations between leaf, pod husk and bean Cd concentrations as found in this study were also reported by Ramtahal et al. (2016). In contrast, leaf Cd concentrations were not correlated with concentrations of Cd in beans and pod husks in a previous study, in which we investigated Cd accumulation by cocoa in a field trial comparing different cocoa production systems in Bolivia (Gramlich et al., 2017). In the latter study, plant Cd concentrations were much lower than in the study presented here and in the study of Ramtahal et al. (2016). The lack of correlation between leaf and bean Cd concentrations may be related to this difference. Possibly, leaf-to-bean transfer of Cd is significantly influenced by leaf Cd concentration only at high concentration levels. Cadmium loading of beans involves xylem-to-phloem transfer, which is generally well controlled by plants (Clemens et al., 2013). However, at elevated Cd concentrations the control might be less effective.

In contrast to Cd, the essential elements Fe and Zn showed no correlations between concentrations in leaves and beans, indicating that their allocation in these parts was controlled by independent physiological factors. Many authors reported competition between Zn and Cd uptake in plants (Choudhary et al., 1995; Dar et al., 2012; Oliver et al., 1994). The positive correlation between leaf Zn and Cd concentrations suggests that competition did not play a major role in the uptake of these two elements by cocoa in our study. The fact that the correlations between Zn and Cd in soils were of similar magnitude as in the leaves may suggest that the uptake of the two elements by the plants was primarily determined by their occurrence and availability in the soil ($R^2 = 0.22$ (Zn_{total}-Cd_{total}), 0.28 (Zn_{DGT}-Cd_{DGT})).

4.3. Correlations of environmental factors with Cd and Zn concentrations in the plants

The DGT-available Cd fraction in the topsoil proved to be the best predictor for plant Cd uptake of all soil parameters analysed in this study, despite quite large differences in the properties of the sampled soils. The DGT method was in particular much better suited to describe the fraction of available soil Cd than AAAcEDTA extraction. The DGT method has been frequently used to predict metal accumulation in annual crops (Zhang and Davison, 2015), but with the exception of two studies to our knowledge not in perennial plants. Puschenreiter et al. (2013) used it to describe the availability of Zn and Cd for uptake by willow (*Salix smithiana*) in a pot trial, and in a recent study we used it to relate the availability Cd and Zn in soil to their accumulation in cocoa leaves in a field trial in Bolivia (Gramlich et al., 2017). The study presented here shows that DGT is not only well suited to predict Cd contents of cocoa leaves, but also of cocoa beans grown on a wide variety of sites with large differences in soil characteristics. The predictive power of DGT for bean Cd may depend on the level of leaf Cd concentrations, comparing the results of this study with those of Gramlich et al. (2017), as discussed before. The role of other soil properties on plant Cd was negligible or minor. The marginal influence of exchangeable soil K and Mg could be explained by ion competition with Cd for uptake by cocoa tree roots.

The highest 'total' and 'available' soil Cd concentrations were found in all regions on alluvial soils. High abundance of Cd in soils developed on sedimentary rocks has been reported also in previous studies (Adriano, 2001). The alluvial soils had relatively low pH values and organic matter contents in comparison to the soils on the Matagalpa Formation. This explains why 'available' soil Cd and plant Cd concentrations were much lower on the Matagalpa Formation than on the alluvial sites, despite similar 'total' soil Cd concentrations (Table 1). The lower 'available' soil and plant Cd concentrations found on Cacaguapa Schist and on Cretaceous Intrusive Rocks, where the soils tended to have even lower pH than the alluvial soils, can probably be attributed to the lower 'total' soil Cd concentration in these soils.

On the alluvial sites in the East of the country not only DGT-available soil Cd concentrations were elevated relative to 'total' soil Cd as on most other alluvial sites, but also plant Cd concentrations relative to DGT-available soil Cd (Figs. 2, 3). These variations in soil-plant transfer show that additional factors were at work that neither DGT, nor any of the other soil parameters accounted for. One reason that could account for the observed variation in soil-plant transfer could be that different cultivars may have been grown in the different parts of the study region. This factor was not included in the study, as there was not reliable information about the cultivars grown, taking into consideration also that cocoa trees are often grafted on root stock of a different cultivar. Differences in heavy metal accumulation among different cultivars have been found in many plant species (Clemens et al., 2013; Grant et al., 1999).

It is interesting to note that DGT-available soil Cd was largely determined by 'total' soil Cd and DTPA-extractable Fe and that in the absence of DGT measurements these two parameters could be used to predict leaf Cd concentrations with similar accuracy as by using DGT-available soil Cd. The predictive value of DTPA-extractable Fe can to a large extent be explained by its strong correlation with soil pH, which is well known as a key factor of soil Cd solubility. In fact, predictions using soil pH instead of DTPA-extractable Fe in the model explained only slightly less of the variance in DGT-available soil Cd (R^2: 0.85 compared to 0.88).

In contrast to the suitability of the DGT-method to predict the availability of Cd to cocoa, the availability of Zn could not be described well in this study, as DGT-available Zn was not correlated to the bean Zn concentrations and only weakly to the Zn in the leaves ($R^2 = 0.22$). While in previous studies DGT has been found to be a useful method to analyse Zn availability for several crop plants (Zhang and Davison, 2015), in a recent study on Zn and Cd uptake by wheat from uncontaminated soils, DGT-available Cd was also correlated much closer to plant Cd than the DGT-available Zn to the Zn in the plants (Grüter et al., 2017). These results indicate that under the studied conditions additional factors are dominating Zn uptake by the plants.

5. Conclusion

DGT-available soil Cd was the best predictor of Cd concentrations in the sampled cocoa beans, pod husks and leaves. It explained most of the influence of the analysed soil parameters, including 'total' soil Cd concentration and geological substrate, on plant Cd concentrations, notably the high Cd uptake from the soils on alluvial substrates. To explain the high Cd uptake in the east of the country, other factors such as the effect of the cocoa cultivars should be considered in future studies.

Supplementary data to this article can be found online at http://dx.doi.org/10.1016/j.scitotenv.2017.08.145.

Acknowledgements

We thank Jesus Humberto Mata for his valuable help in collecting the soil and plant samples taken for this survey. We also thank the 55 farmers who agreed to participate in the survey. We acknowledge the World Food System Centre from ETH Zurich, Switzerland, for funding the project.

References

Adams, M.L., Zhao, F.J., McGrath, S.P., Nicholson, F.A., Chambers, B.J., 2004. Predicting cadmium concentrations in wheat and barley grain using soil properties. J. Environ. Qual. 33, 532–541.

Adriano, D.C., 2001. Cadmium (Chapter 8). Trace Elements in Terrestrial Ecosystems. Springer, New York.

Barberi, F., Carapezza, M.L., Cioni, R., Lelli, M., Menichini, M., Ranaldi, M., Ricci, T., Tarchini, L., 2013. New geochemical investigations in Platanares and Azacualpa geothermal sites (Honduras). J. Volcanol. Geotherm. Res. 257, 113–134.

Bouyoucos, G.J., 1962. Hydrometer method improved for making particle size analyses of soils. Agron. J. 54, 464–465.

Chavez, E., He, Z.L., Stoffella, P.J., Mylavarapu, R.S., Li, Y.C., Moyano, B., Baligar, V.C., 2015. Concentration of cadmium in cacao beans and its relationship with soil cadmium in southern Ecuador. Sci. Total Environ. 533, 205–214.

Choudhary, M., Bailey, L.D., Grant, C.A., Leisle, D., 1995. Effect of Zn on the concentration of Cd and Zn in plant tissue of two durum wheat lines. Can. J. Plant Sci. 75, 445–448.

Clemens, S., Aarts, M.G.M., Thomine, S., Verbruggen, N., 2013. Plant science: the key to preventing slow cadmium poisoning. Trends Plant Sci. 18, 92–99.

Dar, S.R., Thomas, T., Dagar, J.C., Lal, K., Mir, A.H., Kumar, A., Mir, H., Bakshi, M.R., Mehboob, S., Singh, D., 2012. Zinc and cadmium availability as affected by zinc fertilization and saline water irrigation in wheat (*Triticum aestivum* L.) grown on cadmium polluted soil. Afr. J. Agric. Res. 7, 4996–5004.

European Food Safety Authority, 2012. Cadmium dietary exposure in the European population. EFSA J. 10, 2551.

Evangelou, M.W.H., Robinson, B.H., Günthardt-Goerg, M.S., Schulin, R., 2012. Metal uptake and allocation in trees grown on contaminated land: implications for biomass production. Int. J. Phytorem. 15, 77–90.

Fageria, N.K., Baligar, V.C., Clark, R.B., 2002. Micronutrients in crop production. Adv. Agron. 77, 185–268.

FAO soil classification map of Honduras, 1998. Centro Internacional de Agricultura Tropical - CIAT. Cali, Colombia.

Fauziah, C.I., Rozita, O., Zauyah, S., Anuar, A.R., Shamshuddin, J., 2001. Heavy metals content in soils of peninsular Malaysia grown with cocoa and cocoa tissues. Malays. J. Soil Sci. 5, 47–58.

Fromm, I., 2013. Organic chocolate for the Swiss market: contract farming in the cocoa sector in Honduras (Chapter 7). In: da Silva, C.A., Rankin, M. (Eds.), Contract Farming for Inclusive Market Access. Food and Agriculture Organization of the United Nations (FAO), Rome (pp.).

Gramlich, A., Tandy, S., Andres, C., Chincheros Paniagua, J., Armengot, L., Schneider, M., Schulin, R., 2017. Cadmium uptake by cocoa trees in agroforestry and monoculture systems under conventional and organic management. Sci. Total Environ. 580, 677–686.

Grant, C.A., Bailey, L.D., McLaughlin, M.J., Singh, B.R., 1999. Management factors which influence cadmium concentrations in crops (Chapter 7). In: McLaughlin, M.J., Singh, B.R. (Eds.), Cadmium in Soils and Plants. Springer Netherlands, Dordrecht, pp. 151–198.

Grüter, R., Costerousse, B., Bertoni, A., Mayer, J., Thonar, C., Frossard, E., Schulin, R., Tandy, S., 2017. Green manure and long-term fertilization effects on soil zinc and cadmium availability and uptake by wheat (*Triticum aestivum* L.) at different growth stages. Sci. Total Environ. 599–600, 1330–1343.

Hart, J.J., Welch, R.M., Norvell, A.N., Kochian, L.V., 2002. Transport interactions between cadmium and zinc in roots of bead and durum wheat seedlings. Physiol. Plant. 116, 73–78.

He, Q.B., Singh, B.R., 1994. Crop uptake of cadmium from phosphorus fertilizers: I. Yield and cadmium content. Water Air Soil Pollut. 74, 251–265.

ISRIC, 2002. In: van Reeuwijk, L.P. (Ed.), Procedures for Soil Analysis. International Soil Reference and Information Center (ISRIC), sixth edition.

Kabata-Pendias, 2001. Trace Elements in Soils and Plants. third edition. CRC Press, Boca Raton, Florida.

Kirkham, M.B., 2006. Cadmium in plants on polluted soils: effects of soil factors, hyperaccumulation, and amendments. Geoderma 137, 19–32.

Köleli, N., Eker, S., Cakmak, I., 2004. Effect of zinc fertilization on cadmium toxicity in durum and bread wheat grown in zinc deficient soil. Environ. Pollut. 131, 453–459.

Kottek, M., Grieser, J., Beck, C., Rudolf, B., Rubel, F., 2006. World map of the Köppen-Geiger climate classification updated. Meteorol. Z. 15, 259–263.

Kummerow, J., Kummerow, M., Souza Da Silva, W., 1982. Fine-root growth dynamics in cacao (Theobroma Cacao). Plant Soil 65, 193–201.

Kwawukume, P.S., Essuman, A., Kwawukume, J., 2014. A re-designed kiln for ashing cocoa pod husk (CPH) in cocoa growing areas of Ghana. J. Front. Constr. Eng. 3, 30–36.

Lakanen, E., Ervio, R., 1971. A comparison of eight extractants for the determination of plant available micronutrients in soils. Acta Agralia Fenn. 123, 223–232.

Lettens, S., Vandecasteele, B., De Vos, B., Vansteenkiste, D., Verchelde, P., 2011. Intra- and inter-annual variation of Cd, Zn, Mn and Cu in foliage of poplars on contaminated soil. Sci. Total Environ. 409, 2306–2316.

Lindsay, W.L., Norvell, W.A., 1978. Development of a DTPA soil test for zinc, iron, manganese, and copper. Soil Sci. Soc. Am. J. 42, 421–428.

Manton, W.I., 2013. Nonnutritive constituents in chocolate and cocoa (Chapter 6). In: Watson, R.R., Preedy, V.R., Zibadi, S. (Eds.), Chocolate in Health and Nutrition. Humana Press, New York, Heidelberg, Dordrecht, London, pp. 73–87.

McLaughlin, M.J., Singh, B.R., 1999. Cadmium in soils and plants: A global perspective (Chapter 1). In: McLaughlin, M.J., Singh, B.R. (Eds.), Cadmium in Soils and Plants. Springer Netherlands, Dordrecht, pp. 1–9.

Mertens, J., Van Nevel, L., De Schrijver, A., Piesschaert, F., Osterbaan, A., Tack, F.M.G., Verheyen, K., 2007. Tree species effect on the redistribution of soil metals. Environ. Pollut. 149, 173–181.

Mite, F., Carrillo, M., Durango, W., 2010. Avances del monitoreo de presencia de cadmio en almendras de cacao, suelos y aguas en Ecuador. Congreso Ecuatoriano de la Ciencia del Suelo, 17–19 de Noviembre del 2010. Santo Domingo.

Mounicou, S., Szpunar, J., Andrey, D., Blake, C., Lobinski, R., 2003. Concentrations and bioavailability of cadmium and lead in cocoa powder and related products. Food Addit. Contam. 20, 343–352.

Nan, Z., Li, J., Zhang, J., Cheng, G., 2002. Cadmium and zinc interactions and their transfer in soil-crop system under actual field conditions. Sci. Total Environ. 285, 187–195.

Netherlands Ministry of Foreign Affairs, 2016. CBI Product Factsheet: Cocoa in Switzerland. CBI Market Intelligence, The Netherlands.

Oliver, D.P., Hannam, R., Tiller, K.G., Wilhelm, N.S., Merry, R.H., Cozens, G.D., 1994. The effect of Zn fertilization on cadmium concentration in wheat grain. J. Environ. Qual. 23, 705–711.

Pérez, A.L., Anderson, K.A., 2009. DGT estimates cadmium accumulation in wheat and potato from phosphate fertilizer applications. Sci. Total Environ. 407, 5096–5103.

Puschenreiter, M., Wittstock, F., Friesl-Hanl, W., Wenzel, W.W., 2013. Predictability of the Zn and Cd phytoextraction efficiency of a Salix Smithiana clone by DGT and conventional bioavailability assays. Plant Soil 369, 531–541.

Ramtahal, G., Yen, I.C., Ahmad, N., Bekele, I., Bekele, F., Maharaj, K., Wilson, L., Harrynanan, L., 2015. Prediction of soil cadmium bioavailability to cacao (*Theobroma cacao* L.) using single-step extraction procedures. Commun. Soil Sci. Plant Anal. 46, 2585–2594.

Ramtahal, G., Yen, I.C., Bekele, I., Bekele, F., Wilson, L., Maharaj, K., Harrynanan, L., 2016. Relationships between cadmium in tissues of cacao trees and soils in plantations of Trinidad and Tobago. Food Nutr. Sci. 7, 37–43.

Ross, S.M., 1994. Sources and forms of potentially toxic metals in soil-plant systems (Chapter 1). In: Ross, S.M. (Ed.), Toxic Metals in Soil-plant Systems. John Wiley and Son Ltd, Chichester, pp. 3–26.

R-Development Core Team, 2015. R: A Language and Environment for Statistical Computing. R Foundation for Statistical Computing, Vienna.

Sanaeiostovar, A., Khoshgoftarmanesh, A., Shariatmadari, H., Afyuni, M., Schulin, R., 2012. Combined effect of zinc and cadmium levels on root antioxidative responses in three different zinc-efficient wheat genotypes. J. Agron. Crop Sci. 198, 276–285.

Smolders, E., 2001. Cadmium uptake by plants. Int. J. Occup. Med. Environ. Health 14, 177–183.

The European Commission, 2014. Commission regulation (EU) No 488/2014 of 12 May 2014 amending regulation (EC) No 1881/2006 as regards maximum levels of cadmium in foodstuffs. Off. J. Eur. Union.

United States Environmental Protection Agency, 1996. Method 3050B: Acid Digestion of Sediments, Sludges, and Soils. Selected Analytical Methods for Environmental Remediation and Recovery (SAM). Revision 2.

US Geological Survey, 2016. The earth surface processes team response to hurricane mitch: the Honduras geologic mapping project. http://geology.er.usgs.gov/eespteam/GISLab/honduras/honduras_project_introduction.htm, Accessed date: 10 August 2016.

USA Department of Agriculture, 2007. Forest inventory and analysis. National core field guide, Version 4.0.

Zarcinas, B.A., Ishak, C.F., McLaughlin, M.J., Cozens, G., 2004. Heavy metals in soils and crops in southeast Asia. 1. Peninsular Malaysia. Environ. Geochem. Health 26, 343–357.

Zhang, H., Davison, W., 1995. Performance-characteristics of diffusion gradients in thin-films for the in-situ measurement of trace-metals in aqueous-solution. Anal. Chem. 67, 3391–3400.

Zhang, H., Davison, W., 2015. Use of diffusive gradients in thin-films for studies of chemical speciation and bioavailability. Environ. Chem. 12, 85–101.

Zhang, H., Davison, W., Knight, B., McGrath, S.P., 1998. In situ measurements of solution concentrations and fluxes of trace metals in soils using DGT. Environ. Sci. Technol. 32, 704–710.