Unprecedented high dust ingestion estimates for the general population in a mining district of DR Congo

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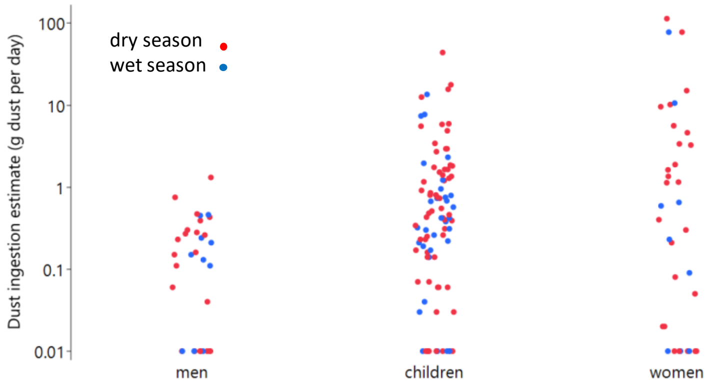
Abstract

The mining of metals in low income countries is often associated with high exposure to dust that contributes to metal exposure. Here, dust ingestion estimates were made from fecal excretion of inert tracers with corrections for dietary contribution. The study took place in the cobalt mining area of Lubumbashi (DR Congo) and involved 120 non-occupationally exposed participants in the dry season, with 51 of these being repeated in the rainy season. For each participant, duplicate meals (0-96 h), feces (24-120 h) and indoor/outdoor dust (<250 µm) were collected. The dust ingestion estimates (g day^{-1}) were derived from Nb, Ti and V as best tracers and were 0.28 (geometric mean), 3.3 (mean) and 13 (P95); these values are almost a factor ten above currently accepted estimates for the general population in high income countries. Mean dust ingestion in the dry season was twice that of the rainy season and the P95s were significantly higher in children (3-15 years) than in male adults and toddlers, geophagy (>40 g day^{-1}) was suspected in three individuals. These data explain the previously reported extreme cobalt exposures in children and support the need to manage dust in the metal mining operations.



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Introduction

Children can ingest soil and dust unintentionally through hand-to-mouth behavior. Such soil and dust ingestion can be an important vector for their exposure to environmental contaminants, with well-known examples lead (Pb) and organohalogenated compound such as PCBs and PBDEs [1–4]. Most soil or dust ingestion estimates are available for toddlers and children because of their more intense hand-to-mouth behavior. Some children deliberately eat soil (soil-pica behavior) and some adults also report to eat soil or ingest clay according to local cultures (geophagy; especially by pregnant women). The soil and dust estimates for children are typically <100 mg day^{-1} but measured values are highly depending on the analytical method that yielded the estimate[5]. The US Environmental Protection Agency (EPA) compiled data from different dust ingestion studies and recommended ingestion values for indoor settled dust, soil and outdoor settled dust, grouped by age. The proposed soil and dust ingestion are 50 mg day^{-1} for adults, 100 mg day^{-1} for age 1-21 years and 200 mg day^{-1} as an upper percentile for 3-6 year old children. Values of 1 g day^{-1} are proposed for soil pica behavior and 50 g day^{-1} for geophagy [5]. The most recent compilation of such data highlighted that most observation were from developed countries in the northern hemisphere, i.e. USA, UK and The Netherlands[5,6]. More recent data from Canada, China, and Taiwan is generally corroborating the means and upper percentiles of the earlier studies with some higher P90 values (90th percentile) found for rural people with a wilderness lifestyle[7–10].

It is striking that no dust estimates have been made for developing countries where people often live in houses with no hard floor, spend a large time outdoors and where high dust concentrations are present during the dry season. A risk assessment of street dust composition of Angola suggested that dust ingestion may be an important source of toxic elements such as Pb and As and called for data on dust ingestion estimates in such environments[11]. Metal exposure in mining areas was monitored via toenail composition in the Zambian Copperbelt Province[12] and correlation analysis suggested that dust exposure is an important vector of toxic trace elements. A similar biomonitoring in a copper mining and processing area in Western-Uganda showed, however, that toenail metal concentrations were explained by extraneous soil entrapment in the (cleaned) toenails but overall suggested that metal exposure via dust was an important but yet unquantified pathway[13]. Human biomonitoring of children living around a Pb-Zn mine showed that the metal levels in urine and feces were higher in younger children (0-3 years old) than in older children (4- 7 years old) and that this was associated with the contamination in their home environment, i.e. soil concentrations. [14]. However, the precise pathway of the exposure was not assessed. A strong case can be made for measuring dust ingestion in the Copperbelt in the Democratic Republic of Congo (DRC) that contains some of the richest cobalt (Co) deposits in the world. The high human exposure to Co and other trace elements in that area was first documented in 2009, especially among children under 14 years[15]. We have identified exposure routes by quantitatively linking Co ingestion via the diet and dust to urinary Co concentrations, thereby relying on an existing toxicokinetic model and a default dust ingestion estimate, merely assumed due to lack of data[16]. For children the observed urinary Co concentrations largely exceeded the predictions and the right skewed urinary Co data for children was unexplained. We concluded that better dust ingestion estimates were needed. In a recent study, we found again very high exposures to Co, especially among children with urinary Co concentrations more than 1000-fold above reference values[17]. In that study, strong correlations were found between concentrations of Co in urine or blood and Co concentrations in surface dust, and these correlations were markedly stronger for children than for adults.

The most common way to quantify soil and dust ingestion is measuring fecal excretion of insoluble tracers of dust and soil, i.e. chemical elements that are almost completely eliminated via feces and that are mainly present in soil and dust, not in the diet, personal care products or pharmaceuticals. Binder et al.[18] were the first to use this method with Al, Ti and Si as tracers. Over the years, many alterations have been made to this methodology by correcting for dietary contribution (the so-called mass balance approach), by selecting different tracers based on their dietary contributions to the fecal excretion, addressing different sampling durations and selecting tracers on the basis of their homogeneity within dust size classes[5,6]. Currently, the Best Tracer Method (BTM) is most often used. The BTM uses the median of values of a number of tracers that rank low in the order of increasing diet/dust tracer concentration ratio. The latter ranking logically depends on dietary habits of the participants and on other site specific conditions[19].

We performed a dust ingestion study based fecal excretion of tracers corrected for dietary contribution. This was made in a large group of members of the general population of Lubumbashi in DR Congo. Lubumbashi is the capital of a region with intense mining activities and, consequently, a high degree of environmental pollution by trace metals, especially in areas close to mines and metal processing industries. As in most low-income countries, the degree of exposure to suspended dust in Lubumbashi is considerable and, visibly, much higher than in countries with a high proportion of domestic and urban surfaces. At the outset, we selected to sample in two seasons, i.e. dry and rainy season, to contrast the environment. This region has a dry season that lasts about 5 months (rainfall < 5 mm/month) and the dust is visibly present in the air and covers the environment, including crops. We selected participant of different socioeconomic status (SES), gender and age, as these might affect the outcome. Over ten different dust tracers were considered and the heterogeneity in dust composition was addressed.

Material and methods

Study design

For this survey, 120 participants were monitored for 120 hours during the dry season (August 2016 till October 2016). Part of the original participant group (n=51) was monitored again during the rainy season (December 2016 till February 2017). The participants were recruited in the town of Lubumbashi and in the territory of Kipushi, DR Congo and were collected in several different household. For each participant, four different samples were collected: the total diet (food and drinks) of 96 consecutive hours, feces (96 h) collected 24 h after the start of diet collection to account for gut transit time and dust samples from both inside and outside the house of the participant. A cumulative 96 h collection of feces was selected over 24 h collection to reduce daily variations in transit times and dietary habits, this period had been used in other nutritional studies with metals[20].

Participants

In the dry season, 120 participants were recruited and consisted of 31 persons from the urban area and 89 persons living in the mining districts in and around the city of Lubumbashi, i.e. Kilongo, Congo Kiwele, Kabetsha, Kasapa CDM, Kawama Ruashi and Kawama village. During the rainy season, 51 of the original participants were revisited to obtain samples, 13 in the urban areas and 38 in the mining districts. The age, height and weight of the participants were recorded. The social status was derived from a questionnaire for all but four participants. The head of the household was interviewed in the local language. Participants were classified with a high SES if they ate at least three meals a day. An intermediate SES was granted if they ate two meals, while participants with a low SES reported eating only one meal each day. The participant were also grouped by age as toddlers (age 2-3 y), children (4-15 y) and adults (16 years and older). The study received prior ethical approval from the Ethical Committee of the Faculty of Medicine of the University of Lubumbashi, and all participants consented to participate in the study after having received appropriate information. Table SI 1 gives the description of the participants.

Sample collection

A trained interviewer explained participants or their parents how to collect meals, drinks and fecal sample. The collection of duplicate meals and drinks, further collectively termed duplicate meals, was made during 96 consecutive hours (t=0-96 h). Participants were asked to eat and drink normally and to collect a duplicate portion of all food and drinks consumed in 5 L polyethylene (PE) containers with drinks and food being collected in separate containers. At the end of each day, these containers were collected by the lab staff and stored at room temperature pending processing. The participants received the equivalent of 4 US$ to cover the costs of doubling their diet. They also received the equivalent of 4US$ after every successfully completed day of sampling to stimulate the completion of the study. Depending on the participant group, the collection of diets started at different times of the week. This was done to account for different dietary habits during the week or weekend.

The 96 h fecal collection started 24 h after the start of the diet collection (t=24-120 h). Stools were collected in 3 L PE containers used personally by each participant. Children were supervised by adults to avoid contamination by dust or incorrect sampling. Each stool container was closed with a lid and was collected by the lab staff at the end of each day and samples were frozen.

Samples of surface dust were taken per household using a dustpan on a random day during the sampling period to avoid additional cleaning of the house by the owners. Both indoor and outdoor surface dust was sampled in the dry season whereas only an indoor sample was collected during the rainy season. The dust samples were sieved over 250 µm sieve and that sieved fraction was stored in polyethylene mini-grip bags pending analysis. The outdoor surface soil is defined here as dust since the environmental setting does not justify to distinguish soil from dust for the exposure, hence the term dust is furthermore used as the collective term for soil and dust.

Sample pre-treatment

The food and drinks samples (0-96 h) of each participant were combined in a large shallow PE container. Occasional bones, wrappings and fruit peels were removed from the collected duplicate meal. Intact fruit such as apples or small fish were, however, retained as these items are ingested completely. The food and drinks were homogenized with stainless-steel scissors and a stainless-steel electrical mixer (Orbit, Gorgia). The total wet weight was recorded and subsamples, 5-10% of the total wet weight, were taken. The subsamples were oven-dried (VWR DRY-Line) at 70°C and the dry weight was recorded. Afterwards, the dried subsample was milled with a food-processor (Geepas) and then ground to a fine powder with pestle and mortar. The samples were transferred in duplicate to sealed PE mini-grip bags for storage and transport.

The frozen daily stools of four days were combined per participant and dried in a ventilated oven (VWR VENTI-Line) at 105°C. The dried stools were then milled with a food-processor (Geepas) to a fine powder and stored in duplicate in PE mini-grip bags.

Chemical analyses

Oven dried and ground fecal and diet samples were weighed and ashed in a muffle furnace (Nabertherm L 9/11/SW B410) at 500°C for 4 h to remove organic matter and their ash content were subsequently weighed. Total elements in the ash were measured after four acid decomposition as described in the Supporting information. Analysis of over 10 candidate tracers, including the six ones that will be used in this analysis (Ti, V, Y, Nb, La and Ce) digest solutions were made with Inductively Coupled Plasma Mass Spectrometry (ICP-MS; Agilent 7700x). Instrumental details and details on the Quality Control and Quality Assurance program are given in Annex 1 of the Supporting Information.

Dust samples of the dry season were ashed at 500°C for 4 hours. The dust samples of the rainy season (i.e. only indoor samples) were not ashed due to insufficient amount of sample. The digestion and analyses was equal as performed on ashed fecal samples. All tracer concentrations in dust are expressed on ash weight basis (dry season) or as received basis (rainy season). The average % ash of indoor dust was 87%, that of outdoor dust 93%, this means that a small error is introduced by either or not ashing.

Statistical analysis

Data analysis was performed using JMP Pro 12 (SAS Institute Inc.). Most data failed the Shapiro-Wilk test for normality and were log-transformed. Data were analyzed with ANOVA to test effect of age, season, SES and gender, and their two way interactions. Quantile regression with the maximum likelihood approach was used to identify factors explaining the 0.95 quantile (P95). The effect of season was also tested pairwise for the subset of participants that were monitored in both seasons (n=51).

Results and discussion

Participants and the fecal, diet and dust samples

The cohort consisted of 8% toddlers, 51% children and 41% adults, with a 54/46 female/male ratio. Only 5% of the participants had a high SES according to the number of daily meals (3/day), 31% had only two meals per day and 64% had a low SES with only one meal reported per day (Table SI 1).The average body weight was 13 kg for toddlers 27 kg for children and 65 kg for adults; the average BMI of adults was 24.

The daily stool weight (dry weight) of participants ranged 2-109 g day^{-1} with a geometric mean of 28 g day^{-1}. The mean value for adults was 36 g day^{-1} which corresponds with values found in literature and suggests that sampling was rather complete. Mean stool weight for adults from low income countries is 39 g day^{-1}, significantly above that of high income countries, i.e. 30 g day^{-1} [21]. The stool weight was unaffected by gender or season but it increased significantly with age, (Figure SI 1) and there was a smaller effect of SES, i.e. low SES participants had larger stool weight (22 g day^{-1}) than those with intermediate SES (16 g day^{-1}). The effects of SES on stool weight is likely related to differences in diet, i.e. higher fiber intake in peopled with low SES. A metabolic study with toddlers of a low income country showed that stool weight ranged 11-16 g day^{-1} on a casein control diet but that stool weight and fecal energy loss at least doubled for a diet on cassava, the energy loss by fecal excretion was largely related to undigested fibers and starch in the feces[22]. It is well established that increased dietary intake of fibers increases stool weight[21].

Duplicate diets were collected here to identify the intake of inert tracers via the diet. The mean daily diet dry weight ranged 100-800 g day^{-1}, the geometric mean diet dry weight being only 245 g day^{-1}, and was affected by age category, mean values increasing from 210 g day^{-1} for toddlers to

290 g day^{-1} for adults. The mean values for the small group of high SES participants (n=8) was 315 g day^{-1}. All these mean values are below dietary intake values for a reference energetic intake of about 2000 kcal day^{-1}, i.e. approximately 500 g dry weight day^{-1} for a cereal dominated diet that is typical for the general population in this region. In our previous study[16] performed in the same region but with food questionnaires based on weekly recalls, we obtained a median energy intake of 1650 kcal day^{-1}, or about 400 g day^{-1}. Along the same lines, a 24 h dietary recall of adult women in Kisangani (eastern DR Congo) yielded about 1900 kcal day^{-1} [23]. Consequently, we speculate that, despite our instructions and financial incentives, the duplicate meals were incompletely sampled in the present study, also because stool weights were within the expected range as described above. This shortcoming of the study will be taken forward into the discussion of dust ingestion estimate values.

The dust samples were collected on a household basis. Since we recruited several participants per household, the number of dust samples was below that of participants, i.e. 31 indoor samples and 47 outdoor samples in the dry season and 29 indoor samples in the rainy season and no outdoor samples in the rainy season, the latter because wet soil ingestion was considered less important than the dry indoor dust.

The quality assurance of analyses included a certified solid reference material that was included in the acid digests and ICP-MS. Table SI 1 shows the results of the analyses of the basalt reference material, showing that all six tracers that will be used further were recovered within 13% of the certified values. Other tracers were also measured but some did not yield acceptable recoveries (Zr, Ta) and others that did show good recoveries (e.g. Al, Cr, Nd, U,…) were not considered as candidate tracers for dust ingestion estimates but will be briefly referred to below.

Fecal ash fraction

The fecal ash fraction is shown in Figure SI 2 and can be used as a first indication of excessive dust ingestion. After ashing, some of the fecal samples were clearly orange, the color of the soil and dust in Katanga, indicating a high amount of dust in the feces. Some samples contained small, visible stones. The fecal ash fraction ranged 8-77%. The overall geometric mean value was 16%, which corresponds with fractions found in different human studies, i.e. about 17-20% [24] and 14-22%,[25] both studies showing the smallest fraction in the presence of fiber intake due to dilution with undigested fibers. The geometric mean of the daily fecal ash excretion was 5.2 g day^{-1}, in line with a reference daily fecal ash excretion of about 5 g day^{-1} [24]. However, the distribution of fecal ash fraction was highly right skewed (skewness +3.3), with some very large ash fractions (> 30%) among women and children, especially during the dry season (Figure 1). As substantiated in Figure SI 3, the fecal ash fraction was significantly ($p<0.01$) higher in the dry season than in the wet season when assessed by paired t-test in participants providing data in both seasons. The three highest values were observed in female adults, one of these was identical in the dry and rainy season with 40-80 g fecal ash per day, presumably as a result of geophagy, i.e. eating soil or clay, which is a frequent habit among (pregnant) women in many African countries[26].

Tracer concentrations in diet, feces and dust and selection of best tracer

All tracers Nb, Ti, V, Ce, Y, La were above the limit of quantification in all diet, dust and fecal samples. The tracer concentrations ranked diet<feces<dust, either expressed on oven dry weight basis (Table 1) or on ash weight basis (Table SI 2). This shows that the tracers in the fecal samples are mainly derived from the dust, not from food. The tracer concentrations in the dust samples were not different between inside and outside samples, the only exception being Nb for which outside dust contained slightly more Nb than inside dust. Most houses in this region had no hard floor and inside dust is, therefore, almost identical to outside dust. The tracer concentrations in the dust did

not vary largely among different samples, the standard deviations of log transformed concentrations were about 0.2 for all tracers, i.e. 68% of samples had concentrations within a factor 1.6 ($=10^{0.2}$) of the mean. The tracer concentrations in the dust were significantly different between the mining and non-mining zones and among areas within these zones, but the means differed less than one order of magnitude (details not shown).

Two indices are used here to select the most suitable tracer. The first index is the ratio of the concentration of the tracer in the diet to that in the dust, the tracer with the lowest ratio allows most sensitively detecting the dust ingestion. The diet-dust concentration ratios were calculated for each participant, and the geometric means of the diet-dust concentration ratios ranked Nb<Ti<V<Ce<Y<La (Table 1). Niobium (Nb) proved the most suitable tracer, because dietary concentrations of Nb are more than 1000-fold below those in dust. The second index is the ratio of daily dietary tracer ingestion D-Tr (µg day^{-1}) to the daily fecal excretion F-Tr (µg day^{-1}) of the corresponding person, the lowest ratio being the most preferred index to measure dust ingestion estimate. That ratio was calculated per individual and was, again, lowest for Nb for which, on average, 24% of the daily excretion was derived from the dietary sources. However, here only 116 data are available for fecal Nb concentrations because that element was only measured in part of the samples in contrast all other tracers detected for 165 subjects.

We had also measured other tracers with ICP-MS. The quality assurance and/or reproducibility (of analytical replicates) for the quantification of Zr, Ta, Cr, Sb and Pb were considered not good enough to include it in the analyses. Considering all successfully detected elements, the diet-dust concentration ratios of first 11 tracers ranked Nb< Ti< V< Ce< Fe< Y< Nd< Cu< U< La<<Al (details not shown). The ranking of Fe and Cu as potential tracers is unexpected as these elements are nutrients for plants, however this is explained by their high concentrations in the dust of that

region, however such nutrients should not be used as tracers for dust ingestion as they can be absorbed. The rather low ranking of Al is in contrast with the USA studies where Al has often been ranked among the best candidate tracers[6]. In the sections below, we will proceed with the six tracers that are described in Table 1.

Dust ingestion estimates

Dust ingestion can be estimated with or without correction for dietary contribution. The estimate with correction for dietary contribution is defined in this work as the *external* dust ingestion estimate. The external daily dust ingestion estimate of a person (g day^{-1}) is the ingestion of dust that is not present in the duplicate diet of the corresponding test person, formally

$$m_{DUST-external} = \frac{F\text{-}Tr - D\text{-}Tr}{[Tr_{DUST}]} \qquad (1)$$

where F-Tr is the daily fecal tracer excretion (µg day^{-1}), D-Tr the daily dietary intake of the tracer (µg day^{-1}) and $[Tr_{DUST}]$ the tracer concentration in the dust (µg g^{-1}). This will be calculated below using either the arrhythmic average tracer dust concentration of the entire region (=method 1) or the corresponding location specific one (method 2, thereby averaging local indoor and outdoor dust). Negative dust ingestion estimates occur when the fecal excretion rate exceeds the dietary intake. Here, these negative rates are replaced by 0.01 g day^{-1} and the number of these non-detectable estimates (as defined here) are noted.

The total dust ingestion estimate $m_{DUST-total}$ (g day^{-1}) is the value without correction for dietary sources of the tracer, formally

$$m_{DUST-total} = \frac{F\text{-}Tr}{[Tr_{DUST}]} \qquad (2)$$

The total dust estimate (=method 3) assumes that the diet can also contain dust, i.e. the ingredients of the food or drinks do not contain intrinsic tracers (i.e. tracers are not present within plant or animal products) but that all tracers present in the prepared food are derived from dust that was present on food items or added during processing and preparation. In the present study, most food consisted of locally processed maize and cassava and was often prepared in open pots.

Table 2 and Figure 1 show the dust ingestion estimates as obtained with the three methods and the six tracers. A positive external dust rate was estimated for about 63-82% of participants depending on the tracer, i.e. the fecal tracer excretion was generally higher than the tracer intake in most subjects. The dust ingestion estimates vary maximally by a factor of about 3 with the type of tracer used and the variation is less than 1.5-fold when using either Nb, Ti or V, i.e. the tracers with the lowest diet:dust concentration ratios. These three tracers also yielded generally lower estimates and the estimates clearly increased starting from La (rank 4), suggesting a bias towards overestimation of dust ingestion estimate (Table 2). The difference in dust ingestion estimate between method 1 (average dust composition) and method 2 (location specific dust composition) also proved rather small, being factor 1.5 at most. This is consistent with the relatively low variability of the dust composition within the tested region as discussed above. The variation in dust ingestion estimates among tracers found here is markedly smaller than that found in other studies that typically have a factor of five uncertainty[5]. We believe that we obtained much more consistent estimates of dust ingestion when using different tracers than in previous studies because of the much larger amounts of dust ingested by our participants than in hitherto studied populations. The entire dataset suggests that a limiting tracer method (LMT: dust estimate based on the tracer with lowest estimate, here Nb) is not strictly defensible, also considering several missing Nb data, and that a weighed estimate may be made, i.e. a combination of tracers as in the Best Tracer Method (BTM). To adopt the

BTM, we propose here to use median dust ingestion estimates of Nb, Ti and V, ranking 1-3 in diet:dust concentration ratio. The BTM data with method 1 will be used further in the discussion as the Nb data in method 2 was incomplete, moreover methods 1&2 yield almost identical data for Ti and V.

The difference between the total dust estimate (method 3) and external dust ingestion estimate (methods 1-2) is, however, large and is up to factor 5 for geometric mean estimates with smaller effects at the P95 values (Table 2). The total dust estimate considers that the fecal tracers are entirely due to contaminating dust, i.e. tracers that are present on food items are assumed to originate from external dust. This is quite plausible in this region where food harvesting, processing and cooking occur in dusty environments. The concentrations of some tracers in the diet are larger than those in other regions and, therefore, suggest dust contamination of the diet. For example, dietary intakes of Y, Ce and L of the toddlers (1-3 years) in our study were at least 7-fold higher than those of the 1-4 years old children of a Superfund site in USA [19]. The major diet here is maize flour. Vanadium concentrations in cereal products have been monitored in France[27] and are 0.05 µg V g^{-1} (range 0.01-0.08 µg g^{-1}) whereas the diet here contains 0.17 µg g^{-1} (geometric mean, Table 1) and no other food groups in the French diet study (except some fish samples) exceed the mean dietary V concentration here, suggesting an important dust contribution to the diet. It has been estimated that about 13-17 % of Pb (a poorly available element) in maize or cassava was derived from dust in food items collected in Uganda, that estimate used the V as a tracer for dust[13]. Kribek et al. [28] measured metals in cassava leaves collected in the Zambian Copperbelt and identified dust particles adhering to the leaves by SEM; washing leaves lowered trace metals concentrations in cassava leaves by over factor four. The Cu concentrations in the washed leaves could exceed 100 mg Cu kg^{-1} dry weight, a large value that unlikely reflect uptake by root and which suggests that washing does not remove all dust.

Factors explaining dust ingestion estimates

The dust ingestion estimates derived with the BTM are, in g day^{-1}, 0.28 (geometric mean), 3.3 (mean) and 13 (P95). The geometric mean and/or the P95 values of dust estimates were only marginally affected by the choice to replace the undetected values by the arbitrary value of 0.01 g day^{-1} or by the suspected incomplete sampling of the duplicate data. This is illustrated by a sensitivity analysis presented in Annex 2 of the Supporting Information. The dust ingestion estimates are extremely right skewed (skewness >6; Figure 2). This is due to four outliers that can be detected in Figure 1: three subjects were adult women and one was a 12 y-old child, one women was the same subject in the dry as in the rainy season, suggesting that this person was taking clay supplements or ate soil (geophagy). The further statistical analysis was done after removal of these 4 outliers, thus yielding 159 participants with complete data (Table 3). Their data were still highly skewed(skewness 3.4), but a normal distribution (skewness -0.06) was obtained after logarithmic transformation of all data where dust ingestion estimates were above the detection limit (82 % of data). The dust ingestion estimates were unaffected by season when the entire dataset was considered, however, paired analyses on subjects included twice revealed that, on average, dust ingestion in the dry season was double ($p<0.05$) that in the rainy season (Figure SI 4). Regression analysis also revealed that the log transformed dust ingestion estimate decreased with increasing age ($p<0.05$) but that there were no effects of body weight or length. By ANOVA, age category, SES, season and gender, and their two-way interactions were not significant, although children and female adults tended to have the largest dust ingestion estimates (Table 3). Six different groups were, therefore, made based on the three age classes and two genders, revealing that the geometric mean dust ingestion estimate was larger for female children than for adult males (Tukey test, $\alpha=0.05$) and, more importantly, that the P95 (of log transformed dust intake estimates) was markedly and significantly ($p<0.001$) higher in children and in female adults than in the other groups as detected by quantile regression analysis on these six age-gender groups. The SES had weak and inconsistent effects on dust ingestion estimates. An analysis on SES and gender effects per age class showed that the P95 of the dust intake estimate of adults from low or intermediate social classes was about 5-fold higher than for adults from the high social class; the differences were even more marked for women for whom that difference was about 10-fold; no such SES effects were found in the other age groups.

The findings of this study can now be taken forward to improve exposure assessments in polluted sites in similar settings. For example, our Co exposure study in this region[16] found adequate predictions for adults when considering their diet and commonly accepted default values for dust ingestion, however, the measured Co exposure (urinary Co) for children was five times higher than expected by their diet and a default dust ingestion of 0.2 g dust day^{-1} [16]. Using the mean dust ingestion estimate for children calculated here (i.e. 1.7 g dust day^{-1}) leads to a three-fold higher Co intake for the average child, thus corresponding more closely with the observed urinary concentrations (details not shown). This reasoning is certainly also applicable for some very high values of urinary Co values when one considers the P95 of dust intake estimates, which yield a 10-fold higher calculated exposure and predicted urinary Co concentrations of about 200 µg Co g^{-1} creatinine, values that are exceptionally found in individuals (e.g. data for children: geometric mean, P75 and max are 28, 63 and 370 µg Co g^{-1} creatinine[16]).

Taken together, this study has revealed unprecedented, but not unexpected high estimates of dust and soil intake for people living in a generally dusty and metal-polluted area. We believe that our findings of high involuntary dust ingestion can be generalized to populations living in similar environments in low income countries, especially when the dry season leads to high dust concentrations. In combination with appropriate assessments of the bioavailability of soil borne metals or metalloids relative to the diet such as developed for Pb, Cd and As[29], our estimates of dust ingestion can be used for improving human risk assessments for toxic agents in mining affected regions in low income countries. Data of biaccessible concentrations of Co and Cu in soil and dust collected in the Zambian Copperbelt already indicated a risk by taking the default 100 mg daily dust ingestion for young children[30], clearly an underestimated risk given current new data.

Associated content

Supporting Information

Supporting information contains additional graphs and tables and details of analytical methodologies. This material is available free of charge via the Internet at http://pubs.acs.org.

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This paper was written through contributions of all authors. all authors have given approval to the final version of this manuscript.

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Table 1.Tracer concentrations in the diet, dust and fecal samples and the concentration or intake/excretion ratio. Data are geometric means of all concentrations and of concentration or intake ratios of all participants in both seasons. Tracers are ranked from low to high diet/dust concentration ratio, the lower values are most suitable for dust ingestion estimates.

	Nb	Ti	V	Ce	Y	La
	$\mu g\ g^{-1}$					
Diet	0.020	6.1	0.17	0.10	0.041	0.071
Inside dust	19	5700	88	43	15	17
Outside dust	30	5800	85	42	13	17
Feces	0.76	220	4.0	3.0	1.1	1.8
Diet to dust concentration ratio x 1000 (-)	0.73	0.93	1.81	2.16	2.72	3.82
Daily dietary intake to fecal excretion ratio (-)	0.24	0.29	0.45	0.35	0.39	0.41

Table 2. Dust ingestion estimates (in g dust/day, geometric mean, GM, and 95th percentile, P95) using different tracers and according to the use of average or location specific dust composition, and the use of a correction or not for tracers ingested with the diet. Tracers are ranked from low (Nb) to high (La) diet/dust concentration ratio (see Table 1). The lower values are most suitable for dust ingestion estimates. The best tracer method (BTM) uses data of the first three ranked tracers.

	Nb	Ti	V	Ce	Y	La	BTM[§]
	Method 1:external dust ingestion estimate (g/day) corrected for dietary intake and using average dust composition						
GM	0.28	0.27	0.21	0.41	0.40	0.48	**0.28**
P95	9.8	15	13	25	23	34	**13**
n (% of n >DL)[$]	113 (82)	161 (80)	161 (71)	161(82)	161 (77)	161 (78)	163(82)
	Method 2:external dust ingestion estimate (g/day) corrected for dietary intake and using location specific dust composition						
GM	n.d.°	0.25	0.15	0.40	0.36	0.44	**0.26**
P95		17	15	36	28	41	**18**
n (% of n >DL)[$]		152 (78)	106 (63)	149 (80)	151(74)	149 (75)	151 (79)
	Method 3:total dust ingestion estimate (g/day) uncorrected for dietary intake and using average dust composition						
GM	0.67	0.79	0.98	1.5	1.7	2.3	**0.87**
P95	10	15	14	27	24	35	**14**
n (% of n >DL))	114	162	162	162	163	162	164

[$] DL=detection limit is reached if dust ingestion estimate <0, i.e. when fecal tracer excretion exceeds dietary tracer intake;

[§] BTM=Best Tracer Method defined here as the median dust ingestion estimate based on first three traces (Nb, Ti, V) or mean of Ti and V for method 2. °n.d.=no data, the number of location specific correct combination was considered too small to be used.

Table 3. External dust ingestion estimates of the general population in Lubumbashi based on the Best Tracer Method after exclusion of 4 subjects with an excessive (>40 g/day) dust ingestion estimate. The data are sorted by age groups (toddlers: 2-3 year, children 4-15 year, adults 16 years and more) and gender.

age group	gender	n	dust ingestion estimate (g day^{-1})			
			geometric mean*	median	mean	P95$^{\$}$
Toddler	F	5	0.28^{AB}	0.30	0.51	$1.2^{\circ B}$
	M	5	0.64^{AB}	0.42	0.86	$1.9^{\circ B}$
Child	F	49	0.35^{A}	0.43	1.8	10.1^{A}
	M	36	0.29^{AB}	0.40	1.6	13.8^{A}
Adult	F	32	0.27^{AB}	0.35	2.3	12.1^{A}
	M	32	0.09^{B}	0.15	0.22	0.95^{B}
All data		159	0.25	0.30	1.4	9.5

°maximum value for Toddlers; *Tukey HSD test;$^{\$}$detected with quantile regression on the six age/gender groups

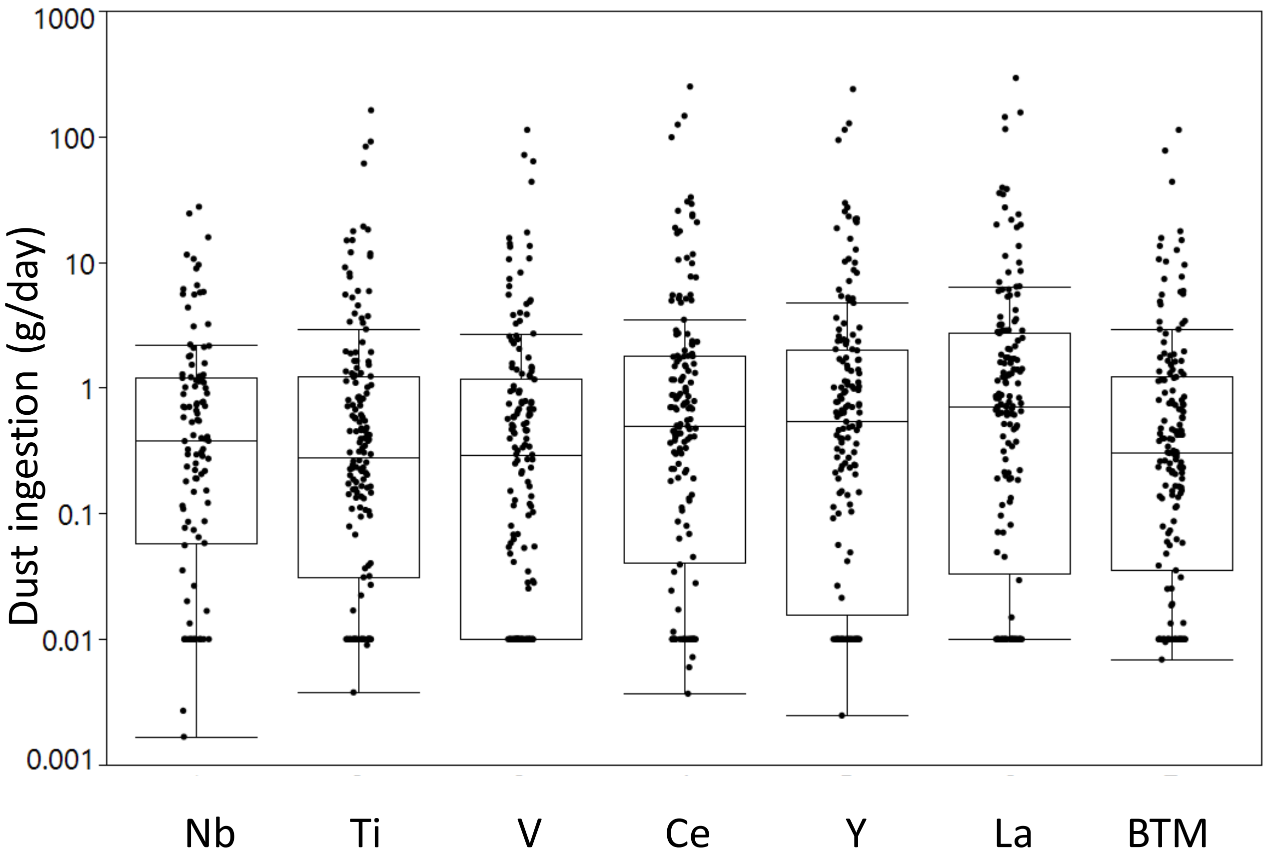

Figure 1. Daily external dust ingestion estimates in the general population in Lubumbashi using different tracers. All data are calculated with the regional average dust composition (=method 1). The Best Tracer Method (BTM) is the median of estimates of the first three tracers (Nb, Ti and V) that ranked lowest in diet:dust ratio. Negative dust ingestion are considered as non-detected and are replaced by 0.01 g day^{-1}.

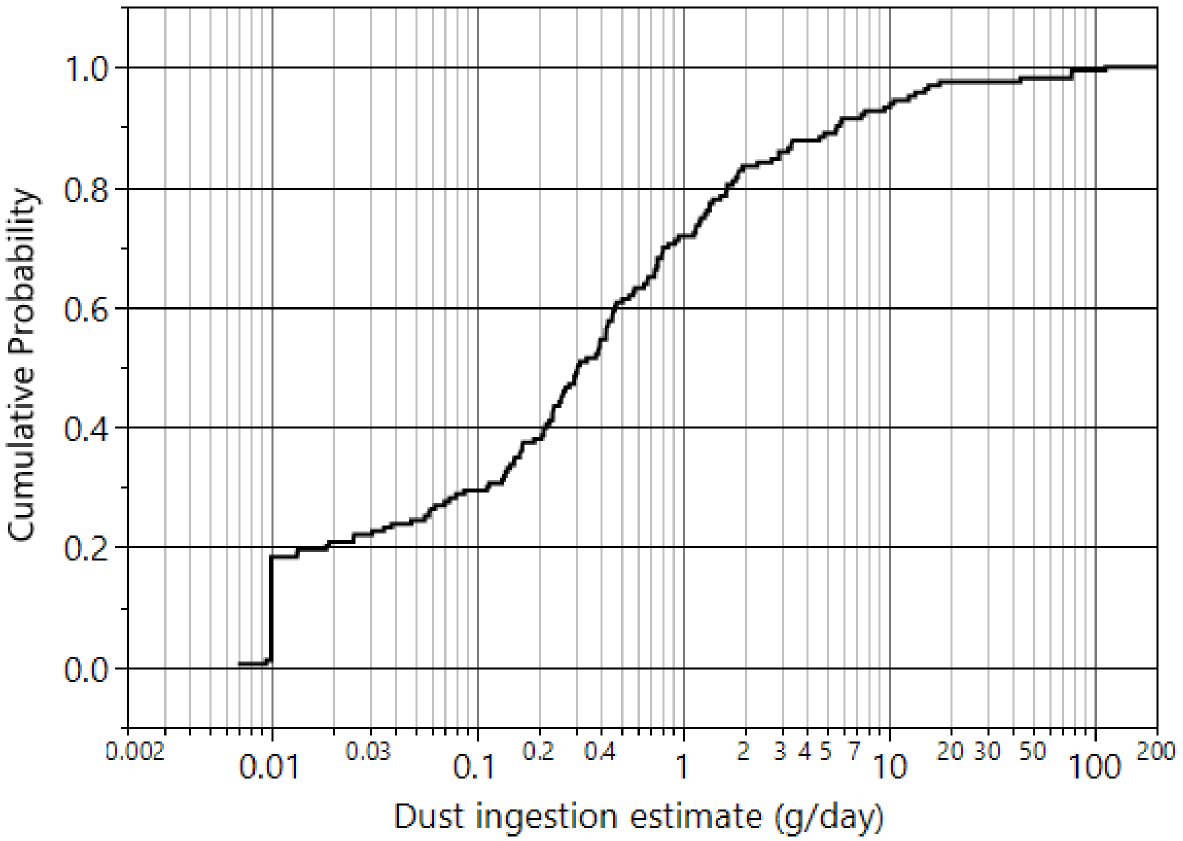

Figure 2. Cumulative probability of the external dust ingestion estimates in the general population of Lubumbashi, data based on the Best Tracer Method and including all data (n=163), the negative dust estimates (18 % of the data) were replaced by a positive low estimate of 0.01 g day^{-1}.