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W.I. Hagens | J.P.A. Lijzen | A.J.A.M. Sips | A.G. Oomen

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W.I. Hagens[1]
J.P.A. Lijzen[2]
A.J.A.M. Sips[1]
A.G. Oomen[1]

Contact:
Werner Hagens
[1] Centrum voor Stoffen en Integrale Risicoschatting (SIR)
[2] Laboratorium voor Ecologische Risicobeoordeling (LER)
Rijksinstituut voor Volksgezondheid en Milieu (RIVM)

Werner.Hagens@rivm.nl

This investigation has been performed by order and for the account of DGM-BWL, within the framework of Risico's in relatie tot bodemkwaliteit (M/711701); Implementatie biobeschikbaarheid mens

RIVM, P.O. Box 1, 3720 BA Bilthoven, the Netherlands Tel +31 30 274 91 11 www.rivm.nl

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Rapport in het kort

De bioaccessibility en relatieve biobeschikbaarheid van lood uit bodems onder nuchtere en gevoede condities: De afleiding van een correctie factor voor de "gemiddeld fysiologische status".

Het RIVM heeft meer en nauwkeurigere gegevens gegenereerd om te berekenen hoeveel lood mensen, kinderen in het bijzonder, binnenkrijgen via de bodem. De bodem in Nederland is op een groot aantal locaties, vooral in binnensteden, verontreinigd met lood. Een hoge loodconcentratie in de bodem vormt een (potentieel) risico voor de gezondheid. Vooral kinderen lopen gevaar omdat ze grotere hoeveelheden grond via hun handen inslikken en de effecten van lood groter zijn voor kinderen.

De fysiologische status van de mens, nuchter, gevoed of gemiddeld, is van invloed op de mate waarin het menselijk lichaam lood uit de bodem opneemt, en daarmee op de risicobeoordeling. Zo wordt lood minder opgenomen na een maaltijd (gevoede condities) dan als iemand niet heeft gegeten (nuchtere condities). Voorheen waren de nuchtere condities het uitgangspunt. Aangezien de gemiddelde conditie een realistischer uitgangspunt is, zijn de gegevens voor de risicobeoordeling daarvoor gecorrigeerd.

Tot op heden was de correctiefactor voor de gemiddelde voedingscondities afgeleid met 11 bodems. In dit onderzoek is de correctiefactor afgeleid van een veel grotere hoeveelheid bodems (45). Met de correctiefactor op basis van dit onderzoek kunnen de gegevens voor nuchtere condities worden gecorrigeerd. Deze nieuwe kennis kan worden gebruikt voor normstelling voor lood in de bodem en voor locatiespecifieke risicobeoordeling van bodemverontreiniging.

Trefwoorden:
Relatieve biobeschikbaarheid, bioaccessability, lood, gevoed, nuchter,

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Summary

Many sites in the Netherlands are contaminated with lead due to (historical) use and misuse. This lead can form a potential health risk. Especially young children are at risk since children ingest larger quantities of soil particles than adults because of hand-to-mouth behaviour. Moreover, from literature it is known that children are specifically susceptible to the effects of lead.

From literature, it is known that lead is better absorbed in fasted conditions and, hence, better bioavailable, than in fed conditions. Therefore, the choice of the physiological condition (i.e. fasted, fed or average fed/fasted state) that is used to estimate the bioavailability of lead from soil influences the value of the relative bioavailability factor to be used in current risk assessment. Dutch policy makers have indicated that they would encourage a more “average physiological state” condition to be used in risk assessment of lead in soil.

With the RIVM *in vitro* digestion model an estimation of the bioavailability of lead from soil can be made. To estimate the bioavailability of lead from soil for the “average physiological state”, the same soil should be analyzed with the *in vitro* digestion method for both fasted and fed conditions. However, another possibility is to derive an “average physiological state” correction factor (CF_{APS}) to estimate the bioaccessibility of lead from soil for the “average physiological state” based on a conversion of the bioaccessibility for only fasted (or fed) conditions.

The objective of this research is to derive an “average physiological state” correction factor (CF_{APS}) that can be used to estimate the relative bioavailability factor (*Rel* F) of lead from soils for average physiological conditions based on *in vitro* determined bioaccessibility of lead for only fasted (or fed) conditions. This *Rel* F_{APS} can be used as input for *Sanscrit* (sanerings criterium model) to assess the potential health risk of the lead contaminated soil (for a child under average physiological conditions). Previously, the “average physiological state” correction factor (CF_{APS}) was derived for only 11 separate soils (with 2 different food types). By order of the Ministry of Housing, Spatial Planning and Environment, this correction factor is now further substantiated by investigation of 45 additional soils.

In conclusion, the new dataset of 70 samples (Table 5) results in a smaller difference between the bioaccessibility of lead from soils for fasted and fed conditions. As a consequence, the ratio fed-fasted (α) and its derived “average physiological state” correction factor (CF_{APS}; Formula 3) are higher for the given percentiles compared to the current dataset (based on 11 soils). As the current data set is based on 11 soils only, a conservative P80 value of CF_{APS} (=0.855) was chosen for the scientific foundation of the intervention value of lead in soil (NOBO notes, VROM 2006). Based on the new dataset (70 samples), the P80 value of $CF_{APS,\ NEW}$ will become 0.922. However, since the new dataset with 70 samples results in a better foundation of the CF_{APS}, a lower percentile could be chosen by policy makers (P50-P80), depending on the level of conservatism that is desired.

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Contents

1 Introduction

Many sites in the Netherlands are contaminated with lead due to (historical) use and misuse of lead containing products and waste. This lead can form a potential health risk. Especially young children are at risk since children ingest larger quantities of soil particles than adults because of hand-to-mouth behaviour (Oomen *et al.,* 2006 and references therein). Moreover, it is known that children absorb lead better than adults. It is reported that lead is (partly) absorbed by the same mechanism as calcium. Calcium is better absorbed in children than in adults as growth demands more calcium (references in Oomen *et al.,* 2006). Also from a toxicological point of view, lead already affects children at low doses, resulting, among others, in impaired neurobehavioral functioning and decreased haemoglobin levels (IPCS, 1995; Baars *et al.,* 2001).

1.1 Bioavailability and bioaccessibility

According to the general interpretation in pharmacology, oral bioavailability (F) is defined as the fraction of an orally administered dose that reaches the systemic circulation. Oral bioavailability (F) can be divided into 3 different major processes (formula 1; Oomen *et al.,* 2006).

(1) $F = F_B \times F_A \times F_H$

After ingestion of soil, the total bioavailability (F) depends on the amount of contaminant that is released from the matrix (i.e. soil) during the digestion in the gastrointestinal tract (Bioaccessibility; F_B). This released fraction is considered to represent the total amount of contaminant available for transport across the intestinal epithelium and able to reach the portal vein (absorbed fraction; F_A). Metabolization of the contaminant may occur in the intestinal epithelium and/or in the liver (first-pass effect). The fraction that is not metabolized (F_H) will be transported throughout the body and represents the bioavailable fraction (F). Please note that if lead is the contaminant, no metabolization is expected, resulting in a F_H fraction of 1 (for more information: Oomen *et al.,* 2006).

1.2 Risk assessment

In standard risk assessment of contaminated soils, the potential risk (for children) might be overrated. This overestimation could lead to unnecessary remediation of sites resulting in high clean up costs and possible social unrest in the contaminated area. Site specific information concerning the oral bioavailability of lead from soil by the human body can make the risk assessment more realistic.

Insight into the uptake process of lead can be obtained by simulating the human digestion process. With *in vitro* digestion models, the potential risk can be estimated by predicting the amount of contaminant (i.e. lead) that, for physiological conditions, would be released from the matrix (i.e. soil) after ingestion (bioaccessibility; F_B) and would, in principle, be ready for uptake in the body (Oomen *et al.,* 2006). With the RIVM *in vitro* digestion model, risk assessment of contaminated sites can be refined by including site specific bioaccessibility information. In a previous report, the scientific basis of this *in vitro* digestion method has been described (Oomen *et al.,* 2006).

From literature, it is known that lead is better absorbed in fasted conditions and, hence, better bioavailable, than in fed conditions (James *et al.,* 1985; Heard *et al.,* 1983; Heard *et al.,* 1982; Blake *et al.,* 1983). Also, the bioaccessibility appears to be higher for fasted conditions (Oomen *et al.,* 2006; Lijzen *et al.,* 2006). Based on the higher bioaccessibility and higher absorption of lead for fasted conditions, the fasted state is the most conservative state to assess a relative bioavailability factor of lead from soil (i.e gives the highest relative bioaccessibility factor). Therefore, the choice of the physiological condition (i.e. fasted, fed or average fed/fasted) that is used to estimate the bioaccessibility of lead from soil influences the value of the relative bioavailability factor to be used in current risk assessment.

Dutch policy makers have indicated that they would encourage a more “average physiological state” condition to be used in risk assessment of lead in soil (NOBO notes, VROM 2006). The rationale behind this “average physiological state” condition is that a child will not always be totally fed or fasted when contact with soil occurs. As exposure to lead via soil is assumed to be a chronic process, the “average physiological state” condition would therefore be more realistic.

The RIVM *in vitro* digestion model is developed for fasted conditions (for the estimation of the bioavailability of contaminants in soil) and fed conditions (for the estimation of the bioavailability of substances from food). Up till now, the bioaccessibility studies for lead contaminated soils with the *in vitro* digestion model were performed mostly for fasted conditions. To estimate the “average physiological state” for the bioaccessibility of lead from soil, the same soil should be analyzed with the *in vitro* digestion method for both fasted and fed conditions. This implicates that all determinations of the bioaccessibility of lead from soil requires twice as high costs and time as determination of the bioaccessibility for only fasted or fed conditions. Therefore, it can also be chosen to determine the bioaccessibility only for fasted conditions, resulting in a conservative bioaccessibility value, but with less experimental costs. A less conservative approach would be to derive an “average physiological state” correction factor (CF_{APS}) to estimate the bioaccessibility of lead from soil for the “average physiological state” based on a conversion of the bioaccessibility for only fasted or fed conditions. This approach is applicable if a constant difference between the bioaccessibility of lead from soil for fasted and fed conditions is assumed. The applicability of this "average physiological state" correction factor will result in a reduction of experimental time and cost.

Oomen *et al*. (2006) described the difference in bioaccessibility for fasted and fed conditions for 11 soils. For a robust and realistic determination of this "average physiological state" correction factor, additional research is needed. Moreover, an estimate should be made of the time a child is fasted and fed. For instance, if a child eats 3 meals per day, and conditions can be considered fed for 2 hours after the meal, the child is fed for 6 hours per day. It can be assumed that a young child (1-4 years) sleeps 12 h per day, which is not included as potential play time, but merely indicates that the potential play time during a day is the other 12 h. Hence, the child is assumed to be in a fed state for 6 of the 12 h (half of the time) and in the fasted state for the other half of the time. Therefore, the weighted average bioavailability factor should be based on half the time fed and half the time fasted conditions (Oomen *et al.,* 2006).

1.3 "Average physiological state" correction factor

Up until now, the information on bioaccessibility of lead from soil for fed conditions (spaghetti and breakfast meal) was available for only 11 soils. The ratio between the bioaccessibility for fed and fasted conditions (= "α") can be calculated with the following formula (2). The fed/fasted ratios for the 11 soils and their percentile values are listed in Table 1.

(2) $F_{B\ soil,\ Fed} = \alpha \times F_{B\ soil,\ Fasted}$

$F_{B\ soil,\ Fed}$ = Bioaccessibility of lead from soil as estimated for fed conditions

$F_{B\ soil,\ Fasted}$ = Bioaccessibility of lead from soil as estimated for fasted conditions

α = ratio bioaccessibility fed/fasted from soils

As previously indicated, it is assumed that a child is in a fed state for half of the time and in the fasted state for the other half of the time. Therefore, the "average physiological state" correction factor can be derived based on the "ratio fed/fasted" with the following formula (3) (Lijzen *et al.,* 2006). The percentile values for the "average physiological state" correction values are listed in Table 1.

(3) $CF_{APS} = (1 + \alpha)/2$

CF_{APS} = "Average physiological state" correction factor

α = ratio bioaccessibility fed/fasted from soils

1 = the fasted conditions are fixed at 1

With this "average physiological state" correction factor, the bioaccessibility of lead from soil for fasted or fed conditions can be converted to the bioaccessibility of lead for average physiological state conditions following formula (4)

(4.1) $F_{B\ soil,\ APS} = F_{B\ soil,\ Fasted} \times CF_{APS}$

(4.2) $F_{B\ soil,\ APS} = F_{B\ soil,\ Fed}\ /\ CF_{APS}$

Table 1: All available data of 11 previously tested soils for both fasted and fed condition (Oomen *et al.,* 2006). The ratio fed/fasted is given for the combined data of fed conditions with a breakfast and spaghetti meal. Furthermore, the percentile values for the ratio fed/fasted (α) and for the "average physiological state" correction values (CF_{APS}) are derived.

Bioaccessibility ratio for fed/fasted conditions					
soil number	**breakfast**	**spaghetti**	**Percentile**		
1	1.00	0.46		**Ratio fed/fasted conditions α**	**"Average physiological state" correction factor CF_{APS}**
2	0.73	0.33			
3	*no data **	*no data **			
4	0.59	0.21			
5	0.52	0.15			
6	0.51	0.54			
7	0.50	0.54	**P50**	0.56	0.781
8	0.71	0.85	**P60**	0.59	0.797
9	0.60	0.67	**P70**	0.66	0.832
10	0.70	0.65	**P80**	0.71	0.855
11	0.58	0.34	**P90**	0.74	0.871
12	0.36	0.74	**P95**	0.85	0.923

*No fed/fasted ratio could be derived from soil 3

The P50, P80 and P90 of the ratio fed/fasted are 0.56; 0.71 and 0.74 respectively (Table 1; Lijzen *et al.,* 2006). In the NOBO (NOrmstelling BOdem) workgroup, it was provisionally decided to use the P80 percentile for the scientific foundation of the intervention value of lead in soil (ratio fed/fasted, α=0.71; "average physiological state" correction factor, CF_{APS}=0.855). At that moment, only the data of the 11 soils were available. A conservative percentile (P80) was chosen rather then a P50 (or another value) to ensure safe application (no underestimation of the potential risk) of this correction factor (NOBO notes, VROM 2006). If the bioaccessibility of lead for both fed and fasted conditions is known for more soils, a lower percentile could be chosen that might reflect a more realistic factor (Lijzen *et al.,* 2006).

1.4 The relative bioavailability factor

In the body, not all bioaccessible lead (release of lead from the matrix) will become bioavailable (uptake of lead in the body). Therefore, the *in vitro* determined bioaccessibility value of lead from

soil ($F_{B,soil}$) should be converted to a bioavailability value (F) of total lead in the body (see also formula 1).
The bioavailability of lead from soil F_{soil} is not equal to the bioavailability of dietary lead based on the studies that were used for deducting the Maximum Permissible Risk (MPR_{human}) for lead F_{MPR} (IPCS 1995; FAO/WHO 1993; Ziegler *et al.,* 1978; Ryu *et al.,* 1983; Baars *et al.,* 2001).
To account for the difference in bioavailability of dietary lead and lead from soil, the relative bioavailability factor (*Rel* F) can be calculated. This relative bioavailability factor (*Rel* F) for lead from soil is calculated by dividing the bioavailability of lead from soil (F_{soil}) by the bioavailability of lead from the MPR studies (F_{MPR}). The relative F is therefore a ratio of two bioavailability values (Oomen *et al.,* 2006).

(5.1) Relative $F = F_{soil} / F_{MPR}$
(5.2) Relative $F = (F_{A,\ soil} \times F_{B,\ soil} \times F_{H,\ soil}) / F_{MPR}$

As indicated in section 1.1, if lead is the contaminant, no metabolization is expected, resulting in a $F_{H,\ soil}$ fraction of 1. In addition, the "average physiological state" for the absorption of soil ($F_{A,}$ soil = 0.8) and the F_{MPR} (= 0.4) are derived in Oomen *et al.* (2006).If these values are introduced in formula 5.2, the relationship between relative bioavailability factor and bioaccessibility of lead from soil becomes:

(6.1) *Rel* $F_{fasted} = 0.8 \times F_{B,\ soil} / 0.4$ ($= 2 \times F_{B,\ soil,\ fasted}$)
(6.2) *Rel* $F_{APS} = F_{B,soil,\ fasted} + F_{B,soil\ fed}$
(6.3) *Rel* $F_{APS} = 2 \times F_{B,\ soil,\ fasted} \times CF_{APS}$

Formula 6.1 describes the *Rel* F_{fasted} of lead from soil for fasted bioaccessibility conditions. Up till now, the derived *Rel* F of all soils is based on the bioaccessibility ($F_{B,soil\ fasted}$) of lead for fasted conditions (formula 6.1). This is thus a conservative value.
Formula 6.2 describes the calculation of *Rel* F_{APS} based on bioaccessibility F_B of lead for fed and fasted conditions. Please note that for formula 6.2, the bioaccessibility of lead per soil should be measured for both fasted and fed conditions.
To derive the *Rel* F_{APS} based on the bioaccessibility of lead for fasted conditions, the average physiological state correction factor (CF_{APS}) can be taken into account (formula 6.3).

1.5 Current use of bioavailability in risk assessment

Dutch risk assessment for lead is based on a criterion laid down by the FAO/WHO (1993) and the IPCS (1995). The recommendation is to avoid lead blood levels above 50 µg/l, resulting in a provisional tolerable weekly intake (PTWI) of 25 µg Pb/kg body weight per week, i.e. or a Tolerable Daily Intake (TDI) of 3.6 µg Pb/kg body weight per day (Baars *et al.,* 2001). This criterion is based on the (MPR) bioavailability ($F_{,MPR}$) of 40% of dietary lead for children (Ziegler *et al.,* 1978).

The Dutch intervention value for lead in soil is currently 530 mg Pb/kg dry matter. However, based on the TDI in humans of 3.6 μg Pb/kg body weight/day, and the *Sanscrit* (sanerings criterium model) exposure scenario (Otte *et al.,* 2007), based on "living with a garden", the intervention value for a child should be 301 mg Pb/kg dry matter (Van den Berg *et al.,* 1995; Lijzen *et al.,* 2006). In this calculation a relative bioavailability factor (*Rel* F) of 1 is used. In a meeting in 2006 at the Ministry of VROM it was provisionally decided to keep the generic intervention value of lead in soil at 530 mg/kg (NOBO notes, VROM 2006). The difference between 301 mg/kg and 530 mg/kg can be explained mainly by:

1. The assumption that children daily ingest on average 100 mg soil (instead of 150 mg before).
2. A correction for a lower bioavailability of lead from soil relative to the bioavailability of dietary lead. This intervention value correction factor amounts 0.74 (instead of 1.0 before). This generic intervention correction factor of 0.74 is based on two components (A and B) (Oomen *et al.,* 2006; Lijzen *et al.,* 2006)
   - A: The P80 percentile of all the relative bioavailability factors determined for soil for fasted conditions (*Rel* F_{fasted}) up till 2006 (=0.87).
   - B: The P80 percentile of the "average physiological state" correction factor (CF_{APS}) of the human gastrointestinal tract, based on 11 soils (=0.854).

   Intervention value correction= p80 "Rel F_{fasted}" x *P80 "CF_{APS}"= 0.87* x *0.855 = 0.74*

Additional information on the derivation of this intervention value correction factor and information to assess the human health risk for specific sites can be found in the reports of Lijzen *et al*. (2006) and Oomen *et al.* (2006).

1.6 Objective

The objective of this research is to derive an "average physiological state" correction factor (CF_{APS}) that can be used to estimate the bioaccessibility of lead from soil for "average physiological state" conditions based on the bioaccessibility determined for fasted conditions. With *Sanscrit*, (Otte *et al.,* 2007) the bioaccessibility for average physiological state conditions can be used to asses the potential health risk of lead contaminated soils.

Previously, the "average physiological state" correction factor (CF_{APS}) was derived from fasted and fed data obtained with the *in vitro* digestion model for 22 samples (11 separate soils with 2 different food types). By order of the Ministry of Housing, Spatial Planning and Environment, this correction factor is now further substantiated by investigation of 45 additional soils. With this study, the CF_{APS} can be determined more reliable, as the number of soils used for the calculation is increased.

2 Materials and methods

2.1 The soils

In 2007, ninety lead contaminated soils were collected for the project "stedelijke ophooglagen". These samples were taken from the "stedelijke ophooglaag", a specific layer present in the soil of relatively old urban cities. This layer is contaminated with lead due to historical usage, former industrial activity, and rubbish. The purpose of this project is to establish a generic factor that correlates the bioavailability of lead from "stedelijke ophooglagen" with their specific soil characteristics. More information concerning this ongoing project "stedelijke ophooglagen" can be found in the "projectplan stedelijke ophooglagen" (Hagens, 2007a) and in the "voortgangsrapportage 2007" (Oomen 2007). The ninety lead contaminated soils were taken from in total 17 Dutch cities. Per city, up to 4 locations were samples and per location, 2 samples were taken. From the 90 soil, the bioaccessibility of lead for fed conditions was estimated for only the 45 odd numbered soil samples and used in this report. This selection of these 45 soil samples represents a balanced set of lead contaminated soils with one soil sample (of the two samples taken per location) for each sampled contaminated location of the project "stedelijke ophooglagen".

The bioaccessibility of lead for fasted and fed conditions was also determined for the reference soil Montana Soil as control sample (both 0.6 and 0.06 grams for fasted conditions; 0.4 and 0.04 for fed conditions). It is noted that the solid-to-liquid ratio for the digestion model for fasted and fed conditions are similar. For fasted conditions 0.06 and 0.6 g soil per digestion tube result in a solid-to-liquid ratio of 1(g soil):958 (l digestion fluid) and 1:96, respectively. For fed conditions 0.04 and 0.4 g soil per digestion tube result in a solid-to-liquid ratio of 1:1063 and 1:106, respectively. The rationale for these solid-to-fluid ratios is described in Oomen *et al.* (2006).

The old dataset of the 11 soils that were used previously to derive the "average physiological state" correction value weighted fed/fasted correction value are also included in this report. It should however kept in mind that for these old *in vitro* bioaccessibility experiments, 0.4 grams(fed) and 0.6 grams (fasted) were used. In 2006, it was decided to preferably use 0.06 grams (fasted) and 0.04 grams (fed) of soil as this represents the amount of daily ingested soil better. Furthermore, this small amount of soil will not result in a pH shift during the *in vitro* digestion experiments which increased reproducibility (Oomen *et al.,* 2006). Therefore, 0.04 grams (fed) and 0.06 grams (fasted) were used for the 45 soils in the new experiments. Even so, the food used for the fed conditions was the spaghetti and breakfast (breakfast conform the study of Maddaloni *et al.,* 1998) for the 11 old soils and Olvarit 15M52 (Nutricia®, the Netherlands) for the 45 new soils.

2.2 *In vitro* digestion model

The RIVM *in vitro* digestion model can simulate both fasted and fed conditions of the human gastrointestinal tract. However, the differences in physiology between fasted and fed state changes the bioaccessibility of contaminants, as pH, salt and enzyme concentrations are different. In the present research, the digestion model simulating fasted and fed condition was used to study the bioaccessibility of lead from 45 soils for both conditions, i.e. simulating the physiological conditions before and shortly after consumption of a meal. The RIVM *in vitro* digestion model for fasted conditions is described by Oomen *et al.* (2003) and Sips *et al.* (2001). The development of the *in vitro* digestion model simulating fed conditions is described by Versantvoort *et al.* (2004; 2005).

2.2.1 Fasted conditions

The *in vitro* digestion model for fasted conditions starts by introducing 9 ml of saliva of pH 6.5 ± 0.2 to 0.06 g (or 0.6 g) of soil (dry weight). This mixture is rotated head-over-heels for 5 minutes at 55 rpm. Subsequently, 13.5 ml of gastric juice (pH 1.07 ± 0.07) is added, and the mixture is rotated for 2 hours. The pH of the mixture is measured. The mixture of saliva and gastric juice usually has a pH of about 1.2, and the allowed pH interval in the presence of soil is 1.5 ± 0.5. Finally, 27 ml of duodenal juice (pH 7.8 ± 0.2) and 9 ml bile (pH 8.0 ± 0.2) are added simultaneously, and the mixture is rotated for another 2 h. The pH of this mixture with intestinal juices is measured. The allowed pH interval is 6.0 ± 0.5, also depending on the soil. All digestive juices are heated to 37 ± 2 °C. Mixing is done in a rotator that is also heated to 37 ± 2 °C. At the end of the *in vitro* digestion process, the digestion tubes are centrifuged for 5 minutes at 3000 g, yielding the chyme (the supernatant) with and the digested soil (the pellet).

The bioaccessibility values of the 90 soil samples were measured for fasted state (in duplo) with 0.06 gram of soil. This work was conducted as part of the project “stedelijke ophooglagen”. Of this group of 90 samples, only the 45 odd numbered soil samples were used in this study. The bioaccessibility of the 11 old soils (fasted state) were performed with 0.6 gram soil. In addition, a blank (duplo) was measured (fasted conditions, no soil) that was used to correct for the background concentration of lead during the experiments.

2.2.2 Fed conditions

The *in vitro* digestion simulating fed conditions starts by introducing 0.04 g (or 0.4 g) of soil (dry weight) to 6 ml stimulated saliva (pH 6.8 ± 0.2) and 4.5 g food (spaghetti, breakfast [conform the study of Maddaloni *et al.,* 1998], or Olvarit [product number 15M52, Nutricia®, the Netherlands]). Immediately, 12 ml of stimulated gastric juice (pH 1.30 ± 0.02) is added and the mixture is rotated head-over-heels (55 rpm) for 2 h. The pH of the gastric fluid is determined, and the allowed interval is 2.5 ± 0.5. Subsequently, 12 ml stimulated duodenal juice (pH 8.1 ± 0.2), 6 ml stimulated bile (pH 8.2 ± 0.2), and 2 ml sodium bicarbonate (84.7 g/l) are added simultaneously. The mixture is rotated for another 2 h and the pH of the chyme was determined, with the allowed pH-interval 6.5 ± 0.5.

Separation of chyme and pellet was obtained by centrifugation at 3000 g for 5 minutes. The whole process is performed at 37 ± 2 °C. Samples can be taken from the stomach and intestinal phase to obtain information on the bioaccessibility of the contaminant.

The bioaccessibility of the 45 new soils (fed conditions) were measured (in duplo) with 0.04 gram of soil per experiment. In contrast, the bioaccessibility of the 11 old soils (fasted state) were performed with 0.4 gram soil. In addition, a blank (duplo) was measured (fed conditions, no soil) that was used to correct for the background concentration of lead during the experiments.

2.3 Analysis

2.3.1 11 old soils

The total lead concentration in the 11 old soils (2006 data) was previously detected by a method based on the destruction of the soil with diluted aqua regia (1:3 with water) followed by microwave assisted destruction. This procedure dissolves lead in the hot and acid mixture. The lead concentration was detected with ICP-MS.

2.3.2 45 new soils

The total lead concentration in the 45 new soils (2008 data) was detected by a method based on the destruction of the soil with undiluted aqua regia followed by microwave assisted destruction. This procedure dissolves lead in the hot and acid mixture. The lead concentration was detected with ICP-AES. These experiments were carried out by ALcontrol BV (Hoogvliet, The Netherlands) according AS3000; NEN5709; AS3010; NEN6966 and NEN6961.

2.3.3 Chyme

After the digestion experiment, the supernatant (=chyme) was collected. The concentration of lead in chyme for both old en new soils (fed and fasted conditions) was detected with ICP-MS.

2.4 Calculating the factor fed/fasted

The amount of lead in the chyme after the *in vitro* digestion for fasted and fed conditions is expressed as a percentage of the total amount of lead present in the soil. This gives a bioaccessibility (expressed in percentage for both fasted and fed conditions). Per soil, the bioaccessibility for both fasted and fed conditions is determined in duplo. As a result, the bioaccessibility for both fasted and fed conditions per soil is expressed as an average of the duplo values. To calculate, per soil, the ratio fed / fasted (α), the average bioaccessibility of the fed state was divided by the average bioaccessibility of the fasted state. This ratio was used to calculate the "average physiological state" correction factor (CF_{APS}).

The old dataset of the 11 soils that were used previously to derive the “average physiological state” correction factor (CF_{APS}) are also included in this report. The *in vitro* digestion experiments for the 11 soils were performed for fed conditions with both spaghetti and breakfast (Oomen *et al.,* 2006).

2.5 Statistics

Statistical analyses to investigate whether the old dataset with 0.6 gram soil (fasted) and 0.4 gram soil (fed) differed from the new dataset with 0.06 gram soil (fasted) and 0.04 gram soil (fed) were performed by a two-tailed Student’s t-test. To investigate whether the type of food (spaghetti, breakfast and Olvarit 15M52) significantly effects the bioaccessibility, a one-way ANOVA followed by the LSD Post-Hoc test was conducted. P-values lower than 0.05 were considered statistically significant.

3 Results

3.1 New bioaccessibility data

The bioaccessibility of lead for fasted and fed situations was calculated according the guideline for determining the oral bioavailability of lead from soil (Hagens *et al.,* 2007b). For each soil, the lead concentration in chyme (bioaccessible fraction) for fasted and fed conditions was divided by the total lead content of the soil (as measured by ALcontrol BV).
Since all experiments were performed in duplo, the average bioaccessibility for fasted and fed conditions was calculated.

Table 1 shows the results of the *in vitro* digestion model with the 45 selected soils and the reference soil (Montana Soil 2710). The lead content of the chyme (in µg), the total lead content of the soil (in µg/gram dry soil) and the bioaccessibility for fasted and fed conditions (= lead content of chyme divided by total lead content; as %) of each soil (in duplo) is represented in the various columns. The ratio fed/fasted (α) is calculated per sample.

Table 1: Calculated bioaccessibility data of the 45 soils (+ reference Montana Soil 2710) for fasted and fed conditions. For each soil, the ratio fed/fasted (α) is given.

Soil Number	Total Pb in soil	Pb (µg) in chyme (fasted)				Bioaccessibility FASTED (%)				Pb (µg) in chyme (fed)				Bioaccessibility FED (%)				Ratio fed/fasted (α)
	µg/gr-ds	duplo I	duplo II	avarage	stdv	duplo I	duplo II	avarage	stdv	duplo I	duplo II	avarage	stdv	duplo I	duplo II	avarage	stdv	
MS2710 (0.4g)	5532 by NIST	348.2	649.6	518.4	154.4	6.3	11.7	9.0	3.9	553.5	990.8	772.1	309.3	10.0	17.9	14.0	5.6	**1.55**
MS2710 (0.04g)	5532 by NIST	2240.8	2241.9	2422.5	313.9	40.5	40.5	40.5	0.0	2185.6	2065.8	2125.7	84.7	39.5	37.3	38.4	1.5	**0.95**
MS2710 (0.04g)	5532 by NIST	1970.0	2350.9	2355.4	387.6	35.6	42.5	39.1	4.9	2291.0	1917.0	2104.0	264.5	41.4	34.7	38.0	4.8	**0.97**
1	1800	1110.7	1108.2	1109.4	1.8	61.7	61.6	61.6	0.1	1132.8	771.2	952.0	255.7	62.9	42.8	52.9	14.2	**0.86**
3	2300	1222.6	1353.8	1288.2	92.8	53.2	58.9	56.0	4.0	866.6	798.9	832.8	47.9	37.7	34.7	36.2	2.1	**0.65**
5	1300	446.7	694.8	570.7	175.5	34.4	53.4	43.9	13.5	402.6	335.2	368.9	47.7	31.0	25.8	28.4	3.7	**0.65**
7	1600	541.7	477.5	509.6	45.4	33.9	29.8	31.9	2.8	369.2	569.2	469.2	141.5	23.1	35.6	29.3	8.8	**0.92**
9	900	904.5	648.4	776.5	181.1	100.5	72.0	86.3	20.1	338.0	492.0	415.0	108.9	37.6	54.7	46.1	12.1	**0.53**
11	810	293.6	312.4	303.0	13.3	36.2	38.6	37.4	1.6	248.1	303.1	275.6	38.9	30.6	37.4	34.0	4.8	**0.91**
13	480	238.7	162.8	200.7	53.7	49.7	33.9	41.8	11.2	256.6	90.1	173.4	117.7	53.5	18.8	36.1	24.5	**0.86**
15	710	308.9	326.4	317.6	12.4	43.5	46.0	44.7	1.7	209.1	258.6	233.8	35.0	29.4	36.4	32.9	4.9	**0.74**
17	920	506.1	508.1	507.1	1.4	55.0	55.2	55.1	0.2	290.7	174.7	232.7	82.0	31.6	19.0	25.3	8.9	**0.46**
19	1700	595.2	352.5	473.8	171.6	35.0	20.7	27.9	10.1	332.0	442.7	387.3	78.3	19.5	26.0	22.8	4.6	**0.82**
21	1100	297.6	350.3	324.0	37.2	27.1	31.8	29.5	3.4	309.5	123.0	216.3	131.9	28.1	11.2	19.7	12.0	**0.67**
23	390	341.4	372.4	356.9	21.9	87.5	95.5	91.5	5.6	164.7	217.0	190.8	37.0	42.2	55.6	48.9	9.5	**0.53**
25	560	276.8	260.9	268.9	11.3	49.4	46.6	48.0	2.0	195.1	301.7	248.4	75.4	34.8	53.9	44.4	13.5	**0.92**
27	810	200.1	265.3	232.7	46.1	24.7	32.7	28.7	5.7	174.5	167.9	171.2	4.6	21.5	20.7	21.1	0.6	**0.74**
29	530	292.2	264.2	278.2	19.8	55.1	49.8	52.5	3.7	226.6	159.8	193.2	47.2	42.7	30.2	36.5	8.9	**0.69**
31	610	291.1	138.2	214.7	108.1	47.7	22.7	35.2	17.7	160.3	201.8	181.0	29.3	26.3	33.1	29.7	4.8	**0.84**
33	1400	819.3	897.2	858.2	55.1	58.5	64.1	61.3	3.9	519.9	360.0	440.0	113.1	37.1	25.7	31.4	8.1	**0.51**
35	2300	841.7	248.3	545.0	419.6	36.6	10.8	23.7	18.2	460.4	535.6	498.0	53.1	20.0	23.3	21.7	2.3	**0.91**
37	1200	331.6	251.1	291.3	56.9	27.6	20.9	24.3	4.7	176.2	175.5	175.8	0.5	14.7	14.6	14.7	0.0	**0.60**
39	460	274.2	321.2	297.7	33.2	59.6	69.8	64.7	7.2	184.9	423.0	304.0	168.4	40.2	92.0	66.1	36.6	**1.02**
41	520	480.3	342.2	411.3	97.7	92.4	65.8	79.1	18.8	179.7	137.0	158.4	30.2	34.6	26.3	30.5	5.8	**0.39**
43	1000	893.9	845.9	869.9	34.0	89.4	84.6	87.0	3.4	698.4	406.0	552.2	206.8	69.8	40.6	55.2	20.7	**0.63**
45	640	331.2	329.6	330.4	1.1	51.8	51.5	51.6	0.2	399.7	238.6	319.1	113.9	62.5	37.3	49.9	17.8	**0.97**
47	1300	821.7	668.4	745.1	108.4	63.2	51.4	57.3	8.3	443.5	450.5	447.0	5.0	34.1	34.7	34.4	0.4	**0.60**
49	1100	848.3	769.9	809.1	55.5	77.1	70.0	73.6	5.0	460.7	404.7	432.7	39.6	41.9	36.8	39.3	3.6	**0.53**
51	370	268.0	235.6	251.8	22.9	72.4	63.7	68.1	6.2	118.8	78.0	98.4	28.8	32.1	21.1	26.6	7.8	**0.39**
53	480	481.2	455.3	468.3	18.3	100.3	94.9	97.6	3.8	192.7	121.7	157.2	50.2	40.1	25.4	32.7	10.5	**0.34**
55	2800	442.2	380.7	411.5	43.5	15.8	13.6	14.7	1.6	235.7	103.4	169.5	93.5	8.4	3.7	6.1	3.3	**0.41**
57	1900	1100.7	1444.3	1272.5	243.0	57.9	76.0	67.0	12.8	806.4	595.2	700.8	149.3	42.4	31.3	36.9	7.9	**0.55**
59	660	707.8	216.9	462.4	347.1	107.2	32.9	70.1	52.6	651.3	192.0	421.7	324.8	98.7	29.1	63.9	49.2	**0.91**
61	1300	371.9	401.0	386.4	20.6	28.6	30.8	29.7	1.6	270.2	179.1	224.6	64.4	20.8	13.8	17.3	5.0	**0.58**
63	4800	722.4	594.0	658.2	90.8	15.0	12.4	13.7	1.9	481.4	457.1	469.3	17.2	10.0	9.5	9.8	0.4	**0.71**
65	1300	328.4	305.7	317.0	16.0	25.3	23.5	24.4	1.2	233.9	189.5	211.7	31.4	18.0	14.6	16.3	2.4	**0.67**
67	1700	361.2	565.6	463.4	144.5	21.2	33.3	27.3	8.5	44.5	217.0	130.8	121.9	2.6	12.8	7.7	7.2	**0.28**
69	680	434.2	347.6	390.9	61.2	63.9	51.1	57.5	9.0	172.2	172.3	172.2	0.1	25.3	25.3	25.3	0.0	**0.44**
71	860	356.6	281.6	319.1	53.0	41.5	32.7	37.1	6.2	216.8	143.4	180.1	51.8	25.2	16.7	20.9	6.0	**0.56**
73	920	357.2	385.2	371.2	19.8	38.8	41.9	40.3	2.1	292.8	187.5	240.2	74.4	31.8	20.4	26.1	8.1	**0.65**
75	3500	1538.5	1558.0	1548.2	13.8	44.0	44.5	44.2	0.4	290.6	567.5	429.0	195.8	8.3	16.2	12.3	5.6	**0.28**
77	410	199.6	135.7	167.6	45.2	48.7	33.1	40.9	11.0	91.6	66.1	78.8	18.0	22.3	16.1	19.2	4.4	**0.47**
79	560	265.3	285.4	275.3	14.3	47.4	51.0	49.2	2.5	189.3	118.0	153.6	50.4	33.8	21.1	27.4	9.0	**0.56**
81	660	330.1	371.0	350.6	29.0	50.0	56.2	53.1	4.4	266.4	181.7	224.1	59.9	40.4	27.5	33.9	9.1	**0.64**
83	670	183.2	644.5	413.9	326.2	27.3	96.2	61.8	48.7	326.7	196.5	261.6	92.0	48.8	29.3	39.0	13.7	**0.63**
85	580	394.2	382.0	388.1	8.7	68.0	65.9	66.9	1.5	187.5	210.9	199.2	16.5	32.3	36.4	34.3	2.9	**0.51**
87	630	267.9	290.9	279.4	16.3	42.5	46.2	44.4	2.6	210.2	125.0	167.6	60.3	33.4	19.8	26.6	9.6	**0.60**
89	610	439.3	332.6	385.9	75.4	72.0	54.5	63.3	12.4	310.5	282.0	296.2	20.2	50.9	46.2	48.6	3.3	**0.77**
																	average :	**0.67**
																	stdv :	**0.23**

Stdv: Standard deviation

3.2 Old bioaccessibility data

The data of the 11 soils that were used previously to derive the ratio fed/fasted (Oomen *et al.,* 2006) are also included in this report. The *in vitro* digestion experiments of these 11 soils were performed for fed conditions with both spaghetti and breakfast. Hence, the bioaccessibility for fasted and fed conditions and the ratio fed/fasted (α) are represented in table 2 for both types of food.

Table 2: Previously assessed bioaccessibility data of 11 soils for fasted and fed (breakfast and spaghetti) conditions.

Soil number	Bioaccessibility FASTED (%)				Bioaccessibility FED (%)				Ratio fed/fasted (α)
	Fasted				Breakfast				
	duplo I	*duplo II*	avarage	*stdv*	*duplo I*	*duplo II*	avarage	*stdv*	
PU1	*62.6*	*61.4*	62.0	*0.9*	*63.58*	*59.90*	61.7	*2.6*	**1.00**
PU2	*61.1*	*51.3*	56.2	*6.9*	*33.75*	*48.01*	40.9	*10.1*	**0.73**
PU3	*no data **				*32.28*	*27.28*	29.8	*3.5*	*no data **
PU4	*63.3*	*34.4*	48.8	*20.4*	*27.48*	*29.68*	28.6	*1.6*	**0.59**
PU5	*14.7*	*5.1*	9.9	*6.8*	*5.68*	*4.54*	5.1	*0.8*	**0.52**
PU6	*22.2*	*21.4*	21.8	*0.6*	*11.01*	*11.25*	11.1	*0.2*	**0.51**
PU7	*16.0*	*15.3*	15.6	*0.5*	*7.49*	*8.19*	7.8	*0.5*	**0.50**
PU8	*12.2*	*12.7*	12.5	*0.4*	*8.74*	*8.95*	8.8	*0.1*	**0.71**
PU9	*12.2*	*12.7*	12.4	*0.3*	*7.24*	*7.73*	7.5	*0.3*	**0.60**
PU10	*11.5*	*12.4*	11.9	*0.6*	*8.41*	*8.40*	8.4	*0.0*	**0.70**
PU11	*15.6*	*26.3*	20.9	*7.6*	*13.52*	*10.70*	12.1	*2.0*	**0.58**
PU12	*14.2*	*14.9*	14.6	*0.5*	*0.00*	*10.60*	5.3	*7.5*	**0.36**
								average :	**0.62**
								stdv :	**0.17**
Soil number	**Fasted**				**Spaghetti**				**Ratio fed/fasted (α)**
	duplo I	*duplo II*	avarage	*stdv*	*duplo I*	*duplo II*	avarage	*stdv*	
PU1	*62.6*	*61.4*	62.0	*0.9*	*17.14*	*39.44*	28.3	*15.8*	**0.46**
PU2	*61.1*	*51.3*	56.2	*6.9*	*17.63*	*19.86*	18.7	*1.6*	**0.33**
PU3	*no data **				*27.18*	*27.01*	27.1	*0.1*	*no data **
PU4	*63.3*	*34.4*	48.8	*20.4*	*11.15*	*9.27*	10.2	*1.3*	**0.21**
PU5	*14.7*	*5.1*	9.9	*6.8*	*1.61*	*1.30*	1.5	*0.2*	**0.15**
PU6	*22.2*	*21.4*	21.8	*0.6*	*12.24*	*11.18*	11.7	*0.8*	**0.54**
PU7	*16.0*	*15.3*	15.6	*0.5*	*9.31*	*7.67*	8.5	*1.2*	**0.54**
PU8	*12.2*	*12.7*	12.5	*0.4*	*10.25*	*10.96*	10.6	*0.5*	**0.85**
PU9	*12.2*	*12.7*	12.4	*0.3*	*8.48*	*8.21*	8.3	*0.2*	**0.67**
PU10	*11.5*	*12.4*	11.9	*0.6*	*7.43*	*8.05*	7.7	*0.4*	**0.65**
PU11	*15.6*	*26.3*	20.9	*7.6*	*7.52*	*6.74*	7.1	*0.5*	**0.34**
PU12	*14.2*	*14.9*	14.6	*0.5*	*10.66*	*11.01*	10.8	*0.2*	**0.74**
								average :	**0.50**
								stdv :	**0.22**

*: No bioaccessibility data for fasted conditions are available for soil number 3. Hence, no ratio fed fasted could be derived.

Stdv: Standard deviation

3.3 The combination of old en new data

The derivation of an “average physiological state” correction factor (CF_{APS}) will be more reliable if all available data will be used. Therefore, it is suggested to combine the bioaccessibility data for fasted and fed conditions of the 11 old soils and the 45 new soils.
An advantage will be that the derived CF_{APS} could be applied for various soil characteristics. Moreover, the use of different food (Olvarit, breakfast and spaghetti) results in a more realistic CF_{APS}. The rationale for this more realistic CF_{APS} is that a child will be exposed to several different soil types after eating several different types of food.
Table 3 shows the different datasets (I, II and III) that are available up till now with their ratio fed/fasted (α) per soil (70 soils).

Table 3: The ratio fed-fasted (α) determined per soil in the present study (dataset I: Olvarit) and previous studies (dataset II: Breakfast and dataset III: Spaghetti).

Food Type:	**Olvarit**			**Breakfast**	**Spaghetti**
Dataset:	**I**			**II**	**III**
1.55*	0.82	0.63	0.28	1.00	0.46
0.95	0.67	0.97	0.44	0.73	0.33
0.97	0.53	0.60	0.56	0.59	0.21
0.86	0.92	0.53	0.65	0.52	0.15
0.65	0.74	0.39	0.28	0.51	0.54
0.65	0.69	0.34	0.47	0.50	0.54
0.92	0.84	0.41	0.56	0.71	0.85
0.53	0.51	0.55	0.64	0.60	0.67
0.91	0.91	0.91	0.63	0.70	0.65
0.86	0.60	0.58	0.51	0.58	0.34
0.74	1.02	0.71	0.60	0.36	0.74
0.46	0.39	0.67	0.77		

Dataset I: 0.06 (fasted) and 0.04 (fed) gram soil except * : 0.6 (fasted) and 0.4 (fed) gram soil.

Dataset II + III : 0.6 (fasted) and 0.4 (fed) gram soil

Grey: Reference soil Montana 2710

The old and new data have been statistically analysed

- With a two-tailed Student’s T-test, no significant difference is observed between data obtained with **0.6 and 0.4 gram** soil as starting material (respectively fasted and fed conditions of dataset II and III, including one reference soil, indicated with * in table 3) **versus 0.06 and 0.04 gram** soil as starting material (respectively fasted and fed conditions of dataset 1. excluding 1 reference soil indicated with * in table 3) in the *in vitro* digestion method.
- With an ANOVA test, no significant difference was observed between dataset II (breakfast) and dataset III (spaghetti).
- With an ANOVA test, no significant difference was observed between dataset I (Olvarit) and dataset II (breakfast).
- With an ANOVA test a significant difference ($P = 0.019$) was observed between dataset I (Olvarit) and dataset III (spaghetti) (significant difference if $P < 0.05$).

Although dataset I (Olvarit) is significantly different than dataset III (spaghetti) , the use of all available data will result in a more general "average physiological state" correction factor (CF_{APS}), since this value includes several soils with different soil characteristics and several types of food. In addition, the outcome in ratio fed/fasted value (α) and the derived CF_{APS} for the different combinations of datasets is highly comparable (Table 4). Therefore, the relevance of this significant difference is questioned.

3.4 The "average physiological state" correction factor

Based on the datasets listed in table 3, various percentile values for the ratio fed/fasted (α) and the derived "average physiological state" correction factor (CF_{APS}) are calculated for different combinations of the datasets (Table 4). In addition, the average, standard deviation and number of included soils per combined datasets are given in table 4.

Table 4: Percentile values of the ratio fed/fasted (α) and the "average physiological state" correction factor (CF_{APS}) for the various combinations of datasets. In addition, the average, standard deviation and number of included soils per combined datasets are given. The different datasets are listed in table 3.

Food type	Olvarit		breakfast and spaghetti		Olvarit and Breakfast		All data	
Dataset*	I		II + III		I + II		I + II + III	
Avarage	0.67		0.55		0.66		0.64	
Stdv	0.23		0.20		0.22		0.23	
Number of data	48		22		59		70	
Percentile	Ratio fed/fasted (α)	"Average physiological state" correction factor (CF_{APS})	Ratio fed/fasted (α)	"Average physiological state" correction factor (CF_{APS})	Ratio fed/fasted (α)	"Average physiological state" correction factor (CF_{APS})	Ratio fed/fasted (α)	"Average physiological state" correction factor (CF_{APS})
P50	0.64	0.821	0.56	0.781	0.63	0.817	0.62	0.809
P60	0.67	0.837	0.59	0.797	0.67	0.834	0.66	0.828
P70	0.76	0.882	0.66	0.832	0.73	0.866	0.72	0.859
P80	0.89	0.946	0.71**	0.855**	0.86	0.930	0.84	0.922
P90	0.93	0.966	0.74	0.871	0.93	0.964	0.92	0.961
P95	0.97	0.984	0.85	0.923	0.97	0.986	0.97	0.984

*:The datasets are listed in Table 3.

**: The P80 percentile of the old dataset (II and III). This value is provisionally in use (NOBO notes, VROM 2006).

Indicated in grey: The combination of datasets I, II and III (all available data).

Stdv: Standard deviation.

Note that the percentile values for the combined datasets II+III are already discussed in the introduction of this letter report. In the NOBO workgroup, it was provisionally decided to use the P80 percentile (α=0.71; CF_{APS}=0.855) as indicated in Table 4 (NOBO notes, VROM 2006). As indicated in section 3.3, the inclusion of all available data (dataset I, II and III; indicated in grey) will result in a more general CF_{APS}.

4 Discussion

To assess the bioavailability of lead from soil, an *in vitro* digestion model can be used. In default setting at the RIVM, only the fasted physiological state is mimicked. However, from literature, it is known that lead is better absorbed for fasted conditions than for fed conditions (James *et al.,* 1985; Heard *et al.,* 1983; Heard *et al.,* 1982; Blake *et al.,* 1983). This results in an overestimation (worst-case assumption) of the bioavailability of lead from soil since human exposure to soil does not always occur in fasted conditions. This leads to an overestimation of the potential health risk. Dutch policy makers have indicated that they would encourage a more "average physiological state" condition to be used in the derivation of the bioavailability of lead from soil (NOBO notes, VROM 2006). The rationale behind this "average physiological state" situation is that a child will not always be fasted when contact with soil occurs. The "average physiological state" condition would therefore be more realistic since it will mimic the average physiological condition of a child.

Up until now, the information of bioaccessibility of lead from soil for the fed condition was available for only 11 soils with two different fed conditions (breakfast and spaghetti meal). The P50, P80 and P90 of the ratio fed/fasted (α) are 0.56; 0.71 and 0.74 respectively (Lijzen *et al.,* 2006). Based on this limited data set, the NOBO decided to preliminary use the P80 percentile (α=0.71; CF_{APS}=0.855) (NOBO notes, VROM 2006). With this study, the CF_{APS} can be determined more reliable, as the number of soils used for the calculation is increased Moreover, the application of this CF_{APS} will result in a reduction in experiments, time and cost since the bioaccessibility of lead for fasted or fed conditions can be converted to the "average physiological state" conditions (see also formula 4). As already stated earlier, this approach is only applicable if a constant difference between the bioaccessibility of lead from soil for fasted and fed conditions is assumed. Indeed, table 3 indicates that roughly, this assumption is correct.

The objective of this research was to derive an "average physiological state" correction factor (CF_{APS}) that can be used to estimate the relative bioavailability factor (*Rel* F) of lead from soils for average physiological conditions based on *in vitro* determined bioaccessibility of lead for only fasted (or fed) conditions. This *Rel* F_{APS} can be used as input for *Sanscrit* (Otte *et al.,* 2007) to assess the potential health risk of the lead contaminated soil (for a child under average physiological conditions).

This letter-report provides the needed additional bioaccessibility data of lead from soil for both fasted and fed conditions. With this new data, combined with the previous dataset, the derived CF_{APS} is based on 70 different Dutch soils with a range of soil and lead characteristics. Moreover, the fed condition is estimated with 3 different relevant meals, including breakfast and spaghetti. The rationale to combine the old data with the new data is that a child will play at different locations and hence, be exposed to several different soil types and lead characteristics. Moreover, a child will be exposed during the entire "estimated potential play time" of 12 hours per day (Oomen *et al.,* 2006). This includes playing after eating breakfast, lunch and diner (= different types of food). Taken together, the total dataset (2008) results in a more realistic CF_{APS}, applicable for different soils and various meals.

The percentile values for both the ratio fed/fasted conditions (α) and for the CF_{APS} (see also Tables 4 and 5) are rather similar for the different combinations of datasets. Therefore, **we suggest using all available data possible since this will result in a more realistic CF_{APS}, based on 70 soils (table 5). The percentile values given in table 5 for the "newly proposed dataset" (= 2008 dataset) can all be used in risk assessment. However the decision which percentile should be used in risk assessment is a decision that should be made by policy makers, depending on the level of protection that is required.**

This letter report will also be included in a RIVM report in (expected in 2008). This report describes the results of the project "stedelijke ophooglagen". This RIVM report will report percentile values of the bioaccessibility and relative bioavailability of lead from soils for fasted conditions. With this report, policy makers can choose a realistic percentile values for both $\mathbf{CF_{APS}}$ **and *Rel*** $\mathbf{F_{fasted,\ stedelijke\ ophooglaag}}$ based on average or median (P50) conditions or on a more conservative value (P60-P95) depending on the level of certainty that is required. Both chosen percentile values will derive a generic realistic "intervention value correction factor" for soil from "stedelijke ophooglagen" (see section 1.5).

5 Conclusions

In conclusion, the new dataset of 70 samples (Table 5) results in a smaller difference between the bioaccessibility of lead from soils for fasted and fed conditions. As a consequence, the ratio fed-fasted (α) and its derived "average physiological state" correction factor (CF_{APS}; Formula 3) are higher for the given percentiles compared to the current dataset (based on 11 soils). As the current data set is based on 11 soils only, a conservative P80 value of CF_{APS} (=0.855) was chosen for the scientific foundation of the intervention value of lead in soil (NOBO notes, VROM 2006). Based on the new dataset (70 samples), the P80 value of $CF_{APS,\ NEW}$ will become 0.922. However, since the new dataset with 70 samples results in a better foundation of the CF_{APS}, a lower percentile could be chosen by policy makers (P50-P80), depending on the level of conservatism that is desired.

Table 5: Summary of results for policy makers: Percentile values of the ratio fed/fasted (α) and the "average physiological state" correction factor (CF_{APS}) for the current (provisional) dataset (old; data till 2006) that is used in risk assessment and for the newly proposed dataset (new + old; all available data = 2008 dataset). In addition, the average, standard deviation and number of included soils per combined datasets are given. The different datasets are listed in table 3.

Dataset*	Current provisional dataset (2006), used in risk assessment II + III		Newly proposed dataset (2008), used in risk assessment I + II + III	
Avarage	0.55		0.64	
Stdv	0.20		0.23	
Number of data	22		70	
Percentile	**Ratio fed/fasted (α)**	**"Average physiological state" correction factor** (CF_{APS})	**Ratio fed/fasted (α)**	**"Average physiological state" correction factor** (CF_{APS})
P50	0.56	0.781	0.62	0.809
P60	0.59	0.797	0.66	0.828
P70	0.66	0.832	0.72	0.859
P80	0.71**	0.855**	0.84	0.922
P90	0.74	0.871	0.92	0.961
P95	0.85	0.923	0.97	0.984

*: The datasets are listed in Table 3

**: The P80 percentile of the 2006 dataset (II and III). This value is provisionally used in the derivation of the current intervention value of lead (NOBO notes, VROM 2006; Oomen *et al*., 2006)

Stdv: Standard deviation

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National Institute
for Public Health
and the Environment

P.O. Box 1
3720 BA Bilthoven
The Netherlands
www.rivm.com