Alternative Methods for the Prediction of Relative Bioavailability of Arsenic in Mining Soils

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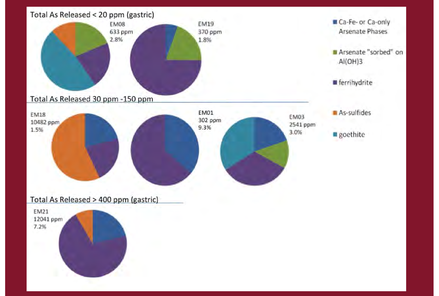
ABSTRACT

The current "gold standard" for evaluation of the oral bioavailability of arsenic (As) in soils is the juvenile swine in vivo method, which is approved by the US EPA for use in human health risk assessments (HHRA). Risk assessment calculations typically utilize default oral toxicity values, which are based on ingestion of readily soluble forms of As such as sodium arsenate (NaAs). However, mining soils in California are relatively high in iron hydroxide phases which bind As strongly, resulting in reduced solubility. The in vivo method compares the absorption of As in mining soils to NaAs. Absorption is quantified by measuring As excreted in urine over a 48 hour period. This data is plotted into a dose response curve and the ratio of the slope of the line between the two substances yields a value that represents the relative bioavailability (RBA) of the test sample to NaAs. This value can then be used to adjust the toxicity criteria utilized in HHRA's. The difficulty with in vivo studies is that they are cost prohibitive. The aim of the current study is to develop in vitro methods coupled with mineralogical characteristics to accurately predict in vivo RBA results. A total of 24 soil samples were collected from the Empire Mine State Historic Park (EMSHP) in California to assist in the development of these methods. Soils were homogenized and screened down to less than 250 um fractions and distributed to the investigative team for various analyses. Six of these 24 samples were identified as being of particular interest based on various criteria including total As content (302-12,041 mg/kg), predicted in vitro bioaccessibility (IVBA) (1.5-9.3%), and mineralogical content. These 6 soils were then analyzed using the in vivo method. RBA's ranged from 3.8 to 19.6%. While the IVBA underestimated RBA, the two values correlated relatively well with an r^2 value of 0.82. Additional bench top methods, including sequential chemical extractions and geochemical analyses, are being developed aimed at better predicting RBA results.

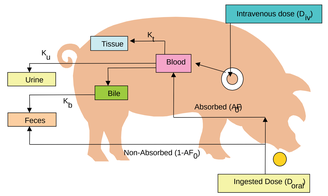
INDENTIFYING SOIL SAMPLES FOR *IN VIVO* ANALYSIS



Minerology as detected using X-Ray Absorption Fine Structure (XAFS) analysis. % represents in vitro bioaccessiblity of arsenic (IVBA).

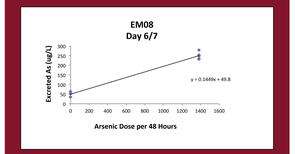
IN VIVO ANALYSIS OF RELATIVE BIOAVAILABILITY IN JUVENILE SWINE

Materials and Methods:



Absorbed As Estimated as a funtion of As released into Urine

Group Number	Sample	Arsenic (mg/kg)
1	EM01	302
2	EM03	2541
3	EM08	633
4	EM18	10482
5	EM19	370
6	EM21	12041
7	Sodium Arsenate	10
8	Negative Control	0



Urinary Excretion Factor (UEF) is the Slope
Relative Bioavailability $(RBA)_{(x)} = UEF_{(x)} / UEF_{(NaAs)}$

Bioavailability for Arsenic in Test Soils

Test Material	90% Confidence Interval			
	RBA Day 6/7	RBA Day 9/10	RBA day 12/13	All Days
EM01	26.8 (20.3-33.4)	29.2 (24.1-34.3)	15.0 (7.8-22.3)	**23.7 (10.9-36.5)**
EM03	17.0 (13.4-20.6)	15.9 (13.0-18.8)	12.9 (11.2-14.6)	**15.3 (11.7-18.8)**
EM08	20.3 (18.4-22.2)	19.5 (14.2-24.8)	17.7 (12.3-23.2)	**19.2 (16.9-21.4)**
EM18	6.8 (5.8-7.7)	4.4 (2.2-6.5)	3.8 (1.3-6.2)	**4.0 (3.3-4.6)**
EM19	13.8 (11.1-16.4)	11.7 (9.5-13.9)	9.8 (7.2-12.5)	**11.7 (8.3-15.2)**
EM21	23.5 (19.1-28.0)	26.0 (22.3-29.8)	19.6 (14.2-25.1)	**23.0 (17.6-28.5)**

Using Bioavailability to Assess Human Health Risk

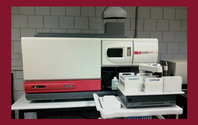
	As (mg/kg)	Default Risk	RBA	Adjusted Risk	RBC ($1x10^{-5}$)	RBC ($1x10^{-4}$)
EM01	302	7.E-04	0.237	2.E-04	1.8	179
EM03	2541	6.E-03	0.153	9.E-04	2.8	278
EM08	633	1.E-03	0.192	3.E-04	2.2	221
EM18	10482	2.E-02	0.04	1.E-03	10.6	1062
EM19	370	9.E-04	0.117	1.E-04	3.6	363
EM21	12041	3.E-02	0.23	7.E-03	1.8	185

Adjusted Risk= Default Risk x RBA
Residential Scenario Soil Ingestion Pathway Only

IN VITRO METHODS FOR PREDICTING BIOAVAILABITY

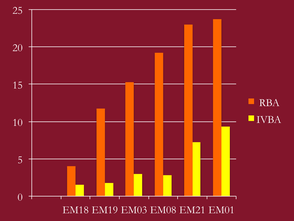
OSU *in vitro* Gastrointestinal Method

Simulated Human Gastric System

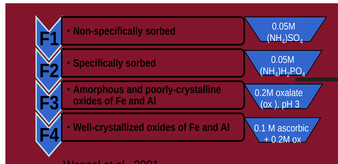


Metal Analysis by ICP

$$\text{In vitro Bioaccessible As (IVBA As), \%} = \frac{\text{Bioaccessible As, mg/kg}}{\text{Total As, mg/kg}} \times 100\%$$



Sequential Chemical Extraction Procedure



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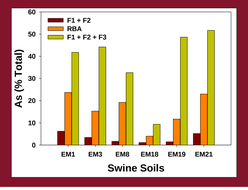

$\sum$ F1-F3 > RBA As > $\sum$ F1-F2

SUMMARY

- Relative Bioavailability (RBA) of Arsenic in Mining Waste Soils ranged from 3.8 to 19.6%
- OSU *in vitro* Bioaccessibility correlates but under predicts *in vivo* RBA
- Sequential Chemical Extraction
  - Fraction 1 + 2 under predicts RBA
  - Fraction 1 + 2 + 3 over predicts RBA

NEXT STEPS

- Evaluate cataloged soils using Sequential Chemical Extractions to determine when F1 + F2 + F3 no longer over predicts RBA. Hundreds of soils available that have previously been evaluated *in vivo*
- Develop a Fraction "2.5" to improve predictability
- Identify of 6 additional site soils to test *in vivo*

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