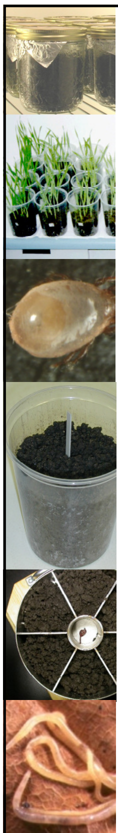

THE USE OF SOIL ECOTOXICOLOGICAL TESTS TO EVALUATE SOIL QUALITY

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http://www1.ci.uc.pt/imar/unit/people/cvs/cv/j_p_sousa.php
http://www.facebook.com/labsolos

Soil Ecology
& Ecotoxicology Lab

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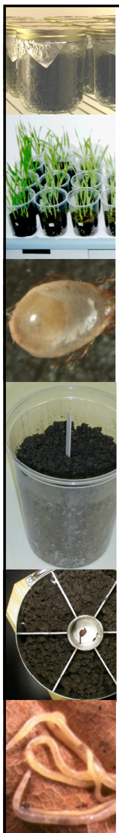
Outline of presentation

- *Importance of Ecotoxicology in Ecological Risk Assessment (ERA)*
- *Major tools (tests) used in Ecotoxicology*
- *Case study from Brazil*
- *Evolution of Soil Ecotoxicology in Brazil and perspectives*



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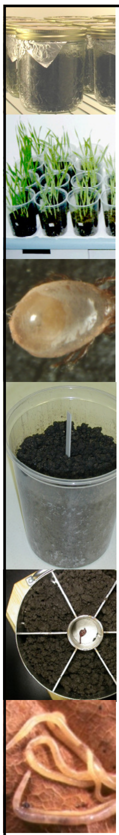
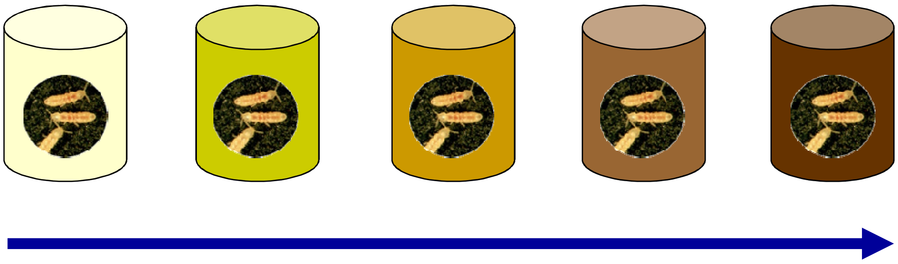
"All substances are poisons. There is none which is not a poison. The right dose differentiates the poison from a remedy"

Paracelcius (1493-1541)



DOSE is the KEY

Control



Increasing concentration of the contaminant or of the contaminated medium;

Determination of NOEC or ECx

Toxicology | Chemistry | Ecology

↓

EcoToxicology → ***Science describing the toxic effects on living organisms, especially in populations and communities within ecosystems (Thuhaut, 1969)***

↓

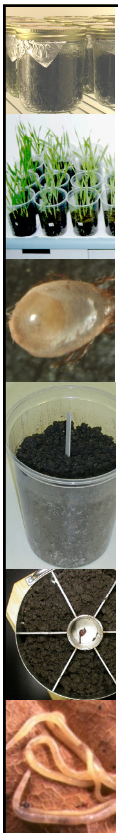
Risk Assessment → ***Science whose task is to assess, monitor and to predict the fate of foreign substances in the environment (Moriarty, 1988)***

↑

Involves Chemical characterization and monitoring and Biological testing and ecological monitoring

↓

Management of the environment



What is *Ecological Risk Assessment* ?

Sutter et al. (2000)

ERA is the process of collecting, organizing and analyzing environmental data to estimate the probability of adverse effects towards ecological receptors (e.g., species, populations, communities, processes) or ecosystems due to contamination.

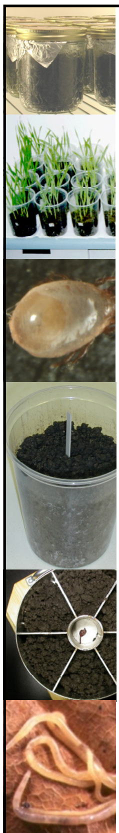
Ecological Risk Assessment - ERA

Prospective ERA

- Assessing future risks;
- Used to evaluate the risk of chemical substances (e.g., pesticides) or soil amendments (e.g. sludge application)
- 'Substance testing'

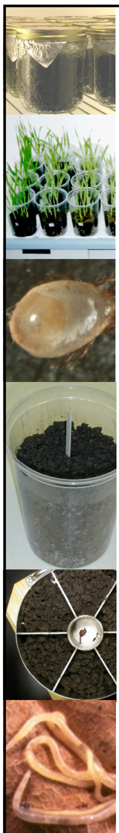
Retrospective ERA

- Assessing or ongoing risks;
- Used to assess risk of contaminated sites
- 'Direct toxicity assessment'



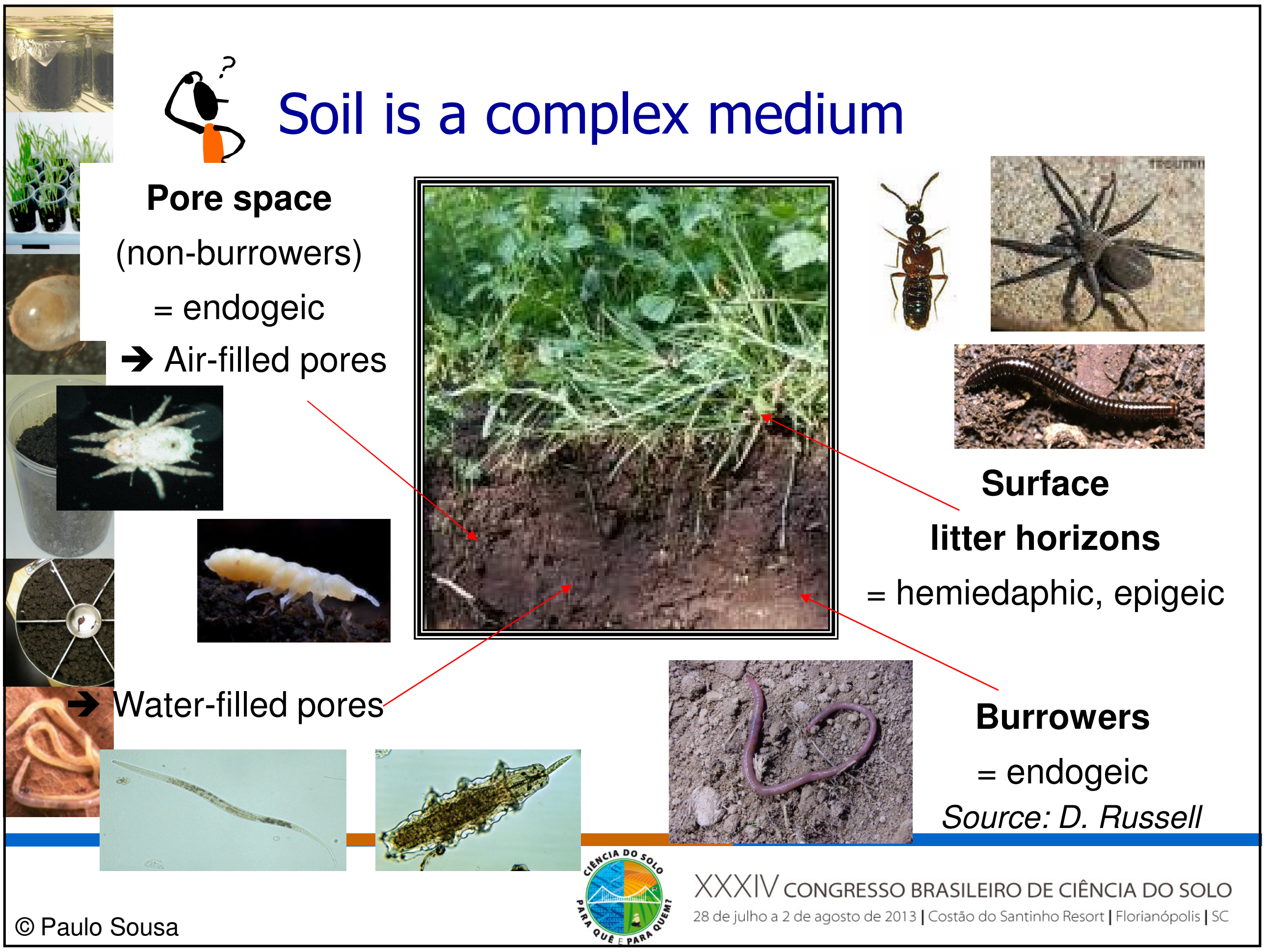
Protection goals
(what do we want to protect?)

- Soil quality and sustainability
- Ability of soil do deliver ecosystems services (Provisioning, Regulatory and Supporting)
- Ecosystem services (and underlying processes) should be protected or restored on a permanent sustainable basis
- Aim has to be established by regulatory actions (e.g. EU biodiversity strategy 2020/2050; EU Soil Thematic Strategy, CONAMA 420/09)



Protection goals
(what do we want to protect?)

Biodiversity (structural and functional) ➲ Ecological processes ➲ Ecossystem services


Soil is a complex medium
Pore space
(non-burrowers)
= endogeic
➔ Air-filled pores
➔ Water-filled pores
Surface
litter horizons
= hemiedaphic, epigeic
Burrowers
= endogeic
Source: D. Russell
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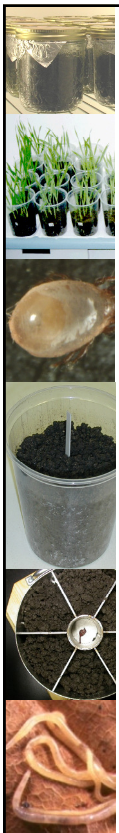
© Paulo Sousa



Protection goals
(what do we want to protect?)

Biodiversity (structural and functional) ➲ Ecological processes ➲ Ecossystem services

But maybe we don't need to protect every species/population at every place at every time!

Which protection goals ?

Which functions / services to protect? ?

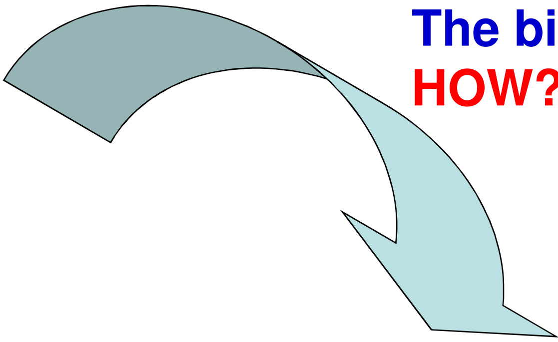
Do we want a multifunctionality of the soil to all its extent?

Intended land-use of the site has to be taken into consideration

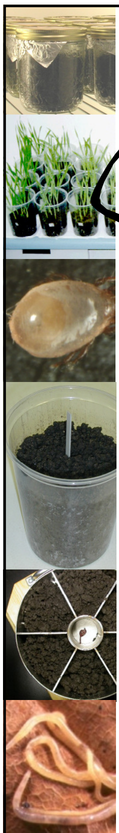
Suitability for Use

Risk-based Land Management

Which functions / services to protect ?



The big question!
HOW?



Which functions / services to protect ?

HOW?

Use of <u>ECOSYSTEM SERVICES</u> concept to tailor the risk assessment

Using endpoints that are linked to Ecosystem Services needed at a certain site

Improves communication with stakeholders
Makes ERA results more perceptible

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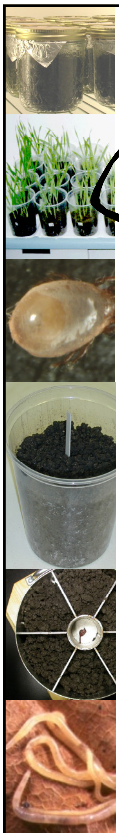
Which functions / services to protect ?

	Ecosystem service	Land use				
		Nature	Agriculture	Parks	Allotment gardens	Industrial sites
Supporting	Nutrient cycling	+	+	+/-	+	+/-
	Soil formation	+	+	+/-	+/-	-
	Primary production	+	+	+/-	+	-
Regulating	Climate regulation	+	+	+/-	+/-	-
	Disease suppression	+	+	+/-	+	-
	Water regulation purification	+	+	+/-	+/-	+

Source: Faber and Wan Wensem, STOTEN (2012)



Use of Ecosystem Services concept

HOW?

Type of land use

↓

Required ecosystem services

↓

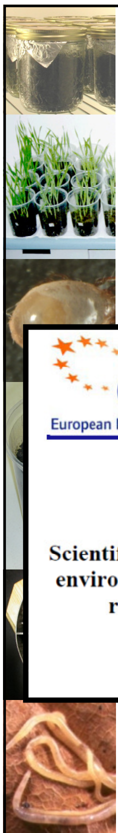
Ecological requirements

↓

Indicator endpoints

→ measurement parameters (ecotoxicological evaluations *sensu lato*)

Use of Ecosystem Services concept



Contents lists available at ScienceDirect

Science of the Total Environment

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

ELSEVIER

Development of a framework based on an ecosystem services approach for deriving specific protection goals for environmental risk assessment of pesticides

Karin M. Nienstedt [a,1], Theo C.M. Brock [b], Joke van Wensem [c], Mark Montforts [d], Andy Hart [e], Alf Aagaard [a,2], Anne Alix [f], Jos Boesten [b], Stephanie K. Bopp [a,*], Colin Brown [g], Ettore Capri [h], Valery Forbes [i,3], Herbert Köpp [j], Matthias Liess [k], Robert Luttik [d], Lorraine Maltby [l], José P. Sousa [m], Franz Streissl [a], Anthony R. Hardy [n]

efsa

European Food Safety Authority

SCIENTIFIC OPINION

Scientific Opinion on the development of specific protection goal options for environmental risk assessment of pesticides, in particular in relation to the revision of the Guidance Documents on Aquatic and Terrestrial Ecotoxicology (SANCO/3268/2001 and SANCO/10329/2002)[1]

EFSA Panel on Plant Protection Products and their Residues (PPR)[2, 3]

e at ScienceDirect

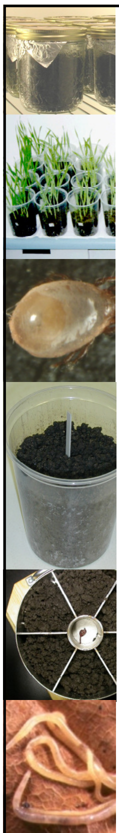
Science of the Total Environment

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

ELSEVIER

Elaborations on the use of the ecosystem services concept for application in ecological risk assessment for soils

J.H. Faber [a,*], J. van Wensem [b]



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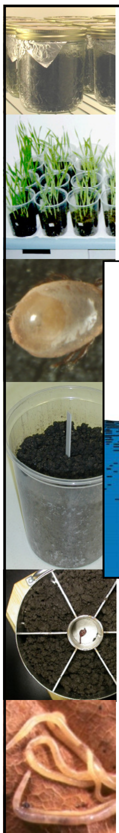
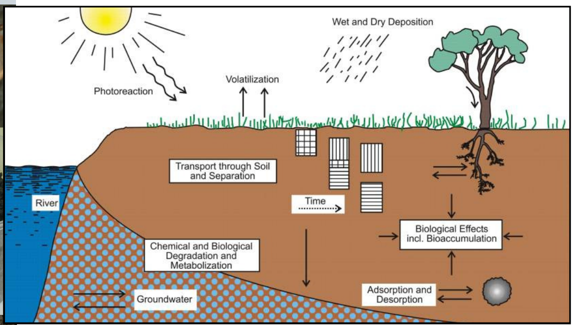


RA=Chemistry + Ecotoxicology

Fate of chemicals

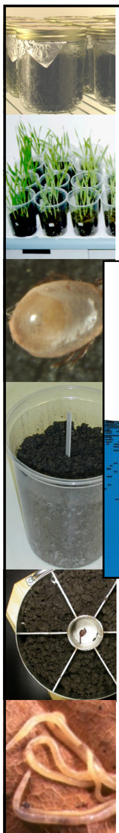


- Chemical concentration in different compartments;
- Modelling
- Comparison with legislated values

Effects of chemicals

- Ecotoxicological testing at different levels of biological organization;
- Ecological monitoring

Risk Assessment



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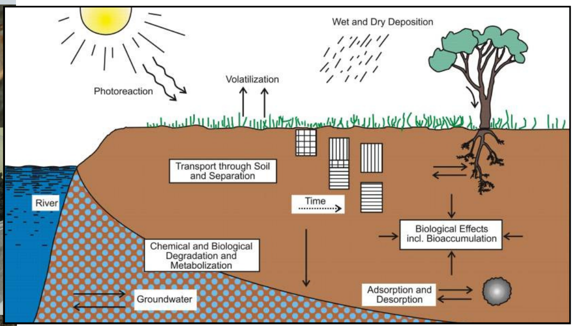

- Chemical concentration in different compartments;
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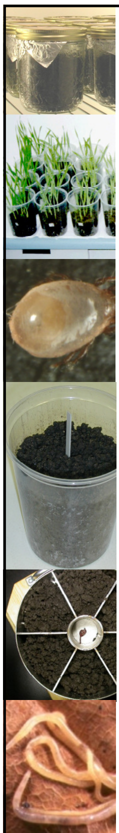
Risk Assessment

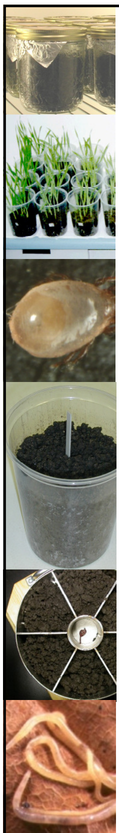


No !

Effects of chemicals

- Total concentration does not reflect bioavailability;
- Chemical analysis does not take into account the interactions between contaminants;
- No legislated threshold values for many chemicals and when existing many are generic values;
- Ecotoxicological /ecological data may help to eliminate false-positive and false negative risk values

So, no true RA without measuring effects



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Chain of Ecotoxicological Evidence

TOXICOLOGICAL
(Response of Individuals)

Laboratory tests

Altered Physiology

Death

Reduced Growth

Reproductive Effects

SHORT-TERM

LONG-TERM

Symbiotic Dissociation

Population Decline

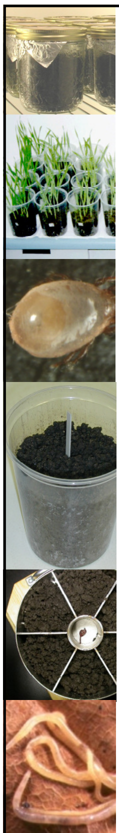
Community Disruption

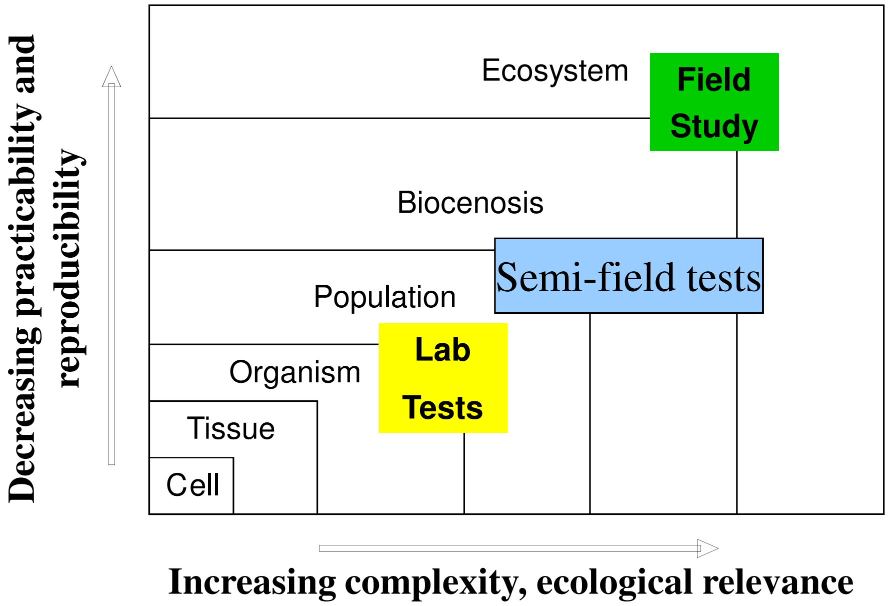
Semi-field & Field tests

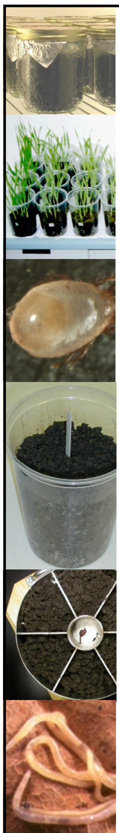
ECOLOGICAL
(Community & Population Responses)

Adapted from Gunderson et al. 1997

Tests at different level of Biological Organization







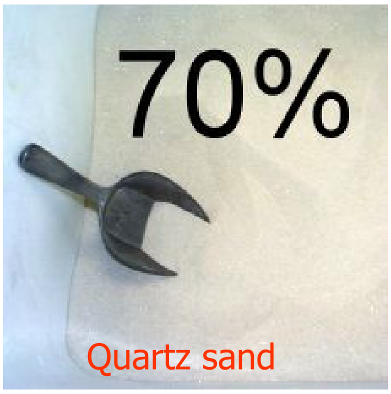
What do we assess when testing in soil ?

Soil habitat function (ISO 15799)
ability of soils/soil materials to serve as a habitat for micro-organisms, plants, soil-living animals and their interactions (biocenosis)
Bioassays made with the soil matrix

Soil retention function (ISO 15799)
ability of soils/soil materials to adsorb pollutants in such that they cannot be mobilized via the water pathway and translocated into the food chain or groundwater
Bioassays made with soil eluates

OECD Artificial Soil

In 1984, OECD defined an artificial mixture for earthworm tests.



Quartz sand

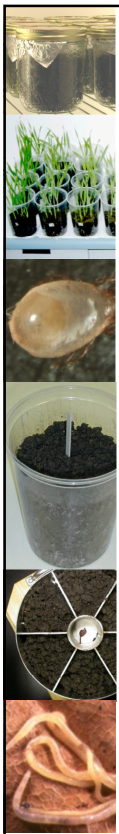
Kaolin clay

Sphagnum peat,
finely ground

$CaCO_3$ is added to adjust the pH to 6.0 ± 0.5

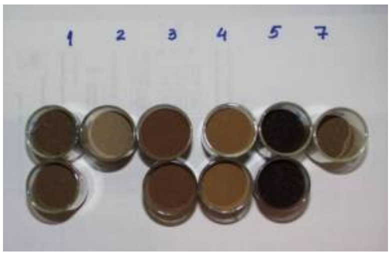
In the tropics peat is replaced by coconut shell fibre (SAT)

Soil invertebrates: Increasing Ecol. Relevance

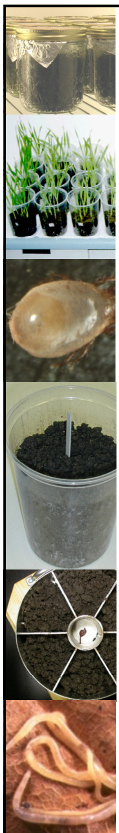


To gain ECOLOGICAL RELEVANCE the transition should be made …

OCDE artificial soil



Natural soils



ALL STARS: Earthworm tests I

Earthworm Reproduction Test (*Eisenia andrei; E. fetida*)

ISO 11268-2:1998

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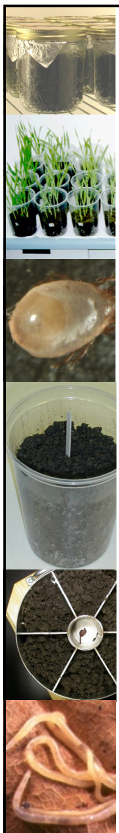
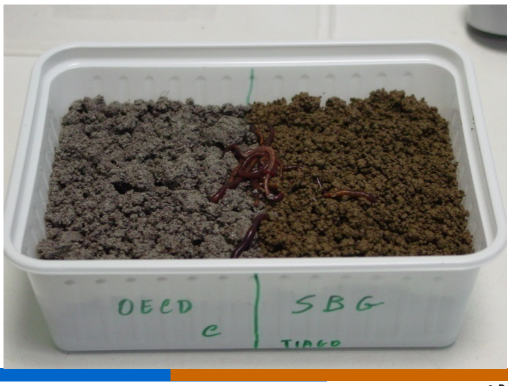


ALL STARS: Earthworm tests II

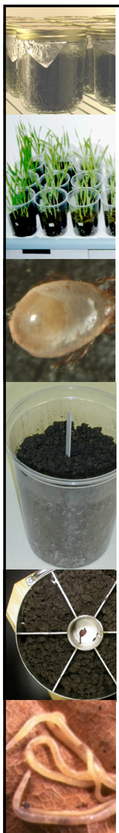
Earthworm Avoidance Toxicity Test (*Eisenia andrei; E. fetida*)

ISO 17512-1:2005

Start: 0 h



End: 48 h



ALL STARS: Enchytraeid test

Enchytraeid Reproduction Toxicity Test (*Enchytraeus albidus*)
ISO 16387:2004

- ***More ecologically relevant than earthworms***
- ***Good substitute test when earthworm test is not possible (difficult soils)***
- ***Other species (e.g., E. crypticus) can be selected (less sensitive to soil properties)***

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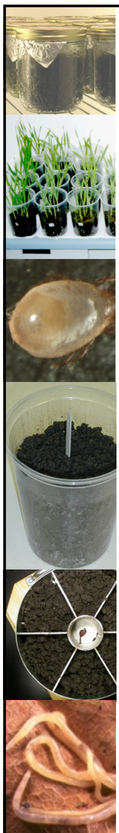
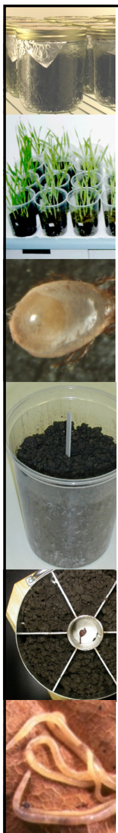


ALL STARS: Collembola tests I

Collembola Reproduction Toxicity Test (*Folsomia candida*)

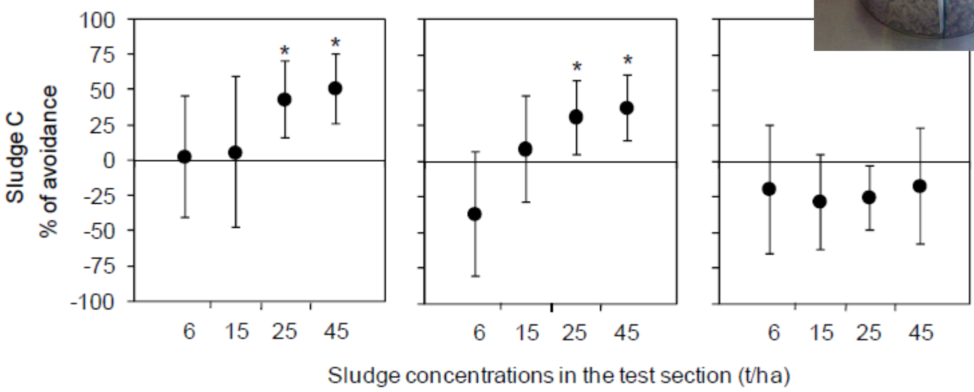
ISO 11267:1998

- ***Other species (e.g., F. fimetaria; Onychiurus folsomi) can be selected (sexual reproduction; increase ecological relevance)***



ALL STARS: Collembola tests II

Collembola Avoidance Toxicity Test
(*Folsomia candida*)
ISO 17512-2 (2011)



Natal da Luz et al., Chemosphere, 2009

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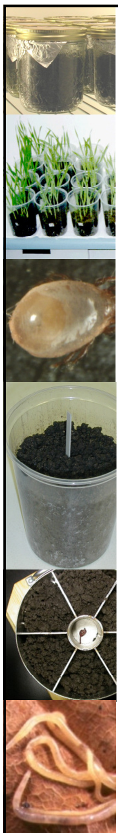


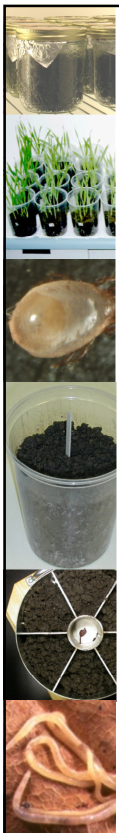
Other Soil invertebrate Tests

Predatory mite ***H. aculeifer*** **reproduction test**
OECD 226: 2008

Nematodes

Caenorhabditis elegans **survival test**
ASTM E 2172-01: 2001

Caenorhabditis elegans **reproduction test**
ISO 10872: 2010



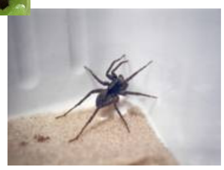
Other Soil invertebrate Tests

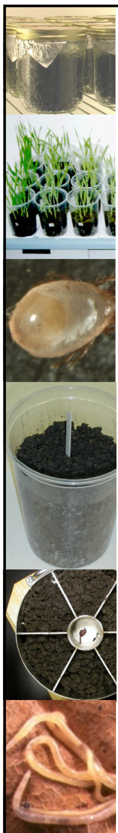
Snail Growth Toxicity Test (*Helix aspersa*)
ISO 15952: 2003

Carabid beetle Test (*Poecilus cupreus*)
BBA VI, 23-2.1.8: 1991

Staphilinid beetle Test (*Aleochara bilineata*)
BBA VI, 23-2.1.10: 1994

Spider Acute Test (*Pardosa* sp.)
BBA VI, 23-2.1.9: 1994





Other Soil invertebrate Tests

Isopod Growth Test (*Porcellio scaber*)
Løkke & Van Gestel 1998

Isopod Reproduction Test (*Porcellio scaber*)
Løkke & Van Gestel 1998

Isopod Feeding Test (*Porcellio scaber*)
Drobne 1997

Centipede Test (*Lithobius mutabilis*)
Mortality & growth
Løkke & Van Gestel 1998

Diplopod Test (*Brachydesmus superus*)
Mortality & reproduction
Løkke & Van Gestel 1998

Invertebrates: Invertebrate tests in the tropics

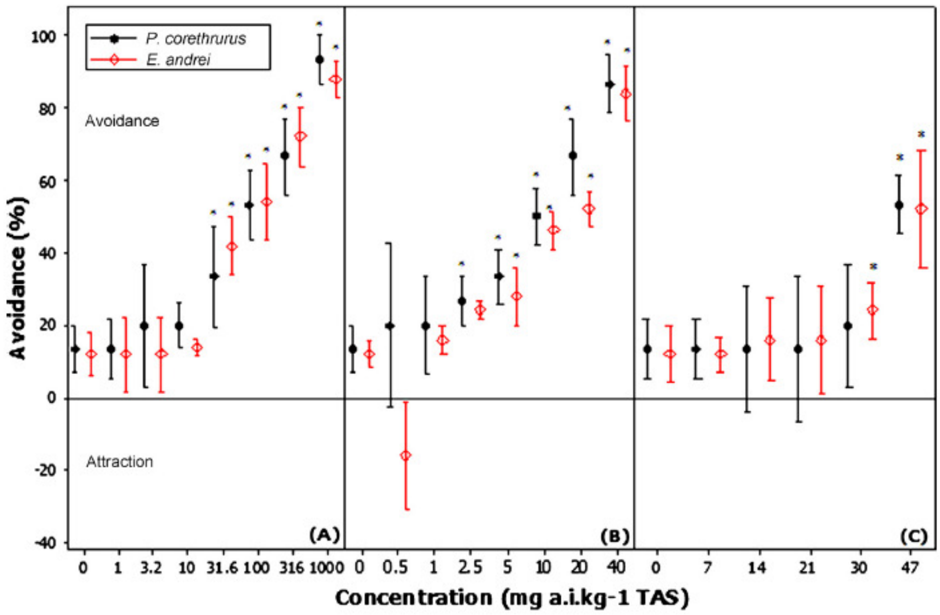
-Tropical strains of temperate
earthworms: *E. fetida, E. andrei*

-Endemic earthworm species
Perionyx excavatus, *Pontoscolex corethrurus, Eudrilus eugeniae*)

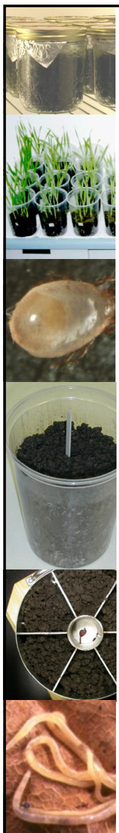
-Tropical strains of Mediterranean
isopods: *Porcellionides pruinosus*

-Endemic millipeds (*Trigoniulus corallinus*)
or isopods (*Circoniscus ornatus, Cubaris murina*)

Invertebrates: Invertebrate tests in the tropics



Buch et al., Applied Soil Ecology, 2013



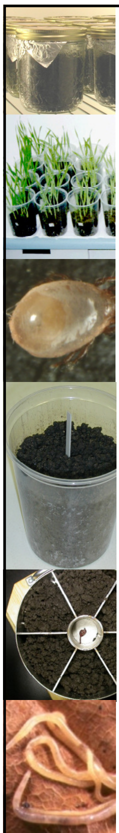
Soil invertebrates: Test battery

Pesticide	Organism	EC50 (95% IC) (mg ai/kg dw soil)	EC50 (mol ai/kg dw soil)
azoxystrobin (403.4 g/mol)	***F. candida***	**187 (130 – 243)**	**4.64E-04**
	E. crypticus	177 (120 – 135)	4.39E-04
	E. andrei	**42.0 (23.2 – 60.8)**	**1.04E-04**
chlorothalonil (265.9 g/mol)	***F. candida***	**31.7 (24.6 – 38.8)**	**1.19E-04**
	E. crypticus	**141 (111 – 170)**	**5.30E-04**
	E. andrei	40.9 (30.1 – 51.7)	1.54E-04
ethoprophos (242.3 g/mol)	***F. candida***	**0.027 (0.024 – 0.031)**	**1.11E-07**
	E. crypticus	**64.8 (34.9 – 94.8)**	**2.67E-04**
	E. andrei	8.3 (3.6 – 13.0)	3.43E-05

Different species present different sensitivities

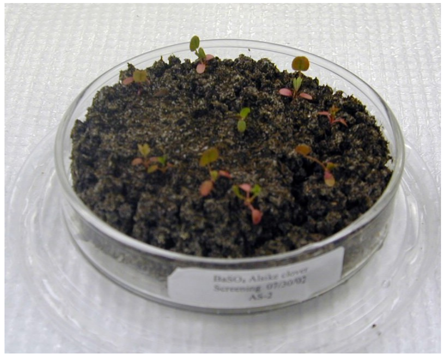
To decrease uncertainty due to different sensitivities, testing on several test species is needed!

Leitão et al. (submited)



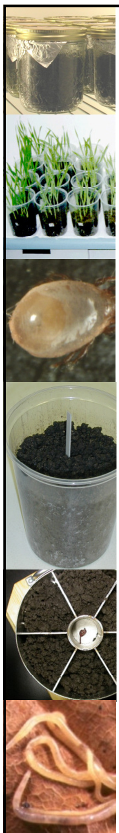
ALL STARS: Plant tests

Plant seed germination and/or growth
Monocots and Dycots
ISO 11269-2:2005



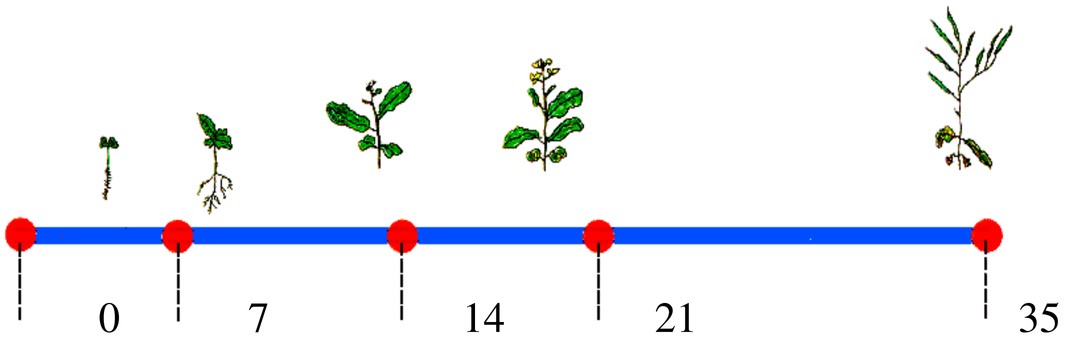
Small soil volumes
Short-term tests (~ 7 d)
Emergence

Larger soil volumes
Longer-term tests (14-21 d)
Growth (root and shoot length and dry mass)

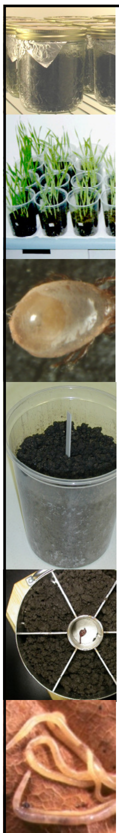


Plant tests: Full life cycle test

Chronic plant test: ISO 22030: 2005



sowing (10 seeds per vessel)	thinning (8 seedlings)	harvesting (4 plants)	pollination	harvesting (4 plants)



ALL STARS: Microbial tests

Different tests have different degrees of sensitivity.
A battery of assays, integrating different ecosystem parameters and different indicators, should be used!

They evaluate effects on the Natural microbial community

This battery should integrate measures of:

- Microbial diversity
- Microbial biomass
- Microbial activity (incl. nutrient cycling)

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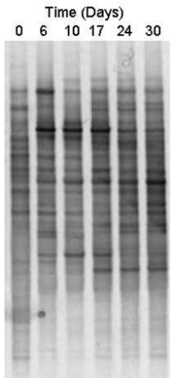
Microbial tests: Microbial diversity



Genetic diversity

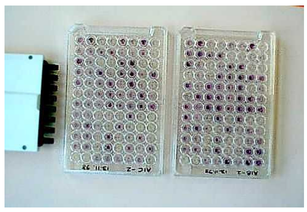
TRFLP, PCR-DGGE (changes in dominant bacterial, fungal, Archea communities)

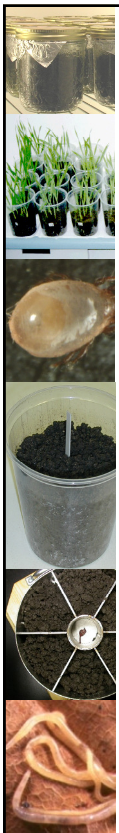
Functional diversity

BIOLOG assay; MicroResp assay(metabolic fingerprinting; based on rate of substrate use; able to discriminate stressed communities)

Structural diversity

PLFA's (combines bacterial and fungal diversity; marker lipid profiles able to measure changes in dominant microbial communities)





Microbial tests: Microbial biomass

Indirect methods are used more often

Chloroform Fumigation-Extraction (ISO 14240-2: 1997)

Substrate Induced Respiration (SIR) (ISO 14240-1: 1997)

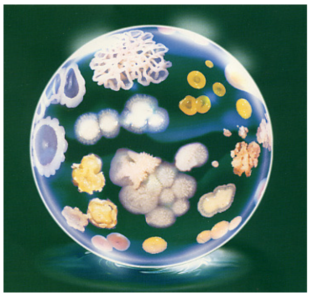
IRGA: Measurement of soil respiration

Microbial tests: Microbial activity

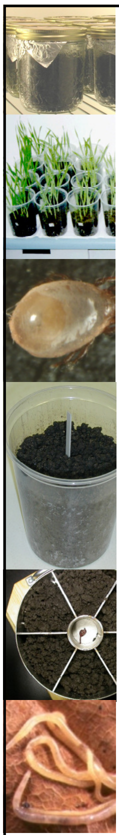
Soil respiration
Respirometry (CO_2 production);
Respiratory quotient (qCO_2) (soil resp. / Cmic);

Soil enzymes (e.g.)
Dehydrogenase (general microbial activity);
Beta-glucosidase, Cellulase, Phenol-oxidase (C cycling);
Urease, Amidase (N cycling);
Phosphatase (P cycling);
Arylsulphatase (S cycling)

Nitrogen transformations
N-mineralization;
Nitrification / Denitrification;
N-fixation



Molecular methods
Metagenomics
Transcriptomics



ALL STARS: Functional tests

Bait-lamina tests (activity of the soil fauna *in situ*)
ISO 18311 (draft 2011)

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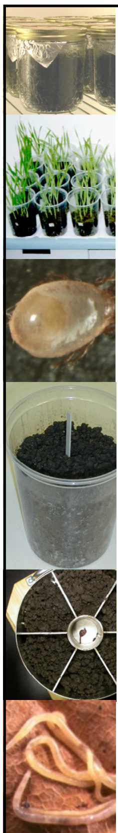
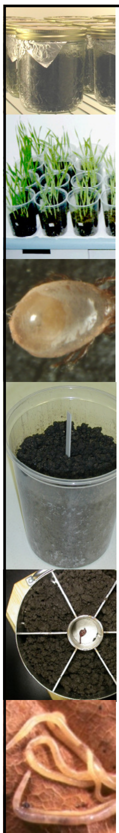


ALL STARS: Functional tests

Litter bag test OECD guidance 56: 2006



ALL STARS: Semi-field methods

A Model ecosystem segments ("microcosms"):

Artificially assembled units with added organisms

A1 Controlled conditions

A2 Field conditions

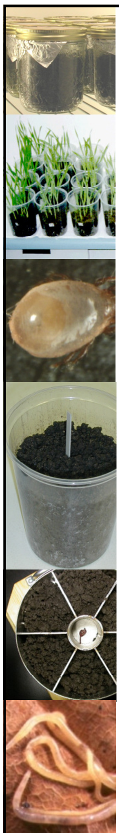
B Ecosystem segments from the field ("mesocosms"):

Intact soil cores with natural communities

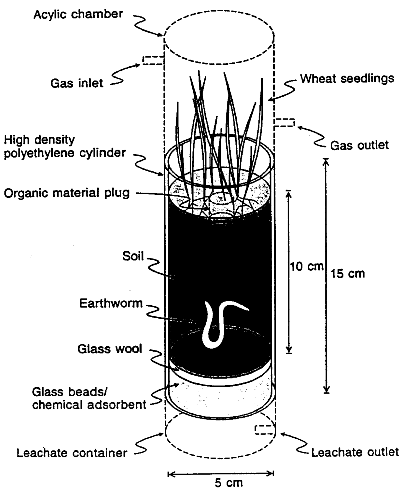
B1 Controlled conditions

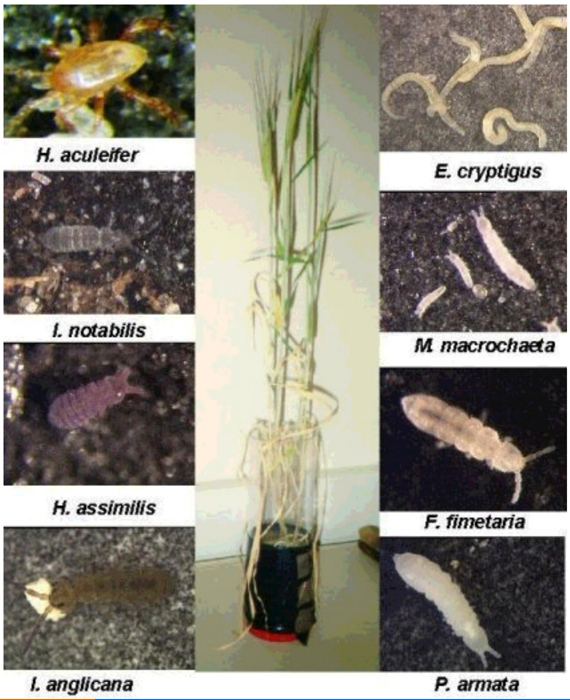
B2 Field conditions

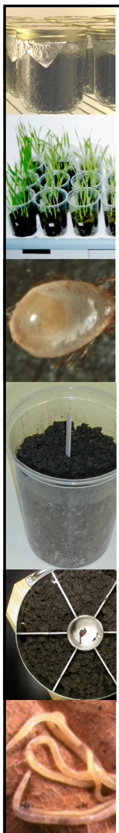
ALL STARS: Semi-field methods



Microcosms



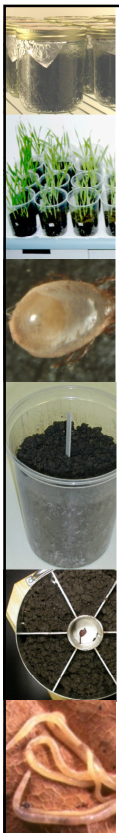




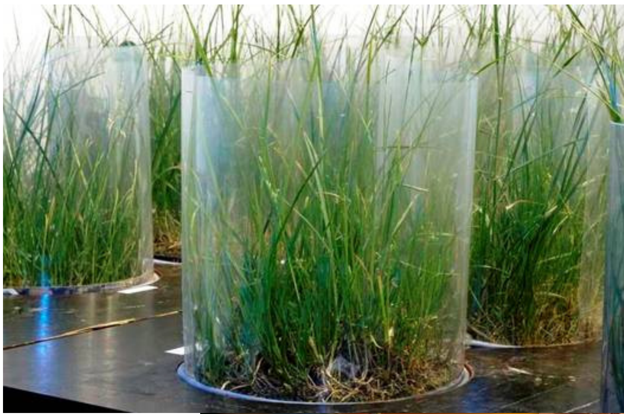
ALL STARS: Semi-field methods

Mesocosms: Terrestrial Model Ecosystems

TME extraction in the field



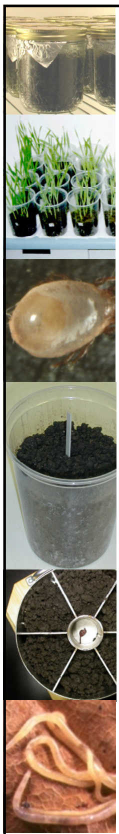
ALL STARS: Semi-field methods



Transparent top sleeve

Leachate bottles

Movable TME carts



ALL STARS: Semi-field methods

Potential Endpoints

Ecosystem *structure*

Chemical residues
- soil, leachate, plant

Nutrients
- soil, leachate

Soil organisms
- nematodes
- arthropods
- enchytraeids
- earthworms
- microorganisms

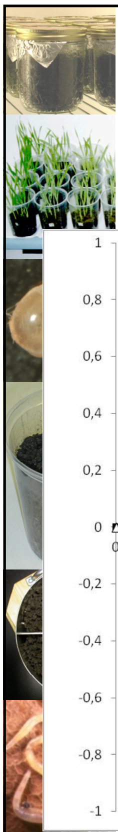
Plants
- biomass

Ecosystem *function*

OM decomposition

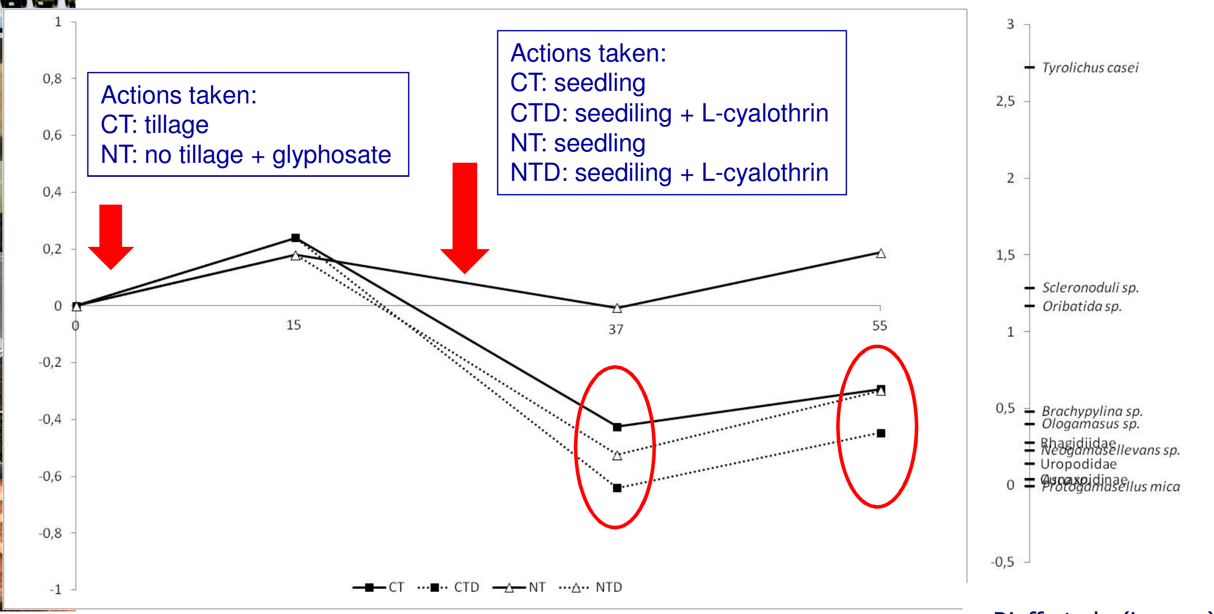
Feeding activity (bait lamina)

Microbial activity (e.g.)
- enzymes
- respiration
- diversity
- molecular methods



Semi Field Methods: TME Case study

Mites on Conventional vs. Now tillage croping system with maize



Rieff et al., (in prep)

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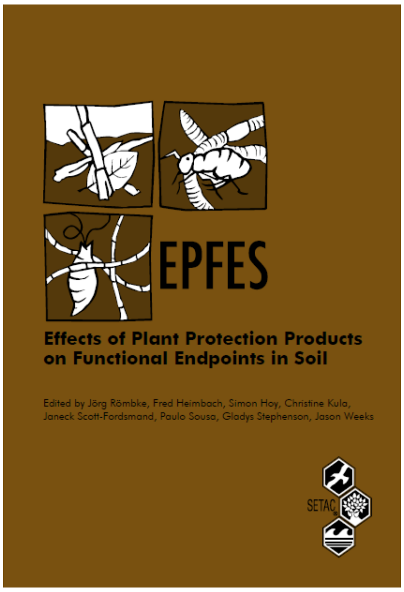
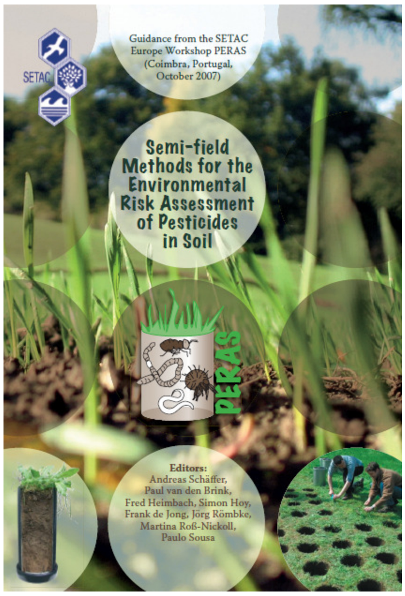
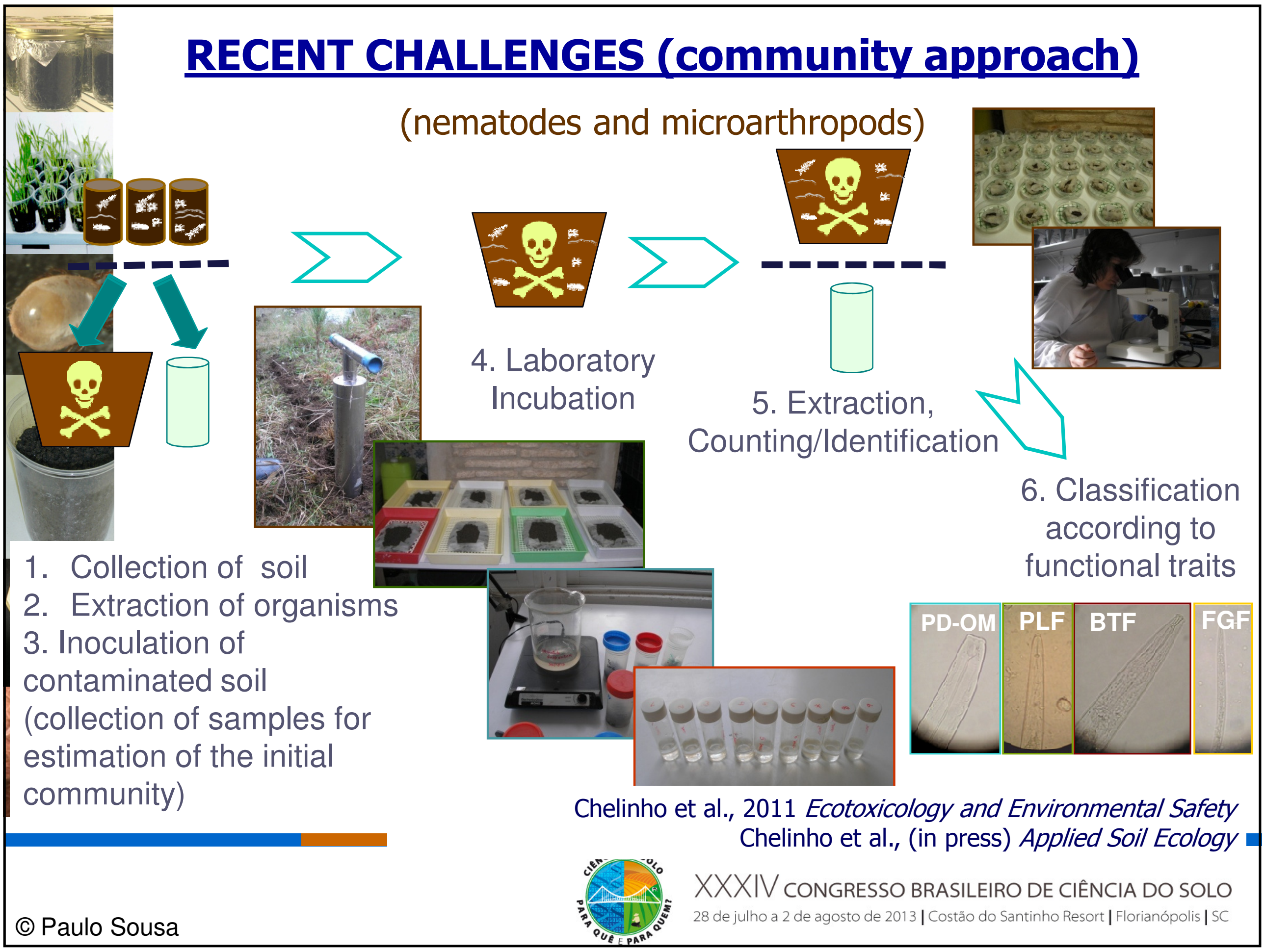
To know more about higher tier tests






RECENT CHALLENGES (community approach)
(nematodes and microarthropods)
1. Collection of soil
2. Extraction of organisms
3. Inoculation of contaminated soil (collection of samples for estimation of the initial community)
4. Laboratory Incubation
5. Extraction, Counting/Identification
6. Classification according to functional traits
PD-OM
PLF
BTF
FGF
Chelinho et al., 2011 Ecotoxicology and Environmental Safety
Chelinho et al., (in press) Applied Soil Ecology
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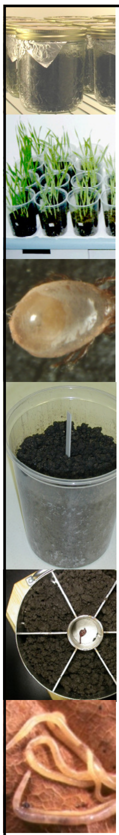
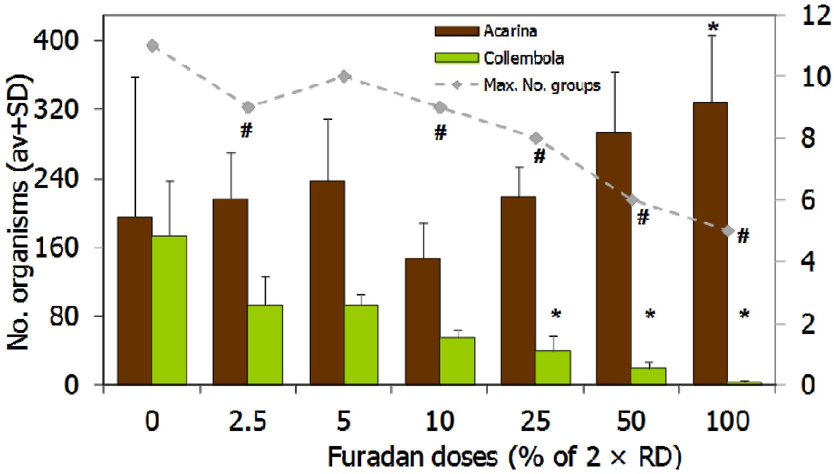
Some data for microarthropods in tests in Brazil





Chelinho et al. (in press) *Applied Soil Ecology*

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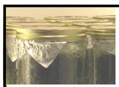
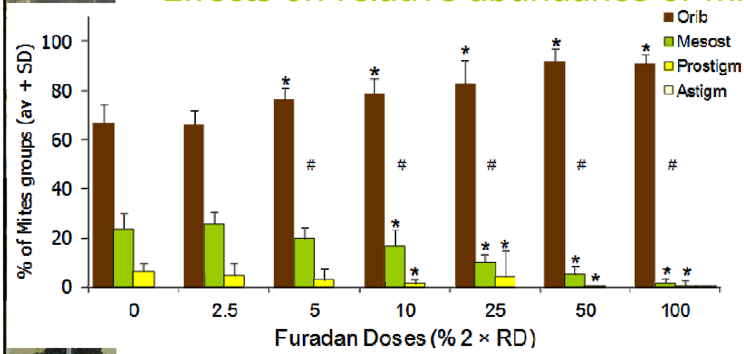


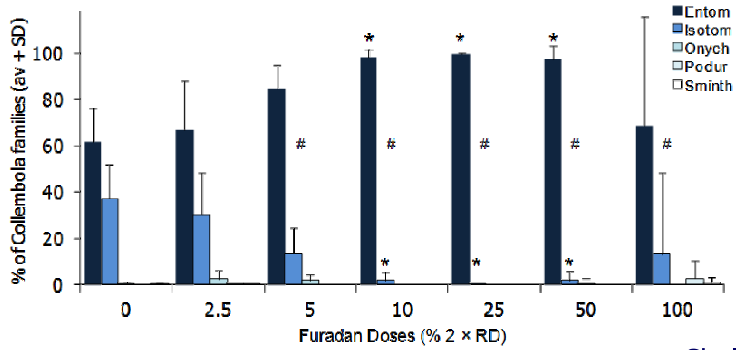
Effects on relative abundance of mite and collembolan groups (BR)



↑ Oribatida ↓ Mesostigmata

- ➢ Decrease in the number of competitors for organic matter and potential preys

↓ Prostigmata



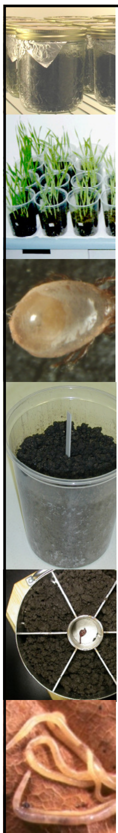
↑ Entomobryidae ↓ Isotomidae

- Collembola and mite communities negatively affected at all doses except the lowest

Chelinho et al. (in press) *Applied Soil Ecology*

Outline of presentation

- *Importance of Ecotoxicology in Ecological Risk Assessment (ERA)*
- *Major tools (tests) used in Ecotoxicology*
- ***Case study from Brazil***
- *Evolution of Soil Ecotoxicology in Brazil and perspectives*



ECOLOGICAL RISK ASSESSMENT OF CONTAMINATED SITES

Case study – Santo Amaro (Brazil)

Júlia Niemeyer & J. Paulo Sousa
IMAR – CMA
Laboratory of Soil Ecology and Ecotoxicology
Universidade de Coimbra, Portugal
jps@zoo.uc.pt
http://www1.ci.uc.pt/imar/unit/people/cvs/cv/j_p_sousa.php
http://www.facebook.com/labsolos

Soil Ecology & Ecotoxicology Lab

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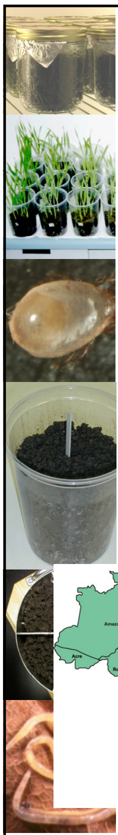
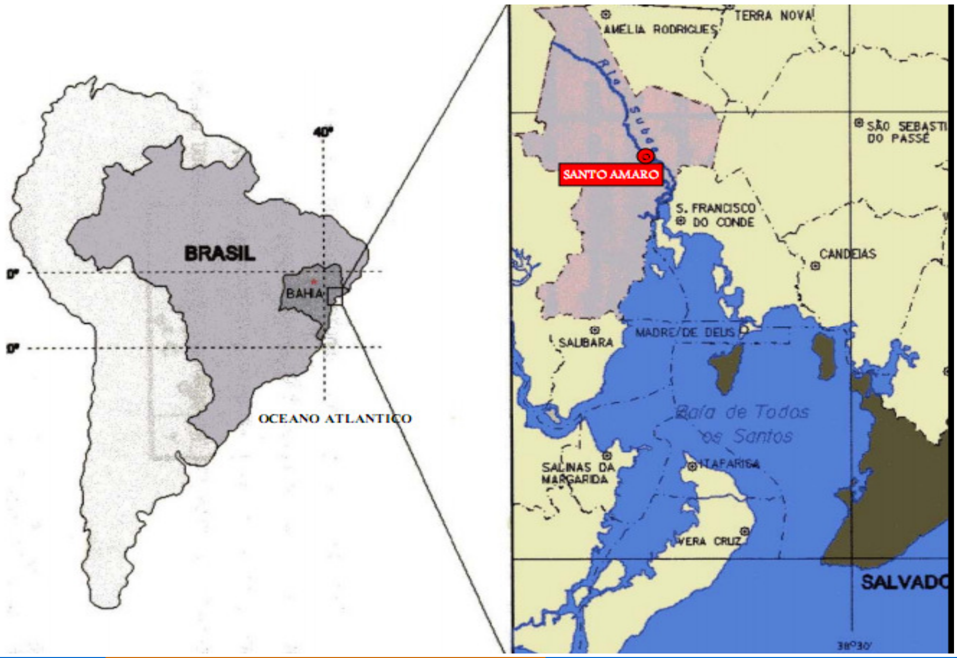
Old Plumbum smelter
Santo Amaro, Bahia, Brasil



Considered by the Brazilian Government a priority area for environmental monitoring

Old Plumbum smelter
Santo Amaro, Bahia, Brasil



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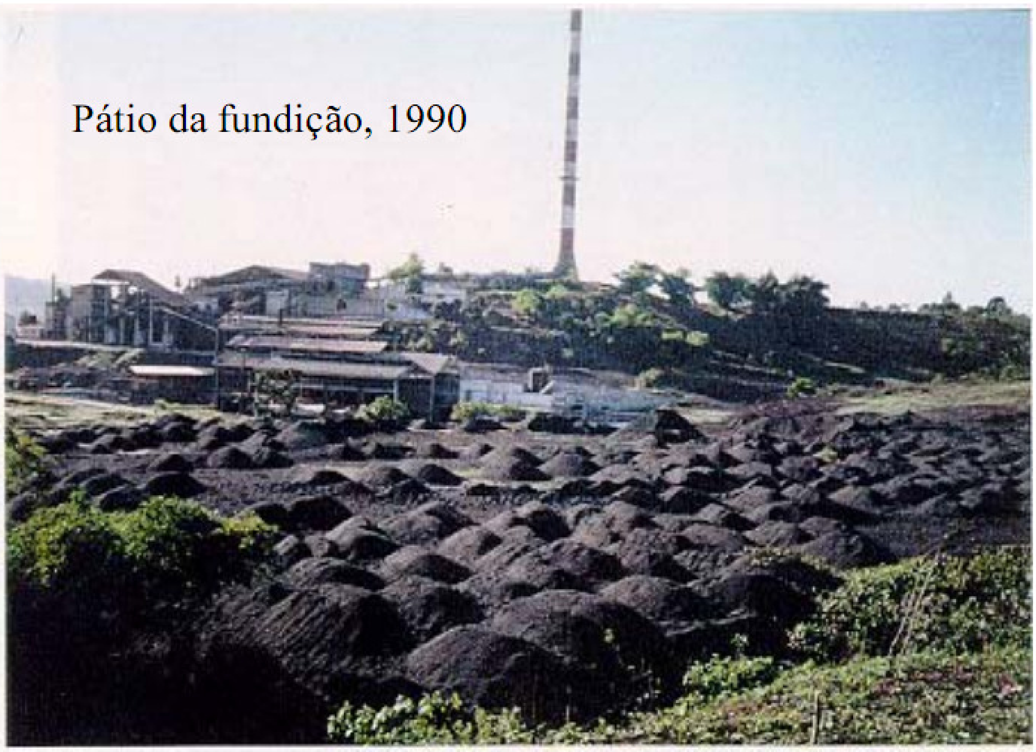
HISTORY of the SITE

- 1960 – Start of smelting activities
- 1980 – Environmental agency from Bahia demands measures to control pollution and decrease productivity
- 1991 – New pollution control measures imposed by the agency
- 1993 – Closure of the smelter



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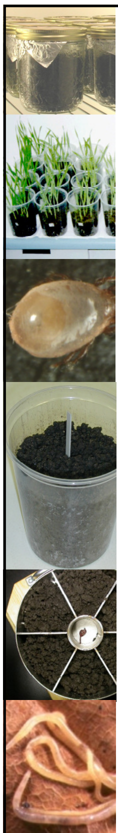
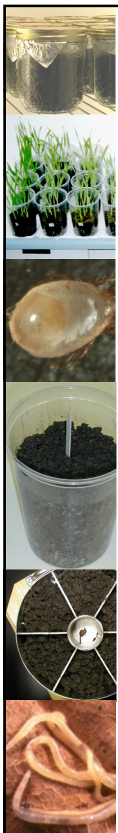
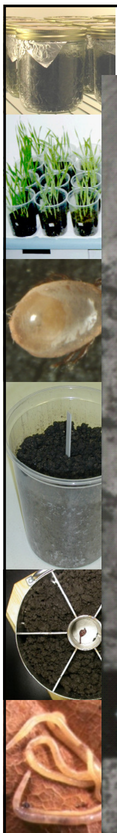



Depósitos de escória na área da fundição, 1998


Depósitos de escória
"encapsulada", 2002





Road – East boundary of the area
Subaé river
Smelter facilities
Lagoon connecting the Subaé river
Lagoon
Urban area

Conceptual Model

• The site is limited by the urban area of Sto Amaro (South) and by agricultural and pasture areas at North

• Relevant contaminants **Pb, Cd, Cu, Zn**

• Actions planned for the smelter area:

(1) In places with a high risk the residues will be removed and encapsulated in a single site

(2) In places with low risk, the residues will stay in place

(3) Major goal is to re-naturalize the area

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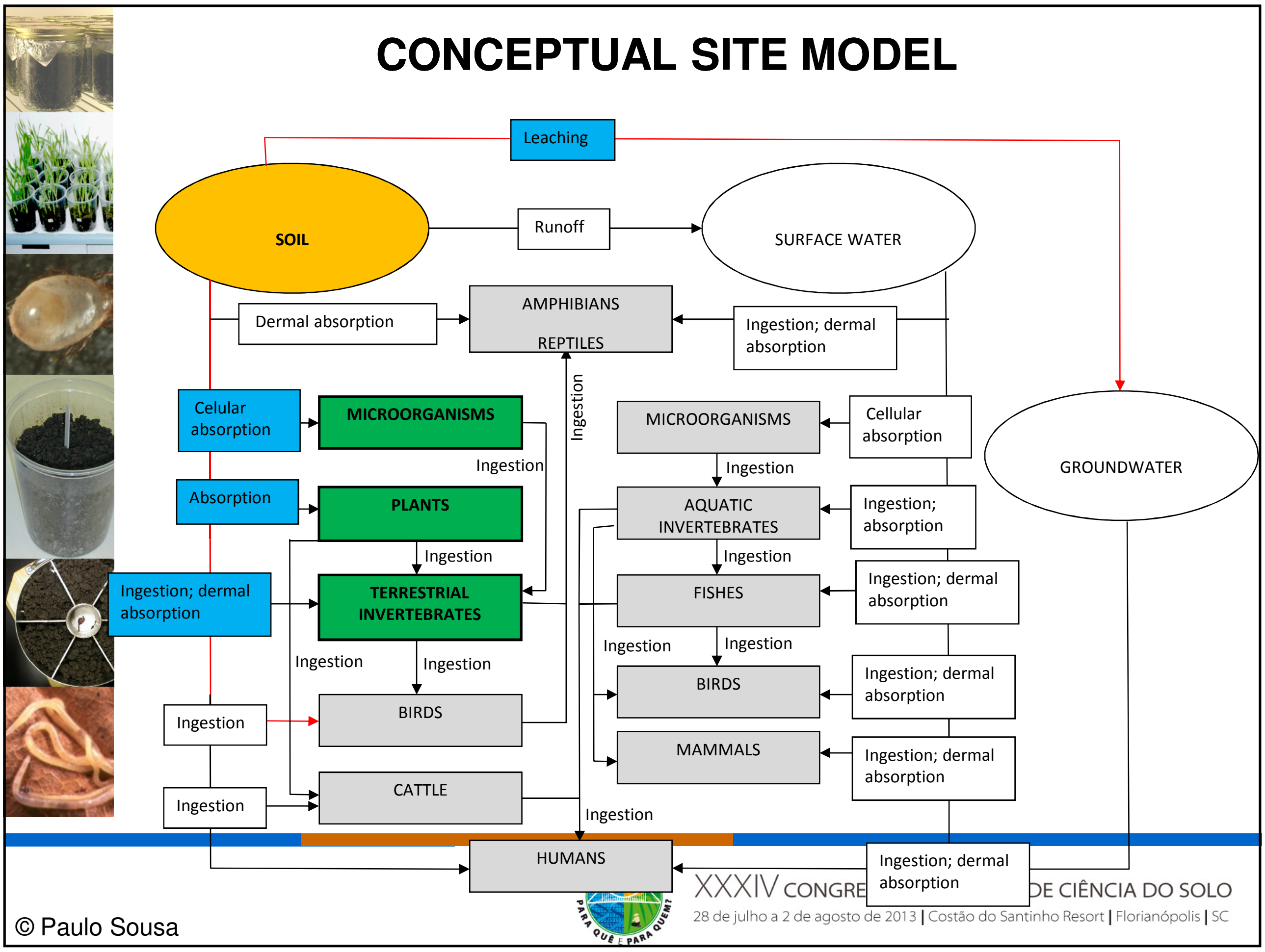
CONCEPTUAL SITE MODEL
Leaching
SOIL
Runoff
SURFACE WATER
Dermal absorption
AMPHIBIANS
REPTILES
Ingestion; dermal absorption
Ingestion
Celular absorption
MICROORGANISMS
Ingestion
MICROORGANISMS
Cellular absorption
GROUNDWATER
Ingestion
Absorption
PLANTS
AQUATIC INVERTEBRATES
Ingestion; absorption
Ingestion
Ingestion
Ingestion; dermal absorption
TERRESTRIAL INVERTEBRATES
FISHES
Ingestion; dermal absorption
Ingestion
Ingestion
Ingestion
Ingestion
Ingestion
BIRDS
BIRDS
Ingestion; dermal absorption
MAMMALS
Ingestion; dermal absorption
CATTLE
Ingestion
Ingestion
HUMANS
Ingestion; dermal absorption
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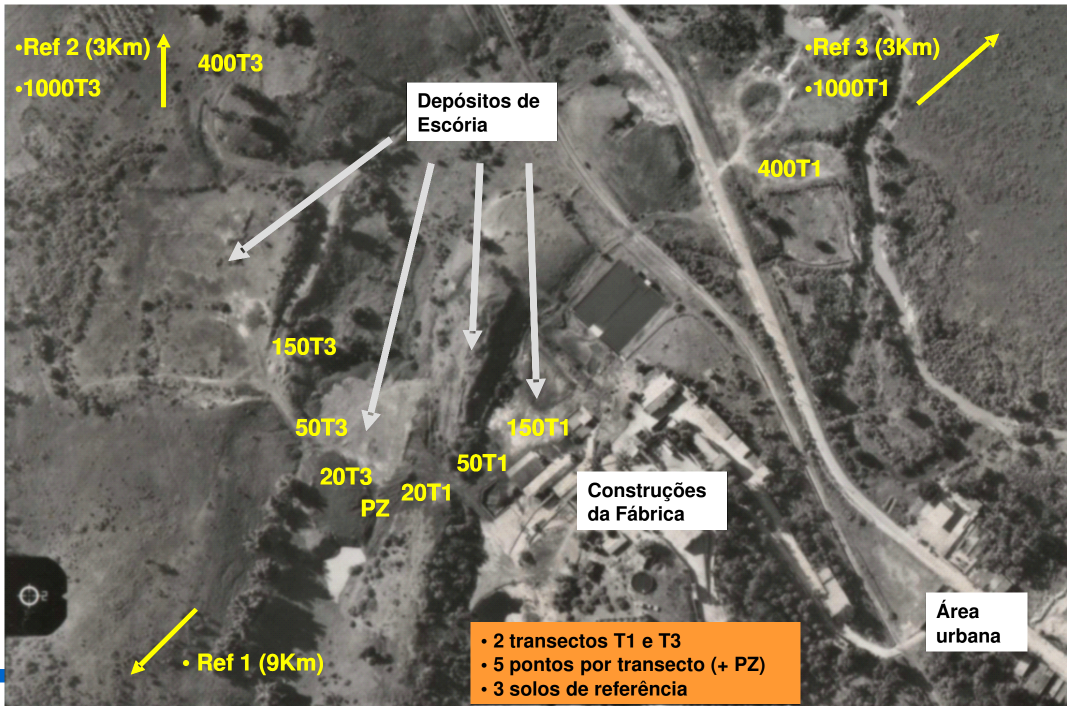
How to plan the sampling strategy for the area?



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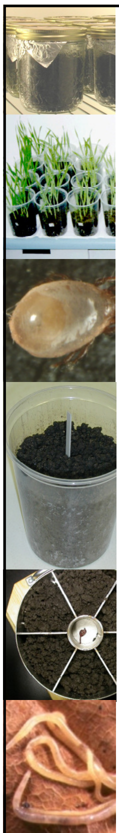
Soil properties

Soil group	Coarse sand (%)	Fine sand (%)	Sand (total) (%)	Silt (%)	Clay (%)	Texture (USDA)	pH (KCl 1:5 v:v)	Organic matter (%)	CEC (meq 100g)	WHC (%)
Group 1										
Ref. 1	2,3	8,6	10,9	42,1	47,0	Silty Clay	7,1	1,1	34,16	53,78
P1000T1	2,5	21,8	24,3	19,9	55,8	Clay	3,7	2,0	43,20	59,95
P20T3	11,4	30,0	41,4	22,3	36,3	Clay Loam	6,8	1,9	42,16	67,73
P400T3	6,5	8,6	15,1	52,4	32,5	Silt Clay Loam	7,1	1,9	35,84	56,67
Group 2										
Ref. 2	50,9	38,5	89,4	2,8	7,7	Loamy Sand	4,9	1,0	37,60	27,53
P0	43,2	31,3	74,5	11,9	13,6	Sandy Loam	6,7	0,3	38,56	44,12
P20T1	48,0	13,8	61,8	19,0	19,3	Sandy Loam	7,1	0,2	37,28	46,4
P150T1	56,2	21,1	77,4	12,3	10,3	Sandy Loam	6,7	2,1	21,28	28,55
P50T3	69,2	9,1	78,3	10,4	11,3	Sandy Loam	7,2	2,8	16,56	22,05
Group 3										
Ref. 3	22,2	15,0	37,2	11,1	51,7	Clay	6,1	3,9	36,48	60,75
P50T1	25,2	13,4	38,6	29,0	32,4	Clay Loam	6,7	1,1	38,16	54,51
P400T1	19,6	23,9	43,5	20,2	36,3	Clay Loam	6,8	5,1	37,44	58,93
P150T3	8,4	15,2	23,5	21,4	55,1	Clay	6,8	2,5	49,20	61,76
P1000T3	10,3	19,5	29,8	29,8	40,4	Clay Loam	7,0	5,7	42,72	57,57

Niemeyer et al. JSS, 2010

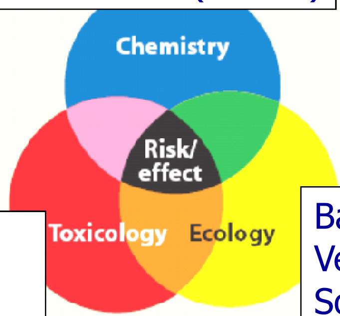


SITE SPECIFIC RISK ASSESSMENT

Santo Amaro da Purificação (Bahia, Brazil)

Screening phase (Tier 1)

Toxic Pressure (msPAF)



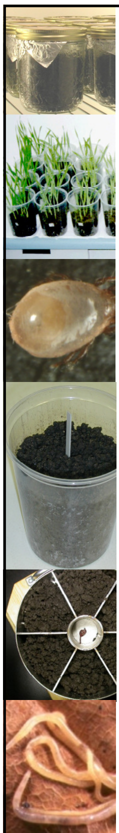
Microtox assays
Daphnia assays
Avoidance *Eisenia*
Avoidance *Folsomia*

Bait lamina
Vegetation cover
Soil respiration

ChLoE – Total metal Concentrations

Soil group	Pb	Cd	Cu	Zn	Cr	Ni	Fe	Mn
Group 1								
Ref. 1	16	<0,2	66	94	77	54	45000	840
P1000T1	23	<0,2	60	80	62	46	48000	360
P20T3	308	<0,2	56	420	78	60	49000	672
P400T3	179	0,3	44	90	59	46	34000	760
Group 2								
Ref. 2	13	<0,2	18	24	16	28	2900	34
P0	1264	<0,2	76	3800 $^{(2.8)}$	72	57	52000	674
P20T1	133	<0,2	56	220	80	56	41000	780
P150T1	37460 $^{(10.4)}$	771 $^{(9.8)}$	594 $^{(1.6)}$	42200 $^{(33.5)}$	57	70	110000	1720
P50T3	26074 $^{(7.1)}$	62	3196 $^{(8.2)}$	95940 $^{(73.5)}$	80	40	117000	5880
Group 3								
Ref. 3	152	<0,2	40	260	59	40	53000	820
P50T1	164	<0,2	60	240	80	58	43000	720
P400T1	961	8,8	60	840	64	48	35000	540
P150T3	2200	12	108	3300	84	58	56000	678
P1000T3	99	<0,2	56	156	84	52	49000	568

Numbers in superscript indicate those soils whose metal concentration exceeded the corrected Dutch HC50 (based on EC50 values) benchmarks according to Rutgers et al. (2008) (Ex: the [Pb] at P150T1: 37460 (10.4), indicates that [Pb] was 10.4 times higher than the HC50corPb).

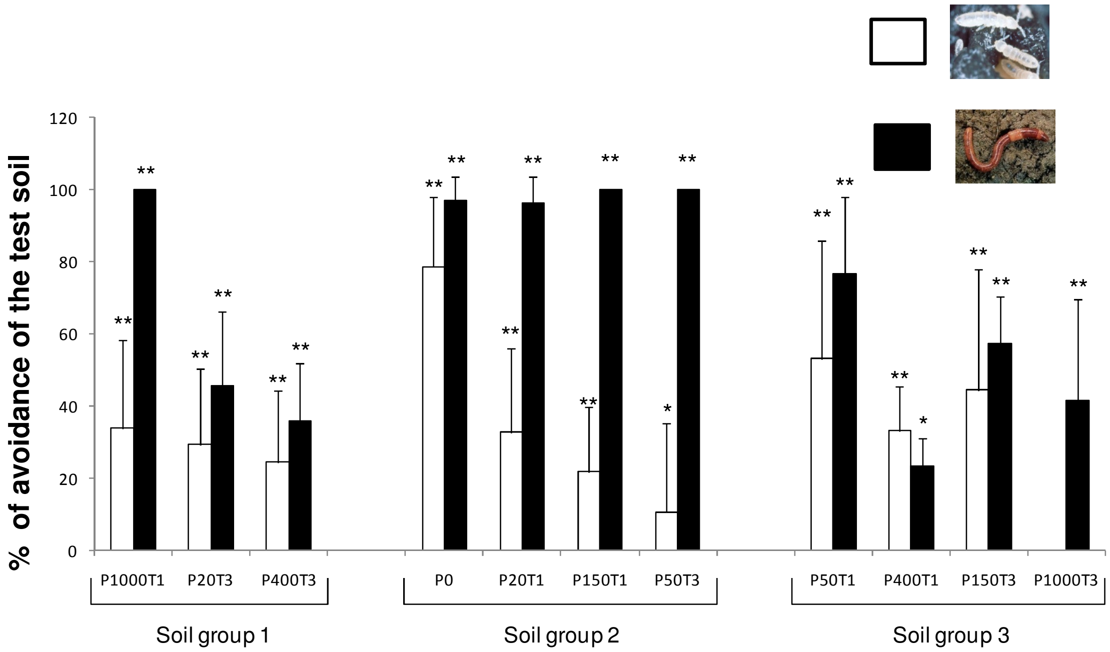


ChLoE – Risk values (Tier 1)

Soil group	Pb	Cd	Cu	Zn	Cr	Ni	Combined ChLoE
	Chemical Line of Evidence (ChLoE)						
Group 1							
P1000T1	0,00	0,00	0,00	0,00	0,00	0,00	**0,00**
P20T3	0,08	0,00	0,00	0,13	0,01	0,02	**0,23**
P400T3	0,05	0,00	0,00	0,01	0,00	0,01	**0,05**
Group 2							
P0	0,29	0,00	0,12	0,73	0,08	0,05	**0,85**
P20T1	0,00	0,00	0,07	0,11	0,08	0,02	**0,25**
P150T1	0,88	0,84	0,60	0,98	0,07	0,10	**1,00**
P50T3	0,83	0,45	0,90	1,00	0,10	0,02	**1,00**
Group 3							
P50T1	0,05	0,00	0,05	0,02	0,04	0,04	**0,19**
P400T1	0,20	0,13	0,04	0,20	0,02	0,02	**0,49**
P150T3	0,30	0,15	0,08	0,47	0,02	0,02	**0,72**
P1000T3	0,00	0,00	0,03	0,00	0,03	0,02	**0,06**

Niemeyer et al. JSS, 2010

EcLoE – Avoidance tests



Niemeyer et al. JSS, 2010

EcLoE – Tests with aquatic organisms

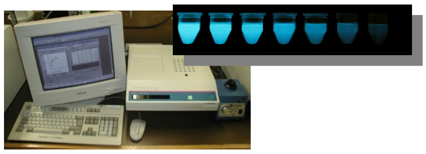
Shaked for 18 h
Centrifuged at 3000 rpm
Stored at 4 ºC

Eluate
1:4 (soil:water)

Daphnia acute tests (ISO 6341)

Toxicity only at 150T1

EC_{50} = 68%



Vibrio fischeri

Toxicity only at m 50T3

EC_{50} = 8.6%

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EcLoE – Risk values (Tier 1)

Soil group	Avoidance *Eisenia*	Avoidance *Folsomia*	*Daphnia* acute	Microtox	Combined EcLoE
	Ecotoxicological Line of Evidence (EcLoE)				
Group 1					
P1000T1	1,00	0.33	0.02	0,00	**0.72**
P20T3	0.46	0.3	0,00	0,00	**0.21**
P400T3	0.36	0.25	0,00	0,00	**0.17**
Group 2					
P0	0.97	0.79	0,00	0,00	**0.72**
P20T1	0.96	0.33	0,00	0,00	**0.6**
P150T1	1,00	0.22	0.32	0,00	**0.73**
P50T3	1,00	0.11	0,00	0.89	**0.82**
Group 3					
P50T1	0.76	0.54	0,00	0,00	**0.43**
P400T1	0.23	0.32	0,00	0,00	**0.15**
P150T3	0.57	0.45	0,00	0,00	**0.30**
P1000T3	0.42	0,00	0,00	0,00	**0.13**

Niemeyer et al. JSS, 2010

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Ecological LoE (ELoE)

Soil group	Bait lamina (% pierced holes)	Vegetation cover (%)	Soil respiration (ug CO_2 /g soil / day)
Overall Ref	48.6 ± 13.9	81.3 ± 21.0	139.4 ± 106.4
Group 1			
P1000T1	45.3 ± 16.1	67.5 ± 15.0	164.0 ± 79.1
P20T3	30.4 ± 15.4	32.5 ± 12.6 ***	82.6 ± 15.8
P400T3	10.3 ± 6.7 ***	97.5 ± 5.0	165.2 ± 41.3
Group 2			
P0	18.4 ± 14.3 ***	22.5 ± 22.2 ***	34.9 ± 7.8 ***
P20T1	17.8 ± 10.2 ***	30.0 ± 16.3 ***	35.1 ± 7.1 ***
P150T1	7.3 ± 8.1 ***	30.0 ± 42.4 ***	49.2 ± 6.6 **
P50T3	11.8 ± 5.7 ***	20.0 ± 14.1 ***	52.2 ± 12.6 **
Group 3			
P50T1	19.8 ± 6.8 ***	57.5 ± 12.6	41.4 ± 2.4 **
P400T1	61.5 ± 23.8	100.0 ± 0.0	234.9 ± 83.3
P150T3	5.5 ± 6.9 ***	57.5 ± 9.6	60.5 ± 9.2
P1000T3	26.3 ± 17.5 *	100.0 ± 0.0	n.d.

n.d. - not determined

Niemeyer et al. JSS, 2010

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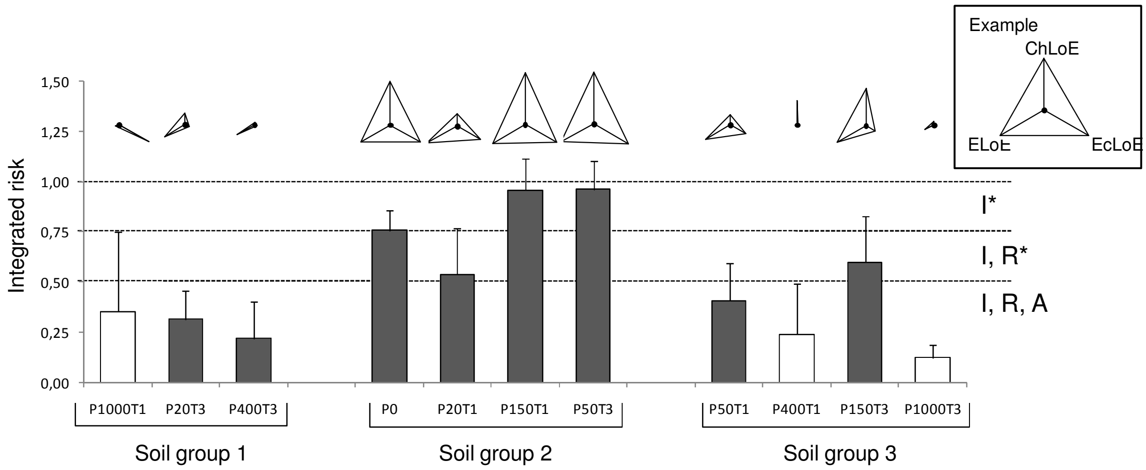
ELoE – Risk values (Tier 1)

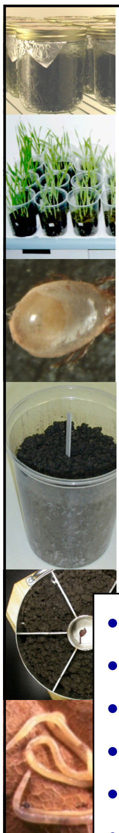
Soil group	Ecological Line of Evidence (ELoE)			
	Bait lamina	Vegetation cover	Soil respiration	Combined ELoE
Group 1				
P1000T1	0,00	0,17	0,00	**0,06**
P20T3	0,37	0,60	0,41	**0,47**
P400T3	0,79	0,00	0,00	**0,40**
Group 2				
P0	0,44	0,72	0,75	**0,66**
P20T1	0,63	0,63	0,75	**0,68**
P150T1	0,85	0,63	0,65	**0,73**
P50T3	0,76	0,75	0,63	**0,72**
Group 3				
P50T1	0,59	0,29	0,70	**0,56**
P400T1	0,00	0,00	0,00	**0,00**
P150T3	0,89	0,29	0,57	**0,67**
P1000T3	0,46	0,00	n.a.	**0,19**

Niemeyer et al. JSS, 2010

Integrated Risk Values (Tier 1)



Niemeyer et al. JSS, 2010

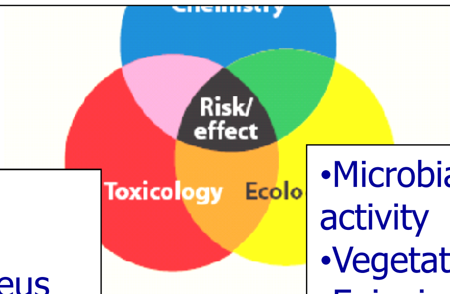


SITE SPECIFIC RISK ASSESSMENT

Santo Amaro da Purificação (Bahia, Brazil)

Detailed evaluation (Tier 2)

- Toxic Pressure (msPAF)
- Comparison of extractable metals with water quality standards



- Reproduction of *Eisenia*
- Reproduction of *Folsomia*
- Reproductions of Enchytraeus
- Plant growth
- Reproduction of *Daphnia* assays
- Algae growth

- Microbial biomass and activity
- Vegetation composition
- Epigeic arthropod communities
- Litter decomposition

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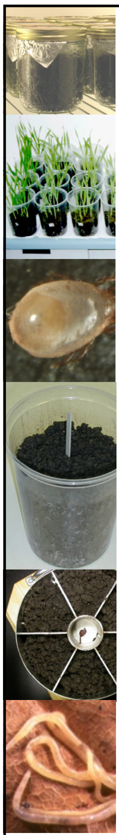
Total and extractable metals

Sites	Total (mg/kg)								Extractable (mg/l) in 0.01 M $CaCl_2$ (1:10; v:v)							
	Pb[a]	Cd[a]	Cu[a]	Zn[a]	Cr	Ni	Fe	Mn	Pb	Cd	Cu	Zn	Cr	Ni	Fe	Mn
Group 1																
Ref 1	16	<0.2	66	94	77	54	45000	840	<0.1	<0.01	<0.8	<0.2	<0.8	<1.4	<1.1	<0.5
P1000T1	23	<0.2	60	80	62	46	48000	360	0.1	0.4	<0.8	1.9	<0.8	1.6	<1.1	71
P20T3	308	<0.2	56	420	78	60	49000	672	<0.1	<0.01	<0.8	<0.2	<0.8	<1.4	<1.1	<0.5
P400T3	179	0.3	44	90	59	46	34000	760	<0.1	<0.01	<0.8	<0.2	<0.8	<1.4	<1.1	<0.5
Group 2																
Ref 2	13	<0.2	18	24	16	28	2900	34	<0.1	<0.01	<0.8	0.2	<0.8	<1.4	<1.1	0.8
P0	1264	<0.2	76	3800 (2.8)	72	57	52000	674	<0.1	<0.01	<0.8	<0.2	<0.8	<1.4	<1.1	<0.5
P20T1	133	<0.2	56	220	80	56	41000	780	<0.1	<0.01	<0.8	<0.2	<0.8	<1.4	<1.1	<0.5
P150T1	37460 (10.4)	771 (9.8)	594 (1.6)	42200 (33.5)	57	70	110000	1720	2.2	7.3	<0.8	1.3	<0.8	<1.4	<1.1	<0.5
P50T3	26074 (7.1)	62	3196 (8.2)	95940 (73.5)	80	40	117000	5880	<0.1	<0.01	<0.8	1.0	<0.8	<1.4	<1.1	<0.5
Group 3																
Ref 3	152	<0.2	40	260	59	40	53000	820	<0.1	0.28	<0.8	<0.2	<0.8	<1.4	<1.1	1.3
P50T1	164	<0.2	60	240	80	58	43000	720	<0.1	<0.01	<0.8	<0.2	<0.8	<1.4	<1.1	<0.5
P400T1	961	8.8	60	840	64	48	35000	540	<0.1	<0.01	<0.8	<0.2	<0.8	<1.4	<1.1	<0.5
P150T3	2200	12	108	3300	84	58	56000	678	<0.1	<0.01	<0.8	0.2	<0.8	<1.4	<1.1	<0.5
P1000T3	99	<0.2	56	156	84	52	49000	568	<0.1	<0.01	<0.8	<0.2	<0.8	<1.4	<1.1	0.9



Aquatic tests
(*Retention function*)

Eluate
1:4 (soil:water)

Reproduction of *Daphnia*
(ISO 6341)

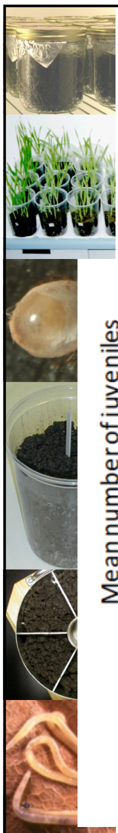
Algae inhibition growth
(ISO 8692)

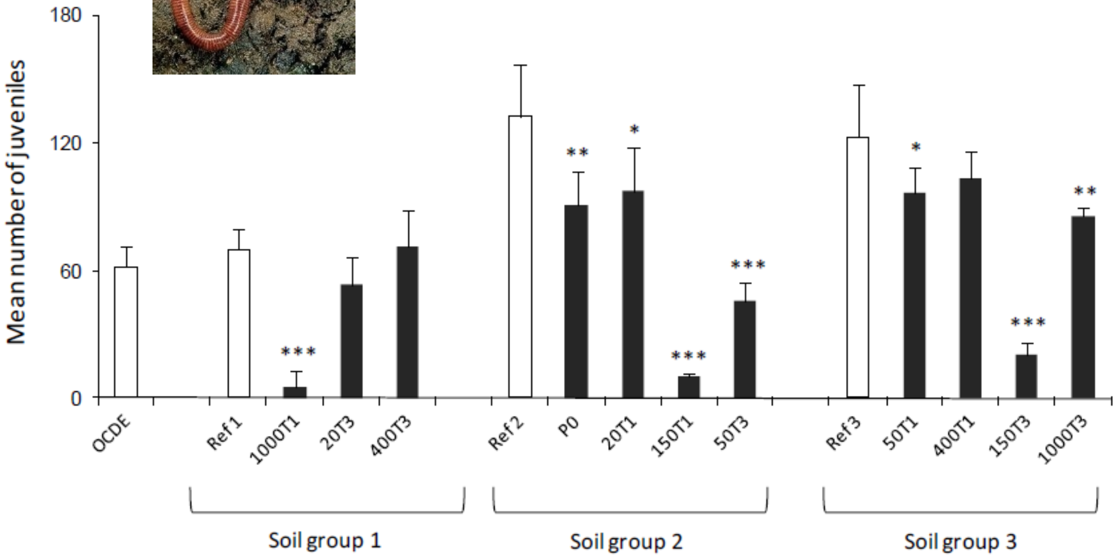
No chronic toxicity was observed.

Integrated Risk values (Tier 2 - retention function)

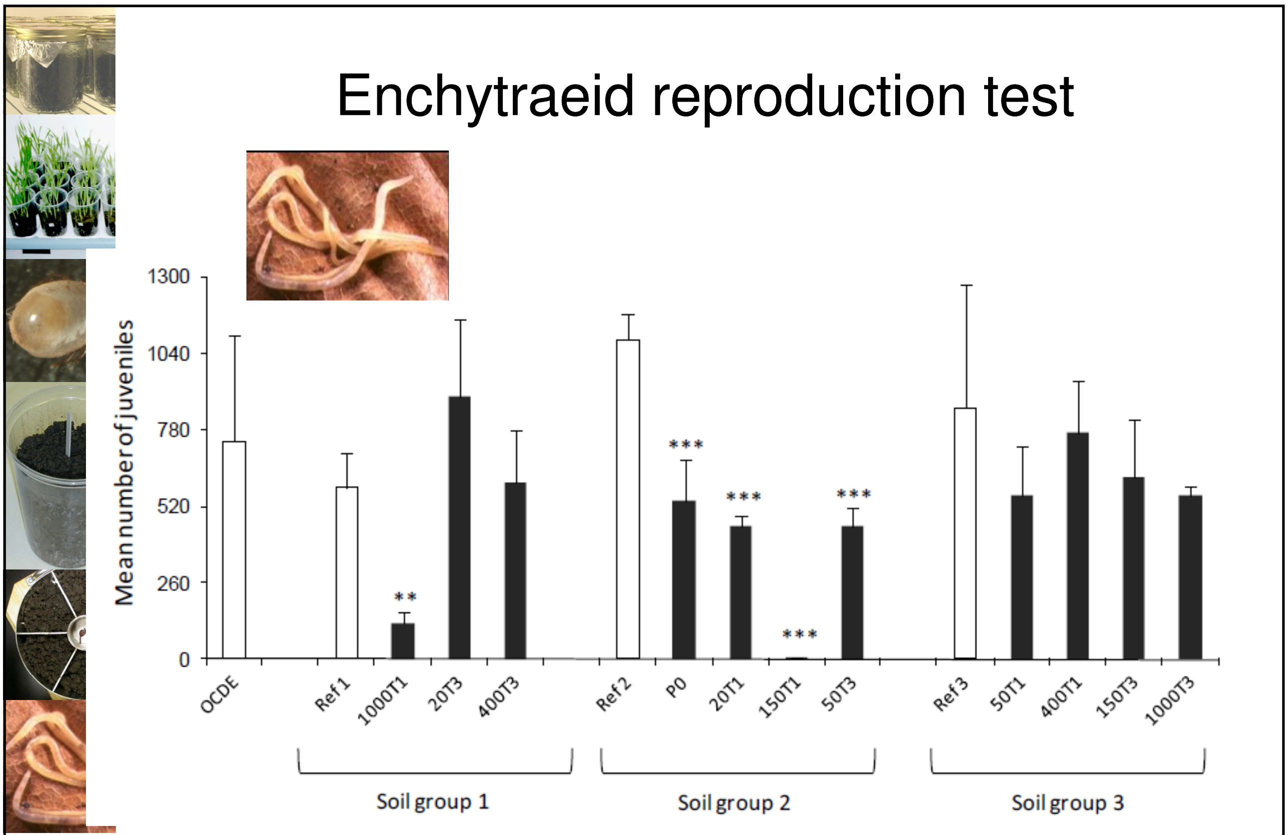
	Chem LoE (Extractable metals)	Growth *P. subcapitata*	Reprod. *D. magna*	**Combined EcLoE**	**IR Retention Function**
Group 1					
1000T1	0.99	0.00	0.00	0.00	0.88
20T3	0.00	0.00	0.00	0.00	0.00
400T3	0.00	0.00	0.00	0.00	0.00
Group 2					
P. Zero	0.00	0.00	0.00	0.00	0.00
20T1	0.00	0.00	0.00	0.00	0.00
150T1	1.00	0.00	0.00	0.00	0.99
50T3	0.44	0.00	0.00	0.00	0.25
Group 3					
50T1	0.00	0.00	0.00	0.00	0.00
400T1	0.00	0.00	0.00	0.00	0.00
150T3	0.00	0.00	0.00	0.00	0.00
1000T3	0.00	0.00	0.00	0.00	0.00

Earthworm reproduction test





Enchytraeid reproduction test



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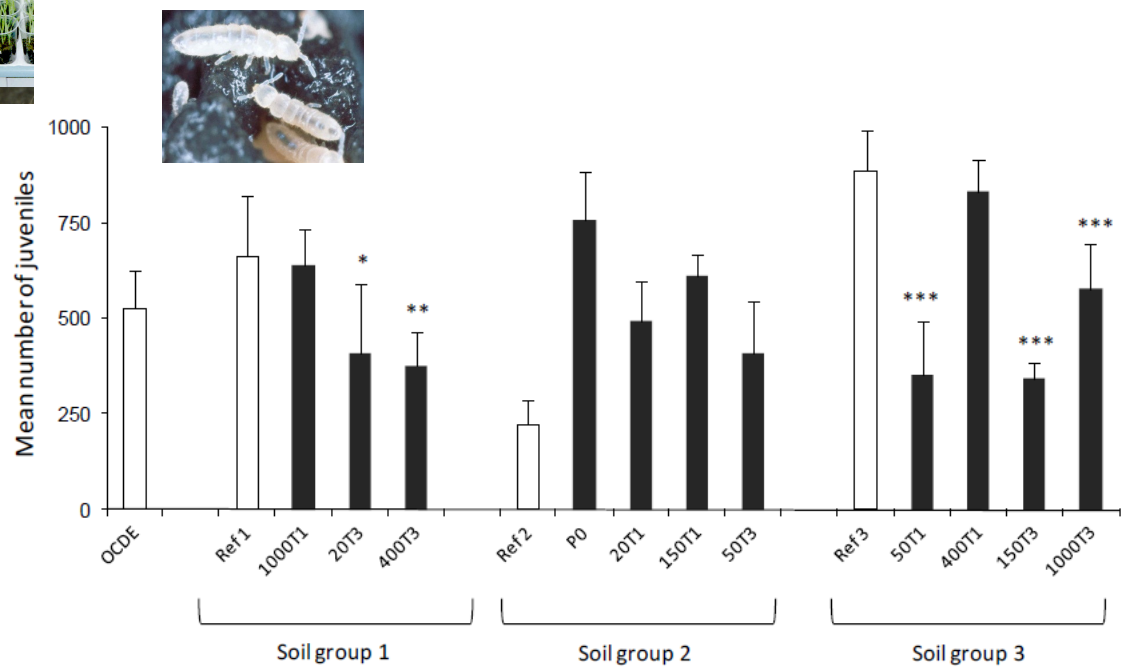
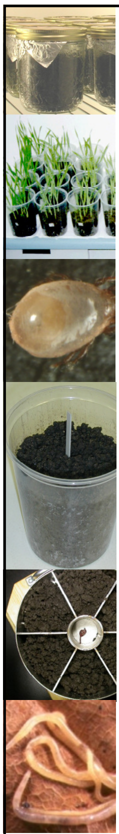
Collembola reproduction test





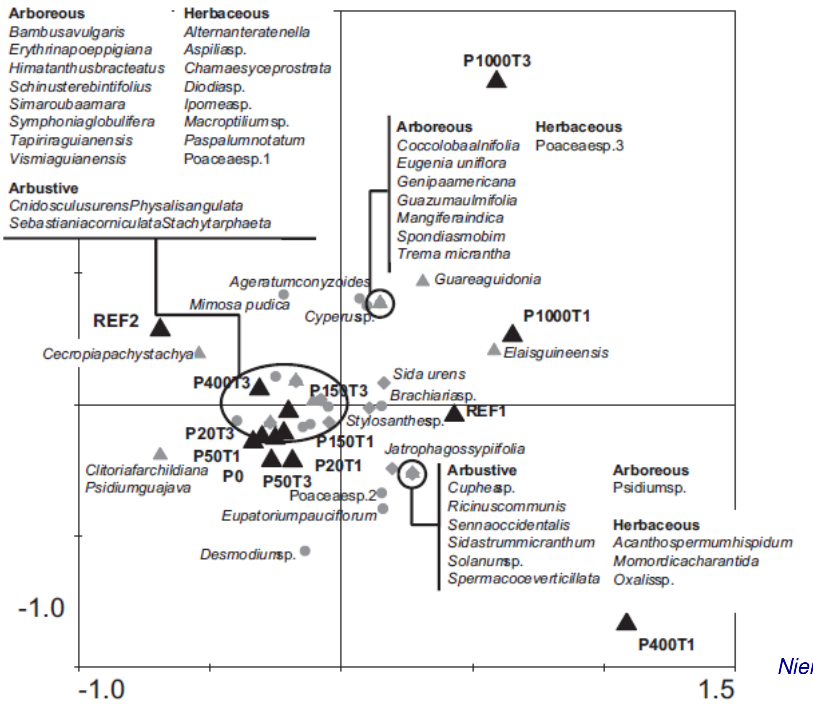
Plant growth test

Soil groups	Shoot length of plants (cm)		Biomass of plants (g; dry weight)	
	A. sativa n=4	*B. rapa* n=4	*A. sativa* n=4	*B. rapa* n=4
Ref 1	31.7 ± 1.8	3.3 ± 0.2	0.17 ± 0.01	0.17 ± 0.04
P1000T1	39.5 ± 5.3	2.7 ± 0.2**	0.31 ± 0.04	0.07 ± 0.02***
P20T3	18.4 ± 1.0***	2.7 ± 0.2**	0.11 ± 0.01**	0.07 ± 0.01***
P400T3	23.0 ± 1.9**	2.3 ± 0.3***	0.16 ± 0.01	0.06 ± 0.01***
Ref 2	25.2 ± 3.9	2.7 ±0.2	0.17 ± 0.02	0.09 ± 0.02
P0	26.3 ± 2.1	3.7 ±0.4	0.23 ± 0.02	0.24 ± 0.02
P20T1	25.2 ± 2.3	2.9 ± 0.0	0.20 ± 0.03	0.14 ± 0.01
P150T1	19.8 ± 1.9*	2.7 ± 0.2	0.14 ± 0.04	0.04 ± 0.01**
P50T3	19.5 ± 3.3*	2.9 ± 0.2	0.17 ± 0.01	0.06 ± 0.01
Ref 3	34.8 ± 3.7	4.7 ±1.2	0.26 ± 0.05	0.45 ± 0.17
P50T1	25.3 ± 2.9**	4.3 ±0.2	0.17 ± 0.03*	0.32 ± 0.05
P400T1	42.7 ± 3.0	7.3 ± 0.4	0.29 ± 0.05	0.91 ± 0.08
P150T3	28.9 ± 1.0*	3.1 ± 0.1**	0.16 ± 0.03*	0.16 ± 0.02***
P1000T3	30.9 ± 5.3	4.8 ± 0.2	0.24 ± 0.05	0.30 ± 0.04*

Chemical and Ecotoxicological Risk values (Tier 2)

	Chem LoE (total metals)	Reprod. *F. candida*	Reprod. *E. crypticus*	Reprod. *E. andrei*	Shoot lenght of *A. sativa*	Shoot lenght of *B. rapa*	Dry W *A. sativa*	Dry W *B. rapa*	Combined EcLoE
Group 1									
1000T1	0.00	0.03	0.96	0.93	0.00	0.17	0.00	0.58	0.63
20T3	0.23	0.38	0.00	0.23	0.42	0.15	0.32	0.59	0.32
400T3	0.05	0.43	0.00	0.00	0.28	0.28	0.08	0.63	0.28
Group 2									
P0	0.85	0.00	0.39	0.31	0.00	0.00	0.00	0.00	0.12
20T1	0.25	0.00	0.59	0.26	0.00	0.00	0.00	0.00	0.16
150T1	1.00	0.00	0.99	0.92	0.17	0.00	0.14	0.53	0.71
50T3	1.00	0.00	0.59	0.66	0.14	0.00	0.00	0.27	0.29
Group 3									
50T1	0.19	0.04	0.00	0.21	0.27	0.09	0.36	0.17	0.17
400T1	0.49	0.00	0.00	0.16	0.00	0.00	0.00	0.00	0.02
150T3	0.72	0.06	0.00	0.83	0.17	0.33	0.29	0.58	0.41
1000T3	0.06	0.61	0.00	0.30	0.11	0.00	0.00	0.21	0.21

Ecological surveys (plant community composition)



Niemeyer et al. EES, 2012

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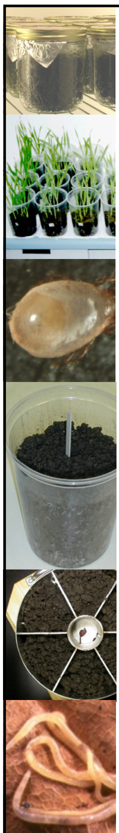
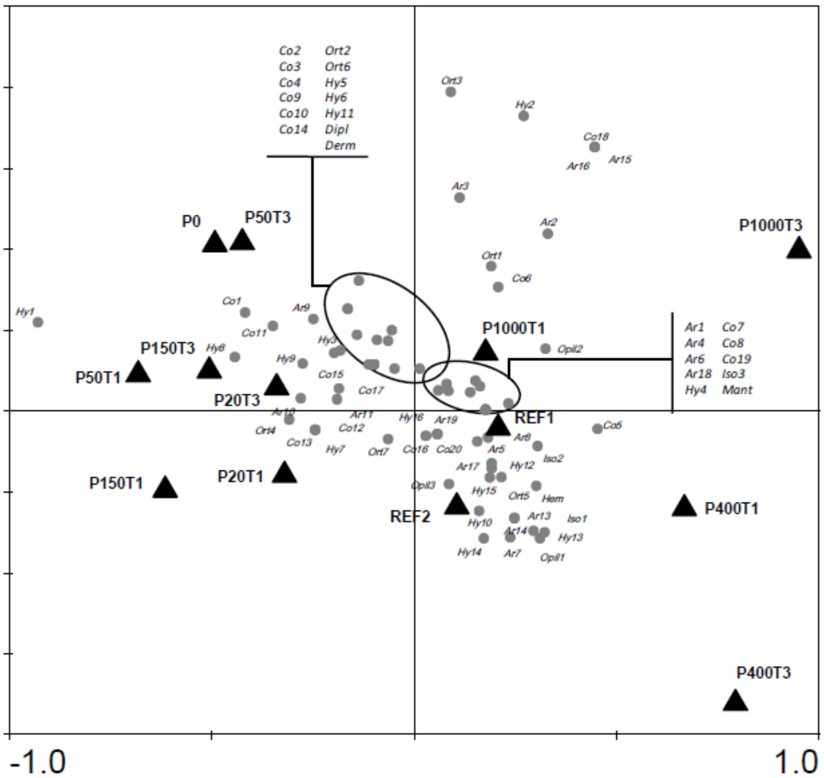
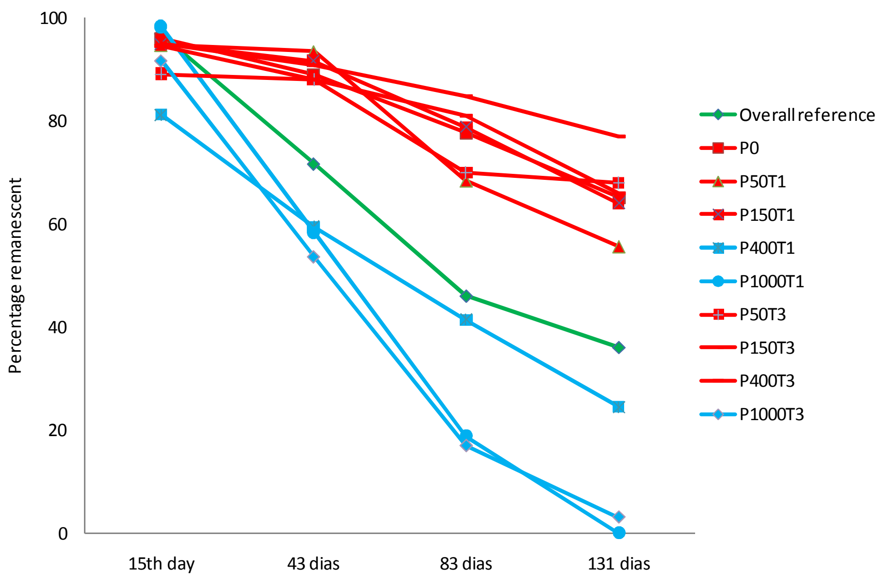


Ecological surveys (invertebrates)



Niemeyer et al. EES, 2012

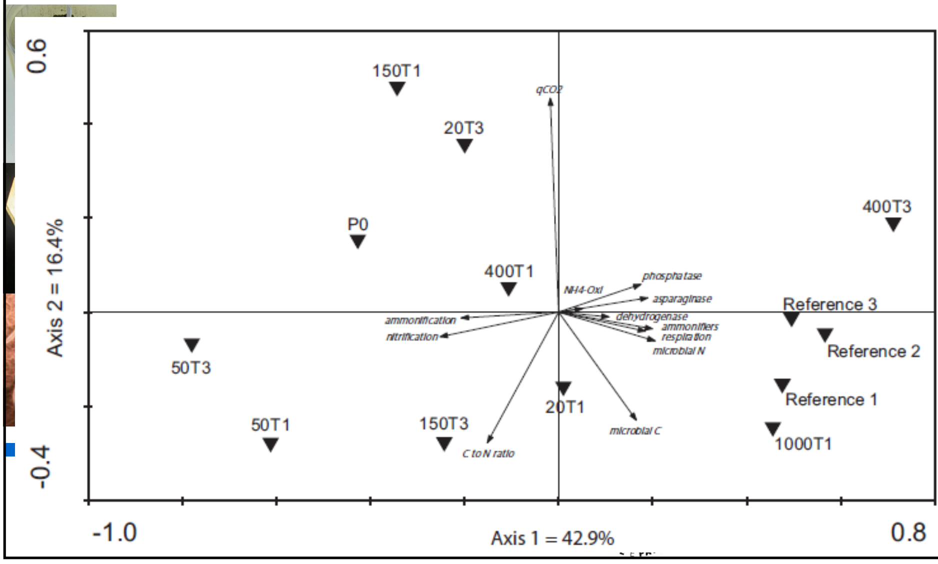
Ecological surveys (organic matter decomposition)



Soil groups	Decomposition rate[a] k (monthly) $n=4$
Overall reference	0.266 ± 0.144
P0	0.047***
P20T1	n.d.
P20T3	n.d.
P50T1	0.063***
P50T3	0.041***
P150T1	0.044***
P150T3	0.025***
P400T1	0.166*
P400T3	0.042***
P1000T1	0.452
P1000T3	0.383

Niemeyer et al. EES, 2012

Sites	MBC (µg/g)	MBN (µg/g)	C/N	qCO_2 (mg CO_2-C/g biomass C/h)	Dehydrogenase (µg PNP/g/d)	Acid phosphatase (ug PNF/g/h)	Asparaginase (µg N-NH_4^+/g/h)	Ammonification (µg N/g/day)	Ammonifiers (log of MPN/g)	Nitrification rate (%)	Nitrifiers NH_4^+ oxidizers (log MPN/g)
Ref. 1	431.2 ± 109.7	36.6 ± 7.6	12.10 ± 3.41	2.8	9.6 ± 1.2	383.2 ± 58.1	76.7 ± 5.2	0.4 ± 0.1	6.8 ± 0.5	6.3 ± 2.7	
Ref. 2	374.3 ± 108.8	33.4 ± 7.5	11.90 ± 5.24	1.6	6.8 ± 2.4	618.5 ± 32.0	36.2 ± 20.2	1.1 ± 0.4	7.0 ± 0.3	3.1 ± 1.6	2.3 ± 0.6
Ref. 3	1121.7 ± 279.4	80.3 ± 14.3	13.83 ± 1.14	2.7	5.1 ± 2.4	849.7 ± 43.0	141.7 ± 22.8	0.6 ± 0.9	6.8 ± 0.1	2.2 ± 1.6	2.9 ± 0.0
Overall reference	642.4 ± 416.1	50.1 ± 16.2	12.61 ± 1.06	2.5 ± 0.6	7.2 ± 2.3	617.1 ± 233.2	84.9 ± 53.2	0.7 ± 0.3	6.9 ± 0.1	3.9 ± 2.2	2.3 ± 0.7
P0	178.1 ± 55.1***	5.4 ± 2.8***	36.12 ± 10.35	2.5 ± 1.3	0.7 ± 0.4**	269.0 ± 22.1**	53.8 ± 29.5	1.7 ± 0.2	4.7 ± 0.9***	15.2 ± 9.6**	2.4 ± 0.1
P20T1	252.4 ± 142.3**	12.8 ± 4.6***	20.37 ± 9.89	1.8 ± 0.6	1.3 ± 1.9**	196.4 ± 33.9***	15.9 ± 18.4***	1.1 ± 0.5	5.3 ± 0.3***	12.4 ± 2.1*	2.5 ±± 0.1
P20T3	170.3 ± 174.1 **	18.6 ± 3.5***	9.64 ± 10.68	10.5 ± 7.8	1.4 ± 1.0**	443.4 ± 9.3	71.0 ± 12.7	1.5 ± 0.3	6.2 ± 0.2	13.3 ± 3.5*	2.4 ± 0.2
P50T1	412.9 ± 31.4	11.0 ± 4.2***	41.10 ± 15.23	1.1 ± 0.1	1.2 ± 2.0**	235.7 ± 50.3**	11.2 ± 19.5***	1.8 ± 0.3	5.5 ± 0.5***	17.1 ± 4.9***	2.6 ± 0.1
P50T3	461.7 ± 20.1	22.0 ± 4.5*	21.62 ± 4.76	1.3 ± 0.3	2.1 ± 0.5*	450.3 ± 45.4	32.5 ± 35.1**	1.8 ± 0.3	5.6 ± 0.5***	13.5 ± 4.5*	2.2 ± 0.5
P150T1	115.5 ± 87.0***	9.3 ± 1.3***	12.03 ± 9.24	12.6 ± 15.9	3.3 ± 0.5	355.3 ± 166.0*	22.8 ± 11.8***	0.4 ± 0.2	5.6 ± 0.1***	8.7 ± 2.8	2.4 ± 0.3
P150T3	543.6 ± 160.8	26.6 ± 3.1**	20.85 ± 7.62	1.3 ± 0.4	2.1 ± 1.1*	651.2 ± 150.7	37.0 ± 12.4*	1.5 ± 0.3	6.6 ± 0.6	10.2 ± 0.5	2.3 ± 0.0
P400T1	797.3 ± 193.3	83.0 ± 21.2	9.62 ± 0.43	3.3 ± 0.4	16.8 ± 3.7	573.1 ± 133.3	91.7 ± 32.9	0.8 ± 0.1	7.0 ± 0.3	−0.2 ± 5.9	2.8 ± 0.0
P400T3	805.3 ± 216.2	59.7 ± 26.5	14.18 ± 2.82	2.3 ± 0.3	1.5 ± 1.1**	792.0 ± 34.5	97.8 ± 16.6	0.4 ± 1.0	6.4 ± 0.1	−3.3 ± 3.17	2.9 ± 0.1
P1000T1	1098.1 ± 184.1	51.1 ± 22.0	23.79 ± 9.79	1.71 ± 0.7	4.8 ± 6.2	515.6 ± 353.5	71.57 ± 18.8	0.6 ± 1.8	6.4 ± 0.4	1.9 ± 7.4	2.8 ± 0.0
P1000T3	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.



Ecological surveys (microbial parameters)

Niemeyer et al. ASE, 2012

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Ecological surveys (microbial parameters)
Variance partitioning

Variance partitioning of microbial and biochemical data according to total metal content and vegetation cover. Values expressed in percentage of total variation excluding covariables (soil moisture, soil organic carbon, and mineral nitrogen contents).

Variables	Variation explained (%)	*p*
Variation of microbial and biochemical parameters (excluding covariables)	66.8[a]	
Covariables	33.2	
Metals and Vegetation (total)	52.2	0.002
Metals	49.1	0.002
Vegetation	25.3	0.002
Metals (pure)	26.9	0.002
Vegetation (pure)	3.1	0.002
Shared	22.2	

[a] Expressed as % of total variation.

Niemeyer et al. ASE, 2012

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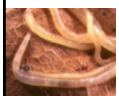
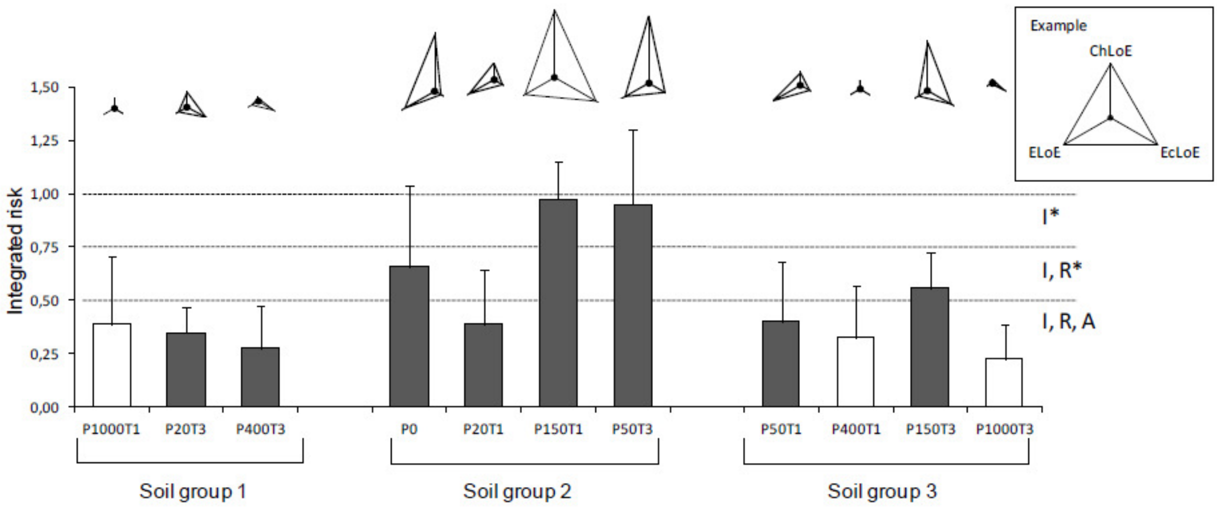
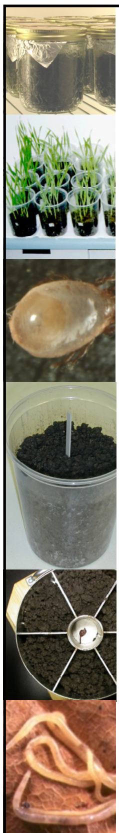
ELoE – Risk values (Tier 2)

Soil groups	Microbial parameters							Surface dewlling arthropods					Decomp	Combined ELoE
	MBC	MBN	Asparaginase	DHA	Ac Fosf	Amon	Nitrif	Araneae	Hymenoptera	Coleoptera	Orthoptera	Other Orders		
Group 1														
1000T1	0.41	0.02	0.16	0.34	0.16	0.15	0.00	0.86	0.42	0.32	0.42	0.45	0.41	0.37
20T3	0.73	0.63	0.16	0.81	0.28	0.54	0.00	0.63	0.50	0.45	0.24	0.26	n.d.	0.46
400T3	0.20	0.16	0.13	0.80	0.22	0.39	0.00	0.65	0.12	0.26	0.29	0.60	0.84	0.44
Group 2														
P. Zero	0.72	0.89	0.37	0.90	0.56	0.60	0.74	0.76	0.41	0.27	0.25	0.76	0.82	0.69
20T1	0.61	0.74	0.81	0.82	0.68	0.34	0.69	0.86	0.57	0.74	0.18	0.29	n.d.	0.63
150T1	0.82	0.81	0.73	0.54	0.42	0.48	0.55	0.69	0.57	0.82	0.53	0.76	0.84	0.69
50T3	0.28	0.56	0.62	0.70	0.27	0.62	0.71	0.88	0.49	0.48	0.47	0.53	0.84	0.62
Group 3														
50T1	0.36	0.78	0.87	0.83	0.62	0.61	0.77	0.90	0.66	0.13	0.22	0.18	0.76	0.67
400T1	0.19	0.40	0.57	0.57	0.07	0.12	0.00	0.48	0.23	0.67	0.25	0.49	0.38	0.37
150T3	0.15	0.47	0.56	0.70	0.05	0.53	0.62	0.99	0.52	0.07	0.29	0.18	0.91	0.47
1000T3	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	0.54	0.45	0.44	0.13	0.29	0.31	0.38



Integrated Risk Values (Tier 2)

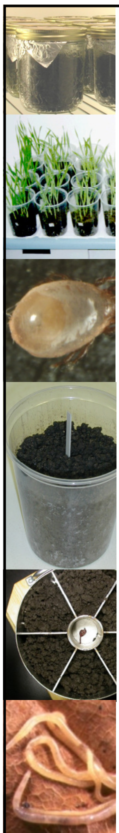




Major outcome

- Results from tier 2 confirmed the risk pointed by tier 1, with points outside the smelter presenting low or environmentally acceptable risk values, and points inside the smelter area presenting high (or very high) risk values, especially in sites associated with tailing deposits.

- The low toxicity in eluate tests indicated high adsorption of metals in soil, probably favored by neutral pH, content and type of clay, and ageing, and consequently no potential risk on retention function in most of points.



Major outcome

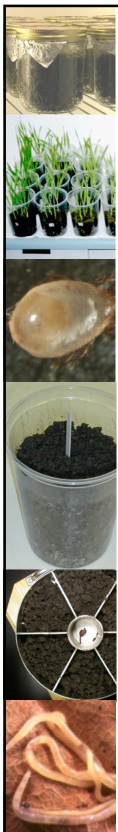
➢In general, the present results indicated that the failed recovery of the area by covering tailings with soil from another site, and the consequently failure in revegetating the area, created inappropriate conditions for the establishment of plant, microbial and animal communities in some of the sites inside the area.

➢So, besides the direct effects of metal contamination seen on ecotoxicological effects, also indirect effects are visible from the presence of these contaminants, compromising the functioning of the ecosystem inside the smelter area.

Major outcome

- High risk values in habitat function above 0.75 inside the smelter area indicate the need to proceed with some remediation action, such as encapsulation of tailing and recovery of vegetation of the smelter area.

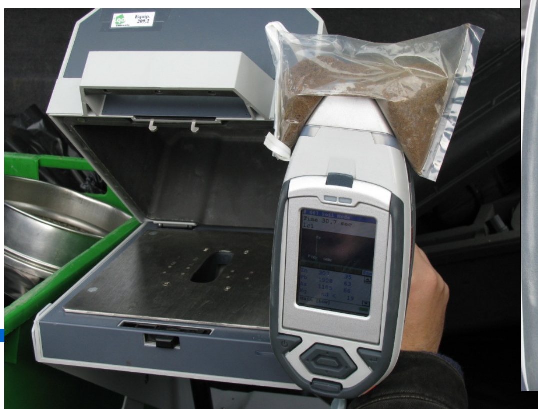
- These actions not only could improve soil conditions and ecosystem functioning, but they could mainly avoid the transport of contaminants to other environmental compartments, namely via dust dispersal to outside the area, or via surface runoff to the existing temporary ponds and the Subaé river.



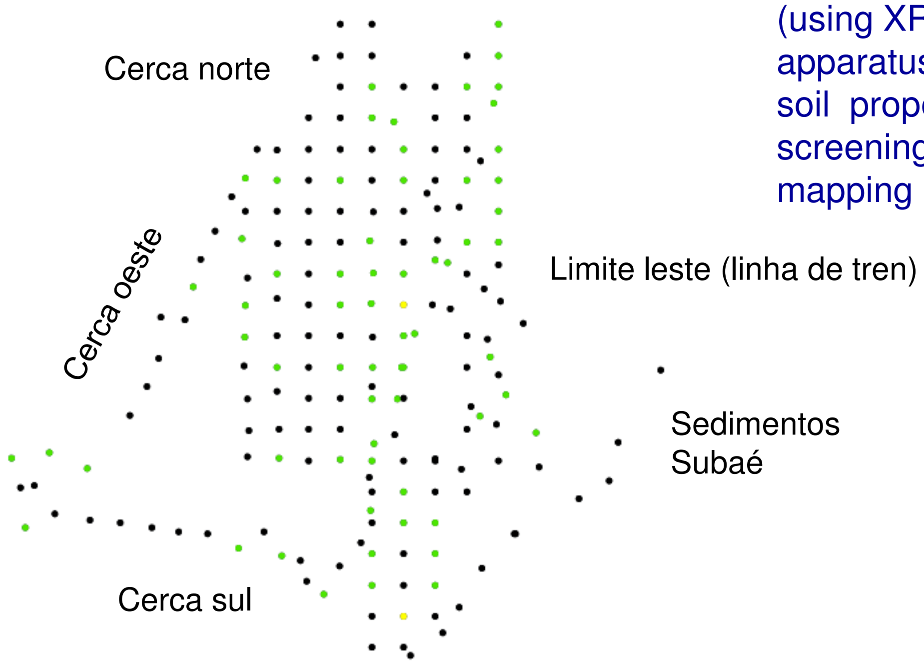
Ongoing activities

- Evaluate effects of runoff contamination to the Subaé River – preliminary chemical and ecotoxicological results confirm contamination of sediment of the river downstream the pipeline entrance.

- Extensive metal (using XRF apparatus) and soil properties screening for mapping



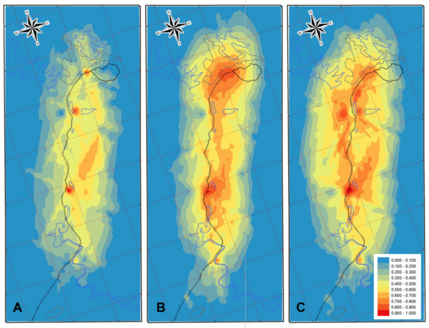
Ongoing activities



- Extensive metal (using XRF apparatus) and soil properties screening for mapping

Ongoing activities

➢ Modeling results from different LoEs and individual and combined risk values with metal and soil properties - link with previous extensive screening to create a risk map for the area



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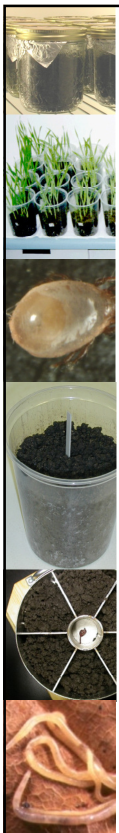


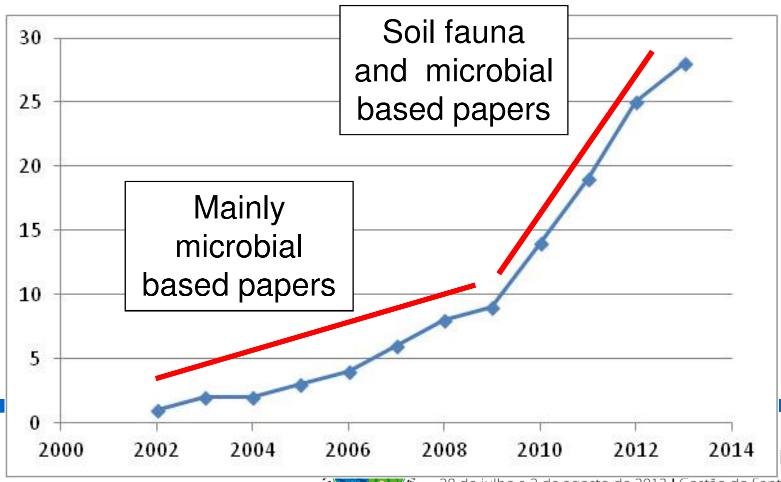
Outline of presentation

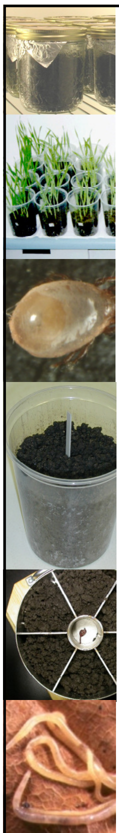
- *Importance of Ecotoxicology in Ecological Risk Assessment (ERA)*
- *Major tools (tests) used in Ecotoxicology)*
- *Case study from Brazil*
- ***Evolution of Soil Ecotoxicology in Brazil and perspectives***

Soil ecotoxicology in Brazil

In 2005 only 1 team doing research in ecotoxicology
Currently around 10 teams are doing active research in soil ecotoxicology

Papers publish in ISI journals (effect assessment)





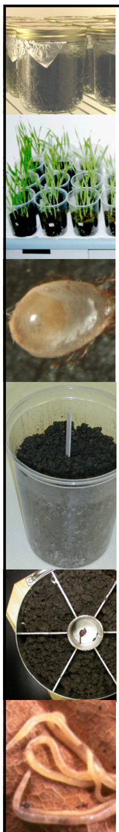
My opinion on future lines of research

- ➤ RETROSPECTIVE RISK ASSESSMENT

Moving beyond the VRQ for CONAMA 420 and derive ecotoxicologically and region specific prevention and investigation values using representative soils from each region;

Create a reference soil library with ecotoxicological data

Train professionals (public and private sectors) on ecological risk assessment



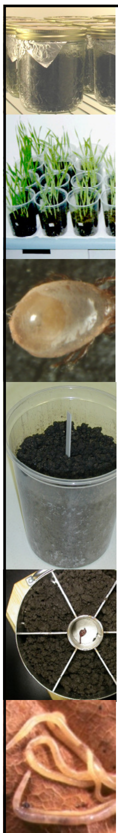
My opinion on future lines of research

➢ PROSPECTIVE RISK ASSESSMENT

Further research on using tropical and sub-tropical Brazilian species to use mainly on higher tier testing (also applicable to retrospective RA)

Improve pesticide risk assessment attending region specific characteristics and particular exposure pathways (e.g., more efforts on studies on soil-water interfaces)

Improve risk assessment of wastes/products (sludges, manures, mineral products, etc)



Team Work !

Thank you very much !