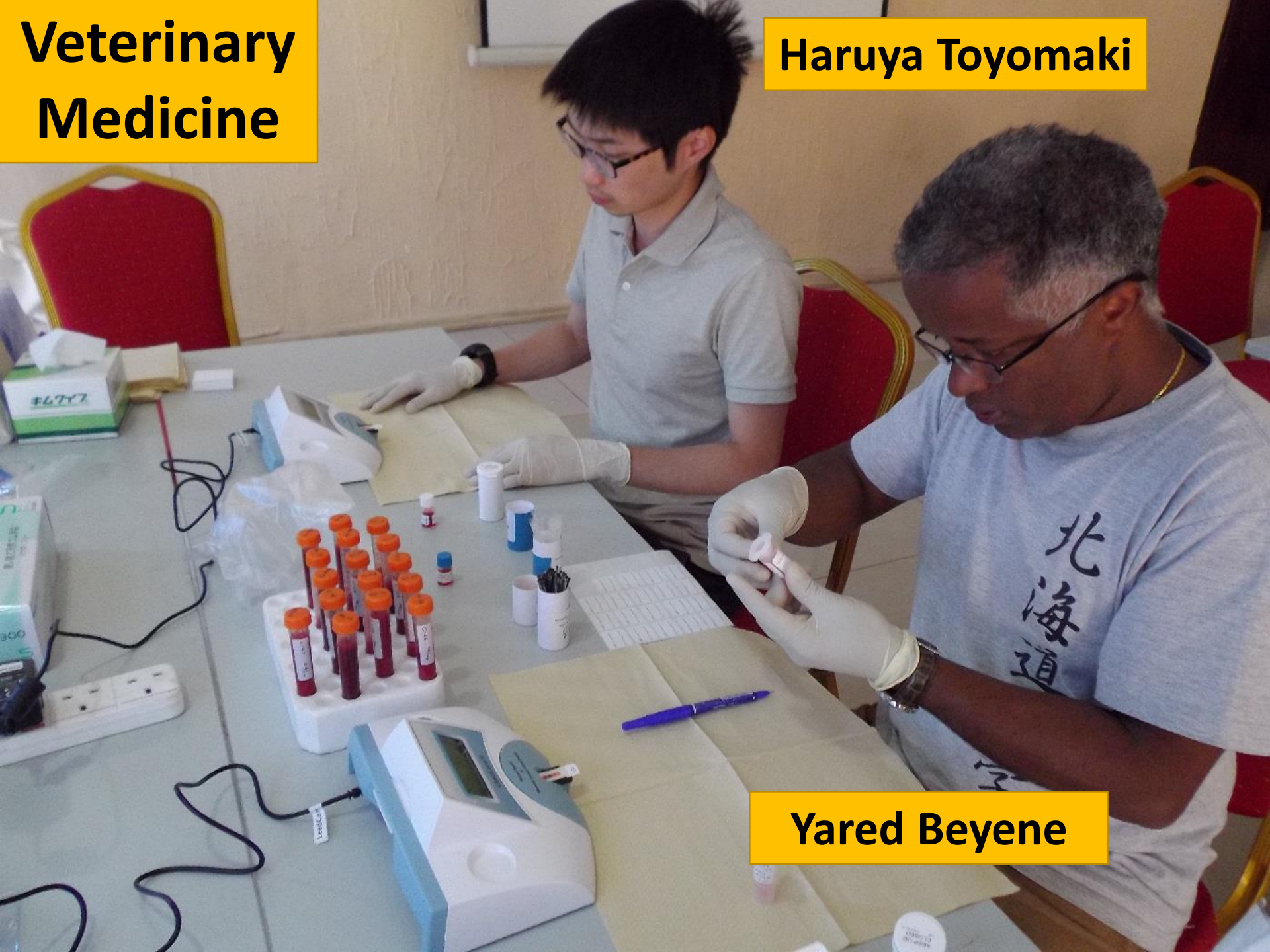
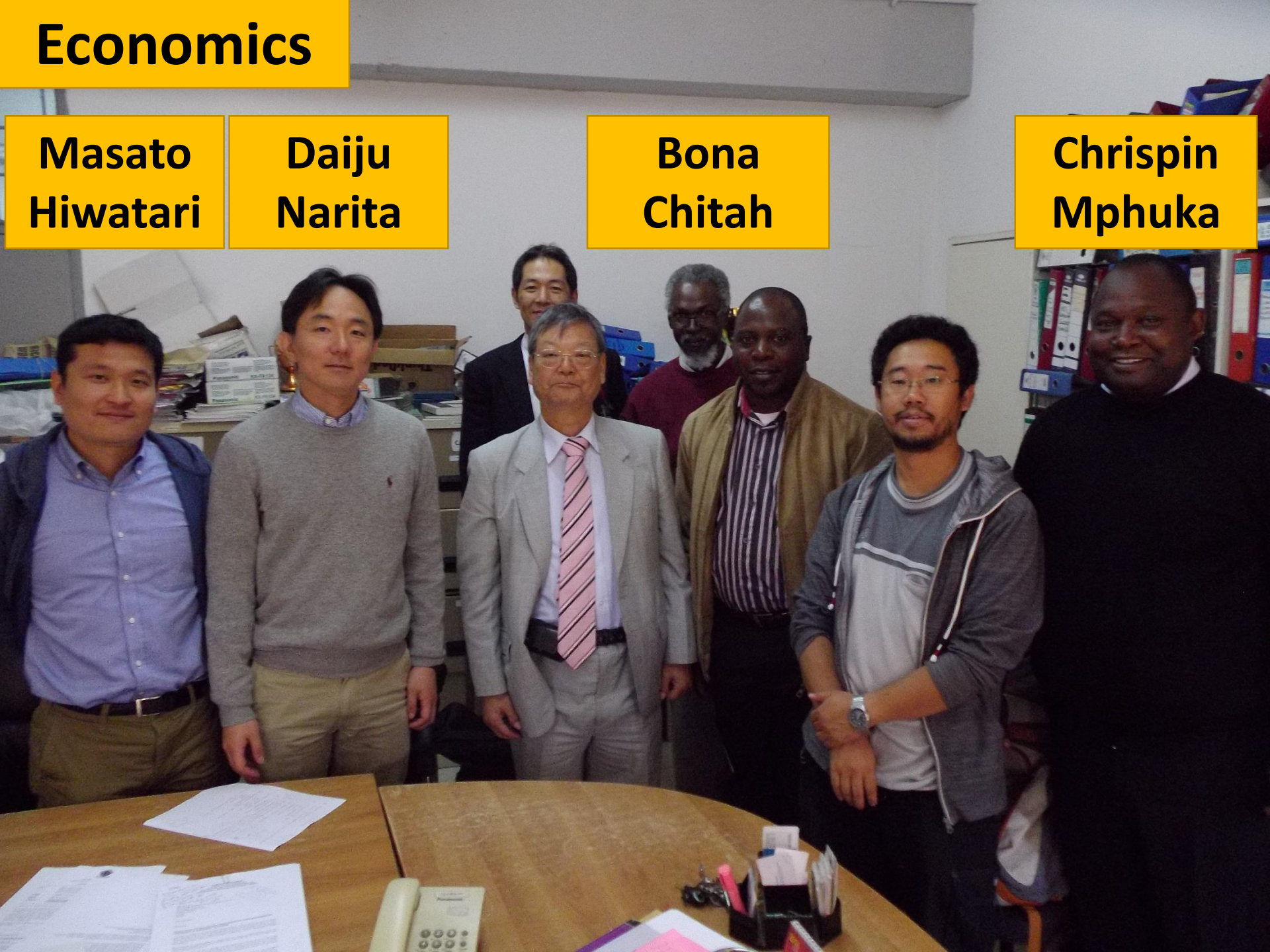
KAMPAI ~TEAM 2 PAST RESEARCH~

Veterinary Medicine
~Dean Dr. Kennedy Choongo~


Veterinary Medicine
Haruya Toyomaki
Yared Beyene

Economics



Medicine & Nursing

Takeshi Saito

Nosiku Sipilanyambe Munyinda

Wakako Fujita

Nancy Zyongwe

Harukazu Tohyama

Sandra Shanunga

Takahiko Yoshida

Education

Robert Serphell

Beatrice Matafwali

Joe Kalima

Gabliel Walubita

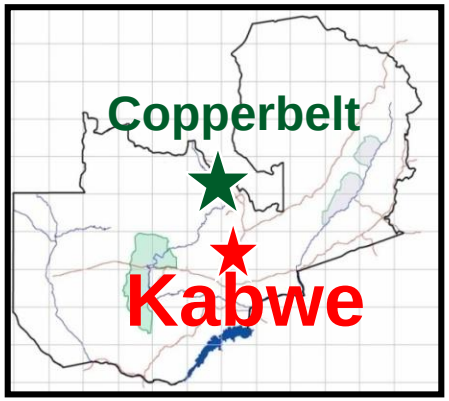
Background -Mining areas in Zambia-

- The rapid economic progress in Africa has contributed to the problem of environmental pollution.
- Major economic activity in Zambia is mining, and this is mainly done in the Copperbelt region (Cu-Co) and Kabwe (Pb-Zn).

Mine in the Copperbelt region

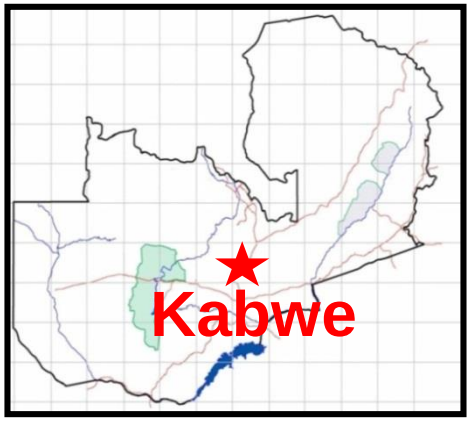
Mining wastes are directly discarded into the Kafue River in the Copperbelt region.

Mine in Kabwe



Background -Kabwe City-

- Kabwe city re-ranked among 'the 10 most polluted places in the world' (Blacksmith Institute, 2014).
- However, no studies have been done to assess metal pollution levels in edible tissues of chicken and cattle from Kabwe.
- No recent study is conducted in human.

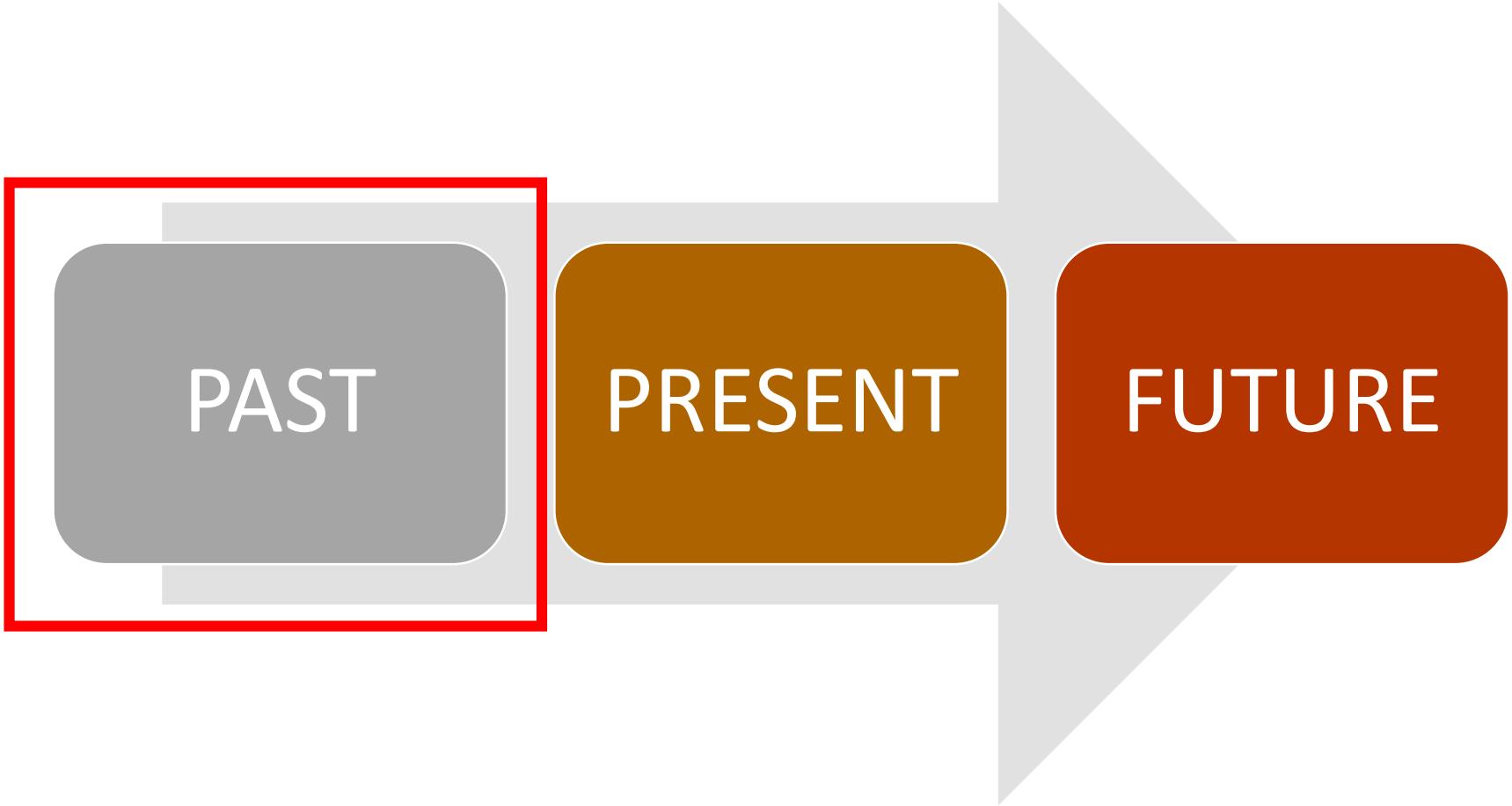


The purpose of the study is

To assess the metal pollution levels in soil, domestic animals and children in Kabwe.

Clarify if soil Pb contamination is source for animals and human, using Stable Pb isotopes.

Research achievement



Outline of PAST study

Pb pollution in **children?**

2012~2015

2009~2011

Pb & Cd contamination in **chicken**

goat

Pb & Cd contamination in **cattle**

Soil contamination and diffusion

2008~2009

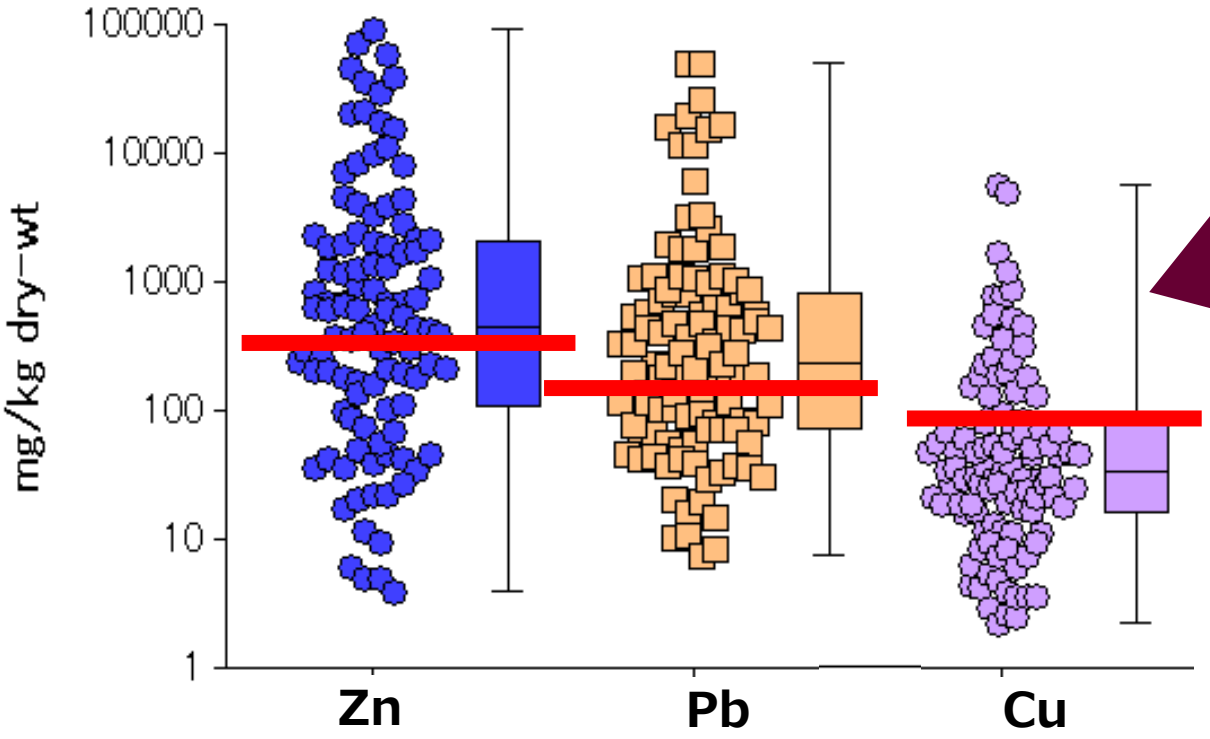
Kabwe Pb & Zn Mining

Soil sampling location in Kabwe (n=101, May 2009)

- GPS coordination
- Microwave digestion (HNO_3)
- Measured by AAS or ICP-MS

(Modified from Google Earth)

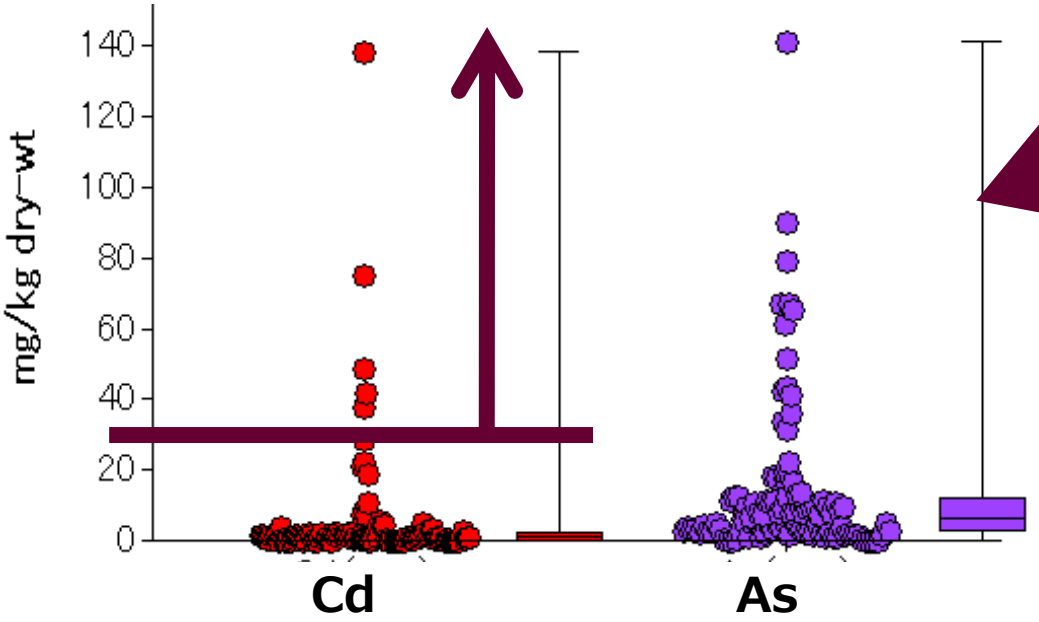
Zn, Pb and Cu concentrations in Kabwe soils



Zn, Cu and Pb concentrations in soil exceeded the FAO Limits.

	Zn	Pb	Cu
Range	5 – 91,000	9 – 51,000	2 – 5,000
Median	610	280	40
FAO Limits	500	150	100

Cd and As in Kabwe soil

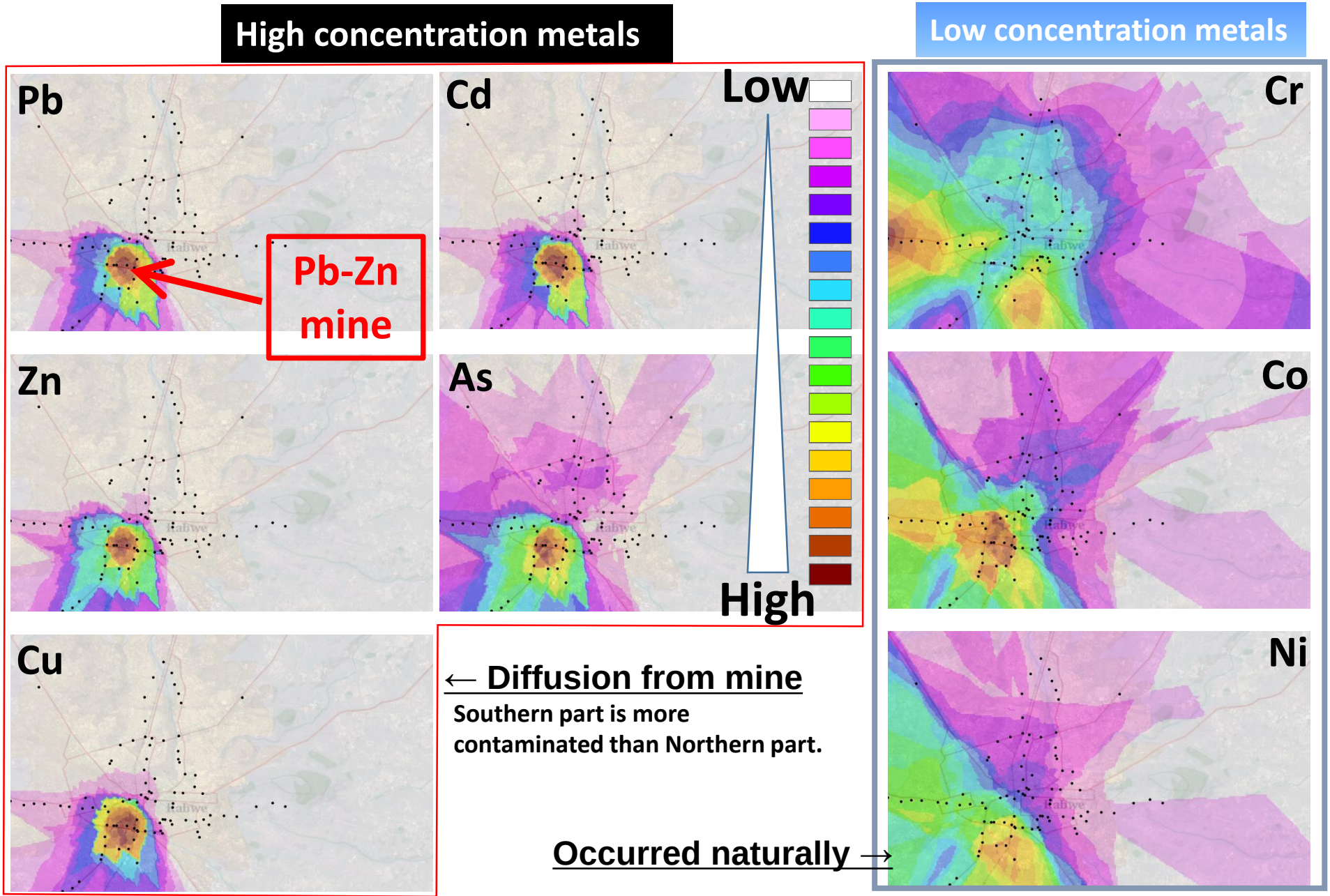


Cd and As concentrations also exceed the FAO Limits.

	Cd	As
Range	0.01 - 140	0.04 - 140
Median	1	7
FAO Limits	1.6	18

Itai-itai disease **(public hazard in Japan)**
→ Soil Cd: 25-50 ppm

GIS (Geographic Information System) analysis



(ArcGIS ver 9.3, ESRI)

Outline of PAST study

Pb & Cd contamination in chicken

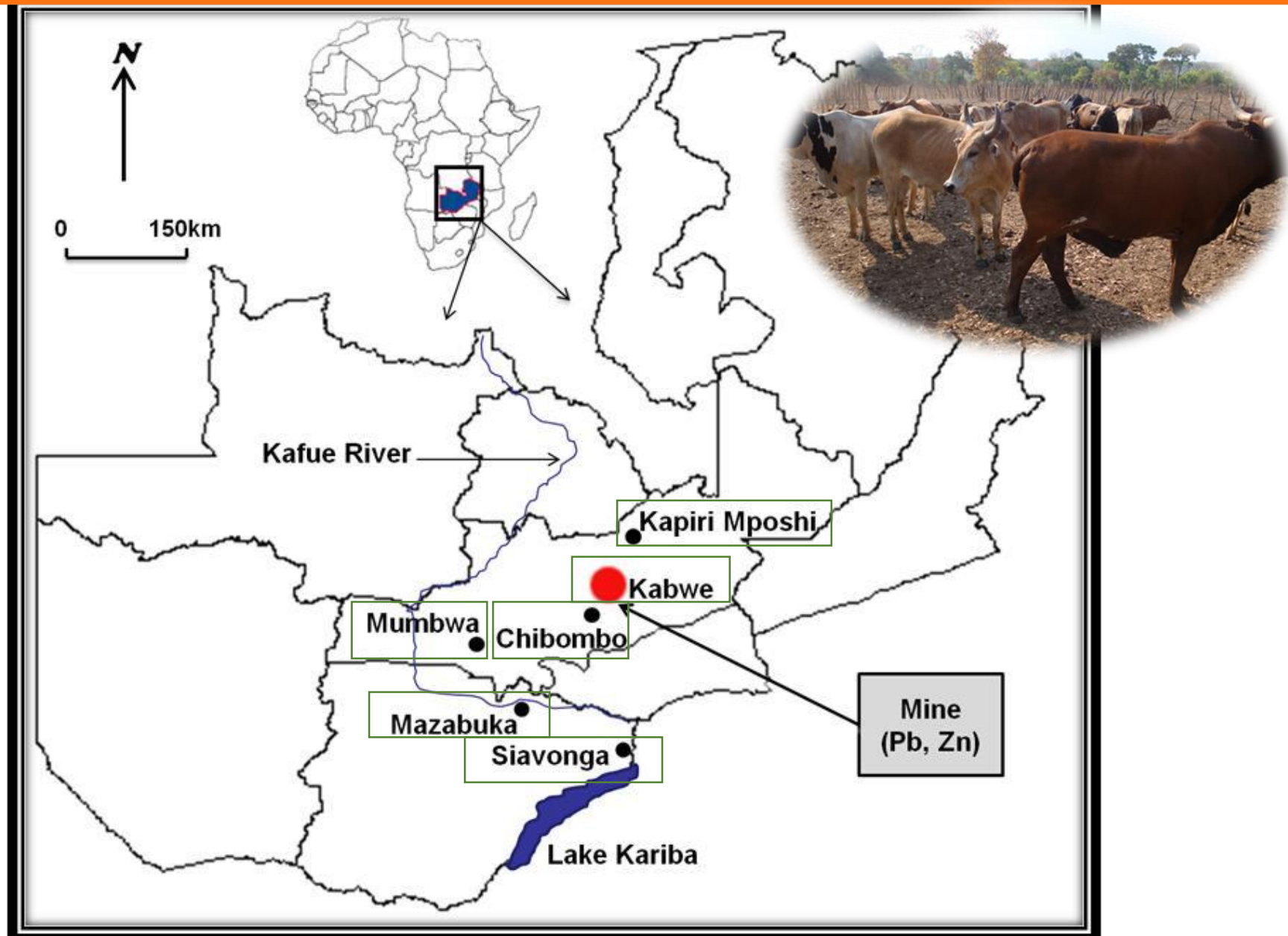
2009~2011

nation
usion

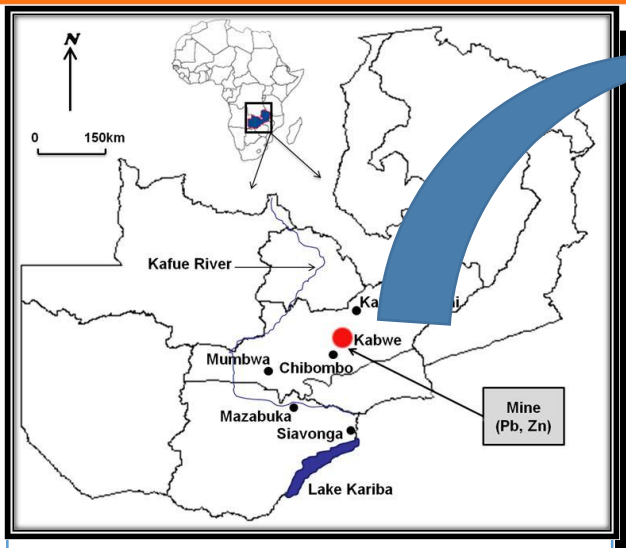
Pb & Cd contamination in cattle

Kabwe Pb & Zn Mining

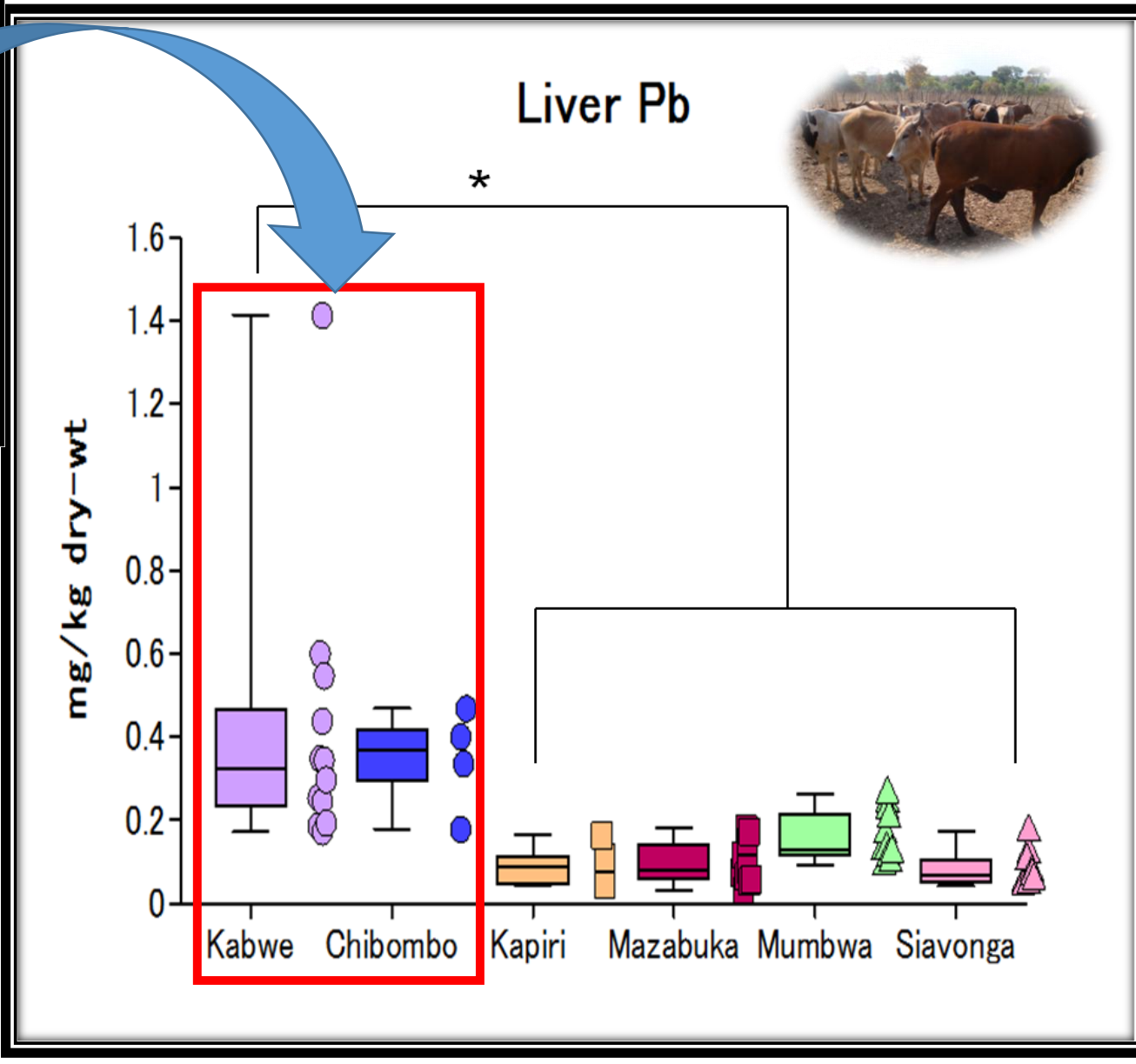
6 towns were selected for cattle sampling



Regional difference of Pb in cattle liver in Zambia

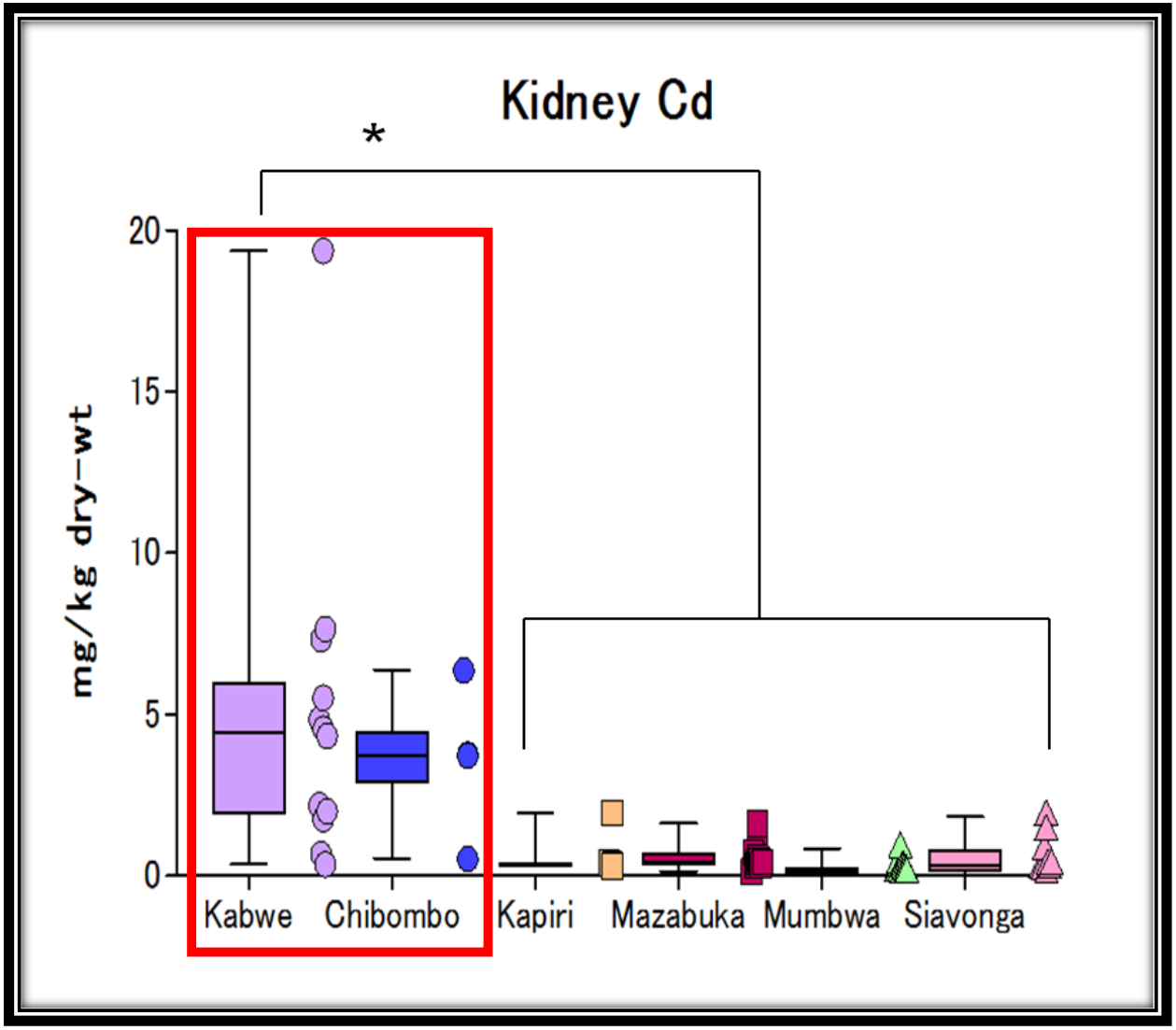


- **Kabwe**
  - ➢ **higher levels of Pb in the liver**
- **Chibombo**
  - ➢ **Pb levels were similar to Kabwe**
- **Other towns**
  - ➢ **Lower Pb levels**
- **Asterisk: $p < 0.001$**

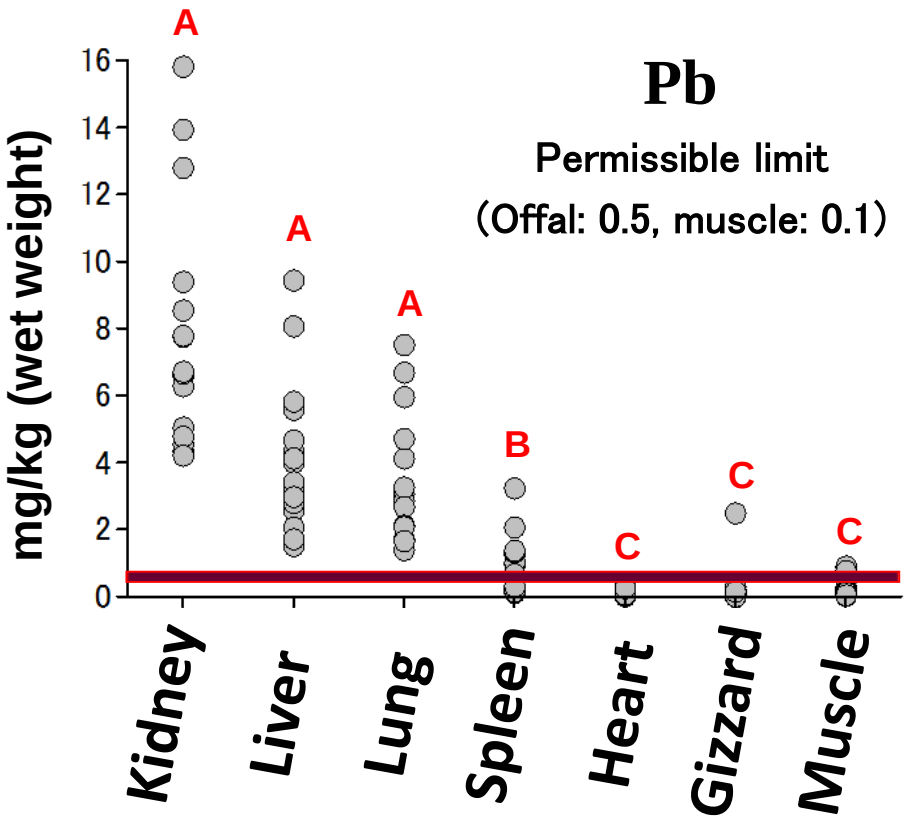


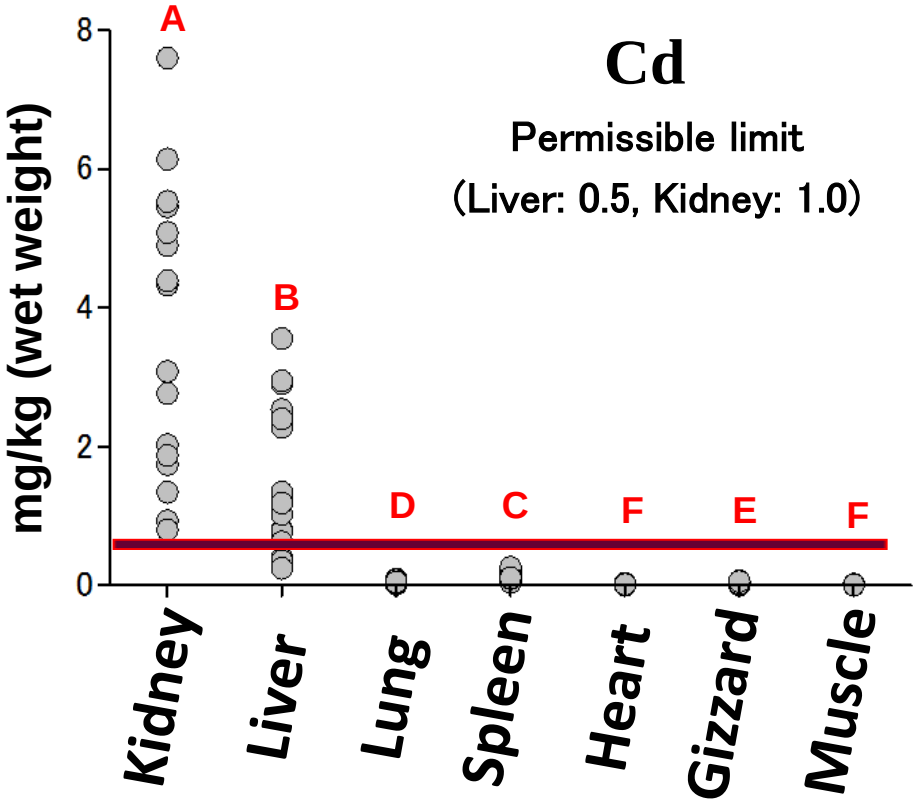
Regional difference of Cd in cattle kidney in Zambia

- **Kabwe**
  - ➢ **Higher levels of Cd in kidneys**
- **Chibombo**
  - ➢ **Cd levels were similar to Kabwe**
- **Other towns**
  - ➢ **Lower Cd levels**
- **Asterisk: $p < 0.001$ (Dunettee test)**



High Pb and Cd levels in muscle and offal of Free-range chickens (n=17) in Kabwe





(Tukey test)

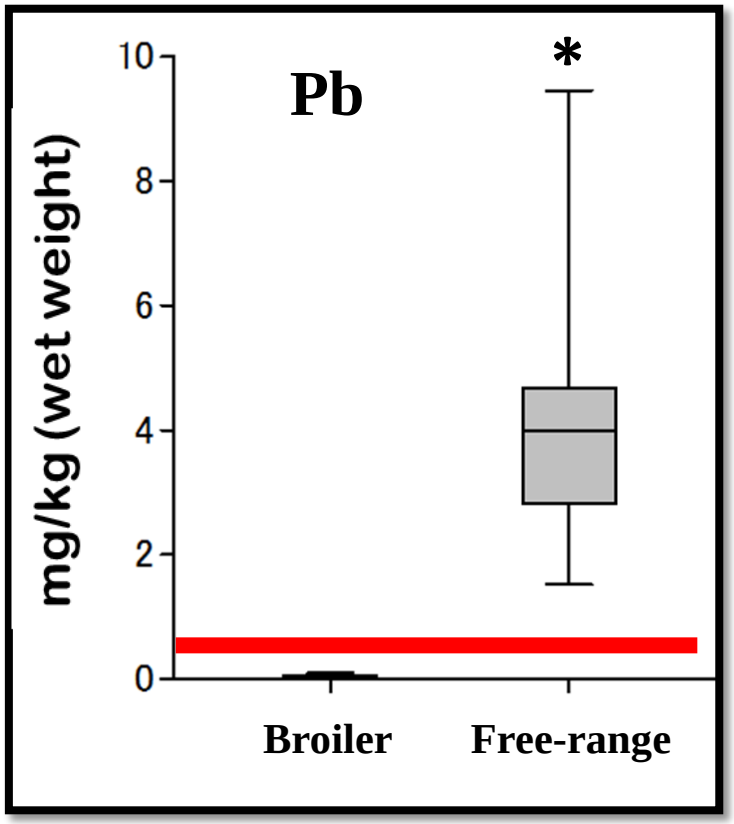
Scavenging Free-range chicken

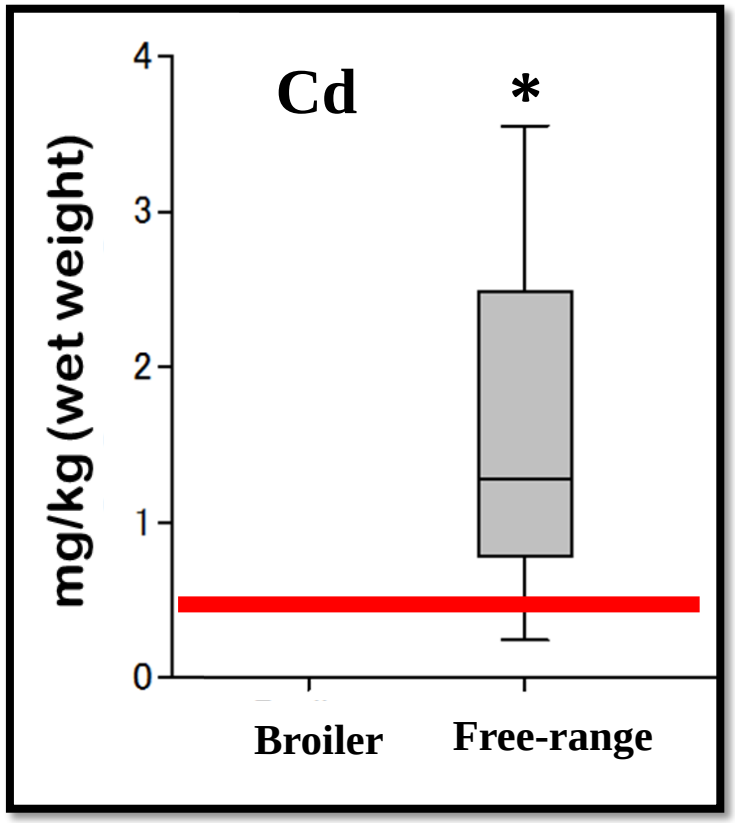
Exceeded the permissible limits

Pb and Cd comparison in liver of Broiler (n=32) and Free-range (n=17) in Kabwe

Broiler  → **Kept indoors and fed commercial feed**

Free-range  → **Roam and scavenge for food in the mine townships**





Outline of PAST study

Pb pollution in children?

2012~2015

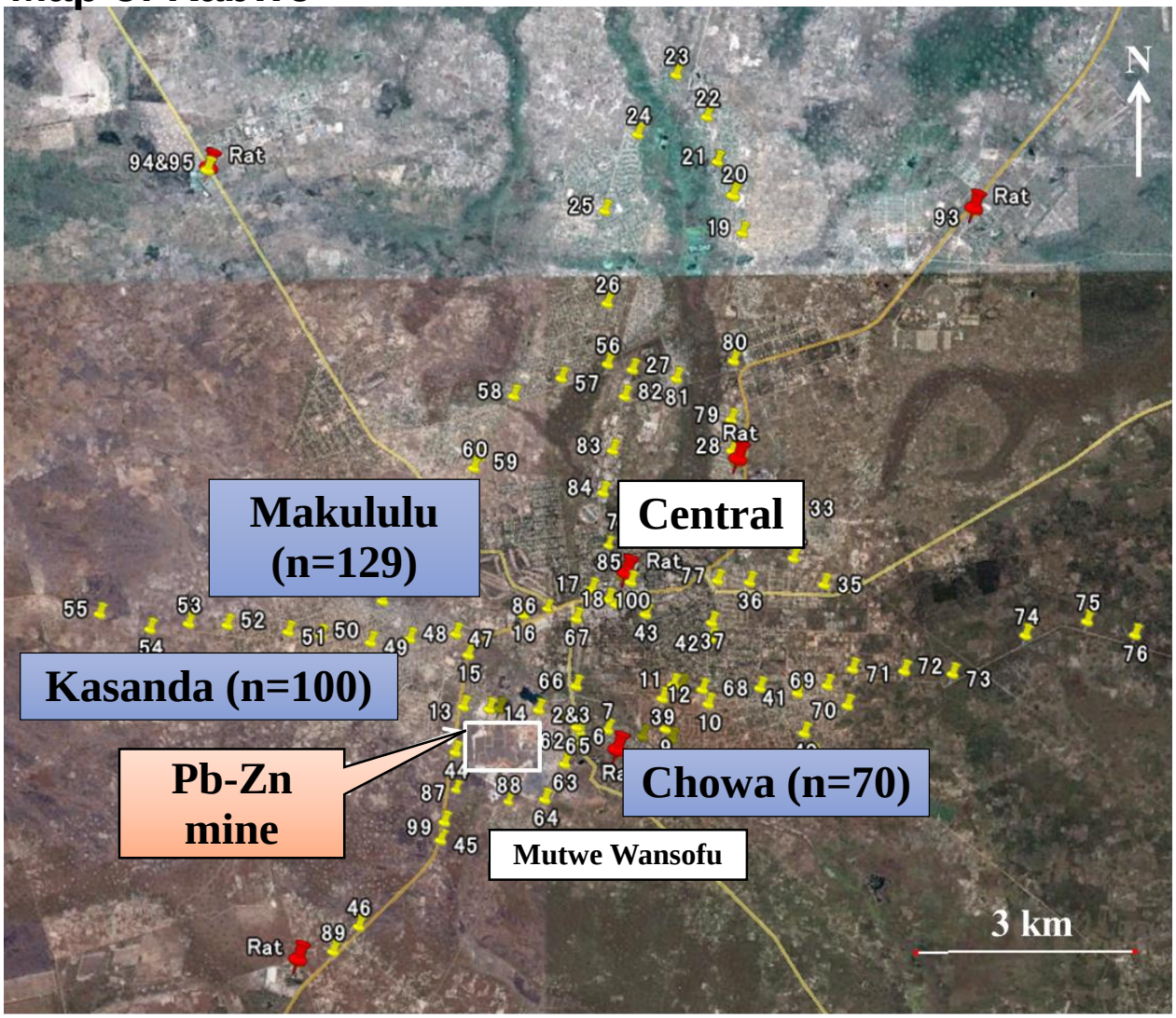
nation

usion

Kabwe Pb & Zn Mining

Human sample collection from Kabwe mine area was done in May 2012

Map of Kabwe



- We collected samples from local communities around Pb-Zn mine.
- 0-7 years old children were main participants.
- 3 Health Centres (Makululu, Kasanda, Chowa area)
- 299 blood
- 250 urine
- 250 feces

(It was difficult to collect urine and fecal samples from all participants)

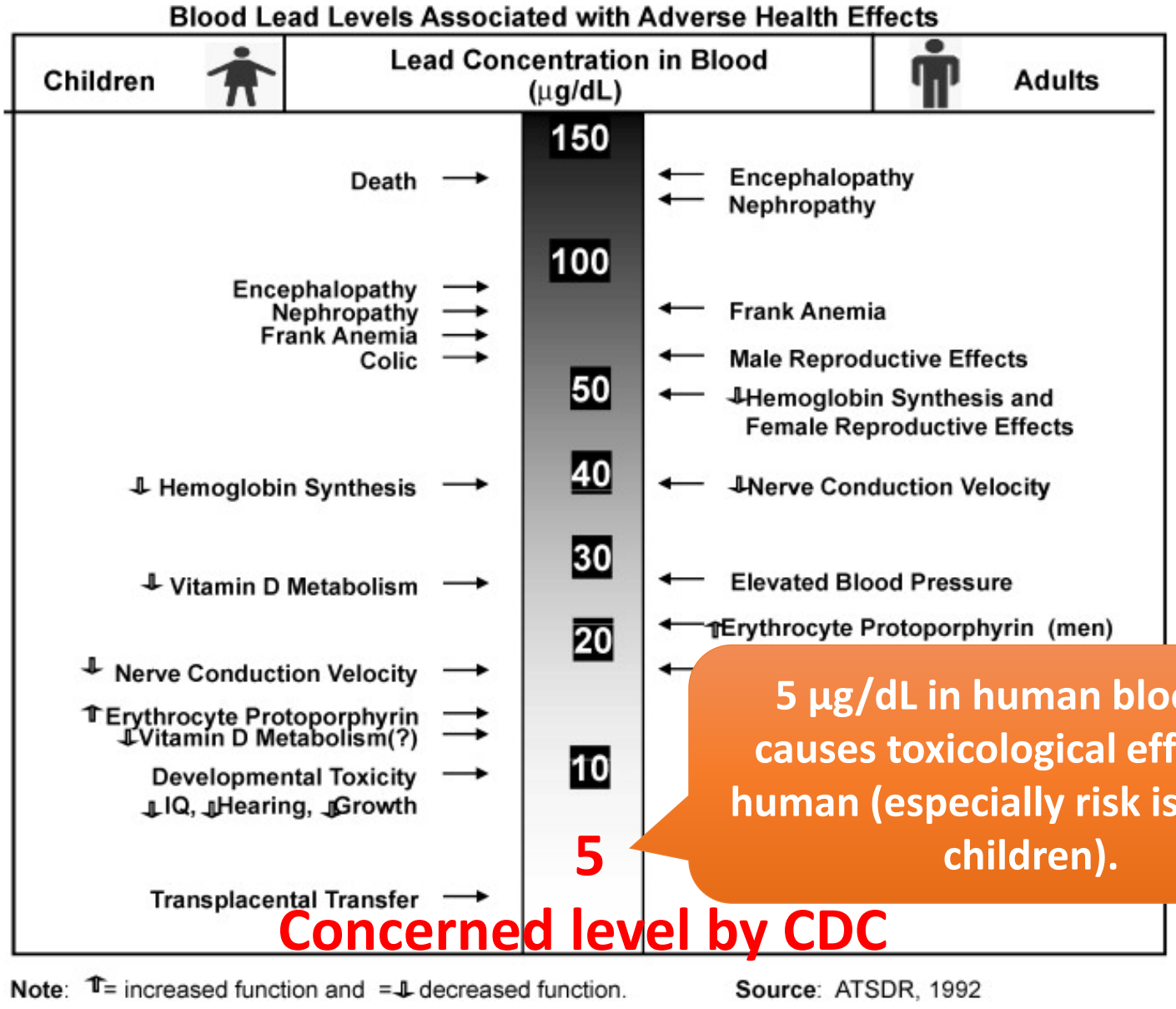
One of the Health Centres where we collected samples

Metal exposure through road dust ?

Some children are barefoot

Scrap metal scavenging at mine

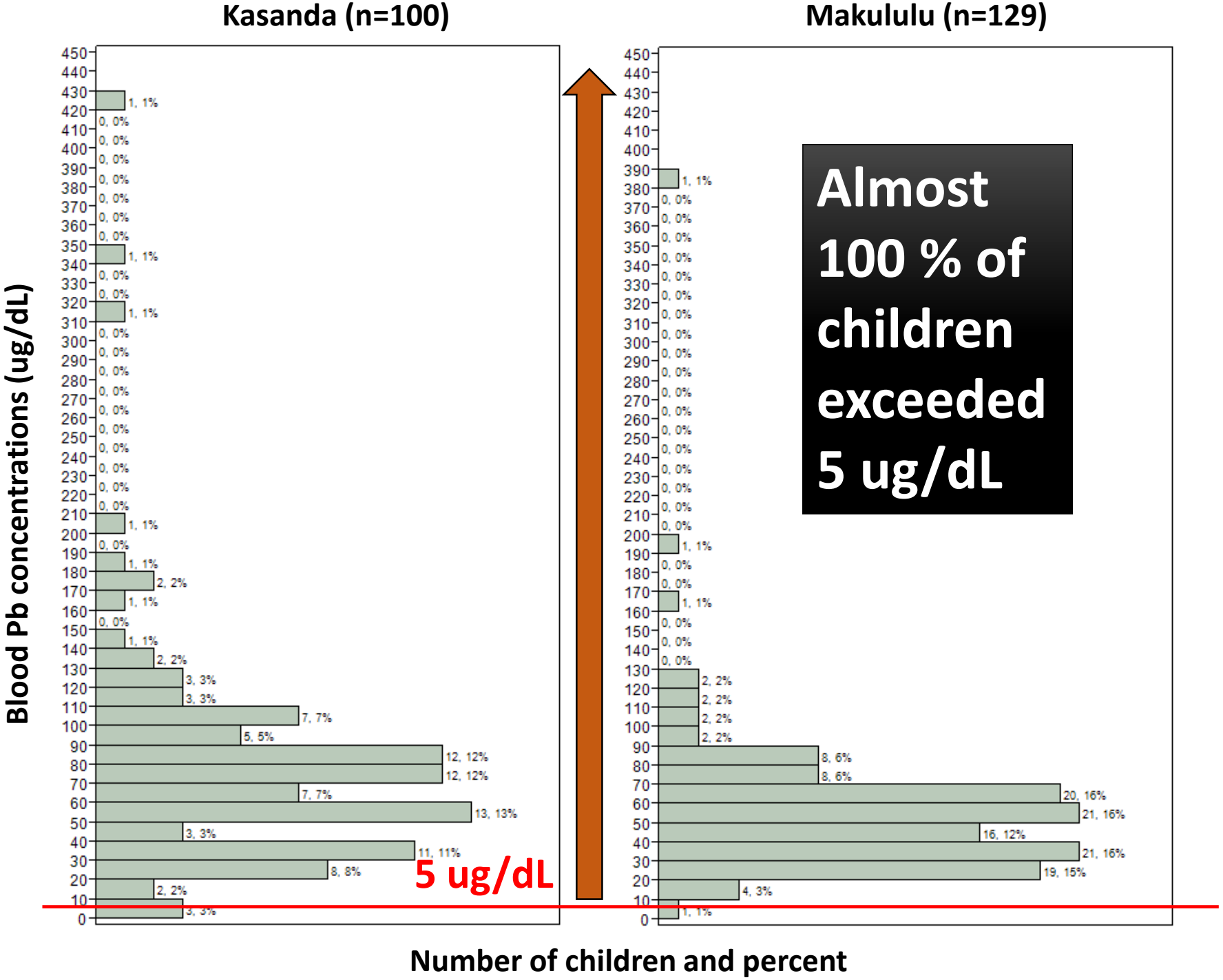
Blood Pb concentration in children in Kabwe



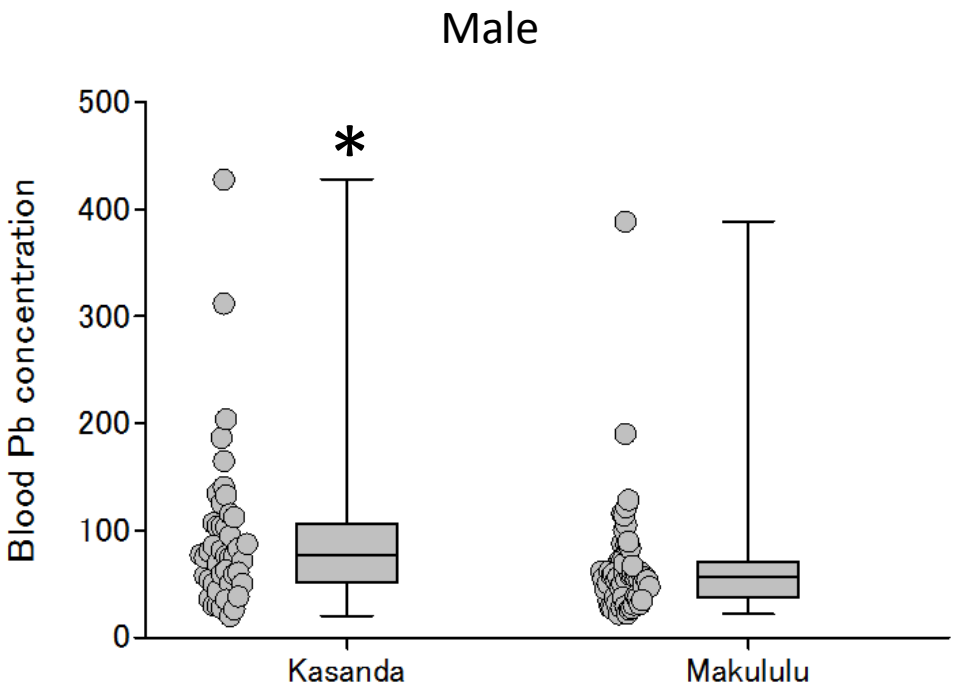
5 µg/dL in human blood, Pb causes toxicological effects on human (especially risk is high for children).

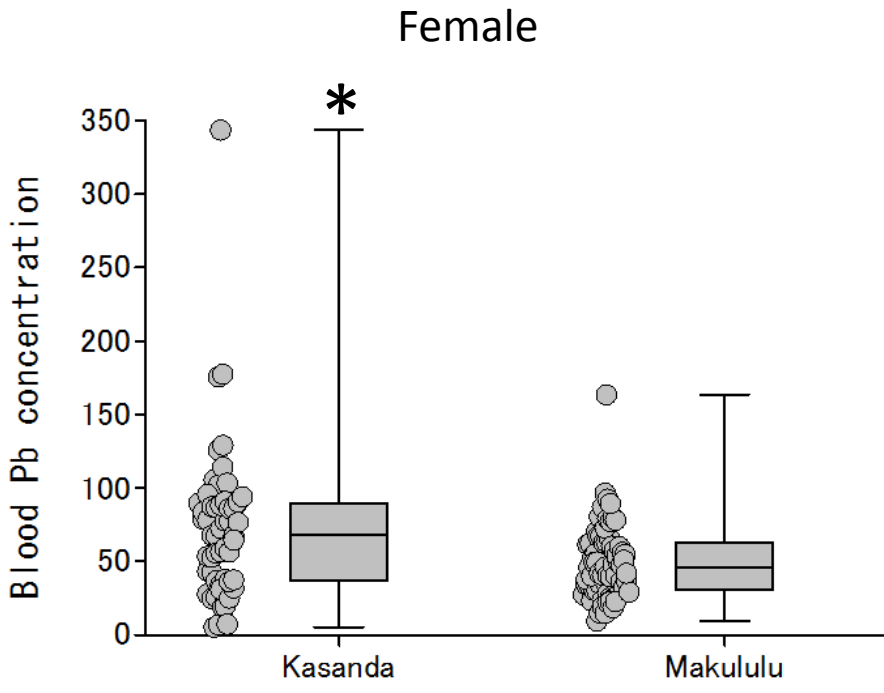
Concerned level by CDC

Fig. 1. Blood-lead levels associated with adverse health effects.


Kasanda (n=100)
Makululu (n=129)
Blood Pb concentrations (ug/dL)
Number of children and percent
5 ug/dL
Almost 100 % of children exceeded 5 ug/dL

BPb: Kasanda vs Makululu (Male and female analyzed separately)

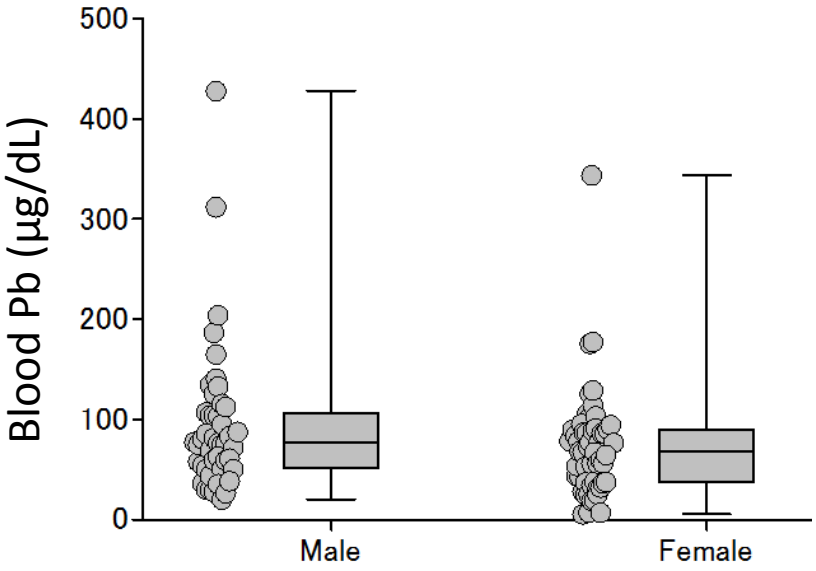




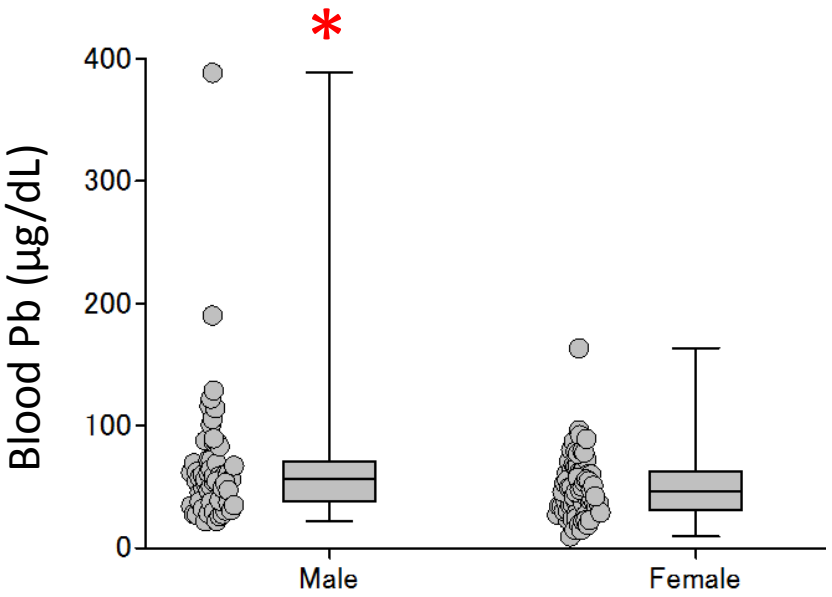
Kasanda was significantly higher in both male and female.

Sex difference (boys vs. girls)

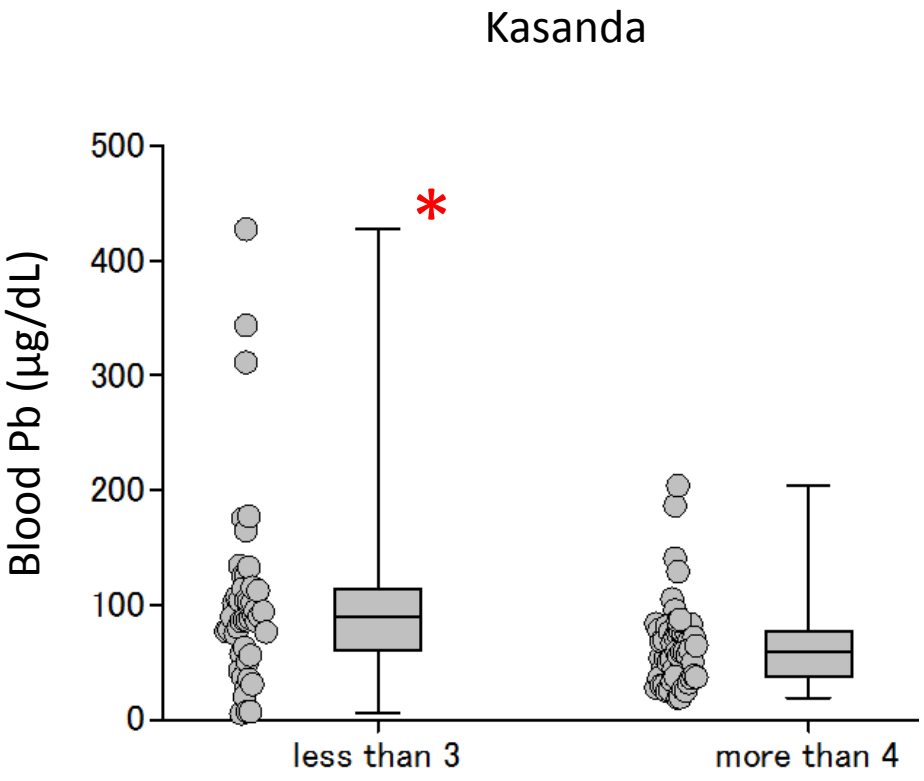
Kasanda
（No difference）

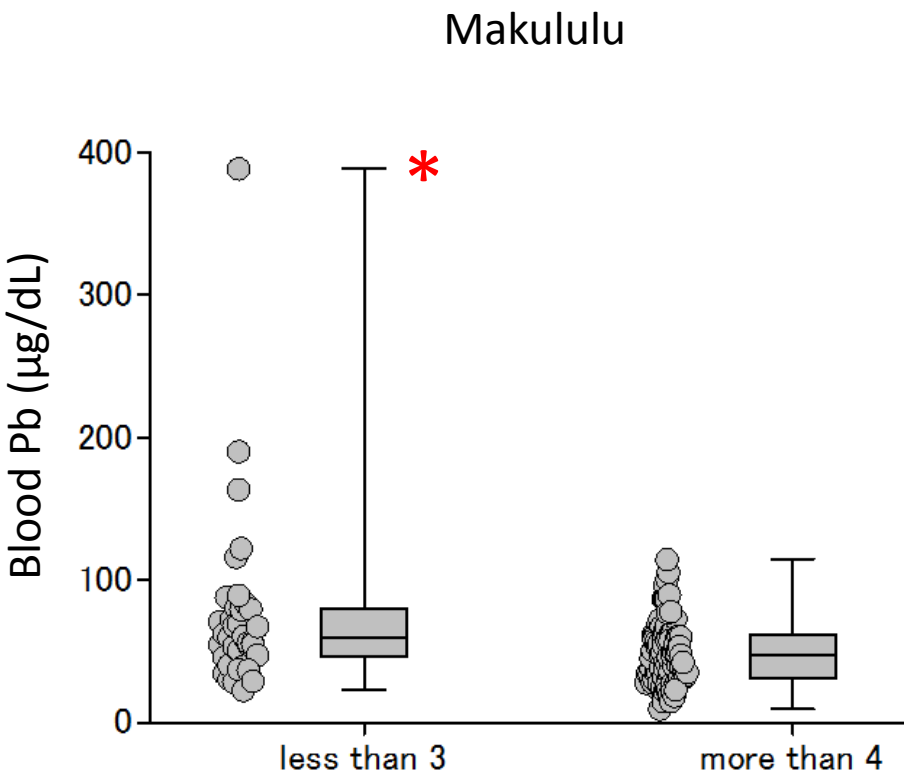


Makululu
（Male was high）

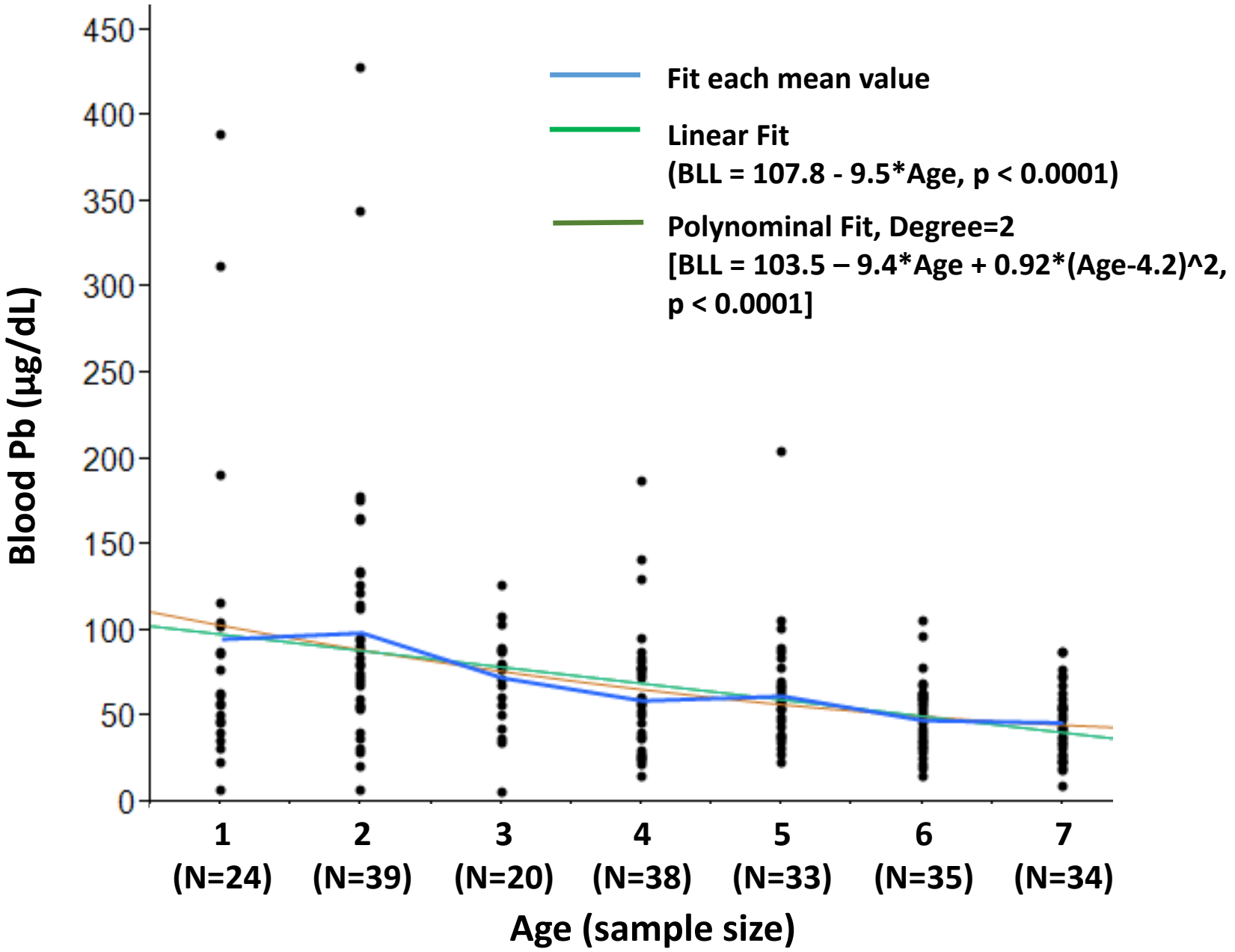


Age difference (0-3 years vs 4-7 years)

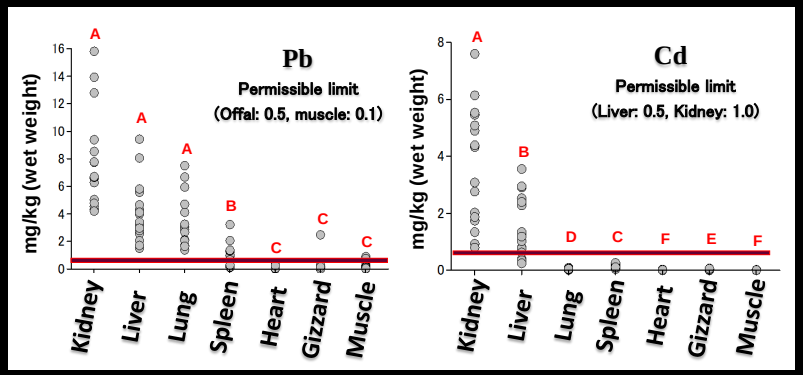




Correlation between Age & blood Pb

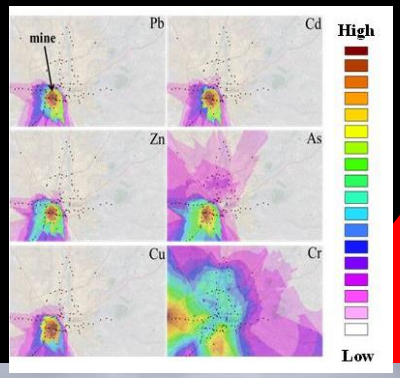


Summary of PAST study



Pb & Cd contamination in chicken

2009~2011



2008~2009

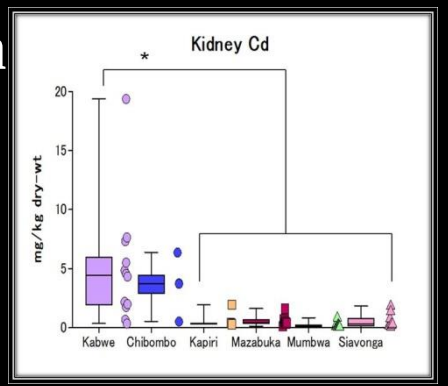
Soil contamination and diffusion

Kabwe Pb & Zn Mining

Pb pollution in children

2012~2015

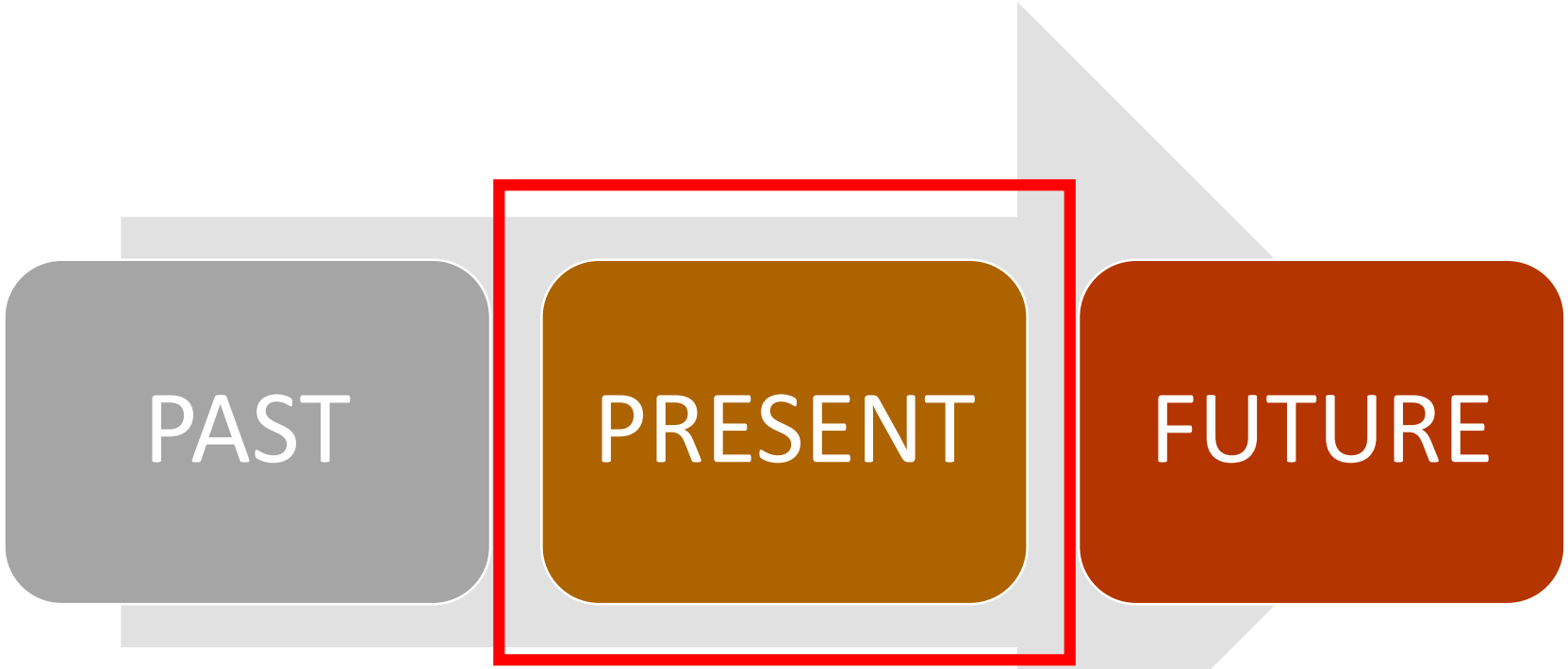
Pb & Cd contamination in cattle



Publications on PAST research

1. **Nakata H & Nakayama SMM** et al. Reliability of stable Pb isotopes to identify Pb sources and verifying biological fractionation of Pb isotopes in goats and chickens. Environ Pollut. 208(Pt B):395-403 (2016)
2. **Yabe J** et al. Lead poisoning in children from townships in the vicinity of a lead-zinc mine in Kabwe, Zambia. Chemosphere119:941-7 (2015)
3. M'kandawire E et al. Molecular characterization and mRNA expression of cytochrome P450 1A1 and cytochrome P450 3A in liver of Kafue Lechwe (Kobus leche kafuensis) as potential biomarkers of pollution of the Kafue River basin, Zambia. Engineering 6:51-58 (2014)
4. **Nakayama SMM**, et al. Accumulation and biological effects of metals in wild rats in mining areas of Zambia. Environmental Monitoring and Assessment 185:4907-4918 (2013)
5. **Yabe J** et al. Metal distribution in tissues of free-range chickens near a lead-zinc mine in Kabwe, Zambia. Environ Toxicol Chem. 2013 Jan;32(1):189-192.
6. **Nakayama SMM**, et al. GIS-based source estimation of Cu pollution in Lake Itezhi-tezhi and metal accumulation profiles in Oreochromis spp. from both field and laboratory studies. Arch Environ Contam Toxicol. 2013 Jan;64(1):119-129.
7. **Nakayama S.M.M**, et al. Metal contaminated soil from mining area caused metal accumulation and biological responses in rats. JJVR, 61:S77-S79 (2013)
8. **Yabe J** et al. Accumulation of metals in the liver and kidneys of cattle from agricultural areas in Lusaka, Zambia. J Vet Med Sci. 2012 Oct;74(10):1345-7.
9. M'kandawire E, et al. The nucleotide sequence of metallothioneins (MT) in liver of the Kafue lechwe (Kobus leche kafuensis) and their potential as biomarkers of heavy metal pollution of the Kafue River. Gene. 2012 Sep 15;506(2):310-6.
10. Ikenaka Y et al. Effects of environmental lead contamination on cattle in a lead/zinc mining area: changes in cattle immune systems on exposure to lead in vivo and in vitro. Environ Toxicol Chem. 2012 Oct;31(10):2300-5.
11. **Nakayama SMM,** et al. Metal and metalloid levels and bio-accumulation characteristics in soil, sediment, land plants and hippopotami (Hippopotamus amphibius L) from the South Luangwa National Park, Zambia. Ecotox Environ Saf. 2012 Jun;80:333-8.
12. **Yabe J** et al. Uptake of lead, cadmium, and other metals in the liver and kidneys of cattle near a lead-zinc mine in Kabwe, Zambia. Environ Toxicol Chem. 2011 Aug;30(8):1892-7.
13. **Nakayama SMM** et al. Metal and metalloid contamination in roadside soil and wild rats around a Pb-Zn mine in Kabwe, Zambia.Environmental Pollution 159 (2011) 175-18.
14. Ikenaka Y, et al. Heavy metal contamination of soil and sediment in Zambia. African Journal of Environmental Science and Technology 4 2010（11）:729-739.
15. **Yabe J,** et al. Current levels of heavy metal pollution in Africa. J Vet Med Sci. 2010 Oct;72(10):1257-63.
16. **Nakayama** , et al. Heavy metal accumulation in lake sediments, fish (Oreochromis niloticus and Serranochromis thumbergi) and crayfish (Cherax quadricarinatus) in Lake Itezhi-tezhi and Lake Kariba, Zambia. Arch Environ Contam Toxicol. 2010 Aug;59(2):291-300.

Research achievement



Research activities

JUNE 17th ~ JULY 7th 2016

Exposure Pathway ?

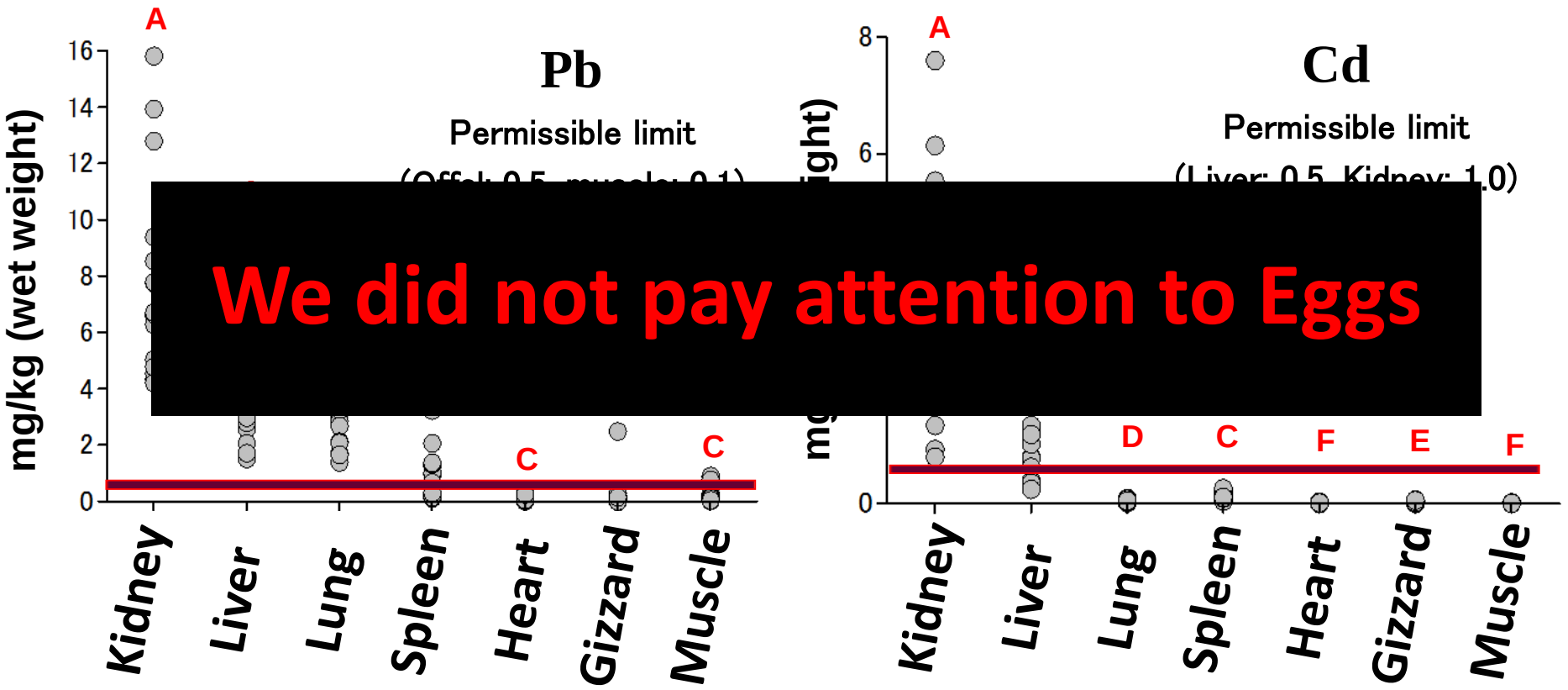
Blood Pb level exceeding the limit in 100% of Children

Soil contamination and diffusion

Water contamination

Plant/Vegetable contamination

High Pb and Cd levels in muscle and offal of Free-range chickens (n=17) in Kabwe



We did not pay attention to Eggs

(Tukey test)

Scavenging Free-range chicken

Exceeded the permissible limits

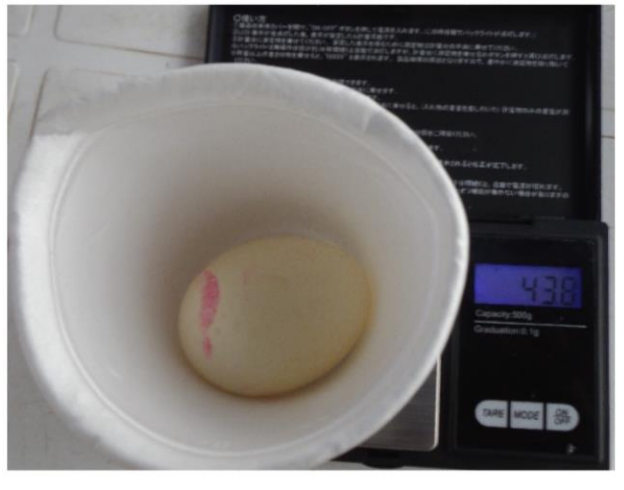
Eggs of Village Chicken

Commercial

Control site (Nakoli)

Mining area (Mutwe Wansofu)

Small



Dog Blood Sampling


• Share home environment
• Pb exposure ?
• Easy to collect

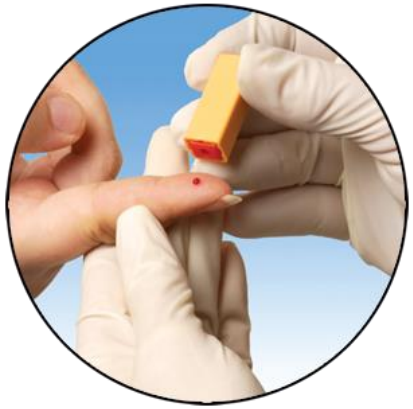
Home visit with children to get GPS coordinates of dog owners

Lecture of LEADCARE
by Dr. Jack (Pure Earth) in Kabwe

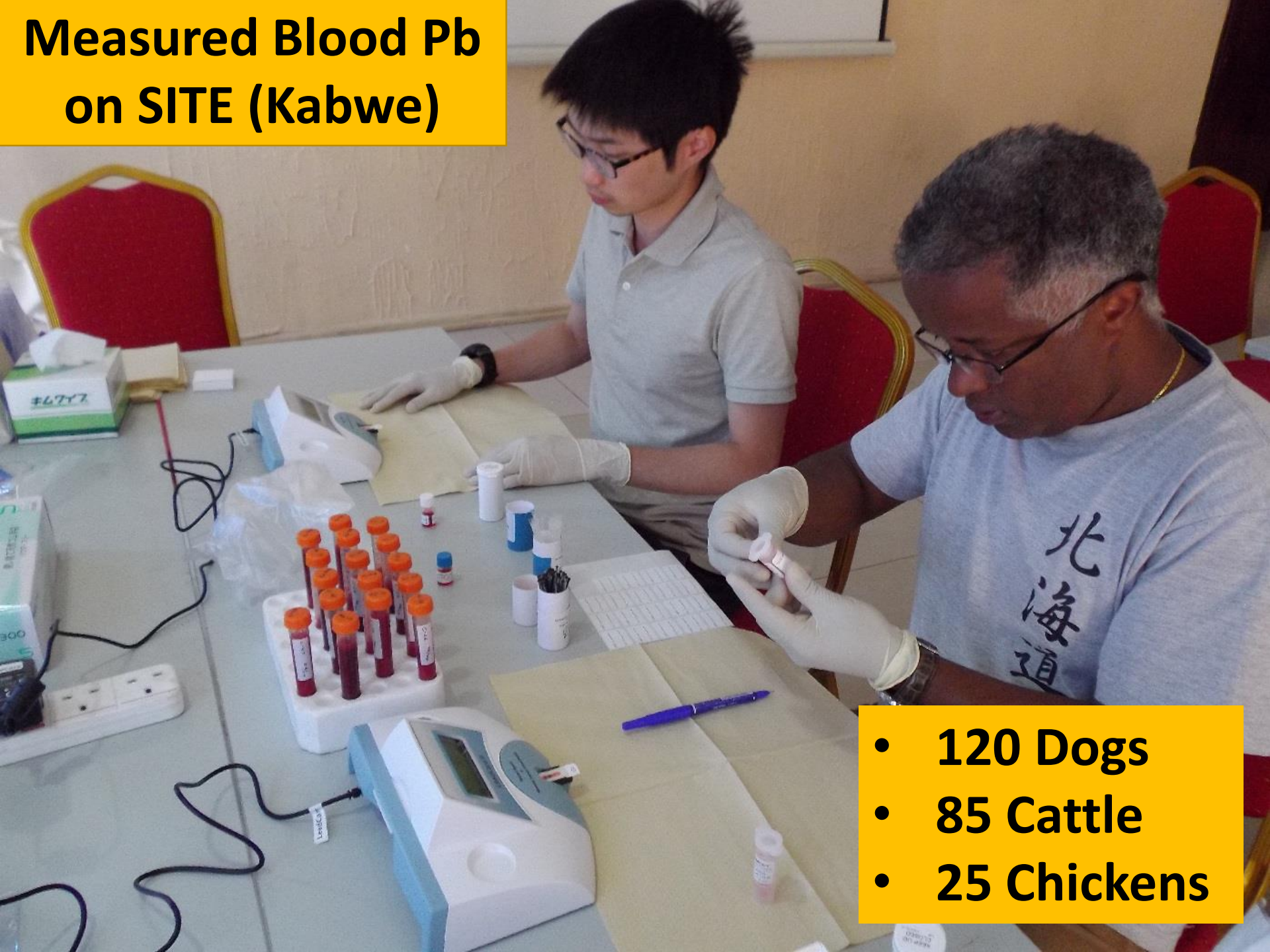
LeadCare® II

ONLY

- **1 Drop of Blood**
- **3 minutes**
- **Blood Pb level**
- **Less invasive**

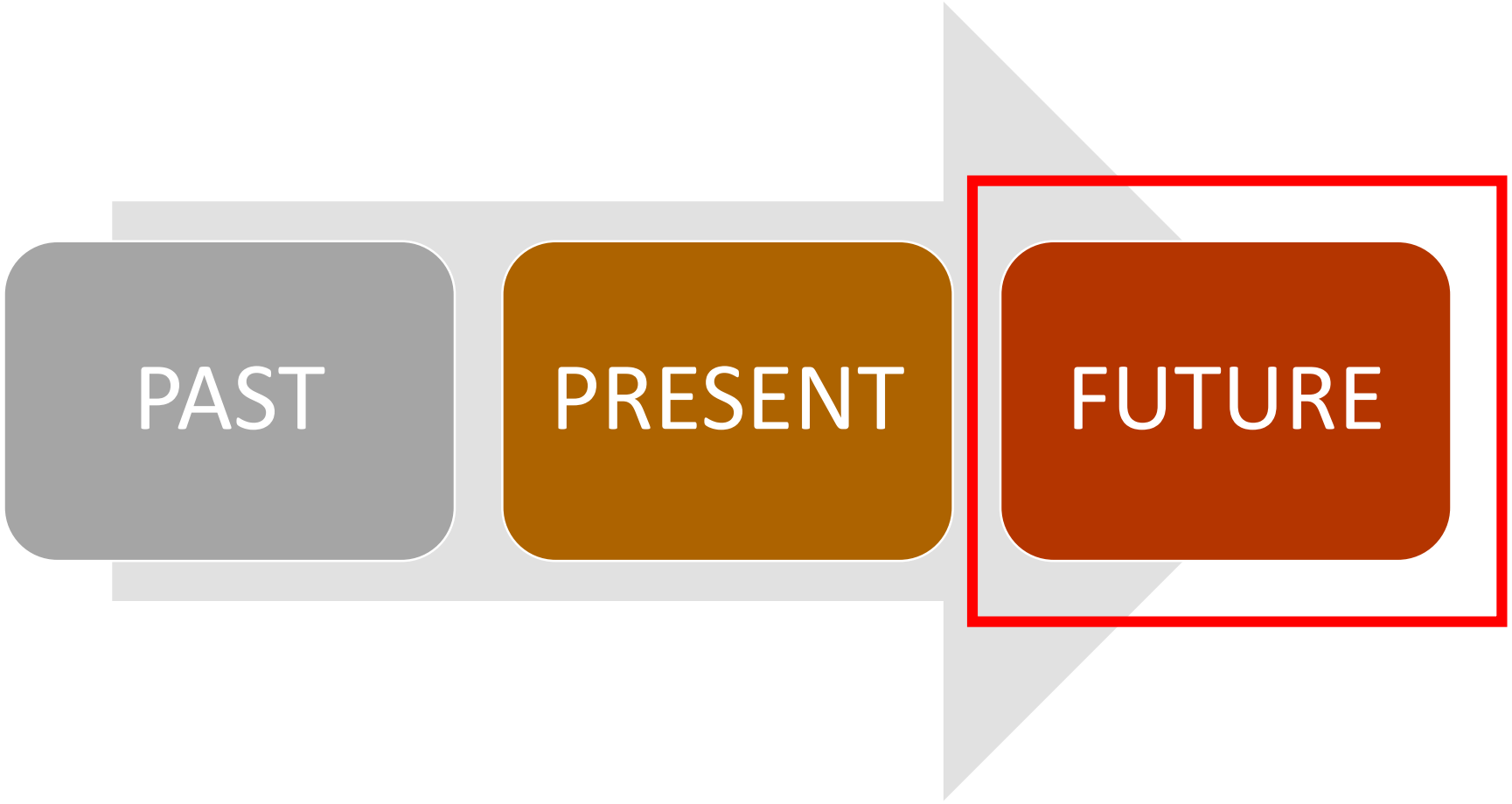


Measured Blood Pb on SITE (Kabwe)



- 120 Dogs
- 85 Cattle
- 25 Chickens

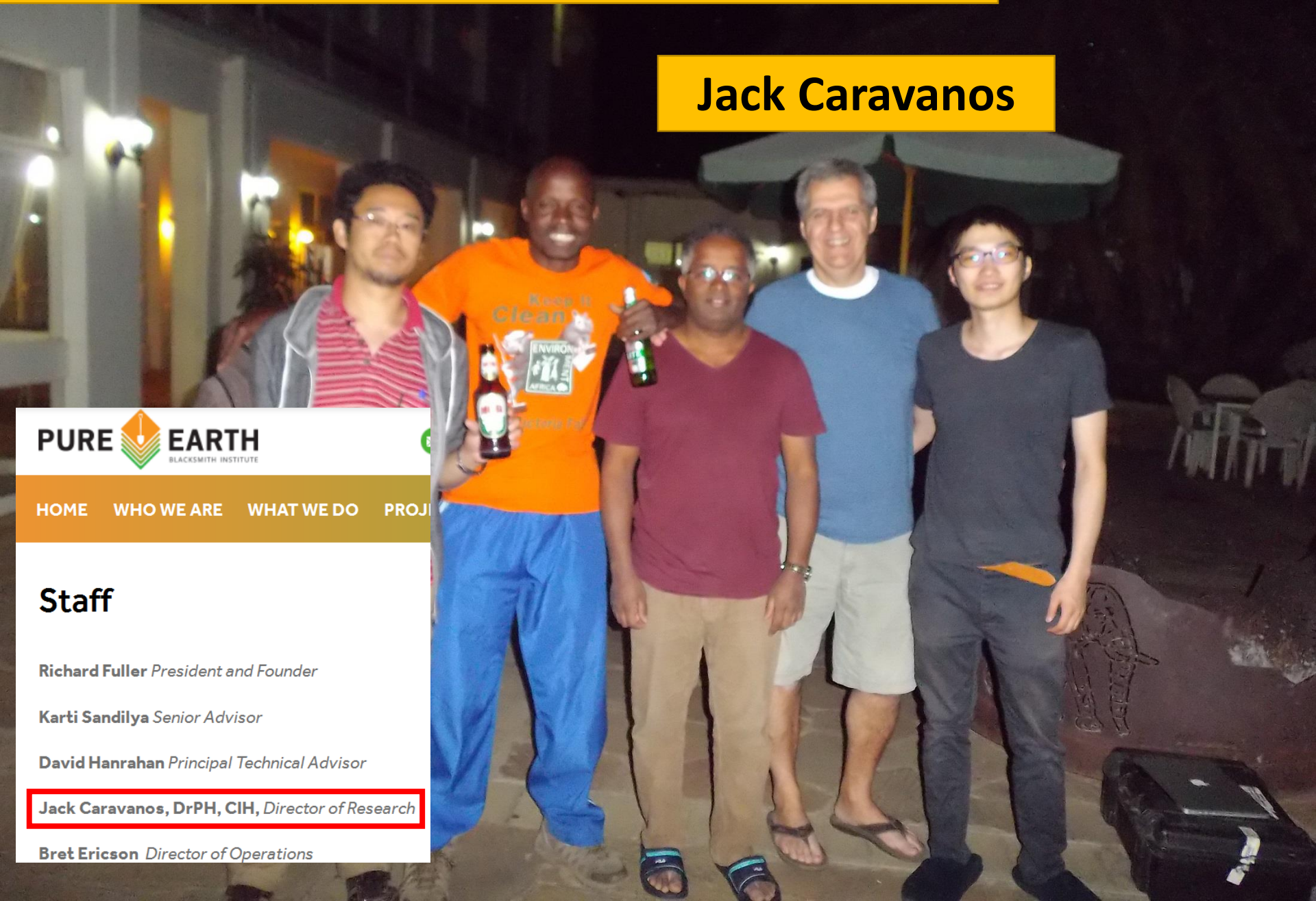
Research achievement



Co-Operation

Pure Earth ~Director of Research~

Jack Caravanos



PURE EARTH
BLACKSMITH INSTITUTE

HOME WHO WE ARE WHAT WE DO PROJ

Staff

Richard Fuller *President and Founder*

Karti Sandilya *Senior Advisor*

David Hanrahan *Principal Technical Advisor*

Jack Caravanos, DrPH, CIH, *Director of Research*

Bret Ericson *Director of Operations*

Pure Earth ~Chowa area Remediation~

KAMPAI PROJECT - OUTPUT 2

Objectives

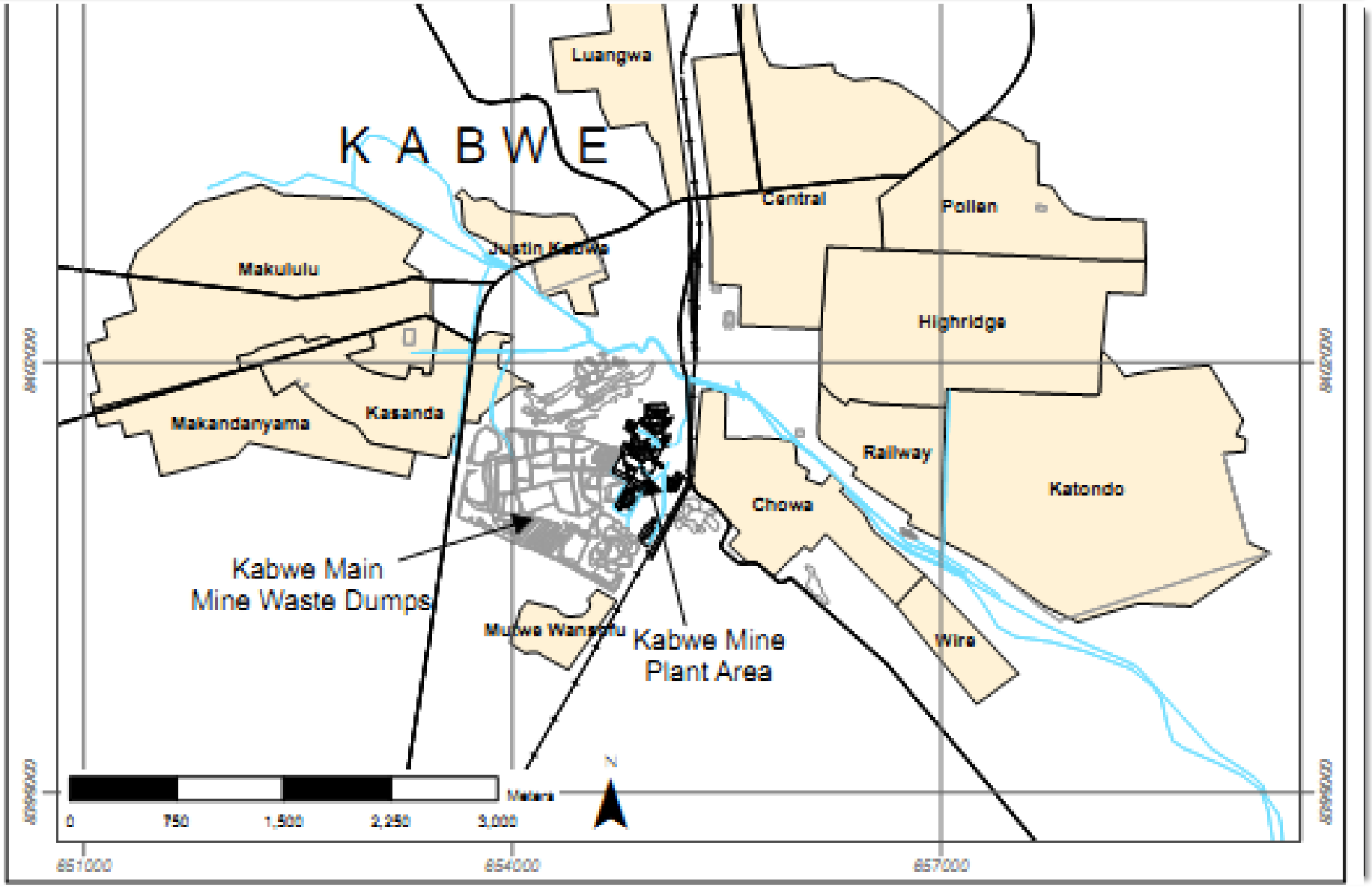
Main objective

- Assess lead (Pb) concentrations, stable Pb isotope ratios and levels of selected biomarkers as well as establish the health and socio-economical impact Pb contamination in Kabwe

Specific objectives

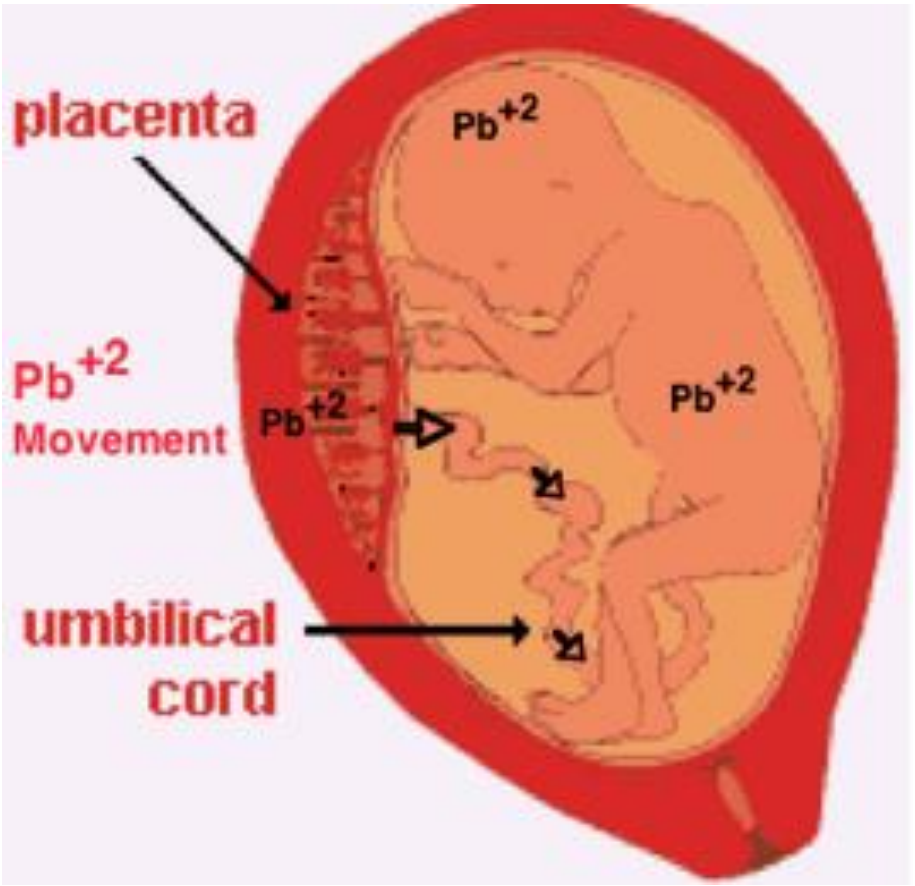
- Measure blood Pb concentrations in children and pregnant mothers in communities around the mine in Kabwe
- Measure Pb concentrations in breast milk
- Measure biomarkers to determine the effects of Pb exposure
- Neurodevelopment assessment of Pb exposure in children
- Assess the e

Study site


K A B W E
Luangwa
Central
Pollan
Justin Kabwe
Makululu
Highridge
Makandanyama
Kasanda
Railway
Chowa
Katondo
Kabwe Main
Mine Waste Dumps
Mudwe Wansofu
Kabwe Mine
Plant Area
Wire
N
Meters
0
750
1,500
2,250
3,000
651000
654000
657000

Blood Pb concentrations in children, maternal blood and breast milk

- Lead crosses the placenta
  - Maternal and umbilical cord blood Pb levels have a strong correlation
  - Blood Pb levels of the infant is similar to that of the mother
- Milk Pb levels
  - Pb levels in breast milk increase with level in maternal blood
  - Concentrations in breast milk indicates prenatal exposure
- Fetuses and infants are at the highest risk of Pb neurotoxicity



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Blood Pb measurements

Lead Care II

ICP-MS

- Blood lead analysis will be conducted annually
- Impact of remediation measures will be evaluated

Stable Pb isotope ratios and biomarkers

- Stable Pb isotope ratios
  - ICP-MS
  - To clarify source of exposure
- Blood biochemistry
  - COBAS Ready - blood chemical analyzer
  - To determine health effects
- Plasma/Urine biomarker
  - HPLC-UV
  - To determine markers of exposure and effect

Neurodevelopment Assessment

- Children under 3 years of age
  - Infants and their mothers will be followed up for a period of 48 months at birth and thereafter every year
  - The Ages and Stages Questionnaire (ASQ) will be used to measure the cognitive, motor, language and behaviour of infants from one to 42 months of age

Neurodevelopment Assessment: 3-18 years old

- Zambia Child Assessment Tool (ZamCAT)
  - For preschool children to assess language, cognition and fine motor skills
- Neuropsychological Assessment Tool (NEPSY)
  - For 3-16 year old to assess subtle central nervous system deficits (social, academic and behavioural difficulties)
- Universal Nonverbal Intelligence Test (UNIT)
  - For 5 to 17 year old to assess general intelligence using nonverbal means
- Kaufman's Assessment Battery for Children (KABC-II)
  - For 7 through 18 year old to assess cognitive and mental processes (Sequential Processing, Simultaneous processing, Learning and Planning Ability)

Economic impact assessment

- Economic assessment is a new area in this field
- The impact of lead in Kabwe will be “quantitatively” estimated to establish a model for new assessment system

Ethical Considerations

- The following specific standards will be upheld:
  - Ethical clearance will be sort from UNZABREC and MOH
  - Written informed consent will be obtained from parents
  - Minimum risk will be upheld in the drawing of biologic samples by the local medical personnel
  - Samples (cord blood, breast milk, blood, hair and urine) will be disposed off according to stipulated guidelines
  - The presence of the parent/caregiver during the neurodevelopmental testing will also be a requirement
  - Respondents will be assured of confidentiality of the results
  - Parents/guardians of affected children will be referred for further assessment and therapy for the child