Group 2 progress report

at General Meeting

on 7th Apr 2017

Today's Group2 Presentation Topics

- Group2 overall progress (Nakata)
- Exposure assessment team (Nakata)
- Economic assessment team (Hiwatari, Narita)
- Neurodevelopment (ND)/QOL analysis team (Fujita)

Group2 overall progress (2017 Jan~Mar)

- Meeting & Trip to Zambia (Feb~Mar 2017)
  - Exposure assess team (Nakayama, Yared, Andrew, Nakata)
  - QOL team (Fujita, Yoshida, Yamasaki)
  - Economic assess team (Hiwatari, Narita, Yamada)

- New member, Dr. Peter Hangoma (Sch. of Public Health) joined in Economic team

- Team structure was updated (next slide)

- Broad Survey in July~Aug 2017 was designed and arranged

- Monitoring sheet (1st term. May 2016~Jan 2017)

- Nakata will be transferred Zambia at end of Apr

➢ Team structure was updated

Team Leader: John Yabe / Shouta Nakayama

(A) Health Risk Assessment

Exposure Assessment

John Yabe (UNZA, Vet)
Kaampwe Muzandu (UNZA, Vet)
Andrew Kataba (UNZA/HU)
Tembo Backsion (UNZA)

Shouta Nakayama (HU, Vet)
Yared Beyene (HU, Vet)
Haruya Toyomaki (HU, Vet)
Hokuto Nakata (HU, Vet)
Takeshi Kuritani (HU, Sci)

Neurodevelopment / QOL analysis

Nosiku Sipilanyambe Munyinda (UNZA, Medical)
Doreen Sakala (MOH)
Nancy Zyongwe (UNZA, Medical)
Sandra Shanungu (Arise Africa)

Wakako Fujita (Wakayama Med)
Takeshi Saito (HU, HS)
Harukazu Tohyama (HU, HS)
Takahiko Yoshida (Asahikawa Med)
Shojiro Yamasaki (HU, HS)
Natsumi Nagai (HU, HS)

IQ analysis

(Robert Serphell (UNZA, Special Education))
Beatrice Matafwali (UNZA, Special Education)
Gabliel Walubita (UNZA, Special Education)
Joe Kalima (UNZA, Special Education)

(B) Economic Assessment

Bona Chitah (UNZA, Economics)
Chrispin Mphuka (UNZA, Economics)
Peter Hangoma (UNZA, Public Health)

Masato Hiwatari (HU, Economics)
Daiju Narita (HU, Economics)
Daichi Yamada (HU, Economics)

Statistician from Medical School Mr. Munbi Chola

- Sub-group化してメンバーを確認
- Sub-groupに日本・ザンビアのリーダー配置（IQチームはザンビアのみ）

➢ Broad Survey in July~Aug 2017 was designed and arranged

【2016年末までの決定事項（→2017年の進捗）】

1. ①2017年7~8月頃に、チーム2のサブグループが同時にKabweの広範囲を対象とした大規模調査（②汚染域のDeep Surveyは後で）
→日程の大枠は確定

2. UNZA医学部のエコノミストPeter Hangoma氏をメンバーに追加（医学・経済がある程度両方分かる方）
→正式に参画

3. 12月末に、サブグループごとに、調査アウトライン（PROPOSAL）を作成し、日本・ザンビア側のTV会議
→大枠は決定。詳細をザンビア側と検討中。

4. 2017年1月にSTATISTIANとディスカッションして、サンプルサイズ（N数）の決定
→決定

5. 経済班の統計データ等をもとに、ランダムサンプリングの対象を決める。
→データは収集済み。対象選定中。

【Decision by end of 2016 (→Progress in 2017 so far)】

1. ①Broad Survey in Whole Kabwe by (all) subgroup in Group2 & ②Deep survey around mines (Hot spots) in the future.
→Approximate schedule for ① was decided.

2. Dr. Peter Hangoma (Health economist) will join in economic group.
→Officially joined.

3. Research (survey) protocol will be created and discussed between Z/J side.
→The outline was decided. Further details are still under discussion.

4. Sample number will be decided by Jan 2017.
→Decided.

5. Sampling target will be decided according to economic statistical data.
→Still under discussion.

➢ Broad Survey in July~Aug 2017 was designed and arranged (cont'd)

6.「1回の調査のみなら1000世帯」、「世銀などによる工事前後のデータ比較ができるなら500世帯程度必要」
→世銀工事に依存せず、①で1000世帯を対象とする。

7. Health assessの対象は、各世帯で「父or/and母、子供1~2人」で2000~4000名？
→父、母、子供2人（7歳以上と未満）として、計4000名を対象

8. 父親の骨中の鉛濃度測定を行い、長期暴露の指標として経済アセスに用いる（骨中濃度測定が不可な場合は、BLLで代用）。
→X-ray式骨中濃度測定器は見積もりなど依頼中。骨中濃度が測定できた場合でも、BLLも測定する。

【今後の予定】
◆4月～7月に、3月のPre調査の問題点など修正
◆7月～8月頃に第1回大規模調査
◆10月以降、調査データのまとめ、サンプルを用いた実験データ収集・解析

6. If we do ① before/after the WB remediation, 500 households is enough for the economic assess. But if one time, 1000 households is needed.

→1000 households will be target for ①.

7. Target for health assess is "father or/and mother, 1~2 child" in each household = 2000~4000 specimens?
→Father, mother, and two children (above/less than 7 years old) = 4000 specimens.

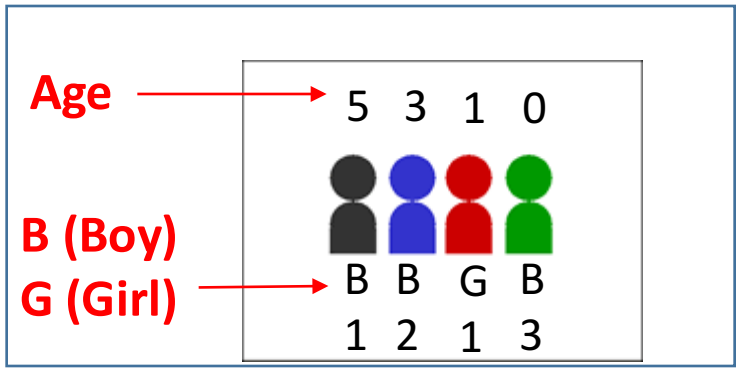
8. Bone Pb level in father can be a long-time (life-time) exposure/accumulation.
→ Purchase of X-ray bone Pb analyzer is still on the way. Both BLL and bone level data should be gotten.

【Future Plan】
◆ Further detail plan for ① will be discussed and finalized in Apr~Jul
◆ ①Broad Survey in Jul~Aug
◆ Lab experiment and data analysis after Oct

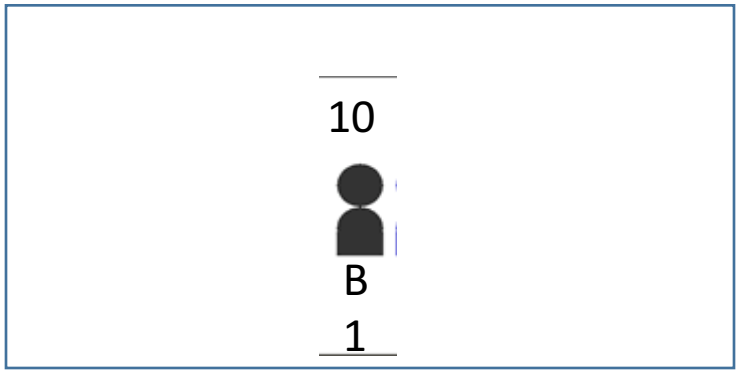
The challenging in previous meeting

1000世帯の中でも、子供の人数や男女比が家庭ごとに異なる
Among 1000 households, child numbers & boys/girl ratio is different in each house.

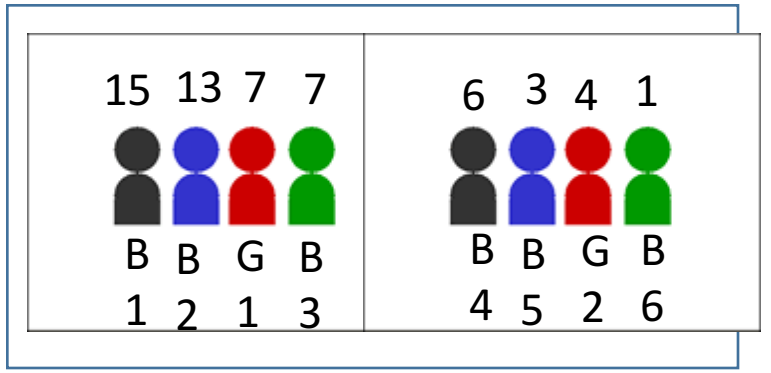
Ex. House No. 1 (3 Boys and 1 Girl)



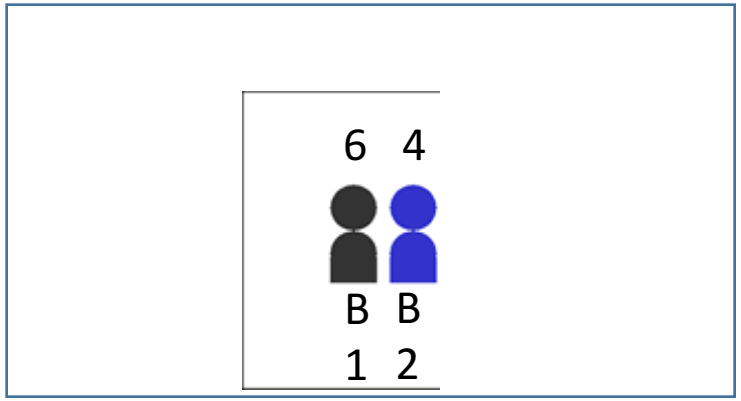
House No. 2 (1 Boy)



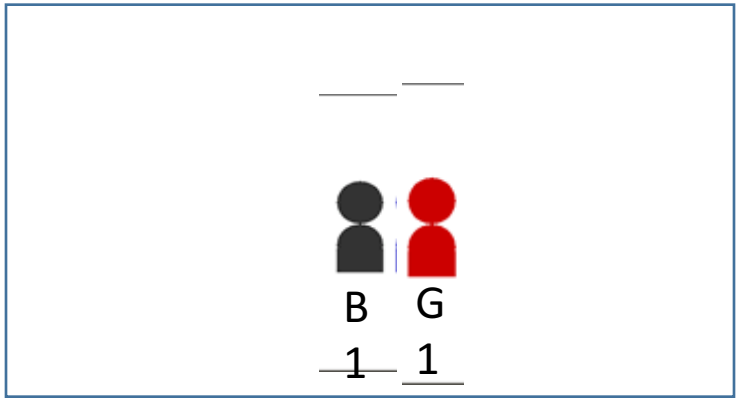
House No. 3 (6 Boys and 2 Girls)



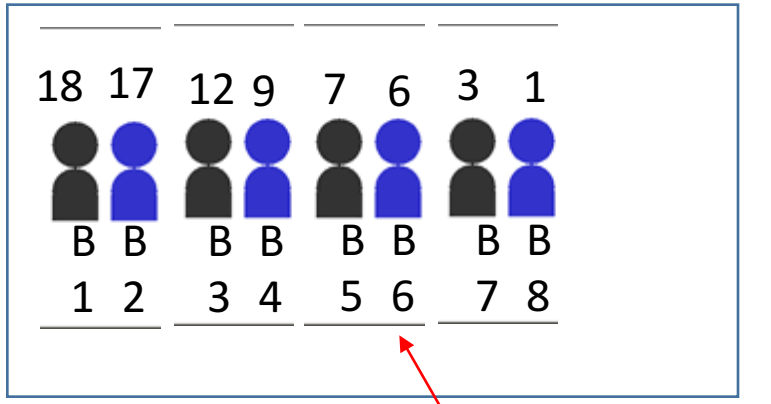
House No. 998 (2 Boys)



House No. 999 (1 Boy and 1 Girl)



House No. 1000 (8 Boys)



仮に経済チームがランダムに1000世帯を選んだとして、その中から、どのように血液サンプリングの対象の子供をバランスよく選ぶのか？ After Economics team randomly selected 1000 household, how can we choose target children for blood collection?

Which child should be targeted???

Today's Group2 Presentation Topics

- Group2 overall progress (Nakata)
- Exposure assessment team (Nakata)
- Economic assessment team (Hiwatari, Narita)
- Neurodevelopment (ND)/QOL analysis team (Fujita)

Sample number, team arrangement, rough schedule

- Assuming 10~20 min for 1 sample collection (from past experience in Oct 2016 pre-sampling)
- Average 10 households (40 persons/samples) per day
- 17th July ~ 21st July: Preparation and practice
- 24th July ~ 30th Aug: Sample collection (N=4000) with 28 working days and 5 Saturdays (half day, optional)
- Four teams (Each team has one Zambian, one Hokudai member, two nurses, and two lab technician)
  →（Yabe, Muzandu, Andrew, Zyambo）×（Nakayama, Yared, Toyomaki, Nakata）

40 samples × 4 teams × 30 days = 4800 samples

- Questionnaire (2 person)
- Blood collection (2 person)
- Blood Dispense and Centrifuge (1 person)
- LeadCareII BLL measurement (1 person)

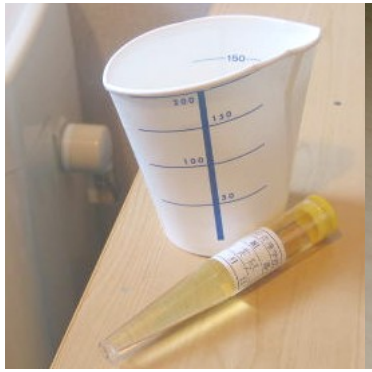
- 8 Nurses & Lab Technicians (2 and 2 in each team) based in Kabwe
- Community Health Worker (CHW) in Kabwe

← will arrange by Yabe, Muzandu, Nakata, Odagiri

Proposal (Exposure assessment sub-team)

Sampling Target

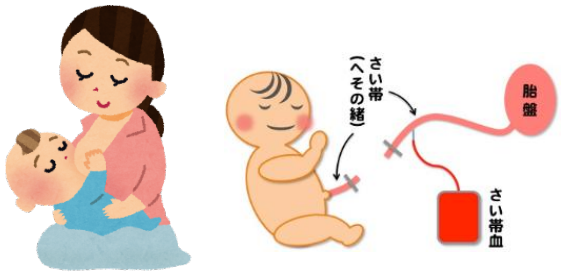
- Blood with heparin (at health center)
  - 5 mL for father, mother, and child above 7 years
  - 2.5 mL for child less than 7 years
- Urine (up to 10 mL) using plastic cup and 15 mL tube
- Feces (2~3 g) using stool container



Plastic cup and 15 mL tube

Stool container with spoon cap

- Breast Milk (1~3 mL for early stage, 5~10 mL for later lactation stage)
- Umbilical cord blood (5~10 mL?, with Syringe)
- Bone Pb (X-ray, still under arrangement)



Proposal – blood sample

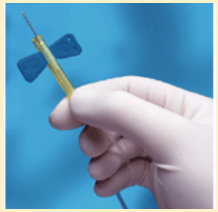
Each data is shared with other sub-groups.
* Father - with Economic (Bone Pb is more appropriate), ND, IQ team
* Mother - with QOL, ND, IQ team
* Child - with ND, IQ team

Father & Mother

【Blood collection】

5 mL with heparin

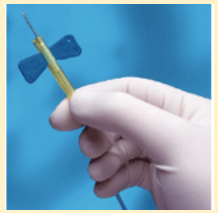
Child above 5 years



Transfer

5 mL with heparin

Child less than 5 years



Transfer

2.5 mL with heparin

Transfer: 2 mL × 2 tubes
(in 2 mL tube)

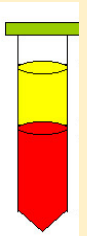
【Pb conc. & Isotope analysis】

LeadCare II

【BLL measurement】
(with only 50 uL)

1 mL of
remaining blood

Centrifuge
(4000 rpm, 10 min)



Plasma Transfer
0.5 mL × 1 tube
(in 1.5 mL tube)

【Blood biochemistry】

Transfer: 2 mL × 1 tubes
(in 2 mL tube)

【Pb conc. & Isotope analysis】

LeadCare II

【BLL measurement】
(with only 50 uL)

3 of LeadCare® II will be purchased until August sampling

Points to be discussed/decided

- How to decide the target area/household (by economic assess team)
- How to collect Paired Blood of mother and child and Milk, umbilical cord blood → Need to discuss with ND team (Nosiku-san)
- Where do we measure bone Pb? (at clinic in parallel with blood collection?) How many min per sample?
  → Arrange by Yabe & Dr. Linda, 3-min for one measurement
- How is overall flow for sampling? How many types and what timing are questionnaires taken?

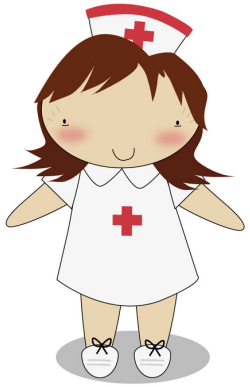
- How to call people to come to the clinic for blood collection?
  → Invitation letter & Reminder by CHW
- Need to arrange transport for participant from their house to clinic for sampling?
- How to employ our own nurse & lab tech for the sampling?
  → Arrange by Yabe, Muzandu, Nakata, Odagiri
- Where collected samples will be stored in Kabwe? Freezer in Kabwe will be necessary.
  → Freezer will be installed in somewhere Kabwe? How often do we need to transfer the samples from the Kabwe freezer to UNZA?

Rough schedule

- Selection of target households (By Economics team, statistical excel data)
- Decide a schedule of sampling for each household & make "Schedule Table"
- Make "Invitation letter" for each household with note of sampling day
- Deliver "Invitation letter" in advance by CHW
  (Reminde the day before sampling)
- Employ 8 Lab Techs & 8 Nurses & CHW
- Prepare consumables & Test kits for LeadCare analyzer (BLL tests)

Future Arrangement

- Overall matter → Nakayama & Yabe
- Bone Pb XRF → Yabe & Dr. Linda
- Nurse, Lab Tech, CHW employment → Nakata, Yabe, Odagiri, Muzandu
- Pb isotope ratio → Nakata, Dr. Kuritani (from Faculty of Science), Haruya
- Biomarkers, etc → Yared, Andrew



Schedule Table

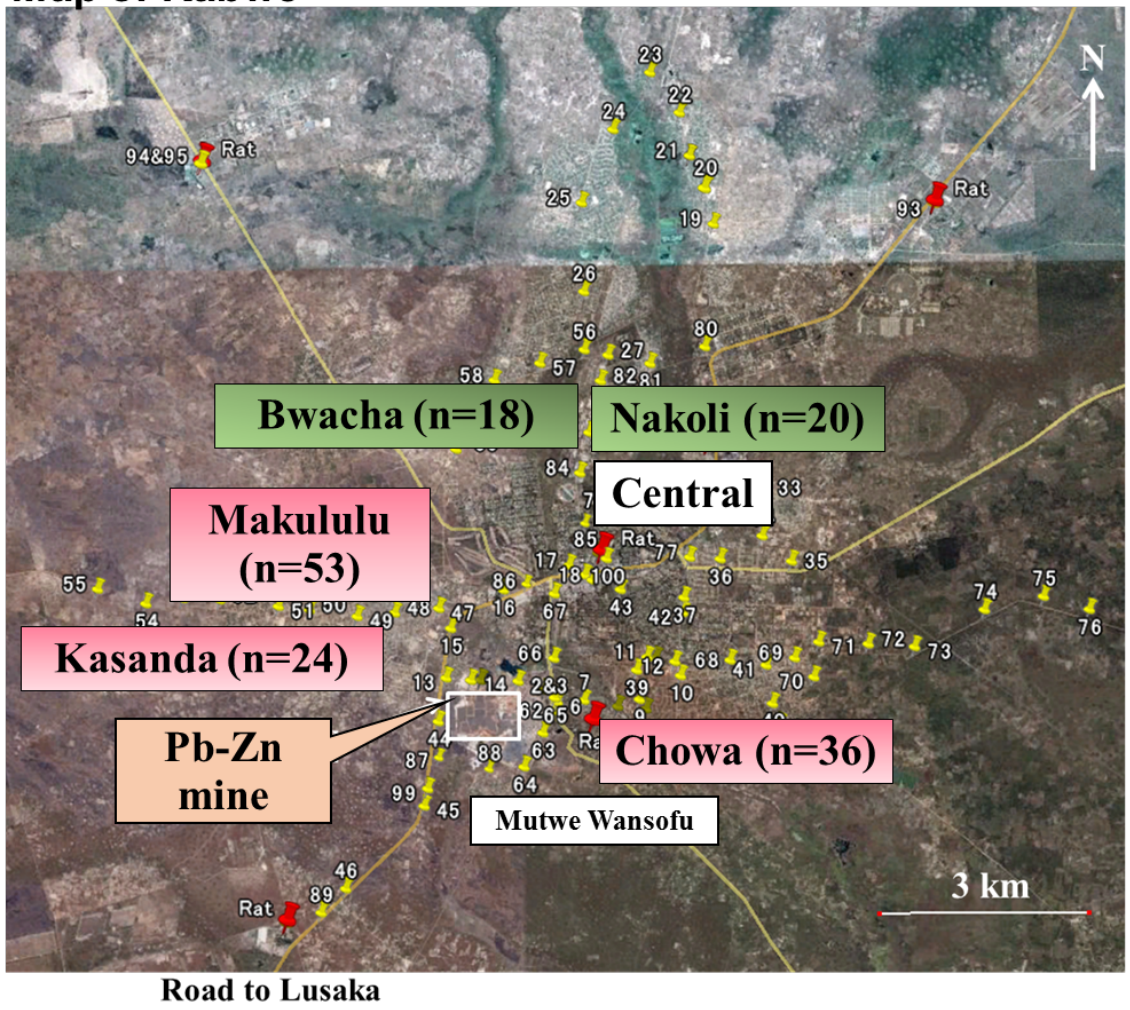
Note;
This table is just an Image.

Date
House hold number
Target
Code ID
Name
Group number
Health Center

Date	Health center	Household number	Code	Name	Team leader
2017/Aug/01	Chowa	CHOWA_#1			Yabe
2017/Aug/01	Chowa	#1			Yabe
2017/Aug/01	Chowa	#1			Yabe
2017/Aug/01	Chowa	#1			Yabe
2017/Aug/01	Chowa	#2			Yabe
2017/Aug/01	Chowa	#2			Yabe
2017/Aug/01	Chowa	#2			Yabe
2017/Aug/01	Chowa	#3			Yabe
2017/Aug/01	Chowa	#3			Yabe
2017/Aug/01	Chowa	#3			Yabe
2017/Aug/01	Chowa	#3			Yabe
2017/Aug/01	Chowa	#3			Yabe
Date	Health center	Household number	Code	Name	Team leader
2017/Aug/01	Kasanda	Kasanda_#1			Nakata
2017/Aug/01	Kasanda	#1			Nakata
2017/Aug/01	Kasanda	#1			Nakata
2017/Aug/01	Kasanda	#1			Nakata
2017/Aug/01	Kasanda	#2			Nakata
2017/Aug/01	Kasanda	#2			Nakata
2017/Aug/01	Kasanda	#2			Nakata
2017/Aug/01	Kasanda	#3			Nakata
2017/Aug/01	Kasanda	#3			Nakata
2017/Aug/01	Kasanda	#3			Nakata
2017/Aug/01	Kasanda	#3			Nakata
2017/Aug/01	Kasanda	#3			Nakata
Date	Health center	Household number	Code	Name	Team leader
2017/Aug/01	Makululu	Makululu_#1			Yared
2017/Aug/01	Makululu	#1			Yared
2017/Aug/01	Makululu	#1			Yared
2017/Aug/01	Makululu	#1			Yared
2017/Aug/01	Makululu	#2			Yared
2017/Aug/01	Makululu	#2			Yared
2017/Aug/01	Makululu	#2			Yared
2017/Aug/01	Makululu	#3			Yared
2017/Aug/01	Makululu	#3			Yared
2017/Aug/01	Makululu	#3			Yared
2017/Aug/01	Makululu	#3			Yared
2017/Aug/01	Makululu	#3			Yared

子供１５０名を対象とした血中鉛濃度調査（２０１６年１０～１１月）
Blood Pb in 150 children (Oct 2016)

Map of Kabwe



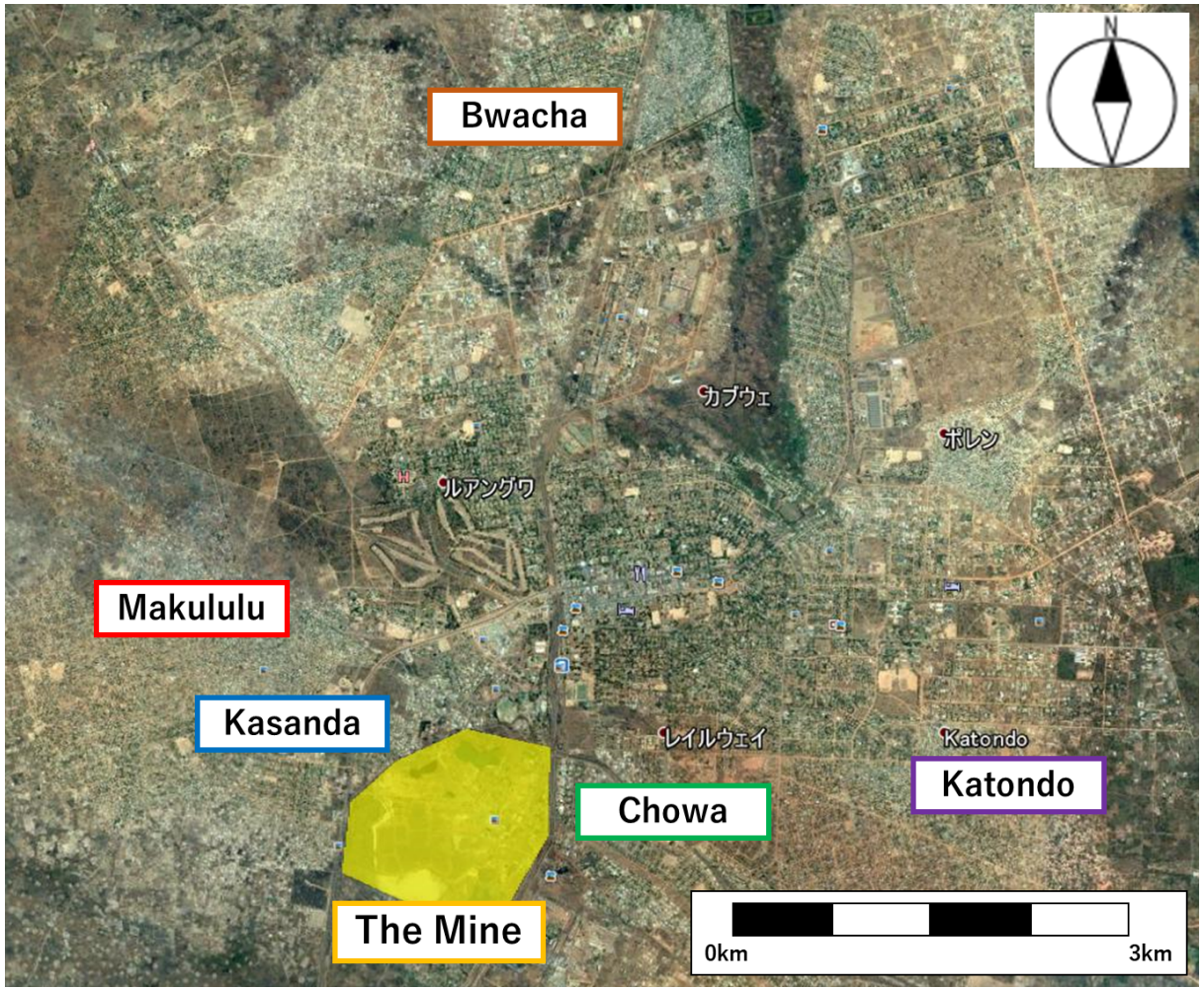
鉱床の近く(Makululu, Kasanda, Chowa)では、ほぼ100%の子供が10 µg/dLを超える鉛濃度を示した。

一方、鉱床から離れたエリア（Bwacha, Nakoli）では、上記エリアよりも濃度が低いが、鉛に曝露されていることが分かった。

→　血中鉛濃度Mappingが必要

データ非公開

Human sampling in Jan-Mar of 2017



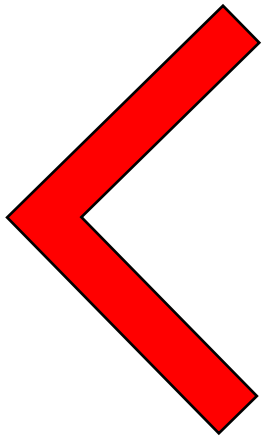
- Mothers and Infants at 5 clinics
- Blood samples of mothers and infants
- Breastmilk samples
- Fecal samples of infants

Blood Lead Levels (BLLs) in Mothers and Infants

Mothers (N = 426)

Infants (N = 417)

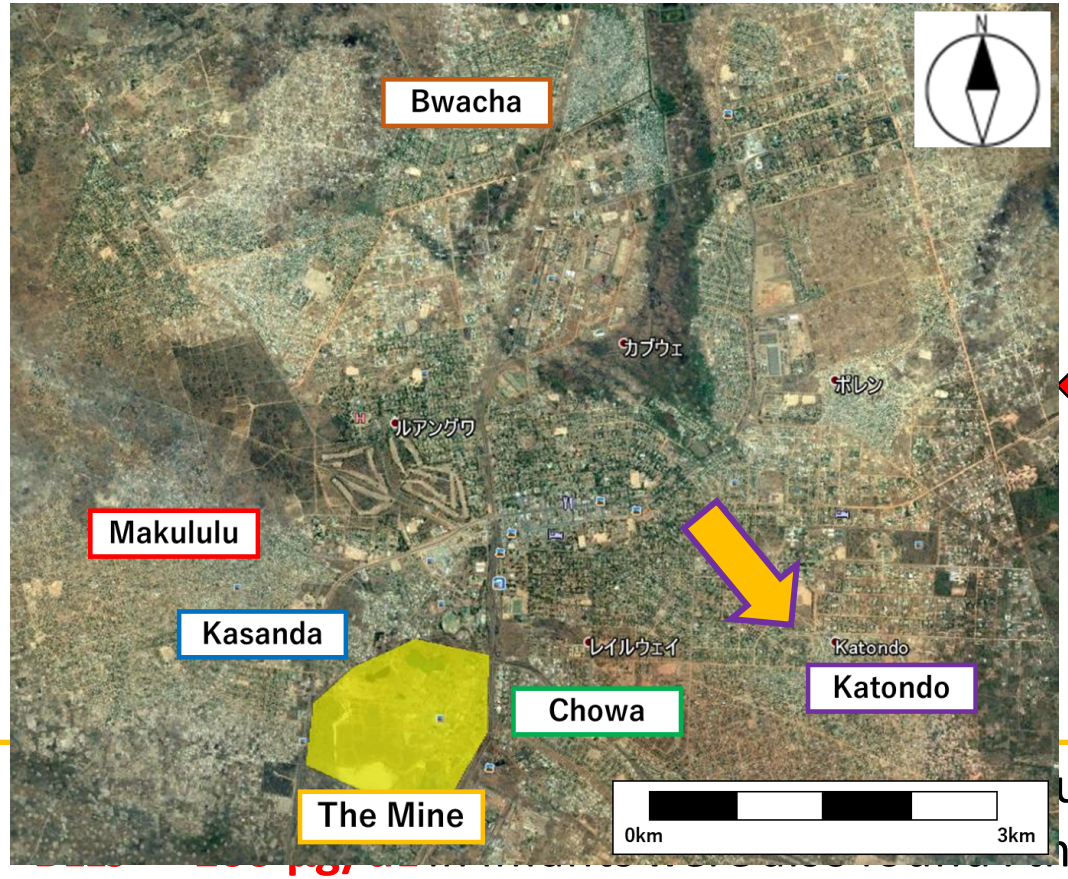
データ非公開



データ非公開

- **BLLs > 45 µg/dL** in mothers and infants were found: this level should be treated with **Chelation Therapy**
- **BLLs > 100 µg/dL** in infants were also found : this level can cause **encephalopathy and deaths in children**
  → **No clinical case has been reported in Kabwe**
- BLLs in infants were **significantly higher** than those in mothers ($p < 0.01$)

Blood Lead Levels (BLLs) in Mothers and Infants



Infants (N = 417)

データ非公開

- [redacted]und: this level should be treated with **Chelation Therapy**
- [redacted]his level can cause **encephalopathy and deaths in children**

  → **No clinical case has been reported in Kabwe**

- BLLs in infants were **significantly higher** than those in mothers ($p < 0.01$)
- The first time to collect samples in Katondo

  → **BLLs in Katondo were similar to Chowa**

Correlation between BLLs of paired mothers and infants

データ非公開

There was a **significant positive correlation** between BLLs of paired mothers and infants ($p < 0.01$, $\rho = 0.6$)

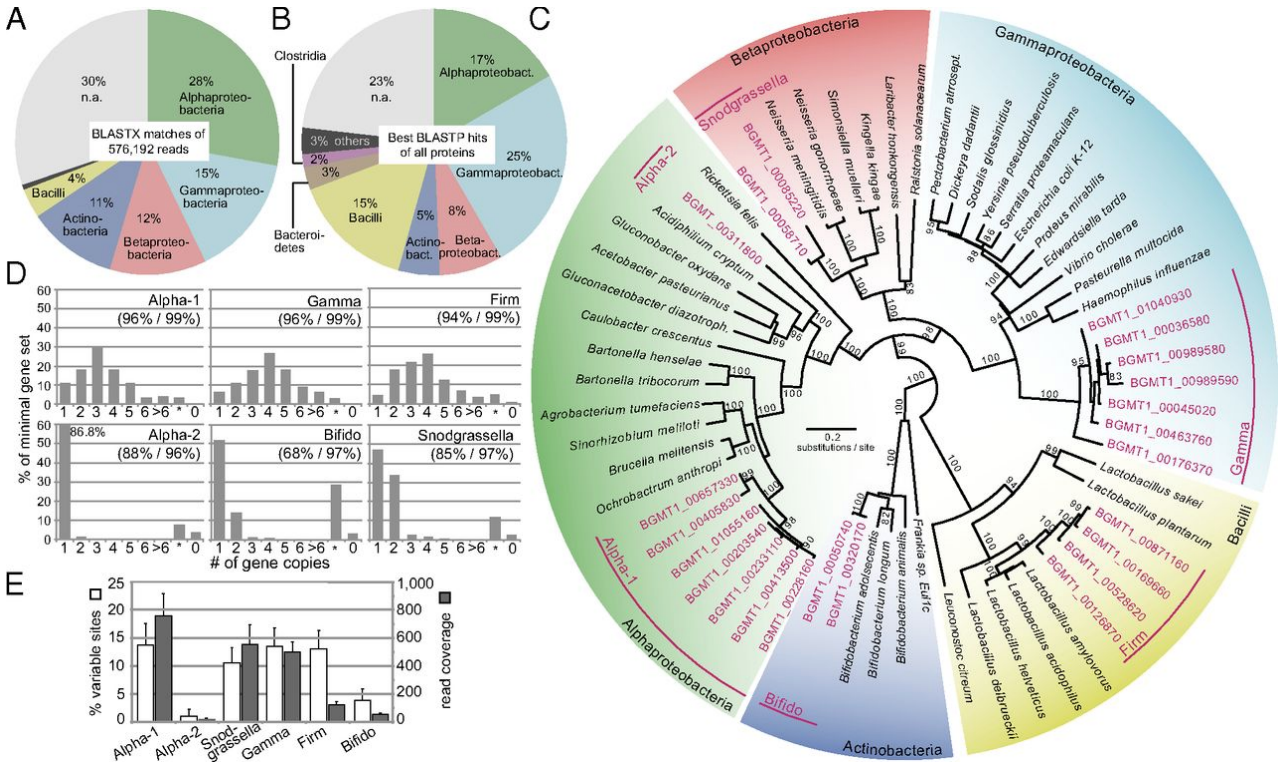
Accurate Pb isotope analysis using NEPTUNE plus

データ非公開

- **Different isotope ratios from low and high polluted area**
- **Similar isotope ratios in soil and chicken kidney at each site**

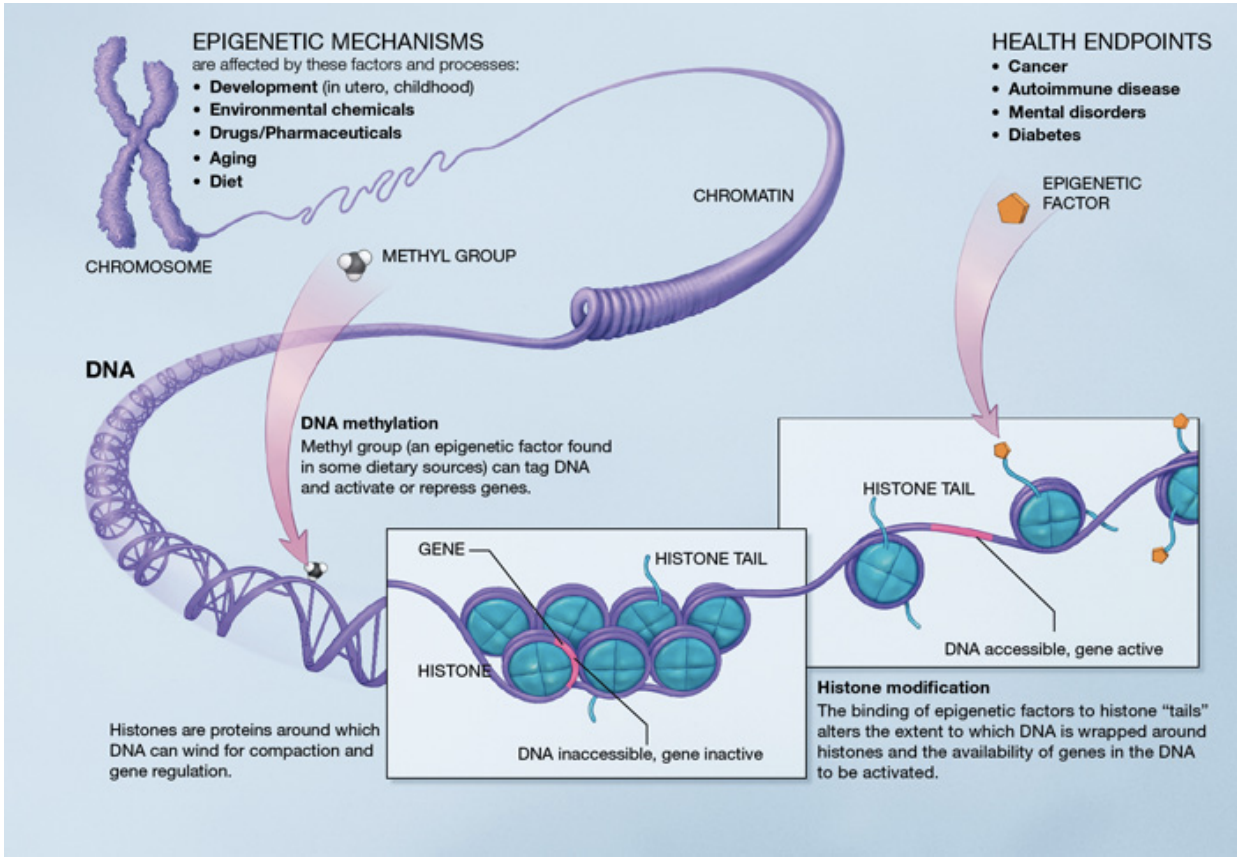
New topics

- Gut microbiota with Group1 (rat and human feces)



Philipp Engel et al., 2012

- Epigenetic analysis



National Institutes of Health - http://commonfund.nih.gov/epigenomics/figure.aspx.