

Malawi MoH/ICOSA: Mapping Protocol

Integrated Control of Schistosomiasis and Intestinal Helminths in sub-Saharan Africa (ICOSA)

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This protocol was produced using material from several individuals within the ICOSA consortium.

ABSTRACT

Schistosomiasis is a highly focalised disease which has been shown to have hugely varied distribution within a small radius. Therefore clear mapping data to establish regions of high and low prevalence is essential in order to assign a treatment plan that reduces worm burden significantly and decreases morbidity associated with schistosomiasis and soil transmitted helminths. In order to produce the most cost-effective, yet sufficiently detailed mapping strategy, a number of alternative mapping methodologies have been considered. The proposed strategy, in our opinion, is a time efficient and cost-effective method which is also sensitive to budget limitations and the potential economical and logistical strains placed on the Ministry. We believe this protocol will reduce costs without losing specificity for directed treatment to schools with the highest burden of disease.

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Background

Mapping is proposed in 10 districts, which are yet to implement mass treatment as shown in Appendix A. The districts are: Balaka, Blantyre, Chiradzulu, Lilongwe, Mwanza, Mzimba (divided administratively into North and South Mzimba), Neno (formerly a part of Mwanza district), Ntcheu and Ntchisi. A National Mapping survey was conducted in Malawi in 2002, sampling children from 30 schools across the country. Though published, there are some concerns regarding the reliability of the data, particularly the *S. mansoni* prevalences, due to technical limitations (e.g. use of thick smears, high technicians' workload, limited time). Unpublished MoH surveys for *S. haematobium* have been conducted in 2003, 2008 & 2010 using urine filtration methodology. The strategy used in districts currently under treatment is a reduced version of WHO mapping guidelines, sampling 50 children in each of five schools per district. A restricted number of schools were sampled due to cost limitations. In the districts yet to implement treatment, the availability of recent prevalence surveys ranges from 0 - 5 schools per district.

Existing prevalence studies in the 10 new districts are not extensive (being based on 1-5 schools), and thus prevalence estimates that could be calculated would be doubted for their precision. Since schistosomiasis is a highly focalised disease, such estimates have a great deal of uncertainty associated with them. It is inadvisable to use such estimates to categorise a district into a prevalence band in order to determine treatment. To avoid potentially wasteful chemotherapy rounds in future years, further mapping is essential.

Existing WHO guidelines for mapping refer to sampling within ecological zones. If it were possible in Malawi to categorise schools into definitive ecological zones that ensured fairly uniform levels of infection in each zone, this would be an ideal way to proceed. In practice, the rich background information required to predict schistosomiasis prevalence to any degree of accuracy is not easily available.

A significant division in Malawi is between highland and lowland (including the lakeshore) regions. All districts to be mapped in this round fall into the highland category, as all lowland districts have already begun implementing treatment. Moreover schistosomiasis is thought to be present throughout the district as water sources are mostly small in nature. However, data available for Malawi does allow that the 10 newly implementing districts can be split into urban and rural sub-districts through the use of educational districts. Educational districts mostly correspond with the overall administrative districts, except for the addition as separate districts of the major cities (Lilongwe, Blantyre, Zomba and Mzuzu), and the separation of the large rural district of Lilongwe into East and West regions. Since urban/rural classification is a significant predictor of S. haematobium infection in Blantyre (Kapito-Tembo et al., 2009), it is right to use this sub-district classification in the mapping protocol. Three of the districts under consideration have significant urban centres (Lilongwe, Blantyre and Mzimba North (Mzuzu City) - see Appendix A). Ideally, it would also be useful to split or stratify the sample by altitude and other predictive factors. This information is not easily available for schools since we lack their GPS coordinates. We would also be able to map separately for Lilongwe Rural East and West, providing we have sufficient time and resources. Though we have no evidence that the ecological zone may differ between these areas, the populations and potential savings in PZQ, is large, and thus may present efficient use of resources if differences are identified. Moreover, Lilongwe District is thought to be the major area of S. mansoni infection in Malawi (in other areas, S. haematobium is thought to dominate), which is sometimes seen to be more focally distributed.

We should map the smallest administrative region at which it is logistically possible to change treatment strategy. It is understood that it will be possible to change treatment strategy by educational district, hence this is the region considered here.

Proposed Mapping Strategy

Primary Objective

The primary aim of this mapping exercise is to categorise sub-districts of Malawi that have not yet started preventive chemotherapy into WHO prevalence categories (Table 1) for *S. haematobium* and *S. mansoni* in order that targeted treatment may commence.

Table 1: WHO guidelines for treatment of schistosomiasis

Category	Prevalence	Act	ion
High-risk	≥50%	Treat all school-age children once a year	Also treat adults considered to be at risk
Moderate-risk	≥10% but <50%	Treat all school-age children once every two years	Also treat adults considered to be at risk
Low-risk	<10%	Treat all school-age children twice during their primary schooling	Praziquantel should be available in dispensaries and clinics

Secondary Objectives

- To classify sub-districts according to prevalence for the soil-transmitted helminths (STHs): hookworm, *Ascaris* and *Trichuris*;
- To enhance knowledge of the spatial distribution of schistosomiasis and STH infections in Malawi;
- To assist planning of the longitudinal preventive chemotherapy evaluation (PCE) strategy for the national schistosomiasis control programme.

Baseline Data Collection for Monitoring of Impact of PCT

This protocol aims to collect prevalence data only for mapping and treatment decisions. Baseline data collection for monitoring of effectiveness and health impact of PCT will be conducted separately using longitudinal cohorts (see separate protocol - PCE strategy).

Selection of Schools

Type of Schools

The surveys will be conducted in primary schools:

- Prevalence in older primary school students should be representative of prevalence in the community as ageprevalence curves plateau in this age group.
- The higher primary school enrolment in Malawi (UNESCO World Data on Education 2010/11) as compared to secondary school ensures that the majority of children of the desired age group will be included in the sampling frame, minimising selection bias.
- Primary schools present a convenient platform for conducting surveys and delivering treatment to at-risk individuals.

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Sampling Frames

The sampling frame is a list of all primary schools in the geographical survey area, from which the survey schools will be selected. A separate sampling frame is required for each of the 'Sampling Areas' defined in Appendix A.

- The sampling frame should be as recent and complete as possible (to avoid bias).
- The sampling frame should be stored in electronic (computerised) format, preferably as a Microsoft Access database, or alternatively as a Microsoft Excel spreadsheet.
- If possible, the sampling frame should include:
- 1. the name of each school
- 2. the educational district in which each school is located
- 3. the identification code of each school
- 4. the location of each school (geographic co-ordinates)
- 5. the total number of students in each school
- As a minimum requirement, numbers 1 and 2 above should be included.

A database of all the schools in Malawi has been provided and the SCI biostatistician has selected the schools based on the sampling criteria, including school list and enrolment figures (see file "Schools for Malawi mapping"). Geographic coordinates are not available for schools in Malawi.

Number of Schools and Pupils to be Selected

We propose drawing separate samples from each 'Mapping Area' listed (see Appendix A). Within each Mapping Area 20 schools will be randomly selected with a further random sample of 30 pupils per school. This will allow us to estimate prevalence with a 95% confidence interval within +/- 10 percentage points (see Appendix B for details of how the sample size was calculated). For example, we shall select 20 schools from 'Blantyre-Rural'. This yields a total mapping sample of 600 pupils in each of 13 sub-districts i.e. 7800 pupils across 260 schools (if we take a single sample from Lilongwe Rural District) or **8400 pupils across 280 schools**, if we sub-divide Lilongwe Rural into the East and West educational districts.

Selection of Schools

The surveys will be conducted in primary schools. The method to select primary schools within each Mapping Area is simple random sampling:

- From the sampling frame for each Mapping Area, a numbered list of all primary schools should be compiled, from 1 N, where N is the total number of primary schools in the Mapping Area.
- Generate n random numbers between 1- N, where n is the number of primary schools in the district to be sampled. This can be done using the =RANDBETWEEN() fuction in Excel.
- Select primary schools with identification numbers corresponding to the randomly generated numbers.

This has been carried out by the SCI biostatistician and is listed in file 'Schools for Malawi mapping'.

Data to be collected

The requirements from each individual are contained in Table 2. The use of Hemastix provides a means of assessing *S. haematobium* prevalence that is comparable to urine filtration (Lengeler et al., 1993) in sensitivity but is more cost effective. However, due to potential contamination from blood in urine due to other causes (menstruation, infection), specificity can be low. Thus all positive haemastix will be confirmed by urine filtration. For *S. mansoni* and STH, Kato-Katz is required. Since we are only collecting prevalence information, we need only see one egg of the relevant species across two Kato-Katz slides to record a positive result. It is proposed that we test for both *Schistosoma* species in all mapping districts since previous evidence as to where *S. mansoni* does not occur is out of date. We understand that there has been recent treatment with ivermectin and albendazole as part of the LF programme (October 2011). It may therefore be inappropriate to map for STHs. However, since Kato-Katz slides are being prepared, it is proposed to test for prevalence of STHs also, thus providing extra monitoring for STH levels. A single slide for STH is sufficient per individual.

Table 2: Data requirements for mapping

ID	Sex	Age	Microscopist initials	At least one egg across 2 slides:				Urine filtration	
				S. mansoni	hookworm	Ascaris	Trichuris	result (for positive urine dipstick results)	

Data will be collected on paper case report forms. A database has been developed and netbooks have been ordered so that data entry can occur by a Malawian team. This will require the team to be trained in the use of the database. The electronic data will then be sent to the SCI biostatistician for analysis.

References

BROOKER, S., KABATEREINE, N. D., CLEMENTS, A. C. & STOTHARD, J. R. 2004. Schistosomiasis control. *Lancet*, 363, 658-9.

JOHN, R., EZEKIEL, M., PHILBERT, C. & ANDREW, A. 2008. Schistosomiasis transmission at high altitude crater lakes in western Uganda. *BMC Infect Dis*, 8, 110.

LENGELER, C., MSHINDA, H., MORONA, D. & DESAVIGNY, D. 1993. Urinary schistosomiasis: testing with urine filtration and reagent sticks for haematuria provides a comparable prevalence estimate. *Acta Trop*, 53, 39-50.

Protocols

General

1. Arriving at the school

- The school information form (see file 'ICOSA Mapping School Form Malawi) should be completed by the team leader.
- The GPS co-ordinates of the school should be entered. Remember also to re-read and re-enter the coordinates at the end of the visit.
- Data on latrines present in the school should be recorded on 'ICOSA Mapping Latrine Form Malawi' by visual inspection.

2. Selecting the class to be surveyed

- Students should be selected from Standard 6 first; we need children between 10-14 years old.
- If too few boys or girls are present in Standard 6, higher Standards should be surveyed, until enough students are available to make up at least 15 boys and 15 girls. (If this is still too few students, Standard 5 should be included).

3. Selecting the students

- The students should be separated into Standards and assembled in lines one line of boys and one line of girls for each Standard to be surveyed.
- If more than the required number of students is present, they should be selected randomly. It is very important to sample randomly to ensure that results are accurate.
- In order to select randomly, it is first necessary to calculate the sampling interval (*SI*) for each Standard/gender group (i.e., the number of positions in the line after which a child is selected).
- *SI* = the total number of students in the line divided by the number of students to be surveyed in that Standard/gender group, rounded to the nearest whole number. Example A. If there are 6 boys in Standard 6 7 boys in Standard 7 and 9 boys in Standard 8, all boys in Standard 6 & 7 are selected so we have 13 boys and 2 additional boys are required from Standard 8). *SI* for boys in this class is 9/2 = 4.5, rounded up to 5 Example B. If there are 60 girls in Standard 6, a sample of girls from this class needs to be selected. *SI* for girls in this class is 60/15 = 4.
- Select an arbitrary "start" number between 1 and the *SI*, which corresponds to the position of the first student to be selected. Subsequent students are selected by adding *SI* to the position of the previously selected child (in other words, if *SI* = 5, every 5th child is selected). Continue to the end of the line.

Here is a worked example of selection of students within a school: Let's say there are 12 boys and 12 girls in Standard 6. All of these children are selected. 3 additional boys and 3 additional girls from Standard 7 would be required to make up the total sample of 15 boys and 15 girls. Since there are more students in this class than the 3 boys and 3 girls required, a random sample of these children is required. Examples of selecting such a random sample are shown in **Figure 1.** In **Example 1**, there are 12 boys and 10 girls in Standard 7 (labelled 13 year old class). In **Example 2**, there are 6 boys 9 girls in Standard 7. *SI* for "example 1" boys = 12/3 (4), "example 1" girls = 10/3 (3.3), "example 2" boys = 6/3 (2) and "example 2" girls = 9/3 (3). To select the random start numbers, select arbitrary numbers between 1 and *SI* for each line. Let's say the numbers selected were 3 & 1 ("example 1") and 1 & 2 ("example 2"). The first student selected corresponds to these positions in the line. Every 4th student after that is selected for "example 1" boys, every 3rd student for "example 1" girls, every 2nd student for "example 2" boys and every 3rd student for "example 1" girls, every 2nd student for "example 2" boys and every 3rd student for "example 1" girls, every 2nd student for "example 2" boys and every 3rd student for "example 1" girls, every 2nd student for "example 2" boys and every 3rd student for "example 1" girls, every 2nd student for "example 2" boys and every 3rd student for "example 1" girls, every 2nd student for "example 2" boys and every 3rd student for "example 1" girls.

4. Collecting the samples

- Each student is asked for consent to provide urine and stool samples.
- The student is given empty stool and urine containers and is instructed on how to collect sufficient amounts of urine and stool for testing. Urine samples should be collected ideally between 10am and 12pm.
- The team leader registers the student, labels the specimens with an identification number and enters the student's personal details on the Case Record Form (see file 'ICOSA Mapping Pupil Form Malawi').
- The student submits the stool specimen to the "Kato-Katz" table and proceeds to the "urine" table where the urine sample is submitted.
- Additional detailed instructions for assigning ID numbers and completed record forms is shown in Appendix C.

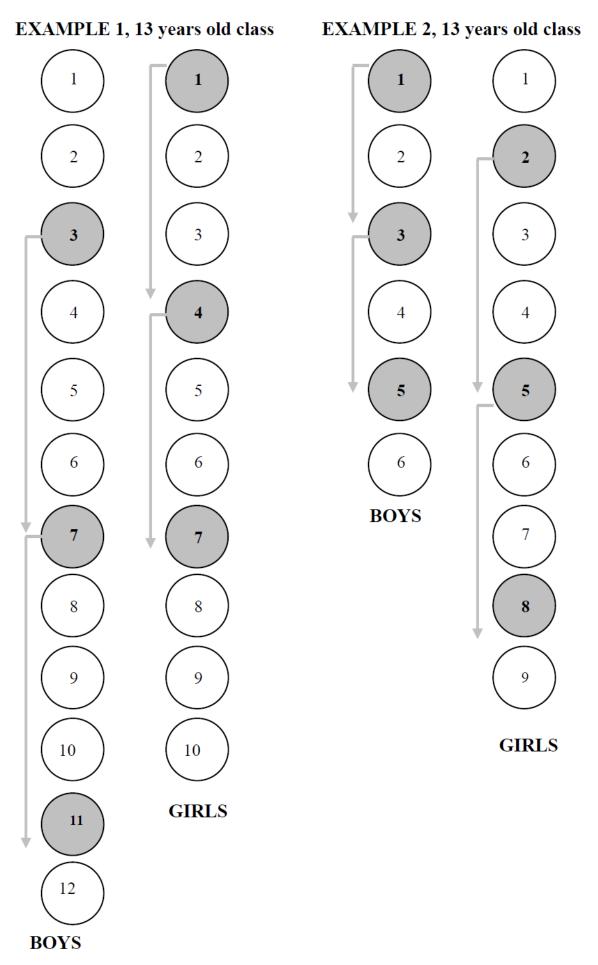


Figure 1. Example: selection of students using a systematic random sample.

Parasitology

Safety precautions

- The stool and urine should be considered potentially infectious.
- Wear gloves whenever handling urine and stool samples plus lab coats when handling stool samples.
- Benches, instruments and equipment should be routinely decontaminated with disinfectants after use.
- Materials contaminated with infectious waste should be disinfected before disposal.
- Drinking or eating during laboratory procedures is prohibited.
- Appropriate disinfectant(s) should be used for disposal of contaminated cotton wool, wooden spatulas and specimen containers and for cleaning of workbenches.
- Used specimen containers must be disinfected before washing.

Equipment

- Stool and urine sample collection: gloves, stool and urine containers.
- Testing for micro-haematuria: gloves, reagent strips (Hemastix).
- Kato-Katz: template (41.7mg), cellophane, applicator stick, stool sieving mesh, wooden spatula, Whatman blotting paper, glass slides, methylene blue, eosin, glycerol, tissue paper, insecticide.

Microscopic examination

- Microscope, Case Record Forms.
- Disinfectants and waste disposal: Chloroxylenol (Dettol), chlorhexidene (JIK), medicated soap, methylated spirits.
- Waste container (containing disinfectant).

Testing for micro-haematuria

- Take a reagent strip and cut it into two (lengthwise) with a pair of scissors.
- Dip one half of the reagent strip into the urine sample for a few seconds. (The other half of the reagent strip will be used for the next student's specimen).
- Wait for about 2 minutes and compare the colour of the reagent strip to the colours on the label of the reagent strip container.
- Write down the result on the Case Record Form next to the appropriate ID number.
- Discard the used reagent strip in the waste container.
- Tests scored as positive or trace positive should be verified by urine filtration.

Testing for *S. haematobium* eggs by urine filtration.

- All urine samples which are 'positive' or 'trace positive' should be subject to urine filtration.
- Once one egg for *S. haematobium* is identified, record this as a '1' on the Case Record Form. If no eggs are seen, record '0'.

Preparing Kato-Katz reagents

- 1. Weigh out 0.5g of Methylene Blue powder.
- 2. Dilute it in 100ml of distilled water (this is the "stock solution").
- 3. Dilute 50ml of glycerine in 50ml of distilled water.

4. Take 1 ml of Methylene blue stock solution and add it to 100ml of the 50% glycerine solution (this is the "working solution").

5. Cut cellophane into 25mm x 30mm pieces and soak them overnight in the working solution.

Stool processing and Kato-Katz

- 1. The stool sample is processed immediately while still fresh.
- 2. Place two glass slides alongside each stool sample.
- 3. On one end of the slide place a sticker labelled with the ID number from the stool sample container.
- 4. On the other end of the slide, place a sticker labelled with the date and slide number (1 or 2).
- 5. Place a template on each of the labelled slides.
- 6. Take a small portion of stool from the sample container using a wooden spatula.

- 7. Smear the stool sample on a sieve until sufficient specimen is obtained for processing.
- 8. Take a small portion of sieved stool with a plastic applicator and transfer it to the slide templates.
- 9. Remove the templates from the glass slides and place them in the disinfectant container for later washing.
- 10. Take a piece of prepared cellophane with forceps and place it over each slide's specimen.

11. Press the slides over blotting paper until there is even spread of the specimen (such that it is transparent) and place the prepared slide into a slide box.

12. Within one hour of preparation, examine the slides under the microscope for hookworm eggs. Once one egg is identified, record this as a '1' on the Case Record Form. If none are seen, record '0'.

13. After at least 24 hours, examine the slides under the microscope for *S. mansoni*, *Ascaris lumbricoides* and *Trichuris trichiura* eggs.

Microscopic examination for S. mansoni and STH

1. Take a Kato-Katz slide, put a little amount of eosin on the slide and place it under microscope using x 10 objective.

2. Read ALL fields of the slide using the vertical 'zig zag' scheme.

3. Once one egg for each species is identified, record this as a '1' on the Case Record Form. If none are seen for a given species, record '0'.

Immediate treatment for each sampled school

Each infected child should be treated with the appropriate drug. Whether children outside the sample are treated depends on the extent of infection within the sample, country policy and the number of tablets available at mapping stage. Mapped schools may require exclusion from future monitoring samples.

<u>Albendazole</u>: 400mg tablet. For treatment of *Ascaris lumbricoides Trichuris trichiura* and hookworm species infections. Single dose treatment

Age	Dose
Under 1 year	Do not treat
1 – 2 years	½ tablet
Children over 2 years	1 tablet
Adults	1 tablet

<u>Praziquantel:</u> 600mg tablet. For treatment of infection with *Schistosoma mansoni and S. haematobium.* Single dose treatment. The standard dose is 40mg/kg body weight. Height can also be used to determine dose required according to the WHO praziquantel dose pole.

Children under 5 years of age are unlikely to be infected with *S. mansoni and S. haematobium*, but praziquantel is safe for use children aged 1 to 5 years. However no children younger than 6 years old will be included in this study.

Weight (kg)	Height (cm)	Number of 600mg tablets
15 – 22.5	94 – 110	1
22.5 – 30	110 – 125	1 1⁄2
30 - 37.5	125 – 138	2
37.5 – 45	138 – 150	2 1/2
45 - 60	150 – 160	3
60 – 75	160 - 178	4
> 75	> 178	5

Appendix A

Areas to be mapped are districts shown in hatched on the map below. Distance from Lilongwe to Blantyre is 311km. The districts vary in size and population numbers. For example Mzimba District (N & S) combined is 10,430km², Ntchisi District is 1655 km², Lilongwe District is 6,159 km², Ntcheu is 3,424 km², Balaka 2,193 km², Mwanza & Neno (formerly one district but now split into 2) 2259 km², Blantyre 2012 km² and Chiradzulu 767 km².



District	Mapping	No. of	Population size	SAC (enrolled &	PZQ need (SAC)
	Area	schools		non-enrolled)	(sampling area)
Mzimba N	Mzimba N	252	819, 297	261, 359	653, 398
	Rural		(with Mzimba S)	(with Mzimba S)	(with Mzimba S)
Mzimba N	Mzuzu		181, 690	48, 029	120, 073
	City				
Mzimba S	Mzimba S	290	819, 297	261, 359	653, 398
			(with Mzimba N)	(with Mzimba N)	(with Mzimba N)
Ntchisi	Ntchisi	139	258, 499	78, 715	196, 788
Lilongwe	Lilongwe	202	1, 116,180	428, 523	1, 071, 308
	Rural East		(with Rural West)	(with Rural West)	(with Rural West)
Lilongwe	Lilongwe	236	1, 116,180	428, 523	1, 071, 308
	Rural		(with Rural East)	(with Rural East)	(with Rural East)
	West				
Lilongwe	Lilongwe	136	868, 800	235, 985	589 <i>,</i> 963
	City				
Ntcheu	Ntcheu	240	528, 088	164, 074	410, 185
Balaka	Balaka	160	360, 252	116, 260	290, 650
Mwanza	Mwanza	49	99, 434	31,610	79, 025
Neno	Neno	69	130, 611	40, 686	101,715
Blantyre	Blantyre	178	373, 843	116, 400	291,000
	Rural				
Blantyre	Blantyre	172	783, 296	208, 252	520, 630
	City				
Chiradzulu	Chiradzulu	88	305,692	96,822 6	242, 065

Appendix B

To map the remaining districts of Malawi, we propose a variant of the WHO guidelines, which state: '200-250 individuals should be an adequate sample for each ecologically homogeneous area in order to evaluate prevalence and intensity'. We are proposing a cluster sample of schools and therefore need to inflate the sample by the 'design effect' to incorporate the between school variance that exists across ecological zones. We have explored suitable values of roh, the intra-cluster correlation, using data from Uganda (S. mansoni) and Burkina Faso (S. haematobium). This work has revealed values of roh of 0.33 for S. mansoni prevalence across a substantial area of Uganda and 0.35 for S. haematobium prevalence across Burkina Faso. Since we are interested in estimating prevalence at district rather than national level, it is reasonable to assume that roh will be lower since each district is likely to contain less ecological variability. Sample size calculations have therefore been based on roh=0.2. From our calculations we have valued several sample size options for pupils/school and schools/district in terms of cost and in each educational district we propose to take a random sample of 20 schools; 30 pupils per school. This will allow us to estimate prevalence with a 95% confidence interval within +/- 10 percentage points. If the resulting 95% confidence interval is above or contains 50%, treatment for that district will be annual. If prevalence is significantly below 50% but above 10% (or the confidence interval contains 10%), treatment can be biennial for that district. Extra care will be needed when calculating confidence intervals for district prevalence; they will need to be inflated according to the design effect that results from a cluster sample. If the amount of variation between schools are lower than has been estimated (focality of distribution), confidence limits on estimates will be narrower.

Appendix C: Standard Operating Procedure: ICOSA Data Collection forms

Form 1: ICOSA - Mapping School Form

The Mapping Form is critical for the programme to succeed; it will allow us to gather background information required for the programme. This form must be filled out upon arrival at each of the schools that are participating in the activities of the Schistosomiasis and Soil Transmitted Helminth programme.

Section A: Site Details

Site Details should be filled out on arrival at the location as outlined on the forms.

1. Date of Visit: To be filled on the day of Mapping activities following:

Day (DD) – Month (MMM) – Year (YYYY)

2. Team Leader Initials: The data collector will record his/her initials in the allocated spot on the form:

Team Leader Initials|___|__|Example: John Jones Smith|J|J|S|

- 3. District Name: Record the name of the District here in BLOCK Capitals to ensure it is easy to read
- 4. Educational District Name: Record the name of the Educational District here in **BLOCK Capitals** to ensure it is easy to read. This is our 'Mapping Area'.
- 5. Educational District Code: Fill in the district code (DDD) in accordance with the assigned codes (see 'Schools for Malawi mapping'). This is a 3 digit number: 001 013.

Section B: GPS

The GPS device is likely to arrive with the default setting of degrees and minutes. At first use, you will need to change this to decimal degrees (instructions for the GARMIN eTrex[®] H):

- 1. Press PAGE and switch to the menu page
- 2. Select SETUP and press ENTER
- 3. Select UNITS and press ENTER
- 4. Select POSITION FRMT and press ENTER
- 5. Select the decimal degrees format hddd.ddddd and press ENTER.

GPS coordinates must be recorded on site at arrival and departure (stand in the same place for each recording).

Section C: School Details

School information will be gathered on site through conversations with the Headteacher who will assist you in the mapping activities.

1. School Name: Record the name of the school here in BLOCK Capitals to ensure it is easy to read

- School Code: Fill in the school code (SSS) in accordance with the assigned codes (this is a 3 digit code: 001 020. Schools are numbered (arbitrarily) 1 to 20 within each Mapping Area (to be assigned by field team on arrival in 'Mapping Area'/District).
- 3. Name of Headteacher: Record the name of the Headteacher here in **BLOCK Capitals** to ensure it is easy to read
- 4. Have pupils in the school received deworming treatment in the last year?: Write the corresponding number in available space.

1=Yes 0=No 2=Don't know

- 5. Lowest Standard Taught: Write the corresponding number to the lowest Standard taught in the school in the available space.
- 6. Highest Standard Taught: Write the corresponding number to the highest grade taught in available space

Section D: Enrolment Numbers

Record the enrolment in the available space. The headteacher will be able to assist you with this section. Example:

D. Enrolment numbers					
	Boys Enrolled	Girls Enrolled			
Total	1. 1 8 4	2. 1 6 3			
Standard 6	3. 5 0	4. 4 5			
Standard 7	5. 7 5	6. 6 3			
Standard 8	7. 5 9	8. 5 5			

Additional data on the number and condition of latrines present will be recorded on form 'ICOSA Mapping latrine form'.

Form 2: ICOSA - Mapping Pupil Form

The Mapping Pupil Form is critical for the mapping programme to succeed; it will allow us to gather background information required for the programme. This form must be filled out as samples are collected from students. The pots that these samples are collected in must be recorded with the ID number so that the analysis looking for eggs, haematuria and the dipstick results can be recorded next to the correct individual.

At the School

- Fill out the top of the form
 - o Date of Survey
 - School Code SSS (from 'ICOSA Mapping School Form')
 - Page Number
- ID Number: This will be recorded using the district code (DDD), the school code (SSS) and the individual number (NN) which numbers 1-99; where the first student will be have an NN = 01
 - Label slides or take the corresponding sticker and put in the ID Number location.
 - E.g. The following would be the ID Number for the district with code 001, the school with code 001 and first student (01):

(DDD.SSS.NN) |0|0|1|.|0|0|1|.|0|1|

- Identical ID stickers should also be stuck on the child's sample containers.
- Sex: Record the gender of the student as either **M** for Male and **F** for Female
- Age: Record the age of the student in years.
 - E.g. |**1**|**1**|
- Take the sample containers (Urine and Faecal) and label with ID number (written or sticker)

At the school/In the lab

- Microscopist Initials: The lab technician who is handling the samples must record his/her initials in the provided location.
 - Lab Technicians Initials
 - Example: John Jones Smith
 |J|J|S|
- Egg Detection (stool): Slides will be read for hookworm on the day of collection. Slides will be read for the follow species of parasitic helminths at least 24 hours later: *Schistosoma mansoni, Ascaris lumbricoides* and *Trichuris trichiura*.
 - A simple presence/absence test is being used for the mapping activities. If one egg is found of a species it must be recorded using the result key on the form.
 - E.g. 1=Yes, 0=No
- Haematuria (Macro) will be recorded visually using the following codes:
 - 1=Yes, 0=No

- Urine Dip Stick: The results of the dip stick test will be recorded as follows:
 - \circ 0 = none, 1 = trace haemolysed, 2 = trace non-haemolysed, 3 = +, 4 = ++, 5 = +++
 - If positive use urine filtration to detect presence of eggs

Urine filtration (if dip stick tests positive): A simple presence/absence test; if one egg is found, record *1=yes* otherwise, record *0=no*. If the original dip stick test was negative then leave this field blank.

Draft Logistics (this can be optimised during the survey as required)

Proposed Logistics

No of teams

Team A: Consisting of 2 vehicles and 2 Sub-teams. Each sub-team consists of team leader, 2 central level technicians and 1 district technician. Accompanied by 1-2 Ugandan technician & Dr Wendy Harrison (for one week). Team B: Consisting of 2 vehicles and 2 Sub-teams. Each sub-team consists of team leader, 2 central level technicians and 1 district technician. Accompanied by 1-2 Ugandan technicians.

Draft timetable for each week:

Monday: Courtesy call to district officials. Sub-team 1: School 1&2. Sub-team 2: School3&4. Collect samples (morning), perform haemastix, urine filtration and review KK slides (for hookworm).

Tuesday: Subteam 1: Schools <u>5</u>&6. Subteam 2: Schools 7&8. Technicians can review KK slides from schools 1-4(for S. mansoni and other STHs – either with the collection team, or at the district laboratory as appropriate). Perform haemastix, urine filtration and prepare KK slides and read for hookworm (schools 5-8).

Wednesday: Subteam 1: Schools 9&10. Subteam 2: Schools 11&12.

Thursday: Subteam 1: Schools 13&14. Subteam 2: Schools 15&16.

Friday: Subteam 1: Schools 17&18. Subteam 2:19&20

Saturday: Review KK slides (*S. mansoni* schools 17-20) (these could also be reviewed on Monday morning if preferred). Travel to new district.

Sunday: Rest day.

Thus it is proposed that each sub-team visits 2 schools/day. It is suggested that two technicians accompany the team leader to perform haemastix, urine filtration, KK preparation and hookworm slide reading at the schools. Potentially, faecal samples could be collected at one school and taken to subsequent school. One technician can remain at a central location to continue *S. mansoni, Trichuris* and *Ascaris* reading. Team can return to central location after lunch, such that all technicians can read KK slides in the afternoon. Ugandan technicians to assist in schools or at central location as needed for training and efficiency.

Useful information

Primary schools are open in the morning until 1pm. Older classes usually stay the longest. In some schools, timings are staggered to share classrooms. Class size per Standard varies considerably but can be in the order of 50-250 students.

Draft Timetable (for discussion)

There are 13 'Mapping Areas' each with 20 schools to be sampled

N.B. Monday 5th March is a public holiday and schools are closed. Schools close 23rd March for Easter holidays

Team A: Lilongwe Rural East, Lilongwe Rural West, North Mzimba, South Mzimba, Mzuzu City, Ntchisi Team B: Balaka, Mwanza & Neno, Ntcheu, Blantyre, Blantyre City, Chiradzulu

Monday 13th and Tuesday 14th February: Training exercise in Lilongwe.

Wednesday 15th February – Friday 17th February: Collect data from Lilongwe City schools, as part of training exercise as appropriate. Potentially begin mapping survey in Lilongwe Rural districts (it should be possible with four teams to collect data from 24 schools; perhaps more if more of the training people are included).

Saturday 18th February: Dr Narcis to return to Uganda.

Mapping survey

Sunday 19th February: Travel day Week 1 (w/c 20th Feb): Team A Mzimba South; Team B: Ntcheu Week 2: (w/c 27th Feb)Team A Mzimba North; Team B: Balaka Week 3: (w/c 5th March) (N.B. 4 days) Team A Mzuzu City Team B: Blantyre City (N.B. schools are likely to be closer together in urban area). N.B. Ugandan technicians to return to Lilongwe for flight 11th March Week 4: (w/c 12th March) Team A: Lilongwe East (or Ntchisi); Team B: Mwanza & Neno Week 5: (w/c 19th March) Team A: Lilongwe West; Team B: Blantyre Rural Schools close: 23rd March (Week 6: Team A: Ntchisi Team B: Chiradzulu)

Monday 23rd March – Friday 27th March: Entry of mapping data in Malawi. Training to be provided in data entry and use of database by SCI biostatistician. Data analysis for one district will be conducted in Malawi by biostatiscian to demonstrate and explain output of mapping data analysis.

Notes: Calculations are based on the ability of each sub-team to visit two schools per day, and thus 2 sub-teams to complete a survey of 20 schools in one week. If it is possible to visit more schools in one day (e.g. in urban districts), this could be completed in a shorter time frame. In order to reduce the time taken, we plan to take a joint stratified sample from the small districts of Mwanza and Neno. This may mean we are only able to make treatment recommendations for these two districts together, but the statistician advises that if variability between schools is small, we may still be able to calculate the prevalence in the two districts independently. I have also suggested sampling in Lilongwe City as part of training, as it would be good to practice throughput through schools, and we are short of time. I have suggested sampling in the urban areas in week 4 (as presumably the distances between schools will be smaller) due to the holiday on the Monday. The Lilongwe district is thought to have the highest prevalence of *S. mansoni*, so we may want to rearrange timetable so that the Ugandan technicians can assist in these districts. Since we are interested in presence/absence, reviewing slides will take longer in low prevalence areas than high prevalence areas.

We clearly have a problem in that we are missing a week for collection in one of the northern/central districts and one of the southern districts (funds for a sixth week have been included in the budget but there are only five weeks following the training until the schools close for holidays). I think we must go ahead with data entry in the week commencing 26th March as the data will be required for the baseline data collection protocol development, which must also be carried out before the MDA in June.

Possible solutions for discussion:

- Data could be collected in two rural mapping areas after the schools have closed. Mapping teams could go to the villages and sample children outside the schools. This is possible logistically, but I am concerned that it will make it very difficult to collect a random sample for example, the children that present may be those who are healthier (potentially underestimating the prevalence of schistosomiasis), or those who know they have blood in their urine and desire treatment (hence overestimating the prevalence). If there was a way of randomly selecting the children while the schools are open from the class list, this could be averted, but would it be logistically possible?
- We could return to the schools following the Easter holidays to collect data in these districts. This is feasible from a data entry point of view (we may need to exclude these districts from the monitoring of longitudinal cohorts) but would have implications for fuel useage.
- For the northern/central teams: Could we somehow cover one of the Lilongwe Rural districts in the training week? Or not split this district into two? Or do some of the schools in the training week and visit more than 2 schools some days to make up time?

• For the southern districts: could we recruit an additional team in Blantyre to cover the Blantyre rural district e.g. from the university (potentially accompanied by one of the Ugandan technicians to ensure protocol/training is followed), which would allow our team to go to another district and collect the data from Blantyre rural before they return to Lilongwe?