Methods

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Project title: OneHealthWater: Drinking-water under a "One Health" lens – quantifying microbial contamination pathways between livestock and drinking-water

Data set: Longitudinal household and microbiological survey of livestock-related risk factors for microbial contamination of household stored water in Siaya County, Kenya
Version 2, completed 31 Mar 2020

Study site

Fieldwork took place in ten villages in Siaya County, Kenya, a rural site on the shores of Lake Victoria, which hosts a Health and Demographic Surveillance System (Odhiambo et al. 2012) and where residents participate in several ongoing studies of livestock and human health (Thumbi et al. 2015). These villages are among a sub-sample of 33 that also participate in an ongoing Population and Population-Based Infectious Disease Surveillance platform and are the focus of an ongoing Population-Based Animal Syndrome Surveillance (PBASS) study (Thumbi et al., 2015).

Study and sample design

These data were collected through a longitudinal, observational study of livestock-related risk factors for contamination of point-of-consumption water with faecal indicator bacteria. The study comprised a questionnaire survey of participating households, coupled with direct observation of relevant conditions and behaviours in the home, as well as microbiological testing of stored and source water for faecal indicator bacteria. Eligible study participants were adult members of households participating in the ongoing PBASS study, whose households included children aged 6-59 months as the cohort at greatest risk of diarrhoeal disease. The sample size was powered to detect differential proportions of contaminated drinking-water using preliminary effect size estimates from Ghana (Wardrop et al. 2018), in the absence of evidence from Siaya. In Ghana, approximately 70% of water samples were contaminated in non-cattle keeping households, 90% were contaminated in cattle-keeping households (H1) and the proportion of contamination variance in cattle-keeping households explained by other covariates was estimated at 0.3. Within the study population, 55% of households own cattle (Thumbi et al. 2015). Based on these assumptions and a Type 1 error rate of 0.05, and a desired power of 0.9, a power calculation using the G* Power software indicated a required sample size of 196 households, which we rounded to 240 to allow for households declining to participate or dropping out of the study. Prior to recruitment, the study design was registered with the International Standard Randomised Controlled Trial Number (ISRCTN) registry (Reference number: ISRCTN69058168).

Ethical approval

Ethical approval for the study was obtained from the Faculty of Social and Human Sciences, University of Southampton (reference: 31554; approval date: 12/02/2018) and the Kenya Medical Research Institute (reference: KEMRI/SERU/CGHR/091/3493, approval date: 17/10/2017).

Field team recruitment and training

The field team was recruited from an existing group of field officers who had slightly over five years' experience in conducting field based research. The field team was subjected to intense training of the data collection tools prior to actual data collection. The training involved a detailed review of the questionnaires to ensure a deeper understanding of the questions, possible answers and correct translations from English to Dholuo. A pre-test of the questionnaires on CommCare (Mobile based data collection application) was done to ensure proper functioning of the data collection App and a smooth flow of the questions. The team was thoroughly trained on questionnaire administration techniques, water sample collection and transportation to the laboratory. After successful training, the study was piloted in a few households and appropriate adjustments made to the data tools.

After completion of the first round of data collection, a refresher training of the field team was done before commencing the second round of data collection. The team also reviewed the questions from the first round, in some cases adding or revising response categories and elsewhere inserting a small number of additional questions. Fields resulting from these revisions are flagged in the data dictionary for the household survey.

Pictorial guides were used to aid in the classification of both water source and sanitation types.

Following piloting, the Joint Monitoring Programme (JMP) core question concerning the main source of drinking-water (WHO / UNICEF 2006) was adapted to include an addition response category for water kiosks. Following a team review and follow-up site visits after wet season fieldwork, it emerged that some households were fetching water from broken pipes. Others had adapted their water supplies to cope with intermittent supplies by storing piped and rainwater in the same tank. Specific response categories were introduced for such sources in the dry season.

The number of questions concerning water security was expanded in the light of initial fieldwork and piloting, which suggested the study sites experienced significant water insecurity.

Participant recruitment and questionnaire administration

Eligible households were randomly selected from lists of those participating in the PBASS study. After seeking informed consent, questionnaire interviews were conducted in the Dholuo language with adult respondents during an initial (12th March to 24th May 2018), and follow-up sampling visit (20th November 2018 to 18th February 2019). To assess domestic contacts with animals, interviewers observed the presence of livestock (e.g. cattle, goats, poultry), dogs and cats in the compound during interview and observed evidence of animal presence in the home (e.g. faeces; feathers; footprints). Interviewers also assessed whether the drinking-water container could be accessed by any animals and where chicken coops, asked whether poultry were permanently confined in such coops. The respondent was also asked about whether drinking-water was stored separately for children versus adults. To measure other known risk factors for stored water contamination, water storage characteristics (e.g. whether containers were covered) were observed, and respondents were asked about any water treatment or cleaning of storage vessels. Interviewers also asked about sanitation facilities and handwashing behaviours, observing whether soap was available. The questionnaire survey also covered the use of multiple water sources for multiple purposes, given the study's emphasis on interactions of people and livestock around water sources. Finally, questions were asked relating to water security, such as water storage tanks, adaptations to water scarcity, and direct and indirect costs associated with fetching water.

After completing the questionnaire, the survey team requested that the respondent fetch a sample of the stored drinking-water, including a second sample of any water stored separately for children. The interviewer observed how the respondent collected the sampled water. Samples were also tested in situ for their residual free chlorine levels using a strip test (SenSafe®). Approximately 6 to 8 water samples

from households were collected per day. Approximately 500 ml of water was collected and poured into sterile polyethylene one-litre bottles (Fisher Scientific, UK).

For both visits, a separate survey team also visited the sources of stored drinking-water reported by households, collecting a water sample where source water was still available. Sources used by more than one household that had already been visited were not resampled during this process. Depending on the type of source, water samples were poured in polyethylene one litre bottles (Fisher Scientific, UK) or the bottles were dipped in the source (using sterile gloves and/or strings).

Physico-chemical and microbiological water testing methods

Free residual chlorine: This was tested in situ using SenSafe free chlorine Water Check test strips, capable of detecting 0, 0.05 0.1, 0.2, 0.4, 0.6, 0.8, 1.2, 1.5, 2.0, 2.6, 4.0, and >6.0 ppm (mg/L) of free chlorine. The method is approved by the US Environmental Protection Agency (ITS Method 99-003), as published in the 2007 Federal Register. No calibration is necessary for these strips.

Faecal indicator bacteria: All sample bottles were kept in a cooled container (4°C) and shipped within four hours to the Kenyan Medical Research Institute (KEMRI) laboratories in Kisian (Kenya). Samples were either processed immediately or kept in the fridge (4°C) and processed within 24 hours.

Because of the ethical and logistical issues raised by collecting the very large water sample volumes required to enumerate pathogenic Cryptosporidium sp., we assessed microbiological water quality via faecal indicator bacteria (FIB), namely *E. coli* and intestinal enterococci.

Enumeration of FIB followed ISO standard methods (ISO 9308-1:2014 for *Escherichia coli* and total coliforms, and ISO 7899-2:2000 for Intestinal enterococci) and was performed using the membrane filtration technique. Since during pilot studies some water samples generated Too Numerous To Count (TNTC) results for 10 ml volumes and almost all for 100 ml volumes, it was decided also to filter 0.1, 1 ml and 10ml volumes for the samples from visit 1. Subsequently, data from visit 1 suggested that bacterial contamination levels were lower than in pilot fieldwork, so 100ml samples were additionally processed in visit 2, thereby enabling assessment of *E. coli* compliance with the WHO guideline value (not detectable in 100ml). Therefore, across the two visits, for each water sample and for each FIB group investigated, four volumes (0.1, 1, 10 and 100 ml) were poured into a filtration unit containing approximately 10 ml of quarter-strength Ringer's (QSR) solution and then filtered through a 0.45µm pore-size cellulose nitrate filter (Thermo Scientific) using a vacuum pump (Fisher®). Four filters, one for each volume of the filtered water, were placed onto coliform chromogenic agar (CCE) agar (Difco®) in Ø 55mm petri dishes (Fisher®). Plates were then incubated upside down for 24 ± 2 hours at 37.0 ± 0.5°C. Colonies that showed shades of dark-blue to violet were counted as *E. coli*, while those that appeared pink to red-coloured were recorded as presumptive coliforms (total coliforms) that were not *E. coli* (ISO 9308-1:2014). In addition, four filters from each volume of the filtered water, were placed onto Slanetz and Bartley agar (Oxoid®) in Ø 55mm petri dishes (Fisher®). Plates were then incubated upside down for 48 ± 2 hours at 37.0 ± 0.5 °C, and raised colonies that showed shades of red, maroon and pink were counted as presumptive intestinal enterococci (ISO 7899/2:2000).

Data management, processing, quality control, linkage and anonymisation

Data management: All field-based data were collected using Android devices via the CommCare data management system (https://www.dimagi.com/commcare/). Laboratory-based enumeration of faecal indicator bacteria took place with only partial knowledge of sample origins, so as to reduce potential for technician bias in interpreting bacteria counts. Some fields, notably water volumes and costs, were automatically calculated within CommCare.

Quality control: Range checking was built into data capture screens, as was logical branching within data collection forms. Linkage of water sample laboratory records and field records was undertaken to check for orphan records and duplicates. Bacterial counts were examined for digit preference following data collection, but there was no evidence of this.

Calculated fields: Several fields are automatically calculated, such as those relating water consumption and water-related expenditure. Calculations are documented in the data dictionary.

Anonymisation: Personal data such as participant or field team member names and contact details have been removed, the original household identifiers (a composite unique ID consisting of the household ID and compound ID) have been replaced with unique pseudo-identifiers, and comments in the data file have been screened for inadvertent disclosure of personal data. Local place names, village names, and other geocodes have also been removed.

Data structure, linkage & related data resources: The field *UniqueID* contains a unique ID for each household, repeated for the two visits. The ten villages are also uniquely numbered in the *VillageID* field. Each water sample also has a unique ID. The field *SampleID* may be used to link household stored water characteristics in the OHW_HHSurvey_Data table to the OHW_FaecalBacteria_Data table; *ChildSampleID* serves the same purpose for water stored at home separately for children. Note that all sample IDs should have be 8 characters long. Some sample IDs (in the laboratory file) are only 7 characters long as a trailing zero was not entered. A trailing zero should be added to these IDs prior to linkage.

The fields *StoredWaterSrcID* (the specific source of water stored in the home when households were visited) and *WaterSourcesID* (a water source used for domestic purposes, but not necessarily the source of water stored in the home at the time of the visit) may be used to link the OHW_HHSurvey_Data to the related data set on water sources and their characteristics. This data set may be found via this persistent identifier: http://doi.org/10.5255/UKDA-SN-853860 . The field *SampleID* in this related data set may also be used to link the characteristics of water sources from which samples were taken to the microbiological test results in the laboratory, held in the OHW_FaecalBacteria_Data table.

Note that the participants in this study also took part in the PBASS study, but PBASS data (e.g. concerning household demographics, goods and services), which were collected with separate funding, remain the property of KEMRI.

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