

# BMJ Open Environmental and behavioural exposure pathways associated with diarrhoea and enteric pathogen detection in 5-month-old, periurban Kenyan infants: a cross-sectional study

Kelly K Baker <sup>1</sup>, Jane Awiti Odhiambo Mumma,<sup>2</sup> Sheillah Simiyu,<sup>3</sup> Daniel Sewell,<sup>4</sup> Kevin Tsai,<sup>1</sup> John David Anderson <sup>5</sup>, Amy MacDougall,<sup>6</sup> Robert Dreibelbis,<sup>6</sup> Oliver Cumming <sup>6</sup>

**To cite:** Baker KK, Mumma JAO, Simiyu S, *et al.* Environmental and behavioural exposure pathways associated with diarrhoea and enteric pathogen detection in 5-month-old, periurban Kenyan infants: a cross-sectional study. *BMJ Open* 2022;**12**:e059878. doi:10.1136/bmjopen-2021-059878

► Prepublication history and additional supplemental material for this paper are available online. To view these files, please visit the journal online (<http://dx.doi.org/10.1136/bmjopen-2021-059878>).

Received 14 February 2022  
Accepted 05 October 2022



© Author(s) (or their employer(s)) 2022. Re-use permitted under CC BY-NC. No commercial re-use. See rights and permissions. Published by BMJ.

For numbered affiliations see end of article.

## Correspondence to

Dr Kelly K Baker;  
kelly-k-baker@uiowa.edu

## ABSTRACT

**Objectives** The aim of this study was to test whether household environmental hygiene and behavioural conditions moderated associations between diarrhoea and enteric pathogen detection in infants 5 months of age in Kenya and pathogen sources, including latrine access, domestic animal co-habitation and public food sources.

**Design** Cross-sectional study utilising enrolment survey data of households participating in the Safe Start cluster-randomised controlled trial.

**Setting** Kisumu, Kenya.

**Participants** A total of 898 caregivers with 5-month (22 week  $\pm$  1 week) aged infants were enrolled in the study and completed the enrolment survey.

**Primary and secondary outcome measures** Outcomes were (1) caregiver-reported 7-day diarrhoea prevalence and (2) count of types of enteric viruses, bacteria and parasites in infant stool. Exposures and effect modifiers included water access and treatment, cohabitation with domestic animals, sanitation access, handwashing practices, supplemental feeding, access to refrigeration and flooring.

**Results** Reported handwashing after handling animals (adjusted odds ratio (aOR)=0.20; 95% CI=0.06 to 0.50) and before eating (aOR=0.44; 95% CI=0.26 to 0.73) were strongly associated with lower risk of caregiver-reported diarrhoea, while cohabitation with animals (aOR=1.54; 95% CI=1.01 to 2.34) living in a household with vinyl-covered dirt floors (aOR=0.60; 95% CI=0.45 to 0.87) were strongly associated with pathogen codetection in infants. Caregiver handwashing after child ( $p=0.02$ ) or self-defecation ( $p=0.03$ ) moderated the relationship between shared sanitation access and infant exposure to pathogens, specifically private latrine access was protective against pathogen exposure of infants in households, where caregivers washed hands after defecation. In the absence of handwashing, access to private sanitation posed no benefits over shared latrines for protecting infants from exposure.

**Conclusion** Our evidence highlights eliminating animal cohabitation and improving flooring, postdefecation and food-related handwashing, and safety and use of cow

## STRENGTHS AND LIMITATIONS OF THIS STUDY

- ⇒ We rely on objective molecular-based outcome measures of infant exposure to enteric pathogens, a large sample size and documentation of many household environmental and behavioural living conditions.
- ⇒ We explicitly analyse household environmental and behavioural living conditions as intermediate factors that can modify the transmission of enteric pathogens between sources of human and animal in the household and infants.
- ⇒ We included supplemental foods given to the infants within the last 24 hours as a potential transmission pathway.
- ⇒ Our 7-day self-reported diarrhoea and pathogen detection exposure outcomes occurred in recent time prior to observation of household living conditions, which limits causal inference.
- ⇒ Our statistical power to examine effect modification hypotheses was limited in cases where living conditions varied little across the population, for example, primary water sources.

milk sources as interventions to prevent enteric pathogen exposure of young infants in Kenya.

**Trial registration number** NCT03468114

## INTRODUCTION

More than a third of all children in low-income and middle-income countries experience infections by one or more pathogens within the first year of life.<sup>1 2</sup> Longitudinal studies of enteric pathogen prevalence show that the proportion of children in low-income communities shedding pathogens climbs rapidly after birth and is sustained through at least 24 months of age.<sup>2 3</sup> One enteric infection episode can increase susceptibility to re-infection or co-infection through

pathogen–pathogen interactions in the gut, downregulation of protective immune or microbiome responses<sup>4</sup> and enteric enteropathy of intestinal tissue.<sup>3</sup> Infants who experience early onset of enteric infections (eg, before 6 months of age) may be more vulnerable to enteropathy and malnutrition conditions that increase enteric infection incidence rates.<sup>5</sup>

Multiple inter-related environmental exposure pathways can contribute to pathogen transmission. Literature identifying targeted interventions that prevent exposure of infants in the first months of life, beyond breast feeding, is scarce. Evidence on impact of improving household safe drinking water sources and/or treatment and storage, access to basic latrines and handwashing with soap on overall enteric pathogen detection and diarrhoea in children under 2 years of age is mixed, with some rigorously controlled and well-powered trials of interventions reporting little impact, even with high levels of behavioural compliance.<sup>1 6 7</sup> One explanation for the trial results is that interventions did not target the most important conditions resulting in pathogen transmission. Most handwashing interventions focus on infant caregivers, not on <5 year siblings and playmates.<sup>8</sup> Infant food safety depends on handwashing, but also hygienic conditions of the food preparation and feeding environment<sup>9 10</sup> and safety of ready-to-eat roadside and packaged foods. The presence of rodents, flies and domestic animals in the immediate household or compound can contaminate soil and surfaces in areas where infants dwell with faeces containing pathogens,<sup>11 12</sup> and those pathogens could persist longer in the environment if household flooring is dirt or another type of permeable material.<sup>13</sup> Similarly, the presence of older siblings who could transmit pathogens through child–child interaction or open defecation on floors could contaminate surfaces.<sup>14</sup> The benefits of maintaining hygiene conditions in one's own household can be derailed if hygiene conditions of communal compound living spaces and infrastructure (eg, shared latrines) are poor.<sup>15–17</sup> Access to a latrine within a compound is not a guarantee that all compound residents will consistently use it for personal defecation or child faeces disposal.<sup>17</sup> Sharing latrines is an established risk factor for childhood diarrhoea.<sup>15</sup> At a community scale, hygiene of one's household does not protect infants in daycare or heavily polluted community settings, or if pollution from the community is blown, flooded, or transported by feet into compound grounds.<sup>18–20</sup>

The goal of this study was to test for effect modification by intermediate household environmental (eg, flooring, refrigeration) and behavioural (eg, handwashing, water treatment) conditions in the relationship between human and animal faecal contamination sources and pathogen exposure of 5-month-old infants in Kisumu, Kenya. We utilised enrolment data for infants in the Safe Start Trial,<sup>21</sup> a cluster-randomised trial of a food hygiene behaviour change intervention to estimate associations between water, animal, sanitation, hygiene, food and flooring conditions with caregiver-reported

7-day diarrhoea prevalence in infants, and with count of enteric pathogen types in infant stool as determined by molecular diagnostic assays. We then conducted a moderation analysis to examine hypotheses of proposed environmental conditions and human behaviours that could influence the size and strength of association between faecal sources and enteric pathogens in infants.

## METHODS

### Study setting/ethics

This cross-sectional study uses baseline data collected from 5-month-old (22 weeks  $\pm$  1 week) infants and their caregivers at the point of enrolment into Safe Start cluster-randomised controlled trial of an infant food hygiene behaviour change intervention in Kisumu, Kenya (Clinical Trials identifier: NCT03468114, Pre-results stage as of 18 Oct. 2022). The formative work and trial protocol, including the estimation of sample size for evaluating trial impact, are described elsewhere.<sup>21–25</sup> Kisumu is a city of approximately 490 000 people (Kisumu county integrated development plan 2013–2017) in the western region of Kenya. The study site includes communities in two low-income periurban neighbourhoods in Kisumu. The study was approved by the scientific and ethical review committees at Great Lakes University of Kisumu (Ref. No. GREC/010/248/2016), London School of Hygiene and Tropical Medicine (Ref. No. 14695) and University of Iowa (IRB ID 201804204).

### Study design and participants

Caregivers with 5-month-old infants living in the catchment area of participating Community Health Volunteers (CHVs) participated in a survey and provided a sample of infant stool for microbial analysis at enrolment into the trial. We defined eligibility of an infant as being 22 weeks ( $\pm$ 1 week) of age, as verified by birth registration card, who resided in one of the two study neighbourhoods. We enrolled caregivers who were responsible for care of the infant during the day and were at least 18 years of age.

### Patient and public involvement

Study participants and CHVs provided input on the goals, design and implementation of the Safe Start Study through prestudy knowledge dissemination meetings and formative research.<sup>21 25</sup>

### Outcomes

The outcomes for this study are: (1) 7-day caregiver-reported diarrhoea prevalence prior to enrolment, where diarrhoea is defined as three or more loose, watery stools in the previous 24 hours; and (2) the sum count of enteric pathogens detected in infant stool.

### Data and sample collection

The study was described in the caregiver's natural language, and a signed copy of the consent form was left for their records. On verification of eligibility and consent, caregivers were interviewed to collect data

about household socioeconomic conditions, access to water and sanitation infrastructure, animal ownership and hygiene practices. In anticipation that infant breast feeding and feeding practices may vary day to day and may be subject to response bias or recall bias, we asked caregivers about overall dietary history and foods given to infants in the last day. Caregivers were then provided several sterile commercially produced diapers and a sterile Ziploc bag, and asked to use these until the infant defecated. Diapers prevented cross-contamination via, for example, caregivers collecting infant faeces with dirt attached from the ground. Caregivers were instructed to fold the diaper, place it into the storage bag and store it in a cool dark place out of the reach of children and animals. The research team returned within 24 hours to collect the diaper, placed it in a cooler on ice packs and transported it to the laboratory within 5 hours of collection from the household. If the infant did not defecate in the first 24 hours, the team returned each day up to 5 days after enrolment to assess whether the infant had defecated. If no faeces could be collected, the infant was de-enrolled from the study.

### Nucleic acid extraction

Lab technicians unwrapped diapers in biosafety cabinets and used sterile stool collection scoops to transfer 200 mg of stool into Zymobiomics Shield Collection tubes, which were vortexed on a bead beater for 20 min and then processed according to the manufacturer's instructions for the ZymoBIOMICS DNA/RNA extraction mini-kit (Zymo Research, Irvine, California, USA). One molecular-grade water only sample was prepared each day of sample processing as a process contamination control. Approximately half ( $n=383$ ) of samples were spiked with  $3\ \mu\text{L}$  of  $1.8 \times 10^6$  CFU/ $\mu\text{L}$  of live bacteriophage MS2 to serve as a process control to assess for inhibition and efficiency in DNA and RNA recovery. Samples were transported on dry ice to the University of Iowa and stored at  $-80^\circ\text{C}$  until analysis.

### TaqMan Array card analysis

A total of 23 gene targets of pathogen of interest in the TaqMan assays were used to assess pathogen presence in infants. Pathogen gene targets were Adenovirus 40–41 Fibre, Adenovirus broad species Hexon, Rotavirus NSP3, Norovirus GI ORF 1–2, Norovirus GII ORF 1–2, *Aeromonas aerolysin* toxin *aerA*, *Campylobacter jejuni*/*C. coli* (*cadF*), Enterohemorrhagic *Escherichia coli* (EHEC) 0157 *rdh*, Enterococcal *E. coli* (EAEC) *aatA* and *aaiC*, Enteropathogenic *E. coli* (EPEC) *bfpA* and *eaec*, Enterotoxigenic *E. coli* (ETEC) *elt* and *est*, *Clostridioides difficile* *tcdB*, *Salmonella enterica* *ttt*, *Shigella* spp *virG*, *Vibrio cholerae* *hlyA*, *Giardia duodenalis* Assemblage A triosephosphate isomerase (TPI), *Giardia duodenalis* Assemblage A triosephosphate isomerase (TPI), *Cryptosporidium* spp 18S, *C. hominus* LIB13 and *C. parvum* LIB13.<sup>26 27</sup> For each sample,  $40\ \mu\text{L}$  of extract was mixed with  $5\ \mu\text{L}$  of nucleic acid-free water,  $50\ \mu\text{L}$  of 2X RT-buffer,  $0.6\ \mu\text{L}$  of 50 mg/

$\text{mL}$  bovine serum albumin (to reduce inhibition) and  $4\ \mu\text{L}$  of 25X AgPath enzyme from the AgPath-ID One-Step Reverse Transcription-Polymerase Chain Reaction (RT-PCR) kit (Thermo Fisher, Waltham, Massachusetts, USA) and pipetted into a well on a compartmentalised TaqMan card that included primer and probe assays in duplicate for each gene. TaqMan assays were completed in either a ViiA7 or QuantStudio8 instrument (Thermo Fisher, Waltham, Massachusetts, USA) for cycling conditions:  $45^\circ\text{C}$  for 20 min and  $95^\circ\text{C}$  for 10 min, followed by 40 cycles of  $95^\circ\text{C}$  for 15 s and  $60^\circ\text{C}$  for 1 min. A subset of samples that included both low and high Cq results were analysed on both machines and compared to confirm that results did not vary between machines, before proceeding with further use of both machines. We defined a sample as positive if a gene target amplified within a 35 Cycle threshold (Cq35). If multiple gene targets were used to detect one type of pathogen, amplification of either gene (EAEC *aaiC/aatA*, EPEC *bfpA/eaec*, ETEC *elt/est*) was considered positive for the general type of pathogen. The pathogen-specific detection patterns used to define the pathogen count variable for this analysis are described elsewhere.<sup>28</sup>

### Data analysis

Independent variables representing point sources of faeces that could contaminate the environment with enteric pathogens included household latrine design and location of the latrine, sharing of a latrine, ownership of domestic animals, whether animals are typically kept inside the household and observation of rodents or their faeces in the household. Improved sanitation was defined as a flush, pour flush, ventilated pit latrine or pit latrine with an impermeable slab, according to WHO/UNICEF JMP criteria.<sup>29</sup> Although they do not represent point sources of faeces, food ingredients and especially animal-based ones, as well as community drinking water sources could be sources of pathogens that originate outside the household. Thus, primary and secondary water sources and whether the primary water source is intermittent, and feeding the infant solid or liquid (other than breastmilk) foods were also defined as pathogen sources. A basic water source was defined as a piped tap to household or compound, a public tap, tube well, borehole, protected spring, protected hand dug well or rainwater, according to WHO/UNICEF Joint Monitoring Programme criteria, available within 30 min round trip.<sup>29</sup> Independent variables representing intermediate environmental conditions or behaviours that could modify pathogen transmission pathways and prevent exposure included household flooring (soil/surfaces), treating drinking water after collection, prior and recent (in the last day) breastfeeding status, presence of a handwashing station with soap and water and self-reported handwashing at critical times. Three of these critical handwashing times are behaviours that could modify hand cleanliness after touching human faeces or animals, while

three were focused specifically on modification of food sources being prepared within the household.

Potential confounders included in this analysis are marital status of caregiver, maternal education, household wealth, presence of multiple children under 5 years of age, infant preterm birth status, rotavirus vaccination status and prior or current breastfeeding practices. Principal components analysis with Promax rotation of 15 household assets (bicycle, motorbike, car, refrigerator, mobile phone, wrist or pocket watch, wall clock, radio, cassette or CD player, television, DVD player, microwave oven, presence of grates on the windows and doors, use of electricity for lighting and use of propane or electricity for cooking) resulted in a household wealth variable which was stratified into five quintiles.

All analysis were conducted with R software V.4.0.3 (R Foundation for Statistical Computing, Vienna, Austria). Associations between exposures and the binary indicator for 7-day caregiver-reported diarrhoea were evaluated using logistic regression. Bivariate associations were evaluated between each exposure and self-reported diarrhoea, and fully adjusted relationships between the exposures and the outcome was evaluated by running a single model with the confounder variables listed above and all exposure variables included. Associations between exposures and the ordinal categorical pathogen count variable were estimated using ordinal logistic regression. As with caregiver-reported diarrhoea, we evaluated bivariate and fully adjusted relationships.

In both fully adjusted models, multicollinearity among the exposure variables resulted in non-identifiable or nearly-non-identifiable model, resulting in the exclusion of some redundant exposure variables. In addition, some confounder variables had insufficient variability to have estimable effects and hence were removed. Finally, caregivers reporting a lack of breast feeding were so rare that this variable was excluded from the data. Logistic regression results are reported as ORs, with 95% CIs, for having diarrhoea, and ordinal logistic regression results are reported as ORs for having a higher count of pathogen types in stool versus fewer pathogen types. A random effect was initially included in models to adjust for spatial clustering, but was removed due to lack of variation in outcomes between villages.

Our moderation analyses tested whether hygiene of intermediate exposure pathways modified the relationships between point sources of pathogen contamination and infant health outcomes.

#### Hypothesis 1

Lack of access to a latrine with a barrier between users and excreta, and/or sharing unclean latrines with others can result in infant exposure to pathogens in human faeces<sup>15 20 30</sup> through faeces being tracked by feet onto household floors, where infants play and place objects or hands that have been on the floor in their mouth. We hypothesise that floor type moderates the effect of latrine access on pathogens in infants such that permeable dirt

floors that absorb liquids and sustain microbial growth, and that are harder to clean and disinfect, increase the association between human sanitation and pathogens in children compared with impermeable floors, like vinyl, concrete, or tile.

#### Hypothesis 2

Lack of or sharing latrine access can result in infant exposure to pathogens when caregivers do not wash hands after self-defecation or cleaning a child and then place hands in the infant's mouth. We hypothesise that hand-washing after defecation or child defecation moderates the effect of latrine access on infant health, such that human sanitation will be associated with pathogens in children among caregivers not washing hands after self or child defecation but not among caregivers who wash hands after self/child defecation.

#### Hypothesis 3

Like with latrines, an association between domestic animals or rodents and enteric infections in children could be caused by exposure of infants to floors contaminated with animal faeces.<sup>12</sup> We hypothesise that floor type can moderate this risk such that a dirt floor increases the association between domestic animals kept in or near the household or the presence of rodents and pathogens in children compared with households with impermeable floors.

#### Hypothesis 4

Zoonotic transmission of pathogens to infants could also occur through hands of caregivers who touch domestic animals or their faeces, and then place hands in the infant mouth. We hypothesise that washing hands after handling animals moderates the effect of animal or rodent presence in the household on pathogens in children such that caregivers not washing hands after handling animals increases the association between animals or rodents and pathogens in infants compared with caregivers who wash hands after touching animals.

#### Hypothesis 5

Animal or human faeces contamination on hands can be introduced into infant food during preparation or feeding and ingested by the infant while handwashing after defecation and animal handling could prevent transmission. We hypothesise that food-related handwashing also moderates the association between faeces sources (latrine access and the presence of animals or rodents in the household) and pathogens in infants such that caregivers (1) washing hands before preparing food, (2) washing hands before eating or (3) washing hands before feeding the infant decreases the association between latrines, domestic animals and rodents with pathogens in infants, compared with caregivers who do not wash hands at these times.

### Hypothesis 6

Infant supplemental foods include street foods prepared by vendors, raw fruits and prepackaged commercial products (eg, pasteurised milk) that could contain contamination. Cooking (eg, porridge), washing (eg, fruit) or storing these foods can mitigate or enhance these external food system-based pathogen transmission risks.<sup>31</sup> We hypothesise that access to a refrigerator for food storage moderates the pathway between supplemental foods and pathogens in infants such that a lack of refrigeration increases the association between supplemental food and pathogens in infants compared with households with refrigeration.

### Hypothesis 7

Similarly, reliance on unsafe water sources can increase the chances of pathogen infection through water, but water treatment can reduce or eliminate this contamination. We hypothesise that filtering, boiling or chlorinating water after collection moderates the association between type of water source and pathogens in infants such that not treating drinking water increases the association between household drinking water source and pathogens in infants compared with households who treat their drinking water.

These hypotheses were tested one at a time by adding an interaction term to the fully adjusted pathogen count model; the large number of potential confounders and exposure variables prohibited the simultaneous inclusion of all interaction terms being estimable. For each hypothesis listed above we ran an ordinal logistic regression model of the form

$$\text{Pathogens} = \text{Confounder variables} + \text{Exposure variables} + \text{Hypothesis – specific interaction term.}$$

Moderation effects were then tested using a likelihood ratio test comparing the additive model excluding the interaction term to the full model including the interaction term.

## RESULTS

### Characteristics of study participants

A total of 898 infant–caregiver pairs were enrolled in the study and completed the enrolment survey. The majority of caregivers were married, with secondary-level education, and had multiple young children (table 1). The preterm birth rate was high at 16% of infants. Most infants had received two doses of oral rotavirus vaccine, as verified by infant health registration cards. Access to a basic drinking water source and a latrine of improved design was common, although a third of caretakers used secondary water sources and nearly all households shared their latrine with multiple other families. Most households had cleanable (ceramic, tile, concrete or wood) floors or covered a dirt floor with rugs or vinyl. Of the 12.8% of households who owned animals, 82.5% (10.5% of all caregivers) kept their animals inside the household.

**Table 1** Characteristics of caregivers and their 5-month-old infants enrolled into the Safe Start Study in Kisumu, Kenya between 2018 and 2019

Socio-economic, environmental, and behavioural living conditions	% (n)
Caregiver is single vs Married	11.8 (105)
Caregiver has primary or no education vs secondary or higher education	39.1 (348)
Household wealth Quintile	
Poorest	19.6 (174)
Low-middle	19.7 (175)
Middle	20.7 (184)
Upper-middle	20.0 (178)
Highest quintile	20.0 (178)
More than one child under 5 years lives in household	35.6 (320)
Infant was born preterm (<37 weeks gestational age) vs not preterm (≥37 gestational weeks age)	16.0 (140)
Rotavirus vaccination series*	
No vaccination	1.2 (10)
One dose	1.6 (13)
Both doses	97.2 (803)
Infant ever breastfed vs never breastfed	99.8 (885)
Infant still breastfed vs weaned	99.7 (884)
Breastfed in the last day vs not breastfed in last day	57.5 (516)
Exposure variables	
Household relies on Improved latrine vs unimproved latrine or open defecation†	86.6 (770)
Sharing latrine	
Private household latrine	10.9 (98)
1–5 households share	17.7 (159)
>5 households Share latrine	71.4 (641)
Household owns animals vs does not own animals	12.8 (114)
Animals sleep inside household vs do not sleep in household	10.5 (94)
Rodents or rodent droppings observed vs no droppings	69.7 (620)
Live rodent, n=500	
Droppings, n=93	
Rodent tracks, n=25	
Other, n=1	
None, n=269	
Household Flooring	
Unfinished dirt floor	8.4 (75)
Unfinished floor covered with vinyl sheets or carpet	23.2 (206)
Finished floor (ceramic, concrete, tile, wood)	68.4 (608)

Continued



Table 1 Continued

Socio-economic, environmental, and behavioural living conditions	% (n)
Household primarily relies on basic drinking water source vs limited or unimproved source†	99.2 (885)
Household uses limited/unimproved secondary water source vs improved or no secondary source	32.0 (287)
Household does not treat drinking water vs treats water by boiling, Water Guard, or Aqua Tabs	43.8 (393)
Infant fed water in the last day vs did not give water	18.8 (169)
Infant ever fed milk	
Never	71.3 (632)
Ultra heat treated (UHT)	17.5 (155)
Fresh packed milk	8.9 (79)
Local cow or goat	1.0 (9)
Powdered formula	1.4 (12)
Infant has been fed solid food before vs never fed solid food	4.7 (42)
Infant fed milk in the last day	
UHT vs no UHT	11.2 (101)
fresh packed vs no fresh packed milk	2.0 (18)
local cow milk vs no local milk	0.8 (7)
ATM Milk vs no ATM milk	3.5 (31)
Milk Bar vs no Milk Bar	8.4 (75)
Other milk-based liquid vs no other liquid	4.0 (36)
Observed designated handwashing area with soap and water vs no handwashing area or no area with soap and water	6.6 (59)
Reported Handwashing at critical times	
After cleaning an infant that defecated	58.8 (528)
After self-defecation	88.0 (790)
After handling animals	12.0 (108)
Before preparing food	59.1 (531)
Before feeding child	60.5 (543)
Before eating	69.7 (626)
Total enrolled infants	n=889

\*Missing 64.  
†Defined according to WHO/UNICEF Joint Monitoring Programme criteria.  
UHT, ultra-high temperature.

Of the domestic species owned by households, chickens (73 of 89 households) and cats (45 of 49) represented the vast majority of animals kept in the household, although many other households kept animals in the compound yard (online supplemental table 1). Almost all caregivers reported that the infant was still breast fed, but only half reported giving the infant breastmilk in the last 24 hours.

Almost a third of infants consumed cow or goat milk by 5 months of age, with Long Life ultra-high temperature (UHT) pasteurised milk being the most common supplemental food. In the last day prior to the interview, a fifth of infants were given water and a third cow's milk, with UHT being the most common. Only ~5% of caregivers reported feeding infants solid foods. While 332 caregivers indicated they had a handwashing station in the household, only 74 of these had water present at the time of observation and only 59 (6.6% of all caregivers) had soap and water. Yet handwashing at critical times was reported by over 60% of caregivers for after self-defecation or infant defecation, and for preparing food, feeding the infant and personal eating. In contrast, just 12.0% reported washing hands after handling animals.

Variables excluded from multivariable exposure models due to low variation in subgroups included rotavirus vaccination, infant ever breast fed, infant still breast fed, primary drinking water source, infant has been fed solid food before and ever feeding the infant local milk or other milk type ever (UHT and fresh packed milk were only types with sufficient data). Although nearly all caregivers reported prior or ongoing breast feeding of the infant, only 57.5% of caregivers reported breast feeding the infant within the last day, so this variable was used to control for confounding of exposure–outcome relationships by breast feeding. Variables excluded due to collinearity included animals kept inside the household (strongly predicted by animal ownership) and feeding the infant milk in the last day (proxy for history of milk feeding).

### Exposure pathways and 7-day caregiver-reported diarrhoea history

A total of 862 out of 898 caregivers interviewed at enrolment had complete data for all exposure variables of interest and self-reported 7-day diarrhoea in infants. There were no discernible differences between individuals with and without full exposure data. Diarrhoea prevalence among 5-month-old infants in Kisumu at enrolment was 14.9% (134). Associations with potential confounders are reported in online supplemental table 2. In the full multivariable model, washing hands after handling animals and before eating were strongly associated with a fivefold and 2.27-fold lower odds of reported diarrhoea, respectively (table 2). In bivariate models, use of improved latrine was also protective against diarrhoea, but this relationship was weakened after adjusting for other wash conditions. Odds of diarrhoea were higher for infants fed UHT and fresh packed milk, but these relationships were weakened after adjusting for other exposure pathways.

### Exposure pathways and enteric pathogens in infants

Data on both the exposures of interest and pathogen detection outcome were available for 773 infants. Genes for at least one of 23 assessed enteric pathogens were detected in stool in 88.9% (n=707) of infants, with two

**Table 2** ORs and CIs from logistic regression models of caregiver-reported 7-day diarrhoea in 5-month infants and potential faeces sources and exposure pathways

Exposure pathways	Bivariate OR (95% CI)	Adjusted OR (95% CI)
Improved latrine vs unimproved latrine/open defecation	<b>0.52 (0.32 to 0.84)</b>	0.66 (0.38 to 1.17)
Sharing latrine		
Private household latrine	Ref	Ref
1–5 Households share latrine	1.64 (0.77 to 3.74)	1.71 (0.69 to 4.53)
>5 Households share latrine	1.61 (0.84 to 3.39)	1.62 (0.70 to 4.10)
Owens animals vs do not own animals	1.23 (0.71 to 2.04)	–
Animals sleep inside household vs do not sleep in household	1.40 (0.79 to 2.39)	1.64 (0.84 to 3.08)
Rodents or rodent droppings present vs not present	1.46 (0.96 to 2.26)	1.14 (0.69 to 1.92)
Household flooring		
Unfinished dirt floor	1.22 (0.62 to 2.25)	0.76 (0.35 to 1.55)
Covered unfinished floor	1.07 (0.68 to 1.65)	0.86 (0.52 to 1.39)
Finished floor	Ref	Ref
Limited/unimproved secondary water source vs improved or no secondary source	0.69 (0.45 to 1.03)	0.65 (0.40 to 1.03)
Drinking water is not treated in the household vs water is treated	1.34 (0.93 to 1.92)	1.24 (0.82 to 1.88)
Infant fed water in the last day vs no water	1.51 (0.97 to 2.31)	1.23 (0.69 to 2.15)
Infant fed milk in the last day		
UHT vs no UHT	<b>2.08 (1.25 to 3.37)</b>	–
Fresh packed vs no fresh packed milk	1.14 (0.26 to 3.52)	–
Local cow milk vs no local milk	0.95 (0.05 to 5.62)	–
Automated Milk Dispenser (ATM) vs not ATM milk	0.84 (0.25 to 2.19)	–
Milk bar vs no milk bar	1.77 (0.97 to 3.08)	–
Owens refrigerator for food storage vs no refrigerator	0.73 (0.40 to 1.26)	0.96 (0.44 to 2.00)
Handwashing area with soap and water vs no area or area with water only	0.89 (0.38 to 1.81)	1.24 (0.45 to 3.17)
Reported handwashing at critical times vs no handwashing at these times		
After cleaning an infant that defecated	0.78 (0.54 to 1.14)	1.13 (0.72 to 1.78)
After self-defecation	0.60 (0.37 to 1.02)	0.80 (0.45 to 1.46)
After handling animals	<b>0.25 (0.09 to 0.56)</b>	<b>0.20 (0.06 to 0.50)</b>
Before preparing food	0.77 (0.53 to 1.12)	1.33 (0.82 to 2.19)
Before feeding child	0.70 (0.48 to 1.01)	0.81 (0.51 to 1.28)
Before eating	<b>0.51 (0.35 to 0.74)</b>	<b>0.44 (0.26 to 0.73)</b>

For binary independent variables, the reference category is specified after 'vs'. '–' refers to a variable not estimated in multivariate model due to limited subgroup data or correlation with another exposure variable. Adjusted model includes independent variables of interest and confounders. Statistically significant results ( $p \leq 0.05$ ) are in bold type. UHT, ultra-high temperature.

or more pathogens detected in 66.9% ( $n=532$ ) of infants (median: 2 (25%–75%, IQR: 1–3)), based on a Cq35 threshold. The ordinal categorical model outcome variable was categorised as one of 0, 1, 2, 3, 4, 5 or  $\geq 6$  types of pathogens detected, due to scarce data for higher pathogen counts. Associations with confounders are reported in online supplemental table 3. In the multivariable ordinal regression model, living in a household with a vinyl-covered dirt floor was associated with a 1.67-fold lower chance of having more pathogen types detected in infant stool compared with infants in households with cement or tile floors (table 3). Infants fed any type of milk had a 1.42-fold to 2.32-fold increased chance of a higher count in pathogen diversity in infant stool versus infants never given milk. Keeping animals in the house posed a 1.54-fold higher odds of higher pathogen count compared with not keeping animals, while washing hands

after handling animals was counterintuitively associated with a 1.52-fold increase in odds compared with not washing hands after handling animals.

### Moderation of pathogen exposure pathways

After adjusting for confounders, moderation hypothesis tests for exposures and pathogen infections revealed:

1. There was no evidence that floor type modified the relationship between basic latrine access ( $p=0.93$ ) or latrine sharing ( $p=0.49$ ) and pathogen count in infants.
2. There was no evidence that handwashing after personal defecation ( $p=0.91$ ) or after cleaning a child ( $p=0.96$ ) modified the relationship between basic latrine access and pathogen count. There was moderate evidence for the hypothesis that handwashing after self-defecation ( $p=0.03$ ) or after child defecation ( $p=0.02$ ) modified the relationship between latrine sharing and pathogen

**Table 3** ORs and CIs from ordinal regression models of detecting one additional enteric pathogen type in faeces of 5-month infants and potential faeces sources and exposure pathways

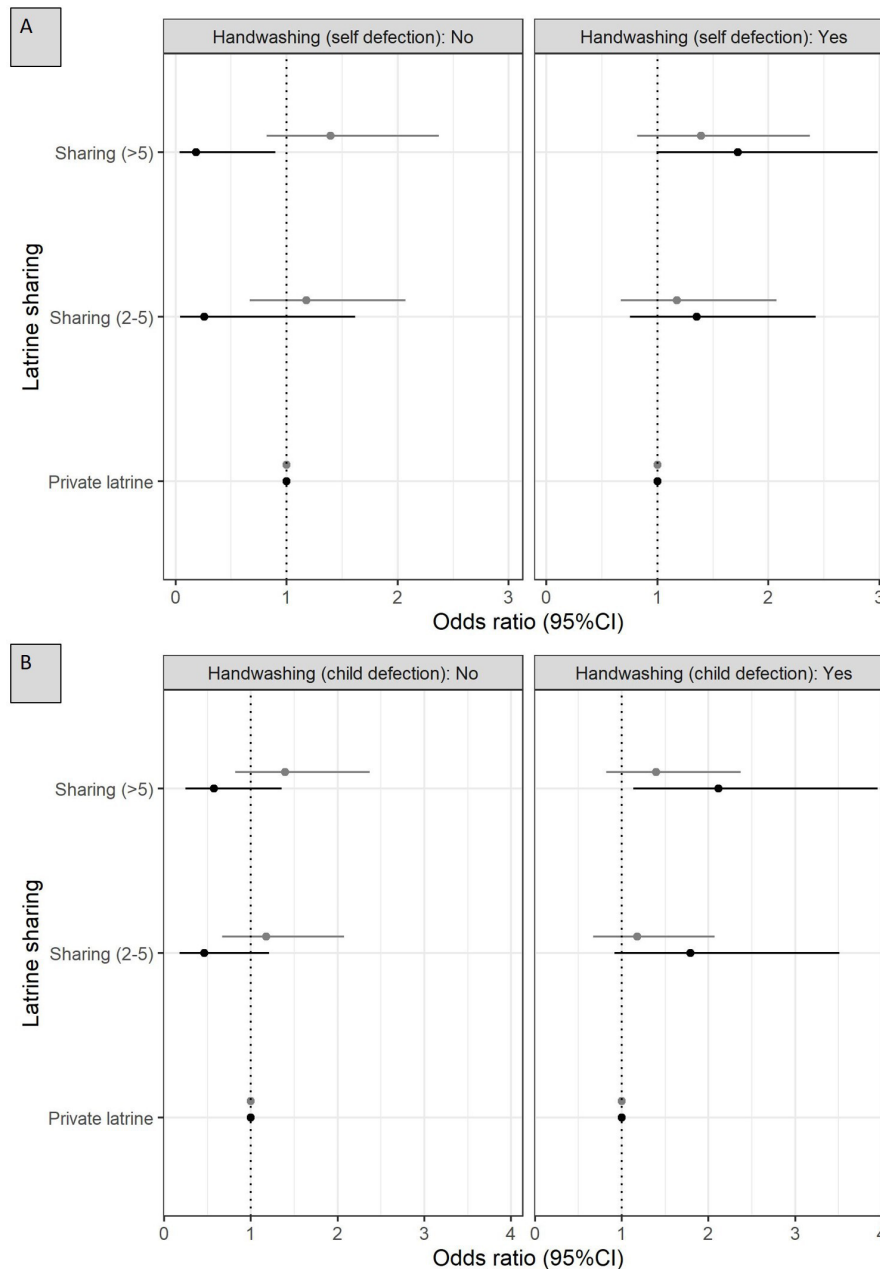
Exposure	Bivariate OR (95% CI)	Adjusted OR (95% CI)
Improved latrine vs unimproved/open defecation	<b>0.64 (0.45 to 0.92)</b>	0.79 (0.53 to 1.19)
Sharing latrine		
Private latrine	Ref	Ref
1–5 Households share latrine	1.09 (0.67 to 1.76)	1.18 (0.67 to 2.07)
>5 Households share latrine	1.43 (0.95 to 2.17)	1.18 (0.82 to 2.37)
Owens animals vs do not own animals	1.36 (0.95 to 1.95)	–
Animals sleep inside household vs do not sleep in household	<b>1.48 (1.01 to 2.17)</b>	<b>1.54 (1.01 to 2.34)</b>
Rodents or rodent droppings present vs not present	1.29 (0.99 to 1.69)	1.18 (0.76 to 1.61)
Household Flooring		
Unfinished fl.21 (0.77 to 1.90) <sup>o</sup>	1.21 (0.77 to 1.90)	0.87 (0.53 to 1.42)
Covered unfinished floor	<b>0.62 (0.45 to 0.83)</b>	<b>0.60 (0.45 to 0.87)</b>
Finished floor	Ref	Ref
Limited/unimproved secondary water source vs improved or no secondary source	0.98 (0.76 to 1.27)	0.98 (0.72 to 1.33)
Drinking water is not treated in the household vs water is treated	1.08 (0.84 to 1.38)	1.03 (0.78 to 1.36)
Infant fed water in the last day vs no water	1.30 (0.95 to 1.78)	0.98 (0.66 to 1.44)
Infant ever fed milk		
Never	Ref	Ref
UHT	<b>1.53 (1.10 to 2.14)</b>	1.42 (0.99 to 2.06)
Fresh packed milk	<b>1.53 (1.00 to 2.37)</b>	1.51 (0.96 to 2.38)
Other	2.74 (0.76 to 9.51)	2.32 (0.99 to 5.37)
Infant ever fed solid food vs no solid food	0.65 (0.37 to 1.15)	–
Infant fed milk in the last day		
UHT vs no UHT	<b>1.64 (1.11 to 2.44)</b>	–
Fresh packed vs no fresh packed milk	0.85 (0.35 to 2.07)	–
Local cow milk vs no local milk	2.09 (0.54 to 7.97)	–
Automated Milk Dispenser (ATM) vs not ATM milk	0.84 (0.42 to 1.67)	–
Milk bar vs no milk bar	1.13 (0.72 to 1.79)	–
Owens refrigerator for food storage vs no refrigerator	0.74 (0.52 to 1.05)	0.99 (0.62 to 1.57)
Handwashing area with soap and water vs no area or area with water only	0.71 (0.44 to 1.15)	0.93 (0.50 to 1.73)
Handwashing at critical times vs no handwashing at these times		
After cleaning an infant that defecated	0.78 (0.61 to 1.01)	0.94 (0.70 to 1.28)
After self-defecation	0.73 (0.49 to 1.08)	0.88 (0.57 to 1.36)
After handling animals	1.35 (0.94 to 1.93)	1.52 (0.98 to 2.37)
Before preparing food	0.81 (0.63 to 1.04)	0.78 (0.57 to 1.08)
Before feeding child	1.08 (0.84 to 1.40)	1.17 (0.87 to 1.56)
Before eating	0.99 (0.75 to 1.30)	1.07 (0.75 to 1.52)

For binary independent variables, the reference category is specified after 'vs'. '–' refers to a variable not estimated in multivariate model due to limited subgroup data or correlation with another exposure variable. Adjusted model includes independent variables of interest and confounders. Statistically significant results ( $p < 0.05$ ) are in bold type. UHT, ultra-high temperature.

count (figure 1A and B, respectively). Sharing a latrine with 2 to 5 (OR=1.35; 95% CI: 0.75 to 2.43) or >5 other households (OR=1.72; 95% CI: 0.99 to 2.98) was associated with higher pathogen count in infants relative to households with private latrines among caregivers who washed hands postdefecation. Sharing a latrine with 2 to 5 (OR=1.79; 95% CI: 0.92 to 3.51) or >5 households (OR=2.11; 95% CI: 1.13 to 3.94) versus private latrines

was also associated with higher pathogen detection in infants among caregivers who washed hands after child defecation. However, pathogen count was lower in households sharing a latrine with 2 to 5 (OR=0.26; 95% CI: 0.04 to 1.62) or >5 other (OR=0.18; 95% CI: 0.04 to 0.90) households among caregivers who did not wash after defecation, or after child defecation (OR=0.47; 95% CI: 0.18 to 1.21; and OR=0.58; 95% CI:

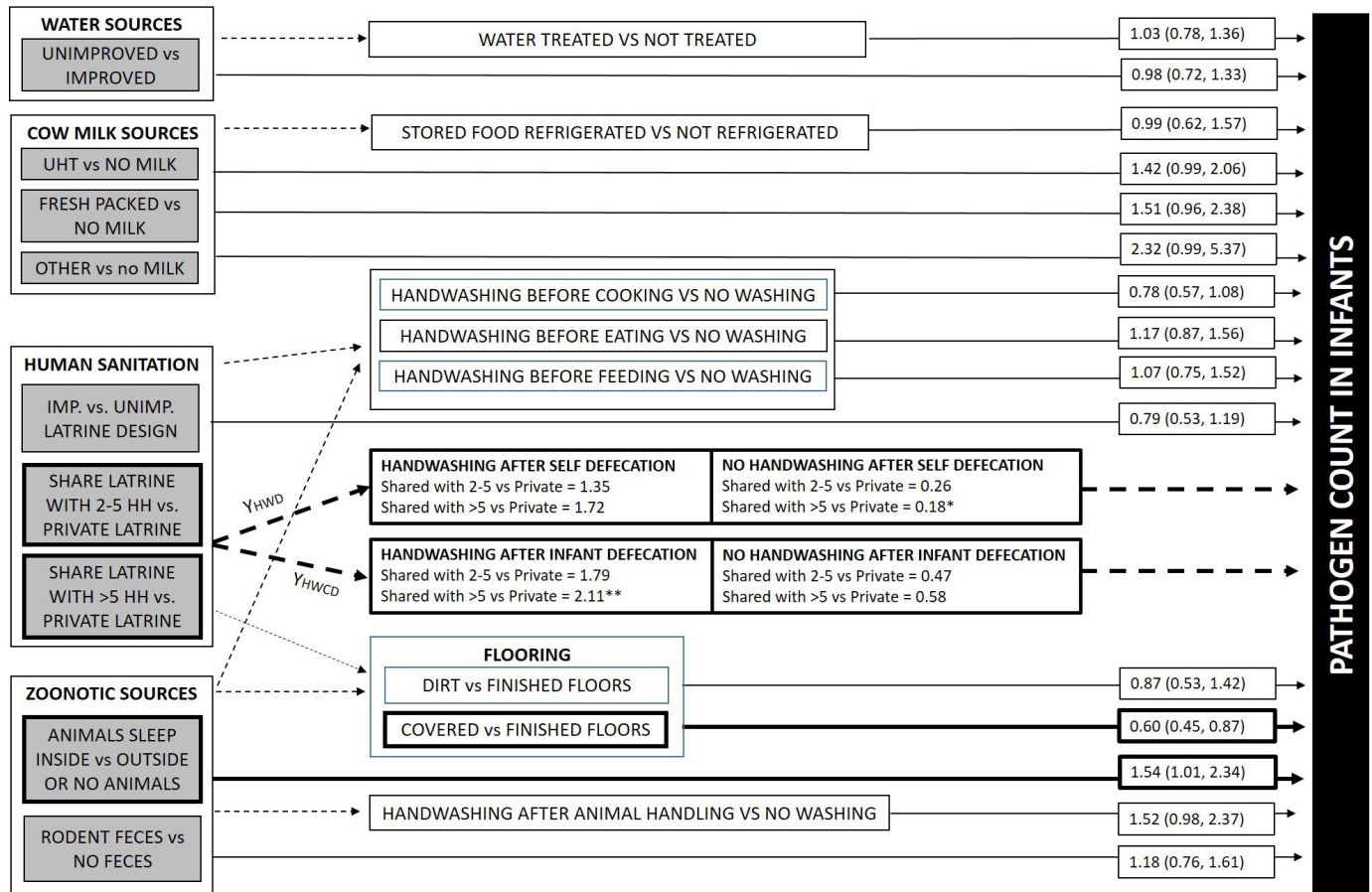




**Figure 1** Relationship between latrine sharing and pathogen count for handwashing after personal defecation and after child defecation (figure 1A and B, respectively). The OR estimate is indicated by a circle, with lines indicating the 95% CI in that estimate. The ORs shown in grey are from the main model with no interaction terms. The ORs in black are from the interaction model.

0.24 to 1.36, respectively). If sanitation is viewed as the effect modifier, then higher pathogen count was associated with not washing hands after defecation (OR=8.43; 95% CI: 1.71 to 41.56) or child defecation (OR=3.59; 95% CI: 1.35 to 9.51) in households with private latrines. The association between pathogen count and not washing hands after defecation or after child defecation was weaker in households sharing latrines with 2 to 5 (OR=2.18; 95% CI: 0.71 to 6.72 and OR=1.67; 95% CI: 0.78 to 3.57, respectively) or >5 (OR=1.54; 95% CI: 0.77 to 3.09, and 2.06; 95% CI: 1.07 to 3.97, respectively) other households.

3. There was no evidence that floor type modified the relationship between owning domestic animals, and by proxy keeping them in the household ( $p=0.60$ ) and pathogen count in infants.
4. There was no evidence that handwashing after handling animals modified the relationship between keeping animals inside and pathogen count ( $p=0.28$ ).
5. There was no evidence that washing hands before food preparation modified the association between pathogen count and latrine access ( $p=0.74$ ), sharing a latrine ( $p=0.49$ ), or animals sleeping inside ( $p=0.24$ ). Similarly, there was no evidence that washing hands be-



**Figure 2** Relationships between human sanitation, animal sanitation and public food and water sources that could introduce enteric pathogens into the household, and the housing conditions that can modify pathogen transmission, with the outcome of higher (vs lower) count of pathogen species detected in 5-month-old infants. Shaded boxes on the left represent the various hypothesised sources of enteric pathogens for this community in Kisumu, while clear boxes represent intermediate/modifying household conditions. Dashed lines indicate a hypothesised pathway from sources to intermediate household conditions, while non-dashed lines indicate hypothesised direct relationships with the outcome. The adjusted OR and 95% CIs reported in table 3 are reported here according to their hypothesised path. Since effect modification was detected for shared latrines and handwashing after self or child defecation, subgroup pathways indicated by bold dashed lines and associations are reported. The term  $Y_{HWD}$  refers to a subgroup analysis of shared sanitation and pathogen count conditional on whether the caregiver reported washing his/her hands after self-defecation. Similarly,  $Y_{HWCD}$  refers to a subgroup analysis of shared sanitation and pathogen count conditional on whether the caregiver reported washing his/her hands after cleaning a child that defecated. Statistically significant associations between pathogen sources or household conditions with pathogen are indicated by bolded lines and box outlines. UHT, ultra-high temperature.

fore feeding a child modified the association between pathogen count and latrine access ( $p=0.54$ ), sharing a latrine ( $p=0.61$ ) or owning animals ( $p=0.94$ ). And, there was no evidence that washing hands before self-eating modified the association between pathogen count and latrine access ( $p=0.15$ ), sharing a latrine ( $p=0.47$ ), or owning animals ( $p=0.63$ ).

- There was no evidence that access to household refrigeration modified the effect of milk type on pathogen count ( $p=0.17$ ).
- We could not investigate modification of treating water on unsafe primary water sources and pathogens, but among the 99.8% of households using a basic water source, there was no evidence that treating water by chlorination or boiling affected pathogen count in children ( $p=0.97$ ).

**Figure 2** summarises the relationships identified between human sanitation and animal presence conditions that could introduce pathogens into the household, the housing conditions that can transmit pathogens, and higher versus lower pathogen count in infants.

## DISCUSSION

The goal of this study was to identify risk factors linked to diarrhoea prevalence and enteric pathogen detection among young infants (<6 months old) in Kisumu, Kenya, and examine whether improved household hygiene environments (eg, flooring, refrigeration for safe food storage) and behaviours (eg, handwashing, water treatment) modified infant exposure to enteric pathogens from human and animal vectors. The very high prevalence

of enteric pathogen detection and codetection in Kisumu infants at 5 months age highlighted the critical need for exposure prevention interventions in early infancy, similar to other studies in low-income countries.<sup>2</sup> Our evidence implicated handwashing after handling animals and before eating and living in a household with vinyl-covered dirt floors (vs finished floors) as strong factors reducing the risk of self-reported diarrhoea at 5 months of age, while owning and cohabitation with animals and feeding infants processed cow milk were associated with increased risk of exposure to pathogens. Additionally, our evidence suggested that the benefits of private sanitation are limited if post-defecation handwashing is not practiced.

Latrine access was not a strong risk factor for diarrhoea or pathogens in infants after adjusting for other environmental and behavioural conditions, although sharing latrines was implicated in pathogen exposure when examined in the context of mitigating postdefecation handwashing behaviours. The interaction between these two conditions was influenced by strong benefits from postdefecation handwashing in households with private sanitation, and sustained pathogen exposure risks to infants from shared latrines, even when caregivers were washing hands after touching faeces. Infants usually have no direct contact with latrines designed for able-bodied children and adults, although shared latrines are a risk factor for moderate and severe diarrhoea in children across countries and studies.<sup>15 32 33</sup> Pathogen transmission from shared latrines could occur through pathways not examined in this study, like fly density, flooding, or soil on shoes or feet moving between the latrine and the compound.

Household flooring type has been minimally explored in enteric disease literature, but finished flooring has been linked to decreased risk from diarrhoea in Egypt,<sup>13</sup> and lower prevalence of *Ascaris lumbricoides* and soil-transmitted helminths in Kenya and Bangladesh in children under 5 years.<sup>30</sup> In our study, we used household flooring construction as a proxy for floor hygiene, based on the premise that covered or finished floors are easier to clean and less absorbent for sustaining pathogens. Type of household flooring did not modify the relationship between shared sanitation and pathogens in infants in this study. In fact, we counterintuitively observed higher pathogen counts in infants in households with the highest standard of finished flooring (mostly concrete) versus vinyl-covered dirt floors. This could mean concrete, which is porous, can sustain pathogen contamination similar to dirt floors or that floor type was a poor indicator of floor hygiene. Caregivers who cover dirt floors with vinyl or carpet may have been more concerned about hygiene issues posed by living in undeveloped household structures, and clean them more often than caregivers who are not concerned about dirt floors or who have finished floors that they perceive to be safe. We did not ask caregivers about floor cleaning practices, which might be a better indicator on the role of floors in transmission.

Owning and sharing living spaces with animals was associated with higher pathogen counts. Cohabitation between infants and animals in domestic settings makes infant contact with animals or their faeces likely, such as from ingestion of contaminated household drinking water.<sup>34</sup> Additionally, domestic animals belonging to one's neighbours often wander through spaces of households who do not own animals,<sup>35</sup> meaning ownership is not a comprehensive indicator of animal exposure. While handwashing after animal handling was not associated with pathogen detection count and did not moderate the strong association between owning animals and pathogen count, it was protective against infant diarrhoea. Owning animals may influence infant exposure via multiple pathways, with caregiver hands having relatively little importance as a pathway. We did not assess caregiver washing of infant hands, but infants place their own hands into their mouth frequently.<sup>19 36</sup> Touching of animals, animal faeces or contaminated floors followed by placing hands, dirt or faeces in the mouth may be a more important mechanism linking animal presence and pathogens in infants.

Information on the role of supplemental foods in infant infection in the first months of life is also scarce, possibly due to the assumption that breast feeding is the only or primary source of infant food at this stage. Like many self-reported behaviours where respondents are aware of sanctioned and disapproved behaviours, caregivers may over report breast feeding to avoid censure. While nearly all caregivers said they still breast feed 5-month infants, only 57% reported breast feeding within the last day, reinforcing that supplemental foods are important exposure pathways for Kisumu infants at ages when exclusive breast feeding is encouraged. Those not breast feeding relied mostly on cow's milk, especially Long Life UHT milk. Urbanisation of low-income cities has led to increased numbers of people living in low-income neighbourhoods and more women working outside of the home. More women working outside the home has led to increased demand for convenient supplemental foods, like packaged milk, that are palatable to very young infants and can be used by secondary caregivers for feeding.<sup>37-41</sup> Our data indicated packaged milk-based foods were sources of pathogen exposure, prompting the question as to whether the risks from UHT milk consumption derive from food supply chains or unhygienic household food management.

Animal-based foods, like cow milk, can pose a risk for zoonotic enteric pathogen transmission<sup>42</sup> although pasteurisation is an effective means for making milk safer. In a parallel study, we showed enteric pathogens do occur in processed milk products in Kenya, but households contribute more to infant food contamination than food products.<sup>31</sup> Pasteurised milk products may be risk factors in this study due to postpurchasing contamination from household surfaces, hands or utensils or storing milk for prolonged periods after opening. Food-related handwashing did not modify human and animal source-to-infant transmission, although washing before eating was



generally protective for infants. Other practices related to the boiling, feeding and storage of food were not studied here, but may explain or moderate food-borne pathogen exposure. Forthcoming manuscripts from the Safe Start Trial will reveal whether promotion of handwashing during food preparation and feeding, boiling food, storing foods in closed containers and feeding infants from dedicated containers can reduce diarrhoea and enteric pathogen detection rates in this study population.

Additional limitations of the study include inability to assess the causal relationships between exposure conditions and outcomes, although it is unlikely the pathogen status of infants—to which caregivers were unaware—caused a change in household environments or behaviours. We also relied on self-reported response about breast feeding, handwashing and feeding behaviours and 7-day diarrhoea symptoms in infants, which are all vulnerable to reporting bias. Self-reported handwashing was reported at much higher frequencies than observations of functional handwash stations would indicate. Enumerators visually verified as many conditions, such as water sources, latrines, handwashing station and flooring as feasible. In the case of handwashing, we may have undermeasured functional handwashing stations due to households storing soap and water in other secure locations. Postdefecation handwashing, especially among households relying on shared latrines, may occur outside the premises that were inspected. Additional strengths include the use of an ordinal distribution representing pathogens codetection patterns for identifying many exposure risk factors that would have remained masked by using a simplistic pathogen presence/absence outcome. The utility of using microbial diversity in pathogen contamination for distinguishing between high and low risk conditions in settings where exposure is the norm has also been demonstrated in environmental studies of soil, water and food.<sup>18 31</sup> We spiked around half of all stool specimens with MS2 virus as a sample transport and extraction process control. This was adequate for surveillance of cold chain failures that would affect batches of specimens transported en masse, but limited our ability to detect poor extraction efficiency among individual specimens. This study was performed among low-income periurban and urban Kenya households and the results may not be generalisable to rural households in Kenya, to higher-income households, and to low-income households in other countries. For example, animal ownership was relatively low in our study population compared with rural areas, where zoonotic transmission may be a more important source of pathogen transmission. Middle-income households are more likely to own a clean private latrine, a plentiful water supply and soap, as well as other indicators of household hygiene like a refrigerator. Such households are typically not recruited into diarrhoea disease studies unless they happen to live in a poorer neighbourhood but presumably, these living conditions prevent their infants from experiencing the high baseline level of enteric infections and diarrhoea as observed here.

In summary, our results point to a need for interventions that limit the presence of animals in households, increase handwashing after handling animals and using the latrine, and before eating, and promote safe management of milk-based infant foods to reduce the high population prevalence of enteric disease in <6-month infants. Interventions must target commercial packaged cow milk as one of the most common supplemental foods, including packaged milk products that are rapidly growing in popularity in urban populations.

#### Author affiliations

<sup>1</sup>Department of Occupational and Environmental Health, The University of Iowa, Iowa City, Iowa, USA

<sup>2</sup>Great Lakes University of Kisumu, Kisumu, Kenya

<sup>3</sup>African Population and Health Research Center, Nairobi, Kenya

<sup>4</sup>Department of Biostatistics, The University of Iowa, Iowa City, Iowa, USA

<sup>5</sup>Consultant, Austin, Texas, USA

<sup>6</sup>Department of Disease Control, London School of Hygiene & Tropical Medicine, London, UK

**Contributors** KKB and OC conceived of this study concept. JAOM, OC, KKB, JDA and RD designed the parent study. SS and KT collected data. JAOM and KKB supervised data collection. JAOM managed data assimilation and curation. KKB and DS performed the analysis and drafted the manuscript. AM provided additional statistical review. All authors reviewed and submitted comments on the final manuscript. KKB is responsible as guarantor for the finished work, including the conduct of the study, access to the data, and the decision to publish.

**Funding** The SHARE Research Consortium (Grant Number ITDCHA2310), funded by the United Kingdom Department for International Development (DFID) funded the Safe Start Trial.

**Competing interests** None declared.

**Patient and public involvement** Patients and/or the public were involved in the design, or conduct, or reporting or dissemination plans of this research. Refer to the Methods section for further details.

**Patient consent for publication** Not applicable.

**Ethics approval** The study was approved by the scientific and ethical review committees at the Great Lakes University of Kisumu (Ref. No. GREC/010/248/2016), London School of Hygiene and Tropical Medicine (Ref. No. 14695) and University of Iowa (IRB ID 201804204). Participants gave informed consent to participate in the study before taking part.

**Provenance and peer review** Not commissioned; externally peer reviewed.

**Data availability statement** Data are available upon reasonable request. This study includes enrolment data from a clinical trial. De-identified human data will be made available upon request once the primary results of the trial are published.

**Supplemental material** This content has been supplied by the author(s). It has not been vetted by BMJ Publishing Group Limited (BMJ) and may not have been peer-reviewed. Any opinions or recommendations discussed are solely those of the author(s) and are not endorsed by BMJ. BMJ disclaims all liability and responsibility arising from any reliance placed on the content. Where the content includes any translated material, BMJ does not warrant the accuracy and reliability of the translations (including but not limited to local regulations, clinical guidelines, terminology, drug names and drug dosages), and is not responsible for any error and/or omissions arising from translation and adaptation or otherwise.

**Open access** This is an open access article distributed in accordance with the Creative Commons Attribution Non Commercial (CC BY-NC 4.0) license, which permits others to distribute, remix, adapt, build upon this work non-commercially, and license their derivative works on different terms, provided the original work is properly cited, appropriate credit is given, any changes made indicated, and the use is non-commercial. See: <http://creativecommons.org/licenses/by-nc/4.0/>.

#### ORCID iDs

Kelly K Baker <http://orcid.org/0000-0002-3774-2628>

John David Anderson <http://orcid.org/0000-0003-0840-9890>

Oliver Cumming <http://orcid.org/0000-0002-5074-8709>

## REFERENCES

- Rogawski McQuade ET, Platts-Mills JA, Gratz J. Impact of water quality, sanitation, handwashing, and nutritional interventions on enteric infections in rural Zimbabwe: the sanitation hygiene infant nutrition efficacy (shine) trial. *J Infect Dis* 2019.
- Platts-Mills JA, Babji S, Bodhidatta L, et al. Pathogen-Specific burdens of community diarrhoea in developing countries: a multisite birth cohort study (MAL-ED). *Lancet Glob Health* 2015;3:e564–75.
- Rogawski ET, Liu J, Platts-Mills JA, et al. Use of quantitative molecular diagnostic methods to investigate the effect of enteropathogen infections on linear growth in children in low-resource settings: longitudinal analysis of results from the MAL-ED cohort study. *Lancet Glob Health* 2018;6:e1319–28.
- Shen S-S, Qu X-Y, Zhang W-Z, et al. Infection against infection: parasite antagonism against parasites, viruses and bacteria. *Infect Dis Poverty* 2019;8:49.
- Walson JL, Berkley JA. The impact of malnutrition on childhood infections. *Curr Opin Infect Dis* 2018;31:231–6.
- Null C, Stewart CP, Pickering AJ, et al. Effects of water quality, sanitation, handwashing, and nutritional interventions on diarrhoea and child growth in rural Kenya: a cluster-randomised controlled trial. *Lancet Glob Health* 2018;6:e316–29.
- Luby SP, Rahman M, Arnold BF, et al. Effects of water quality, sanitation, handwashing, and nutritional interventions on diarrhoea and child growth in rural Bangladesh: a cluster randomised controlled trial. *Lancet Glob Health* 2018;6:e302–15.
- Watson JA, Ensink JHJ, Ramos M, et al. Does targeting children with hygiene promotion messages work? the effect of handwashing promotion targeted at children, on diarrhoea, soil-transmitted helminth infections and behaviour change, in low- and middle-income countries. *Trop Med Int Health* 2017;22:526–38.
- Islam MS, Mahmud ZH, Gope PS, et al. Hygiene intervention reduces contamination of weaning food in Bangladesh. *Trop Med Int Health* 2013;18:250–8.
- Kirk MD, Angulo FJ, Havelaar AH, et al. Diarrhoeal disease in children due to contaminated food. *Bull World Health Organ* 2017;95:233–4.
- Emerson PM, Lindsay SW, Alexander N, et al. Role of flies and provision of latrines in trachoma control: cluster-randomised controlled trial. *Lancet* 2004;363:1093–8.
- Zambrano LD, Levy K, Menezes NP, et al. Human diarrhea infections associated with domestic animal husbandry: a systematic review and meta-analysis. *Trans R Soc Trop Med Hyg* 2014;108:313–25.
- Wright CE, el Alamy M, DuPont HL, et al. The role of home environment in infant diarrhea in rural Egypt. *Am J Epidemiol* 1991;134:887–94.
- Chard AN, Baker KK, Tsai K, et al. Associations between soil-transmitted helminthiasis and viral, bacterial, and protozoal enteroinfections: a cross-sectional study in rural Laos. *Parasit Vectors* 2019;12:216.
- Baker KK, O'Reilly CE, Levine MM, et al. Sanitation and Hygiene-Specific risk factors for moderate-to-severe diarrhea in young children in the global enteric multicenter study, 2007–2011: case-control study. *PLoS Med* 2016;13:e1002010.
- Simiyu S, Swilling M, Cairncross S, et al. Determinants of quality of shared sanitation facilities in informal settlements: case study of Kisumu, Kenya. *BMC Public Health* 2017;17:68.
- Ritter RL, Peprah D, Null C, et al. Within-Compound versus public Latrine access and child feces disposal practices in low-income neighborhoods of Accra, Ghana. *Am J Trop Med Hyg* 2018;98:1250–9.
- Medgyesi D, Sewell D, Senesac R, et al. The landscape of enteric pathogen exposure of young children in public domains of low-income, urban Kenya: the influence of exposure pathway and spatial range of play on multi-pathogen exposure risks. *PLoS Negl Trop Dis* 2019;13:e0007292.
- Medgyesi D, Brogan J, Sewell D, et al. Where children play: young child exposure to environmental hazards during play in public areas in a transitioning internally displaced persons community in Haiti. *Int J Environ Res Public Health* 2018;15:1682.
- Harris M, Alzua ML, Osbert N, et al. Community-Level sanitation coverage more strongly associated with child growth and household drinking water quality than access to a private toilet in rural Mali. *Environ Sci Technol* 2017;51:7219–27.
- Mumma J, Simiyu S, Aseyo E, et al. The safe start trial to assess the effect of an infant hygiene intervention on enteric infections and diarrhoea in low-income informal neighbourhoods of Kisumu, Kenya: a study protocol for a cluster randomized controlled trial. *BMC Infect Dis* 2019;19:1066.
- Tsai K, Simiyu S, Mumma J, et al. Enteric pathogen diversity in infant foods in low-income neighborhoods of Kisumu, Kenya. *Int J Environ Res Public Health* 2019;16. doi:10.3390/ijerph16030506. [Epub ahead of print: 12 02 2019].
- Davis E, Cumming O, Aseyo RE, et al. Oral contact events and caregiver hand hygiene: implications for Fecal-Oral exposure to enteric pathogens among infants 3–9 months living in informal, Peri-Urban communities in Kisumu, Kenya. *Int J Environ Res Public Health* 2018;15. doi:10.3390/ijerph15020192. [Epub ahead of print: 24 01 2018].
- Aseyo RE, Mumma J, Scott K, et al. Realities and experiences of community health volunteers as agents for behaviour change: evidence from an informal urban settlement in Kisumu, Kenya. *Hum Resour Health* 2018;16:53.
- Simiyu S, Czerniewska A, Aseyo ER, et al. Designing a food hygiene intervention in low-income, Peri-Urban context of Kisumu, Kenya: application of the trials of improved practices methodology. *Am J Trop Med Hyg* 2020;102:1116–23.
- Johnson HL, Liu J, Fischer-Walker C, et al. Estimating the distribution of causes of death among children age 1–59 months in high-mortality countries with incomplete death certification. *Int J Epidemiol* 2010;39:1103–14.
- Liu J, Kabir F, Manneh J, et al. Development and assessment of molecular diagnostic tests for 15 enteropathogens causing childhood diarrhoea: a multicentre study. *Lancet Infect Dis* 2014;14:716–24.
- Liu J, Gratz J, Amour C, et al. A laboratory-developed TaqMan array card for simultaneous detection of 19 enteropathogens. *J Clin Microbiol* 2013;51:472–80.
- UNICEF. WHO. Progress on sanitation and drinking water - 2015 update and MDG assessment. Available: [http://www.wssinfo.org/fileadmin/user\\_upload/resources/JMP-Update-report-2015\\_English.pdf](http://www.wssinfo.org/fileadmin/user_upload/resources/JMP-Update-report-2015_English.pdf)
- Benjamin-Chung J, Crider YS, Mertens A, et al. Household finished flooring and soil-transmitted helminth and Giardia infections among children in rural Bangladesh and Kenya: a prospective cohort study. *Lancet Glob Health* 2021;9:e301–8.
- Hoffmann V, Simiyu S, Sewell D. Influence of milk product safety and household food hygiene on bacterial contamination of infant food in peri-urban Kenya. *MedRxiv* 2020.
- Heijnen M, Cumming O, Peletz R, et al. Shared sanitation versus individual household latrines: a systematic review of health outcomes. *PLoS One* 2014;9:e93300.
- Fuller JA, Clasen T, Heijnen M, et al. Shared sanitation and the prevalence of diarrhea in young children: evidence from 51 countries, 2001–2011. *Am J Trop Med Hyg* 2014;91:173–80.
- Barnes AN, Anderson JD, Mumma J, et al. The association between domestic animal presence and ownership and household drinking water contamination among peri-urban communities of Kisumu, Kenya. *PLoS One* 2018;13:e0197587.
- Barnes AN, Mumma J, Cumming O. Role, ownership and presence of domestic animals in peri-urban households of Kisumu, Kenya. *Zoonoses Public Health* 2018;65:202–14.
- Kwong LH, Ercumen A, Pickering AJ, et al. Hand- and Object-Mouthing of rural Bangladeshi children 3–18 months old. *Int J Environ Res Public Health* 2016;13. doi:10.3390/ijerph13060563. [Epub ahead of print: 04 06 2016].
- Mumma JAO, Cumming O, Simiyu S. Infant food hygiene and childcare practices in context: findings from an urban informal settlement in Kenya. *Am J Trop Med Hyg* 2019.
- Haider R, Begum S. Working women, maternity entitlements, and breastfeeding: a report from Bangladesh. *J Hum Lact* 1995;11:273–7.
- Rea MF, Venâncio SI, Batista LE, et al. Determinants of the breastfeeding pattern among working women in São Paulo. *J Hum Lact* 1999;15:233–9.
- Aggarwal A, Arora S, Patwari AK. Breastfeeding among urban women of low-socioeconomic status: factors influencing introduction of supplemental feeds before four months of age. *Indian Pediatr* 1998;35:269–73.
- Hirani SAA, Karmaliani R. The experiences of urban, professional women when combining breastfeeding with paid employment in Karachi, Pakistan: a qualitative study. *Women Birth* 2013;26:147–51.
- Kirk MD, Pires SM, Black RE, et al. World Health organization estimates of the global and regional disease burden of 22 foodborne bacterial, protozoal, and viral diseases, 2010: a data synthesis. *PLoS Med* 2015;12:e1001921.