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Food hygiene intervention to improve food hygiene behaviours, and reduce food contamination in Nepal: an exploratory trial

By
Om Prasad Gautam

Thesis submitted in accordance with the requirements for the degree of
Doctor of Philosophy (PhD) of the
University of London
September 2015

Disease Control Department (DCD)
Faculty of Infectious and Tropical Diseases (ITD)
London School of Hygiene and Tropical Medicine (LSHTM)
University of London

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ABSTRACT

Objectives: This thesis describes a study that designed, implemented and assessed the effect of a food hygiene intervention on mothers’ food hygiene practices as primary outcomes, and the impact of the interventions on the level of microbiological contamination in food as a secondary outcome. An additional objective was to explore whether food hygiene interventions can be integrated into nutrition, health and water, sanitation and hygiene (WASH) strategies and programmes in Nepal.

Methods: The theoretical and practical approach of Behaviour Centred Designed was employed. In step-A: Assess, a systematic literature review was conducted to identify sectoral knowledge and programmatic gaps on food hygiene and sectoral policy documents analysis was done as part of gray literature review to ascertain whether food hygiene interventions can be integrated into Nepal’s health, WASH and nutrition programming. In step-B: Build, formative research was carried out to identify and prioritise key food hygiene behaviours, and inform the intervention design. In step-C: Create, a scalable food hygiene intervention package was designed and tested using a novel approach to behaviour change employing emotional drivers and changing behaviour settings. In Step-D, the intervention was Delivered by female food hygiene motivators in four intervention clusters over a period of three months while four clusters acted as a control group in a rural area of Nepal. In Step E: Evaluate, a Cluster Randomized, Before-After study with Control (BAC) was employed. Behavioural outcomes were measured before and after the intervention in 239 households with a child aged 6-59 months in four intervention and four control clusters. The microbiological contamination in commonly-used child foods was measured in a sub-sample of 80 households.

Results: Systematic review identified the need for research into improving food hygiene behaviour to reduce contamination in food and improve health outcomes in low-income settings. Nepal’s policy environment can enable the integration of food hygiene promotion within ongoing WASH, nutrition and health programmes. Five key food hygiene risk behaviours were prioritized, and likely determinants of behaviour change were identified through formative research. The motivational and creative food hygiene intervention package was designed and delivered in community settings. The intervention was effective in significantly improving multiple food hygiene behaviours. The 5 targeted food hygiene
behaviours were rare at baseline. Forty five days after the 3 months intervention, key behaviours were more common in the intervention group than in the control group (43% [SD14] vs. 2% [SD 2], p=0.02). The difference of differences was an increase in mean proportion of 42% (p=0.02). The intervention appeared to be equally effective in improving all five behaviours and in all intervention clusters. Commonly-used child foods from the intervention and control clusters were heavily contaminated with total coliforms and E. coli during child feeding at baseline and the behavioural intervention was effective in significantly reducing the contamination in the intervention group during follow-up. After adjusting for baseline, the intervention reduces the mean coliform count by -2.00 log_{10} cfu/gm (p=0.020) and E. coli by -1.00 log_{10} cfu/gm (p=0.083). Contamination in water was low as compared to food at baseline and did not improve after the intervention. 

**Conclusion:** This systematic approach employing emotional drivers and change in behavioural settings substantially altered multiple food hygiene behaviours and reduced microbial contamination in commonly-used child food in Nepal. Ingestion of microbes by children can only be eliminated if the food hygiene intervention deals with all key behaviours. This study responds to an important evidence gap. Current evidence, to which this study has contributed, is sufficient to merit prioritisation of food hygiene by those concerned with designing more effective WASH, health and nutrition programmes. The work suggests that interventions on food hygiene should have a higher priority than those on water treatment, which is not currently the case in development projects. The BCD approach provided a theory of change and a useful process framework for the design, delivery and evaluation of the intervention. Additional research is needed to test the impact of such interventions on both behaviour and health outcomes. Further tests would help to determine if the intervention can be replicated in diverse settings and at large scale and so add value to existing programmatic responses to poor WASH and undernutrition. The implementation of a tested food hygiene package through a scalable pilot was identified as a next step towards demonstrating the delivery of hygiene interventions through existing service delivery mechanisms in Nepal.
ACKNOWLEDGEMENTS

Firstly, my special gratitude and sincere appreciation goes to my PhD supervisor, Dr Valerie Curtis, Director, Hygiene Centre at London School of Hygiene and Tropical Medicine (LSHTM) for her scientific guidance and support, and for being a good mentor throughout the process.

I would like to thank all advisory committee members namely Prof Sandy Cairncross, Dr Wolf-Peter Schmidt, Dr Sue Cavill and Prof Claudio E. Lanata for their continuous support and technical inputs at various stages of my research work. I am also indebted to the input and inspiration provided by upgrading committee members including Dr Jayne Webster, Dr Jim McCambridge, and Dr Joe Brown. I am grateful to many LSHTM ITD/DCD staff members and colleagues including Dr Robert Aunger, Dr Jeroen Ensink, Dr Jane Bruce, Mrs Eileen Chappell, Ms Alexandra Chitty, Mrs Joanna Esteves Mills and Ms Chantelle Thomas for their appropriate support. I am very thankful to my colleague Ms Yael Velleman for her proofreading support. I would like to thank Mr Oliver Cumming and Ms Katie Greenland for their feedback on systematic review. Thanks are due also to Prof Richard Carter and Mr Erik Harvey for understanding the importance of food hygiene for the WASH sector and for committing partial funding on behalf of WaterAid for this work.

I would like to thank the UK Department for International Development (DFID) for funding my PhD and research through the SHARE Consortium, and to WaterAid UK for co-funding my field work. I am grateful for the support of the Ministry of Health and Population, Child Health Division in Nepal for their collaboration and cooperation for the trial. Thanks go also to SHARE for providing me the opportunity to share work with a wider audience through hosting my blog on the SHARE website.

Special thanks goes to creative team members Mr Anil K. Thaman, Mr Sujain Choudhary, Mr Khem Raj Shrestha, Mr Buddhiman Shrestha, Mr Pariskrit Shrestha, Mr Naseeb Pandey and the production team for their involvement and support in designing the intervention package.

I would like to thank all the mothers (especially mothers of young children) from the study area of Kavre district, Nepal, who participated and contributed to different phases of the study.
Thanks to all key informants from the Ministry of Health and Population, Ministry of Urban Development, Ministry of Education, Ministry of Agriculture, and representatives from member organizations of the International Association of NGOs and donor agencies, who participated in the policy discussions and debates.

Thanks to all social leaders, teachers, female food hygiene motivators, female community health volunteers, food hygiene observers, video recorders, research assistants (Ms Kamala KC, Ms Nirmala Bista, Ms Prakriti Pant, Ms Lauren D’mello-Guyett), project coordination committee members (including Mr Achyut KC, Ms Biddhya Tamang, Mr Thamu Narayan Bhandari), local health institutions and VDC staff for their involvement and support for the study in field.

Thanks to Dr Shyam Raj Upreti, Director, Child Health Division, Ministry of Health and Population for his continuous cooperation and support on behalf of MoHP Nepal. I would like to thank Ex. Hon’ble health member, National Planning Commission Dr Chet Raj Pant, Ex. Hon’ble member of Kavre constituency Mr Krishna Prasad Sapkota and Ex. Country representative for WaterAid in Nepal Mr Ashutosh Tiwari for their attendance in the programme and their encouragement.

Thanks go to Dr Sameer Dixit and his team from the Centre for Molecular Dynamics Nepal (CMDN), Intrepid Lab for their support in assessing food microbiology, and ENPHO lab for their work to assess water samples. Thanks also to the District Public Health Office, Kavre for their coordination support.

I thank my wife Mrs Shreejana Dhungana and son Shishir Gautam for their inspiration, continuous support and cooperation throughout the PhD process. I am indebted to my parents who have always inspired and motivated me.

Finally, I would like to extend my earnest gratitude to many others who supported me during various phase of the study tenure.

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<td>BCD</td>
<td>Behaviour Centred Design</td>
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<td>BCP</td>
<td>Behavioural Control Point</td>
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<td>CBO</td>
<td>Community Based Organization</td>
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<td>CCP</td>
<td>Critical Control Point</td>
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<td>CDC</td>
<td>Centre for Disease Control</td>
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<tr>
<td>CFU</td>
<td>Colony-forming unit</td>
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<td>CHD</td>
<td>Child Health Division</td>
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<td>CLTS</td>
<td>Community Led Total Sanitation</td>
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<td>CMDN</td>
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<td>HACCP</td>
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<td>Information Education and Communication</td>
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<td>IFH</td>
<td>International Scientific Forum on Home Hygiene</td>
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<td>Acronym</td>
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<td>WASH</td>
<td>Water, Sanitation and Hygiene</td>
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1. Introduction

Globally, the epidemiological transition is shifting the burden of diseases from communicable to non-communicable, however low-income countries are afflicted with multiple problems. Things are getting better to reduce the child morbidity and mortality, but we have not cracked all of the problems. Food hygiene is one that has been neglected, yet it could be one of the biggest outstanding problems we still have to solve. This thesis highlights where the problems are and tried narrowing both the evidence and programmatic gaps on food hygiene.

This chapter includes the study background, the evidence gap and knowledge on food hygiene, justification, aims, objectives, research questions, framework to guide the study, and overall chapter outlines.

1.1 Background

1.1.1 Why food hygiene matters
Poor food hygiene is likely to contribute gastro-enteric infections, food-borne diseases, diarrhoeal diseases and childhood undernutrition among infants and young children, however the magnitude of the problem is difficult to quantify due to lack of evidences from low-income settings. Various interventions can prevent the transmission of disease-causing agents including improving the quality (and quantity) of water[1, 2], sanitation[2, 3], handwashing with soap[4-6] and exclusive breastfeeding[7]. However, interventions to address disease transmission through food have been largely overlooked in research and programming in low-income settings. Simple and replicable food hygiene interventions are needed that can be implemented within water, sanitation and hygiene (WASH), health, and nutrition programmes.

1.1.2 Food-borne infections are a universal problem
Preventable and treatable food-borne diseases are a major cause of illness in high and low-income settings alike. Whilst it is hard to ascribe exact numbers to the problem, it is estimated that each year 1 in 6 Americans (or 48 million people) falls ill from foodborne diseases, with 128,000 hospitalized, and 3,000 dying[8]. In the United Kingdom, 1 in 5 people develop gastrointestinal illnesses annually; in 2007 approximately one million people suffered a foodborne illness, which led to 20,000 receiving hospital treatment and 500 deaths, at a total
cost of nearly £1.5 billion[9]. UK data also suggest that 39% of outbreaks are due to inappropriate food storage, 31% are due to inadequate cooking, and 20% are due to cross-contamination in the kitchen[10]. In Australia, 5.4 million fall sick annually from eating contaminated food and up to 20% of this illness results from unsafe food handling behaviours[11].

In low income settings data are far more scarce. The UN Food and Agriculture Organization (FAO) in 2003 summarized that the data for low-income settings (developing countries) are particularly weak and out-dated, although it is recognised that a wide range of bacterial infections are widespread[12]. Most knowledge about food-borne infections in low-income settings is therefore based on expert opinion and biological plausibility, rather than field data[13]. Very few, if any, disease outbreaks have been reported as caused by food contamination in developing countries, indicating poor availability of data regarding the incidence foodborne diseases, rather than the absence of such diseases[14]. Further gathering of field-based evidence while putting in place preventive measures in low-income settings is therefore vital. In 2006, the World Health Organization launched the Initiative to Estimate the Global Burden of Foodborne Diseases[15]; its final report will be released in 2015.

**1.1.3 Diarrhoeal diseases burden and importance of food hygiene:**

Diarrhoeal diseases are the second leading cause of death from disease among children under five globally [16]. In 2011, preventable diarrhoeal episodes lead to an estimated 700,000 child deaths[17]. According to some experts, up to 70% of diarrhoeal episodes in developing countries are caused by pathogens transmitted through food[18, 19]. Contaminated weaning foods are potentially a major contributor in low-income settings[20], although observational studies were inconclusive[14]. Around 72% of death associated with diarrhoea happen in the first two years of life [17] and diarrhoea risk increases during the infant weaning period[17, 21, 22], with potentially long-lasting effects, as in many countries weaning and complementary foods are prepared under unhygienic conditions and infants may be exposed to infective doses of food-borne pathogens[20], including bacteria, viruses and parasites. Previous review revealed 23% increased risk of diarrhoea in children when unhygienic child stool disposal was practised[23].

Various pathogens are implicated in food-borne infections and diarrhoea, including *Salmonella, Vibrio cholera, Shigella, Escherichia coli (E. coli), Campylobacter, Norovirus,*
Streptococci, Bacillus cereus and Clostridium botulinum\cite{24, 25} and enteric viruses such as rotavirus\cite{26, 27}. There is significant variation in the type of pathogens isolation by age-groups and geographic settings; in the Netherlands, Norovirus and Salmonella were found to be the most common causes of foodborne outbreaks\cite{28}, whereas in Bolivia, enteropathogenic E. coli, Shigella and Vibro cholera were commonly detected\cite{29}. Pathogenic strains of E. coli alone may account for up to 25\% of diarrhoeal disease episodes in developing countries\cite{30}. The most common cause of severe and fatal diarrhoea worldwide is rotavirus\cite{17}. The symptoms of food-borne illness range from mild to life-threatening. While diarrhoea is the most common gastrointestinal illness, kidney and liver failure, brain and neural disorders, and even death can also result from food contamination.

In the last 20 years, there has been significant reduction in the prevalence of childhood diarrhoea mortality worldwide. However, in many low-income countries the disease burden remains high and there are persistent gaps in knowledge surrounding the role of food hygiene in diarrhoea prevention to effectively inform public health policy\cite{31}. Among the recommended strategies for controlling diarrhoeal diseases in developing countries, food hygiene promotion has not been a priority. Previous initiatives for the prevention of diarrhoeal diseases and reviews have focused mostly on water supply, sanitation and handwashing with soap interventions, but what is lacking is knowledge about the role and effectiveness of food hygiene intervention.

**1.1.4 Childhood undernutrition and importance of food hygiene:**
Repeated episodes of diarrhoea in early life can have a long-lasting and irreversible impact on an individual’s nutritional status \cite{32}, by causing loss of appetite, mal-absorption, increased nutrient loss and increased metabolism\cite{33}. Diarrhoea risk increases during the infant weaning period\cite{21, 22}. A quarter (25\%) of stunting can be attributed to five or more episodes of diarrhoea before the age of two \cite{17}. The risk to nutrition from faecal pathogens is not limited to diarrhoea; ingestion of faecal bacteria may cause environmental enteropathy\cite{34}, as children living in contaminated environments and poor sanitary conditions are exposed to a large number of pathogens that can cause damage to the gut, reducing nutrient absorption and increasing loss of nutrients. It is estimated that a substantial proportion of malnutrition is due to impaired intestinal absorptive function resulting from multiple and repeated enteric infections\cite{35}. Figure 1, from Leonardo Mata’s study among the Children of Santa Maria Cauque, Guatemala, shows that infectious diseases such as diarrhoea can affect children’s
growth once weaning is initiated [20, 36, 37]; it also demonstrates that the cumulative effect of illness leads to growth faltering.

A study of Gambian children showed a mean deficit of 1.2kg by the first year of age, and it was estimated that diarrhoeal diseases contributed to almost half of this deficit[37]. In contrast, another study in The Gambia showed 43% linear growth failure by increased intestinal permeability where diarrhoea was not associated with such growth failure[38]; this might be due to environmental enteropathy. Evidence from Bangladesh shows that among children aged <24 months, diarrhoea duration increases progressively as nutritional status decreases, and that this effect is apparent for moderately malnourished children (60-74% weight-for-age) as well as for severely malnourished children (<60% weight-for-age)[39]. Contaminated weaning foods are thought to be a major contributor to diarrhoeal diseases and malnutrition in low income settings[14, 25].

The nutrition sector has traditionally placed greater emphasis on ‘nutrition specific’ interventions, resulting in a somewhat limited evidence base on the impact of the latter on tackling the underlying determinants of foetal and child nutrition and

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1 ‘Nutrition-specific’ programmes such as breastfeeding support and promotion, food fortification, micronutrient supplementation and treatment of severe malnutrition, address the immediate determinants nutrition. ‘Nutrition-sensitive’ programmes such as agriculture, water and sanitation, education and employment, healthcare, support for resilience and women’s empowerment, address the underlying determinants of nutrition.
development. Improving food hygiene practices should therefore contribute to reducing childhood undernutrition in low-income settings.

1.1.5 Early exposure to contaminated food:
WHO recommends that infants be exclusively breastfed for the first six months of life, and that from six months up to two years or older, they should receive safe complementary food[41]. Exclusive breastfeeding has a protective effect on diarrhoea morbidity and mortality[7]; however, compliance with exclusive breastfeeding in low-income settings is low (exclusive breastfeeding among infants younger than six months was only 39% in 2010, as shown in Figure 2) [42]. In most countries, breast-fed children aged between 4 to 6 months are given complementary food (such as water, juice, solid and semi-solid food) and are thus potentially exposed to food-borne pathogens in early life[43].

![Figure 2: Trends in exclusive breastfeeding among infants younger than six months. Note: Trend analysis based on 66 countries covering 74% of the developing world population (excluding China). Source: Cai et al, 2012)](image)

1.1.6 Weaning/complementary foods and factors contributing to contamination:
Complementary and weaning foods given to children aged 6-23 months in low-income settings are thought to be frequently contaminated and children who, until then, have only consumed milk may be exposed to infective doses of foodborne pathogens[25]. Studies show that 16% of sampled weaning food fed to children in Zimbabwe[44], 41% in Bangladesh[45] and 44% in rural Egypt [46] was contaminated with E. coli; and 68% of weaning food samples in Liberia[47] were contaminated with total faecal coliforms. Biologically acidified, and non-acidified baby milk and a millet gruel prepared and stored at ambient temperatures over a
period of 8 hrs increased in total colony count and in number of *Bacillus cereus*, *Clostridium welchii*, *Staphylococcus aureus* and *Escherichia coli* in West Africa [48]. Milk products report high level of Total Faecal Coliforms, as they provide a nutritious lactose available environment for growing coliforms [21, 48]. A study conducted in Peru in 1982-1984 showed that infants had nearly 10 episodes of diarrhoea in their first year of life, with weaning foods suggested as a major transmission route[49].

Various factors contribute to making complementary food unsafe and increase the risk of foodborne infection among children. Such factors have previously been studied [47, 50, 51] and reviewed [14, 20, 52, 53], and include: hot climate [54]; poor storage practices due to lack of refrigeration; insufficient cooking time [20, 54]; time elapsed between meal preparation and feeding [47, 49, 55, 56]; inadequate re-heating[57, 58]; washing utensils in contaminated water[53]; use of unsterilized and dirty feeding bottles for children[14, 59]; and limited maternal awareness about the link between diarrhoea and improper food handling. Other factors that may lead to contamination include environmental contamination due to lack of sanitation, presence of children’s faeces in the home environment and unhygienic management of child’s stools in low-income settings [60, 61]; use of unclean serving utensils; and not washing hands prior to cooking and feeding. Unhygienic practices related to bottle feeding, premastication and mashing have been found to be related to a high level of bacterial contamination[62]. Previous studies mainly focused on identifying the factors associated with microbial contamination in food, however few went so far as to design interventions to remedy this situation.

1.1.7 Factors contributing to microbial growth in food:

Microbes in food can multiply quickly over time if food is kept at ambient temperatures. Studies have found differences in coliform and *Enterobacteriaceae* counts between food stored for less than four hours and food stored for four or more hours[63]. Counts of bacteria including *E. coli* and *S aureus* increased from $10^4$ to above $10^8$ after 24 hours at $37^\circ$C[47, 49, 56, 64], and counts of *Shigella flexneri* increased 2-3 log within 6 hours[65]. A study in Peru showed that within one hour of preparation, faecal bacteria increased three-fold [49] and significant multiplication of faecal coliforms occurred when there was a delay of more than four hours between preparation and consumption of food[66]; such food fed to infants was rarely reheated[67]. Temperatures of 20-40$^\circ$C are optimal for bacterial growth in food;
temperatures below 6°C or higher than 60°C inhibit growth[14]. Improper storage and handling of cooked food could therefore contribute substantially to food-borne illnesses. Contamination may depend on the type of food and hygiene practices such as preparation and storage[25, 68]. Control of time factors during cooking, food storage and feeding need therefore to receive special attention in the promotion of health and hygiene[52, 56]. Previous studies also assessed the antimicrobial effect of fermentation in weaning foods. In the Ghanaian village, the extent of contamination was significantly higher in the unfermented dough than in fermented dough when examined for gram-negative bacilli (GNB) immediately after preparation and during storage [69].

1.1.8 Food as a critical pathway for transmission of faecal pathogens:
The F-diagram (Figure 3) shows food as a critical pathway by which faecal pathogens are ingested by a new host[70, 71]. Food is potentially important for disease transmission because pathogens in food have an easy route into the digestive system, and because some gastro-enteric pathogens can multiply in food and thereby increase the dose ingested, as we have discussed[72]. Once faecal contamination is present in food, other factors such as temperature and feeding practices contribute to the growth of bacteria and/or micro-organisms. To cause diarrhoea, pathogens need to enter the body through the mouth.

![Figure 3: Faecal-oral routes of disease transmission (Source: Wagner and Lanoix 1958[71] and LSHTM-UNICEF-ODA-INFDC,1997[70]).](image-url)

Whilst the transmission pathways are known, the relative importance of each remains unknown[71, 73] and may be context-specific. Safe sanitation, water and handwashing have been shown to reduce the risk of diarrhoeal infection in multiple studies[74]; however,
interventions targeting foodborne transmission as a key pathway have rarely been explored [75].

WASH interventions such as safe water provision and use of latrines act as primary barriers for faecal-oral transmission of pathogens. Previous systematic reviews suggest that improved water quality and excreta disposal reduce the risk of diarrhoea by 17% and 36% respectively[3]. Hygiene practices, primarily handwashing with soap at critical times, act as secondary barriers. Evidence from observational studies, randomized controlled trials and reviews suggests that handwashing with soap can reduce childhood diarrhoea by 30-47%(4, 5, 76); some observational studies suggest that handwashing before preparing food is a particularly important opportunity to prevent childhood diarrhoea[77]. Although hygiene is high on the agenda in the WASH sector, hygiene interventions have often been limited to promotion of handwashing with soap, neglecting the need for and importance of food hygiene.

1.1.9 The home as an important location for infection and food hygiene:
The home has been identified as an important location for acquiring foodborne diseases [10, 78-80], due largely to specific food hygiene practices [49]. 7% to 47% of all food-borne infections are thought to originate in the home, including in high-income settings[80]. A study in Japan found bacterial contamination levels in kitchens to be higher than on bathroom surfaces[81]. Hazard analyses carried out in households in the Dominican Republic reported that kitchen knives and blenders were contaminated with Salmonella spp., and in Viet Nam, the risk of diarrhoea was significantly higher by cooking places (cooking in table and other areas)[82]. A study in rural Kenya showed that 44% of kitchen dishes were hygienically unsafe[83]; and water used for preparing weaning foods as well as feeding bowls were a major source of faecal contamination in weaning-food in The Gambia[84]. Further, many households in low-income settings cook once daily, and food is re-heated for the evening meal. The temperature used is often below 60°C and large quantities of bacteria may grow in the food following cooking that are not killed when the food is re-heated[14]. Since young children eat more often than adults, they are especially exposed to reheated foodstuffs. Hygiene practices of care givers of children at home, especially in the kitchen, are important because they have most contact with children and children’s food[85].

Children’s feaces are thought to pose a greater public health risk than those of adults because they tend to contain higher concentrations of pathogens. Young children are often allowed to
practice open defecation in the household yard in few low income settings believing that the infant stools to be harmless[86]. Presence of flies in the environment may facilitate to further contaminate food in domestic environment. Safe disposal of child faeces remains a somewhat overlooked facet of WASH, and of sanitation programmes more specifically[86]. Young children are at risk of being exposed to pathogens in the domestic environment. All these factors make the home a prime location for foodborne infections.

1.1.10 Hazard analysis and critical control points (HACCP):

HACCP[87] is a systematic approach for assuring the production and processing of safe food. Hazard Analysis and Critical Control Points (HACCP) methods are conventionally used to reduce microbiological contamination in food mostly in industrial settings, not for studies. However, the WHO/FAO Expert Committee on Food Safety recommended that the HACCP approach be used in homes in low-income settings to attain insights into hazards associated with food preparation and applicable preventive measures[88, 89]. This approach includes seven principles: i) conducting a hazard analysis and identifying hazards; ii) determining critical control points; iii) establishing critical limits; iv) establishing monitoring procedures; v) establishing corrective action; vi) establishing verification procedures; and vii) establishing record-keeping and documentation procedures[87].

Whilst these principles have been applied in many studies to identify hazards, assess microbial contamination and identify control points [51, 64, 89-92], there are still few examples of the use of HACCP for the design and testing of hygiene programmes in domestic settings. Small-scale studies show that weaning food hygiene activities following the HACCP approach can help to identify measures for improving home food safety[64, 92, 93].

1.1.11 Food hygiene behaviour change:

Changing people’s behaviour can be a difficult and complex undertaking[94]. Traditional hygiene promotion approaches have focused on educating people about health, germs and the value of using soap, for example; however, such approaches have rarely resulted in positive, sustained behaviour change[5, 13, 95-97]. These approaches fail to account for the fundamental role of broader structural determinants, such as cultural or social norms[95] as well as environmental constraints. The lack of suitable behaviour change approaches possibly explains the observed discrepancies between people’s knowledge about food safety and their actual practices[98]. Habits and previous experiences have been suggested as possible reason
for unsafe food behaviours[99]. An emic approach[100] is needed to establish the factors important for determining behaviour and forming habits. It is increasingly recognised that hygiene behaviour is determined by a range of factors and is deeply-rooted in the environment in which the behaviour takes place[95, 101]. Interventions focused on emotional drivers such as nurture, disgust and affiliation have achieved significant improvement in targeted behaviours[102].

Food hygiene involves multiple behaviours, making effective food hygiene promotion challenging. The very limited evidence base and scarcity of food hygiene interventions in low-income settings further compounds this challenge. Further, while motivating people to start practicing a new positive behaviour can be relatively straightforward, getting them to keep practicing this behaviour on a daily basis may be much harder. Part of the answer for achieving long term, sustained behaviour change may be found in the science of habit formation. Behaviour will not occur unless motivation is extremely high. Habits are environmentally triggered and behaviour repetition induces a motivational shift away from intentional control [103]. Critical environmental cues must be immediately available (without seeking or effort) to practice behaviours, such as designated places for handwashing with soap and water in or near the latrine and food preparation and cooking areas. This can help new practices to become part of the daily routine. Studies have shown that compliance of handwashing is higher when soap and water is immediately available [104]. There is currently no evidence that food hygiene practices can be changed at scale by community level promotional campaigns. There is therefore a need for a motivational intervention to improve food hygiene routines.

1.1.12 Food hygiene: an agenda for research
Recognising the prospect that interventions to promote food hygiene practice may be of considerable importance in the control of diarrhoea, the WHO Diarrhoeal Diseases Control Programme identified two research agendas in the 1980s and 1990s to: i) determine the factors associated with an increased or decreased risk of faecal contamination of foods and to, ii) design intervention studies to measure the impact of promoting specific changes in food hygiene behaviours, faecal contamination and, if possible, on growth and diarrhoeal morbidity[58]. The need for multi-disciplinary approaches from epidemiology, nutrition, microbiology, social sciences (anthropology and economics) and communication to execute such studies was identified[58]. Various attempts have been made to assess risk factors but
there is still very limited evidence to support the design and execution of intervention studies on food hygiene.

In 2000, the World Health Assembly identified prevention and control of foodborne diseases as a public health priority. Recent efforts such as the WHO initiative to estimate the global burden of foodborne diseases[15], and a number of regional declarations and strategy papers have tried to harmonise efforts to reduce foodborne infections. In 2003, the Regional Committee for Africa adopted a resolution on food safety. South East Asian countries in 1998 committed to a 10-point strategy to reduce the burden of foodborne diseases[105]. However, it is still unclear whether actions towards achieving these strategies led to any progress. In order to improve the situation, the WHO launched the Five Keys to Safer Food, in which ‘five food hygiene messages’ were translated into 40 languages [106]; however, the scale and effectiveness of those interventions has not yet been reported. Two intervention studies using the HACCP approach targeting household settings and focusing on food microbiology [92, 93] demonstrated some success, but have limitations for scaling-up, as interventions relying on inter-personal communication with frequent households visits may be expensive if expanded to nationwide scale. A few household trials tested behaviours[107] but a strategy to design full-scale interventions is yet to be adopted. Food hygiene clearly deserves a higher level of research and programme priority.

1.2 Rationale and justification for conducting a food hygiene study in Nepal

Nepal is one of the least developed countries in the world[108] and a country of diversity in terms of topography (mountains, hills and terai (plain)), ethnicity and language, comprising approximately 125 castes or ethnic groups speaking some 123 dialects or languages and following different traditions, including regarding food habits.

Although Nepal has made some progress in reducing child mortality from 162 per 1,000 live births in 1990 to 54/1,000 live births in 2011[109], the basic determinants for child health such as hygiene and sanitation are poor. According to national records, 38% of Nepalese people still practice open defecation and 15% do not have access to water services[110, 111]. Coverage statistics often mask deficits in terms of the quality of water, use of sanitation facilities and sustainability of the available services. According to NDHS 2011, 48% of
households had soap and water [112]; however, compliance of handwashing with soap practices has not been measured. Communicable diseases such as acute respiratory infections, diarrhoea, cholera, typhoid, hepatitis-A and skin diseases are prevalent[113]. Diarrhoea is the second leading cause of under-five mortality and diarrhoea outbreaks, particularly due to cholera, are common. In 2009, a single outbreak caused 371 deaths in mid and far-west Nepal[114] and investigative reports have ignored the role played by inadequate food hygiene. Children aged 6-23 months are most susceptible to diarrhoea and the prevalence of bloody diarrhoea is highest among children age 12-23 months in Nepal[112]. Nepal experiences seasonal variation in diarrhoeal diseases, which peak when temperatures increase and rainfall occurs (Figure 4).

Similarly, Nepal is among the top ten countries in the world in terms of stunting prevalence [115], and in the top twenty in terms of numbers of stunted children globally[116]. 41% of Nepal’s children under-five are stunted, 11% suffer from wasting, 29% are underweight and nearly one in two are anaemic in the year 2011[112]. Despite some progress, Nepal is still facing severe challenge in reducing diarrhoeal diseases and childhood undernutrition(Table 1).

Table 1: Trends in childhood mortality and children’s nutritional status in Nepal

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<td>13</td>
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<tr>
<td>Underweight (low weight-for-age)**</td>
<td></td>
<td>43</td>
<td>39</td>
<td>29</td>
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Note: * deaths per 1,000 live births, ** % of children <5yrs, based on WHO Child Growth Standards. Source: Nepal MDG progress report 2005 and NDHS, 2011[112]

In Nepal, complementary foods are often introduced early; around 30% of Nepali children receive complementary food before the age of 6 months (median duration of exclusive breastfeeding is 4.2 months) [109]. In recent years, the food security agenda has been prioritised by the state, demonstrated through a three-year interim plan[117] that recognises food security as a basic element of human rights, and through the interim constitution of Nepal, which recognises food as a basic human rights. However, food hygiene aspects are not linked with any of these initiatives.
Among the recommended strategies for controlling diarrhoeal diseases in Nepal, various interventions have been implemented such as community-based integrated management of childhood illnesses (CB-IMCI), Vitamin-A supplementation, Water, Sanitation, and Hygiene (WASH), immunisation, breastfeeding and micronutrient supplementation; but food hygiene promotion has been under-prioritised. It is generally assumed that poor food hygiene behaviours increase the level of microbiological contamination in food and hence the risk of diarrhoeal diseases; however, there is little rigorous quantitative evidence to support this premise in low-income settings including in Nepal. The introduction of a food hygiene intervention in such a challenging environment to improve the above situation is imperative.

No studies have been undertaken in Nepal to establish whether the introduction of simple food hygiene interventions can change mothers’ food hygiene behaviours, and reduce microbes in food and childhood diarrhoea. This study was designed to fill this evidence gap by designing and delivering a food hygiene intervention, and exploring the extent to which such interventions can be integrated into WASH, health and nutrition programmes.
1.3 Aims and objectives of the study

1.3.1 Aims:
The main aim of this study was to design, implement and assess the effect of a scalable food hygiene intervention on mothers’ food hygiene practices, and to assess the impact of the intervention on the level of microbiological contamination of food fed to young children in Nepal. The study further explores whether food hygiene interventions can be integrated into nutrition, health and WASH strategies and programmes in Nepal.

1.3.2 Objectives
The study was conducted in two phases, with the following objectives:

Phase I objectives
Formative research:
• To document current food hygiene behaviour and its environmental and psychological determinants among mothers.
• To assess levels of microbiological contamination in food fed to young children (6-59 months) in rural Nepal
• To identify the critical control points of food contamination while preparing, handling, and storing food, and feeding the child at home and prioritise key behaviours.

Phase II objectives
Cluster Randomized, Before-After study with Control:
• To design, test and introduce a simple, focused, feasible and replicable food hygiene intervention targeting mothers with a young child (aged 6-59 months)
• To measure compliance with and effect of the intervention on food hygiene behaviours/practices as a primary outcome
• To measure the effect of the food hygiene intervention on the levels of microbiological contamination in food as a secondary outcome.

Sectoral policy analysis and debate:
• To explore whether food hygiene interventions can be integrated into nutrition, health and WASH strategies and programmes in Nepal.
Note: Measuring the effect of the food hygiene intervention on the period prevalence of diarrhoea (with one week re-call period), would have required a much larger sample and was beyond the scope of this study.

1.3.3 Research questions:
- What are the underlying causes and determining factors for unhygienic food practices among mothers in rural Nepal?
- Can the identification of critical control points help to facilitate prioritising key food hygiene behaviours?
- Do scalable and replicable food hygiene interventions have any effect on mothers’ food hygiene practices?
- Do mothers’ food hygiene practices have an effect on the level of microbiological contamination of child food?
- How can food hygiene interventions be integrated into health, nutrition and WASH strategies and programmes in Nepal?

1.4 Study Framework
The study employed two conceptual frameworks - i) Behaviour Centred Design (BCD)[118] underpinned by Evo-Eco theory of behaviour change[101] to design, deliver and evaluate the food hygiene intervention; and ii) Hazard Analysis and Critical Control Points (HACCP)[87] to identify control points and prioritize behaviours.

The BCD approach was developed by a team at the Environmental Health Group of the London School of Hygiene and Tropical Medicine (LSHTM), and builds on evolutionary and environmental psychology as well as marketing practice, to design and test imaginative and provocative behaviour change interventions[118]. BCD has been used in designing, implementing and evaluating successful behaviour change interventions such as a handwashing with soap (SuperAmma) trial in India [101, 102, 118]. BCD involves five steps (Figure 5): A - Assess; B - Build; C - Create; D - Deliver; and E - Evaluate the intervention. This study followed all steps, and went beyond these to include policy implications.
In BCD, the Evo-Eco approach offers a new way of understanding the psychological and environmental determinants of behaviour, helps to generate insights into the drivers of deeply habitual and normative behaviours and behavioural settings, provides design principles for the campaign, and guides learning about the mechanism of change that led to the intervention’s success. BCD thus offers a theory of change and a process guide. Figure 6 shows the way in which the BCD approach and Theory of Change (details presented in chapter III) was used to design, implement and evaluate the food hygiene intervention study in Nepal. Steps A and B were completed in the first phase and steps C, D, and E were done in the second phase of the study.
Figure 6: BCD process to design, implement and evaluate a food hygiene trial in Nepal

The five step process:

Assess: Step A involved the collection and analysis of scientific and local knowledge of the topic. The first step was to check out what is known / unknown about the behaviour change and identified target audience, to understand current behaviours, when and where they are practiced, what might change behaviours and how might the target audience be affected? The knowledge and programmatic gaps on food hygiene were assessed through literature review (systematic search) of existing studies. Additional lessons from past experiences such as from handwashing studies[102] and other international initiatives on food hygiene were drawn[106]. This provided an agenda for the formative research. I also examined sectoral policies and strategies in Nepal to ascertain whether food hygiene can be integrated into the existing policy and programme framework as part of gray literature review.
**Build: Step B** involved conducting **formative research** in the context of rural villages in Nepal to build knowledge of existing food hygiene behaviours, to identify behavioural determinants including motives and environment (physical, social, biological), identify critical control points, prioritise key behaviours and inform the design of the intervention package. The formative research was conducted using Evo-Eco theory of behaviour change[101] and HACCP principles[87] and details are highlighted in the methods chapter III.

**Create: Step C** involved the **design of the intervention package** involving a creative team, comprising a variety of professional experts from private sector, programme development, social marketing, social and community mobilization, behaviour change and communication, curriculum design, research, product innovation and graphic art fields. A motivational intervention package was developed based on insights from the formative research, past experience and key programme principles. Making an explicit use of theory to develop an intervention prior to testing, and incorporating insights from the theory into an explicit model of how the intervention might alter behaviour, or affect other links in the causal chain between intervention and outcome, should lead to better-developed interventions, and also to better-designed evaluations[119].

**Deliver: Step D** involved the **delivery of the intervention package**. The intervention was delivered in Cluster Randomised fashion over three months in all intervention clusters through community/ group events and household visits by locally-recruited and trained female food hygiene motivators. Emphasis was given to changing behaviour using emotional drivers and change in behavioural settings.

**Evaluate: Step E** involved the overall **evaluation of the intervention effects** using a Cluster Randomized, Before-After study with Control (BAC). The compliance with and effect of the intervention on mothers’ food hygiene behaviours were measured as primary outcome, and the effect on the levels of microbiological contamination in commonly-used child food was measured as a secondary outcome before and after the intervention in both the intervention and control groups.

The details about the HACCP approach are described in chapter-III.
1.5 Thesis outline:
This thesis consists of seven chapters and follows the paper-style format for PhD thesis.

Chapter I includes the introduction, the evidence gap and knowledge on food hygiene, justification, aims, objectives, research questions, framework to guide the study, and overall chapter outlines. Chapter II covers the literature review and summarises published food hygiene intervention initiatives and the association of food hygiene with levels of microbial contamination, diarrhoeal diseases and childhood undernutrition. This chapter also includes sectoral policy analysis in Nepal as part of gray literature review and briefly discusses whether food hygiene can be integrated into sector strategies and programmes. Chapter III presents the overall methods used in the study. Chapter IV (Paper-1) presents the findings obtained from the formative research. It highlights prevalent food hygiene practices and their determinants, the critical and behavioural control points and the food hygiene behaviours that we identified as key. Chapter V (Paper-2) presents the design, delivery and evaluation of the intervention and covers the effect on our five key food hygiene behaviours as primary outcomes. Chapter VI (Paper-3) presents the effect of the intervention on the level of microbiological contamination in commonly-used child foods as a secondary outcome. Chapter VII includes the overall summary discussion, the lessons learned, recommendations including implications for future research, and conclusions.
1.6 References:


8. CDC, *CDC Estimates of Foodborne Illness in the United States*, 2011, Centers for Diseases Control and Prevention, Atlanta, USA.


23. EHP, *Children’s Feces Disposal Practices in Developing Countries and Interventions to Prevent Diarrheal Diseases: A Literature Review*, 2004, USAID.


89. WHO, *Application of the Hazard Analysis Critical Control Point (HACCP) System for the Improvement of Food Safety. WHO-supported case studies on food prepared in homes, at


105. WHO/SEARO, 10-Points; Regional Strategy for Food Safety in the South-East Asia Region. WHO regional office for South East Asia (SEARO), 1999((updated 07 June 2004)).


114. EDCD, Preparedness and Management of Diarrhoea Outbreak, EDCD Presentation. Department of Health Services, Epidemiology and Diseases Control Division, 2010.
CHAPTER - II
A systematic review of interventions to improve food hygiene, and their effects on contamination of food, diarrhoea and undernutrition among infants and children

2.1 ABSTRACT:

Objective: To examine the epidemiological evidence for the impact of food hygiene interventions on food hygiene behaviour, microbial quality of child food, nutritional status and diarrhoeal disease. In addition, to explore whether food hygiene interventions can be integrated into nutrition, health and WASH strategies and programme in Nepal?

Methods: The Medline, Embase and Global Health databases were electronically searched. Primary inputs of interest included food hygiene behaviour change interventions in the domestic setting and outcomes of interest included sustained food hygiene behaviours, microbial contamination of child food, diarrhoeal diseases and childhood undernutrition. Only 5 of 418 identified studies met the criteria for inclusion in the systematic review; two intervention and three observational studies. In addition to systematic review, Government policies, strategies, and programme implementation guidelines from Nepal’s health, WASH, nutrition and food technology sectors were reviewed as gray literature.

Results: The two intervention studies conducted in Bangladesh and Mali using Hazard Analysis and Critical Control Points (HACCP) principles in domestic settings proved effective at reducing the faecal contamination of weaning food. Two small-scale household level behaviour trials conducted in Guatemalan and Brazil assessed the feasibility of practicing targeted behaviours generated encouraging results, but were inconclusive on whether these behaviours can be effectively targeted and sustained over time through public health intervention. Only one observational study conducted in Bangladesh attempted to demonstrate a correlation between food hygiene practices and diarrhoea morbidity and growth of children as an intervention effect, but the evidence for this association was too weak to draw any conclusions about the links. Through gray literature, it was evident that Nepal’s policy environment can enable the integration of food hygiene promotion within ongoing WASH, nutrition and health programmes.

Conclusion: More food hygiene interventions are needed to provide evidence on whether community-based food hygiene interventions can improve behaviour, reduce microbial contamination of child food and improve health outcomes in low-income settings. Questions also remain as to how such interventions can be scaled. The establishment of an accountable ‘institutional home’ for hygiene promotion and the development of an overarching food safety policy including national hygiene framework would enhance the policy environment for integration of food hygiene through different sectoral programme in Nepal.

Key words: food hygiene, microbiology, diarrhoea, childhood undernutrition, behaviours, policy, review
2.2 INTRODUCTION:
Preventable and treatable foodborne diseases remain a major cause of illness both in high and low-income settings. The magnitude of this problem has often been quantified in high-income settings [1-3], but lack of data means that the burden of foodborne illness in low-income settings is not known. Policies are consequently based on extrapolation and assumptions derived from expert opinion and biological plausibility [4].

Diarrhoeal diseases remain the second leading global cause of death among children [5, 6]. Global malnutrition contributes to 45% of child deaths worldwide: a staggering 3.1 million young lives are lost each year [7]. Children in low-income countries are particularly vulnerable to growth faltering during the period when complementary foods are initiated [8]. One hypothesis suggests that a condition known as environmental enteropathy may account for a significant proportion of childhood under-nutrition. This condition is caused by sustained ingestion of faecal bacteria and is thus associated with poor sanitation and hygiene [9].

Current efforts to address the diarrhoeal disease burden in low-income settings include promotion of exclusive breastfeeding, water supply, sanitation promotion, handwashing with soap, measles vaccination, ORT and zinc supplementation and rotavirus vaccination. Food hygiene has been largely ignored as a strategy to control diarrhoea among the vulnerable age group of children 6 to 59 months. Though there is a growing recognition in the nutrition sector of the need for ‘nutrition-sensitive interventions’ [7], nutrition programmes have traditionally placed greater emphasis on ‘nutrition specific interventions’ [10], resulting in limited focus on hygiene and food hygiene. A recent review paper urged professionals working on nutrition to be informed on issues of clean water and safe food environments, and to utilize their expertise to develop effective strategies[11].

Food is often a vehicle for the transmission of pathogens of faecal origin and a medium for their growth. The introduction of un-hygienically prepared weaning foods exposes the child to enteric pathogens [12, 13]. Factors that are associated with contamination of weaning and other child foods have been previously studied [13-22] and reviewed [11-13, 21]. The reviews highlighted the need for interventions to reduce contamination of weaning food in households in low-income countries.
Possible food related risk factors for diarrhoea and undernutrition have also been explored, but to a lesser extent [13, 17, 23]. Studies have also used the HACCP approach to investigate the processes and procedures that contribute to microbial contamination in low-income domestic settings and to identify points where controls could be applied to prevent contamination [14, 24-26]. Despite knowledge on the causes of contamination of child food and the association of contaminated food with diarrhoea, studies with diarrhoea and nutritional outcomes frequently fail to consider factors related to food hygiene as possible risk factors.

The Water, Sanitation and Hygiene (WASH) sector also intervenes to prevent diarrhoea. Recent effectiveness trials in this area have mostly focused on sanitation [27] and handwashing with soap [28]. Previous reviews related to the health impact of WASH have mostly considered the impact of water (mainly quality) [29-31], sanitation [30, 31] and handwashing with soap [32, 33]. Food hygiene is not considered, yet previous reviews on food in low-income settings have generally concluded that weaning food prepared in unhygienic conditions is frequently and heavily contaminated with pathogens and is thus a major risk factor for diarrhoeal diseases and the cause of associated malnutrition in low-income countries [12, 13]. Proponents have strongly advocated for more focus on food hygiene and development of appropriate interventions in low-income countries [17].

It has been postulated that food, not water, may be the most important route of transmission of diarrhoeal diseases in low-income countries [17]. Despite the risk posed by contaminated food, no systematic reviews of the impact of food hygiene interventions on people’s behaviour, microbial reduction, and health outcomes in low-income domestic settings have been conducted. This systematic review was carried out to describe and understand the effect of food hygiene interventions on behaviour change, microbial quality of child food, diarrhoeal disease and nutritional status.

Food hygiene promotion is an important, but neglected intervention within public health, nutrition and water, sanitation and hygiene (WASH) programmes in Nepal. This might be due to policy and programming gaps, competing priorities, or lack of experience and knowledge on how to change people’s food hygiene behaviours in Nepal. Together with systematic search it is therefore government policies, strategies, and programme implementation guidelines from Nepal’s health, WASH, nutrition and food technology sectors were reviewed as gray literature search with an objective to explore the potential for the integration of food hygiene
interventions into Nepal’s nutrition, health and WASH strategies and programmes. Findings obtained from gray literature were triangulated through key informant interviews with policy and programme development professionals and a policy debate was organized among policy makers, programme implementers, donors and researchers. However, details methods and findings obtained from gray literature review (i.e. policy and strategy analysis) are only presented in this chapter. The summary reports including details methods, and findings obtained from key informant interviews, and policy debate are reflected in Annex A.

2.3 METHODS:

Two prong methods were used: A) ‘Systematic literature review’ to examine the epidemiological evidence for the impact of food hygiene interventions on food hygiene behaviour, microbial quality of child food, nutritional status and diarrhoeal disease; and B) ‘Nepal specific gray literature’ review to explore whether food hygiene interventions can be integrated into nutrition, health and WASH strategies and programme in Nepal. The specific methods used for each review are as follows:

A) Systematic literature review

2.3.1 Selection criteria

Only peer reviewed journal articles or studies meeting the following definition were eligible for inclusion.

<table>
<thead>
<tr>
<th>Inclusion criterion definition:</th>
</tr>
</thead>
<tbody>
<tr>
<td>Any weaning, infant, child or complementary food hygiene interventions or programme which intends to improve unhygienic food practices and seeks to prevent, control, eliminate/reduce the contamination in weaning/infant/complementary/child’s food, diarrhoeal diseases and childhood undernutrition</td>
</tr>
</tbody>
</table>

Any studies with food hygiene practices as the exposure of interest and outcomes on improved and/or sustained food hygiene practices, microbial contamination of weaning or child food, diarrhoeal diseases or childhood undernutrition were included. Studies dealing with any age group and type of population were included as long as they were dealing with domestic food. Only interventions that were designed to improve food hygiene within the household – whether the intervention itself was at the household or community level - were included, and
all interventions to improve food hygiene in commercial or industrial settings (including street vendors and restaurants) were excluded. Any papers focused on interventions related to dietary intake instead of food hygiene promotion were excluded. The geographic location was limited to low-income settings because this was the specific area of interest. The World Bank’s operational classification of economies (as of 1 July 2014) was used to limit low-income settings. Decision was made whether specific study location meets the criteria (GNI per capita of $1,045 or less) regardless of whether countries is in low or middle-income countries. Only intervention studies (experimental study design); randomized control trials and quasi-experimental studies (household and cluster randomisation), case control and cohort studies were reviewed. Cross-sectional studies were excluded unless they included behavioural trials of improved food hygiene practices. Due to the limited number of food hygiene intervention studies identified, all comparative designs were considered regardless of whether or not allocation was random or a trial was followed by an observational study.

2.3.2 Type of intervention:
The included studies evaluated food hygiene interventions to improve behaviour or reduce food contamination, diarrhoeal diseases and childhood undernutrition. No restriction was applied to the frequency, type of behaviours (within food hygiene), intensity of the intervention or duration of intervention. Studies related to food supply, food scarcity and micronutrients that did not contain food hygiene components as the exposure of interest were also excluded. Studies related to diarrhoea and or childhood undernutrition outcomes that did not consider food hygiene as an exposure of interest were also excluded from the list.

2.3.3 Search strategy
The Medline, Embase and Global Health databases were electronically searched using key words and search strings (Table 1). Databases were searched for journal articles published between 1970 and 24 September 2012 initially and researched again for new articles up to 21 September 2014. The search strategy was only prepared in English and all publications were restricted to those only written in English with limits on human subject and studies from low-income settings. Additional papers were identified by searching the reference lists of retrieved papers. The review was conducted in accordance with PRISMA guidelines for systematic reviews [34] and a PRISMA checklist was completed. Only specific results relevant to the aims of this paper were extracted from included studies. These findings are presented in the
results section of this paper. PRISMA checklist and data collection forms are attached in CD-R, Annex-I.

Table-1: main search strings for all database and crude results

<table>
<thead>
<tr>
<th>String ID</th>
<th>Search String</th>
<th>Results (21 September 2014)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Medline</td>
</tr>
<tr>
<td>1</td>
<td>((infant or child or children or weaning or complementary) adj3 food)</td>
<td>12221</td>
</tr>
<tr>
<td>2</td>
<td>((food adj3 (hygiene or handling or preparation or storage or feeding or quality or safety or intervention or programme)) or HACCP method* or critical control point*)</td>
<td>30428</td>
</tr>
<tr>
<td>3</td>
<td>food adj4 (hygiene* or unhygienic or unhygiene) adj4 (behaviour* or practice*)</td>
<td>84</td>
</tr>
<tr>
<td>4</td>
<td>((food adj3 (contamination or microbiology or microbes or microbial or microorganism)) or bacteriological quality)</td>
<td>54722</td>
</tr>
<tr>
<td>5</td>
<td>((Diarrhoea or diarrhea or diarrhoeal) adj3 (diseases or disease or incidence or infection or illness or prevalence))</td>
<td>7527</td>
</tr>
<tr>
<td>6</td>
<td>(stunting or wasting or underweight or malnourish* or undernutrition) adj3 (infant or child or children)</td>
<td>3182</td>
</tr>
<tr>
<td>7</td>
<td>2 or 3</td>
<td>30431</td>
</tr>
<tr>
<td>8</td>
<td>1 and 7</td>
<td>630</td>
</tr>
<tr>
<td>9</td>
<td>4 or 5 or 6</td>
<td>65221</td>
</tr>
<tr>
<td>10</td>
<td>8 and 9</td>
<td>154</td>
</tr>
<tr>
<td>11</td>
<td>limit 10 to (english language and humans)</td>
<td><strong>122</strong></td>
</tr>
<tr>
<td></td>
<td>Total</td>
<td><strong>414</strong></td>
</tr>
</tbody>
</table>

2.3.4 Data extraction

Records were first excluded on the basis of their abstracts and were then assessed for eligibility using inclusion and exclusion criteria. Duplicates were removed at this time. Relevant data, including a brief description of the study (study title, publication date, study design, locations and settings, objectives, study focus and conclusions) were extracted. Final
decisions were made on inclusion based on relevance and study design. Abstracts were then assessed for relevance and the full text was reviewed for further evaluation and inclusion. Papers were grouped according to study type and all included studies were assessed on methodological quality. The following data were extracted for all studies: study title, design, location, objectives, sample size, main exposure, main outcomes, summary measures and main results. The electronic data search, screening, extraction, review and write-up was done by a single author (myself) and the relevancy of included articles and quality was checked by two independent reviewers. Almost 45 working days were used to complete the systematic review process and draft write-up.

**B) Nepal specific gray literature review:**

**2.3.5 Multiple sector policy, and strategy documents review**

All policy and strategy documents issued between 1990-2013 by the health, nutrition, WASH and food technology and quality control sectors in Nepal were reviewed using an operational definition for food hygiene. Altogether nine key documents for health sector including the “NHSP-II, 2010-2015”[35]; three key nutrition sector documents, including the ‘MSNP, 2013-2017”[36]; six key WASH documents, including the “Sanitation and Hygiene Master Plan, 2011”[37]; and four key food technology & quality control documents, including the “Food Act and Minimum Quality & Quality Standard for Food”[38, 39] were reviewed. Findings obtained from each sector policy review are summarized in result section.

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**Operational definition of “Food Hygiene” for policy and strategy analysis**

‘Food hygiene’ refers to key behaviours that reduce microbial contamination and growth, and therefore offer protection from diarrhoeal diseases and foodborne infection. These include thorough cooking practices, cleanliness of serving utensils, hand washing with soap before feeding/eating, proper storage of cooked food, thorough re-heating before feeding, and water and/or milk treatment, in domestic or other (e.g. schools, hospitals, child care centre) settings. Industrial food safety and the service sector (restaurants, street vendors) were excluded from policy review. Relevant sector policies and strategies were reviewed for any statements, strategic points, approaches or actions related to **food hygiene behaviours**.
2.4 RESULTS:
Summary of the results are also grouped in two categories; A) Results obtained from ‘Systematic Literature Review’; and B) results obtained from ‘Nepal specific gray literature review’.

A) Systematic literature review:

2.4.1 Study selection and procedures:
Figure 1 presents a flow diagram of the search strategy and number of studies screened for eligibility and included in the review. The database search strategy resulted in 418 titles and abstracts, including 4 identified through reference search and duplicates. After screening, 36 studies were thoroughly assessed for eligibility. Five of the studies assessed met the inclusion criteria and were included in the present review (Table 2).

Figure 1: PRISMA flow of information through the different phases of a systematic review
2.4.2 Study design:
Detailed information on the characteristics of the studies included in this systematic review is presented in Table 2. Two studies were intervention studies (randomized controlled trials) with microbiological outcomes on weaning food in the domestic setting [40, 41], one was a longitudinal study (cohort study) with control and was focussed on cleanliness, diarrhoea and child growth as outcomes of interest [42], while two were observational studies followed by behavioural trials to test the feasibility of improving behaviours [43, 44].

2.4.3 Settings and study participants:
The five studies were conducted in Bangladesh (2 studies) [40, 42], Mali [41], Guatemala [44], and in Brazil[43]. The Bangladesh intervention study by M.S. Islam et al. (2013) [40] was conducted in a rural setting using 60 randomly-selected households with children aged 6-18 months with bacterial contamination in weaning food and water as the main outcomes of interest. The Mali intervention by O. Toure et al. (2013) [41] was conducted in a peri-urban (slum) setting using 60 randomly selected households with bacterial contamination in food as the main outcome of interest. The other Bangladesh study was a cohort (longitudinal) design, conducted by NU. Ahmed et al. (1993) [42] in rural Bangladesh using 85 households with children aged 0-18 months in both the intervention and control groups. Childhood diarrhoea, cleanliness practices and nutritional status were the main outcomes of interest. The Brazilian study by CMG Monte et al. (1997)[43] was conducted in an urban slum using 75 households with mothers and with adoption of the advocated behaviours as the main outcome of interest. The Guatemalan study by S de Tejada and F. Cano (1995)[44] was conducted in an urban setting using 25 families with children aged between 12 and 23 months. Once again, adoption of the advocated behaviours was the main outcome of interest (Table 2).
<table>
<thead>
<tr>
<th>Author / date</th>
<th>Study title</th>
<th>Study design</th>
<th>Geographic location</th>
<th>Sample size</th>
<th>Main objectives</th>
<th>Main exposure</th>
<th>Main outcomes</th>
<th>Summary measures</th>
<th>Main results</th>
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</thead>
<tbody>
<tr>
<td><strong>Intervention studies with control</strong></td>
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<tr>
<td>M.S. Islam, 2013</td>
<td>Hygiene intervention reduces contamination of weaning food in Bangladesh</td>
<td>RCT</td>
<td>Bangladesh</td>
<td>60 households (30 intervention and 30 control)</td>
<td>Measure the impact of a hygiene intervention on the contamination of weaning food in Bangladesh</td>
<td>Hygiene training / intervention using HACCP approach</td>
<td>Bacterial contamination in weaning food and water (faecal coliforms and faecal streptococci)</td>
<td>Mean (log-transformed bacteria) difference</td>
<td>Intervention following HACCP substantially reduced faecal bacteria in weaning food. The difference in faecal coliforms (FC) and faecal streptococci (FS) before and after intervention is statically significant (p&lt;0.001) in food and water at point of use.</td>
</tr>
<tr>
<td>O. Toure, 2013</td>
<td>Piloting an intervention to improve microbiological food safety in Peri-Urban Mali</td>
<td>RCT</td>
<td>Mali</td>
<td>60 households (30 intervention and 30 control)</td>
<td>Test the hygiene promotion programme to find out their impact on microbial reduction in food</td>
<td>Hygiene training and demonstration using HACCP approach</td>
<td>Bacterial contamination in food (thermotolerant coliforms)</td>
<td>Total load of bacteria in weaning food</td>
<td>Thermotolerant coliform (TTC) &gt;100/gm in 55% and 86% sampled collected after cooking (cooled after cooking) and after 3-6hr storage during baseline. After intervention contamination detected (&gt;10 TTC/gm) only in 17% and 4% sampled collected after cooking and after-heating (stored food re-heated).</td>
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<tr>
<td><strong>Longitudinal studies (cohort studies) with control</strong></td>
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<tr>
<td>N.U. Ahmed, 1993</td>
<td>A longitudinal study of the impact of behavioural change intervention on cleanliness, diarrhoeal morbidity and growth of children in rural Bangladesh</td>
<td>Cohort study (Longitudinal study)</td>
<td>Rural Bangladesh</td>
<td>185 intervention and 185 control households</td>
<td>To reduce childhood diarrhoea by modifying hygiene behaviours, using solutions from within the community</td>
<td>Theme I - Ground sanitation, Theme II - personal hygiene Theme III - Food hygiene</td>
<td>Childhood diarrhoea, cleanliness practices and nutritional status</td>
<td>Prevalence – Diarrhoea, Proportion - cleanliness, Z scores - nutritional status</td>
<td>Higher adoption of behaviours (including food hygiene) were associated with better cleanliness status, lower diarrhoea and malnutrition rate in intervention site. Between-site longitudinal analysis showed that the intervention site had substantially higher cleanliness scores, lower diarrhoeal morbidity, and better growth status compared to those of the control site, with differences increasing over time.</td>
</tr>
<tr>
<td>Author / date</td>
<td>Study title</td>
<td>Study design</td>
<td>Geographic location</td>
<td>Sample size</td>
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<td>Main exposure</td>
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<td>Main results</td>
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<tr>
<td>C.M.G. Monte, 1997</td>
<td>Designing educational messages to improve weaning food hygiene practices of families living in poverty</td>
<td>Observational study including household trial</td>
<td>Urban slum, drought-prone region of northeastern Brazil</td>
<td>75 mothers (each of 15 non-practising mothers from 5 groups invited to adopt behaviours)</td>
<td>Whether or how weaning food hygiene practices can be improved in slum conditions</td>
<td>Educational messages to promote behaviours using household trial</td>
<td>Adoption of advocated behaviours</td>
<td>Frequencies / proportions</td>
<td>All initiated the advocated behaviours and most (50-80%) sustained the new behaviours (handwashing 73%, using boiled water for reconstituting powdered milk 80%, feeding gruel by spoon rather than bottle feeding 53%, not storing gruels and milk 80%) up to one month.</td>
</tr>
<tr>
<td>S.S. de Tejada, 1995</td>
<td>Weaning-food hygiene in a Guatemalan town</td>
<td>Observational study including household trial</td>
<td>Guatemalan town of Ciudad Vieja, Sacatepequez province</td>
<td>25 mothers involved in testing handwashing and 15 on re-heating behaviour</td>
<td>Identify specific activities contribute to the bacterial contamination of weaning food; with the ultimate aim of changing them through education intervention</td>
<td>Demonstration of hand-washing and re-heating staple foods for two weeks</td>
<td>Adoption of behaviours</td>
<td>Frequencies</td>
<td>After one week testing, handwashing was adopted enthusiastically (after a project introduced a small hose which was connected to the cement basin and family perceived this is simple, easy-to-use device and soap used also increased). In two-weeks trial, re-heating beans was easier than for tortillas (14/15 and 11/15 adopted respectively)</td>
</tr>
</tbody>
</table>
2.4.4 Interventions:
The targeted food hygiene behaviours and nature of the interventions varied considerably between the studies (Table 3). The Bangladesh and Mali intervention studies [40, 41] targeted the same 4 behaviours for improvement. Both delivered interventions through household-level hygiene training and demonstration using the HACCP approach and were delivered over a three and four week period respectively. The longitudinal study in Bangladesh [42] targeted three themes including Food Hygiene which included eight behaviours. The food hygiene behaviours were promoted together with sanitation and personal hygiene with the assistance of village leaders through a ‘clean life’ campaign delivered by local project workers and volunteer mothers for about 7 months. Household trials in the Brazil study [43] targeted four specific behaviours and mothers were taught how to perform the recommended behaviours using educational techniques. The Guatemalan study[44] only targeted two behaviours. Behaviours were promoted through demonstration of hand-washing for a week and re-heating staple foods for two weeks.
### Table 3: Targeted food hygiene behaviours by included studies

<table>
<thead>
<tr>
<th>Targeted behaviours</th>
<th>Studies</th>
</tr>
</thead>
<tbody>
<tr>
<td>Washing hands with safe water and soap before food preparation or child feeding,</td>
<td>Bangladesh[40].</td>
</tr>
<tr>
<td>after cleaning child’s bottom and after using latrines</td>
<td>Mali[41] intervention studies</td>
</tr>
<tr>
<td>Use of safe water to wash utensils and prepare food</td>
<td></td>
</tr>
<tr>
<td>Cooking and re-heating food until boiling</td>
<td></td>
</tr>
<tr>
<td>Covering the food with lid during storage</td>
<td></td>
</tr>
<tr>
<td>Do not use any feeding bottles, but if you do then soak them in strong salt water</td>
<td>Bangladesh[42] (cohort - longitudinal study)</td>
</tr>
<tr>
<td>or wash the bottles in hot water and boil the nipple before feeding</td>
<td></td>
</tr>
<tr>
<td>Prepare only the quantity of bottle mixture that the baby can drink at one time</td>
<td></td>
</tr>
<tr>
<td>Use only Tube well water for drinking and mixing with food</td>
<td></td>
</tr>
<tr>
<td>Wash both hands (care takers and child’s hands) and eating plates</td>
<td></td>
</tr>
<tr>
<td>Do not feed leftover food to a child</td>
<td></td>
</tr>
<tr>
<td>Keep all food covered</td>
<td></td>
</tr>
<tr>
<td>Store clean pots, pan and plates upside down or covering them</td>
<td></td>
</tr>
<tr>
<td>Cover water pitchers</td>
<td></td>
</tr>
<tr>
<td><strong>Ground sanitation and personal hygiene- not related to food hygiene</strong></td>
<td></td>
</tr>
<tr>
<td>Handwashing before and after defined events</td>
<td>Brazil[43]</td>
</tr>
<tr>
<td>Boiling water for reconstituting powdered milk</td>
<td></td>
</tr>
<tr>
<td>Feeding gruel by cup and spoon rather than bottle-feeding</td>
<td></td>
</tr>
<tr>
<td>Not storing prepared gruels and milks</td>
<td></td>
</tr>
<tr>
<td>Washing children’s hands before eating</td>
<td>Guatemalan town[44]</td>
</tr>
<tr>
<td>Thorough re-heating of staple food</td>
<td></td>
</tr>
</tbody>
</table>

#### 2.4.5 Summary measures:

Meta analysis was not appropriate as there was substantial heterogeneity in the study designs and outcomes measures. Different studies included different measures (Table 2). Out of five studies, only two randomly allocated the intervention to see the effect of the food hygiene intervention on microbiological contamination in selected weaning food. The outcome measures varied; one trial measured thermotolerant coliforms (TTC) [41] and the other assessed faecal coliforms and faecal streptococci in weaning food and water [40]. Only one
of the studies reported correlations between intervention (including food hygiene) and diarrhoea and undernutrition [42], although the allocation of the intervention was not random. The two observational studies measured the feasibility of adopted behaviours [43, 44].

2.4.6 Study quality:

The identified studies were of varying quality. Quality issues included study design, rigorous reporting of results, methods of outcomes measurement, the process followed to select behaviours for promotion, consistency on the number of behaviours, use of randomized control trial protocols, reporting biases, behaviour observation biases, small sample sizes, reporting of programme components, collection of samples for microbiology and use of behaviour change principles.

Neither of the intervention studies with microbiology included in this review (Bangladesh and Mali) reported the randomization process [40, 41]. Both intervention studies were efficacy rather than effectiveness trials [40, 41]. In the Bangladesh study food samples were taken at different points of time, but the authors did not mention how they minimized the influence on mother’s behaviours while doing the repeated measurement [40]. Of the 5 studies included in the review, only the Brazil study justified the sample size [43]. Only the cohort study in Bangladesh highlighted the measures taken to minimize observer bias [42] and no study attempted to blind participants to the outcome measures. The Brazil and Guatemalan studies had objectives around assessing the feasibility of change but did not assess the effectiveness of the intervention [43, 44].

Though there was variation in the behaviours that were targeted, all followed certain principles to prioritize behaviours. Bangladesh and Mali intervention studies used Hazard Analysis and Critical Control Points (HACCP) to determine the behaviours to target [40, 41]. The Bangladesh cohort study used a community based trial [42], the Brazil study used rapid ethnographic assessment, community survey and structured observation [43], while the Guatemalan study used behavioural and microbiological analysis [44]. Only two intervention studies reported the sustainability of the effect of the intervention up to three months after the intervention [40, 41]. Other studies recorded outcomes either longitudinally [42], during or immediately after the trial [42-44]. Only the cohort study from Bangladesh reported diarrhoea outcomes, but this study relied on self-reported diarrhoea and failed to report potential
sources of bias [42]. The same study reported Weight-for Age-Z-scores, which were calculated according to National Centre for Health Statistics and WHO standards, but the intervention was not randomly assigned. No study mentioned the availability of a protocol [40-44]. No studies reported the response rate, meaning that the issue of selection bias cannot be fully evaluated. Only the Mali intervention study reported the reach of the intervention and explicitly mentioned that the tested intervention was far from being affordable and effective at scale which would limit uptake by public health authorities and donors in low-income countries [41].

2.4.7 Outcomes:
Two intervention studies reported microbial outcomes [40, 41], one cohort study reported on diarrhoea and growth of children [42] and two observational studies assessed the feasibility of behaviours through household trial [43, 44]. The study outcomes are summarized in table 2.

Behavioural outcomes:
The study conducted in Brazil [43] tested the feasibility of adoption of four advocated food hygiene behaviours (handwashing, boiling water, spoon feeding, and not storing gruels and milk) through household trials in five groups using carefully designed and delivered educational messages. They found that all promoted behaviours were initiated by all, and 60% of the participants sustained and practiced them every time during a one-month period. Among all advocated behaviours, spoon-feeding was the most difficult behaviour to adopt to completely, as their babies were already accustomed to bottle feeding [43]. The study in Guatemala [44] tested two behaviours among a few mothers and households: washing children’s hands before eating and re-heating staple food (beans and tortillas). After demonstration, handwashing was adopted enthusiastically, especially after the project introduced a cement basin and the family perceived this was a simple, easy-to-use device. Soap use also increased. In the two-week trial of re-heating, 14 out of 15 households adopted the reheating recommendations for beans, and 11 out of 15 adopted the suggestions for tortillas[44].
**Microbial outcomes:**

The Mali intervention study [41] found that 3 weeks’ training using the HACCP approach was effective in improving home food safety. Before the intervention, thermotolerant coliform (TTC) contamination levels exceeded 100 per gram in 55% of food samples cooled after cooking (prior to child service), and in 86% of samples of food stored prior to child service. After the intervention, contamination was detected (i.e. >10 TTC/g) in less than 17% of the food samples cooled (prior to child service) after cooking and in only 4% of food samples reheated after storage and cooled prior to child service. The reduction in faecal contamination was highly significant (p<0.001). Their follow-up visit after three months also produced better results on food microbiology; only up to 17% of food samples failed to meet their standard (<10TTC/g)[41]. The intervention study conducted in Bangladesh followed a very similar approach and procedures to assess the effect of the intervention reported that weaning foods were heavily contaminated with Faecal Coliforms (FC) and Faecal Streptococci (FS) during baseline. The FC and FS counts were 1.84 log$_{10}$ and 1.92 log$_{10}$ colony-forming unit (cfu/g) respectively in intervention households and 0.86 log$_{10}$ and 1.33 log$_{10}$ cfu/g respectively in control households. Food samples were collected and analyzed after mothers had received a four-week training. The FC and FS count immediately after intervention in the intervention group dropped to 0.10 log$_{10}$ and 0.09 log$_{10}$ cfu/g (p<0.001), respectively. The Authors also reported the significant reduction in water contamination both at source and point-of-use (PoU)[40].

**Diarrhoea and childhood undernutrition outcomes:**

The cohort study in Bangladesh found that higher intervention adoption rates were associated with lower diarrhoea and growth status in the intervention group. The food hygiene knowledge score (0.43, p<0.001) and overall (including sanitation, personal hygiene and food hygiene) adoption score (0.56, p<0.001) were negatively correlated with diarrhoea (-0.25, p=0.001) and positively correlated with cleanliness (0.43, p<0.001) and weight-for-age Z-score (0.13, p<0.05). The adoption rates of food hygiene behaviours were not reported. Knowledge of germ theory, speeches delivered by leaders, and mobilisation of volunteer mothers from the villages were related to the observed improvements.
B) Nepal specific gray literature review:

2.4.8 Programme linked with diarrhoea control – desk review:

Table 4 summarises the sector specific programmes on the prevention and management of diarrhoeal diseases in Nepal. Different sector agencies (government and non-government agencies) are implementing their own set of intervention and those are directly and indirectly contributing to diarrhoeal control initiatives. Food hygiene has not been explicitly included in any of these programmes. Potential opportunities to integrate food hygiene into these programmes were explored and discussion centred on how best to integrate it.

<table>
<thead>
<tr>
<th>Health</th>
<th>Nutrition</th>
<th>WASH</th>
</tr>
</thead>
<tbody>
<tr>
<td>Treatment:</td>
<td>Prevention:</td>
<td>Prevention:</td>
</tr>
<tr>
<td>• Oral Rehydration Therapy (ORT), Zinc, continued feeding</td>
<td>• Infant and young child feeding (exclusive breastfeeding)</td>
<td>• Water supply and point of use promotion</td>
</tr>
<tr>
<td>• Oral rehydration solution</td>
<td>• Micronutrient supplementation (Vitamin-A, MNP/Baal vita powder in 15 districts)</td>
<td>• Sanitation promotion (total sanitation)</td>
</tr>
<tr>
<td>• Antibiotics for dysentery</td>
<td>• Bi-annual deworming tablet to children 12-59 months</td>
<td>• Hygiene promotion: mostly ‘handwashing with soap’</td>
</tr>
<tr>
<td>• Case management</td>
<td>• Households food security (promote kitchen gardening...)</td>
<td>• Other hygiene aspects (personal and domestic hygiene, waste management)</td>
</tr>
<tr>
<td>• Outbreak management</td>
<td>• School health and nutrition</td>
<td></td>
</tr>
<tr>
<td>Prevention:</td>
<td>• Project based programme: such as SUAAHARA in 20 districts (IYCF, WASH promotion, food security)</td>
<td></td>
</tr>
<tr>
<td>• Measles vaccination</td>
<td></td>
<td></td>
</tr>
<tr>
<td>• Community Based - IMCI/Newborn Care Programme</td>
<td></td>
<td></td>
</tr>
<tr>
<td>• Health promotion</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>


2.4.9 Policy and strategy analysis:

Water, Sanitation and Hygiene (WASH) Sector:

Six key policy / strategy documents provide the legal and programmatic basis for WASH in Nepal. The ‘National sanitation policy and guidelines’ of 1994[45] guides planning and implementation of sanitation programmes in an integrated manner with health, water supply and local development programmes. In 1999, the “Local Self Governance Act” [46] established the legal basis for the devolution of responsibilities and authorities to local government for physical infrastructure development, including water and sanitation systems. With the adoption of the Millennium Development Goal target to halve the proportion of people without sustainable access to safe drinking water and sanitation by 2015, Nepal set
access targets of 73% for water and 53% for sanitation. The national target for water and sanitation access for all by 2017 is reflected in the Government’s Tenth Development Plan, 2002-2007 [47].


The “National Urban Policy” was issued in 2007, indentifying poor sanitation, environmental degradation, and lack of services for the urban poor as areas requiring urgent attention. The “Three Year Interim Plan 2007-2010”[50] highlighted the need for a water supply and sanitation sector-wide approach (SWAp). It recognized both schools and community-led approaches for total sanitation promotion; but in reality hygiene remained a low priority and was often implemented as a one-off project activity. The “Urban Water Supply and Sanitation Policy 2009”[51] stressed the implementation and use of hygienic latrines at households and in all urban areas by the year 2017. This document reflects that by 2009 there was a growing recognition of the need for guiding documents for sanitation and hygiene programme.

The most recent guiding document for the sector is the “Sanitation and Hygiene Master Plan 2011”[37] developed to create an enabling policy environment for achieving national targets through collaborative efforts in an accelerated manner. It emphasises decentralised planning and implementation by generating and mobilising local resources and synergising the efforts of sector and non-sector actors. It articulates detailed sanitation and hygiene related programmes and activities and includes ‘food hygiene’ under activities to be conducted once communities have become Open Defecation Free (ODF); this, however, is limited to handwashing, and covering food and water. A national guiding framework for hygiene was not in existence.
**Health sector:**

The *National Health Policy of 1991* [52] intended to extend the primary health care system to the rural population to reduce infant and child mortality. Environmental Health was incorporated as one of the promotive health services, it also focused on testing food and drinking water within the hospitality industry. A diarrhoea control programme was introduced, but the aspects of domestic food hygiene and the role of food hygiene to prevent diarrhoea were not mentioned. The nutrition component focused only on food supplementation, without explicit mention of the importance of food hygiene.

Similarly, *The Second Long-term Health Plan, 1997-2017* [53] included preventive and promotive aspects. Essential healthcare services included disease-specific programmes such as diarrhoea, as well as environmental health and sanitation and air pollution. Food hygiene was mentioned alongside water supply, solid waste disposal, sewerage, and excreta disposal. However food hygiene was not included in the action plan and the absence of food hygiene and WASH indicators and targets resulted in poor implementation. Further, *The Elements of Essential Health Care Services (2000)* [54] included Environmental Sanitation and Hygiene as one of twenty main interventions, but again failed to mention food hygiene.

The ‘*Health Sector Strategy: An Agenda for Reform*’ document of 2004 [55] set priorities for meeting the MDGs, reducing poverty and improving health. The package of Essential Health Care Services (EHCS) included preventive and curative services, but left out hygiene and food hygiene. The Behaviour Change Communications (BCC) strategy set out in the document to support EHCS also failed to include hygiene or WASH promotion. The *Nepal Health Sector Programme-Implementation Plan-I (NHSP-IP-I), 2004-2009* [56], the first overarching health sector plan focused on increasing coverage and raising the quality of EHCS, mainly family health, safe motherhood, child health and control of communicable diseases did not include WASH or hygiene.

The *Three Year Interim Plan 2007-2010* [50] prioritized the principles of primary health services and public health. Access to quality drinking water and sanitation was mentioned as part of drinking water projects and programmes to secure food for the rural poor, yet the importance of food hygiene for nutrition was not highlighted.
With the growing realization of need of WASH for diarrhoea control in Nepal, in 2010 WASH was explicitly included as a cross-cutting theme under the Nepal Health Sector Programme-II, 2010-2015\(^5\) under the ‘Environmental Health and Hygiene’ theme. It aimed to improve water quality with particular emphasis on water quality surveillance, and promote hygiene and sanitation. However, the lack of WASH indicators and budget allocation resulted in the poor implementation of these programmes. NHSP-II also includes nutrition and health education and communication programmes, but these do not incorporate food hygiene.

**Nutrition sector:**

In the nutrition sector the first National Nutrition Policy and Strategy, 2004\(^5\) was the main policy document guiding interventions until 2012. It recognized the need for good hygiene practices and clean environments for reducing intestinal worms and highlighted improved food hygiene, food safety, safe water and sanitation. However, the detailed hygiene components were not operationalised and programmes were not implemented. Instead, only the ‘nutrition specific’ interventions were prioritised and implemented.

During 2009/10, the Nutrition Assessment and Gap Analysis (NAGA)\(^5\) assessed the determinants for undernutrition and identified strengths, weaknesses, and gaps. It suggested the need for a national nutrition architecture and a multi-sectoral approach through an agreed nutritional determinants model. NAGA recommended interventions on sanitation and hygiene through health centres and communities and reducing the risk of infection from unprotected water sources and poor sanitation and waste disposal, and encouraged coordination efforts to promote handwashing across multiple sectors and programmes.

The momentum resulting from NAGA led to the formulation of a Multi-Sector Nutrition Plan (2013-2017)\(^3\), the latest guiding document and milestone for nutrition in Nepal. The goal of the plan is to reduce maternal, infant and young child undernutrition by one third. It also aims to significantly reduce undernutrition over ten years, so that it no longer impedes Nepal’s human capital and socio-economic development. The plan opens policy amendment opportunities in all sectors (health, WASH, education, agriculture) and includes the promotion of nutrition ‘specific’ and ‘sensitive’ services. WASH related activities include young child feeding, promotional campaigns to increase practices of handwashing with soap at critical times, open defecation free campaigns (ODF), water safety plans, and water safety
at the point-of-use. The responsibility for implementing these activities lies with the Ministry of Urban Development.

**Food technology, safety and quality control:**

Food safety in Nepal is the responsibility of the Department of Food Technology and Quality Control (DFTQC) in the Ministry of Agriculture Development (MoA). This is the government agency responsible for implementing legislation to ensure safety in human and animal food supplies. The DFTQC monitors food quality, contamination and nutrient value at all stages of the processing and distribution steps, but at a limited scale (mainly urban and import/export food) but nothing related to food safety in the home. Our policy analysis revealed that as at the end of 2013 no ‘food safety policy’ existed in Nepal.

DFTQC functions are guided by *Food Rules* (most recently amended in 2007) [60], *The Food Act (1996)* [61] and the *Minimum Essential Quality Standards for Food and Animal Food (2011)* [39]. These documents cover issues such as institutional roles, food analysis and inspections, packed food labelling, food seller regulations, use of additives, licensing, prohibitions on production, sale or distribution, and minimum quality standards of human and animal food processed drinking water and baby formula. None of these documents included hygiene and its impact on the quality of food and role in protecting public health.
Table 5: Summary of individual policy/strategy documents included for the review

<table>
<thead>
<tr>
<th>SN</th>
<th>Policy / strategy document</th>
<th>Sector</th>
<th>Year</th>
<th>Ownership</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Sanitation and Hygiene Master Plan, 2011</td>
<td>WASH</td>
<td>2010</td>
<td>GoN, SCNSA</td>
</tr>
<tr>
<td>2</td>
<td>MPPW, Urban Water Supply and Sanitation Policy</td>
<td>WASH</td>
<td>2009</td>
<td>GoN, MPPW</td>
</tr>
<tr>
<td>3</td>
<td>Three Year Interim Plan, Approach Paper, 2007-2010</td>
<td>All</td>
<td>2007</td>
<td>GoN, NPC</td>
</tr>
<tr>
<td>4</td>
<td>National Drinking Water Quality Standards in Nepal</td>
<td>WASH</td>
<td>2005</td>
<td>GoN, DWSS</td>
</tr>
<tr>
<td>7</td>
<td>Local Self Governance Act, 1999</td>
<td>All</td>
<td>1999</td>
<td>GoN</td>
</tr>
<tr>
<td>8</td>
<td>National Sanitation Policy 1994, Government of Nepal</td>
<td>All</td>
<td>1994</td>
<td>GoN</td>
</tr>
</tbody>
</table>

**Health Sector**

<table>
<thead>
<tr>
<th>SN</th>
<th>Policy / strategy document</th>
<th>Sector</th>
<th>Year</th>
<th>Ownership</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Nepal Health Sector Programme-Implementation Plan II (NHSP-IP 2), 2010-2015</td>
<td>Health</td>
<td>2010</td>
<td>GoN, MoHP</td>
</tr>
<tr>
<td>2</td>
<td>Nepal Health Sector Programme – implementation plan, 2004-2009</td>
<td>Health</td>
<td>2004</td>
<td>GoN, MoHP</td>
</tr>
<tr>
<td>3</td>
<td>Health Sector Strategy: An Agenda for Reform, 2004</td>
<td>Health</td>
<td>2004</td>
<td>GoN, MoHP</td>
</tr>
<tr>
<td>4</td>
<td>Elements of Essential Health Care Services: Main Interventions or Program Components, 2000</td>
<td>Health</td>
<td>2000</td>
<td>GoN, MoHP</td>
</tr>
<tr>
<td>6</td>
<td>National Health Policy, 1991</td>
<td>Health</td>
<td>1991</td>
<td>GoN, MoHP</td>
</tr>
</tbody>
</table>

**Nutrition sector**

<table>
<thead>
<tr>
<th>SN</th>
<th>Policy / strategy document</th>
<th>Sector</th>
<th>Year</th>
<th>Ownership</th>
</tr>
</thead>
<tbody>
<tr>
<td>2</td>
<td>Nepal Nutrition Assessment and Gap Analysis, 2009</td>
<td>Nutrition</td>
<td>2009</td>
<td>GoN, MoHP</td>
</tr>
</tbody>
</table>

**Food quality and standard sector**

<table>
<thead>
<tr>
<th>SN</th>
<th>Policy / strategy document</th>
<th>FQSS</th>
<th>Year</th>
<th>Ownership</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Minimum Essential Quality Standard for Food and Animal Food (2068)</td>
<td>FQSS</td>
<td>2011</td>
<td>GoN, DFTQC/ MoA</td>
</tr>
<tr>
<td>2</td>
<td>Food Act (third amendment 1992)</td>
<td>FQSS</td>
<td>1992</td>
<td>GoN</td>
</tr>
<tr>
<td>3</td>
<td>GoN, Food Rules, 2027 (latest amendment in 01 October 2007)</td>
<td>FQSS</td>
<td>1970</td>
<td>GoN</td>
</tr>
</tbody>
</table>

*FQ=Food quality and standard sector*
2.5 DISCUSSION:

2.5.1 Systematic literature review:
Overall our review found that few studies have assessed the effect of food hygiene interventions in low-income settings. However, all five studies that were identified studies reported some positive results. The small number of studies makes it difficult to draw any conclusions about the effectiveness of food hygiene interventions on improvement of food hygiene behaviours, reduction in microbes, diarrhoeal diseases and childhood undernutrition. The methodological flaws in many of the studies complicate the interpretation of results and indicate a need for more research.

The review nevertheless suggests that it may be feasible to change food hygiene behaviours [41, 43, 44]. However there were no randomized trials with behavioural outcomes and those with microbial contamination as outcomes failed to quantify behaviour change [40], or only reported changes observed by the trainers [41]-a likely source of bias. Randomised controlled trials on the microbiological effectiveness of food hygiene measures at household levels which used HACCP suggest that substantial reductions in microbial contamination in weaning food are possible [40, 41] but gave no indication of whether such reductions can be achieved in a community-based intervention. One study offered some evidence that improving food hygiene can reduce diarrhoeal diseases and improve child growth, but confounding and bias cannot be ruled out in these cohort studies. [42]. No randomized food hygiene intervention trials with diarrhoeal and nutritional outcomes were identified.

All studies prioritised different behaviours. The WHO five key behaviours for safer food initiatives prioritizes five few key behaviours [62], however, different behaviours may be practiced sub-optimal in different contexts. Though several food hygiene behaviours may be amenable to change, how such improvement can be made through the design and delivery of interventions in different programme delivery contexts is yet to be determined. There is therefore a need to identify the specific behaviours that are critical to minimize the infection at different transmission pathways.

Most studies used educational, messaging and demonstration methods to improve behaviours. Evidence from other behaviour change initiative suggests that simply lecturing and messaging has low effectiveness [4, 63-65] whilst programmes based on theory and
behavioural science and that creatively target emotions and the settings in which behaviour takes place are likely to be more successful [66, 67]. Any future design and delivery of ‘food hygiene interventions’ must be based on scientific principles and emerging understanding of behaviour change.

Measurement of behaviour is always difficult and likely to introduce bias in studies. Previous studies showed that the presence of observers has consistently biased results[68]. However due to a lack of gold standard measures, past studies have considered observation to be a more accurate measure than self-report in handwashing domain [32]. There is still a lack of studies with robust methods to measure the food hygiene behaviours precisely. It is therefore essential to develop food hygiene promotion interventions that are effective to achieve long-lasting behaviour change and those must take this into account by trying to improve measurement techniques and reduce bias.

The intensively tailored domestic food hygiene interventions using the HACCP approach such as in Mali and Bangladesh showed that faecal contamination in weaning food can be reduced at the household level [40, 41]. Both studies assumed that their intervention improved the targeted behaviours and that such improvement had an impact on the reduction of contamination. However, the degree to which the behaviours were improved and how were not reported or discussed. Though these interventions were effective at a small scale, it is not clear whether such interventions would be equally effective if they were scaled up, or indeed whether such interventions are feasible to implement in community settings. Both studies failed to report any details on the programme components, which limits potential opportunities for further replication in different settings. These studies did not report the interaction of their outcomes with socio-demographic variables of the study populations, including other factors such as availability of sanitation facilities, water, soap, cooking fires, refrigerators, designated cooking places, presence of child faeces in the home environment, and availability of an appropriate pot for re-heating. HACCP approach used as a research technique not a community level intervention. There is a need for the development of scalable community-based food hygiene interventions to determine the extent to which ‘food hygiene interventions’ can improve behaviours and reduce contamination in food, diarrhoeal diseases and childhood undernutrition.
Despite one study reporting plausible effects on diarrhoeal morbidity and child growth status [42], it is difficult to attribute this to food hygiene since the interventions targeted a variety of behaviours (sanitation, personal hygiene and food hygiene) and the allocation of the intervention was not random. Reported diarrhoea was recorded at each visit, which is again subject to reporting bias [69]. Since participating mothers knew that the intervention was about reducing diarrhoeal diseases, it is highly unlikely to report the entire occurrence of diarrhoea morbidity [69, 70]. The effect of food-borne contamination on child health during the critical window of weaning and long term development needs to be better understood. In particular, the relationship between foodborne pathogens and malnutrition needs to be better understood. More community-based behaviour change interventions that address these methodological flaws are therefore needed.

This review has a number of limitations. For example, we only reviewed studies in the English language focusing on low-income settings. We only reviewed published studies and did not search for grey literature from other sources such as government or donor reports. Studies had many methodological flaws including reporting of partial results, failing to report participants eligibility, biases in outcomes measurements, inconsistency on the number of targeted behaviours, not including randomization procedures, not revealing or reporting the availability of trial protocols, not reporting sample size calculation, not reporting programme components, potential influence on intended behaviours while collecting microbes sample and not using behaviour change principles were major challenges in the review process. Future research should address these methodological flaws.

Current global approaches to tackling diarrhoeal diseases and childhood undernutrition are often limited to curative or supply-oriented responses, with insufficient attention allocated to preventive measures such as food hygiene. Current evidence is insufficient; hence simple, and scalable food hygiene interventions that target key behaviours and can be implemented in community settings need to be developed and tested to determine their success at changing important food hygiene –related behaviours and the influence of these behaviours on health outcomes in low-income settings.
2.5.2 Nepal specific gray literature review:
While the existing policy framework offers important entry points for hygiene promotion in existing WASH, health and nutrition programmes, no sector in Nepal has set implementation guidelines for hygiene or has developed an understanding on the key hygiene behaviours that require addressing, nor have they an effective operational approach to Behaviour Change. No government or other integrated or cross-sectoral food hygiene interventions were identified.

The health sector policy / strategy environment getting conducive to integrate hygiene into ongoing health programme. The institutional roles and responsibilities for the health sector in relation to hygiene are yet to be clearly established and implemented. The NHSP-III (currently in progress of formulation) offers an opportunity to address these and make health sector accountable for hygiene/ food hygiene promotion.

In WASH sector, the development of “Sanitation and Hygiene Master Plan 2010-2015”[37] ensured enabling policy environment for hygiene including some aspects of food hygiene promotion. However, the absence of an operational guideline or framework for Master Plan implementation remains an important barrier to progress. There is a need of overarching “national hygiene framework” in-line with Master Plan with clear institutional responsibilities for hygiene promotion so that the importance of behaviour change aspects fully realized and institutionalise.

The NAGA initiatives in Nepal [59] led to the formulation of a Multi-Sector Nutrition Plan (2013-2017) [36] and MSNP was as an ideal opportunity to embed food hygiene in different sector strategic plans and ongoing programmes. Though, MSNP does not explicitly mention food hygiene but can be framed under the hygiene component.

No food safety policy existed in Nepal until the end of 2013. No national standard for commonly-used household foods such as for Rice, Jaulo, Dhindo, Curry are set. An overarching ‘food safety policy’ to respond the needs of Nepal’s diverse settings including rural population is required.
2.6 REFERENCES:

1. CDC, CDC Estimates of Foodborne Illness in the United States, 2011, Centers for Diseases Control and Prevention, Atlanta, USA.
11. Marino, D., Water and food safety in the developing world: global implications for health and nutrition of infants and young children. Journal of the AMERICAN DIETETIC ASSOCIATION, 2007 Nov(107(11)): p. 1930-4...


CHAPTER – III

Methods for the design and implementation of the formative research, food hygiene intervention trial and its evaluation

3. Overall methods:
This chapter describes the overall methods used in the two different phases of this study. This includes the methods for the Formative Research (Chapter IV - Step B), the Creation (design) and Delivery of the food hygiene intervention package (steps C and D), and the Cluster Randomized, Before-After study with Control (Chapter V and VI - step E; evaluation of the intervention for primary and secondary outcomes). Due to the paper style format of this thesis, some of the descriptions of the methods given in this section are repeated in the relevant chapters/papers.

3.1 Study setting:
The study was conducted in two rural hill local administrative units (Village Development Committees – VDCs) of Kavre District, Nepal (Figure 1). Each VDC consists of nine wards (clusters) with an estimated total population of approximately 5,500 in each VDC. Kavre district consists of 87 Village Development Committee (VDCs) and 3 municipalities with a total population of 385,218. The majority of the population belongs to the Tamang group (a historically deprived group in Nepal) followed by other major ethnic groups: Brahmin, Chhetri, Newar, Pahari and Bishwokarma. The overall literacy rate in Kavre district was 56%, and only 39% women were literate in 2011. Pneumonia and diarrhoea are the first and second leading causes of death among children under five in the district respectively[1], and around 41% children are stunted[2].

Two VDCs - Baluwapati Deupur and Nayagaun - were selected as the study location. The area was selected taking into account the geographic location (remote hill location and mountainous terrain – 2,195m elevation), socio-economic variability (low-income communities, mixed settlements with a majority from marginalised ethnicities), low sanitation coverage and high diarrhoeal disease prevalence. The landscape is dominated by mountains and hills, and terrace agriculture is widely practiced. Most households subsist through agriculture, small-scale household animal farming, and selling of milk and agricultural
produce. The population is primarily comprised of ethnic minorities (Tamang 63%) and Brahmin/chhetri (28%) speaking multiple languages (Tamang and Nepali respectively). Almost half of the population relies on piped water supply from unprotected sources, and the other half on surface water. Forty four percent practice open defecation, and animal and child faeces are often present in household environment. All houses, including kitchen surfaces, are made of mud and stone; most households cook on the floor using firewood for cooking fuel. In most households, the only room on the ground floor serves multiple functions such as a kitchen (cooking, feeding, keeping waste materials), bedroom, sitting room for the entire family, and sometimes also keeping animals. The majority of the households and kitchens are small, dark and over-crowded.

The **formative research** was conducted in two randomly-selected wards (ward 1 and 4) out of nine in Baluwapati Deupur VDC between April and June 2012. All households with a child aged 6-59 months in the two wards became the study population.

The **Cluster Randomized, Before-After Study with Control** was conducted in eight randomly-selected wards (hereafter referred to as clusters) from the 18 eligible wards of the two VDCs (Baluwpati Deupur and Nayagaun). Inclusion and exclusion criteria were used to select the eight clusters. The trial was conducted between October 2012 and December 2013. The total participant population ranged between 417 – 786 people (75 – 141 households) in each cluster. Households with at least one child aged 6-59 months in the eight clusters became the study population.

Only households with children aged 6-59 months were included in the study due to their increased vulnerability to diarrhoea [3, 4] from consumption of contaminated food and other infections. Despite the fact that many children under 6 months were consuming foods other than breast milk, it was decided to exclude them in order to not contradict WHO recommendations on exclusive breastfeeding [5] by discussing food hygiene.
Figure 1: Study site for Formative Research and Cluster Randomized, Before-After study with Control study in Nepal.
3.2 Ethical approval for all the studies:

Ethical approval to conduct the Formative Research (FR) and Cluster Randomized, Before-After study with Control (BAC) was granted by the Research Ethics Committee of the London School of Hygiene and Tropical Medicine (LSHTM), and the Nepal Health Research Council (NHRC) separately.

Informed consent was obtained from all participants during the formative research. The consent forms were verbally presented to all participating mothers and grandmothers in local language to receive consent for their voluntary participation. The signature (those who were literate) or witnessed (those who were illiterate) was obtained from all participants before proceeding to formative research. Video recording assistants were made aware of all ethical issues prior to data collection. The informed consent forms are included in CD-R annex-II.

Informed consent was obtained from all recruited participants during the intervention trial. A written consent form was provided to or read-out to the participants stating giving details of what was required of them as follows:

- During baseline (social & demographic information)
- While recruiting individual household for the intervention trial
- During baseline and follow-up – behaviour observation
- During compliance measurement (after completion of follow-up measurement)

No judgmental statements were made on opinions or experiences shared by respondents in any interview, discussion or observation. All videos and photographs were taken with the prior verbal permission of the participants. To preserve anonymity, all findings were presented without ascribing names or identifiable personal descriptions. The different informed consent forms are included in CD-R annex-III. The ethical approval letters for the formative research and intervention trials from LSHTM and NHRC are attached in Annex B, C, D and E. Local administrative bodies (Office of the Village Development Committee) and Health Institutions were pre-informed and involved in the study. A project coordination committee was formed in each VDC under the chairmanship of the VDC secretary with representation from local health institutions, local social leaders, teachers and PI of the study (myself) to establish institutional responsibility and get local consensus. The study funder had no role in intervention design, implementation, measurement of outcomes and writing the report.
3.3 Detailed methods - Formative Research (step B):

The formative research served to inform the design of the food hygiene intervention package. The research objectives were to document current food hygiene behaviours and their determinants, to assess the level of contamination in commonly-used child food, to identify critical behavioural control points, and to prioritise key food hygiene behaviours.

Study design:

The formative research primarily employed qualitative and microbiological testing methods supplemented with anthropological and quantitative (survey) methods.

Study participants/households and sampling frame:

Mothers in households with a child aged 6 to 59 months in the two study clusters of Baluwapati Deupur were invited to participate in the study. Altogether, 68 households were eligible and consented to participate.

The sample size for qualitative studies depends on the objectives, study techniques and diversity of the study population [6-8]. The qualitative nature of the formative research primarily and resulting richness of the information gathered for each objective and tools informed the necessary sample size, as detailed in Table 1. All 68 eligible households with a mother having a child aged 6-59 months from two study clusters were included in the assessment of behavioural outcomes. Thirty participating households were randomly selected from amongst the 68 for measurement of the presence of bacteria in food.

Conceptual frameworks:

Two conceptual frameworks were used in the design of the formative research. To identify behaviours likely to cause risk of pathogens transfer, the Hazard Analysis and Critical Control Points approach was used (HACCP)[9-11]. The Evo-Eco theoretical framework[12] was used to understand food hygiene-related behaviours in context.

HACCP is a systematic approach for identification, assessment and control of hazards, involving six steps: i) identify hazards and assess their severity and risk; ii) determine critical control points; iii) specify criteria to ensure control; iv) monitor critical control points; v) take
corrective action; and vi) monitoring and verification. Only steps relevant to the study objectives were used (identify hazards, assess risk, determine control points, and specify criteria for control points). In a nutshell, a flow diagram of the steps in food preparation is constructed and critical control points were identified.

![HACCP components](image)

**Figure 2: HACCP components [9]**

Behaviour Centred Design is a systematic approach for designing behaviour change interventions, underpinned by Evo-Eco theory which was developed and is used at LSHTM [12-15]. The approach pinpoints key behaviours, seeks to identify psychological (habitual, motivated or planned), bodily and environmental (social, biological and physical) causes of behaviour, taking into account behavioural settings, which are akin to theatres of performance with roles, scripts, routines, props, purposes and particular histories[16]. This framework depicts behaviours as a dynamic interaction between bodies, environments and the brain. The environment presents some challenges or opportunities to the brain, which produces potential responses to that challenge, and the body, which produces behaviours that change the environment. When a behaviour is performed the resulting reward provides feedback which determines the likelihood of repeat and of habit formation[17].
Data collection techniques, instruments and methods:

Table 1 briefly summarises the tools, techniques and methods used in the formative research, which included microbiological methods to determine food contamination, behavioural study methods with focus on anthropological and consumer research techniques to understand risk behaviours in social and physical context, and hazard analysis methods to analyse potential hazards and identify control points. Formative research data collection tools are attached in CD-R annex-IV.
<table>
<thead>
<tr>
<th><strong>Methods</strong></th>
<th><strong>Instruments</strong></th>
<th><strong>Total participants / Households (HH)</strong></th>
<th><strong>Strengths</strong></th>
<th><strong>Weaknesses</strong></th>
</tr>
</thead>
</table>
| Mother’s daily routine | • Daily routine analysis  
  • Moment in Life Analysis | • 30 households (n=30)  
  • 30 households (n=30) | Useful to extract common pattern of behaviours in the community | Cultural sensitivity need to be considered. Anthropological skills needed |
| Video filming | • Video cameras and checklist | • 30 households (n=30) | Useful to extract behavioural missed opportunities and capture real life behaviour | Parsing analysis is time and resource consuming |
| ‘Teach the Researcher’ session | • Observation  
  • Observation checklists | • 5 households (n=5) | Useful to identify the physical, biological and social barriers to perform intended behaviours. Offers anthropological insights for researchers. | Subject to observer’s effect if objective is known. Time consuming |
| In-depth interviews / household survey | • Questionnaires & checklists | • 68 households (n=68) | Easy to reach with wider population within short time administering questionnaires | May not be useful to extract behavioural determinants |
| Household observation | • Observation & checklists | • 68 households (n=68) | Offers direct access to social phenomenon. Directness and avoids the self-report problem. Gold standard tool | Resource and time intensive. Susceptible to observer bias and effect. Need to explicitly define the behaviour |
| Focus group | • FGDs Checklists  
  • Barrier mapping | • 9 FGDs with mothers (n=68) | Time efficient. Suitable for groups with less formal | Limited number of questions can be addressed. >one researcher needed. |
<table>
<thead>
<tr>
<th>Methods</th>
<th>Instruments</th>
<th>Total participants / Households (HH)</th>
<th>Strengths</th>
<th>Weaknesses</th>
</tr>
</thead>
<tbody>
<tr>
<td>discussions (FGDs)</td>
<td>• Motives stories</td>
<td>• 2 FGDs with grandmothers (n=15) • 11 motivational exercises (n=83)</td>
<td>education. Instant verification of data by group and easy to understand social norms trend.</td>
<td>Challenge to conclude minority opinion and maintain confidentiality</td>
</tr>
<tr>
<td>Food sample collection and testing</td>
<td>• Sample collection sterile containers, 3M PetriFilm <em>E. coli</em> and <em>Coliforms</em> plates, Thermometer • Lab protocol</td>
<td>105 samples from 30 HHs: • 30 immediately after cooking • 30 during feeding • 30 after 5 hours storage • 15 after re-heating</td>
<td>Good indicator to establish links between behaviour and microbial contamination. Sampling at critical times would suggest whether behavioural control measure can be applied</td>
<td>Collecting samples, maintaining adequate temperature in field, finding an appropriate media and lab to support sample processing needs a lot of preparation, involves time and huge cost.</td>
</tr>
<tr>
<td>Water sample collection and testing</td>
<td>• Sample collection sterile containers, membrane filter, EMB agar, pH meter</td>
<td>• 30 ready to serve water from 30 HHs</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Milk testing</td>
<td>• Sample collection sterile containers, EMB agar / 3M PetriFilm, pH meter</td>
<td>• 13 ready to serve milk from 13 HHs</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Jad (homemade alcohol) testing</td>
<td>• Sample collection sterile containers, EMB agar</td>
<td>• 12 ready to serve Jad from 12 HHs</td>
<td></td>
<td></td>
</tr>
<tr>
<td>HACCP analysis and identification of control points</td>
<td>• Behaviours and microbes assessment • Food flow diagram • Hazard mapping • Identification of CCP/BCP</td>
<td>• Multiple steps and actions</td>
<td>Logical exercise to identify control points and prioritize key food hygiene behaviours.</td>
<td>This is an intensive exercise involving experts from multiple field (behaviour, microbiology) including community people and researcher.</td>
</tr>
</tbody>
</table>
**Behavioural study methods:**

- **Observation of daily routines:** The daily routines of 30 randomly-selected mothers were recorded using the ‘Day in Life Analysis (DILO)’ method. The daily routine of mothers was assessed from early morning until evening paying particular attention to the place of cooking, feeding and eating. Specific actions during cooking were assessed using a ‘Moment in the Life Analysis (MILO)’ approach. Behaviours that are linked with food hygiene and or that are likely to re-contaminate food/water/milk during cooking/boiling are recorded.

- **Video recordings:** To identify commonly-practiced food hygiene behaviours and assess missed opportunities, food preparation, feeding and storage behaviours were filmed in 30 randomly-selected households. Local women were recruited and trained to use hand-held cameras. Different female workers were recruited from different castes / ethnic groups to ensure cultural sensitivity (women from certain castes cannot enter the kitchen of a woman from another caste). The videos recorded mothers preparing, cooking, handling, storing, feeding, and reheating child foods. Filming began prior to commencement of food preparation and followed all aspects of mother’s work and behaviours for about two to three hours, except when privacy was required (for bathing, defecation or urination). Data obtained from video filming were parsed, transcribed and analyzed to identify the missed opportunities against various food hygiene related behaviours.

- **‘Teach the researcher’ sessions:** To gain an emic perspective, five mothers were asked at different times to teach the researcher how to cook their child’s food, while the researcher observed closely the mother’s behaviours during food preparation, cooking, child feeding, storage, and reheating of the child’s food. The physical, and biological barriers to performing food hygiene behaviours in kitchen environment were recorded and obtained evidences were used to triangulate information obtained using other tools. This is a new tool which I have tested in-order to gain an emic view of food preparation behaviour.

- **In-depth interviews and household surveys:** 68 in-depth interviews were carried out with mothers in each household, using a questionnaire containing structured and open-ended questions concerning socio-demographic data, food preparation, handling, feeding, storage and re-heating, common diseases among children, their exposure to health messages, and
their knowledge and attitudes. This exercise was useful to assess prevalent knowledge on food hygiene and practices of mothers, including those related to: handwashing with soap before preparing food, and before feeding child; use of serving utensils, protection of food from insects; flies; animals; cooking temperature; reheating practice and temperature; food storage; keeping food at a safe temperature; use of safe water and milk for cooking/boiling and feeding to child; common food used for children and their access to safe water; and food hygiene etc.

- **Observation**: The physical environment and visible level of cleanliness were assessed in all 68 households, documenting the cleanliness of the kitchen and utensils, the household compound including yard and toilets, and cleanliness of mothers’ and children’s hands. All aspects were assessed using operational definitions and checklists.

- **Focus group discussions (FGDs)**: nine FGDs with mothers and two with grandmothers were conducted to identify commonly-used child foods, understand the food chain supply mechanism, and identify socio-cultural practices around food preparation, handling, storage, feeding and re-heating. Each focus group discussion spanned 1:30 to 2 hours, at a convenient time either before going to the field in the morning (8:00 to 10:00) or during lunch in the afternoon (12:30 to 14:30), with refreshments provided. The FGD were recorded using a voice recorder (Olympus). The recordings and notes were later compiled, translated and transcribed after the FGD.

- **Motivational exercise**: Eleven motivational exercises were performed with mothers and grandmothers to assess immediate motives for the key behaviours. Seven different pictures demonstrating common motives for each behaviour (attraction, nurture, disgust, status/respect, affiliation, purity and disease) were shown and stories were articulated around those pictures to identify the emotional drivers. Mothers were then asked to rank the pictures according to how likely these were to motivate them to practice key behaviours. Some quotations indicative of the rationales used to rate particular motive as important for good food hygiene behaviours were as follows:
  - Nurture: “Mothers good food hygiene behaviours symbolize expression of their love of their children for child’s bright future”
  - Attraction: “Everybody likes safe/clean food, loves people with clean hands, loves eating in clean serving utensils”
  - Affiliation (norms): “If others do I will do, if they don't I will not do it. I would
always practice good food hygiene like everyone else is doing- if they don't, I won't too”

- Status/respect: “People will see me and respect me if I practice good food hygiene behaviours. I will get the respect of my whole family if I practice good food hygiene behaviours”.
- Disgust: “I will feel ‘Shikshik’ to eat/feed food that are briefly cooked, food accessed by flies/dust/dirt. I will feel Shikshik if I offer food to my child without washing hands with soap. If hands smell, village people won't like me”.
- Purity: “I don't feel pure in mind, feel contaminated, Shik, Shik if I don't clean kitchen regularly”
- Diseases: “If I don’t practice good food hygiene practices, my child will get sick of diseases like diarrhoea, and other diseases. I always practice good food hygiene behaviours to protect my child from diseases”.

**Microbiological study methods:**

To study the microbiological contamination of foods the first step was to identify which foods to sample. We identified commonly-used child foods through focus group discussions and household surveys. We found that households used a variety food items for daily consumption and in ceremonies and festivals. There was no specific weaning or child food for children aged 6-59 months. The majority of households just served the same solid food consumed by other family members, sometimes softened by mashing or with the addition of milk or water.

Commonly-used solid foods were *bhat* (boiled rice) or *dhido* (maize, wheat or millet porridge) served with *dal* (pulses) or vegetables (green or dry). Commonly-used child foods, namely rice or *dhindo* along with *dal* or vegetables (whichever was being cooked on the day the sample was collected) were sampled to ensure consistent sampling for microbial assessment across the selected households. 105 food samples were collected from 30 randomly-selected households at four different stages. Water samples from all 30 households, milk samples from 13 households and Jad (homemade alcoholic brew) samples from 12 household were collected by laboratory technicians once during child feeding at mid-day. Total coliforms (TC) and *E. coli*, the WHO-recommended indicator organisms for measuring faecal contamination of samples [18], were quantified in all food and drink samples.

The food sample collection stages were as follows:

- **First food sample collection (30 samples):** immediately after cooking (5 to 15 minutes), directly from cooking vessels.
- **Second food sample collection (30 samples):** during feeding, from mother’s/child’s hand (first feeding immediately after cooking, from mother’s hand if mother is feeding or from child’s hand if child is self-feeding).
- **Third food sample collection (30 samples):** stored or leftover food after five hours’ storage (±15min) directly from storage containers.

- **Fourth food sample collection (15 samples):** immediately after reheating (0 to 5 minutes), directly from re-heating vessels.

**Sampling procedures:**

50 gram food samples were collected, using 60ml sterile and labelled plastic containers with tight-fitting lids, using sterile wooden spatulas, when needed. Temperature was recorded just before collecting each sample using sterile thermometers. Similarly, 250ml water samples were collected using sterile and labelled containers and 50ml milk and Jad samples were collected using labelled containers. All samples were stored on ice in an insulated cooler box immediately after collection, and transported to the laboratory within four hours of collection, maintaining a temperature below 6°C. All samples stored overnight at the lab were maintained at the same temperature and processed early in the morning the following day. Each household from which food samples were collected received NRs 200 (~2US$), the approximate equivalent cost of the amount of food collected.

**Hazard analysis:**

Following the behavioural and microbial assessments, a thorough food flow diagram (food supply chain from collection of raw materials to feeding) was developed based on reported and observed patterns of food material processing, food preparation, storage and feeding. Potential sources of hazards and possible contamination and recontamination were documented. The risk behaviours and actual levels of contamination were then compared.

**Formative research study management and staff:**

I led the formative research as Principal Investigator. In addition, two research assistants were recruited and trained to support field data collection and supervision work. Five video recorders and 4 field data collectors were locally recruited and trained to collect data using various tools. Data collectors submitted filled form to research assistant by the end of the each day. Forms were reviewed on the same very day in the field and data quality was checked. Video recorders also handed over data card after completion of the recording. I have reviewed and parsed all the films, transcribed and analyzed using simple format. Food, water, milk, and Jad samples were collected using 4 trained lab technicians (sample collectors). Samples were analysed at local lab in Kathmandu using 4 trained lab technicians and a microbiologist. A
necessary coordination and technical inputs were received from my PhD supervisor, and PhD advisory committee members (as needed).

**Data analysis:**

**Behavioural analysis:**

Interviews and FGDs were recorded and transcribed. Videos were parsed and used to identify missed opportunities for food hygiene. The food flow diagram was used to visualise likely hazards and identify critical control and behavioural control points. The findings were organized following the common themes of behavioural and environmental determinants using Evo-Eco principles [12]. Main themes includes i) behavioural determinants such as daily routine, common child food, food preparation, utensil cleanliness, food feeding and handwashing, storage, re-heating practices, and water and milk feeding etc, ii) environmental determinants such as social, physical, and biological, and iii) other relevant findings were categorised under specific themes such as associated motives and barriers, hazard analysis and determination of control points. Mostly deductive methods was used to analyze data using defined themes using Evo-Eco principles but some of the emerging themes were also considered using inductive formative research methods. For analysis of the determinants of behaviour, intentions, knowledge, motivations, habits and routines, objects employed, contamination, and physical and social settings including environmental factors were considered under each theme. Relevant verbatim are presented in the respective section.

The first step in analyzing the script data was to determine which events tend to follow from others in the reported daily pattern of activity in this population. If there was similarity between the routines of different women, at least in terms of a common order of events, then such routines could be compounded or aggregated, and any intervention targeting particular points within a routine can be expected to work similarly for the target population. Data obtained from video material were transcribed and analysed to identify the missed opportunities for practicing various food hygiene related behaviours.

Quantitative data (observations, survey, motives mapping, mothers’ daily routine) was entered and analysed using the IBM SPSS Statistics 19 and the frequencies of relevant behaviours were reported along with qualitative information. Qualitative data was analyzed using ATLAS.ti 7 Qualitative Data Analysis Software using specific themes.
Microbiological analysis:

Total coliforms (TC) and E. coli colony counts per gram of food (cfu/gram of food) and cfu/ml of milk were quantified using 3M™ PetriFilm[19] E. coli and Coliform Count Plate[20] media. Membrane filters and Eosine Methylene Blue (EMB) agar media were used to quantify total coliforms and E.coli in cfu/100ml of water and cfu/ml of Jad in a specialised laboratory. Sample processing, inoculation, incubation and interpretation (colony counts) were done by trained lab technicians with the support from microbiologist. Colony counts raw data were first entered into excel sheet in lab and results were handed over to me. I have thoroughly monitored and supervised all the process in the lab as per protocol. After entering raw data into SPSS by myself, food samples with total coliforms and E. coli counts of <10 cfu/grm, between 10 and 100 cfu/grm, and >100 cfu/grm of food were categorised as low risk, medium risk and high risk respectively. The colony counts were log-transformed (log_{10}) to compare the mean counts at four different stages for food. The temperature of food samples and pH of milk and water samples were assessed. The specific procedures applied to sample processing, interpretations and quality control are as follows:

<table>
<thead>
<tr>
<th>Food, milk, water and Jad processing, interpretation and quality control procedures:</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Food sample processing:</strong> Of each 50 gram food sample, 10 grams were weighed and suspended in a vial containing 90ml of sterile Maximum Recovery Diluent (1:10 ratio). The sample was vortexed for a maximum of one minute. Multiple serial dilutions 10^{-1} to 10^{-4} were carried out. 1ml of the food suspension was then transferred to the middle of the labelled 3M Petrifilm, E-Coli and Coliforms Count Plates [20] with the sterile barrier filter tip of a micropipette. The cover was rolled down slowly on the inoculated media to avoid trapping any bubbles. The sample was spread by pressing gently with the spreader. The inoculated petrifilm was allowed to stand at room temperature for at least one minute before incubation to allow the media to gelify. The inoculated petrifilm plates were incubated to detect the total coliforms for 24 hours +/- 1 hour at 37°C +/- 1°C, and to detect the E. coli, plates were incubated for 48 hours +/- 1hour at 37°C +/- 1°C. The reading was done following AOAC, international all foods method 991.14 as recommended by 3M PetriFilm. All colonies with gas (blue and red) at 24 hours were counted as confirmed coliforms, and only blue colonies with gas at 48 hours were counted as E. coli. Such individual colonies were counted and used to calculate the total coliforms and E. coli load as colony forming units (cfu) per gram of food respectively. The same methods were used for processing milk samples.</td>
</tr>
</tbody>
</table>
Water and Jad processing: Membrane filters with 0.45μm pore size were used to filter the water samples. The filter was then transferred onto the surface of dried Eosine Methylene Blue (EMB) agar. The EMB plates were then incubated at 37°C +/- 1°C for 24 hours in an inverted position. The pH of each water sample was measured and recorded before filtering. All colonies on the EMB agar presenting as dark purple nucleated colonies with green metallic sheen were recorded as E. coli, and dark purple nucleated colonies without green metallic sheen were recorded as total coliforms. Such individual colonies were counted and used to calculate the E. coli and total coliforms load as colony forming units (cfu) per 100ml of water respectively. The Jad samples were diluted in sterile Maximum Recovery Diluent (MRD) to 10^{-2} to 10^{-6} depending on the countability of the overnight culture. 0.1ml of diluted Jad samples were then transferred onto the surface of dried Eosine Methylene Blue (EMB) agar. Incubation and interpretation was done using the same method used in the analysis of the water samples to count E. coli and coliforms loads as colony forming units (cfu) per ml of Jad respectively.

Quality control: The specific techniques were used to ensure the data generated would be valid and a reliable representation of the contaminant levels in this community. One blank 3M PetriFilm was placed in the incubator and in the laboratory; for every 25 samples assayed. If the blank samples showed any TFC bacterial growth, contamination is present. Positive controls (spiked with E. coli and Kliebsiella spp) were also run to test the media, to show positive TFC bacterial growth. If the results were negative, either the media is flawed or the equipment such as the incubator temperature is incorrect. No contamination occurred.

Critical and Behavioural Control Point identification (CCP&BCP) and behaviour prioritisation:
Critical and behavioural control points were identified as points at which control measures can be applied, and whether loss of control could result in a potential hazard. Control points were used to assess whether specific behaviours impact on bacterial destruction and reduce their propagation. Decisions to prioritise key behaviours were made based on the identified control points, depending on whether behavioural control measures could be applied and whether relevant behaviours were adversely practiced during the formative research. Formative research results and findings are discussed in chapter IV. A five minute video documenting prevalent food hygiene behaviours was also produced and attached in CD.
3.4 Design of the food hygiene intervention package (step C):

After completion of the formative research, the next step was to create a food hygiene intervention package.

Based on the formative research findings, a ‘creative brief’ (Annex F) was produced providing study background and rationale, study context, prioritised behaviours and reasons for prioritisation, the physical, social and biological settings of the study area and participants, basis for intervention design (simplicity, scalability, target behaviours, emotional drivers and behaviour settings, branding, positioning, delivery mechanism, scale, target audience, etc.), prevalence of behaviours and scope of work for the creative team. Past experiences, in particular from small-scale weaning food studies in Mali[21], Bangladesh[22], Brazil[23], the SuperAmma handwashing trial in India [15], and the WHO “five key behaviours for safe food” initiative[24] were carefully examined before designing the package.

A multidisciplinary creative team was assembled in Nepal, bringing individual expertise from the private sector, programme development, marketing, social and community mobilisation, behaviour change and communication, curriculum design, research, and art fields. The creative brief and a visit to the study area allowed the team to understand the context, feasibility, setting and socio-cultural aspects of the intervention.

Figure 4 highlights the logical flow of ‘Theory of Change’ [25] for the food hygiene intervention which was used while systematically design the intervention. Theory of change guided the process of change by outlining causal linkages such as defining inputs, activities, outputs, outcomes and impact. The reduction of microbiological contamination in food as impact and the improvement in five key food hygiene behaviours as outcomes were determined. Various innovative activities linking with emotional drivers such as nurture, disgust, affiliation and status were identified through the creative process to implement within the behaviour environment such as in physical, social and biological – as a function to influence behaviours. The whole food hygiene intervention package and its implementation modality became the input.
A prototype package was developed through an interactive process based on settings and prioritised barriers and motivations. The emphasis was placed on simplicity, feasibility, scalability, cultural acceptability and the use of user-friendly language. We focused on using novel rather than traditional behaviour change approaches, which have often been ineffective [26, 27]. The operational feasibility of the package was assessed and all tools were pre-tested in the field. The package was finalised after incorporating stakeholder feedback on the prototype.

Figure 4: Theory of change for the food hygiene intervention

Figure 5: Work with creative team
The promotion package was designed to be implemented over a three–month period through six events (2 community event and 4 group events) followed by six door-to-door household visits. Each event was designed around a specific motivational theme: ‘Nurture’ (bringing affection and happiness into the family through ensuring an enabling food hygiene environment for the successful child); ‘Disgust’ (piquing disgust by highlighting the risks of poor food hygiene); ‘Social Respect’ (ensuring social respect in the community with a desire to be an ‘ideal mother’); and ‘Affiliation’ (creating a group consensus and moral attraction towards food hygiene behaviours as part of a social desire to establish group norms). The campaign’s theory of change was that mothers would identify with a central ‘ideal mother’ character, who practiced safe hygiene so as to be respected in the community (status motive). In addition Nurture, Disgust and Affiliation as possible key levers of change, and recognised the need to disrupt daily food preparation habits that were held in place by tradition, routine and the social and physical settings of kitchens in which the target behaviours took place.

Six key behaviours were identified through formative research, of which five adversely-practiced food hygiene behaviours were prioritised for targeting through the food hygiene intervention trial:

- 1: Cleaning of child food serving utensils using soap/ash just before serving food
- 2: Handwashing with soap by mother before feeding, and by child before eating
- 3: Proper storage of cooked food in containers with a tight-fitting lid (to prevent contact with flies/dust)
- 4: Thorough re-heating of leftover/stored food just before feeding child (≥70°C)
- 5: Serving of treated water or boiled milk

Figure 6 highlights the six identified behaviours (from 0 to 5) and five prioritised targeted behaviours (from 1 to 5). Behaviour 0 – thorough cooking – was dropped because it was found that food was almost always properly cooked at the outset.
A programme implementation guide for the implementation of the promotion package was developed. The campaign’s identity was built around the ‘Ideal Mother’ as an inspirational character, creating psychological motivation for all mothers to attain this ideal. The campaign slogan was “Safe Food, Healthy Child”, supported by tailored branding consisting of a logo and colour-scheme exclusively identified with the campaign. Mothers with a child aged 6-59 months were the primary target audience as they are the main actors in food preparation and child feeding. However, efforts were made to involve other family members in the campaign including grandmothers.

The package was designed to be delivered by locally recruited female ‘Food Hygiene Motivators’ (FHM), in similar capacity to that of Nepal’s Female Community Health Volunteers (FCHVs), in order to ensure future scalability. 15 FHM (3 of whom were already FCHVs) were recruited locally to implement the campaign. While selecting the food hygiene motivators in each intervention cluster, first preference was given to Female Community Health Volunteers (FCHVs) as they had previous exposure of leading some form of health promotion programme. Out of four already available FCHVs by default in four intervention clusters, three of them were selected as food hygiene motivators and remaining 12 motivators were locally recruited through the process supported by project coordination committees. All
food hygiene motivators have at least school leaving certificate (SLC) education and or similar capacity with the Nepal’s Female Community Health Volunteers (FCHVs).

A five-day residential training programme was organised for all motivators in order to enhance their food hygiene knowledge, improve their technical capacity, develop skills to conduct community events and household visits, and enhance their interpersonal communication skills. The training aimed to ensure that FHMs could implement the food hygiene promotion package independently. We paid attention to motivating the motivators as Champions of food hygiene:

- Two food hygiene motivators were declared as best food hygiene motivators based on their performance; for example how quickly they were able to declare the majority of their mothers as ‘ideal mothers’ and able to declare respective cluster as ‘safe food hygiene zone’.
- They received 5 days skilled based training certificate, three months work experience certificate which can be used for future references and also received a token of appreciation.
- They received monthly monetary incentives
- They had chance to be a role-model in the society thereby to increase their social status.

Figure 7: Food hygiene motivators including study team; FHM training.

A small pre-trial pilot of procedures, techniques and materials was carried out over a period of 15 days in order to allow FHMs to practice the intervention package in a non-study area. This helped to identify difficulties in executing the intervention and build the confidence of the FHMs.
The package of tools, activities and approaches are briefly described in Step D (delivery of campaign in subsequent section). To deliver and evaluate the effect of the intervention on mothers’ food hygiene behaviours and on the level of contamination in food, a Cluster Randomized, Before – After study with Control (BAC) was implemented. The trial methods are set out in the following section.
3.5 Detailed methods - Cluster Randomized, Before – After study with Control (BAC)

This section sets out the methods of the food hygiene intervention trial. The primary outcome was the effects of the intervention on mother’s food hygiene behaviours and the secondary outcome was the effects of intervention on the level of microbial contamination in food.

Study design:

The trial employed a Cluster Randomized, Before-After study with Control (BAC) design. Cluster randomization was employed because the intervention was applied at a community level. Although the study followed procedures related to true-experimental design (cluster randomized trial) during randomization, the outcomes were analysed using BAC design due to the small number of clusters (eight).

Selection of study clusters, study population and recruitment:

**Village / cluster level sampling:**

Eight clusters were randomly selected from 18 eligible clusters in the two VDCs (Baluwapati Deupur and Nayagaun) using the inclusion and exclusion criteria outlined in Table 2.

<table>
<thead>
<tr>
<th>Inclusion criteria</th>
<th>Exclusion criteria</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cluster (wards):</td>
<td>Cluster (Ward)</td>
</tr>
<tr>
<td>• Clusters within the two selected VDCs</td>
<td>• Clusters in which formative research was conducted</td>
</tr>
<tr>
<td>• Clusters having &gt;30 households with a child aged 6-59 months</td>
<td>• Clusters in which pre-trial activities were conducted</td>
</tr>
<tr>
<td>• Clusters with heterogeneous population settlements (minimum two ethnic caste groups)</td>
<td>• Any clusters (village) geographically attached to one another (adjoining villages/clusters were randomly excluded to avoid contamination)</td>
</tr>
</tbody>
</table>

**Household level sampling:**

A list of eligible households with a child aged 6-59 months in the eight clusters was obtained from local health institutions and physically verified with the help of Female Community Health Volunteers and locally-recruited data collectors. Within the eight clusters, households with children aged below 6 months and above 5 years were excluded. Within each cluster a sample of 29-30 households having at least one child aged between 6-59 months was
randomly selected to participate in the study. Random selection from the list of eligible households was carried out by the PI of the study (myself) using Microsoft Excel. The final sample included 239 households from eight clusters. Written informed consent was received from mothers in all participating households.

**Social, demographic and economic information collection:**
Detailed social, demographic and economic information was collected from each eligible household using a closed-ended structured questionnaire. Mothers with a child aged 6-59 months were the respondents. This information included details such as the number of children aged 6-59 months, mother’s level of education, profession, ethnicity, household income and source, access to basic services including water and sanitation facilities, availability of refrigerators and soap at home, types of cooking fuel used, etc. Information was also collected regarding feeding practices, such as types of food fed to young children, feeding times, etc. Socio-demographic information data collection tools are included in CD-R annex V.

**Randomization:**
Figure 9 shows the flow diagram of the trial. After socio-demographic information collection and baseline outcome measurement, the eight clusters were allocated into intervention and control clusters in a 1:1 ratio (four intervention and four control clusters). Allocation of intervention and control clusters was random and each cluster had an equal chance of being in either group. For randomization, eight clusters were listed (1 to 8) and sent to UK (one of my study advisers) for random allocation using a random number generated using Microsoft Excel. Written informed consent (witness informed consent in case of illiterate mothers) was obtained from each household before cluster randomization (after selecting eligible clusters using inclusion criteria). Four clusters were included in each group since increasing the number of clusters can increase the plausibility in a BAC study; i.e. one independent cluster per arm is better than none, two are better than one, and three are better than two[28]. After randomisation, there were 120 (30 HHs per cluster × 4 cluster) and 119 (29/30 HHs per cluster × 4 cluster) households with a child aged 6-59 months in the intervention and control groups respectively. The four intervention clusters received the food hygiene intervention; no intervention was delivered in the four control clusters. The baseline and post-intervention follow-up outcome assessment was conducted 6 weeks (45 days) before and 6 weeks (45 days) after completion of the intervention in both groups. Behavioural outcomes were measured in
all 239 households, but microbial contamination in commonly-used child food and water was assessed using samples taken from a randomly-selected subsample of 80 (40 intervention and 40 control) households.

**Masking:**

Intervention group knew that they had been received the intervention, hence it was impossible to fully mask (blind) the study participants. The primary outcomes of interest (composite performance = the mean proportion of mothers sustaining all key food hygiene behaviours) were measured by food hygiene observers not connected with the intervention implementation, in order to reduce observer bias. To minimise the impact of the observers’ presence on mothers’ behaviour, mothers were told that the purpose of the observation was to document their daily routine. Participating mothers thus saw no connection between the outcome measurement and the food hygiene campaign. Food and water samples were collected by trained lab technicians unconnected to the study. Another set of independent microbiologist and lab technicians processed, homogenised, inoculated and interpreted the results at a certified laboratory in Kathmandu. Food sample collectors and lab technicians were blinded as to the intervention status of the samples. Lab technicians didn’t know whether they are analysing samples from control or intervention group. No further masking of participants or food hygiene observers was possible due to the nature of the intervention.
The food hygiene campaign was implemented over three months, during the May-August 2013 rainy season. The campaign was delivered by female Food Hygiene Motivators through six events (two community and four group events) followed by six door-to-door household visits, totalling 12 ‘exposures’ for each mother. The community events were organized in public places and group events mostly in private household compounds. To deliver the campaign more effectively in small groups, each cluster was divided into three sub-groups based on population settlements (10-30 household) and at least one FHM was responsible for each sub-group. Every 15 days, one joint community/group event (in each cluster) or sub-group event was held followed by door-to-door household visits by FHMs. Events lasted an average of 3.5 hours (min. 2.5hrs to max. 4.5hrs). Household visits lasted 1.5hrs on average (min. just over 1hr to max. 3hrs). Additionally, half-day school sessions were organised targeting students and teachers in four Government schools within each of the four intervention clusters.

A summary of the intervention components is provided in Table 3 below. The campaign implementation details can be accessed online at (http://www.sharereresearch.org/NewsAndEvents/Detail/om_blog_food_hygiene_study). A 19-minute video documentary detailing the actual delivery of community and group events and household visits is available online and digitally (https://www.youtube.com/watch?v=gPj6lyN0ZCU). Video documentary is also attached in CD-R.

The intervention package was delivered as follows.

- **Change agents:** locally recruited and trained female ‘food hygiene motivators’
- **Period:** May – August 2013
- **Activities per cluster:** two community events, four group events and six door-to-door household visits.
- **Approach and branding:** campaign approach, logo, slogan (Safe Food, Healthy Child), colour scheme and programme song (installed in all mothers’ mobile as ring tone).
- **Identity/unifiers:** ‘Ideal mother’ as an inspirational character, creating psychological motivation for all mothers to attain this.
- **Guiding document**: a programme implementation guideline and tools
- **Primary target audience**: mothers having a child aged 6-59 months and grandmothers
- **Behaviour change principles**: Each event was designed around a specific motivational theme such as ‘Nurture’, ‘Disgust’, ‘Social Status’, and Affiliation. The campaign changed the physical and social settings of the kitchen (behavioural settings).

- **Activities and tools**: Detailed activities/tools presented in Table 3, few key activities / tools included **situation analysis** through storytelling, video-clip and behaviour illustration demonstration, **behaviour reinforcement** using 3D flipcharts, illustrations highlighting key messages and motives, programme ringtone (highlighting key messages) installation in mothers’ phone, **use of emotional drivers** such as through ‘child life game’, ‘letter exchange’, ‘family drama’ (to create **nurture motive**), ‘puzzle games to reinforce the cues placed in the kitchen’, ‘folk songs’, ‘commitment’, ‘competitions’ (to create an **affiliation motive**), ‘hot potato’ games, ‘3M PetriFilm demonstration’, ‘glo-germ’, ‘disgust exercise’ (to create a **disgust motive**), disgusting pictures demonstration to avoid social exclusion; **habit formation** through engaging activities and demonstration such as cooking and re-heating, use of illustrated baby bib with the message “did you wash your hands with soap before feeding me?” as reminder for handwashing with soap, watch-dog exercise for cross-learning and monitoring, **competitions and pledging** to establish **social norms** such as ‘**clean kitchen**’, ‘**ideal mother**’, ‘**safe food hygiene zone**’, social/public pledging such as placement of ‘ideal mother photos and safe food hygiene zone billboard in the junction of the village’ (to create a **social status** motive), rallies chanting “Ideal Mother Hi Hi,
Diarrhoea Bye Bye!” public commitments and identification of volunteers mothers from each group for **sustainability monitoring**.

![Image](image.png)

*Figure 11: Campaign delivery*

We also set out to change the behavioural settings for the 5 key behaviours. This involved not just changing the physical context of the kitchen, but changing the script in mother’s heads and the pressure that she feels to carry out the behaviour from local social codes. Settings are needed to meet the social and physical need of the people and people’s behaviours are also determined by physical and social settings. Mothers were continuously practicing pre-determined behaviours in the kitchen therefore it was essential to change the behavioural setting. The undermining of behaviour settings demonstrates the dynamics, purposeful coercive nature of the ecological environment with respect to people when they are components of behaviour settings[29].

The change in behavioural setting used as a concept – includes the physical ‘stage’ the script in mothers heads, the social norms of behaviour belonging to that settings, the infrastructure and the objects[12]. Behaviour within particular settings are predictable, if one knew only the physical and social context, and the role of an individual played[30]. Using this concept, attempt was made to slightly change in kitchen settings to encourage and reinforce targeted behaviour.
The key feature was ‘kitchen makeovers’ to change behavioural settings. Based on the idea of settings as an important determinant of behaviour([12, 14]), mother’s including respective family members met in a group to carry out a ‘kitchen makeover’: they discussed the changes that they wanted to make to support the new behaviours and transform their kitchen areas.

Kitchen makeovers used coloured bunting for kitchen demarcation using coloured ribbon and behaviours flag after cleaning, danglers placed at eye level in kitchen as visual reminder for all behaviours, and the fan, ideal mother board, and branded fire-blowing instrument (dhungro) also placed in kitchen. The use of such tools from the outset was expected to encourage behaviour change and transform the kitchen environment from a one-room household used for cooking, eating, running of the household, sleeping, and at times keeping animals to a beautiful, bright and special small kitchen in one corner of the room, with a partition to keep animals away. All households in intervention cluster got a makeover. The social component of the makeover including public commitment to make kitchen clean created a new ritual.

*Figure 12: Kitchen makeover (demarcation using bunting) and use of eye danglers*
<table>
<thead>
<tr>
<th>Events / visits</th>
<th>Purpose</th>
<th>Key content</th>
</tr>
</thead>
<tbody>
<tr>
<td>First community event (3hrs)</td>
<td>Raise awareness of, generate interest in, and elicit commitment to the campaign and the five food hygiene behaviours.</td>
<td>Distributed invitation card a day before the event. Programme ritual (put-up back-drop banner, nail cutting, handwashing with soap, putting on programme badges) initiated. Programme jingle introduced. Campaign objectives described by social leader. Situation contextualised via situational analysis – story, flex with pictures, video clip (disgust motive exploited). Five food hygiene behaviours and their benefits presented. ‘Ideal mother’ introduced as a source of inspiration. Public commitment oath taken, and certificates distributed. Public rally chanting - “safe food, healthy child, we want ideal mother’.</td>
</tr>
<tr>
<td>First household visits (3hrs)</td>
<td>Remind mothers of public commitment; change settings reinforcing the desired behaviours (particularly kitchen cleanliness).</td>
<td>Programme jingle installed on mothers' phones. Kitchen compared with 'ideal kitchen' using clean kitchen illustration. Kitchen demarcated with ribbons and flags reminding mothers of the food hygiene behaviours. Danglers placed at eye-level (round illustration of all behaviours, ideal mother board, dhungro – a branded fire blowing instrument). Importance of food hygiene behaviours refreshed via a brief talk using a 3D flip chart. Three-month work plan formulated to ensure each mother meets the public commitment.</td>
</tr>
<tr>
<td>First group event (3hrs)</td>
<td>Reinforce programme ritual; establish group norms/habits for all behaviours; and generate interest in having clean kitchens.</td>
<td>Programme ritual carried out. Mothers' experiences of changes in their kitchens shared. Group norms elicited via cooking demonstration. Benefits of five food hygiene behaviours reiterated via visual aids (3M PetriFilm, Glo-germ lotion before feeding). Bibs with the message “did you wash your hands before feeding me?” distributed as reminder/reward for HWWS. ‘Clean kitchen’ competition announced (putting-up clean kitchen indicators in the village).</td>
</tr>
<tr>
<td>Second household visits (2hrs)</td>
<td>Reinforce correct food hygiene behaviours with the view to these becoming habitual.</td>
<td>Mothers’ preparation of food observed and corrected where necessary. Importance of five food hygiene behaviours reiterated (used 3M PetriFilm, glo-germs, bib, plastic bucket for handwashing, kettle for boiling water). Mothers reminded about ‘clean kitchen’ competition.</td>
</tr>
<tr>
<td>Second group event (3hrs, 15min)</td>
<td>Increase mothers' confidence; link food hygiene behaviours with affiliation, nurture and status; generate interest in becoming an 'ideal mother'.</td>
<td>Programme ritual carried out. Obstacles faced by mothers shared and strategies for overcoming these discussed. 'Child Life Game' played – the future that mothers want for their children discussed and linked to the five food hygiene behaviours (nurture motive). Puzzle game played to encourage kitchen cues (social respect motive). Folk song composed by mothers conveying key food hygiene messages - affiliation elicited. 'Ideal mother' competition announced. Behaviour reminder ‘fan’ reflecting five behaviours and ideal mother sticker distributed.</td>
</tr>
<tr>
<td>Third household visits (2hrs)</td>
<td>Establish reheating and boiling as social norms; ensure a conducive family environment exists to practice behaviours</td>
<td>Mothers’ food reheating practices observed and corrected where necessary (noting re-heated temperature, motivated to use appropriate vessel to re-heat food and kettle to boil water). Family meeting held to promote food hygiene behaviours (using 3D flipchart). Mothers reminded about ‘clean kitchen’ and 'ideal mother' competitions (visual cues). Anonymous visits performed (by field staff and coordination committee).</td>
</tr>
<tr>
<td>Third group event (3hrs)</td>
<td>Show that implementing the five food hygiene behaviours will avoid disgust and social exclusion and will increase social prestige / happiness.</td>
<td>Programme ritual carried out. Mothers participated in disgust exercises (glo-germs used in food, plate, bowl, glass, spoon) and games (hot potato game using disgusting and safe pictures to demonstrate social inclusion and exclusion). 'Safe food hygiene zone' competition announced. 'Clean kitchen' competition winner announced and publically commended, thereby conferring prestige. Participants and guests visited winner’s house to encourage and share learning.</td>
</tr>
<tr>
<td>Fourth household visits (2hrs)</td>
<td>Create peer pressure, build confidence and reduce observer bias in observation of mothers' 5’ behaviours</td>
<td>Peer-review (watch-dog) exercise carried-out (element of secrecy entailed) by peer mother. Observer mother reported back practices. Mothers reminded about 'ideal mother' and 'safe food hygiene zone' competitions. Mothers three month work plans reviewed. Anonymous visits performed (by field staffs and coordination committee).</td>
</tr>
<tr>
<td>Fourth group</td>
<td>Reiterate that implementing the five behaviours</td>
<td>Programme ritual carried out. Advice provided by mothers to a fictional mother (Dhukhimaya) experiencing social, environmental and attitudinal</td>
</tr>
<tr>
<td>Events / visits</td>
<td>Purpose</td>
<td>Key content</td>
</tr>
<tr>
<td>-----------------</td>
<td>---------</td>
<td>-------------</td>
</tr>
<tr>
<td>event (2hrs, 30min)</td>
<td>food hygiene behaviours will increase social prestige and status; encourage men to participate</td>
<td>barriers to adopting food hygiene behaviours. A drama (family member role play) showed how to become an ideal mother and tackling social, attitudinal and physical barrier. 'Ideal mother' competition winners announced and publically commended (ideal mother photo placed in the junction of the village), thereby conferring prestige. Men involved in the event and celebration.</td>
</tr>
<tr>
<td>Fifth household visits (1hr)</td>
<td>Reinforce food hygiene behaviours; mothers’ self-evaluate their food hygiene behaviours</td>
<td>Mothers’ work plans reviewed. Mothers' food hygiene behaviours observed (ongoing progress). Mothers' performance self-evaluated publically. 'Safe food hygiene zone' indicators reinforced.</td>
</tr>
<tr>
<td>Second community event (4hrs)</td>
<td>Ensure food hygiene behaviour change is sustained post-intervention by further entrenching behaviours as social norms and prestige-conferring practices</td>
<td>Programme ritual carried out. Response received from Dhukhimaya linking food hygiene behaviours to child health and social status. Encouraging social norms by re-performing folk songs. Mothers volunteer (who had at least non-formal education) to continually monitor the community’s food hygiene behaviours. Mothers publically re-pledge their commitment to sustainable food hygiene behaviour change (appreciation certificate distributed). Experiences of stakeholders heard. Remarks from social leaders, guests, representatives link food hygiene as to social respect. 'Safe food hygiene zones’ declared and billboards erected at each entry point of the cluster. Group photo session performed. Community rally chanting “we want ideal mother, ideal mother hi-hi, diarrhoea bye-bye” and using local music and programme song. Intervention formally closed.</td>
</tr>
<tr>
<td>Sixth household visits (1hr)</td>
<td>Entrench food hygiene behaviours into mothers’ daily routine and identify any remaining barriers to these practices; ensure sustainability</td>
<td>Sustainability work plans formulated by mothers. Ease of implementation of food hygiene behaviours analysed by participants (pile sorting exercise using illustrations) and feedback provided. Sustained behaviour change pledged by entire families. Household visits formally end.</td>
</tr>
</tbody>
</table>

**Process documentation:**

Details of the food hygiene campaign, key learnings, and actions taken throughout the campaign implementation period were documented. With an aim to generate interest, provide greater insights on how to design, implement and evaluate food hygiene behaviour change interventions and share key successes/challenges among WASH, Health and nutrition sector professionals, academicians, development practioners and donor communities, a blog was set-up on the SHARE website and shared every month through their newsletter ([http://www.shareresearch.org/NewsAndEvents/Detail/om_blog_food_hygiene_study](http://www.shareresearch.org/NewsAndEvents/Detail/om_blog_food_hygiene_study)). The daily work of FHMs was monitored by locally recruited two campaign assistants. Three review meetings were held together with FHMs and campaign assistants in three months and one final review meeting was held at the end of the campaign to document their learning / experiences. Throughout the campaign implementation period, mothers’ experience and knowledge was also recorded verbatim (Annex G), case studies documented (see Annex H for an example) and relevant images were captured (Annex I). A project coordination committee was formed at
the beginning of the intervention, which included representatives from VDCs, local health institutions, local social leaders, teachers, and the PI of the study (myself). The project coordination committee met four times (inception phase, launching, mid-term review, and post-completion review). Committee members were also involved in campaign monitoring during implementation. Before actual implementation of the trial, list of activities were performed which are captured and now presented in CD-R annex VI.

3.5.2 Outcome assessment (step E):
This section includes the sample size for primary and secondary outcomes and details the methods used to assess:

- The effect of the food hygiene intervention on mothers’ food hygiene behaviours as a primary outcome;
- The effect of the food hygiene intervention on levels of microbiological contamination in food as a secondary outcomes; and
- The reach of the intervention (compliance measurements)

Sample size
The sample size required to measure the primary and secondary outcomes before and after the intervention in both groups was estimated using STATA 12.0.

Sample size for primary outcomes (behavioural outcomes): A sample size of eight clusters with a minimum of 28 households per cluster for two sample comparisons of proportions (two-sided test) using 95% confidence interval (p<0.05), 90% power, 5% loss to follow-up (0.05), 1.29 design effect, allowed for detecting a difference of 20% in cluster prevalence of key food hygiene behaviours between the control and intervention arms. Thorough re-heating practices were used as a base because this practice required the greatest sample size as compared to other behaviours (assuming 7% thorough reheating practices in the control group and 27% in the intervention group).
Table 4: Summary parameters for sample size calculation – primary outcomes

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Estimate</th>
</tr>
</thead>
<tbody>
<tr>
<td>Type I error ((\alpha)): significance level (p-value)</td>
<td>0.05 (95% confidence level)</td>
</tr>
<tr>
<td>Type II error ((\beta)): power of test</td>
<td>90% (Type II error of 10%)</td>
</tr>
<tr>
<td>Proportion observed in control group ((m_1))</td>
<td>0.07 (7% thorough re-heating)</td>
</tr>
<tr>
<td>Proportion expected in intervention group ((m_2))</td>
<td>0.27 (27% thorough re-heating)</td>
</tr>
<tr>
<td>Ratio of sample sizes (exposed : unexposed)</td>
<td>1</td>
</tr>
<tr>
<td>Types of test</td>
<td>Two sided test</td>
</tr>
<tr>
<td>Estimated loss to follow-up</td>
<td>0.05 (5% loss to follow-up)</td>
</tr>
<tr>
<td>Design effect (DE)</td>
<td>1.29</td>
</tr>
<tr>
<td>Required sample size: n1 (control group) = 111 and n2 (intervention group) = 111</td>
<td></td>
</tr>
</tbody>
</table>

Actual sample size used: Control 119 and Intervention 120

\[ \text{sampsi .07 .27, alpha(0.05) power(0.90) \times 1.29 \times 0.05} \]

**Sample size for secondary outcomes (microbiological outcomes):** A sample size of eight clusters with 10 households per cluster (80 households in total) for two sample comparisons of means (two-sided test) allowed for detecting the cluster mean differences in the level of microbes (TC) in commonly-used child food during feeding by 1.0 \(\log_{10}\) cfu/gram between control and intervention households (TC 2.30 \(\log_{10}\) mean cfu/gram in control households, and 1.40 \(\log_{10}\) mean cfu/gram in intervention households), with 90% power using 95% confidence interval (p<0.05). Food sampling during feeding was considered as the basis for analysis because it provided an overall random sampling opportunity.
Table 5: Summary parameters for sample size calculation – secondary outcomes

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Estimate</th>
</tr>
</thead>
<tbody>
<tr>
<td>Type I error ($\alpha$): significance level (p-value)</td>
<td>0.05 (95% confidence level)</td>
</tr>
<tr>
<td>Type II error ($\beta$): power of test</td>
<td>90% (Type II error of 10%)</td>
</tr>
<tr>
<td>Observed mean total coliforms in food from control group (m1):</td>
<td>$2.30 \log_{10}$ mean cfu/grm of food</td>
</tr>
<tr>
<td>$\log_{10}$ mean cfu/grm of food</td>
<td></td>
</tr>
<tr>
<td>Expected mean total coliforms in food from intervention group (m2):</td>
<td>$1.40 \log_{10}$ mean cfu/grm of food</td>
</tr>
<tr>
<td>$\log_{10}$ mean cfu/grm of food</td>
<td></td>
</tr>
<tr>
<td>Expected standard deviation (SD1): $\log_{10}$ cfu/grm</td>
<td>1.14</td>
</tr>
<tr>
<td>Expected standard deviation (SD2): $\log_{10}$ cfu/grm</td>
<td>0.90</td>
</tr>
<tr>
<td>Ratio of sample sizes (exposed : unexposed)</td>
<td>1</td>
</tr>
<tr>
<td>Types of test (two sample comparison of mean)</td>
<td>Two sided test</td>
</tr>
<tr>
<td>Estimated required sample size: n1=28, n2=28</td>
<td></td>
</tr>
<tr>
<td>Actual sample size used: n1 = 40, n2=40</td>
<td></td>
</tr>
</tbody>
</table>

Additional food samples were also tested at various stages to assess the effect of the intervention on the level of contamination in food at different behavioural control points. For other stages of food sampling, only seven households per cluster (28 households from the four intervention clusters and 28 households from the four control households) were included for collecting samples during baseline and follow-up as shown in Table 6.
Table 6: food and water sample collection stages and total collected samples

<table>
<thead>
<tr>
<th>Group</th>
<th>Baseline</th>
<th></th>
<th></th>
<th>Follow-up</th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Immediately</td>
<td>During</td>
<td>After</td>
<td>After</td>
<td>Immediately</td>
<td>During</td>
</tr>
<tr>
<td></td>
<td></td>
<td>after</td>
<td>feeding</td>
<td>5hrs</td>
<td>reheating</td>
<td>cooking</td>
<td>feeding</td>
</tr>
<tr>
<td></td>
<td></td>
<td>cooking</td>
<td></td>
<td>storage</td>
<td></td>
<td></td>
<td>storage</td>
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<tr>
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<td>28</td>
<td>28</td>
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<td>56</td>
<td>56</td>
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</table>

|          |          | 40 |          |          | 40 |          |          |
|          |          | 40 |          |          | 40 |          |          |
| Total     | 80 | 80 |          |          |          |          |          |

Altogether, 248 food and 80 water samples were tested during baseline and again during follow-up, and total coliforms and *E. coli* in food and water samples were quantified.

**Behavioural outcome assessment (primary outcome):**

The primary outcome measure was the proportion of mothers practicing all prioritised food hygiene behaviours when observed at mid-day. Structured observations[15, 26] were carried-out to observe all key behaviours, to ascertain the: i) proportion of mothers cleaning child food serving utensils using soap/ash; ii) proportion of mothers washing both their hands with soap before feeding child, and washing the child’s hands before eating; iii) proportion of households practicing proper storage of cooked food with a tight-fitting lid and no visible flies/dust/dirt in the food; iv) proportion of mothers thoroughly re-heating leftover/stored food and maintaining adequate temperature (≥70⁰C); v) proportion of households serving treated water to their children.

Behaviours were assessed 6 weeks (45 days) before and 6 weeks (45 days) after the intervention period. 25 female food hygiene observers were recruited, trained and mobilised to carry out the structured observation of food hygiene behaviours, with at least three observers responsible for each cluster. Observations were made in all intervention households.
(n=120 with no loss to follow up) and control (non-intervention) households (n=119 with two lost to follow up) at baseline and follow-up (post-intervention). The same observers were utilised during baseline and follow-up.

Data on the targeted key food hygiene behaviours were collected through direct observation. Observations were carried out between 1:00pm and 5:00pm (four hours of continuous observation), when the behaviours of interest (in particular the feeding of children with leftover or stored food) were likely to be seen. The observations took place in both groups simultaneously, and were completed within 12 days. Observers only observed one household per day, and recorded only the behaviour of mothers with a child aged 6-59 months in the respective households. Precautions were taken to avoid observer bias and observer influence on behaviours. Observers were asked to minimise chatting during observations or discussing their findings with other observers. If needed, observers were only allowed to discuss recent natural social, ritual or cultural activities to make the observation situation as normal as possible. Behaviour observers and food hygiene motivators were not the same individuals, and behaviour observers were kept uniformed about the purposes and activities of the programme. They may, however, have spotted some residual campaign materials in intervention villages.

A structured observation checklist was used to record all behaviours (attached in CD-R, annex-VII); however, in front of mothers observers used only a notebook, using codes to record their observations and write a short description of behaviours. An essential criterion for observers to start their observation was the physical presence of the participant mother at the household at the time of observation; another was the existence of leftover, stored or cooked food. In the absence of one or both of the above, observers skipped to the next household and returned on another day to complete the observation. Observers were randomly assigned to observe behaviours within the cluster. After completion of the observation in each day, food hygiene observers had to handover the observation checklist to the research assistant. Research assistant ensured that there was no missing field in observation checklist, data were properly recorded and forms were completely filled. Observer’s behaviour was also closely monitored and they were not allowed to discuss the observed behaviours amongst themselves. Anonymous supervision was done during data collection for at least 20% of the households to ascertain whether compliance was met.
In order to reduce the inter-observer variation error, intensive training was provided for all observers and they were also involved in pre-testing the observation checklist in field. The key operational definition for each behaviour was standardised and relevant images were shared with observers in order to familiarise the different possible behaviour beforehand. Data consistency was also checked by research assistant using different observation forms used by different observers on the same day. The repeated measurement by same observers or by two or more observers for same households was not done with a fear of introducing observers’ effect/bias.

Data entry was done by fully trained data assistant and work was routinely monitored, emphasizing the importance of accuracy when entering data. All field data were double entered into SPSS and later transferred into STATA for analysis. Emphasis was given to ensure that the data entered and analysed was consistent with the behaviours observed by the observers. In addition, I randomly selected almost 30% of the filled data form (observation checklist) and cross-verified the data entered into the system. The data error rate was almost zero.

**Observed behaviours:**

i) Stored all cooked/leftover food in container/s with a tight-fitting lid and no flies or visible dirt-dust accessing stored food: yes/no

ii) Stored/leftover food re-heated before serving to child and maintained at adequate temperature ($70^\circ$ c): yes/no

iii) Serving utensils (plates, bowl/glass, spoon) washed using soap/ash just before serving child food: yes/no

iv) Mother washed both her hands with soap just before feeding child, and both of the child’s hands were washed before eating food: yes/no

v) Served only treated water to their children: yes/no

**Microbial outcomes assessment (secondary outcomes):**

Commonly-used child food and water samples were collected from 80 randomly-selected households from the eight clusters with a child aged 6-59 months (10 households per clusters - 40 from the intervention clusters and 40 from the control clusters) at different stages of
preparation. Total coliforms (TC) and Escherichia Coli (E. coli), the WHO-recommended indicator organisms for measuring faecal contamination of samples[18, 19], were detected and quantified for each sample during baseline and follow-up. 3M<sup>TM</sup> Petrifilm<sup>TM</sup> (as recommended by the Centers for Disease Control and Prevention (CDC)[19]) E. coli and total coliforms count plates[20] media were used to quantify the microbes in food. Membrane filtration followed by incubation at 37<sup>0</sup>C (+/-1<sup>0</sup>C) on HiCrome Colifroms Agar w/SLS media (M1300-500G) was used for water, according to standard methods. The TC and E.coli counts were expressed as Colony Forming Units per gram (cfu/gm) for food, and cfu/100ml for water. Samples were collected by trained lab technicians unconnected with the intervention team, who arrived separately and unannounced. Another set of independent microbiologist and lab technicians processed, homogenised, inoculated and interpreted the results at a certified laboratory in Kathmandu. Both teams were blinded to the details of clusters and allocation to the intervention and control groups. Altogether, 248 food samples and 80 water samples were collected during baseline, and the same amount from the same households after the intervention. Each household from which food samples were collected received NRs 200 (~2US$) each time, as a reward for the donation of the food. All collected samples were processed, homogenised, inoculated, incubated and analysed using standard operating guidelines prepared for the study as detailed below.

**Sample collection stages and timing:**
Commonly-used child foods were collected at four different stages during baseline and follow-up periods, and water samples were collected once each during baseline and follow-up.

**Food samples:**
- **First stage:** immediately after cooking (5 to 15 minutes), directly from cooking vessels
- **Second stage:** during feeding, from mothers’/child’s hand (first feeding immediately after cooking, from mother’s hand if mother is feeding or from child’s hand if child is self-feeding).
- **Third stage:** leftover food stored from stage one after five hours storage (±15min) directly from stored containers
- **Fourth stage:** the same leftover food stored in stage three, immediately after reheating (0 to 5 minutes), directly from re-heating container/vessels
Water samples:
- Water: during feeding directly from serving utensils (during mid-day feeding)

**Sampling procedures:**
50 gram food samples were collected each time, using 60ml sterile and labelled plastic containers with tight-fitting lid, using a sterile wooden spatula when needed. Temperature was recorded just before collecting each sample using sterile thermometers. Similarly, 250ml water samples were collected using sterile and labelled containers. All samples were stored on ice in an insulated cooler box immediately after collection, and transported to the laboratory within four hours of collection, maintaining a temperature below 6°C. All samples stored overnight were maintained at the same temperature and processed early in the morning the following day.

**Laboratory procedures and interpretation - food:**
Of each 50 gram food sample, 10 grams were weighed and suspended in a labelled sterile vial containing 90ml of sterile Maximum Recovery Diluent - MRD (1:10 ratio). The sample was vortexed for a maximum of one minute to dislodge bacterial cells from the food particles. The sample was then allowed to stand for one minute to let heavy food particles settle. Multiple serial dilutions $10^{-1}$ to $10^{-4}$ were carried out. For plating, 1ml of the food suspension was transferred from the surface of the container to the middle of the labelled 3M Petrifilm, E. coli and Coliforms Count Plates using the sterile barrier filter tip of a micropipette. The cover was rolled down slowly on the inoculated media to avoid trapping any bubbles. The sample was spread by pressing gently with the spreader. The inoculated Petrifilm was allowed to stand at room temperature for at least one minute before incubation, to allow the media to gellify. The inoculated Petrifilm plates were incubated to detect the coliforms for 24 hours +/- 1 hour at 37°C +/- 1°C, and to detect the E. coli, plates were incubated for 48 hours +/- 1hour at 37°C +/- 1°C. The reading was done following AOAC, the international all foods method 991.14, as recommended by 3M PetriFilm. All colonies with gas (blue and red) at 24 hours were counted as confirmed coliforms, and only blue colonies with gas at 48 hours were counted as E. coli. Such individual colonies were counted and used to calculate the total coliforms and E. coli load as colony forming units (cfu)/gram of food respectively.

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1 www.3M.com/microbiology
Laboratory procedures and interpretation - water:
Membrane filters and HiCrome Coliform Agar w/SLS media were used to detect TC and *E. coli* from water samples simultaneously using the membrane filtration method, providing a definitive count of colony forming units (cfu) per 100ml of sampled water. The sample was filtered through a sterilised membrane filter with a 0.45μ pore size. To test the sample, 10ml of sterile water was poured into the filtration unit; 1ml of the sample was poured in, followed by 20-25ml of sterilised water for equal distribution in the same filtration. 50ml and 100ml samples were poured directly into the filtration unit without sterile water. The membrane filter was removed aseptically after filtration using sterile forceps, and placed on the media plate. Samples were then incubated at 37°C (+/-1°C) on HiCrome Coliform agar, selective for TC and *E. coli* for 24 hours before cultural characteristics were observed. Results were interpreted by counting the number of colonies on the surface of the membrane filter and calculating CFU/100ml. All colonies presenting as ‘dark blue to violet’ were recorded as *E. coli*, and ‘salmon to red’ as total coliform. Individual colonies were counted and used to calculate the *E. coli* and TC load as colony forming units (cfu)/100ml of water.

Reach assessment (compliance measurement):
To ascertain whether or not the intervention reached the intended target groups and to assess the exposure and reach of the intervention, assessment was undertaken in all clusters after measuring all outcomes of interest during follow-up in both groups (120 intervention households and 117 control households). Compliance measurement was conducted using completely different sets of field data collectors during October – November 2013. Compliance measurement team was not connected with trial implementation, evaluation and project management team except me. Using structured close-ended questionnaires, mothers were asked whether they have heard of or participated in campaign activities to ascertain the intervention reach in the intervention group, and to detect any cross-contamination between the intervention and control groups during the trial period. Additionally, mothers were asked questions to explore whether change in behaviours over time has a role in changing social norms. Compliance measurement checklists are attached in CD-R annex-VIII.
**Statistical analysis:**

**Statistical analysis for behavioural outcomes:**

The Primary outcomes were the comparison of cluster-level mean of observed proportion of all five key food hygiene behaviours as a composite performance score between groups - before and after the intervention. Cluster level analysis was used since the allocation of the intervention was done at cluster level. Analysis at cluster level is suitable as opposed to household level analysis to account for the small number of clusters, and this approach is commonly used to account for lower level clustering with expectation of increasing between-cluster variability[31]. The prevalence of all key food hygiene behaviours was calculated at cluster level during baseline and follow-up, and recoded to measure the primary outcome. As a secondary analysis, a comparison of all individual behaviours was conducted at cluster level by different groups during baseline and follow-up. The small number of clusters requires the use of the non-parametric test two-sample Wilcoxon rank-sum test (Mann-Whitney U)[32] to compare group means and for the estimation of the statistical support. This test does not rely on the assumption of normality and is resistant to outliers[31].

The effect size of the intervention was measured with the difference-of-differences i.e [Follow-up – baseline]_{intervention} minus [follow-up – baseline]_{control}. Further, sub-group analysis was conducted for behavioural outcomes stratified by religion, caste/ethnicity, educational level, economic status and types of cooking fuel. Statistical support for effect modification was assessed by computing the difference in food hygiene behaviours (composite performance) between sub-groups within each cluster, comparing mean difference of differences between intervention and control clusters, following methods described by Cheung and colleagues[33]. The effect of the intervention did not significantly differ due to presence or absence of the other included risk variables hence there was no need of further multivariate analysis. The intra-class correlation coefficient was calculated using STATA loneway command. Data were entered into a spreadsheet and SPSS, and statistical analysis was performed using SPSS 19 and STATA 12.

**Statistical analysis for microbial outcomes:**

The primary analysis was focused on the differences in mean count (mean of the log transformed bacterial count) of TC and *E. coli* cfu/gram of food and cfu/100ml of water by intervention and control arms at cluster level during baseline and follow-up at various stages. Cluster level analysis was used since the allocation of intervention was done at cluster
level[31]. Given the number of clusters a non-parametric test was used, the two-sample Wilcoxon’s rank sum test (Mann-Whitney), which does not rely on the assumption of normality and that is resistant to outliers[31], for estimating statistical significance[34]. The TC and E. coli colony counts were log-transfered (log_{10}), and analysis was done to compare the differences in means by group. The effect of the intervention was therefore the summary means of the log-transformed microbe counts (TC and E. coli cfu/gram in food, cfu/100ml in water) compared between the control and intervention groups during baseline and follow-up. Food samples with total coliforms and E. coli counts 0 cfu/grm (non-detectable level), <10 cfu/grm, in-between 10-100 cfu/grm and >100 cfu/grm of food categorized as no risk, low risk, medium risk and high risk respectively. The effect size of the intervention was measured using difference-of-differences analysis i.e. [follow-up – baseline]_{intervention} minus [follow-up – baseline]_{control}. The original laboratory data were entered into Excel and then transferred into STATA (version 12) for analysis.

3.5.3 Overall trial management structure:

Figure 13 summarises the overall management structure of the food hygiene intervention trial in Nepal led by myself with the support from various team members.
3.5.4 Research and Programme Budget:
The total financial resources used to meet the overall objectives of this study within two years were GBP 66,589. Of the total budgetary requirements, GBP 10,000 allocated from my own PhD SHARE scholarship funding, GBP 38,781 from SHARE consortium call-C application grant and GBP 17,808 received from WaterAid UK. My time involvements are not included in any of these costs. Table 7 summarises the total cost breakdown by different budget line times. The cost effectiveness analysis was not done as part of the study outcomes, however I have tried to split budget into two parts i.e. i) research only cost and ii) food hygiene intervention trial programme cost which might be a interest for researchers and programme implementators accordingly. The total programme expenditure (excluding research) per person in the intervention group was only £18.30 and per household expenditure was £103.54.

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<th>Budget line items</th>
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<td>Formative research - all inclusive</td>
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3.6 References:


20. 3M-PetriFilm, *Petrifilm™ E. coli and Coliform Count Plates International Guide*. 3m PetriFilm. www.3M.com/microbiology or https://www.msu.edu/course/fsc/441/3mc&ec.html


**RESEARCH PAPER COVER SHEET**

**SECTION A – Student Details**

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<tr>
<td>Principal Supervisor</td>
<td>Dr Val Curtis</td>
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<td>Thesis Title</td>
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**SECTION B – Paper already published**

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**SECTION C – Prepared for publication, but not yet published**

| Where is the work intended to be published? | American Journal of Tropical Medicine and Hygiene |
| Paper’s authors in the intended authorship order: | Om P. Gautam, Valerie Curtis |
| Stages of publication | Not yet submitted |

**SECTION D – Multi-authored work**

| Give full details of your role in the research included in the paper and in the preparation of the paper | I led the design of the study, its conduct in the field, the analysis and the write-up. |

Student Signature: ____________________________ Date: 25 September 2015

Supervisor Signature: ____________________________ Date: 25 September 2015
CHAPTER – IV

(Paper 1)

Study Title:
Food hygiene practices of rural women and microbial risk for children: formative research in Nepal

Authors:
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Key words:
Food hygiene, behaviours, microbes, diarrhoea, mother, children, formative research
ABSTRACT:

Aim: Formative research was conducted in a rural hill setting in Nepal during April-June 2012 to inform the design of an intervention to promote safe food hygiene practices.

Methods: A variety of methods underpinned by Evo-Eco theory and Hazard Analysis Critical Control Points (HACCP) were used to pinpoint key risk behaviours and their environmental and psychological determinants in 68 households with a mother having a child aged 6-59 months. These included video recordings, observation of daily routine, teach the researcher sessions, in-depth interviews, observations, focus group discussions, motives mapping, microbiological assessment and identification of critical control points.

Results: Physical settings, especially the kitchen, form a challenging environment for mothers in rural hill settings of Nepal to practice adequate food hygiene behaviours. Prevalent food hygiene practices of mothers were inadequate, leading to frequent exposure of young children to highly contaminated food, water and milk. We identified 6 critical control points and of these 5 needed improving.

Conclusion: Five key adversely-practiced food hygiene behaviours are suggested for prioritisation. While designing a food hygiene intervention package, consideration should be given to the physical, biological and social environment and immediate motives behind each practice should be taken into consideration while framing key messages. The creative and engaging tools/activities should be designed around common motivational themes as drivers of behaviour change such as nurture, disgust, affiliation and social status/respect.
INTRODUCTION:

Poor food hygiene is likely to be an important contributor to high rates of infectious diseases in resource-poor settings. According to some estimates, up to 70% of diarrhoeal episodes in developing countries are caused by pathogens transmitted through food [1, 2]. Children are particularly vulnerable to diarrhoeal infections during the weaning period [3-5] when protection from breast milk reduces and complementary food is introduced. High rates of diarrhoeal infections contribute to problems of undernutrition [6-8], where a quarter of stunting can be attributed to a child having 5 or more episodes of diarrhoea before the age of two [5]. Environmental enteropathy arising from subclinical exposures to faecal pathogens [7] may be a further cause of poor growth in children residing in unhygienic environments. However, there are few studies of microbial food contamination and fewer studies on interventions to improve this situation in the countries with high diseases burdens.

Whilst poor food hygiene has often been implicated as a source of diarrhoea pathogens in industrialized countries [9-12], very little is known about food hygiene in resource-poor settings. Here, mothers face an uphill task to prepare and serve uncontaminated food in conditions where water may not be available on tap, with few sanitation facilities, where surfaces are hard to clean, where cleaning products are scarce and in warm climates where refrigerators are uncommon. Such studies as there are point to high levels of microbial contamination in foods fed to young children [3, 13-18] and food preparation as risk factor for diarrhoeal diseases [10, 17, 19].

Improving the food hygiene is therefore one of a number of priorities for reducing the burden of infections in developing countries. However, before we can proceed to large scale intervention, more needs to be understood about food hygiene behaviours. We need to better understand which behaviours predispose to food contamination, what new behaviours can feasibly replace them, and how families can best be encouraged to practice safe food preparation.
This formative research study used a variety of theory-based methods to investigate food hygiene behaviour in a rural hill area of Nepal. The objective was to identify key food hygiene behaviours, their determinants and inform the design of an intervention.

**METHODS:**

**Study site and sampling:**
The formative research was conducted in Kavre District, Nepal. Here, diarrhoea is the second leading cause of child death and around 41% children are stunted [20]. The district is steeply hilly and the population subsist through small scale agriculture and dairy farming. The population’s ethnic origins are mixed with a majority (63%) belong to Hill Aadiwasi/Janajaati (Tamang) followed by Brahmin/Chhetri (28%). Around 70% of households had piped water and 44% practiced open defecation. Two wards which were not scheduled to be part of the intervention trial were selected and all 68 households with children aged 6-59 months in the two wards agreed and consented to participate in the study. Ethical approval was granted by the Research Ethics Committee of the London School of Hygiene and Tropical Medicine (LSHTM), and the Nepal Health Research Council (NHRC).

**Conceptual frameworks:**
The study used two conceptual frameworks to conduct formative research. To identify behaviours likely to cause risk of pathogen transfer we used a modified version of Hazard Analysis and Critical Control Points (HACCP) [21-23]. HACCP is a systematic approach to the identification, assessment and control of hazards [21]. We used only the steps relevant to our objectives such as identification of hazards associated with food preparation, handling and feeding, assessing risk, and determining critical control points where control measures would be applied.

To understand food hygiene-related behaviour in context we used Behaviour Centred Design underpinned by the Evo-Eco theoretical framework, a systematic approach to designing behaviour change interventions developed at LSHTM [24-27]. The approach has 5 steps; A-
Assess, B-Build, C-Create, D-Design and E-Evaluate. Formative research was the B-Build step in the process - the approach pinpoints key behaviours, seeks to identify causes for the behaviours that are psychological (habitual, motivated or planned), bodily and environmental (social, biological and physical) and pays attention to behavioural settings, which are akin to theatres of performance with roles, scripts, routines, props, purposes and particular histories[28].

**Study methods and instruments/tools:**

The study used microbiological methods to investigate food contamination and anthropological and consumer research techniques to understand risk behaviours in social and physical context. The following are summaries of methods and instruments/tools used in the study.

**Microbiological methods and instruments/tools:**

**Microbial assessment:** Total coliforms (TC) and *E. coli*, the WHO-recommended indicator organisms for measuring faecal contamination [29], were quantified in commonly-used child food, water, milk and Jad (alcoholic brew) from 30 randomly-selected households. One hundred and five food samples were collected at four different stages (30 immediately after cooking, 30 during feeding, 30 after five hours of storage and 15 immediately after-reheating). Thirty samples of ready-to-serve water, 13 of milk and 12 of Jad were collected, transported, homogenized, inoculat ed and incubated, and results were interpreted using standard operating procedures and appropriate media.

**Hazard analysis and critical control points:** A food flow diagram (food supply chain from collection of raw materials to feeding) was developed based on observed patterns of food preparation, storage and feeding. Potential sources of hazards and possible re-contamination were documented. Critical control points were identified based on their role in bacteria destruction, survival, and or propagation. Behaviours were prioritised for intervention based on whether control points acted as barriers and whether respective behavioural actions could be applied as control measures.
**Behavioural methods and instruments/tools:**

**Video recordings:** Food preparation behaviour was filmed in 30 randomly-selected households. Local women were trained to use hand-held video cameras to follow all aspects of child food preparation, cooking, handling, storing, feeding and re-heating. Filming was continuous for two to three hours, except when privacy was required.

**Observation of daily routines:** The daily work routine of 30 randomly selected mothers starting from early morning (wake-up) to the end of the day (going to bed) was recorded using ‘Day in Life Analysis (DILO)’ tool. This tool helps to assess whether food hygiene behaviours are part of their daily routine work. Specific actions during cooking time were assessed using ‘a Moment in the Life Analysis (MILO)’ tool.

**‘Teach the researcher’ sessions:** Five mothers were asked to teach the researcher how to cook their child’s food, while the researcher observed closely the mother’s behaviour actions during food preparation, cooking, feeding the child, storage, and reheating.

**In-depth interviews and household surveys:** In-depth interviews (IDI) were carried out with all sampled mothers (68) in each household concerning their socio-demographic characteristics, access to basic services, common child food, the current source of knowledge, common infections etc.

**Observation:** The physical environment and levels of cleanliness were assessed in 68 households by observation using standardised definitions and checklists.

**Focus group discussions (FGDs):** nine FGDs with mothers and two with grandmothers were conducted to identify commonly-used child foods, to understand the food chain supply mechanism, and to identify socio-cultural practices around food.

**Motives mapping:** Eleven motivational exercises were performed to assess immediate motives. Seven different pictures demonstrating common motives (attraction, nurture, disgust, status, respect, affiliation, purity and disease) were shown and stories were articulated around
those pictures. After discussion, mothers were then asked to rank the pictures according to how likely these were to motivate them to practice key behaviours.

Data analysis:

Microbiological analysis:
Fifty grams of food in each stage, 250 ml water, 50 ml milk and 50 ml of Jad were collected and transported to the laboratory maintaining a temperature below $6^\circ$C. Total coliforms (TC) and E. coli colony counts per gram of food (cfu/gram of food), cfu/100ml of water, cfu/ml of milk and Jad were recorded in the laboratory. 3M-PetriFilm, E. coli and Coliforms Count Plates [30, 31] was used to detect total coliforms and E. coli in food and milk samples. Membrane Filters (0.45μ pore size) and Eosine Methylene Blue (EMB) agar media was used to detect the same total coliforms and E. coli in water and Jad samples. The counted colonies were log-transformed ($\log_{10}$) to compare the mean counts at four different stages for food. The count of TC and E. coli are categorized as $<10$ cfu/gm, 10-100 cfu/gm and $>100$ cfu/gm of food to observe risk. The temperature of food samples and pH of milk, water and Jad samples were assessed. Specific methods applied for samples processing and interpretation are described and available online.

Behavioural analysis:
Interviews and FGDs were recorded and transcribed, videos were parsed and used to identify missed opportunities, food follow diagram used to visualize likely hazards and identified critical and behavioural control points. Quantitative data (observations, survey, motive mapping) was entered and analyzed using IBM SPSS statistics 19. The findings were organized following the categories of behavioural determinants in the Evo-Eco checklist[24]. Behaviour (planned, motivated and habitual), Environment (social, physical and biological), settings and other relevant findings are presented under specific themes and as relevant summary statistics are presented in frequencies.
RESULTS:

Household and participant characteristics:
The social and demographic characteristics of the study population are presented in Table 1. Respondents ranged in age from 17-43 years (mean 28 years). Majority of the study household belonged to Tamang (75%) followed by Brahmin/Chhetri (19%) and dalit (6%) cast group. The majority of the mothers had no or informal education (mothers who attended non-formal education lead by Government as part of literary improvement plan such as adult education), and the majority of households earned less than $100 US per month. Most of the respondents were either housewives (50%), or dependent on agricultural work (27%). Sixty eight households had 84 children aged 6-59 months. Ninety four percent of household had soap available (mostly laundry soap). Figure 1 highlights the environmental settings.

Behaviour related to food hygiene, its determinants and level of microbes in food

Daily routine of mothers in behavioural settings:
Most mothers followed a similar daily routine of rising at 5am, followed by defecation, fetching water, lighting the fire, cleaning the animal shed, feeding animals, sweeping inside and outside the house, preparing tea, and feeding snacks to children. This was followed by either domestic or field work (~3 hours). Though cooking time varied by caste groups, the majority of the mothers prepared lunch (the first main meal) between 9am and 11am. Most mothers (82%) needed at least one hour to prepare food. After feeding the child and eating lunch, afternoon work included domestic work such as sweeping, laundry, feeding animals, caring for animals or work in the field (~3-4hrs). Food given to children in the afternoon mostly comprised leftovers of food cooked in the morning, often stored in the same cooking vessel or in a bowl/plate near/around the cooking area at ambient temperature. Half of mothers mentioned having some leisure time in the afternoons. Most mothers cooked dinner between 6 and 7pm and then fed their children the freshly prepared food. After a short leisure or tidying up period the day ended at around 8 to 9pm for all mothers. The key food hygiene behaviours related to child food are overshadowed by mother’s daily routine so the daily routine was an important determinant of how mothers perform behaviours. Most mothers did not link their daily routine with the food hygiene behaviours suggest that those are yet to be
formed. Below we describe what was fed to children and then pinpoint the food hygiene issues identified at the stages of preparation, storage feeding, reheating etc.

**Child Food**

As reported, female children are weaned at 5 months and male children at 6 months in rural Nepal. Weaning starts with a ‘rice feeding ceremony (pasne)’, mostly using rice pudding. After the first day of weaning, the majority of the mothers/carers reported feeding their children the same food that adults and older children consume daily. The study found that majority of the households served solid food to children, including those being weaned (6-23 months). Table 2 shows the types of food fed to young children in Nepal. During or immediately after feeding, all mothers said that they offered water to their children to drink and sometimes milk (43%); several Tamang families also offered Jad. Liquid foods included milk and water, semi solid foods included Jaulo—a rice based porridge and the main solid food was rice with pulses and/or vegetable curry. Other foods included *dhindo* (maize- or wheat-flour porridge), *roti* (maize- or wheat-flour flatbread) with *dal* (pulses), *lito* (roasted rice-maize or millet-flour with ghee and sugar), *khichari* (rice, pulses, turmeric powder and vegetables), fruits (banana, orange, etc.), boiled eggs and breast milk.

**Food preparation and cooking:**

Mothers or grandmothers generally cooked for and fed children. Though foods were usually prepared twice a day children were fed on average 4-5 times a day; all households therefore fed stored or leftover food to children. Mothers were multitasking while cooking, making cross-contamination of food likely; such tasks included feeding animals, sweeping or wiping the kitchen, washing utensils, using the toilet, doing laundry, cleaning the child’s bottom, breastfeeding, and wood or water collection. Other behaviours likely to cause contamination were tasting curry using fingers instead of a spoon, grinding spices without cleaning the grinding stone or washing hands, re-using unwashed cooking vessels, etc. Around 90% and 94% of opportunities for handwashing with soap were missed before and during cooking respectively (Figure 2). During cooking around 50% of mothers also missed the opportunity to cover food to protect it from kitchen dust. However, food sample collected immediately after cooking found to be less contaminated as compared to other times (Table 3, Figure 3). The total coliforms and *E. coli* mean counts in 30 food samples collected immediately after cooking were 0.14 log<sub>10</sub> cfu/gm and 0.01 log<sub>10</sub> cfu/gm respectively. High levels of TC (>100
cfu/gram) were counted in only one sample at this stage. All samples were collected within 5-15 minutes of cooking, and 77% samples had a recorded temperature of >60°C with a mean temperature of 66°C [SD 9].

**Cleanliness of cooking and serving utensils:**
Shortly after cooking, foods are served in plates or bowls. Observations showed that only 16% of mothers cleaned serving utensils thoroughly using ash or soap just before serving food to children, although half (47%) considered such practice as good and it differed among the different ethnic groups. Video analysis shows around 84% of the opportunities to clean serving utensils just before offering food were missed by mothers. The Majority used ash to wash their cooking vessels as it is readily available in all households. The use of soap was considered a luxury. Since the majority of households did not have a kitchen rack on which to place washed utensils, these were placed on the mud floor. Flies, animal faeces and dust or dirt are therefore likely to contaminate and re-contaminate washed utensils.

**Feeding and handwashing with soap:**
Almost all mothers fed the child using their hands (81%). If child fed themselves, they did the same. Many mothers allowed the child to carry food around while eating. Although 18% of mothers believed that hands should be clean to maintain good food hygiene practices, only 7% reported the use of soap to wash their hands before cooking and before feeding their child. Soap and water was available in 94% and 100% households respectively. Up to 88% and 91% of mother and child handwashing opportunities before feeding were missed by mothers according to video analysis respectively. Use of water to wash hands and varying practices was commonly reported; a participating mother mentioned: “I always wash my child’s hands with water before feeding but if their grandmother is feeding, she never does this”. The second round of samples was taken during child feeding and samples were collected from mothers’ and children’s hands. The mean food temperature during feeding was 37°C [SD 6]. The TC and *E. coli* mean counts increased during feeding and reached 1.48 log_{10} cfu/gm and 0.67 log_{10} cfu/gm respectively. Of the 30 samples tested at this stage, high levels of TC (>100 cfu/gram) were counted in 14 samples, (47%) and high levels of *E. coli* were counted in 5 (15%) samples.
Storage of cooked leftover food:
Generally food was cooked in the morning and stored inadequately (without tight-fitting lid) at ambient temperature. All households reported storing leftover food for subsequent feeding, and such practice was observed in 73% of households. Although 63% of households covered food for storage, the majority put a dirty ladle over the stored food which attracted flies, dust, dirt and animals. Only 43% covered food appropriately. Appropriate food storage and protection of food from flies and dust were considered as good food hygiene practices by only 13% and 4% of mothers respectively. Cooked food stored up to 12 hours was considered to be fresh. Such stored food was served to children multiple times during the day. Due to lack of refrigerators the leftover foods were either stored in the cooking vessel or in a small bowl or plate on the floor near the kitchen for multiple feeding. The third samples were taken after 5 hours of storage. The mean temperature of stored food was 32\(^0\)C [SD 3]. The TC and E. coli mean counts increased substantially at this stage and reached 2.03 log\(_{10}\) cfu/gm and 1.29 log\(_{10}\) cfu/gm respectively. Of the 30 samples tested at this stage, high levels of TC (>100 cfu/gram) were counted in 22 (73%) samples and high levels of E. coli were counted in 11 (37%) samples.

Re-heating stored/ leftover food before feeding:
Observation revealed that only half of households reheated stored food, and that only 19% of food reheated reached an adequate temperature (>60\(^0\)C). Of those who reheated food, some used a frying pan (Cauldron - Karai) and some used a serving steel bowl. Mothers reported that the use of curry, vegetable soup, milk, water or ghee for reheating food will protect the food or pot from burning. None listed thorough reheating as a good food hygiene practice, and a statement made by one mother represents a common practice: “I do not reheat food because it will take time and energy, and it will burn the food”. The fourth batch of samples was taken immediately after re-heating from those households in which reheating was practiced. Samples were collected within 0-5 minutes of reheating, and only 19% of samples had a temperature of >60\(^0\)C (with mean 51\(^0\)C[SD 13]). The mean counts of TC and E. coli in 15 food samples collected immediately after reheating were 1.39 log\(_{10}\) cfu/gm and 0.96 log\(_{10}\) cfu/gm respectively. Of the 15 samples tested at this stage, high levels of TC and E. coli (>100 cfu/gram) were counted only in 7 (23%) and 3 (10%) of the samples.
**Water and Milk feeding:**

Together with food or immediately after food, young children were always offered either water or home-produced cow/buffalo milk. Water from communal or household taps (delivering was from unprotected sources) were commonly collected in gagri (thin copper or brass vessel), that are left uncovered (47%), or are covered with a bowl (53%). None of the households boiled water before serving it to the children and none of them re-boiled milk when serving it at different times during the day. As revealed from video footage, all water treatment and 95% of milk boiling opportunities were missed by mothers. A few households even gave their children never-boiled milk to drink. Water, milk and Jad samples were collected during mid-day. The mean TC and *E. coli* counts in 30 water samples collected during feeding were $1.62 \log_{10} \text{ cfu/100ml}$ and $0.92 \log_{10} \text{ cfu/100ml}$ respectively, with 7.27 mean pH. Milk samples were heavily contaminated with TC and *E. coli*, with mean counts of $5.15 \log_{10} \text{ cfu/ml}$ and $1.26 \log_{10} \text{ cfu/ml}$ respectively, and with 5.89 mean pH. Twelve Jad samples were tested and no total coliforms or *E. coli* were isolated. The Jad was acidic and its mean pH was 2.9 (minimum 2 and maximum 3.7) scale.

**Environment based determinants of the behaviours:**

**Social environment:**

Study found that the social environment determined specific behaviours such as different food hygiene practices associated with different ethnic groups. Brahmin/Chhetri used cow dung to smear on kitchen floors after daily eating and only wipes kitchen wall twice a year. It is Tamang custom to only wash cooking and serving utensils once a day in the late morning. A wooden spoon was used to mix flour while cooking *dhindo* but was never washed; instead, remnants were peeled or scraped off before use. Brahmin and Tamang also reported only washing the milk storage container twice in a month. The majority of Tamang families also fed Jad (alcoholic brew) to children under five as a substitute for water and milk. A mother from this ethnic group said: “I feed Jad to the child so that they don’t cry and disturb my work; at the same time, Jad kills the worms in the child’s stomach”. Apart from family and local community the primary sources of new information were reported as teachers and female community health volunteers (FCHVs). Most mothers attended monthly credit group meetings. Most mothers listened to local radio during leisure time but very few had television at home.
**Physical environment:**
Most households used pipe water connections (unprotected spring sources), around 57% practiced open defecation and none had refrigerators. Study found that the physical settings more importantly the kitchen offered a challenging environment for mothers to practice food hygiene behaviours. All houses were made of mud and stone, with kitchen surfaces made of mud, and most households cooked on the floor using firewood. Most of the households did not have a separate kitchen, hence one room in the ground floor served for cooking, eating, running of the household, sleeping and sometimes for keeping animals such as chickens, goats and buffaloes. This room was often dark and smoky. Only 7% of households had a cupboard in which to store washed kitchen utensils. Figure 1 shows the overall cleanliness status of entire study settings.

**Biological environment:**
Animal faeces were observed on the ground in 79% of compounds and the majority (65%) of households disposed of child faeces in adjacent fields or open spaces. Study found that biological environment had role to determined key behaviours. Such as flies were present throughout the village, particularly in kitchens making contamination more likely. Only 34% of compounds, 45% of toilets, 57% of the kitchens, and 67% of the stored utensils was classed as visibly ‘clean’ (Figure 1). Almost all households collected waste kitchen water to feed animals which attracted flies that also landed on kitchen surfaces, utensils and food. Hands could not be washed in this container because mothers believed that animals should not be given soapy water. Only 28% of children’s and 40% of mother’s hands were classed as visibly ‘clean’.

**Does perceived risk have any effect on behaviour?**
Of 84 children from 68 households, 39 (46%) were reported to have been sick in the past month. The reported prevalence of diarrhoea in the past week was 24%. The perceived causes and risk factors for diarrhoea varied among mothers. One third of mothers mentioned flies and dust, a quarter mentioned contaminated food and other causes were not washing hands with soap before feeding child, ‘evil eye’. During FGDs, mothers reported that they first seek the services of traditional healers when their child had diarrhoea (local names - pakhala, jhadabanta, ruipa - cherpa) and if the situation becomes worse they do seek support from FCHVs and only take the child to a nearby health institution.
Barriers and motives to practice behaviours:

The barriers, the immediate motives and the motivational drivers for key behaviours such as cooking food thoroughly, cleanliness of serving utensils using ash or soap, handwashing in the kitchen before feeding/eating, covering food, reheating stored food before feeding, and treatment of water or milk before feeding are reported in Table 4. Mothers were pre-determined to perform routine behaviours in their environmental settings. The understanding of behavioural determinants and identification of barriers was a basis to explore what would motivate mothers to address some of those barriers through the motivational exercise using imaginary story. Mothers were reported almost similar motives to practice specific behaviour but different sets of motives for different behaviour. The key emotional drivers were linked with each specific motives and we found that the ‘nurture, disgust, affiliation and status’ were the common motivational drivers. The key motivational drivers for each behaviour are presented in Table 4.

Hazard analysis and determination of control points:

A food flow diagram (Figure 4) from collection of raw materials to cooking to storage, to feeding was drawn. Potential sources of hazards and possible re-contamination were documented for each key step (Figure 4, Table 5). Critical control points were identified based on their role in bacteria destruction, survival, and or propagation using microbial evidences (Table 3). Behavioural control points were identified whether respective behavioural actions could be applied as control measures (Figure 4, Table 6).

As presented in Figure 4, the study identified ‘cooking’ and ‘re-heating’ of food, and boiling of water and milk as critical control points through which bacteria destruction can be expected if behavioural measures such as thorough cooking, thorough re-heating and boiling water/milk were applied correctly respectively. Similarly, storage, use of serving utensils and feeding/eating using hands or spoon are identified as behavioural control points in which proper storage of cooked food with tight lid, cleanliness of serving utensils (plates, bowls and spoons) using ash/soap, and handwashing with soap before feeding and eating identified as a corrective behavioural control measures.
DISCUSSION:
This study showed that the prevalent food hygiene behaviours of mothers were inadequate as assessed using multiple methods. Optimal behaviours that would mitigate faecal oral transmission of microbes in commonly used child food such as cleanliness of serving utensils, handwashing with soap before feeding/eating, proper storage, through re-heating and boiling milk/water before offering were uncommon among mothers (Figure 2 and Table 4). Young children in rural Nepal were exposed to microbes in food, water and milk. Although various factors and multi-tasking behaviours of mothers were thought to be responsible for re-contamination of food, water and milk, the HACCP approach was useful to identify six critical and behavioural control points. The identification of critical control points (CCPs) is particularly important and can facilitate appropriate targeting of resources and prevention efforts[32]. The immediate motives and emotional drivers for practicing behaviours were identified for each behaviour.

Our findings showed that the physical settings, especially the kitchen, present a challenging environment for mothers to properly practice adequate food hygiene behaviours. Pertinent aspects of the physical environment included the lack of proper kitchen settings, lack of infrastructure, and difficulty in meticulously cleaning the kitchen surfaces compounded by an unclean household environment. This is compounded by the biological environment (heavy presence of flies, animal accessing in kitchen and presence of animal faeces in compound etc) and the social environment (different ethnic groups followed different practices) and traditions (feeding family food to children etc). A review article demonstrates that weaning foods prepared under unhygienic conditions are frequently heavily-contaminated with pathogens and thus are a major factor in the cause of diarrhoeal diseases and associated malnutrition [17]. In Viet Nam, the risk of diarrhoea was different by cooking places [33]. Our study suggests that food hygiene behaviours are underpinned by physical, social and biological settings. Initiatives to improve food hygiene behaviours in such environments should consider changes in the physical, social and biological setting, most importantly the kitchen setting though it would be challenging.

Thorough cooking was identified as a critical control point when appropriate temperature is maintained. Our findings show that food may have become contaminated/re-contaminated on
multiple occasions during cooking due to multitasking by mothers during cooking. Despite all these risks, thorough cooking practices eliminated microbial contamination (TC and E. coli) risks even though mothers were unaware of the benefits of thorough cooking. The thorough cooking is one of the most important critical control points if a high level of temperature (>60°C) is maintained which kill vegetative forms of pathogenic bacteria[34]. Previous studies using HACCP also found that traditional cooking was very effective in eliminating faecal contamination [35]. Reinforcement and encouragement using immediate motives would be useful for sustaining thorough cooking practices.

Handling food increases the risk of contamination and our hazard mapping showed that the foods were subject to contamination during serving and feeding/eating yet many mothers missed opportunities to properly wash their/child’s hands and serving utensils before serving food. Food samples taken immediately after cooking from plates or bowls after hand contact showed increased levels of contamination. This shows that promoting thorough cooking alone is insufficient to prevent microbial contamination at the point of feeding. It was likely because both the behaviours were rare in the study settings. A main reported and observed barrier for not washing hands in the kitchen was that all households collect kitchen wastewater in a container that is then fed to animals. Families feared that if they used soap to wash their hands, the animal will not drink the soapy water. Hence mitigating this barrier may increase the frequency of handwashing during cooking and before feeding the child. In addition mothers gave other various reasons which are presented as ‘verbatim’ as table 7. While handwashing with soap can prevent infection and save many lives, it is still rare in many countries [26] and handwashing before feeding children and before handling food was practiced less than handwashing at other times [36]. We suggest that promotion of food hygiene should include cleanliness of serving utensils and handwashing with soap before feeding and eating. Previous studies suggest that mothers quickly adopt handwashing with soap as compared to spoon feeding practices [37]. The immediate motives for practicing handwashing and cleanliness of utensils might be useful to reinforce such practices.

Leftover cooked food was kept ambient temperatures after the first feeding, making the multiplication of organisms likely as none of the households had a refrigerator. Food samples tested after five hours of storage had a higher level of contamination compared to any other time. Lukewarm temperature, the environment (flies/dust/dirt/storage location), inadequate
utensil storage, and storage without a tight-fitting lid all make these high levels of contamination possible. Previous studies also reported increased bacteria counts from $10^4$ to above $10^8$ after 24h at $37^0C$ [38-42], three-fold increased within 1 hour of storage[40] and significant multiplication of faecal coliforms when there was a delay of more than 4 hours between preparation and consumption of food [43]. Avoiding storage of cooked food was difficult in the study setting; hence storage of leftover food with a tight-fitting lid to avoid from additional contamination from flies, dust and dirt, and animals was the only possible measure that can be applied at this stage in the study settings. The identified immediate motives would be an useful key messages to encourage this habit.

Offering stored food without reheating or briefly reheating was common in the study setting and various barriers to reheat food were reported. Bacteria were not killed in sufficient quantities in briefly reheated food suggesting that reheating practices were inadequate and temperature had major role to kills bacteria. The practice of touching food with hands, putting additional water to make food wet and not maintaining proper temperature also add more potential contamination. The Cauldron was identified as an appropriate object for thoroughly reheat food, yet many households did not use it for this purpose. Thorough re-heating of stored food just before offering to the child was identified as a control point and immediate motives to practice thorough re-heating would be used as key messages to encourage such habit.

None of households treated water before serving to children and majority of water samples were contaminated with total coliforms and failed to meet WHO and Nepal national water quality standards[44]. Contamination was likely caused by contamination at source (unprotected spring), storage practices (uncovered containers), and contaminated serving utensils (glasses or bowls). The kettle was identified as an appropriate object with which to boil water, but was mostly used for tea preparation. Similarly, milk was also heavily contaminated, due to unhygienic milking (location, washing gutter using contaminated water etc), inadequate boiling, inadequate storage (storing in ambient temperatures without covering), inadequate handling, and not re-boiling before serving. The most feasible options identified to make water and milk safe were boiling in that setting at household level. Boiling water was identified as one of the preferred household water treatment options and its effectiveness was previously tested[45]. The motives identified to boil water and milk just
before feeding child could encourage such practice. Jad had non-detectable level TC and *E. coli* and all Jad samples were more acidic which might have affected the level of microbiological contamination.

While identifying the control points, it was recognized that each of the points can offers critical control measures (killing bacteria) and behavioural control measures (reducing contamination). The food flow diagram and mapping show that cooking/boiling, storing and re-heating food offers best possible critical control points. In addition to critical control points, this study indentified behavioural control points in between cooking and feeding practices; failure to address these control points in fact increases the level of contamination in food either through food serving utensils (plates, bowls) or through hands.

Behavioural assessment, identification of determinants, assessment of microbiological contamination in food and finally identification of critical and behavioural control points offered sufficient information for prioritising multiple food hygiene behaviours to address all transmission pathways in the next phase, focusing on critical control and behavioural control points. We suggested the following six key behaviours should be prioritised as control measures for behaviour action.

- Thorough cooking of child food
- Cleanliness of serving utensils using ash or soap just before serving cooked or re-heated food
- Handwashing with soap by mothers before feeding, and by child before eating
- Proper storage of leftover cooked food with a tight-fitting lid to protect from flies, dust or dirt
- Thorough re-heating of any stored / leftover food just before feeding child
- Treatment of water and milk before serving to child

To design and implement a food hygiene promotion intervention, we recommend to prioritize five key behaviours, since the majority of mothers already practice thorough cooking in this setting, a practice socially and culturally rooted in the rural settings. Details and rationale regarding the five prioritised behaviours are presented in table 6.
Our formative research had several limitations. Due to the qualitative nature of the study, it was not possible to perform statistical analysis and establish causal associations with the contributing factors. We have best estimated the behavioural outcomes using multiple tools rather than structured observation for all behaviours following one identical method. To best feed the information for the design of the intervention (for next phase), we have explored in-depth understanding and further probe the causes of practicing adverse behaviours which might introduced certain level of bias but use of different tools triangulated the outcomes by various ways. Our evidence may not be generalisable to the urban context of Nepal. The samples were taken during pre-monsoon season hence the microbes presented in this study therefore be much lower than would be typical during the wet seasons in such a contaminated environment. We have only included total coliforms and E.coli as indicators of presence of faecal matter in food, water and milk but it would have been ideal to analyze the presence of faecal pathogens and human-specific bacteriodales species by using sensitive molecular techniques. Such limitations do not negate the implications of this study.

Evidences are mounting that current efforts are insufficient to prevent diarrhoeal diseases in low-income settings; this may be because most programmes exclude food hygiene interventions and therefore fail to address critical transmission pathways. Our findings suggest that current WASH interventions will not effectively eliminate the faecal-oral transmission of microbes unless control measures applied in these points. The HACCP [23] approach including anthropological and consumer research techniques was useful to identify six critical and behavioural control points and to prioritized five key adversely practiced behaviours that are suggested for prioritisation as a control measure and for the design of an intervention. Our next step will be to design a simple and scalable food hygiene intervention targeting key prioritised behaviours for cost-effective implementation in normal community settings using behaviour change principles / approaches. The study also concluded that while designing a food hygiene intervention package, consideration should be given to slight changes in the physical, biological and social environment particularly the kitchen. The immediate motives behind each practice should be taken into consideration while framing key messages. The specific tools should be designed around common motivational themes as drivers of behaviour change such as nurture, disgust, affiliation and social status/respect. This study also tested the detailed methodology to conduct formative research on food hygiene in rural settings that can be applied in other low-income settings.
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Table 1: Social and demographic characteristics of the participants and households (n = 68)

<table>
<thead>
<tr>
<th>Variable</th>
<th>Mean (range)</th>
<th>Variable</th>
<th>Frequency (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mother’s age</td>
<td>28 (17 - 43) years</td>
<td>Main source of HHs income:</td>
<td></td>
</tr>
<tr>
<td>Family members per house</td>
<td>7 (3-18) members</td>
<td>Agriculture</td>
<td>39 (57%)</td>
</tr>
<tr>
<td>Children’s age (n=84)</td>
<td>28 (6-58) months</td>
<td>Self-employment</td>
<td>6 (9%)</td>
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<td>Education level of mothers:</td>
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<td>Manual labor</td>
<td>9 (13%)</td>
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<td>None</td>
<td>18 (27%)</td>
<td>Business/shop</td>
<td>3 (4%)</td>
</tr>
<tr>
<td>Informal</td>
<td>32 (47%)</td>
<td>Others</td>
<td>11 (16%)</td>
</tr>
<tr>
<td>Primary</td>
<td>7 (10%)</td>
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</tr>
<tr>
<td>Secondary</td>
<td>9 (13%)</td>
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<td></td>
</tr>
<tr>
<td>Higher Secondary</td>
<td>2 (3%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cast/Ethnicity of mothers:</td>
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<td></td>
</tr>
<tr>
<td>Brahmin/Chhetri/Thakuri</td>
<td>13 (19%)</td>
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<tr>
<td>Hill Aadiwasi/Janajaati</td>
<td>51 (75%)</td>
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<td>Hill Dalit</td>
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<td>Religion:</td>
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<tr>
<td>Hinduism</td>
<td>18 (27%)</td>
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<td>Buddhism</td>
<td>50 (74%)</td>
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<td>Occupational status of mothers:</td>
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<tr>
<td>Housewife</td>
<td>34 (50%)</td>
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<td>Un-skilled labor</td>
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<tr>
<td>Agriculture</td>
<td>18 (27%)</td>
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<td>Business</td>
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</tr>
<tr>
<td>Others</td>
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<td>Households with refrigerator (observed)</td>
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<tr>
<td>No</td>
<td>67 (99%)</td>
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<tr>
<td>Yes</td>
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<td>Observed human faeces in compound</td>
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<td>Visibly clean compounds</td>
<td>34 (57%)</td>
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<td>Child’s hands visibly clean (observed)</td>
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<td>Visibly clean toilet (n=29)</td>
<td>43 (74%)</td>
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<tr>
<td>Visibly clean compounds</td>
<td>34</td>
<td>Visibly clean Kitchen</td>
<td>45 (67%)</td>
</tr>
<tr>
<td>Visibly clean garbage around the house</td>
<td>40</td>
<td>Visibly clean Utensils</td>
<td>57 (96%)</td>
</tr>
<tr>
<td>Observed animal faeces in compound</td>
<td>79</td>
<td>Visibly clean Kitchen</td>
<td>67 (96%)</td>
</tr>
</tbody>
</table>

Figure 1: Environmental cleanliness during observations (n=68)
Table 2: Types of food fed to young children (n=84 children)

<table>
<thead>
<tr>
<th>Age group</th>
<th>Food type*</th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Liquid</td>
<td>Semi-solid</td>
<td>Solid</td>
<td>Total</td>
</tr>
<tr>
<td>6 - 23 months</td>
<td>1 (2)</td>
<td>8 (19)</td>
<td>33 (79)</td>
<td>42 (100)</td>
</tr>
<tr>
<td>≥24 - 59 months</td>
<td>0 (0)</td>
<td>0 (0)</td>
<td>42 (100)</td>
<td>42 (100)</td>
</tr>
<tr>
<td>Total</td>
<td>1 (1)</td>
<td>8 (10)</td>
<td>75 (89)</td>
<td>84 (100)</td>
</tr>
</tbody>
</table>

*value in parenthesis are percentage

Figure 2: Observed missed opportunities to practice correct food hygiene behaviours (data from video analysis)
Table 3: Microbes in commonly used child food at different stages and in water and milk samples

<table>
<thead>
<tr>
<th>Samples</th>
<th>Microbes</th>
<th>3M PetriFilm</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Total coliforms (TC)</td>
<td>E. coli</td>
</tr>
<tr>
<td>Food sampling stages</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Immediately after cooking directly from vessels (n=30)</td>
<td>Mean (Log10 cfu/gram) [range]</td>
<td>0.14 [0.00 - 2.08]</td>
</tr>
<tr>
<td></td>
<td>SD</td>
<td>0.42</td>
</tr>
<tr>
<td>During feeding from mother's hands (n=30)</td>
<td>Mean (Log10 cfu/gram) [range]</td>
<td>1.48 [0.00 - 2.94]</td>
</tr>
<tr>
<td></td>
<td>SD</td>
<td>1.18</td>
</tr>
<tr>
<td>After 5 hours storage, directly from container (n=30)</td>
<td>Mean (Log10 cfu/gram) [range]</td>
<td>2.03 [0.00 - 3.32]</td>
</tr>
<tr>
<td></td>
<td>SD</td>
<td>1.14</td>
</tr>
<tr>
<td>Immediately after re-heating from re-heating container (n=15)</td>
<td>Mean (Log10 cfu/gram) [range]</td>
<td>1.39 [0.00 - 2.83]</td>
</tr>
<tr>
<td></td>
<td>SD</td>
<td>1.21</td>
</tr>
<tr>
<td>Water samples</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Water during feeding (n=30)</td>
<td>Mean (Log10 cfu/100ml)</td>
<td>1.62 [0.00 - 3.18]</td>
</tr>
<tr>
<td></td>
<td>SD</td>
<td>0.86</td>
</tr>
<tr>
<td>Milk samples</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Milk during feeding (n=13)</td>
<td>Mean (Log10 cfu/ml) [range]</td>
<td>5.15 [0.00-7.96]</td>
</tr>
<tr>
<td></td>
<td>SD</td>
<td>2.17</td>
</tr>
</tbody>
</table>

Total count of TC and E.coli log-transformed. Range (min - max of log transformed count). SD (standard deviation)
Immediately after cooking (n=30) | During feeding (n=30) | After 5hrs storage (n=30) | After re-heating (n=15)
---|---|---|---
Total Coliforms (cfu/gram)
>100 (CFU/grm) & 1 & 14 & 22 & 7
10-100 (CFU/grm) & 29 & 12 & 6 & 6
<10 (CFU/grm) & 4 & 2 & 2 & 4

Figure 3: Counts of microbes in commonly used child food in different stages
Table 4: Mapping - barriers and motives to practice food hygiene behaviours

<table>
<thead>
<tr>
<th>Food hygiene behaviours</th>
<th>Prevalent practices (%)*</th>
<th>Reported barriers by mothers</th>
<th>Reported immediate motives by mothers</th>
<th>Motivationa l drivers</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cooking / thorough cooking</td>
<td>77%</td>
<td>Following traditional habit to cook food only twice a day. Child wanting to eat the same family food so no separate cooking. Time constraints to cook. Saving cooking fuel. Multitasking during cooking</td>
<td>Following rituals - traditional cooking routine habits. Thoroughly cooked food test better. Hot foods don’t cause stomach pain to child.</td>
<td>Affiliation and nurture</td>
</tr>
<tr>
<td>Cleanliness of serving utensils using ash/soap</td>
<td>16%</td>
<td>Rightly un-availability of ash/soap in the yard. Utensils put in mud floor due to lack of kitchen rack. Flies, animal faeces and dust/dirt may re / introduce contamination. Use of old cloths to rinse just before putting food</td>
<td>Cleaning utensils using ash makes them shiny, clean and removes all the dirt like soap. Ash is readily available, don’t have to buy it. Using soap removes stain / oil</td>
<td>Status / respect, disgust</td>
</tr>
<tr>
<td>Handwashing with soap before feeding and eating</td>
<td>7%</td>
<td>No handwashing station in kitchen. Mothers exposure to work during feeding (domestic/agricultural). Soap cost money. All households collect kitchen wastewaster in a container that is then fed to animals, families feared that if they used soap to wash their hands, the animal will not drink the soapy water. Belief on handwashing takes time</td>
<td>Washing hands removes visible dust/dirt/bad odour and produce good smell. Child looks smart if hands are clean. Water readily available</td>
<td>Disgust, nurture and status / respect</td>
</tr>
<tr>
<td>Proper storage of cooked food</td>
<td>43%</td>
<td>Lack of refrigerator, regular electricity. No cupboard to store food. Tradition to use same cooking vessel or bowl/plate to store food. Use of mat to cover food</td>
<td>Proper cover with lid protects food from flies/ dust and prevents from animals (goats, hens, cats)</td>
<td>Disgust and affiliation</td>
</tr>
<tr>
<td>Thorough re-heating</td>
<td>19%</td>
<td>Time and energy required. Difficulties in lighting the fire and fire smoke. Beliefs on no need to re-heat in summer. Fear of burning food. Not having adequate pots. Fear of destroying test and nutritious value.</td>
<td>Re-heated food keeps child’s abdomen warm and protects from abdominal pain. Re-heated food would be testier</td>
<td>Affiliation, nurture</td>
</tr>
<tr>
<td>Milk and water treatment (boiling)</td>
<td>0% (mid-day feeding)</td>
<td>Water: Fear that child doesn’t like the taste of boiled water. Time constraint. Felt no need to boil water in summer Milk: Felt no need to re-boil milk. Fear of wreck the milk if boiled. No proper vessel to boil milk frequently. Believed that raw milk have more nutritious value</td>
<td>No extra fire needed: use leftover fire to boil water. Don’t need to be physically present to boil water like milk. Boiled water when anybody become ill. Boiled milk is testier. Almost all believed milk is ‘nectar’ (Amrit) and special for god to render</td>
<td>Nurture, affiliation, social status / respect</td>
</tr>
</tbody>
</table>

*Various tools were used to triangulate and best estimate the practices*
### Table 5: Hazard analysis - rice, vegetable and milk processing and potential sources of contamination

<table>
<thead>
<tr>
<th>Steps</th>
<th>Possible source of contamination</th>
</tr>
</thead>
<tbody>
<tr>
<td>Rice from the home/market</td>
<td>Field, processing and handling, storage container</td>
</tr>
<tr>
<td>Inspect rice</td>
<td>Hands, utensil, environmental exposure (dirt/dust)</td>
</tr>
<tr>
<td>Wash rice and discard water</td>
<td>Container to collect rice, water, hands, utensil, dirt and dust</td>
</tr>
<tr>
<td>Cook rice</td>
<td>Utensil, time, temperature, water, duration, location, fuel used</td>
</tr>
<tr>
<td>Store rice</td>
<td>Cover, location, animal exposure, ambient temperature, utensil, flies</td>
</tr>
<tr>
<td>Vegetables from kitchen garden/market</td>
<td>Field, utensil, hands, environmental exposure</td>
</tr>
<tr>
<td>Wash and cut vegetables</td>
<td>Water, bowl/plates, knife, chopping board, flies</td>
</tr>
<tr>
<td>Put oil in the pot (oil from market)</td>
<td>Utensil, hands, environmental contamination (dirt/dust), smoke/ash</td>
</tr>
<tr>
<td>Grind coriander seeds, turmeric powder, chilli, garlic, ginger etc in the stone</td>
<td>Contaminated spices, hand and utensil contamination to peel and cut garlic/ginger, rock and mortar, water, flies</td>
</tr>
<tr>
<td>Cook vegetables</td>
<td>Spices (Masala), water, utensil, temperature, duration, location, environmental exposure, cover, flies</td>
</tr>
<tr>
<td>Add water and salt</td>
<td>Water, water vessel, salt, temperature, salt container</td>
</tr>
<tr>
<td>Stir and take out from stove</td>
<td>Spoon, cover, animal exposure, environmental exposure</td>
</tr>
<tr>
<td>Store curry</td>
<td>Cover, location, animal exposure, ambient temperature, utensil, flies</td>
</tr>
<tr>
<td>Milking cow / buffalo</td>
<td>Milking container, hands, water, cow/buffalo, location, flies/insects</td>
</tr>
<tr>
<td>Milk processing</td>
<td>Filter (rare practice) using sieve or cloth, boil temperature, utensil</td>
</tr>
<tr>
<td>Milk storage</td>
<td>Inadequate cover, environmental exposure (flies, dirt), storage utensil, animal exposure,</td>
</tr>
<tr>
<td>Wash hands before feeding</td>
<td>Hands, water, techniques</td>
</tr>
<tr>
<td>Serve food with curry or milk</td>
<td>Utensils/spoon, plates/bowls, water, hands, glass</td>
</tr>
<tr>
<td>Feeding to child</td>
<td>Mother’s hands, child’s hands, spoon, bowls/plates, environmental, water, milk, Jad, flies</td>
</tr>
<tr>
<td>Store with cover or without cover</td>
<td>Duration, location, inadequate cover, flies/insects, animal exposure, dirt/dust, ambient temperature</td>
</tr>
<tr>
<td>Briefly reheat/no re-heat before feeding</td>
<td>Duration, utensils, temperature, hands</td>
</tr>
<tr>
<td>Feeding re-heated or cold food</td>
<td>Hands, spoon, bowls/plates, environmental, water, milk, Jad, flies</td>
</tr>
</tbody>
</table>
Table 6: Five key prioritized behaviours and reasons for promotion

<table>
<thead>
<tr>
<th>Illustrations</th>
<th>Key behaviours</th>
<th>Key behavioural actions</th>
<th>Reasons for promotion</th>
</tr>
</thead>
<tbody>
<tr>
<td><img src="image1" alt="Illustration" /></td>
<td>Cleanliness of child food serving utensils</td>
<td>Cleaning of serving utensils using ash/soap before serving cooked/re-heated food into serving utensil</td>
<td>Serving utensils likely to carry microbes. Cleaning them before serving food reduces contamination. Current practices are inadequate and microbial levels increased when serving utensils used</td>
</tr>
<tr>
<td><img src="image2" alt="Illustration" /></td>
<td>Handwashing with soap by mother before feeding and child before eating</td>
<td>Mothers wash both hands using soap before feeding child. Child both hands are washed using soap before eating food</td>
<td>Hands likely to carry faecal microbes. Handwashing with soap can reduce microbes contamination on hands. Low soap use observed, and increased microbe levels obtained when food was mixed using hands</td>
</tr>
<tr>
<td><img src="image3" alt="Illustration" /></td>
<td>Proper storage of cooked food</td>
<td>Store cooked food in containers with a tight-fitting lid. Protect stored food from flies / dust / dirt</td>
<td>Flies and dust/dirt are likely to carry faecal microbes and contaminate food. Flies were accessing food and other possible contamination routes were observed. No refrigerator found. Stored foods were heavily contaminated. Tight-lid may protect stored food from flies / dust / dirt and cross-contamination</td>
</tr>
<tr>
<td><img src="image4" alt="Illustration" /></td>
<td>Thorough re-heating of leftover/stored food</td>
<td>Thoroughly re-heat leftover / stored food before feeding to child. Maintain temperature at least at 70°C while re-heating</td>
<td>Thorough re-heating/cooling (70°C) kills most vegetative forms of microbes. Due to absence of refrigeration, all food is stored in ambient temperature, though cooking was adequate but re-heating practice are inadequate and microbes were found in re-heated food.</td>
</tr>
<tr>
<td><img src="image5" alt="Illustration" /></td>
<td>Milk / water treatment</td>
<td>Boil milk before feeding to child (at any time). Only serve treated water</td>
<td>Improperly boiled milk stored in ambient temperature w/o cover likely to carry microbes; contamination also likely in stored water. Heavy contamination noted in both milk and water</td>
</tr>
</tbody>
</table>

*Note: Thorough cooking was one of the main critical control points however not prioritized as many mothers currently practicing such behaviour.*
Mothers gave many reasons during formative research why they should wash hands, why not & difficulties, and why not reheat food & few reasons why they should. The following table gives some of the verbatim quotes from the mothers:

<table>
<thead>
<tr>
<th>Verbatim in relation to handwashing at different times</th>
<th>Verbatim related to ‘why not and difficulties’</th>
</tr>
</thead>
<tbody>
<tr>
<td>• ‘I wash my hands with water if I see any dirt / dust in my hands’</td>
<td>• “In kitchen we don’t use soap because we feed waste water to animals, if we use soap they will not drink. I told everyone to wash hands out if needed.’’</td>
</tr>
<tr>
<td>• ‘I wash my hands with soap after cleaning utensils because my hands would be black if I don’t use soap after cleaning them’</td>
<td>• “Sometimes do not get the time to wash hands, sometimes there is no soap in the house, sometimes there is no money to buy soap and sometimes forget to wash the hands”</td>
</tr>
<tr>
<td>• “If we wash hands our children will also learn that behaviour”</td>
<td>• “I always use water to wash hands but findings soap always is quite difficult”</td>
</tr>
<tr>
<td>• ‘I wash hands because it removes the dirt and germs’</td>
<td>• “I wash my hand before feeding the child so why to wash child’s hand before feeding”</td>
</tr>
<tr>
<td>“Unless we put water in the hands before eating/feeding we are not satisfied, this is our tradition”</td>
<td>• “We have soap but not everyone in the house uses it”</td>
</tr>
<tr>
<td>• ‘We have a child who had difficulties to walk, speak and eat. I have to wash his hands with water before he eats food. We don’t have money to buy the soap’’</td>
<td>• ”When I washed my hands and feet with soap &amp; water it caused allergy so I wash my hands and feet with ash and water”</td>
</tr>
<tr>
<td>• ‘Washing hands with soap before cooking and feeding child can ensure child’s food safety in the kitchen’’</td>
<td>• “’What’s the harm of washing hands with mud or cow dung if there is no soap and ash? If available, I prefer ash but young daughters wants soap’.”</td>
</tr>
<tr>
<td>• “I always wash child's hands with water before feeding to baby but if grandmother is feeding, she never does this”.</td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Reasons for not re-heating or heating the stored food</th>
<th>Reasons for not re-heating the food</th>
</tr>
</thead>
<tbody>
<tr>
<td>Why to re-heat food?</td>
<td>“No one comes in time to eat food and child wants food frequently so how many times to reheat the cooked food”</td>
</tr>
<tr>
<td>• “Hot food is good for the health, and it doesn’t cause stomach pain”</td>
<td>• “I don’t re-heat because it will take time and energy and it will burn”</td>
</tr>
<tr>
<td>• “In winter, it is nice if the food is warm”</td>
<td>• “If I re-heat food, all nutritious value will destroy and child will have diarrhoea after eating that food, it is therefore, I won’t re-heat the food”</td>
</tr>
<tr>
<td>• “Re-heated food is always tasty and child will eat more”,</td>
<td>• “If we re-heat cooked food long, it will be sticky and all nutrients value will go away”</td>
</tr>
<tr>
<td></td>
<td>• “For long re-heated food, we considered as leftover food, hence I always briefly re-heat if I had to do so in cold season. That way it will also be quicker to feed child straight away”</td>
</tr>
<tr>
<td></td>
<td>• “Who bothers to reheat the food? Child eat the cold food by themselves at any times without our any support”</td>
</tr>
<tr>
<td></td>
<td>• “In winter we reheat the food before eating but in summer if the food is reheated it becomes sour”</td>
</tr>
<tr>
<td></td>
<td>• “If child starts crying, I can immediately provide leftover food”</td>
</tr>
<tr>
<td></td>
<td>• “If I cook food in the morning, it will be sufficient up to evening for child. I don’t re-heat the food in summer because child can’t eat hot food”</td>
</tr>
<tr>
<td></td>
<td>• “I just briefly re-heat the food because no one can eat the hot food”</td>
</tr>
</tbody>
</table>
Figure 4: Food flow diagram of commonly used child food - likely hazards, critical and behavioural control points

- **Fresh milk**: Initial contamination likely (in field, while processing, handling, storing)
  - Contamination from utensils likely (unclean utensils, knives etc)
  - Environmental contamination / cross contamination likely (dirt/dust, location, animals/flies/insects)
  - Hands contamination likely
  - Water contamination likely

- **Rice**: Critical Control Point (CCP) and Behaviour Control Point (BCP):
  - Bacteria destruction
  - Bacteria propagation

- **Spices**
  - Methi, Turmeric powder, Garlic, Salt

- **Oil**

- **Fresh milk**

- **Filter**

- **Boil**

- **Cook**

- **Wash**

- **Inspect**

- **Cut**

- **Peel**

- **Sinki or veg**

- **Water**

- **Store in ambient temp**

- **Serve/Feed (serve to child)**

- **Put into bowls/plates and mix the food**

- **Temperature check: Dipping fingers into the mixed food**

- **Feed/serve with hands (common) or spoon (few)**
## RESEARCH PAPER COVER SHEET

### SECTION A – Student Details

<table>
<thead>
<tr>
<th>Student</th>
<th>Om Prasad Gautam</th>
</tr>
</thead>
<tbody>
<tr>
<td>Principal Supervisor</td>
<td>Dr Val Curtis</td>
</tr>
<tr>
<td>Thesis Title</td>
<td>Effect of an intervention to change food hygiene behaviour in Nepal</td>
</tr>
</tbody>
</table>

### SECTION B – Paper already published

<table>
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<tr>
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</thead>
<tbody>
<tr>
<td>When was the work published?</td>
<td></td>
</tr>
<tr>
<td>If the work was published prior to registration for your research degree, give rationale for its inclusion</td>
<td></td>
</tr>
<tr>
<td>Have you retained the copyright for the work?</td>
<td>Was the work subject to academic peer review</td>
</tr>
</tbody>
</table>

### SECTION C – Prepared for publication, but not yet published

| Where is the work intended to be published? | THE LANCET Global Health |
| Paper’s authors in the intended authorship order: | Om P. Gautam, Wolf P. Schmidt, Sandy Cairncross, Sue Cavill, Valerie Curtis |
| Stages of publication | Not yet submitted |

### SECTION D – Multi-authored work

| Give full details of your role in the research included in the paper and in the preparation of the paper | I led the design of the study, the protocol development, the intervention design, the trial set-up and its conduct in the field, the evaluation, the analysis, and the write-up. |

Student Signature: ____________________________  Date: 25 September 2015

Supervisor Signature: ____________________________  Date: 25 September 2015
CHAPTER – V

(Paper 2)

Study Title:
Effect of an intervention to change food hygiene behaviour in Nepal

Authors:
Om Prasad Gautam¹, Wolf-Peter Schmidt¹, Sandy Cairncross¹, Sue Cavill², Valerie Curtis¹

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Key words:
Food hygiene, behaviours, intervention, microbes, diarrhoea, mother, children
ABSTRACT

Background: Poor food hygiene is likely to be a major contributor to child morbidity and mortality. In this study we report on the results of a trial of an intervention to improve five food hygiene behaviours amongst mothers of children aged 6-59 months in rural Nepal.

Methods: A food hygiene intervention was designed and tested using Behaviour Centred Design- a systematic 5 step process to: A, Assess, B, Build, C, Create, D, Deliver and E, Evaluate the intervention. In step A, the current state of knowledge and the parameters for the intervention were established. In step B, formative research was carried out to understand key behaviours and identify likely drivers of change. In step C, an intervention package was created and piloted and in step D, it was delivered by female hygiene motivators over a period of 3 months. The intervention targeted five behaviours and employed four emotional drivers of behaviour whilst changing the physical and social settings of kitchens. In step E the effect of the package on behaviour was evaluated via a cluster-randomised before-after study with a control group. Four villages were randomly selected to receive the intervention and four served as a control with 29-30 households with a child aged 6-59 months in each cluster. The primary outcome was the cluster level mean difference in proportion of mothers directly observed practicing all five food hygiene behaviours.

Results: The five targeted food hygiene behaviours were rare at baseline (composite performance of five behaviours in intervention 1% [SD 2] and in control groups 2% [SD 2]). Six weeks after the intervention the target behaviours were more common in the intervention group than in the control group (43% [SD14] vs. 2% [SD 2], p=0.02) during follow-up. The difference of differences was thus an increase in the mean proportion of mothers practicing all five targeted behaviours of 42% (p=0.02). The intervention appeared to be equally effective in improving all five behaviours in all intervention clusters.

Conclusion: This study shows that a theory-driven intervention employing emotional motivators and changing behaviour settings was capable of substantially altering multiple food hygiene behaviours in Nepal.
INTRODUCTION:

Despite recent progress[1], diarrhoeal disease still kills 700,000 under-five children every year globally[2]. Interventions that can prevent the transmission of the agents of gastro-enteric infection include improving water supply, sanitation facilities, handwashing with soap, and exclusive breastfeeding. However, the role of food hygiene in disease transmission has been largely overlooked. One estimate suggests that up to 70% of diarrhoeal episodes in developing countries may be caused by pathogens transmitted through food[3, 4]. Contaminated weaning foods are thought to be a major contributor to diarrhoeal diseases and malnutrition in low-income contexts [5, 6] though observational studies have been inconclusive[7]. Interventions to improve food hygiene practices are a likely candidate for public health investment, whether in the context of Water, Sanitation and Hygiene (WASH), nutrition or other preventive health programmes. However, it is not yet clear whether and how food hygiene practices can be improved in the places where safe practices are most needed – in kitchens in low-income settings. This study was designed to explore whether a systematically designed, theory-driven, scalable intervention could improve food hygiene behaviours in a context of poor household infrastructure, low socio-economic development and high disease burdens, namely rural Nepal.

Inadequate food hygiene may be accountable for diarrhoeal diseases in infants and young children. High rates of diarrhoeal disease in childhood also predispose to malnutrition among young children [8-10]. Diarrhoea risk increases during the infant weaning period in low income settings [5, 11-13] and child growth often falters after the initiation of weaning [9]. Weaning foods are often prepared in unhygienic conditions and infants who, until then have consumed only breast milk, may be exposed to infective doses of food-borne pathogens[5]. Foods prepared in unhygienic conditions are an important factor in the transmission of diarrhoeal agents to children under five years old in low resource settings[5, 14]. Foods also provide a route for the transmission of the agents of Environmental Enteropathy, which may be a cause of child malnutrition in developing countries[15, 16]. However, research into food hygiene has been neglected and, partly as a result, diarrhoea and undernutrition prevention programmes tend to prioritise breastfeeding promotion, food and micronutrient supplementation and immunisation rather than food hygiene and safety[17] in low-income settings.
Developing country homes provide many challenges to safe food hygiene practice[18, 19] including: high ambient temperatures[14, 20], lack of refrigeration, poor storage facilities[21], inadequate sanitation and presence of animals in kitchens leading to environmental faecal contamination[16, 22], lack of running water[23], cooking fuel scarcity[5], hard-to-clean surfaces, often compounded by heavy female workloads and poor access to information on safe hygiene. Poor practices that have been documented include time gaps between meal preparation and feeding[6, 24, 25], the use of unclean utensils[26, 27], washing utensils in contaminated water[23], allowing flies to access foods, not washing hands prior to food handling and feeding[28] and the use of dirty cloths for wiping hands/utensils. A number of studies to date have assessed risk factors and microbial contamination in food in developing countries [11, 29-32]; but only a few have reported designing and testing interventions to counter the problem in domestic settings [33, 34].

Changing people’s behaviour is a difficult and complex undertaking. Behaviours are determined by a wide array of factors, not just knowledge. Interventions on hygiene behaviour that focused on emotional drivers (such as nurture, disgust, affiliation and status) have had more success than those that have promoted the health benefits [35-38]. Building on the success of previous handwashing intervention in India[35] we used Behaviour Centred Design (BCD)[39], a process of designing behaviour change intervention underpinned by Evo-Eco theory[40] to design and evaluate a food hygiene behaviour change intervention in the challenging context of rural Nepal. Though Nepal’s health indicators are improving[41], diarrhoea is still the second most important cause of death in under-fives. 41% of children are stunted[41] and Nepal ranks among the top twenty countries for absolute numbers of stunted children [42]

This paper focuses on the design and evaluation of the food hygiene intervention with mothers’ food hygiene practices as a primary outcome. Other papers in this series concern the formative research that informed the design of the intervention and the impact on microbiological contamination of child food.
METHODS:

Study site and population:
The study was conducted in Kavre District, Nepal, a rural hilly area, between October 2012 and December 2013. Eight wards (clusters) were randomly selected from 18 eligible wards from two adjacent Village Development Committees (VDCs). To be included in the study wards had to be: rural (up to ~2,195m elevation), have a heterogeneous population (minimum two ethnic/caste groups), be geographically separated have more than 30 households with a child aged 6-59 months, have low sanitation coverage, have high diarrhoea prevalence according to local health institution data and be a cluster where formative research had not been conducted. The reference populations of the selected wards ranged from 417 - 786 people. All households with at least one child aged 6-59 months in eight clusters became the study population.

In Kavre, houses are made of mud and stone. In most households, the single ground floor room serves for cooking, sitting, sleeping and sometimes also for keeping animals. Most food preparation takes place on the floor and firewood is the main sources of fuel. Half of the population uses piped water from unprotected springs, the other half surface water and the majority of the households have no toilet. Animal and child feaces are visibly present throughout the study villages.

Recruitment, randomization and masking:
Figure 1 shows the flow diagram for the trial. Within each cluster 29-30 households having at least 1 child aged between 6-59 months were randomly recruited to participate in the study. The final sample included 239 households from eight clusters. Written informed consent was received from the mothers in all participating households. Detailed social, demographic and economic information was collected from each household using a closed-ended structured questionnaire.

The clusters were then randomized into four interventions and four control clusters. The intervention clusters received the food hygiene promotion intervention and no intervention was delivered in the control clusters. During baseline, there were 120 households with a child aged 6-59 months in the four intervention clusters, and 119 in the four control clusters.
Outcome measurement was carried out approximately 45 days before and 45 days after completion of the three months’ intervention. The primary outcome of interest (the proportion of mothers sustaining all key food hygiene behaviours) was measured by food hygiene observers not connected with the intervention. It was not possible to fully blind the study participants however mothers were told that the purpose of the observation was to document their daily routines.

**Intervention design and delivery:**
The food hygiene behaviour change motivational package was designed on the basis of formative research following the steps of Behaviour Centred Design (BCD) and Theory of Change. The key target behaviours were pinpointed through formative research applying the principles of Hazard Analysis and Critical Control Point (HACCP)[43]. BCD involves five 5 steps: A Assess, B: Build, C: Create, D: Deliver and E: Evaluate[39]:

**Step A (assess):** The first step involved the collection and analysis of scientific and local knowledge concerning food hygiene behaviour to define target behaviours, the parameters of the intervention and the questions to be answered in the Formative Research. We carried-out a systematic review of literature on food hygiene (presented elsewhere), examined past experience, in particular small scale weaning food studies in Mali[34], Brazil[44] and Bangladesh[33], other hygiene interventions, learning particularly from the successful SuperAmma handwashing trial in India[35], and the WHO five key behaviours for safer food initiative[45]. We consulted colleagues in government and NGOs to establish parameters concerning how to ensure the replicability and scalability of the intervention in the context of Nepal. Our assessment of knowledge gaps provided the agenda for our formative research.

**Step B (build):** Formative research was conducted to gain insights into specific behaviours; actors; and behavioural determinants including habits, motives and plans, and social, physical and biological factors in the kitchen and village environment (the key elements of the BCD model[39, 40]). To provide additional information so as to decide which of many candidate food hygiene behaviours to prioritize, we carried-out a HACCP assessment[43] supplemented with food testing for microbial contamination. This work led us to identify five key behaviours to target:

- Cleaning of child food serving utensils using soap/ash before serving food
- Handwashing with soap by mother before feeding, and by child before eating
• Storage of cooked food in containers with a tight-fitting lid
• Thorough re-heating of leftover/stored food before feeding the child (temp ≥70°C)
• Serving only treated water to the child

A further critical control point: the thorough cooking of foods to be fed to a child was also identified. However, this was already common practice so did not need to be targeted. Figure 2 illustrates the 5 targeted behaviours.

**Step C (create):** An ad hoc creative team with expertise in marketing, graphic design, product innovation, programme development and behaviour change was brought together to design the intervention informed by formative research findings. Our creative brief required an intervention package that employed the motives of affiliation and disgust and that changed behavioural settings (social and physical determinants of behaviour) [40, 46]. Key principles were that the intervention should be recognisable, feasible to implement by local health extension agents and have a reasonable possibility of replication at larger scale. Prototypes of the intervention components were developed and pretested in several iterations in non-study areas and the package was finalized after incorporating government and NGO stakeholder feedback. Package included novel activities linked with emotional drivers and behavioural settings to encourage five key behaviours.

**Step D (deliver):** The food hygiene promotion package was delivered through six events followed by six door-to-door household visits by 15 local women who were trained and mobilised as ‘Food Hygiene Motivators’ over a period of three months during May-August 2013 (see Table 1). The primary target audience were mothers having a child aged 6-59 months. The campaign’s theory of change was that mothers would identify with a central ‘ideal mother’ character, who practiced safe hygiene so as to be respected in the community (status motive). In addition we identified Nurture, Disgust and Affiliation as possible key levers of change, and recognised the need to disrupt daily food preparation habits that were held in place by tradition, routine and the social and physical settings of kitchens in which the target behaviours took place.

The campaign activities were implemented by FHMIs following training and using a written manual. The implementation details can be accessed online at (http://www.sharereresearch.org/NewsAndEvents/Detail/om_blog_food_hygiene_study) and a YouTube video documentary can be accessed at (https://www.youtube.com/watch?v=gPj6IyN0ZCU).
Nurture based activities included: child life game, letter exchange, family drama. Affiliation based activities included: folk song, puzzle game, peer review, cookery demonstration. Disgust based activities included: glo-germs, 3M PetriFilm, hot potato game, disgust exercise. Social and physical settings of kitchens were disrupted by holding makeover parties where the kitchen was redecorated using coloured bunting, danglers placed at eye level in kitchen, designated a clean food hygiene zone and neighbours agreed to practice new hygiene rituals. Interest were generated through storytelling, video-clip and illustration demonstration and behaviour were reinforced highlighting key principle messages and immediate motives using illustrations. Habit formation through demonstration such as cooking & re-heating, use of a baby bib that has illustration and messages i.e. “did you wash your hands with soap before feeding me?” Social norms elicited through competitions and pledging such as ‘clean kitchen’, ‘ideal mother’, ‘safe food hygiene zone’, public commitment, social/public pledging such as placement of ideal mother photos and safe food hygiene zone bill board in the junction of the village, rally chanting “ideal mother Hi Hi, diarrhoea Bye Bye!”. Images from the campaign are shown in Figure 3.

**Step E (evaluate):** Below we describe the evaluation of the intervention.

**Outcome assessment:**

The primary outcome, i.e., the proportion of mothers practicing all five target behaviours – was measured by structured observation[35, 37]. The proportion of mothers: i) cleaning child food serving utensils using soap/ash; ii) washing both hands with soap before feeding child and washing the child’s hands before eating; iii) storing cooked food with tight lid and no visible flies/dust/dirt in the food; iv) thoroughly re-heating leftover/stored food at adequate temperature (≥70°C); v) serving treated water to their children was assessed by direct observation (and temperature measurement). Assessments took place 45 days before and 45 days after the intervention period. Twenty-five independent female food hygiene observers were recruited and trained to carry out the structured observation of food hygiene behaviours. Observations were made in all intervention households (n=120, with no loss to follow up) and control (non-intervention) households (n=119 with two lost to follow up) at baseline and follow-up (post-intervention). Observations were carried out between 1:00pm and 5:00pm (4 hrs of continuous observation), when the behaviours of interest were likely to be seen. Observations took place in both groups simultaneously, and were completed within 12 days. Observers recorded the behaviour of mothers with a child aged 6-59 months in the respective
households. Observers were kept blind to the study objectives and independent of the intervention. A structured observation checklist was used to record all behaviours. The reach of the intervention was also assessed post-intervention to ascertain exposure density and to check for contamination of the control group.

**Sample size:**
We calculated that a sample size of eight clusters with a minimum of 28 households per cluster for two sample comparisons of proportions (two-sided test) using 95% confidence interval (p<0.05), 90% power, 5% loss to follow-up (0.05), 1.29 design effect would allow us to detect a difference of 20% (7% in control group, 27% in intervention group) in cluster prevalence of target behaviours (thorough re-heating as a basis) between the control and intervention arms.

**Statistical analysis:**
Our primary outcome was the comparison of cluster-level means proportion of the observed practice of all five behaviours as a composite performance score before and after the intervention. We used cluster-level analysis since the intervention was allocated by cluster [47]. As a secondary analysis, we compared all individual behaviours at cluster level by different groups during baseline and follow-up. Since we only had 8 clusters (4 in intervention and 4 in control), we used a non-parametric test i.e. the two-sample Wilcoxon rank-sum test (Mann-Whitney U)[48] to compare cluster-level means and to estimate statistical support. This test does not rely on the assumption of normality and is resistant to outliers[47]. The effect size of the intervention was calculated by difference-of-differences i.e. [Follow-up – baseline]_{intervention} minus [follow-up – baseline]_{control}. Sub-group analysis stratified by religion, caste/ethnicity, educational level, economic status and types of cooking fuel was carried out. Statistical support for effect modification was assessed by computing the difference in food hygiene behaviours (composite performance) between sub-groups within each cluster, comparing the mean difference of differences between intervention and control clusters, following the method described by Cheung and colleagues[49]. The intraclass correlation coefficient was calculated using the STATA ‘loneway’ command. Data were entered into a spreadsheet and SPSS, and statistical analysis was performed using SPSS 19 and STATA 12.
Ethics:
Ethical approval for the study was granted by the ethics committees of the London School of Hygiene and Tropical Medicine, UK, and the Nepal Health Research Council. Informed consent was obtained from all participants. To preserve anonymity, findings were presented without ascribing personal description. Funders played no role in this study.

RESULTS:
Social and demographic characteristics:
Table 2 shows that intervention and control clusters and households had similar social and demographic characteristics. Clusters ranged in size from 75 to 141 households (417 to 786 people). The mean age of participating mothers was 27 years, and the majority lacked formal education. Over 50% of mothers in both groups belonged to the Hill Aadiwaasi/Janajaanati ethnic group (part of the historically deprived Tamang). Around one third of mothers were of the Brahmin/Chhetri caste, and 8% were hill Dalit. Most households earned less than 10,000 Nepali Rupees (NRs) per month (~100US$/month), mainly from agriculture. Only half of the participating households had latrines and around 65% of households reported disposing of their child’s faeces in fields. Animal faeces were observed in 86% of household compounds. Soap was observed in more than 80% of households in both groups. Only one household had a refrigerator.

Feeding practices:
The intervention and control group did not differ significantly by feeding practices at baseline. Around 58% of children had received supplementary food before the age of six months. Children were fed with different types of liquid (water, cow or buffalo milk), semi-solids (jaulo, lito-made from roasted rice flour, ghee and sugar), solids (rice, dhido - a type of porridge with curry/dal/vegetable), dry food (beaten rice, popcorns) and snacks (dry noodles). Some ethnic group also fed jad (an alcoholic brew). The majority of households (86%) fed the same staple food to their children that they themselves consumed daily. Nine in ten households cooked only twice a day, in the late morning and late evening, but children were mostly fed four times a day with stored or leftover food. Food was cooked mostly by mothers and/or grandmothers and fed by hand to children.
Reach of the intervention:

Following the intervention, all mothers had heard of and participated in the campaign, compared with almost none in control cluster (see Table 3). Out of 12 expected exposures (two community events, four group events and six household visits) during the three-month campaign period, 90% of mothers were exposed at least ten times. All intervention group mothers were able to describe the five key behaviours that ‘ideal mothers’ should practice.

Effects of the intervention on food hygiene behaviour:

Figure 4 shows that the cluster average of mothers performing all five target behaviours was low in both intervention and control groups at baseline (1% [SD 2] vs 2% [SD 2]). Following the campaign the key behaviours were more common in the intervention group than in the control group (43% [SD 14] vs. 2% [SD 2], p=0.020; see Figure 4/Table 4). After adjusting for the baseline prevalence measured in both the groups, the effect size of the intervention (as a difference of differences) was an increase in the mean proportion of target behaviours of 42% (p=0.020); see Table 4.

Target behaviours improved in all intervention clusters from 0% to 30% (p=0.002) in cluster 1, from 0% to 37% (p=0.001) in cluster 2, from 0% to 63% (p<0.001) in cluster 7 and from 3% to 43% (p=0.001) in cluster 8 (Figure 5). There was no difference among the control clusters.

Figure 6 and Table 4 gives the changes in each targeted behaviour. The proportions of mothers practicing individual behaviours in intervention and in control group at baseline were almost similar. After the intervention there was a major improvement in all behaviours in the intervention groups, but not in the control group. After adjusting for the baseline prevalence measured in both the groups, the effect size of the intervention (as a difference of differences) was an increased in the mean proportion of 54% (p=0.021) for cleanliness of child food serving utensils using soap or ash just before serving food behaviour. The campaign was equally successful to improve handwashing with soap behaviour. The effect size of the intervention for washing both hands with soap and water (HWWS) just before feeding child by mother was an increased in the mean proportion of 64% (p=0.021) and handwashing with soap by children before eating was an increased in the mean proportion of 63% (p=0.021). Similarly, the effect size of the intervention was an increase in the mean proportion of 69%
(p=0.021) for households storing cooked or leftover food in containers with tight fitting lids and no flies or no visible dirt or no dust present in stored food behaviour.

The effect size of the intervention was an increase in the mean proportion of 85% (p=0.020) for the behaviour ‘thoroughly re-heating stored or leftover food before feeding their child, and maintaining ≥70°C temperature’. When re-heated, food mean temperature was 54°C [min 30°C – max 75°C] in the intervention and 61°C [min 35°C – max 78°C] in the control group during baseline and during follow-up, the mean temperature of re-heated food was 76°C (minimum 55°C – maximum 92°C) in the intervention group. The effect size of the intervention for ‘water treatment practices’ was an increase in the mean proportion of 74 percent-point (p=0.021). Although milk was not included in the universal analysis, our results showed that in those households that served milk, more than 84% and 18% boiled milk before serving to their children during follow-up in the intervention and control group respectively. Though there appeared to be some difference in effect size with education, ethnicity and type of cooking fuel, statistical tests failed to support this.

The pile sorting exercise during reach measurement helped to rank behaviours according to the ease of practicing the behaviours by intervention group mothers. Proper food storage, handwashing with soap and cleanliness of serving utensils were ranked as relatively easy to perform behaviours by 55%, 49% and 34% of mothers respectively. The boiling water and re-heating food were ranked as difficult to performed behaviours by 41% and 48% respectively.

The intra-class correlation coefficient of key food hygiene behaviours (effect of all behaviours) at village level was 0.000 at baseline and 0.043 at follow-up. At household level, the intra-class correlation coefficient was 0.000 during baseline and 0.475 during follow-up period.

**DISCUSSION:**

This study suggests that it is possible to change entrenched food hygiene habits, even in environmentally challenging conditions such as pertain in rural Nepal. We attribute the apparent success of the intervention to the use of a systematic process employing global and local knowledge, behavioural theory and a creative process to design a tailored intervention
targeting emotional drivers of food hygiene behaviour as well as changing food preparation settings.

An alternative interpretation of the study results is that the changes in behaviour that were observed were not representative of real behaviour, but were due to reactivity on the part of observed mothers. In effect, mothers may have been anxious to demonstrate to observers that they had learnt the lessons of the campaign by adhering closely to target behaviours during the observation period, but did not actually change their daily food hygiene routines. We think that this is unlikely because: a) observers were not connected by mothers to the intervention and mothers were told that the observation was to monitor their daily routine; b) observers were not connected to programme implementation, c) behaviours were only observed twice during the study period; and d) random spot checks and triangulation while collecting microbial samples. Limited information on the evaluation of food hygiene intervention using direct observation is available, but several studies that have been conducted to measure the effectiveness of handwashing with soap interventions were through direct observation such as in Bangladesh[38] and India[35, 37]. Those studies claimed that the differential reactivity was either none-existent or very low if participants make no link between the intervention and the outcome measurement process. The intended behaviours (HWWS) remained very low in control groups, even after several rounds of observation[35].

The fact that the intervention was equally effective across targeted behaviours and across clusters and in differing socio-economic settings suggests that the improvements were due to the effects of the intervention itself and this is a robust intervention capable of being generalised. Emphasis was placed on motivating mothers rather than educating them using a creative approach and behaviour change science. Several authors have called for the use of more creative and innovative techniques to change public health related behaviour [35, 37, 38, 50, 51]. We also paid attention to the training and motivation of our outreach workers (FHM)s as studies have suggested that the quality of interventions improves if implementation team is skilled[35].

The question of which elements of the intervention were most effective cannot be determined from this quantitative study. Hence we do not know if the kitchen makeovers or the activities based on nurture, disgust, affiliation, or status were the active ingredients. We suspect that the food preparation setting disruption activity was particularly effective, involving as it did a
transformation in the physical environment (repainting, bunting, danglers as behaviour reminder, kitchen tools), the script (mothers committing to behave in a new way), social control (commitment made in front of their neighbours) and the changing designation/purpose of the setting (from kitchen corner to safe food hygiene zone). Figures 3 (right top picture) show a typical transformation from all-purpose room used for cooking, eating, running the household, sleeping, and, at times, keeping animals to having a beautiful, bright and special small kitchen in one corner. The settings idea is a powerful one that was laid out in the 1950s by ecological psychologists[46], but that has since been neglected. Yet Roger Barker showed that knowing settings can predict behaviour 90% of the time, and all behaviour takes place in settings[52]. This concept could be useful for changing health-related behaviour.

The use of emotional motivators such as nurture, disgust, affiliation and social status may have motivated the key behaviours. Our focus on the positive emotional rewards of each behaviour (becoming an ideal mother, shiny serving utensils, child’s warm tummy, tasty food, social approval) probably helped to reinforce each behaviour, making them part of the daily food preparation routine. To our knowledge this is the first study to attempt to use emotional drivers to affect food hygiene behaviours, though they have been shown to work on other hygiene behaviours such as handwashing in low [35, 36, 53] and high income [54, 55] countries.

This study faced the particular challenge of trying to change five different behaviours at the same time. We addressed this through identifying and disrupting the behavioural setting that was common for all of these practices (the food preparation setting) and by using drivers that could be associated with all of the behaviours (disgust, nurture, affiliation and status). We further suspect that performing one behaviour serve as a reminder to perform another. For example, mothers practicing cleanliness of serving utensils just before feeding their children were highly likely to remember to wash their hands just before feeding, as both activities happen simultaneously. Many of the target behaviours happened in sequence; for example, immediately after re-heating the food, the mother served the food using serving utensils, then washed her hands and stored the leftover food properly. It may thus be easier to change multiple behaviours when they are practiced in similar settings and in sequence, when the practice of one can cue another.
The intervention was relatively intense, with 6 events and 6 door-to-door contacts. Based on our detailed process evaluation (paper forthcoming), the intervention could be further simplified for wider scaling-up. This study, however, provides proof of principle that food hygiene can be improved in challenging environments provided that interventions are based on a careful process involving Formative research, behavioural theory and imaginative and motivating creative campaigns. Behaviour Centred Design provides a simple process framework for the design of such interventions (the ABCDE steps) as well as theoretical basis for identifying key drivers of and a theory of change for behaviour. This study has been able to minimise persistent knowledge and programmatic gap on food hygiene by designing and implementing effective food hygiene intervention. This package can be delivered through public health, WASH, and nutrition programmes in low-income settings however it is as yet unclear whether the large-scale replication of the package will achieve the same degree of behaviour change. It now remains to streamline the intervention to a form that can be delivered on a mass scale across the country, and indeed in the many other countries where such interventions are needed.

**LIMITATIONS OF THE STUDY:**

As with most behaviour change interventions in communities [35, 37] it was not feasible to fully blind participants or observers. Participants in the intervention group were aware that they were receiving the intervention. Data collectors were also not fully blinded as they may also have been able to spot materials that remained in villages at follow up. However, we went to great lengths to make sure that mothers did not connect the presence of observers with the intervention. Observers were different people from those that had carried out the intervention and mothers were told that the observers monitoring their daily routine. It is possible that mothers in the intervention group displayed more reactivity and hence better behaviour when observers were present, thus inflating the results.

Whilst the study targeted mothers with a child aged 6 months and over because this is the age at which complementary foods are supposed to be introduced according to international guideline. However, learnt that mothers generally introduce food other than breast milk at a much earlier age. This is likely to be an important risk factor for infection and malnutrition. Whilst promotional efforts should clearly focus on exclusive breast feeding before six months of age, if complementary foods are to be introduced at all, they should be prepared safely. This is something of a dilemma for policy and a topic requires further exploration.
Though the results of this study suggest that the overall intervention package achieved improved food hygiene behaviour, it is hard to draw conclusions concerning which tools had the greatest impact and which motivational drivers or settings modifications had the greatest effect, or whether the results were due to a combined effect. More light should be shed on this from the analysis of data from the process evaluation (yet to be carried out and not reported here). The study was conducted in the rainy season which is the peak period of diarrhoea incidence and also cultivation season for farmer. Besides the cultivation season, the level of community participation in the intervention was high. Avoiding cultivation seasons could have been offered better participation. The diarrhoeal outcomes of the study were not reported due to limited sample size. The cost effectiveness analysis of the intervention was not determined so we can’t conclude whether improving food hygiene is cost effective intervention. Any future studies on food hygiene should include diarrhoeal outcomes and ‘cost benefit’ analysis.

ACKNOWLEDGEMENT:
This study was funded by the UK Department for International Development (DFID), through the SHARE Consortium and co-funded by WaterAid. The authors would like to thank all the mothers, social leaders, teachers, female food hygiene motivators (FHM), research assistants, project coordination committee members, local health institutions and VDCs staffs for their involvement in this study. We thank the creative and production team members. We are grateful for the support of the MoHP/CHD in Nepal for their non-funding collaboration and cooperation for the study. We would like to thank Miss Yael Velleman and colleagues from LSHTM, SHARE consortium, WaterAid.
REFERENCES:


Eight eligible clusters

**VDC - A**
(9 clusters)

- Four clusters
- Inclusion and exclusion criteria applied

**VDC - B**
(9 clusters)

- Four clusters

**Recruitment:** 30HHs from each cluster (total 239HHs)
**Baseline Socio-demographic information** survey in all HHs

**Baseline outcomes measurement** (total 239HHs)
(structured observation of behaviours, microbial sampling, diarrhoea (self report))

**Cluster randomisation**

- 4 intervention clusters (120HHs)
- 4 control clusters (119HHs)

Three months food hygiene intervention

No intervention

**Follow-up measurement (45 days after)**
(120HHs in intervention and 117 in control)
(behaviours observations, microbes, diarrhoea)

Figure 1: Flow diagram of the trial

0. Thorough cooking

1. Cleanliness of serving utensils using soap/ash

2. Handwashing with soap before feeding (mother) and before eating (child)

3. Proper storage of cooked food

4. Thorough re-heating of leftover / stored food (temp ≥ 70°C)

5. Water/milk treatment (boiling)

Key food hygiene behaviours

Figure 2: Five key prioritized food hygiene behaviours (from 1-5)
Figure 3: Images from the campaign materials and events
<table>
<thead>
<tr>
<th>Events / visits</th>
<th>Purpose</th>
<th>Key content</th>
</tr>
</thead>
<tbody>
<tr>
<td>First community event (3hrs)</td>
<td>Raise awareness of, generate interest in, and elicit commitment to the campaign and the five food hygiene behaviours.</td>
<td>Distributed invitation card a day before the event. Programme ritual (put-up back-drop banner, nail cutting, hand washing with soap, putting on programme badge etc...) initiated. Programme jingle introduced. Campaign objectives described by social leader. Situation contextualised via situational analysis – story, flex with pictures, video clip (disgust motive exploited). Five food hygiene behaviours and their benefits presented. ‘Ideal mother’ introduced as a source of inspiration. Public commitment oath taken, and certificates distributed. Public rally chanting - “safe food, healthy child, we want ideal mother”.</td>
</tr>
<tr>
<td>First households visits (3hrs)</td>
<td>Remind mothers of public commitment; change settings reinforcing the desired behaviours (particularly kitchen cleanliness)</td>
<td>Programme jingle installed on mothers' phones. Kitchen compared with 'ideal kitchen' – using clean kitchen illustration. Kitchen demarcated with ribbons and flags reminding mothers of the food hygiene behaviours. Danglers placed at eye-level (round illustration of all behaviours, ideal mother board, dhungro – a branded fire blowing instrument). Importance of food hygiene behaviours refreshed via a brief talk using a 3D flip chart. Three month work plan formulated to ensure each mother meets the public commitment.</td>
</tr>
<tr>
<td>First group event (3hrs)</td>
<td>Reinforce programme ritual; establish group norms/habits for all behaviours; and generate interest in having clean kitchens.</td>
<td>Programme ritual carried out. Mothers' experiences of changes in their kitchens shared. Group norms elicited via cooking demonstration. Benefits of five food hygiene behaviours reiterated via visual aids (3M PetriFilm, Glo-germ lotion before feeding). Bibs with the message “did you wash your hands before feeding me?” distributed as reminder / reward for HWWS. ‘Clean kitchen’ competition announced (putting clean kitchen indicators in the village).</td>
</tr>
<tr>
<td>Second household visits (2hrs)</td>
<td>Reinforce correct food hygiene behaviours with the view to these becoming habitual.</td>
<td>Mothers' preparation of food observed and corrected where necessary. Importance of five food hygiene behaviours reiterated (used 3M PetriFilm, glo-germs, bib, plastic bucket for handwashing, kettle for boiling water). Mothers reminded about ‘clean kitchen’ competition.</td>
</tr>
<tr>
<td>Second group event (3hrs, 15min)</td>
<td>Increase mothers' confidence; link food hygiene behaviours with affiliation, nurture and status; generate interest in becoming an 'ideal mother'.</td>
<td>Programme ritual carried out. Obstacles faced by mothers shared and strategies for overcoming these discussed. 'Child Life Game' played – the future that mothers want for their children discussed and linked to the five food hygiene behaviours (nurture motive). Puzzle game played to encourage kitchen cues (social respect motive). Folk song composed by mothers conveying key food hygiene messages - affiliation elicited. 'Ideal mother' competition announced. Behaviour reminder ‘fan’ reflecting five behaviours and ideal mother sticker distributed.</td>
</tr>
<tr>
<td>Third household visits (2hrs)</td>
<td>Establish reheating and boiling as social norms; ensure a conducive family environment exists to practised behaviours</td>
<td>Mothers' food reheating practices observed and corrected where necessary (noting re-heated temperature, motivated to use appropriate vessel to re-heat food and kettle to boil water). Family meeting held to promote food hygiene behaviours (using 3D flip chart). Mothers reminded about ‘clean kitchen, and ‘ideal mother’ competitions (visual cues). Un-identical visits performed (by field staff and coordination committee)</td>
</tr>
<tr>
<td>Third group event (3hrs)</td>
<td>Show that implementing the five food hygiene behaviours will avoid disgust and social exclusion and will</td>
<td>Programme ritual carried out. Mothers participated in disgust exercises (glo-germs used in food, plate, bowl, glass, spoon) and games (hot potato game using disgusting and safe pictures to demonstrate social inclusion and exclusion). 'Safe food hygiene zone' competition announced. 'Clean kitchen' competition winner announced and publically commended,</td>
</tr>
<tr>
<td>Fourth household visits (2hrs)</td>
<td>Increase social prestige and happiness. Participants and guests visited winner’s house to encourage and share learning.</td>
<td></td>
</tr>
<tr>
<td>-------------------------------</td>
<td>---------------------------------------------------------------------------------------------------------</td>
<td></td>
</tr>
<tr>
<td>Fourth group event (2hrs, 30min)</td>
<td>Create peer pressure, build confidence and reduce observer bias in observation of mothers’ 5 behaviours. Peer-review (watch-dog) exercise carried out (element of secrecy entailed) by peer mother. Observer mother reported back practices. Mothers reminded about ‘ideal mother’ and ‘safe food hygiene zone’ competitions. Mothers three month work plans reviewed. Un-identical visits performed (by field staffs and coordination committee).</td>
<td></td>
</tr>
<tr>
<td>Fifth household visits (1hr)</td>
<td>Reiterate that implementing the five food hygiene behaviours will increase social prestige and status; encourage men to participate. Programme ritual carried out. Advice provided by mothers to fictional mother (Dhukhimaya) experiencing social, environmental and attitudinal barriers to adopting food hygiene behaviours. A drama (family member role play) showed how to become an ideal mother and tacking social, attitudinal and physical barrier. ‘Ideal mother’ competition winners announced and publically commended (ideal mother photo placed in the junction of the village), thereby conferring prestige. Men involved in the event and celebration.</td>
<td></td>
</tr>
<tr>
<td>Second community event (4hrs)</td>
<td>Reinforce food hygiene behaviours; mothers self-evaluate their food hygiene behaviours. Mothers’ work plans reviewed. Mothers’ food hygiene behaviours observed (ongoing progress). Mothers’ performance self-evaluated publically. ‘Safe food hygiene zone’ indicators reinforced. Programme ritual carried out. Response received from Dhukhimaya linking food hygiene behaviours to child health and social status. Encouraging social norms by re-performing folk song etc. Mothers volunteer (those who had non-formal education) to continue monitoring community’s food hygiene behaviours. Mothers publically re-pledge their commitment to sustainable food hygiene behaviour change (appreciation certificate distributed). Experiences of stakeholders heard. Remarks from social leaders, guests, representatives link food hygiene as to social respect. ‘Safe food hygiene zones’ declared and bill boards erected at each entry point of the cluster. Group photo session performed. Community rally chanting “we want ideal mother, ideal mother hi-hi, diarrhoea by-bye) and using local music and programme song. Intervention formally closed.</td>
<td></td>
</tr>
<tr>
<td>Sixth household visits (1hr)</td>
<td>Entrench food hygiene behaviours into mothers’ daily routines and identify any remaining barriers to these practices; ensure sustainability. Sustainability work plans formulated by mothers. Ease of implementation of food hygiene behaviours analysed by participants (pile shorting exercise using illustrations) and feedback provided. Sustained behaviour change pledged by entire families. Household visits formally end.</td>
<td></td>
</tr>
</tbody>
</table>

For more details about programme activities components, follow blog: [http://www.sharereresearch.org/NewsAndEvents/Detail/om_blog_food_hygiene_study](http://www.sharereresearch.org/NewsAndEvents/Detail/om_blog_food_hygiene_study)
Table 2: Social and demographic characteristics of the study population at baseline

<table>
<thead>
<tr>
<th>Variable</th>
<th>Control (n=119HH)</th>
<th>Intervention (n=120HH)</th>
<th>P-value*</th>
</tr>
</thead>
<tbody>
<tr>
<td>Village/cluster HHs size (mean, range)</td>
<td>95 (83-112)</td>
<td>99 (75-141)</td>
<td></td>
</tr>
<tr>
<td>Number of clusters</td>
<td>4 (100%)</td>
<td>4 (100%)</td>
<td></td>
</tr>
<tr>
<td>Selected HHs per cluster (mean, range)</td>
<td>30 (29-30)</td>
<td>30 (30-30)</td>
<td></td>
</tr>
<tr>
<td>Family size (mean, SD)</td>
<td>5.8 (2.3)</td>
<td>5.9 (2.1)</td>
<td></td>
</tr>
<tr>
<td>Mothers' age (mean, range)</td>
<td>27 (18-50)</td>
<td>27 (19-43)</td>
<td></td>
</tr>
<tr>
<td>Number of children (6-59 months)</td>
<td>143 (100%)</td>
<td>150 (100%)</td>
<td></td>
</tr>
<tr>
<td><strong>Religion:</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hinduism</td>
<td>48 (40%)</td>
<td>59 (49%)</td>
<td>0.553</td>
</tr>
<tr>
<td>Buddhism</td>
<td>71 (60%)</td>
<td>59 (49%)</td>
<td></td>
</tr>
<tr>
<td>Others</td>
<td>0 (0%)</td>
<td>2 (2%)</td>
<td></td>
</tr>
<tr>
<td><strong>Education level of mothers:</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>None or informal</td>
<td>62 (52%)</td>
<td>58 (48%)</td>
<td></td>
</tr>
<tr>
<td>Primary (up to 5th grade)</td>
<td>23 (19%)</td>
<td>27 (23%)</td>
<td>0.816</td>
</tr>
<tr>
<td>Secondary (up to 10th grade)</td>
<td>26 (22%)</td>
<td>24 (20%)</td>
<td></td>
</tr>
<tr>
<td>Higher secondary or university</td>
<td>8 (7%)</td>
<td>11 (9%)</td>
<td></td>
</tr>
<tr>
<td><strong>Caste/ethnicity of mothers:</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Brahmin/Chhetri/Thakuri</td>
<td>34 (29%)</td>
<td>46 (38%)</td>
<td>0.743</td>
</tr>
<tr>
<td>Hill Aadiwaasi / Janajaati</td>
<td>76 (64%)</td>
<td>64 (53%)</td>
<td></td>
</tr>
<tr>
<td>Hill dalit</td>
<td>9 (8%)</td>
<td>10 (8%)</td>
<td></td>
</tr>
<tr>
<td><strong>Monthly HHs income in NRs:</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>&lt;10,000 NRs</td>
<td>70 (59%)</td>
<td>63 (52%)</td>
<td></td>
</tr>
<tr>
<td>≥10,000 to &lt;20,000 NRs</td>
<td>30 (25%)</td>
<td>37 (31%)</td>
<td>0.622</td>
</tr>
<tr>
<td>≥20,000 NRs</td>
<td>19 (16%)</td>
<td>20 (17%)</td>
<td></td>
</tr>
<tr>
<td><strong>Types of cooking fuel:</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Firewood</td>
<td>111 (93%)</td>
<td>104 (87%)</td>
<td>0.421</td>
</tr>
<tr>
<td>Gas cylinder</td>
<td>3 (3%)</td>
<td>3 (3%)</td>
<td></td>
</tr>
<tr>
<td>Bio-gas</td>
<td>5 (4%)</td>
<td>13 (11%)</td>
<td></td>
</tr>
<tr>
<td><strong>Main water source for drinking:</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Piped water to tap in yard, plot</td>
<td>57 (48%)</td>
<td>62 (52%)</td>
<td>0.810</td>
</tr>
<tr>
<td>Surface Water</td>
<td>62 (52%)</td>
<td>58 (48%)</td>
<td></td>
</tr>
<tr>
<td>Toilet / latrine at households</td>
<td>64 (54%)</td>
<td>60 (50%)</td>
<td>0.703</td>
</tr>
<tr>
<td>Soap observed at HHs</td>
<td>100 (84%)</td>
<td>96 (80%)</td>
<td>0.519</td>
</tr>
<tr>
<td>Refrigerator at households</td>
<td>1 (1%)</td>
<td>1 (1%)</td>
<td>0.995</td>
</tr>
</tbody>
</table>

* p-value from Chi2 test after clustering (cluster level analysis)
Table 3: Reach of the intervention - post intervention measurement

<table>
<thead>
<tr>
<th>Variable</th>
<th>Control (n=117 HH)</th>
<th>Intervention (n=120 HH)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Heard of food hygiene intervention?</td>
<td>1%</td>
<td>100%</td>
</tr>
<tr>
<td>Participated in food hygiene campaign?</td>
<td>0%</td>
<td>100%</td>
</tr>
<tr>
<td>Participated in &gt;10 events/HH visits (N=12)</td>
<td>0%</td>
<td>90%</td>
</tr>
<tr>
<td><strong>Exposure by intervention cluster in &gt;10 events/HH visits</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cluster 1</td>
<td>-</td>
<td>97%</td>
</tr>
<tr>
<td>Cluster 2</td>
<td>-</td>
<td>90%</td>
</tr>
<tr>
<td>Cluster 7</td>
<td>-</td>
<td>90%</td>
</tr>
<tr>
<td>Cluster 8</td>
<td>-</td>
<td>83%</td>
</tr>
<tr>
<td><strong>Participated in competitions?</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Clean kitchen competition</td>
<td>0%</td>
<td>100%</td>
</tr>
<tr>
<td>Ideal mother competition</td>
<td>0%</td>
<td>100%</td>
</tr>
<tr>
<td>Safe food hygiene zone</td>
<td>0%</td>
<td>99%</td>
</tr>
<tr>
<td><strong>Made public commitment to practice behaviours?</strong></td>
<td>0%</td>
<td>95%</td>
</tr>
<tr>
<td><strong>Made public commitment to sustain behaviours?</strong></td>
<td>0%</td>
<td>93%</td>
</tr>
<tr>
<td><strong>Reported that the ideal mother should practice following behaviours?</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cleanliness of serving utensils</td>
<td>1%</td>
<td>100%</td>
</tr>
<tr>
<td>HWWS before feeding and eating</td>
<td>1%</td>
<td>100%</td>
</tr>
<tr>
<td>Proper storage of leftover food</td>
<td>1%</td>
<td>100%</td>
</tr>
<tr>
<td>Thoroughly re-heat leftover/stored food</td>
<td>1%</td>
<td>100%</td>
</tr>
<tr>
<td>Treat water and boil milk before serving</td>
<td>1%</td>
<td>100%</td>
</tr>
<tr>
<td><strong>Reported believes on change in social norms over time in village (after 3 months intervention)</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Is cleaning serving utensils just before feeding common?</td>
<td>2%</td>
<td>91%</td>
</tr>
<tr>
<td>Is HWWS before feeding child common?</td>
<td>8%</td>
<td>97%</td>
</tr>
<tr>
<td>Is storing food in container with a lid common?</td>
<td>21%</td>
<td>98%</td>
</tr>
<tr>
<td>Is re-heating food before eating common?</td>
<td>9%</td>
<td>98%</td>
</tr>
<tr>
<td>Is boiling/treating water before drinking common?</td>
<td>3%</td>
<td>85%</td>
</tr>
</tbody>
</table>
Figure 4: key food hygiene behaviours before and after intervention, by study group
(Mean proportion of all key food hygiene behaviours (composite performance - cluster level analysis): i) serving utensils are washed using soap/ash just before putting child food, ii) mother washed her both hands with soap just before feeding child and child’s both hands are washed before eating food, iii) stored all cooked/leftover food in container/s with a tight lid and no flies / no visible dirt-dust accessing stored food, iv) stored/leftover food are re-heated before serving to child and maintained adequate temperature (70°C), v) served only treated water for their children when observed). *p-value from Wilcoxon rank-sum test

% difference = 0%

Figure 5: key food hygiene behaviours before and after intervention, by study clusters
(Proportion of mothers sustaining all key food hygiene behaviours by cluster: i) serving utensils are washed using soap/ash just before putting child food, ii) mother washed her both hands with soap just before feeding child and child’s both hands are washed before eating food, iii) stored all cooked/leftover food in container/s with a tight lid and no flies / no visible dirt-dust accessing stored food, iv) stored/leftover food are re-heated before serving to child and maintained adequate temperature (70°C), v) served only treated water for their children when observed).
Figure 6: Prevalence of key food hygiene behaviours before and after the food hygiene campaign in intervention group (n=120)
Table 4: Changes in mother's food hygiene behaviours from baseline to follow-up period (direct comparison and difference of differences)

<table>
<thead>
<tr>
<th>SN</th>
<th>Key food hygiene behaviours</th>
<th>Baseline*</th>
<th>Follow-up*</th>
<th>Effect size (difference of differences)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Intervention (n=120)</td>
<td>Control (n=119)</td>
<td>P-value</td>
<td>Intervention (n=120)</td>
</tr>
<tr>
<td>All</td>
<td>Proportion (in mean) of mothers sustaining all key food hygiene behaviours (combination of all key behaviours)</td>
<td>1% (2 SD)</td>
<td>2% (2 SD)</td>
<td>0.4047</td>
</tr>
<tr>
<td>Individual behaviours (5 key behaviours)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>Proportion of mothers cleaning child food serving utensils using soap / ash just before putting child's food</td>
<td>3% (4 SD)</td>
<td>6% (6 SD)</td>
<td>0.544</td>
</tr>
<tr>
<td>2.a</td>
<td>Proportion of mothers washing both hands with soap &amp; water before feeding child</td>
<td>5% (4 SD)</td>
<td>7% (3 SD)</td>
<td>0.457</td>
</tr>
<tr>
<td>2.b</td>
<td>Proportion of child washed both hands with soap and water before eating food</td>
<td>5% (4 SD)</td>
<td>5% (4 SD)</td>
<td>0.883</td>
</tr>
<tr>
<td>3</td>
<td>Proportion of households stored cooked/leftover food in containers with a tight-fitting lid and no flies / no visible dirt/dust in stored food</td>
<td>24% (17 SD)</td>
<td>26% (16 SD)</td>
<td>0.885</td>
</tr>
<tr>
<td>4</td>
<td>Proportion of mothers thoroughly reheating leftover/stored food and maintaining 70 or &gt;70 degree centigrade temperature</td>
<td>3% (4 SD)</td>
<td>6% (3 SD)</td>
<td>0.240</td>
</tr>
<tr>
<td>5</td>
<td>Proportion of households treating water before serving to child</td>
<td>1% (2 SD)</td>
<td>3% (2 SD)</td>
<td>0.155</td>
</tr>
</tbody>
</table>

Note:
Data collapsed to analyzed at cluster level * Mean proportion
** Wilcoxon rank-sum test (Mann-Whitney)
RESEARCH PAPER COVER SHEET

SECTION A – Student Details

<table>
<thead>
<tr>
<th>Student</th>
<th>Om Prasad Gautam</th>
</tr>
</thead>
<tbody>
<tr>
<td>Principal Supervisor</td>
<td>Dr Val Curtis</td>
</tr>
<tr>
<td>Thesis Title</td>
<td>Hygiene behaviour reduces faecal contamination of child food in Nepal</td>
</tr>
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SECTION B – Paper already published

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<tbody>
<tr>
<td>When was the work published?</td>
<td></td>
</tr>
<tr>
<td>If the work was published prior to registration for your research degree, give rationale for its inclusion</td>
<td></td>
</tr>
<tr>
<td>Have you retained the copyright for the work?</td>
<td>Yes</td>
</tr>
<tr>
<td>Was the work subject to academic peer review</td>
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</tr>
</tbody>
</table>

SECTION C – Prepared for publication, but not yet published

<table>
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<tr>
<th>Where is the work intended to be published?</th>
<th>PLOS ONE</th>
</tr>
</thead>
<tbody>
<tr>
<td>Paper’s authors in the intended authorship order:</td>
<td>Om P. Gautam, Wolf P. Schmidt, Joe Brown, Valerie Curtis</td>
</tr>
<tr>
<td>Stages of publication</td>
<td>Not yet submitted</td>
</tr>
</tbody>
</table>

SECTION D – Multi-authored work

| Give full details of your role in the research included in the paper and in the preparation of the paper | I led the design of the study, the protocol development, the intervention design, the trial set-up and its conduct in the field, the evaluation, the analysis, and the write-up. |

Student Signature: _____________________ Date: 25 September 2015

Supervisor Signature: ____________________________ Date: 25 September 2015
Study Title:
Hygiene behaviour reduces faecal contamination of child food in Nepal

Authors:
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Key words:
Food hygiene, weaning food, behaviours, intervention, microbes, mother, children
ABSTRACT

Introduction: Few interventions have targeted food hygiene in low income settings. This study assessed the effect of an intervention to improve food hygiene behaviour on the level of microbiological contamination in commonly used child foods in rural Nepal.

Methods: A total of 120 households in each of four intervention clusters were exposed to a three-month food hygiene intervention, while in four control clusters 119 households received no intervention. The intervention targeted five food hygiene behaviours: thorough reheating of child foods, cleaning of serving utensils, handwashing with soap before feeding/eating, storing cooked food in a container with a tight-fitting lid and boiling water and milk. In 40 of each of the intervention and control households we collected a total of 248 samples of commonly used foods during baseline, and a similar number at follow up. Food samples were collected immediately after cooking, during feeding, after 5 hours of storage and immediately after reheating; water samples were collected only when the child was found to be drinking water. Total coliforms and E. coli were enumerated in all samples.

Results: At baseline, the mean counts of total coliforms in commonly used child foods during child feeding were $2.78 \log_{10}$ and $2.48 \log_{10}$ colony-forming unit (cfu)/gram in the intervention and control households respectively. The E. coli mean counts were $1.68 \log_{10}$ and $1.36 \log_{10}$ cfu/gm respectively. Post intervention, the total coliforms mean count was reduced to $0.61 \log_{10}$ cfu/gm ($p<0.020$) and the E. coli mean count was reduced to non-detectable levels ($p<0.013$) in intervention arm. After adjusting for baseline contamination, the intervention reduced (as a difference of differences measure) the mean coliform count by $-2.00 \log_{10}$ cfu/gm ($p=0.020$) and the E. coli count by $-1.00 \log_{10}$ cfu/gm ($p=0.083$). Contamination in water was low as compared to child food during baseline in both the groups and remained low after intervention however water treatment alone was not sufficient to reduce the level of contamination in water during feeding.

Conclusion: The behavioural intervention was effective in reducing faecal indicator bacteria (mean count of total coliforms and E. coli cfu/gm) in food samples. The actual ingestion of microbes by children can only be reduced or eliminated if food hygiene interventions deal with all key behaviours. Food is likely more important than water as a route of transmission of microbial contamination and promotion of water treatment should be accompanied with safe household water storage practices.
INTRODUCTION:

Diarrhoea is the second leading disease cause of death for children under the age of five globally[1] and in Nepal[2]. Contaminated weaning foods are potentially a major contributor to diarrhoea in low-income settings[3, 4]. Unhygienic preparation and feeding of weaning foods often exposes infants in low-income settings to pathogens of faecal origin. Diarrhoea risk increases during the infant weaning period[5-7].

Previous studies examining the prevalence of faecal indicator bacteria (FIB) in weaning foods have reported high levels: an estimated 16% of weaning foods fed to village children in Zimbabwe[8], 41% in Bangladesh[9], and 44% in Egypt[10] were contaminated with Escherichia coli (E. coli). In Liberia, 68% of tested adult and weaning foods were contaminated with coliforms[11]. Pathogenic strains of E. coli are a common cause of moderate to severe diarrhoea [12] accounting for up to 25% of diarrhoea episodes in low-income settings[13]. The ingestion of faecal bacteria may cause environmental enteric dysfunction, exacerbating malnutrition and resulting in poor growth in early life[14] and the weaning period (6-23 months ) is the developmental age most critical for stunting[15]. Previous studies have estimated that the level of contamination in foods can be higher than in drinking water in low-income settings[16].

The home is an important location for acquiring food-borne disease[17]. Studies to date have identified risk factors for food contamination such as hot climate[16], poor storage facilities, environmental faecal contamination[18], cooking fuel scarcity[3], time gaps between meal preparation and feeding[19], and washing utensils in contaminated water[20]. Other likely risk factors include environmental conditions such as lack of sanitation, animal presence in the kitchen, lack of refrigeration, poor storage (flies accessing food), hard-to-clean surfaces, and lack of running water and cleaning products, and poor knowledge concerning food contamination.

A study in rural Kenya showed that 44% of dishes were hygienically unsafe[21]; and water used to prepare weaning food and the feeding bowl was a major source of faecal contamination in the Gambia [22]. Personal hygiene practices (handwashing, water treatment) of mothers were significantly related to a high level of bacterial contamination of drinking water and weaning foods in Thailand [23]. The identification of behaviours
responsible for ingestion of microbes in child food in low-income settings and improving those behaviours through scalable interventions is therefore vital.

Interventions targeting foodborne transmission as a key pathway have rarely been tested [24], despite their probable importance. Food makes an ideal vector for the transmission of enteric pathogens, providing a fertile medium for persistence and potentially proliferation [25] of diarrheagenic bacteria. Once contamination occurs, the level of microbes in food can grow quickly if food is stored at ambient temperatures. Temperatures of 20-40°C are optimal for enteric bacterial growth in food, with 37°C (body temperature) being optimal; temperatures below 6 or higher than 60°C inhibit growth[26, 27], though microbes have different tolerance for changing environmental conditions and conditions for growth. Studies have found increases in coliforms and Enterobacteriaceae counts in food stored for four hours or more[28]. Counts of bacteria including E. coli and S. aureus increased from 10⁴ to above 10⁸ after 24 hours at 37°C[4, 11, 29, 30], and counts of Shigella flexneri increased 2-3logs within 6 hours[31]. Such conditions are likely in low-income settings, as food is often cooked once a day and then re-heated, often to inadequate temperatures.

The Hazard Analysis and Critical Control Points (HACCP)[32] process has been used mostly in economically rich countries to evaluate food safety. It has been used to assess the level of microbial contaminations in food and factors associated with this have been assessed by many studies [5, 23, 33, 34], and the framework holds promise for low-income countries as well[35] where controls could be applied to prevent and reduce contamination causing disease [30, 33, 36, 37]. Few studies built on the HAACP process to design and test small scale hygiene programmes in domestic settings that proved to be effective in reducing the contamination of weaning food [37, 38], despite the promise of such strategies to lower exposure to disease-causing microbes among children.

The food hygiene intervention was designed on the basis of detailed formative research into the behaviours and the determinants of these behaviours. The intervention package was implemented in intervention clusters over a period of three months focusing adversely practiced five key behaviours. In this paper we report the effect of the intervention on microbiological contamination in foods commonly fed to children aged 6-59 months.
METHODS:

Study site:
The study was conducted in eight clusters which were randomly selected from two rural hill administrative units of Kavre District, Nepal, between October 2012 and December 2013. The area is rural and hilly (2,195 m elevation) and the mainly subsistence farming population are from mixed ethnic backgrounds with low sanitation coverage and high prevalence of diarrhoeal disease.

Selection of participants and randomization:
Figure 1 shows the flow diagram of the trial. A total of 239 households with a child aged 6-59 months from eight clusters were recruited (29-30 from each cluster). After collecting baseline information, the clusters were randomized into intervention and control arms. Four intervention clusters with 120 households received the food hygiene intervention over three months, while the four control clusters with 119 households received no intervention. Microbial contamination in commonly-used child food and water was assessed in a random sample of 10 households from each cluster (40 intervention and 40 control households in total) at baseline and from the same households after completion of the intervention.

Selection of food for sample collection:
Foods commonly fed to children were identified during the formative research and baseline assessment (figure 2). There were no particular foods prepared especially for children aged 6-59 months in this setting. Instead, adult foods were often softened by mashing, or by adding vegetable soup or milk. Foods served varied seasonally, depending on their availability. Among the 293 children in 239 households at baseline, 86% received the same solid foods as consumed by the rest of the family. Of 102 children in the weaning age group (6-24months), 70% received the same solid food as the rest of the family, 21% received food that had been softened, and 9% received liquid food. The commonly-used solid foods were bhat (boiled rice), dhido (maize, wheat or millet porridge), roti (maize- or wheat-flour bread), served with dal (stewed pulses), vegetables (green or dried), and sometimes with milk. Occasionally used semi-solid complementary foods were lito (roasted rice, maize or millet flour cooked in oil/ghee with sugar and water), jaulo (soup made with soaked rice fried in oil/ghee with fenugreek, water, salt and turmeric powder), khichari (soaked rice cooked with pulses, turmeric powder and vegetables), and rice pudding (rice cooked in milk with sugar). Solid or semi-solid foods were mostly offered with a drink of water (often untreated) or unpasteurised
buffalo or cow milk. A few households also served locally brewed Jad (homemade alcoholic brew made from fermented rice, wheat, maize or spent millet grain) to their children. To ensure consistent sampling for microbial assessment across the selected households, we sampled the most regularly served foods: rice or dhido along with dal or vegetables (whichever was being cooked on the day the sample was collected) from all households. In addition, water and milk served to children were also chosen for microbial assessment. Since milk was not served in all households, we excluded milk from our analysis.

**Prioritised behaviours and pathways:**
Figure 3 shows the five food hygiene behaviours that were targeted by the intervention. We also identified thorough cooking as a key control point but observed that foods were always thoroughly cooked at initial preparation. The prioritised behaviours were:

- **Behaviour 1:** Cleaning of child food serving utensils using soap/ash just before serving food
- **Behaviour 2:** Handwashing with soap by mother before feeding, and by child before eating
- **Behaviour 3:** Proper storage of cooked food in containers with a tight-fitting lid (to prevent contact with flies/dust)
- **Behaviour 4:** Thorough re-heating of leftover/stored food just before feeding child (≥70°C)
- **Behaviour 5:** Serving of treated water or boiled milk

**Summary of intervention and delivery:**
The intervention campaign was delivered by trained food hygiene motivators during May-August 2013. Based on prior formative research and theory, the intervention used motivational themes including ‘Nurture’, ‘Disgust’, ‘Social Respect/Status’ and ‘Affiliation’ and change in behavioural settings such as kitchen. The intervention was delivered through six events and six door-to-door visits per household by local trained food hygiene motivators (described elsewhere). The intervention used innovative and motivational approaches and avoided educational messaging. These included storytelling, video-clips, kitchen makeovers, danglers placement at eye level, baby bibs, games, a ringtone with the campaign song for mothers, cookery demonstrations, visualisations of hand contamination (‘glo-germ demonstration’), competitions (‘clean kitchen’, ‘ideal mother’, ‘safe food hygiene zone’), public commitments, social/public pledging and rallies. The campaign implementation details can be accessed online at [http://www.shareresearch.org/NewsAndEvents/Detail/om_blog_food_hygiene_study](http://www.shareresearch.org/NewsAndEvents/Detail/om_blog_food_hygiene_study) and
19 minute YouTube video documentary detailing the food hygiene intervention trial can be accessed at (https://www.youtube.com/watch?v=gPj6IyN0ZCU).

Microbial assessment
Total coliforms (TC) and Escherichia Coli (E. coli) [39, 40], were detected and quantified for each food sample during baseline and follow-up. 3M™ Petrifilm™ [40] E. Coli and Coliform Count Plates[41] media were employed according to manufacturers’ instructions (following AOAC, the international all foods method 991.14, as recommended by 3M PetriFilm). Membrane filtration followed by incubation at 37°C (+/- 1°C) on HiCrome Coliform Agar w/SLS media (M1300–500G) was used for water, according to standard methods. The TC and E. coli counts were expressed as colony forming units/gram (cfu/gm) for food, and cfu/100ml for water. Samples were collected by trained lab technicians unconnected with the intervention team, who arrived separately and unannounced. Another set of independent microbiologists and lab technicians processed samples and interpreted the results at a certified laboratory in Kathmandu. Both teams were blinded with respect to sample origin (intervention or control). Altogether, 248 food samples and 80 water samples were collected during baseline, and the same amount from the same households after the intervention. Each household received a small sum to recompense the value of the food collected.

Food and water sample collection
Food samples were collected at four different stages of preparation, storage and feeding: (i) first stage - directly from cooking vessels 5 to 15 minutes after cooking; (ii) second stage - from the mother or the child’s hand during feeding; (iii) third stage - leftover food taken directly from containers after an average of five hours’ storage; (iv) fourth stage - the same leftover stored food immediately after reheating, directly from the re-heating container. Food temperatures were noted in each time samples were collected because the level of contamination is dependent on the food temperature. The water samples were collected during feeding directly from serving utensils.

Sample size:
Altogether 248 food samples and 80 water samples were collected during baseline and the same during follow-up. The Sample size was calculated so as to be able to detect a mean difference in the level of total coliforms in child foods (during feeding) of 1.0 log_{10} cfu/gram between control and intervention households (TC 2.30 log_{10} mean cfu/gram in control
households, and 1.40 log$_{10}$ mean cfu/gram in intervention households), with 90% power using 95% confidence interval (p<0.05).

**Sampling procedure:**
From each household, in each stage of sampling, 50 grams of food was collected into 60ml sterile and labelled plastic containers with tight-fitting lids, using a sterile wooden spatula when needed. The temperature was recorded just before collection using a sterile thermometer. Similarly, 250ml of water were collected using sterile and labelled containers from each household. All samples were transferred to an insulated cooler box containing ice immediately after collection, and transported to the laboratory within four hours of collection, maintaining a temperature below 6°C. The detailed adopted laboratory procedures to process, count and interpret total coliforms and *E. coli* load as colony forming units (cfu)/gram of food and cfu/100ml of water is available.

**Data analysis:**
We used cluster level analysis since the intervention was allocated by cluster[42]. Given the number of clusters (four each in the intervention and control arms), we used a non-parametric test, the two-sample Wilcoxon’s rank sum test (Mann-Whitney), which does not rely on the assumption of normality and that is resistant to outliers[42], for estimating statistical support [43]. The TC and *E. coli* colony counts were log-transformed (log$_{10}$), and differences in means by group were calculated. The effect of the intervention was therefore calculated as the summary means of the log-transformed microbe counts (TC and *E. coli* cfu/gram in food, cfu/100ml in water) compared between the control and intervention groups during baseline and follow-up in each stages. The size of the effect of the intervention was measured using difference-of-differences analysis i.e. (follow-up – baseline)$_{intervention}$ minus (follow-up – baseline)$_{control}$. The original laboratory data were entered into Excel and then transferred into STATA (version 12) for analysis.

**Consent and blinding:**
Informed consent was obtained from all participants beforehand. To preserve anonymity, findings were presented without ascribing personal description. Both teams (sample collection and lab processing) were blinded with respect to intervention to allocation. Ethical approval for the study was granted by the ethics committees of the London School of Hygiene and Tropical Medicine, UK, and the Nepal Health Research Council.
RESULTS:

Social, demographic and economic characteristics:
The characteristics of the 80 households from which food and water samples were collected are shown in Table 2. Intervention and control cluster households had broadly similar social, demographic and economic characteristics. The maternal age ranged from 19 to 50 years with a mean of 27 years. Around 50% of mothers in both group either had none or informal education. Over 50% of mothers in both groups belonged to the hill aadiwassi/janajaati group (part of the historically deprived Tamang ethnic group). More than 50% of households in both groups earned less than 10,000 NRs (~100US$/month) per month. The majority used firewood as their main cooking fuel. Around 50% of households untreated piped water, and 50% used surface water for drinking. Collected water was usually stored for a long time in large, mostly uncovered containers (gagri). Around 50% of the population in both groups defecated in the open. Soap was observed in the majority of households. None of the households had a refrigerator.

Food hygiene practices and effect of the intervention:
The effect of the intervention on mothers’ food hygiene behaviours is reported elsewhere. Briefly; the five key food hygiene behaviours were observed during baseline and follow-up. The composite performance of all behaviours was rare at baseline in both the intervention (1%) and control (2%) groups. Following the three-month intervention, the level of performance of all observed behaviours rose to 43% in the intervention group and remained unchanged (2%) in the control group. The intervention improved all five key behaviours.

Effects of the intervention on the reduction of microbial contamination (total coliforms and E. coli):
Table 3 shows that commonly-used child foods from both the intervention and control households were heavily contaminated with total coliforms (TC) and E. coli at baseline, with no significant differences between groups in all stages. During follow-up, the mean TC and E. coli count in the food samples was significantly reduced in the intervention group in all stages. Levels of contamination remained almost similar to baseline in the control group. Figures 4 to 7 shows the proportion of food samples that had non-detectable level of TC and E. coli (<1cfu/gm food) in both the groups during baseline and follow-up. There was however only a small reduction in the level of contamination in water samples collected at the point-of-use
directly from feeding cup/bowl during follow-up (Table 3 and Figure 8). The effect of the intervention on the level of contamination in child food at different stages presented below.

**Contamination of child food immediately after cooking:**
Table 3 shows that, the TC mean counts in food samples collected immediately after cooking at baseline were 1.14 log$\text{_{10}}$ cfu/gm and 0.80 log$\text{_{10}}$ cfu/gm (p=0.386) and the *E. coli* mean counts in the same samples were 0.52 log$\text{_{10}}$ and 0.27 log$\text{_{10}}$ cfu/gm (p=0.554) in the intervention and control groups respectively. During follow-up the mean TC count reduced significantly and the *E. coli* mean count reduced to non-detectable level in the intervention group. After adjusting for the baseline contamination in both groups, the intervention (as a difference of differences measure) reduced the mean TC count by -0.98 log$\text{_{10}}$ cfu/gm (p=0.148) and *E. coli* mean count by -0.53 log$\text{_{10}}$ cfu/gm (p=0.083). The proportion of food samples that had non-detectable level of TC and *E. coli* (<1cfu/gm food) increased at follow-up in the intervention group (Figures 4 to 7). Table 4 shows the mean temperature of the collected food samples during this stage in both the groups.

**Contamination of child food during feeding:**
At baseline, the TC mean counts were 2.78 log$\text{_{10}}$ and 2.48 log$\text{_{10}}$ cfu/gm, and the *E. coli* mean counts were 1.68 log$\text{_{10}}$ and 1.36 log$\text{_{10}}$ cfu/gm in the intervention and control groups respectively. During follow-up, the TC mean count dropped significantly and the *E. coli* mean count reduced to non-detectable level in intervention group as compared to control. After adjusting for the baseline contamination in both groups, the intervention reduced the mean TC count by -2.00 log$\text{_{10}}$ cfu/gm (p=0.020) and *E. coli* by -1.00 log$\text{_{10}}$ cfu/gm (p=0.083). The proportion of food samples that had non-detectable level of TC and *E. coli* (<1cfu/gm food) increased at follow-up in the intervention group (Figures 4 to 7). The mean and range of temperature of the collected food samples during child feeding was almost the same in both groups during baseline and follow-up as shown in Table 4.

**Contamination of child food after 5hrs storage:**
Samples collected in the third stage (after five hours of storage) during baseline were heavily contaminated in both groups. The TC mean counts were 3.39 log$\text{_{10}}$ and 2.46 log$\text{_{10}}$ cfu/gm, and the *E. coli* mean counts were 2.26 log$\text{_{10}}$ and 1.44 log$\text{_{10}}$ cfu/gm in the intervention and control groups respectively with no statistical differences between the groups. Post intervention, the TC mean count and the *E. coli* mean count was significantly reduced in intervention group.
After adjusting for the baseline contamination, the intervention reduced the mean TC count by $-2.14 \log_{10} \text{cfu/gm (p=0.020)}$ and $E. coli$ mean count by $-1.33 \log_{10} \text{cfu/gm (p=0.248)}$. Figures 4 to 7 shows the proportion of food samples that had non-detectable level of TC and $E. coli$ (<1cfu/gm food) at baseline and follow-up. As shown in Table 4, there was little variation in the mean and range of temperature of the collected food samples after five hours of storage in both groups.

**Contamination of child food immediately after re-heating:**
The TC mean counts in food samples collected immediately after re-heating were $3.55 \log_{10}$ and $3.02 \log_{10} \text{cfu/gm}$, and the $E. coli$ mean counts were $2.25 \log_{10}$ and $1.76 \log_{10} \text{cfu/gm}$ in the intervention and control groups respectively at baseline. Post intervention, the TC mean count and the $E. coli$ mean count was significantly dropped in the intervention group. After adjusting for the baseline contamination in both groups, the intervention reduced the mean TC count by $-1.90 \log_{10} \text{cfu/gm (p=0.083)}$ and $E. coli$ mean count by $-1.11 \log_{10} \text{cfu/gm (p=0.083)}$. The proportion of food samples that had non-detectable level of TC and $E. coli$ (<1cfu/gm food) increased at follow-up in the intervention group (Figures 4 to 7). Table 4 shows the variation in the mean and range of temperature of the collected food samples immediately after re-heating during baseline in both groups. During follow-up, the mean temperature was $77^\circ \text{c (range of min 68}^\circ \text{c to max 93}^\circ \text{c)}$ in the intervention group possibly due to the effect of the intervention, whereas in control group, the mean temperature remained $32^\circ \text{c (range of min 20}^\circ \text{c to max 68}^\circ \text{c)}$.

**Contamination in water during feeding:**
The level of contamination in water was low as compared with levels of contamination in food. During baseline, there were no differences in the TC and $E. coli$ mean counts by group in water samples collected at mid-day during child feeding. At follow up, there was a significant difference in the mean count. The TC count was $2.33 \log_{10} \text{cfu/100ml (p=0.148)}$, and the $E. coli$ mean count was $1.67 \log_{10} \text{cfu/100ml water (p=0.083)}$ in the intervention group. After adjusting for the baseline contamination in both groups, the intervention (as a difference of differences measure) reduced the mean TC count by $-1.06 \log_{10} \text{cfu/100ml (p=0.148)}$, and $E. coli$ mean count by $-1.18 \log_{10} \text{cfu/100ml (p=0.043)}$. Figure 8 shows the proportion of water samples that had non-detectable level of coliforms (<1cfu/100ml water) during baseline and follow-up.
DISCUSSION:
The intervention was successful in achieving broad reductions of total coliforms and *E. coli* in commonly-used child food in the intervention group during follow-up. Our results demonstrate the public health importance of community level food hygiene intervention targeting multiple behaviours using emotional drivers of behaviour change and change in kitchen settings to effectively reduce the faecal contamination in child food. There was only weak evidence to observe the effect of intervention on microbial reduction in water sample (Table 3).

Baseline data suggested that food hygiene practices of mothers contribute to high levels of bacterial contamination in food. Young children in rural Nepal are frequently exposed to high levels of faecal bacteria through the food and water they consume daily. Our intervention was effective in reducing the presence of faecal contamination in food and in reducing FIB counts in food, at all stages (<1 log\(_{10}\) cfu/gm of food) and most importantly at the time of child feeding in the intervention group during follow-up. Our findings suggest that all five behaviours are critical to control microbes and failing to practice any may lead to an increase in microbial exposure. For example, promoting thorough cooking and re-heating can reduce microbes, but use of unwashed serving utensils may re-introduce contamination if this behaviour is not addressed in concert. Similarly, even when food is thoroughly cooked or re-heated and serving utensils have been washed, if the mother’s or child’s hands have not been washed prior to feeding or eating, further contamination may be introduced. Further, while proper storage does not eliminate microbes, it can protect from cross-contamination or ingestion of additional contamination from flies, dust, dirt, animals, hands, or via other pathways. Stored food protects from cross-contamination, but it should be re-heated thoroughly before serving to eliminate microbes that may have grown in storage. In addition, if children are given contaminated water together with safe food, all other food hygiene measures practiced may be in vain. These results suggest that interventions must be holistic and comprehensive to effect a sustained barrier against foodborne microbial contamination at the household level.

*Importance of thorough cooking to reduce microbes in food:*
The mean TC and *E. coli* counts were low in the samples collected immediately after cooking (within 5-15min) compared with other stages in both groups during baseline and follow-up, since the sufficiently high cooking temperature (>70\(^{\circ}\)C) is likely to have inactivated microbes.
Despite the exclusion of this behaviour as a priority behaviour from the intervention, the intervention was effective in reducing contamination in food immediately after cooking. This might be due to the careful links and emphasis on re-heating behaviours and encouragement of thorough cooking practices throughout the intervention period. The study confirms that thorough cooking (≥70°C temp) kills most TC and E. coli in food, and that any food hygiene intervention should include thorough cooking as one of the key behaviours.

**Importance of cleanliness of serving utensils and handwashing with soap before eating/feeding to reduce microbes in food:**

Many utensils were found to be kept on mud floors, exposed to flies, dirt and dust, unprotected from contamination via a variety of potential pathways. Similar practices have been observed in rural settings in other countries [21, 44]. Hands (both the child’s and the mother’s) are likely to carry faecal microbes, and using hands for feeding or eating without washing them with soap is likely to increase the risk of pathogen transmission and associated diseases among children. Samples collected when mothers put cooked food in serving utensils and used their hands or a spoon to mix the food (within 30min after cooking) showed that the levels of TC increased by 2 log_{10} cfu/gm and E. coli count increased by 1 log_{10} cfu/gm at this stage during baseline, suggesting a fundamental role of serving utensils and hands in the transmission of microbes in food (see Table 3). We observed that the probable means of introducing microbes into the food at this stage is either the serving utensils or the mother’s hands. The samples collected during follow-up in the intervention group during child feeding were less contaminated compared with the control group, possibly due to performance of behaviours increasing cleanliness of serving utensils and handwashing with soap.

Our earlier results suggested that the intervention was effective in improving both the cleanliness of serving utensils and handwashing with soap before feeding / eating behaviours in the intervention group during follow-up, and that this contributed to significant reduction in mean TC and E. coli counts during child feeding. Faecal contamination of hands and cleanliness of serving utensils undoubtedly a significant threat to health and significant reduction of bacteria in intervention group confirms its importance as highly effective intervention. Since samples were collected only during feeding, we could not discern whether the cleanliness of serving utensils or handwashing with soap had greater impact in reducing the level of contamination. Observational studies have suggested that handwashing before preparing food is a particularly important opportunity to prevent childhood diarrhoea[45]. Our
experience in addressing multiple food hygiene behaviours suggests that in order to secure impact in terms of reducing food contamination in low-income settings, the importance of handwashing with soap just before feeding/eating and cleanliness of serving utensils should be equally emphasised.

**Importance of proper storage of cooked / leftover food in a container with a tight-fitting lid to reduce cross-contamination:**

Cooking once or twice a day and feeding a child multiple times was a common practice. Refrigeration of cooked food to maintain appropriately low temperatures, or continuously keeping food sufficiently heated was not viable options due to lack of availability and affordability of refrigerators or other technologies that allow food continuous re-heating at low cost. Similarly, completely eliminating the use of stored food was impractical given the need for child feeding throughout the day (3-4 times) and the multiple tasks performed by primary care givers that prevent them from being able to cook several times a day. The only remaining option was storing food in a container with a tight-fitting lid and re-heating before feeding despite the fact that the temperatures of 20-40°C are optimal for bacterial growth in food and that temperatures below 6 or higher than 60°C inhibit growth[26]. At baseline, food cooked in the morning and stored for five hours was heavily contaminated with TC and _E. coli_ (Table 3). On average, there was an increment of mean TC count by 2 log₁₀ cfu/gm, and of the mean _E. coli_ count by 1.5 log₁₀ cfu/gm during baseline, within five hours of storage. During the follow-up period, contamination recorded in the intervention group was low as compared to control group within the same time frame (5 hours of storage). Improved storage practices and clean kitchen settings due to kitchen makeover potentially contributed for low level of contamination. Proper storage possibly protected food from additional contamination from flies, dust, dirt, insects, animals, and other sources however such practice was not enough to eliminate already contained bacteria. The higher levels of contamination recorded in the control group compared to those in the intervention group can be ascribed to stored food being left uncovered and the flies might have transmitted faecal contaminants / microbes into food [3] or through other exposure due to poor storage practices[46]. Our study suggests that appropriate storage prevents food from additional possible contamination but such practice is not sufficient to eliminate bacteria in food. Therefore, promotion of proper storage should go along with re-heating practices.
**Importance of thorough re-heating of stored/leftover food to destroy microbes:**

Thorough re-heating and cooking (maintaining $\geq 70^\circ C$) kills most microbes\[26\] and potentially contributes to the reduction of foodborne infections. During baseline, thorough re-heating practices were rare in both groups, and the temperature used for re-heating was below the recommended level (mean temperature $40^\circ C$). Table 3 shows that the so-called re-heated food had an even higher level of contamination during baseline; this was because food was usually re-heated briefly (without maintaining adequate temperature), re-heated in a bowl which potentially re-contaminated the food, and mixed by hand during re-heating, thereby contributing to further contamination. The intervention was effective in reducing the level of contamination in food immediately after re-heating during follow-up and there were significant differences in the level of food contamination between the intervention and control groups (Table 3). Almost 89% food samples were free (<1cfu/gm food) from TC and 96% free from *E. coli* in intervention group during follow-up. The intervention was effective in improving thorough re-heating practices (result reported elsewhere) and majority of households maintained adequate temperature (mean temperature $77^\circ C$) while re-heating food during follow-up in the intervention group potentially destroyed the count of TC and *E. coli* in food. Our findings is consistent with results of other studies using HACCP, which were also effective in reducing faecal contamination when re-heating was applied adequately [30, 36, 37]. We strongly believe that any food hygiene intervention should include thorough re-heating practices as one of the key behaviours that potentially contribute to the reduction of microbes in food, diarrhoeal diseases and would impact on health in low-income settings.

**Microbial contamination in water and its importance in food hygiene:**

Serving water together with or just after feeding is common in many countries, as well as in the study area. There is abundant evidence to show that water treatment can reduce contamination and potentially contribute to diarrhoeal disease reduction. Studies has found that stored water are more contaminated than water at the source\[47\], and that treating household stored water had a more significant effect on reducing diarrhoea than source treatment\[48\]. Stored water is served without treatment (table 3). The water samples which were tested when mother offered for drinking during mid-day are contaminated with TC and *E. coli* at the level of 2.45 log10 cfu/100ml and 1.81 log10 cfu/100ml in intervention group at baseline respectively. The level of contamination was similar in the control group. Our intervention focused on improving water treatment practices (boiling), and the intervention was effective in improving water treatment behaviours significantly in the intervention group. During follow-up little reduction was observed in the level of contamination. Although the
proportion of samples that had no level of coliforms or *E. coli* improved, there was no significant difference in contamination levels in water between the groups. This can be explained by the fact that the intervention focused on motivating mothers to adopt water treatment practices. In reality, once treatment has been done by boiling water during the morning meal time, the treated water was then stored in a bottle or jug that are not commonly washed or covered. This leaves treated water at risk of re-contamination by hands, flies, dust or dirt, storage container [49-51]. Other explanation for none significant results could be due to relatively low sample size and large effect size anticipated for water. Based on these findings, we would suggest that the intervention on water treatment should be accompanied with safe household water storage practices and therefore a hygiene promotion package should include activities / motivations on use safe household storage together with treatment. More strikingly, the food samples were heavily contaminated then water showing that food might be the major source of faecal contamination in rural setting.

**Limitations:**
Microbial contamination of food and water was assessed as a secondary outcome of the main study. The food samples were collected at four stages, while water was only sampled once during feeding. The second food samples were taken when cooked food was put into serving utensils and mixed using hands to feed child (during feeding); uncertainty remains as to which particular behaviour - handwashing or cleanliness of serving utensils - had a greater influence on reducing contamination. It would have been worth taking swabs of utensils along with food samples at that stage. It was not possible to fully blind the study participants and data collectors due to the nature of the intervention. Although sample collectors and laboratory technicians at field and laboratory levels were blinded concerning the details of clusters and allocation to the intervention and control group, the presence of ‘outsiders’ taking samples may subject behaviours to outsider bias; we went to great lengths to de-link the sample collection from the food hygiene campaign, but the risk of bias cannot be entirely ruled out. To reduce differential reactivity, we had restricted the study to assessing outcomes after only 45 days of the intervention. Mothers were not aware of the links between the food sample collection and the food hygiene campaign. On the other hand, despite the good results achieved by this study, we may not able to draw conclusions on which particular tools have been most effective in delivering the results. The study environment was hugely contaminated with human and animal faeces. Microbial source tracking would have identified the source of contamination in these settings and assessment of pathogens using highly specific molecular techniques could help to set up a response strategy as well as foodborne diseases surveillance.
for large scale diseases prevention programmes. Outcomes were measured 45 days after completion of the three months intervention but unable to measure the sustained outcomes after six months or after a year. Despite these limitations, we believe that assessing microbial contamination as a secondary outcome in the behaviour change intervention study supports the results obtained from the behavioural (primary) outcome.

**Conclusion:**
The food hygiene intervention was shown to be effective in reducing the level of contamination (mean count of total coliforms and *E. coli* cfu/gm) in the samples of child food. We propose that changing multiple food hygiene behaviours is fundamental to achieving a reduction of microbes in commonly used child food at different key stages of cooking, feeding, storage and re-heating. This is achievable through applying a simple and motivational package implemented by community promoters with active involvement of mothers. This study confirms the importance of food hygiene promotion for the reduction of microbial contamination in food and practicing all five behaviours acts as barrier for microbe transmission. The actual ingestion of microbes by children can only be reduced or eliminated if a food hygiene intervention deals with all key behaviours. Undermining one or more behaviours may therefore lead to negative outcomes; all five food hygiene behaviours should therefore be promoted and practiced. By looking the different pathways for food contamination and subsequent outcomes, we conclude that existing interventions to reduce diarrhoeal disease transmission and improve nutritional outcomes are not sufficient to eliminate the faecal-oral transmission of bacteria among infants and young children, as many interventions ignore the most critical pathways by which faecal pathogens can be ingested such as through food.

**ACKNOWLEDGEMENT:**
This study was funded by the UK Department for International Development (DFID), through the Sanitation and Hygiene Applied Research for Equity (SHARE) Consortium, and co-funded by WaterAid UK. The authors would like to thank all the mothers from study area in Kavre district Nepal, children, social leaders, teachers, female health motivators (FHM), research assistants, project coordination committee members, local health institutions and VDCs staff for their involvement in this study. We thank the CMDN/Intrepid Nepal Pvt. Ltd and ENPHO and laboratory technicians for their assistance. The study funder has had no role in intervention design, implementation, and measurement of outcomes or communication of results.
REFERENCES:


41. 3M-PetriFilm, *Petrifilm™ E. coli and Coliform Count Plates International Guide*. 3m PetriFilm. www.3M.com/microbiology or https://www.msu.edu/course/fsc/441/3mc&ec.html


Eight eligible clusters

Recruitment: 30HHs from each cluster (total 239HHs)
Baseline Socio-demographic information survey in all HHs

Baseline measurement (total 239HHs)
(structured observation of behaviours-all, microbial sampling (8*10=80HHs), diarrhoea [self report]-all)

Cluster randomisation

4 intervention clusters (120HHs) 4 control clusters (119HHs)
Three months food hygiene intervention No intervention

Follow-up measurement (45 days after)
(120HHs in intervention and 117 in control)
(behaviours observations-all, microbes sampling (8*10=80HHs), diarrhoea-all)

Figure 1: Flow diagram of the trial

<table>
<thead>
<tr>
<th>Types of food</th>
<th>Images of commonly used child food items</th>
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<tr>
<td>Semi-solid Food</td>
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</tr>
<tr>
<td>Liquid Food</td>
<td><img src="image3.png" alt="Liquid Food Images" /></td>
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</tbody>
</table>

Figure 2: Commonly used child food in rural Nepal (examples)
Figure 3: Behaviours to control/eliminate contamination in food (from 1-5)

1. Cleaning of child food serving utensils using soap/ash
2. Handwashing with soap by mother before feeding, and by child before eating
3. Proper storage of cooked/ leftover food in containers with a tight-lid
4. Thorough cooking/re-heating of leftover/stored food just before feeding child (temp ≥ 70°C)
5. Water/milk treatment (boiling)
Table 2: Social and demographic characteristics of the population from which food samples were collected

<table>
<thead>
<tr>
<th>Variable</th>
<th>Control (n=40HHs)</th>
<th>Intervention (n=40HHs)</th>
<th>P-value*</th>
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<td>4 (100%)</td>
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<td>Total HHs per group</td>
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<td>Mothers' age (mean, range)</td>
<td>27 (19-50)</td>
<td>28 (19-40)</td>
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</tr>
<tr>
<td><strong>Religion:</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hinduism</td>
<td>17 (43%)</td>
<td>22 (55%)</td>
<td>0.505</td>
</tr>
<tr>
<td>Buddhism</td>
<td>23 (57%)</td>
<td>18 (45%)</td>
<td></td>
</tr>
<tr>
<td><strong>Education level of mothers:</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>None or informal</td>
<td>21 (52%)</td>
<td>21 (53%)</td>
<td></td>
</tr>
<tr>
<td>Primary (up to 5th grade)</td>
<td>6 (15%)</td>
<td>9 (22%)</td>
<td>0.409</td>
</tr>
<tr>
<td>Secondary (up to 10th grade)</td>
<td>11 (28%)</td>
<td>6 (15%)</td>
<td></td>
</tr>
<tr>
<td>Higher secondary or university</td>
<td>2 (5%)</td>
<td>4 (10%)</td>
<td></td>
</tr>
<tr>
<td><strong>Caste/ethnicity of mothers:</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Brahmin/Chhetri/Thakuri</td>
<td>11 (28%)</td>
<td>15 (38%)</td>
<td>0.672</td>
</tr>
<tr>
<td>Hill Aadiwaasi / Janajaati</td>
<td>26 (65%)</td>
<td>20 (50%)</td>
<td></td>
</tr>
<tr>
<td>Hill dalit</td>
<td>3 (7%)</td>
<td>5 (12%)</td>
<td></td>
</tr>
<tr>
<td><strong>Monthly HHs income in NRs:</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>&lt;10,000 NRs</td>
<td>22 (55%)</td>
<td>23 (58%)</td>
<td></td>
</tr>
<tr>
<td>10,000 to &lt;20,000 NRs</td>
<td>11 (28%)</td>
<td>10 (25%)</td>
<td>0.956</td>
</tr>
<tr>
<td>≥20,000 NRs</td>
<td>7 (17%)</td>
<td>7 (17%)</td>
<td></td>
</tr>
<tr>
<td><strong>Types of cooking fuel:</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Firewood</td>
<td>39 (97%)</td>
<td>35 (88%)</td>
<td>0.169</td>
</tr>
<tr>
<td>Gas cylinder</td>
<td>1 (3%)</td>
<td>5 (12%)</td>
<td></td>
</tr>
<tr>
<td>Bio-gas</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Main water source for drinking:</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Piped water to tap in yard, plot</td>
<td>20 (50%)</td>
<td>20 (50%)</td>
<td>1.000</td>
</tr>
<tr>
<td>Surface Water</td>
<td>20 (50%)</td>
<td>20 (50%)</td>
<td></td>
</tr>
<tr>
<td>Toilet / latrine at households</td>
<td>20 (50%)</td>
<td>22 (55%)</td>
<td>0.746</td>
</tr>
<tr>
<td>Soap observed at HHs</td>
<td>30 (75%)</td>
<td>34 (85%)</td>
<td>0.352</td>
</tr>
<tr>
<td>Refrigerator at households</td>
<td>0 (0)</td>
<td>0 (0)</td>
<td></td>
</tr>
</tbody>
</table>

* p-value from Chi2 test after clustering (cluster level analysis)
Table 3: Microbes in commonly used child food (mean log10 cfu/gm) and water (mean log10 cfu/100ml)- before and after the food hygiene intervention (cluster level-difference of differences analysis)

<table>
<thead>
<tr>
<th>Sampling stages</th>
<th>Microbes</th>
<th>Before (baseline)</th>
<th>After (follow-up)</th>
<th>Effect size (difference of differences)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Intervention mean (SD)</td>
<td>Control mean (SD)</td>
<td>p-value*</td>
<td>Intervention mean (SD)</td>
</tr>
<tr>
<td>immediately after cooking, directly from vessels (n=56)</td>
<td>Coliforms (log10 mean cfu/gm)</td>
<td>1.14 (0.63)</td>
<td>0.80 (0.53)</td>
<td>0.386</td>
</tr>
<tr>
<td></td>
<td>E. coli (log10 mean cfu/gm)</td>
<td>0.52 (0.40)</td>
<td>0.27 (0.43)</td>
<td>0.554</td>
</tr>
<tr>
<td>during feeding, from mother’s/child’s hand (n=80)</td>
<td>Coliforms (log10 mean cfu/gm)</td>
<td>2.78 (0.21)</td>
<td>2.48 (0.77)</td>
<td>0.248</td>
</tr>
<tr>
<td></td>
<td>E. coli (log10 mean cfu/gm)</td>
<td>1.68 (0.35)</td>
<td>1.36 (0.83)</td>
<td>0.386</td>
</tr>
<tr>
<td>after 5hrs storage, directly from stored container (n=56)</td>
<td>Coliforms (log10 mean cfu/gm)</td>
<td>3.39 (0.27)</td>
<td>2.46 (0.93)</td>
<td>0.148</td>
</tr>
<tr>
<td></td>
<td>E. coli (log10 mean cfu/gm)</td>
<td>2.26 (0.23)</td>
<td>1.44 (1.22)</td>
<td>0.248</td>
</tr>
<tr>
<td>immediately after re-heating, directly from re-heating container (n=56)</td>
<td>Coliforms (log10 mean cfu/gm)</td>
<td>3.55 (0.48)</td>
<td>3.02 (0.76)</td>
<td>0.386</td>
</tr>
<tr>
<td></td>
<td>E. coli (log10 mean cfu/gm)</td>
<td>2.25 (0.26)</td>
<td>1.76 (0.93)</td>
<td>0.563</td>
</tr>
</tbody>
</table>

Microbes in ready to feed water

| Water during feeding                                      | Coliforms (log10 mean cfu/100ml) | 2.45 (0.19)       | 2.49 (0.28)       | 0.772                   | 2.33 (0.92)       | 3.43 (0.49) | 0.148 | -1.06 | 0.148 |
|                                                       | E. coli (log10 mean cfu/100ml)    | 1.81 (0.32)       | 1.74 (0.18)       | 0.772                   | 1.67 (0.74)       | 2.78 (0.52) | 0.083 | -1.18 | 0.043 |

Cluster level analysis: data collapsed by cluster to analyze at cluster level
* Wilcoxon rank-sum test
** difference of differences = {follow-up - baseline}intervention minus {follow-up - baseline}control
Table 4: The mean temperature of commonly used child food during sampling (temp in degree centigrade)*

<table>
<thead>
<tr>
<th>Group</th>
<th>Period</th>
<th>Immediately after cooking (5-15min): mean (min-max)</th>
<th>During feeding (0min): mean (min-max)</th>
<th>After 5hrs storage (±15min): mean (min-max)</th>
<th>Immediately after reheating (0-5min): mean (min-max)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Intervention</td>
<td>Baseline</td>
<td>78 (65-90)</td>
<td>33 (22-42)</td>
<td>25 (19-32)</td>
<td>41 (30-78)</td>
</tr>
<tr>
<td></td>
<td>Follow-up</td>
<td>73 (62-88)</td>
<td>39 (22-50)</td>
<td>23 (18-35)</td>
<td>77 (68-93)</td>
</tr>
<tr>
<td>Control</td>
<td>Baseline</td>
<td>72 (60-87)</td>
<td>34 (22-45)</td>
<td>26 (23-30)</td>
<td>39 (30-75)</td>
</tr>
<tr>
<td></td>
<td>Follow-up</td>
<td>76 (60-94)</td>
<td>36 (20-53)</td>
<td>22 (18-31)</td>
<td>32 (20-68)</td>
</tr>
</tbody>
</table>

* cluster level analysis
Figure 4: Total coliform (cfu/gm) in commonly used child food during baseline and follow-up in intervention group.

Figure 5: Total coliform (cfu/gm) in commonly used child food during baseline and follow-up in control group.
Figure 6: E. coli (cfu/gm) in commonly used child food during baseline and follow-up in intervention group

Figure 7: E. coli (cfu/gm) in commonly used child food during baseline and follow-up in control group
Figure 8: Total coliform and E. coli (cfu/100ml) in ready to feed water - before and after food hygiene intervention in intervention group
7. Discussion, conclusion and recommendations:

This chapter includes a summary of research findings, overall conclusions and recommendations, including implication for future research.

Chapter I highlighted the fact that food hygiene has been under prioritized as part of diarrhoea and childhood undernutrition prevention and control programmes despite the fact that the incidence of diarrhoeal diseases is especially high after complementary feeding is initiated[1-4]. Increased investment in food hygiene is thus critical to child health and development. It noted that the potential factors associated with the contamination of weaning and child foods have previously been studied [5-14] and reviewed [2, 9, 13, 15] and a few studies also explored possible risk factors for diarrhoea and undernutrition [8, 9, 16]. A few studies used the HACCP approach to investigate the processes and procedures that contribute to microbial contamination in domestic settings and to identify points where controls could be applied [5, 6, 17, 18]. Further, effectiveness measurement trials of WASH interventions have mostly focused on sanitation[19] and handwashing with soap[20]; and reviews of the health impact of WASH have mostly considered the impact of water[21-23], sanitation[22, 23], handwashing with soap [24, 25] and not food hygiene. Previous reviews related to food safety in low-income settings mostly concluded that weaning food prepared in unhygienic conditions is frequently heavily contaminated with pathogens and is therefore a major factor in the cause of diarrhoeal diseases and associated malnutrition in low income countries[2, 9]; these reviews called for the application of food hygiene interventions in developing countries[8]. The importance of the issue as well as its relative neglect shows the need for food hygiene interventions in low-income settings, and that this should be built on modern behaviour change approaches rather than assuming that health education will produces the behavioural changes required.
7.1 Summary of research findings:
The main objective of this thesis was to design and implement a food hygiene intervention, and assess its effect on mothers’ food hygiene practices and on the level of microbiological contamination in food. An additional objective was to explore whether food hygiene interventions can be integrated into nutrition, WASH and health strategies and programmes in Nepal. The research findings presented in this thesis were obtained from a systematic review (Chapter II), formative research (Chapter IV), and Cluster Randomized, Before-After study with control (Chapter V and VI), and are summarised in this section.

Chapter II presented the evidence obtained from the systematic review of food hygiene intervention. No systematic review has previously been conducted on food hygiene interventions and their impact on people’s behaviour, microbial contamination and health outcomes. Medline, Embase, and Global Health datasets were searched and only five studies[26-30] met the review inclusion criteria. Interventions using HACCP principles focusing on domestic settings were effective in reducing faecal contamination in child foods [26, 27]. However, none of these studies reported the details of behaviour change programme components, and the feasibility of scaling up food hygiene behaviour change interventions in community settings is yet to be explored. Two small-scale household level behavioural trials assessed the feasibility of targeting a few behaviours, but were inconclusive as to whether these could be implemented and sustained through public health interventions [29, 30]. Results obtained from a longitudinal (cohort) study showed correlations between food hygiene, diarrhoea morbidity and child growth, but were inconclusive[28].

Many aspects of the relationship between food hygiene and child health remain unclear and there is limited evidence about how best to design and deliver food hygiene behaviour change interventions. It is unclear which factors, or behaviours pose the greatest risk of food contamination and should therefore be targeted by interventions seeking to reduce these risks; it is also unclear how such interventions can be implemented in a normal community setting to have an impact on improving behaviours, reducing contamination and diarrhoeal diseases. The review thus showed that there was a need for a scalable food hygiene interventions to investigate both programmatic and research gaps.
Spurred by this need in this research we developed designed and delivered an intervention using the approach of Behaviour Centred Design (BCD) underpinned by Evo-Eco theory and Theory of Change.

Chapter IV described the findings obtained from the formative research conducted using Evo-Eco theory\[31\] and HACCP principles\[18, 32\] to pinpoint the key food hygiene behaviours in the study area and their determinants and inform intervention design. We found that living space, especially the kitchen, presented a challenging environment for mothers and other family members to practice adequate food hygiene behaviours. The physical environment had the following problems – dark and difficulties to meticulously clean the kitchen surface and practice behaviours, the single room used for multiple purposes, the biological environment had the following problems - heavy presence of flies and animals in the kitchen, and presence of animal faeces in the compound, and the social environment had the following problems - different ethnic groups following different practices and long held food preparation traditions.

In the study area, prevalent food hygiene practices of mothers were inadequate and young children were frequently exposed to high levels of faecal bacteria through food, water and milk consumed daily. Around 47%, 30% and 92% respectively of food, water and milk samples were heavily contaminated with total coliforms. The HACCP approach was used to identify six critical and behavioural control points.

The behavioural assessment, identification of behavioural determinants, assessment of microbiological contamination in food and finally identification of critical and behavioural control points offered sufficient information for prioritising multiple food hygiene behaviours. Of the six identified behaviours, five were selected for targeting through a hygiene intervention, excluding thorough cooking, which is already practised by most mothers in the study area. The behaviours were selected to address most of the critical contamination pathways through which pathogens are likely to be introduced and ingested in food, and to support the reduction of contamination in food. The immediate motives and barriers to practicing all key behaviours including the role of kitchen settings were identified, and motivational drivers of behaviour change such as ‘disgust’, ‘nurture’, ‘affiliation’ and ‘social status’ were determined. Understanding all these behavioural determinants through formative research helped to provide the brief for an innovative, creative, motivational food hygiene intervention package.
Chapters III and V described the design and delivery of the food hygiene intervention. The intervention package was designed working with a creative team. Results from the formative research, a creative brief created based on the formative research findings, and past experiences from Bangladesh[26], Mali[27], India[20] and other settings and hygiene behaviours provided a useful basis for the intervention design process. The prototype package was designed and pre-tested in the field. Local female food hygiene motivators (FHCs) were trained to deliver the intervention independently, following programme guidelines. Their skills to deliver the package were enhanced through the engagement in pre-trial activities conducted as part of intervention package pre-testing in a real-life scenario.

The intervention package was delivered using a campaign approach over three months through six events followed by six household visits. The campaign emphasised changing the physical and social nature of kitchen settings and employed emotional drives. The theory of change for the campaign is shown in chapter III (Figure 4). As guided by theory, different engaging, attractive and appealing activities using programme guide and collateral were implemented in the behavioural settings (physical, social and biological environment) to motivate mothers using nurture, disgust, affiliation and status as a key motivational drivers. The campaign had an appealing desire to be ideal mothers, to be respected as such in their community, to bring happiness to the family by ensuring an environment that supports the success of their child, and by piquing their disgust through highlighting the risks of poor food hygiene with an expectation that such emotion leads to behaviour change resulted reduction in low level of contamination in child food. Previous behaviour change studies on handwashing have shown that traditional behaviour change promotion methods are often ineffective and highlighted the need for creative and innovative techniques [20, 33, 34]. As improved knowledge of the need to practice better hygiene does not necessarily translate into practice, the campaign focused on motivating mothers to improve behaviours rather than educating them to improve knowledge. Very few immediate motivational messages were communicated, to improve knowledge. As recorded through the process documentation, kitchen makeover with eye danglers’ placement in kitchen as behaviour reminders was helpful to disrupt daily food preparation, storage and feeding habits and instrumental to continuously reinforcing / reminding corrective behaviours. The competitions (clean kitchen, ideal mother and safe food hygiene zone), public pledging such as putting ideal mothers photos in the junction of the village, activities linked with emotions such as child life game, glo-germs, hot potato, drama, the provision of bib, and folk song were repetitively mentioned exciting activities by mothers.
Chapters V and VI discussed the effect of the food hygiene intervention measured using a Cluster Randomized, Before-After study with Control design. The proportion of mothers practicing all prioritised food hygiene behaviours was measured by structured observation[20, 35] in all four randomly-selected intervention clusters (120 households with no loss to follow-up) and four control cluster (119 households with two loss to follow-up) before and after the intervention as primary outcomes. The effect of the intervention on the level of faecal bacterial contamination in food and water was measured as secondary outcomes of the study in sub-samples of the study population (40 households in the intervention clusters and 40 households in the control clusters). The intervention and control cluster households had broadly similar social, demographic and economic characteristics. BAC studies are more convincing if the intervention and control groups start off with roughly similar baseline[36].

The intervention campaign accomplished a wide reach (90% exposed >10 times out of 12 targeted exposure) in the intervention group. The food hygiene intervention demonstrated that it is possible to change entrenched and habitual food hygiene behaviours in poor rural settings like Nepal. Results indicate that the food hygiene intervention campaign achieved an increase in the mean proportion of target behaviours of 42% (p=0.020). The intervention was equally effective in significantly improving the set of five key behaviours identified through the formative research process, as measured at baseline and follow up. The effect of the intervention was almost equal across the intervention clusters with few variations. The intervention was also successful in reducing the presence of total coliforms and E. coli in commonly-used child food. Low levels of microbes in food were observed at four stages (<1 log_{10} cfu/gm of food) and most importantly at the time of child feeding in the intervention group during follow-up.

The study results suggest that multiple behaviours can be targeted by behaviour change interventions and, and show that emotional motivators can work to change people’s behaviours. We suspect however, that the most important part of the intervention may have been the change in the physical and social settings in the kitchen which is kitchen makeovers using demarcation bunting, eye danglers etc marking as they did a major change for mothers in the way they thought about and used kitchens both physically and socially.
We failed to show a significant change in the microbiological quality of the drinking water following the intervention. This may be due to campaign focus only on water treatment but not on storage after treating water. However, contamination levels in water were at a low level at the start and remained low as compared to food suggesting that food is one of the prime sources of microbial exposure to young children in low-income settings like rural Nepal.

The success of the intervention can be attributed to many factors within the intervention. Various novel elements including the change in physical and social settings of the kitchen including behaviour reminders placement, and the use of emotional drivers through socially and culturally relevant creative and appealing activities possibly motivated mothers to change behaviours in the new kitchen environment. Triggering messages linked with immediate motives were used to encourage behaviours. The effective implementation of the package by the active, motivated and skilled FHM and overwhelming involvement of mothers including grandmothers in the events might have encouraged mothers to continuously perform behaviours and establish social norms. Previous studies also suggest that the quality of interventions improves if the implementation team is skilled[20] and hygiene behaviours are performed if communities are actively involved[37]. Despite the good results achieved by this trial, we are not able to draw conclusions as to which particular tools have been most effective in delivering the results, which motivational drivers had the greatest effect, or whether the results were due to a combined effect, including the change in physical settings.

The experience gained in this study of dealing with multiple behaviours and results suggests that the promotion of multiple behaviours may have a complementary and mutually-reinforcing effect, if all behaviours are addressed in one setting and practiced in sequence. Since most behaviours dealt with the kitchen environment, it was easier to change one settings (as changing settings can be challenging in the programme context). Most prioritised behaviours happen in sequence, making it easier for mothers to remember subsequent behaviours while performing one behaviour. Findings also suggest that all five key food hygiene behaviours are critical to controlling or eliminating the faecal contamination levels in commonly-used child food. For example, promoting thorough cooking and re-heating can kill most microbes, but the use of unwashed serving utensils to serve cooked food may re-introduce contamination if this behaviour is not addressed. Similarly, even when food is thoroughly cooked or re-heated and serving utensils have been washed, if the mother’s or child’s hands have not been washed prior to feeding or eating, further contamination may be
introduced. Additionally, while proper storage does not kill microbes, it protects from cross-contamination or ingestion of additional contamination from flies, dust, dirt, animals, hands etc. Proper storage in a container with a tight-fitting lid can protect from cross-contamination, but such food should be reheated thoroughly before serving. Lastly, if children are given contaminated water or milk together with safe food, all other food hygiene measures practiced may be in vain. Undermining one or more behaviours may lead to negative outcomes. All five hygiene behaviours should therefore be practiced.

The food hygiene package was relatively labour intensive to deliver through six events and six household visits but it can still be simplified to make it something more scalable. The total exposure can be reduced to fit only those activities which were more appealing, emotional and attractive without diluting the overall principles.

The gray literature review of the Nepal’s health, WASH, nutrition and food technology sectors policies are described in Chapter-II and summary of the policy makers consultations and view are described in Annex A to ascertain whether food hygiene can be integrated into sectoral strategies and programmes in Nepal. The quick analysis showed that Nepal has a favourable policy environment for incorporating food hygiene into ongoing health, nutrition and WASH programmes, but that none of these sectors has set implementation guidelines for hygiene or has developed an understanding on the key hygiene or food hygiene behaviours that require addressing. Nor do they have the scalable package to deliver behaviour change programmes. There is growing recognition of the importance of hygiene overall and food hygiene in particular for maximising the health benefits of ongoing sanitation, water supply, nutrition and health programmes. Failure to integrate food hygiene into ongoing initiatives was identified as a missed opportunity.

In the WASH sector, food hygiene is thought to be included as part of the ongoing hygiene promotion initiatives as part of post-ODF promotion and total sanitation movement, and there is need for an overarching “national hygiene framework” in-line with the Sanitation and Hygiene Master Plan, with clear institutional responsibilities for hygiene promotion so that the importance of behaviour change aspects can be fully realised and institutionalised. In the nutrition sector, although the guiding policy and strategy 2004[38] recognised the need for good hygiene practices and improved food hygiene, in practice ‘nutrition specific’ interventions have been prioritised over nutrition sensitive-interventions. The Nepal Multi-

How behaviour change programmes can be designed and delivered in community settings to achieve significant improvements in behaviours and reduce food contamination were partially answered by sharing this food hygiene trial components and results. However, questions related to how such an intervention can be delivered through regular health, WASH and nutrition programmes in a cost effective way are yet to be answered. A policy debate held as part of this study suggested that a food hygiene intervention can be piloted in a few districts through ongoing CB-IMCI or health promotion or immunization programmes, child feeding programmes as well as hygiene promotion programmes delivered by the Health, WASH and Nutrition sector. An opportunity to integrate food hygiene as one component of the ongoing pilot initiative to integrate hygiene into the national immunisation programme[40] was discussed and representatives from collaborative partners of this initiative shared their commitment and willingness.

7.2 Overall conclusion
Overall the study suggests that it is possible to change food hygiene behaviour and improve the microbiological quality of food in challenging contexts such as rural areas of Nepal. Factors that led to the success of the intervention included, using theory and process to guide intervention design and testing, using emotional rather than educational approaches and changing the settings of behaviour. On top of this we paid close attention to the quality of the delivery of the intervention through careful training and monitoring of the intervention. The campaign was relatively intensive and work now needs to be done to create interventions that can be delivered through existing institutions cost-effectively at scale. The policy environment in Nepal is now primed for this to happen.

Successfully implemented simple and motivational food hygiene intervention in community setting targeting multiple behaviours suggested that getting mothers to change long-held routine kitchen practices need clever new ways, informed by theory, practical novel approaches of behaviour change such as use of emotional drivers and change in behavioural settings. The improvement of all key safe food hygiene practices may possibly contribute to
the prevention and control of diarrhoea and malnutrition. However, the effect of food-borne contamination on child health during the critical window of weaning and long term development still needs to be better understood. In particular the relationship between food-borne pathogens and health outcomes (diarrhoea, malnutrition), its cost effectiveness analysis and long term sustainability needs more evidence. This would inform policy at national and international levels. Future challenges include the simplification of the intervention; the testing of its long-term effects in different contexts; the scaling up of the intervention with the determination of its impact on health status; and the integration of the intervention into normal public health, WASH and nutrition programmes in low-income settings. The scaling-up of food hygiene intervention is required to influence relevant ministries as well as to answer some of the unanswered questions around food hygiene. Effective collaboration with relevant ministries is required. Key collaborative action points may include working closely with WASH programmes where hygiene components have been included but have not specifically focussed on food hygiene, working with nutrition programmes already targeting improvements in complementary feeding practices at scale but have not focused on food hygiene and working closely with health sector already targeting public health intervention to prevent diarrhoeal diseases but have not specifically focussed on food hygiene.

Based on the findings presented in this thesis, it is possible to conclude the following:

- Despite the fact that food hygiene is likely to be a major contributor to poor health in developing countries, it has been rarely studies and even more infrequently targeted.
- Identification of critical and behavioural control points is a useful approach to research and identify key food hygiene behaviours.
- The six food hygiene behaviours (thorough cooking, cleanliness of serving utensils, handwashing with soap by mothers before feeding and by child before eating, proper storage, thorough re-heating and treatment of water/milk before serving) are key behaviours in the food hygiene domain. Among six key behaviours, adversely practiced behaviours can be prioritised for improvement through intervention.
- Understanding of physical, social, biological environmental settings and behavioural determinants, motives around specific behaviours through formative research, and the application of this understanding to tool design is essential for designing a socially and culturally appropriate and effective intervention package capable of achieving an effective change in behaviours.
The involvement of a creative team in the intervention design process is helpful to design an innovative and creative promotional package that catches the attention of the target audience.

Significant improvements in five key targeted food hygiene behaviours indicate that a theory driven, systematically designed scalable food hygiene intervention employing motivations and changing behavioural settings can substantially alter multiple food hygiene behaviours in low-income settings like Nepal.

The adoption of all key food hygiene behaviours is fundamental to the reduction of microbial contamination in food in low-income settings. Undermining one or more behaviours increases risk, mainly during cooking, feeding, storage and re-heating or boiling.

The promotion of water treatment options alone may not ensure the consumption of water of sufficient quality; any household point-of-use water treatment promotion initiatives should include safe household water storage practices.

Getting mothers to change long-held routine kitchen-based food hygiene practices will always be difficult. Innovative and creative ways and approaches to change behaviour are needed.

It is as yet unclear whether the large-scale replication of the package will achieve the same degree of behaviour change and microbial reduction in commonly used child food. However, current results demonstrate the public health importance of food hygiene.

By understanding the possible pathways for food contamination and subsequent outcomes, it is possible to conclude that existing interventions to reduce diarrhoeal disease transmission and improve nutritional outcomes are not sufficient to eliminate the faecal-oral transmission of bacteria among infants and young children, as many interventions ignore the most critical pathways by which faecal pathogens can be ingested, i.e. food.

Nepal has a favourable policy environment to incorporate food hygiene into ongoing health, nutrition and WASH programmes. Failure to integrate will constitute a missed opportunity to improve child health in Nepal.

Any policy and strategy analysis studies should contain information about the financial resources or human resources or any other issues that would determine how much these policies are actually put in place and run and resourced to offer better conclusion.
Motivating mothers and other family members to achieve sustainable improvement in key food hygiene behaviours could be an important intervention in the control and reduction of foodborne infection, diarrhoea and childhood undernutrition in Nepal.

7.3 Recommendations:

Programmatic recommendations:

• Food hygiene interventions must be carefully designed based on behaviour change principles addressing behavioural settings and understanding determinants of behaviour change. Programmes that creatively target emotions, motives and the settings in which behaviours take place are likely to be more successful. Future design of any behaviour change intervention should consider these principles.

• The intervention which was designed for this study and implemented in community settings with the support from food hygiene motivators (FHM) was effective to improve multiple behaviours and reduce food contamination. However, it is yet unclear whether large scale replication of this package will achieve the same degree of behaviour change and microbial reduction in food. The next step therefore should be to refine the package and test whether it can be equally successful at large scale in different settings. This would also include working closely with the nutrition, WASH and health sectors.

• Establishment of an accountable ‘institutional home’ for hygiene promotion and development of an overarching food safety policy including a national hygiene framework would enhance the policy environment for integration of hygiene through different sectoral programmes in Nepal.

• Governments, donors and practitioners should increase financing of food hygiene interventions. The damage caused by diarrhoea and undernutrition to a child before the age of five is irreversible and goes on to predict a range of health and other outcomes later in life. Increased investment in food hygiene is thus critical to child health and development in many low-income settings like Nepal.

• Better integration across the nutrition, health and WASH sectors is needed, and food hygiene should be mainstreamed. Evidence-based guidelines that better reflect the complex determinants of food hygiene behaviours and suggest greater cross-sectoral collaboration should be developed in low-income settings like Nepal.
• Evidence-based international guidelines that better reflect the complex determinants of food hygiene behaviours and suggest greater cross-sectoral collaboration, are essential. Ongoing discussions on the post-2015 development framework provide the perfect setting and crucial range of stakeholders required for such efforts. Guidelines formulated could then be adapted to the national context through a participatory process that engages all relevant stakeholders.

• Overall, strategies to control diarrhoeal diseases and improve childhood undernutrition in low-income settings should prioritise food hygiene behaviour change initiatives.

Future research recommendations:

• Additional high quality food hygiene intervention studies with particular emphasis on transmission pathways and health outcome is needed to further test scalable behaviour change intervention in different settings that yield tangible results and build on existing programmatic responses to poor WASH and undernutrition.

• The inclusion of microbial indicators in behaviour change intervention trials can provide more convincing results; it is therefore recommended that future behaviour change studies include such indicators to minimise existing knowledge and evidence gap in this sector.

• Although the food hygiene intervention was effective in reducing food contamination, the transmission pathways of all the major enteric pathogens should be more clearly established through future research to better target interventions.

• The effect of food-borne contamination on child health during the critical window of weaning and long term development needs to be better understood. In particular the relationship between food-borne pathogens, diarrhoea and malnutrition needs more evidence. This would inform policy at national and international levels. Any future food hygiene studies should address this issue for better programmatic influence.

• The cost effectiveness and benefit analysis of any public health intervention is equally imperative to recommend whether those interventions can be implemented at scale. Any future food hygiene intervention study should address these aspects thoroughly.

• Future efforts to improve public health, reduce food-borne infection, diarrhoeal diseases and childhood undernutrition need to adequately address food hygiene thereby to block the possible contamination pathway.
7.4 Disclaimer:
This study was funded by Department for International Development (DFID) through SHARE Consortium and co-funded by WaterAid UK. However, the views expressed do not necessarily reflect the Department’s and WA’s officials policies. I declare that I have no conflicts of interest.
7.5 References:

PhD-related links and publications:


- **Poster Presentation** in Research Students' Poster Day at LSHTM on 05 Feb 2014. Poster included findings from food hygiene intervention trial. Received "best poster - acrtect award" for demonstrating the use of innovative and creative science in research ([http://www.shareresearch.org/Resource/Details/food_hygiene_intervention_poster](http://www.shareresearch.org/Resource/Details/food_hygiene_intervention_poster))

- **Web-publications (food hygiene):**
  - **Om's Blog:** reporting progress on a food hygiene intervention to improve food hygiene behaviours, and reduce food contamination and diarrhoeal disease burden in Nepal. [http://www.shareresearch.org/NewsAndEvents/Detail/om_blog_food_hygiene_study](http://www.shareresearch.org/NewsAndEvents/Detail/om_blog_food_hygiene_study)
  - **Video documentary (19min) on ‘Food Hygiene Intervention’** describing actual trial, 2015. [https://www.youtube.com/watch?v=gPi6IyN0ZCU&feature=youtu.be](https://www.youtube.com/watch?v=gPi6IyN0ZCU&feature=youtu.be)
  - **YouTube video of Om** on ‘Why food hygiene matters’ produced by hygiene centre LSHTM and available in hygiene Centre website ([http://www.youtube.com/watch?v=6lMzOMVe-s](http://www.youtube.com/watch?v=6lMzOMVe-s))
  - **A podcast providing details on Om's food hygiene work** made publicly available ([http://soasradio.org/content/nepal-investigating-sanitation-health-and-food-hygiene](http://soasradio.org/content/nepal-investigating-sanitation-health-and-food-hygiene))
ANNEX A:

Policy vs practice: can food hygiene be integrated into health, nutrition and WASH programming in Nepal?

Summary

To explore whether food hygiene interventions can be integrated into nutrition, health and WASH strategies and programme in Nepal, an assessment was carried out during Sept – December 2013. Government policies, strategies, and programme implementation guidelines from Nepal’s health, WASH, nutrition and food technology sectors were reviewed and details are highlighted in Chapter II as part of ‘Nepal specific gray literature review’. Findings obtained from policy / strategy review were triangulated through key informant interviews with policy and programme development professionals. To establish consensus findings from this trial in Nepal and summary of policy brief were shared, and a debate on study findings was facilitated among policy makers, programme implementers, donors and researchers.

We found that Nepal’s policy environment can enable the integration of food hygiene promotion within ongoing WASH, nutrition and health programmes, and that there is growing recognition of the importance of food safety/hygiene. This recognition is not, however, reflected in existing programme. Failure to integrate food hygiene into ongoing programmes was identified as a ‘missed opportunity’.

The establishment of an accountable ‘institutional home’ for hygiene promotion and the development of an overarching food safety policy including national hygiene framework would enhance the policy environment for integration of food hygiene through different sectoral programme in Nepal. The need of a scalable pilot under normal service delivery programme was recognised.
INTRODUCTION:

Food hygiene promotion is an important, but neglected intervention within public health, nutrition and water, sanitation and hygiene (WASH) programmes. In Nepal this might be due to policy and programming gaps, competing priorities, or lack of experience and knowledge on how to change people’s food hygiene behaviours. In this context, we have previously tested and documented the effects of a food hygiene intervention in Nepal (result presented elsewhere). In this paper, we explore whether such types of interventions can be integrated into Nepal’s nutrition, health and WASH strategies and programmes.

Nepal has made some progress in reducing child mortality from 162/1,000 live births in 1990 to 54/1,000 live births in 2011[1], however the basic determinants for child health remain poor. Diarrhoeal diseases are still the second leading cause of under-five mortality in Nepal. With around 41% of under-five Nepali children stunted and 29% underweight [1], Nepal is among the top ten countries in terms of stunting prevalence, and in the top twenty in terms of absolute numbers of stunted children globally[2]. Despite these challenges, great emphasis is placed on dietary intake and supplementation ('nutrition specific' interventions) in Nepal’s nutrition programmes over ‘nutrition sensitive’ interventions such as sanitation and hygiene promotion. Despite the importance of exclusive breastfeeding in the first six months for protecting child health, around 30% of Nepali children received complementary food before the age of 6 months[3]. It is therefore plausible that young Nepali children are exposed to microbes through contaminated food in their early life. Food hygiene plays a potentially important role in childhood diarrhoea and undernutrition in Nepal.

While hygiene is recognised as a critical component of WASH programmes in order to achieve health outcomes, current efforts usually do not prioritise hygiene and much less food hygiene. WASH sector data shows that sanitation coverage in Nepal increased from 6% to 62% between 1990 and 2011[4], while monitoring/survey data shows sanitation use as 35% in 2011[5]. No national hygiene target has been set, nor are there estimates on hygiene practices; however, NDHS data shows that 48% of households have soap and water at the place where family members washed their hands[1]. The combination of low sanitation use and lack of hygiene practices is likely to lead to transmission of faecal pathogens through food. The lack

1 ‘Nutrition-specific’ programmes address the immediate determinants of foetal and child nutrition and development, whilst ‘nutrition-sensitive’ programmes address the underlying determinants. [link to source]

http://www.unicef.org/ethiopia/3_Nutrition_sensitive_interventions_and_programmes_how_can.pdf
of credible data makes the magnitude of food-borne infections and associated consequences difficult to estimate. Nonetheless, the introduction of a food hygiene intervention for reducing food-borne infections, diarrhoeal diseases and undernutrition is likely to be important, and requires an enabling policy, strategy and programme implementation and monitoring environment.

This document explores the potential for the integration of food hygiene interventions into Nepal’s nutrition, health and WASH strategies and programmes. Details obtained from ‘policy / strategy’ analysis included in Chapter II as part of Nepal specific gray literature review and details obtained from key informants interviews and policy debate are reflected as Annex A in this document.

**METHODS:**

Four approaches were used for policy content analysis and identification of food hygiene integration entry points: desk review, key informant interviews, development of a policy brief and policy debate and dissemination. Written consent was obtained from key informants. Ethical approval was obtained from the ethics committee of the LSHTM and NHRC, Nepal. Figure 1 shows the four approaches used for the policies and strategies content analysis and identification of food hygiene integration entry points.

![Figure 1: Different approaches used to assess whether food hygiene can be integrated into national policy and programme](image-url)

**Figure 1: Different approaches used to assess whether food hygiene can be integrated into national policy and programme**
**Desk review (policies and strategies analysis):**
All policy and strategy documents issued between 1990 - 2013 by the health, nutrition, WASH and food technology and quality control sectors in Nepal were collected and reviewed as part of grey literature review and details are already presented in Chapter II.

**Key informant interviews:**
To identify current practices on implementation of food hygiene-related interventions and perceptions around the feasibility of integration, semi-structured key informant interviews were conducted with 16 policy and programme development professionals, including high level professionals from government health, nutrition, food technology and quality control and WASH ministries and departments, as well as relevant donor and INGO staff.

**Policy brief:**
A policy brief highlighting policy statements, current gaps and possible integration entry points was developed based on the reviewed documents and key informant perceptions. The brief was used in a subsequent policy debate forum.

**Policy debate using food hygiene trial findings and policy synthesis:**
A national level workshop was organised bringing together around 60 policy makers, programme implementers, donors and researchers. The importance of food hygiene was highlighted, and the summary policy brief was presented and shared. The tested food hygiene intervention package, implementation modality and its effectiveness were presented and shared. Based on the findings from the policy analysis, interviews, and using evidence from the full study, a policy debate was conducted among the attendees. The debate focused on whether food hygiene can be integrated into different sector strategies and programmes, and ways for doing so through existing service delivery mechanisms. Verbal commitments made by high level officials from different ministries and donor and INGOs were recorded, as well as the consensus reached during the debate. Figure 2 shows few images from the key approached taken such as desk review, policy debate and dissemination of trial evidences.
Analysis:

Qualitative evidence obtained using different methods was analysed under different themes. Sector-specific vertical programmes linked with diarrhoea control were summarised under treatment and prevention themes. Evidence obtained from policy / strategy analysis was summarised under policy and strategy themes as part of grey literature in Chapter II. Results obtained from key informant interviews were summarised under the theme of feasibility of integrating hygiene into sector programmes. The richness of the information after triangulating evidence from the desk review with key informant interviews led to the development of a policy brief. The policy brief and findings obtained from food hygiene intervention trial set the agenda for the multi-sector policy debate. The consensus developed in policy debate forum and evidence obtained from review and key informant interviews helped to draw a conclusion. Specific results are discussed in this paper (annex-A).
RESULTS:
Findings obtained from policy / strategy analysis and review are presented in Chapter II as part of ‘Nepal specific gray literature review’ process. This section only covers the findings obtained from key informant interviews, and policy debate using main trial findings and policy brief.

Feasibility of integrating food hygiene into sectoral programmes:

WASH sector perceptions:
WASH informants felt that although hygiene has been included in some recent policies/strategy, water and sanitation policy has had far more attention. Since 2008, the International Year of Sanitation, WASH activities gained momentum under the coordination of the Steering Committee for National Sanitation Action (SCNSA) and National Sanitation and Hygiene Steering Committee (NSHSC) in Nepal. All believed that although the Sanitation and Hygiene Master Plan offers a strong starting point for hygiene interventions, implementation guidelines and frameworks are needed to operationalise the Plan. Numerous approaches were used to promote sanitation and hygiene in Nepal, ranging from simple public messaging campaigns to long-term participatory programmes but almost none on food hygiene. A lack of operational research on hygiene hinders evidence-based planning and programming. The lack of institutional ownership for hygiene promotion was seen by informants as a major challenge. One planner stated: “we have tried our best to incorporate hygiene into our water and sanitation programmes, but we never monitored whether such integration resulted in behaviour change; we still count taps and toilets”. One donor referred to the prevailing assumption within the WASH sector that hygiene and food hygiene promotion is the role of the health and nutrition sectors, while health sector and nutrition actors believe that hygiene is the responsibility of the WASH sector; the result is continued under-prioritisation of hygiene. Hygiene efforts are also undermined by the absence of a solid epidemiological evidence base. Nonetheless, most informants felt that food hygiene could be included as part of ongoing hygiene promotion, as well as within the national Total Sanitation Movement and ODF promotion follow-up activities.

Health sector perceptions:
Based on this evaluation, health sector stakeholders do recognise the importance of food hygiene for infection prevention and improved child health. However, institutional
responsibilities regarding hygiene are not the mandate of any specific government health authority, leading to a fragmented approach. Following the cholera outbreak in 2009, as the death toll increased and the scale of the outbreak became clearer, stakeholders gradually realised the need for collective efforts for diarrhea control. Since then there has been a marked increase in attention to sanitation and hygiene within the health sector however lack of explicit budget allocation and indicators for WASH in NHSP-II was major challenge. There was consensus that NHSP-II opens a conducive policy environment to work on hygiene including food hygiene and few initiatives such as formation of Environmental Health and Hygiene Technical Committee under NHEICC, leadership in celebrating global handwashing days and broadcasting of WASH messages through national media were cited as work progress. Informants felt that food hygiene promotion could be incorporated into ongoing diarrhoea control initiatives such as community-based integrated management of childhood illness (CB-IMCI), using Female Community Health Volunteers (FCHVs), ongoing health promotion (national health education information and communication programme NHEICC); however, the way in which hygiene could be incorporated into these programmes remained unclear. One high level official said that though opportunities have been missed in the past, the food hygiene will be a major component in a pilot project which will integrate hygiene into the national immunisation programme [6].

**Nutrition sector perceptions:**
In the nutrition sector informants argued that the lack of a responsible institution for hygiene impedes progress. The discussions revealed that the MSNP succeeded in increasing the accountability of different actors in nutrition and offered an opportunity to amend policies, allocate resources and prioritise hygiene promotion. However, not all sectors have acted on this opportunity. The current proposed behaviour change component in MSNP includes only an awareness-raising programme and a short training course, which are unlikely to be sufficient for changing population hygiene behaviours. Programme implementators concluded that the MSNP implementation guide should include food hygiene as a part of the child feeding strategy and promotional interventions. Informants further agreed that although the nutrition-specific aspects of the Plan are clear, the nutrition-sensitive components, including hygiene promotion, such as which aspects of hygiene should be prioritise and how behaviour change could be achieved, lack clarity. Since the endorsement of the MSNP, several projects to improve food security and maternal and child nutritional have been initiated with the
support of development partners such as an integrated nutrition, hygiene and sanitation, agriculture and health services promotion programme in order to address the key factors affecting nutritional status of women and children under two years in Nepal [7].

**Food Technology and Quality Control sector perceptions:**
This evaluation revealed that the DFTQC does not currently address domestic food hygiene despite the growing recognition of its importance. As stated, their ongoing nutritional programmes, including mass media campaigns, have focused on the nutritious value of foods rather than contamination. Quality assurance initiatives were generally on-off exercises limited to urban and industrial settings. There was a common belief that the DFTQC’s current work is not linked with health outcomes, as it operates under the Ministry of Agricultural Development and establishing functional coordination with the health sector is challenging. Although current work is well-placed to respond to growing public concerns regarding food safety in urban settings, it is unable to respond to the needs of the rural population, which is most affected by the outcomes of poor food hygiene. One respondent further retreated, ‘institutional mandates and its positioning needs revision/re-location’.

**Policy debate and dissemination of Nepal’s food hygiene trial package:**
The food hygiene trial and its implications for policy were discussed at the national forum.

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**Food hygiene trial in Nepal**

**Food hygiene trial in Nepal** was conducted to design, deliver and evaluate the effect of an intervention on the level of food hygiene behaviours of mothers and food contamination [8]. This consisted of a motivational package targeting five key food hygiene behaviours using emotional drivers rather than cognitive appeals as well as emphasis on change in behaviour settings. Four clusters received the intervention over a period of three months whilst four clusters remained a control group. Outcomes were measured in 239 households with a child aged 6-59 months. 43 percent of mothers were able to maintain all 5 key behaviours and intervention was successful to improve the microbiological indicators in child food significantly. The results suggest that it is possible to substantially improve multiple food hygiene behaviour and reduce the risk of microbiological contamination of child foods through scalable community level interventions.
The feasibility of implementing such a programme through the normal service delivery set-up, its cost-effectiveness and likely scalability were discussed. The participants felt that it should be piloted in few districts to answer the question as to how food hygiene improvements can be delivered through normal government channels. The debate around existing policies, opportunities and gaps concluded that the health, WASH, and nutrition sector has favourable policy environment to incorporate food hygiene as part of their hygiene activities and that further opportunities can be utilized while amending sectoral policy in the spirit of MSNP.

DISCUSSION:

Changing in hygiene behaviour for the achievement of health and nutrition targets has been largely overlooked in Nepal. While the existing policy framework offers important entry points for hygiene promotion in existing WASH, health and nutrition programmes, no sector has set implementation guidelines for hygiene or has developed an understanding on the key hygiene behaviours that require addressing, nor have they an effective operational approach to Behaviour Change. No government or other integrated or cross-sectoral food hygiene interventions were identified. We found growing recognition of the importance of hygiene overall and food hygiene in particular for maximising the health benefits of ongoing sanitation, water supply, nutrition and health programmes. Failure to integrate food hygiene into ongoing initiatives was identified as a missed opportunity. To answer whether food hygiene can be integrated into ongoing Health, WASH and nutrition programme, the need for a scalable pilot initiative through ongoing routine programme was recognised.

The health sector policy / strategy environment getting conducive to integrate hygiene into ongoing health programme. The institutional roles and responsibilities for the health sector in relation to hygiene are yet to be clearly established and implemented. The NHSP-III (currently in progress of formulation) offers an opportunity to address these and make health sector accountable for hygiene/ food hygiene promotion. Health sector actors recognized that tackling diarrhoeal diseases requires a comprehensive package of preventive and curative interventions[9]. Though ongoing programme link doesn’t include food hygiene component but it can be integrated into ongoing diarrhoea control initiatives such as through IMCI, health promotion and immunisation programme in Nepal. How to integrate food hygiene into normal service delivery mechanism needs piloting.
In WASH sector, the development of “Sanitation and Hygiene Master Plan 2010-2015”\[10\] ensured enabling policy environment for hygiene including some aspects of food hygiene promotion. However, the absence of an operational guideline or framework for Master Plan implementation remains an important barrier to progress. Common monitoring indicators and monitoring system and institutional responsibilities for hygiene promotion have not been defined in any of the documents. Though food hygiene thought to be included as part of the ongoing hygiene promotion initiatives from WASH sector as part of post ODF promotion and total sanitation movement, there is a need of overarching “national hygiene framework” in-line with Master Plan with clear institutional responsibilities for hygiene promotion so that the importance of behaviour change aspects fully realized and institutionalise.

The NAGA initiatives in Nepal \[11\] led to the formulation of a Multi-Sector Nutrition Plan (2013-2017) \[12\] and MSNP was identified as an ideal opportunity to embed food hygiene in different sector strategic plans and ongoing programmes. Though, MSNP does not explicitly mention food hygiene but can be framed under the hygiene component. Nutrition specific interventions have been prioritised over nutrition sensitive programme in Nepal. A hygiene framework that better reflects the complex determinants of health and nutrition is needed, and should illustrate the benefits of, and opportunities for, multi-sectoral collaboration. Ongoing discussions concerning the policy amendment by different sector in-line with MSNP\[12\] and current sectoral leading documents \[10, 13\] provide the perfect opportunity for each sector to materialize such initiative. Multi-sectoral agreement on common hygiene / food hygiene indicators and allocation of appropriate resources through different sectors should also be a programmatic priority. The more integrated approach \[7\] are needed for the improvement of nutritional status of Nepali children and those programmes should offer an ideal entry points for mainstreaming food hygiene components.

No food safety policy existed in Nepal until the end of 2013. No national standard for commonly-used household foods such as for Rice, Jaulo, Dhindo, Curry are set. An overarching ‘food safety policy’ to respond the needs of Nepal’s diverse settings including rural population is required. The DFTQC in coordination with health, nutrition, WASH and agriculture sector should take lead for this initiative.
Policy debate concluded that Nepal has favourable policy environment to incorporate food hygiene into ongoing Health, Nutrition and WASH programme. Such conclusions warrant the need of scalable food hygiene package and sharing of our recently tested food hygiene trial components and its results filled such gap. Concerned related to how could behaviour change programme design and delivered in community settings to have significant improvements in behaviours and reduce food contamination answered. However, questions related to how it can be delivered through normal service delivery programme from different sector in cost effective way remained unanswered. Further debate to answer this question on the same forum concluded that it can be piloted in few districts through ongoing any CB-IMCI or health promotion or immunization programme from health sector, any hygiene promotion programme from WASH and any child feeding programme from nutrition. Respective ministries and relevant donors should take lead to materialise such need. One of immense opportunity to integrate food hygiene as one of the major component into ongoing pilot initiatives i.e. ‘integrating hygiene into the national immunisation programme’[6] discussed and representative from collaborative partners for such initiative shared their commitments and willingness. This pilot opportunity however limits the scope of food hygiene intervention only among the target group that are coming into health institutions for vaccinating their under one year children. This initiative however will not limit other sector/partners to incorporate food hygiene into their programme. Failure to integrate will constitute further missed opportunity to improve child’s health in Nepal.

Several barriers to this approach have been identified such as: i) lack of responsible institution to lead the hygiene promotion, ii) lack of food safety policy and hygiene framework to guide multiple sector, iii) continued vertical health programming, iv) under-prioritisation of hygiene within the WASH sector, v) continued promotion of nutrition-specific programme over nutrition-sensitive, vi) lack of common indicators and monitoring on hygiene/food hygiene across different sectors, and vii) lack of accountability for hygiene resulting in inadequate funding and implementation. Integrating food hygiene through Health, Nutrition and WASH programme should therefore strategically address these barriers as well. The Government, I/NGOs and donor agencies working in health, WASH and nutrition sector in Nepal should work in coordinated manner to priority food hygiene behaviour change programme thereby to address prevalent diarrhoeal and childhood undernutrition problem in Nepal.
REFERENCES:


ANNEX B:

Ethical approval from LSHTM Ethical Review Board to conduct formative research

Observational / Interventions Research Ethics Committee

14 May 2012

Gan Prasad Gautam
Research Degree Student, ITD
LSHTM

Dear Dr Gautam

Study Title: Food Hygiene intervention to improve food hygiene behaviours, reduce food contamination and diarrhoeal diseases burden in Nepal
LSHTM ethics ref: 6164

Thank you for your email of 6 May 2012, responding to the Committee’s request for further information on the above research and submitting revised documentation.

The further information has been considered on behalf of the Committee by the Chair.

Confirmation of ethical opinion

On behalf of the Committee, I am pleased to confirm a favourable ethical opinion for the above research on the basis described in the application form, protocol and supporting documentation as revised, subject to the conditions specified below.

Conditions of the favourable opinion

Approval is dependent on local ethical approval having been received where relevant.

Approved documents

The final list of documents reviewed and approved by the Committee is as follows:

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After ethical review

Any subsequent changes to the application must be submitted to the Committee via an E2 amendment form.

Yours sincerely,

Professor Andrew J Hall
Chair

Improving health worldwide
ANNEX C:

Ethical approval from NHRC, Nepal to conduct formative research

16 May 2012

Mr Om Prasad Gautam
Principal Investigator
London School of Hygiene and Tropical Medicine (LSIHM, UK)

Ref: Approval of Research Proposal entitled “Food Hygiene intervention to improve food hygiene behaviors, reduce food contamination and diarrheal diseases burden in Kavre District of Nepal”

Dear Mr. Gautam,

It is my pleasure to inform you that the above-mentioned proposal submitted on 15 April 2012 (Reg. no. 38/2012; please use this number during further correspondence) has been approved by NHRC Ethical Review Board on 99 May 2012 (2069-01-27). This approval is only for formative research for now.

As per NHRC rules and regulations, the investigator has to strictly follow the protocol stipulated in the proposal. Any change in objective(s), problem statement, research question or hypothesis, methodology, implementation procedure, data management and budget that may be necessary in course of the implementation of the research proposal can only be made so and implemented after prior approval from this council. Thus, it is compulsory to submit the detail of such changes intended or desired with justification prior to actual change in the protocol.

If the researcher requires transfer of the bio samples to other countries, the researcher should apply to NHRC for the permission.

Further, the researchers are directed to strictly abide by the National Ethical Guidelines published by NHRC during the implementation of their research proposal and submit progress report and full or summary report upon completion.

As per your research proposal, your total research budget is US$ 7,878.06 and NHRC processing fee is NRs 8,530.00.

If you have any question, please contact our research section.

Thank You

[Signature]

Dr. Shanker Pratap Singh
Member Secretary

Nepal Health Research Council
Estd. 1991

Ref. No. 207

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Website: http://www.nhrc.org.np, Email: nhrc@nhrc.org.np

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ANNEX D:
Ethical approval to conduct Cluster Randomized, Before-After study with Control from LSHTM Ethical Review Board

Observational / Interventions Research Ethics Committee

On Prasad Gautam
Research Degree Student
DC / ITD
LSHTM

24 May 2013

Dear Mr. Gautam,

Study Title: Food hygiene intervention to improve food hygiene behaviours, and reduce food contamination in Nepal: an exploratory trial
LSHTM ethics ref: 0390

Thank you for your email of 13 May 2013, responding to the Interventions Committee’s request for further information on the above research and submitting revised documentation.

The further information has been considered on behalf of the Committee by the Chair.

Confirmation of ethical opinion

On behalf of the Committee, I am pleased to confirm a favourable ethical opinion for the above research on the basis described in the application form, protocol and supporting documentation as revised, subject to the conditions specified below.

Conditions of the favourable opinion

Approval is dependent on local ethical approval having been received, where relevant.

Approved documents

The final list of documents reviewed and approved by the Committee is as follows:

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<td>Informed Consent form – Household recruitment for trial</td>
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<td>Structure observation checklist: observed behaviours</td>
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<td>Baseline Survey – Household Questionnaires</td>
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<td>Standard operating guidelines to collect, process, inoculate, incubate and interpret food, milk and water samples</td>
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<td>Food, milk and water sample collection forms / details</td>
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Improve health worldwide
After ethical review

Any subsequent changes to the application must be submitted to the Committee via an E2 amendment form. All studies are also required to notify the ethics committee of any serious adverse events which occur during the project via form E4. An annual report form (form E3) is required on the anniversary of the approval of the study and should be submitted during the lifetime of the study. At the end of the study, please notify the committee via form E5.

Yours sincerely,

[Signature]

Professor John DH Porter
Chair
ethics@lshtm.ac.uk
http://intras.lshtm.ac.uk/management/committees/ethics/
ANNEX E:

Ethical approval to conduct Cluster Randomized, Before-After study with Control from NHRC Nepal

31 March 2013

Mr. Om Prasad Gautam
Principal Investigator
London School of Hygiene and Tropical Medicine (LSHTM) UK

Ref: Approval of Research Proposal entitled Food hygiene intervention to improve hygiene behaviors and reduce food contamination in Kavre district of Nepal: an exploratory trial

Dear Mr. Gautam,

It is my pleasure to inform you that the above-mentioned proposal submitted on 28 February 2013 (Reg. no. 24/2013 please use this Reg. No. during further correspondence) has been approved by NHRC Ethical Review Board on 29 March 2013 (2069-12-16).

As per NHRC rules and regulations, the investigator has to strictly follow the protocol stipulated in the proposal. Any change in objective(s), problem statement, research question or hypothesis, methodology, implementation procedure, data management and budget that may be necessary in course of the implementation of the research proposal can only be made so and implemented after prior approval from this council. Thus, it is compulsory to submit the detail of such changes intended or desired with justification prior to actual change in the protocol.

If the researcher requires transfer of the bio samples to other countries, the investigator should apply to the NHRC for the permission.

Further, the researchers are directed to strictly abide by the National Ethical Guidelines published by NHRC during the implementation of their research proposal and submit progress report and full or summary report upon completion.

As per your research proposal, NHRC processing fee is NRs. 52,200.00.

If you have any questions, please contact the research section of NHRC.

Thanking you.

[Signature]

Dr. Shanker Pratap Singh
Member Secretary
ANNEX F: Creative Brief (briefing note for creative team)

**General:**

| Title: | Food hygiene intervention to improve food hygiene behaviours and reduce food contamination in Nepal |
| Contact: | Om Prasad Gautam, PI of the study, LSHTM, UK |
| Site: | Kavre district, Baluwapati Deupur and Nayagaun VDCs |

**Background and rationale:**

Preventable and treatable food-borne diseases are a major cause of illness globally. Inadequate food hygiene is likely to account for a substantial proportion of foodborne infections including diarrhoea among infants and young children. Very few intervention studies have been carried out and there has been little effort to undertake food hygiene interventions for the reduction of childhood diarrhoea and malnutrition. A simple and replicable food hygiene intervention, which can be implemented by the WASH/health/nutrition sectors is needed.

**Context and prioritized behaviours:**

A ‘formative research - FR’ was conducted to inform the design of the intervention. Using Evo-Eco theory, HACCP method as well as various tools, prevailing level of food hygiene behaviours, motives, determinants, level of contamination in food, critical control as well as behavioural control points were identified. Food hygiene behaviours are underpinned by social, physical and biological settings, including level of awareness, cultural beliefs and individual psychology/attitude. The low level of maternal education combined with the physical setting, social norms, cultural values/beliefs, and environmental factors influenced mother’s ability to maintain food hygiene practices. Although the extent to which food hygiene practices varies, we have prioritized five behaviours which are critical.

<table>
<thead>
<tr>
<th>Five key behaviours</th>
<th>Reasons for promotion</th>
</tr>
</thead>
<tbody>
<tr>
<td>Thorough re-heating of leftover/stored food</td>
<td>Thorough re-heating/cooking (70°C) kills most microbes. Due to absence of refrigeration, all food is stored in ambient temperature, though cooking was adequate re-heating practice were inadequate</td>
</tr>
<tr>
<td>Cleanliness of child food serving utensils</td>
<td>Serving utensils likely to carry microbes. Cleaning them before serving food reduces contamination. Current practices are inadequate and microbial levels increased when utensils used (FR)</td>
</tr>
<tr>
<td>Handwashing with soap before feeding (mother) and before eating (child)</td>
<td>Hands likely to carry faecal microbes. Handwashing with soap can reduce microbe contamination on hands. Low soap use observed, and increased microbe levels obtained when food was mixed using hands</td>
</tr>
</tbody>
</table>
**Five key behaviours**

<table>
<thead>
<tr>
<th>Proper storage of cooked food</th>
<th>Reasons for promotion</th>
</tr>
</thead>
<tbody>
<tr>
<td>Flies and dust/dirt are likely to carry faecal microbes and contaminate food. Flies were accessing food and other possible routes were observed. No refrigerator. Stored food was heavily contaminated</td>
<td></td>
</tr>
<tr>
<td>Milk / water treatment</td>
<td>Improperly boiled milk stored in ambient temperature w/o cover likely to carry microbes; contamination also likely in stored water. Heavy contamination noted in both milk and water</td>
</tr>
</tbody>
</table>

*Note: Thorough cooking is one of the main critical control points however not prioritized as many mothers currently practice this behaviour.*

**Basis for intervention design (things to remember):**

<table>
<thead>
<tr>
<th>Intervention package</th>
<th>Simple, scalable, re-producible and replicable</th>
</tr>
</thead>
<tbody>
<tr>
<td>Behaviours</td>
<td>Five key food hygiene behaviours - multiple behaviours as above</td>
</tr>
<tr>
<td>Intervention focus</td>
<td>Use of emotional drivers (motives), change in settings (physical, bio and socio)</td>
</tr>
<tr>
<td>Intervention approach</td>
<td>Campaign style with identity (positioning, branding)</td>
</tr>
<tr>
<td>Intervention duration</td>
<td>Three months (May-August 2013) during rainy season</td>
</tr>
<tr>
<td>Frequency of events</td>
<td>6 community events and 6 household visits</td>
</tr>
<tr>
<td>Intervention delivery</td>
<td>Community and households level intervention by FCHVs/similar capacity</td>
</tr>
<tr>
<td>Primary target group</td>
<td>Households with a mother having a child aged 6-59 months</td>
</tr>
<tr>
<td>Secondary target group</td>
<td>All family members including grandmothers and villagers</td>
</tr>
<tr>
<td>Total Households</td>
<td>162HHs with a child aged 6-59 months in four intervention arm (450 indirect HHs in intervention area)</td>
</tr>
</tbody>
</table>

**Study settings and socio-economic context:**

Food hygiene intervention will be conducted in typical rural settings of Kavre, Nepal, at the adjoining Baluwapati Deipur and Nayagaun VDCs. VDCs with a multi-ethnic/lingual society comprising different caste and ethnic groups with majority (>60%) belong to the Hill Aadiwasi/Janajaati following different religions and cultural dimension. Majority of the target population (mothers) had none/informal education and almost all households are poor (majority earns <100$ per month). Majority (~90%) used pipe water connection (unprotected spring sources), around 50% practiced open defecation and none of them had refrigerator. More than 50% of households keep animals in the same house/compound in which they live, cook and eat and animal faeces observed almost in all HH’s surroundings (>80%). Flies were present throughout the village, particularly in kitchens. Firewood was the main source of cooking fuel and most HH cook on the floor which is made of mud. Majority (94%) owns soap for bathing and washing clothes. Most households served solid food (rice with
curry/vegetables or milk) to children, including those being weaned (6-23months). Around 24% children aged between 6-59months had diarrhoea in the last week.

Prevalent practices, barriers and motives related to five key prioritized food hygiene behaviours

<table>
<thead>
<tr>
<th>Thorough re-heating</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Current practices:</strong> Cooking once or twice but feeding multiple times (4-5 times) in a day. All households fed stored/leftover food to children. Only half of the households reheated stored food and that only 19% maintained adequate temp. Re-heated food was heavily contaminated when tested. Few used frying pan (Cauldron - Karai) and some used serving steal bowl to re-heat the food.</td>
</tr>
<tr>
<td><strong>Barriers:</strong> i) time and energy required, ii) difficulty in lighting the fire and fire smoke, iii) beliefs on no need to re-heat in summer, iv) fear of burning food, v) not having adequate pots, vi) fear of destroying test and nutritious value.</td>
</tr>
<tr>
<td><strong>Motives:</strong> Believed re-heated food keeps child’s abdomen warm, no abdominal pain, and it will be testier. Use of curry/vegetables soup or milk or water or ghee to re-heat food as it will protect from burning the food/pot.</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Cleanliness of child food serving utensils</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Current status:</strong> Only 16% cleaned serving utensils thoroughly using ash/soap before serving food to children and this practice differs by caste. A wooden spoon was used to mix flour while cooking dhindo but was never washed; instead, remnants were peeled or scraped off before use. Ash and soap were available almost in all households.</td>
</tr>
<tr>
<td><strong>Barriers:</strong> Majority didn’t have rack to put washed utensils hence put on the mud floor. Flies, animal faeces and dust/dirt cause contamination and re-contamination of washed utensils. Rightly unavailability of ash/soap in the yard</td>
</tr>
<tr>
<td><strong>Motives:</strong> Used ash because it makes the utensils shiny, clean and removes all the dirt like soap, ash is readily available and they don’t have to buy it.</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Handwashing with soap before feeding by mother and before eating by child</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Current status:</strong> Almost all mothers fed the child using their hands. Many mother allowed child to carry food around while eating. Ash and soap were available in 100% and 94% households respectively but only 7% of mothers used soap to wash hands before feeding child.</td>
</tr>
<tr>
<td><strong>Barriers:</strong> i) proximity of HW station in the kitchen, ii) mothers frequent exposure to work (domestic dust/field/agricultural), iii) soap cost money, iv) no use of soap in waste water collecting container (used for animals), vi) lack of knowledge on importance of HWWS, vii) belief on washing hands takes time</td>
</tr>
</tbody>
</table>
| **Motives:** i) wash hands to remove visible dust/dirt/bad odour, ii) wash hands for good smell, iii) child looks smart if hands are clean, iv) water is readily available so commonly used, vi) tradition of pouring
water in hands before eating. As revealed from motivational exercises, disgust, nurture and respect/status were motivational factors to wash hands.

**Proper storage of cooked food**

*Current status:* Generally food was cooked in the morning and stored at ambient temperatures with inadequate storage practices (without tight-fitting lid). Though 63% covers food while storing but majority of them put unclean ladle over the stored food which attracts flies/dust/dirt/animal. Cooked food store up to 12hrs considered fresh.

*Barriers:* Lack of refrigerator. Majority don’t have cupboard to store food hence either stored in same cooking vessel or in small bowl/plate on the floor. Few HHs also used mat to cover food.

*Motives:* Proper cover protects food from flies/ dust and prevents from animals (goats, hens, cats). Few mothers prefer frying pan to store the leftover food.

**Milk and water treatment (boiling)**

*Current practices:* None of the households boiled water before serving it to the children and none of them re-boiled milk when served at different times. A few HHs even gave their children raw (un-boiled) milk to drink.

*Barriers:* For water: child doesn’t like the taste of boiled water, didn’t have time to boil the water, felt no need to boil water in summer. For milk; felt no need to re-boil milk, frequent boiling may wreck the milk, no proper vessel to boil milk frequently, some believed that raw milk will have more nutritious value hence offered without boiling it.

*Motives:* Few HHs only boil water when the child/family members suffered from common cold. Majority mentioned ‘cold water’ taste is better than ‘hot water’. For milk, i) few believed that frequently boiled milk is testier, ii) almost all believed milk is ‘nectar’ (*Amrit*) and special for god to render.

**Scope of work for creative team:**

- Produce intervention package, detail session plan, curriculum including communication strategies to facilitate the promotion of food hygiene intervention package
- Develop prototype intervention package and discuss with stakeholders
- Test prototype intervention package, ensure feasibility and acceptability and finalize the package
ANNEX G:

Verbatim: voices from field during trial implementation period

Voices (comments) recorded during campaign implementation period suggest that the campaign approach was working and implemented activities had become the part of the community and family discussion. The campaign also influenced secondary target audiences. Some of the key voices (comments) made by mothers, social leaders and food hygiene motivators during campaign implementation period are included in this section:

Voices from Mothers:

“I felt like crying while listening to ‘Sani Maiya’s story’ (referring to a story highlighted in the programme). I didn’t realize how fast tears rolled down my cheeks. I wish it will never happen to anyone, even my enemies”.

25 years old participating mother from one of the intervention clusters

“In child’s life game, it was very sad to see ‘Heera’ moving backwards/regressing due to his mother’s poor food hygiene behaviours”. I will never forget this in my life and will never let my child down like Heera”.

33 years old participating mother in one of the intervention clusters

“I fear that my child will also turn out to be Heera. He doesn’t eat well. He only craves for dry food and junk food. People will back bite about my child just like Heera”.

24 years old participating mother from one of the intervention clusters

“My son had frequent diarrhoea like Sanimaiya’s child. If this programme wasn’t implemented, I am sure I would be living the life like Sanimaiya but these days I am doing all five behaviours and my child is also fine.”

30 years mother from one of the intervention clusters

“Yesterday night, my son, while calling (on phone) to his father, asked “Baba, did you take your dinner?” Father replied, “yes I did”, then son asked “did you wash your hands with soap properly before taking your food?”, father again replied “yes I did son”, then son again asked “did you also put on baby bib?”, I laughed a lot. Later, I spoke with my husband and mentioned about the programme and bib, he was quite happy”.

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“Kitchen makeover made my kitchen look so different—beautiful, bright and especial, I feel like sitting in the kitchen all the time and cleaning it over and again”.

*24 years old participating mother in one of the intervention clusters*

“We used to move in and out frequently to fetch water, wash dishes, throw waste water, wash hands etc. But after a bucket, tub and soap is placed inside the kitchen, the kitchen chores have become hassle free. Now, we can do it in the tub. It saves our time and energy. Whenever an outsider visits our home, he/she is also amazed to see the hand washing station inside the kitchen”.

*30 years old participating mother from one of the intervention clusters*

“During the initial days of the programme, I wasn’t concerned about the programme much. Eventually after many household’s kitchens were declared as “Clean Kitchen” and other mothers incorporated the five key behaviours in daily lives, I was scolded by fellow mothers for being the odd one out. My neighbours pressed me a lot and some of them encouraged too. I was ashamed of my own deeds and started to clean my house and practice five key behaviours. I felt the impulse to continue these behaviours after receiving rewards”.

*27 years old participating mother from one of the intervention clusters*

“Before the introduction of this programme, I used to take a nap during my spare time but nowadays, cleaning kitchen has become my hobby. I scrub the kitchen with mud and find different ways to make my kitchen clean & beautiful during spare time”.

*24 years old participating mother from one of the intervention clusters*

“Putting everything in order makes kitchen look beautiful and attractive and I’ll also get a reward and prize for winning ‘Clean Kitchen’ competition”.

*22 years old participating mother from one of the intervention clusters*

“One day, our goat’s head was stuck in a cooking vessel while accessing leftover food in the kitchen and we were about to cut the goat’s head but these days we can’t see them in kitchen anymore”.
“Yesterday, my daughter (A, a 3-years old daughter of participating mother) asked her grandmother when she started feeding “Oye Batulee! did you wash your hands with soap and water before feeding me?”

25 years old participating mother from one of the intervention clusters

“I was about to feed my child (B, a 4 years old son) but he became angry and told me ‘I won’t eat anything without putting on the baby bib, do you understand?’ Then I was so nervous and I never forgot to put it on afterwards”.

25 years participating mother from one of the intervention clusters

“My son had been suffering from intermittent diarrhoea throughout the year since last two years. As soon as I participated in this programme and started practicing the food hygiene behaviours, the diarrhoea has gone and hope will never happen again.

23 years old participating mother from one of the intervention clusters

“Before implementation of this programme, my child used to suffer from diarrhoea frequently. He had lost weight and always looked like sick. After the programme implementation, there has been no incidence of diarrhoea and he hasn’t been taken to traditional healers as well”.

32 years old participating mother from one of the intervention clusters

“My kitchen was so dirty before. Nowadays, I am so happy because flies do not appear in my kitchen and kitchen looks very clean and nice”.

26 years old participating mother from one of the intervention clusters

“I used to keep my goats and chicken inside the home near to kitchen but nowadays I have made a separator for them. This has made my kitchen clean and beautiful”.

22 years participating mother from one of the intervention clusters

“Before attending this program, I used to cook food in the same unwashed vessel for about two-three days, but nowadays I wash them every time when I cook food”.

30 years mother from one of the intervention clusters
“My mother-in-law never did those behaviours, I thought it’s difficult to change her attitude but after getting prize in Clean Kitchen Competition, she started making kitchen and surroundings clean as well as performing all five behaviours along with me. Nowadays, I found my house clean and bright after returning from school”.

26 years mother (teacher) from intervention cluster

“My kitchen looks so beautiful after makeover; I got confused if it was mine”.

29 years mother from intervention cluster

“My child had persistent diarrhoea during rainy season in last couple of years, but surprisingly in this rainy season he hasn’t got any diarrhoeal episode”

24 years old mother from intervention cluster

“My cooking vessels used to left around without cleaning for about 2-3 days as I was roaming around, taking rest without even thinking of washing them frequently, but now, I clean-up cooking as well as serving utensils more frequently because I want to make my son like Krishna”.

19 years mother from one of the intervention clusters

“After this programme, I have started doing these five behaviours for my own child’s future and reciprocally also getting certificate and prizes as well as my photo is put-up in the village as an ideal mother. Now I became the leader of our group too. I had never thought such will happen in my life and in our village”.

30 years mother from one of the intervention clusters

“I used to re-heat rice briefly in bowl, now I realized how much tasty it is when thoroughly re-heated using cauldron. Now onwards, I will always thoroughly reheat leftover food before I eat and serve to my child”.

27 years mother from intervention cluster

“I will make my son like Krishna (a character of ‘child’s life game’) as he was successful in his life because his mother practiced all five behaviours when he was child”.

33 years mother from intervention cluster
“This programme encouraged us to speak in public places, in mass gatherings and also to dance besides motivating to perform five key food hygiene behaviours”.

_26 years mother from intervention cluster_

“During this season last year, I had seen many people were being carried to hospital due to diarrhoea. Many people, especially from Singhe were seriously ill and they had to be rushed to the hospital immediately, whether they were in farm or fields. Such incidence is rarely seen this year. Even in my neighborhood, this season, the incidence of diarrhoea isn’t heard until now. I never thought that kitchen cleanliness and practicing such simple key food hygiene behaviours would do such wonders”.

_32 years mother from intervention cluster_

“Nowadays, if I don’t wash my hands with soap and water, I feel that there’s a huge amount of dirt and germs as we’d seen after putting Glo-germs. Therefore, I feel the urge to wash my hands every time immediately after handling something dirty and not only before feeding child”.

_28 years mother from intervention cluster_

“My child agrees to have meal only after putting on programme bib. He has even started to eat more food”.

_22 years mother from intervention cluster_

“I could have saved NRs 50,000 thousand if this programme had been launched last year because I had to spend that much money when my child was hospitalized due to severe diarrhoea and pneumonia last year”.

_27 years mother from intervention cluster_

“Flies are dramatically reduced in my house after kitchen makeover, now it feels good”.

_23 years mother from intervention cluster_

“I am sure the reason behind attaching these flags and ribbons are due to holy worship (Lakh batti baleko – lighting 100 thousand lights) in your home, looks so especial”.

_A relative of our participating mother_
“Our daughter-in-laws used to wash cooking vessels once a week before but now they have started washing them twice a day, this programme really helped to bring our in-laws in right track”.

60 years grandmother of participating mother

“Today we have excluded few mothers from our game because they occurred to hold disgusting play cards during hot potato game. I had a fear that community people will start excluding me in the same way as they did in this game because I didn’t win in the clean kitchen competition. I therefore, started cleaning my kitchen as well as requested my children to do the same in my absence”.

26 years mother from one of the intervention clusters

“Oh, the black spot on the 3M PetriFilm should be the reflection of flies defecating in food”.

40 years mother from intervention cluster during one of the events

“Boiling water is the easy task because we don’t have to be physically present while boiling water and we can also use the leftover coal”.

30 years mother from intervention cluster

“Hand washing with soap not only made my hands clean but also removed the dirt from cracked skins of the hands which was not yet possible by washing hands with water only”.

32 years mother from intervention cluster

Voices from secondary audiences:

“I have pasted the sticker ‘Do you want to be an Ideal Mother?’ besides my mother’s bed so that she won’t forget about “Ideal Mother” competition”.

10 years old daughter of a participating mothers from intervention cluster

‘I always clean animal (buffalo, cow) dung but never washed my hands with soap, I am not sure how much germs I had eaten before, but from today onwards, I promise to wash my hands with soap at least before eating so that I can’t eat them anymore”.

65 years old man after seeing germs in his hands using Glo-germ
“I am sure, this is the picture of my own house. I have placed milk and milk container without covering. I feel ashamed of myself. I need to go now and cover the container with proper lid”.

**While playing hot potato game, a non-participating mother from intervention cluster gets the play card of poor storage and immediately goes home to correct her behaviour**

“You have displayed the picture of all ‘Ideal Mothers’ in this village. Don’t you have the provision of displaying pictures of ‘Ideal Fathers’?”

**40 years old local from intervention cluster while displaying Ideal Mothers’ picture**

“My daughter-in-law is very much lazy, she never cleans kitchen so we didn’t get prize, but from today onwards, I will always clean my kitchen thereby to win the ‘Clean Kitchen Competition’. We are ashamed in front of all the villagers”.

**75 years grandmother from participating household after declaration of clean kitchen winner**

“There has been a sudden increase of queue in the nearby community tap stand nowadays because mothers bring utensils to clean every day. I thought this programme brought new things for them!”

**25 years man from one of the intervention clusters**

“After implementation of this programme, there has been very few incidence of diarrhoea among under five children in our village. I am bringing too less number of ORS packets and Zinc tablets from Sub-Health Post these days”.

**40 years old Female Community Health Volunteer (FCHV) of one of the intervention clusters**

**Voices from teachers / social leaders:**

“Continuous monitoring and motivation are the keys to make the programme successful and the success of this programme is the effect of these two keys”.

**Ex. hon’ble member of Constituent Assembly from respective constituency on his remarks during closing ceremony of the programme**
“All mothers (especially in-laws) in the village always felt reluctant to sing and dance in-front of male members of their family even in festive seasons/gatherings such as Teej (a holi festival for Hindu women), marriage ceremony etc, but it is surprising to observe that they can now dance, sing the song and also can give a lecture like a political leader in such public gatherings. This programme really empowered participating mothers in the village”.

A social leader from intervention cluster

“There used to be faeces all over the yard and children’s food were often accessed by chickens in village. Now, there has been a tremendous change which I never thought off”.

58 years local teacher from intervention cluster

“I had been seeing these behaviours since my childhood. I was afraid that mothers won’t change their behaviours. However, after implementation of this programme, I am surprised to see how mothers have been changing their behaviours”.

A local school teacher from one of the intervention households

“Like in urban settings, they had a hand washing station inside the kitchen which I found easy because we can wash hands inside even it is raining outside”.

35 years male relative of participating mother in intervention cluster

“I am very glad to observe that this programme not only helped to change the mothers’ behaviours but also sharpened the capacity of our sisters i.e. Food hygiene motivators”.

A social leader and project coordination committee member

“I have realized that changing people’s behaviour needs continuous effort and determination from programme team and people’s themselves”

A social leader and teacher from intervention cluster

“I am curious how you influenced our mothers through the programme so effectively. Mothers talked about this programme all the time- wherever they go. If we invite them for any meetings they hardly come, but if FHMs invite them, not only mothers but all villagers come”.

A principal, secondary school in intervention cluster
ANNEX H:

A case study

‘From a shy helpless mother to empowered volunteer leader’

On a breezy March midnight, a newly recruited mother for the study (before the intervention), was crying silently and her mother-in-law was consoling her. Her 16 months old child was on his death bed, he had become unconscious that night after suffering from intermittent diarrhoea for a month. The child used to throw out every time after eating and pass loose watery stool 4-5 times a day. He had even stopped eating or feeding on mother’s milk. “We had lost our hopes on his survival” recalled her mother-in-law. He miraculously survived that night and was rushed to traditional healer early morning then to nearby health institution later in the afternoon.

A shy 32 years old mother caring her children in a small room of a house (where she had to cook, feed children, sleep, and keep animals (goats, chickens inside), the dark spot in the kitchen and flies in and around are quite scary, the leftover food was half covered and it was difficult to separate the colour of rice due to flies accessing them) clearly had no idea about the major contributor of her child’s poor health nor did her family members. Upon questioning about why (before programme intervention), she amused us by telling “you must’ve thought that my child suffered from diarrhoea due to poor sanitary condition, but it’s not so. The whole village is like this, children are dirty but only my child suffered from diarrhoea. It’s due to Lagani/Nepaley (linking with evil eye). If not, how was my child cured (for some time) by the traditional treatment”? Despite the superstitious belief, she was baffled by the fact that her child who was born 3.5kg weighed mere 7kg at 16th month of his life. His growth rate had slowed down after the introduction of complementary food.

Her house lies at the midst of the Tamang settlement. They didn’t have toilet and family members defecating openly so did their children. She cleaned the cooking vessels only 2-3 times a week and cleanliness of serving utensils just before serving food was out of question. Handwashing with soap before feeding children was a far cry. She used to cook food twice a day and stored cooked food in an ambient temperature to feed her children four-five times a day. However, the storage of leftover food wasn’t proper. Flies were seen everywhere mostly
in the kitchen. The reheating of leftover food was not done in any seasons including winter and untreated water consumed from the nearby source.

It might be destiny or sheer luck, her cluster was randomly selected as a programme intervention cluster. During the programme kick-off, her child was again suffering from diarrhoea and she could relate herself to Sanimaiya story (a story telling activity; the lead character of a story recited during the event). “I thought Sanimaiya was me and my plight, my story is being told. It is a very heart touching story and the programme is exceptional too”. The story was an eye-opener to her and she realized that “five key food hygiene behaviours” were the formula for a healthy child. Then she determined not to miss any session of the programme and attended programme regularly. During first household visit, the makeover of her kitchen was done, kitchen area was demarcated and separated with ribbon, flags, kitchen cues were maintained and several eye danglers were put-up to make the kitchen look bright and beautiful. Moreover, she was very excited about her kitchen makeover and said, “very soon, I’ll bring all the necessary kitchen utilities and make my kitchen as beautiful as it is shown in the clean kitchen picture and Brahmin fellow mothers will not get chance to backbite me”. And she did it! She was among the few mothers who practiced all five behaviours within three weeks of programme kick off. In the next visit talk, she mentioned, scrubbing kitchen floor decreased the number of flies and it must be due to the practice of five key food hygiene behaviours that her children have stopped suffering from diarrhoea.

Not only that, she maintained clean kitchen by following all indicators and won both “Clean Kitchen” competition and “Ideal Mother” title in first time. Her family members though ignored initially became quite supportive when they realized that the child gradually started eating more food and reduced diarrhoea after practicing five key food hygiene behaviours. They were most captivated by the use of “Glo germs” in their hands to see dirt, germs and varying microbes in food at different stages through PetriFilm. Furthermore, she told that the programme is very interesting and attractive. She said “the games such as hot potato, role play, folk song, child’s life game, puzzle game are very fun and stimulate us a lot of things. I wish this programme had come earlier, I could have saved my first child”, she opined. The accumulated effort of all members of this sub-cluster had made it possible to declare this area as “Safe Food Hygiene Zone”.

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Her mother in-law was also very positive about the programme and supported her daughter in-law to participate in the programme, started practicing five key behaviours by family members and maintained a clean kitchen. She admitted that this programme brought a lot of positive changes in her life. She said “it not only made our children healthy but had other advantages too. My daughter in-law and other family members have become so enthusiastic about cleanliness of kitchen and surrounding. Our house has become cleaner than those of Brahmins. I feel very proud.”

She is now a volunteer leader of the sub-cluster (group) leaves no stones unturned to encourage mothers sustaining the five key food hygiene behaviours. She motivates them during every meeting, either at field, farms or elsewhere and goes for timely monitoring. She aspires to make her child like “Krishna” and forever indebted to “Safe Food, Healthy Child” trial that changed her life completely.

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ANNEX I: Relevant images from food hygiene intervention trial

First Community Event

1. Story telling: sanimaiya’s story

2. Situation analysis using pictures

3. Video show – situation contextualization

4. Introduction of five key food hygiene behaviours

5. Mothers ways of realizing five key behaviours

6. Public commitments – oath taking

7. Public rally chanting - “safe food, healthy child, we want ideal mother”

8. Commitment certificate distribution
First Household Visits

1. Kitchen demarcation using bunting (kitchen separation using ribbon and flags showing each behaviour)

2. Eye danglers placed at eye-level (round illustration of all behaviours, ideal mother board, dhungro – a branded fire blowing instrument).

3. Change in kitchen settings: transformation of kitchen environment in to new, beautiful, bright and clean kitchen settings

4. Importance of food hygiene behaviours refreshed via a brief talk with 3D flip chart
First group event

1. Programme ritual such as cutting nail, washing hands with soap etc at the beginning of the event

2. Mothers' experiences of changes in their kitchens shared.

3. Group norms elicited via cooking demonstration

4. Use of Glo-germs before feeding

5. Bibs distributed as reminder/reward for HWWS (bib message - did you wash your hands before feeding me?)

6. Visual aids demonstration (3M PetriFilm)
Second household visits

1. Importance of five food hygiene behaviours reiterated
2. Use of plastic bucket for handwashing in kitchen
3. Installation of programme song ring tone in mother’s mobile
4. Use of bib before feeding children re-enforced
5. Food hygiene motivator explaining the importance of eye dangler
6. Poster including illustration explaining clean kitchen competition indicators disseminated in each HH
Second group event

1. Second group event – programme venue including back-drop banner

2. 'Child Life Game' played - nurture motive

3. Folk song composed by mothers conveying key food hygiene messages - affiliation elicited

4. Mothers singing and dancing in folk song competition

5. Puzzle game played to encourage kitchen cues (social respect motive)

6. Public pledging for folk song competition winner group
**Third household visits**

1. Mothers' food reheating practices observed

2. Noted re-heated tempt and encouraged mothers to maintain appropriate temp

3. Benefits of thorough re-heating communicated showing 3M PetriFilm
Third group event

1. Game (hot potato game using safe pictures – demonstrated social inclusion)

2. Game (hot potato game using disgusting pictures – demonstrated social exclusion)

3. Disgust exercise using Glo-germs in serving utensils

4. Mothers participated in disgust exercises (Glo-germs used in food, plate, bowl, glass, spoon)

5. 'Clean kitchen' competition winner announced and pledged

6. Participants / guests visited winners’ house to encourage and share learning.
Fourth household visits

1. Importance of clean kitchen reinforced

2. Reminding mothers about 'ideal mother' and 'safe food hygiene zone' competitions

3. One of the clean kitchen winner’s household

4. Handwashing station in one of the participating household

5. Un-identical visits performed (by field staffs and coordination committee).
Fourth group event

1. Dhukhimaya’s letter (innovative letter exchange)

2. Mothers participating in role play (drama) exercise

3. 'Ideal mother' competition winners announced

4. A drama (family member role play) to become an ideal mother avoiding social, attitudinal and physical barrier

5. 'Ideal mother' competition winners publically commended

6. Ideal mother photo put-up in the junction of the village = prestige conferred
Second Community Event (closing)

1. Guests attending second community event

2. Mothers offered garlands and tikas to study team during closing event

3. Public commitments from mothers to sustain behaviours

4. Group photo. All mothers received appreciation certificates during closing event

5. Appreciation certificates distributed to project coordination committee members

6. Mothers participating in community event

7. Public rally chanting - “we want ideal mother, ideal mother hi-hi, diarrhoea bye-bye”

8. Rally after declaration of ‘safe food hygiene zones’ and bill boards erected at each entry point of cluster
Microbiological assessment in food and water samples:

1. Samples collection in field
2. Sample transportation
3. Sample homogenization in lab
4. Sample processing in lab
5. Multiple serial dilutions
6. Inoculation
7. Incubation
8. Colony count: *E. coli* and total coliforms
Annex J: Collaboration with Child Health Division, MoHP Nepal

To,
Mr. Om Prasad Gautam
PhD Student
London School of Hygiene and Tropical Medicine (LSHTM)
London, UK

Subject: Collaboration on ‘Food hygiene intervention to reduce diarrhoeal diseases burden and food contamination in Nepal’ study.

Dear Mr. Gautam,

First of all, I am very pleased to hear that you are doing PhD in London School of Hygiene and Tropical Medicine (LSHTM), London, UK.

We have reviewed your e-mails about the forthcoming research study in Nepal on ‘Food hygiene intervention to reduce diarrhoeal diseases burden and food contamination in Nepal’ as part of your PhD research. The study which you want to do and the research questions you have prepared are very much relevant in the context of Nepal’s diarrhoeal diseases control initiatives as well as other foodborne infections. We also feel that the intervention which you will be developing as part of your research design will be useful to any food hygiene promotion initiatives as well as diarrhoeal disease control programme. Diarrhoeal diseases are still the second leading cause of childhood mortality in Nepal and food hygiene behaviours possibly playing role to make high mortality and morbidity. Therefore, the findings of your study and intervention design will be very much useful to our on-going diarrhoea disease prevention programme.

In this context, Ministry of Health and Population, Child Health Division would be very much happy to support for your study and would like to collaborate in this research. As part of our collaborative action, MoHP/CHD will not cover any financial support but we will provide technical support which you may need/required.

I hope this study and collaborative action will add significant contribution in policy and programmatic design in diarrhoeal diseases control program in Nepal.

I wish you good luck with your PhD work and research.

Looking forward to work together!

With regards,

Dr. Shyam Raj Upreti
Director
Child Health Division, DoHIS
Ministry of Health and Population, Teku, Nepal
ANNEX K: Appreciation letter from collaborator

It is our great pleasure to appreciate Mr. Om Prasad Gautam, PhD Fellow at London School of Hygiene and Tropical Medicine, UK for the successful implementation of “Food Hygiene Intervention Trial” in Kavre District, Nepal during 2012-2013. We are very much thankful to Mr Gautam for selecting Kavre District as one of his study site and implementing highly needful public health campaign targeting to mothers to improve their food hygiene behaviours.

This collaborative work between Mr. Om Gauatam and Ministry of Health and Population, Child Health Division and Central Regional Health Directorate as well as local health institutions has made significant improvement in the village to improve behaviours and in fact reduce workload of local health institution regarding the treatment of diarrhoeal cases.

As compared to past years, very few diarrhoeal cases have been reported this year from the respective villages where the intervention was implemented. This is a remarkable impact of the trial. This trial not only made public health benefits but also change certain rooted social norms in the village relating to food hygiene practices. During monitoring visits, we found that villagers’ kitchen were much more cleaned, they were following and practicing key food hygiene behaviours. The learning and evidences from this study will contribute to enhance diarrhoea control efforts especially to address “food hygiene” aspects.

We would also like to highlight that our collaboration with Mr. Gautam’s ‘food hygiene intervention trial’ study was fruitful to minimize evidence gap, enhanced capacity and generate new knowledge to improve household food hygiene in Nepal.

On behalf of Ministry of Health and Population, Child Health Division, we would like to present a letter of appreciation to Mr. Om Prasad Gautam for his valuable contribution as well as for successful completion of the trial.

We wish him a very successful completion of his PhD and best of luck for his future endeavour,

Sincerely,

Dr. Shyam Raj Uperti
Chief, EPI, Child Health Division
Ministry of Health and Population, Nepal

Date: 07 January 2014
ANNEX L: Appreciation letter from local stakeholders

Letter of appreciation

Mr. Om Prasad Gautam, PhD student at London School of Hygiene and Tropical Medicine, UK successfully implemented ‘Food Hygiene Intervention Trial’ study in Baluwapati Deupur and Nayagaun VDC, Kavre, Nepal dated on 2013 AD. We are very much fortunate that Mr. Gautam selected our VDC as one of his study site and implemented such a noble, creative and highly needful public health campaign mainly targeting to mothers to improve their food hygiene behaviours. We are very much pleased with Mr. Gautam because he involved Ministry of Health and Population, District (Public) Health Office, Local Health institution since the inception phase of the trial. We have learned a lot from him on food hygiene and our capacity has been enhanced on the aspect of implementing behaviour change programme.

The trial has made significant differences in the village and in-fact reduce our workload to treat diarrhoeal cases. As compared to past few years, we are getting very few diarrhoeal cases this year because of the impact of the trial. This trial not only made public health benefits but also change certain rooted social norms in the village relating to food hygiene practices. During our monitoring visits, we can see that, villager’s kitchen are much more clean nowadays, they have been following & practicing key food hygiene behaviours. Villagers together with local institutions including sub-health post (SHP), village development committee office (VDC) and all local political parties also took an ownership on the programme for the overall social, economic and public health benefit of the people. Mr. Gautam together with his team put greater effort to motivate mothers to change their unhygienic behaviours. In addition, three months intervention trial provided a greater opportunity for all of us to participate, monitor, learn as well as to change our own mindset on the ways to implement behaviour change programme. In this context, we would also like to inform you that, we will use our enhanced capacity and new knowledge on food hygiene in other areas too.

On behalf of Ministry of Health and Population, District (Public) Health Office, Baluwapati Deupur Sub-Health Post, Kavre, we would like to present a letter of appreciation to Mr. Om Prasad Gautam for his valuable contribution as well as for successful completion of the trial.

We wish him a very successful completion of his PhD and best of luck for his future endeavour.

Sincerely,

Biddhaya Tamang
In-charge
Baluwapati Deupur Sub-Health Post
Baluwapati, Deupur, Kavre, Nepal
Date: 09 December 2013
ANNEX M: Appreciation letter from local stakeholders

04 December 2013

To whom it may concern,

This is to certify that Mr. Om Prasad Gautam successfully conducted ‘Food Hygiene Promotion Intervention Study’ in Baluwapat Deupur and Nayagaun VDC, Kavre, Nepal as part of his PhD research field work.

Since the beginning of the study, Mr Om established a very good coordination with all local stakeholders and ensured strategic involvement of the stakeholders in the intervention clusters. With Mr Om’s immense knowledge on public health & social mobilization skills as well as the efforts made by his dedicated team in the field, the trial was successfully conducted in various wards of Baluwapat and Nayagaun VDC, Kavre district, Nepal. We are very much glad to observe that, the trial made huge differences in mother’s food hygiene knowledge and behaviours in the village. As a result of implementation of motivational package, majority households / mothers were able to win the clean kitchen competition, ideal mother competition as well as safe food hygiene zone declaration title. On behalf of project coordination committee and VDC office, we thank Mr Om for the opportunity he has provided for all the coordination committee members as well as VDC staffs to closely observe, and monitor the programme during the implementation period. We are very much confident that, the trial will definitely produce better results.

On behalf of Ministry of Local Development, Village Development Committee as well as Project Coordination Committee, we would like to thank Mr Om for the implementation of his study in our highly deprived settings and making differences in people’s life. We would like to congratulate Mr Om for the successful completion of the trial in the field.

In this regards, we wish him for the very success in his future work and looking forward to his arrival in Nepal after completion of his PhD from such a prestigious institution in UK.

Sincerely,

Tharu Narayan Bhandari
VDC Secretary,
Baluwapat Deupur VDC
Kavre, Nepal