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Christopher W. Wegner

Date

Human and Animal Behaviors as Risk Factors for Diarrheal Disease in Rural Madagascar

By

Christopher W. Wegner Master of Public Health

Environmental Health

Thomas R. Gillespie, PhD Committee Chair

> Paige Tolbert, PhD Committee Member

> Sarah Zohdy, PhD Committee Member

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By

Christopher W. Wegner

B.S. University of California, Irvine 2010

## Thesis Committee Chair: Thomas R. Gillespie, PhD

An abstract of A thesis submitted to the Faculty of the Rollins School of Public Health of Emory University in partial fulfillment of the requirements for the degree of Master of Public Health in Environmental Health 2013

#### Abstract

## Human and Animal Behaviors as Risk Factors for Diarrheal Disease in Rural Madagascar By Christopher W. Wegner

**Purpose**: Ecological factors and human and animal behavior play a key role in facilitating transmission of enteric waterborne and zoonotic pathogens in low- and middle-income countries. While a number of studies have focused on epidemiological or environmental determinants of infection, the influence of human behavior on disease transmission has been less investigated. To address this gap in research we examined transmission dynamics using a mixed-methods approach focusing on water, sanitation, and hygiene behaviors commonly associated with increased risk of enteric waterborne and zoonotic disease transmission.

**Methods**: From June to August 2012, survey questionnaires asking about basic demographics and water, sanitation, hygiene, and livestock practices were given to villagers (n=190) in three rural villages of Ranomafana National Park, Madagascar. Concurrently, ecological structured observations of four key transmission events were conducted. Chi-square tests, univariate regression analysis, and multivariate regression analysis were performed to test for associations with reported diarrhea in the past four weeks.

**Results**: Villagers in Ambodiaviavy (23.1%) reported the highest prevalence of reported diarrhea, followed by Ambatolahy (19.2%) and Ankialo (17.1%). Univariate regression analysis revealed significant (p-value < 0.05) association between various risk factors (i.e. medication practices, defecation practices, water treatment, foot hygiene, etc.) and reported diarrhea. Multivariate regression analysis showed animals given medication (OR=5.71, 95%CI-2.01, 16.22), not treating drinking water (OR=19.13, 95%CI-2.92, 125.20), and trading with other villages (OR=5.83, 95%CI-1.39, 24.49) had highest effect on reported diarrhea adjusting for other covariates. Observations shed light on known and unknown transmission pathways.

**Conclusions**: Our findings suggest that differences in human hygiene and livestock interactions lead to a disproportionate incidence of diarrhea, with direct observations revealing previously unknown pathways of transmission within these rural villages. By using direct observations in conjunction with survey collection we gain a better understanding of waterborne and zoonotic disease transmission dynamics in this rural African setting. In summary, these methods allow close examination of infectious disease transmission, and demonstrate the utility of a mixed-methods approach in elucidating risk factors for diarrheal disease in rural Madagascar.

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## **INTRODUCTION**

Diarrheal disease caused by waterborne and zoonotic enteric pathogens has become an arresting global public health problem, yet it is not completely understood how, when, and why human enteric infections occur in high risk communities. We know that enteric pathogens have found a comfortable transmission *modus operandi* at the human, animal, water and food interface. This niche utilizes a variety of commonly known mechanisms and feedbacks to support transmission cycles in susceptible communities afflicted by outbreak, sporadic, and endemic conditions. Some of these mechanisms include ecological and social drivers, gene-environment interactions, and life-course trajectories[1].

However, scientific understanding of these mechanisms and their role in supporting waterborne and zoonotic disease transmission is still evolving[2]. Likewise, the emergence and re-emergence of waterborne and zoonotic diseases is welldocumented, yet sources of most gastrointestinal infections remain unknown despite popular conceptions regarding infectious disease etiology[2]. Moreover, the influence of interacting risk factors that facilitate disease transmission and diarrheal disease infection in diverse settings has been less investigated. How human behavior, human relationships with livestock and domestic animals, livestock and domestic animal behavior, water and food all interact to fuel transmission dynamics at the community level is very complex and not fully understood. Attempting to explain disease by observing each of these risk factors separately only gives a one-dimensional understanding of how transmission dynamics work, yet even scientific knowledge of these risk factors alone is still not completely understood. Our ability then to pinpoint specific human and animal behavior predictors from a web of these interacting risk factors is lacking at its most fundamental level. This gap in knowledge needs to be addressed in order to better understand and potentially mitigate the global burden of diarrheal disease in different cultural and environmental settings.

In low- and middle-income countries where enteric bacterial and parasitic pathogens are the leading cause of infectious diarrhea[3], rural communities may be particularly vulnerable to endemic waterborne and zoonotic enteric disease. A system of interacting components including anthropologically influenced landscape ecology, human ecology, and human-created environments, coupled with natural perturbations and natural disasters mediates transmission cycle dynamics. These intermediate steps causally modify disease, but infection is also concomitantly influenced by the populations and traits of humans, livestock, and domestic animals[1]. For instance, human and livestock interaction frequency vary according to their respective behavioral patterns in addition to their socially and culturally defined relationship[4]. The behavioral patterns of humans and livestock are what fundamentally cultivate that crucial moment when exposure to a pathogen takes place or when a transmission cycle is facilitated to persist through space and time. For instance, a villager might drink water straight from a pump that is contaminated or give that water to livestock who might then become infected. Likewise, transmission between humans and animals (livestock and domestic animals) may be further enhanced through multiple environmental reservoirs including water and soil when poor economic and water, sanitation, and hygiene conditions exist. This situation is even further complicated by high risk sub-groups, such as children under the age of five

who may be exposed to a greater enteric waterborne and zoonotic pathogen load due to age related behaviors such as geophagy[4]. Unfortunately, many low- and middleincome countries have yet to achieve the separation of drinking water or food from sewage, demonstrating our current understanding of this phenomenon as a pale reflection of the true size of the problem[5].

From a prevention perspective, it is crucial to begin developing an understanding of waterborne and zoonotic enteric disease transmission dynamics in low-income rural communities; and to determine the most probable pathways and routes of transmission that result in diarrheal disease. If possible, determining the transmission potential from all sources and interactions can tell us something about where to appropriately apply control and prevention strategies in rural communities with endemic diarrheal disease. The island of Madagascar is famous for its biodiversity and high degree of endemism, once containing the highest number of primate species than any other region of its size in the world [6, 7]. In the rural southeastern region of Madagascar is Ranomafana National Park (RNP), one of Madagascar's best-known and most well studied park in the country. This park is home to flora and fauna that can be found in no other place on the planet. In addition, it serves as a model for subsequent parks and reserves, both in the country and abroad. However, rural villages in and around the Ranomafana rainforest, including other large parts of the island, are plagued by diarrheal disease with relatively unexplained etiology and transmission risk factors. These villages are surrounded by pristine forest, forest fragments, and forest-village eco-tones where human, livestock, wildlife, and domestic animal interaction frequency vary according to ecological and social drivers of health. Poor water, sanitation, and hygiene practices are rife in these villages and there is

an extreme lack of access to needed healthcare[8]. This created a rare opportunity to understand waterborne and zoonotic enteric disease transmission dynamics and explain the causal risk factors of diarrheal disease in these villages.

Anthropological and epidemiological research on zoonotic pathogens among people, livestock, and water in and around RNP will shed light on the explanation, prevalence, and severity of this issue while informing rational public health and conservation intervention strategies in this region and similar regions around the world. A mixed-methods approach is being employed to ensure valid and efficient measurement of behavior risk factors related to transmission dynamics and syndromic diarrhea. In this study our goal is to integrate two complimentary perspectives in order to reduce bias and develop a degree of counterbalancebetween the separate quantitative and qualitative approaches. As part of a long term initiative in RNP, by studying the condition and daily lives of the communities in RNP we hope to improve their health and wellbeing, as well as that of the livestock, wildlife, and environments they depend on. If successful, this study may influence future studies and intervention programs in RNP.

## BACKGROUND

## **Infectious diseases**

Infectious diseases are ubiquitous throughout the earth and have a major impact on every living creature. Today, they are a continuing danger to everyone – remaining among the leading causes of death and disability worldwide[9]. The emergence and reemergence of infectious diseases, likewise, creates a significant burden on global economies and public health[10]. As such, a little over 25% of the 57 million annual deaths that occur worldwide are estimated to be directly related to infectious diseases, which does not include death that occurs as a consequence of past infections or complications associated with chronic infections[9]. Typically, those at an economic disadvantage in low- and middle-income countries are disproportionately affected by the burden of morbidity and mortality associated with infectious diseases. This health disparity is predominantly felt by high-risk groups such as infants and children, as well indigenous and disadvantaged minority populations[9].

#### **Enteric waterborne and zoonotic diseases**

Infectious diseases take on many shapes, sizes, and forms. Chief among them are emerging and re-emerging infectious diseases, which are infectious diseases that are new to a population or have already been occurring in a population, but are increasing in incidence or geographic range [11]. The reasons behind why and how infectious diseases either infiltrate a new host population or increase in dissemination have been widely discussed in the literature [5, 9-11]. Between 1940 and 2004 though, 60.3% of the 335 emerging infectious disease events were caused by zoonotic pathogens, infections in animals that are transmitted to humans[9, 10]. In fact, 64% of the > 1400 documented human pathogens are considered zoonotic[12]. Zoonotic diseases are caused by a wide variety of bacterial, viral, and parasitic pathogens, which all have multiple transmission characteristics. For instance, many zoonotic pathogens possess biological features that permit them to be waterborne or foodborne pathogens as well. Waterborne zoonotic diseases such as Giardia, Cryptosporidium, Campylobacter, norovirus, rotavirus, and *Escherichia coli* result in over 3.5 million deaths per year[13]. Indeed, foodborne zoonotic diseases affect one-third of the global population where some of the most important emerging infectious diseases of this kind include Salmonella, Campylobacter,

enterohaemorrhagic, *E. coli*, *Toxoplasma* and *Cryptosporidium*[14]. These diseases share common risk factors and are the enteric pathogens responsible for much of the diarrheal disease in developed and developing countries. To compound matters, the past ten years have seen an increase in the number of zoonotic viral disease worldwide[12].

#### **Clinical features of diarrheal disease**

Diarrhea is an alteration in the natural bowel movement typically characterized by an increase in water content, volume, or frequency[15]. Diarrhea can be further classified into distinct syndromes, which include dysentery, acute diarrhea, and diarrhea with extended duration[16]. Acute diarrhea is typically defined as 3 or more soft or liquid bowel movements within 14 days or less. Persistent diarrhea usually lasts 14 days or more. Clinical features can include fever, abdominal pain, bloody stool, nausea, vomiting, fecal evidence of inflammation, and heme-positive stool[15]. Clinical clues may suggest a specific enteric pathogen and aid in attributing distinct and interacting risk factors to communities with emerging or re-emerging diarrhea disease. For instance, persistent diarrhea lasting over fourteen days should prompt an investigation into *Giardia* and *Cryptosporidium* regarding known reservoirs, potential vehicles, common sociocultural and behavior risk factors, as well as socio-economic and ecological drivers[15].

#### **Global diarrheal disease**

Infectious diarrhea is one of the leading causes of morbidity and mortality in developing countries. Responsible for an estimated average of 3.2 diarrheal episodes annually and 2.5 million deaths per year in children under the age of five, diarrhea clearly places a disproportionate burden on the health of this high risk group[15, 17]. The burden of diarrheal disease lays mainly in South Asia and sub-Saharan Africa where roughly 90% of deaths are due to pneumonia and diarrhea, totaling 38 million annual diarrheal DALYs (Disability Adjusted Life-Years) and 26 million annual diarrheal DALYs respectively[18, 19]. However, estimated diarrhea-proportional under-five mortality due to diarrhea is the largest in Africa demonstrating the substantial health onus due to diarrheal disease in this region of the world[20]. In Africa, diarrheal disease is ranked in the top four diseases of burden for all four regions of the continent. In the Southern region it is ranked 3<sup>rd</sup> while in the Eastern region it is ranked 4<sup>th</sup>[21].

#### **Risk factors for diarrheal disease transmission**

Globally, approximately 90% of deaths due to diarrhea have been attributed to unsafe water, inadequate sanitation and poor hygiene[19]. This is troubling considering over 780 million people are without access to improved sources of drinking water and 2.5 million lack improved sanitation[22]. 653 million of those without access to improved sources of drinking water, however, live in rural areas. In addition, 1.8 billion rural dwellers (72% of global total) lack access to improved sanitation, driving home the significance of urban-rural disparities[23]. Understanding the convergence of water, sanitation, and hygiene practices in rural areas – specifically in Africa where diarrheal disease is most prevalent and where a preponderance of mortality transpires – and concurrent changes in human behavior and animal (livestock and domestic) relationships is paramount to the sustainable reduction in global diarrheal disease.

Determinants of infectious disease morbidity and mortality include both distant and proximal level risk factors. Distant level factors include socioeconomic factors, such as income, social status, and education, which are mediated by environmental and behavioral risk factors. These factors are, in turn, causally linked to more proximal level risk factors of mortality, such as undernutrition, infectious disease, and injury[24]. Human behavior however, plays a central role in the epidemiology of waterborne and zoonotic enteric disease transmission. There is a bewildering array of cultural, religious, ethnic, age, and gender related variables that influence and complicate human behavior in every part of the world. This can include personal hygiene, the use of proper sanitary infrastructure, and appropriate treatment of drinking water. Contamination of water and food sources by human waste and the simultaneous ingestion of these contaminated necessities of life further complicate and fuel acquisition and dissemination. In addition, close association with domestic animals and livestock provide greater opportunities for transmission. Animal behavior and the role of animals in different societies vary widely, which affect transmission dynamics. Also, the beliefs around food, how they are prepared and different eating habits determine the range of pathogens communities may be exposed to[4].

For instance, rotavirus provides a perfect example for zoonotic disease transmission dynamics. In many low- and middle-income countries, close contact between humans and livestock and domestic pets, including dogs and cats, can increase the chances of zoonotic enteric viral pathogen acquisition. Close contact can be exacerbated by a monsoon climate, rain, and flooding, which can increase exposure to potentially infected animals. Furthermore, it is common for farmers to come into direct contact with livestock and domestic animal feces, dust, and effluent. The lack of proper handwashing and hygiene behavior overall, no doubt plays a role in these vehicles of transmission causing directly or indirectly 5.5 million infections per year just through farm workers' exposure to animal rotaviruses[25]. However, children are also a high risk group for these types of events as it was concluded in one study that children were the reservoir for rotavirus infections in humans[26]. Despite these findings and their relevance to similar zoonontic diseases, still very little is known regarding how human behavior, livestock and domestic animal behavior, and environmental vehicles interact to create short- and long-term disease transmission cycles.

#### Survey versus structured observation methodology

The type of instruments most commonly used to study risk factors related to diarrheal disease have traditionally been quantitative methods such as survey questionnaires[27]. Questionnaire-based surveys though, may be inadequate at gathering data related to food and handwashing practices, and have been found to overestimate rates of handwashing[28, 29]. In general, this self-reported style of investigation may be limited in its efficacy, scope, and accuracy for studying human behavior, as it can only render an imperfect or partial picture of disease and its etiology[30, 31].

What quantitative data lacks is where qualitative data thrives. Methods such as direct structured observations bring realism to applied research and primary data collection. They also allow use to change directions as new information comes to light that places our emphasis on a new aspect of the study[31]. This flexible study design allows the researcher to place attention, as well as the amount of attention, on a constellation of social, cultural, religious, and behavioral variables that a survey is not capable of capturing[31].

Observations have been used before in hygiene behavior studies with varying levels of success [27, 30, 32, 33]. Once an observer has been properly trained and standardization has been completed between observers, observation techniques such as spot-checks can be time-saving and a sound economic alternative [33]. Generally observers use a line-item checklist of behaviors, or behavior proxies in order to be less invasive[33].

Ethnographic excursions into infectious disease transmission and diarrheal disease should be utilized more often to investigate and uncover environmental-behavioralcomplexes that are difficult to quantify. Such a technique can complement environmental epidemiological research and bring the researcher close to the people and phenomena that make up the daily practices that shape social action[31].

#### Madagascar

The Republic of Madagascar is the fourth largest island located off of the east coast of Africa in the Indian Ocean. The majority of its 22,005,222 people live in rural areas (71%) and earn their living fishing, farming, and herding zebu cattle[34]. It's been estimated that roughly 90% of the country's native vegetation has been burnt down due to the slash and burn technique used to clear brush and forest for crop production[7, 35]. This level of deforestation and subsequent erosion pose major threats to Madagascar's rich biodiversity of the forests, including influencing forces on that of its population's health and economy.

With 50% of its population living below the poverty line, access to health care poses a serious challenge [36]. The most common causes of morbidity and mortality in Madagascar include lower respiratory infection, malaria, and diarrhea, accounting for 51% of deaths among children under the age of 5[34, 36]. It's been estimated that the number of child deaths (under 5 years of age) is approximately 93,000 [24]. Life expectancy at birth is 66 years and the annual number of under-5 deaths is estimated at

44,000[37]. 41% of the total population use improved drinking water sources (rural-29%, urban-71%), while only 11% of the total population use improved sanitation facilities (rural-10%, urban-15%)[37]. According to the 2003-2004 Demographic Household Survey, 10% of children under five had one or more episodes of diarrhea in two weeks preceding the survey[36]. Despite the substantial burden of diarrheal disease on the Malagasy, very little research has been conducted to quantify incidence or understand the risk factors and etiology of this disease.

#### Ranomafana

The population health for the site of the current study, Ranomafana National Park, has been the site for health investigations previously. In 1995, Kightlinger et al. found ascariasis and trichuriasis to be common in 18 rural rainforest villages where conditions conducive to transmission and persistence, such as inadequate sanitation, poor hygiene, lack of footwear humid tropical climate, inaccessible health care, and poverty were evident[8]. The girls were observed to have higher egg and worm counts, and significantly higher prevalence and mean worm burdens than the boys. Children were generally more heavily infected, which was most likely due to increased exposure to infective eggs[8]. In both scenarios distinct gender role difference and age specific behaviors played a role.

In 1997, Hardenbergh studied dietary and anthropometric data collected from 0 to 9 year old children (n=613) from two adjacent cultivator communities[38]. Males were found to fall below the international growth reference cutoff indicating chronic undernutrition and poor health. Their diet lacked calories, riboflavin, niacin, and calcium from the high carbohydrate, low fat, and minimal protein intake. Hence, boys' intake was concluded to be insufficient relative to the international recommendations. Overall, the diet in RNP was found to be deficient in many areas with respect to international recommendations and activity levels[38].

The most recent study was published by Randremanana et al. from the Pasteur Institute in 2012[39] They assessed the etiology of infant diarrheal disease across fourteen districts in Madagascar. This cross-sectional study utilized survey interviews and collected clinical data and fecal samples from 2,802 children with a median age of 20.3 months. A majority of these children presented a diarrheal syndrome where *Giardia lamblia*, *Ascaris lumbricoides*, *Trichomonas intestinalis*, *Entamoeba histolityca*, *Campylobacter*, *Escherichia coli*, and rotavirus were all prevalent in the sample population[39]. This study highlighted the need for active surveillance programs on the island and further studies on the risk factors facilitating disease transmission cycles and infection.

The present study is based off a pilot study analyzed and reported by Bodager in 2012[40]. This study was conducted in the summer 2011 and set out to determine specific ecological and behavioral factors associated with diarrheal disease in two rural communities in RNP, Ambodiaviavy and Ankialo. Surveys were given to each subject in the ten random households chosen to partake in the study. In addition, sample collection of fecal material from each subject who was surveyed, as well as a variety of livestock and domestic animals, were conducted and analyzed for *Cryptosporidium* by Bodager[40]. Results showed risk factors related to diarrhea in the human population included water use, meal-time practices, and defecation practices. 14.7% of the subjects in Ambodiaviavy and 12.5% of the subjects in Ankialo tested positive for

*Cryptosporidium*. Lastly, there was evidence of zoonotic disease transmission between humans and pigs in the Ankialo community[40].

This present study looks to improve upon this pilot study by delving into more specific behavioral risk factors that we believe directly influence and fuel waterborne and zoonotic enteric disease transmission in these communities. In order to do so, we purpose that using a mixed-methods approach with an improved survey questionnaire and direct structured observations will capture a more realistic and deeper understanding of why, how, and where infectious disease transmission takes place. This study is vital to painting a more accurate picture of what goes on in these villages and will assist in future studies, as well as in long-term health projects. Findings from this study may also elucidate the validity and utility of using qualitative methods in investigating social-cultural and behavioral risk factors of infectious diseases. Global environmental health investigations into infectious disease risk factors would certainly benefit from qualitative methods that give a voice to individuals and communities.

## **PROJECT AIMS AND OBJECTIVES**

In Madagascar, diarrheal disease is a tremendous burden to the health and wellbeing of its native population and a potential force influencing the economics and ecology of infectious disease[41]. Regrettably, the etiology and risk factors facilitating transmission of diarrheal disease causing pathogens has not been rigorously investigated on the island. This represents a formidable challenge to the Malagasy and a situation in which the lack of knowledge on risk factors associated with waterborne and zoonotic enteric disease transmission, the agents responsible for infectious diarrhea, also reflects what is known at a global scale. Rural villages in Ranomafana are subjects of their historical and economic predispositions, as well as their own ecological systemization and socio-cultural fabric of society. Infectious diseases appear to be rife in a very accommodating setting where a perfect storm of environmental, behavioral, and economic factors facilitate waterborne and zoonotic enteric disease transmission.

Furthermore, the methods used to investigate this phenomenon have traditionally utilized household survey questionnaires, which, used alone, can be limited in its efficacy, scope, and accuracy[30]. The use of direct structured observations has been found to be a valid and useful method for determining the frequency of different behaviors and can serve as a qualitative compliment to other quantitative and qualitative methodologies[30].

The aim of this study is to examine transmission dynamics using a mixed-method approach focusing on water, sanitation, and hygiene behaviors commonly associated with increased risk of waterborne and zoonotic enteric disease transmission. Using behavioral survey data and direct structured observations in three rural communities in Ranomafana National Park, Madagascar, we examined hygienic behavioral patterns, livestockhandling practices, and underlying environmental factors and their association with syndromic diarrhea. We utilized survey data on water collection and use, sanitation, hygiene, livestock, food preparation, and health practices, and structured observation data on human sanitation, hygiene, water and food practices and livestock behavior. By using direct observations in conjunction with survey collection we gain a better understanding of waterborne and zoonotic disease transmission pathways and narrow in on conventional and uncommon co-determinants that facilitate this process.

#### **Objectives and hypothesis**

*Objective 1*: To determine if human behaviors are associated with human diarrheal disease in three rural villages via survey questionnaire.

H<sub>o</sub>: There are no human behaviors associated with human diarrheal disease in the study villages.

*Objective 2*: To determine if observed human behaviors are associated with diarrheal disease prevalence in the study villages via structured observations.

H<sub>o</sub>: The rates of observed human behaviors are not related with diarrheal disease prevalence in the study villages.

*Objective 3*: To determine if observed livestock and domestic animal behaviors are associated with diarrheal disease prevalence in the study villages via structured observations.

H<sub>o</sub>: The rates of observed livestock and domestic animal behaviors are not related with diarrheal disease prevalence in the study villages.

*Objective 4*: To determine if structured observations will reveal previously unknown transmission pathways or routes in the study villages.

H<sub>o</sub>: Structured observations will not reveal previously unknown transmission pathways in the study villages.

#### **Relevance to environmental public health**

The field of environmental health is rife with co-interactions and intertwined hazards between humans, animals, and the environment that in-turn have short- and longterm effects on the health of the these very same humans, animals, and environments. If allowed to go unchecked, the accumulating and synergistic results from these forces has the potential to further increase the imbalance at a local level, but may extend to adjacent populations under the right conditions. Diarrheal disease is an infamous harbinger of morbidity and mortality in Madagascar and in many parts of the world. Studying the antecedent causes of waterborne and zoonotic disease transmission will increase the knowledge about how the interface of environmental and behavioral risk factors influence proliferation of this pressing environmental health related disease. Infectious diarrhea has been externally linked to similar environmental and behavioral risk factors that determine whether or not diarrhea becomes endemic in rural communities that may have very diminutive access to healthcare. Furthermore, direct observations to understand of how humans and animals behave within their built environment and landscape can open up new avenues of investigating environmental health issues.

#### **METHODS**

#### Study area

The study took place in Ranomafana National Park (RNP), a 43,500-hectare World Heritage Site (21°02'–21°25' S and 47°18'–47°37' E) [7, 42]. RNP resides in a southeastern submontane rainforest area of Madagascar[35] and possesses one the richest and most endangered biotas in the world[7]. The altitude within RNP ranges from 600m to 1,478m[42]. Climate conditions in the park are seasonal with an average rainfall of 2.5-4 m of rain/year[35]. We conducted our field study during the cooler, dry season from June 1 to August 1, 2012.

## **Study population**

The study population consisted of three villages out of the roughly 65 rural villages in or around RNP: Ambatolahy (21°25'020 S, 47°42'950 E & N=215), Ambodiaviavy (21°26'391 S, 47°48'486 E & N=337), and Ankialo (21°08'062 S, 47°20.638 E & N=350). These three villages differed in their location within and around the park, as well as in apparent cultural practices, economic status, proximity to forest, and landscape features. Ambatolahy and Ambodiaviavy were surrounded by tropical rainforest and rainforest fragments. Ankialo was surrounded by rice paddy fields and invasive species of plants and trees (secondary forest). Sanitation and hygiene practices in these villages are poor and open defecation is commonly practiced. Livestock and domestic animals typically were kept in the household at night and released outside during the day. Animal feces material was visible throughout the villages.

#### **Survey questionnaire**

During June and July 2012, survey questionnaires were administered to the three participating villages. Villages, households, and human subjects selected during a 2011 pilot study were used for this study[40]. Informed consent was acquired by all human subjects prior to the survey and structured observations. Human subjects were anonymously assigned unique identifications (i.e. 1-2-HS-3; designates the village (1), the household (2), human subject (HS), and individual (3)). Two different surveys were conducted in all three villages. Both surveys were developed based on results from the 2011 pilot study in order to examine key issues in more depth[40]. A method known as

"translation/back-translation" was employed to increase reliability of translated surveys[43].

Surveys were conducted in Malagasy (native Madagascar language) by three trained and qualified staff members of Centre ValBio. The three surveyors (1 male, 2 females) were Malagasy natives who worked for the Health and Hygiene Team at Centre ValBio. They had previously conducted survey interviews in multiple studies, but were still individually trained to administer this study's survey questionnaires. The surveyors were randomly assigned to each household in all three villages.

The first survey was given to each individual in the household (Ambatolahy (n=54), Ambodiaviavy (n=66), and Ankialo (n=70)). Subjects pregnant at the time of the survey were exempt from taking the survey. The head-of-household typically answered questions on behalf of subjects under the age of five. The individual survey included 72 questions exploring an array of diarrhea risk-associated behavior variables including: basic demographics, economic status indicators, trading of goods and livestock, interaction with livestock and domestic animals, water, sanitation, and hygiene practices, food and diet, health status, and perceived health problems.

The second survey questionnaire administered was a household-level survey given to the head-of-household (Ambatolahy (n=10), Ambodiaviavy (n=10), and Ankialo (n=10)). This survey made inquiries into 50 different variables including: home characteristics, agriculture practices, livestock ownership and storage, economic status indicators, latrine use, and water and sanitation practices. The household-level survey was not fully analyzed in this study.

## **Structured observations**

During the same time period structured observations were conducted at all three villages (Ambatolahy-(142.26 total person-hours), Ambodiaviavy-(124.17 total person-hours), and Ankialo-(59.97 total person-hours). Four key events were chosen to observe multiple human and animal behaviors during village visits as likely to carry risk of diarrheal disease transmission. Initial unstructured observations were undertaken at Ambatolahy and Ambodiaviavy on separate days in order to generate pre-coded field entry forms and establish a protocol. Additional unstructured observations were conducted in order to ensure the field data entry forms were extensively pre-tested to check for reliability, replicability, ease of administration, and consistency between and within observers. The pre-coded forms contained structured categories, while at the same time allowing for qualifying comments. We were unable to do an unstructured observation or piloted structured observations in Ankialo due to time constraints and the remoteness of the village.

Surprise visits were judged to be inappropriate in this cultural setting. Therefore, prior to conducting observations in each village, we invited the entirety of the village to a discussion regarding our presence and study. The exact purpose and hypothesis of the study was not disclosed and words were chosen carefully to ensure village members understood that we were there simply to conduct research on the general health and wellbeing of the village. Unique identifications of households and human subjects were never revealed to the researchers.

Observations were conducted by four American researchers (2 males, 2 females) who were trained during the initial unstructured observation and pilot periods or by shadowing researchers during actual data collection. Daily debriefing meetings were held at the end of each observation day in order to discuss key findings and prepare for the next day. Researchers conducted observations in intervals of one to six hours and collected data on no more than two events at a time. The four key events observed included: 1) handwashing and bathing (79.07 total person-hours), 2) livestock behavior (154.99 total person-hours), 3) food preparation and storage (6 meals per village), and 4) collecting water and cleaning (91.34 total person-hours).

The *handwashing and bathing* and *collecting water and cleaning* events typically took place at a pump, spring, stream, river, or bathroom where researchers collected data. For *livestock behavior* and *food preparation and storage* observations, researchers roamed in separate and distinct areas of the village. The exception to this was Ankialo, where researchers typically collected data at clusters of two or three households due to the increased distance between households. Centre ValBio staff set up times for researchers to observe meals (breakfast, lunch, and dinner) as part of food preparation and storage observations. Gender and age (child = < 15, adult =  $\geq$  15) was collected for all events except livestock behavior. Subjects observed to be under the age of fifteen years old were considered children while subjects observed to be fifteen years of age and older were considered to be adults. Prior to beginning structured observations our local translator assisted us in determining the best guess for whom we should consider a child versus an adult. During unstructured observations we tested this with the aid of the translator to assure accuracy during structured observations.

For handwashing and bathing, at the time of an event data was collected on location, whether a bucket or cup was used, if soap was used, body parts cleaned (i.e. arms, hands, legs, feet, face, head, core), if the subject brushed his/her teeth, and what the subject used to brush with. For collecting water and cleaning, at the time of an event data was collected on location, collection container (i.e. jerry can, bucket, metal, cup, and water bottle), whether or not the collection container was cleaned before use, whether or not soap was used, and if another object was cleaned. Data collected for livestock behavior included species (canine, chicken, duck, feline, geese, pig, rabbit, turkey, and zebu), water interaction and location, food interaction, human interaction, animal interaction, entry of household, entry of kitchen, fecal interaction, and interaction description. Data collected for food preparation and storage events included a stepwise description of each event in the cooking or preparation scheme, whether or not animals or children interaction with the food preparer, food, or stored water, and a description of the interaction. In Ambodiaviavy and Ankialo we collected data on whether or not the subject was wearing shoes, sandals, or was barefoot.

#### Data analysis

Data analysis for the survey questionnaire was carried out using SAS 9.3 (SAS Institute Inc., Cary, NC). The outcome was reported diarrhea in the past four weeks, a binary variable with a "yes" or "no" response. In the original survey there were two questions that separately asked if the subject had had diarrhea in the past four weeks with blood and a subsequent question that asked if the subject had had diarrhea without blood in the past four weeks. These two variables were collapsed for the analysis to produce one dependent variable designated as reporting diarrhea with and without blood in the past four weeks. The categorical variables were compared to the village variable using the  $\chi^2$  and Fisher's exact tests in order to explain differences between villages. The categorical variables were also compared to reported diarrhea using the  $\chi^2$  and Fisher's exact tests to assess which variables were associated. A univariate regression analysis was used for variables that showed a statistically significant association with diarrhea in order to determine their sign and significance with reported diarrhea. Additional multivariate regression was executed using reported diarrhea with and without blood in the past four weeks as the dependent binary outcome variable. The independent variables used in the modeling included the variables that were statistically significant in the univariate regression analysis and cogent with *a priori* knowledge about the association of each variable with syndromic diarrhea. The explanatory variables that made it into the final models were selected because of their consistent statistical significance and sign in the coefficient estimates throughout different model structures. Correlation between variables was tested using Pearson's correlation coefficient in order to assess collinearity.

For the structured observations SAS 9.3 (SAS Institute Inc., Cary, NC) was used to calculate basic frequencies and percentages of observations and their categories. Qualifying comments were used to support results from the survey. Food preparation and storage was not analyzed quantitatively, but qualifying comments were used to support survey results.

#### **Ethical considerations**

This project was submitted to Emory University's Institutional Review Board and found exempt from further review because it did not meet the definition of "research" involving "human subjects" as set forth in Emory University policies and procedures and federal rules.

#### RESULTS

#### **Characteristics of all study subjects**

Of the 193 human subjects that took the survey in all three villages, 190 were included in the analysis. Three women were pregnant at the time of the study and were therefore, not allowed to be surveyed. Nearly 44% of sample population was female with Ambodiaviavy possessing the highest number of female subjects surveyed (47.0%) and Ankialo the lowest (40.0%). The mean age of all subjects was 23.4 (SD – 17.9) and were similar between villages (Ambatolahy-23.8(19.4), Ambodiaviavy-24.3(18.5), Ankialo-22.4(16.2%)). A little over 6% of the sample population was under the age of 5 with 35.3% of the total population between the ages of 5 and 14. Ambatolahy had the highest number of subjects under the age of 15 (44.5%), followed by Ankialo (42.9%) and then Ambodiaviavy (39.4%). There was a statistically significant difference (p-value <0.0001, Table 1) in tribe between the three villages where 100% of subjects surveyed in Ankialo were Betsileo in. Nearly all subjects in Ambatolahy were Betsileo (98.2%) with one subject identifying as Vakinakaratra, while respondents in Ambodiaviavy were mixed with roughly 60% identifying as Betsileo and 40% as Tanala. A majority of all subjects reported being employed and/or a student (87.9%). There was a statistically significant difference (p-value = 0.0195) in farming as a primary profession with nearly half of all subjects reporting farming as one of their primary professions. Just under a third of subjects from Ambatolahy reported farming as a primary profession in contrast to Ankialo where over a half of subjects reported farming as a primary profession (55.7%). In addition, 37.9% of all subjects reported being a student, 22.1% reported being a homemaker, 16.8% reported "other" as one of their primary professions, and approximately 10.0% reported trading as a primary profession (animal-2.1%, not animal-(7.9%). Nearly two-thirds of all subjects make no income on a weekly basis and (12.6%) of

income earners make over 20,000 Malagasy Ariary (\$1.00 = 2260.23 AR) (Table 1). Subjects were not asked to report education level, but Bodager reported only two subjects from Ankialo had completed "some university" and one subject from Ambodiaviavy finished "some technical school"[40].

#### Differences in daily life, behaviors, and responsibilities between villages

Subjects were asked an array of questions about their daily life, behaviors, and responsibilities in order to gain a better understanding of potential diarrhea related risk factors prevalent in these three villages. Furthermore, knowing the prevalence of these risk factors give insight into key differences between the three villages that might better explain what has a greater effect on syndromic diarrhea across and within these villages.

Trading is an important part of life in these rural villages, however only 12.2% of all subjects reported trading goods or services with other villagers while only 10.6% of all subjects reported trading goods or services with other villages in the area (Table 2). Trading with other villages typically takes place on Sunday's, the traditional market day in this region of Madagascar.

Many household responsibilities thought to be associated with transmission of infectious diseases were not responsibilities for a majority of all subjects who were surveyed. Close to 90% of subjects in Ambodiaviavy and Ankialo reported that tending livestock was "not a responsibility", while roughly 80% in Ambatolahy reported the same. There was a statistically significant relationship between collecting water and all three villages (p-value = 0.0086) where 72.2%, 89.4%, and 77.1% of reported subjects in Ambatolahy, Ambodiaviavy, and Ankialo respectively, reported this as "not a responsibility". Cooking was also not a responsibility for nearly 80% of all subjects.

Likewise, hunting was only a responsibility for a little over 5% of all subjects, but was statistically significant across villages (p-value = 0.0249) however. In addition, 67.9% of all subjects did not report house and yard cleaning as a responsibility with 16.7% of those surveyed in Ambatolahy, 10.6% in Ambodiaviavy, and 7.1% in Ankialo reporting they cleaned the house and yard "once a day". Lastly, a little over 60% of all subjects said using animal feces was not a task they participated in. Ankialo subjects reported the fewest amount of people who didn't use animal feces (48.6%) with Ambodiaviavy reporting the highest amount of individuals who don't use animal feces (Table 2).

Subjects were asked about their defecation practices, which showed a statistically significant relationship (p-value <0.0001) across villages. Roughly 70% of villagers in Ambatolahy reported defecating in a latrine with the remainder revealing they practice open defecation. Ankialo showed very similar results with Ambodiaviavy reporting the opposite with nearly four-fifths of villagers practicing open defecation. Likewise, subjects were asked to report all of their bathing practices, which proved to be statistically significant for all six types of bathing practices across the villages. Nearly 70% of all subjects reported bathing in a waterway (p-value <0.0001), which included rivers, streams, springs, canals, ponds, and lakes. Individuals in Ankialo had the fewest subjects report bathing in a waterway (34.3%) with the other two villages reporting similar figures of around 90%. The remainder of bathing practices including bathing in the field, behind the house, "everywhere", and "don't bathe", were mostly practiced by villagers in Ankialo (Table 3).

The villagers' water practices were probed in order to understand possible transmission dynamics with water as a potential reservoir for enteric pathogens. Only one-fifth of all subjects reported treating their drinking water before they drank it, which proved to be statistically significant across villages (p-value = 0.0107). In fact, just fewer than 10% of subjects in Ambatolahy and one-quarter of subjects in Ankialo treated their drinking water, while one-third of subjects in Ambodiaviavy reported drinking water that was treated before. Of those who reported treating their drinking water, 60.0%, 93.8%, and 93.8% in Ambatolahy, Ambodiaviavy, and Ankialo respectively, used the boiling method to treat their drinking water before drinking. No one reported using the local chlorine solution for treating their drinking water. Typically subjects across all three villages treated their drinking water less than once per week (59.5%) with nearly 30% treating their drinking water more than once a week. Only five subjects, from Ambodiaviay, reported treating their drinking water. Moreover, to take water from their storage containers, most individuals (97.4%) used a cup as one of their reported techniques for doing so. Close to 100% of all subjects reported using their stored water for drinking. Using stored water for bathing (p-value <0.0001), washing meat to cook (pvalue = 0.0553) washing dishes and utensils (p-value = 0.0134), giving to animals (pvalue <0.0001), and "other" (p-value = 0.0010) all showed a statistically significant relationship with the villages (Table 4).

Subjects were asked about their handwashing rituals, one of the most studied hygiene practices in water, sanitation, and hygiene research. Handwashing more than once a day was very common for 96.8% of all villagers. Not only is the frequency of handwashing important to preventing infection and the spread of pathogens, but the timing of handwashing is just as critical to breaking the transmission cycle. Therefore, we asked subjects to report all the times they wash their hands as an open-ended question

without reading any possible times out loud. We found a statistically significant difference (p-value = 0.0422) between villages who reported washing their hands before food preparation (Ambatolahy-38.9%, Ambodiaviavy-27.3%, & Ankialo-18.6%). Only two subjects reported washing their hands after preparing food. Likewise, only one subject reported washing their hands before collecting food while not one villager reported washing after collecting food. There was however, a statistically significant difference (p-value < 0.0001) of washing hands after defecating across villages where 72.2% of subjects in Ambatolahy reported washing their hands at this time, and only 30.3% of those from Ambodiaviavy, and 31.4% from Ankialo, reported washing their hands after defecating. Nearly a quarter of all subjects reported handwashing after working with livestock including 32.9% of subjects in Ankialo, 22.2% in Ambatolahy, and 16.7% in Ambodiaviavy. It appears that handwashing before eating is the most popular time for these three villages with 85.3% of all subjects reporting this time. Ambodiaviavy villagers reported the highest at roughly 90% of all those surveyed and Ankialo reported the lowest at 80%. Contrastingly, 72.6% of all subjects reported that they did not wash their hands after using animal feces. In addition, roughly half of all subjects declared they did not wash their hands after farming in the field. A little over a half over of all subject reported washing their hands during an "other" time. These times mostly included "after playing", "after wipe baby's butt", "every morning", "before going to school", and "after school" (Table 5).

In addition to water, the use of soap makes handwashing more effective in terms of preventing diarrheal disease[44, 45]. Supplementary questions were asked to assess this integral step when handwashing. Assuredly, just over 80% of all subjects reported

sometimes having soap in the household, which was statistically significant (p-value = 0.0010) across villages. Nearly a quarter of subjects surveyed in Ambatolahy and Ankialo reported always having soap in the household while only 4.6% of Ambodiaviavy subjects reported the same. With regards to how frequent villagers use soap (p-value = 0.0188), nearly one-fifth of all subjects admitted they don't wash their hands with soap with Ankialo possessing the highest at 21.4% of subjects surveyed. It was more common for the reporting villagers to sometimes handwash with soap, where 71.8% sometimes did compared to about 10% always using soap when handwashing. Additional utilization of soap beyond handwashing was statistically significant across villages when washing for market day (p-value = 0.0096), and washing kitchen items (p-value < 0.0001). Furthermore, a majority of all subjects use soap for bathing their body, washing hair, or washing clothes. The previous variables regarding soap may be influenced by the availability of soap for purchase, which doesn't appear to be a problem as most villagers procured their soap from a store in their village, another villager, or from the market. As an interesting aside, nearly 80% of all subjects said they would like to learn to make soap (Table 6).

Diet and personal habits play a major role in overall health and can influence an individual's susceptibility to infection from an enteric pathogen. Along with water, villagers were asked to report additional liquids they consume, as well as how much rice they eat during harvesting season, and their smoking, drinking, and chewing tobacco habits. Over 90% of all subjects reported drinking juice and rice water. To make rice water, villagers would pour water in their large pots after cooking rice in it, allowing the water to become very hot before serving. There was a statistically significant difference

(p-value = 0.0006) of tea drinkers across villages with Ambodiaviavy possessing highest percent of tea drinkers (78.8%). There was also a statistical significant relationship between beer drinkers (p-value = 0.0002) and toaka gasy (moonshine) drinkers (p-value < 0.0001) across the three villages.. Over half of all subjects reported drinking "other", which was also statistically significant (p-value = 0.0121). Most of the other drinks reported were milk or a variety of herbal and medicinal teas. Rice is a staple crop for both diet and income for the Malagasy, especially in the Ranomafana area of Madagascar. It wass not surprising to see that 92% of all subjects ate rice three times a day, which showed to be statistically significant (p-value < 0.0001) among all villages. Whereas Ankialo and Ambatolahy reported either eating rice three times a day or sometimes more than three times a day, 15.2% of those surveyed in Ambodiaviavy reported eating rice only twice a day. This may be a reflection of the mixture of tribes in this village (Table 1). With regards to personal habits, a majority of all villagers reported that they didn't smoke, chew tobacco, or drink alcohol(Table 7).

As reported by Kightlinger et al. in 1995, parasites were a major concern for villages in Ranomafana[8]. One of the most significant risk factors exposing individuals to these parasites is walking around barefoot where the ground contains high concentrations of these diarrhea causing pathogens. While many of those surveyed reported using a taxi as a primary mode of transportation, close to 100% of villagers reported an additional mode of transportation was by foot. This is alarming because roughly half of all subjects in Ambodiaviavy and Ankialo reported not having shoes or sandals (Table 8). Unfortunately, we were unable to ask this question in Ambatolahy where many villagers were observed to be barefoot as well.

The villager's medication practices were asked and these questions revealed that a similar percentage of villagers in each village had taken medication in the past four weeks. In addition to that, all three villages reported similar percentages for those who symptoms improved from the medication, which was 88.2%, 97.2%, and 97.1% for Ambatolahy, Ambodiaviavy, and Ankialo respectively. Moreover, there was a statistically significant difference (p-value = 0.0017) for those who reported giving their animals medication in the past four weeks across villages. Ankialo reported the highest at 38.6% with Ambatolahy the lowest at 11.8%. This may be a function of the amount of animals owned by the individual villagers surveyed or the villages as a whole (Table 9). The survey did not inquire into what the villagers took the medication for or why they gave medication to their animals.

A key behavior that can conceivably fuel zoonotic disease transmission is the handling of feces. In these three villages around half of the villagers surveyed revealed that they had handled human feces in the past four weeks while there was a statistically significant difference (p-value = 0.0027) of handling animal feces between villages. Ankialo villagers reported the highest at 75.7% with the other two villages both close to approximately 50%. Not surprisingly, chicken feces, pig feces, and zebu (cattle) feces were the most reported animal feces handled by villagers. Ankialo reported the highest for each of these species with handling pig feces statistically significant (p-value < 0.0001) across villages (Table 10).

### Examination of basic characteristics of those who reported diarrhea

The primary outcome of interest is reported diarrhea with and without blood in the past four weeks. This was answered in two parts with one question asking about blood without diarrhea and the proceeding question about diarrhea with blood. These two categories were collapsed into one reported diarrhea variable because too few subjects reported having diarrhea with blood to perform separate valid statistical analyses. Subsequently, from all three villages 37 out of the 187 (19.8%) subjects who responded to the two questions about syndromic diarrhea reported having diarrhea in the past four weeks. In Ambatolahy, 19.2% or 10 out of the 54 subjects who responded to the two questions reported having diarrhea in the past four week. Villagers in Ambadoiaviavy reported the highest prevalence of diarrhea with 23.1% or 15 out of 65 subjects who responded the lowest prevalence with 12 out of 70 (17.1%) subjects revealing they had diarrhea in the past four weeks (Table 11).

There was a slight gender difference between those who reported having diarrhea (female-51.4% vs male-48.7%) with females in Ambodiaviavy containing the highest prevalence of females with diarrhea. Ambatolahy had the highest percentage of men who reported having diarrhea compared to the other villages. In Madagascar, when children turn 15 years of age they are considered an adult. Of those who reported diarrhea nearly half were 14 years of age or younger. One subject in Ambatolahy and one in Ankialo were four years of age or younger, a high risk group for diarrhea. There was a statistically significant association (p-value = 0.0020) between tribe and reported diarrhea with the Betsileo comprising just over two-thirds of subjects reporting diarrhea. Farmers, students, and homemakers reported the highest prevalence of diarrhea with farmers reporting the highest with 10.2% of all subjects, followed by students and then homemakers. Those who made no income reported over half of the diarrhea with the remainder of cases

almost evenly dispersed among the other four categories of income (Table 11).

## Differences in lifestyle and behaviors among those reporting diarrhea

Trading goods and services with other villagers (p-value = 0.0085), as well as with other villages (p-value = 0.0081) showed a statistically significant association with reported diarrhea. Although less than half of reported diarrhea cases were by those who traded with other villagers or villages, Ambodiaviavy reported the most, followed by Ankialo and then Ambatolahy. A majority of those who had diarrhea in the past four weeks also reported that they were not responsible for tending livestock, collecting water, cooking, or hunting. Seven villagers in Ambodiaviavy reported house and yard cleaning from more than once a day to less than once a week. Four villagers in Ankialo who reported diarrhea also house and yard clean once a week. Although not statistically significant (p-value = 0.5150) more villagers with diarrhea reported using animal feces as a household responsibility than any other household responsibilities with 10.8% of Ambodiaviavy villagers using animal feces less than once a week. Five Ankialo villagers with diarrhea also reported using animal feces less than once a week while two subjects reporting from Ambatolahy use animal feces less than once a week (Table 12).

Defecation behavior was statistically significant (p-value = 0.0053) with reported diarrhea as two-thirds of those who reported having diarrhea practiced open defecation compared to using a latrine as an alternative. 100% of villagers with diarrhea from Ambodiaviavy practiced open defecation while Ankialo villagers with diarrhea mostly used a latrine. Subjects with diarrhea from Ambatolahy were about split, but still reported more individuals practicing open defecation. A larger proportion of villagers who bathed in a bathroom did not report having diarrhea while those who bathed in a waterway also reported having diarrhea. Ambodiaviavy reported the highest subjects with diarrhea and washed in waterways, followed by Ambatolahy and then Ankialo. In addition, very few subjects reported having diarrhea when they bathed in the field, behind the house, "everywhere", or did not bathe(Table 13).

Treating drinking water was also statistically associated (p-value = 0.0066) with reported diarrhea as all but two subjects – one from Ambodiaviavy and one from Ankialo - reported diarrhea while not treating their drinking water. The two who treated their drinking water used boiling as their treatment technique with only one responding that they only sometimes (< once a week) treated their drinking water. A substantial proportion (94.6%) of those with diarrhea reported using a cup to take water from their water storage container. There was a statistically significant relationship between those who used an "other" object to take water from their storage container and reported diarrhea. Nearly 100% of those who reported other said they used a cup. Virtually all those who reported having diarrhea used their stored water for drinking, while a considerable number of those with diarrhea used their stored water for washing dishes and utensils, cooking, washing meat to cook, and washing fruit and vegetables. Furthermore, there was a significant association (p-value = 0.0329) between villagers who used stored water for an "other" purpose and reported diarrhea. Nearly all those who responded "other" said they used the stored water for washing their face and hands with a few using it to brush their teeth (Table 14).

The frequency of handwashing did not prove to be very influential on reported diarrhea (Table 15). However, the times at which villagers did handwash showed to be more telling of whether or not someone reported having diarrhea. Coincidently, 75.7% of

those who reported diarrhea, but only 15.5% of subjects surveyed overall, also reported washing their hands before eating (Table 15). Although not statistically associated, almost all of those who reported diarrhea also responded that they did not wash their hands before and after food preparation, before and after food collection, before and after using the bathroom, before and after working with livestock, after eating, before and after breastfeeding, before and after using animal feces, and before and after farming in the field (Table 16).

100% of those who reported diarrhea in Ambatolahy and Ambodiaviavy sometimes had soap in the household compared to always or never having soap. A quarter of those with diarrhea in Ankialo always had soap in the household. A similar story was reported with frequency of washing hands with soap where a majority of those with diarrhea sometimes did, except eight subjects with diarrhea also reported they do not wash their hands with soap. Moreover, a majority of subjects with diarrhea in addition to handwashing also used soap for washing hair, bathing their bodies, washing clothes, and washing for market day (Table 17).

Dietary and personal habits didn't seem to be associated with reported diarrhea. A majority of those with diarrhea drank tea, juice, coffee, rice water in addition to water. In addition, almost all villagers with diarrhea ate rice three times and typically never smoked, chewed tobacco, or drank alcohol. However, there was a statistically significant relationship between both drinking toaka gasy (moonshine) (p-value = 0.0241) and drinking "other" (p-value = 0.0081) and reported diarrhea (Table 18).

The villagers primary mode of transportation was statistically associated (p-value = 0.0256) between taking a taxi-brousse and reported diarrhea with roughly 73% of those

with diarrhea taking a taxi-brousse. Though not statistically significant, 89.2% of those with diarrhea reported their primary mode of transportation to be by foot. Correspondingly, twice as many villagers with diarrhea did not have shoes or sandals than those who did. Having shoes or sandals was statistically associated with reported diarrhea (p-value = 0.0250) (Table 19).

The use of medication proved to have a statistically significant relationship with reported diarrhea. A majority of humans with diarrhea reported taking medication in the past four weeks with Ambodiaviavy containing the highest proportion, followed by Ambatolahy and then Ankialo. This had a highly statistical significant association (p-value = 0.0016) with reported diarrhea. However, whether or not symptoms improved from that medication was not statistically associated. Likewise, subjects that reported diarrhea also gave their animals medication in the past four weeks about half the time, which showed a statistically significant association (p-value = 0.0007). Once again, whether or not symptoms improved from taking the medication was not significant (Table 20).

Handling human or animal feces did not predict reported diarrhea amongst villagers. A moderate amount of those who reported diarrhea did however handle human feces (54.1%) or animal feces (70.2%). Villagers with diarrhea mostly reported handling chicken feces, followed by zebu and then pig fecal material (Table 21).

# Univariate regression analysis

The risk factors that had a statistically significant association with reported diarrhea with or with blood in the past four weeks from the Chi-square or Fisher's exact tests were further tested in a univariate regression analysis. This analysis exhibited multiple risk factors to be statistically significant predictors of reported diarrhea not adjusting for possible confounders or covariates at  $\alpha$ =0.05 as depicted in Table 22.

Not surprisingly, not treating drinking water had the largest effect on reported diarrhea where treating drinking water proved to be protective against diarrhea. Not using "other" for the use of soap had the second largest effect on reported diarrhea, although this was not statistically significant. Taking medication and giving medication to an animal in the past four weeks resulted in a statistically significant effect on reported diarrhea, although without temporality taking medication may not be a valid predictor. Open defecation was of the expected sign and significance. Trading with other villagers and villages increased the odds of diarrhea compare to those who didn't.

The type of liquids villagers were drinking had an interesting relationship with reported diarrhea. Drinking herbal and medicinal teas was protective while not drinking these teas increased the odds of reporting diarrhea by 2.69. Furthermore, drinking toaka gasy (moonshine) also proved to be protective against reported diarrhea. Subjects who reported they don't drink toaka gasy were 2.51 more likely to report having diarrhea in the past four weeks.

Surprisingly, using a taxi-brousse as a primary mode of transportation showed a statistically significant association with reported diarrhea where the odds of reporting diarrhea was 2.43 times higher for those who used a taxi-brousse compared to those who did not. On the same note, villagers in Ambatolahy and Ankialo were 2.70 times more likely to report diarrhea when they didn't have shoes or sandals compared to those who did.

Two demographic variables engendered an important relationship with reported

diarrhea. Being in the Betsileo tribe reduced the odds of reporting diarrhea by a factor of 0.27 whereas being Tanala increased the odds of reported diarrhea by 3.78 times compared to not identifying with that tribe. Similarly, being a homemaker increased the odds of villagers reporting diarrhea in the past four weeks by 2.36 compared to those who did not report being a homemaker as a primary profession.

# **Multivariate regression analysis**

Multiple regression analysis was performed to investigate the relationship of explanatory variables on reported diarrhea and unravel the distinct influence of these variables in the presence of each other. This procedure was executed as a strictly exploratory measure in an attempt to peak through the noise for significant predictors. Variables were assessed for their sign and significance at  $\alpha$ =0.05. All of the variables included in the model were tested for collinearity and found to be weakly or moderately correlated (0 < | r | < 0.7).

The first model in Table 23 shows the four demographic variables as not statistically significant (p-value > 0.05) in this analysis. Age had a negative relationship with reported diarrhea and had a very neutral effect on reported diarrhea by a factor of 0.99. Females were 2.01 times more likely to report having diarrhea compared to males while those who reported being a homemaker as a primary profession were 2.70 times more likely to report diarrhea than those who didn't report being a homemaker. None of the villages were statistically significant. Not surprising, living in Ambatolahy proved to be protective when compared to living in Ambodiaviavy. However, villagers in Ambatolahy were 1.04 times more likely to report having diarrhea in the past four weeks compared to villagers in Ankialo. Likewise, villagers in Ambodiaviavy were also 1.36

times more likely to report having diarrhea in the past four weeks when matched against villagers in Ankialo.

The two water, sanitation, and hygiene behavior variables had contrasting effects on reported diarrhea. The odds of reporting diarrhea were 2.68 times higher for villagers that practiced open defecation than those who did not. This association was not statistically significant however. Expectedly, the odds of reporting diarrhea were 19.13 times more likely among those who didn't treat their drinking water. While this was statistically significant the 95% confidence interval was still very wide.

The results also indicated that trading goods or services with other villages increased the odds of reporting diarrhea in the past four weeks by 5.83. This was statistically significant and positively associated with reported diarrhea. Lastly, animals taking medication in the past four weeks appears to be positively and significantly associated with reporting diarrhea, with an odds ratio of 5.71. The first model is represented below:

Pr(Reported diarrhea) = -5.9165 + 1.7420\*Animal taken medication in past 4 weeks - 0.0109\*Age + 0.9863\*Open defecation + 2.9511\*Don't treat drinking water + 1.7626\*Trade with other villages + 0.6969\*Female +

0.9919 \* Homemaker + 0.0433 \* Ambatolahy + 0.3036 \* Ambodiaviavy

The second model in Table 24 includes similar variables except it includes weekly income as a proxy for socio-economic status. The four demographic variables appear to have a similar association with age negatively and not significantly associated with reporting diarrhea. The effect of females on reporting diarrhea lowered from the previous model to an odds ratio of 1.93, but remained not statistically significant. Villagers that

reported homemaker as a primary profession remained positively and not significantly associated with reported diarrhea, with a similar odds ratio. Location also remained positively yet not significantly associated.

The two water, sanitation, and hygiene behavior variables also maintained similar associations from the model in Table 23. Open defecation remained not statistically significant and had a slightly lower odds ratio of 2.47. Likewise, villagers who didn't treat their drinking water maintained a positive and statistically significant association with a slightly lower odds ratio of 17.87.

In addition, trading goods or services with other villages appears to be positively and significantly associated with reported diarrhea either. The odds ratio only substantially increased to 8.09. Villagers whose animals had taken medication in the past four weeks showed a statistically significant relationship and slightly higher effect on reporting diarrhea with an odds ratio of 6.90.

Lastly, results also indicated that weekly income was not statistically associated with reporting diarrhea in the past four weeks. This may indicate that differences between those in different categories of income might have been explained by other factors in the model. Villagers making no income, 1AR - 4,999AR, and 10,000AR - 19,999AR were positively associated, while villagers making 5,000AR - 9,999AR were negatively associated with reporting diarrhea. This second model is represented below:

Pr(Reported diarrhea) = -6.6911 + 1.9319\*Animal taken medication in past 4 weeks - 0.00133\*Age + 0.9046\*Open defecation + 2.8829\*Don't treat drinking water + 2.0906\*Trade with other villages + 0.6588\*Female + 0.9536\*Homemaker + 0.1923\*Ambatolahy + 0.3546\*Ambodiaviavy + 0.8070\*0 AR (No Income) + 0.1554\*1 AR - 4,999 AR - 0.3725\*5,000 AR -9,999 AR + 0.5491\*10,000 AR - 19,999 AR

## **Perceived health problems**

Subjects were asked to report what they perceived to be biggest health problems in their household in order to gain some insight into their knowledge, attitude, and perception of health for future studies and interventions. These questions were openended with the first question asking what they thought the biggest health problem in their household was. The next question asked what they thought the second biggest health problem in their household was with the final question asking what they thought the third biggest health problem in their household was. Surveyors only wrote down one answer per question.

In Ambatolahy, villagers reported respiratory illness as their number health problem followed by malaria and then headaches. Diarrhea was considered fifth among the number one health problems. Respiratory illness was also considered the second biggest health problem in the household. Headaches and malaria were the next two health problems again as well. Once again, respiratory illness was considered the third biggest health problem in the household, but this time followed by diarrhea and then malaria (Table 25).

As portrayed in Table 26, the biggest health problems in Ambodiaviavy were slightly different. The biggest health problem was still respiratory illness by quite a bit with diarrhea and malaria considered as well. The second biggest health problem this time was headaches followed closely by malaria and then asthma. Diarrhea was considered the third biggest health problem with malaria and respiratory illness coming in second and third respectively.

Villagers in Ankialo also reported slightly different health problems than the other two villages. Intriguingly, headaches were reported to be the biggest health problem in the household, which was tied with toothaches. Diarrhea was ranked seventh among the biggest health problems. The second biggest health problem was considered headaches again with respiratory illness right after and then diarrhea. The third biggest health problem was reported to be respiratory illness followed by diarrhea and then malaria.

## Handwashing and bathing observations

In total, we did handwashing and bathing observations for a combined 46.53 hours of time in Ambatolahy, 19.12 hours in Ambodiaviavy, and 13.42 hours in Ankialo between four observers. This was done in order to understand what time of the day villagers typically washed their hands, how often, what other parts of their body were being washed, if they used soap, and if they brushed their teeth. We also took that time to observe whether or not villagers were wearing shoes or sandals in order to understand what foot hygiene might be like in the villages.

In Ambatolahy, there were a total of 402 observations observed between four observers. A third of the villagers observed were male adults, followed by female adults and male children accumulating roughly a quarter of the observation each. When villagers in Ambatolahy washed they typically washed their feet, hands, legs, arms, or face either separately or in combination. Soap was only observed to be used in 2.0% of the observations. In Ambatolahy, villagers typically bathed or washed from one of two pumps. In some of the observations, villagers were seen washing or bathing in a spring adjacent to the two pumps. 46 villagers were observed brushing their teeth. Of those who did, roughly one-third used their finger, a third used a toothbrush only, and another third used a toothbrush with toothpaste. Unfortunately, footwear in this village was not recorded.

Handwashing and bathing observations in Ambodiaviavy totaled at 251. This time, just under half of the villagers observed were female adults with male adults representing roughly one-fifth. Female children accounted for just under one-fifth as well, while male children represented the lowest at 14.3%. In roughly 70% of the observations villagers washed their hands either separately or in combination with another body part. Feet, legs, arms, and face were also observed to be washed around half the time either separately or in combination with other body parts. A slightly higher percentage of villagers observed used soap, but only 16 of those observed used it still. As in Ambatolahy, handwashing and bathing almost always takes place at one of four pumps in the village. Observations at one of the pumps were not included in the analysis. Five villagers were observed washing at the river that snaked through their rice paddy fields. 38 villagers or 15.4% of the observations were witnessed to have brushed their teeth. Of those who did nearly all were observed to have only used their finger. In two of the observations they used a toothbrush only and in the other three they used a toothbrush with toothpaste. A majority of those observed were barefoot with one-tenth of the observations revealing the use of shoes. Only one villager observed wore sandals and two were unknown.

Not surprisingly, there were only 9 observations in the village of Ankialo due to the landscape and built environment of this village. Spring water that flowed into the rice paddy fields were the main source of water for this village. They did not have any pumps

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that we observed or were told about by the villagers. In all of the observations, feet and legs were washed. Hands, arms, and the face were also body parts typically washed from these observations. Only female villagers were observed washing with children contributing five observations compared to four who were adults. In two of the observations the villager was seen brushing his or her teeth. In one instance the villager only used a finger while in the other the villager only used a toothbrush. Lastly, only one villager was recorded as barefoot while the other eight were not recorded or unknown.

#### **Collecting water and cleaning observations**

Observing villagers collecting water and cleaning yielded 55.15 hours of observation time in Ambatolahy, 22.77 hours in Ambodiaviavy, and 13.42 hours in Ankialo. We watched villagers collect water because we wanted to see where they were collecting what could have been their drinking water, as well as the water they would use for cooking, cleaning, and bathing in their household. It was important to see if they washed out the containers they were using to collect this water and who was actually collecting the water. We also wanted to see if villagers were washing other items that were visibly dirty with material potentially contaminated by pathogens.

In Ambatolahy, we observed 294 separate incidences related to collecting water and cleaning. Roughly two-fifths of those were female adults while female children, male children, and male adults made up roughly 20% each. In a little over half of the observations a plastic bucket was used to collect water and in one-tenth of the observations villagers drank the water straight from the water source. About half of the time we observed villagers cleaning these buckets out with their hands and water. In 81 of the observation we witnessed the villager clean another object, which mostly included dishes and utensils, clothing, and some food. However, in only 5.1% of all observations in this village did the villager actually use soap to clean out the bucket, jerry can, or cup used to collect water or the other item cleaned. Once again, most this activity took place at one of the two functioning pumps in the village. Although, some of this activity also took place in the river upstream of the village, and in separate springs above the pumps that potentially feed into the pumps.

In Ambodiaviavy we witnessed 377 observations related to collecting water and cleaning. Once again, the majority of observations were of female adults, followed by female children at around one-fifth, and then male children and male adults. A majority of the observations involved the use of a plastic bucket to collect water at one of the pumps in the village. Jerry cans and cups were also used to collect water with only four individuals drinking water straight from one of the pumps. Villagers in this village were observed to have cleaned additional objects fewer times than their counterpoints in Ambatolahy. These objects were clothing two-thirds of the time with dishes or utensils the other third of the observations. Unfortunately, soap was not used 3.7% of the time. Results also show that 95.5% of those observed were barefoot at the time observations were taking place.

There was an increase in observations for Ankialo villagers with 30 collecting water and cleaning observations. Yet again, the majority of observations were female adults at 56.7%. This was succeeded by female children at 23.3%, male adults at 13.3% and then male children at 6.7%. In almost all of the observations where water was collected a plastic bucket was used instead of a jerry can or cup. An additional object was cleaned in eight of the observations with half of all observations resulting in the object or

plastic bucket being cleaned with the villager's hands and water. Despite this, in only five observations did the villager use soap as an additional cleaning agent. The additional objects cleaned were mostly clothes and one involving food. Contrary to Ambodiaviavy, the villagers observed were wearing sandals in 27 of the observations with the remainder barefoot.

# Livestock and domestic animal behavior observations

Observing livestock and domestic animal behavior and their interaction with humans, other animals, different waterways, food, and feces allowed us to see the relationship the villagers had with these animals, as well as the relationship these animals had with their environment. From conducting these observations we were able to piece together a story of these potential enteric pathogen reservoirs and transmitters of zoonotic disease through space and time where a simple survey questionnaire would fall short of capturing. In total, we collected a combined 39.58 hours of observations in Ambatolahy, 82.28 hours in Ambodiaviavy and 33.13 hours in Ankialo. We observed eight different animals throughout our field study, which included zebu (cattle), dogs, chickens, ducks, cats, fish, geese, and pigs. Through the course of these observations we witnessed a variety of known transmission pathways, but also never before seen transmission pathways that were very specific to either these villages or Madagascar as a whole.

Residents of Ambatolahy own an assortment of poultry, which made up a majority of the animals observed in this village. Many of the human interactions were with male children, especially from pigs, cats, chickens, and ducks. Chickens interacted with the widest variety of gender and age combinations. Poultry also had the most amount of interaction with water sources, which mostly consisted of springs located throughout the village. Chickens and ducks were also observed to interact with food more frequently than any other animal. Geese, followed by chickens and then pigs interacted with other animals more often whether making direct contact or eating close with each other. Most likely due to the density of chickens, they were observed to interact with feces the most. Chickens also entered the household more frequently along with cats who are considered the best means for ridding the household of rodents. Lastly, chickens and geese were once again observed to play the largest role in entering the kitchen structures where villagers cook and store goods.

Livestock and domestic animal interaction were once again mostly dominated by chickens in Ambodiaviavy. Additionally, zebu, dogs, and pigs also played a pivotal role in this village' observations. Similar to Ambatolahy, chickens interacted with humans the most with male children the most frequent point of human contact. There was very little contact or interaction with water sources, although chickens were observed interacting with one specific pump more than any other water source. Not surprisingly, a little over half of the observations were of chickens interacting with food. During the study period in this village every household laid out different foods or crops to dry out in the sun leaving it vulnerable to animals. Chickens also interacted with other animals and fecal material more than any other animal with dogs representing the second highest amount of interactions. A little over one-tenth of observations were also of chickens entering households while roughly 3% of observations were of them also entering kitchen structures.

Ankialo was also heavily dominated by chickens compared to the influence of any other animal. Ankialo was unique when compared to the other two villages though because villagers sheltered their livestock. Poultry and pigs were typically kept on the first story of the household, which was in addition to households having a significant amount of space between each other. This reduced the chance for observing animal behavior or their interaction with other sources. In this village both chickens and zebu interacted with humans more than other animals. There was very little animal interaction with any water source, although ducks interacted with a local stream four times. The sheer presence and influence of chickens also dominated interaction with food, other animals, feces, and entering households and kitchens. Kitchens in this village were typically on the second or third floor, but chickens did make their way up there.

#### DISCUSSION

This study gave us a unique opportunity to build upon the 2011 pilot study by combining emic and etic approaches to understanding socio-cultural influenced behaviors as risk factors for diarrheal disease in these three villages. The mixed-methods approach we deployed allowed us to see the intricacies of these villagers' daily lives using direct observations, which added richness to the results from the survey questionnaires. The survey questionnaire enabled us to see which of these individual behaviors separately influenced and determined reported diarrhea. In addition, this field season provided a rare instance for us to assess the impact of how the built environment, landscape, and topography's interactive role with human and livestock behavior influences transmission dynamics.

We set out to investigate the risk factors that determined syndromic diarrhea in rural villages of a submontane rainforest area in Madagascar. The prevalence of diarrhea reported by the study sample population seems to reflect the differing level of water,

sanitation, and hygiene practices in the three villages. While there were many underlying cultural and behavioral similarities between the villages that influenced the prevalence of diarrheal disease in this area, the roles that each villager played within the family unit and the village as a whole determined the degree and type of risk factor that potentially exposed them to diarrhea causing pathogens. It must also be noted that the season played a formidable role in setting the collective agenda for each village. We were there during the dry season, a time when the entire family resided in their homes within the confines of the village. This undoubtedly impacted their daily activities, potentially fostering the necessary conditions for waterborne and zoonotic enteric disease transmission cycles. In addition, the biodiversity of the forest and the relationship that each of these villages has with the forest also seemed to impact their daily lives and activities. We attempted to discern how each of these distant level risk factors influenced the more proximal level risk factors through the use of structured observations while simultaneously recording the frequency of risk related behaviors that illustrated the pattern of their day-to-day routine. It is hypothesized that the convergence of human behavior, animal behavior, and their interaction with certain environmental vehicles determined which transmission pathway would persist long enough, even if momentarily, to cause exposure.

The multivariate regression analysis indicated one key water behavior that was a statistically significant determinant of reported diarrhea. The finding that villagers who did not treat their drinking water had a higher chance of reporting diarrhea during the previous four weeks highlights the overall lack of health protective water, sanitation, and hygiene behaviors in these villages. Very few villagers reported that they treated their drinking water and those who did typically treated it less than once a week. Considering

they were observed to have collected water once to multiple times a day with plastic buckets that were very rarely cleaned with soap, this is a very telling human behavior that is very likely a determinant of diarrheal disease in these villages. This is consistent with a meta-analysis revealing that improving water quality (microbial safety), especially through point-of-use treatment, was more effective at reducing diarrheal disease than previously thought[46].

Stored water was typically used for drinking, washing fruits, vegetables, and meat, or cooking purposes, indicating that the chance for ingesting or being exposed to contaminated water and food through this pathway was highly plausible. The absence of a water treating technique was confirmed through meal time observations where water used in the cooking process was never observed to have been boiled before being used to clean food or utensils. However, we cannot rule out the possibility that water was boiled before we entered the household.

Observations revealed that the primary sources of drinking water for all three villages were unprotected. In Ambatolahy, where villagers have 24-hour access to water, the primary sources of water were two pumps and an adjacent river. Results from the 2011 pilot study water samples revealed that the two primary pumps tested positive for *Salmonella* and *E. coli* while the main stream flowing through one of the pumps tested positive for *Salmonella* and *E. coli* and *Salmonella* as well[47]. In addition, the river and household buckets tested positive for *E. coli*[47]. We observed these pumps in great length to find a high occurrence of use at both these pumps where handwashing, bathing, cleaning of items including food, and water collection took place in high frequency during early morning and early evening time periods. There were four pumps in Ambodiaviavy that

were also primary sources of water with behaviors at these pumps similar to those in Ambatolahy; the one caveat being that pumps in Ambatolahy had running water all day whereas the pumps in Ambodiaviavy were turned on for an hour and a half in the morning and once again in the evening. All four pumps and household buckets tested positive for *E. coli*, *Shigella*, and *Salmonella*[47]. The adjacent river tested positive for *E*. *coli*[47]. Villagers in Ambodiaviavy reported treating their drinking water more often, yet had the highest prevalence of diarrhea suggesting recontamination after treatment or over reporting of "good" hygiene behaviors. Ankialo was noticeably different in that there were no pumps and water was mostly procured from springs that fed into the rice paddy fields. The 2011 pilot study also indicated that water samples taken from various water sources in Ankialo tested positive for E. Coli, Shigella, and Salmonella[47]. Despite having the second lowest proportion of villagers who reported treating their drinking water and the highest proportion of villagers reporting they treated their drinking water less than once a week, villagers in Ankialo had the lowest proportion of reported diarrhea possibly suggesting that contaminated drinking water may have been a minor determinant of diarrheal disease in this village, if at all. However, if their water sources are still contaminated with the bacterial pathogens they tested positive for in 2011 then drinking water may still be a determinant of diarrheal disease in this village.

The one caveat to treating their drinking water was the post-meal tradition of making rice water. As reported by a majority of villagers, it's customary for Malagasy to eat rice three times a day because rice is their most abundant staple crop. At the end of the meal it was common to fill the rice pot with water to heat up for roughly 10-20 minutes. The amount of time the water was heated and allowed to steep varied and it

could not be verified if this was an effective "boiling" method for drinking water treatment. Although there was no statistically significant association, all but one subject who reported having diarrhea in the past four weeks also drank rice water. However, it is difficult to conclude too much from this as nearly the entire sample population reported drinking rice water.

Boiling water in these villages also inflicted an additional severe health concern for villagers. Typically wood was used as fuel for fire because they could not afford coal or an alternative method for heat. When they cooked, smoke from the fire would engulf their homes due to the lack of ventilation, potentially causing chronic respiratory disorders. The only metric for this phenomenon though were the questions asking about their biggest health problems to which respiratory illness was frequently regarded as one of the top health problems in all three villages. In conjunction with malnutrition[38], chronic respiratory illness could potentially induce a degree of immunosuppression, potentially putting those in these three villages at an increased risk of diarrheal infection. The role that livestock play in these rural villages is central to their health, wealth, and status in the community. This is reflected both in the high density of poultry, dogs, cats, and pigs within certain villages and relative management practices of these animals. This relationship and reliance on livestock, as well as the type of care they give these animals, causally links the health of the villagers with that of their animal counterparts. This assertion is reflected by the evidence of zoonotic disease transmission from the previous year's study [40] and from the statistically significant association between animals being given medication in the previous four weeks and reported diarrhea. The univariate analysis showed a strong relationship with reported diarrhea, but showed an even

stronger effect when adjusting for multiple predictors and confounders that included age, sex, village location, and weekly income.

One potential explanation for this association is the emergence of antimicrobial resistance in these villages. The emergence and dissemination of livestock resistance to antimicrobial agents in strains of waterborne and zoonotic bacteria in rural Africa is not well established, but evidence for animal-to-human spread of antibiotic resistant bacteria is nevertheless mounting [48]. Some of the potential pathways observed that might be facilitating transmission of antimicrobial-resistant zoonotic bacteria from livestock to humans in these villages included direct contact with treated livestock, fecal contamination of food and water, migration of zebu between villages, trading livestock, handling of livestock feces, the use of livestock feces in fertilizer, and food consumption, which have all been documented in studies previously[49]. It's also possible that this association was confounded by simple zoonotic disease transmission where viral and parasitic pathogens or bacterial pathogens that are not antibiotic resistant were responsible. Without clinical data on livestock and humans it's difficult to confirm this relationship. Future studies on fecal samples taken during the 2011 pilot study and this year's field study should investigate this.

There was a sea of transmission pathways by which zoonotic disease transmission could take place in these villages though. In all three villages, a high number of poultry were observed to have roamed throughout the village during the day. In Ambatolahy and Ambodiaviavy where the majority of household structures were built to within one to three meters of each other, the density of poultry and dogs was very high. This was observed to increase the likelihood of interaction between these animals with reservoirs and humans.

In all three villages we observed a considerable amount of fecal material from poultry, pigs, zebu, and dogs on the ground surrounding the household and kitchen structures. Undoubtedly, this fecal material served as a major reservoir in multiple transmission pathway scenarios. Typically children would play outside and walk through the fecal material or touch the ground, increasing the likelihood of fecal-oral transmission. The same can also be said for men and women who stayed in the village during the day. Homemakers were in fact significantly associated with reported diarrhea in the univariate analysis, but were not significant in the multivariate analysis. In addition, only half of those surveyed reported having shoes or sandals, yet a majority of those observed were barefoot. Indeed, twice as many of those without shoes or sandals reported having diarrhea in the previous four weeks with the univariate analysis also showing a statistically significant association between not having shoes or sandals with reported diarrhea. Even though not having shoes or sandals did not make it to the final model, it is highly likely that going barefoot is a probable risk factor for diarrheal disease in these rural villages. Further investigation needs to be conducted before this can be said conclusively. Although, in 1995, Kightlinger et al. noted the same thing[8].

Fecal matter in the villages could potentially make its way into households and kitchens through animals and humans tracking it in. Chickens and dogs were observed to have entered both household and kitchen structures numerous times and sometimes during cooking and meal-time periods. Both of these animals could have shed hair and dust as feces-contaminated fomites once inside the households and kitchens, which has been shown with astrovirus, adenovirus, and rotavirus[50]. This could have potentially

contaminated the uncovered stored water and food in the kitchens exposing villagers to reservoirs of diarrhea causing pathogens.

During field observations we also witnessed run-off from rain mix with the fecal matter on the ground, which could have potentially made its way into households or exposed those who walked through this water barefoot. In Ambatolahy and Ambodiaviavy the area comprising the bulk of housing was typically uphill of the adjacent rivers that flowed by these villages, which meant run-off would flow into these rivers potentially contaminating the rivers, as well as the area in between. In areas of the village where water did not flow downhill large puddles would form, in some instances creating cesspools of contaminated water.

Handling animal feces for various purposes was also moderately prevalent in all three villages, showing a statistically significant association across villages. While not significantly associated with reported diarrhea, many villagers reported handling chicken, duck, pig, and zebu feces, animals most commonly associated with zoonotic disease transmission[51-53]. A moderate proportion of those who reported handling animal feces also reported having diarrhea in the past four weeks. This type of hygiene behavior represents the most direct pathway for fecal-oral transmission reported in these villages even though this behavior was rarely observed. In Ankialo however, adult men were frequently seen translocating manure to their fields for fertilizer, sometimes using their hands or touching shovels that had been touching zebu feces.

One of the most common prevention strategies for preventing the fecal-oral spread of diarrhea causing pathogens is handwashing, where it has been estimated that introducing handwashing with soap in a community could reduce the risk of diarrhea by 47%; especially in the most vulnerable and underserved populations[54]. However, in this rural region of Madagascar, and specifically in these three villages, handwashing with soap was a fairly limited hygiene behavior. Nearly all villagers reported washing their hands more than once a day with a majority (71.8%) also reporting they "sometimes" used soap. Handwashing in Ambatolahy and Ambodiaviavy took place at specific times of the day at one of the pumps in the village. In the morning most villagers washed their hands and some other part of the body before going on with the day's activities. This usually coincided with collecting water multiple buckets of water to be used throughout the day. Villagers may have over-reported the use soap during handwashing, as this was very rarely observed.

Handwashing is the most beneficial and cost-effective hand-hygiene behavior when done at certain times or events[54]. The most cited time that villagers washed their hands was before eating, a very critical time for washing hands. Unfortunately, an overwhelming majority of villagers didn't report washing their hands at other critical times including before preparing food, before collecting food, after going to the bathroom, before or after handling children, after working with livestock, before breastfeeding, after using animal feces, and after working in the field. Understandably, many of those surveyed may not have had these as responsibilities throughout the day. Oddly though, handwashing before eating was statistically associated with reported diarrhea at  $\alpha = 0.10$  ( $\chi^2$  p-value = 0.0751). This may be the result of four possible scenarios. One would be improper handwashing technique such as the lack of handwashing with soap, which was confirmed by the structured observations. The second involves the possibility of foodborne disease as a result of contaminated food. Likewise, the water villagers used to wash their hands may have been contaminated, which is fairly likely considering the water results from the 2011 pilot study[47]. Lastly, this may also be due to some type of sampling error not accounted for in the sampling design or analysis.

Another interesting finding regarding handwashing is that villagers an overwhelming amount of the time reported having diarrhea in the previous four weeks in the absence of handwashing at all of the critical times listed in Table 16. The lack of handwashing at crucial times in conjunction with the lack of soap may act as a primary behavior determining whether or not villagers in these three villages acquire infectious diarrhea.

Multivariate regression also indicated that trading goods or services with other villages was a statistically significant determinant of diarrheal disease, as opposed to trading goods or services within the village, which was statistically significant in a univariate analysis but did not make it in the final model. As mentioned previously, Sunday is the traditional market day for villages in this area of RNP where rural villages walk to the nearest town holding the market. In this scenario the translocation of food or fomites contaminated with enteric pathogens is very likely. The important aspect of this to focus on though is what is being traded. Unfortunately, we were unable to do structured observations at one of these events to confirm what exactly was being traded, how frequently trading was going on, and how many villages were involved in the trading process at these markets. As with all trading though, goods move fast whether from country to country or village to village[55]. This raises the risk for foodborne or enteric pathogens to translocate from one village to another village or one region of

Madagascar to another. Future studies, should study this phenomenon in more detail to understand the dynamics and consequences of trading goods between villages that may result in the spread of novel waterborne, foodborne and zoonotic enteric diseases.

Open defecation and being a homemaker as a profession were not significant in the multivariate model when adjusting for other covariates. This may be a result of confounding by other risk factors not in the model or a function of the other variables in the model contributing a stronger effect with reported diarrhea. Be as it may, open defecation may be associated with diarrhea via different causal linkages. While conducting observations, we set a primary objective of observing the events assigned for that time period, but also kept an eye out to see if villagers used the latrines as a secondary objective. In Ambatolahy, there were ten private latrines located throughout the village that were locked. Only once was a latrine observed to have been used in this village. In Ambodiaviavy, there was one private latrine and two public latrines, which were never observed to have been used. However, in Ankialo most of the households owned a private latrine, but were also never observed to have been used. It's highly probable that the villagers used the latrines when we weren't watching though. As they reported in the survey, nearly half of all villagers used a latrine to defecate. Yet, a majority of those who reported having diarrhea practiced open defecation. This may have been due to the fact that villagers who practiced open defecation typically did so at the edge of the village in a slightly forested area with banana trees and bushes. This was only observed in Ambodiaviavy where villagers were spotted walking into a forested area five to seven meters away from the nearest household structure to presumably defecate or urinate. Were we unable to confirm this in order to uphold their privacy, but

conversations with the surveyors led us to believe that these small forested areas were where "open defecation" typically took place in these villages. Assuming this is true, a majority of the villagers in these villages were barefoot, didn't wash their hands with soap, and did not use toilet paper. These hygiene behaviors would certainly put these villagers at a greater risk of acquiring an enteric pathogen.

Homemakers in these rural villages took on multiple duties throughout the day that exposed them to diarrhea causing pathogens. As well, adult women mostly played the role of homemaker in each household, which combined may have accounted for why this was not a significant predictor of reported diarrhea when adjusting for other covariates. Homemakers were typically in charge of house and yard cleaning, food preparation, cooking, collecting water, and sometimes even farming or livestock duties. Observations revealed that these responsibilities mostly took place in the village during the day, and usually in the presence of livestock and domestic animals. This meant increased interaction with these animals or fomites that came from these animals, including feathers, hair, and dust. Again, most villagers were observed to be barefoot, which meant the longer an individual walked within the village the higher the risk of making direct contact with animal feces.

Similarly, the separate role that men played compared to women and the role that children played compared to adults, determined their activities and behaviors throughout the day. This is what ultimately defined the level and quantity of interacting risk factors that lead to the exposure of diarrhea causing pathogens. While not statistically significant determinants of reported diarrhea, age, gender, village location, and weekly income certainly had separate and potentially modifying influence on the covariates and reported diarrhea.

Male and female children who remained in the village during the day were observed to have the co-highest exposure to animal feces and livestock in general. The high risk group, children under the age of fifteen, comprised half of those who reported diarrhea. They were also observed more often throughout the village, which is most likely due to their sheer density compared to that of adults. Interestingly, female children were more likely to help out adult women with the daily household responsibilities including food preparation, collecting water, etc. while the male children typically watched over the infants in the family or tended to the livestock. Because children were lower to the ground and played games that involved touching the ground, or in some cases the chickens, this may have put them at increased risk of coming into contact with fecal material, the animals, and food that was put out in the sun to dry. In Ambodiaviavy, there were multiple observations of infants walking, crawling, and sitting naked on dried beans and rice.

Adult women most likely held the co-highest exposure to animal feces and livestock compared to children and adult men. As previously described, most adult women were homemakers, but in many cases also participated or were in charge of the farming and livestock duties. Adult men however, were a majority of the time observed to wake up early in the morning, wash themselves at the pump (Ambatolahy and Ambodiaviavy) and take off to work in the field or head to their job. In many cases adult men wouldn't return back to the village or the household until dinner.

The difference in diarrhea prevalence among villages and that of the associating risk factors is a product of each village's distinct overall water, sanitation, and hygiene

culture, as well as socio-economic status (SES). Ankialo reported the lowest prevalence of diarrhea disease and correspondingly the results of the survey also showed they had the lowest quantity of risk factors for diarrheal disease. While Ankialo villagers traded with each other and other villages more often while handling human and animal feces the most, which together may have been the most probable determinants of diarrheal disease in this village, they also practiced open defecation less frequently, didn't bathe in waterways as often, treated their drinking water more frequently, and washed their hands at critical times more often with soap. In fact, they were the only village to have been observed using a tippy-tap for handwashing, which is an improved hardware for handwashing[56]. In addition, there were more students and farmers in this village, which may be a reflection of the socio-economic status of the village. Of the three villages this village was noticeably wealthier due to the larger houses, which were typically three stories. Villagers in Ankialo on average owned more livestock, especially zebu, which is a better proxy for socio-economic status in rural Madagascar than income. Homes in this village were very spread out compared to the other two villages, which meant the density of animals per square-meter was substantially less. As a result of this, livestock from different households rarely interacted, which may lower the chance for animal-to-animal and animal-to-human zoonotic disease transmission to happen. However, the first story of almost each house was dedicated to sheltering poultry and pigs, yet this did not appear to increase the frequency of interaction between livestock and human. However, this may be a bit presumptuous. Villagers here had larger areas for crops, especially rice paddy fields, and in many instances most homes or pair of homes had a latrine and bathroom. The lack of observations in these villages was not due to the lack of observation hours, but instead

reflects the lack of interactions by which waterborne and zoonotic enteric disease transmission can occur. The space between the houses and the environment, specifically the large rice paddy fields may play a decisive factor in the pathogen load within households and the village at large; ultimately acting as a barrier to the spread of pathogens

Ambatolahy reported the second highest prevalence of diarrhea among the three villages. This village reported trading amongst each other and with other villages the least, while treating their water least frequently and defecating in the open the second most of the villages. They washed their hands more frequently than villagers from Ankialo, while washing at critical times second most compared to the other villages. In addition, they used soap more often when handwashing while handling human and animal feces less frequently than the other villages too. However, they had the fewest farmers and second fewest students. This village was unique compared to the other two villages because this village was located right next to the main highway that passed through RNP. This afforded them increased access to travel as shown by their increased use of taxi-brousses. This village's location was extremely close to the national park and as such, many of the men and some women worked as tour-guides, local hotel staff, or employees of the local research facility, Centre ValBio. Working in the forest may have increased their exposure to wildlife, which represent additional vehicles of zoonotic diseases[57]. Because of their location to and employment at Centre ValBio, some of the villagers may have had a more advanced understanding of water, sanitation, and hygiene practices, which may have biased the survey results unfortunately. However, the close proximity of the homes meant that livestock interacted more frequently with humans, food, and water sources, which is

quite possibly increasing the chance for waterborne and zoonotic disease transmission. The topography of the land is hilly with some of the homes sitting on top of the hill while the majority of the households sit at the bottom. The springs and rice paddy terraces at the top of the hill are frequently used to clean laundry and by animals for drinking, which may be contaminating this water. The water from these sources then flows down to the main pump at the entrance of the village, which a majority of villagers use for bathing, colleting water, and cleaning items. Observations reveal frequent use of the two pumps, which is concerning considering these pumps may be contaminated with bacteria.

Ambodiaviavy had the highest prevalence of diarrhea and was characterized by the highest combination of interacting risk factors contributing to diarrheal disease in the village. Oddly, they reported treating their water the most and more frequently, although this was never observed and they may have considered rice water a form of treatment when answering the question. This is a limitation that needs to be addressed in future surveys given in this area. They traded amongst each other and with other villages the second most while defecating in the open more frequently. They reported washing their hands more frequently, yet not at the most critical times and less often with soap than the other two villages. Villagers handled human and animal feces second most and had the lowest number of students. The difference in this village was the proximity of households to each other and the density of livestock in the village, which certainly facilitated a higher chance for dissemination and acquisition of waterborne and zoonotic enteric diseases through multiple pathways. Observations revealed a higher incidence of livestock interaction with humans, food, and water sources. Also, the chance for handwashing was limited to morning and night when the pumps were turned on. In

addition, when it rained in this village the water either formed puddles in the village or drain down to the banana trees where villagers were hypothesized to openly defecate.

Weekly income was used as a proxy for SES in this community, but was not a statistically significant determinant of reported diarrhea. It did however have an effect on some of the other predictors by making them stronger predictors of diarrhea, such as animals given medication in the previous four weeks, not treating drinking water, and trading with other villages. This result may be due the weakness of this variable as a proxy for SES. It's also quite plausible that SES does not confound the relationship between the predictors and reported diarrhea. However, the difference seen between these villages would suggest that SES is at play with regards to their water, sanitation, and hygiene practices.

Surprisingly, the difference in health perception nearly matched the prevalence of diarrhea in these three villages. Ambodiaviavy reported the highest prevalence of diarrhea and, likewise reported diarrhea the most compared to the other villages in what they regarded as the first, second, and third biggest health problem in their households. Ankialo had the lowest diarrhea prevalence, but considered diarrhea to be one of their biggest health problems more so than Ambatolahy. Overall, the perception of diarrhea as a major health problem is not on equal footing with respiratory illness or malaria. This might represent a barrier to addressing diarrhea through preventative interventions if they do not prioritize diarrhea or the risk factors associated with it.

The structured observations served as a great method for studying these populations and gaining more realistic and tangible insight into transmission dynamics within these villages. Observations revealed unknown pathways that would have been

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impossible to capture by survey questionnaire alone. The most notable event happened during a livestock interaction observation period in Ambodiaviavy. The following is an excerpt from the livestock interaction observation form on July 6, 2012 at 8:27AM:

"[H]olding zebu in front of house, holding to calm it, before pregnancy ritual Pregnancy Ritual: Prior day pregnant woman had craving for zebu, ritual performed then where woman selected which part of animal she wanted to eat, which was bought at the market. Zebu brought to the room pregnant woman was in, held down, and woman's mouth and noise where forced against zebu's nostril so she could only breathe though the nostril, for about 3 minutes with a few breaks. People were gathered in the room. There was a bowl with inner intestines/lining/feces of zebu, solids were rung out and then placed asides. Liquids in a new bowl were handed to pregnant woman, she drank them. Adult male took out plates of various zebu organs and cut 1cmx1cmx1cm slices of raw liver. After pregnant woman finished drinking liquid, another adult female began to ring out solids again into a bowl to make more liquid. Pregnant woman ate 6 pieces of liver, then followed with liquid after. Adult male ate one piece of liver. Other woman poured remaining solids and liquids on pregnant woman, a lot spilled on flood (she was sitting on floor). Woman poured using her hands, then ran out of house, gagged, and rinsed hands with water from a cup, no soap."

Conversations with our translator revealed that this pregnancy ritual was quite common in this cultural setting, but had never been seen or recorded previously by any researcher or staff at Centre ValBio. This type of event was not carried out with concern of proper sanitation or hygiene and represents a potential catalyst for zoonotic disease transmission. An additional observation that represents a different type of danger was seen in Ankialo. The following is an excerpt from the collecting water and cleaning observation form on July 19, 2012 at 9:15AM:

"[P]laying with syringe, getting water from plastic watering can, baby." This event involved a male child and a female child playing with a syringe that had been used to inoculate an animal previously. The two children were filling it up with water and squirting each other and an infant child. It was not known whether or not anyone was stabbed with the syringe, but it's quite possible this may have happened when the observer was not watching moments later. Syringes were observed at some of the households in piles of trash throughout Ankialo as well. Villagers did report medicating their animals in the previous four weeks more than the other two villages.

The reality of what goes in these villages day-to-day and hour-to-hour is not something that could have been understood by a researcher through a simple or even indepth survey. The level of detail required in this setting would have been white-washed, inhibiting the ability of the researcher to dig through the noise to realize the different uniting risk factors that could potentially drive transmission cycles in this rural environment. The frequency of interactions observed in each village was judged to be a precise indicator of whether or not human behaviors were associated with the prevalence of diarrhea in a village. Similarly, they were also an indicator of whether or not livestock and domestic animal behaviors were associated with the prevalence of diarrhea in a village. The absence of both human and animal observations in Ankialo was associated with the reported diarrhea prevalence by those surveyed and the village at large. Likewise, the frequency of human and livestock observations in Ambatolahy was also associated with the reported diarrhea prevalence by those surveyed and the village as a whole. The same can definitively be said for Ambodiaviavy, the village with the highest frequency of human and animal observations and prevalence of reported diarrhea.

## Limitations

This field season was an opportunity to continue working with the villages that took part in the 2011 pilot study. It would have been ideal to increase the number of villages in the study or increase the number of households per village in order to increase the sample size. Increasing the sample size would have increased statistical power, but would also have given us an opportunity to observe additional villages. Indeed, the small sample size may have influenced the variance of the estimates, resulting in artificial effects. However, this should only have had a slight effect on the estimates, if any. This could have been addressed by treating the random sample design as a cluster sample design. Under this scenario we could have created weights adjusting for the sample size in each village to apply to each procedure in the statistical analysis.

Another shortcoming in our study is the Hawthorne effect. It was difficult to tell if our presence altered their behaviors or patterns of daily activities. It is possible, but a challenge to verify, whether their behavior remained the same at the beginning and changed over time or if their behavior changed in the beginning and normalized over time. This was not tested statistically due to time constraints. In addition, intra- and interobserver variation was not tested for agreement between observations. However, conducting open observations followed by unstructured observations in Ambatolahy and Ambodiaviavy should have considerably reduced the variation in observations between and within observers. Moreover, we were unable to observe individuals in the households we surveyed due to ethical constraints. Linking individual observation data with the corresponding survey data would have been advantageous for two reasons. It would have allowed us to check the degree of agreement between what was reported and what was observed using the kappa statistic [58]. Second, it would have given us a more comprehensive understanding of how individual behavior influences transmission dynamics in these types of settings. Adding to this, there were many instances where we were unable to observe certain events due to ethical or cultural considerations. Some of these events were vital to understanding the type and frequency of behaviors known to facilitate the spread and acquisition of diarrhea causing pathogens. Finally, we were limited to the amount of hours we were able to do to observations. Ideally, daily observations would take place over a longer period of time.

There were a few risk factors we were unable to measure that would have been beneficial to our investigation on zoonotic disease transmission and diarrheal disease prevalence. For instance, more detailed information about the villager's diarrheal episodes including the duration of diarrheal episode, the type of diarrhea (i.e. color, consistency, smell, watery, presence of absence of blood, etc.), the signs and symptoms of diarrheal disease, and whether or not medication was taken to treat the disease would have been ideal. More detailed information regarding personal hygiene, livestock and domestic animal relationships, and meal time practices would have added a new level of richness and authenticity.

Lastly, this field season took place during the three month dry season. The remainder of the year is the wet season, which even fewer researchers have conducted

studies during. During the wet season villagers migrate to their fields in order to work, which we hypothesize would change transmission dynamics quite considerably. Future studies should include field seasons during the months from September to April.

# **CONCLUSION**

This study set out to investigate human and livestock behaviors as risk factors for zoonotic disease transmission and diarrheal disease in a rural African setting. Zoonotic disease transmission is one of the most pressing factors contributing to emerging and reemerging diseases around the world. Likewise, enteric foodborne, waterborne, and zoonotic diseases plague some of the utmost vulnerable and underserved populations in the world. It is vital that we are able to pinpoint risk factors, or the combination of risk factors, that specifically determine whether or not a community or region is susceptible to endemic zoonotic disease transmission. The consequence of which is diarrheal disease, a debilitating disease that is of major concern for high risk groups such as children under the age of five, those who are immunocompromised, and adults above 65 years of age[59]

To tackle the complexities of transmission dynamics and diarrheal disease we utilized a mixed-methods approach to build on the results from the 2011 pilot study in Ranomafana National Park, Madagascar. The prevalence of reported diarrhea was the highest in Ambodiaviavy, followed by Ambatolahy, and the lowest in Ankialo. Risk factors reported in the survey questionnaire and observed during structured observations concur with the prevalence of reported diarrhea by each village. Across all villages, there was variety of culturally influenced water, sanitation, and hygiene behaviors in conjunction with livestock and domestic animal behavior that showed statistically significant associations with reported diarrhea. Specifically, animals given medication in the previous four weeks, not treating drinking water, and trading with other villages were all significant determinants of diarrheal disease in all three villages.

The associations found by the survey questionnaire were confirmed by observations that revealed what human and animal behavior risk factors were prevalent in these villages. The lack of handwashing at critical junctures, as well as deficiency in the use of soap is of great cause for concern. In addition, it's hypothesized that zoonotic antimicrobial-resistant bacteria dissemination and acquisition by humans is the explanation behind the link between animals given medication and human reported diarrhea. This is an alarming assertion, but clinical tests need to be performed in order to test this hypothesis. Observations and survey results revealed the lack of water treatment and its potential role in determining diarrhea. Furthermore, trading goods and services in other villages is also a significant determinant of diarrhea disease due to the potential translocation of infectious diseases from one village to another.

The role that livestock play in these villages is fundamental to their economic survival, but they are also causally linked to their health. Animal fecal material acts as a reservoir that plays multiple roles in the transmission dynamics of these villages. For instance, many villagers reported handling animal feces, a behavior most prevalent in Ankialo. Likewise, water is a potential vehicle in Ambatolahy and Ambodiaviavy. Water sample results from the 2011 pilot study show that water sources may be contaminated with bacteria, which may be the result of fecal contamination from animals or humans. Age and gender roles also determine an individual's daily activities and routine, which predisposes one to a specific level of daily risk to diarrheal disease acquisition. The difference in diarrheal disease prevalence was explained by the differing water, sanitation, and hygiene cultures unique to each of the three villages. Ambodiaviavy reported and was observed to have the poorest water, sanitation, and hygiene practices, which ultimately explained their reported diarrhea prevalence. Further, while SES may provide a moderating effect on behaviors, using weekly income as a proxy for SES did not show a demonstrable effect on the other predictors of reported diarrhea. Reported perception of health problems largely agreed with that of diarrhea prevalence across villages with Ambodiaviavy reporting the highest diarrheal prevalence while affirming that diarrhea was one of their biggest health problems.

Structured observations revealed unknown transmission pathways and routes never before observed or recorded in these villages. In Ambodiaviavy, a rare pregnancy ritual involving the ingestion of raw zebu organs was observed. In Ankialo, two children were observed playing with a syringe that was previously used to inoculate an animal. Known pathways and routes of transmission were observed in great detail, highlighting the use of observations to elucidate diverse components about transmission dynamics in these villages. Conducting structured observations confirmed that human and animal behaviors were associated with diarrheal disease prevalence in the villages of RNP. This however cannot be confirmed statistically.

The utilization of a mixed-methods approach in this rural setting was unprecedented in Madagascar and substantially increased the strength of this study. Despite certain shortcomings including a small sample size, limitations of the observations, additional unmeasured risk factors, and the inability to account for seasonality, results from the survey and structured observations painted a detailed picture of life in these villages. Even though the survey was a snapshot of time, the observations opportunistically filled crucial behavioral gaps and illuminated key time periods, locations, and events where transmission and infection could happen in these villages. Diarrheal disease may be a pressing health problem in this region of Madagascar, but this study has shed light on those water, sanitation, and hygiene risk factors that determine this disease, paving the way for future interventions.

#### Recommendations

This study increased the depth and detail of knowledge about specific water, sanitation, and hygiene behaviors that impact waterborne and zoonotic disease transmission and diarrheal disease in these villages. Many studies have assessed appropriate interventions aimed at population health improvements that address these issues[46]. These interventions include hygiene, sanitation, water supply, water quality, or multiple interventions. In order to develop proper and site specific interventions in this rural Malagasy setting, local desirability, feasibility, sustainability, cost-effectiveness and health benefits need to be considered. The Health and Hygiene team at Centre ValBio have worked with villages in and around RNP before addressing water, sanitation, and hygiene issues, but to seemingly no avail. No doubt, this is a daunting task given the conditions of villages in RNP and the cultural barriers that might limit some of the most effective interventions used today. Nevertheless, given that some level of improved water, sanitation, and hygiene practice would greatly benefit those in these villages, it is recommended Centre ValBio and other partner organizations continue to work with villagers to reduce the health burden of waterborne and zoonotic diseases. Briefly, hardware such as point-source water treatment and handwashing stations might prove to

be effective and sensible interventions in these villages. However, numerous studies reported behavior change as a more effective intervention than improved infrastructure[46]. Therefore, educating villagers on why simple tasks such as keeping animals out of the house and kitchen, as well as the promotion of hand soap as a desirable consumer product could prove to be highly effective interventions.

Future studies should address the knowledge, attitudes, and practices of these villagers on water, sanitation, hygiene, and livestock issues in order to gauge if these or other interventions would be adopted more wholeheartedly by these villages. If results from our questions asking about their biggest health problems are any indicator, respiratory illness and malaria might take precedence over diarrheal disease. However, results from this study indicate the potential for the emergence or re-emergence of zoonotic diseases in these rural villages of Madagascar. Additionally, future studies need to incorporate the influence of wildlife on transmission dynamics in RNP. Further deforestation and climate change may inadvertently increase the interaction between wildlife, such as lemurs, and the inhabitants of these villages.

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# Tables

		Ambatolahy	Ambodiaviavy	Ankialo	OVERALL	
		N=54	N=66	N=70	N=190	
Variable		n (%)	n (%)	n (%)	n (%)	p-value*
Gender						-
	Female	25 (46.3)	31 (47.0)	28 (40.0)	84 (44.2)	0.6905
	Male	29 (53.7)	35 (53.0)	42 (60.0)	106 (55.8)	
Mean A	ge (SD)	23.8 (19.4)	24.3 (18.5)	22.4 (16.2)	23.4 (17.9)	-
Age						
	$\leq$ 4 years old	5 (9.3)	5 (7.6)	3 (4.3)	13 (6.8)	
	5 - 14 years old	19 (35.2)	21 (31.8)	27 (38.6)	67 (35.3)	
	15 - 32 years old	14 (25.9)	20 (30.3)	23 (32.9)	57 (30.0)	0.9515
	33 - 53 years old	11 (20.4)	14 (21.2)	12 (17.1)	37 (19.5)	
	> 53 years old	5 (9.3)	6 (9.1)	5 (7.1)	16 (8.4)	
Tribe						
	Betsileo	53 (98.2)	39 (59.1)	70 (100.0)	162 (85.3)	
	Tanala	0 (0.0)	27 (40.9)	0 (0.0)	27 (14.2)	< 0.0001
	Vakinakaratra	1 (1.9)	0 (0.0)	0 (0.0)	1 (0.5)	
Employe	ed and/or student	45 (83.3)	56 (84.9)	66 (94.3)	167 (87.9)	0.1022
Primary	Profession*					
	Business/trade (not animal)	5 (9.3)	3 (4.5)	7 (10.0)	15 (7.9)	0.4531
	Business/trade (animal)	1 (1.9)	0 (0.0)	3 (4.3)	4 (2.1)	0.2044
	Civil servant	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	-
	Other professional	2 (3.7)	0 (0.0)	0 (0.0)	2 (1.1)	0.0797
	Farmer	17 (31.5)	34 (51.5)	39 (55.7)	90 (47.4)	0.0195
	Animal care	3 (5.6)	7 (10.6)	6 (8.6)	16 (8.4)	0.6109
	Centre Valbio	3 (5.6)	0 (0.0)	1 (1.4)	4 (2.1)	0.1013
	Ranomafana National Park	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	-
	Tourist guide	1 (1.9)	0 (0.0)	0 (0.0)	1 (0.5)	0.2842
	Hotel employee	1 (1.9)	0 (0.0)	0 (0.0)	1 (0.5)	0.2842
	Teacher	0 (0.0)	1 (1.5)	0 (0.0)	1 (0.5)	0.6316
	Health care	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	-
	Student	20 (37.0)	21 (31.8)	31 (44.3)	72 (37.9)	0.3219
	Homemaker	10 (18.5)	18 (27.3)	14 (20.0)	42 (22.1)	0.4477
	Other	17 (31.5)	9 (13.6)	6 (8.6)	32 (16.8)	0.0023
Weekly	Income - Malagasy Ariary (AR)	Ť				
	0 AR (No Income)	33 (61.1)	40 (60.6)	44 (62.9)	117 (61.6)	
	1 AR - 4,999 AR	4 (7.4)	5 (7.6)	2 (2.9)	11 (5.8)	
	5,000 AR - 9,999 AR	6 (11.1)	6 (9.1)	10 (14.3)	22 (11.6)	0.8720
	10,000 AR - 19,999 AR	5 (9.3)	7 (10.6)	4 (5.7)	16 (8.4)	
	≥ 20,000 AR	6 (11.1)	8 (12.1)	10 (14.3)	24 (12.6)	

TABLE 1. Basic demographics of human subjects by villag

\* Subjects asked to report all professions

† \$1 = 2260.23 AR

		Ambatolahy	Ambodiaviavy	Ankialo	OVERALL	
		N=54	N=66	N=70	N=190	
Variable		n (%)	n (%)	n (%)	n (%)	p-value
Trade with oth	er villagers	3 (5.6)	9 (13.6)	11 (15.9)	23 (12.2)	0.1782
Trade with oth	er villages	3 (5.6)	6 (9.2)	11 (15.7)	20 (10.6)	0.1876
Tending livest	ock					
No	ot a responsibility	43 (79.6)	60 (90.9)	64 (91.4)	167 (87.9)	
Μ	ore than once a day	3 (5.6)	0 (0.0)	0 (0.0)	3 (1.6)	
Oı	nce a day	3 (5.6)	1 (1.5)	3 (4.3)	7 (3.7)	0.3054
3 t	to 5 times a day	2 (3.7)	2 (3.0)	2 (2.9)	6 (3.2)	
Oı	nce a week	1 (1.9)	1 (1.5)	1 (1.4)	3 (1.6)	
Le	ess than once a week	2 (3.7)	2 (3.0)	0 (0.0)	4 (2.1)	
Collecting wat	er					
No	ot a responsibility	39 (72.2)	59 (89.4)	54 (77.1)	152 (80.0)	
Μ	ore than once a day	4 (7.4)	0 (0.0)	0 (0.0)	4 (2.1)	
	nce a day	5 (9.3)	5 (7.6)	4 (5.7)	14 (7.4)	0.0086
3 t	to 5 times a day	4 (7.4)	1 (1.5)	6 (8.6)	8 (4.2)	
Oı	nce a week	2 (3.7)	1 (1.5)	7 (10.0)	12 (6.3)	
Le	ess than once a week	0 (0.0)	0 (0.0)	2 (2.9)	0 (0.0)	
Cooking			· · ·	. ,		
No	ot a responsibility	43 (79.6)	55 (83.3)	50 (71.4)	148 (77.9)	
	ore than once a day	2 (3.7)	1 (1.5)	1 (1.4)	4 (2.1)	
Oı	nce a day	2 (3.7)	2 (3.0)	4 (5.7)	8 (4.2	0.6946
3 t	to 5 times a day	2 (3.7)	1 (1.5)	6 (8.6)	9 (4.7)	
Oı	nce a week	5 (9.3)	5 (7.6)	7 (10.0)	17 (9.0)	
Le	ess than once a week	0 (0.0)	2 (3.0)	2 (2.9)	4 (2.1)	
Hunting						
No	ot a responsibility	48 (88.9)	66 (100.0)	66 (94.3)	180 (94.7)	
Μ	ore than once a day	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	
Oı	nce a day	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0.0249
3 t	to 5 times a day	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	
Oı	nce a week	1 (1.9)	0 (0.0)	2 (2.9)	3 (1.6)	
Le	ess than once a week	5 (9.3)	0 (0.0)	2 (2.9)	7 (3.7)	
House and yar	d cleaning					
No	ot a responsibility	36 (66.7)	44 (66.7)	49 (70.0)	129 (67.9)	
Μ	ore than once a day	2 (3.7)	1 (1.5)	0 (0.0)	3 (1.6)	
Oı	nce a day	9 (16.7)	7 (10.6)	5 (7.1)	21 (11.1)	0.2939
3 t	to 5 times a day	2 (3.7)	6 (9.1)	3 (4.3)	11 (5.8)	
Oı	nce a week	3 (5.6)	6 (9.1)	12 (17.1)	21 (11.1)	
Le	ess than once a week	2 (3.7)	2 (3.0)	1 (1.4)	5 (2.6)	
Using animal fo	eces					
-	ot a responsibility	35 (64.8)	47 (71.2)	34 (48.6)	116 (61.1)	
	ore than once a day	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	
Oı	nce a day	1 (1.9)	1 (1.5)	0 (0.0)	1 (1.1)	0.0583
	to 5 times a day	0 (0.0)	0 (0.0)	3 (4.3)	3 (1.6)	
	nce a week	4 (7.4)	3 (4.6)	10 (14.3)	17 (9.0)	
Le	ess than once a week	6 (25.9)	15 (22.7)	23 (32.9)	52 (27.4)	

TABLE 2. Trading and household responsibilities of human subjects by village

	Ambatolahy	Ambodiaviavy	Ankialo	OVERALL	
	N=54	N=66	N=70	N=190	
Variable	n (%)	n (%)	n (%)	n (%)	p-value*
Defecate					
Latrine	38 (70.4)	13 (19.7)	50 (71.4)	101 (53.2)	< 0.0001
Open	16 (29.6)	53 (80.3)	20 (28.6)	89 (46.8)	
Bathe†					
Bathroom	5 (9.3)	6 (9.1)	19 (27.1)	30 (15.8)	0.0046
Waterway‡	49 (90.7)	59 (89.4)	24 (34.3)	132 (69.5)	< 0.0001
Field	0 (0.0)	0 (0.0)	11 (15.7)	11 (5.8)	< 0.0001
Behind house	1 (1.9)	1 (1.5)	11 (15.7)	13 (6.8)	0.0011
Everywhere	0 (0.0)	0 (0.0)	4 (5.7)	9 (4.7)	< 0.0001
Don't bathe	0 (0.0)	0 (0.0)	9 (12.9)	4 (2.1)	0.0371

TABLE 3. Defecation and bathing practices of human subjects by village

†Subjects asked to report all bathing practices with some subjects reporting more than one

‡Waterway includes rivers, streams, springs, canals, ponds, and lakes

TABLE 4. Water practices	of human	subjects	by village
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	Ambatolahy	Ambodiaviavy	Ankialo	OVERALL	
	N=54	N=66	N=70	N=190	
Variable	n (%)	n (%)	n (%)	n (%)	p-value*
Treat drinkning water	5 (9.3)	21 (32.3)	17 (24.6)	43 (22.8)	0.0107
Drinking water treatment technique					
Boil	3 (60.0)	15 (93.8)	15 (93.8)	33 (89.2)	
Filter through cloth/gauze	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	
Settle	0 (0.0)	1 (6.3)	0 (0.0)	1 (2.7)	0.0606
Chlorine/Sur'eau	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	
Other	2 (40.0)	0 (0.0)	1 (6.3)	3 (8.1)	
Frequency of treating drinking water					
Sometimes (< once per week)	2 (40.0)	10 (50.0)	13 (76.5)	25 (59.5)	
Often (> once per week)	3 (60.0)	5 (25.0)	4 (25.5)	12 (28.6)	0.0781
Always (every time you drink)	0 (0.0)	5 (25.0)	0 (0.0)	5 (11.9)	
Use to take water from storage container	†				
Bucket	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	-
Cup	54 (28.4)	64 (33.7)	67 (35.3)	185 (97.4)	0.4426
Hands	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	-
Pour directly from container	0 (0.0)	2 (1.1)	7 (3.7)	9 (4.7)	0.0247
Other	20 (10.5)	22 (11.6)	22 (11.6)	64 (33.7)	0.8046
Use stored water‡					
Drinking	54 (100.0)	66 (100.0)	69 (98.6)	189 (99.5)	1.0000
Bathing	3 (5.6)	5 (7.6)	34 (48.6)	42 (22.1)	< 0.0001
Cooking	42 (77.8)	46 (69.7)	50 (71.4)	138 (72.6)	0.5897
Washing fruit and vegetables	42 (77.8)	45 (68.2)	42 (60.0)	129 (67.9)	0.1095
Washing meat to cook	41 (75.9)	45 (68.2)	39 (55.7)	125 (65.8)	0.0553
Washing dishes and utensils	46 (85.2)	47 (71.2)	43 (61.4)	136 (71.6)	0.0134
Washing clothes	1 (1.9)	4 (6.1)	2 (2.9)	7 (3.7)	0.5611
Giving to animals	3 (5.6)	4 (6.1)	32 (45.7)	39 (20.5)	< 0.0001
Other	15 (27.8)	32 (48.5)	43 (61.4)	90 (47.4)	0.0010

\*Calculated by Chi-square test or Fisher's exact test

†Subjects asked to report all ways of taking water from storage container

\$Subjects asked to report all uses for stored water

TABLE 5. Handwashing practices of human subje
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		Ambatolahy	Ambodiaviavy	Ankialo	OVERALL	
		N=54	N=66	N=70	N=190	
Variable		n (%)	n (%)	n (%)	n (%)	p-value
Frequency	of washing hands					
	More than once a day	52 (96.3)	66 (100.0)	66 (94.3)	184 (96.8)	
	Once a day	1 (1.9)	0 (0.0)	4 (5.7)	5 (2.6)	
	3 to 5 times a week	1 (1.9)	0 (0.0)	0 (0.0)	1 (0.5)	0.0540
	Once a week	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	
	Never	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	
	Other	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	
Handwash	ing times†‡					
	Food preperation					
	Before	21 (38.9)	18 (27.3)	13 (18.6)	52 (27.4)	0.0422
	After	1 (1.9)	1 (1.5)	0 (0.0)	2 (1.1)	0.5322
	Food collection					
	Before	1 (1.9)	0 (0.0)	0 (0.0)	1 (0.5)	0.2842
	After	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	-
	Bathroom					
	Before	0 (0.0)	2 (3.0)	0 (0.0)	2(1.1)	0.1992
	After	39 (72.2)	20 (30.3)	22 (31.4)	81 (42.6)	< 0.000
	Dealing with kids					
	Before	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	-
	After	2 (3.7)	3 (4.6)	9 (12.9)	14 (7.4)	0.1028
	Working with livestock					
	Before	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	-
	After	12 (22.2)	11 (16.7)	23 (32.9)	46 (24.2)	0.0815
	Eating			× ,	~ /	
	Before	46 (85.2)	60 (90.9)	56 (80.0)	162 (85.3)	0.2001
	After	3 (5.6)	0 (0.0)	1 (1.4)	4 (2.1)	0.1013
	Breastfeeding	× ,	× ,		× ,	
	Before	0 (0.0)	1 (1.5)	0 (0.0)	1 (0.5)	0.6316
	After	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	-
	Using animal feces					
	Before	1 (1.9)	0 (0.0)	0 (0.0)	1 (0.5)	0.2842
	After	13 (24.1)	13 (19.7)	26 (37.1)	52 (27.4)	0.0647
	Farming in the field			× /		
	Before	1 (1.9)	0 (0.0)	0 (0.0)	1 (0.5)	0.2842
	After	20 (37.0)	37 (56.1)	39 (55.7)	96 (49.5)	0.0641
	Other	27 (50.0)	33 (50.0)	42 (60.0)	102 (53.7)	0.4111

†Subjects asked to report all handwashing times

\$Surveyors did not read options out loud

	Ambatolahy	Ambodiaviavy	Ankialo	OVERALL	
	N=54	N=66	N=70	N=190	
Variable	n (%)	n (%)	n (%)	n (%)	p-value*
Soap in the household					
Always	12 (22.6)	3 (4.6)	18 (26.5)	33 (17.7)	
Sometimes	41 (77.4)	62 (93.9)	50 (73.5)	153 (81.8)	0.0010
Never	0 (0.0)	1 (1.5)	0 (0.0)	1 (0.5)	
Frequency of washing hands with soap					
Always	10 (19.2)	1 (1.5)	7 (10.0)	18 (9.6)	
Sometimes	34 (65.4)	53 (80.3)	48 (68.8)	135 (71.8)	0.0188
Do not wash hands with soap	8 (15.4)	12 (18.2)	15 (21.4)	35 (18.6)	
Additional use of soap†					
To wash for market	29 (53.7)	40 (60.6)	55 (78.6)	124 (65.3)	0.0096
Washing clothes	48 (88.9)	55 (83.3)	60 (85.7)	163 (85.8)	0.6865
Washing kitchen items	37 (68.5)	26 (39.4)	18 (25.7)	81 (42.6)	< 0.0001
Bathing bodies	53 (98.2)	61 (92.4)	65 (92.9)	179 (94.2)	0.3369
Washing hair	52 (96.3)	61 (92.4)	63 (90.0)	176 (92.6)	0.4197
Other	10 (18.5)	8 (12.2)	14 (20.0)	32 (16.8)	0.4367
Source of soap‡					
Market	14 (25.9)	53 (80.3)	63 (90.0)	130 (68.4)	< 0.0001
Other villager	0 (0.0)	32 (48.5)	29 (41.4)	61 (32.1)	< 0.0001
Store in the village	51 (94.4)	52 (78.8)	46 (65.7)	149 (78.4)	< 0.0001
Donation	0 (0.0)	0 (0.0)	2 (2.9)	2 (1.1)	0.3337
Other	1 (1.9)	0 (0.0)	0 (0.0)	1 (0.5)	0.2842
Would learn to make soap	41 (77.4)	51 (79.7)	55 (80.9)	147 (79.5)	0.8915

TABLE 6. Use of soap practices of human subjects by villages

†Subjects asked to report all uses of soap

\$Subjects asked to report all sources of soap

		Ambatolahy	Ambodiaviavy	Ankialo	OVERALL	
		N=54	N=66	N=70	N=190	
Variable		n (%)	n (%)	n (%)	n (%)	p-value*
Drink o	ther than water†					
	Tea	31 (57.4)	52 (78.8)	33 (47.1)	116 (61.1)	0.0006
	Juice	46 (85.2)	64 (97.0)	69 (98.6)	179 (94.2)	0.0054
	Coffee	35 (64.8)	34 (51.5)	34 (48.6)	103 (54.2)	0.1707
	Beer	13 (24.1)	36 (54.6)	41 (58.6)	90 (47.4)	0.0002
	Toaka gasy	11 (20.4)	18 (27.3)	48 (68.6)	77 (40.5)	< 0.0001
	Rice water	51 (94.4)	66 (100.0)	66 (94.3)	183 (96.3)	0.1171
	Only water	53 (98.2)	64 (97.0)	66 (94.3)	184 (96.8)	0.8771
	Other	24 (44.4)	32 (48.5)	48 (68.6)	104 (54.7)	0.0121
Freque	ncy of eating rice during harvest time					
	Less than once a day	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	
	Once	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	
	Twice	0 (0.0)	10 (15.2)	0 (0.0)	10 (5.3)	< 0.0001
	Three times	53 (100.0)	54 (81.8)	66 (95.7)	173 (92.0)	
	More than three times	0 (0.0)	2 (3.0)	3 (4.4)	5 (2.7)	
Smoke						
	Never	50 (94.3)	62 (93.9)	64 (91.4)	176 (93.1)	
	Sometimes (≤once per week)	0 (0.0)	1 (1.5)	2 (2.9)	3 (1.6)	0.3795
	Often (3-5 times per week)	2 (3.8)	0 (0.0)	0 (0.0)	2(1.1)	
	Daily	1 (1.9)	3 (4.6)	4 (5.7)	8 (4.2)	
Che win	g tobacco					
	Never	36 (67.9)	51 (78.5)	62 (88.6)	149 (79.3)	
	Sometimes (≤once per week)	0 (0.0)	1 (1.5)	0 (0.0)	1 (0.5)	0.0363
	Often (3-5 times per week)	1 (1.9)	1 (1.5)	0 (0.0)	2 (1.1)	
	Daily	16 (30.2)	12 (18.5)	8 (11.4)	36 (19.2)	
Alcohol	l					
	Never	41 (77.4)	40 (61.5)	20 (28.6)	101 (79.3)	
	Sometimes (≤once per week)	7 (13.2)	22 (33.9)	46 (65.7)	75 (39.9)	< 0.0001
	Often (3-5 times per week)	3 (5.7)	2 (3.1)	4 (5.7)	9 (4.8)	
	Daily	2 (3.8)	1 (1.5)	0 (0.0)	3 (1.6)	

†Subjects asked to report all liquids they drink

#### TABLE 8. Transportation and foot hygiene practices of human subjects by village

	Ambatolahy	Ambodiaviavy	Ankialo	OVERALL	
	N=54	N=66	N=70	N=190	
Variable	n (%)	n (%)	n (%)	n (%)	p-value*
Primary mode of transportation†					
Cart	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	-
Taxi-brousse	51 (94.4)	36 (54.6)	22 (31.4)	109 (57.4)	< 0.0001
Private car	5 (9.3)	0 (0.0)	0 (0.0)	5 (2.6)	0.0016
Motorcycle	0 (0.0)	0 (0.0)	4 (5.7)	4 (2.1)	0.0371
Bike	4 (7.4)	5 (7.6)	1 (1.4)	10 (5.3)	0.1707
Foot	51 (94.4)	60 (90.9)	68 (97.1)	179 (94.2)	0.2847
Other	5 (9.3)	5 (7.6)	3 (4.3)	13 (6.8)	0.4816
Have shoes or sandals					
Yes	n.a.	39 (59.1)	32 (45.7)	71 (52.2)	0.1186
No	n.a.	27 (40.9)	38 (54.3)	65 (47.8)	

n.a. = not available

\*Calculated by Chi-square test or Fisher's exact test

†Subjects asked to report all modes of transportation

	Ambatolahy	Ambodiaviavy	Ankialo	OVERALL	
	N=54	N=66	N=70	N=190	
Variable	n (%)	n (%)	n (%)	n (%)	p-value*
Taken medication in past 4 weeks	34 (63.0)	36 (54.5)	35 (50.0)	105 (56.1)	0.3512
Symptoms improved from medication	30 (88.2)	35 (97.2)	34 (97.1)	99 (94.3)	0.2782
Animal taken medication in past 4 weeks	6 (11.8)	13 (19.7)	27 (38.6)	46 (24.6)	0.0017
Animal symptoms improved from medication					
Yes	4 (66.7)	12 (92.3)	19 (70.4)	35 (76.1)	0.2636
No response	2 (33.3)	1 (7.7)	8 (29.6)	11 (23.9)	

TABLE 9. Medication practices for human subjects and their animals by village

	Ambatolahy	Ambodiaviavy	Ankialo	OVERALL	
	N=54	N=66	N=70	N=190	
Variable	n (%)	n (%)	n (%)	n (%)	p-value*
Handled human feces	25 (46.3)	31 (47.0)	35 (50.0)	91 (47.9)	0.9038
Handled animal feces	28 (51.9)	32 (49.2)	53 (75.7)	113 (59.8)	0.0027
Species†					
Cat	1 (1.9)	0 (0.0)	3 (4.3)	4 (2.1)	0.0562
Chicken	24 (44.4)	22 (33.3)	37 (52.9)	83 (43.7)	0.0713
Dog	1 (1.9)	1 (1.5)	0 (0.0)	2 (1.1)	0.1985
Duck	6 (11.1)	6 (9.1)	14 (20.0)	26 (13.9)	0.1462
Geese	2 (3.7)	0 (0.0)	6 (8.6)	8 (4.2)	0.0052
Pig	10 (18.5)	9 (13.6)	34 (48.6)	53 (27.9)	< 0.0001
Rabbit	5 (9.3)	0 (0.0)	0 (0.0)	5 (2.6)	0.0016
Rodent	1 (1.9)	0 (0.0)	1 (1.4)	2 (1.1)	0.2105
Turkey	1 (1.9)	0 (0.0)	0 (0.0)	1 (0.5)	0.2842
Zebu	15 (27.8)	25 (37.9)	28 (40.0)	68 (35.8)	0.3373

TABLE 10. Feces handling practices of human subjects by village

\*Calculated by Chi-square test or Fisher's exact test

†Subjects asked to report all species

		Ambatolahy N=52	Ambodiaviavy N=65	Ankialo N=70	OVERALL N=187	
Predictor	vs. reported diarrhea	n (%)	n (%)	n (%)	n (%)	p-value*
Reporte	d diarrhea†‡	10 (19.2)	15 (23.1)	12 (17.1)	37 (19.8)	0.6833*
Gender						
	Female	4 (7.7)	9 (13.9)	6 (8.6)	19 (10.2)	0.3410
	Male	6 (11.5)	6 (9.2)	6 (8.6)	18 (9.6)	
Age						
	$\leq$ 4 years old	1 (1.9)	0 (0.0)	1 (1.4)	2 (1.1)	
	5 - 14 years old	5 (9.6)	6 (9.2)	5 (7.1)	16 (8.6)	
	15 - 32 years old	1 (1.9)	4 (6.2)	4 (5.7)	9 (4.8)	0.7420
	33 - 53 years old	3 (5.8)	3 (4.6)	0 (0.0)	6 (3.2)	
	> 53 years old	0 (0.0)	2 (3.1)	2 (2.9)	4 (2.1)	
Tribe						
	Betsileo	10 (18.5)	4 (6.1)	12 (17.1)	26 (14.0)	0.0020
	Tanala	0 (0.0)	11 (16.7)	0 (0.0)	11 (5.9)	
Employe	d and/or student	8 (15.4)	14 (21.5)	11 (15.7)	33 (17.7)	1.0000
Primary	Profession					
	Business/trade (not animal)	0 (0.0)	0 (0.0)	2 (2.9)	2 (1.1)	0.7392
	Business/trade (animal)	0 (0.0)	0 (0.0)	1 (1.4)	1 (0.5)	1.0000
	Civil servant	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	-
	Other professional	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1.0000
	Farmer	4 (7.7)	8 (12.3)	7 (10.0)	19 (10.2)	0.5592
	Animal care	0 (0.0)	2 (3.1)	3 (4.3)	5 (2.7)	0.1815
	Centre Valbio	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1.0000
	Ranomafana National Park	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	-
	Tourist guide	1 (1.9)	0 (0.0)	0 (0.0)	1 (0.5)	0.1979
	Hotel employee	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1.0000
	Teacher	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1.0000
	Health care	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	-
	Student	4 (7.7)	6 (9.2)	6 (8.6)	16 (8.6)	0.4604
	Homemaker	2 (3.9)	7 (10.8)	4 (5.7)	13 (7.0)	0.0301
	Other	1 (1.9)	2 (3.1)	3 (4.3)	6 (3.2)	0.8716
Weekly	Income - Malagasy Ariary (AR)					
	0 AR (No Income)	6 (11.5)	8 (12.3)	8 (11.4)	22 (11.8)	
	1 AR - 4,999 AR	1 (1.9)	3 (4.6)	0 (0.0)	4 (2.1)	
	5,000 AR - 9,999 AR	1 (1.9)	0 (0.0)	3 (4.3)	4 (2.1)	0.4060
	10,000 AR - 19,999 AR	2 (3.9)	2 (3.1)	0 (0.0)	4 (2.1)	
	$\geq$ 20,000 AR	0 (0.0)	2 (3.1)	1 (1.4)	3 (1.6)	

TABLE 11. Basic demographics of human subjects that reported diarrhea by village

† Three subjects did not respond to the set of question inquiring into whether or not subjects had diarrhea with blood and seperately without blood

‡ Syndromic diarrhea with and without blood

\*\*P-value for location (villages #1-3 combined) vs. reported diarrhea

	Ambatolahy	Ambodiaviavy	Ankialo	OVERALL	
	N=52	N=65	N=70	N=187	
Predictor vs. reported diarrhea	n (%)	n (%)	n (%)	n (%)	p-value
Trade with other villagers	1 (1.9)	5 (7.7)	3 (4.4)	9 (4.8)	0.0085
Trade with other villages	2 (3.9)	3 (4.7)	3 (4.3)	8 (4.3)	0.0081
Household responsibility - Tending livestock					
Not a responsibility	10 (19.2)	14 (21.5)	11 (15.7)	35 (18.7)	
More than once a day	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	
Once a day	0 (0.0)	0 (0.0)	1 (1.4)	1 (0.5)	0.7162
3 to 5 times a day	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	
Once a week	0 (0.0)	1 (1.5)	0 (0.0)	1 (0.5)	
Less than once a week	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	
Household responsibility - Collecting water					
Not a responsibility	9 (17.3)	14 (21.5)	8 (11.4)	31 (16.6)	
More than once a day	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	
Once a day	0 (0.0)	1 (1.5)	2 (2.9)	3 (1.6)	0.8105
3 to 5 times a day	1 (1.9)	0 (0.0)	1 (1.4)	2(1.1)	
Once a week	0 (0.0)	0 (0.0)	1 (1.4)	1 (0.5)	
Less than once a week	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	
Household responsibility - Cooking	. ,			· · /	
Not a responsibility	8 (15.4)	13 (20.0)	9 (12.9)	30 (16.0)	
More than once a day	0 (0.0)	1 (1.5)	0 (0.0)	1 (0.5)	
Once a day	1 (1.9)	0 (0.0)	1 (1.4)	2 (1.1)	0.6839
3 to 5 times a day	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	
Once a week	1 (1.9)	0 (0.0)	2 (2.9)	3 (1.6)	
Less than once a week	0 (0.0)	1 (1.5)	0 (0.0)	1 (0.5)	
Household responsibility - Hunting	. ,			. ,	
Not a responsibility	9 (17.3)	15 (23.1)	11 (15.7)	35 (18.7)	
More than once a day	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	
Once a day	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0.8096
3 to 5 times a day	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	
Once a week	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	
Less than once a week	1 (1.9)	0 (0.0)	1 (1.4)	2 (1.1)	
Household responsibility - House and vard cleaning					
Not a responsibility	9 (17.3)	9 (13.9)	8 (11.4)	26 (13.9)	
More than once a day	0 (0.0)	1 (1.5)	0 (0.0)	1 (0.5)	
Once a day	0 (0.0)	1 (1.5)	0 (0.0)	1 (0.5)	0.2655
3 to 5 times a day	0 (0.0)	2 (3.1)	0 (0.0)	2 (1.1)	
Once a week	0 (0.0)	1 (1.5)	4 (5.7)	5 (2.7)	
Less than once a week	1 (1.9)	1 (1.5)	0 (0.0)	2 (1.1)	
Household responsobility - Using animal feces					
Not a responsibility	7 (13.5)	7 (10.8)	6 (8.6)	20 (10.7)	
More than once a day	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	
Once a day	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0.5150
3 to 5 times a day	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0.0100
Once a week	1 (1.9)	1 (1.5)	1 (1.4)	3 (1.6)	
Less than once a week	2 (3.9)	7 (10.8)	5 (7.1)	14 (7.5)	

TABLE 12. Trading and household responsibilities of human subjects that reported diarrhea by village

	Ambatolahy	Ambodiaviavy	Ankialo	OVERALL	
	N=52	N=65	N=70	N=187	
Predictor vs. reported diarrhea	n (%)	n (%)	n (%)	n (%)	p-value*
Defecate					
Latrine	4 (7.7)	0 (0.0)	8 (11.4)	12 (6.4)	0.0053
Open	6 (11.5)	15 (23.1)	4 (5.7)	25 (13.4)	
Bathe					
Bathroom	1 (1.9)	0 (0.0)	3 (4.3)	4 (2.1)	0.3329
Waterway	9 (17.3)	14 (21.5)	4 (5.7)	27 (14.4)	0.5581
Field	0 (0.0)	0 (0.0)	1 (1.4)	1 (0.5)	0.6957
Behind house	0 (0.0)	1 (1.5)	0 (0.0)	1 (0.5)	0.4698
Everywhere	0 (0.0)	0 (0.0)	2 (2.9)	2 (1.1)	1.0000
Don't bathe	0 (0.0)	0 (0.0)	2 (2.9)	2 (1.1)	0.1758

TABLE 13. Defecation an	nd bathing practice	s of human subject	s that reported	l diarrhea by village

TABLE 14. Water practices	of human subjects that	t reported diarrhea by village

	Ambatolahy	Ambodiaviavy	Ankialo	OVERALL	
	N=52	N=65	N=70	N=187	
Predictor vs. reported diarrhea	n (%)	n (%)	n (%)	n (%)	p-value*
Treat drinking water					
Yes	0 (0.0)	1 (1.6)	1 (1.5)	2(1.1)	0.0066
No	10 (19.2)	13 (20.3)	10 (14.5)	33 (17.8)	
Drinking water treatment technique					
Boil	0 (0.0)	1 (6.3)	1 (6.3)	2 (5.4)	
Filter through cloth/gauze	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	
Settle	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1.0000
Chlorine/Sur'eau	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	
Other	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	
Frequency of treating drinking water					
Sometimes (< once per week)	0 (0.0)	0 (0.0)	1 (5.9)	1 (2.4)	
Often (> once per week)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1.0000
Always (every time you drink)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	
Take water from storage container					
Bucket	0 (0.0)	0 90.0)	0 (0.0)	0 (0.0)	-
Cup	10 (19.2)	14 (21.5)	11 (15.7)	35 (18.7)	0.2572
Hands	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	-
Pour directly from container	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0.2087
Other	4 (7.7)	9 (13.9)	5 (7.1)	18 (9.6)	0.0202
Use stored water					
Drinking	10 (19.2)	15 (23.1)	11 (15.7)	36 (19.3)	0.1979
Bathing	1 (1.9)	0 (0.0)	6 (8.6)	7 (3.7)	0.5644
Cooking	6 (11.5)	13 (20.0)	9 (12.9)	28 (15.0)	0.5975
Washing fruit and vegetables	6 (11.5)	12 (18.5)	9 (12.9)	27 (14.4)	0.4178
Washing meat to cook	6 (11.5)	13 (20.0)	9 (12.9)	28 (15.0)	0.1367
Washing dishes and utensils	8 (15.4)	13 (20.0)	10 (14.3)	31 (16.6)	0.0578
Washing clothes	1 (1.9)	2 (3.1)	0 (0.0)	3 (1.6)	0.1406
Giving to animals	1 (1.9)	2 (3.1)	8 (11.4)	11 (5/9)	0.1379
Other	4 (7.7)	3 (4.6)	5 (7.1)	12 (6.4)	0.0329

	Ambatolahy	Ambodiaviavy	Ankialo	OVERALL	
	N=52	N=65	N=70	N=187	
Predictor vs. reported diarrhea	n (%)	n (%)	n (%)	n (%)	p-value*
Frequency of washing hands					
More than once a day	10 (19.2)	15 (23.1)	10 (14.3)	35 (18.7)	
Once a day	0 (0.0)	0 (0.0)	2 (2.9)	2 (1.1)	
3 to 5 times a week	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0.4059
Once a week	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	
Never	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	
Other	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	
Handwashing times					
Food preparation					
Before	3 (5.8)	3 (4.6)	4 (5.7)	10 (5.4)	0.9646
After	0 (0.0)	1 (1.5)	0 (0.0)	1 (0.5)	0.3574
Food collection					
Before	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1.0000
After	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	-
Bathroom					
Before	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1.0000
After	6 (11.5)	3 (4.6)	3 (4.3)	12 (6.4)	0.1976
Dealing with kids					
Before	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	-
After	0 (0.0)	0 (0.0)	2 (2.9)	2(1.1)	0.7403
Working with livestock					
Before	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	-
After	1 (1.9)	1 (1.5)	5 (7.1)	7 (3.7)	0.5215
Eating					
Before	8 (15.4)	12 (18.5)	8 (11.4)	28 (15.0)	0.0751
After	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0.5865
Breastfeeding					
Before	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1.0000
After	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	-
Using animal feces					
Before	1 (1.9)	0 (0.0)	0 (0.0)	1 (0.5)	0.1979
After	2 (3.9)	2 (3.1)	4 (5.7)	8 (4.3)	0.3888
Farming in the field					
Before	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1.0000
After	2 (3.9)	8 (12.3)	5 (7.1)	15 (8.0)	0.1864
Other	6 (11.5)	7 (10.8)	6 (8.6)	19 (10.2)	0.6631

TABLE 15. Handwashing practices of human subjects that reported diarrhea by village

	Ambatolahy	Ambodiaviavy	Ankialo	OVERALL	
	N=52	N=65	N=70	N=187	
Predictor vs. reported diarrhea	n (%)	n (%)	n (%)	n (%)	p-value*
Handwashing times					
Food preparation					
Before	7 (13.5)	12 (18.5)	8 (11.4)	27 (14.4)	0.9646
After	10 (19.2)	14 (21.5)	12 (17.1)	36 (19.3)	0.3574
Food collection					
Before	10 (19.2)	15 (23.1)	12 (17.1)	37 (19.8)	1.0000
After	10 (19.2)	15 (23.1)	12 (17.1)	37 (19.8)	-
Bathroom					
Before	10 (19.2)	15 (23.1)	12 (17.1)	37 (19.8)	1.0000
After	4 (7.7)	12 (18.5)	9 (12.9)	25 (13.4)	0.1976
Dealing with kids					
Before	10 (19.2)	15 (23.1)	12 (17.1)	37 (19.8)	-
After	10 (19.2)	15 (23.1)	10 (14.3)	35 (18.7)	0.7403
Working with livestock					
Before	10 (19.2)	15 (23.1)	12 (17.1)	37 (19.8)	-
After	9 (17.3)	14 (21.5)	7 (10.0)	30 (16.0)	0.5215
Eating					
Before	2 (3.9)	3 (4.6)	4 (5.7)	9 (4.8)	0.0751
After	10 (19.2)	15 (23.1)	12 (17.1)	37 (19.8)	0.5865
Breastfeeding					
Before	10 (19.2)	15 (23.1)	12 (17.1)	37 (19.8)	1.0000
After	10 (19.2)	15 (23.1)	12 (17.1)	37 (19.8)	-
Using animal feces					
Before	9 (17.3)	15 (23.1)	12 (17.1)	36 (19.3)	0.1979
After	8 (15.4)	13 (20.0)	8 (11.4)	29 (15.5)	0.3888
Farming in the field					
Before	10 (19.2)	15 (23.1)	12 (17.1)	37 (19.8)	1.0000
After	8 (15.4)	7 (10.8)	7 (10.0)	22 (11.8)	0.1864
Other	4 (7.7)	8 (12.3)	6 (8.6)	18 (9.6)	0.6631

TABLE 16. Human subjects that reported diarrhea and did not handwash at these times by village

	Ambatolahy	Ambodiaviavy	Ankialo	OVERALL	
	N=52	N=65	N=70	N=187	
Predictor vs. reported diarrhea	n (%)	n (%)	n (%)	n (%)	p-value*
Soap in the household					
Always	0 (0.0)	0 (0.0)	3 (4.4)	3 (1.6)	
Sometimes	10 (19.6)	15 (23.1)	9 (13.2)	34 (18.5)	0.2028
Never	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	
Frequency of washing hands with soap					
Always	0 (0.0)	0 (0.0)	2 (2.9)	2 (1.1)	
Sometimes	6 (12.0)	14 (21.5)	7 (10.0)	27 (14.6)	0.6953
Do not wash hands with soap	4 (8.0)	1 (1.5)	3 (4.3)	8 (4.3)	
Additional use of soap					
To wash for market	4 (7.7)	13 (20.0)	10 (14.3)	27 (14.4)	0.2400
Washing clothes	8 (15.4)	13 (20.0)	10 (14.3)	31 (16.6)	0.7312
Washing kitchen items	6 (11.5)	7 (10.8)	4 (5.7)	17 (9.1)	0.5597
Bathing bodies	10 (19.2)	14 (21.5)	9 (12.9)	33 (17.7)	0.2322
Washing hair	10 (19.2)	14 (21.5)	10 (14.3)	34 (18.2)	1.0000
Other	1 (1.9)	0 (0.0)	1 (1.4)	2 (1.1)	0.0484
Source of soap					
Market	6 (11.5)	11 (16.9)	10 (14.3)	27 (14.4)	0.5581
Other villager	0 (0.0)	7 (10.8)	3 (4.3)	10 (5.4)	0.4178
Store in the village	9 (17.3)	13 (20.0)	8 (11.4)	30 (16.0)	0.6217
Donation	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1.0000
Other	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1.0000
Would learn to make soap	8 (15.7)	12 (19.1)	11 (16.2)	31 (17.0)	0.4343

TABLE 17. Use of soap practices of human subjects that reported diarrhea by village

		Ambatolahy	Ambodiaviavy	Ankialo	OVERALL	
		N=52	N=65	N=70	N=187	
Predictor	vs. reported diarrhea	n (%)	n (%)	n (%)	n (%)	p-value*
Drink of	ther than water					
	Tea	5 (9.6)	11 (16.9)	4 (5.7)	20 (10.7)	0.3361
	Juice	9 (17.3)	14 (21.5)	12 (17.1)	35 (18.7)	1.0000
	Coffee	8 (15.4)	5 (7.7)	7 (10.0)	20 (10.7)	0.9953
	Beer	2 (3.9)	7 (10.8)	4 (5.7)	13 (7.0)	0.1047
	Toaka gasy	1 (1.9)	1 (1.5)	7 (10.0)	9 (4.8)	0.0241
	Rice water	9 (17.3)	15 (23.1)	12 (17.1)	36 (19.3)	1.0000
	Only water	9 (17.3)	14 (21.5)	11 (15.7)	34 (18.2)	0.0929
	Other	2 (3.9)	3 (4.6)	8 (11.4)	13 (7.0)	0.0081
Frequen	cy of eating rice during harvest time					
	Less than once a day	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	
	Once	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	
	Twice	0 (0.0)	1 (1.5)	0 (0.0)	1 (0.5)	0.6517
	Three times	9 (17.6)	14 (21.5)	12 (17.4)	35 (18.9)	
	More than three times	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	
Smoke						
	Never	8 (15.7)	15 (23.1)	11 (15.7)	34 (18.3)	
	Sometimes (≤once per week)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0.3071
	Often (3-5 times per week)	1 (2.0)	0 (0.0)	0 (0.0)	1 (0.5)	
	Daily	0 (0.0)	0 (0.0)	1 (1.4)	1 (0.5)	
Chewing	g tobacco					
	Never	8 (15.7)	10 (15.6)	10 (14.3)	28 (15.1)	
	Sometimes (≤once per week)	0 (0.0)	1 (1.6)	0 (0.0)	1 (0.5)	0.1557
	Often (3-5 times per week)	0 (0.0)	1 (1.6)	0 (0.0)	1 (0.5)	
	Daily	1 (2.0)	3 (4.7)	2 (2.9)	6 (3.2)	
Alcohol						
	Never	7 (13.7)	13 (20.3)	5 (7.1)	25 (13.5)	
	Sometimes (≤once per week)	1 (2.0)	2 (3.1)	6 (8.6)	9 (4.9)	0.1356
	Often (3-5 times per week)	1 (2.0)	0 (0.0)	1 (1.4)	2(1.1)	
	Daily	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	

TABLE 18. Dietary and personal habits of human subjects that reported diarrhea by village

TABLE 19. Transportation and foot hygiene	practices of human subjects	that reported diarrhea by village

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	Ambatolahy	Ambodiaviavy	Ankialo	OVERALL	
	N=52	N=65	N=70	N=187	
Predictor vs. reported diarrhea	n (%)	n (%)	n (%)	n (%)	p-value*
Primary mode of transportation					
Cart	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	-
Taxi-brousse	9 (17.3)	11 (16.9)	7 (10.0)	27 (14.4)	0.0256
Private car	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0.5848
Motorcycle	0 (0.0)	0 (0.0)	2 (2.9)	2 (1.1)	0.1758
Bike	2 (3.9)	0 (0.0)	0 (0.0)	2 (1.1)	1.0000
Foot	8 (15.4)	14 (21.5)	11 (15.7)	33 (17.7)	0.2322
Other	2 (3.9)	1 (1.5)	0 (0.0)	3 (1.6)	0.7234
Have shoes or sandals					
Yes	n.a.	5 (.7.7)	4 (5.7)	9 (6.7)	0.0250
No	n.a.	10 (15.4)	8 (11.4)	18 (13.3)	

n.a. = not available

	Ambatolahy	Ambodiaviavy	Ankialo	OVERALL	
	N=52	N=65	N=70	N=187	
Predictor vs. reported diarrhea	n (%)	n (%)	n (%)	n (%)	p-value*
Taken medication in past 4 weeks	7 (13.5)	13 (20.0)	9 (12.9)	29 (15.5)	0.0016
Symptoms improved from medication	6 (18.2)	13 (37.1)	9 (25.7)	28 (27.2)	1.0000
Animal taken medication in past 4 weeks	2 (4.1)	7 (10.8)	7 (10.0)	16 (8.7)	0.0007
Animal symptoms improved from medication					
Yes	1 (16.7)	7 (58.3)	6 (22.2)	14 (31.1)	0.2924
No response	1 (16.7)	0 (0.0)	1 (3.7)	2 (4.4)	

TABLE 20. Medication practices for human subjects and their animals of those who reported diarrhea by village

TABLE 21. Feces	handling prac	tices of humar	i subiects that	reported diarrh	ea by village

	Ambatolahy	Ambodiaviavy	Ankialo	OVERALL	
	N=52	N=65	N=70	N=187	
Predictor vs. reported diarrhea	n (%)	n (%)	n (%)	n (%)	p-value*
Handling human feces	7 (13.5)	7 (10.8)	6 (8.6)	20 (10.7)	0.3796
Handled animal feces	6 (11.5)	9 (14.1)	11 (15.7)	26 (14.0)	0.1422
Species					
Cat	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0.5865
Chicken	5 (9.6)	5 (7.7)	9 (12.9)	19 (10.2)	0.3564
Dog	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1.0000
Duck	1 (1.9)	3 (4.6)	3 (4.3)	7 (3.7)	0.3249
Geese	1 (1.9)	0 (0.0)	1 (1.4)	2 (1.1)	0.6585
Pig	2 (3.9)	3 (4.6)	8 (11.4)	13 (7.0)	0.2667
Rabbit	2 (3.9)	0 (0.0)	0 (0.0)	0 (0.0)	0.2572
Rodent	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1.0000
Turkey	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	-
Zebu	3 (5.8)	6 (9.2)	6 (8.6)	15 (8.2)	0.4559

TABLE 22. Univariate regression analysis of medication practices, tribe, water, sanitation, and hygiene practices, transportation, and profession\*

Predictors†	Likelihood Ratio χ²	p-value	Coefficient Estimate (β)	Standard Error	χ² p-value	Unadjusted Odds Ratio	(95% Confidence Interal)
Animal taken medication in past 4 weeks	10.39	0.0013	1.3107	0.4010	0.0011	3.71	(1.69 - 8.14)
Taken medication in past 4 weeks	10.75	0.0010	1.3144	0.4314	0.0023	3.72	(1.60 - 8.67)
Tribe							
Betsileo	8.15	0.0043	-1.3296	0.4511	0.0032	0.27	(0.11 - 0.64)
Tanala	8.15	0.0043	1.3296	0.4511	0.0032	3.78	(1.56 - 9.15)
Use latrine	7.88	0.0050	-1.0567	0.3882	0.0065	0.35	(0.16 - 0.74)
Open defecation	7.88	0.0050	1.0567	0.3882	0.0065	2.88	(1.34 - 6.16)
Treat drinking water	9.32	0.0023	-1.8255	0.7509	0.0151	0.16	(0.04 - 0.70)
Don't treat drinking water	9.32	0.0023	1.8255	0.7509	0.0151	6.21	(1.42 - 27.04)
Trade with other villages	5.89	0.0153	1.2838	0.5087	0.0116	3.61	(1.33 - 9.79)
Drink other than water - Other							
Drinking other	7.03	0.0080	-0.9909	0.3824	0.0096	0.37	(0.18 - 0.79)
Not drinking other	7.03	0.0080	0.9909	0.3824	0.0096	2.69	(1.27 - 5.70)
Trade with other villagers	5.92	0.0150	1.2127	0.4807	0.0116	3.36	(1.31 - 8.63)
Remove stored drinking water - Other	5.16	0.0232	0.8576	0.3752	0.0223	2.36	(1.13 - 4.92)
Drinking toaka gasy	5.36	0.0206	-0.9208	0.4169	0.0272	0.40	(0.18 - 0.90)
Not drinking toaka gasy	5.36	0.0206	0.9208	0.4169	0.0272	2.51	(1.11 - 5.69)
Have shoes or sandals‡	5.07	0.0243	-0.9916	0.4523	0.0283	0.37	(0.15 - 0.90)
Don't have shoes or sandals ‡	5.07	0.0243	0.9916	0.4523	0.0283	2.70	(1.11 - 6.54)
Transport by taxi-brousse	5.19	0.0228	0.8865	0.4047	0.0285	2.43	(1.10 - 5.36)
Homemaker	4.33	0.0374	0.8587	0.4031	0.0332	2.36	(1.07 - 5.20)
Use water in storage container - Other							
Use other	4.64	0.0312	-0.814	0.3874	0.0356	0.44	(0.21 - 0.95)
Not using other	4.64	0.0312	0.814	0.3874	0.0356	2.26	(1.06 - 4.82)
Use of soap - Other							
Use other	5.48	0.0192	-1.4759	0.7551	0.0506	0.23	(0.05 - 1.00)
Not using other	5.48	0.0192	1.4759	0.7551	0.0506	4.38	(0.10 - 19.22)

\*Dependent variable is reported diarrhea with and without blood

†Predictors of diarrhea that resulted in statistically significant p-value from Chi-square test or Fisher's exact test

‡Question only asked in Ambodiaviavy and Ankialo

TABLE 23. Multivariate regression analysis of demographics, medication practices, water, sanitation, and	
hygiene practices, location, and profession*	

	Coefficient	Standard		Adjusted Odd	8
Predictors †	Estimate (β)	Error	χ <sup>2</sup> p-value	Ratio	(95% Confidence Interal)
Intercept	-5.9165	1.2004	< 0.0001	-	-
Animal taken medication in past 4 weeks	1.7420	0.5329	0.0011	5.71	(2.01 - 16.22)
Age‡	-0.0109	0.0160	0.4975	0.99	(0.96 - 1.02)
Open defecation	0.9863	0.5542	0.0751	2.68	(0.91 - 7.94)
Don't treat drinking water	2.9511	0.9586	0.0021	19.13	(2.92 - 125.20)
Trade with other villages	1.7626	0.7324	0.0161	5.83	(1.39 - 24.49)
Female vs. Male	0.6969	0.5645	0.2171	2.01	(0.66 - 6.07)
Homemaker	0.9919	0.6662	0.1365	2.70	(0.73 - 9.95)
Ambatolahy vs. Ambodiaviavy	0.0433	0.6117	0.9435	0.77	(0.21 - 2.81)
Ambatolahy vs. Ankialo	-	-	-	1.04	(0.32 - 3.46)
Ambodiaviavy vs. Ankialo	0.3036	0.6319	0.6309	1.36	(0.39 - 4.68)

 

 Model
 Pr(Reported diarrhea) = -5.9165 + 1.7420\*Animal taken medication in past 4 weeks - 0.0109\*Age + 0.9863\*Open defecation + 2.9511\*Don't treat drinking water + 1.7626\*Trade with other villages + 0.6969\*Female

\*Dependent variable is reported diarrhea with and without blood

†Predictors chosen were statistically significant from previous models or chosen because of a priori knowledge

‡Age is a continuous variable

TABLE 24. Multivariate regression analysis of demographics, medication practices, water, sanitation, and hygiene practices, location, profession, and weekly income\*

	Coefficient	Standard		Adjusted	
Predictors†	Estimate (β)	Error	χ² p-value	Odds Ratio	(95% Confidence Interal
Intercept	-6.6911	1.4877	< 0.0001	-	-
Animal taken medication in past 4 weeks	1.9319	0.5796	0.0009	6.90	(2.22 - 21.50)
Age‡	-0.00133	0.00171	0.9382	1.00	(0.97 - 1.03)
Open defecation	0.9046	0.5646	0.1091	2.47	(0.82 - 7.47)
Don't treat drinking water	2.8829	0.9827	0.0033	17.87	(2.60 - 122.61)
Frade with other villages	2.0906	0.8615	0.0152	8.09	(1.50 - 43.78)
Female vs. Male	0.6588	0.5816	0.2573	1.93	(0.62 - 6.04)
Homemaker	0.9536	0.7270	0.1897	2.60	(0.62 - 10.79)
Ambatolahy vs. Ambodiaviavy	0.1923	0.6334	0.7614	0.85	(0.23 - 3.17)
Ambatolahy vs. Ankialo	-	-	-	1.21	(0.35 - 4.19)
Ambodiaviavy vs. Ankialo	0.3546	0.6518	0.5864	1.43	(0.40 - 5.11)
) AR (No Income) vs. 1 AR - 4,999 AR	0.8070	0.8459	0.3401	1.92	(0.22 - 16.47)
) AR (No Income) vs. 5,000 AR - 9,999 AR	-	-	-	3.25	(0.38 - 27.63)
) AR (No Income) vs. 10,000 AR - 19,999 AR	-	-	-	1.29	(0.17 - 9.62)
0 AR (No Income) vs. ≥ 20,000 AR	-	-	-	2.24	(0.43 - 11.76)
1 AR - 4,999 AR vs. 5,000 AR - 9,999 AR	0.1554	1.2106	0.8978	1.70	(0.12 - 24.79)
I AR - 4,999 AR vs. 10,000 AR - 19,999 AR	-	-	-	0.68	(0.05 - 9.16)
I AR - 4,999 AR vs. ≥ 20,000 AR	-	-	-	1.17	(0.11 - 12.53)
5,000 AR - 9,999 AR vs. 10,000 AR - 19,999 AR	-0.3725	1.1474	0.7454	0.40	(0.04 - 3.99)
5,000 AR - 9,999 AR vs. ≥ 20,000 AR	-	-	-	0.69	(0.07 - 6.53)
10,000 AR - 19,999 AR vs. ≥ 20,000 AR	0.5491	1.1535	0.6340	1.73	(0.18 - 16.61)

Model **Pr(Reported diarrhea)** = -6.6911 + 1.9319\*Animal taken medication in past 4 weeks - 0.00133\*Age + 0.9046\*Open defecation + 2.8829\*Don't treat drinking water + 2.0906\*Trade with other villages + 0.6588\*Female + 0.9536\*Homemaker + 0.1923\*Ambatolahy + 0.3546\*Ambodiaviavy + 0.8070\*O AR (No Income) +

\*Dependent variable is reported diarrhea with and without blood †Predictors chosen were statistically significant from previous models or chosen because of *a priori* knowledge ‡Age is a continuous variable

TABLE 25. First, second, and third biggest health problems of human subjects in Ambatolahy*, n (%)	

Number #1		Number #2	Number #3		
Respiratory Illness	13 (24.1)	Respiratory Illness	12 (22.2)	Respiratory Illness	12 (22.2)
Malaria	10 (18.5)	Headache	7 (13.0)	Diarrhea	10 (18.5)
Headache	8 (14.9)	Malaria	7 (13.0)	Malaria	10 (18.5)
I don't know	7 (13.0)	Eye disease	6 (11.1)	Joint and muscle pain	3 (5.6)
Diarrhea	6(11.1)	I don't know	3 (5.6)	Headache	2 (3.7)
Blindness	3 (5.6)	Vertigo	3 (5.6)	Mumps	2 (3.7)
Toothache	2 (3.7)	Mumps	2 (3.7)	Eye disease	1 (1.9)
Acute viral or bacterial infection	1 (1.9)	Sinusitus	2 (3.7)	High blood pressure	1 (1.9)
Epilespy	1 (1.9)	Toothache	2 (3.7)	I don't know	1 (1.9)
High blood pressure	1 (1.9)	Fever	1 (1.9)	Jaundice	1 (1.9)
Joint and muscle pain	1 (1.9)	High blood pressure	1 (1.9)	Stomachache	1 (1.9)
No problem	1 (1.9)	Influenza	1 (1.9)	Abdominal colic	0 (0.0)
Abdominal colic	0 (0.0)	Joint and muscle pain	1 (1.9)	Abdominal pain	0 (0.0)
Abdominal pain	0 (0.0)	Nosebleed	1 (1.9)	Acute viral or bacterial infection	0 (0.0)
Asthma	0 (0.0)	Sore throat	1 (1.9)	Asthma	0 (0.0)
Cardiovascular disease	0 (0.0)	Stomachache	1 (1.9)	Blindness	0 (0.0)
Ear infection	0 (0.0)	Abdominal colic	0 (0.0)	Cardiovascular disease	0 (0.0)
Eye disease	0 (0.0)	Abdominal pain	0 (0.0)	Ear infection	0 (0.0)
Fatigue	0 (0.0)	Acute viral or bacterial infection	0 (0.0)	Epilespy	0 (0.0)
Fever	0 (0.0)	Asthma	0 (0.0)	Fatigue	0 (0.0)
Goitre	0 (0.0)	Blindness	0 (0.0)	Fever	0 (0.0)
Hemerrhoids	0 (0.0)	Cardiovascular disease	0 (0.0)	Goitre	0 (0.0)
Impaired vision	0 (0.0)	Diarrhea	0 (0.0)	Hemerrhoids	0 (0.0)
Influenza	0 (0.0)	Ear infection	0 (0.0)	Impaired vision	0 (0.0)
Jaundice	0 (0.0)	Epilespy	0 (0.0)	Influenza	0 (0.0)
Mental illness	0 (0.0)	Fatigue	0 (0.0)	Mental illness	0 (0.0)
Migraine	0 (0.0)	Goitre	0 (0.0)	Migraine	0 (0.0)
Mumps	0 (0.0)	Hemerrhoids	0 (0.0)	Myocardial Infarction	0 (0.0)
Myocardial Infarction	0 (0.0)	Impaired vision	0 (0.0)	No problem	0 (0.0)
Nosebleed	0 (0.0)	Jaundice	0 (0.0)	Nosebleed	0 (0.0)
Polycythemia	0 (0.0)	Mental illness	0 (0.0)	Polycythemia	0 (0.0)
Sinusitus	0 (0.0)	Migraine	0 (0.0)	Sinusitus	0 (0.0)
Skin disease	0 (0.0)	Myocardial Infarction	0 (0.0)	Skin disease	0 (0.0)
Sore throat	0 (0.0)	No problem	0 (0.0)	Sore throat	0 (0.0)
Stomachache	0 (0.0)	Polycythemia	0 (0.0)	Toothache	0 (0.0)
Vertigo	0 (0.0)	Skin disease	0 (0.0)	Vertigo	0 (0.0)
No answer	0 (0.0)	No answer	3 (5.6)	No answer	9 (16.7)

\*Subjects were asked to report biggest health problem seperately, followed by second biggest health problem seperately, and then their third biggest health problem seperately. These were all separate, open-ended questions.

TABLE 26. First, second	l. and third biggest health	problems of human subje	ects in Ambodiaviavy*, n (%)

Number #1		Number #2	Number #3		
Respiratory Illness	18 (27.3)	Headache	13 (19.7)	Diarrhea	19 (28.8)
Diarrhea	7 (10.6)	Malaria	12 (18.2)	Malaria	11 (16.7)
Malaria	7 (10.6)	Asthma	8 (12.1)	Respiratory Illness	6 (9.1)
Asthma	6 (9.1)	Respiratory Illness	4 (6.1)	Headache	3 (4.6)
No problem	6 (9.1)	Toothache	3 (4.6)	Influenza	2 (3.0)
Stomachache	5 (7.6)	Fever	2 (3.0)	Acute viral or bacterial infection	1 (1.5)
I don't know	4 (6.1)	Joint and muscle pain	2 (3.0)	Asthma	1 (1.5)
Headache	3 (4.6)	Vertigo	2 (3.0)	Cardiovas cular disease	1 (1.5)
Influenza	2 (3.0)	Diarrhea	1 (1.5)	Fever	1 (1.5)
Joint and muscle pain	2 (3.0)	Ear infection	1 (1.5)	Joint and muscle pain	1 (1.5)
Toothache	2 (3.0)	High blood pressure	1 (1.5)	Sore throat	1 (1.5)
Acute viral or bacterial infection	1 (1.5)	Stomachache	1 (1.5)	Stomachache	1 (1.5)
Epilespy	1 (1.5)	Abdominal colic	0 (0.0)	Vertigo	1 (1.5)
Hemerrhoids	1 (1.5)	Abdominal pain	0 (0.0)	Abdominal colic	0 (0.0)
Myocardial Infarction	1 (1.5)	Acute viral or bacterial infection	0 (0.0)	Abdominal pain	0 (0.0)
Abdominal colic	0 (0.0)	Blindness	0 (0.0)	Blindness	0 (0.0)
Abdominal pain	0 (0.0)	Cardiovascular disease	0 (0.0)	Ear infection	0 (0.0)
Blindness	0 (0.0)	Epilespy	0 (0.0)	Epilespy	0 (0.0)
Cardiovas cular dise as e	0 (0.0)	Eye disease	0 (0.0)	Eye disease	0 (0.0)
Ear infection	0 (0.0)	Fatigue	0 (0.0)	Fatigue	0 (0.0)
Eye disease	0 (0.0)	Goitre	0 (0.0)	Goitre	0 (0.0)
Fatigue	0 (0.0)	Hemerrhoids	0 (0.0)	Hemerrhoids	0 (0.0)
Fever	0 (0.0)	I don't know	0 (0.0)	High blood pressure	0 (0.0)
Goitre	0 (0.0)	Impaired vision	0 (0.0)	I don't know	0 (0.0)
High blood pressure	0 (0.0)	Influenza	0 (0.0)	Impaired vision	0 (0.0)
Impaired vision	0 (0.0)	Jaundice	0 (0.0)	Jaundice	0 (0.0)
Jaundice	0 (0.0)	Mental illness	0 (0.0)	Mental illness	0 (0.0)
Mental illness	0 (0.0)	Migraine	0 (0.0)	Migraine	0 (0.0)
Migraine	0 (0.0)	Mumps	0 (0.0)	Mumps	0 (0.0)
Mumps	0 (0.0)	Myocardial Infarction	0 (0.0)	Myocardial Infarction	0 (0.0)
Nosebleed	0 (0.0)	No problem	0 (0.0)	No problem	0 (0.0)
Polycythemia	0 (0.0)	Nosebleed	0 (0.0)	Nosebleed	0 (0.0)
Sinusitus	0 (0.0)	Polycythemia	0 (0.0)	Polycythemia	0 (0.0)
Skin disease	0 (0.0)	Sinusitus	0 (0.0)	Sinusitus	0 (0.0)
Sore throat	0 (0.0)	Skin disease	0 (0.0)	Skin disease	0 (0.0)
Vertigo	0 (0.0)	Sore throat	0 (0.0)	Toothache	0 (0.0)
No answer	0 (0.0)	No answer	16 (24.2)	No answer	17 (25.8)

\*Subjects were asked to report biggest health problem seperately, followed by second biggest health problem seperately, and then their third biggest health problem seperately. These were all separate, open-ended questions.

TABLE 27. First, second, and third biggest health problems of human subjects in Ankialo\*, n (%)

Number #1		Number #2	Number #3		
Headache	16 (22.9)	Headache	18 (25.7)	Respiratory Illness	16 (22.9)
Toothache	16 (22.9)	Respiratory Illness	14 (20.0)	Diarrhea	14 (20.0)
Respiratory Illness	15 (21.4)	Diarrhea	6 (8.6)	Malaria	9 (12.9)
Abdominal pain	4 (5.7)	Malaria	6 (8.6)	Headache	6 (8.6)
Joint and muscle pain	3 (4.3)	Toothache	6 (8.6)	Fever	4 (5.7)
Asthma	2 (2.9)	Fever	5 (7.1)	Joint and muscle pain	4 (5.7)
Diarrhea	2 (2.9)	Joint and muscle pain	4 (5.7)	Toothache	3 (4.3)
Impaired vision	2 (2.9)	Fatigue	2 (2.9)	Acute viral or bacterial infection	1 (1.4)
Stomachache	2 (2.9)	Stomachache	2 (2.9)	Mental illness	1 (1.4)
Acute viral or bacterial infection	1 (1.4)	Acute viral or bacterial infection	1 (1.4)	Migraine	1 (1.4)
Fever	1 (1.4)	Skin disease	1 (1.4)	Stomachache	1 (1.4)
Goitre	1 (1.4)	Vertigo	1 (1.4)	Abdominal colic	1 (1.4)
I don't know	1 (1.4)	Abdominal colic	0 (0.0)	Abdominal pain	0 (0.0)
Malaria	1 (1.4)	Abdominal pain	0 (0.0)	Asthma	0 (0.0)
Polycythemia	1 (1.4)	Asthma	0 (0.0)	Blindness	0 (0.0)
Vertigo	1 (1.4)	Blindness	0 (0.0)	Cardiovas cular dise as e	0 (0.0)
Abdominal colic	0 (0.0)	Cardiovascular disease	0 (0.0)	Ear infection	0 (0.0)
Blindness	0 (0.0)	Ear infection	0 (0.0)	Epilespy	0 (0.0)
Cardiovascular disease	0 (0.0)	Epilespy	0 (0.0)	Eye disease	0 (0.0)
Ear infection	0 (0.0)	Eye disease	0 (0.0)	Fatigue	0 (0.0)
Epilespy	0 (0.0)	Goitre	0 (0.0)	Goitre	0 (0.0)
Eye disease	0 (0.0)	Hemerrhoids	0 (0.0)	Hemerrhoids	0 (0.0)
Fatigue	0 (0.0)	High blood pressure	0 (0.0)	High blood pressure	0 (0.0)
Hemerrhoids	0 (0.0)	I don't know	0 (0.0)	I don't know	0 (0.0)
High blood pressure	0 (0.0)	Impaired vision	0 (0.0)	Impaired vision	0 (0.0)
Influenza	0 (0.0)	Influenza	0 (0.0)	Influenza	0 (0.0)
laundice	0 (0.0)	Jaundice	0 (0.0)	Jaundice	0 (0.0)
Mental illness	0 (0.0)	Mental illness	0 (0.0)	Mumps	0 (0.0)
Migraine	0 (0.0)	Migraine	0 (0.0)	Myocardial Infarction	0 (0.0)
Mumps	0 (0.0)	Mumps	0 (0.0)	No problem	0 (0.0)
Myocardial Infarction	0 (0.0)	Myocardial Infarction	0 (0.0)	Nosebleed	0 (0.0)
No problem	0 (0.0)	No problem	0 (0.0)	Polycythemia	0 (0.0)
Nosebleed	0 (0.0)	Nosebleed	0 (0.0)	Sinusitus	0 (0.0)
Sinusitus	0 (0.0)	Polycythemia	0 (0.0)	Skin disease	0 (0.0)
Skin disease	0 (0.0)	Sinusitus	0 (0.0)	Sore throat	0 (0.0)
Sore throat	0 (0.0)	Sore throat	0 (0.0)	Vertigo	0 (0.0)
No answer	1 (1.4)	No answer	4 (5.7)	No answer	9 (12.9)

\*Subjects were asked to report biggest health problem seperately, followed by second biggest health problem seperately, and then their third biggest health problem seperately. These were all separate, open-ended questions.

#### TABLE 28. Handwashing and bathing data from structured observations

	Ambatol	ahy		Ambodi	aviavy	Ankialo			
	Dates: Ju	Dates: June 12, 18, 19, & 21		Dates: J	uly 9, 10, &	Dates	July 19, 20,	21, & 2	
	Total tim	e: 46.53 hou	rs	Total tin	<i>ie:</i> 19.12 hou	irs	Total time: 13.42 hours		
	n	(%)	n/hr	n	(%)	n/hr	n	(%)	n/hr
Total observations	402	(100.0)		251	(100.0)		9	(100.0)	0.7
Observer		. ,			. ,			. ,	
#1 - Male	140	(34.8)	3.0	89	(35.5)	4.7	8	(88.9)	0.6
#2 - Female	106	(26.4)	2.3	118	(47.0)	6.2	1	(11.1)	0.1
#3 - Male	63	(15.7)	1.4	44	(17.5)	2.3	0	(0.0)	0.0
#4 - Female	93	(23.1)	2.0	0	(0.0)	0	ů 0	(0.0)	0.0
Gender and Age	75	(23.1)	2.0	0	(0.0)	0	0	(0.0)	0.0
Female Child	65	(16.2)	1.4	48	(19.1)	2.5	5	(55.6)	0.4
Male Child	99		2.1	48 36	. ,	1.9	0	. ,	0.4
		(24.6)			(14.3)			(0.0)	
Female Adult	108	(26.9)	2.3	112	(44.6)	5.9	4	(44.4)	0.3
Male Adult	129	(32.1)	2.8	55	(21.9)	2.9	0	(0.0)	0.0
Body Part Washed*									
Feet	210	(52.2)	4.5	138	(55.0)	7.2	9	(100.0)	0.7
Legs	128	(31.8)	2.8	123	(49.0)	6.4	9	(100.0)	0.7
Buttocks	2	(0.005)	0	0	(0.0)	0	0	(0.0)	0.0
Core	0	(0.0)	0	3	(1.2)	0.2	0	(0.0)	0.0
Arms	94	(23.4)	2.0	139	(55.4)	7.3	4	(44.4)	0.3
Hands	200	(49.8)	4.3	177	(70.5)	9.3	8	(88.9)	0.6
Face	117	(29.1)	2.5	157	(62.6)	8.2	3	(33.3)	0.2
Mouth	6	(0.01)	0.1	3	(1.2)	0.2	0	(0.0)	0.0
Head	8	(2.0)	0.2	7	(2.8)	0.4	0	(0.0)	0.0
Hair	8 1		0.2	0	(0.0)	0.4	0		0.0
	1	(0.002)	0.0	0	(0.0)	0	0	(0.0)	0.0
Water Source †		(18.0)		10	(4.4.8)			(0.0)	
Pump #1	169	(42.0)	3.6	42	(16.7)	2.2	0	(0.0)	0.0
Pump #2	107	(26.6)	2.3	0	(0.0)	0	0	(0.0)	0.0
Pump #3	0	(0.0)	0	73	(29.1)	3.8	0	(0.0)	0.0
Pump #4	0	(0.0)	0	127	(50.1)	6.6	0	(0.0)	0.0
River	0	(0.0)	0	5	(2.0)	0.3	0	(0.0)	0.0
River next to village	1	(0.3)	0.0	0	(0.0)	0	0	(0.0)	0.0
River upstream from village	0	(0.0)	0	0	(0.0)	0	0	(0.0)	0.0
Stream	0	(0.0)	0	0	(0.0)	0	2	(22.2)	0.1
Spring	0	(0.0)	0	0	(0.0)	0 0	4	(44.4)	0.3
Spring above pump #1	0	(0.0)	0	0	(0.0)	0	0	(0.0)	0.0
				0		0			
Spring above pump #2	36	(9.0)	0.8		(0.0)		0	(0.0)	0.0
Spring below pump #1	0	(0.0)	0	1	(0.4)	0.1	0	(0.0)	0.0
Spring below pump #2	5	(1.2)	0.1	0	(0.0)	0	0	(0.0)	0.0
Spring below pump #3	0	(0.0)	0	1	(0.4)	0.1	0	(0.0)	0.0
Spring to next pump #1	65	(16.2)	1.4	0	(0.0)	0	0	(0.0)	0.0
Spring to next pump #2	8	(2.0)	0.2	0	(0.0)	0	0	(0.0)	0.0
Spring pool on hill	5	(1.2)	0.1	0	(0.0)	0	0	(0.0)	0.0
Rice paddy field water	0	(0.0)	0	0	(0.0)	0	3	(33.3)	0.2
House	1	(0.3)	0.0	2	(0.8)	0.1	0	(0.0)	0.0
Bucket or Cup									
Yes	10	(2.5)	0.2	55	(21.9)	2.9	0	(0.0)	0.0
No	391	(97.3)	8.4	196	(78.1)	10.3	9	(100.0)	0.7
Water bottle	1	(0.3)	0.0	0	(0.0)	0	0	(0.0)	0.0
Soap Used	1	(0.5)	0.0	0	(0.0)	0	0	(0.0)	0.0
•	0	(2.0)	0.2	16	(6.4)	0.0	0	(0,0)	0.0
Yes	8	(2.0)	0.2	16	(6.4)	0.8	0	(0.0)	0.0
No	391	(97.3)	8.4	234	(93.2)	12.2	9	(100.0)	0.7
Unknown	3	(0.8)	0.1	1	(0.4)	0.1	0	(0.0)	0.0
Brushed Teeth									
Yes	46	(11.4)	1.0	38	(15.4)	2.0	2	(22.2)	0.1
No	356	(88.6)	7.7	213	(84.9)	11.1	7	(77.8)	0.5
Brushed Teeth With									
Finger only	14	(3.5)	0.3	33	(13.2)	1.7	1	(11.1)	0.1
Tooth brush only	16	(4.1)	0.3	2	(0.8)	0.1	1	(11.1)	0.1
Tooth brush & toothpaste	10	(4.1)	0.3	3	(0.8)	0.1	0	(0.0)	0.0
	2								
Unknown	2	-(0.0)05	0.0	0	(0.0)	0	0	(0.0)	0.0
Footwear <sup>‡</sup>	-	10 C	6		(22 -			/	. ·
Barefoot	0	(0.0)	0	223	(88.8)	11.7	1	(11.1)	0.1
Shoes	0	(0.0)	0	25	(10.0)	1.3	0	(0.0)	0.0
Sandals	0	(0.0)	0	1	(0.4)	0.1	0	(0.0)	0.0
Unknown	402	(100.0)	8.6	2	(0.8)	0.1	8	(88.9)	0.6

\* Observers recorded all body parts washed for each individual observation †Ambatolahy only had pump #1 and pump #2, while Ambodiaviavy had pumps #1-4, and Ankialo did not have any pumps ‡Footwear was only recorded in Ambodiaviavy and Ankialo

	Ambatol	ahy		Ambodia	aviavy		Ankialo		
	Dates: J	une 12, 18, 19	9, 20, & 21	Dates : Ju	ıly 9,10, & 1	1	Dates: Ju	ıly 19, 20, 21	, & 22
	Total tim	e: 55.15 hour	s	Total time: 22.77 hours		s	Total time: 13.42 ho		urs
	n	(%)	n/hr	n	(%)	n/hr	n	(%)	n/h
Fotal observations	294	(100.0)	5.3	377	(100.0)	16.6	30	(100.0)	2.2
Observer									
#1 - Male	42	(14.3)	0.8	167	(44.3)	7.3	25	(83.3)	1.9
#2 - Female	127	(43.2)	2.3	142	(37.7)	6.2	5	(16.7)	0.4
#3 - Male	46	(15.7)	0.8	68	(18.0)	3.0	0	(0.0)	0.0
#4 - Female	79	(26.9)	1.4	0	(0.0)	0	0	(0.0)	0.0
Gender and Age									
Female Child	58	(19.7)	1.1	75	(19.9)	3.3	7	(23.3)	0.5
Male Child	61	(20.8)	1.1	35	(9.3)	1.5	2	(6.7)	0.1
Female Adult	116	(39.5)	2.1	248	(65.8)	10.9	17	(56.7)	1.3
Male Adult	58	(19.7)	1.1	19	(5.0)	0.8	4	(13.3)	0.3
Container									
Bucket	156	(53.1)	2.8	298	(79.1)	13.1	20	(66.7)	1.5
Jerry Can	4	(1.4)	0.1	26	(6.9)	1.1	0	(0.0)	0.0
Cup	15	(5.1)	0.3	16	(4.2)	0.7	0	(0.0)	0.0
Mouth	34	(11.6)	0.6	4	(1.1)	0.2	0	(0.0)	0.0
Other	87	(29.6)	1.6	37	(9.8)	1.6	10	(33.3)	0.7
Water Source†									
Pump #1	123	(41.8)	2.2	43	(11.4)	1.9	0	(0.0)	0.0
Pump #2	138	(46.9)	2.5	0	0.0	0	0	(0.0)	0.0
Pump #3	1	(0.3)	0.0	112	(29.7)	4.9	0	(0.0)	0.0
Pump #4	0	(0.0)	0	206	(54.6)	9.0	0	(0.0)	0.0
River	0	(0.0)	0	16	(4.2)	0.7	0	(0.0)	0.0
River next to village	1	(0.3)	0.0	0	(0.0)	0	0	(0.0)	0.0
River upstream from village	8	(2.7)	0.1	0	(0.0)	0	0	(0.0)	0.0
Stream	0	(0.0)	0	0	(0.0)	0	2	(6.7)	0.
Spring	0	(0.0)	0	0	(0.0)	0	19	(63.3)	1.4
Spring above pump #1	1	(0.3)	0.0	0	(0.0)	0	0	(0.0)	0.0
Spring above pump #2	13	(4.4)	0.2	0	(0.0)	0	0	(0.0)	0.0
Spring below pump #1	0	(0.0)	0	0	(0.0)	0	0	(0.0)	0.0
Spring below pump #2	0	(0.0)	0	0	(0.0)	0	0	(0.0)	0.0
Spring below pump #3	0	(0.0)	0	0	(0.0)	0	0	(0.0)	0.0
Spring to next pump #1	1	(0.3)	0.0	0	(0.0)	0	0	(0.0)	0.0
Spring to next pump #2	0	(0.0)	0	0	(0.0)	0	0	(0.0)	0.0
Spring pool on hill	5	(1.7)	0.1	0	(0.0)	0	0	(0.0)	0.0
Rice paddy field water	0	(0.0)	0	0	(0.0)	0	7	(23.3)	0.5
House	1	(0.3)	0.0	0	(0.0)	0	2	(6.7)	0.1
Cleaned before use		()			()			( ,	
Yes	112	(38.1)	2.0	233	(61.8)	10.2	15	(50.0)	1.1
No	89	(30.3)	1.6	107	(28.4)	4.7	5	(16.7)	0.4
Soap used		(2012)			()		-	()	
Yes	15	(5.1)	0.3	14	(3.7)	0.6	5	(16.7)	0.4
No	174	(59.2)	3.2	261	(69.2)	11.5	22	(73.3)	1.6
Additional objects cleaned		(***=)			(0,1_)			()	
Yes	81	(27.6)	1.5	26	(9.6)	1.1	8	(26.7)	0.6
No	213	(72.5)	3.9	341	(90.5)	15.0	22	(73.3)	1.6
Object cleaned	213	(, 2.0)	5.7	5-11	(30.0)	10.0		(10.0)	1.0
Clothing	17	(5.8)	0.3	19	(5.0)	0.8	6	(20.0)	0.4
Dishes/Utensils	33	(11.2)	0.6	8	(2.1)	0.8	0	(0.0)	0.0
Food	10	(3.4)	0.0	0	(0.0)	0.4	1	(3.3)	0.1
Other	10	(50.3)	2.7	349	(92.6)	15.3	23	(76.7)	1.
Footwear	140	(30.3)	2.1	577	()2.0)	13.5	23	(10.7)	1.
Barefoot	0	(0.0)	0	360	(95.5)	15.8	3	(10.0)	0.2
Shoes	0	(0.0)	0	16	(4.2)	0.7	0	(10.0)	0.0
Sandals	0	(0.0)	0	10	(0.3)	0.7	27	(0.0)	2.0
Unknown	0 294	(100.0)	5.3	0	(0.3) (0.0)	0.0	0	(90.0)	2.0

	Ambatola	•	Dates: June Total time:	vations = 6 e 18,19, 20, 8 39.58 hours	ž 24			
	Bovine	Canine	Chicken	Duck	Feline	Fish	Geese	Porcine
	N=18	N=23	N=196	N=179	N=40	N=0	N=130	N=51
	n(%)	n(%)	n(%)	n(%)	n(%)	n(%)	n(%)	n(%)
Observer								
#1 - Male	6 (0.9)	2 (0.3)	55 (8.6)	27 (4.2)	0 (0.0)	0 (0.0)	16 (2.5)	16 (2.5)
#2 - Female	11 (1.7)	14 (2.2)	89 (14.0)	108 (17.0)	0 (0.0)	0 (0.0)	36 (6.7)	35 (5.5)
#3 - Male	1 (0.2)	7 (1.1)	52 (8.2)	44 (6.9)	40 (6.3)	0 (0.0)	78 (12.2)	0 (0.0)
Human interaction								
Female Child	0 (0.0)	1 (0.2)	5 (0.8)	0 (0.0)	4 (0.6)	0 (0.0)	0 (0.0)	0 (0.0)
Male Child	1 (0.2)	6 (0.9)	5 (0.8)	13 (2.0)	18 (2.8)	0 (0.0)	1 (1.4)	19 (26.8
Female Adult	0 (0.0)	0 (0.0)	2 (0.3)	1 (0.2)	3 (0.5)	0 (0.0)	1 (0.2)	0 (0.0)
Male Adult	0 (0.0)	1 (0.2)	6 (2.8)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1 (0.2)
Unknown	0 (0.0)	0 (0.0)	1 (0.2)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Water Interaction								
Pump #1	0 (0.0)	0 (0.0)	1 (0.2)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Pump #2	0 (0.0)	0 (0.0)	2 (0.3)	0 (0.0)	0 (0.0)	0 (0.0)	3 (0.5)	0 (0.0)
Pump #3	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1 (0.2)	0 (0.0)
Pump #4	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
River	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	3 (0.5)	0 (0.0)
River next to village	0 (0.0)	0 (0.0)	1 (0.2)	2 (0.3)	0 (0.0)	0 (0.0)	3 (0.5)	0 (0.0)
River upstream from village	1 (0.2)	2 (0.3)	0 (0.0)	1 (0.2)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Stream	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Spring	5 (0.8)	0 (0.0)	0 (0.0)	5 (0.8)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Spring above pump #1	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Spring above pump #2	0 (0.0)	1 (0.2)	8 (1.3)	21 (3.3)	0 (0.0)	0 (0.0)	14 (2.2)	0 (0.0)
Spring above pump #3	0 (0.0)	0 (0.0)	10 (1.6)	6 (0.9)	0 (0.0)	0 (0.0)	0 (0.0)	14 (2.2
Spring above pump #4	0 (0.0)	1 (0.2)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Spring below pump #1	0 (0.0)	0 (0.0)	7(1.1)	6 (0.9)	0 (0.0)	0 (0.0)	7(1.1)	0 (0.0)
Spring below pump #2	0 (0.0)	4 (0.6)	33 (5.2)	38 (6.0)	0 (0.0)	0 (0.0)	56 (8.8)	0 (0.0)
Spring below pump #3	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Spring to next pump #1	0 (0.0)	0 (0.0)	2 (0.3)	0 (0.0)	0 (0.0)	0 (0.0)	4 (0.6)	0 (0.0)
Spring to next pump #2	0 (0.0)	0 (0.0)	4 (0.6)	24 (3.8)	0 (0.0)	0 (0.0)	2 (0.3)	0 (0.0)
Spring pool on hill	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Rice paddy field water	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Rice paddy spring above pool	5 (0.8)	0 (0.0)	3 (0.5)	32 (5.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Puddle	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
House	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Food Interaction	0 (0.0)	0 (0.0)	70 (11.0)	31 (4.9)	1 (0.2)	0 (0.0)	9 (1.4)	0 (0.0)
Animal Interaction	0 (0.0)	0 (0.0)	23 (3.6)	4 (0.6)	0 (0.0)	0 (0.0)	28 (4.4)	11 (1.7
Feces Interation	1 (0.2)	0 (0.0)	6 (0.9)	0 (0.0)	0 (0.0)	0 (0.0)	1 (0.2)	3 (0.5)
Enter Household	0 (0.0)	3 (0.8)	5 (0.8)	0 (0.0)	14 (2.2)	0 (0.0)	0 (0.0)	0 (0.0)
Enter Kitchen	0 (0.0)	0 (0.0)	8 (1.3)	0 (0.0)	0 (0.0)	0 (0.0)	2 (0.3)	0 (0.0)

TABLE 30. Livestock interaction data from structured observation data in Ambatolahy

		Ambodia	viavy	Total obser	vations =	= 1032			
				Dates: July	4, 5, 6, 8,	9, & 10			
				Total time:	82.28 hou	rs			
		Bovine	Canine	Chicken	Duck	Feline	Fish	Geese	Porcine
		N=27	N=42	N=916	N=17	N=0	N=0	N=0	N=30
		n(%)	n(%)	n(%)	n(%)	n(%)	n(%)	n(%)	n(%)
Observ	er								
	#1 - Male	3 (0.3)	17 (1.7)	249 (24.2)	7 (0.7)	0 (0.0)	0 (0.0)	0 (0.0)	5 (0.5)
	#2 - Female	21 (2.0)	16 (1.6)	262 (25.4)	1 (0.1)	0 (0.0)	0 (0.0)	0 (0.0)	21 (2.0
	#3 - Male	3 (0.3)	9 (0.9)	405 (39.2)	9 (0.9)	0 (0.0)	0 (0.0)	0 (0.0)	4 (0.4)
Human	interaction								
	Female Child	1 (0.1)	0 (0.0)	2 (0.2)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
	Male Child	2 (0.2)	1 (0.1)	51 (4.9)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	5 (0.5)
	Female Adult	0 (0.0)	0 (0.0)	14 (1.4)	6 (0.6)	0 (0.0)	0 (0.0)	0 (0.0)	2 (0.2)
	Male Adult	4 (0.4)	1 (0.1)	15 (1.5)	2 (0.2)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
	Unknown	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Water I	interaction								
	Pump #1	0 (0.0)	0 (0.0)	2 (0.2)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
	Pump #2	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
	Pump #3	0 (0.0)	0 (0.0)	7 (0.7)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1 (0.1)
	Pump #4	0 (0.0)	5 (0.5)	17 (1.7)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	2 (0.2)
	River	0 (0.0)	0 (0.0)	0 (0.0)	2 (0.2)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
	River next to village	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
	River upstream from village	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
	Stream	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
	Spring	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
	Spring above pump #1	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
	Spring above pump #2	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
	Spring above pump #3	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
	Spring above pump #4	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
	Spring below pump #1	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
	Spring below pump #2	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
	Spring below pump #3	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
	Spring to next pump #1	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
	Spring to next pump #2	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
	Spring pool on hill	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
	Rice paddy field water	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
	Rice paddy spring above pool	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
	Puddle	0 (0.0)	0 (0.0)	5 (0.5)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
	House	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Food Ir	teraction	11 (1.1)	9 (0.9)	576 (55.8)	4 (0.4)	0 (0.0)	0 (0.0)	0 (0.0)	2 (0.2)
	Interaction	3 (0.3)	19 (1.8)	77 (7.5)	2 (0.2)	0 (0.0)	0 (0.0)	0 (0.0)	12 (1.2
	nteration	2 (0.2)	2 (0.2)	36 (3.5)	1 (0.1)	0 (0.0)	0 (0.0)	0 (0.0)	4 (0.4)
	Iousehold	1 (0.1)	5 (0.5)	123 (11.9)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	3 (0.3)
	Kitchen	0 (0.0)	1 (0.1)	33 (3.2)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	2 (0.2)

TABLE 31. Livestock interaction data from structured observation data in Ambodiaviavy

	Ankialo		Total obse	rvations =	= 234			
			Dates : July	y 19, 20, 21	, & 22			
			Total time:	· 33.13 hou	ırs			
	Bovine	Canine	Chicken	Duck	Feline	Fish	Geese	Porcine
	N=11	N=4	N=176	N=33	N=1	N=2	N=0	N=7
	n(%)	n(%)	n(%)	n(%)	n(%)	n(%)	n(%)	n(%)
Observer								
#1 - Male	2 (0.9)	4 (1.7)	24 (10.3)	7 (3.0)	0 (0.0)	2 (0.9)	0 (0.0)	5 (2.1)
#2 - Female	9 (3.9)	0 (0.0)	114 (48.7)	8 (3.4)	0 (0.0)	0 (0.0)	0 (0.0)	1 (0.4)
#3 - Male	0 (0.0)	0 (0.0)	38 (16.2)	18 (7.7)	1 (0.4)	0 (0.0)	0 (0.0)	1 (0.4)
Human interaction								
Female Child	1 (0.4)	0 (0.0)	1 (0.4)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1 (0.4)
Male Child	8 (3.4)	3 (1.3)	3 (1.3)	1 (0.4)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Female Adult	0 (0.0)	0 (0.0)	3 (1.3)	7 (3.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Male Adult	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Unknown	0 (0.0)	0 (0.0)	1 (0.4)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Water Interaction								
Pump #1	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Pump #2	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Pump #3	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Pump #4	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
River	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
River next to village	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
River upstream from village	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Stream	0 (0.0)	0 (0.0)	0 (0.0)	4 (1.7)	0 (0.0)	2 (0.9)	0 (0.0)	0 (0.0)
Spring	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Spring above pump #1	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Spring above pump #2	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Spring above pump #3	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Spring above pump #4	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Spring below pump #1	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Spring below pump #2	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Spring below pump #3	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Spring to next pump #1	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Spring to next pump #2	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Spring pool on hill	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Rice paddy field water	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Rice paddy spring above pool	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Puddle	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
House	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Food Interaction	0 (0.0)	0 (0.0)	55 (23.5)	5 (2.4)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Animal Interaction	2 (0.9)	1 (0.4)	45 (19.2)	14 (6.0)	0 (0.0)	0 (0.0)	0 (0.0)	6 (2.6)
Feces Interation	0 (0.0)	0 (0.0)	12 (5.1)	2 (0.9)	0 (0.0)	0 (0.0)	0 (0.0)	2 (0.9)
Enter Household	0 (0.0)	0 (0.0)	57 (24.4)	0 (0.0)	1 (0.4)	0 (0.0)	0 (0.0)	0 (0.0)
Enter Kitchen	0 (0.0)	0 (0.0)	2 (0.9)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)

TABLE 32. Livestock interaction data from structured observation data in Ankialo