RISK ASSESSMENT OF NOROVIRUS INFECTION CONSIDERING INDIRECT IMPACTS OF SEASONAL FLOOD IN SOUTHEAST ASIA

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Abstract

Floods are not only the dangerous disaster situation threatening human lives, but they also can pose additional risks for human health. Less attention has been paid to indirect health impacts of floods such as health problems than direct impacts (e.g. loss of life, destructions of property and agricultural crops). This is due to difficultly to prove the damage to human health via indirect routes such as contaminated water and food, although floods are likely to impact on human health physically and psychologically. In terms of physical health impacts, floods are potentially able to cause gastroenteritis diseases outbreaks. While norovirus (NoVs) is well known as a leading causal pathogen for acute gastroenteritis in developed countries, structured data on the prevalence of the disease in developing countries are not available thus far. Oysters are often associated with NoV infection due to its feature to accumulate the virus from surrounding water environment. If oysters are contaminated by NoVs transported from urban area by floodwater, urban flood potentially increases the risk of foodborne diseases by consuming oysters. On the other hand, based on the above feature, NoV contamination in oysters was reported to be well correlated to number of cases of gastroenteritis in some developed countries. In terms of psychological impact, in some areas of developing countries hit by flooding frequently, people seem to adapt their lives to the floodwater so well. As an indicator of the psychological impact, people's feeling to a disease is important in the estimation of its burden but this relationship has not been investigated especially for common diseases with a low fatality.

Hue City, located in central of Vietnam, is known as a flood city. About two-thirds of the city's residents are likely exposed to flooding when a heavy rainy occurs. Since the city has no wastewater treatment plant, domestic wastewater is discharged directly into drainage channels, although a part of wastewater passes through septic tanks. The channels flow directly into the Perfume River. Oysters cultivated in the downstream of the river are probably affected by contaminants from the city where urban flooding occurs regularly. This area has potential to show indirect impacts of flood as above described and so it was selected for the field of this study. The present study aims (1) to estimate the prevalence of NoVs in cases of acute gastroenteritis in

developing countries; (2) to reveal the epidemic of gastroenteritis in Hue city by monitoring NoV contamination in oysters collected in a downstream lagoon; (3) to assess the risk of NoV infection due to the contaminated oysters; (4) to test the hypothesis that frequent exposures to flood can change the feeling about gastroenteritis; and (5) to propose a new method to estimate burden of gastroenteritis considering the feeling about the disease.

To estimate the prevalence of NoVs in developing countries, relevant studies were identified by searching PubMed and Web of Science for the period from January 1, 1990 through March 31, 2016. Studies performed in developing countries with a study period of at least 12 months and which provided information on PCR-confirmed NoVs prevalence in patients diagnosed with acute gastroenteritis were included. A meta-analysis was conducted on NoVs prevalence, focusing on viral genogroups GI and GII, in cases of acute gastroenteritis. Using evidence from 178 articles, the estimated NoVs prevalence among 148,867 patients with acute gastroenteritis was 17% (95% CI: 15-18%). The prevalence decreased from 18% (95% CI: 16%-20%) for upper middle-income countries to 15% (13%-18%) and 6% (3%-10%) for lower middle- and low-income countries, respectively. There were no significant differences in NoVs prevalence by age group (under 5 years, 5 years and over, and mixed ages) or severity of symptoms as defined by community, outpatient, or inpatient setting. The pooled prevalence of NoV GII (15%, 95% CI: 13-17%) was significantly higher than that of NoV GI (1%, 95% CI: 1-1%) in patients with acute gastroenteritis.

As a potential indicator for the epidemic of gastroenteritis in Hue city, the level of NoV contamination in oysters collected in the lagoon receiving urban drainage from Hue City for 17 months (August 2015 to December 2016) was investigated. A total of 34 oyster samples were collected at two sampling sites in the lagoon. NoV GI was more frequently detected than GII (positive rate 79% vs. 41%). Maximum concentrations of GI and GII were 2.4×10^5 and 2.3×10^4 copies/g, respectively. Co-contamination with GI and GII was observed in 35% of samples. NoV GII concentration was higher at station A in the flood season than in the dry season (P= 0.04, Wilcoxon signed-rank test). Six genotypes (GI.2, GI.3, GI.5, GII.2, GII.3, and GII.4) were identified in both wastewater and oyster samples, and genetically similar or identical sequences were obtained from the two types of samples. These

observations suggest that urban drainage and seasonal flooding contribute to NoVs contamination of oysters in the downstream. On the other hand, due to this impact of flood, monitoring of NoVs concentration in oysters seemed ineffective to reveal the epidemic of gastroenteritis in the city. Estimated risk of NoV infection due to consuming oysters harvested in the lagoon was quite high (36.3 to 72.2%).

As another indirect impact of flood, its psychological impact on human health was investigated by a cross-sectional study involving 293 people living in flooding areas and 365 people in non-flooding areas in T. T. Hue province, Vietnam, and three provinces in Cambodia. A questionnaire was developed for this investigation. As the result, the participants felt that diarrhea, severe diarrhea, cough, fever, skin problems, and eye problems happened more frequently during/after flooding. The feeling about all those diseases of the people living in the flooding areas, except eye problems, was significantly different from that of the people living in non-flooding areas in Cambodia ($P < 0.05$). The same results were observed in Vietnam for diarrhea and cough. In Vietnam, factors associated with the feeling about diseases that happened more frequently during and after flood, were age and education. According to this finding, a new method was proposed to incorporate psychological impact of floods into health burden of gastroenteritis, which was estimated by Disability Adjusted Life Years (DALYs), by modifying the disability weight.

Overall, the findings from the study demonstrated that seasonal floods were able to adversely cause indirect impacts on human health not only physically but also psychologically. The prevalence of NoVs in patients with acute gastroenteritis in developing countries was 17%. The urban drainage surely contributed to the oyster contamination with NoVs in the study area however there was no significant correlation between NoV concentration in oysters and the number of diarrhea cases recorded in the largest hospital in the city during the study period. This is due to poor surveillance system in Vietnam as well as virus transportation enhanced by flood. A considerable risk of NoV infection due to consuming oysters contaminated with NoVs induced by flood-water mixed with domestic wastewater from Hue City was noticeable. Also, our findings suggest that, among those living in the flooding areas, floods can change their feelings to gastroenteritis. More supports to those who are

frequently affected by floods with considering of the change of their feelings could contribute to reduction of adverse impacts of floods.

【和訳】

洪水は、人命を脅かす危険な災害であるだけでなく、それに接触する人々に疾病のリスクももたらす。洪水の直接的な影響（例：人命の損失、財産や農産物の喪失）に比べて、健康問題のような洪水の間接的な健康影響には、これまで注目がされてこなかった。これは、洪水が人の健康に身体的および心理的な影響を与えそうであるにも関わらず、汚染された水や食品のような間接的な経路での人の健康への損害を立証することが難しいためである。身体的な健康影響という点では、洪水は潜在的に胃腸系疾患の流行を引き起こすことがある。ノロウイルス（NoV）は、先進国での急性胃腸炎の主たる原因病原体として良く知られているが、途上国でのこの疾病の罹患率に関する構造化されたデータは、現在のところ利用可能でない。牡蠣は、ウイルスをため込みやすい性質により、NoV 感染にしばしば関連づけられる。牡蠣が洪水により都市域から運ばれてきた NoV に汚染されるとすれば、都市洪水は牡蠣の喫食による食中毒のリスクを増加させる。一方で、幾つかの先進国では、牡蠣のこの性質にもとづいて、牡蠣の NoV 汚染が胃腸炎の患者数とよく相関することが報告されている。心理的な影響に関しては、洪水に頻繁に見舞われる地域では、人々は洪水にうまく生活を適応させているように見える。疾病負荷を推定するために、疾病に対する感覚は重要であるものの、特に胃腸炎のように死亡率が低く、ありふれた疾病に対してこの関係について研究が行われてこなかった。

ベトナム中部に位置するフエ市は、洪水に見舞われる都市として有名である。豪雨が起こると、都市住民の約３分の２が洪水に曝される。フエ市には下水処理場がないため、都市下水は直接（一部は浄化槽を通過して）、排水路に放流される。その排水路はパフューム川につながっている。その川の下流に生息する牡蠣は、洪水が定期的に起こる都市からの汚染物質による影響を受けているだろう。上記の背景のもとで、この都市を本研究のフィールド

に選定した。この研究の目的は、(1)途上国における急性胃腸炎の患者の中で NoV 罹患率を推定すること、(2)下流のラグーンで牡蠣の NoV 汚染をモニタリングすることで、この地域の胃腸炎流行を明らかにすること、(3)汚染された牡蠣を食べることによる NoV 感染のリスクを評価すること、(4)洪水に頻繁に見舞われることで胃腸炎に対する感覚が変わるという仮説を検証すること、そして、(5)胃腸炎に対する感覚を考慮した胃腸炎による疾病負荷の推定手法を提案することである。

途上国における NoV 罹患率を推定するために、1990 年 1 月から 2016 年 3 月までに発表された関連研究を、PubMed と Web of Science から同定した。少なくとも 12 ヶ月に渡って途上国で行われた研究で、急性胃腸炎と診断された患者の中での PCR で確認された NoV 罹患率に関する情報を提供している研究が選ばれた。急性胃腸炎の患者の中で、遺伝子群 GI と GII に着目しながら、NoV 罹患率に関するメタ解析が行われた。178 の論文からのエビデンスを用いて、148,867 人の患者の間での NoV 罹患率は 17%（95%CI: 15-18%）と推定された。罹患率は、中程度で上位の所得の国の 18%（16-20%）から、中所得で下位の 15%（13-18%）、低所得の 6%（3-10%）に低下した。年齢層や症状の重さによる NoV 罹患率の違いはなかった。急性胃腸炎患者の間でプールされた NoV GII の罹患率（15%、95% CI: 13-17%）は、NoV GI のそれ（1%、95% CI: 1-1%）よりも有意に高かった。

幾つかの先進国ではその有効性が報告されていることから、フエ市における胃腸炎流行の潜在的な指標として、同市からの下水を受容するラグーンで採取された牡蠣の NoV 汚染レベルを 17 ヶ月間（2015 年 8 月～2016 年 12 月）に渡って調査した。ラグーンの 2 地点で 1 回に 34 個の牡蠣試料を採取した。NoV GI（検出率 79%）は NoV GII（41%）よりも高い頻度で検出された。GI および GII の最大濃度は、それぞれ 2.4×10^5 および 2.3×10^4 copies/g であった。35%の試料では、両方の遺伝子群が検出された。河口に近い地点での NoV GII の濃度は雨季に乾季よりも高かった（p=0.04）。牡蠣試料と都市下水試料の両方から 6 つの遺伝子型（GI.2、GI.3、GI.5、GII.2、GII.3、GII.4）が

同定され、２つの試料からは類似した、あるいは同一の遺伝子配列が得られた。この結果は、都市下水と洪水が、下流での牡蠣の NoV 汚染に貢献していることを示している。一方で、この洪水による影響のために、牡蠣の NoV 濃度のモニタリングは、この地域の胃腸炎流行を明らかにするには効果的ではないようであった。なお、このラグーンで採取された牡蠣を食べることによる NoV 感染のリスク推定値は非常に高かった（１ヶ月のリスクとして 0.36～0.72）。

もう１つの洪水の間接影響として心理面への影響を、ベトナムのフエ省とカンボジアの３つの県において、洪水域の住民 293 人と非洪水域の住民 365 人を巻き込んだクロスセクショナル研究によって調査した。この調査のために、質問票を作成した。参加者は、洪水の間あるいは洪水後に、下痢症、ひどい下痢症、風邪、熱、皮膚病、眼病がより多く発生すると感じていた。これらの疾病に対する感覚は、眼病を除いて、カンボジアでは洪水域の住民と非洪水域の住民の間で有意に異なっていた。ベトナムでの調査でも、下痢症と風邪に関して同じ結果が得られた。これらの結果は、洪水の経験が、疾病に対する感覚を変化させることがある事実を示している。なお、ベトナムにおいては年齢と教育が、洪水によって疾病に対する感覚を変えるときの影響因子として見いだされた。この知見にしたがって、胃腸炎による疾病負荷（障害調整損失年（DALYs）で推定される）に洪水による心理的な影響を組み込む新しい方法を提案した。

全体的に見て、本研究からの知見は、季節的な洪水が人間の身体的な健康だけでなく心理的にも間接影響を与えうることを示した。途上国の急性胃腸炎の患者の中での NoV の罹患率は 17%であり、先進国と同等のレベルにあった。フエ市では、都市排水が確かに牡蠣の NoV 汚染に貢献していたが、牡蠣の NoV 濃度と下痢症患者数に相関はなかった。これは、ベトナムにおける貧弱なサーベイランスシステムと、洪水によって促進されるウイルスの輸送によるものである。フエ市からの下水が混じった洪水が運んだであろう NoV に汚染された牡蠣を食べることによる高い感染リスクは、特筆すべきものであ

った。また、我々の知見は、洪水が起こる地域の住民の間で、洪水が胃腸炎に対する感覚を変化させうることを示した。洪水に頻繁に見舞われる人々に対するこの感覚の変化に配慮した支援は、洪水の健康影響の低下に貢献するだろう。

CONTENTS

CHAPTER Page

LIST OF TABLES

LIST OF FIGURES

CHAPTER 1

INTRODUCTION

1.1 Flood and its impacts

Flood is one of the most common natural disasters to affect human life around the world. The International Disaster Database defines flood as "A condition that occurs when water overflows the natural or artificial confines of a stream or other body of water or accumulates by drainage over low-lying areas" (EM-DAT 2017).

Flood impacts can be divided into direct and indirect damages (Messner and Meyer 2006). Direct damage is caused by floodwaters that directly affect humans and the environment through loss of life, trauma, destruction of property, destruction of agricultural crops, and changes to ecological systems. In the two consecutive extreme flooding events of 2004 and 2007, direct damages were accounted for a total output loss of 249,611 million and 148,408 million in Bangladeshi Taka (BDT), respectively. In most cases, output losses from direct flood damages occur regionally in agricultural, industrial, construction, and housing services (Haque and Jahan 2015). Feyen et al. (2012) calculated the expected annual damage (EAD) from river flooding events in Europe to be EUR 6.4 billions; in the future, this EAD may increase to EUR 14–21.5 billions (based on 2006 values) depending on climate scenarios. The direct economic losses from the major flood events in Europe between 2003 and 2009 were approximately EUR 17 billions (European Environmental Agency 2010).

Indirect damages happen after the initial flooding events. For example, a flood may contaminate environmental water. If this water is then used for irrigation, the harvested food may also be contaminated. The risk of diarrhea and other diseases increases with consumption of this food. In Italy, for example, indirect damage from a flood event

accounted for a significant percentage of total losses. Indirect damage was estimated to be in the range of EUR 3.3 to 8.8 billion (based on 2000 values), which was approximately one-fifth of the estimated cost of direct damage (Carrera et al. 2015). The detail of indirect impacts of flood will be discussed in chapter 2.

1.2 Why focus on impacts of flood in terms of infectious diseases?

Outbreaks of infectious diseases are as one of the major consequences of flooding events to human. Infectious diseases, such as malaria and diarrhea, were increased after a flood event in Mozambique (Kondo et al. 2002). In Bangladesh, respondents to a study by Kunii et al. (2002) reported health problems that included fever (43%), diarrhea (27%), and respiratory infections (14%); children under 5 years of age were found to be more susceptible to diarrhea after a flood event than the older age groups. An increase in diseases of eyes, skin, and gastrointestinal tract was reported after a flood in Taiwan (Huang et al 2016). Outbreaks of leptospirosis were reported in Rio de Janeiro (Barcellos and Sabroza 2001) and in the Philippines (Easton 1999) after flood events.

1.3 What kind of previous studies in flood-related diseases?

Flood negatively affects human's physical health and psychology. Direct damage of flood affects humans and the environment. For example, in Bangladesh, 15,000 people are killed each year by floods (Malilay 1997). Flooding water may contaminate the local water and food supplies and damage the sewage system, resulting in contamination and increase the potential for communicable diseases (Du et al. 2010). If urban flooding occurs in areas with combined sewer systems, flood water may pose health risks to citizens exposed to pathogens in these waters (Veldhuis et al. 2010; De Man et al. 2014). Increasing waterborne and

foodborne diarrheal diseases have been reported in India, Brazil, Bangladesh, Mozambique, USA, and Taiwan following flooding episodes (Cairncross & Alvarinho, 2006; Huang et al. 2016). Mental health problems such as anxiety, stress, psychological distress, probable anxiety, probably depression, probable post-traumatic stress disorder, were reported following flood events (Carroll et al. 2010; Paranjothy et al. 2011; Hetherington et al. 2017).

1.4 What are the knowledge gaps related to floods in terms of infectious diseases?

As above mentioned, less attention has been paid to indirect health impacts of floods than direct impacts. This is due to difficultly to prove the damage to human health via indirect routes such as contaminated water and food, although floods are likely to impact on physical health and psychology.

In terms of physical health impacts, floods are potentially causing gastroenteritis outbreaks. Acute gastroenteritis contributes significantly to the burden of disease worldwide. A previous study (Liu et al. 2012) indicated that 801,000 worldwide mortalities in children under 5 years of age are associated with diarrheal disease. The incidence and mortality rates of diarrhea are much higher in low-income than in middle- and high-income countries (Walker et al. 2013). While norovirus (NoVs) is well known as a leading causal pathogen for acute gastroenteritis in developed countries, structured data on prevalence in developing countries are not available thus far. The detail is going to be discussed in chapter 3.

Regarding gastroenteritis outbreaks, oysters are one of the important vehicles for transferring foodborne pathogens since they have a high capacity to accumulate enteric viruses and bacteria (Metcalf et al. 1979; Rippey 1994; Le Guyader et al. 2012; Wang, et al. 2014; Pu et al. 2016). In fact, recently, there have been increasing outbreaks of human NoVs associated with consumption of oysters (Fitzgerald et al. 2014; Lodo et al. 2014; Loury et al.

2015; Wang et al. 2015). If oysters are contaminated by NoVs transported from urban area by floodwater, urban flood potentially increases the risk of foodborne diseases by consuming such contaminated oysters. On the other hand, based on this feature of oysters to accumulate enteric viruses, NoV contamination in oysters was reported to be well correlated to number of cases of gastroenteritis in some developed countries (Ueki et al. 2005; Pu et al. 2016). The detail will be discussed in chapter 4.

On the other hand, in terms of psychological impacts by floods, in some areas of developing countries hit by flooding frequently, people seem to adapt their lives to the floodwater so well. As an indicator of the psychological impacts, people's feeling to disease is important in the estimation of its burden, especially for common diseases with a low fatality. The detail will be discussed in chapter 5.

1.5 Study area

Hue city, located in central of Vietnam, is known as a flood city. The annual rainy season is from August to January, with a flood season from October onwards. About two-thirds of the city's residents are likely exposed to flooding when a heavy rain occurs (NCAP, 2005). Similar to most cities in developing countries, the city has no wastewater treatment plant. Domestic wastewater is discharged into drainage channels after passage of some of the wastewater through septic tanks. The channels flow directly into the Perfume River. Oysters cultivated in the downstream of the river are probably affected by contaminants from the city, where urban flooding occurs regularly. This area has potential to show indirect impacts of flood as above described and so it was selected for the field of this study.

1.6 Objectives of the study

The present study aims (1) to estimate the prevalence of NoVs in cases of acute gastroenteritis in developing countries; (2) to reveal the epidemic of gastroenteritis in Hue city by monitoring NoV contamination in oysters collected in a downstream lagoon; (3) to assess the risk of NoV infection due to the contaminated oysters; (4) to test the hypothesis that frequent exposures to flood can change the feeling about gastroenteritis; (5) to propose a new method to estimate burden of gastroenteritis considering the feeling about the disease.

1.7 Outline of the study

This study involves six chapters (Figure 1.1). Besides the introduction of this study that has been mentioned, the second chapter will presented about literature review on indirect impacts of flooding on human health. In chapter 3, the prevalence of NoVs in the case of gastroenteritis in developing countries will be investigated by means of systematic review and meta-analysis. In chapter 4, the application of monitoring NoVs contaminated with oysters to identify the epidemic of gastroenteritis in Hue City, Vietnam will be discussed, and the risk of NoV infection due to consuming the contaminated oysters will be also assessed. Chapter 5 will introduce the impacts of flood on psychology. In this chapter, the feeling about infectious diseases of people living in flood areas and non flood areas will be quantified, and the factors associated with the feeling will also be verified. In addition, a new method will be proposed to incorporate psychological impact of floods into health burden of gastroenteritis using Disability Adjusted Life Years (DALYs), by modifying the disability weight. Finally, the conclusions and recommendations will be presented based on all results of this study in chapter 6.

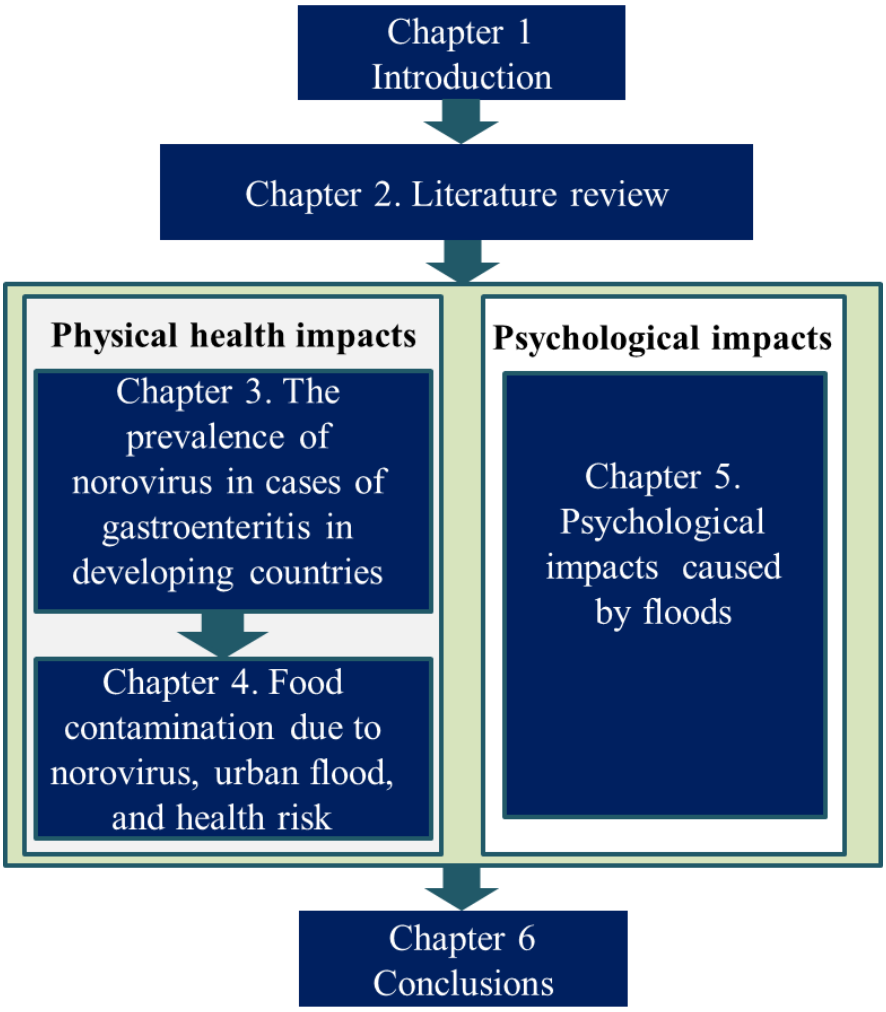

Figure 1.1 Outline of the study

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CHAPTER 2

LITERATURE REVIEW

Contents of this chapter are as follows.

(1) Indirect impacts of floods in terms of physical health

(2) Evaluation risk of the food-borne gastroenteritis related to flood

(3) Indirect impacts of floods in terms of psychology

2.1 Indirect impacts of floods in terms of physical health

2.1.1 Introduction

The concept of flood and its related to infectious diseases were mentioned in the previous study. In this chapter, the literature on the impact of flooding on human health, with a focus on indirect damage from floodwaters are pointed out.

2.1.2 Scope of impacts of flood in the world

With global climate change, floods have become one of the most common natural disasters to affect human life worldwide. Floods can have a wide range of effects on humans, including economical and ecological impacts; they can also damage homes, public buildings, and infrastructure. From 2005 to 2016, more than 1900 flood events were reported, caused an estimated 64,000 deaths, more than 888 million individuals affected globally and more than USD 3 billion in damages (EM-DAT 2016) (Table 1). Floods also are associated with an increased risk of water- and vector-borne diseases (World Health Organization, Flooding and Communicable Diseases Fact Sheet).

Table 2.1 Flood impacts worldwide from 2005 to 2016 (adapted from EM-DAT, 2016)

Continent	Flood events (*n*)	Deaths (*n*)	Individuals affected (*n*)	Damage (USD)
Africa	467	8,767	32,789,561	4,065,233
Americas	380	6,775	41,420,998	54,280,165
Asia	791	47,337	809,539,983	215,121,084
Europe	239	1,087	4,076,482	56,952,740
Oceania	50	186	727,030	12,536,747
Total	1,927	64,152	888,554,054	342,955,969

2.1.3 Direct impact of floods on human health

A number of studies have examined the effects of floodwaters on human health (Du et al. 2010; Ten Veldhuis et al. 2010; Bharti et al. 2003; Cairncross and Alvarinho 2006; Abaya et al. 2009; Campanella 1999; Schwartz et al. 2006; Kondo et al. 2002; Kunii et al. 2002; Barcellos and Sabroza 2001; Easton 1999; Ceuppens et al. 2014; Phanuwan et al. 2006; De Man et al. 2014; Kazama et al. 2011; Cann et al. 2013; Aggarwal and Krawczynski 2000; McCarthy et al. 1994; Corwin et al. 1999; Hau et al. 1999; Vachiramon et al. 2008; Schnitzer et al. 2007). Floodwaters may contaminate local water sources and food supplies and damage sewage systems, which increases the potential for communicable diseases (Du et al. 2010). Many pathogens can survive in domestic wastewater (Godfree and Godfrey 2008). Thus, if urban flooding occurs in areas with combined sewer systems, floodwaters may pose health risks for citizens who are exposed to pathogens in these waters (Ten Veldhuis et al. 2010). For example, pathogenic *Leptospira* sp. (the causative agent of

leptospirosis) can be transmitted to humans and animals by direct contact with urine from infected rodents or through contaminated floodwater (Bharti et al. 2003).

Diarrheal diseases alone (many of which are foodborne illnesses) kill 2.2 million people globally every year (World Health Organization 2014). Increases in waterborne and foodborne diarrheal diseases have been reported in India, Brazil, Bangladesh, Mozambique, and the United States following flood events (Cairncross and Alvarinho 2006). Diarrheal diseases were also reported after a flood in the Gambella region of Ethiopia (Abaya et al. 2009). Campanella (1999) reported an increase in acute diarrhea and acute respiratory disease in Nicaragua after Hurricane Mitch and its associated flooding. In Bangladesh after the 1988, 1998, and 2004 floods, species associated with flood-related diarrheal epidemics included *Vibrio cholera*, rotavirus, enterotoxigenic *Escherichia coli*, *Shigella*, and *Salmonella* (Schwartz et al. 2006).

Vollaard et al. (2004) reported that flooding was a risk factor for diarrheal illnesses caused by *Salmonella enterica* serotype Paratyphi A (paratyphoid fever). Flooding significantly increased the pathogen prevalence in water samples from 0% to 50% (p = 0.001). The average concentration of *E. coli* increased 10-fold from 0.48 log MPN/100 mL (standard deviation [SD]: 0.54 log MPN/100 mL) to 1.46 log MPN/100 mL (SD: 0.43 log MPN/100 mL; Mann-Whitney U test, $P < 0.001$). However, flooding had no significant impact on the prevalence of coliforms or enterococci (Mann–Whitney U test, P = 0. 207 and 0.541, respectively) (Ceuppens et al. 2014). Furthermore, Liu et al. (2016) reported that floods effect on bacillary dysentery for 3 weeks with a cumulative risk ratio of 1.52 (95% confidence interval [CI]: 1.08–2.12). Schnitzer et al. (2007) demonstrated that the risk for gastrointestinal disease during floods was related to contact with floodwaters.

Numerous studies have attempted to evaluate the risk of infection from exposure to contaminated floodwater (De Man et al. 2014; Kazama et al. 2011). De Man et al. (2014) assessed the risks of infection from exposure to urban floodwater in the Netherlands during 23 events in 2011 and 2012 using quantitative microbial risk assessment (QMRA). The results showed that *Campylobacter jejuni* was the most prevalent species in urban floodwater samples (61%, range: 14 to >103 MPN/L), followed by *Giardia* spp. (35%, 0.1–142 cysts/L), enteroviruses (35%, 103–104 pdu/L), *Cryptosporidium* (30%, 0.1–9.8 oocysts/L), and NoVs (29%, 102–104 pdu/L). When children were exposed to floodwaters originating from combined sewers, storm sewers, or rainfall-generated surface runoff, the mean risks of infection per event were 33%, 23%, and 3.5%, respectively. For adults, those risks were 3.9%, 0.58%, and 0.039%, respectively. The annual risk of infection was also calculated for flooding from different urban drainage systems. An exposure frequency of once every 10 years to flooding originating from combined sewers resulted in an annual risk of infection of 8%, which was equal to the risk of infection due to flooding originating from rainfall-generated surface runoff 2.3 times per year. However, these annual infection risks would likely increase with a higher frequency of urban flooding due to heavy rainfalls, as foreseen in climate change predictions.

Kazama et al. (2011) used a dose–response model with coliform bacteria to estimate the impact of flooding on public health. They found the annual average risk of infection during medium-sized flood events to be 0.21. The risk from groundwater use ranges from 0.12 to 0.17 in inundation areas and reaches as high as 0.23 outside the inundation areas. A high risk of waterborne disease was found in residential areas, and the annual average risk during small flood events was 0.94.

Phanuwan et al. (2006) showed that people who are exposed to floodwaters via contamination of drinking water sources or direct contact have a higher risk of viral infection. High concentrations of enterovirus, hepatitis A virus, NoVs, and adenovirus were found in water sampled from the Ciliwung River, Jakarta, Indonesia. All of these viruses were detected in one out of three groundwater wells in the flooded area; however, no viruses were found in groundwater samples in non-flooded areas or tap water samples. Furthermore, in a systematic review of 83 studies, Cann et al. (2013) identified the most common waterborne pathogens following extreme water-related weather events, such as flooding and heavy rainfall: *Vibrio* spp. (21.6%) and *Leptospira* spp. (12.7%).

Aggarwal and Krawczynski (2000) reported that hepatitis E virus outbreaks often follow heavy rains and floods, when water sources become contaminated. Flood-related outbreaks of hepatitis A and E viruses were reported in other studies as well (McCarthy et al. 1994; Corwin et al. 1999; Hau et al. 1999).

The prevalence of skin diseases during flood events was evaluated by Vachiramon et al. (2008). The authors found that eczema was the most prevalent dermatosis (34.5%); the great majority of other cases were irritant contact dermatitis. Sixteen individuals presented with itchiness and skin maceration in the web spaces of the toes.

In contrast, other studies did not demonstrate any harmful effects from floods (Fenske et al. 2001; Rohayem et al. 2006; Ceuppens et al. 2014; Aavitsland et al. (1996); Malilay 1997; Greenough et al. 2001; Sedyaningsih-Mamahit et al. 2002). Fenske et al. (2001) studied the aftermath of the Odra flood (the summer of 1997). Regular bacteriological investigations during the Odra flood showed no harmful conditions. The researchers cultivated fibroblast-like cells from ultrafiltrated 10-L water samples, demonstrating that the conditions of the Odra flood hindered the occurrence of *Enterovirus*. A comparison of the virus contents of

the Odra lagoon and water from the beaches of Usedom in both June/July 1997 and August 1997 showed fewer infectious units in the water during the flood.

Rohayem et al. (2006) assessed the risk of viral disease transmission during floods, specifically the viral burden in flooded areas of the city of Dresden (Germany) in August 2002. The authors found no increased risk for the transmission of viral diseases through water contact in flooded areas.

Epidemics of waterborne diseases were not found to follow floods in Norway (Aavitsland et al. 1996) or the United States (Malilay 1997; Greenough et al. 2001). Sedyaningsih-Mamahit et al. (2002) investigated an outbreak of hepatitis E virus in Indonesia, but found no climatic influences (flood or drought) on virus transmission in the epidemic.

2.1.4 Indirect impact of floods on human health via food contamination

Floodwaters can affect human health indirectly via food sources. Floodwaters are able to carry contaminants such as viruses, bacteria, human and animal excrement, and chemicals away from the ground and even upstream. When food items, including food crops on agricultural land, come into contact with this water, the food becomes hazardous to consume and has health risks for humans.

Flooding events have been linked to crop damage and contamination with pathogens, hazardous chemicals, and pesticides via surface runoff, remobilization of contaminated river sediments, and contaminated upstream terrestrial areas, such as grazing areas (Tirado et al. 2010; Gelting et al. 2011). The major contributors to the viral contamination of food are human sewage and feces, infected food handlers, and animals (FAO/WHO 2008), which all may be influenced by climate-induced changes. For example, flooding can result in the overflow of untreated human sewage, resulting in an increased likelihood of enteric virus contamination during the production of fresh produce and molluscan shellfish (FAO/WHO

2008). Some pathogens can persistently survive in the environment; for example, poliovirus can survive on lettuce for 23 days after the flooding of outdoor plots with wastewater (Tierney et al. 1977). This may lead to an increased health risk for people who consume food after flood events.

Many studies have revealed that flood events are associated with the contamination of food (Rojas et al. 2010; Le Guyader et al. 2008; Orozco et al. 2008; Castro-Ibanez et al. 2015; Orozco et al. 2008, Donnison and Ross 2009; Liu et al. 2013). Some outbreaks of fascioliasis in Cuba have been linked to the flooding of lettuce fields (Rojas et al. 2010). Following a flood event in France, Le Guyader et al. (2008) found that cases of gastroenteritis were associated with oyster consumption. Shellfish may become

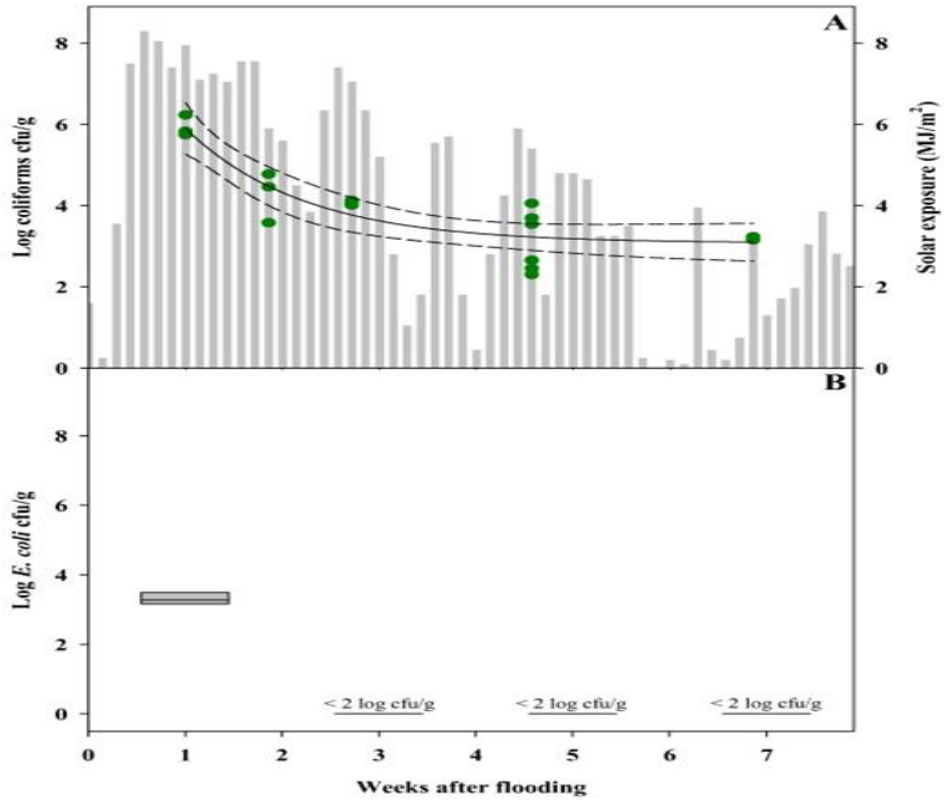

Fig. 2.1 (A) Changes in coliforms (log cfu/g) in lettuce (dots) and solar exposure (bars) after a flooding event. The solid line represents the best-fitted equation and the dotted lines are confidence bands generated by nonlinear regression analysis. (B) Boxplot of *E. coli* (log cfu/g) in lettuce after a flooding event. The bottom and top of the box represent the 25th and 75th percentiles (Castro-Ibanez et al. (2015).

contaminated after flooding, so the gastroenteritis outbreak was likely caused by the consumption of contaminated oysters. Five different viruses were detected in the shellfish: Aichi virus, NoVs, astrovirus, enterovirus, and rotavirus were detected in the shellfish. Of 205 cases of gastroenteritis, eight stool samples were positive for multiple enteric viruses and one stool sample had seven different enteric viruses.

The effects of flooding on the occurrence of *Salmonella* in a hydroponic tomato farm were investigated by Orozco et al. (2008), who found *Salmonella* and *E. coli* in samples of tomatoes. Most of the *Salmonella newport* strains were isolated from tomato samples collected during or immediately after a flood, not before the flood. Similarly, Castro-Ibanez et al. (2015) reported that flooding was a major risk factor for the microbial contamination of leafy greens. The authors obtained coliform and *E. coli* counts from lettuce at 1, 3, 5, and 7 weeks after flooding. High levels of *E. coli* (> 3 log cfu/g) were found in lettuce samples taken 1 week after flooding. The *E. coli* concentrations found in the lettuce correlated well with the levels observed in irrigation water and soil. Therefore, floodwater seems to be the most likely vector of the *E. coli* contamination in this study (Fig. 11.1). In addition, using multiplex polymerase chain reaction, lettuce samples were found to be positive for *Salmonella* spp. and verotoxigenic *E. coli* (O145, O111, O103, and O126) at 1 week after the flood event. Kawasaki et al. (2012) reported that the concentration of dimethylarsinic acid in Japanese rice grains was very low under aerobic conditions, but increased during continuous flooding. In the field experiment, the concentration of arsenic was higher during 3 weeks of flooding than in the case of intermittent irrigation.

With regard to the indirect effects of climate change on the ecology of *E. coli* O157 and *Salmonella*, intensive precipitation might be an intermediate contamination pathway for pathogens from manure on livestock farms and from grazing pastures via increased surface

and subsurface runoff. When crops are irrigated with this water, contamination may increase. Flooding as a result of extreme rain events can transport pathogens from surface water to fresh produce and could contaminate entire fields (Orozco et al. 2008; Donnison and Ross, 2009; Liu et al. 2013). Because wastewater includes many pathogenic microorganisms, the main of health risks to humans from flooding are from the consumption of crops grown in fecally contaminated soil and from the ingestion of contaminated water. Samples of agricultural soil revealed contamination by fecal waste from municipal wastewater and livestock operations after a 1999 hurricane in the United States (Casteel et al. 2006).

Floodwater also affects human health through food contaminated by chemical hazards (Lake et al. 2015; Lake_et al. 2014). Polychlorinated dibenzo-*p*-dioxins and furans (dioxins; PCDD/Fs) and polychlorinated biphenyls (PCBs) can be transferred from the environment to humans. The main route of transfer is via food—approximately 90% of human intake of PCDD/Fs and PCBs occurs this way (Liem et al. 2000). Lake et al. 2015 demonstrated that regular river flooding events transfer PCDD/Fs and PCBs to the environment (soil and grass) of industrial river catchments. Such contamination can be transferred to food. Although flooding can be a mechanism for transferring PCDD/Fs and PCBs to food, the impact varies by food type (e.g., an effect was seen for beef but not lamb). In other words, PCDD/Fs and PCBs were transferred by a flood into meat and thereby into the human food chain.

The sediments of many river systems are contaminated PCDD/Fs or PCBs around the world (Hilscherova et al. 2003; Jiamo et al. 2003; Umlauf et al. 2005). Within such areas, Lake et al. (2014) found that farming on flood-prone land may be an additional source of elevated PCDD/F and PCB levels in beef. High cadmium values were observed in wheat, lettuce,

and potatoes from the floodplain of the Meuse River after a flood event during the winter of 1993–1994. The human health risks associated with heavy metal contamination of the soil, and indirectly the food chain, seemed very low. The most important exposure risks were linked to cadmium and lead levels in soils that had a flooding frequency of once every 2 years. For lead, the most important exposure pathway was the ingestion of soil, whereas ingestion of locally grown vegetables was the principal exposure pathway for cadmium (Albering et al. 1999).

However, floodwater does not seem to be a source of *Listeria monocytogenes* contamination. Castro-Ibanez et al. (2015) examined the microbial contamination of lettuce samples that were collected 1, 3, 5, and 7 weeks after a flooding event. They only detected *L. monocytogenes* in two lettuce samples collected 3 weeks after the flood event, even though it is known to grow well on leafy greens. None of the tested samples taken 1 week after flooding were positive for *E. coli* O157:H7. Ceuppens et al. (2014) examined the influence of environmental factors on the microbiological parameters of lettuce farming. In this study, flooding had no effect on the concentrations of *E. coli*, coliforms, or enterococci (Mann–Whitney U test, $p = 0.332$, 0.143, and 0.541, respectively).

Fig. 2. 2 A simulation of floods and food as potential carriers of disease between urban and rural areas in Hue City, Vietnam (Watanabe et al. 2014).

The risk of diseases such as diarrhea can be transmitted between urban and rural areas via flood and foods. The risk of diarrhea can be spread by floods from urban areas to rural areas, where agricultural products are produced. These products, which were contaminated by polluted urban floodwaters, may then be sold to urban residents. This transfer of health risk agents between urban areas and rural areas was examined by Watanabe et al. (2014) in a study about the microbial contamination of agricultural fields that were affected by seasonal floods around Hue City, Vietnam (Fig. 2.2). In Hue City, inhabitants have a high risk of infection from seasonal flooding because they are frequently exposed to the floodwaters, which are easily contaminated with pathogens from urban drainage. The floodwater eventually flows out of the urban area and continues downstream, carrying with it various contaminants.

A priority in flood management is the protection of urban residents, but this may not be extended to the surrounding rural areas. Many rural areas are used as agricultural fields to produce fresh vegetables for urban dwellers. Watanabe et al. (2014) investigated the prevalence of *E. coli* on lettuce as a fresh vegetable and in soil samples from 29 fields and four sites in four villages. One of the villages was located upstream from the city for comparison. The authors found no clear differences in the contamination levels of the four villages before the seasonal flooding. After the flood, contamination was lowest in the village that was farthest downstream from the city. Multiple linear regression analysis demonstrated that the upstream village ($P < 0.1$) and manure use ($P < 0.05$) were significant contributors to contamination after the flood, whereas there were no significant factors

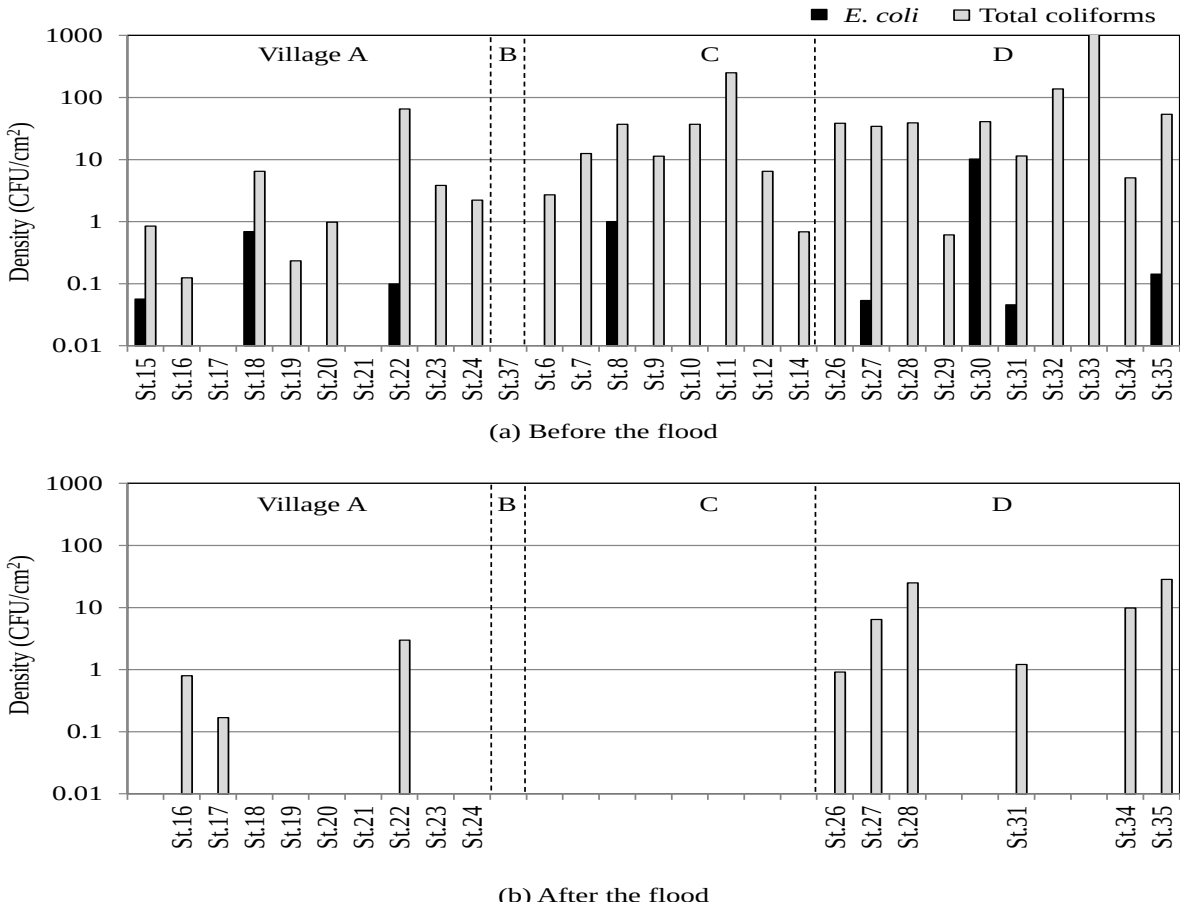

Fig. 2.3 Microbial contamination of vegetable samples collected before and after a seasonal flood (Watanabe et al. 2014).

before the flood. Seasonal flooding washed contamination from the fields, as demonstrated by the relatively low level of contamination in the most remote village. Manure, which may be used improperly, had more of an effect on the microbial contamination of agricultural fields than the seasonal flood (Fig. 2.3).

2.2 Evaluation risk of the food-borne gastroenteritis related to floods

Norovirus are the most common cause of gastroenteritis worldwide and the leading cause of food-borne gastroenteritis (Ramani at el. 2014). NoV belongs to the family *Caliciviridae* and has a positive-sense single-stranded RNA genome. NoVs are classified into seven genogroups (GI to GVII; Vinje 2015). GI, GII, and GIV infect humans (Zheng et al. 2006; Glass et al. 2009). The major mode of NoV transmission is the fecal-oral route involving direct or indirect ingestion of contaminated food or water (Glass et al. 2009).

NoV concentrations in oysters have a strong seasonal trend with higher concentrations observed during the winter months (Flannery et al. 2012; Rajko-Nenow et al. 2012; Le Mennec et al. 2017). After heavy rains cause flooding, contamination of oyster-producing areas by NoVs can occur because of untreated wastewater effluent or sewage treatment failure (Astrom et al. 2009; Wang & Deng 2012; Grodzki et al. 2012). Some studies in developed countries revealed that the same genotype of NoVs were detected in cases and oysters cultivated downstream of residential areas (Ueki et al. 2005; Pu et al. 2016).

Several molecular epidemiological studies have been conducted for NoVs circulating in human populations in developing countries including Vietnam (My et al. 2013b), Bangladesh (Rahman et al. 2016), Thailand (Supadej et al. 2017), India (Menon, et al.

2016), Nicaragua (Bucardo et al. 2014), and Peru (Saito et al. 2014). However, environmental data are extremely limited in those countries.

The Quantitative microbial risk assessment (QMRA) procedure (Haas et al. 1999), consists of four steps, which is including hazard identification, exposure assessment, dose-response relationship, and risk characterization, was used to evaluate risk infection due to consuming oysters contaminated with NoVs by consumers.

The dose of pathogen per exposure [pathogen intake (number of the pathogen) per day] was calculated separately for average dose and maximum dose using Eq.2.1 and Eq.2.2, respectively, as follows.

$D_{Mean} = C_v \times M_w \times M_c$ (Eq.2.1)

$D_{Max} = C_v \times Max_w \times Max_c$ (Eq.2.2)

Where, D is dose; C_v is concentration of virus per digestive tissue; M_w is mean of weight of digestive tissue; Max_w is maximum of weight of DT; M_c is mean of number of oysters consuming at a time; Max_c is maximum amount of oysters consuming at a time.

The probability of infection for NoVs was calculated using a Fractional Poisson dose-response model (Mesner et al. 2014).

Model P (D_{Mean}, τ) = $\tau \times (1-e^{-D/\mu})$ (Eq.2.3)

Model P (D_{Max}, τ) = $\tau \times (1-e^{-D/\mu})$ (Eq. 2.4)

Where, τ is the fraction of susceptible subjects; μ is the mean aggregate size; $e^{-D/\mu}$ is the probability of receiving an exact dose of 0. Until now, the parameter τ and μ have not been available for Vietnamese people. These parameters were based on a previous study in American people (Mesner et al. 2014) with $\tau = 0.722$ and $\mu = 1106$.

The monthly probability of infection for NoVs was calculated separately for mean dose and maximum dose using Eq.2.5 and Eq.2.6, respectively.

$P_{Mean}(D_{Mean}, \tau) = 1 - (1 - P(D_{Mean}, \tau))^{n}$ (Eq. 2.5)

$P_{Max}(D_{Max}, \tau) = 1 - (1 - P(D_{Max}, \tau))^{n}$ (Eq. 2.6)

Where, n: time per month

2.3 Indirect impacts of floods in terms of psychology

People living in flooding area have suffered many infectious diseases occur after flood events such as diarrhea, skin & eye infections, malaria, respiratory infectious diseases, and leptospirosis, leishmaniasis, hepatitis, dengue fever, pink eye, dermatitis, coughs, sore throat, and general sickness (Kondo et al. 2002; Carroll et al. 2010; Amilasan et al. 2012; Baqir et al. 2012). On the other hand, mental health problems such as anxiety, stress, psychological distress, probable anxiety, probable depression, probable post-traumatic stress disorder, were reported following flood events (Carroll et al. 2010; Paranjothy et al. 2011; Hetherington et al. 2017), or increase the risk of stressors, that are indirectly associated with the flooding such as economic problem, worry about the reoccurrence of floods on who are affected, as the risk of secondary stressors (Stanke et al. 2012). As such, quantify feeling of people affected by flooding which is an indicator of psychology, is important for improving prevention and mitigation to the impact of flooding on psychological and mental health.

Some previous studies have shown that sex, age, and education are potential risk factors linked to poorer mental health (Ginexi et al. 2000; Tunstall et al. 2006; Collins et al. 2013).

2.4 Summary of this chapter

1) With global climate change, the frequency of floods has increased in recent decades. The direct health impacts of these floods are generally easier to measure than their indirect health impacts, emphasizing the need for measures to prevent the indirect health impacts. Although there have been some studies to the contrary, this review of the literature clearly shows that floods and food are potential carriers of disease agents between urban and rural areas. The indirect damage from flooding should be addressed in food safety management and research, as well as in health risk assessments.

2) Psychological impacts caused by floods were pointed out based on many previous studies

3) Risk evaluation process and a Fractional Poisson dose-response model have been proposed for evaluating the risk of NoVs infection due to the contaminated oysters.

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CHAPTER 3

THE PREVALENCE OF NOROVIRUS IN CASES OF GASTROENTERITIS IN DEVELOPING COUNTRIES

3.1 Introduction

To estimate the prevalence of NoVs in cases of gastroenteritis in developing countries, a systematic review and meta-analysis were performed based on 178 articles that were published from 1990 to 2016.

Norovirus is an important cause of nonbacterial gastroenteritis worldwide (Patel et al. 2008; Glass et al. 2009). According to Ahmed et al. (2014) global NoV prevalence in community cases of gastroenteritis was 24%, while the prevalence in outpatients and patients admitted to hospitals for treatment of symptoms associated with NoV were 20% and 17%, respectively. NoVs has surpassed rotavirus as the predominant cause of acute gastroenteritis in children in countries where rotavirus vaccination has been introduced (McAtee et al. 2016; Hemming et al. 2013). Although seven genogroups of NoV (GI-GVII) have been identified (Vinje 2015), two genogroups (GI and GII) are the main causes of NoV-associated gastroenteritis; a recent increase in NoV GII-associated gastroenteritis was reported not only in developed countries (Tu et al. 2008; Eden et al. 2010; Mathijs et al. 2011; Vega et al. 2011; Motomura et al. 2016) but also in developing countries (Tan et al. 2015; Alam et al. 2016).

There is a considerable difference in access to safe water, sanitation, and hygiene between developed and developing countries (World Health Organization and UNICEF, 2012;

Thapar et al. 2004), which may correlate with a higher risk of NoV-associated gastroenteritis in developing countries. Patel at el. (2008) estimated that NoV in cases of gastroenteritis in developing countries was 12% among children under 5 years old. However, this estimation was based on datasets from 11 studies in only seven developing countries; the prevalence varies by country depending on location, age group, economic status, severity of symptoms, and epidemic conditions. Considering these factors, Ahmed et al. (2014) performed a meta-analysis of the prevalence of NoV in cases of gastroenteritis on a global scale but mentioned that a clear data gap remains for determining the NoV prevalence in developing countries due to lack of high quality reports on the prevalence in these countries. We searched the relevant reports from developing countries defined by World Bank as low-income and middle income countries (World Bank 2015), using the databases other than those used by Ahmed et al. (2014), including the most recent publications. Using a large number of papers identified, this study aimed to estimate the prevalence of NoVs in cases of acute gastroenteritis in developing countries from 1990 to 2016 by means of systematic review and meta-analysis.

Due to inadequate access to safe drinking water, poor sanitation and hygiene, I hypothesized that the prevalence of NoVs in case of acute gastroenteritis in developing countries is higher than that in developed countries.

3.2 Methods

Search strategy and selection criteria

The procedure to search and select studies relevant to this review was similar to Ahmed et al. (2014). A systematic search was performed using PubMed and Web of Science databases of studies published between January 1, 1990 and March 31, 2016. The search terms used were as follows: “Noroviruses”, “Norwalk-like Viruses”, “Norwalk like Viruses”, “Small

Round-Structured Viruses", "Round-Structured Viruses, Small", and "Small Round Structured Viruses". Full-text articles were assessed and selected if they met the following inclusion criteria: used PCR-based diagnostics for all stool specimens from patients, enrolled patients who presented with symptoms of acute gastroenteritis, were performed in developing countries as defined by the World Bank (2015), and had a study period of at least 12 months.

We excluded papers which did not have an English abstract, did not show the number of patients with acute gastroenteritis or patients positive for NoV, or percentages that could be used for calculating prevalence. If the same data were repeated in multiple studies, only the most complete study was considered. Additionally, papers, which reported neither diarrhea nor vomiting as common symptoms of gastroenteritis, were also excluded.

Data extraction

From the eligible articles, we obtained the following information: first author, title, journal, year of publication, country, period and duration of study, surveillance setting, number of patients with acute gastroenteritis, number of patients positive for NoV, number of patients without acute gastroenteritis, number of cases tested without acute gastroenteritis but positive for NoV (asymptomatic NoV infection), and age group. Data were stratified by age of subjects, surveillance setting, and country income level categorized according to World Bank classifications (World Bank 2015). For age, the data were stratified into two groups: under 5 years, and 5 years and over. The data unable to be stratified in this manner were not stratified and treated as "mixed age". For surveillance setting, the data were stratified into four different settings as a proxy for severity of symptoms: "community" indicating patients with relatively mild symptoms which were only recognized by community cohort studies, "outpatients", "inpatients" including those who visited an emergency department, and "other

setting", which included studies that contained no specific setting or unstratified data. For income level, data were stratified based on estimates of gross national income (GNI) per capita for 2014 countries were classified into high income, upper middle-income, lower middle-income, or low-income groups (World Bank 2015). In addition, where possible, the data were stratified by NoV genogroups as well. Studies performed during December 1, 2002 to January 31, 2003 or December 1, 2006, to January 31, 2007 were considered as reporting pandemic data, while the remaining studies were considered as reporting endemic data according to the definition of a previous review (Ahmed et al. 2014).

Statistical analysis

A meta-analysis was conducted for NoV prevalence in cases of acute gastroenteritis for the prevalence of NoV GI and GII and asymptomatic NoV infection. Heterogeneity between studies was evaluated using the I^2 test. All analyses were performed using STATA software version 14 (Stata Corp, College Station, TX). *P*-values < 0.05 were considered statistically significant.

3.3 Results

A literature search of two electronic databases identified 8,280 relevant articles for the present systematic review (Fig. 3.1). We narrowed down the relevant articles through assessment of titles and abstracts and reviewed 539 full-text articles for eligibility; 377 articles were excluded based on exclusion criteria (83 were from high-income countries, 74 were outbreak reports, 11 were written in foreign language, 19 discussed laboratory methods, 11 did not use PCR-based diagnosis, 8 were laboratory reports, 17 included insufficient data, 3 reported traveler's diarrhea, 69 had a study period < 1 year, 37 reported no data, and 45 were unable to be assessed). A total of 162 papers met the inclusion criteria, which included 85 studies from Ahmed et al. (2014) and full data were extracted from all

included studies. The final dataset consisted of 178 papers from 46 countries; a majority of studies were from China (n = 57), Brazil (n = 22), India (n = 18), and Thailand (17) (Fig.3. 2). Study countries were categorized into upper middle- (n = 19), lower middle- (n = 21), and low-income countries (n = 6) (Table 3.1).

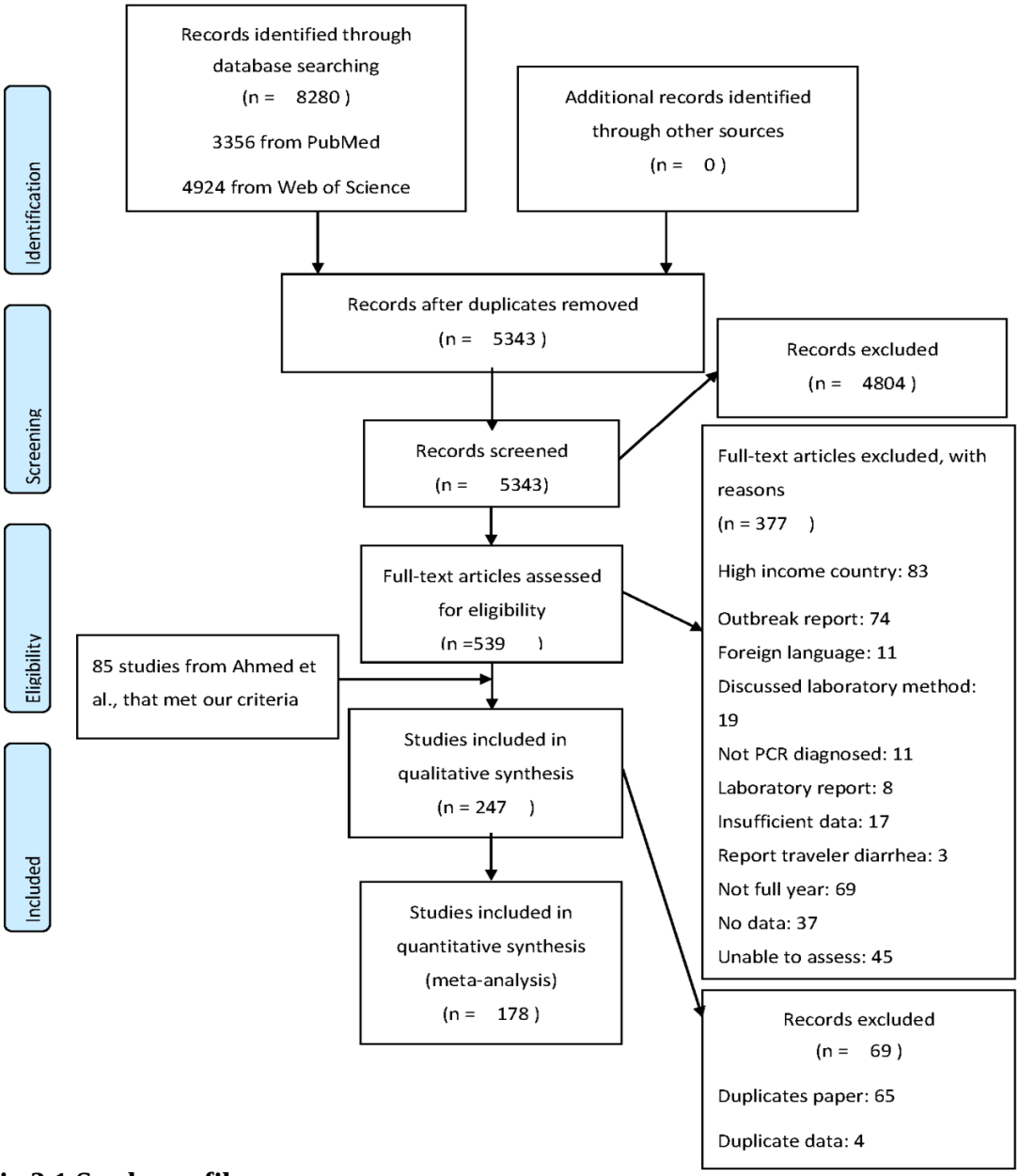

Fig 3.1 Study profile

From: Moher D, Liberati A, Tetzlaff J, Altman DG, The PRISMA Group (2009). Preferred Reporting Items for Systematic Reviews and Meta-Analyses: The PRISMA Statement. PLoS Med 6(7): e1000097. doi:10.1371/journal.pmed1000097

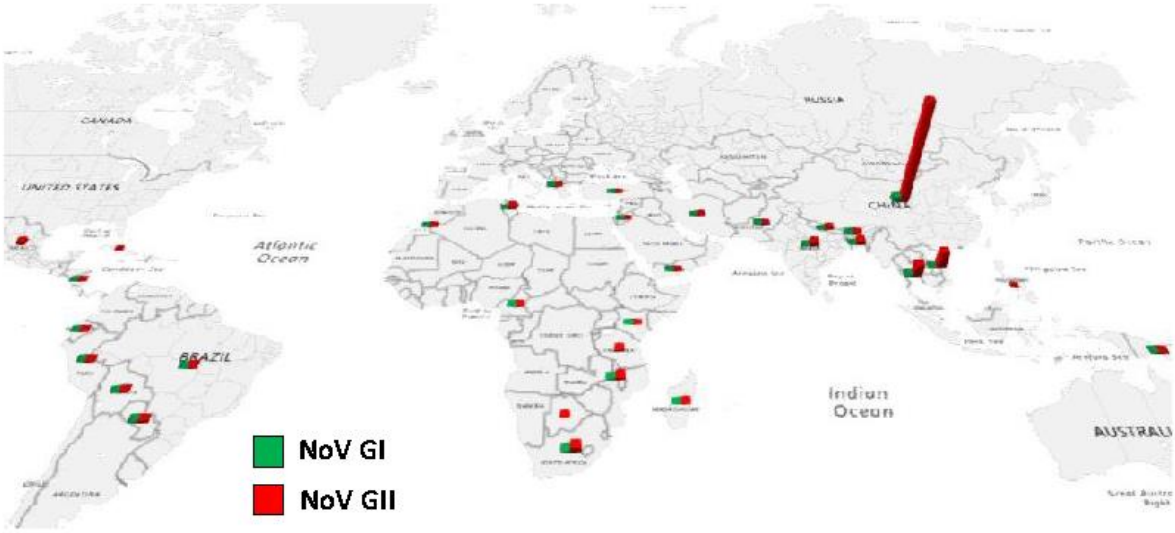

Fig 3.2 Countries reported on NoV GI and GII prevalence in meta-analysis

Table 3. 1. Distribution of data among various strata.

	Number of studies (n=178)	Cases tested (n=148,867)
Age groups		
<5 years	114	87,684
>= 5 years	22	7,545
Mixed	72	53,638
Setting		
Inpatient	118	69,290
Outpatient	43	58,312
Community	15	5,025
Other	32	16,240
Country income level		
Low income	6	14,577
Lower middle income	21	31,232
Upper middle income	19	103,058
Pandemic period		
Included	54	34,473
Not included	154	114,394

A pooled analysis of all 178 articles revealed the NoV prevalence among 148,867 patients with acute gastroenteritis was 17% (95% CI: 15-18; $P < 0.01$ test for heterogeneity (P_h)) as shown in Table 3.2. There were no differences in NoV prevalence among the three age groups assessed ($P_h = 0.727$). Country income level significantly affected the NoV prevalence; a decrease from 18% in upper middle- to 15% in lower middle- and 6% in low-income countries ($P_h < 0.001$) was noted. Prevalence during pandemics was significantly different from that under endemic time periods ($P_h < 0.001$). There were no significant differences in prevalence characterized by severity of symptoms defined by community (16%), inpatient (17%), or outpatient settings (16%) ($P_h = 0.897$). Related to the severity, NoV prevalence was noted in 5% of patients without gastroenteritis (95% CI: 4-8%; $I^2 = 95.25\%$; $P_h < 0.001$) (Table 3.3).

Table 3. 2 NoV prevalence in patients with gastroenteritis.

	I^2 (%)	*P*	Effect size (95% CI)
Age group			
<5 years	97.73	<0.01	0.16 (0.14-0.18)
>= 5 years	95.26	<0.01	0.17 (0.13-0.21)
Mixed	98.84	<0.01	0.17 (0.14-0.21)
Heterogeneity between groups	0.727		
Setting			
Inpatient	98.40	<0.01	0.17 (0.14-0.19)
Outpatient	98.46	<0.01	0.16 (0.14-0.19)
Community	95.74	<0.01	0.16 (0.11-0.21)
Other	97.96	<0.01	0.18 (0.14-0.22)
Heterogeneity between groups	0.897		
Country income level			
Low income	98.01	<0.01	0.06 (0.03-0.10)
Lower middle income	97.61	<0.01	0.15 (0.13-0.18)
Upper middle income	98.30	<0.01	0.18 (0.16-0.20)
Heterogeneity between groups	<0.01		
Data in pandemic period			
Included	97.65	<0.01	0.17 (0.14-0.20)
Not included	98.29	<0.01	0.17 (0.15-0.18)
Heterogeneity between groups	<0.001		
Overall	**98.27**	**<0.01**	**0.17 (0.15-0.18)**

Table 3.3. NoV prevalence in patients without gastroenteritis.

	I^2 (%)	*P*	Effect size (95% CI)
Age group			
<5 years	92.04	<0.01	0.06 (0.04-0.08)
>= 5 years	99.58	<0.01	0.04 (0.02-0.06)
Mixed	98.02	<0.01	0.05 (0.01-0.13)
Heterogeneity between groups	0.411		
Setting			
Inpatient	94.67	<0.01	0.06 (0.03-0.10)
Outpatient	90.86	<0.01	0.04 (0.02-0.07)
Community	93.44	<0.01	0.07 (0.02-0.14)
Heterogeneity between groups	0.161		
Country income level			
Lower middle income	92.13	<0.01	0.04 (0.02-0.07)
Upper middle income	96.98	<0.01	0.08 (0.04-0.12)
Heterogeneity between groups	<0.001		
Data in pandemic period			
Included	82.72	<0.01	0.10 (0.05-0.17)
Not included	95.75	<0.01	0.05 (0.03-0.07)
Heterogeneity between groups	0.036		
Overall	**95.25**	**<0.01**	**0.05 (0.04-0.08)**

The pooled analysis revealed a higher prevalence of NoV GII (15%; $I^2 = 98.57\%$, $P < 0.001$) than NoV GI (1%; $I^2 = 92.91\%$, $P < 0.001$) in patients with acute gastroenteritis, as shown in Tables 3.4 and 3.5. The prevalence of NoV GII, which was the predominant genogroup, in patients with acute gastroenteritis was assessed by country income, pandemic or endemic classification, and setting as an indicator of symptom severity. NoV GII prevalence was lower in studies only during pandemic periods (13%, 10-15) than in whole studies including endemic periods (17%; 14-19; P_h <0.01). The NoV GII prevalence was higher in upper middle-income countries (16%; 14-19; P_h<0.001) than lower middle- (14%; 11-18;

P_h<0.001) or low-income countries (6%; 3-11; P_h<0.001). There were no differences in the NoV GII prevalence (P_h = 0.557) by study setting (as an indicator of symptom severity).

Table 3.4. NoV GI prevalence in patients with acute gastroenteritis.

	I^2 (%)	***P***	**Effect size (95% CI)**
Age group			
<5 years	90.72	<0.01	0.01 (0.00-0.01)
>= 5 years	89.74	<0.01	0.02 (0.01-0.04)
Mixed	94.94	<0.01	0.01 (0.01-0.02)
Heterogeneity between groups	0.041		
Setting			
Inpatient	92.5	<0.01	0.01 (0.01-0.01)
Outpatient	82.92	<0.01	0.00 (0.00-0.01)
Community	95.00	<0.01	0.03 (0.00-0.08)
Other	94.46	<0.01	0.02 (0.01-0.04)
Heterogeneity between groups	0.003		
Country income level			
Low income	91.1	<0.01	0.00 (0.00-0.01)
Lower middle income	91.00	<0.01	0.01 (0.00-0.01)
Upper middle income	90.91	<0.01	0.01 (0.01-0.01)
Heterogeneity between groups	0.097		
Data in pandemic period			
Included	93.34	<0.01	0.01 (0.00-0.02)
Not included	92.79	<0.01	0.01 (0.01-0.01)
Heterogeneity between groups	0.674		
Overall	**92.91**	**<0.01**	**0.01 (0.01-0.01)**

Table 3.5. NoV GII prevalence in patients with acute gastroenteritis.

	I^2 (%)	*P*	Effect size (95% CI)
Age group			
<5 years	98.05	<0.01	0.15 (0.12-0.17)
>= 5 years	95.36	<0.01	0.15 (0.10-0.19)
Mixed	98.98	<0.01	0.16 (0.13-0.21)
Heterogeneity between groups	0.704		
Setting			
Inpatient	98.78	<0.01	0.15 (0.12-0.18)
Outpatient	98.73	<0.01	0.17 (0.13-0.21)
Community	87.13	<0.01	0.13 (0.09-0.18)
Other	96.35	<0.01	0.17 (0.13-0.21)
Heterogeneity between groups	0.557		
Country income level			
Lower income	98.53	<0.01	0.06 (0.03-0.11)
Lower middle income	97.88	<0.01	0.14 (0.11-0.18)
Upper middle income	98.38	<0.01	0.16 (0.14-0.19)
Heterogeneity between groups	0.001		
Data in pandemic period			
Included	97.58	<0.01	0.13 (0.10-0.15)
Not included	98.50	<0.01	0.17 (0.14-0.19)
Heterogeneity between groups	<0.01		
Overall	**98.57**	**<0.01**	**0.15 0.13-0.17)**

3.4 Discussion

NoV is recognized as an important cause of nonbacterial gastroenteritis worldwide. Although acute gastroenteritis may occur more frequently in developing countries due to poor sanitation and hygiene, NoV prevalence is still unknown due to a lack of published well-structured data. Our meta-analysis indicates that the NoV prevalence in cases of acute gastroenteritis in developing countries was 17%.

Our review used a similar design for the systematic review as Ahmed et al. (2014), with small modifications. For example, we stratified economic status based on the country income level (World Bank 2015) instead of WHO mortality stratum (Beaglehole et al. 2003) in order to highlight differences among developing countries. We also added an additional analysis of the data stratified by NoV genogroup.

NoV prevalence in this study was similar to that in Ahmed et al. (2014). They estimated the global prevalence of NoV in cases of gastroenteritis to be 18% (95% CI: 17%-20%) with a clearly remaining gap in developing countries. Our finding thus was not consistent with the previous reports that the prevalence of NoV in case of gastroenteritis in developing countries should be higher than in developed countries due to limited access to safe water, sanitation, and hygiene (World Health Organization and UNICEF 2012; Thapar et al. 2004). Poor surveillance about NoV and the tendency of people not to use medical services for mild diseases such as gastroenteritis caused by NoV could cause the underestimation of the prevalence in developing countries. Even in this situation, we found much higher estimates of NoV prevalence than those reported in Africa (11%) (Kabue et al. 2016) and Latin America (15%) (O'Ryan et al. 2017), indicating that the other regions targeted in the present study (i.e., Asia and the Middle East) probably have a higher prevalence.

The present study also revealed a significant difference in NoV prevalence based on genogroups GI (1%) and GII (15%). The much higher prevalence estimate of NoV GII is supported by many previous studies (Kabue et al. 2016; O'Ryan et al. 2017; Siebenga et al. 2009; Zheng et al. 2010) which show that 37% to 100% of NoV-associated acute gastroenteritis is associated with this genogroup.

By age, prevalence was similar in patients with acute gastroenteritis under 5 years (16%; 14%-18%), 5 years and over (17%; 13%-21%), and of mixed ages (17%; 14%-21%). Our finding was consistent with Ahmed et al. (2014), who pointed out that global NoV prevalence in under 5 years, 5 years and over, and mixed ages was similar [18% (95% CI: 15%-20%), 18% (95% CI: 13%-24%) and 19% (95% CI: 17%-21%), respectively].

By country income, prevalence decreased as income decreased. Since low-income countries have more pathogens to cause acute gastroenteritis than other countries, the proportion of NoV causing acute gastroenteritis in these countries was lower than countries of other income statuses (Ahmed et al. 2014). Another possible reason for this trend is that people in low-income countries tend not to use medical services for mild gastroenteritis caused by NoV because of the economic burden. This could lead to underestimation of NoV prevalence in those countries, especially in outpatient and inpatient settings.

Prevalence was similar between studies from pandemic time periods (17%; 14%-20%) and studies from endemic time periods (17%; 15%-18%). Our finding was consistent with Ahmed et al. (2014), which estimated the prevalence in pandemic time periods (2002-2003 and 2006-2007) and the whole periods including endemic time was 19% (95% CI: 16%-22%) and 18% (95% CI: 16%-20%), respectively.

By setting, prevalence was similar between patients with acute gastroenteritis in community (16%; 11%-21%), inpatient settings (17%; 14%-19%), and outpatient settings (16%; 14%-

19%). Our results contradict that of previous studies. Ahmed et al. (2014) estimated that global NoV prevalence was lower in community settings compared to outpatient and inpatient settings. A possible reason for the difference between our finding and the previous finding are that we stratified country setting by income instead of WHO mortality stratum (Beaglehole et al. 2003), and high-income countries were not included in the final analysis. This is supported by O'Ryan et al. (2017) estimating that NoV prevalence in Latin America was 15% in the community (95% CI: 11%-21%), 14% in outpatient settings (95% CI: 10%-19%), and 16% in hospital settings (95% CI: 12%-21%).

There are several strengths and potential limitations in the present study. Our systematic review, based on a reasonably large number of studies (178 studies in 46 developing countries worldwide), provided a more probable estimate of NoV prevalence in developing countries by summarizing data with a wide range of estimates and study designs. It also provided the prevalence of NoV GI and GII independently. Weaknesses include the stratification of age into only two categories. To determine prevalence in other age groups, such as young adults or the elderly, an additional meta-analysis would need to be performed. In addition, the overall prevalence is potentially overestimated because the data in the pandemic periods 2002-2003 and 2006-2007, due to the emergence of new GII.4 variants, were included in the analysis, as pointed out by Ahmed et al. (2014). Moreover, as stated above and exemplified in Fig.3.1, we could not assess 45 relevant studies, and their exclusion may have biased our results. Finally, since number of cases was not limited in order to collect as many studies as possible in developing countries, some studies reporting a few cases of gastroenteritis may have produced a bias in the estimated prevalence.

In conclusion, the estimated prevalence of NoV in cases of acute gastroenteritis was 17% in developing countries. This review can be used to estimate the burden of NoV-associated acute gastroenteritis in developing countries, which is currently unclear due to poor

diagnosis and surveillance systems, and the estimation may enhance the development of human NoV vaccines.

3.5 Summary of this chapter

As the results, following conclusions were obtained:

1) The estimated NoV prevalence among 148,867 patients with acute gastroenteritis in developing countries was 17% (95% CI: 15-18%)

2) The prevalence decreased from 18% (95% CI: 16%-20%) for upper middle-income countries to 15% (13%-18%) and 6% (3%-10%) for lower middle- and low-income countries, respectively.

3) There were no significant differences in NoV prevalence by age group (under 5 years, 5 years and over, and mixed ages) or severity of symptoms as defined by community, outpatient, or inpatient setting.

4) The pooled prevalence of NoV GII (15%, 95% CI: 13%-17%) was significantly higher than that of NoV GI (1%, 95% CI: 1%-1%) in patients with acute gastroenteritis.

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CHAPTER 4

FOOD CONTAMINATION DUE TO NOROVIRUS, URBAN FLOOD, AND HEALTH RISK

4.1 Introduction

As mentioned in the previous chapter, the prevalence of NoVs in cases of gastroenteritis in developing countries was lower than those in developed countries because of the poor surveillance system in developing countries, and people in developed countries tended to go to see specialists in hospitals more than those in developing countries. To overcome these limitations, we tried to conduct a field survey in Hue City. As a potential indicator for the epidemic of gastroenteritis, monitoring NoV contamination in oysters, which was reported effectively in developed countries (Ueki et al. 2005; Pu et al. 2016), was applied to predict the epidemic NoVs in cases of gastroenteritis in the city. In addition, risks of NoV infection due to consuming oysters in the city was also estimated.

The objectives in this chapter are to reveal the epidemic of gastroenteritis in Hue city by monitoring NoV contamination in oysters collected in a downstream lagoon receiving urban drainage from the city for 17 months (from August 2015 to December 2016), and assess the risk of NoV infection due to consuming oysters harvested in the lagoon.

4.2 Research area and sample collection

Similar to most cities in developing countries, Hue City, Vietnam, has no wastewater treatment plants. Domestic wastewater is discharged into drainage channels after passage of some of the wastewater through septic tanks. The channels flow directly into the Perfume River. Wild oysters harvested downstream of the city could be susceptible to contamination by NoVs discharged in the urban wastewater. In Vietnam, oysters are often consumed raw,

fried, dried, or baked, with a mean consumption of 4.9 g/person/day (Nguyen et al. 2012). Vietnam has a tropical monsoon climate with the rainy and dry seasons. There is a possibility of an increased risk of NoV infections associated with consumption of raw oysters during the rainy season. Despite this public health concern, no quantitative data on NoV concentrations in oysters and wastewater have been reported in Vietnam.

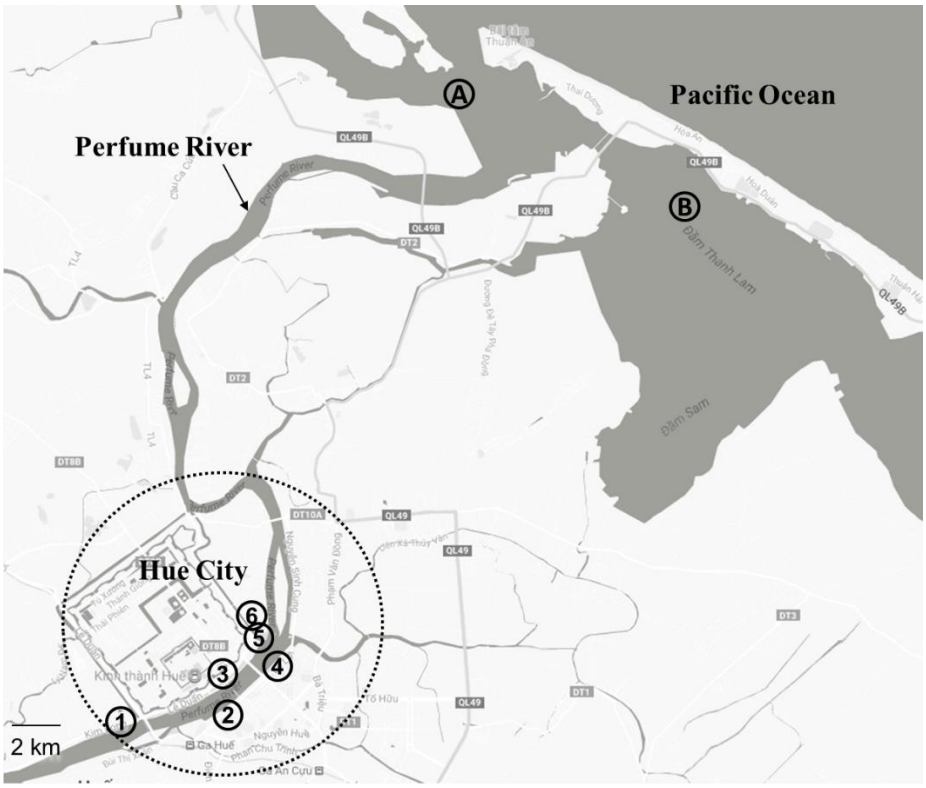


Fig. 4.1 Sampling sites. The Perfume River flows across Hue City and reaches the Pacific Ocean. Oyster samples were collected at the stations A and B in the lagoon. Wastewater samples were collected at six sites in Hue City. 1, Kim Long; 2, Le Loi; 3,Coopmark; 4, Doi Cung; 5, Trinh Cong Son; 6, Bach Dang.

Hue City is located at 16°28′00″N, 107°34′45″E in the central part of Vietnam, on the banks of the Perfume River (Fig. 4.1). The city has 354,124 inhabitants as of 2015. It has a tropical monsoon climate with an average annual rainfall of about 3,000 mm. The rainy season usually begins in August and ends in January, with a flood season from October onwards. The dry season is from March to August. The average temperature is 25°C.

Municipal wastewater collection and treatment systems have not been established, although some houses are equipped with septic tanks. In the Kinh Thành (citadel) area of the city, 56% of wastewater is treated by septic tanks (Lieu et al. 2010). Domestic wastewater is directly or indirectly discharged into drainage channels that empty into the Perfume River. Therefore, drainage and flood waters are likely the most probable sources of viruses detected in oysters.

Wild oyster (*Crassostrea ariakensis*) samples were collected every month from August 2015 to December 2016 at two sites (station A and B, Fig. 4.1) in a lagoon located at the mouth of the Perfume River. Nine individual oysters were collected at each sample site, for a total of 306 individual oysters. Station A is located close to the river mouth. Station B is a commercial oyster harvesting area located approximately 6 km east of station A. The oysters at station B are anticipated to be contaminated by the flow from the river, especially in the rising tide, and by domestic wastewater discharge from some households along the lagoon bay.

Wastewater samples were collected from six sites of the urban drainage channels in Hue City from June to December 2016 (1, Kim Long; 2, Le Loi; 3, Coopmark; 4, Doi Cung; 5, Trinh Cong Son; 6, Bach Dang; Fig. 4.1). Approximately 100 to 500 mL of wastewater samples were collected from the outfalls of the drain tunnels adjacent to the Perfume River every month. Oyster and wastewater samples were transported to the laboratory in the Department of Microbiology & Carlo Urbani Center, College of Medicine and Pharmacy, Hue University, within 1 h under refrigeration (<10°C).

4.3 Materials and Methods

Viral extraction

Oyster samples

Oysters were opened using an autoclaved shucking knife. Afterward, the digestive tissue of the three individual oysters was collected and transferred into a 2-mL tube using a pair of autoclaved scissors and weighed. Viruses were extracted from the composite digestive tissue sample in triplicate (i.e., three composite samples from nine individual oysters per site). Ten microliters of murine NoV (MNV, S7-PP3 strain) stock solution kindly provided by Dr. Yukinobu Tohya (Nihon University, Kanagawa, Japan), and two stainless beads (3.2 mm diameter; Tomy Seiko, Tokyo, Japan) were added to each composite digestive tissue sample. One milliliter of enzyme solution (6.24 mg/mL amylase, 6.24 mg/mL lypase, and 0.252 mg/mL proteinase K) was added. The samples were mashed using a Mini BeadBeater (BSP 3110BX; BioSpec Products, Bartlesville, OK, USA) at 4800 oscillations/min for 2 min. The samples were then incubated at 37°C for 60 min for digestion by enzymes, and further incubated at 60°C for 15 min for deactivation of the enzymes. The samples were centrifuged at 9100 × *g* for 12 min. The supernatants (approximately 3.0-3.5 mL) were collected and stored at −80°C.

Wastewater samples

Viral extraction was performed using mixed cellulose ester membrane (type HA; 0.45 μm pore size and 90 mm diameter; Millipore, Billerica, MA, USA) as previously described (Katayama et al. 2002). Briefly, after adding $MgCl_2.6H_2O$ to the collected wastewater samples to a final concentration of 25 mM, 100-500 mL of the solution was filtered through the membrane. This was followed by 200 mL of 0.5 mM H_2SO_4 (pH 3) and 10 mL of 1 mM NaOH (pH 10.8). The filtrate (virus concentrate) was dispensed in a 50-mL

tube along with 100 µL of 50 mM H_2SO_4 and 100 µL of 100× TE buffer, and mixed well to neutralize. The virus concentrate samples were stored at −80°C.

RNA extraction and reverse transcription

After adding 500 µL of 400 mM citric buffer (pH 2.5) to the 500 µL supernatants prepared from the oyster samples, each mixture was centrifuged at 9100 × g for 12 min. Viral RNA was extracted from 300 µL of the supernatant or virus concentrate from wastewater samples using a Direct-zol RNA MiniPrep (Zymo Research, Irvine, CA, USA) following the manufacturer's protocol. Undiluted and ten-fold diluted RNA extracts were prepared for each sample to evaluate the occurrence of inhibition in reverse transcription (RT) and real-time PCR. cDNA was obtained through RT reaction using iScript Advance cDNA Synthesis Kit (Bio-Rad, Hercules, CA, USA) on a Veriti 96-well Thermal Cycler (Thermo Fisher Scientific, Waltham, MA, USA).

NoV quantification by real-time PCR

Real-time PCR assays were carried out to quantitatively detect NoV GI and GII using SsoFast Probes Supermix (Bio-Rad) on a CFX96 Touch Real-Time PCR Detection System (Bio-Rad) with previously reported primers and probes (Kazama et al. 2016). Forward primer COG1F, reverse primer COG1R, and probes RING1-TP(a) and RING1-TP(b) (Kageyama et al. 2003) were used for NoV GI. Forward primers COG2F and ALPF, reverse primer COG2R (Kageyama et al. 2003), and probe RING2AL-TP (Ministry of Health, Labour and Welfare, 2007) were used for NoV GII. MKMNVF and MKMNVR primers and MKMNV-TP probe were used for MNV (Hata et al. 2011). Twenty microliters of reaction mixture contained 5 µL of cDNA, 10 µL of SsoFast Probes Supermix (Bio-Rad), primers, and probe with the concentrations specified in Table S1. All samples were analyzed

using duplicate wells. The reaction temperature and time were 95°C for 30 s and 49 cycles of 95°C for 15 s and 56°C for 60 s.

The number of NoV genome copies was determined based on the standard curve prepared from a log dilution series of GI and GII DNA standard (range, 10^1-10^6 copies per well). Cycle quantification (C_q) value of 40 was set for a limit of quantification (LOQ) (Bustin et al. 2009). When the C_q value over 40 was obtained, the result was reported as under LOQ. When all replicates were negative, the result was marked as not detected (ND). The number of NoV genome copies in digestive tissue was calculated by considering the sample dilution and quantitation processes. All samples were assessed for extraction efficiency using MNV as a process control. The extraction efficiency was calculated by dividing the amount of MNV in the virus concentrate by the amount of MNV spiked into samples. When the extraction efficiency exceeded 1%, the concentrations of NoV GI and GII were calculated and those of three composite samples were reported for each month (ISO/TS 15216-1:2017).

Pyrosequencing

Thirteen oyster and wastewater samples were selected for pyrosequencing considering the results of real-time PCR and agarose gel electrophoresis after semi-nested PCR. For oyster samples, three samples for GI (September, October, and November 2016 in station A) and four samples for GII (October and December 2016 in both stations) were selected. Three wastewater samples were selected for GI analysis and four samples for GII analysis. Collection was done in the Doi Cung, Le Loi, and Kim Long drainage channels from September to December 2016.

Semi-nested PCR was performed as described previously (Kazama et al. 2016). Briefly, COG1F, G1SKR, G1SKF, and G1SKR were used for GI, and COG2F, G2SKR,

G2SKF, and G2SKR were used for GII (Kojima et al. 2002; Kageyama et al. 2003). In the first PCR, amplification was performed in a 50-μL reaction mixture containing 15 μL of cDNA, 25 pmol of both forward and reverse primers, and 25 μL of NebNext High Fidelity 2× PCR Master Mix (BioLab. Middlebury, CT, USA). In the second PCR, the 100 μL reaction mixture consisted of 2 μL of the first PCR product, 50 pmol of both forward and reverse primers, and 50 μL of NebNext High Fidelity 2× PCR Master Mix (BioLab). The first and second amplification conditions were: denaturation at 98°C for 30 s followed by 25 cycles at 98°C for 10 s, 50°C for 30 s, and 72°C for 30 s and a final extension step at 72°C for 30 s. PCR products were visualized using agarose gel electrophoresis. PCR products with an expected length were gel-purified using QIAquick Gel Extraction Kit (Qiagen, Valencia, CA, USA) following the manufacturer's protocol.

Pyrosequencing was performed using the GS Junior system (Roche Applied Science, Penzberg, Germany). Two unique adaptors were ligated to the 5' and 3' ends of the amplicons and fusion PCR was performed with the purified products. The fusion primers were described previously (Kazama et al. 2016). Forward primers included FLX Titanium Primer A (25-mer sequence used for the sequencing), Multiplex Identifier (10-mer sequence for barcoding each sample), and G1SKF or G2SKF sequences. Reverse primers included FLX Titanium Primer B (25-mer sequence used for the sequencing) and G1SKR or G2SKR sequences. The 10 μL volume of the purified nested-PCR products was subsequently added to 90 μL of a reaction mixture containing 50 μL of NebNext High Fidelity 2× PCR Master Mix (BioLab) and 50 pmol of both forward and reverse primers. PCR conditions were an initial denaturation at 98°C for 30 s; 5 amplification cycles of denaturation at 98°C for 10 s, annealing at 50°C for 30 s, and extension at 72°C for 30 s; and a final extension of 72°C for 30 s. The fusion PCR products were purified using a QIAquick PCR Purification Kit

(Qiagen) with QIAcube (Qiagen). Measurement involved the DNA concentration using the Quant-iT PicoGreen dsDNA Assay Kit (Thermo Fisher Scientific) with infinite M1000 PRO (TECAN, Mannedorf, Switzerland). Four to six samples with different Multiplex Identifier sequences were mixed for the individual pyrosequencing runs using the GS Junior system (Roche Applied Science) with the Titanium emPCR Kit (Lib-L) and the GS Junior Titanium Sequencing Kit (Roche Applied Science) according to the manufacturer's protocol (Kazama et al. 2016).

Bioinformatic analysis

The analysis was performed as previously described (Kazama et al. 2016). Briefly, QIIME 1.8.0 software (Caporaso et al. 2010) was used. The split_library.py software package with a minimum quality score parameter of 25 was used for quality filtering and primer sequence removal. The software package denoiser.py was used to correct sequences with incorrect nucleotides in the nested-PCR and/or pyrosequencing steps. After the split_library.py package was used to remove reverse primers, Perseus software (Quince et al. 2011) was applied to remove chimeric sequences. Sequences were then clustered into operational taxonomic units (OTUs) based on a minimum 97% similarity in nucleotide sequence using the pick_otus.py package, and the pick_rep_set.py package was applied to select a representative sequence of each OTU. Norovirus Genotyping Tool Version 1.0 (Kroneman et al. 2011) was applied to identify genotypes and variants of the representative sequences. If the genotyping tool was not able to assign, the sequence was recorded as “not assigned”. In each oyster and wastewater sample, a rarefaction curve created by the Analytic Rarefaction 2.0 software (http://strata.uga.edu/software/) was applied to evaluate the OTU diversity of the NoV strains (Kazama et al. 2016).

Phylogenetic analysis

To compare the nucleotide sequences obtained from oyster and wastewater samples, phylogenetic analysis was performed for NoV GII.2, GII.3, and GII.4 by MEGA7 (Kumar et al. 2016) using the neighbor-joining method after calculating genetic distance using the Kimura two-parameter gamma model for GII.2 and GII.4, and the Jukes–Cantor model for GII.3. A bootstrapping test was done with 1000 duplicates.

Nucleotide sequence accession numbers

Nucleotide sequence data from oyster and wastewater samples have been deposited in the DDBJ/EMBL/GenBank databases under accession number DRA005981.

Comparison with epidemic of gastroenteritis

To the best of our knowledge, there are no available data on NoV-related gastroenteritis in Hue City. We obtained the number of patients affected by diarrhea and gastroenteritis caused by unknown pathogens, including NoVs, recorded in the Hue Central Hospital from August 2015 to December 2016 (unpublished) and analyzed the correlation with the NoV concentration in oyster samples.

Statistical analyses

The difference between NoV concentrations in the flood season and dry season was tested by Wilcoxon signed-rank test. In addition, the correlation between NoV concentration in oyster samples and the number of diarrhea and gastroenteritis patients was tested by Spearman's Rank correlation. The difference between NoV GI and GII concentrations in wastewater samples was tested using the Student *t* test. $P < 0.05$ was regarded as significant. All statistical analyses were performed with R software version 3.1.2.

4.4 Results

NoV concentration in oyster samples

A total of 34 oyster samples were collected from the two stations during the 17-month study period. Each sample consisted of three composite samples. Thus, 102 composite samples were examined for NoV GI and GII by qPCR. The recovery rates of MNV for oyster samples ranged from 1.3-79% (geometrical mean: 10%, N=102; the average of standard deviation among the triplicates of extraction: 0.1), which allowed us to quantify NoV genomes in the oyster samples. Among the 34 samples extracted in triplicates, for GI, 13 samples presented positive or negative results in all three extractions, and 12 samples were positive in one extraction (Table 4.1).

Table 4.1 Variability of NoV detection in triplicate extractions from oyster samples

NoV	No. of samples with the following no. of positive extractions:				Total no. of samples
	3	2	1	0	
GI	6	9	12	7	34
GII	1	2	11	20	34

For GII, most of the samples (21) gave the same results for three extractions, followed by positive in one extraction and positive in two extractions. Of the 34 monthly-collected samples, NoV GI and GII were detected in 27 (79%) and 14 (41%) samples, respectively (Fig. 4.2). Co-contamination with NoV GI and GII was observed in 12 (35 %) samples. At station A, NoV GI and GII were identified in 76% and 53% of the samples, respectively. NoV GI (82%) was more frequently detected at station B compared to station A, while NoV GII (29%) was less frequently detected. There was the variability of NoV concentrations among some composite samples in the same month, and the larger variability was observed

mainly in the flood season (e.g. NoV GI in October 2015 presented from not detected to 2.4 × 10^5 copies/g digestive tissue).

NoV GI at the station A was continuously detected in oysters from August 2015 to April 2016, except in January 2016, with a maximum concentration up to 2.7 × 10^5 copies/g digestive tissue in October and November 2015 in the flood season. GI was not detected from May to July 2016 and then was detected continuously from August to December 2016. The maximum concentration of NoV GI at the station B was 2.1 × 10^4 copies/g digestive tissue in August 2015, decreased from September 2015 to February 2016, and continuously detected from May to December 2016 (Fig. 4.2A). NoV GII concentration at the station A was also higher between September 2015 and January 2016, except in October 2015, with a maximum concentration of 2.7 × 10^4 copies/g digestive tissue in November 2015, and then was not detected from May to September 2016. NoV GII was again detected in October 2016 and December 2016 (up to 2.3 x 10^4 copies/g digestive tissue). NoV GII at the station B was sporadically detected in four of the 17 months (Fig. 4.2B). During the 17-month study period, the NoV GII concentration at the station A was higher in the flood season than in the dry season (P = 0.04; Wilcoxon signed-rank test). The concentration of NoV GI was not significantly different between the two seasons (P = 0.28; Wilcoxon signed-rank test).

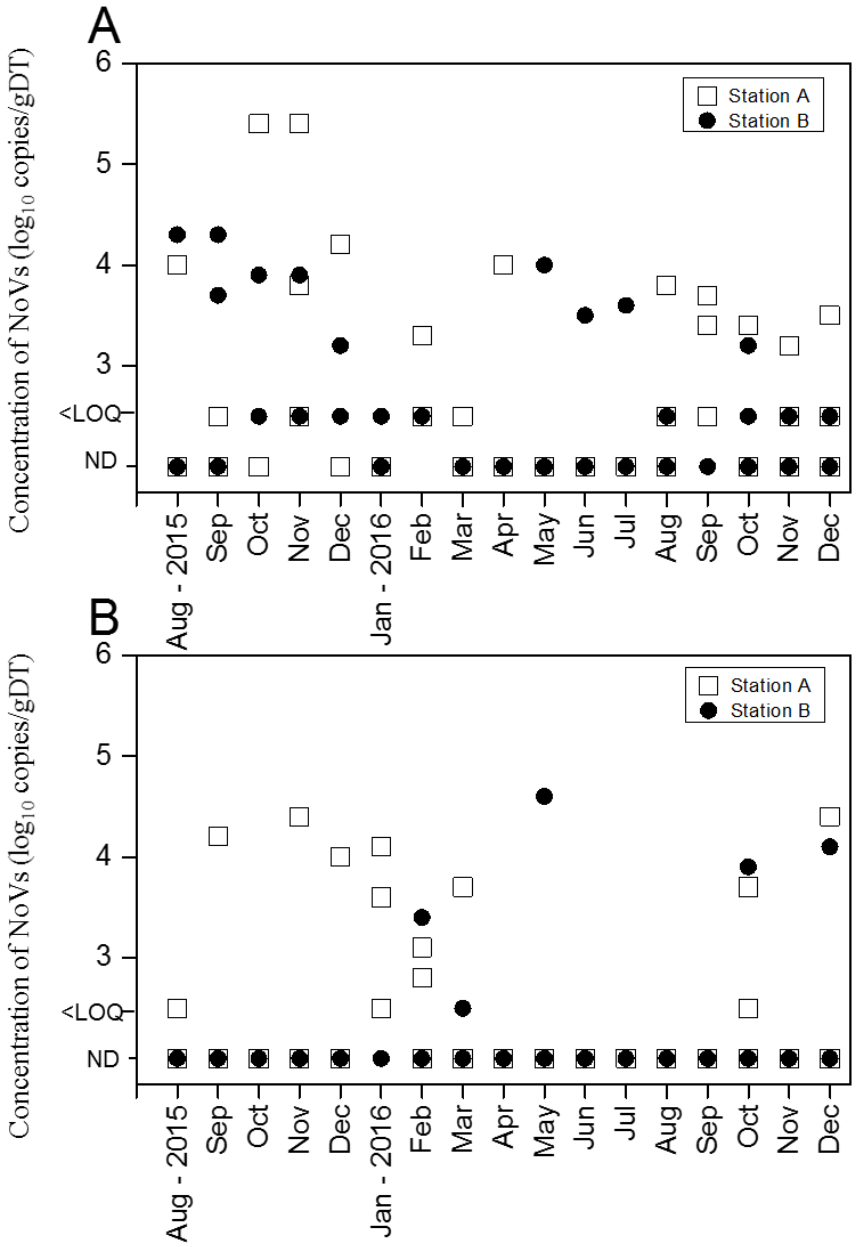


Fig. 4.2 Concentrations of NoV GI (A) and GII (B) in oyster samples. Three different composite samples were made and analyzed for each month. LOQ, limit of quantification; ND, not detected.

Comparison with gastroenteritis cases

No significant correlation was evident between the NoV concentration in oysters and the number of diarrhea cases caused by unknown pathogens reported in the largest hospital in Hue City during the study period (Spearman's correlation coefficient of 620, $P = 0.2$).

NoV concentration in wastewater samples

The recovery rates of MNV in the wastewater samples ranged from 1.5-24% (geometric mean: 5.6%, N=39). All RNA extracts from wastewater samples were inhibited and the results from ten-fold diluted extracts were used for quantification. The NoV genome was quantified in all 39 samples and the positive rate of NoV GI and GII was 87% and 95%, respectively. NoV GI was continuously detected from June to December 2016, with a maximum concentration of 5.6×10^2 copies/mL in July 2016 at the Le Loi site. The concentration of NoV GII ranged from 1.3×10^1 to 3.1×10^3 copies/mL. NoV GII was continuously detected from June to December 2016, with a maximum concentration of 3.1×10^3 copies/mL in July 2016 at the Bach Dang site **(**Fig. 4.3**)**. The concentration of NoV GII was significantly higher than NoV GI during June to December 2016 ($P = 0.006$).

NoV genotypes detected in oysters and wastewater

The genotype distribution of NoV strains was determined in seven wastewater and six oyster samples using pyrosequencing (Table 4.2). A total of 107,341 NoV sequence reads were obtained with mean reads of 8,300 and 13,740 per sample for NoV GI and GII, respectively. All rarefaction curves reached a plateau, except for NoV GI in the oyster sample at the station A in September. The results indicated that pyrosequencing runs were deep enough to evaluate the diversity of NoVs. All genotypes detected in the wastewater samples were also observed in the oyster samples (Table 4.2). In the wastewater samples, GI genotypes were detected once in September 2016. The rate of sequence reads was the

highest for GI.2 (72.9%), followed by GI.5, and GI.3. GII genotypes were detected from September, October, and December. GII.4 presented the highest sequence read rate in September (98.6%), but GII.2 was prevalent in October and December. As for oyster samples, GI.2 and GI.5 were detected in October with a rate of sequence reads of 84.3% and 14.3%, respectively (Table 4.2). GII.2, GII.3, and GII.4 were detected in three of four samples, and GII.3 presented the highest sequence read rate.

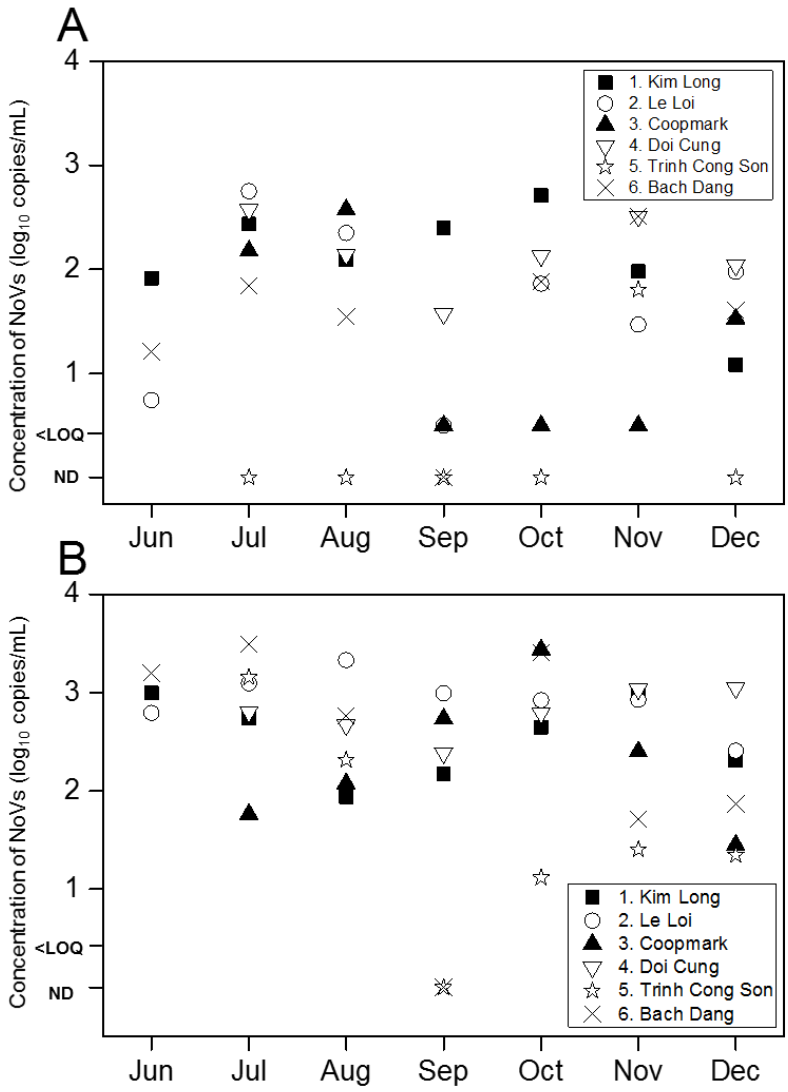


Fig.4.3 Concentrations of NoVs in wastewater samples collected in Hue City in 2016. (A: NoV GI; B: NoV GII). LOQ, limit of quantification; ND, not detected.

NoV GII.2, GII.3, and GII.4 sequences obtained from wastewater and oyster samples were phylogenetically compared with those available in GenBank (Fig. 4.4). For NoV GII.2 (nine strains, Fig. 4.4A), three strains in wastewater and three strains in oyster were grouped into a cluster of the GII.P16-GII.2/Marseille-E15061 strain (KY817510), which caused a gastroenteritis outbreak in France (Bidalot et al. 2017). The other three strains of NoV GII.2 in wastewater samples had nucleotide sequences similar to the 15-DS-4/2015/KT962983. Concerning NoV GII.3 (five strains, Fig. 4.4B), all wastewater sequences (two strains) and oyster sequences (three strains) were grouped into a cluster of a NoV strain found in Japan (Noshiro 2011/1/2011/JP/AB685707). Two distinct clusters of the NoV GII.4 sequences (11 strains, Fig. 4.4C) were identified. In cluster 1, the NoV strains in wastewater samples (three strains) and oyster samples (three strains) displayed a close relationship with Riviera 2008 in the United States (GQ413969). In cluster 2, NoV strains in the oyster samples (two strains) were grouped into a cluster of GII.4 Sydney 2012 strain (JX459908). The other three strains in the wastewater samples clustered into a distinct and separate branch from the GII.4 Sydney 2012 strain.

Table 4.2 NoV genotypes detected in wastewater and oysters in 2016[a]

Genotype	Wastewater							Oysters					
	Doi Cung	Le Loi	Kim Long	Doi Cung	Le Loi	Kim Long	Doi Cung	Station A				Station B	
	Sep		Oct	Nov		Dec		Sep	Oct	Nov	Dec	Oct	Dec
GI.2	72.9	–	ND	ND	–	–	ND	ND	84.3	ND	–	–	–
GI.3	4.6	–	ND	ND	–	–	ND	100	1.4	ND	–	–	–
GI.5	22.5	–	ND	ND	–	–	ND	ND	14.3	ND	–	–	–
GII.2	–	1.4	86.3	–	ND	99.4	–	–	39.0	–	33.3	ND	32.4
GII.3	–	ND	8.8	–	ND	0.2	–	–	58.0	–	64.8	ND	66.2
GII.4	–	98.6	4.9	–	ND	0.4	–	–	3.0	–	1.9	ND	1.3

[a] The numbers represent the rate (%) of sequence reads assigned to each genotype divided by the total number of reads in each genogroup.

ND, not detected in pyrosequencing; "–", not applied to nested-PCR and/or pyrosequencing.

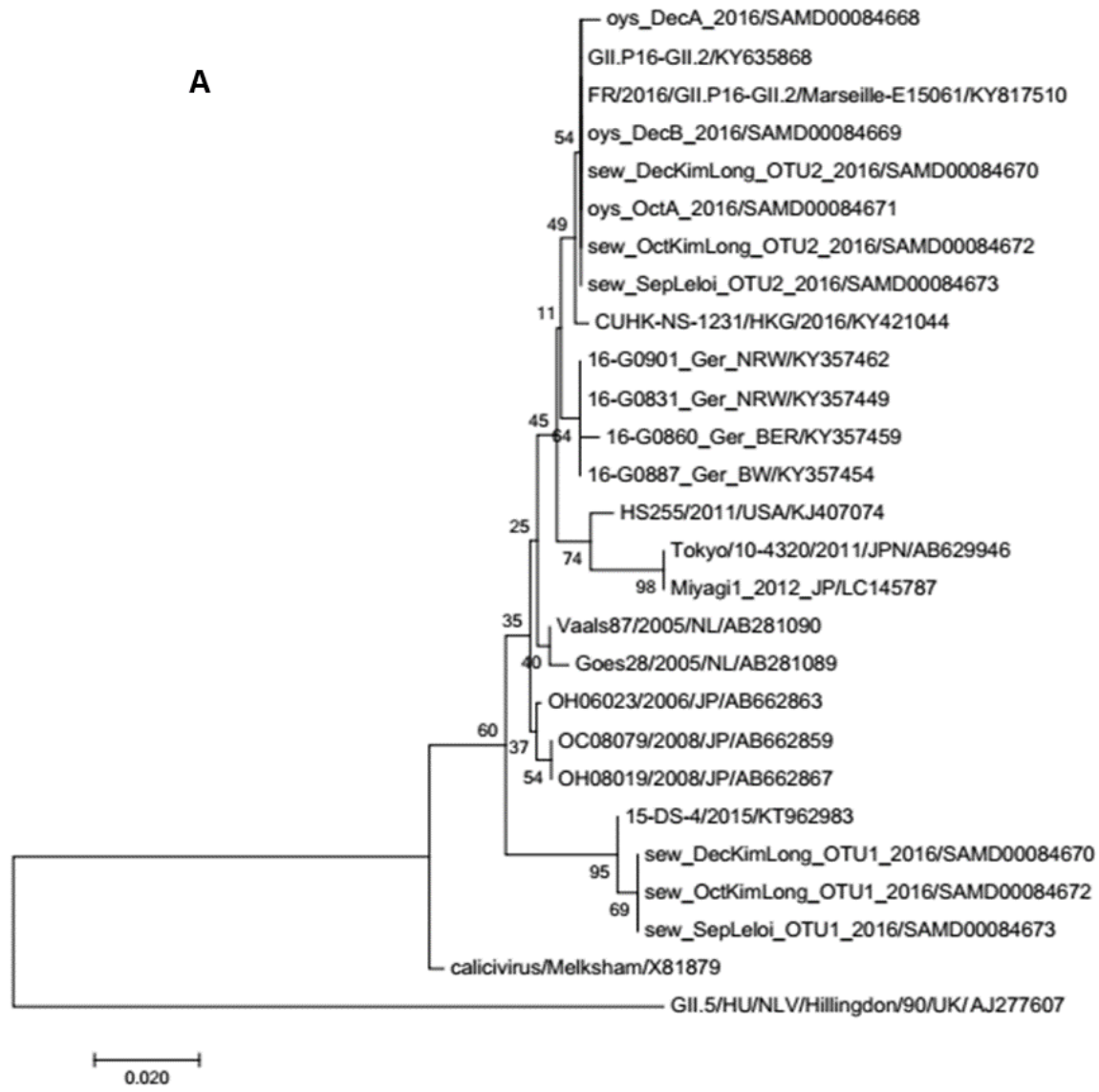
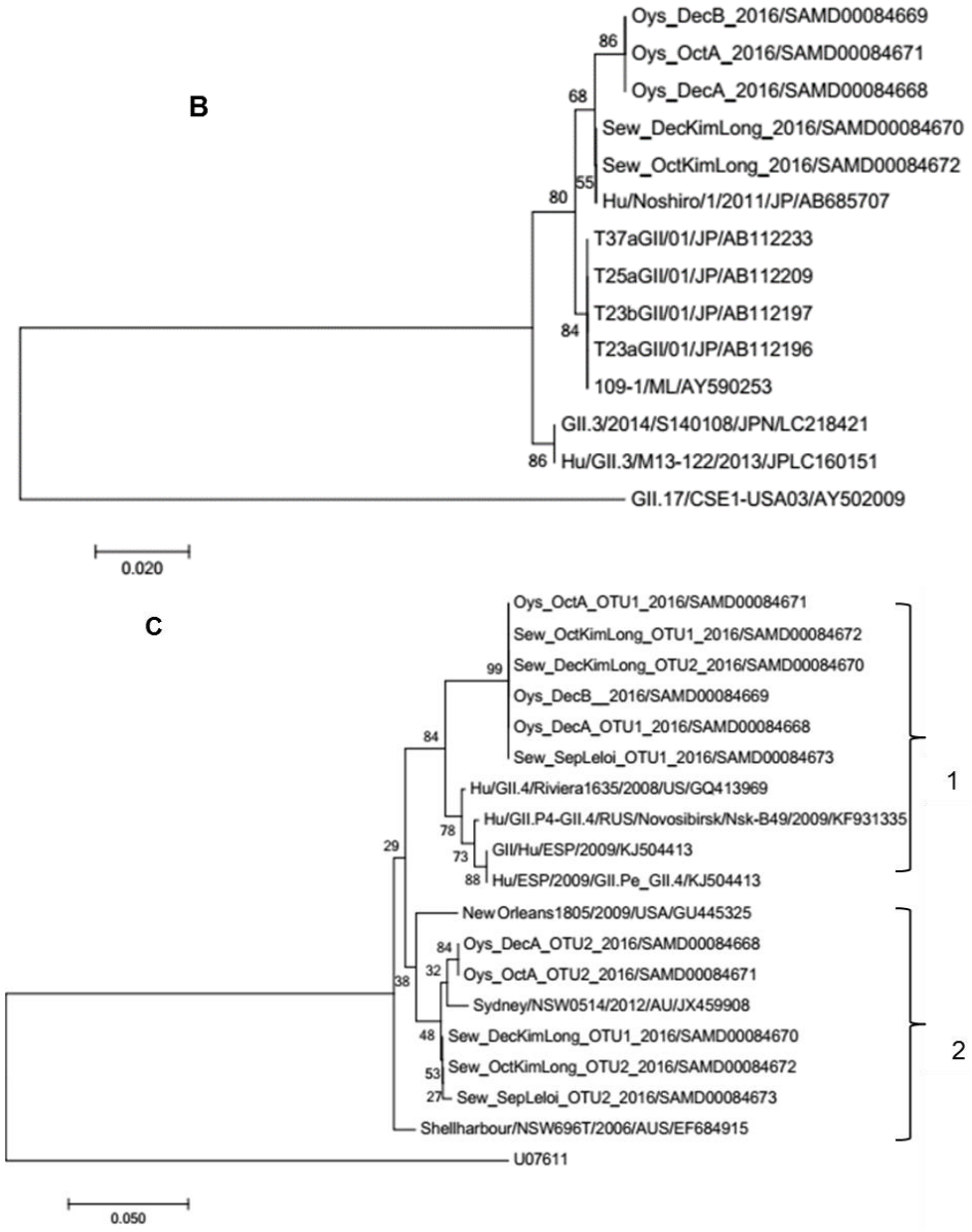


Fig. 4.4 Phylogenetic tree of GII.2 (A), GII.3 (B), and GII.4 (C) sequence detected in wastewater and oyster samples. The obtained sequences are designated with names starting sampling date (month), followed by station (A: station A, B: station B) or sampling sites of wastewater, then followed by "Oys" (oyster) or sewage. The reference sequences are designated with names starting the name of strain, year, country, and accession number.



4.5 Discussion

NoVs are the most common cause of foodborne gastroenteritis worldwide. Data from developed countries demonstrate that NoVs excreted from infected individuals can contaminate oysters cultivated downstream of residential areas (Ueki et al. 2005; Pu et al. 2016). In this study, NoV contamination in oysters was investigated in Hue City, Vietnam, to clarify the understanding of the level and sources of NoV contamination in developing countries. NoV GI and GII were detected in 79% and 41% of samples, respectively, with a maximum concentration of 2.4×10^5 and 2.3×10^4 copies/g digestive tissue, respectively. GII concentration in oyster samples at station A

was significantly higher in the flood season than in the dry season. GI concentration fluctuated regardless of the seasons. Six genotypes including GI.2, GI.3, GI.5, GII.2, GII.3, and GII.4 were identified in both wastewater and oyster samples. Genetically similar or identical sequences were obtained from the two types of samples. These observations suggest that urban drainage and seasonal flooding contribute to the oyster contamination with NoVs in the study area.

In this study, we found a significantly higher concentration of NoV GII in the flood season than in the dry season. Flooding water could bring NoVs from the urban area to the downstream area in which oysters were harvested. This led some individual oysters, depending on their own capacity, increasingly accumulated NoVs during flooding time more than the other time. This finding related to seasonal flooding is consistent with the results reported in some previous studies. Seasonal flooding was associated with transferring NoVs from wastewater to oysters (Astrom et al. 2009; Wang & Deng 2012; Grodzki et al. 2012), and a NoV outbreak due to direct exposure to sewage containing flood water (Schmid et al. 2005). The present and previous observations suggest that seasonal flooding can contribute to the contamination of oysters with NoVs. In contrast, we did not observe the clear seasonal peak during the winter months, as has been reported in developed countries located in the temperate zone (Flannery et al. 2012; Rajko-Nenow et al. 2012; Le Mennec et al. 2017). The maximum NoV concentrations in wastewater samples were 10^2 and 10^3 copies/mL for GI and GII every month from June to December, and the immune status of population against NoV infection may be different in the study area with a tropical monsoon climate (Czerkinsky and Holmgren 2009; Qadri et al. 2013).

The NoV concentration is consistent with those reported in some previous studies. In Ireland, Rajko-Nenow et al. (2013) reported that the concentration of NoVs in oysters ranged from 3.1×10^3 to 1.3×10^5 copies/g for GI and from 2.4×10^4 to 1.7×10^5 copies/g for GII. Lowther et al. (2012) found that the maximum concentration of NoV GI and GII in the United Kingdom was 1.6×10^4 copies/g and 1.8×10^4 copies/g, respectively. The positive rate of NoVs in the oyster samples presently collected in Vietnam (79% and 41% for GI and GII, respectively) was higher than commercial oysters reported in France (2% and 8% for GI and GII, respectively, Schaeffer et al. 2013), Australia (GI, not detected and GII, 1.7%, Brake et al. 2014), and higher than wild oysters reported in the United States (55%, Gentry et al. 2009). This may be because wild oyster samples were collected in the lagoon receiving untreated domestic wastewater from a large urban population of 350,000.

In the present study, NoV contamination in oysters at the two sampling sites showed a different trend. NoV GI was detected more frequently than NoV GII at the two stations, and the difference in the positive rate was more obvious at station B than at station A. This difference is probably due to the location of the sampling sites. Station A is located close to the mouth of the Perfume River, which flows through the center of Hue City. It is readily affected by drainage from the city. Station B is located at the center of lagoon and so might be less affected than station A. Another possible explanation includes the difference in viral particles between NoV GI and GII. Researchers have reported a higher stability of NoV GI in water (Lysen et al. 2009), a longer half-life of GI in oysters (Le Mennec et al. 2017), and the specific binding of GI strains to oyster digestive tissues, which leads to increased bioaccumulation (Le Guyader et al. 2006).

Although we tried to compare the NoV concentration in oysters with the number of diarrhea cases recorded in the largest hospital in the city during the study period (i.e. from August 2015 to December 2016), no significant correlation was observed. The possible reason for this inconsistency includes the virus transportation enhanced by flood. Floodwaters are able to flush drainage channels in the urban area and NoVs are transported downstream more than in drier seasons. This probably leads to the increased NoV concentration in oysters behind or regardless of the occurrence of NoV infections in the city. Another possible explanation is the characteristics of the epidemiological data. As the data were the sum of inpatient and outpatient diarrhea cases caused by unknown pathogens in the largest hospital in the city, severe diarrhea cases would be encountered. In contrast, NoVs cause a self-limited gastrointestinal illness and not all infected individuals see a specialist in a hospital.

Environmental surveillance of NoVs was conducted for the first time in Vietnam and the data presented in this study reveal the co-circulation of several NoV genotypes in Hue City. A total of six genotypes including GI.2, GI.3, GI.5, GII.2, GII.3, and GII.4 were detected in both wastewater and oyster samples. Detection of the same NoV genotypes in both wastewater and oyster samples was consistent with previous studies. For example, GI.5, GII.3, and GII.4 were detected in wastewater and oysters in Ireland (Rajko-Nenow et al. 2013), and GII.3 and GII.4 were found in Japan (Pu et al. 2016). In this study, GII.2, GII.3, and GII.4 strains found in wastewater and oyster samples clustered together, indicating a close relationship between the prevalent NoV strains in those samples. Although papers reporting clinical surveillance of NoVs are limited in Vietnam, diverse genotypes of NoVs (i.e., GI.3, 4, and 5, and GII.2, 3, 4, 6, 7, 9, 12, 13, and 17) have been identified. GII.4 is the predominant genotype in Vietnam as reported elsewhere (Trang et

al. 2012; My et al. 2013a, 2013b; Hoa-Tran et al. 2017). However, GII.2 strains predominated in the wastewater samples in October and December 2016. Although the polymerase genotype of NoV strains was not determined in this study, the obtained GII.2 sequences were closely related to the GII.P16-GII.2 variant isolated in outbreaks in France (Bidalot et al. 2017). The GII.P16-GII.2 variant was found in recent gastroenteritis cases in some countries including Japan (Motomura et al. 2016), Germany (Niendorf et al. 2017), and Thailand (Supadej et al. 2017). Our observations suggest that the GII.P16-GII.2 variant is spreading worldwide including Vietnamese water environment in the 2016-2017 epidemic season.

The present results have to be interpreted within the context of strengths and potential limitations. This is the first report about the simultaneous detection of NoVs in oysters and wastewater in Vietnam. NoV GI and GII in the oyster samples were investigated in a 17-month period. NoVs in the urban drainage of the Hue City were investigated from June to December 2016 as the most probable source of viruses detected in the oysters. Distribution of NoV genotypes in the oyster and wastewater samples was also analyzed using pyrosequencing. This may help us to understand the molecular epidemiology of NoVs in Hue City, a typical city in monsoon Asia affected by frequent seasonal flooding. We believe that our results will improve knowledge about the ecology of NoVs in urban settings and in places where seasonal flooding and/or failure in wastewater treatment have potential impact on food- or water-borne NoV gastroenteritis. We did not find a significant correlation between NoV concentration in oysters and the number of diarrhea cases recorded in the Hue Central Hospital. Further data collection is needed to clarify this relationship by determining the occurrence of NoV infections in humans. Moreover, our findings have important implications in food safety management and public health relevance, and represent an excellent initial step toward understanding the epidemiology of NoV infections in developing countries.

4.6 Evaluation of the infectious risk

In Vietnam, oysters are often consumed raw, made soups, fried, dried, or baked. Based on the questionnaire survey, the detail information related to consuming oysters in Thua Thien Hue province was shown in table 4.3.

Table 4.3 Consuming oysters among people in Thua Thien Hue province

Consuming oyster	Number	Percentage
Yes	56	18.3
No	250	81.7
If yes, type of oyster	(n=56)	
Raw oyster	1	1.8
Undercooked oyster	6	10.7
Cooked oyster	46	82.1
Undercooked and cooked oyster	3	5.4
Average of weight of DT of one piece oyster	0.15 g	-
Average of amount of oysters eaten at a time	5,1 pieces	-
Maximum of weight of DT of a piece oyster	0.41 g	-
Maximum of oysters eaten at a time	10 pieces	-

DT: Digestive tissue

The oysters were harvested at the two stations in this study. Normally, only oysters harvested in station B were for the commercial purposes. Risks of NoVs infection due to consuming oysters harvested at the station B ranged 36.3 % to 72.2 % (Table 4.4). Higher risk of infection was observed in the flooding season.

Table 4.4 Monthly risk of NoVs infection due to consuming raw oysters

	Risk of infection	
	P_{Mean}	P_{Max}
August 2015	0.722	0.722
September	0.722	0.722
October	0.719	0.722
November	0.717	0.722
December	0.473	0.720
January 2016	-	-
February	-	-
March	-	-
April	-	-
May	0.721	0.722
June	0.652	0.722
July	0.681	0.722
August	-	-
September	-	-
October	0.507	0.721
November	0.442	0.717
December	0.363	0.705

The present risks have to be interpreted within the context of potential limitations. The calculation based on the number copies of genes so that could include the inactive viruses in the oysters. In addition, consuming raw oysters in the study area were not so common. These lead to the risk of NoV infection in the population may be lower than our results.

4.7 Summary of this chapter

1) NoV GI and GII were detected in 79% and 41% of samples, respectively, with a maximum concentration of 2.4×10^5 and 2.3×10^4 copies/g digestive tissue, respectively. The positive rate was higher than in some developed countries.

2) GII concentration in oyster samples at station A was significantly higher in the flood season than in the dry season. The seasonal flood contributed to the contamination of oysters with NoVs.

3) A significant correlation was not observed between the NoV concentration in oysters and the number of diarrhea cases.

4) Six genotypes were identified in both wastewater and oyster samples, and GII.2, GII.3, and GII.4 strains found in samples clustered together indicating a close relationship between those samples.

5) The risk of NoV infection due to consuming the oysters was quite high (from 36.3% to 72.2%).

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CHAPTER 5
PSYCHOLOGICAL IMPACTS CAUSED BY FLOODS

5.1. Introduction

As mentioned in chapter 1 and 2, floods are potentially able to affect on psychology with respect to their indirect impacts. As an indicator of the psychological impacts, people's feeling to disease is important in the estimation of its burden, especially for common diseases with a low fatality. The objectives in this chapter are to test the hypothesis that facing floods frequently can reduce the burden of gastroenteritis by acceptance of this disease and to propose a new method to estimate the burden of gastroenteritis considering the feeling about the disease.

Southeast Asia is a region suffered from flood disasters regularly. It is estimated that 9.6 million people in this region were affected by the flooding (Bean, 2011). Vietnam and Cambodia are monsoon Asia countries affected seasonal flooding frequently. In Vietnam, a flooding event in 2016 caused 26 deaths, and 111851 houses have been flooded. And the most affected provinces were Thua Thien Hue, Quang Nam, Quang Ngai and Binh Dinh province (The United Nations in Vietnam, 2016). In Cambodia, from late September to October 2013, more than 200 people were killed, and over 1.5 million people have suffered flooding (International Federation of Red Cross and Red Crescent Societies, 2013). However, data on psychological impacts associated with floods in these countries are extremely limited. This study was carried out in Thua Thien Hue province, Vietnam, and three provinces in Cambodia including Kampong Chhnang, Kampong Thom, and Battambang.

5.2. Methods

Study design and setting

A cross-sectional study was conducted in two countries of Southeast Asia, including Vietnam and Cambodia. In Vietnam, Thua Thien Hue province (324 households) was selected. In Cambodia, Kampong Chhnang, Kampong Thom, and Battambang province (334 households) were selected. The study was conducted in August to September 2016 in Vietnam and September 2016 to May 2017 in Cambodia. These study sites consist of flood area and non-flood area.

In Vietnam, the annual precipitation in Thua Thien Hue province is 3200 mm and it comes mainly in the rainy season from September to December, which always triggers flooding. While in

Cambodia, the annual precipitation of the three provinces mentioned above averages about 1500 mm, and the wet season in these provinces is from May to October.

Participants

A convenience sampling method was used. People were recruited if they (1) were willing to participate in the study (2) living in the area less than 1 year; and (3) had to be at least 18 years old. People were excluded if they: (1) had serious physical or psychological damage; (2) refused the offer to join; and/or (3) did not have time to enroll in the study and complete the questionnaire.

Measurements and Instruments

Data were collected via face-to-face interviews, which were conducted by well-trained our team researchers who each had experience working on research related to infectious diseases, with supported by village health workers. We visited the households and invited the principal of the households or any other people to participate in the survey.

Outcomes

In order to quantify the feeling of people who facing flooding frequently to infectious diseases, we developed a questionnaire to collect information about participant's socio-demographic, flood experience and feeling about infectious diseases.

Socio-demographic measurements were including gender, age, and education level.

Flood experience were including their experiences with flooding in their house (yes, no).

Feeling about infectious diseases: we asked respondents about their feeling to infectious diseases/symptoms that happened more frequently during/after flooding. The diseases/symptom were classified into diarrhea, severe diarrhea (indicated by at least a symptom including bloody diarrhea, vomiting, high fever, persistent diarrhea – lasts 14 days or longer and/or dehydration, WHO), cough, fever, eye problem, skin problem, and other. We also asked the participants to compare their feeling between several diarrhea and non-severe diarrhea with some common diseases, which one more severe or uncomfortable was. The common diseases consisted of broken hand bones, gouty, low back pain, dental caries, pharyngitis, asthma, headache, insomnia (cases), alcohol dependence syndrome, hypertensive heart diseases, hepatitis B, diabetes, peptic ulcer, and dengue fever.

Ethical consideration

This study was approved by the Human Ethics Committee of Graduate School of Medicine and Faculty of Medicine, The University of Tokyo. Both paper and electronic data sets were stored securely. After clearly introducing the survey, we asked them to sign on a prepared consent form if they agreed to participate. Participants could refuse to participate or withdraw from the interview at any time.

Data analysis

To compare feeling about infectious diseases between people from the flood area and non-flood area, the data obtained in the survey was statistically analyzed using Chi-square test.

The multivariate logistic regression model was applied to investigate the factors associated with feeling diseases happened more frequently during/after flooding. Diseases, which people felt that happened more frequently during/after flooding, were considered the outcome variables.

Meanwhile, area (flooding/non-flooding), sex (male/female), age and education level (not yet/ primary school/secondary school/ high school/higher) were independent variables. We used Bayesian Model Average (BMA) for selecting the optimal model based on the lowest value of Bayesian Information Criterion (BIC) and the highest posterior model probability. The strength of association between the dependent variables and the independent variables were estimated using the odds ratio (OR) with 95% confidence interval.

Data analysis was performed in R version 3.1.2 and the Statistical Package for the Social Science (SPSS), version 19.0 (IBM, Armonk, NY, USA). Significance was set at $P < 0.05$.

Modified disability weight

A new method was proposed to incorporate the psychological impacts of floods into the burden of the gastroenteritis, which was estimated by DALYs, by means of modifying the disability weight.

We modified disability weight using the following Equations (Eq.)

1. $$Modified\ DW = Original\ DW \times \frac{more\%}{50\%} \quad \text{(Eq.5.1)}$$

2. $$Modified\ DW = Original\ DW \times \frac{more\%}{50\%} \times \frac{(100\% - less\%)}{50\%} \quad \text{(Eq.5.2)}$$

3. $$Modified\ DW = \sum_{k=1}^{n} \text{original DW} \times \frac{\text{same \%}}{\sum_{k=1}^{n}(\text{same\%})} \quad \text{(Eq.5.3)}$$

Where, n is number of common diseases

To prevent potential bias, if value of % more and % less of the comparison between non-severe diarrhea and severe diarrhea to the common diseases were less than 10% or more than 70%, they did not employ to calculate the modified DW of non-severe diarrhea and severe diarrhea.

5.3. Results

5.3.1 Socio-demographic Characteristics

This study included 285 male and 373 female aged 18 years and above, from two countries including Vietnam and Cambodia. In Vietnam, of 324 respondents, mean age was 46.1 (18-76), 41.3 % of them were male, almost 84% of participants were graduated from elementary, secondary and high school. The majority of religious of them was Buddhist (49.2%). As for Cambodia, of 334 respondents, mean age was 40.5 (18-80), 45.2% of them were male. The majority of education level was the primary school (45.5%) (Table 5.1).

Table 5.1. Socio-demographic characteristics of study participants

	Vietnam N= 324		**Cambodia N=334**	
	N	%	N	%
Gender				
Male	134	41.3	151	45.2
Female	190	58.7	183	54.8
Age				
Mean (range)	46.1 (18-76)		40.5 (18-80)	
Education level				
Not yet	8	2.5	70	21.0
Primary school	94	29.0	152	45.5
Secondary school	93	28.7	57	17.1
High School	84	25.9	41	12.2
Higher	45	13.9	14	4.2
Religious				
Buddhist	159	49.2	_	_
Christian	10	3.1	_	_
None	154	47.7	_	_

5.3.2 Flood experience

Table 5.2 shows the flood experience. A haft of Vietnamese participants was in flood area. Whereas participants in Cambodia having flood experience were 60.5%.

Table 5.2. Experienced flood in house within 10 years recently

Country	Flood experience	N	%
Vietnam (N=324)	Non flood area	163	51.3
	Flood area	161	49.7
Cambodia (N=334)	Non flooding area	202	60.5
	Flooding area	132	39.5

5.3.3 Feeling about infectious diseases

The participants in two countries felt that diarrhea, severe diarrhea, cough, fever, skin problem and eye problems happened more frequently during/after flooding (Table 5.3). In Vietnam, the highest frequency was diarrhea (49.1%), followed by some common diseases such as fever (32.7%), dengue fever (18.2%), skin problems (17.9%), and common cold (17.3%). In Cambodia, the highest common diseases were diarrhea (68.2 %), followed by fever, cough, skin problems, eye problems, and severe diarrhea.

Table 5.3. Feeling about diseases/symptoms that happen more frequently during/after flood

Diseases/symptoms	Vietnam (N= 324)		Cambodia (N=334)	
	N	%	N	%
Diarrhea	159	49.1	228	68.2
Severe diarrhea	22	6.8	55	16.5
Cough	50	15.4	185	55.4
Fever	106	32.7	212	63.5
Skin problems	58	17.9	103	30.8
Eye problem	20	6.2	91	27.2
Dengue fever	59	18.2	_	_
Common Cold	56	17.3	_	_
Pharyngitis	3	0.9	_	_
Headache	10	3.1	_	_
Malaria	6	1.9	_	_
Other	13	4.0		
Unknown	36	11.1	_	_

We asked participants from Vietnam and Cambodia to compare severe diarrhea in terms of severe/uncomfortable situation to 14 and 11 common diseases, respectively (Table 5.4). In Vietnam, more than 50% of them felt severe diarrhea was more severe/uncomfortable than dental caries, low back pain, insomnia, and diabetes. However, more than 50% of them felt that severe diarrhea was less severe/uncomfortable than 8 of 14 common diseases (i.e. broken hand bones, headache, pharyngitis, peptic ulcer, hepatitis B, gouty arthritis, hypertensive heart diseases, and asthma). In Cambodia, more than 50% of them felt that severe diarrhea was more severe/uncomfortable than dental caries, dengue fever, diabetes, and hypertensive heart diseases. Whereas more than 50% of them felt that severe diarrhea was less severe/uncomfortable than pharyngitis, hepatitis B, alcohol dependence syndrome, and asthma (Table 5.4).

In parallel, we also asked respondents in Vietnam and Cambodia to compare severe/uncomfortable situation of non-severe diarrhea to 14 and 11 common diseases, respectively (Table 5.5). In Vietnam, 50% of them feel that non-severe diarrhea was more severe/uncomfortable than dengue

fever, headache, pharyngitis and diabetes. Meanwhile, 50% of them felt that non-severe diarrhea was less severe/uncomfortable than all of the common diseases except dengue fever, headache, pharyngitis and diabetes. In Cambodia, 50% of them felt that non-severe diarrhea was more severe/uncomfortable than dental caries, dengue fever, hypertensive heart diseases and asthma. Whereas more than 50% of them felt that non-severe diarrhea was less severe/uncomfortable than hepatitis B, diabetes, and alcohol dependence syndrome (Table 5.5).

Table 5.4. Comparison feeling between severe-diarrhea to some common diseases in terms of severe or uncomfortable level.

Compared situation/ Severe diarrhea	Vietnam								Cambodia							
	Less		Same		More		Total		Less		Same		More		Total	
	n	%	n	%	n	%	n	%	n	%	n	%	n	%	n	%
Dental caries	71	22	16	5	235	73	322	100	19	21.3	24	27.0	46	51.7	89	100
Dengue fever	76	24.1	58	27.0	181	48.9	315	100	9	24.3	8	21.6	20	54.1	37	100
Broken hand bones	167	52.5	25	7.9	126	39.6	318	100	-	-	-	-	-	-	-	-
Headache	266	82.1	16	4.9	42	13.0	324	100	38	39.6	24	25.0	34	35.4	96	100
Low back pain	111	34.9	21	6.6	186	58.5	318	100	37	44.6	16	19.3	30	36.1	83	100
Insomnia (cases)	101	31.4	34	10.6	187	58.1	322	100	-	-	-	-	-	-	-	-
Pharyngitis	254	79.4	14	4.4	52	16.2	320	100	35	51.5	10	14.7	23	33.8	68	100
Peptic ulcer	201	65	34	11	74	23.9	309	100	-	-	-	-	-	-	-	-
Hepatitis B	195	71.2	17	6.2	62	22.6	274	100	3	50.0	1	16.7	2	33.3	6	100
Gouty Arthritis	175	62.1	19	6.7	88	31.2	282	100	9	26.5	9	26.5	16	47.0	34	100
Diabetes	66	22.1	21	7.0	212	70.9	299	100	1	25.0	0	0	3	75.0	4	100
Hypertensive heart diseases	187	59.9	31	9.9	94	30.1	312	100	5	29.4	3	17.6	9	53.0	17	100
Alcohol dependence syndrome	120	49	19	7.8	106	43.3	245	100	18	66.7	3	11.1	6	22.2	27	100
Asthma (cases)	173	57.7	29	9.7	98	32.7	300	100	6	66.7	2	22.2	1	11.1	9	100

Table 5.5. Comparison feeling between non-severe diarrhea to some common diseases in terms of severe or uncomfortable level.

	Vietnam								Cambodia							
Compared situation/ non-severe diarrhea	**Less**		**Same**		**More**		**Total**		**Less**		**Same**		**More**		**Total**	
	n	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**
Dental caries	218	68.1	30	9.4	72	22.5	320	100	54	24.1	36	16.1	134	59.8	224	100
Dengue fever	9	2.7	1	0.3	300	97.0	310	100	15	22.7	8	12.1	43	65.2	66	100
Broken hand bones	295	93.3	4	1.3	17	5.4	316	100	-	-	-	-	-	-	-	-
Headache	96	30.0	52	16.3	172	53.7	320	100	96	36.4	49	18.5	119	45.1	264	100
Low back pain	253	79.3	23	7.2	43	13.5	319	100	86	36.9	54	23.2	93	39.9	233	100
Insomnia (cases)	223	69.7	32	10	65	20.3	320	100	-	-	-	-	-	-	-	-
Pharyngitis	84	26.3	55	17.2	180	56.5	319	100	69	39.2	38	21.6	69	39.2	176	100
Peptic ulcer	289	93.5	5	1.6	15	4.9	309	100	-	-	-	-	-	-	-	-
Hepatitis B	254	92.7	4	1.5	16	5.8	274	100	10	66.7	1	6.6	4	26.7	15	100
Gouty Arthritis	259	92.2	6	2.1	16	5.7	281	100	42	38.5	21	19.3	46	42.2	109	100
Diabetes	15	5.1	5	1.7	276	93.2	296	100	5	55.6	0	0	4	44.4	9	100
Hypertensive heart diseases	287	92.3	4	1.3	20	6.4	311	100	8	20.0	7	17.5	25	62.5	40	100
Alcohol dependence syndrome	200	80.6	18	7.3	30	12.1	248	100	20	51.3	8	20.5	11	28.2	39	100
Asthma (cases)	271	90.3	9	3	20	6.7	300	100	9	25.7	5	14.3	21	60	35	100

5.3.4 Feeling to infectious diseases between people in flood area and non-flood area

The Vietnamese respondents felt diarrhea, severe diarrhea, cough, fever, skin problems, eye problems, dengue fever, common cold, pharyngitis, headache, and malaria happened more frequently during/after flooding. The feeling about diarrhea, cough, common cold, and headache of people living in the flooding areas was significantly different from that of people living in the non-flooding areas (Table 5.6). Meanwhile, in Cambodia, the participants felt that diarrhea, severe diarrhea, cough, fever, skin problems and eye problems happened more frequently during/after flooding. The feeling about all those diseases of the people living in the flooding areas, except eye problems, was significantly different from that of the people living in the non-flooding areas (Table 5.6).

Table 5.6. Feeling about diseases/symptoms that happen more frequently during/after flooding between people in flood area and non-flood area

	Vietnam (N= 324)				**Cambodia (N=334)**			
	Flooding area (N= 161)	Non flooding area (N= 163)	χ^2	***P***	Flooding area (N=132)	Non flooding area (N=202)	χ^2	***P***
Diarrhea	70 (43.5%)	89 (54.6%)	3.69	0.05	106 (80.3%)	122 (60.4%)	13.7	<0.001
Severe diarrhea	11 (6.8%)	11 (6.7%)	0	1	31 (23.5%)	24 (11.9%)	7.0	0.008
Cough	18 (11.2%)	32 (19.6%)	3.76	0.05	90 (68.2%)	95 (47.0%)	13.6	<0.001
Fever	47 (29.2%)	59 (36.2%)	1.45	0.23	103 (78.0%)	109 (54.0%)	18.9	<0.001
Skin problems	30 (18.6%)	28 (17.2%)	0.05	0.82	51 (38.6%)	52 (25.7%)	5.6	0.017
Eye problem	9 (5.6%)	11 (6.7%)	0.03	0.85	41 (31.1%)	50 (24.8%)	1.3	0.25

Dengue fever	32 (19.9%)	27 (16.6%)	0.60	0.44	-	-	-	-
Common Cold	42 (26.1%)	14 (8.6%)	17.34	<0.001	-	-	-	-
Pharyngitis	2 (1.2%)	1 (0.6%)	0.35	0.55	-	-	-	-
Headache	9 (5.6%)	1 (0.6%)	-	0.01	-	-	-	-
Malaria	2 (1.2%)	4 (2.5%)	-	0.68	-	-	-	-

In Vietnam, the respondents felt that common cold and headache happened more frequently during/after flooding, and the feeling about these diseases of the people living in the flood areas was significantly different from that of the people living in the non-flooding areas. More specially, the difference was only in the rural areas in cases of headache (Table 5.7).

When comparing feeling about severe/uncomfortable situation of severe-diarrhea to one of the common diseases, we pointed out that feeling of Vietnamese participants was not significantly different between people were living in flooding areas and non-flooding areas (Table 5.8). In the other side, feeling of Cambodian respondents was not significantly different between people living in the flood areas and the non-flooding areas when comparing their feeling of severe/uncomfortable level of severe-diarrhea to those of the common diseases, aparted from dental caries (P=0.02), headache (P=0.02), low back pain (P<0.001), and pharyngitis (P=0.01) (Table 5.8).

Similarly, comparison of feeling about the severe of non-severe diarrhea to the common diseases, we pointed out that feeling of Vietnamese and Cambodia respondents living in the flooding areas were not significantly different between from that of the people living in the non-flooding areas (Table 5.9).

Table 5.7 Feeling to infectious diseases between people in flood area and non-flood area by location in Vietnam

	Urban area			**Rural area**		
	Flooding area (n=89)	**Non-flooding area (n=75)**	***P***	**Flooding area (n=72)**	**Non-flooding area (n=88)**	***P***
Diarrhea	40 (44.9%)	47 (62.7%)	0.18	30 (41.7%)	42 (47.7%)	0.21
Severe diarrhea	7 (7.9%)	6 (8.0%)	1.0	4 (5.6%)	5 (5.7%)	1.0
Cough	10 (11.2%)	15 (20.0%)	0.31	8 (11.1%)	17 (19.3%)	0.14
Fever	25 (28.1%)	28 (37.3%)	0.57	22 (30.6%)	31 (35.2%)	0.35
Skin problems	14 (15.7%)	20 (26.7%)	0.25	16 (22.2%)	8 (9.1%)	0.07
Eye problem	8 (9.0%)	3 (4.0%)	0.24	1 (1.4%)	8 (9.1%)	0.06
Dengue fever	24 (27.0%)	12 (16.0%)	0.09	8 (11.1%)	15 (17.0%)	0.287
Common Cold	23 (25.8%)	4 (5.3%)	<0.01	19 (26.4%)	10 (11.4%)	0.014
Pharyngitis	1 (1.1%)	0 (0%)	0.36	1 (1.4%)	1 (1.1%)	1.0
Headache	3 (3.4%)	0 (0%)	0.11	6 (8.3%)	1 (1.1%)	0.046
Malaria	2 (2.2%)	4 (5.3%)	0.29	0 (0%)	0 (0%)	-

Table 5.8 Comparison feeling uncomfortable/severe of severe diarrhea to those of some common diseases by area

	Vietnam								Cambodia							
	Flooding area (n=161)			Non-flooding area (n=163)			χ^2	*P*	Flooding area (n=132)			Non-flooding area (n=202)			χ^2	*P*
	Less	Same	More	Less	Same	More			Less	Same	More	Less	Same	More		
Dental caries	39	8	112	32	8	123	1.16	0.56	10	14	23	9	10	23	9.4	0.02
Dengue fever	88	29	39	93	29	37	0.16	0.92	6	3	9	3	5	11	3.2	0.36
Broken hand bones	84	13	61	83	12	65	0.16	0.92	-	-	-	-	-	-	-	-
Headache	20	6	135	22	10	131	1.14	0.56	22	11	17	16	13	17	9.9	0.02
Low back pain	59	9	88	52	12	98	1.29	0.52	24	10	13	13	6	17	17.0	<0.001
Insomnia (cases)	50	16	94	51	18	93	0.12	0.94	-	-	-	-	-	-	-	-
Pharyngitis	27	4	128	25	10	126	2.65	0.27	19	8	8	16	2	15	11.3	0.01
Peptic ulcer	95	22	36	106	12	38	3.57	0.16	-	-	-	-	-	-	-	-
Hepatitis B	84	9	37	111	8	25	5.42	0.07	0	1	2	3	0	0	6.6	0.09
Gouty Arthritis	82	8	47	93	11	41	1.35	0.51	4	5	7	5	4	9	1.3	0.73
Diabetes	104	13	28	108	8	38	2.51	0.28	0	0	2	1	0	1	1.5	0.45
Hypertensive heart diseases	94	14	48	93	17	46	0.34	0.84	1	3	5	4	0	4	6.4	0.09
Alcohol dependence syndrome	58	10	60	62	9	46	1.54	0.46	10	1	3	8	2	3	2.4	0.49
Asthma (cases)	81	13	53	92	16	45	1.54	0.46	4	0	1	2	2	0	4.7	0.19

Table 5.9 Comparison feeling uncomfortable/severe of non-severe diarrhea to those of some common diseases by area

	Vietnam								**Cambodia**							
	Flooding area (n=161)			**Non-flooding area (n=163)**					**Flooding area (n=132)**			**Non-flooding area (n=202)**				
	Less	**Same**	**More**	**Less**	**Same**	**More**	χ^2	***P***	**Less**	**Same**	**More**	**Less**	**Same**	**More**	χ^2	***P***
Dental caries	101	13	43	117	17	29	4.31	0.11	21	16	48	33	20	86	1.6	0.66
Dengue fever	148	0	6	152	1	3	2.04	0.36	6	3	12	9	5	31	2.8	0.41
Broken hand bones	142	2	12	153	2	5	3.24	0.20	-	-	-	-	-	-	-	-
Headache	82	22	53	90	30	43	2.53	0.28	37	21	46	59	28	73	0.31	0.96
Low back pain	121	10	27	132	13	16	3.66	0.16	36	19	39	50	35	54	1.0	0.80
Insomnia (cases)	103	18	38	120	14	27	3.64	0.16	-	-	-	-	-	-	-	-
Pharyngitis	81	28	50	99	27	34	4.86	0.09	25	10	33	44	28	36	5.1	0.16
Peptic ulcer	137	4	11	152	1	4	5.77	0.06	-	-	-	-	-	-	-	-
Hepatitis B	116	3	10	138	1	6	2.98	0.22	3	0	4	7	1	0	7.1	0.07
Gouty Arthritis	124	2	11	135	4	5	3.21	0.20	17	8	14	25	13	32	1.9	0.59
Diabetes	135	2	8	141	3	7	0.28	0.87	1	0	3	4	0	1	2.9	0.23
Hypertensive heart diseases	143	2	11	144	2	9	0.20	0.90	2	3	10	6	4	15	0.75	0.86
Alcohol dependence syndrome	101	10	17	99	8	13	0.52	0.77	10	5	2	10	3	9	4.81	0.18
Asthma (cases)	131	4	12	140	5	8	1.09	0.58	4	2	4	5	3	17	3.9	0.26

In terms of modified disability weight (DW), the ranged scale of the DWs using the Eq.5.2 ranged 0 to 4 × original DW compared to range from 0 to 2 × original DW and 0 to 1 × original DW for the Eq.5.1 and Eq.5.3, respectively. We selected the Eq.5.2 as the formula to calculate the DW of severe diarrhea and non-severe diarrhea. The Eq.5.2 was employed is that it has a larger scale than the Eq.1 and Eq.5.3 with respect to the comparison them to the value of the original DW. The larger scale is likely to be a more potential representation of DW value. In Vietnam, the modified DW for non severe diarrhea and severe diarrhea were 0.024 and 0.096, respectively. In Cambodia, the modified DW for non-severe diarrhea and severe diarrhea were 0.065 and 0.079, respectively. The detail data were shown in table 5.10, table 5.11, table 5.12, and table 5.13. There was no significantly different between the DW of severe diarrhea and non-severe diarrhea in the flooding areas and the non-flooding areas.

Table 5.10 The modified disability weight of non-severe diarrhea in Vietnam

Diseases		**Modified DW for non-severe diarrhea in Vietnam**			
	Original DW	**Total**	**Flooding area**	**Non-flooding area**	***P***
Dental caries	0.081	0.023	0.032	0.016	0.110
Headache	0.029	0.016	0.019	0.014	0.280
Insomnia	0.100	0.025	0.033	0.017	0.160
Pharyngitis	0.070	0.030	0.043	0.023	0.090
Modified DW	-	0.024	0.032	0.018	-

Table 5.11 The modified disability weight of severe diarrhea in Vietnam

Diseases	Modified DW for severe diarrhea in Vietnam				
	Original DW	**Total**	**Flooding area**	**Non-flooding area**	***P***
Broken hand bones	0.1	0.075	0.072	0.078	0.92
Low back pain	0.12	0.184	0.17	0.199	0.52
Insomnia	0.1	0.159	0.162	0.157	0.94
Peptic ulcer	0.003	0.001	0.001	0.001	0.16
Gouty Arthritis	0.13	0.062	0.072	0.072	0.51
Hypertensive heart diseases	0.25	0.119	0.120	0.117	0.84
Alcohol dependence syndrome	0.18	0.159	0.185	0.133	0.46
Dengue fever	0.2	0.081	0.086	0.076	0.92
Asthma	0.04	0.024	0.028	0.02	0.46
Modified DW	-	0.096	0.099	0.095	-

Table 5.12 The modified disability weight of non-severe diarrhea in Cambodia

Diseases	Original DW	Modified DW for non-severe diarrhea in Cambodia			
		Total	Flooding area	Non-flooding area	*P*
Dental caries	0.08	0.027	0.035	0.029	0.66
Low back pain	0.12	0.093	0.108	0.107	0.80
Headache	0.03	0.020	0.023	0.023	0.96
Pharyngitis	0.07	0.060	0.053	0.076	0.16
Diabetes	0.02	0.017	0.004	0.038	0.23
Hepatitis B	0.08	0.114	0.055	0.263	0.07
Gouty Arthritis	0.13	0.103	0.148	0.102	0.59
Hypertensive heart diseases	0.25	0.064	0.044	0.094	0.86
Alcohol dependence syndrome	0.18	0.158	0.373	0.194	0.18
Dengue fever	0.2	0.046	0.097	0.049	0.41
Asthma	0.04	0.015	0.041	0.011	0.26
Modified DW	-	0.065	0.089	0.090	

Table 5.13 The modified disability weight of severe diarrhea in Cambodia

Diseases	Original DW	Modified DW for severe diarrhea in Cambodia			
		Total	Flooding area	Non-flooding area	*P*
Dental caries	0.08	0.027	0.035	0.019	0.02
Low back pain	0.12	0.101	0.179	0.054	<0.001
Headache	0.03	0.021	0.034	0.010	0.02
Pharyngitis	0.07	0.065	0.117	0.028	0.01
Hepatitis B	0.075	0.060	0.000	0.024	0.09
Gouty Arthritis	0.13	0.052	0.074	0.028	0.73
Hypertensive heart diseases	0.25	0.068	0.048	0.000	0.09
Alcohol dependence syndrome	0.18	0.235	0.404	0.000	0.49
Dengue fever	0.2	0.084	0.131	0.072	0.36
Modified DW	-	0.079	0.114	0.026	

The relationship between the risk factors and the feeling of diseases happened more frequently during and after flooding was shown in table 5.14. In Vietnam, factors associated with the feeling non-diseases, severe-diarrhea, and eye problems happened more frequently during/after flooding was education. And age was a factor associated with feeling that non-severe diarrhea and skin problem happened more frequently during/after flooding. The risk of skin problems increased with age. In Cambodia, area was a factor associated with the feeling that non-severe diarrhea, severe diarrhea, cough, fever, skin problems happened more frequently during/ after flood.

Table 5.14 Factors associated with feeling of diseases happened more frequently during/after flooding: Results of multivariable logistic regression analysis in Vietnam and Cambodia.

Diseases	Variable		Vietnam		Cambodia	
			OR (95% CI)	***P* (Wald's test)**	**OR (95% CI)**	***P* (Wald's test)**
Non-severe diarrhea	Age	18-29	Ref.		-	-
		30-39	5.2(1.3-20.6)	0.018	-	-
		40-49	2.0 (0.5-7.4)	0.295	-	-
		50-59	3.4 (1.0-11.5)	0.051	-	-
		60-69	1.5 (0.4-5.3)	0.520	-	-
		>69	2.2 (0.6-8.1)	0.228	-	-
	Education	Not yet	Ref.		-	-
		Primary school	20.9 (2.0-220)	0.012	-	-
		Secondary school	4.1(1.6-10.7)	0.003	-	-
		High school	4.2(1.7-10.6)	0.002	-	-
		Higher	1.6 (0.7-3.9)	0.263	-	-
	Area	Non-flooding	-	-	Ref.	
		Flooding	-	-	2.7 (1.6-4.5)	<0.001
Severe diarrhea	Education level	Not yet	Ref.		-	-
		Primary school	3.5 (0.0-.)	0.999	-	-
		Secondary	4.4 (1.2-15.5)	0.022	-	-

		school				
		High school	5.5 (1.4-21.9)	0.016	-	-
		Higher	5.4 (1.4-21.6)	0.017	-	-
	Area	Non-flooding	-	-	Ref.	
		Flooding	-	-	2.3 (1.3-4.1)	0.006
Cough	Area	Non-flooding	-	-	Ref.	
		Flooding	-	-	2.4 (1.5-3.8)	<0.001
Fever	Area	Non-flooding	-	-	Ref.	
		Flooding	-	-	3.0 (1.8-5.0)	<0.001
Skin problems	Age	18-29	Ref.			
		30-39	0.6 (0.179-2.3)	0.505	-	-
		40-49	1.5 (0.4-6.0)	0.535	-	-
		50-59	1.3 (0.4-4.5)	0.718	-	-
		60-69	3.4 (0.8-14.0)	0.092	-	-
		>69	3.5 (0.8-16.5)	0.107	-	-
	Area	Non-flooding	-	-	Ref.	
		Flooding	-	-	1.8 (1.1-2.9)	0.013
Eye problem	Education	Not yet	Ref.		-	-
		Primary school	0.8 (0.1-7.8)	0.824	-	-
		Secondary school	6.4 (1.2-33.1)	0.027	-	-

	High school	3.9 (0.9-16.4)	0.064	-	-
	Higher	1.3 (0.4-4.1)	0.606	-	-

Note: BMA Model selections by area (flooding/non-flooding), sex, age, and education.

To estimate NoVs in cases of gastroenteritis burden, disability-adjusted life years (DALYs) were calculated based on years of life lost (YLL) and years lived with disability (YLD). The calculation of DALY as follows.

DALY = YLL + YLD (WHO 2017) (Eq.5.4)

YLL = R × N × L1 (Eq.5.5)

YLD = R x modified DW × L2 (Eq.5.6)

In where,

R: Risk of NoV infection; N: number of deaths; L1: standard life expectancy at age of death (in years); L2: average duration of disability (years).

Assumptions for calculating DALY. In this study, cases of NoV infection were including mild and severe cases. The durarion of NoV infection of mild cases and severe cases were 2.1 and 7.2 days, respectively. The number of deaths was 0.8 per 100.000 cases (Gibney et al. 2014). And the assumption of the standard life expectancy at age of death was 50.

The burden of NoV infection of mild cases and severe cases in the population ranged from 0.05 to 0.1 DALY/1000 persons and 0.317 to 0.631 DALY/1000 persons, respectively. The detail data were shown in the table 5.15.

Table 5.15. Estimating the burden of NoV infection in the population

	Risk of infection		**Modified YLD**							**Modified YLL**				**Modified DALY per 1000 persons**		**Modified DALY per 1000 person**	
	P_{Mean}	$P_{Maximum}$	DW	L2 (2.1 days)	YLD mean	YLD Max	L2 (7.2 days)	YLD mean	YLD Max	N (0.8/ 100000 cases)	L1	YLL mean	YLL max	DALY Mean	DALY Max	DALY Mean	DALY Max
				Mild cases			Severe case			Severe case				Mild cases		Severe cases	
Aug 2015	0.722	0.722	0.024	0.006	0.0001	0.0001	0.0197	0.0003	0.0003	0.00001	50	0.00029	0.00029	0.100	0.100	0.631	0.631
Sep	0.722	0.722	0.024	0.006	0.0001	0.0001	0.0197	0.0003	0.0003	0.00001	50	0.00029	0.00029	0.100	0.100	0.631	0.631
Oct	0.719	0.722	0.024	0.006	0.0001	0.0001	0.0197	0.0003	0.0003	0.00001	50	0.00029	0.00029	0.099	0.100	0.628	0.631
Nov	0.717	0.722	0.024	0.006	0.0001	0.0001	0.0197	0.0003	0.0003	0.00001	50	0.00029	0.00029	0.099	0.100	0.626	0.631
Dec	0.473	0.720	0.024	0.006	0.0001	0.0001	0.0197	0.0002	0.0003	0.00001	50	0.00019	0.00029	0.065	0.099	0.413	0.629
Jan 2016	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Feb	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Mar	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Apr	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
May	0.721	0.722	0.024	0.006	0.0001	0.0001	0.0197	0.0003	0.0003	0.00001	50	0.00029	0.00029	0.100	0.100	0.630	0.631
Jun	0.652	0.722	0.024	0.006	0.0001	0.0001	0.0197	0.0003	0.0003	0.00001	50	0.00026	0.00029	0.090	0.100	0.569	0.631
Jul	0.681	0.722	0.024	0.006	0.0001	0.0001	0.0197	0.0003	0.0003	0.00001	50	0.00027	0.00029	0.094	0.100	0.595	0.631
Aug	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Sep	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Oct	0.507	0.721	0.024	0.006	0.0001	0.0001	0.0197	0.0002	0.0003	0.00001	50	0.00020	0.00029	0.070	0.100	0.443	0.630
Nov	0.442	0.717	0.024	0.006	0.0001	0.0001	0.0197	0.0002	0.0003	0.00001	50	0.00018	0.00029	0.061	0.099	0.386	0.626
Dec	0.363	0.705	0.024	0.006	0.0001	0.0001	0.0197	0.0002	0.0003	0.00001	50	0.00015	0.00028	0.050	0.097	0.317	0.616

5.4. Discussion

While it has been well established that flooding events have the capacity to impact on mental health such as anxiety, stress (Carroll et al. 2010; Stanke et al. 2012), it has not been clear whether people in developing countries hit flooding frequently can reduce their psychological stress caused by flood-infectious risk of infection. In this study, the feeling of people living in the flood-affected areas to infectious diseases was quantified and compared it to that of people living in the non-flooded areas to better understand the indirect impacts of floods on human health. We pointed out the participants felt that diarrhea, severe diarrhea, cough, fever, skin problems, and eye problems happened more frequently during/after floods. In Vietnam, the factors associated with the feeling about diseases happened more frequently during and after flood, were age and education. In Cambodia, area was associated with the feeling.

Our results were consistent with some previous studies. Impacts of floods were reported not only in physical health but also in mental health (Few et al. 2004; Ahern et al. 2005; Du et al. 2010; Paranjothy et al. 2011; Tempark et al. 2013; Hetherington et al. 2017). A higher incidence of several diseases including dengue fever, pink eye, dermatitis, and psychological problems (stress, nervous, anxious, sleeplessness, etc.) was reported in places affected by flooding compared those non affected flooding (Bich et al. 2011). Psychological distress increased with people living in the flooding areas compared to those who did not (Reacher et al. 2004).

Age and education were associated with the adverse impact of floods on psychological health (Ginexi et al. 2000; Collins et al. 2013). The present and previous studies suggest that age and education were risk factors related to associated-flood psychological problems. And people living flood area had suffered from psychological problems.

Our finding indicated that feeling of people living in flooding areas was affected by some infectious diseases. And these feelings were as same as to those in non-flooding areas to infectious diseases that more frequently happened during and after flooding, and this was also reported as same as in the two countries. However, two different trends were observed in two countries. In Vietnam, people living in non-flooding areas feeling that diarrhea and cough happened more frequently during and after

flooding was significantly higher than those in flooding areas. This possible that people hit by flooding frequently were more resistant to the common infectious diseases that frequently occurred in their life. In the contrary, in Cambodia, people living in the flooding areas feeling those diseases more happen during and after flooding, significantly higher than that of people living in the non-flooding areas. The different trends of the feeling about diseases happened more frequently during/after flood were possible explained by the difference of the magnitude and frequency of flood events that they were impacted, as well as the culture of each country. The perceived threat of infectious disease exerts associated with attitudes and behavior (Murray et al. 2012). A number analysis about community resilience to natural disasters as flooding should be conducted to more deeply understand its issue.

The present results have to be interpreted within the context of strengths and potential limitations. This is the first report on the feeling of people living in flooding areas and non-flooding areas in two monsoons Asia countries (Vietnam and Cambodia) affected by seasonal flooding frequently. The data were based on the survey using the questionnaire developed by our team. However, this study has some potential weaknesses. The data on the feeling of participants were collected based on their own experiences. This possible caused by memory recall errors. The study was designed as a cross-sectional study, and it is possible to make any causal inference on the relationship between the feeling of people and area. Convenience samples cannot be considered representative of all people who were facing flooding frequently in Vietnam and Cambodia. Nevertheless, our results provide people's feeling to infectious diseases in flooding area and non-flooding area to estimate the potential burden of mental health caused by flooding events. This may help us understand the feeling about infectious diseases of people in monsoon Asia affected by seasonal flood frequently, which has not been well known so far.

In conclusion, our findings suggest that among those living in the flooding areas, floods can change their feelings to gastroenteritis. Further studies on the population should be conducted to deeply estimate the associated-flood psychological problems. The local governments need to develop strategies to minimize and prevent the impacts of floods, more support to people who are impacted flood frequently with considering psychological impacts could contribute to reduction of adverse impacts of flood.

5.5 Summary of this chapter

1. The participants felt that diarrhea, severe diarrhea, cough, fever, skin problems, and eye problems happened more frequently during/after flooding.
2. The feeling about all those diseases of the people living in the flooding areas, apart from eye problems, was significantly different from that of the people living in non-flooding areas in Cambodia ($P < 0.05$). The same results were observed in Vietnam for diarrhea and cough.
3. In Vietnam, factors associated with the feeling about diseases that happened more frequently during and after flood, were age and education.
4. The feeling to diarrhea diseases was not affected by flood
5. Regardless of flood experience, the burden of NoVs in cases of gastroenteritis for mild cases and severe cases in Hue city were from 0.05 to 0.1 DALY/1000 persons, and 0.317 to 0.631 DALY/1000 persons, respectively.

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CHAPTER 6

CONCLUSIONS

6.1 Conclusions

Flood is one of the most common natural disasters to affect human life around the world. Floods not only threaten people's lives, but they also bring additional risks for diseases. The variation of flood frequency due to climate change makes more and more challenges to prevention of our society from its direct and indirect impacts. This study was carried out to identify the indirect impacts of floods on the human health in physical and psychological aspects. Our main findings are shown as follows.

1) The estimated prevalence of NoVs in cases of acute gastroenteritis was 17% in developing countries. This prevalence was comparable to that in developed countries, rejecting my hypothesis. This may be due to the poor surveillance systems, and the tendency of people in developing countries not to use medical services for mild diseases such as gastroenteritis caused by NoVs.

2) Monitoring NoVs concentration in oysters was implemented to identify the epidemic of gastroenteritis in Hue City, Vietnam according to the successful trials in developed countries. NoV GI and GII were detected in 79% and 41% of oyster samples, respectively, with a maximum concentration of 2.4×10^5 and 2.3×10^4 copies/g digestive tissue, respectively. NoV GII concentration in oyster samples at the station located close to the river mouth, was significantly higher in the flood season than in the dry season. NoV GI concentration fluctuated regardless of the seasons. Six genotypes of NoVs including GI.2, GI.3, GI.5, GII.2, GII.3, and GII.4 were identified in both wastewater and oyster samples, and genetically similar or identical sequences were obtained from the two types of samples. These observations suggested that urban drainage surely contribute to the oyster contamination with NoVs in the study area. On the other hand, there was no significant correlation between NoV concentration in oysters and the number of diarrhea cases recorded in the largest hospital in the city during the study period. This is due to poor survellence system in Vietnam as well as virus transportation enhanced by flood.

3) The risk of NoVs infection due to consuming oysters harvested in the lagoon was from 36.3% to 72.2%. A risk of NoVs infection due to consuming oysters contaminated with NoVs induced by the flood-water mixed with domestic wastewater from Hue City was noticeable.

4) In terms of indirect impacts of flood on psychology, the participants felt that diarrhea, severe diarrhea, cough, fever, skin problems, and eye problems happened more frequently during/after flooding in both Vietnam and Cambodia. The feeling about all those diseases, except eye problems, of the people living in the flooding areas was significantly different from that of the people living in the non-flooding areas in Cambodia. The same results were observed in Vietnam for diarrhea and cough. In Vietnam, factors associated with the feeling about diseases that happened more frequently during and after flood, were age and education. People facing floods frequently could not reduce the burden of gastroenteritis by acceptance of this disease.

5) According to this finding, a new method was proposed to incorporate psychological impact of floods into the burden of the gastroenteritis, which was estimated by DALYs, by means of modifying the disability weight. The burden of the gastroenteritis for mild cases and severe cases in Hue City were from 0.05 to 0.1 DALY/1000 persons and 0.317 to 0.631 DALY/1000 persons, respectively. These diseases burdens were lower than those calculated using the original disability weight. This is not due to the impact of flood, since there was no significant difference in the estimated burden among those in flooded and non-flooded areas, but possibly to less impact of people living in developing countries as a result of more frequent occurrence of gastroenteritis than in developed countries.

6.2 Recommendations

The following recommendations are proposed based on our findings and conclusions in this dissertation.

1. Strengthening communication and knowledge regarding of food safety especially during the seasonal flooding may help to prevent future NoVs-associated infection outbreaks.
2. More supports to those who are frequently affected by floods considering psychological impacts could contribute to reduction of adverse impacts of floods.

3. Further studies are needed to clarify the relationship between the epidemic of NoVs in cases of gastroenteritis and NoVs concentration in oysters in the study site, considering some potential factors such as water temperature, salinity, flow direction, etc.

4. Further studies should be conducted to understand more deeply the feeling of people living in flood-affected areas about infectious diseases by application of cohort study.

5. Modification of disability weight for common diseases that happen more frequently in flooding areas should be continued.

APPENDIX 1
APPENDIX FOR CHAPTER 3
Search strategy

Two electronic searches were conducted to find relevant studies. Initial searches were done using PubMed (31.3.2016) using the broad search term: "((((((((((((((((((Noroviruses AND Humans[Mesh])) OR (Norwalk-like Viruses AND Humans[Mesh])) OR (Norwalk like Viruses AND Humans[Mesh])) OR (Small Round-Structured Viruses AND Humans[Mesh])) OR (Round-Structured Viruses, Small AND Humans[Mesh])) OR (Small Round Structured Viruses AND Humans[Mesh])) OR (Small Round Structured Viruses AND Humans[Mesh])) OR (Norovirus* AND Humans[Mesh])) OR (Norovirus* [TIAB] AND Humans[Mesh])) OR (Norwalk-like Virus* AND Humans[Mesh])) OR (Norwalk-like Virus* [TIAB] AND Humans[Mesh])) OR (Norwalk like Virus* AND Humans[Mesh])) OR (Norwalk like Virus* [TIAB] AND Humans[Mesh])) OR (Small Round-Structured Virus* AND Humans[Mesh])) OR (Small Round-Structured Virus*[TIAB] AND Humans[Mesh])) OR (Round-Structured Virus*, Small AND Humans[Mesh])) OR (Round-Structured Virus*, Small [TIAB] AND Humans[Mesh])) OR (Small Round Structured Virus* AND Humans[Mesh])) OR (Small Round Structured Virus* [TIAB] AND Humans[Mesh]) AND (("1990/01/01"[PDat] : "2016/03/31"[PDat]) AND Humans[Mesh])".

No	Searches	Results
PubMed		
1	Noroviruses Filters: Humans	3346
2	Norwalk-like Viruses Filters: Humans	3351
3	Norwalk like Viruses Filters: Humans	3351
4	Small Round-Structured Viruses Filters: Humans	3331
5	Round-Structured Viruses, Small Filters: Humans	3331
6	Small Round Structured Viruses Filters: Humans	3331
7	Norovirus* Filters: Humans	2818
8	Norovirus* [TIAB] Filters: Humans	2626
9	Norwalk-like Virus* Filters: Humans	318
10	Norwalk-like Virus* [TIAB] Filters: Humans	318
11	Norwalk like Virus* Filters: Humans	318
12	Norwalk like Virus* [TIAB] Filters: Humans	318
13	Small Round-Structured Virus* Filters: Humans	173
14	Small Round-Structured Virus*[TIAB] Filters: Humans	168
15	Round-Structured Virus*, Small Filters: Humans	178
16	Round-Structured Virus*, Small	178

	[TIAB] Filters: Humans	
17	Small Round Structured Virus* Filters: Humans	173
18	Small Round Structured Virus* [TIAB] Filters: Humans	168
19	OR/1-18	3498
20	Limit 19 to 1990/01/01 to 2016/03/31	3356

For Web of Science, the used search term was "(Noroviruses) OR (Norwalk-like Viruses) OR (Norwalk like Viruses) OR (Small Round-Structured Viruses) OR (Round-Structured Viruses, Small) OR (Small Round Structured Viruses) OR (Norovirus*) OR (Norwalk-like Virus*) OR (Norwalk like Virus*) OR (Small Round-Structured Virus*, Small) OR (Small Round Structured Virus*)".

No.	Author	Year of Public	Tittle	Journal	Start Date	End Date	Country	Country classify	No. Tested	No.NoV +	GI	GII	GIV	Mix GI and GII	No.Control tested	No.Control NoV +	Age	Setting	Involve pandemic period (1=yes, 0=no)
1	Taylor Wolfaardt et al	1997	Incidence of human calicivirus and rotavirus infection in patients with gastroenteritis in South Africa	J Med Virol	1991/10/01	1995/10/01	South Africa	Upper middle income	1296	32	NA	NA	NA	NA	0	0	Mixed	inpatient	0
2	T.;Jiang Farkas et al	2000	Prevalence and genetic diversity of human caliciviruses (HuCVs) in Mexican children	J Med Virol	1-89	1-91	Mexico	Upper middle income	115	9	NA	NA	NA	NA	66	3	< 5	community	0
3	B. A.;Subekti Oyofo et al	2002	Enteropathogens associated with acute diarrhea in community and hospital patients in Jakarta, Indonesia	Fems Immunology and Medical Microbiology	3-97	8-99	Indonesia	Lower middle income	278	49	NA	NA	NA	NA	102	0	Mixed	other	0
4	D.;Lesmana Subekti et al	2002	Incidence of Norwalk-like viruses, rotavirus and adenovirus infection in patients with acute gastroenteritis in Jakarta, Indonesia	FEMS Immunol Med Microbiol	3-97	6-99	Indonesia	Lower middle income	161	39	NA	NA	NA	NA	102	0	< 5	inpatient	0
4	D.;Lesmana Subekti et al	2002	Incidence of Norwalk-like viruses, rotavirus and adenovirus infection in patients with acute gastroenteritis in Jakarta, Indonesia	FEMS Immunol Med Microbiol	3-97	6-99	Indonesia	Lower middle income	57	6	NA	NA	NA	NA	102	0	> 5	inpatient	0
5	D. S.;Tjaniadi Subekti et al	2002	Characterization of Norwalk-like virus associated with gastroenteritis in Indonesia	Journal of Medical Virology	10-97	9-99	Indonesia	Lower middle income	102	31	NA	NA	NA	NA	20	0	Mixed	inpatient	0
6	G. S.;Doan Hansman et al	2004	Detection of norovirus and sapovirus infection among children with gastroenteritis in Ho Chi Minh City, Vietnam	Archives of Virology	1999/12/01	2000/11/01	Vietnam	Lower middle income	1339	72	4	68	0	0	0	0	Mixed	inpatient	0
7	G. S.;Katayama Hansman et al	2004	Genetic diversity of norovirus and sapovirus in hospitalized infants with sporadic cases of acute gastroenteritis in Chiang Mai, Thailand	J Clin Microbiol	2000/07/01	2001/07/01	Thailand	Upper middle income	105	9	NA	NA	NA	NA	0	0	< 5	inpatient	0
8	U. D.;Li Parashar et al	2004	Human caliciviruses as a cause of severe gastroenteritis in Peruvian children	Journal of Infectious Diseases	1995/02/15	1997/02/14	Peru	Upper middle income	233	30	NA	NA	NA	NA	248	12	< 5	inpatient	0
9	T. G.;Okame Phan et al	2004	Human astrovirus, norovirus (GI, GII), and sapovirus infections in Pakistani children with diarrhea	Journal of Medical Virology	1990	1994	Pakistan	Lower middle income	517	51	12	39	0	0	0	0	< 5	inpatient	0

10	W.;Cunliffe Dove et al	2005	Detection and characterization of human caliciviruses in hospitalized children with acute gastroenteritis in Blantyre, Malawi	Journal of Medical Virology	7-98	6-99	Malawi	Low income	398	26	0	26	0	0	0	0	< 5	inpatient	0
11	M. C.;Rocha Albuquerque et al	2007	Human bocavirus infection in children with gastroenteritis, Brazil	Emerg Infect Dis	1-03	12-05	Brazil	Upper middle income	705	24	NA	NA	NA	NA	0	0	Mixed	other	1
12	Shuvra Kanti et al	2007	Molecular and epidemiological trend of norovirus associated gastroenteritis in Dhaka City, Bangladesh	Journal of Clinical Virology	10-04	9-05	Bangladesh	Lower middle income	917	41	0	41	0	0	0	0	< 5	other	0
13	A.;Donia Fabiana et al	2007	Influence of enteric viruses on gastroenteritis in Albania: epidemiological and molecular analysis	J Med Virol	one year study		Albania	Upper middle income	313	19	8	11	0	0	0	0	Mixed	other	0
14	Pattara; Maneekarn Khamrin et al	2007	Genetic diversity of noroviruses and sapoviruses in children hospitalized with acute gastroenteritis in Chiang Mai, Thailand	Journal of Medical Virology	3-02	12-04	Thailand	Upper middle income	248	35	0	35	0	0	0	0	< 5	inpatient	1
15	Bindhu et al	2007	Human caliciviruses in symptomatic and asymptomatic infections in children in Vellore, South India	Journal of Medical Virology	12-01	12-04	India	Lower middle income	350	53	0	27	0	0	0	0	< 5	inpatient	1
15	Bindhu et al	2007	Human caliciviruses in symptomatic and asymptomatic infections in children in Vellore, South India	Journal of Medical Virology	12-01	12-04	India	Lower middle income	500	38	0	38	0	0	173	7	< 5	community	1
16	T. A.;Yagyu Nguyen et al	2007	Diversity of viruses associated with acute gastroenteritis in children hospitalized with diarrhea in Ho Chi Minh City, Vietnam	J Med Virol	10-02	9-03	Vietnam	Lower middle income	1010	56	0	56	0	0	0	0	Mixed	inpatient	1
17	D. C.;Dove et al	2007	Norovirus infection in children with acute gastroenteritis, Madagascar, 2004-2005	Emerg Infect Dis	5-04	5-05	Madagascar	Low income	237	14	4	10	0	0	0	0	Mixed	inpatient	0
18	Carolin e C et al	2007	Norovirus detection and genotyping for children with gastroenteritis, Brazil	Emerging Infectious Diseases	1-98	5-05	Brazil	Upper middle income	117	18	NA	NA	0	0	0	0	Mixed	inpatient	1
18	Carolin e C et al	2007	Norovirus detection and genotyping for children with gastroenteritis, Brazil	Emerging Infectious Diseases	1-98	5-05	Brazil	Upper middle income	172	24	NA	NA	0	0	0	0	Mixed	outpatient	1
18	Carolin e C et al	2007	Norovirus detection and genotyping for children with gastroenteritis, Brazil	Emerging Infectious Diseases	1-98	5-05	Brazil	Upper middle income	226	33	NA	NA	0	0	0	0	< 5	other	1
18	Carolin e C et al	2007	Norovirus detection and genotyping for children with gastroenteritis, Brazil	Emerging Infectious	1-98	5-05	Brazil	Upper middle	63	9	NA	NA	0	0	0	0	> 5	other	1

				Diseases				income											
19	S. S. R.;Rajendran et al	2008	Closing the diarrhoea diagnostic gap in Indian children by the application of molecular techniques	Journal of Medical Microbiology	1-03	12-03	India	Lower middle income	158	25	NA	NA	NA	NA	99	7	< 5	inpatient	0
20	Filemon et al	2008	Pediatric norovirus diarrhea in Nicaragua	Journal of Clinical Microbiology	3-05	2-06	Nicaragua	Lower middle income	409	45	NA	NA	NA	NA	0	0	Mixed	community	0
20	Filemon et al	2008	Pediatric norovirus diarrhea in Nicaragua	Journal of Clinical Microbiology	3-05	2-06	Nicaragua	Lower middle income	133	20	NA	NA	NA	NA	0	0	Mixed	inpatient	0
21	Preeti et al	2008	Norovirus genotype IIb associated acute gastroenteritis in India	Journal of Clinical Virology	12-05	2-07	India	Lower middle income	192	24	0	24	0	0	0	0	< 5	inpatient	1
21	Preeti et al	2008	Norovirus genotype IIb associated acute gastroenteritis in India	Journal of Clinical Virology	12-05	2-07	India	Lower middle income	44	4	0	4	0	0	0	0	< 5	outpatient	1
22	Miao;Xie Jin et al	2008	Emergence of the GII4/2006b variant and recombinant noroviruses in China	Journal of Medical Virology	2-06	1-07	China	Upper middle income	1110	114	12	102	0	0	0	0	< 5	inpatient	1
23	Rungnapa et al	2008	Genetic diversity of norovirus, sapovirus, and astrovirus isolated from children hospitalized with acute gastroenteritis in Chiang Mai, Thailand	Journal of Medical Virology	5-00	3-02	Thailand	Upper middle income	296	24	7	17	0	0	0	0	< 5	inpatient	0
24	Jerome et al	2008	Detection and characterization of human caliciviruses associated with sporadic acute diarrhea in adults in Djibouti (Horn of Africa)	American Journal of Tropical Medicine and Hygiene	9-02	2-04	Djibouti	Lower middle income	75	14	NA	NA	NA	NA	0	0	> 5	inpatient	1
25	T. A.; Nguyen et al	2008	Norovirus and sapovirus infections among children with acute gastroenteritis in Ho Chi Minh City during 2005-2006	J Trop Pediatr	12-05	11-06	Vietnam	Lower middle income	502	32	0	32	0	0	0	0	Mixed	inpatient	0
26	Girish et al	2008	Genetic diversity of noroviruses and sapoviruses in children with acute sporadic gastroenteritis in New Delhi, India	Journal of Clinical Virology	1-01	12-01	India	Lower middle income	226	36	NA	NA	NA	NA	0	0	Mixed	outpatient	0
27	L. R.;Giuberti et al	2008	Hospitalization due to norovirus and genotypes of rotavirus in pediatric patients, state of Espirito Santo	Mem Inst Oswaldo Cruz	7-04	11-06	Brazil	Upper middle income	68	27	NA	NA	NA	NA	0	0	Mixed	inpatient	0
28	Khira;Gharbi-Khelifi et al	2008	Acute infantile gastroenteritis associated with human enteric viruses in Tunisia	Journal of Clinical Microbiology	1-03	6-05	Tunisia	Upper middle income	380	49	NA	NA	NA	NA	248	3	Mixed	outpatient	0
28	Khira;Gharbi-	2008	Acute infantile gastroenteritis associated with human enteric viruses	Journal of Clinical	1-03	6-05	Tunisia	Upper middle	252	61	NA	NA	NA	NA	1233	5	Mixed	inpatient	0

	Khelifi et al		in Tunisia	Microbiology				income											
29	P.;Dhongade et al	2009	Epidemiological, clinical, and molecular features of norovirus infections in western India	J Med Virol	7-05	6-07	India	Lower middle income	830	89	3	85	0	1	0	0	Mixed	other	1
30	L.;Song Guo et al	2009	Genetic analysis of norovirus in children affected with acute gastroenteritis in Beijing, 2004-2007	J Clin Virol	3-04	11-07	Beijing,China	Upper middle income	1126	100	0	100	0	0	0	0	Mixed	inpatient	1
31	Yu;Cheng Jin et al	2009	Viral agents associated with acute gastroenteritis in children hospitalized with diarrhea in Lanzhou, China	Journal of Clinical Virology	7-05	6-07	China	Upper middle income	544	50	NA	49	NA	NA	0	0	< 5	inpatient	1
32	Aziza H.;Ali Kamel et al	2009	Predominance and Circulation of Enteric Viruses in the Region of Greater Cairo, Egypt	Journal of Clinical Microbiology	3-06	2-07	Egypt	Lower middle income	230	62	NA	NA	NA	NA	0	0	Mixed	outpatient	1
33	L.;Pombubpa Kittigul et al	2009	Molecular characterization of rotaviruses, noroviruses, sapovirus, and adenoviruses in patients with acute gastroenteritis in Thailand	J Med Virol	1-06	2-07	Thailand	Upper middle income	106	13	1	12	0	0	0	0	< 5	inpatient	1
33	L.;Pombubpa Kittigul et al	2009	Molecular characterization of rotaviruses, noroviruses, sapovirus, and adenoviruses in patients with acute gastroenteritis in Thailand	J Med Virol	1-06	2-07	Thailand	Upper middle income	156	4	1	3	0	0	0	0	> 5	inpatient	1
34	M. K.;Chatterjee Nayak et al	2009	A new variant of Norovirus GII.4/2007 and inter-genotype recombinant strains of NVGII causing acute watery diarrhoea among children in Kolkata, India	J Clin Virol	7-06	9-07	India	Lower middle income	111	21	0	21	0	0	0	0	< 5	outpatient	1
35	Khira;Ambert-Balay Sdiri-Loulizi et al	2009	Molecular Epidemiology of Norovirus Gastroenteritis Investigated Using Samples Collected from Children in Tunisia during a Four-Year Period: Detection of the Norovirus Variant GGII.4 Hunter as Early as January 2003	Journal of Clinical Microbiology	1-03	4-07	Tunisia	Upper middle income	788	128	11	117	0	0	0	0	Mixed	other	1
36	T. V.;Natarajan Sowmyanaraya nan et al	2009	Nitric oxide production in acute gastroenteritis in Indian children	Trans R Soc Trop Med Hyg	1-06	12-07	India	Lower middle income	110	13	NA	NA	NA	NA	0	0	< 5	other	1
37	Jin;Yang Xu et al	2009	Molecular Epidemiology of Norovirus Infection Among Children With Acute Gastroenteritis in Shanghai, China, 2001-2005	Journal of Medical Virology	2001/01/01	2005/12/31	Shanghai,China	Upper middle income	484	45	NA	37	NA	NA	0	0	< 5	inpatient	1
38	F.;Nordgren	2010	Asymptomatic norovirus infections in Nicaraguan children and its association	Pediatr Infect Dis J	6-05	9-06	Nicaragua	Lower middle	163	19	4	10	0	4	0	0	< 5	community	0

	Bucardo et al		with viral properties and histo-blood group antigens					income											
39	Wei-xia;Ye Cheng et al	2010	Epidemiological study of human calicivirus infection in children with gastroenteritis in Lanzhou from 2001 to 2007	Archives of Virology	12-01	6-07	China	Upper middle income	1195	86	2	81	NA	0	0	0	< 5	inpatient	1
40	M. S. R.;Victoria Ferreira et al	2010	Surveillance of Norovirus Infections in the State of Rio De Janeiro, Brazil 2005-2008	Journal of Medical Virology	2005	2008	Brazil	Upper middle income	176	80	NA	NA	NA	NA	0	0	> 5	inpatient	1
40	M. S. R.;Victoria Ferreira et al	2010	Surveillance of Norovirus Infections in the State of Rio De Janeiro, Brazil 2005-2008	Journal of Medical Virology	2005	2008	Brazil	Upper middle income	58	12	NA	NA	NA	NA	0	0	> 5	outpatient	1
40	M. S. R.;Victoria Ferreira et al	2010	Surveillance of Norovirus Infections in the State of Rio De Janeiro, Brazil 2005-2008	Journal of Medical Virology	2005	2008	Brazil	Upper middle income	85	46	NA	NA	NA	NA	0	0	< 5	inpatient	1
40	M. S. R.;Victoria Ferreira et al	2010	Surveillance of Norovirus Infections in the State of Rio De Janeiro, Brazil 2005-2008	Journal of Medical Virology	2005	2008	Brazil	Upper middle income	671	224	NA	NA	NA	NA	0	0	< 5	outpatient	1
41	Debora Maria Pires et al	2010	Viral load and genotypes of noroviruses in symptomatic and asymptomatic children in Southeastern Brazil	Journal of Clinical Virology	2-03	6-04	Brazil	Upper middle income	229	40	NA	NA	NA	NA	90	12	< 5	inpatient	0
42	A. L.;Velazquez Gutierrez-Escolano et al	2010	Human caliciviruses detected in Mexican children admitted to hospital during 1998-2000, with severe acute gastroenteritis not due to other enteropathogens	J Med Virol	3-98	12-00	Mexico	Upper middle income	299	20	0	19	NA	0	0	0	< 5	inpatient	0
43	P.;Manekarn Khamrin et al	2010	Emergence of new norovirus variants and genetic heterogeneity of noroviruses and sapoviruses in children admitted to hospital with diarrhea in Thailand	J Med Virol	1-05	12-05	Thailand	Upper middle income	147	10	0	10	0	0	0	0	< 5	inpatient	0
44	L.;Pombubpa Kittigul et al	2010	Norovirus GII-4 2006b variant circulating in patients with acute gastroenteritis in Thailand during a 2006-2007 study	J Med Virol	1-06	2-07	Thailand	Upper middle income	158	73	18	48	0	7	0	0	> 5	other	1

44	L.;Pombubpa Kittigul et al	2010	Norovirus GII-4 2006b variant circulating in patients with acute gastroenteritis in Thailand during a 2006-2007 study	J Med Virol	1-06	2-07	Thailand	Upper middle income	75	35	8	23	0	4	0	0	Mixed	outpatient	1
44	L.;Pombubpa Kittigul et al	2010	Norovirus GII-4 2006b variant circulating in patients with acute gastroenteritis in Thailand during a 2006-2007 study	J Med Virol	1-06	2-07	Thailand	Upper middle income	115	49	10	31	0	8	0	0	< 5	other	1
44	L.;Pombubpa Kittigul et al	2010	Norovirus GII-4 2006b variant circulating in patients with acute gastroenteritis in Thailand during a 2006-2007 study	J Med Virol	1-06	2-07	Thailand	Upper middle income	198	87	20	56	0	11	0	0	Mixed	inpatient	1
45	L. J.;Liu Liu et al	2010	Identification of norovirus as the top enteric viruses detected in adult cases with acute gastroenteritis	Am J Trop Med Hyg	7-07	6-08	Beijing,China	Upper middle income	557	147	25	112	0	0	0	0	> 5	inpatient	0
46	J.;de Villiers Mans et al	2010	Emerging norovirus GII.4 2008 variant detected in hospitalised paediatric patients in South Africa	J Clin Virol	1-08	12-08	South Africa	Upper middle income	245	35	3	31	0	1	0	0	Mixed	inpatient	0
47	K.;Sebunya Mattison et al	2010	Molecular detection and characterization of noroviruses from children in Botswana	J Med Virol	2000/01/01	2006/12/31	Botswana	Upper middle income	74	16	0	16	0	0	26	8	Mixed	inpatient	1
48	V. K.;George Menon et al	2010	Genogroup IIb norovirus infections and association with enteric symptoms in a neonatal nursery in southern India	J Clin Microbiol	1-03	12-06	India	Lower middle income	161	60	NA	NA	NA	NA	148	24	< 5	inpatient	1
49	S. M.;Ganesh Nataraju et al	2010	Emergence of Noroviruses homologous to strains reported from Djibouti (horn of Africa), Brazil, Italy, Japan and USA among children in Kolkata, India	Eur Rev Med Pharmacol Sci	5-08	5-09	India	Lower middle income	53	5	NA	NA	NA	NA	312	2	< 5	outpatient	0
49	S. M.;Ganesh Nataraju et al	2010	Emergence of Noroviruses homologous to strains reported from Djibouti (horn of Africa), Brazil, Italy, Japan and USA among children in Kolkata, India	Eur Rev Med Pharmacol Sci	5-08	5-09	India	Lower middle income	260	5	NA	NA	NA	NA	312	2	< 5	inpatient	0
50	M.;Hassan Rahman et al	2010	Molecular detection of noroviruses in hospitalized patients in Bangladesh	Eur J Clin Microbiol Infect Dis	1-04	12-05	Bangladesh	Lower middle income	189	37	3	13	0	1	0	0	Mixed	inpatient	0
51	M.;Cuevas Abugalia et al	2011	Clinical features and molecular epidemiology of rotavirus and norovirus infections in Libyan children	J Med Virol	10-07	9-08	Libya	Upper middle income	260	36	NA	NA	NA	NA	0	0	< 5	inpatient	0

51	M.;Cuevas Abugalia et al	2011	Clinical features and molecular epidemiology of rotavirus and norovirus infections in Libyan children	J Med Virol	10-07	9-08	Libya	Upper middle income	260	55	NA	NA	NA	NA	0	0	< 5	outpatient	0
52	A.;Luchs Cilli et al	2011	Characterization of rotavirus and norovirus strains: a 6-year study (2004-2009)	J Pediatr (Rio J)	2004	2009	Sao Paulo, Brazil	Upper middle income	89	26	3	16	NA	NA	0	0	< 5	other	1
53	Ying-chun;Hu Dai et al.	2011	Molecular epidemiology of norovirus gastroenteritis in children in Jiangmen, China, 2005-2007	Archives of Virology	9-05	8-07	Jiangmen,China	Upper middle income	881	115	0	115	0	0	0	0	< 5	outpatient	1
54	Yan;Jin Gao et al.	2011	Clinical and Molecular Epidemiologic Analyses of Norovirus-Associated Sporadic Gastroenteritis in Adults From Beijing, China	Journal of Medical Virology	10-07	9-08	China	Upper middle income	403	48	4	44	0	0	0	0	> 5	outpatient	0
55	N. M.;Kirby Kaplan et al.	2011	Detection and molecular characterisation of rotavirus and norovirus infections in Jordanian children with acute gastroenteritis	Arch Virol	1-06	12-07	Jordan	Upper middle income	368	42	4	37	0	1	0	0	Mixed	inpatient	1
56	A.;Al-Eryani Kirby et al.	2011	Rotavirus and norovirus infections in children in Sana'a, Yemen	Trop Med Int Health	11-07	3-09	Yemen	Lower middle income	290	30	10	18	0	2	0	0	< 5	other	0
57	S. M.;Pativada et al.	2011	Molecular epidemiology of norovirus infections in children and adults: sequence analysis of region C indicates genetic diversity of NVGII strains in Kolkata, India	Epidemiology and Infection	11-07	10-09	India	Lower middle income	2495	78	6	72	0	0	0	0	Mixed	inpatient	0
58	A. A.;Kocazeybek et al.	2011	Frequency and phylogeny of norovirus in diarrheic children in Istanbul, Turkey	J Clin Virol	4-08	8-09	Turkey	Upper middle income	238	36	0	36	0	0	0	0	Mixed	inpatient	0
59	F. P.;Ochoa Rivera et al.	2011	Norovirus prevalence in 'pathogen negative' gastroenteritis in children from periurban areas in Lima, Peru	Trans R Soc Trop Med Hyg	9-06	08-7	Peru	Upper middle income	224	39	1	36	0	NA	0	0	< 5	community	0
60	K.;Hassine et al.	2011	Molecular detection of genogroup I sapovirus in Tunisian children suffering from acute gastroenteritis	Virus Genes	1-03	4-07	Tunisia	Upper middle income	788	128	11	117	0	0	0	0	Mixed	other	1
61	M.;Gong Zeng, Z. et al.	2011	Prevalence and genetic diversity of norovirus in outpatient children with acute diarrhea in Shanghai, China	Jpn J Infect Dis	8-08	7-09	Shanghai,China	Upper middle income	910	165	4	161	0	0	0	0	Mixed	inpatient	0
62	S.;Chen et al.	2011	Symptomatic and asymptomatic infections of rotavirus, norovirus, and adenovirus among hospitalized children in Xi'an, China	J Med Virol	3-09	5-10	China	Upper middle income	201	41	0	41	0	0	53	19	< 5	inpatient	0

63	N.;Khamrin et al.	2012	A wide variety of diarrhea viruses circulating in pediatric patients in Thailand	Clin Lab	1-07	12-07	Thailand	Upper middle income	160	24	1	23	0	0	0	0	< 5	inpatient	0
64	Hanan; Mansour et al.	2012	Increase in the detection rate of viral and parasitic enteric pathogens among Egyptian children with acute diarrhea	Journal of Infection in Developing Countries	2005	2007	Egypt	Lower middle income	2112	191	NA	NA	NA	NA	0	0	< 5	outpatient	1
65	M. S.;Xavier Mda et al.	2012	Assessment of gastroenteric viruses frequency in a children's day care center in Rio De Janeiro, Brazil: a fifteen year study (1994-2008)	PLoS One	1994	2008	Rio de Janeiro, Brazil	Upper middle income	130	44	NA	NA	NA	NA	0	0	< 5	community	1
66	F.;Ribas-Aparicio et al.	2012	Molecular characterization of human calicivirus associated with acute diarrheal disease in Mexican children	Virol J	10-05	12-06	Mexico	Upper middle income	414	128	0	72	0	0	0	0	< 5	other	0
67	Shahram - et al.	2012	Relative Frequency of Noroyirus Infection in Children Suffering From Gastroenteritis and Referred to Aboozar Hospital, Ahvaz, Iran	Jundishapur Journal of Microbiology	2008	2009	Iran	Upper middle income	143	9	NA	NA	NA	NA	0	0	< 5	inpatient	0
68	Yabo;Ma Ouyang, et al.	2012	Etiology and epidemiology of viral diarrhea in children under the age of five hospitalized in Tianjin, China	Archives of Virology	4-08	4-09	China	Upper middle income	766	64	NA	NA	NA	NA	0	0	< 5	inpatient	0
69	S.;Mohebbi Romani et al.	2012	Prevalence of norovirus infection in children and adults with acute gastroenteritis, Tehran, Iran, 2008-2009	Food Environ Virol	5-08	5-09	Iran	Upper middle income	293	26	9	17	0	0	0	0	Mixed	inpatient	0
70	N. V.;Luan le Trang, et al.	2012	Detection and molecular characterization of noroviruses and sapoviruses in children admitted to hospital with acute gastroenteritis in Vietnam	J Med Virol	11-07	10-08	Vietnam	Lower middle income	501	180	0	180	0	0	0	0	< 5	inpatient	0
71	Y. H.;Zhou Wang et al.	2012	Molecular epidemiology of noroviruses in children and adults with acute gastroenteritis in Wuhan, China, 2007-2010	Arch Virol	1-07	5-10	Wuhan, China	Upper middle income	1042	277	NA	NA	NA	NA	0	0	< 5	inpatient	0
71	Y. H.;Zhou Wang et al.	2012	Molecular epidemiology of noroviruses in children and adults with acute gastroenteritis in Wuhan, China, 2007-2010	Arch Virol	1-07	5-10	Wuhan, China	Upper middle income	747	189	NA	NA	NA	NA	0	0	> 5	inpatient	0
72	M.;Xu Zeng et al.	2012	Clinical and molecular epidemiology of norovirus infection in childhood diarrhea in China	J Med Virol	8-08	7-09	China	Upper middle income	317	81	0	81	0	0	0	0	Mixed	inpatient	0
72	M.;Xu Zeng et al.	2012	Clinical and molecular epidemiology of norovirus infection in childhood diarrhea in China	J Med Virol	8-08	7-09	China	Upper middle income	4123	1067	13	1051	0	3	0	0	Mixed	outpatient	0

73	Glicelia Cruz et al.	2013	Norovirus Diversity in Diarrheic Children from an African-Descendant Settlement in Belem, Northern Brazil	Plos One	4-08	7-10	Brazil	Upper middle income	81	16	1	11	NA	NA	78	0	< 5	community	0
74	Y.;Li Chen et al.	2013	Viral agents associated with acute diarrhea among outpatient children in southeastern China	Pediatr Infect Dis J	1-10	12-10	China	Upper middle income	811	147	31	116	0	0	0	0	< 5	inpatient	0
75	D.;Bicer et al.	2013	Annual report on norovirus in children with acute gastroenteritis in 2009 and their genotypes in Turkey	Infez Med	1-09	1-10	Turkey	Upper middle income	412	44	NA	NA	NA	NA	0	0	< 5	other	0
75	D.;Bicer et al.	2013	Annual report on norovirus in children with acute gastroenteritis in 2009 and their genotypes in Turkey	Infez Med	1-09	1-10	Turkey	Upper middle income	108	6	NA	NA	NA	NA	0	0	> 5	other	0
76	A.;Arvelo et al.	2013	Prevalence and genetic diversity of norovirus among patients with acute diarrhea in Guatemala	J Med Virol	10-07	8-10	Guatemala	Lower middle income	1875	227	NA	NA	NA	NA	0	0	Mixed	outpatient	0
76	A.;Arvelo Estevez et al.	2013	Prevalence and genetic diversity of norovirus among patients with acute diarrhea in Guatemala	J Med Virol	10-07	8-10	Guatemala	Lower middle income	528	114	NA	NA	NA	NA	0	0	Mixed	inpatient	0
77	M. E.;Martinez et al.	2013	Molecular epidemiology of norovirus strains in Paraguayan children during 2004-2005: description of a possible new GII.4 cluster	J Clin Virol	1-04	12-05	Paraguay	Upper middle income	378	161	34	46	0	4	0	0	< 5	other	0
78	M.;Sdiri-Loulizi et al.	2013	Prevalence and genetic diversity of norovirus infection in Tunisian children (2007-2010)	J Med Virol	4-07	4-10	Tunisia	Upper middle income	407	38	6	32	0	0	0	0	< 5	inpatient	0
79	Vipin Kumar et al.	2013	Short Report: Norovirus Genogroup II Gastroenteritis in Hospitalized Children in South India	American Journal of Tropical Medicine and Hygiene	2005	2006	India	Lower middle income	282	28	0	28	0	0	0	0	< 5	inpatient	0
80	S.;Afrad Nahar et al.	2013	High prevalence of noroviruses among hospitalized diarrheal patients in Bangladesh, 2011	J Infect Dev Ctries	1-11	12-11	Bangladesh	Lower middle income	257	73	15	52	0	0	0	0	Mixed	inpatient	0
81	Akram;Iranpour Najafi et al.	2013	Epidemiological Surveillance of Norovirus Diarrhea in Hospitalized Children with Acute Gastroenteritis in South of Iran	Jundishapur Journal of Microbiology	2008	2010	Iran	Upper middle income	29	1	NA	NA	NA	NA	0	0	> 5	inpatient	0
81	Akram;Iranpour et al.	2013	Epidemiological Surveillance of Norovirus Diarrhea in Hospitalized Children with Acute Gastroenteritis in South of Iran	Jundishapur Journal of Microbiology	2008	2010	Iran	Upper middle income	346	46	NA	NA	NA	NA	0	0	< 5	inpatient	0
82	Pimmnapar;Sirinarumitr et al.	2013	Optimization of one-step real-time reverse transcription-polymerase chain reaction assays for norovirus detection and molecular epidemiology of noroviruses in Thailand	Journal of Virological Methods	12-05	11-06	Thailand	Upper middle income	500	119	3	116	0	0	441	30	< 5	inpatient	0

83	My;Thompson et al.	2013	Endemic Norovirus Infections in Children, Ho Chi Minh City, Vietnam, 2009-2010	Emerging Infectious Diseases	5-09	12-10	Vietnam	Lower middle income	1419	241	3	240	0	0	607	15	< 5	inpatient	0
84	L.;Sun Sai et al.	2013	Epidemiology and clinical features of rotavirus and norovirus infection among children in Ji'nan, China	Virol J	2-11	1-12	china	Upper middle income	502	58	0	58	0	0	0	0	< 5	outpatient	0
84	L.;Sun Sai et al.	2013	Epidemiology and clinical features of rotavirus and norovirus infection among children in Ji'nan, China	Virol J	2-11	1-12	china	Upper middle income	265	22	0	22	0	0	0	0	< 5	inpatient	0
85	L.;Wang Sai et al.	2013	Clinical and molecular epidemiology of norovirus infection in adults with acute gastroenteritis in Ji'nan, China	Arch Virol	6-10	5-11	china	Upper middle income	480	42	2	40	0	0	0	0	> 5	outpatient	0
86	Z.;Qian et al.	2013	Novel Norovirus GII.4 Variant, Shanghai, China, 2012	Emerg Infect Dis	7-11	12-12	Shanghai,China	Upper middle income	748	77	NA	77	NA	NA	0	0	> 5	outpatient	0
87	J. A.;Linhares Ada et al.	2013	Norovirus infection in children admitted to hospital for acute gastroenteritis in Belem, Para, Northern Brazil	J Med Virol	5-08	4-09	Belam,Brazil	Upper middle income	169	69	NA	NA	NA	NA	0	0	< 5	inpatient	0
88	J. A.;Linhares Ada et al.	2013	Group A rotavirus and norovirus display sharply distinct seasonal profiles in Belem, northern Brazil	Mem Inst Oswaldo Cruz	5-08	4-11	Belam,Brazil	Upper middle income	483	171	NA	NA	NA	NA	0	0	< 5	inpatient	0
89	A.;Khamrin et al.	2013	Emergence of norovirus GII/4 2006a and 2006b variants in hospitalized children with acute gastroenteritis in Thailand	Clin Lab	1-06	12-06	Thailand	Upper middle income	156	32	0	32	0	0	0	0	< 5	inpatient	0
90	P. V.;Lam et al.	2013	The dynamics of GII.4 Norovirus in Ho Chi Minh City, Vietnam	Infect Genet Evol	5-09	12-10	Vietnam	Lower middle income	2054	315	11	304	0	0	0	0	< 5	inpatient	0
91	E.;Lopman Trainor et al.	2013	Detection and molecular characterisation of noroviruses in hospitalised children in Malawi, 1997-2007	J Med Virol	7-97	6-07	Malawi	Low income	1941	220	37	185	0	0	0	0	< 5	inpatient	1
92	M.;Cui Zhu, S. et al.	2013	Analysis of the aetiology of diarrhoea in outpatients in 2007, Henan province, China	Epidemiology and Infection	1-07	12-07	china	Upper middle income	1526	56	NA	NA	NA	NA	0	0	Mixed	outpatient	0
93	J. A.;Andersson et al.	2014	Monitoring of seasonality of norovirus and other enteric viruses in Cameroon by real-time PCR: an exploratory study	Epidemiol Infect	9-11	8-12	cameroon	Lower middle income	200	100	45	55	0	0	0	0	Mixed	community	0
94	S.;Bucardo Becker-Dreps et	2014	Etiology of childhood diarrhea after rotavirus vaccine introduction: a prospective, population-based study in Nicaragua	Pediatr Infect Dis J	2010/01/25	2011/01/24	Nicaragua	Lower middle income	333	68	15/63	45/63	0	3-63	106	14	< 5	community	0

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95	N.;Khamrin Chaimongkol, P et al.	2014	Molecular characterization of norovirus variants and genetic diversity of noroviruses and sapoviruses in Thailand	J Med Virol	1-07	1/2010-2011	Thailand	Upper middle income	567	90	1	89	0	0	0	0	< 5	inpatient	0
96	P.;Samoilovich Chhabra, E et al.	2014	Viral gastroenteritis in rotavirus negative hospitalized children <5 years of age from the independent states of the former Soviet Union	Infect Genet Evol	1-09	12-09	four NIS (Armenia, Georgia,Moldova,Ukraina)	Lower middle income	170	43	1	42	0	0	0	0	< 5	inpatient	0
96	P.;Samoilovich Chhabra et al.	2014	Viral gastroenteritis in rotavirus negative hospitalized children <5 years of age from the independent states of the former Soviet Union	Infect Genet Evol	1-09	12-09	twoNIS (Azerbaijan, Belarus)	Upper middle income	325	65	2	63	0	0	0	0	< 5	inpatient	0
97	M.;Oumzil El Qazoui et al.	2014	Rotavirus and norovirus infections among acute gastroenteritis children in Morocco	BMC Infect Dis	1-11	12-12	Morocco	Lower middle income	335	54	2	63	0	0	0	0	< 5	inpatient	0
98	D.;Wasfy Elyan et al.	2014	Non-bacterial etiologies of diarrheal diseases in Afghanistan	Trans R Soc Trop Med Hyg	2009	2010	Afghanistan	Low income	432	3	NA	NA	NA	NA	0	0	< 5	outpatient	0
99	Li-ping; et al.	2014	Prevalence and genetic diversity of noroviruses in outpatient pediatric clinics in Beijing, China 2010-2012	Infection Genetics and Evolution	10-10	12-12	china	Upper middle income	1128	182	NA	177	NA	NA	0	0	Mixed	outpatient	0
100	P.;Wang Liu, et al.	2014	Genetic susceptibility to norovirus GII.3 and GII.4 infections in Chinese pediatric diarrheal disease	Pediatr Infect Dis J	3-09	3-10	Xian, China	Upper middle income	124	34	0	34	0	0	0	0	< 5	inpatient	0
101	L.;Zhong Lu, et al.	2014	Molecular epidemiology of human calicivirus infections in children with acute diarrhea in Shanghai: a retrospective comparison between inpatients and outpatients treated between 2006 and 2011	Arch Virol	2006	2011	Shanghai,China	Upper middle income	674	206	0	206	0	0	0	0	< 5	inpatient	1
101	L.;Zhong Lu, et al.	2014	Molecular epidemiology of human calicivirus infections in children with acute diarrhea in Shanghai: a retrospective comparison between inpatients and outpatients treated between 2006 and 2011	Arch Virol	2006	2011	Shanghai,China	Upper middle income	436	127	1	126	0	0	0	0	< 5	outpatient	1
102	J.;Murray Mans et al.	2014	Human caliciviruses detected in HIV-seropositive children in Kenya	J Med Virol	2-99	6-00	Kenya	Lower middle income	105	18	1	16	0	1	0	0	Mixed	community	0
103	Denisy et al.	2014	Monitoring of Calicivirus Among Day-Care Children: Evidence of Asymptomatic Viral Excretion and	Journal of Medical Virology	10-09	10-11	Brazil	Upper middle income	539	43	NA	NA	NA	NA	0	0	< 5	community	0

			First Report of GI.7 Norovirus and GI.3 Sapovirus in Brazil																
104	M. T.;Bozdayi Mitui et al.	2014	Detection and molecular characterization of diarrhea causing viruses in single and mixed infections in children: a comparative study between Bangladesh and Turkey	J Med Virol	7-05	6-06	Bangladesh	Lower middle income	138	97	6	91	0	0	0	0	< 5	inpatient	0
104	M. T.;Bozdayi Mitui et al.	2014	Detection and molecular characterization of diarrhea causing viruses in single and mixed infections in children: a comparative study between Bangladesh and Turkey	J Med Virol	9-04	12-05	Turkey	Upper middle income	150	15	3	12	0	0	0	0	< 5	inpatient	0
105	S.;Hanevik Moyo et al.	2014	Genetic diversity of norovirus in hospitalised diarrhoeic children and asymptomatic controls in Dar es Salaam, Tanzania	Infect Genet Evol	8-10	7-11	Tanzania	Low income	705	129	0	129	0	0	561	52	< 5	inpatient	0
106	S. M.;Damasio et al.	2014	Acute gastroenteritis and enteric viruses in hospitalised children in southern Brazil: aetiology, seasonality and clinical outcomes	Mem Inst Oswaldo Cruz	9-10	9-11	Brazil	Upper middle income	225	19	NA	NA	NA	NA	0	0	Mixed	inpatient	0
107	Wilaiporn; et al.	2014	Detection of diarrheal viruses circulating in adult patients in Thailand	Archives of Virology	1-08	12-08	Thailand	Upper middle income	332	3	0	3	0	0	0	0	Mixed	inpatient	0
108	K. W.;Maure Soli et al.	2014	Detection of enteric viral and bacterial pathogens associated with paediatric diarrhoea in Goroka, Papua New Guinea	Int J Infect Dis	8-09	11-10	Papua New Guinea	Lower middle income	199	27	10	17	0	0	0	0	< 5	inpatient	0
109	Bicer;Defne Suat et al.	2014	A Retrospective Analysis of Acute Gastroenteritis Agents in Children Admitted to a University Hospital Pediatric Emergency Unit	Jundishapur Journal of Microbiology	1-09	1-10	Turkey	Upper middle income	520	51	NA	NA	NA	NA	0	0	Mixed	inpatient	0
110	G.;Jin Tian et al.	2014	Clinical characteristics and genetic diversity of noroviruses in adults with acute gastroenteritis in Beijing, China in 2008-2009	J Med Virol	8-08	7-09	Beijing,China	Upper middle income	519	136	28	108	0	4	0	0	> 5	inpatient	0
111	N.;Vu Van Trang et al.	2014	Association between norovirus and rotavirus infection and histo-blood group antigen types in Vietnamese children	J Clin Microbiol	9-10	9-12	Vietnam	Lower middle income	807	311	2	284	NA	1	0	0	< 5	inpatient	0
112	Wei;Yang et al.	2014	Surveillance of pathogens causing gastroenteritis and characterization of norovirus and sapovirus strains in Shenzhen, China, during 2011	Archives of Virology	1-11	12-11	Shenzhen,China	Upper middle income	983	210	5	63	NA	NA	0	0	Mixed	outpatient	0
113	Muhammad et al.	2015	Viral Etiologies of Acute Dehydrating Gastroenteritis in Pakistani Children: Confounding Role of Parechoviruses	Viruses-Basel	1-09	12-10	Pakistan	Lower middle income	563	110	NA	NA	NA	NA	0	0	< 5	inpatient	0

114	M. S.;Estevam et al.	2015	The prevalence of norovirus, astrovirus and adenovirus infections among hospitalised children with acute gastroenteritis in Porto Velho, state of Rondonia, western Brazilian Amazon	Mem Inst Oswaldo Cruz	2-10	2-12	Brazil	Upper middle income	591	46	NA	NA	NA	NA	0	0	Mixed	inpatient	0
115	Katherine et al.	2015	The epidemiology and aetiology of diarrhoeal disease in infancy in southern Vietnam: a birth cohort study	International Journal of Infectious Diseases	7-09	12-13	Vietnam	Lower middle income	748	176	7	167	0	2	0	0	< 5	other	0
116	Estienney et al.	2015	Relationship between GII.3 norovirus infections and blood group antigens in young children in Tunisia	Clinical Microbiology and Infection	1-11	8-12	Tunisia	Upper middle income	114	42	0	42	0	0	0	0	Mixed	inpatient	0
117	Sarah-Blythe et al.	2015	Epidemiology and Genetic Characterization of Noroviruses among Adults in an Endemic Setting, Peruvian Amazon Basin, 2004-2011	Plos One	2004/02/01	2011/12/31	Peru	Upper middle income	184	26	6	19	0	1	176	14	> 5	community	1
118	R.;Jroundi Benmessaoud et al.	2015	Aetiology, epidemiology and clinical characteristics of acute moderate-to-severe diarrhoea in children under 5 years of age hospitalized in a referral paediatric hospital in Rabat, Morocco	J Med Microbiol	3-11	3-12	Morocco	Lower middle income	122	1	NA	NA	NA	NA	0	0	< 5	inpatient	0
119	L.;Abente Bodhidatta et al.	2015	Molecular epidemiology and genotype distribution of noroviruses in children in Thailand from 2004 to 2010: a multi-site study	J Med Virol	10/20040to12/2006;3/2008 to8/2010		Thailand	Upper middle income	3621	516	13	503	0	0	3799	181	< 5	outpatient	0
120	Derya; et al.	2015	Eight different viral agents in childhood acute gastroenteritis	Turkish Journal of Pediatrics	1-12	1-13	Turkey	Upper middle income	240	56	NA	NA	NA	NA	0	0	Mixed	other	0
121	Maria Sandra et al.	2015	The prevalence of norovirus, astrovirus and adenovirus infections among hospitalised children with acute gastroenteritis in Porto Velho, state of Rondonia, western Brazilian Amazon	Memorias Do Instituto Oswaldo Cruz	2-10	2-12	Brazil	Upper middle income	591	46	NA	NA	NA	NA	0	0	Mixed	inpatient	0
122	Maria;Castano de los et al.	2015	Norovirus and Rotavirus infection in children aged less than five years in a paediatric hospital, Havana, Cuba	Brazilian Journal of Infectious Diseases	3-10	2-11	Cuba	Upper middle income	88	28	NA	NA	NA	NA	0	0	< 5	inpatient	0
123	Zhiyong;Liu Gao et al.	2015	Increased norovirus activity was associated with a novel norovirus GII.17 variant in Beijing, China during winter 2014-2015	Bmc Infectious Diseases	1-12	3-15	china	Upper middle income	640	191	3	187	0	1	0	0	Mixed	outpatient	0
124	Zhiyong;Li Gao et al.	2015	Human calicivirus occurrence among outpatients with diarrhea in Beijing, China, between April 2011 and March 2013	Journal of Medical Virology	4-11	3-13	China	Upper middle income	287	263	21	237	0	5	0	0	Mixed	inpatient	0
125	Paul A. et al.	2015	Transmission of Norovirus Within Households in Quininde, Ecuador	Pediatric Infectious	2-11	5-12	Ecuador	Upper middle	186	19	17	38	0	5	146	15	< 5	inpatient	0

				Disease Journal				income										
126	Shilpi et al.	2015	Aetiology of childhood viral gastroenteritis in Lucknow, north India	Indian Journal of Medical Research	8-10	7-12	India	Lower middle income	169	4	NA	NA	NA	NA	0	0	< 5	inpatient	0
126	Shilpiet al.	2015	Aetiology of childhood viral gastroenteritis in Lucknow, north India	Indian Journal of Medical Research	8-10	7-12	India	Lower middle income	109	0	NA	NA	NA	NA	0	0	< 5	community	0
127	Jiankang; et al.	2015	Emergence and predominance of norovirus GII.17 in Huzhou, China, 2014-2015	Virology Journal	3-14	2-15	China	Upper middle income	809	193	12	180	0	1	0	0	Mixed	outpatient	0
128	Ralf et al.	2015	Gastrointestinal Infections and Diarrheal Disease in Ghanaian Infants and Children: An Outpatient Case-Control Study	Plos Neglected Tropical Diseases	6-07	10-08	Ghana	Lower middle income	548	91	NA	NA	NA	NA	651	48	Mixed	outpatient	0
129	Xiaofang; et al.	2015	Molecular detection and characterization of sapovirus in hospitalized children with acute gastroenteritis in the Philippines	Journal of Clinical Virology	6-12	8-13	Philippines	Lower middle income	417	25	NA	25	NA	NA	0	0	< 5	inpatient	0
130	B. A.;Trivedi Lopman et al.	2015	Norovirus Infection and Disease in an Ecuadorian Birth Cohort: Association of Certain Norovirus Genotypes With Host FUT2 Secretor Status	J Infect Dis	3-09	3-12	Ecuador	Upper middle income	438	70	NA	NA	NA	NA	1016	181	Mixed	community	0
131	L.;Jia et al.	2015	Molecular characterization and multiple infections of rotavirus, norovirus, sapovirus, astrovirus and adenovirus in outpatients with sporadic gastroenteritis in Shanghai, China, 2010-2011	Arch Virol	1-10	12-11	Shanghai,China	Upper middle income	436	126	0	126	0	0	0	0	< 5	outpatient	0
132	Qing-Bin;Huang Lu et al.	2015	An increasing prevalence of recombinant Gil norovirus in pediatric patients with diarrhea during 2010-2013 in China	Infection Genetics and Evolution	1-10	12-13	china	Upper middle income	2140	1123	5	1118	0	0	0	0	Mixed	inpatient	0
133	Suelen; Modena Paesi et al.	2015	Detection of rotavirus and norovirus in the elderly population of Caxias do Sul, Rio Grande do Sul, Brazil, from 2010 to 2012	Scientia Medica	2010	2012	Brazil	Upper middle income	51	6	NA	NA	NA	NA	0	0	> 5	outpatient	0
134	P. R.;Chitambar et al.	2015	Molecular surveillance of non-polio enterovirus infections in patients with acute gastroenteritis in Western India: 2004-2009	J Med Virol	1-08	12-09	India	Lower middle income	450	24	NA	NA	NA	NA	0	0	< 5	inpatient	0
135	L.;Castano et al.	2015	Norovirus and Rotavirus infection in children aged less than five years in a paediatric hospital, Havana, Cuba	Braz J Infect Dis	3-10	2.2011	Cuba	Upper middle income	88	28	NA	22	NA	NA	0	0	< 5	inpatient	0
136	D.;Deng Tan, L.	2015	High prevalence and genetic diversity of noroviruses among children with	J Med Virol	1-10	12-11	Nanning,China	Upper middle	354	101	0	101	0	0	0	0	< 5	outpatient	0

	et al.		sporadic acute gastroenteritis in Nanning City, China, 2010-2011					income											
137	Hoa-Tran et al.	2015	Molecular epidemiology of noroviruses detected in Nepalese children with acute diarrhea between 2005 and 2011: Increase and predominance of minor genotype GII.13	Infection Genetics and Evolution	11-05	1-11	Nepal	Low income	4437	208	17	90	0	1	0	0	< 5	inpatient	1
137	Hoa-Tran et al.	2015	Molecular epidemiology of noroviruses detected in Nepalese children with acute diarrhea between 2005 and 2011: Increase and predominance of minor genotype GII.13	Infection Genetics and Evolution	11-05	1-11	Nepal	Low income	4437	148	14	133	1	0	0	0	< 5	outpatient	1
138	Corinne N. et al.	2015	A Prospective Multi-Center Observational Study of Children Hospitalized with Diarrhea in Ho Chi Minh City, Vietnam	American Journal of Tropical Medicine and Hygiene	5-09	4-10	Vietnam	Lower middle income	1419	293	NA	NA	NA	NA	609	16	< 5	inpatient	0
139	Xin;Wang Wang et al.	2015	Etiology of Childhood Infectious Diarrhea in a Developed Region of China: Compared to Childhood Diarrhea in a Developing Region and Adult Diarrhea in a Developed Region	Plos One	2010/10/01	2014/09/30	Beijing,China	Upper middle income	1422	139	NA	NA	NA	NA	0	0	< 5	outpatient	0
139	Xin;Wang Wang et al.	2015	Etiology of Childhood Infectious Diarrhea in a Developed Region of China: Compared to Childhood Diarrhea in a Developing Region and Adult Diarrhea in a Developed Region	Plos One	2010/10/01	2014/09/30	Beijing,China	Upper middle income	507	57	NA	NA	NA	NA	0	0	> 5	outpatient	0
140	X.;Han Wu et al.	2015	Prevalence and genetic diversity of noroviruses in adults with acute gastroenteritis in Huzhou, China, 2013-2014	Arch Virol	3-13	2-14	Huzhou,China	Upper middle income	796	211	8	203	0	0	0	0	> 5	outpatient	0
141	Ying;Pan Xue et al.	2015	Epidemiology of norovirus infections among diarrhea outpatients in a diarrhea surveillance system in Shanghai, China: a cross-sectional study	Bmc Infectious Diseases	5-12	2014/04/30	Shanghai,China	Upper middle income	3941	903	94	769	0	40	0	0	Mixed	inpatient	0
142	Wangchuk et al.	2015	Norovirus GII.21 in children with Diarrhea, Bhutan	Emerg Infect Dis	2-10	12-12	Bhutan	Lower middle income	270	64	4	60	0	0	0	0	< 5	other	0
143	Jianxing et al.	2015	Etiology of diarrhea among children under the age five in China: Results from a five-year surveillance	Journal of Infection	1.1.2009	31.12.2013	China	Upper middle income	18266	2162	NA	NA	NA	NA	0	0	< 5	outpatient	0
144	D. M.;Ma Zhang et al.	2015	Clinical epidemiology and molecular profiling of human bocavirus in faecal samples from children with diarrhoea in Guangzhou, China	Epidemiol Infect	7-10	12.2012	China	Upper middle income	1128	9	NA	NA	NA	NA	0	0	Mixed	other	0

145	Jun;Shen Zhang et al	2015	Genotype distribution of norovirus around the emergence of Sydney_2012 and the antigenic drift of contemporary GII.4 epidemic strains	Journal of Clinical Virology	1.2012	12.2013	China	Upper middle income	846	139	22	117	0	0	0	0	> 5	outpatient	0
146	W.;Cui et al	2015	Clinical characteristics and molecular epidemiology of noroviruses in outpatient children with acute gastroenteritis in Huzhou of China	PLoS One	4.2013	4.2014	China	Upper middle income	1346	383	21	362	0	0	0	0	< 5	outpatient	0
147	Amna;Qureshi et al	2016	Genetic characterization of norovirus strains in hospitalized children from Pakistan	Journal of Medical Virology	4.2006	3.2008	Pakistan	Lower middle income	255	41	9	30	0	2	0	0	< 5	inpatient	1
148	Ruta;Patel et al	2016	Characterization of GII.4 noroviruses circulating among children with acute gastroenteritis in Pune, India: 2005-2013	Infection Genetics and Evolution	1.2005	12.2013	India	Lower middle income	1712	130	4	126	0	0	0	0	< 5	inpatient	1
149	Huan;Gao Mai et al	2016	GII.4 Sydney_2012 norovirus infection in immunocompromised patients in Beijing and its rapid evolution in vivo	Journal of Medical Virology	01-7-12	30-6-13	China	Upper middle income	131	9	0	9	0	0	0	0	> 5	outpatient	0
150	J et al	2016	Norovirus diversity in children with gastroenteritis in South Africa from 2009 to 2013: GII.4 variants and recombinant strains predominate	Epidemiology and Infection	4-09	121/2013	South Africa	Upper middle income	5950	837	54	350	NA	NA	0	0	< 5	inpatient	0
151	Casey L.;Webman et al	2016	Burden of Norovirus and Rotavirus in Children after Rotavirus Vaccine Introduction, Cochabamba, Bolivia	Am J Trop Med Hyg?	2010/01/03	2.2011	Bolivia	Lower middle income	272	133	1	90	0	0	71	21	< 5	inpatient	0
152	Ahenda	2012	Viral etiologies of diarrhea among children attending lwak missing hospital in asembo, western kenya	ASTMH	2007/01/01	2010/06/30	Kenya	Lower middle income	206	13	NA	NA	NA	NA	0	0	< 5	outpatient	0
153	Ballard	2013	A case - control study of norovirus induced acute diarrhea among army recruits in peru	ASTMH	2005/10/01	2011/07/01	Peru	Upper middle income	200	28	7	20	0	1	200	16	Mixed	outpatient	1
154	Chen	2012	Monitoring and analysis on pathogens of viral gastroenteritis in Nanhai of Foshan	Modern Preventive Medicine	2009/03/01	2010/02/01	china	Upper middle income	426	22	NA	NA	NA	NA	0	0	Mixed	unknown	0
155	Chen	2012	Molecular epidemiological study on norovirus in infants with diarrhea in Xiamen	Maternal and Child Health Care of China	2010/05/01	2011/04/20	china	Upper middle income	366	81	5	76	0	0	0	0	< 5	unknown	0
156	Deng	2009	[Comparative analysis on clinical manifestations for gastroenteritis caused by norovirus and rotavirus]	Journal of Epidemiology	2002/01/01	2006/12/31	china	Upper middle income	318	79	NA	NA	NA	NA	0	0	Mixed	inpatient	1
157	Fazeli	2010	Hospital based study of prevalence and genotyping of noroviruses and sapoviruses isolated from children with acute gastroenteritis referred to Masih Daneshvari hospital	Gastroenterolo g y and Hepatology from Bed to Bench	2006/02/01	2008/10/01	Iran	Upper middle income	47	10	0	8	0	0	0	0	Mixed	inpatient	1

158	Feng	2008	[Molecular epidemiological study on norovirus among children with acute diarrhea in Guangzhou]	Zhonghua Erke Zazhi	2006/12/01	2007/12/01	china	Upper middle income	1260	257	87	214		44	0	0	Mixed	unknown	1
159	M. E.;Martinez et al	2013	Molecular epidemilogy of norovirus strains in paraguayan children during 2004-2005. Description of possible new GII4 cluste	Journal of Clinical Virology	2004/01/01	2005/12/01	Paraguay	Upper middle income	378	161	68	93	0	0	0	0	< 5	inpatient	0
160	Gomez Santiago	2012	Molecular characterization of human calicivirus associated with acute diarrheal disease in mexican children	Virology Journal	2005/10/01	2006/12/01	Mexico	Upper middle income	414	128	NA	118	NA	NA	0	0	< 5	unknown	0
161	Hansman	2004	Genetic diversity of norovirus and sapovirus in hospitalized infants with sporadic cases of acute gastroenteritis in Chiang Mai, Thailand	J Clin Microbiol	2000/07/01	2001/07/01	Thailand	Upper middle income	105	9	3	3	NA	NA	0	0	< 5	inpatient	0
162	Jalilian	2012	Relative Frequency of Norovirus Infection in Children Suffering From Gastroenteritis and Referred to Aboozar Hospital, Ahvaz, Iran	Jundishapur Journal of Microbiology	2008/01/01	2009/12/31	Iran	Upper middle income	143	9	0	9	0	0	0	0	< 5	inpatient	0
163	Li	2012	[Molecular and epidemiological study on among children under 5 years old in Nanjing]	Chinese Journal of Experimental & Clinical Virology	2010/07/01	2011/06/01	china	Upper middle income	428	58	NA	NA	NA	NA	428	8	< 5	inpatient	0
164	Liao	2011	The characterization of novorius genotypes in children in Chongqing	Journal of Chongqing Medical University	2008/08/01	2009/07/01	china	Upper middle income	806	251	4	247	0	0	0	0	Mixed	unknown	0
165	Lin	2012	Detection and typing assay of norovirus in acute hospitalizations among children less than 5 years old from 2008 to 2009 in Lulong, Hebei province	Chinese J Exp Clin Virol	2008/10/01	2009/08/31	china	Upper middle income	325	37	NA	NA	NA	NA	0	0	< 5	inpatient	0
166	J.;Iturriza et al	2013	Campylobacter infection in children in malawi is common and is frequently associated with enteric virus coinfections	PLoS ONE [Electronic Resource]	1997/07/01	2007/06/01	Malawi	Low income	1659	192	NA	NA	NA	NA	0	0	< 5	inpatient	1
166	J.;Iturriza-Gomara et al	2013	Campylobacter infection in children in malawi is common and is frequently associated with enteric virus coinfections	PLoS ONE [Electronic Resource]	1997/01/07	2007/01/06	Malawi	Low income	282	20	NA	NA	NA	NA	0	0	Mixed	inpatient	1
167	Tung G. et al	2012	Acute Diarrhea in West African Children: Diverse Enteric Viruses and a Novel Parvovirus Genus	Journal of Virology	2008/11/01	2010/02/28	Burkina Faso	Low income	49	7	NA	NA	NA	NA	0	0	< 5	unknown	0
168	Mustafizur et al	2013	Norovirus Variant GII.4/Sydney/2012, Bangladesh	Emerging Infectious Diseases	2011/12/01	2012/12/31	Bangladesh	Lower middle income	795	254	46	168	10	30	0	0	Mixed	inpatient	0

169	M.;Goel-Apaza et al	2014	Multiple Norovirus Infections in a Birth Cohort in a Peruvian Periurban Community	Clinical Infectious Diseases	2007/06/01	2011/04/30	Peru	Upper middle income	1495	341	NA	NA	NA	NA	0	0	< 5	community	0
170	Tan et al.	2010	[Norovirus infection in adults with sporadic gastroenteritis during 2007-2008 in Nanning Municipal]	Zhongguo Jihua Mianyi	2007/01/01	2008/12/31	China	Upper middle income	696	183	3	180	0	0	0	0	>5	outpatient	0
171	Victoria, et al.,	2007	Prevalence and molecular epidemiology of noroviruses in hospitalized children with acute gastroenteritis in Rio de Janeiro, Brazil, 2004.	Pediatr Infect Dis J	2004/01/01	2004/12/31	Brazil	Upper middle income	318	65	1	27	NA	NA	0	0	Mixed	inpatient	0
172	Yao et al.	2012	Pathogens causing viral diarrhea in children aged <=5 years in Xinhuang, Hunan, 2009-2011	Disease Surveillance	2008/01/01	2011/12/31	china	Upper middle income	1063	94	NA	NA	NA	NA	0	0	< 5	inpatient	0
173	Yu et al.	2011	Molecular epidemiological survey on children with viral diarrhea in Tangshan	Maternal and Child Health Care of China	2007/01/01	2010/12/31	china	Upper middle income	1100	258	NA	NA	NA	NA	0	0	Mixed	unknown	0
174	Zeng et al.	2010	[Epidemiological surveillance of norovirus and rotavirus diarrhea among outpatient children in five metropolitan cities]	Zhonghua Erke Zazhi	2008/08/01	2009/07/01	china	Upper middle income	5091	1049	16	1036	0	3	0	0	Mixed	outpatient	0
175	Zhang et al.	2012	Surveillance of viral diarrhea in Shenzhen in 2010	Disease Surveillance	2010/01/01	2010/12/31	china	Upper middle income	925	186	NA	NA	NA	NA	0	0	Mixed	unknown	0
176	Zhang et al.	2009	Norovirus infection among diarrhea cases in Shenzhen, 2008	Disease Surveillance	2008/01/01	2008/12/31	china	Upper middle income	606	147	NA	NA	NA	NA	0	0	Mixed	unknown	0
177	Zheng et al.	2013	Detection of noroviruses from viral infectious diarrhea patients in Jiading, Shanghai	Chinese Journal of Zoonoses	2008/03/01	2010/02/28	china	Upper middle income	572	53	NA	NA	NA	NA	0	0	Mixed	inpatient	0
178	Zhuge et al.	2011	Detection and typing of caliciviruses from patients with acute diarrhea in Hangzhou area	Chinese Journal of Epidemiology	2009/07/01	2010/08/31	china	Upper middle income	920	195	25	170	0	0	0	0	Mixed	unknown	0

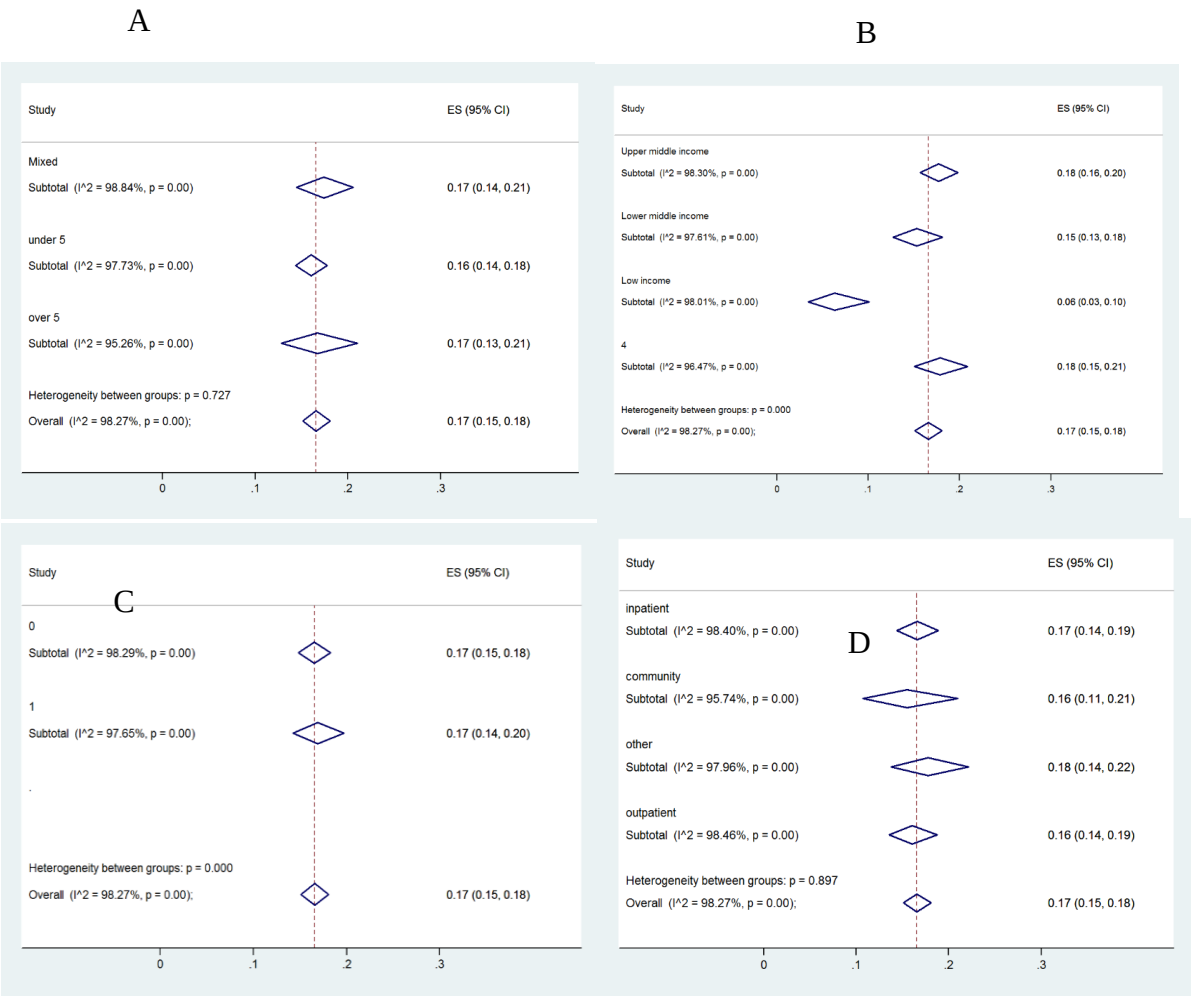

Fig 3.1S. NoV prevalence in patients with gastroenteritis, categorized into following groups: A, Age; B, Country classify; C, pandemic period; D, setting.

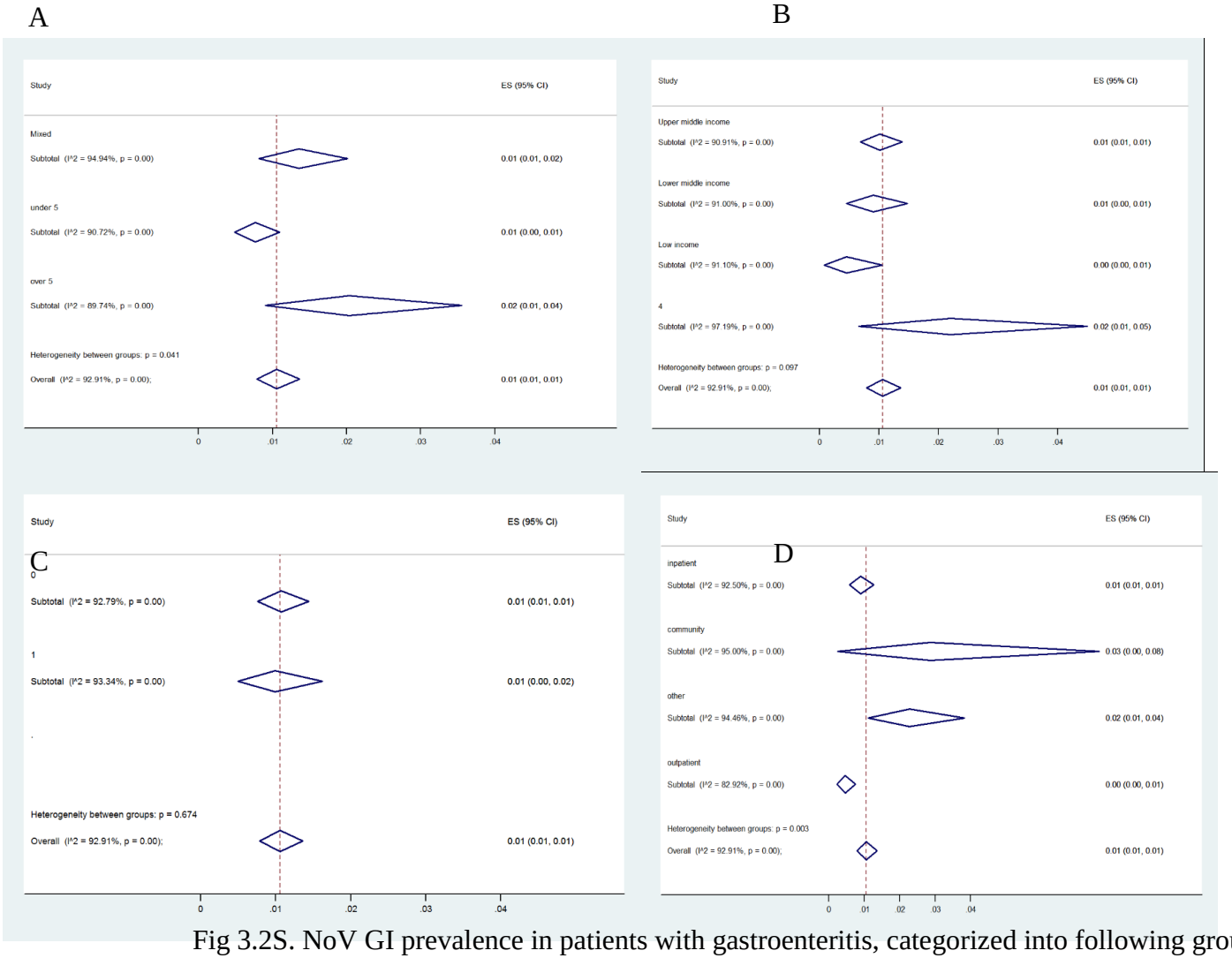

Fig 3.2S. NoV GI prevalence in patients with gastroenteritis, categorized into following groups: A, Age; B, Country classify; C, pandemic period; D, setting.

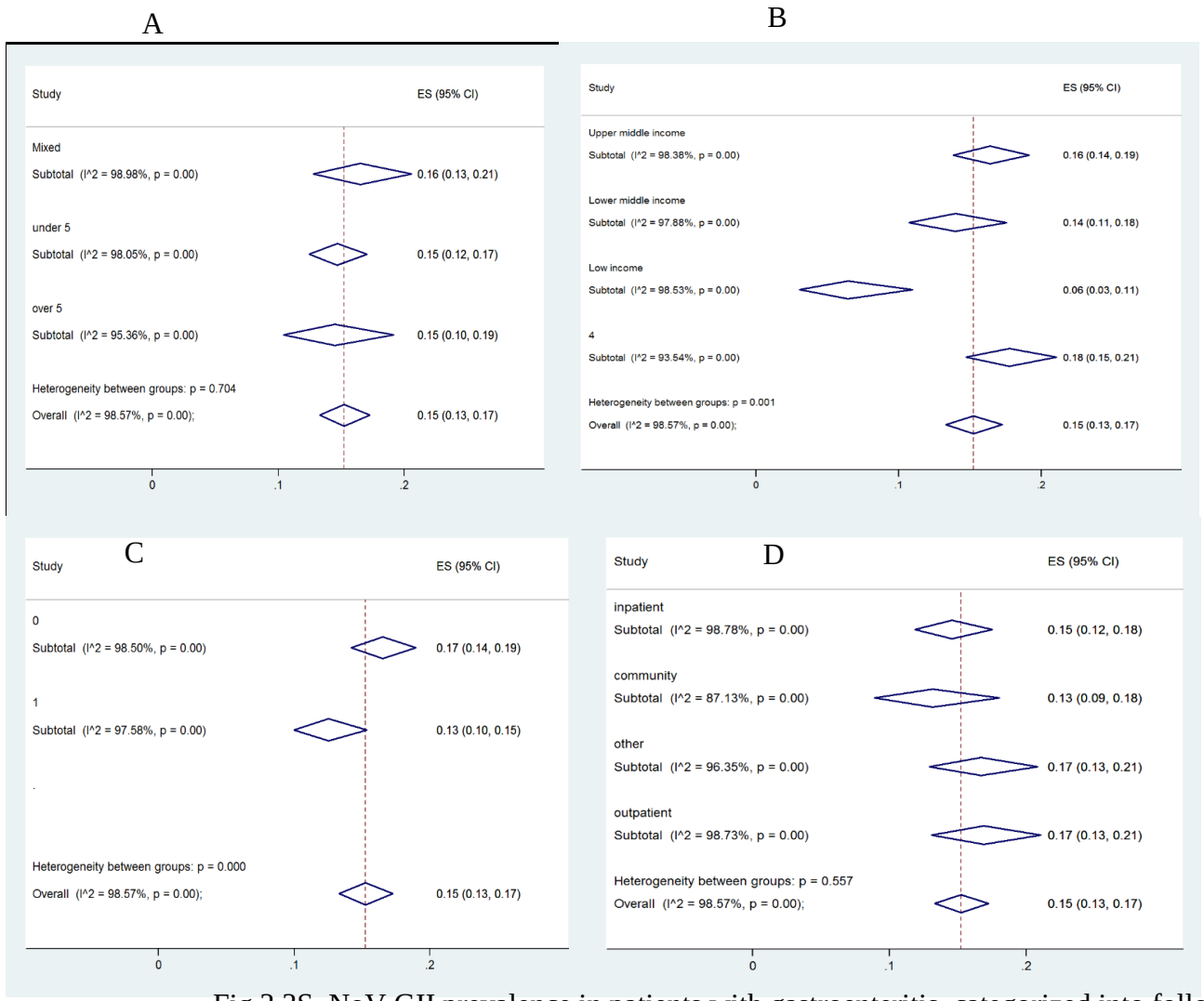

Fig 3.3S. NoV GII prevalence in patients with gastroenteritis, categorized into following groups: A, Age; B, Country classify; C, pandemic period; D, setting.

APPENDIX 2
APPENDIX FOR CHAPTER 4

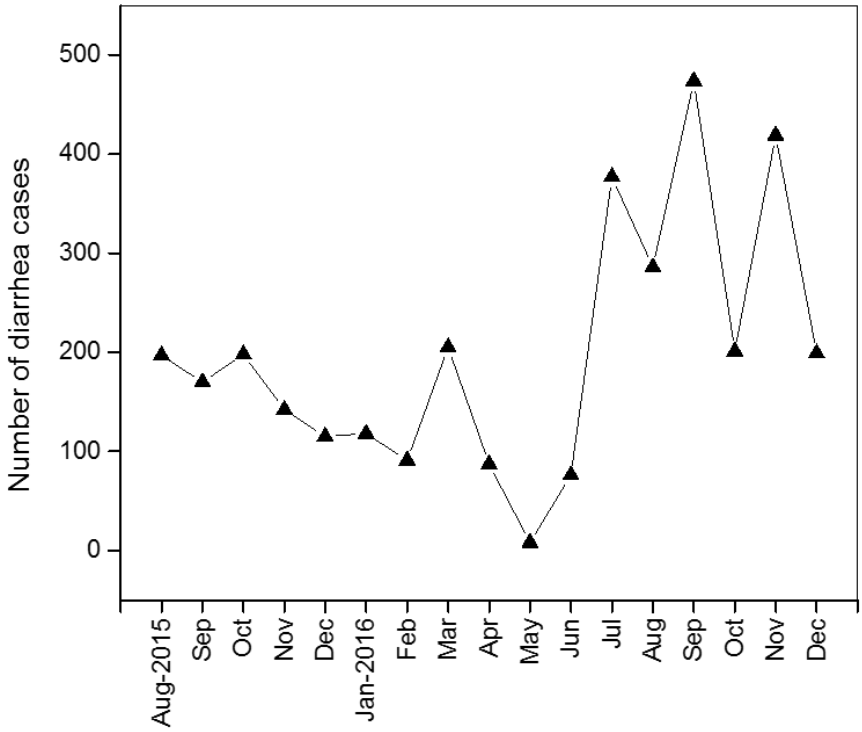

Fig. S4.1 The number of diarrhea cases caused by unknown pathogens in the Hue Central Hospital from August 2015 to December 2016. The cases were the sum of inpatients and outpatients.

Table S4. 1 Nucleotide primers and probes used in real-time PCR

Primer and probe	Sequence (5'-3')[a]	Conc [nM]	Reference
NoV GI			
COG1F	CGYTGGATGCGNTTYCATGA	400	Kageyama et al., 2003
COG1R	CTTAGACGCCATCATCATTYAC	400	
Ring1(a)-TP	FAM-AGATYGCGA/ZEN/TCYCCTGTCCA-IABkFQ	300	
Ring1(b)-TP	FAM-AGATCGCGG/ZEN/TCTCCTGTCCA-IABkFQ	100	
NoV GII			
COG2F	CARGARBCNATGTTYAGRTGGATGAG	400	Kageyama et al., 2003
COG2R	TCGACGCCATCTTCATTCACA	400	
ALPF	TTTGAGTCCATGTACAAGTGGATGCG	400	Japan Food Hygiene Association, 2004
RING2AL-TP	FAM-TGGGAGGGS/ZEN/GATCGCRATCT-IABkFQ	200	
MNV			
MKMNVF	CGGTGAAGTGCTTCTGAGGTT	400	Hata et al., 2011
MKMNVR	GCAGCGTCAGTGCTGTCAA	400	
MKMNV-TP	FAM-CGAACCTAC/ZEN/ATGCGTCAG-IABkFQ	300	

[a] Degenerate bases in primers and probes are as follows: Y, C or T; N, A or C or G or T; R, A or G; B, G or T or C.
FAM, 6-carboxyfluorescein; ZEN, ZEN internal quencher (Integrated DNA Technologies); IABkFQ, Iowa Black fluorescent quencher (Integrated DNA Technologies).

APPENDIX 3
APPENDIX FOR CHAPTER 5

Table S5.1. Optimal models for the dependent variables

Diseases	Optimal model	
	Vietnam	**Cambodia**
Non-Diarrhea	=-2.81 + 0.020*age +0.54*education	= 0.42 + 0.98* Area
Severe diarrhea	= -4.57 + 0.58 * education	= -2 + 0.82 * Area
Cough	=-1.58	= -0.12 + 0.88 * Area
Fever	=-0.57	= 0.16+ 1.11 * Area
Skin problems	= -0.1 – 0.02 * age	= -1.06 + 0.6 * Area
Eye problem	= -1.17 + 1.74 education	= -0.98

APPENDIX 4

IRB APPROVAL, LETTER OF EXPLANATION
INFORMED CONSENT FOR RESEARCH PARTICIPANT

Information Sheet for Participants

1. Title: Research on Utilization of various environmental information for solving health problems in South and Southeast Asian countries (GRENE-Ecohealth project)

2. Project leader:

Chiho Watanabe (Professor)

Department of Human Ecology, School of Medicine, University of Tokyo ○ Role: Supervision of overall project

3. List of other institutes for the project and their roles

○ Role: Field surveys to obtain primary data on population health and the environment; and collection of secondary data from research institutes and hospitals

Integrated Research System for Sustainability Science, University of Tokyo (Japan)
Department of Urban Engineering, University of Tokyo (Japan)
Graduation School of International Health Development, Nagasaki University (Japan)
Department of agriculture, Yamagata University (Japan)
Department of technology, Ehime University (Japan)
Research Institute for Humanity and Nature (Japan)

○ Role: Support for field surveys of staffs from Japan; and Application and examination of ethics in each local area if necessarily

Department of Public Health, Padjdajdaran University (Indonesia)
Institute of Ecology, Pajdajdaran University (Indonesia)
Health Research Department, National Institute of Public Health (Laos) Divina Amalin De La Salle University (Philippine)

BIOPHICS (Cener of Excellence for Biomedical and Public Health Informatics), Mahidol University (Thailand)

Faculty of Public Health, College of Medicine and Pharmacy, Hue University (Vietnam)

4. **Objectives**

Although most of Asian developing countries are currently experiencing rapid and stable economic growth, it may lead negative impact on human health; for example, environmental pollution, climate change including urban heat island phenomena, and social change such as population growth may all have negative effect on population health potentially. However, still very little is known about how these environmental and social changes affect people's health, mainly because of the lack of sufficient information on health and the environment in Asian developing countries.

As a part of the The Third Master Plan for Science and Technology, Japan, DIAS (Data Integration and Analysis System) project was launched from 2006. The project aims at creating an information storage infrastructure, including earth observation, simulation, and socio-economic data based on spatial information techniques, for public benefit applications.

The major purpose of our GRENE-EcoHealth project is to elucidate the relationship between environmental factors and some health/disease outcomes in cities in South and Southeast Asian cities, using rich data from DIAS which captures the environment and society in Asia. More specifically, we attempt to combine earth-observation data from DIAS with more conventional biomedical and/or preventive medicine approaches, such as filed surveys, to clarify the relationship between health and environmental change as well as social changes (lifestyle changes).

GRENE-EcoHealth project consists three subgroups and each has a different goal.

Group1. Atmospheric condition and health

Impact of urban heat and air pollution on the mortality and incidence of cardiovascular and respiratory diseases

Group 2. Vector-born disease

Impact of land use and weather conditions on the prevalence of vector-born disease such as dengue fever

Group 3. Floods and diarrhea incidence

Impact of urban flood on the outbreak of diarrhea which might be caused by infection to E.coli and Hepatitis E

5. Methodology

Our study areas are urban areas in South Asia and South-east Asia, including Bandung in Indonesia, Dhaka in Bangladesh, Bangkok in Thailand, Savannakhét province in Laos, Hue in Vietnam, and Manila in Philippine. In order to analyze the potential relationship between a set of environmental factors (physical factors, particularly) and a group of diseases that are likely affected by the factors, necessary data will be collected through two methods: field work to obtain primary data and secondary data collection.

We are proposing to collect following datset.

Group1. Atmospheric condition and health

(No primary data will be collected) (Secondary data collection)

1.1. Bandung (Indonesia)

Air pollution data will be obtained from LAPAN (Indonesian National Institute of Aeronautics and Space). We also collect climatic data (i.e. rainfall, humidity, wind speed, and air temperature) from meteorological stations in the city. Finally, health event data (i.e. cardiovascular and respiratory diseases) from several health centers will be obtained. For this purpose, permission from the health centers will be obtained.

1.2. Dhaka and neighborhood areas (Bangladesh)

Diarrheal patient data collected by icddr,b (International Centre for Diarrhoeal Disease Research) hospital will be obtained. We will also attempt to obtain HDSS dataset from icddr,b research institute.

Group 2. Vector-born disease

(Primary data collection)

2.1. We will set mosquito traps in city of Bandung to capture mosquitos. RNA analysis will be conducted using dengue fever viruses inside mosquitos.

2.2. In Savannakhét province in Laos we will collect information on local people's population characteristics (e.g. sex and age), date of birth, date and cause of death, and residential migration. This data is called Health and Demographic Surveillance System (HDSS).

2.3. We will set mosquito traps in city of Manila (Philippine) to capture mosquitos. RNA analysis will be conducted using dengue fever viruses inside mosquitos.

(Secondary data collection)

2.4. As secondary data, records on dengue fever patients including their population characteristics and symptoms of dengue fever at local hospitals in Bandung will be collected. For this purpose permission from the hospitals will be obtained.

2.5. As secondary data, records on malaria patients including their population characteristics and symptoms of the disease at local hospitals in Bandung will be collected. For this purpose permission from the hospitals will be obtained.

Group 3. Floods and diarrhea incidence

(Primary data collection)

3.1. We will conduct questionnaire survey in flood risk areas in Bandung to collect information on people's population characteristics, lifestyle, period of flood events and their severity, health events after the flood, and their measure against next potential flood.

3.2 Water samples from bodies of water including a river and side ditch will be collected to identify water pollution levels with chemicals, E.coli, and viruses such as hepatitis E.

3.3. We will conduct questionnaire survey in flood risk areas in Bandung to collect information on people's population characteristics, lifestyle, period of flood events and their severity, health events after the flood, and their measure against next potential flood.

(Secondary data collection)

No secondary data will be collected

——

Thank you very much for your participation.

As mentioned above, the objective of this study is to elucidate the relationship between environmental factors and some health/disease outcomes in cities in South and Southeast Asian cities, particularly those associated with climate change as well as social changes (lifestyle changes). For this purpose it would be highly appreciated if you could participate in our questionnaire survey. The questionnaire was designed to investigate our research questions, and we will ask your name in this questionnaire. However, we anomymize your data in keeping and analyzing it. Hence, the identity of participants will be disclosed.

If you agree to participate our survey, we will forward you a questionnaire sheet. We will ask you some questions on your population characteristics, lifestyle, and behavior against environmental risks and diseases.

The collected information is used for our research purpose only. Your answer would help us to understand how each of environmental risk can affect incidence of diseases.

This study is approved by the Ethics Committee of the University of Tokyo. Your participation for this study is entirely voluntary and you may refuse to answer any question if you choose or may withdraw your consent to participate at any time without penalty. The survey will take just less than 30 minutes. All the information we obtain will remain strictly confidential and your answer will never be identified.

You may ask any question about the study at this time and if you have further questions about this study, please do not hesitate to contact us.

Date: 19th / September/ 2015

Contact investigator

Chiho Watanabe (University of Tokyo), Principal investigator

Toru Watanabe (Yamagata University), Investigator in charge of the survey (Japan).
Nguyen Van Hung (Hue University), Investigator in charge of the survey (Vietnam).
Address and contact (Japan):

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Phone number: +81 (0) 3 5841 3530
Email address: chiho@humeco.m.u-tokyo.ac.jp
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06 Ngo Quyen street, Hue city, Thua Thien Hue province, Vietnam.
Tel: +84-914312951
Email address:drhhung@gmail.com

———————————————————————————————————————

資料 2(同意書・英語)

Informed Consent Form for Participants

To: The Dean of Graduate School of Medicine and Faculty of Medicine, The University of Tokyo

Study Title: **Research on Utilization of various environmental information for solving health problems in South and Southeast Asian countries (GRENE-Ecohealth project)**

Principal Investigator: Chiho Watanabe

I, after reading and having been explained to me the contents of this study, understand what is expected of me as a participant in the study.

I understand:
1. The purpose and procedure of the study
2. The consent of the questionnaire
3. That I will not be placed under any harm of discomfort
4. That I may refuse to answer any question if I don't want to answer
5. That I can withdraw from the study at any time without giving a reason
6. That I can withdraw from the study at any time (during or after study) without any harm or without in any way affecting the health service I receive
7. That any information I provide will be strictly treated in a confidential manner that I will not be identified in the reporting of the result

Date: / /

_____ Signature of the person who receive consent

３．同意撤回書

Retraction Form of Informed Consent

To: The Dean of Graduate School of Medicine and Faculty of Medicine, The University of Tokyo

Study Title: **Research on Utilization of various environmental information for solving health problems in South and Southeast Asian countries (GRENE-Ecohealth project)**

Principal Investigator: Chiho Watanabe

I once understood and agree to participate this study but after re-considering it, I retract my consent.

Date: / /

_____ Signature of the person who receive consent

資料4(質問票)

Research on Utilization of various environmental information for solving health problems in South and Southeast Asian countries

(GRENE-Ecohealth project)

Questionnaire

ID: _____ country/community: _____

Date of the interview:(dd/mm/yy) _____; Interviewer: _____
Participant
Name: _____

Use appropriate section depending on the site/survey.

A) Basic demographical data

Q1.What is your name?

Q2. What is your address?

Q3.What is your gender?

Q4.How old are you?

Q5.What is your highest level of education?

Q6.What is your religious?

Q7.What is your marital status?

Q8.What is your job?

Q9.Could you estimate roughly the annual income of the whole household?

Q10. Income of your spouse?

Q11.What is your family structure?

Q12. Home-ownership?

Q13. How long do you live here?

Q14. How much is your household expenditure?

B) Questions associated with water use and water quality

Q1.How much water do you drink in a day?

Q2.What is the water source for drinking water? Public tap water, public well, private well, or any other source? If you chose well water, how much is the depth of your well?

Q3.Do you boil the water before drinking?

Q4.Do your children drink the same water (in term of source, boiling)?

Q5.Do you use water to make ice? If yes, do you use the same water source to make ice?

Q6.During flooding period, do you use the same drinking water as usual?

Q7.During flooding period, do your children use the same drinking water as usual?

Q8.How often do you take a shower or bath per day? How long for each time?

Q9.How often do your children take a shower or bath per day? How long for each time?

Q10.What is the water source for shower?

Q11.What is the water source for bathing?

Q12.Do your children use the same water for shower or bathing?

Q13.During flooding period, do you shower or bath? If yes, do you use the same water for shower or bathing as usual?

Q14.During flooding period, do your children shower or bath? If yes, do your children use the same water source for shower or bathing as usual?

Q15.How often do you wash your face in per day?

Q16.What is the water source for washing your face?

Q17.How often do your children wash his/her face per day?

Q18.What is the water source for washing your children's face?

Q19.During flooding period, do you use the same water for washing face as usual?

Q20.During flooding period, do your children use the same water for washing face as usual?

Q21.Do you move furniture at the flood time? If yes, how long does it take?

Q22.At the end of flood, do you clean flood water on the floor?

C) Structured check list of 20 hygienic problems in relation to Dengue-like illness.

Q1. Existence of breeding sites for Mosquitoes: Actual

Q2 .Existence of breeding sites for Mosquitoes: Potential Such as abandoned tires, cans, shells of giant clams, cooler boxes, coconut shells, flower pots, boats, and puddles on the ground

Q3 .Existence of breeding sites for Rats: Actual

Q4 .Existence of breeding sites for Rats: Potential holes on the ground

Q5 .Existence of breeding sites for Flies: Actual

Q6 .Existence of breeding sites for Flies: Potential the leavings of a meal or dung

Q7 .Problems with facilities of Shower: Poor drainage

Q8 .Problems with facilities of Shower: No proper building

Q9 .Problems with facilities of Kitchen: Poor drainage

Q10. Problems with facilities of Kitchen: Attracts flies and rats

Q11 .Problems with facilities of Toilet: Lacking= no toilet

Q12 .Problems with facilities of Toilet: Full pit= no proper management of feces

Q13 .Problems with facilities of Toilet: no screen/lid on hole

Q14 .Problems with facilities of Toilet: Door not self-closing

Q15 .Trash site: Trash container without lid

Q16 .Trash site: Improper trash dump

Q17 .Water tank: Without lid or stand

Q18 .Conditions of plot: Pig sty

Q19. Conditions of plot: Cesspool or septic tank

Q20 .Conditions of plot: Yard: Overgrowth of grass

Q21 .Conditions of plot: Yard: Trash/litters found

Q22 .Experience of flood in year 2014

Q23 .Poor drainage after rain

Q24.Have any of your family members contracted Dengue-fever ever? Yes / No If Yes, who did and when it was?

What kind of treatment (including both modern and traditional ones) did he/she get?

If he/she went to hospital/clinic, which hospital/clinic was used?

(If two or more members did, please provide the answer one by one.)

D) Questions regarding the flood-associated health problems.

Q1. Have you experience flood in your house?

If yes, how frequent is your house affected by flood?

If yes, when did the most severe flood come to your house?

If yes, during the most severe flood, how deep water level did you observed in the first floor of your house?

If yes, how many days was your house inundated at that time?

Q2. Does your family have a boat for the flooding time?

Q3. If the first flood starts to be flooded, where does your family stay?

Q4.From how deep water level does your family give up staying in the first floor?

Q5. Have you experience diarrhea during or after flood?

Q6. If yes, how often you experience diarrhea per day?

Q7. Also how long did the symptoms exhibit?

E) Questions regarding mosquito-borne infectious diseases.

Q1. Have you been diagnosed with dengue fever before?

Q2. If yes, what symptoms did you experience?

Mẫu thỏa thuận đồng ý tham gia dành cho người tham gia nghiên cứu

Gửi đến: Trưởng khoa trường sau đại học Y khoa và khoa Y học, đại học Tokyo

Tên nghiên cứu: Nghiên cứu việc sử dụng các thông tin khác nhau về môi trường để giải quyết các vấn đề sức khỏe ở các nước Nam và Đông Nam Á (dự án GRENE-Ecohealth).

Nghiên cứu viên chính: Chiho Watanabe

Tôi, sau khi đọc kỹ và được giải thích về các nội dung của nghiên cứu này, hiểu những điều mong đợi/kỳ vọng khi tham gia vào nghiên cứu này.

Tôi hiểu:

1. Mục đích và quy trình nghiên cứu
2. Thỏa thuận đồng ý của bộ câu hỏi
3. Rằng tôi sẽ không chịu bất kỳ sự khó chịu nào
4. Rằng tôi có thể từ chối trả lời bất kỳ câu hỏi nào nếu tôi không muốn trả lời
5. Rằng tôi có thể rời nghiên cứu vào bất kỳ lúc nào mà không cần đưa ra lý do
6. Rằng tôi có thể rời nghiên cứu vào bất kỳ lúc nào (trong hoặc sau nghiên cứu) mà không chịu bất kỳ sự bất lợi nào hoặc không ảnh hưởng đến các dịch vụ chăm sóc sức khỏe của bản thân
7. Rằng bất kỳ thông tin tôi cung cấp sẽ được giữ bí mật , tôi sẽ không được nêu đích danh trong báo cáo kết quả

Ngày:……../……/……

……Chữ ký của người nhận thỏa thuận

Mẫu rút lại thỏa thuận đồng ý nghiên cứu

Gửi đến: Trưởng khoa trường sau đại học Y khoa và khoa Y học, đại học Tokyo

Tên nghiên cứu: Nghiên cứu việc sử dụng các thông tin khác nhau về môi trường để giải quyết các vấn đề sức khỏe ở các nước Nam và Đông Nam Á (dự án GRENE-Ecohealth).

Nghiên cứu viên chính: Chiho Watanabe

Tôi đã từng hiểu và tham gia nghiên cứu này nhưng sau khi xem xét lại, tôi rút lại sự đồng ý của mình

Ngày:……../……/……

APPENDIX 5
ENGLISH QUESTIONNAIRE VERSION

ID:_____

Date:_____

A. General information of household leader (18-60 years old)	
Q1.	**What is your name?**_____
Q2.	**What is your address?**_____
Q3.	**What is your gender?** □1. Male □2. Female
Q4.	**How old are you?** _____years old
Q5.	**What is your highest level of education?** □1. Illiterate □2. Elementary school □3. Secondary school □4. High School □5.Higher
Q6.	**What is your religious?** □1.Buddhist □ 2. Christian □ 3. Muslim □4. Other_____
Q7.	**What is your marital status?** □1.Married □2. Single □ 3. Divorced □ 4. Other_____
Q8.	**What is your job?**_____
Q9.	**Could you estimate roughly the annual income of the whole household?_____USD**
Q9-1.	**Family structure**

No	**Gender**	**Relationship to household leader**	**Age**	**Job**	**Average monthly Income**
1					
2					
3					
4					
5					
6					
7					
8					
9					
10					

B. Flood experience within 10 years	
Q10.	**Have you experienced flood in your house?** □1.Yes □2. No ⟶ **Go to Q11**
Q10-1.	**How frequent is your house affected by flood?** □1.More frequent □2.Twice a year □3.Every year □4.Once 2 year □5. More rare
Q10-2.	**When did the most severe flood come to your house?**_____
Q10-3.	**During the most severe flood, how deep water level did you observed in the first**

	floor of your house? To your □1.Ankle □2.Knee □3.Leg □4.Stomach □5.Chest □6. Higher than head
Q10-4.	**How many days was your house inundated at that time?**_____day
Q11.	**Does your family have a boat for the flooding time?** □1.Yes □2. No
Q12.	**If the first floor starts to be flooded, where does your family stay?** (Even if no in Q10, please ask them to imagine the flood situation) □1.Still in first floor □2.Loft □3.Second floor □4.Roof □5.Neighbor's house □6.Public facilities □7.Others (please define_____)
Q13.	**From how deep water level does your family give up staying in the first floor?** To your….. □1.Knee □2.Leg □3.Stomach □4.Chest □5.Other
Q14	**What type of floods have influenced on your family recently (10 years recently) ? (multiple choice was acceptable)** □1. Small flood (the peak of floods were lower than the average peak of flood in many years) □2. Medium flood (the peak of floods had reached the average peak of flood in many years) □3. Big flood (the peak of floods was higher than the average peak of flood in many years) □4. Very big flood (the peak of flood has seen high rarely) □5. History flood (the peak of flood was highest)
C. Feeling about infectious diseases	
Q15.	**What diseases/symptoms do you feel that happen more frequently during/after flood?** (Multiple answers are acceptable) □1.Diarrhea □2. Severe diarrhea (including cholera and dysentery) □3. Cough □4. Fever □5. Eye problem (in detail: _____) □6. Skin problem (in detail: _____) □ 7. Other disease/symptom (_____)
Q16.	**Have you experienced severe diarrhea, indicated by bloody diarrhea, vomiting, high fever, persistent diarrhea – lasts 14 days or longer and/or dehydration, in your life?** □ 1.Yes □ 2. No
Q17	**If yes, how many times have you got it?_____**
Q18	**How did you treat it?** □1.Did nothing □2. Used a medicine □3.Visited a doctor/health officer □4. Admitted to hospital/health facility □5. Other (please define_____)
Q19.	**How frequently do you suffer from non-severe diarrhea?** □1.Everyday □2. Once a week □3. Twice a month □4. Once a month □5. Once two months □6. Once three months □7. Twice a year □8. Once a year □9. Rare
Q20	**What month of the year you often has diarrhea ? Multiple answers are acceptable**

	_____
Q21.	**How do you treat it?** □1.Do nothing □2. Using a medicine □3.Visit a doctor/health officer □4. Other(_____)
Q22	**In case of using medicine, how much is roughly estimated cost for the medicine?_____**
Q23.	**How do you feel about the cost for medicine?** □1.Expensive (If checked, how much is the desirable cost?_____) □2.Reasonable
Q24	**In case of visiting doctor, how much is roughly estimated cost for the doctor's diagnosis?_____**
Q25	**How do you feel about the cost for the doctor's diagnosis?** □1.Expensive (If checked, how much is the desirable cost?_____) □2.Reasonable
Q26.	**Have your children experienced diarrhea?** (3 or more loose or liquid stools per day) □ 1.Yes □ 2. No
Q27.	**If yes, what symptoms were followed/including?** (multiple answer are acceptable) □1.Bloody diarrhea □2. Vomiting □3. High fever □4. Persistent diarrhea – lasts 14 days or longer □5. Dehydration □6. Other _____
Q28.	**If yes, how did you treat for him/her?** □1.Did not anything □2.Used medicine □3. Visited a doctor/health officer □4. Admitted to hospital/health facility □5. Other_____
Q29.	**If yes, how much did you pay for the treatment?_____**
Q30.	**If yes, how do you feel about the cost?** □1.Expensive (If checked, how much is the desirable cost?_____) □2.Reasonable
Q31.	**If yes, how many day were you absent from work for taking care your children?_____** days *(This question is only used for responders who have a job)*
Q32.	**Which is more severe or uncomfortable for you?** For example for "Dengue fever": Case a. If he/she feels that **severe diarrhea more** severe or uncomfortable than **dengue fever please circle "more". ---→** (More) /Same/Less Case b. If he/she feels **severe diarrhea less** severe or uncomfortable than dengue fever, please circle "less". -→More /Same/(Less)

Compared situation	**Severe diarrhea (Indicated by at least a symptom including bloody diarrhea, vomiting, high fever, persistent diarrhea – lasts 14 days or longer and/or dehydration)**	**Non-severe diarrhea**

Dengue fever	More/Same/Less	More/Same/Less	
Dental caries	More/Same/Less	More/Same/Less	
Broken bones (hand bones)	More/Same/Less	More/Same/Less	
Headache	More/Same/Less	More/Same/Less	
Low back pain	More/Same/Less	More/Same/Less	
Insomnia (cases)	More/Same/Less	More/Same/Less	
Pharyngitis	More/Same/Less	More/Same/Less	
Peptic ulcer	More/Same/Less	More/Same/Less	
Hepatitis B	More/Same/Less	More/Same/Less	
Gouty Arthritis	More/Same/Less	More/Same/Less	
Diabetes mellitus (cases)	More/Same/Less	More/Same/Less	
Hypertensive heart diseases	More/Same/Less	More/Same/Less	
Alcohol dependence syndrome	More/Same/Less	More/Same/Less	
Tuberculosis	More/Same/Less	More/Same/Less	
Asthma (cases)	More/Same/Less	More/Same/Less	
Malaria	More/Same/Less	More/Same/Less	

Q33. **Did you and your family lose any income cause by absence from work due to diseases/symptoms from the last year to now?**

□1.Yes □2. No

Q34. **How long were you and your family absent from work due to diseases/symptoms from the last year to now?**

No *(is the same as No in Q9-1)*	**Diseases/symptoms**	**Frequency during from <u>the last year to now</u>** (times)	**Number of days loss due to diseases/symptoms <u>per time</u>** *(__day)*	

APPENDIX 6

VIETNAMESE QUESTIONNAIRE VERSION

ID:_____

Ngày:_____

B.	……Chữ ký của người nhận **Thông tin chung về chủ hộ (Tuổi từ 18 đến 60)**
Q1.	**Họ và tên:_____**
Q2.	**Địa chỉ:_____**
Q3.	**Giới tính** □1. Nam □2. Nữ
Q4.	**Anh/chị bao nhiêu tuổi**?_____tuổi
Q5.	**Trình độ học vấn** □1. Mù chữ □2. Tiểu học □3. Trung học cơ sở □4. THPT □5. Mức cao hơn
Q6.	**Tôn giáo** □1.Phật Giáo □ 2. Công Giáo □ 3. Đạo Hồi □4. Khác_____
Q7.	**Tình trạng hôn nhân** □1.Kết hôn □2. Độc thân □ 3. Ly dị/ly hôn □ 4. Khác_____
Q8.	**Nghề nghiệp của anh/chị là gì?_____**
Q9.	**Anh/chị có thể ước tính thu nhập hàng năm của toàn bộ gia đình là bao nhiêu?_____**
Q9-1.	**Cấu trúc gia đình**

No	**Giới tính**	**Quan hệ với chủ hộ**	**Tuổi**	**Nghề nghiệp**	**Thu nhập trung bình hàng tháng**
1					
2					
3					
4					
5					
6					
7					
8					
9					
10					

C.	**Trải nghiệm về lũ lụt trong vòng 10 năm gần đây**
Q10.	**Nhà anh/chị đã từng bị lũ lụt chưa?** □1.Có □2. Không ⟶ **Chuyển đến Q11**
Q10-1.	**Mức độ thường xuyên nhà anh/chị bị lũ lụt là như thế nào?** □1.Thường xuyên □2. Hai lần một năm □3.Hàng năm □4. Hai năm một lần □5. Khác_____
Q10-2.	**Cơn lụt ảnh hưởng nặng nề nhất đến nhà anh/chị là vào năm nào?_____**
Q10-3.	**Lúc đó, mực nước mà anh/chị quan sát được ở tầng trệt của nhà mình là bao nhiêu?** Đến □1.Mắt cá chân □2.Đầu gối □3.Đùi □4.Bụng □5. Ngực □6. Cao hơn đầu

Q10-4.	**Lúc đó, nhà anh/chị bị ngập lụt trong bao nhiêu ngày?_____**ngày
Q11.	**Nhà anh/chị có thuyền/đò cho mùa lũ lụt không?** □1.Có □2. Không
Q12.	Nếu tầng trệt của nhà anh/chị bị lụt, gia đình anh/chị sẽ ở đâu? (Thậm chí **Q10** trả lời là không, vẫn tiếp tục hỏi cho họ tưởng tượng tình trạng lũ lụt) □1.Vẫn ở lại tầng trệt □2.Gác xếp □3.Tầng 2 □4.Mái nhà □5.Nhà hàng xóm □6. Các công trình cộng cộng □7.Khác (Vui lòng xác định_____)
Q13.	**Gia đình anh/chị sẽ di chuyển khỏi tầng trệt khi mực nước lụt dâng lên đến?** □1.Đầu gối □2.Chân □3.Bụng □4.Ngực □5.Khác
Q14.	**Theo anh/chị, những cơn lũ những năm gần đây ảnh hưởng đến gia đình thuộc loại nào ?(có thể trả lời nhiều đáp án) (10 năm trở lại đây)** □1.Lũ nhỏ *(là loại lũ có đỉnh lũ thấp hơn mức đỉnh lũ trung bình nhiều năm)* □2.Lũ vừa *(là loại lũ có đỉnh lũ đạt mức đỉnh lũ trung bình nhiều năm)* □3.Lũ lớn *(là loại lũ có đỉnh lũ cao hơn mức đỉnh lũ trung bình nhiều năm)* □4.Lũ đặc biệt lớn *(là loại lũ có đỉnh lũ cao hiếm thấy)* □5.Lũ lịch sử *(là loại lũ có đỉnh lũ cao nhất từng thấy)*
C. Cảm giác về bệnh truyền nhiễm	
Q15.	Những bệnh/triệu chứng nào anh/chị cảm giác/nghĩ rằng **sẽ xảy ra thường xuyên hơn** trong và/hoặc sau thời gian lũ lụt? (Có thể trả lời nhiều đáp án) □1.Tiêu chảy □2. Tiêu chảy nặng (*Chỉ định bởi tiêu chảy kèm theo ít nhất một triệu chứng bao gồm tiêu chảy ra máu, nôn mửa, sốt cao, tiêu chảy kéo dài - kéo dài 14 ngày hoặc lâu hơn và / hoặc mất nước*) □3. Ho □4. Sốt □5. Các vấn đề về mắt (chi tiết: - _____) □6. Các vấn đề về da (Chi tiết: _____) □ 7. Triệu chứng/bệnh khác (_____)
Q16.	**Anh/chị đã từng bị tiêu chảy nặng, bao gồm ỉa ra máu, nôn mửa, sốt cao, tiêu chảy kéo dài trên 14 ngày, bị mất nước chưa?** □ 1.Có □ 2. Không ⟶ **Chuyển tới Q18**
Q17.	**Nếu có, anh/chị đã bị bao nhiêu lần?_____lần**
Q18.	**Mức độ thường xuyên anh/chị bị tiêu chảy (không nặng) là như thế nào?** □1.Hằng ngày □2. Một lần/tuần □3. Hai lần/tháng □4. Một lần/tháng □5. Một lần/ 2 tháng □6. Một lần/3 tháng □7. Hai lần/năm □8. Một lần/năm □9. Khác(cụ thể_____)
Q19.	**Anh/chị thường xuyên bị tiêu chảy vào thời gian nào trong năm?** (Có thể trả lời nhiều đáp án) □1.Tháng 1 □2. Tháng 2 □3. Tháng 3 □4. Tháng 4 □5. Tháng 5 □6. Tháng 6 □7. Tháng 7 □8. Tháng 8 □9. Tháng 9 □ 10. Tháng 10 □ 11. Tháng 11 □ 12. Tháng 12
Q20.	Theo anh/chị, khi **so sánh** các bệnh dưới đây với **tiêu chảy nặng**, bệnh nào **là nghiêm trọng hơn hay khó chịu hơn** đối với anh/chị**?** ***Ví dụ: Sốt xuất huyết với tiêu chảy nặng*** *Trường hợp 1 (TH1): Nếu người tham gia cho rằng sốt xuất huyết* ***khó chịu/nghiêm trọng hơn*** *so với tiêu chảy nặng -----→* ***Ít hơn*** *TH2: Nếu người tham gia trả lời tiêu chảy nặng* ***ít*** *khó chịu/nghiêm trọng hơn so với sốt xuất huyết-----→* ***Nhiều hơn***

	Tình trạng (Bệnh/triệu chứng) so sánh	**Tiêu chảy nặng** **(*Chỉ định bởi tiêu chảy kèm theo ít nhất một triệu chứng bao gồm tiêu chảy ra máu, nôn mửa, sốt cao, tiêu chảy kéo dài - kéo dài 14 ngày hoặc lâu hơn và / hoặc mất nước*)**	
	Sốt xuất huyết	□1.Nhiều hơn □2. Giống nhau □3. Ít hơn	
	Sâu răng	□1.Nhiều hơn □2. Giống nhau □3. Ít hơn	
	Gãy xương cánh tay	□1.Nhiều hơn □2. Giống nhau □3. Ít hơn	
	Đau đầu	□1.Nhiều hơn □2. Giống nhau □3. Ít hơn	
	Đau thắt lưng	□1.Nhiều hơn □2. Giống nhau □3. Ít hơn	
	Mất ngủ	□1.Nhiều hơn □2. Giống nhau □3. Ít hơn	
	Viêm họng	□1.Nhiều hơn □2. Giống nhau □3. Ít hơn	
	Viêm loét dạ dày	□1.Nhiều hơn □2. Giống nhau □3. Ít hơn	
	Viêm gan B	□1.Nhiều hơn □2. Giống nhau □3. Ít hơn	
	Bệnh gút	□1.Nhiều hơn □2. Giống nhau □3. Ít hơn	
	Tiểu đường	□1.Nhiều hơn □2. Giống nhau □3. Ít hơn	
	Tăng huyết áp	□1.Nhiều hơn □2. Giống nhau □3. Ít hơn	
	Hội chứng nghiện rượu	□1.Nhiều hơn □2. Giống nhau □3. Ít hơn	
	Lao	□1.Nhiều hơn □2. Giống nhau □3. Ít hơn	
	Hen xuyễn	□1.Nhiều hơn □2. Giống nhau □3. Ít hơn	
	Sốt rét	□1.Nhiều hơn □2. Giống nhau □3. Ít hơn	

Q21. Theo anh/chị, khi so sánh **<u>tiêu chảy không nặng</u>** với các bệnh dưới đây, bệnh nào **là nghiêm trọng hơn hay khó chịu hơn** đối với anh/chị**?**

Ví dụ: So sánh với Sốt xuất huyết

Trường hợp 1 (TH1): Nếu người tham gia cho rằng tiêu chảy nặng ***khó chịu/nghiêm trọng <u>hơn</u>*** *so với sốt xuất huyết-----* ***→ Nhiều hơn***

TH2: Nếu người tham gia trả lời tiêu chảy nặng ***<u>ít</u>*** *khó chịu/nghiêm trọng hơn so với sốt xuất huyết-----* ***→ Ít hơn***

Bệnh/triệu chứng so sánh	**Tiêu chảy không nặng**	
Sốt xuất huyết	□1.Nhiều hơn □2. Giống nhau □3. Ít hơn	
Sâu răng	□1.Nhiều hơn □2. Giống nhau □3. Ít hơn	
Gãy xương cánh tay	□1.Nhiều hơn □2. Giống nhau □3. Ít hơn	
Đau đầu	□1.Nhiều hơn □2. Giống nhau □3. Ít hơn	
Đau thắt lưng	□1.Nhiều hơn □2. Giống nhau □3. Ít hơn	
Mất ngủ	□1.Nhiều hơn □2. Giống nhau □3. Ít hơn	
Viêm họng	□1.Nhiều hơn □2. Giống nhau □3. Ít hơn	
Viêm loét dạ dày	□1.Nhiều hơn □2. Giống nhau □3. Ít hơn	
Viêm gan B	□1.Nhiều hơn □2. Giống nhau □3. Ít hơn	
Bệnh gút	□1.Nhiều hơn □2. Giống nhau □3. Ít hơn	
Tiểu đường	□1.Nhiều hơn □2. Giống nhau □3. Ít hơn	
Tăng huyết áp	□1.Nhiều hơn □2. Giống nhau □3. Ít hơn	
Hội chứng nghiện rượu	□1.Nhiều hơn □2. Giống nhau □3. Ít hơn	
Lao	□1.Nhiều hơn □2. Giống nhau □3. Ít hơn	

	Hen xuyễn	□1.Nhiều hơn □2. Giống nhau □3. Ít hơn
	Sốt rét	□1.Nhiều hơn □2. Giống nhau □3. Ít hơn
Q22.	**Anh/chị đã xử lý như thế nào khi bị <u>tiêu chảy nặng</u>?** □1.Không làm gì □2. Sử dụng thuốc mua sẵn □3.Khám bác sĩ/nhân viên y tế □4. Nhập viện/cơ sở y tế □5. Khác (vui lòng xác định_____)	
Q23.	**Anh/chị xử lý như thế nào khi bị <u>tiêu chảy không nặng</u>?** □1.Không làm gì □2. Dùng thuốc □3. Thăm khám bác sĩ/nhân viên y tế □4. Khác (_____)	
Q23-1.	**Nếu dùng thuốc, anh/chị ước tính chi phí cho mua thuốc là mất bao nhiêu tiền_____?**	
Q24.	**Anh/chị nghĩ như thế nào về chi phí thuốc men đó?** □1.Đắt đỏ (Nếu đắt, chi phí anh/chị mong muốn là bao nhiêu tiền_____) □2.Hợp lý	
Q25.	**Nếu đi khám bác sĩ, anh/chị ước tính chi phí cho bác sĩ chẩn đoán là bao nhiêu tiền_____**	
Q26.	**Anh/chị nghĩ như thế nào về chi phí cho việc chẩn đoán của bác sĩ?** □1.Đắt đỏ (Nếu đắt, chi phí anh/chị mong muốn là bao nhiêu tiền_____) □2.Hợp lý	
Q27.	Con anh/chị đã từng bị tiêu chảy chưa? (ỉa phân lỏng từ 3 lần trở lên mỗi ngày) □ 1.Có □ 2. Không	
Q28.	**Nếu có, triệu chứng kèm theo tiêu chảy là gì? (có thể chọn nhiều đáp án)** □1.Ỉa ra máu □2. Nôn mửa □3. Sốt cao □4. Tiêu chảy kéo dài- kéo dài 14 ngày hoặc lâu hơn □5. Mất nước □6. Other _____	
Q29.	**Nếu có, anh/chị đã xử lý như thế nào?** □1.Không làm gì □2. Sử dụng thuốc mua sẵn □3.Khám bác sĩ/nhân viên y tế □4. Nhập viện/cơ sở y tế □5. Khác (vui lòng xác định_____)	
Q30.	**Nếu có, anh/chị đã chữa trị mất bao nhiêu tiền?_____**	
Q31.	**Nếu có, anh/chị nghĩ như thế nào về chi phí chữa trị đó?** □1.Đắt đỏ (Nếu đắt, chi phí anh/chị mong muốn là bao nhiêu tiền_____) □2.Hợp lý	
Q32.	**Nếu có, anh/chị đã nghỉ làm bao nhiêu ngày để chăm sóc cho con?_____** *Câu hỏi này chỉ dùng cho những người có nghề nghiệp)*	
Q33.	**Từ năm ngoái đến nay, anh/chị và gia đình có bị mất thu nhập vì nghỉ làm do bệnh tật gây ra không?** □1.Có □2. Không	
Q34.	**Anh/chị và gia đình phải nghỉ làm mất bao lâu vì bệnh tật từ năm ngoái đến nay?**	

STT *(giống số thứ tự trong mục Q9-1)*	**Bệnh/triệu chứng**	**Tần suất mắc bệnh/triệu chứng trong thời gian từ năm ngoái đến nay (____lần)**	**Số ngày mất vì bênh/triệu chứng mỗi lần (____ngày)**

APPENDIX 7

Academic achievements

Published and Accepted Journal Articles

1. Gia Thanh Nguyen, Jian Pu, Takayuki Miura, Hiroaki Ito, Shinobu Kazama, Yoshimitsu Konta, An Van Le, Toru Watanabe. Oyster contamination with human noroviruses impacted by urban drainage and seasonal flooding in Vietnam, *Food and Environmental Virology*, accepted.
2. Gia Thanh Nguyen, Kevin Phan, Ian Teng, Pu Jian, Toru Watanabe. A systematic review and meta-analysis of the prevalence of norovirus in cases of gastroenteritis in developing countries. *Medicine*. 96:40(e8139), 2017.
3. Erika Ito, Hiroaki Ito, Jian Pu, Nguyen Thanh Gia, Toru Watanabe. Accumulations of Pepper mild mottle virus and Aichi virus in oysters and their potentials as indicator of Norovirus contamination. *Journal of Environmental Engineering Research*, 72(7), 2016 (in Japanese).
4. Dang Thi Anh Thu, Nguyen Thanh Gia, Nguyen Huu Nghi, Nguyen Dinh Minh Man. Musculoskeletal disorder among carpenter in center Vietnam. *Journal of Communication Medicine*. Page: 24-33, 2014.
5. Gia Nguyen Thanh, Srirat Lormphongs, Nantaporn Phatrabuddha. Factors related to hookworm infection among farmers in Phu Xuan sub-district, Phu Vang district, Thua Thien Hue province, Vietnam. *The Public Health Journal of Burapha University.* Vol 8 No 2; Page: 101-111, 2013.
6. Hoang Trong Si, Nguyen Thanh Gia, Nguyen Van Hop, Thuy Chau To, Nguyen Dang Giang Chau, Le Thi Huynh Nhu. Organochlorine pesticides and polychlorinated biphenyls in human breast milk in the suburbs of Hue city, Vietnam. *Journal of Science, Hue University*. No 61; Page: 393-401, 2010.

Submitted Journal Articles

7. Andreas Älgå, Dang Thi Anh Thu, Dell Saulnier, Gia Thanh Nguyen, Johan von Schreeb. Hope for the Best, Prepare for the Worst – An assessment of flood preparedness at primary health care facilities in central Vietnam. *PLOS Currents*, under review.

8. Daniel B Scherman, Adam A Dmytriw, Gia Thanh Nguyen, Nhan Thi Nguyen, Nana Tchantchaleishvili, Christoph Griessenauer, Christopher Ogilvy, Ajith Thomas, Justin M Moore, Kevin Phan. Shunting, Optic Nerve Sheath Decompression and Dural Venous Stenting for Medically Refractory Idiopathic Intracranial Hypertension: Systematic Review and Meta-analysis. *Worldneurosurgery*, under review.

Submitted Book Chapters

1. Gia Thanh Nguyen, Jian Pu and Toru Watanabe. Flood and food as potential carriers of disease in urban and rural areas. In: Health in ecological perspectives in the Anthropocene. *Springer Publishing*. Edited by Chiho Watanabe and Toru Watanabe, under review.

Presentations in international conferences

1. Gia Thanh Nguyen, Jian Pu, Hiroaki Ito, An Van Le, Toru Watanabe. Oyster contamination with human norovirus impacted by urban drainage and flood in central Vietnam. *7th IWA-ASPIRE Conference 2017*, Kuala Lumpur, Malaysia, September 11-14, 2017.

2. Toru Watanabe, Gia Thanh Nguyen, Jian Pu, Kensuke Fukushi. Fecal contamination of foods cultivated at downstream of urban area affected by seasonal flood. Two cases in central Vietnam. *2nd International Forum on Sustainable Future in Asia*, Bali, Indonesia, January 26-28, 2017.

3. Gia Thanh Nguyen, Hiroaki Ito, Jian Pu, Le Van An, Toru Watanabe. One-year monitoring of oyster contamination with human norovirus in a lagoon affected by urban drainage in Vietnam. *12th International Symposium on Southeast Asian Water Environment*, Hanoi, Vietnam, November 28-30, 2016.

4. Gia Thanh Nguyen, Yuri Kanaya, Jian Pu, Toru Watanabe. Modified disability weights for diarrhea diseases based on feeling of flood-affected people in Asian developing countries. *Water and Environment Technology Conference (WET2016)*, Tokyo, Japan, August 27-28, 2016.

5. Susanti Withaningsih, Rina Febriani M.I.L., Gia Thanh Nguyen, Toru Watanabe. Risk and feeling to infectious Diseases in Frequently-Flooded Area of Bandung. *3rd JSPS/GRENE-EcoHealth joint International Symposium on Development of International Network on Health Risk Assessment in Urban Area*, Bali, Indonesia, March 11-12, 2016.

Presentations in domestic conferences

1. Gia Thanh Nguyen, Kanaya Yuri, Toru Watanabe, Feeling to infectious diseases in flood-affected areas: A cross-sectional survey in central Vietnam, 平成27年度土木学会東北支部技術研究発表，平成２８年３月５日，盛岡市.

2. Gia Thanh Nguyen, Dang Thi Anh Thu, Nguyen Huu Nghi, Nguyen Dinh Minh Man. Musculoskeletal disorder among carpenter in center Vietnam. The Conference for Young Scientists of Hue University 2014, Hue city, Vietnam, January 2014.

3. Andreas Älgå, Dang Thi Anh Thu, Gia Thanh Nguyen, Johan von Schreeb. Flood preparedness in several primary health care stations of Thua Thien Hue Province, central Vietnam. International Symposium on Water and Health in Urban Area, Hue city, Vietnam, December 15-17, 2013.

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