

Seasonal Variations in Norovirus Outbreaks Among Passengers and Crew members on Cruise Ships in Europe: A narrative review



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ABSTRACT

Background: Norovirus, a Ribonucleic acid (RNA) virus of the family Caliciviridae, is a human enteric pathogen that is recognized as the leading cause of acute gastroenteritis in children and adults. Worldwide norovirus- Acute gastroenteritis (AGE) outbreaks can occur in a variety of settings, including cruise ships. Seasonal variations may have impacts in outbreaks patterns and outcomes. Commonly, outbreaks of norovirus can occur in restaurants, at tourist destinations and especially aboard cruise ships. Climate change has been shown to influence the incidence and spread of norovirus infections. Understanding norovirus outbreak impact factors is important in preventing major disease and complication, and maintaining the tourism industry among major cruise ships. This study aims to investigate how seasonal variations impact on the spread of infection caused by norovirus outbreaks among passengers and crew on cruise ships in Europe.

Methods: A comprehensive literature review was performed to identify relevant literature to explain how seasonal variations impact the spread of norovirus aboard cruise ships in Europe. The target population for this study was passengers and crew aboard luxury cruise liners in Europe. The inclusion criteria for articles included studies published between 01 January 2006 and 31 December 2017 that addressed norovirus outbreaks and the target population. A total of ten (10) articles were reviewed by one author, and five articles were selected for final review and appraisal process.

Results: Overall, a total of 112 norovirus outbreaks on 73 vessels had been reported in the studies between January 2006 and December 2013, for a total of 2,848 days at risk. There were a total of 880586 passengers and crew aboard the vessels during this period of time. Norovirus outbreaks showed a peak in summer months. Among the 5 studies analyzed, 4 norovirus outbreaks occurred in seasonal (October-April) and 41 outbreaks occurred in off seasonal months (May-September).

Conclusions: This study confirms the importance of identifying the seasonal data norovirus outbreaks on cruise ships in Europe and, at the same time, confirms and compiles the data on a large number of norovirus outbreaks in Europe on passenger' cruise ships. At present, more data

and published studies are needed to confirm an increase in norovirus outbreaks on cruise ships due to seasonal variation and climate.

Key words: Cruise ships, gastroenteritis, viral pathogens, outbreak, epidemiology diseases in EU

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I gained profound, practical knowledge on the importance of the Public Health field through the cruise ship industry. I applied Public Health fundamentals, including infectious disease surveillance, and epidemiological investigation techniques to protect the passengers and crew members from infectious diseases to prevent disease during outbreaks and other health events

I must express my very profound gratitude to my parents for always believing in me and encouraging me throughout my years of studying the Master. This accomplishment would not have been possible without them.

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LIST OF ABBREVIATIONS

AGE	Acute Gastroenteritis
AR	Attack Rate
CASP	Critical Appraisal Skills Programme
CDC	Centers for Disease Control and Prevention
DNA	Deoxyribonucleic acid
EM	Electron microscopy
EU	European Union
IR	Incidence Rate
Kb	Kilobases
ORF	Open Reading Frame
PCR	Polymerase Chain Reaction
PICO	Population, Intervention, Comparison, Outcome
PRISMA	Preferred Reporting Items for Systematic Reviews and Meta-Analysis
RNA	Ribonucleic acid
RT-PCR	Reverse Transcription Polymerase Chain Reaction
TDC	The thoroughness of environmental disinfection cleaning
UV	Ultra Violet
WHO	World Health Organization

CHAPTER 1

INTRODUCTION

Cruise ship tourism has become increasingly popular in the last couple of decades (H Lun, Ressler, Ferson, & A White, 2017). Worldwide, 10 million people travel on cruise ships every year to a variety of destinations (Contribution of Cruise Tourism to the Economies of Europe 2015 Edition, 2015). The European cruise ship industry represents the second largest source of tourism revenue in the world and is also the second most popular cruise destination in the world, receiving some 5.85 million cruise passengers in 2013 (Contribution of Cruise Tourism to the Economies of Europe 2015 Edition, 2015)

Among cruise ships, the most common infectious disease is acute gastroenteritis. Gastroenteritis usually presents as a mild diarrheal illness with additional symptoms of nausea and vomiting. According to “WHO”, gastrointestinal infections are produced by a wide variety of enteropathogens including bacteria, viruses and parasites (WHO,2016). The main family groups causing viral gastroenteritis are: 1) Adenoviridae (adenovirus), 2) Astroviridae (astrovirus), 3) Caliciviridae (noroviruses and sapovirus) 4) Reoviridae (rotavirus) (Arias et al., 2010).

Norovirus accounts for more than 50% of all foodborne and waterborne diarrheal illness, with the rest being attributable to multiple infectious agents that include enteric bacteria (diarrheagenic, *Escherichia coli*, *Shigella* spp, *Campylobacter* spp), and parasites (*Giardia lamblia*, *Entamoeba histolytica*, *Cryptosporidium parvum*)(Simons, Pike, Hulseberg, Prouty, & Swierczewski, 2016)

Norovirus is notorious for causing outbreaks on cruise ships all over the world, affecting hundreds of passengers and as a result leads to considerable morbidity and economic costs for the cruise ship industry (H Lun et al., 2017). Extreme weather variations, such as cold winters, heat waves, floods and temperatures changes are important impact factors for transmission of infectious agents and their survival in natural phenomena (Rohayem, 2009). This can be applied to the transmission of norovirus on cruise ships.

In Europe, significant seasonal variations are an important epidemiological factor on the spread norovirus, with most outbreaks being reported between October and April (Verhoef et al., 2008). The aim of this research is to determine if there is an association between seasonal variations in Europe and the incidence rate of the norovirus (seasonality) among passengers and crew on commercial cruise ships.

CHAPTER 2

LITERATURE REVIEW

BACKGROUND OF NOROVIRUS

Norovirus is an infectious virus. It is recognized as leading cause of AGE worldwide, and it the most common cause of outbreaks (Communicable Disease Network Australia, 2010). Norovirus outbreaks have been reported in a variety of settings nursing homes, hospitals, daycares, university dormitories and schools. This is due to fact that the virus is uniquely suited to areas of close living quarters, shared dining, and low environmental maintenance (Wikswø et al., 2011).

The virus when is visualized by the electron microscopy (EM) is 26 to 34nm in diameter; small, round, with and amorphous surface and ragged outer edge (Communicable Disease Network Australia, 2010).

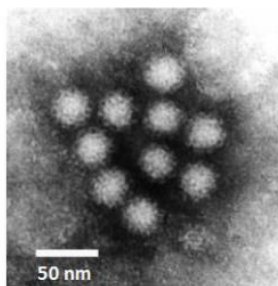


Figure 1 Transmission electron microscopy of Norovirus particles. (Morillo & Tavares Tlmenetsky, 2011)

HISTORY

Human norovirus, previously known as Norwalk virus, was first described by Zahorsky in 1929 as “winter vomiting disease” due to its seasonal predilection and the frequent preponderance of patients which vomiting as a primary symptom (Robilotti, Deresinski, & Pinsky, 2015). However, the agent was not identified until 1968, when an outbreak led to the identification of the virus affected 50% of the students at an elementary school in Norwalk,

Ohio, US, and manifested primarily as nausea, vomiting, diarrhea, and low grade fever (Communicable Disease Network Australia, 2010).

In 2002, norovirus became the official genus name, following further investigations of the viral taxonomy. These investigations of the virus have allowed for a rapid recognition of the causative agent in outbreaks and has changed the understanding of the clinical significance and epidemiology of the infection (Communicable Disease Network Australia, 2010).

TAXONOMY

Norovirus is one of four genera within the Caliciviridae. The other three genera are Lavovirus, Vesivirus and Sapovirus. Sapovirus and Norovirus are the two genera that cause AGE in humans. In 2005, a new virus classification system was established based on the full-length capsid amino acid sequences. Since this classification system was revised, noroviruses have been categorized into at least 6 genogroups (GI-GVI) and further divided into over >40 genotypes (Robilotti et al., 2015). Norovirus continues to evolve in response to the selective pressure exerted by the human immune system (Robilotti et al., 2015).

Only the GI, GII and GIV genogroups have been reported to cause disease in humans, with GII being the most common (90% of the cases), followed by GI (5-10%), whilst GIV is especially rare (Eden et al., 2014). GII, genotype 4 (GII,4), is the cause of 70% of all norovirus infections and has been responsible for all six pandemics of norovirus-associated gastroenteritis (Slmone Guadagnucci Morillo & Tavares Tlmenetsky, 2011). Norovirus is highly infectious due to a combination of low infecting dose (DI $50 < 20$ virus particles), high virus excretion level in humans (10 to 10 copies of RNA per gram of feces), and extended excretion after clinical recovery (Slmone Guadagnucci Morillo & Tavares Tlmenetsky, 2011)

VIROLOGY

Noroviruses (family, genus Norovirus) are genetically diverse group of single stranded ribonucleic acid (RNA) (Communicable Disease Network Australia, 2010), and one of the main characteristics is that they are not enveloped (Hardy, 2005). The RNA genome of the virus

consist of approximately 7700 nucleotides and encodes three open reading frames (ORF) (Hardy et al., 2005). The open reading frame ORF1 encodes for nonstructural polyproteins including a helicase, protease and RNA-dependent RNA polymerase. The ORF2 encodes a viral capsid protein and the ORF3 a small structural protein (Slmone Guadagnucci Morillo & Tavares Tlmenetsky, 2011).

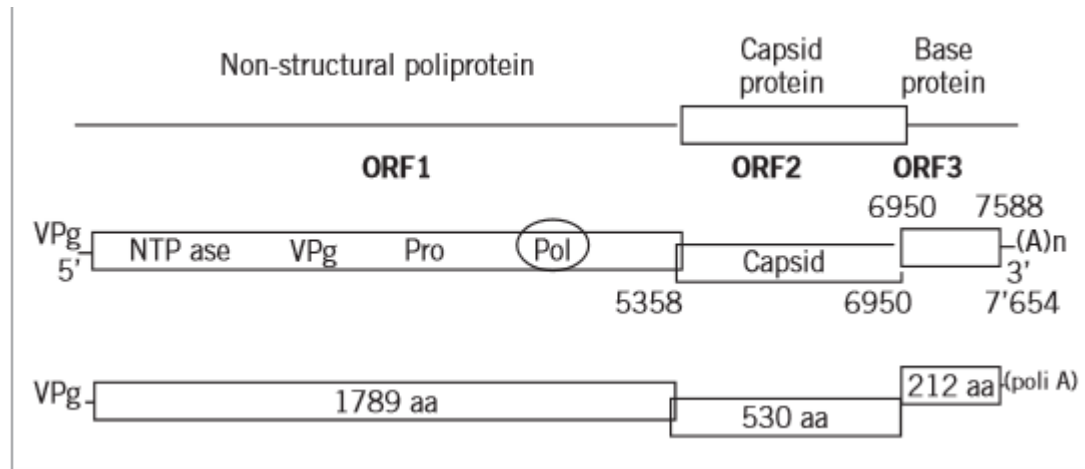


Figure 2 Norovirus genomic organization (Slmone Guadagnucci Morillo & Tavares Tlmenetsky, 2011)

PATHOGENESIS, REPLICATION, CLINICAL FEATURES AND TRANSMISSION

Human caliciviruses cause infection predominantly by fecal-oral routes. Virions are stable in acid and they can survive after passing through the stomach which has significant impact on its infectivity. Noroviruses have the following characteristics, 1) are highly infectious due to the combination of low infecting dose 2) have a high virus excretion level among humans, and 3) continue to have an extended excretion after clinical recovery (Slmone Guadagnucci Morillo & Tavares Tlmenetsky, 2011).

Norovirus has an incubation period of 12 to 48 hours, with symptoms lasting anywhere from 12 to 60 hours. Norovirus infection commonly begins with an acute onset of nausea abdominal cramps, vomiting, and myalgia (Ong, 2013). Non-bloody diarrhea is the most common symptom, occurring in over 90% of the cases (Ong, 2013).

The transmission of the virus can occur through the following general routes: food, water and via person- to- person contact. Norovirus spreads from person to person when , there is direct contact with feces or vomitus from infected people, through contact with contaminated fomites and aerosolized droplets usually from the infected person vomiting (Ong, 2013).

Table 1 Characteristics that influence norovirus transmission

Characteristics	Description
Environment stability	Norovirus particles may be infectious for 2 weeks on environmental surfaces and less than 2 months in water
Vomiting	Vomiting appears to be a particularly effective route of Norovirus spread. Vomiting events may occur and lead to direct transmission (when in public) as well environmental contamination from vomit droplets
Transmission through multiple routes	Norovirus are transmitted via the fecal-oral route and vomit-oral route, and through a number of specific modes, including foodborne, waterborne, environmental and direct person to person spread

Adapted from (Barclay et al., 2014)

CRUISE SHIP INDUSTRY AND NOROVIRUS OUTBREAKS

According to the “State of the art: public health and passenger ships” a cruise ship is defined as a leisure vessel which transport thousands of passengers and crew and that visit multiples places (Mouchtouri et al., 2010). A typical cruise ship carries about 2000 passengers and 800 crew, and the largest ships capacities in excess of 5000 passengers and 2000 crew, and the average cruise lasts longer than 6 days (Marshall, Morris, & Unwin, 2016). This is more than enough time for the incubation of a virus.

Cruise ships were found to be the third most common setting (16%) of norovirus outbreaks in the US, during the period 1996 to 2004 (Mouchtouri et al., 2010). Despite the strict sanitation measures on ships, a large number of norovirus outbreaks have been reported in Europe. Between 1st January and 5th of July 2006, 42 reported AGE outbreaks on 13 different cruise ships sailing in European waters were confirmed or suspected to be caused by Norovirus (Mouchtouri et al., 2010). Almost 1,500 AGE cases were reported among passengers and crew, but no common source was determined (Lopman et al., 2004). A recent investigation in the United States, reported that during 2008 to 2014, a total of 133 cruise ships have reported acute gastroenteritis outbreaks, of which 97% were caused by norovirus (Freeland, Vaughan, & Banerjee, 2016).

CRUISE SHIP CONDITIONS FOR THE EMERGENCE OF NOROVIRUS OUTBREAKS

On a cruise ship the ideal conditions for the emergence of new outbreaks of norovirus exist due to a common source of infection from consumption of food and drinks contaminated before loading or after unloading and the semi-closed environment of the ships facilitates transmission from person- to- person, through the fecal-oral route and environmental contamination aerosols arising from vomiting episodes (Bert et al., 2014). This is demonstrated by a recent investigation of an outbreak on cruise ships that showed an association between illness while on a cruise and shared bathrooms with ill cabin mates who vomit (Mouchtouri et al., 2017)

Investigations on cruise ships have confirmed that norovirus outbreaks can be transmitted person- to- person by different group activities onboard like bingo, at movies theater, live shows and simple handshake. A recent study of handshaking behaviors on cruise ships showed that on board there is a high likelihood that passengers will become familiar with another through handshaking, a common practice among most cultures (Dahl, 2016). Handshakes are an important vehicle of transmission of norovirus because while on-board a cruise ship passenger commonly touch communal surfaces like doorknobs, elevator buttons and railings that can be easily contaminated with the virus after handshake.

OUTBREAK DEFINITION AND ACUTE GASTROENTERITIS CASE

The “WHO” defined an outbreak of gastroenteritis on board cruise ships as an increase in the number of cases of AGE above the number normally occurring in that ship over a defined period of time (WHO, 2011). Furthermore, according to the “CDC”, a reportable case of AGE is defined as a period when there are three or more episodes of loose stools in a 24 hours or vomiting and at least one of the following symptoms: 1) one or more episodes of loose in a 24 hour period 2) abdominal cramps 3) headache 4) muscle aches 5) fever $> 38^{\circ}\text{C}$ (100°F) (Centers for Disease Control and Prevention (CDC), 2002). Any case meeting this description should be reported to the AGE surveillance log on board of the ship.

As described in the “Guidance for the Management of Norovirus Infection in Cruise Ships”; fecal samples (6 and 10) should be taken from a representative sample of those affected collected within 48 hours and submitted to a reputable laboratory for a viral and bacterial analysis (Guidance for the Management of Norovirus Infection in Cruise Ships, 2007). After this analysis an outbreak would be declared if and when greater than three percent ($n=3\%$) of the ship’s passengers or crew members reported AGE to the ship infirmary (Wikswa et al., 2011). The outbreak is confirmed if norovirus was detected in stool samples from greater than two ($n\geq 2$) patients and was considered probable if norovirus was detected in only one ($n=1$) patient’s sample or in more than one environmental sample (Verhoef et al., 2008).

For reporting purposes in Europe, two different thresholds should be used. An initial report should be prepared and sent to the competent authority at ports, when the percentage of reportable AGE cases reaches two percent ($n=2\%$) or more among passengers or two percent ($n=2\%$) or more among crew. A second report should be sent when the number of reportable AGE cases reaches three percent ($n=3\%$) or more among passengers or three percent ($n=3\%$) or more among crew (European Manual for Hygiene Standards and Communicable Diseases, 2016).

The following diagram shows an example of calculating AGE levels to determine an outbreak. This data is taken from a study that analyzed syndromic surveillance data of AGE on cruise ships in order to determine AGE cases-characteristics, incidence and attack rates for cruises with and without outbreaks (Mouchtouri et al., 2017). In this graphic the authors

described the number of AGE cases on board and day which an AGE is probable in the following ports: Cuba, Cyprus, Dominican Republic, Egypt, France, Greece, Italy, Spain and the United Kingdom.

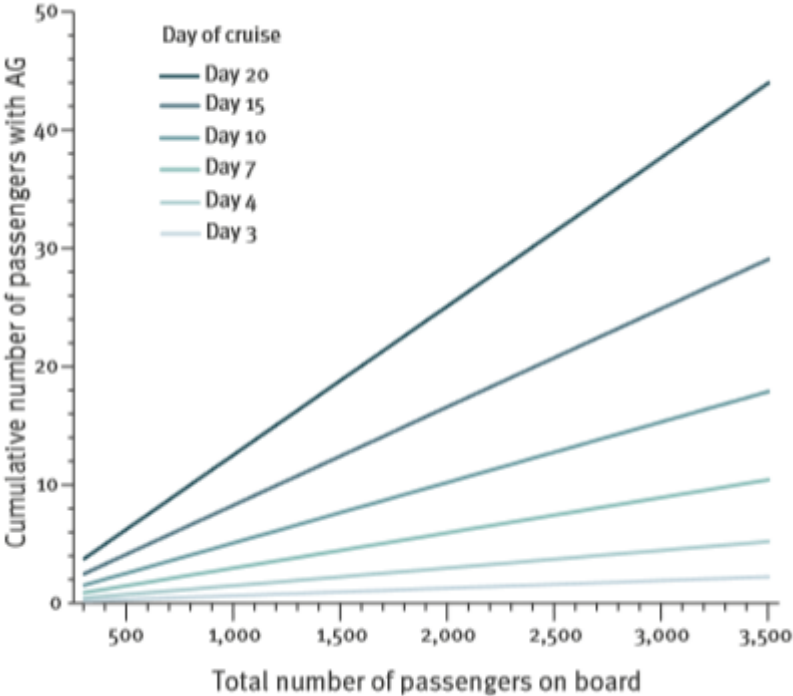


Figure 3 AGE levels needed to declare an outbreak based of days of the cruise of an outbreak (Mouchtouri et al., 2017).

Each cruise vessel should have clearly defined thresholds for determining when there are elevated numbers of probable cases on board. These guidelines should outline specifically when to trigger control measures. The thresholds and guidelines depend on the number of passengers, the length of the cruise and on the itinerary destination of the vessel (EU SHIPSAN ACT joint action, 2016). According to the “European Manual for Hygiene Standards and communicable Disease Surveillance on Passengers Ships”, if AGE surveillance data has shown that if within the first two days of the voyage the incidence of AGE appears to be two cases per every 1,000 passengers, then the probability of having a larger outbreak is 6.82 % (EU SHIPSAN ACT joint action, 2016).

Number of GI cases reported in every 1,000 passengers	Probability of an outbreak (PPV)	ROC Area (95 % CI)
First two days of the cruise		
1	4.63 %	0.743 (0.555-0.932)
2	6.82 %	
3	6.68 %	
4	11.07 %	
First three days of the cruise		
1	3.50 %	0.873 (0.718-1.000)
2	7.61 %	
3	14.64 %	
4	22.76 %	
5	23.10 %	

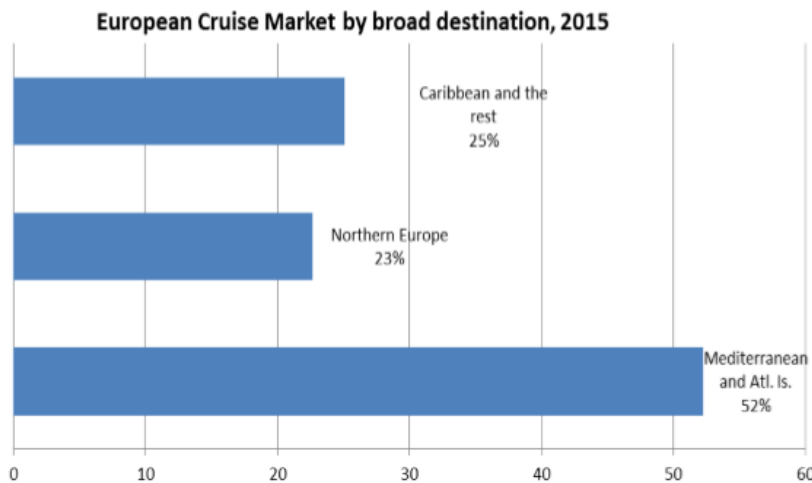
Table 2 Outbreak probability on cruise ships based on the incidence rate within a specified number of days at sea (EU SHIPSAN ACT joint action, 2016)

COMMUNICABLE DISEASE SURVEILLANCE

A recent European Manual for Hygiene Standards and Communicable Diseases Surveillance on Passengers Ships explained that the surveillance of communicable diseases on board cruise ships is important for assessing the burden of communicable diseases and to allow the early detection and management of outbreaks (EU SHIPSAN ACT joint action, 2016). The surveillance of communicable diseases on board cruise ships includes the AGE log. The data in the AGE log for each cruise contains information about cruise dates, name of all ports of call, number of passengers and crew members on board, numbers of travelers with AGE symptoms and their age and sex, their cabin numbers, meal seat in the dining room and the date and time of symptoms onset. (Mouchtouri et al., 2017). Without the AGE log, cruise ships would not be able to enable timely of preventive measures through early detection of outbreaks and at the same time to collect baseline information by season and specific itineraries, in order to determine thresholds for outbreak detection (EU SHIPSAN ACT joint action, 2016).

CRUISE SHIP INDUSTRY IN EUROPE

The cruise industry in Europe is a dynamic source of economic activity providing benefits to virtually all industries and countries throughout Europe (Contribution of cruise Tourism to the economies of Europe 2015 Edition), bringing every year thousands of passengers from different European ports. In 2014, an estimated 6.4 million European residents booked cruises, reporting the majority of the cruise ships visited ports in the Mediterranean, the Baltic Sea and other European regions. The cruise ship industry in Europe transported 29 million



passengers that were recorded through European ports during 2014 (Contribution of cruise Tourism to the economies of Europe 2015 Edition). Outbreaks of norovirus results in a considerable economic cost to the cruise industry.

EUROPEAN CRUISE MARKET

The European cruise market has grown in the recent years. In 2014, the cruise industry had a significant economic impact on Europe that generated direct expenditures of over €16. billion (Contribution of cruise Tourism to the economies of Europe 2015 Edition). The calculation of these direct expenditures includes in the following trends in spending and maintenance €4.55 billion was spent for the construction and maintenance of cruise ships 2), €6.97 billion was spent by cruise lines in support of their operations, €3.64 billion in cruise passenger, €1.48 billion in wages and salaries for crew members, including benefits (Contribution of cruise Tourism to the economies of Europe 2015 Edition)

Industry ①	Expenditures € Million	Jobs	Compensation € Million
Agr., Mining & Constr. ②	€20	171	€4
Manufacturing	€7,988	42,559	€1,707
Non-durable Goods	€2,023	6,744	€246
Durable Goods	€5,975	35,815	€1,461
Wholesale & Retail Trade	€812	11,150	€222
Transportation & Utilities	€3,696	21,902	€805
Hospitality ③	€407	6,421	€143
Financial and Business Services	€1,576	13,419	€463
Personal Services & Govt.	€647	9,336	€266
Subtotal	€15,156	104,958	€3,610
Cruise Line Employees	€1,480	64,873	€1,480
Grand Total	€16,637	169,831	€5,090

Table 3 Economic comparison of European industries including Cruise Line Employees ,2014
(Contribution of cruise Tourism to the economies of Europe 2015 Edition)

A recent report in Europe from 2014, showed that during this year there were 42 cruise lines located in Europe which operated 123 cruise ships. Outside of Europe, 8 more cruise lines participated in the European cruise market (Contribution of cruise Tourism to the economies of Europe 2015 Edition). Among the data presented that in the same year a total of 152 cruise ships were active in Mediterranean waters. These ships carried a potential 3.60 million passengers on 2,478 cruises. Comparatively, among Northern Europe cruise networks, a total of 101 cruises ships were active and carried approximately 1.51 million passengers on 1,184 cruises (Contribution of cruise Tourism to the economies of Europe 2015 Edition).

EUROPEAN CRUISE PASSENGERS

German passengers dominated the European market (Contribution of Cruise Tourism to the Economies of Europe 2015 Edition, 2015). During 2014, a total of 1,8 million passengers were sourced from Germany among cruise ships with itineraries reaching destinations around

the globe. However, European destinations are still the most common (Contribution of cruise Tourism to the economies of Europe 2015 Edition).

Country	Passengers	Share of Total
Germany	1,771,000	27.7%
UK/ Ireland ①	1,644,000	25.7%
Italy	842,000	13.2%
France	593,000	9.3%
Spain	454,000	7.1%
Norway	176,300	2.8%
Switzerland	143,000	2.2%
Austria	122,000	1.9%
Netherlands	109,000	1.7%
Sweden	78 800	1.2%
Belgium	73,000	1.1%
Denmark	37,700	0.6%
Finland	12,200	0.2%
Other Europe	331,000	5.3%
Total	6,387,000	100.0%

Table 4 Number of passengers and country share of total passengers in the cruise industry, 2014 (Contribution of cruise Tourism to the economies of Europe 2015 Edition).

NOROVIRUS SEASONALITY

A study of seasonality of infectious diseases have defined seasonality as “a periodic surge in disease incidence corresponding to seasons or other stereotype calendar periods, and it is characterized of many infectious diseases of public health importance” (Fisman, 2007). Seasonal patterns of norovirus diseases is associated with host-behavioral, host immune function and environmental conditions, such as humidity, temperature cycles, rain patterns and winds have been associated with the increased incidence of norovirus (Ahmed et al., 2013).

Recent investigations of Norovirus, have shown that norovirus outbreaks in Europe for general population are directly correlated to seasonal variations; with a peak incidence during

the winter time (between October and April) and with a common recognition as “winter vomiting disease” (Rohayem et al., 2009) (Ahmed, Lopman, & Levy, 2013). A study of the emergence of new norovirus variants on spring cruise ships have defined the primary norovirus surveillance season as running from May through April of the next year to include a full winter season (Verhoef et al., 2008).

Changes in population behaviors has long been a suspected cause of seasonality in norovirus (Rohayem, 2009). A recent article regarding norovirus outbreak on cruise ships, explained the correlation between the transmission and the host behavior (passengers behavior) (Wikswow et al., 2011). Passengers’ behavior is defined in different functional areas including using the toilet, eating, sightseeing, and the types of touch surfaces.

Another important aspect that relates to the seasonal susceptibility of humans to norovirus infection during winter is low vitamin D levels. During this period of the year, diminished UV radiation reduces vitamin D synthesis within the body (Rohayem, 2009).

For gastrointestinal infections caused by norovirus that are also transmitted by aerosols (vomitus), increased humidity may increase the transmission rate of the virions. This means that changes in the humidity and temperature may influence in the norovirus resistance, transmission and virulence (Rohayem, 2009). Existing published reviews of climate change impacts on food and waterborne diseases, reported that norovirus infections in general population in Germany occurred throughout the year, with a seasonal rise from October to March and a typical peak in November to January (Semenza et al., 2012).

PRINCIPLES OF EPIDEMIOLOGY IN PUBLIC HEALTH

Public Health surveillance involves the systematic collection, analysis, and interpretation of the morbidity and mortality data essential to the planning, implementation and evaluation of public health practice (WHO, 2008). Measuring disease frequency is important in characterizing and understanding outbreaks (WHO, 2008). According to the CDC, rates are the most common way of measuring the frequency of disease outbreaks in a population (WHO, 2008). In the case of cruise ships diseases, such as AGE the rate is calculated as:

$$\frac{\textit{Total number of new cases of AGE in the population at risk}}{\textit{Total number of passengers and crew in the population at risk}}$$

The numerator is new cases of AGE during a specific period-usually during the itinerary of the cruise. While the dominator is the number of individuals on board the cruise ships during that period (CDC Centers for Disease Control and Prevention, 2012). Rates can be expressed per 100, 1000 or per 10000.

Incidence rate or person-time rate is a measure of the frequency of new cases of a disease that incorporates time directly into the denominator (WHO, 2008). The denominator represents the total time passengers and crew members were at risk norovirus infection (CDC Centers for Disease Control and Prevention, 2012). The incidence rates are calculated:

$$\frac{\textit{Total number of new AGE cases reported during a specified period}}{\textit{Total person – time at risk for norovirus exposure}}$$

In outbreak settings, the term attack rate is often used as a synonym for risk. The attack rate is most commonly used in disease outbreak investigations and is a key factor in the formulation of hypotheses of disease origin (WHO, 2008). This number assesses the risk of getting AGE during a specific cruise itinerary (WHO, 2008). Sometimes it is difficult to calculate rates because the population at risk is not known. In that situations, the distribution of cases may help in formulating hypotheses (WHO, 2008).

The attack rates are calculated as:

$$\frac{\textit{Total number of new AGE cases reported}}{\textit{Total number of passengers and crew on board the cruise ship}}$$

THE EPIDEMIC CURVE

The epidemic curve in the Outbreak setting in a cruise ship is defined as a histogram with the number of Norovirus cases on the y-axis and the date of onset of illness on the x-axis (WHO, 2008). The epidemic curve has the following characteristics 1) confirming the existence

of an epidemic 2) identifying the mode of transmission 3) determining the possible period of exposure and or the incubation period of the disease (WHO, 2008).

The shape of an epidemic curve can determine if the epidemic pattern is due to a point of source, common source or person-to-person spread. In the case of common-source Outbreak, a single source of pathogen results in exposure of persons at one point in time (point source), at several points in time (intermittent common source) or over a continuous period (continuous common source) (WHO, 2008). If there is a single source of pathogen but exposure is not confined to one point in time, the epidemic is either an intermittent common-source or a continuous common-source Outbreak (WHO, 2008). In the case of a propagated epidemic the transmission may occur directly (person-to-person spread) or via an intermediate host (WHO, 2008)

CHAPTER 3

METHODOLOGY

A search was conducted to determine if there is an association between seasonal variations in Europe and the incidence rate of norovirus (seasonality) among passengers and crew on commercial cruise ships that were active between 01 January 2006 and 31 December 2017. The research question was formed utilizing the PICO tool (Ayodele, Krallinger, & Mori, 2012).

The PICO model is frequently used in clinical and epidemiological research; where P refers to the target patient population, I refers to intervention used among the population being studied, C refers to comparison studies, and , O refers to possible outcome (Santos, Pimenta, & Nobre, 2007). The PICO method is a useful tool to divide a research question into searchable components and to construct several kinds of research questions (Santos et al., 2007).

Acronym	Definition	Description
P	Patients or problem	Passengers or crew members
I	Intervention	Cruise ship in Europe
C	Control or comparison	Seasonal patterns
O	Expect result	Norovirus seasonality

Table 5 The four components of the PICO strategy with the research question (Santos et al., 2007)

A literature review was conducted to search for articles regarding seasonal variations among norovirus outbreaks on cruise ships in Europe using PubMed. A narrative review was chosen for use in this study because it established relevant scientific information in regards to the research question. The search was conducted between 01 January 2006 and February 2018. The research was carried using terms “norovirus disease outbreaks” AND “cruise ships” AND “seasons” AND “Europe”. Searches results were restricted to articles published from 01 January 2006 and 31 December 2017.

SELECTION OF THE ARTICLES

A screening mechanism was designed to determine whether the articles were eligible for review under specified inclusion criteria. Inclusion criteria were defined as, studies that had explicit geographical restriction, further defined as vessels cruising in EU waters. Inclusion criteria was further defined as studies regarding norovirus outbreaks that were confirmed and detected in stool samples from greater than (2>) patients (passengers or crew) aboard the vessel. Studies of illness in person related to military ships, cargo ships, private sailing boats, and fishing vessels were excluded. The research included papers written in all language (German, France, Spanish, English and Portuguese) and published between 01 January 2006 and 31 December 2017.

Inclusion Criteria	Exclusion Criteria
Published results from academic journal on PubMed with open access	Studies on military ships, cargo ships, fishing, or those on private sailing vessels
Norovirus outbreaks cases sailing in EU waters	Title
Norovirus outbreaks that were confirmed and detected in stool samples from >2 patients (passengers or crew)	Abstract
Full text, any language	Discussion

Table 6 Inclusion and exclusion criteria

Assessment of the retrieved studies was performed in three parts. In the first stage, research results were reviewed individually to select for full-text reports only with open access. Among those with full-text articles available, titles were read and using the inclusion and exclusion criteria, articles was further assessed via their abstract. In the final stage, the research articles were read and assessed for information regarding the research question. Data extraction from the included papers was performed independently, using a standardized data extraction form that included: year of the outbreak, season, geography (place of the outbreak), numbers of

cruise ships involved, numbers of individuals affected, case definition used, age and sex of the passengers and crew involved and any incidence rates or information that could be used to calculate incidence rates.

For this review five articles were included because they met the inclusion criteria (Mouchtouri et al., 2017; Vivancos et al. 2010; Carling et al.2009; Verhoef et al. 2008; Verhoef et al. 2008a). It is important to mention that in the study from Mouchtouri et al., 2017 the five seagoing cruise ships visited multiple countries such as: Cuba, Cyprus, Dominican Republic, Egypt, France, Greece, Italy, Portugal, Spain and the United Kingdom. It did not describe in the review where exactly were the outbreaks. On the other hand, in the article from Carling et al.2009 did not mention or describe the place of the norovirus outbreaks. However, this studies were included in the research.

This research used long-term seasonal patterns to study norovirus outbreaks. This is defined as the normalized proportion of norovirus outbreaks by calendar month of study period (Ahmed et al., 2013). For this research, norovirus surveillance on cruise ships was defined in the northern hemisphere in two periods: 1. Off-seasonal: summer months May, June, July, August and September 2. Seasonal: winter months October, November, December, January, February, March, April (Verhoef et al., 2008).

PRISMA 2009 Flow Diagram was adopted and used as the methodological framework. The flow diagram depicts the flow or information through the different phases of a systematic review. It maps out the number of records identified , included and excluded, and the reason for exclusions (Liberati et al., 2009). The PRISMA protocol was adopted based on to demonstrated the rational for including or excluding articles (Ayodele et al., 2012).

In this review five articles were excluded covered nonhomogeneous locations (one cruise ship study in the Caribbean (Marshall et al., 2016), one Foodborne Outbreak case in Brazil (Morillo et al., 2017), two gastrointestinal data base in European without abstract and full text (Enserink, 2006) (Depoortere, Takkinen, & ECDC Norovirus expert group, 2006), and one systematic review of Norovirus Outbreaks on commercial cruise ships in Italy (Bert et al., 2014). The systematic review study was excluded because most of the norovirus outbreaks occurred on commercial cruise ships after January 1990 and the overwhelming majority of the articles

referred to cruise ships that sailed in the USA. Although some of the included articles described the same outbreak.

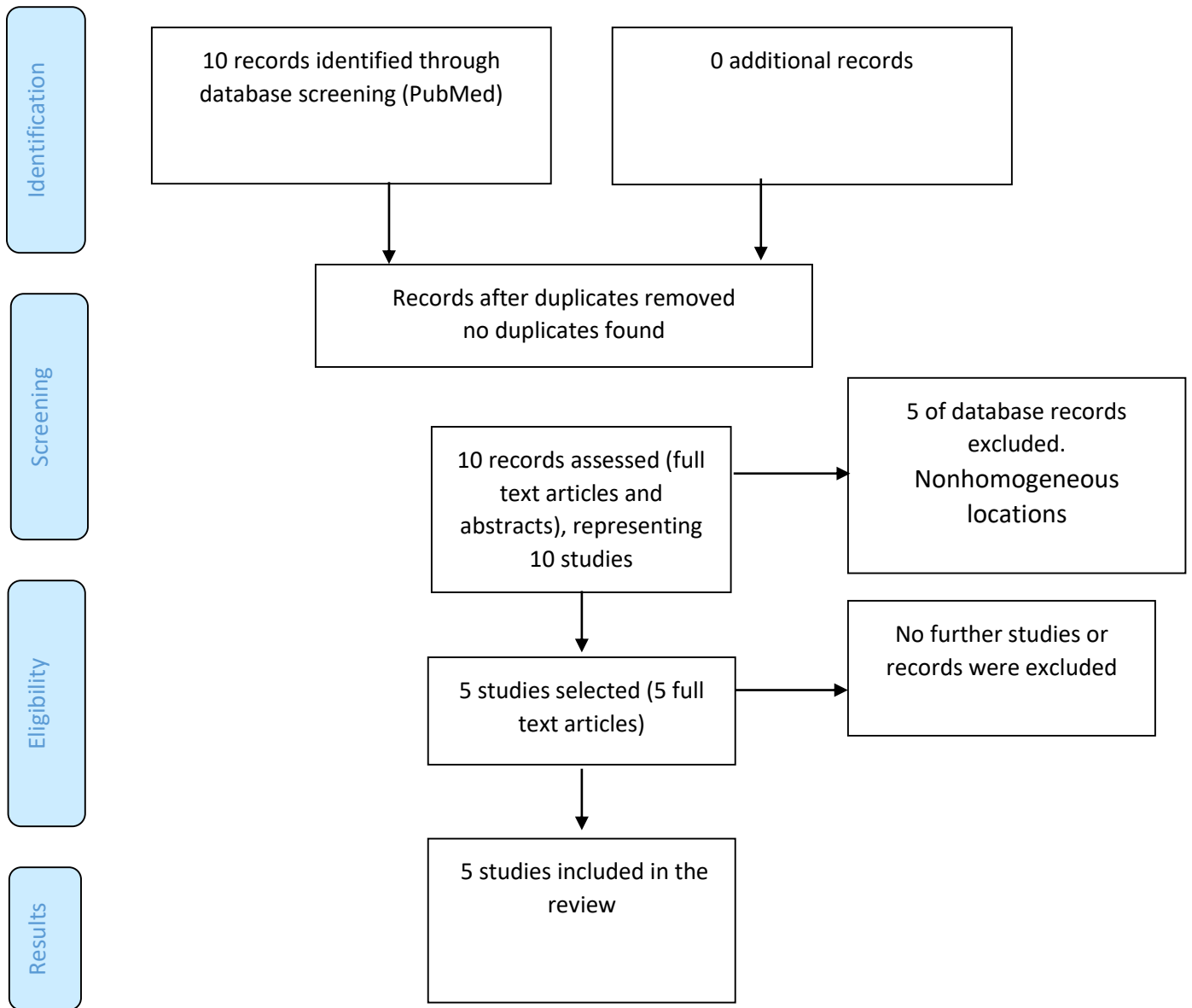


Figure 4. Sample PRISMA (Móher 2009)

Author(s)	Study	Season	Type of the study	Place of the Outbreak	Date of the paper publication
Mouchtouri et al., 2017	Gastroenteritis outbreaks on cruise ships: contributing factors and thresholds for early outbreak detection.	Not reported	Retrospective study	Cuba, Cyprus, Dominican Republic, Egypt, France, Greece, Italy, Portugal, Spain and the United Kingdom	09 Nov 2017
Vivancos et al. 2010	Norovirus Outbreak in a cruise ship sailing around the British Isles: Investigation and multi-agency management of an international outbreak.	October	Retrospective cohort study	British Isles, calling at ports in England, Guernsey, Republic of Ireland, Northern Ireland, Scotland and the Netherlands.	03 March 2010
Verhoef et al. 2008a	Multiple exposures during a norovirus outbreak on a river-cruise sailing through Europe, 2006.	Summertime	Retrospective cohort study	Several European countries	29 February 2008

Verhoef et al. 2008	Emergence of New Norovirus Variants on Spring Cruise Ships and Prediction of Winter Epidemics.	January 1 and August 1 2006. Of the 43 outbreaks, 1 occurred in January 2006; all others occurred from April 24 through July 21. Three outbreaks on 3 ships occurred during the season	The data were obtained retrospectively from outbreak reports	Europe	February 2008
Carling et al.2009	Cruise Ship Environmental Hygiene and the Risk of Norovirus Infection Outbreaks: An Objective Assessment of 56 Vessels over 3 Years.	Not reported	Not reported	Not reported	07 October 2009

Table 7 Characteristics of the studies

CHAPTER 4

RESULTS

RESULTS OF THE REVIEWS

A total of 10 articles were found during the initial search in PubMed. After reading the titles and abstracts, 5 studies were excluded because they did not meet the inclusion criteria in their titles, abstracts or full- texts. The five articles included in this review contained the following study designs: three of them were a retrospective cohort study, in one the data were obtained retrospectively from outbreak reports and the other article does not explicitly state the study design. The study design was important for the research because four reviews were focused on epidemiological report and scientific data.

The five studies included, outbreaks that were between 2006 and December 2013. The time period of the year (season) was discussed in three articles, where two (n=2) studies described outbreaks during fall time and summer seasons, one described one (n=1) an outbreak that occurred in January and 42 outbreaks between April and July (summer season). Finally, the other two (n=2) articles did not report the time period of the outbreak and no information regarding seasonality could be obtained.

Among the four articles included, the place of the outbreak was discussed in European settings. Four (n=4) of the outbreaks were in multiple countries in Europe including Cyprus, France, Greece, Italy, Portugal, Spain, the British Islands, the Netherlands, Switzerland, Germany, and Belgium. One (n=1) study did not report the location of the outbreak.

A total of 112 Norovirus Outbreaks and 73 numbers of vessels were reported in the studies between January 2006 and December 2013.

Study	Numbers of ships involved	Numbers of Outbreaks of Norovirus
Gastroenteritis outbreaks on cruise ships: contributing factors and thresholds for early outbreak detection	5 seagoing cruise ships were involved	9 Outbreaks of AGE could have been caused by Norovirus
Norovirus Outbreak in a cruise ship sailing around the British Isles: Investigation and multi-agency management of an international outbreak	1 ship was involved	1 Outbreak of AGE was caused by Norovirus
Multiple exposures during a norovirus outbreak on a river-cruise sailing through Europe, 2006	1 ship was involved	2 outbreaks of AGE were caused by Norovirus
Emergence of New Norovirus Variants on Spring Cruise Ships and Prediction of Winter Epidemics	13 vessels were involved	43 outbreaks were considered probable norovirus
Cruise Ship Environmental Hygiene and the Risk of Norovirus Infection Outbreaks: An Objective Assessment of 56 Vessels over 3 Years	56 cruise ships were evaluated	57 outbreaks for which a pathogen was identified

Table 8 Study's data

REVIEW PROCESS CONTENT/DATA ANALYSIS

The articles were put through content analysis in order to extract relevant data from the text regarding the research question. Articles were read several times carefully. The data were analyzed using a comparative analysis between seasonal variations in Europe and the incidence rates of Norovirus among passengers and crew.

Mouchtouri et al., described and confirmed the importance the syndromic surveillance data collected to identify attack rate thresholds for early detection to determine AGE case characteristics and incidence and attacks rates for cruises with and without outbreaks and at the same time the study demonstrated the impact of reporting delay on the occurrence of AGE outbreaks on cruise vessels (Mouchtouri et al., 2017).

Vivancos et al., described an epidemiological investigation of a norovirus outbreak during an international cruise sailing around the British Isles in 2008. The study suggested that the transmission of the infection are more likely among those passengers in areas of the ship that are highly transited or used for communal activities and more difficult to clean (Vivancos et al., 2010).

The study by Verhoef, n.d investigated a possible source of norovirus outbreak on a river-cruise which was sourced from food consumed on board, and was impacted by other risk factors like water use, public toilet use and contact with infected people (Verhoef, n.d.). The data was collected using a questionnaire. Furthermore, stool, food and drinking water samples were collected with surfaces swabs. Results showed that consuming tap water and raspberries were the principal source of norovirus infection; however, this could not be proof regarding to the epidemiological data available.

In another study by Verhoef et al., an increase in norovirus on cruise ships identified a common source of infection. Data on the outbreak and viral presence was collected from environmental samples and patient samples. These samples were tested by different institutions using local protocols, primarily reverse transcription-PCR. Among the data collected retrospectively, the research team choses to include the AGE log and accessed the protocols and material for cleaning. Descriptive information indicated that possible contact between boarding

and disembarking passenger groups and cleaning with inappropriate materials for norovirus were the risk factors of the multiples outbreaks in the vessels. The study confirmed that after the first infection is introduced in the cruise ship setting. An outbreak is likely to occur through person-to person transmission (Verhoef et al., 2008).

The last study by Carling et al., reported an evaluation of the thoroughness environmental disinfection cleaning (TDC) of public restrooms on commercial cruise ships and AGE Outbreaks. The data was collected from 6 standardized objects (toilet seat, flush handle or button, toilet stall inner handholds, stall inner door handle, restroom inner, door handle, and a baby changing table surfaces) with high potential for fecal contamination in public restrooms in a cruise ship. The study showed a lack of cleaning on a daily basis and that the environmental hygiene plays an important role in the transmission and perpetuation of norovirus infections on cruise ships (Carling, Bruno-Murtha, & Griffiths, 2009).

OVERALL SEASONALITY

The seasonality data among the articles showed a peak in summer months in cruise ships. Whereas norovirus infections in the general population occurred more frequently during the months of October to March and a typical peak in November to January (Semenza et al., 2012). One possible explanation for the increase of norovirus outbreaks on cruise ships during off-seasonal period is because mostly vessels sail during warmer months of the year and as well in the studies may have been attributed to changes in the behavior of the host (Dowell, 2001). Crowding of individuals in cruise ships is the most common explanation for seasonal infectious diseases, and it certainly has biologic plausibility (Dowell, 2001).

Among the 5 studies analyzed, the following graphic describes that 4 of Norovirus outbreaks occurred in winter season (October-April) and 41 of outbreaks occurred in off season months (May-September), between the years 2006 and 2008. Explanations of seasonality

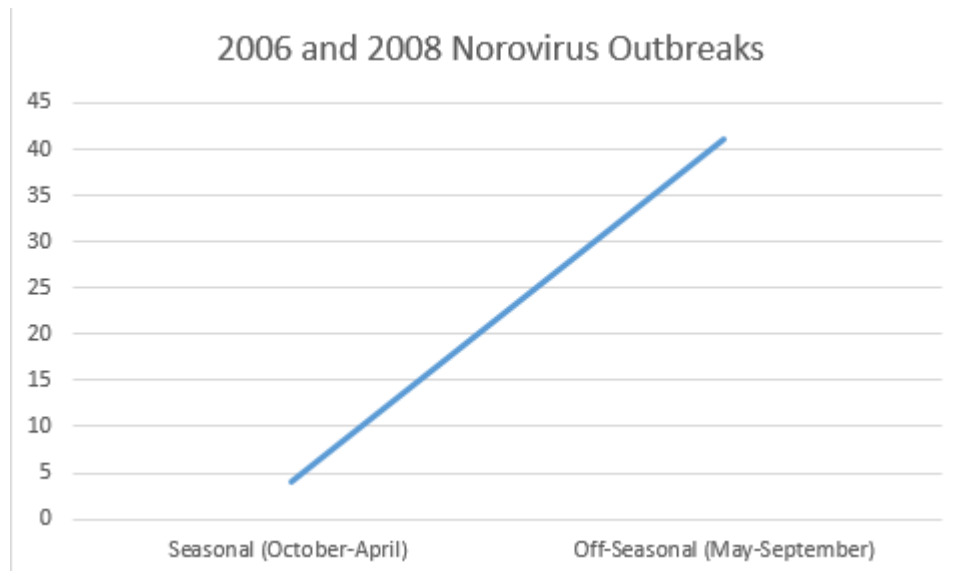


Figure 5 Seasonal patterns of norovirus outbreaks on cruise ships by calendar month of study period

APPRAISAL PROCESS

Critical appraisal skills were utilized for this research because through this tool was possible to have a quick, simple, and easy way to understand and answer the research question. CAPS is recommended propagates a systematic process through which the strengths and weaknesses of a research study can be identified. It also helps in assessing study design and the applicability of studies to a local perspective in the most economical matter (Singh, 2013).

The studies were scrutinized by CASP appraisal questions process. CASP was generated in the United Kingdom to develop an evidence-based approach in health and social care, to analyze the value of the research in relation to the methodological rigor and strength of the evidence (CASP, 2017). In CASP appraisal question process, already had the questions (Systematic Review Checklist) (CASP, 2017). The five articles were tabulated to quantitatively their qualitative values in relation to the direction of the study.

Question	Mouchtouri et al., 2017	Vivancos et al. 2010	Verhoef et al. 2008a	Verhoef et al. 2008	Carling et al.2009
1.Is the study relevant to your research question?	N	Y	Y	Y	N
2. Does the paper address a clearly focused issue?	Y	Y	Y	Y	Y
3. Is the choice of qualitative method appropriate?	Quantitative method used	Quantitative method used	Quantitative method used	Quantitative method used	Quantitative method used
4. Was the author's position clearly described and justified?	Y	Y	Y	Y	Y
5. Was the sampling strategy clearly described and justified?	Y	N	Y	N	Y
6. Was there an adequate description of the method of the data collection given?	Y	Y	Y	Y	Y
7. Were the procedures for data analysis representation described and justified?	Y	Y	Y	Y	Y

8. Are the results credible?	Y	Y	Y	Y	N
9. Can the results be applied to my research question?	N	Y	Y	Y	N
10. Were all important outcomes results considered?	Y	Y	Y	Y	Y
Accept for further use as qualitative study?	Quantitative	Quantitative	Quantitative	Quantitative	Quantitative
Appraisal score	4/11	3/11	2/11	3/11	5/11

Table 9 CASP appraisal questions (CASP, 2017)

RESULTS TO THE RESEARCH QUESTION

In this research it was important to analysis the five articles used for the research questions. Seasonal variations in Europe that are associated with the incidence of norovirus outbreaks in passengers and crew members were identified. In several articles there was no data to summarize season strength by factors as **Off-seasonal**: Summer months May, June, July, August and September. **Seasonal**: winter months October, November, December, January, February, March, April. However; in 3 / 5 studies, the results indicated that the source of norovirus infection was food, water diseases and lack of disinfestation cleaning in toilets areas. Thus, changes in the behavior of the host are involved as an important facilitator of norovirus outbreak during summertime.

Research Question	Mouchtouri et al., 2017	Vivancos et al. 2010	Verhoef et al. 2008a	Verhoef et al. 2008	Carling et al.2009
Is there an association between seasonal variations in Europe and the incidences rates of norovirus among passengers and crew?	N Epidemiological results did not consistently show a wintertime or summertime seasonality for norovirus.	Y Epidemiological results indicated that Food and Water consumption was a risk factor.	Y Epidemiological results indicated that consuming tap water was a risk factor; Study shows a Seasonal factor (October)	Y Detection of a point source was impossible because of the limited investigation of initial outbreaks and share data;	N Epidemiological results showed that the disinfection cleaning in toilet areas was the source of the infection. This study did not show a wintertime and summertime seasonality for norovirus.

Table 10 Results to the research question

TRANSMISSION ROUTE AND SETTING

By reading carefully through each of the chosen studies in Europe; the common question used to investigate the hypothesis if norovirus was a possible source of infection on cruise ships. The studies collected the data by questionnaires and the AGE Log among passengers and crew members that focused on the origin of the AGE Outbreaks, based on the menu consumed on board during the period of the infestation.

Four articles reported that the transmission of norovirus outbreaks were commonly attributed to person-to-person contact between passengers' within closed environments. Despite the strict sanitation measures on ships, in one study the investigators found poor clean of public areas, especially in toilets (common source of infection) and the behavioral risk factors as for the cases in the univariate analyses such as contact with a sick room-mate, consumption of contaminated food and water, delaying reporting of illness to the ship and lack of hand hygiene.

Transmission	Mouchtouri et al., 2017	Vivancos et al. 2010	Verhoef et al. 2008a	Verhoef et al. 2008	Carling et al.2009
Source of the infection	Passengers Behaviors, for example as not report the illness to the ship	Passenger behavior, for example by delaying reporting, concealing symptoms, or lack of hand hygiene	Person to person: contact with sick roommate, and consumption of egg, carrot pie, tap water, and whipped cream	Possible contact between boarding and disembarking passenger groups and cleaning with inappropriate materials for norovirus infection	Enteric pathogens in toilet areas

Table 11 Results common transmission and settings

OVERALL OF NOROVIRUS OUTBREAKS

The study analyzed 112 eligible norovirus outbreaks cases. Results of time period were converted to daily counts. These outbreaks occurred during 2848 days. An outbreak was considered in the study anytime when there was 3% of AGE illness in any passenger or crew population.

During the data abstraction, calculations were limited (e.g. Total number of AGE illness cases occur on that period). Based on the research question, it is important to define the proportion of norovirus outbreaks after the specific exposure, and not the total number of AGE cases.

EPIDEMIOLOGICAL RESULTS

Overall attack rate (AR) and incidence rate (IR) were calculated per 1,000 travelers (including passengers and crew members) and per 1,000 person months respectively. Moreover, incidences rates were calculated with in a period of 30 days. Data among the different articles varied in their attach rates and incidence rates.

Data from Mouchtouri et al., identified that within a period of 30 days 0.0002126 norovirus cases occurred per 1,000 passengers and crew on a cruise ship in Europe. In a second study, Vivancos et al., the results showed that in a period of 30 days, 4.3 norovirus cases occurred per 1,000 travelers on a cruise ship in Europe.

The third study Verhoef et al., indicated that in a period of 30 days, 29.19 norovirus outbreaks cases per 1,000 travelers occurred among passengers and crew members. In the fourth study, Verhoef et al., calculations were limited to addition because the numbers of individuals on board during that period was missed. The last study by Carling et al., reported that in a period of 30 days, 0.16 norovirus outbreaks cases per 1,000 travelers were identified among passengers and crew members. With these results it is possible to see an increase in the overall rate of norovirus cases in Europe on passenger cruise ships.

Study	Numbers of Outbreaks of Norovirus	Numbers of individuals on board during that period	Time Period: Daily Total (days)	Incidence Rate
Mouchtouri et al., 2017	9	869,704	1460	0.000212637
Vivancos et al. 2010	1	1714	4	4.375729288
Verhoef et al. 2008a	2	137	15	29.19708029
Verhoef et al. 2008	43	0	212	
Carling et al.2009	57	9031	1157	0.163654107
	112	880586	2848	

Table 12 Incidence rate of Norovirus Outbreak for passengers and crew members in Europe

The overall monthly attack rate identifies the probability of having a norovirus outbreak among 0.12 % per 1,000 passengers in a period 30 days in Europe on passenger's cruise ships between January 1st 2006 and December 31st 2013. For the articles found, daily attack rate was impossible to calculate because the study reports were not recorded with the dates of onset, and no attack rates were specified for passenger and crew.

The highest monthly attack rates were reported by Verhoef et al., with 14.59 norovirus cases per 1,000 travelers among passengers and crew members. On the other hand, the lowest attack rates were reported by Mouchtouri et al., who demonstrated an attack rate of 0.010 norovirus cases.

Study	Numbers of Outbreaks of Norovirus	Numbers of individuals on board during that period	Overall attack rate
Mouchtouri et al., 2017	9	869,704	0.010348348
Vivancos et al. 2010	1	1714	0.583430572
Verhoef et al. 2008a	2	137	14.59854015
Verhoef et al. 2008	43	0	
Carling et al.2009	57	9031	6.311593401
	112	880586	0.127188032

Table 13. Attack rates from Norovirus Outbreak for passengers and crew members in Europe

CHAPTER 5

DISCUSSION

This research aimed to determine if there is an association between seasonal variations in Europe and the incidence rate of the norovirus (seasonality) among passengers and crew on commercial cruise ships that were active between 01 January 2006 and 31 December 2017. To prospect that seasonal variations influence in the current patterns of norovirus is important to understand some factors that determine seasonality as norovirus appearance and disappearance, the environment changes and the host-behavioral changes.

Noroviruses are highly transmissible, can spread easily, especially in environments where people live in close quarters (Freeland et al., 2016). In Europe, norovirus infections are highly seasonal, with most outbreaks are reported during winter months for the general population (Ahmed et al., 2013). However, the data analyzed showed a peak of norovirus outbreaks in summer months. The seasonality of the norovirus in individuals in cruise ships through the studies have been attributed to changes in the behavior of the host.

The studies do not report a consistent summer or winter seasonality for norovirus outbreak (Mouchtouri et al., 2017, Carling et al.2009). For further studies, it would be interesting to investigate with itineraries, season, latitude, temperature, precipitation, the threshold levels of the outbreak and the surveillance data of each norovirus case, to produce data that will enable the cruise industry to prevent futures outbreaks.

For further development the research would aim to perform a literature review that will be specifically focused on published papers related to norovirus outbreaks and seasonal variations among passengers and crew members in Europe. By reading through several significant and informative research the results of the narrative review in Europe demonstrated how the papers that were published in the recent years, have had a common topic to investigate; the possible source of the norovirus infection.

Another common data collection had found that most research mentioned that cruise ships are highly susceptible to norovirus outbreaks, and that the person-to-person transmission plays an important role. However, Mouchtouri et al. (2017) objected this view by pointing out

that the virus is transmitted due to passenger behaviors. For example, passengers and crew members did not report the illness to the ship immediately, because they did not think it was serious. Every cruise ship company should focus on procedures for management of outbreaks. In the case of the crew members with symptoms, especially food handlers and medical staff, they should be quarantined for at least 48 hours following the resolution of their symptoms. (EU SHIPSAN ACT joint action, 2016).

Vivancos et al., 2010 holds the same opinion as Mouchtouri et al., 2017 that passenger behaviors may contribute to propagation of the virus on board cruise ships. In these two investigations the research found evidence of delay in the reporting of AGE symptoms and the initiating self-isolation. For instance, the initial cases did not report any symptoms for at least 24 hours. More consistent and complete reporting of AGE illness on cruise ships may provide opportunities for education of ill passengers and crew regarding infection control, and enable prompt interventions (Wikswa et al., 2011).

Meanwhile, in the study of Verhoef et al.2008a it was established that the propagation of norovirus outbreaks was due to behavioral risk factors associated with food and waterborne diseases: contact with some sick roommates, and consumption of egg, carrot pie, tap water and whipped cream. This situation illustrated the role of the public health on cruise ships to identify, prevent and minimize risk factors. According to the “European Manual for Hygiene Standards and Communicable Disease Surveillance on Passengers Ships”, foodborne disease prevention may include: 1) The effective implementation and maintenance of food safety management system 2) The standards of food facilities and equipment including durability and ease of cleaning 3) The age of food production facilities and 4) The effective repair, maintenance and condition of food handling facilities and equipment (EU SHIPSAN ACT joint action, 2016).

Despite the strict sanitation measures on ships, in one study the investigators found enteric pathogens in toilet areas due to poor cleaning of public areas, especially in toilets (common source of infection) (Carling et al.2009). European legislation that applies to ships includes safety rules and standards for passenger ships, border measures for public health purposes (Mouchtouri et al., 2010). Prevention inspections may include: 1) food safety and

environmental sanitation inspections on vessels, and 2) surveillance of acute gastroenteritis (AGE) (EU SHIPSAN ACT joint action, 2016).

Incidence rates of norovirus outbreaks cases showed an increase in the overall rate of norovirus cases in Europe on passenger's cruise ships. Also, the overall incidence rate calculations were limited because in one of the studies the numbers of individuals on board during that period was missed.

The overall monthly attack rate identified the probability of having a norovirus outbreak among 0.12 % per 1,000 passengers in a period 30 days in Europe on passenger cruise ships between January 1st 2006 and December 31st 2013. In a recent, "European manual for hygiene Standards and Communicable Diseases" it was reported that if on a cruise ship the AGE surveillance data has shown that during the first two days of the voyage, two passengers reporting AGE every 1,000 passengers, then the probability for having an outbreak is 6.82 % (EU SHIPSAN ACT joint action, 2016).

Daily attack rate was impossible to calculate because the study reports were not recorded with the dates of onset, and no attack rates were specified for passenger and crew. In the future, it would be interesting to study the surveillance data of each norovirus outbreak, to help the cruise ships industry with outbreak prevention based on real surveillance data (Mouchtouri et al., 2017).

In the research overall there were in the 112 norovirus outbreaks on 73 vessels during 2,848 days between January 2006 and December 2013. It would be interested to compare this data with a similar study in other continent during the same period of time.

LIMITATIONS

Limitations are normal barriers but efforts should be put in place to minimize them to a reasonable extent in which any bias would be reflected on the validity and reliability of the study (Ayodele et al., 2012). This research has some limitations that should be acknowledge. For instance, this research focused on published papers related to norovirus outbreaks in Europe, this was due to the limitation in the number of quantitative studies about the topic available. There are only a few scientific papers that cover the research topic in Europe.

The literature research was limited to PubMed, there was difficulty in accessing a large number of scientific literature regarding norovirus outbreaks on cruise ships. Also, the research was limited in the epidemiological measures including the overall incidence rate calculations, the overall daily attack rate and the outbreak frequency. For these measures, it is necessary to have data such as, daily attacks rates specified for passengers and crew, numbers of individuals on board during that period per cruise, the length on the cruise, the surveillance data with number of AGE cases reported in every 1,000 passengers and the epidemic curve with the initial outbreak.

The most important limitation of the research was the lack of information regarding season of the year (summertime and a wintertime seasonality for norovirus outbreak epidemiology), latitude, temperature, and precipitation. Without this information it is still unclear how seasonal variations impacts on the spread of infection caused by norovirus outbreaks among passengers and crew on cruise ships in Europe.

CHAPTER 6

CONCLUSION AND FUTURE RESEARCH

In conclusion, this study confirmed the importance of identifying the seasonality of data for norovirus outbreaks on cruise ships in Europe. Norovirus is a common cause of AGE on cruise ships due to the close setting and can remain infectious on environmental surfaces for a long period of time. Seasonal variations of norovirus are likely to be influenced by environmental and behavioral factors that regulate transmission, virulence and persistence of the virions in host population. Human behavior is one of the highest facilitator of norovirus transmission during summertime aboard of cruise ships.

Norovirus outbreaks on cruise ships is a luxury problem. However, the seasonal data and the epidemiological surveillance of norovirus outbreaks are useful to produce data-driven threshold for identifying an outbreak, and to understand any risk of seasonal factors influencing norovirus outbreak identification, prevention and management. This data is necessary, in order to minimize the impact of the disease, particularly during periods of heightened activity of the virus.

There is a need for additional studies with data from different cruise ship companies in Europe, and the information should be based on the number of cases, number of passengers and crew members and focused in the season (degrees of the seasonality) to identify causes of infection.

This study confirms that norovirus is a common cause of AGE, with a large number of multiple outbreaks in Europe on passenger' cruise ships due to the common source of infection from consumption of food and drinks contaminated before loading or after unloading and the semi-closed environment of the ships facilities that allow transmission from person- to- person, through the fecal-oral route and environmental contamination through aerosols arising from vomiting episodes (Bert et al., 2014). This results explains norovirus epidemiology and that changes in the behavior of the host have been attributed as the most important facilitator of the infection during summertime.

Also, this research confirmed the importance of the role of the public health departments on cruise ships. Public health departments on cruise ships are responsible for identifying, preventing and minimizing public health risks to the passengers and crew members on board. These risks may be infectious (foodborne and waterborne disease), caused by other humans, by animals (pests) or by the environmental conditions.

Future studies should be conducted on a variety of topics including nationality of the passengers and crew member data. German passengers dominate the European market, but others may be impacting various aspects of an outbreak. During 2014, a total of 1,77 million passengers were sourced from Germany with cruise ships itineraries around the globe (Contribution of cruise Tourism to the economies of Europe 2015 Edition). Giving this information, it would be interesting to see the link between seasonal variations, impact assessments and norovirus infections that occur throughout the year on cruise ships given the nationality of the passengers.

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DECLARATION OF INDEPENDENT WORK

“I hereby declare that I am the author of the Master presented. I have written the Master Thesis as applied for previously unassisted by others, using only the sources and references stated in the text.

Date:

Signature:

Eleonora Gamboa Canon

APPENDICES

Ship name:		Voyage number:		Dates	From: _/_/ _	To: _/_/ _	Page:		of	
Total number of passengers on board:		Total number of ill passengers:		Total number of crew on board:			Total number of ill crew:			

Visit date (mm/dd/yy)	Name (Last, First)	Unique Number Identifier*	Age	Gender (M/F)	Pax/Crew	Cabin No.	Date on ship (mm/dd/yy)	Date off ship (mm/dd/yy)	Pax Meal Seat /Crew Pos.	Illness Onset		Diarrhoea		Vomiting	Fever		Abd. cramps	Headache	Myalgia	Stool specimens		Antidiarrhoeal medications (Y/N)	Case fulfils definition of GI for surveillance purposes (Y/N)	Underlying illnesses (specify)	Lab results for...	Positive lab result: (Y/N/NT)	
										Date	Time	Blood Y/N	Y/N		Y/N	Fever degrees °C/°F				Requested	Received						

Appendix 1 Sample AGE Log. (European Manual for Hygiene Standards on Passengers Ships, 2016)

Study	Time Period: Daily Total (days)	Year of the Outbreak	Season	Geography Place of the Outbreak	Numbers of Ships Involved	Numbers of Outbreaks of Norovirus	Numbers of individuals on board during that period	Age of the Passengers and crew involved	Sex of the Passengers and crew involved	Measures	Route of the voyage
1.Gastroenteritis outbreaks on cruise ships: contributing factors and thresholds for early outbreak detection Gastroenteritis outbreaks on cruise ships: contributing factors and thresholds for early outbreak detection	1460	01 January 2010 and 31st December 2013	Not reported	Cuba, Cyprus, Dominican Republic, Egypt, France, Greece, Italy, Portugal, Spain and the United Kingdom	5	9	869,704	The overall age of all travelers was 55.2 or (Standard deviation) 18.3	All travelers: Female (mean) 1,069 or (Standard deviation) 55.3	The overall incidence rate was 2.81 cases per 10,000 traveler-days (95% CI: 0.00–17.60) while the attack rate was 19.37 cases per 10,000 travelers (95% CI: 0.00–127.69).	Different ports in Europe Cyprus, France, Greece, Italy, Portugal, Spain, and United Kingdom

<p>2. Norovirus Outbreak in a cruise ship sailing around the British Isles: Investigation and multi-agency management of an international outbreak</p>	4	Oct-08	Fall time	British Isles	1	1	1714	The average age of the cases was 71.6 years	There were not significant difference between male and female cases	The overall attack rate was only 11.4%	Calling at ports in England, Guernsey, Republic of Ireland, Northern Ireland, Scotland and the Netherlands.
<p>3. Multiple exposures during a norovirus outbreak on a river-cruise sailing through Europe, 2006</p>	15	2006	Summer time. June to July 25 June to 9 July	Several European countries Switzerland, France, Germany, the Netherlands and Belgium	1	2	137	The average age of the population at risk was 57.9	Male 66, female 62	Relative risk was calculated for all questionnaire items. Fisher's exact test was applied if cells in cross tables contained five or fewer records. All results are presented including 95% confidence intervals (95% CI).	Different ports in Europe

4. Emergence of New Norovirus Variants on Spring Cruise Ships and Prediction of Winter Epidemics	212	2006	January 1 and August 1 2006. Of the 43 outbreaks, 1 occurred in January 2006; all others occurred from April 24 through July 21. Three outbreaks on 3 ships occurred during the season.	Europe	13	43	0	Not reported	Not reported	Overall attack rates varied from <1% to 41%. The highest attack rates were 48% for passengers and 19% for crew members.	Different ports in Europe
5. Cruise Ship Environmental Hygiene and the Risk of Norovirus Infection Outbreaks: An Objective Assessment of 56 Vessels over 3 Years	1157	July 2005 through August 2008	Not reported	Not reported	56	57	9031	Not reported	Not reported	Not reported	Not reported
Total	2848				76	112	880,586				

Appendix 2 Overview of the results of articles