CHAPTER I
INTRODUCTION

1.1 Rationale and background

Acute diarrhea caused by Rotavirus infection in children under the age of 5 is a major public health concern worldwide. Additionally, it affects that particular country's society and economics as well (1)

In 2004 A.D., on a global scale, there were approximately 527,000 deaths caused by Rotavirus infection, and over 85% of the deaths occurred in the region of Southern Asia and Africa (2). Patients with acute diarrhea caused by Rotavirus infection can be found all year long, with high occurrences during the period of cold weather. The illness rate of acute diarrhea caused by Rotavirus infection, in developed countries, is caused by Rotavirus 31 – 87% of all the patients, which is of no difference compared to developing countries (3-5). Even though the public health, the sanitation and the living standards of citizens within developed countries are capable of reducing incidents of diarrhea from bacteria or parasites, it cannot reduce the incidents of diarrhea caused by Rotavirus at all (6).

It appears that developed countries in the continents of Europe, US and Australia, totaling to 10 countries, between the year of 1989to1998 A.D., have the rate of illness from diarrhea caused by Rotavirus, per year and per 100,000 child population under the age of 5, which is different from that of Spain's 250 cases, Ireland's 1,080 cases, South America's 2 countries: Venezuela's 3,000 cases and 645 case in Argentina. The ratio of hospital admission for child suffered from Rotavirus will be at the value between 1:19 to 1:80 (7).

The surveillance of the diarrhea cases caused by Rotavirus in Asia by The Asian Rotavirus Surveillance Network (ARSN), whom had examined the stool of children patients admitted to hospitals due to diarrhea, the abundance of Rotavirus was at 45% (1, 8).

In Thailand, it was revealed that the cases of diarrhea in children are caused by Rotavirus by 27 to 34%; with a high infection rate between every year's Octobers for February. It was most discovered in children under the age of 5; the age it was most frequently discovered is 6 months to 2 years (9, 10), Furthermore, Bureau of Epidemiology has a surveillance project on diarrhea caused by Rotavirus and a study of the epidemic strand in 6 hospitals over Thailand, such as Mae-Sord Hospital, Tak Province; Nhong-Kai Hospital, Nhong-Kai province; Prapokklao Hospital, Chantaburi province; Hatyhai Hospital, Songkhla province; Sa-Kaew Hospital, Sa-Kaew province; and Ramatibadee Hospital, Bangkok. It appears that in 2544 to 2546 A.D., there were 4,057 cases of children being admitted into the hospitals due to diarrhea. From 1,950 samples of stool collected to examine for Rotavirus, the Rotavirus was found in 838 cases, equivalent to 43%. Once the strand of Rotavirus was examined, it was disclosed that 54.8% was of G9 strand; while G2, G4, G1 and G3 strands were found in 17.2%, 5.3%, 0.8% and 0.1%, respectively (11) In each year, Thailand has 4.8 million children suffered from diarrhea, 586,000 caused by Rotavirus. 131,000 children suffered from diarrhea caused by Rotavirus had to be admitted into clinics, and 56,000 had to be admitted into hospitals (9, 11, 12)

Because the contract and spread of Rotavirus can happen easily and readily not unlike the virus contraction from the respiratory system, the contract can happen easily between members of the same family, such as adults contracted the virus from a child patient. Adult usually do not symptoms, but will discharge the virus along with the stool, which will become a contamination source to other children. Contraction of virus often happens in a group of hospitals where many child patients cluster together (13). When observing from the aforementioned annual report, the case of diarrhea caused by Rotavirus has caused problems to various hospitals, including the economic and social aspects; because inevitably, the epidemic happens every year. This study was conducted in the pediatric wards, Siriraj Hospital in 2008 A.D., which was the period of Rotavirus' abundance and dispersal. Thus, the researcher is interested in doing retrospective study on information concerning correlation between various factors, medical equipment, and the patients themselves, such as factors on childbirth's profile, factors on the treatments, factors on breastfeeding, which are important factures that may cause Rotavirus capable of spreading from one person to another,

and able to identify the importance of factors and to suggest prevention guideline; in order for personnel providing treatments to patients to understand the risks, to take cautions, so it may become a guideline on the prevention and mitigation of patients' Rotavirus contraction while resting in the hospital.

1.2 Research question

1. Factors in the neonate's profile are such as the gestational age, at the day of birth, which is preterm less than 37 weeks, post term more than 40 weeks, an neonate with unusually low birth weight, births facilitated by equipment, Caesarean section; is there any correlations to the Rotavirus infection?

2. Factors on treatments, such as the placing of Nasogastric tube; are there any correlation to the Rotavirus infection?

3. Factors on breastfeeding, alone formula-fed, being formula-fed and breastfed combined; are there any correlations to the Rotavirus infection?

1.3 Research hypothesis

1.3.1. Factors in birth's profile

- Preterm birth at less than 37 weeks has a correlation to the Rotavirus infection.

- The neonates with unusually low birth weight has a correlation to the Rotavirus infection.

- The type of delivery of equipment or Caesarean section has a correlation to the Rotavirus infection.

1.3.2. Factors on treatments

- The placement of nasogastric tube has a correlation to the Rotavirus infection.

1.3.3. Factors on breastfeeding

- Whether formula-feeding was alone used, or breastfeeding combined with formula-fed were used, there is a correlation to the Rotavirus infection.

1.4 Objective

1. To study factors concerning childbirth's profile, those such as the gestational age at the date of delivery, the birth weight, the type of delivery, and Rotavirus infection in the hospital, conducted in pediatric wards, Siriraj Hospital.

2. To study factors concerning treatments, such as the placement of a nasogastric tube and the Rotavirus infection in the hospital, conducted in pediatric wards, Siriraj Hospital.

3. To study factors concerning breastfeeding, such as those alone breastfed, those formula-fed, those who had the combination of breastfeeding and formula-feeding, with Rotavirus infection in the hospital, in pediatric wards, Siriraj Hospital.

1.5 Conceptual framework

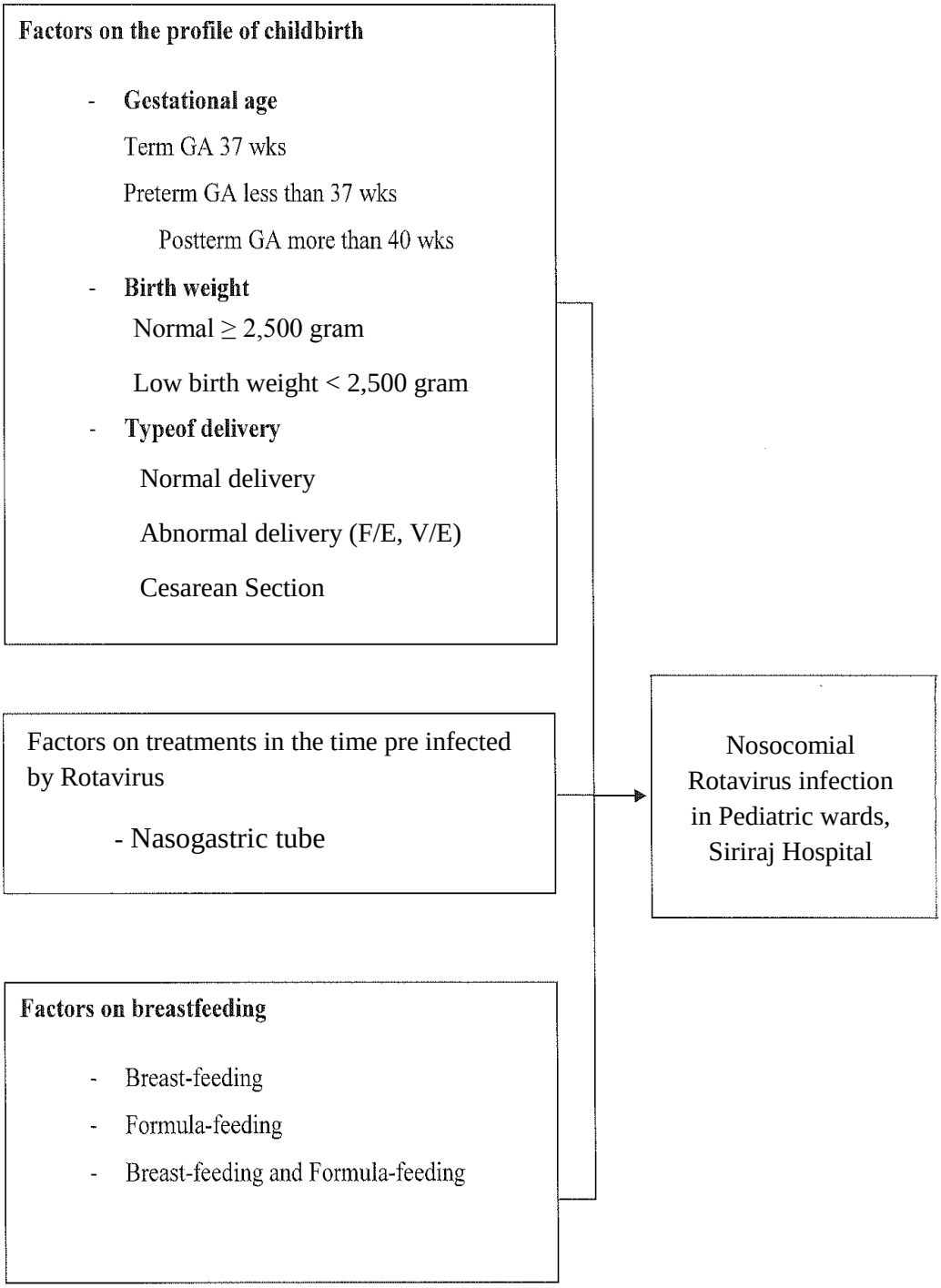

Figure 1.1 Conceptual Framework

1.6 Scope of the study

This study is a study conducted in the 15 pediatric wards of Siriraj Hospital, on those which had Rotavirus infection within 48 hours after birth, or was admitted into the hospital for treatment, or returned to the hospital due to diarrhea within 72 hours after hospital's release. It is a study of the correlations of various factors that may cause diarrhea due to the Rotavirus in the hospital, from the period of July, 24th to November, 10th, 2008 A.D.

1.7 Definition of terms

Factors

Components which may influence the becoming of illness or stimulate the becoming of illness that this study aimed to study, are factors as followed: factors on the child's birth profile, which is made up of gestational age at the date of birth, birth weight, the type of delivery; factors on treatments, such as the placement of Nasogastric tube; factors on breastfeeding, such as those alone breastfed, those alone formula-fed, and those who had a combination of breastfeeding and formula-feeding.

Nosocomial

An infection resulted from the patrons receiving the micro-organism while they were in the hospital. The micro-organism may be an Endogenous organism, or it may be exogenous organisms. As the patients were receiving treatments in the hospital, they would have no symptoms and exhibited no signs of infections, and would not be in the manifestation period of the micro-organism. If the manifestation period of the micro-organism remains unknown, it is considered an infection occurred in the hospital. If it appears that the infection shows itself 48 hours after the patients were treated in the hospital, the infection first found when being treated in the hospital may be an infection in the hospital, which is relevant to the previous hospital treatment, and has returned to the hospital with the infection 72 hours after the hospital's release (14)

Rotavirus

A type of micro-organisms which cause diarrhea. It was discovered by Bishop et al., in 1973 A.D.(15)

Diarrhea

The state in which one excretes liquid feces 3 times or more within a single day, or having loose or blood-infused stool at least once, or discharging fluid fecal matter more than once, within a single day(16).

Pediatric

Pediatric is a field in the health, the growth and the development of children, from the period of infantry, toddlerhood, childhood, up until the teenage; aims on allowing these children to have the opportunity to come to maximum capacity once they reach adulthood.

The children can be categorized as follows:

New Born: Those newly born.

Early neonatal: Neonatal age range. First 7 days (less than 168 hours).

Late neonatal: Neonatal aged between 7 to 28 days.

Infant: Infants aged 29 days to 1 year.

Factors on the birth profile

The birth profile of the study group and the control groups is:

The gestational age at the date of birth,

Using the gestational age, the categories will be:

- Pre-term neonates, gestation age under 37 weeks
- Term neonates, gestation age between 37-39 weeks
- Post-term neonates, gestation age over 40 weeks

Birth weight

Normal birth weight (2,500 grams or more)

Low birth weight (less than 2,500 grams)

Type of delivery, the mode in which each child was delivered,

A normal delivery

The equipment of vacuum extraction (V/E)

The equipment of forceps extraction (F/E)

The equipment of caesarean section (C/S)

Factors on treatments

Nasogastric tube

The placement of the tube from the nose into the stomach, by inserting it through the nostril, through the esophagus to the stomach; often done by doctors or nurses.

1.8 Expected outcomes and benefits of the study

The results of this study are the basis guidelines for the prevention of rotavirus infection in neonates in hospital and for those interested as information or to make further researches.

CHAPTER II
LITERATURE REVIEW

Review of the relevant literature follows

Rotavirus was first studied in animal. In 1983, Dr. Bishop and colleagues (15) found that rotavirus caused acute diarrhea in children in Australia. This finding suggested that rotavirus infection is one of major causes of acute diarrhea both in human, mammals and birds. Additional studies on its biochemistry, antigenicity and molecular properties revealed further information and understandings on the virus structure and its replication. These knowledges and information led to more pathological studies of infected gastrointestinal tracts, clinical appearances, Epidemiology, laboratory tests and development of vaccine against rotavirus.(17)

2.1 Virus classification

Rotavirus is a virus included in the genus *Rotavirus*, in the family Reoviridae with its diameter of 70 nm. Virus particle consisted of 2 layers of capsid, outer layer and inner layer, and the core. Rotavirus is a double stranded RNS virus contains 11 segments of ds RNA which made genome assortment and sequence changes within segment occur easily. Virus replicates in the cytoplasm of the host cells. Serogroups of rotavirus are different and they can be detected by immunofluoresce (IFA), enzyme-link immunosorbent assay (ELISA), and Immune electron microscopy (IEM). There are 6 serogroups of rotavirus (A-F). Group A, B, C infect both humans and animals. Rotavirus group A is the mostly found group that causes acute diarrhea in children. There are 14 serotypes of group a rotavirus. Important serotypes that causes diseases in human are serotype 1, serotype 2, serotype 3, serotype 4, serotype 8 and serotype 9.

In Thailand, during the year 2520-2540 found the outbreak strain of rotavirus Genotype G1 was the most, followed by G2, G4 and G3 respectively over the period 2539 to 2540 were reported. G9 increase of 3 minor of G1 and G2 later Year 2541-2542 report as G1P [8], and the year 2544 to 2546 the most common serotype G9 as much as possible. (18-20)

2.2 Clinical manifestation of acute diarrhea caused by rotavirus

Rotavirus has an incubation period of 24-28 hour. Watery stool with fever and vomiting are major clinical appearances. Watery stool usually recovered within 3-8 days. However, there is also report showed that watery stool could exist up to 22 days. Viruses are most abundant during diarrhea period, considering contagious period. Fever and vomiting usually exist only the first 2 to 3 days. Dehydration caused by rotavirus infection is more severe compared to diarrhea caused by other causes. Most common ages infected are 6 months to 2 years. Due to immunity acquired in childhood, most adults are not susceptible to rotavirus infection and show no or only mild signs and symptoms. (21-23)

Rotavirus can also be found in newborn stool as well. However, rotavirus infection in newborn causes no to mild signs and symptoms due to passive immunity

from mother. Breastfed baby will get mucin from mother which is able to inhibit virus replication. Viruses will attach to mucin, enabling them from attaching to mucous membrane of intestinal tract. Severity and duration of the disease vary due to several factors. Baby with passive immunity or people with history of infection may exhibit only mild or no sign and symptom.(24)

2.3 Rotavirus infection diarrhea diagnosis

Rotavirus infection can be diagnosis by clinical and epidemiologic features. Accurate diagnosis can also be performed by stool examination using enzyme-link immunosorbent assay (ELISA) by polyclonal or monoclonal antibody specific to virus as capture antibody. Specificity and sensitivity of ELISA in Rotavirus diagnosis are comparable to electron microscope. Latex agglutination test yield rapid result with high specificity but low sensitivity. Polyacrylamide gel electrophoresis with silver stain test (PAGE-SS) is useful for virus strain determination as they can reveal pattern of double-stranded RNA of the virus. PAGE-SS is valuable for epidemiology study to determine if the out breaking strains are the same. This test can also distinguish group A Rotavirus from non group A rotavirus.(25) Stool collected on day 1-4 after appearance of symptoms is the most suitable for virus detection although virus can be found in patients stool up to three weeks depends on the duration of diarrhea. Normally virus can still be found in the stool if the diarrhea persists (17).

2.4 Epidemiology of rotavirus (10, 13, 26, 27)

Pattern of rotavirus infection is seasonal occurring in the winter (October – March) in warm countries where the weather is cold and dry. The number of patient is less frequent in summer. In hot countries such as Thailand and Africa's, rotavirus infection is usually found in the first month that starts raining and gradually increases to the maximum number in December and January where the weather is cold and dry.

The most frequent serotypes are serotype1, serotype2, serotype3, serotype4, serotype 8 and serotype 9. Some serotypes can be found many times a year such as serotype 4 with outbreaks in 1982-1983. Serotype 3 is uncommon in Thailand.

Its outbreaks were found in Indonesia and the United Kingdom. Serotype 8 is found in Indonesia and Europe. Serotype 9 is found in USA and Japan occasionally. The other four serotypes can be found globally.

Rotavirus is the most important cause of diarrhea in children aged between 6-24 months. A study in Thailand found that Rotavirus infection is was responsible for 20-50 percent of diarrhea in children. 80-90 percent of children of age more than 3 years old had history of rotavirus infection and antibody against rotavirus could be found for the lifetime, suggesting that there were reinfection without any signs or symptoms.

2.5 How to prevent rotavirus infection (24,28-32)

How to prevent rotavirus infection is as follows.

Avoid physical contact with patient, this method is not practical as some patients show no sign of infection. Virus can also be found in the stool of these patients before exhibition of watery stool. Virus is detectable in the stool 1 day before exhibition of symptoms in 50 percent of child patients, 2 days in 31 percent and 3-5 days in 9 percent. This not only makes prevention of infection far from possible in children patients in hospitals but also increase the chance of infection.

Hand washing can reduce the number of virus, though it is not possible to eliminate all the viruses. Virus is viable for up to 4 hours in the hands and for many days on the body without cleaning. The virus is alive on the floor or dry object with smooth surface such as toys for up to 10 days.

Cleaning objects with alcohol or disinfectants such as hypochlorite, sodium dichlorocyanurate may be more effective in rotavirus elimination than soap. However, it could not completely eliminate the virus as 30 percent of diarrhea was caused by rotavirus infection both in developed and developing countries.

Breastfeeding can prevent an infant from rotavirus infection. A study indicated that the babies those breastfed alone on the first 3 – 4 months can prevent them from infection due to passive immunity from mothers. Mucin from the milk can inhibit multiplication of the virus and the virus would attach to mucin thus prevent them from attaching to the mucous membrane of the intestine.

Probiotics can reduce the duration of the symptoms for 1 to 2 days. Probiotic bacteria will compete with rotavirus on mucous membrane attachment. However, probiotic fails to be used as a preventive medication so taking probiotics to prevent from infection is not advised.

2.6 For the treatment of patients infected with rotavirus

Treatments for rotavirus infection diarrhea are symptomatic and supportive as it would be healed within 3 weeks. The goal of the treatment is to prevent the patients from dehydration. Patients are suggested to sip ORS frequently to replace the loss water by diarrhea and vomiting. In severe cases where water and electrolytes are greatly lost or unable to eat or drink because of vomiting, intravenous fluid replacement is required. Food must be given as soon as dehydration is treated (6) Children can continue their breastfeeding. Formula milk should be diluted before given to the patient. The patients must meet physician immediately if the symptoms are severe. Antibiotics are not necessary as it is viral infection. Immune compromised children should be fed with neutralizing antibody containing milk. (19)

2.7 Contact and pathogenesis

Rotavirus is food borne or direct contact with contaminated devices or instruments used orally. The virus will replicate at the cell membrane on villi of enterocytes in duodenum and upper part of jejunum. Infected cells will disintegrate. Replacing cells have short villi and fail to reabsorb water via glucose dependent sodium channels. In addition, disaccharide level will be low and causes disaccharides congestion. The sugar will consequently be digested by intestinal bacteria into acids. This will affect carbohydrate absorption and lead to watery stool. The symptoms will reduce as the enterocytes are replaced.(28)

2.8 Rotavirus Vaccines

Live attenuated oral rotavirus vaccines are being used by many countries worldwide. They are recommended in vaccination program for children and they are effective in preventing children from rotavirus infection. WHO recommend countries with rotavirus infection risk and high mortality rates to use these vaccines. There are two vaccines recommended by WHO, Rota Teq ™ and Rotarix ™ . Vaccine is given at 4 weeks old. RotaTeq™ is given orally for two doses and Rotarix is given orally for 3 doses. This vaccination is not in immunization schedule for immigrants and old children.(33)

2.9 Factors associated with infectious diseases in hospitals

Study in Hong Kong indicated that hospitalized children under 5 years old had 22% of rotavirus infection. Fisher and colleagues (34) studied nosocomial rotavirus infection without diarrhea and found that the infection was 27 % in developed countries and 23 % in developing countries.

Rotavirus can easily be transferred via fecal-oral route and it is resistant to environment, thus it can cause many infections in hospital or healthcare. Rotavirus infections are reported frequently. Infections in hospitals are usually found in nurseries. In Thailand, each nosocomial infection causes 5 days longer hospitalization. This leads to great loss in the lives of patients, their families, money and other resources. Furthermore, nosocomial infections worsen medical outcomes and decrease reputation of medical staffs and the health cares.(35)

In 1992, Brachman PS and Bennette JV (36) studied rotavirus infection in Boston, Toronto and London. They found that IV fluid replacement and respirator use increased risk of infection due to bad practices.

Direct contact is one of the most important routes of infection. Direct contact with patient, contaminated medical devices or improper use of medical staffs could ultimately spread the virus to other people. (37)

2.10 Principles of infection control in hospitals (14)

Nosocomial infection is a big problem for patient, medical staffs and public. Proper control is essential. Although it could not entirely eradicate the infection, it is still able to reduce infections in the hospital. Factors of infection are controlled as follows,

1. Controlling the spread of rotavirus as well.
   The isolation of infected patients.
   Care instruments thoroughly.
2. Prevention of microorganisms to the patient.
   Disinfection of the instruments.
   Environmental
   Staff the strict requirements on hand washing
3. Strengthening immune to such patients.
   Treatment
   Supplementary feeding
   Vaccination

The standard set by infection control (IC) (47), in controlling the infection within the hospital more strictly and cautiously, such as:

- Expected behavior of personnel on patients' caretaking according to Contact Precaution's regulation
- Expected behavior for personal items of infants and medical equipment
- Expected behavior of maintaining the environment
- Expected behavior for pediatric ward post-delivery/other wards in the submission of stool samples for examination
- Expected behavior for Lactation clinic.
- The managing system for the receiving of newborns during the period which problems of newborns having diarrhea

2.11 Related Research

Khan et al (38) showed that hands were bacteria reservoirs including organisms causing diarrhea. Their study was conducted in Dhaka, Bangladesh by divided members of Shigellosis patients admitted in a clinic. One group received soap and water and was told to wash their hands before their meals and after defecation. Control group didn't received soap and water and was not told to wash their hands. Rectal swab was conducted after ten days. The result showed that secondary case rate in testing group was 2.2% compared with 14.2% in control group. It was concluded that hand washing with soap could reduced secondary case rate up to 84 %. Healthcare education and hand washing promotion reduced diarrhea in children in Guatemala by 14%

Widdowson MA (39)In December 1999, performed a case control study in patient charts to reveal to factors of rotavirus infection in children. It is found that children with preterm birth had greater risk than those with term birth. (OR = 5.2, *p-value* = 0.008)

In addition, Widdowson also found that low birth weight increased the more likely to rotavirus infection (OR=6.1, *p-value* = 0.006). Children underwent Nasogastric feeding also had greater risk of rotavirus infection than children that didn't undergo nasogastric feeding (OR =6.0, *p-value* = 0.006).

In 2003 ,Monthida Weerawikrom (40) Epidemiology and clinical manifestations of rotavirus in children. Research models Descriptive research as a point of education department, inpatient department of Pediatrics. Hospital patient population of children 0-15 years who were treated in intensive care in hospital. Symptoms are watery during November 2545 B.E. - November 2546 B.E. study of diarrhea in children, 101 were found to be infected with Rota virus infection detected 24.7 percent. Rota virus is more common in the winter of clinical symptoms associated with Rota virus infection symptom is watery. Some mucus or blood in the stool. Vomiting May be associated with fever or abdominal pain.

Noppaorn Pawijitra and Piradee Sa-nguankiat (41) studied prevalence of asymptomatic rotavirus infection in Phramongkutkloa Hospital's Nursery. They studied factors involving infection and presence of virus in stool by cross sectioned study from 49 samples in January 2003 A.D. It was found that prevalence of

asymptomatic rotavirus infection were different depended on regions. Factors that may prevent from infection were personal hygiene and socioeconomic status of the family.

Newburg (42) Newburg and colleagues conducted a study entitled Human milk glycans protect infants against enteric pathogens in 2005 A.D. in infants who are breastfed found that breastfeeding reduced infection rate and mortality rate in diarrhea patients. This was due to antibodies and prebiotics received from the mother.

Yolken (24) studied the role of mucin in rotavirus infection prevention and showed that mucin from mother could reduce virus replication, severity and mortality of rotavirus infected diarrhea.

Haley (37) Haley and team found that contacts with medical devices and medical staffs were the most important carrier of disease to patients. Medical devices without proper disinfection made the disease spread more easily.

R. Herruzo (43) In 2008 A.D. Herruzo and colleagues studied the effects of gestational age and the birth weight on rotavirus infection. They found that children with preterm labor and low birth weight had greater more likely to rotavirus infection than children with term labor and normal birth weight (*p-value* <0.05). They also found that the more likely to rotavirus infection increased as the duration of hospitalization increased (*p-value* < 0.01)

Licia L.L. Moreira (44) Moreira and colleagues studied ravel factors involved rotavirus infection in Pediatric teaching Hospital in Brazil in 2002-2005. They found that severe malnutrition patients had greater more likely to rotavirus infection than those were not severe malnutrition. (OR=2.3, *p-value* < 0.05). Nasogastric tube and Peripheral venous catheterization also increased the more likely to Rotavirus infection (OR=3.0, *p-value* < 0.05 and OR=4, *p-value* < 0.001, respectively)

Wanatpreeya Phongsamart (45) Wanatpreeya and colleagues conducted a study A Nosocomial Outbreak of G12 Rotavirus Gastroenteritis in Neonates at a University Hospital in Thailand that were treated in the clinic. With all those who were treated 2, 677cases of rotavirus infection, a total of 117patients, duration of hospitalization of children who are infected than uninfected children statistically significant difference. (*p-value* < 0.001)

CHAPTER III
MATERIALS AND METHODS

3.1 Research design

It is a study of the case, using the design of case-control study to analyze factors concerning the birth profile, such as the gestational age at the date of delivery, the birth weight, the mode of delivery; factors concerning treatments, such as the placement of Nasogastric tube; factors concerning breastfeeding, such as those alone breastfed, those alone formula-fed, those fed with a combination of breastfeeding and formula-feeding, and Rotavirus infection within the hospital.

It is a retrospective study in medical records as previously recorded, illustrating the value as odds ratio (OR), 95% confidence interval (95% CI), and the *p-value.*

The statistical significance was set to be at 0.05.

3.2 Study population

The citizens who participated in this study were patients receiving treatments in 15 pediatric wards of Siriraj hospital, from the period of July 24th to November 10th, 2008 A.D.

3.3 Definition of case and control

The sampling group of this study, whose information was collective retrospectively from the patients' medical record of Pediatric wards, Siriraj Hospital, can be categorized into 2 groups, which are:

The study group, patients in the pediatric wards who had Rotavirus infection, or 48 hours after being admitted into the hospital, or returned to the hospital due to diarrhea within 72 hours after being discharged, who received a diagnosis from a doctor that they were suffering from diarrhea caused by Rotavirus; from July 24th to November 10th, 2008 A.D., which is an infection within the hospital.

The control group, patients who came to receive treatments within the identical ward with the study group, yet was not infected by Rotavirus; and was admitted at the identical period as the study group.

Inclusion criteria

1. Was a patient receiving treatments in pediatric wards, Siriraj hospital, from the period of July 24th to November 10th, 2008 A.D.

2. Has a Rotavirus infection post - child delivery or 48 hours after receiving treatments.

3. Returned to the hospital due to diarrhea within 72 hours after a hospital release.

4. Confirmed to name Rotavirus in stool examination from diarrhea, contact with confirmed cases.

Exclusion criteria

1. Had to record an incomplete information according to the record forms, which were used for the collection of data.

3.4 Sample size estimation

The size of sample groups

Factors concerning being breastfed was one of the factors which had to be analyzed and estimated for its correlation value. From the study by Widdowson MA in December, 1999 A.D., which was a study on the epidemic case of diarrhea caused by Rotavirus in General Hospital Neonatal Medium Care Unit (NMCU), totaling 49 individuals that can be separated to 23 individuals for the case, and 26 individuals for

the control. It was discovered that breastfeeding has the OR value of 0.5, the sample size according to the calculation will be as followed:

The calculation of sample size uses the formulae of Case Control Study following the formulae of (Schelesselman, 1982 A.D.) (46), the level of reliability (1 - α), and the power of test (1 –ß) with the level of reliability at 95%, the power of test 80%.

Formulae $$n_1^* = \frac{[Z_{\frac{\alpha}{2}} \cdot \sqrt{(1+\frac{1}{k})\bar{\pi}(1-\bar{\pi})} + Z_{\beta} \cdot \sqrt{\pi_1(1-\pi_1)+\pi_2(1-\pi_2)/k}\,]^2}{(\pi_1-\pi_2)^2}$$

With $$\pi_1 = \frac{(OR)\pi_2}{(OR)\pi_2+(1-\pi_2)} \quad \text{and} \quad \bar{\pi} = \frac{(\pi_1+\pi_2)}{2}$$

When

n = the sample size in each group, with $n_1 = n_2 = n$

n_1 = sample size of the control group, with the control group's size being $n_2 = n_1$

π_1 = the rate in risk exposure in the experimental group of the ills = 0.65 (39)

π_2 = the rate in risk exposure in the experiment of those not ill = 0.8 (39)

$\bar{\pi}$ = the rate in risk exposure by average amongst the 2 sample groups = 0.75 (39)

OR = Odds Ratio = 0.5 (39)

d = Precision of hypothesis = | π_1 - π_2 | is the absolute error

α = the chance of type I error occurring = 0.05 (2-sided), z 0.025 = 1.96

ß = the chance of type II error occurring = 0.2, z 0.2 = 0.842

$$n_1^* = \frac{[1.96 \cdot \sqrt{\left(1+\frac{1}{1}\right)0.72(1-0.72)} + 0.842 \cdot \sqrt{0.65(1-0.65)+0.8(1-0.8)/1}\,]^2}{(0.65-0.8)^2}$$

$n_1 = n_2$ = at least 139 individuals.

The study group within this research is a study of the spread of Rotavirus infection in the pediatric wards, Siriraj Hospital. In 2008 A.D., there were 117 infected; thus, the study group was made up of 117 neonates. The control group, which was the group of patients who came for treatment at the pediatric wards ward with the study group in pediatric wards, but were not infected, and remained there in the same period as the study group, in the case of 1:3 (46) to increase the statistical power, totaling to 346 neonates.

3.5 Sample selection

The selection of sample groups, in the study group which used the information on Rotavirus patients within the period of July 24th – November 10th, 2008 A.D.; with the epidemic as happened in the wards of pediatric, Siriraj hospital was of an outbreak type, was as follows:

3.5.1 The Case group which used the method of purposive sampling, totaling to 117 cases, by collecting retrospective data of all patients.

3.5.2 The Control group

The method used was to randomly draw numbers from the Running number, using the Table of Random Numbers, utilizing 4 digits numbers and read it top to down the following steps:

Step 1: Ask for a list to admit patients on the ward, and the Department of Pediatrics during the case, the patient must not have rotavirus infection.

Step 2: Running number using the 4 - digit number from 0001 to the full amount control calculated.

Step 3: Use a table of random numbers. Using a pen dipped down to the table and read from the top down below are the last 4 digits in the random selection.

3.6 Research instrument

The tools used in this study were Record form gathering prepared for information form of medical records, record books from pediatric wards, Siriraj Hospital. An information gathering form comprised of the patients' general information, information on clinical symptoms, examination information of the laboratories, information concerning treatments and information on breastfeeding; it was created from studies of documents, theories and various research. It could be categorized into 5 parts as follows:

Part I: Personal information

- Gender
- Age
- The birth profile, History of gestational age at birth, classified into, gestational age preterm birth 37 weeks. Gestational age less than 37 weeks was. Gestational age post term more than 40 weeks.
- The birth weight: the normal weight at 2,500 grams, low birth weight at less than 2,500 grams.
- The type of delivery: normal delivery, caesarean section, uses of equipment (V/E, F/E)
- The period of hospital admission

The character of data records were those with options to record provided, and those needing identifications; there were 7 of them.

Part II: Clinical symptoms

- Body's temperature
- The quality of stool, the frequency in stool discharges
- The character of headaches and vomits

The character of the data records were those with options to record provided and those requesting identifications; there were 3 of them.

Part III: Laboratory findings

- Confirmative examination from the laboratories
- Rotavirus genotype

The character of the data records were those with options to record provided and those requesting identifications; there were 2 of them.

Part IV: Medication (before diagnosis of RV)

- The placement of IV fluid
- The placement of a nasogastric tube
- The provision of Antibiotic after the other infection

The character of the data records were those with options to record provided and those requesting identifications; there were 3 of them.

Part V: Breastfeeding history before diagnosis of RV

Which could be categorized as: those alone breastfed, those breastfed and formula-fed combined, and those alone formula-fed.

The character of data records were those with options to record provided; there were 3 of them.

(See appendix)

3.7 Data collection

3.7.1 The request for permission from Ethics for Research in Human, Faculty of Medicine Siriraj Hospital, as follows:

3.7.1.1. Submit a form requesting for permission in doing a human research to Ethics for Research in Human, Faculty of Medicine Siriraj Hospital.

3.7.1.2. Research contacted and requested for permission in collecting data from the Faculty of Graduate Studies, requesting to collect information from medical records of pediatric wards, Siriraj Hospital.

3.7.1.3. Research requested for permission to use the information as found in medical records from the Siriraj Hospital's director.

3.7.1.4. After being granted permissions, research submitted the documentation to the head of pediatric wards and infection control unit, requesting for permission to collect information as found in medical records.

3.7.2 Data collection was conducted by record form, as follows:

3.7.2.1. After being permitted to use information collected from the sample groups, the research proceeded on collecting information from medical records; by using data collecting form which was created for collected data for research, it separated case group and control group, with details as followed:

Case group

- Gender, age
- Pediatric wards
- The birth profile, History of gestational age at birth, classified into, gestational age preterm birth 37 weeks. Gestational age less than 37 weeks was. Gestational age post term more than 40 weeks.
- The birth weight: the normal weight at 2,500 grams, low birth weight at less than 2,500 grams.
- The type of delivery: normal delivery, caesarean section, uses of equipment (V/E, F/E)
- The period of hospital admission
- Body's temperature
- The quality of stool, the frequency in stool discharges
- The character of headaches and vomits
- An confirmative examination from the laboratories
- Rotavirus genotype
- Alone breastfed
- Breastfed and formula-fed combined
- Alone formula-fed

Control group

- Gender, age
- Pediatric wards
- The birth profile, History of gestational age at birth, classified into, gestational age preterm birth 37 weeks. Gestational age less than 37 weeks was. Gestational age post term more than 40 weeks.
- The birth weight: the normal weight at 2,500 grams, low birth weight at less than 2,500 grams.
- The type of delivery: normal delivery, caesarean section, uses of equipment (V/E, F/E)
- Alone breastfed
- Breastfed and formula-fed combined
- Alone formula-fed

3.7.2.2. Gathered all information collected from the data gathering forms; checked its accuracy and completeness. Once all was done, the data would then be analyzed.

3.8 Data analysis

Brought in the data, whose accuracy and completeness had been re-examined, for its coding form; then performed a statistical analysis by a computer, suing SPSS Version 18.0 program to analyze the data. The data analysis could be categorized as follows:

3.8.1. The study of general information using descriptive statistics

- Used frequency percentage to describe qualitative data, such as gender, pediatric wards, birth profile, and gestational age of the mother at the date of delivery, birth weight at the date of delivery, the profile of hospital admission, Rotavirus genotype, body temperature, treatments and breastfeeding.
- The mean, the standard deviation, the median and the minimum-maximum to describe data, variables age.

3.8.2. The study of factors' correlation using inferential statistics.

3.8.2.1 The test of correlations between factors of interests and the contraction of Rotavirus, analyzed by Univariate Analysis using binary logistic regression to approximate the value of correlation, Crude odds ratio (Crude OR), 95% confident intervals (95% CI), and *p-value.* Factors of interests are such as:

- The birth profile such the gestation age at the date of delivery, the birth weight, the mode of delivery

- Factors on treatment, such as the placement of Nasogastric tube

- Factors concerning breastfeeding, such as being alone breastfed, being alone formula-fed, being formulated-fed and breastfed combined.

3.8.2.2 Via Multivariate Analysis, used binary logistic regression by selecting all dependent and independent variables which do not have collinearity into the model at the same time. By selecting variables through the method of Enter for adjusted confounder, and increased the correlation strengths of various factors and Rotavirus contraction. The Odds ratio value obtained from Multiple Logistic Regression was a value of correlation level that controlled all factor's effect that appeared in, Model (Adjusted OR), the 95% confidence intervals (95%CI) and the value of *p-value.*

3.8.2.3 Set the level of statistical significance to be at ≤ 0.05