1. INTRODUCTION

The nutritional requirements of the normal infant have been studied by investigators over many years and Recommended Dietary Allowances (RDA) for most nutrients have been established (NRC, 1989). Diarrhoeal disease is an important cause of morbidity and mortality in children less than five years of age in tropical developing countries (Walsh and Warren, 1979). In the last 15 years the number of viral, bacterial and parasitic agents identified in persons with diarrhoeal disease in developing countries has doubled. At present an infectious agent can be isolated from most children with acute diarrhoeal disease and more than one potential pathogen can be isolated in 15-30% of children with diarrhoea (Black et al., 1982 and Guerrant et al., 1986).

It matters how infants are fed for their health and development. The evidence for this is strong, notably in relation to the long term health effects of early nutrition.

Infantile Diarrhoea continues to be the chief killer of infants under the age of 1 year in tropical countries. Diarrhoea is the visible symptom of Gastroenteritis. Bacterial colonization of the infant gut may have important influences on the development of gastrointestinal, respiratory and allergic disease (Patricia et al., 1999).

1.1 BREAST-FEEDING

Breast feeding is the normal way of providing young infants with the nutrients they need for healthy growth and development. Colostrum the yellowish, sticky breast milk produced at the end of pregnancy, is recommended by WHO as the perfect food for the newborn. It is packed with antibodies that, increases baby's
immunity. Exclusive breastfeeding is recommended up to 6 months of age (Awasthi et al.,1997).

Mother's milk is the best food for babies. It contains exactly the right amount of nutrients required in the right proportions, to ensure that the baby gets the perfect balanced meal. No manufactured milk, no matter how fortified it may be, comes even close to being as nutritious as breast milk (Atkins. 1992).

1.2 BENEFITS OF BREAST-FEEDING

1. Increases Immunity

   Needless to say, a newborn is extremely delicate and is susceptible to many diseases. It is thus recommended that mothers always breastfeed the baby in the beginning. Colostrum protects the newborn's intestines and strengthens the respiratory system.

2. Increases intelligence

   Breast fed babies have better brain development than their non-breast fed counterparts. Human milk has certain ingredients which are not found in artificial milk which contributes to brain development.

3. Suckling benefits

   Breast fed babies have stronger and better formed teeth. Suckling also improves hand to eye co-ordination.
4. **Protects against infection**

Exclusively breast fed infants have a lower incidence of acute diarrhoeal disease than children that were fed with formula during the same period. IgA in breast milk protects the baby from infection.

Breast feeding provides immediate nurturing and security beyond the womb as well as continuation of nourishment, protection from disease and maternal bonding which makes the transference to the outside world safer and less harsh.

### 1.3 IMPORTANCE OF BREAST FEEDING

Breast feeding is important for the health of babies because they have immature immune system. Breast milk provides the baby with its mother's antibodies and other defence factors. Human milk has many protective factors that can enhance the immune system of the new born child against infection. The antibodies in breast milk are important in the protection against infection because they are active against bacteria, fungi, virus, and other antigenic substances to which the mother has been recently exposed to. Mother's milk contains lactoferrin that has an anti-inflammatory action, lysozyme that lyses bacterial cell walls, oligosaccharides that prevent binding of toxic substances to cells lining the airway and fats that have antibacterial action.

Breast milk is the best and safest food for young infants. Some major benefits are that breast fed infants have fever and less severe episodes of diarrhoea and lower risk of dying from diarrhoea than babies who are not breast fed.(Ginna Wall. 2010).
Breast milk is fortunately still the normal manner of feeding young babies in most rural tropical communities, although decreasing in urban situations.

**Distribution of Immunoglobulins and other soluble substances in the colostrum and milk delivered to the breast fed infant during a 24 hour period** (American Breastfeeding association).

<table>
<thead>
<tr>
<th>Soluble Product</th>
<th>Concentration in mg/day at postpartum</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>&lt; 1 week</td>
</tr>
<tr>
<td>IgG</td>
<td>50</td>
</tr>
<tr>
<td>IgA</td>
<td>5000</td>
</tr>
<tr>
<td>IgM</td>
<td>70</td>
</tr>
<tr>
<td>Lysozyme</td>
<td>50</td>
</tr>
<tr>
<td>Lactoferrin</td>
<td>1500</td>
</tr>
</tbody>
</table>

1.4 **BOTTLE FEEDING**

Despite the nutritional advantages that are associated with breast feeding, some women may be unable to breast feed, either for medical or practical reasons. In such circumstances, bottle feeding is usually recommended. Artificial feeding is usually with some form of modified mammal milk, often cow’s milk. Cow’s milk contains about 2-3 times the content of protein, especially relatively indigestible casein, the same amount of fat, and about as much milk sugar (lactose) as is found in breast milk. Formula milk are dried powdered milk substitutes made in innumerable different forms by large numbers of different companies.
The total protein content of breast milk substitutes should be lowered. Infants fed artificially, whether on the latest whey or casein dominant formulas, have elevated blood urea and amino acid levels. The high protein and salt content of substitutes until the 1970's was linked to hypernatraemic dehydration.

Bottle fed infants, particularly in an unsanitary environment, are at much greater risk of infection free contaminated milk and utensils. The daily energy requirements of the normal newborn infant are three to four times greater than those of adult i.e., 90 - 120 Kcal / Kg versus 30-40 Kcal/kg. These greater needs reflect both the infants, relatively high resting metabolic rate and the special needs for growth and development. However, even in the normal infant, relatively inefficient intestinal absorption contributes to these requirements.

Formula feeding, or bottle feeding, is often presented as a choice equivalent to breastfeeding. In fact there are known and unknown risks associated with the practice. In fact, breastmilk is not ideal but the normal, natural food for babies, adapted over millions of years to meet their unique nutritional and immunological needs. Formula, in comparison, has only been around for about 150 years, and its long term results are still being studied.(Hughes et al., 1996).

Whole cow's milk is not recommended as the main milk drink for infants before the age of 12 months. All the nutrients in breast milk are in perfect balance and the infant can also regulate its own intake, and therefore its weight gain, in a way that is impossible.
**Breast feeding** | **Bottle feeding**
---|---
Nutrition | Not as efficiently utilised as breast milk
Perfect balance of nutrients | Nutritional content depends on proper preparation.
Contains high levels of nutrients | Some babies have difficulty tolerating certain nutrients.
Easily digested and absorbed |  
Content varies according to milk production stage |  
Infant determines amount |  

**Costs**
Free | Pediatrician determines amount
Breast pump | Expensive
Nursing pads, Nursing bras, etc. | Bottles, nipples

**Advantages**
Always the perfect temperature |  
No Preparation time |  
Milk is readily available at any time and any place |  

**Disadvantages**
Mother must be available | Warming formula
Early breastfeeding may be uncomfortable | Preparation time varies
1.5 INFANTILE DIARRHOEA

Infantile diarrhoea falls into a separate category and is a serious illness. Contaminated milk or infection in the alimentary canal may cause diarrhoea in infants. If an infant is breast fed, some of the digestive disorders of the mother may be transmitted to the child.

Diarrhoea is a term used to designate frequent loose movements of the bowels. It is one of the commonest and most troublesome of the disorders of infancy and is the cause of more deaths than any other disease. It is exceedingly common in bottle fed babies than breast fed infants. It is more common and also more fatal among poor children thus making socio-economic status, an important predisposing factor causing diarrhoea.

The baby has a more upset stomach on formula milk.(Hugh et al.,1968).

1.6 POLYMERASE CHAIN REACTION

Quantitative real time PCR assays have been developed to determine the influence of either exclusive breastfeeding or formula feeding on both composition and quantity of the gut microbiota in infants.

To study the gut microbiota composition in a culture independent way, molecular methods have been developed with one based upon the detection of bacterial 16S ribosomal RNA (rRNA) genes. These methods include temperature gradient gel electrophoresis, denaturing gradient gel electrophoresis and fluorescent in situ hybridization. A drawback of these techniques is however, that they are not very suitable for analysis of large number of samples. By contrast, real-time PCR is a technique that is highly suitable for high throughput analysis. Other advantages of real time PCR are that it can be performed quantitatively, and
that it does not require post PCR sample handling, which prevents potential carry over contamination and also results in much shorter assay times.

Recently several assays have been published for the detection of bacterial group and species in faecal samples and gastrointestinal mucosa (George et al., 2006).

1.7 NEED FOR THE STUDY

Infantile diarrhoea is the leading cause of morbidity and mortality in developing countries. Death due to diarrhoea is alarming in today's world. Feeding practice is a serious pre-disposing factor to diarrhoea in infants in the age of 0-5 months.

A microbiological investigation of black infants in South Africa suffering from severe acute summer gastroenteritis revealed enteropathogenic agents in 30 out of 37 patients (Barry et al., 1977).

The new born infant is more susceptible to infections than older children or adults. Breast feeding has been shown to decrease morbidity in gastroenteritis (Labold et al., 2006).

Nutrient intake and absorption are decreased during diarrhoea.

Diet together with other environment factors plays or important role in health and disease.

Enhancing infant formulae to mimic the health promoting benefits of breast milk has been of significant interest in recent years. Differences in the gut microflora of breast fed and formula fed infants have been reported, which may explains disparities in infection rates between these groups (Kain et al., 1989).
*Escherchia coli* was the bacteria isolated in stools of bottle fed infants (Barry *et al.*, 1977) in South Africa. Other bacteria like *Shigella* spp, *Salmonella* spp and *Vibrio cholerae* also account for 30% of diarrhoea in infants (Connor and Barret, 1967).

Diarrhoea caused by *Enteropathogenic E.coli* poses a serious problem to prophylaxis and treatment. So PCR Assay of EPEC strain is of vital necessity for epidemiological studies.

The comparative study was undertaken in urban and rural areas to compare the breast feeding awareness and the association of socio-economic status with diarrhoea.

The awareness of oral rehydration therapy in the treatment of diarrhoea requires more emphasis in rural dwellers as dehydration during diarrhoea can cause death.

### 1.8 THE PRESENT STUDY

Realising the importance of feeding in earlier years this study was undertaken to compare bottle-fed and breast fed infants in urban and rural areas with special reference to isolation, identification and characterisation of diarrhoeal pathogens.
1.9 OBJECTIVES OF THE STUDY

1. To isolate and identify the bacterial pathogens from diarrhoeal faecal samples from breast fed and bottle fed infants suffering from diarrhoea.

2. To characterise the pathogens by PCR.

3. To study the drug resistance of the bacterial strains isolated by Antibiotic sensitivity testing.

5. To compare the bacterial load from infants in urban and rural areas.