The colonization of the infant gut after birth represents a complex succession of bacterial types and is governed by a wide range of external influences such as the mother’s vaginal, faecal and skin floras, the extent of bacterial contamination of the perinatal environment and the feeding regime. The influence of breast feeding and formula feeding on the development of the infant gastrointestinal tract (GIT) microflora has been the subject of numerous studies that have often yielded conflicting results. Earlier studies have reported differences between breast- and formula-fed infants, and in particular that breast-fed infants have a microflora dominated by bifidobacteria. These differences, evident in early comparisons of breast- vs formula-fed infants, appear to have diminished, most probably as a consequence of factors including changes in obstetric practices and environment, advances in bacterial identification and methodology and improvements in formula feeds.

INTRODUCTION

It was originally thought that the main role of the large intestine was one of absorption of salt and water and the excretion of waste materials. However, it is now realized that the gut microflora has an important role in the health of the host (1). Research has shown that if the microflora is suppressed or compromised, the susceptibility of the host to pathogens is increased (2–4). There is also evidence that bacterial metabolism within the gut may have an important role in toxicity of ingested chemicals and in cancer (5–8).

Attempts to modify the composition of the gut microflora by altering the diet have in general been unsuccessful (9), although recently non-digestible oligosaccharides have been shown to exert some effects (10, 11). During early life there are major changes in the microbial ecology of the gastrointestinal tract (12, 13). This has in turn led to interest in the factors that influence the development of the gut microflora in early infancy and in the effects of breast feeding vs formula feeding.

It is often considered that there are differences between breast- and formula-fed infants, and in particular that breast-fed infants have a microflora dominated by bifidobacteria which results in a lower incidence of infant-related diseases such as enterocolitis. However, a critical review of the available data does not always support such a view. Since many of the studies on the role of diet in early development of the microflora have generated controversial and conflicting results, in this review we have considered the development of the microflora in babies and infants and its potential effects on gut integrity and health. We have also tried to identify areas needing further research. This work falls under the following three headings:

1. Development of the gut microflora in infants.
2. The microflora of breast- and formula-fed infants.
3. The development of formula feeds.

DEVELOPMENT OF THE GUT MICROFLORA IN INFANTS

At birth the gastrointestinal tract is sterile (14, 15) and it may take several years to establish a microbial ecosystem similar to that of adults. Only one study has shown viable
microbes present in gastric aspirates of newborn infants delivered by Caesarean section with intact membranes (16). In this study, 3 of 15 babies delivered by Caesarean section had Staphylococcus epidermidis present in their gastric aspirates. The reason for this is uncertain and could possibly be due to contamination during the collection procedure. It is generally accepted that the establishment of the intestinal flora begins after the rupture of the foetal membranes. At this time most bacteria in the intestine are derived from the mother’s faecal and vaginal flora and also from the environment. A wide range of factors are likely to have a significant impact on the types of organisms colonizing the developing gut (Table I) and these may account, at least in part, for inconsistencies between studies. Differences in the isolation and identification procedures for gut anaerobes used in the various studies are also likely to be important reasons for discrepancies.

The majority of studies have focused on the presence of Escherichia coli due to its implications in various disease states of the young infant. Although no longer a common occurrence in industrialized countries, diarrhoeal diseases are among the most frequent causes of death in children from developing countries (17) and enterotoxigenic E. coli strains are the most commonly identified pathogens (18).

During normal delivery, the infant is likely to come in contact with E. coli strains present in the mother’s faecal and vaginal flora. One study showed that the percentage of E. coli in the oronasal cavity of infants increased during longer deliveries (19). However, transmission rates between mother and infant vary between studies. Bettelheim & Lennox-King (20) reported that from 28 mother–infant pairs, 22 infants yielded the same E. coli serotypes as their mothers. In contrast, transmission rates as low as 25% have been reported by others (21, 22). These differences may be attributed to different laboratory identification procedures of E. coli and varying hygienic measures applied during delivery.

Differences have also been shown between maternity wards suggesting transmission to neonates occurs via nurses hands (20, 23). The presence of Klebsiella spp. is increased in adults when exposed to the hospital environment (24). In a study conducted by Gothefer et al. (22), Klebsiella spp. was also a dominant microorganism in the stools of 16 out of 29 infants born in hospital on at least one occasion during the first weeks of life.

It is unlikely that babies born by Caesarean section would come into contact with their mother’s faecal or vaginal flora. This was demonstrated by Bettelheim & Lennox-King (20) who showed that the source of E. coli strains present in 6 of 8 Caesarean delivered babies was probably the nurses hands. In the remaining 2 babies, one had probably been contaminated by the mothers flora because of an emergency Caesarean section after a 16-h labour. The other infant may have acquired E. coli after oxygen administration. Colonization by lactobacilli and coliforms are also delayed in infants delivered by Caesarean section but are established within 1 month (23). However, bacteroides may not have established in infants delivered in this way at 1 month (25). It is unclear whether this delay in colonization would have subsequent detrimental effects on the infant.

Although the newborn infant is exposed to various bacteria, not all are able to establish themselves within the neonatal intestine. Lactobacilli present in maternal vaginal flora did not colonize the intestines of any babies in a study involving 5 mother–infant pairs (26). It would seem that bacteria capable of aerobic growth such as enterobacteria, streptococci and staphylococci are the first to proliferate in the infant gut (27, 28). Initially, there is a positive oxidation–reduction potential in the gut and so facultative and aerobic bacteria reach levels as high as $10^{10}–10^{11}$ bacteria/g in babies compared with adults who have levels in the range of $10^{6}–10^{9}$ bacteria/g (12). When the facultative bacteria increase in numbers they consume oxygen and so lower the redox potential. This allows the anaerobic bacteria to proliferate and facultative bacteria decrease in numbers (12). Anaerobic bacteria most commonly found in the newborn include bifidobacteria, clostridia and bacteroides (12). As availability of oxygen decreases, this restricts the range of substrates that can support the growth of E. coli (29).

### Table II

Factors explaining discrepancies between studies comparing the microflora of breast- and formula-fed infants

1. Buffering capacity of formula feeds
2. Changes in formulation
3. Obstetric practices and environment
4. Bacterial identification and methodology

THE MICROFLORA OF BREAST- AND FORMULA-FED INFANTS

From the above evidence it would appear that the bacteria present in the newborn infant come from a wide variety of sources and this is influenced by mode of delivery and contact with the mother and the environment, leading to individual patterns of colonization. Once feeding commences in neonates, a pattern begins to emerge in the microflora of the gut, and here differences between breast- and formula-fed infants have been reported (Table II).

Early work

At the turn of the century, Tissier (30) identified bifidobacteria as the predominant microorganism in the faeces of breast-fed infants. This composition remained the same until weaning began. The faeces of those infants not breast-fed contained a mixture of microbes in which no single type dominated.
The extensive studies of Bullen et al. (31, 32) in the 1970s also examined the differences between breast- and formula-fed infants. Their findings were similar to those of Tissier (30) in that faecal samples from breast-fed infants had a lower pH and a microflora dominated by bifidobacteria (31). They also demonstrated that acetic acid in the faeces of breast-fed infants is frequently present as an acetate buffer. This effect was not observed in bottle-fed infants (31). This was considered an important factor in restricting the growth of enterobacteria, clostridia and the bacteroides and favouring the proliferation of the acid-tolerant bifidobacteria and lactobacilli.

Breast milk has a low buffering capacity which would make the gut more susceptible to a lowering of pH due to acid production from bacterial fermentation in the colon. The faecal pH of the breast-fed infants in this study was between 5 and 6 whereas formula-fed infants had a faecal pH in the range of 8–9. Bullen et al. (31) suggested that immediately after birth, facultative bacteria such as *E. coli* and *Streptococcus faecium* in both breast- and formula-fed infants would proliferate and this would decrease the pH and Eh. The low pH in the large intestine of the breast-fed babies would be unfavourable for the growth of *E. coli* and so bifidobacteria would then proliferate. However, in the formula-fed infants the high buffering capacity of cow’s milk prevents a lowering of the pH and this would favour the growth of *E. coli* and bacteroides (31). The supplementation of breast-fed infants with formula feeds is a common practice and Bullen (31) noted that this had the same effect as those infants receiving only formula feeds.

### Buffering capacity and advances in formula feeds

Subsequent studies of breast- and formula-fed infants have not always substantiated the earlier work showing preponderance of bifidobacteria in breast-fed infants. There are many reasons for the conflicting evidence presented in different studies (Table II) and these will be discussed further. The buffering capacity of the formula milk would appear to be an important factor influencing microflora development. The above study by Bullen et al. (31) and many others at this time used dried milk preparations. Up until 1974 artificial feeds consisted mainly of unmodified cow’s milk with the addition of sucrose to increase the energy content. Cow’s milk contains higher amounts of protein and phosphorous and this complex results in a high buffering capacity (Table III).

Bullen et al. (32) compared the microflora of breast-fed infants with that of infants fed one of two modified cow’s milk formulas. The only difference between the formulas were the buffering capacities which were either 1.6 or 1.9 times that of breast milk. During the first 4 weeks over 60% of the breast-fed babies produced an acetate buffer which was not seen in either of the 2 formula-fed groups over the same period. The microflora of the breast-fed babies consisted of high counts of bifidobacteria and low counts of bacteroides, clostridia and coliforms. The infants fed the formula with the lower buffering capacity had high counts of bifidobacteria in the first week, though after this time no bacteria species dominated. Those infants fed the formula with a buffering capacity twice that of breast milk had high counts of putrefactive bacteria and low counts of bifidobacteria in their faeces.

A subsequent study carried out in Japan (35) also compared the microflora of breast-fed infants with formula-fed infants. Their findings were consistent with the earlier findings showing that counts of bifidobacteria are higher in breast-fed infants.

More recent studies, however, have not confirmed these results. Rose (36) used formulas with buffering capacities of 1.5 (standard formula) and 1.1 (low protein formula) times that of breast milk. This study showed a significant difference between the buffering capacity of the stools of the infants fed the standard formula compared to those that were either breast-fed or fed the low protein formula. However, this did not have an effect on the bacterial flora. It should be noted that protein and phosphorous content of modern formulas are much lower than those previously used.

### Table III

*Comparison of cow’s milk, human milk and formula feed*

<table>
<thead>
<tr>
<th></th>
<th>Cow’s milk</th>
<th>Human milk</th>
<th>Formula feeds</th>
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</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>Whey&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>SMA Gold</td>
</tr>
<tr>
<td>Energy (kJ/l)</td>
<td>2640</td>
<td>2700–3150</td>
<td>2800</td>
</tr>
<tr>
<td>Protein (g/l)</td>
<td>32</td>
<td>12–14</td>
<td>15</td>
</tr>
<tr>
<td>Casein (g/l)</td>
<td>26</td>
<td>5</td>
<td>6</td>
</tr>
<tr>
<td>Whey proteins (g/l)</td>
<td>6</td>
<td>8</td>
<td>9</td>
</tr>
<tr>
<td>Phosphorous (mg/l)</td>
<td>930</td>
<td>140–150</td>
<td>330</td>
</tr>
</tbody>
</table>

<sup>a</sup> Paul & Southgate (33).
<sup>b</sup> DHSS (34).
<sup>c</sup> Figures compiled from manufacturer’s product information.
Obstetric practices and environmental differences

Low counts of faecal bifidobacteria have been reported in infants fed unmodified cow’s milk between the years 1958 and 1978 (37) and this may be attributed to the routine use of antiseptic creams during delivery in hospitals. Bifidobacteria were not present in 35 maternal cervical cultures in mothers using antiseptic creams (38). In contrast studies carried out in developing countries have reported the ‘classical’ findings of high bifidobacteria (28, 39). Due to less antiseptic procedures and modes of labour it is more likely that these infants would come into contact with their mother’s faecal and vaginal flora. Bifidobacteria may then be more likely to proliferate in the breast-fed infants as described by Bullen et al. (31). Simhon et al. (39) concluded that coliforms and bacteroides were the predominant faecal bacteria in both breast- and formula-fed infants although the collection procedures in this study have been criticized (40). However, it would appear that in recent years studies conducted in infants in industrialized countries have found little difference between the flora of breast- and formula-fed infants.

Different results are also seen between Eastern and Western countries. In Japanese babies (13) bifidobacteria were the predominant faecal bacteria in both breast- and formula-fed infants, although counts of other bacteria including bacteroides, clostridia and enterobacteria were higher in the formula-fed compared to the breast-fed babies. In another Japanese study (35) counts of bifidobacteria as high as $10^{10}–10^{11}$ g/faeces have been reported in breast-fed babies. It was suggested that the ecological differences between countries may be a contributing factor (40).

BACTERIAL IDENTIFICATION AND METHODOLOGY

The enumeration and identification of intestinal bacteria is fraught with difficulties, in part owing to a paucity in selective media for the major anaerobic genera. In particular, there are large differences in the selectivity and specificity of media used for counting bifidobacteria in faeces (41). It is possible, therefore, that changes in the techniques used for enumeration of gut microflora during the last 25 years may have contributed to the conflicting results of studies in this area. Studies using gas liquid chromatography (GLC) which measures bacterial metabolites such as short chain fatty acids (SCFA) have reported low incidences of bifidobacteria in both breast- and formula-fed infants (39, 40). More recently, strain-specific probes were being developed with promising results. Doré et al. (42) have developed a 16S rRNA targeted oligonucleotide probe which was used to enumerate total *Bacteroides* in faecal samples from 4 children. Once developed for the major intestinal anaerobes, especially bifidobacteria, such probes should enable more extensive studies of intestinal microfloras since these probes are rapid and can be used on stored, frozen faecal samples.

Other microflora changes

In addition to the controversy regarding bifidobacteria, there are several other differences reported in the majority of studies in the intestinal microflora between breast- and formula-fed infants. Breast-fed infants in general have lower counts of clostridia and enterococci (12, 26) and higher counts of staphylococci than formula-fed infants (26, 40). However, it must be remembered that there is considerable individual variation. Table IV presents data from various studies examining the faecal flora of breast- and formula-fed infants and shows the diversity in results.

Stark & Lee (12) examined, in a longitudinal study, the composition of the intestinal microflora of 7 breast-fed and 7 formula-fed infants throughout the first year of life (Table V). Although no statistical evaluation was performed differences in the microflora of breast- and formula-fed infants are apparent. However, once weaning commences the differences between breast- and formula-fed infants are less evident and both groups begin to acquire a microflora similar to that of adults within 1 year (12).

Biochemical and physiological changes

Nordin et al. (45) examined microflora associated characteristics in infants as an alternative to bacteriological methods. Microflora associated characteristics (MACs) are defined as the recording of any anatomical structure, biochemical or physiological function which has been influenced by the microflora. Fermentation is the anaerobic breakdown of carbohydrates and proteins that have not been digested in the upper gastrointestinal tract. SCFA are one of the main products of carbohydrate fermentation in the large intestine and contribute to a lowering in intestinal pH. These SCFA may be used as an energy source and also to promote water absorption and so prevent osmotic diarrhoea. Levels of SCFA in young infants are different from adults (46) but become more similar as the infant grows (47). This is probably due to the composition of the infant’s diet. For example, low levels of butyrate may be a result of low levels of starch in the infant diet (1). The levels of SCFA in young infants may also reflect the composition of the infants microflora (47).

Lifshitz et al. (48) noticed a difference in *in vitro* SCFA production between breast- and formula-fed infants, although this was not seen by Parrett & Edwards (49). Their model used a 24-h incubation period compared to 1-h by Lifshitz et al. (48). Fermentation occurs over a much longer period than 1 h and these initial contrasting results may have been overcome with time. However, the infant microflora had a greater capacity to ferment simple sugars and oligosaccharides than more complex carbohydrates compared to adults and this may be important when introducing weaning foods (50).
There are many difficulties in the isolation and identification of faecal bacteria. However, by examining biochemical parameters it would appear that the bacteria required to convert bilirubin to urobilin, degrade mucin and convert cholesterol are not established within the intestine until the second year of life (45). This illustrates that the establishment of the microflora is a gradual process.

### Table IV

<table>
<thead>
<tr>
<th>Study</th>
<th>n</th>
<th>Age of infants</th>
<th>Milk type</th>
<th>Bifidobacteria</th>
<th>Enterococci</th>
<th>Staphylococci</th>
<th>Bacteroides</th>
<th>Clostridia</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bullen et al.</td>
<td>5</td>
<td>14 days</td>
<td>Breast</td>
<td>+</td>
<td>–</td>
<td>ND</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>(32)</td>
<td>4</td>
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<td>Cow’s milk</td>
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</tr>
<tr>
<td>Rose (36)</td>
<td>9</td>
<td>1 day</td>
<td>Breast</td>
<td>+</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
</tr>
<tr>
<td></td>
<td>6</td>
<td></td>
<td>Formula</td>
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<tr>
<td></td>
<td>9</td>
<td>5 day</td>
<td>Breast</td>
<td>–</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
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<td></td>
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<td>Formula</td>
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<tr>
<td></td>
<td>9</td>
<td>6 weeks</td>
<td>Breast</td>
<td>–</td>
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<tr>
<td>Simhon et al.</td>
<td>15</td>
<td>14 days</td>
<td>Breast</td>
<td>–</td>
<td>SR</td>
<td>+</td>
<td>+</td>
<td>SR</td>
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<tr>
<td>(39)</td>
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<tr>
<td>Lundequist et al.</td>
<td>15</td>
<td>3 weeks</td>
<td>Breast</td>
<td>–</td>
<td>–</td>
<td>+</td>
<td>+</td>
<td>SR</td>
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<td>al. (40)</td>
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<tr>
<td>Stark &amp; Lee</td>
<td>7</td>
<td>4 weeks</td>
<td>Breast</td>
<td>SR</td>
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<tr>
<td>(12)</td>
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<tr>
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<td>SR</td>
<td>+</td>
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<td></td>
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<td>Formula</td>
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<tr>
<td>Yoshioka et al.</td>
<td>6</td>
<td>1 day</td>
<td>Breast</td>
<td>SR</td>
<td>+</td>
<td>SR</td>
<td>–</td>
<td>ND</td>
</tr>
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<td>(35)</td>
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<tr>
<td></td>
<td>7</td>
<td>5 day</td>
<td>Cow’s milk</td>
<td>+</td>
<td>–</td>
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<td>ND</td>
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<td></td>
<td>6</td>
<td>1 month</td>
<td>Cow’s milk</td>
<td>+</td>
<td>–</td>
<td>+</td>
<td>–</td>
<td>ND</td>
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<tr>
<td>Roberts et al.</td>
<td>12</td>
<td>1 week</td>
<td>Breast</td>
<td>SR</td>
<td>–</td>
<td>ND</td>
<td>+</td>
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<td>ND</td>
<td>SR</td>
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<td></td>
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<tr>
<td></td>
<td>12</td>
<td>90 days</td>
<td>Breast</td>
<td>+</td>
<td>SR</td>
<td>ND</td>
<td>–</td>
<td>SR</td>
</tr>
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<tr>
<td>Nagendra</td>
<td>PS</td>
<td>3 weeks</td>
<td>Breast</td>
<td>+</td>
<td>ND</td>
<td>ND</td>
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<td>ND</td>
</tr>
<tr>
<td>(44)</td>
<td>6</td>
<td></td>
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</tr>
</tbody>
</table>

SR: same range.
ND: not determined.
PS: pooled sample.
+ = more in breast-fed. – = less in breast-fed.
The development of formula feeds

It has long been recognized by both the medical and scientific communities that breast milk is the ideal food for the growing infant. Breast-fed infants are less susceptible to gastrointestinal infections such as diarrhoea (51) and other disturbances such as acute respiratory infections (52) than formula-fed infants and this is thought to be due to factors present in breast milk (53). Cow’s milk has three times more protein than human milk and has a higher proportion of casein:whey (Table III). The whey fraction of human milk contains immunological factors including α-lactalbumin, lactoferrin, slgA and enzymes such as lysozyme which are individual to each mother and offer protection to the infant (54, 55). Modern formulas have been improved to resemble breast milk and so have lower amounts of protein and electrolytes compared to cow’s milk. They are also supplemented with iron, vitamins A, B group, C, D, E and K. Other modifications are made depending on the manufacturers. Some formulas have casein as the predominant protein while others contain demineralized whey. Even with these advances it is difficult to mimic the qualitative aspects of breast milk. The immunological functions of breast milk including the protective roles of slgA and lactoferrin in human milk have long been established (56, 57). Lactoferrin is a protein present in the whey fraction. It facilitates the absorption of iron and thus may prevent the multiplication in the gut of many facultative and obligate anaerobes that are iron-dependent, although this has yet to be demonstrated in vivo (43). It may be that this is only an in vitro effect, but it is more likely that artificial feeds may require additional factors such as lysozyme, citrate and bicarbonate and in the correct quantities that are present in breast milk (58). These and other components of breast milk may have a synergistic effect not seen in formula feeds. Other advances include the addition of free nucleotides to formula feeds (59) which are present in high concentrations in breast milk, in both the free and bound form. It was previously thought that the addition of nucleotides to infant formula would result in the growth of bifidobacteria. However, in a study conducted by Balmer et al. (60) more infants fed a whey-based formula supplemented with nucleotide monophosphate salts had E. coli as the dominant organism in their faecal flora. More recently nucleotides have been shown to affect foetal small intestinal epithelial cells in culture by affecting proliferation, differentiation and apoptosis of the cells (59). Although these are preliminary results, nucleotides may play an important role in gut maturation.

Non-immunological factors have only recently begun to be investigated. These include glycoproteins, glycolipids and lactose-derived oligosaccharides (61). These dietary carbohydrates and in particular oligosaccharides are of current interest due to their bifidogenic effect within the large intestine (62). Oligosaccharides are being added to adult foods and have been described as prebiotics. Prebiotics are defined as non-digestible food ingredients that beneficially affect the host by selectively stimulating the growth and/or activity of one or a limited number of bacteria in the colon and thus improve host health (63). Human milk contains more than 130 different oligosaccharides, in varying chain length from 3 to 11 units and are the most abundant sugars after lactose. These oligosaccharides are based on 5 monosaccharide residues including sialic acid, N-acetyl-glucosamine, fucose, glucose and galactose. This would support the hypothesis that the reported predominance of bifidobacteria in the flora of breast-fed infants may be due to the presence of galactose-containing oligosaccharides in human milk. The oligosaccharides affect the growth of the intestinal flora and may compete with bacterial receptors on the epithelial cells and so prevent their adhesion (64). It has been shown in vitro that milk fractions depleted of antibodies can block the

Table V
Comparison of faecal microflora of breast fed and formula fed infants (12)
Values represent viable counts (log10 CFU/g wet faeces)

<table>
<thead>
<tr>
<th>Group</th>
<th>4 week</th>
<th>14 week</th>
<th>20 week</th>
<th>12 months</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bifidobacteria</td>
<td>BF</td>
<td>10.6</td>
<td>10.1</td>
<td>10.2</td>
</tr>
<tr>
<td></td>
<td>FF</td>
<td>10.3</td>
<td>10.4</td>
<td>9.8</td>
</tr>
<tr>
<td>Bacteroides</td>
<td>BF</td>
<td>&lt;3.0</td>
<td>&lt;3.0</td>
<td>9.3</td>
</tr>
<tr>
<td></td>
<td>FF</td>
<td>9.3</td>
<td>8.4</td>
<td>8.7</td>
</tr>
<tr>
<td>Clostridia</td>
<td>BF</td>
<td>&lt;3.0</td>
<td>&lt;3.0</td>
<td>5.2</td>
</tr>
<tr>
<td></td>
<td>FF</td>
<td>6.4</td>
<td>5.3</td>
<td>4.7</td>
</tr>
<tr>
<td>Enterobacteria</td>
<td>BF</td>
<td>6.1</td>
<td>5.5</td>
<td>8.1</td>
</tr>
<tr>
<td></td>
<td>FF</td>
<td>9.4</td>
<td>8.3</td>
<td>8.5</td>
</tr>
<tr>
<td>Enterococci</td>
<td>BF</td>
<td>6.3</td>
<td>6.5</td>
<td>8.5</td>
</tr>
<tr>
<td></td>
<td>FF</td>
<td>9.6</td>
<td>9.2</td>
<td>9.5</td>
</tr>
</tbody>
</table>

BF: breast-fed.
FF: formula-fed.
adhesion of bacteria to epithelial cells and this has been attributed to the oligosaccharide fraction (65). In a study conducted by Brand Miller (66), the total concentration of oligosaccharides decreased throughout lactation and the relative concentrations of the individual oligosaccharides varied. An interesting aspect of this study was that the preterm milk of 2 mothers may have been twice as high in oligosaccharide components as full term milk and, therefore, may have been of benefit to the premature infant. In vitro studies suggest that oligosaccharides present in mothers breast milk and which can be detected in the urine of both the mother and the infant, may protect against urinary tract infections in both (65). In 1959 MacGillivray et al. (67) supplemented cow’s milk formula with the indigestible disaccharide lactulose in an attempt to increase lactobacilli in the intestine of formula-fed infants. Lactobacilli counts increased although this did not decrease E. coli counts in the stools.

Probiotics which are defined as live microbial food supplements which are beneficial to health (68) are also being used in attempts to promote health in human infants. Lactobacillus GG was administered to premature infants but although the intestine was colonized by the Lactobacillus GG, there were no clinical benefits observed (69). A similar observation was seen by Stanbridge et al. (70) and the colonization was associated with a small increase in ethanol excretion, although it is unlikely that this would have any clinical benefits. However, in older infants (4–45 months) Lactobacillus GG in the form of fermented milk and freeze dried powdered shortened the duration of diarrhoea compared to controls, without causing mucosal disruption (71). Clearly more research is required to establish whether pre- and probiotics have a beneficial role in gut maturation.

Conclusions and future work

The differences in bifidobacterial counts apparent in early comparisons of breast- vs formula-fed infants appears to have diminished, most probably as a consequence of improvements in the formula feeds which bring them closer in nutritional aspects to breast milk. There are, however, still differences in gut flora most notably in clostridia, enterococci, staphylococci and enterobacteria. Models of the infant gut are required to further investigate the maturation of the gut as well as dietary and physiological factors which determine bacterial colonization. This will facilitate the development of formula feeds to further resemble breast milk by, for example, the inclusion of oligosaccharides and this may in turn contribute to a more bifidogenic effect in formula-fed infants. Advances are also being made in techniques for identifying bacteria, particularly strain-specific probes and their rapid molecular biology-based techniques. The development of such rapid techniques should greatly improve our capacity to conduct large scale surveys of faecal microflora in the developing gut and to evaluate dietary effects. It should also be remembered that functional changes in microflora (MACs) are also usually rapid and simple to perform and can provide more meaningful results in terms of implications for health than simple bacterial counts.

The parallel development of increasingly sophisticated methods of analysis of the components of breast milk, the improvements in the technology of producing formula milks with chemical properties closer to breast milk, and the major developments in molecular biological methods for identifying gut bacteria are likely to yield major advances in formula milks.

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REFERENCES

The gut microflora of infants