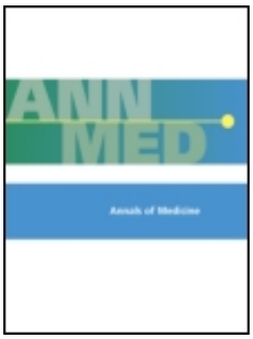

Annals of Medicine

ISSN: 0785-3890 (Print) 1365-2060 (Online) Journal homepage: informahealthcare.com/journals/iann20

Healthy gut microflora and allergy: factors influencing development of the microbiota

Pirkka V Kirjavainen & Glenn R Gibson

To cite this article: Pirkka V Kirjavainen & Glenn R Gibson (1999) Healthy gut microflora and allergy: factors influencing development of the microbiota, Annals of Medicine, 31:4, 288-292, DOI: 10.3109/07853899908995892

To link to this article: https://doi.org/10.3109/07853899908995892

Healthy gut microflora and allergy: factors influencing development of the microbiota

Pirkka V Kirjavainen and Glenn R Gibson[1]

In humans, microbial colonization of the intestine begins just after birth. However, development of the normal flora is a gradual process, which is initially determined by factors such as compozition of the maternal gut microflora, environment and possibly also by genetic aspects. A number of variables, such as the degree of hygiene, mode of delivery, use of antibiotics or other medication and a need for nursing in incubators, can all have a substantial effect on microbial colonization and development. Current knowledge on the significance and impact of such alterations on the health of the infant is poor. However, the essential role of the gut microflora in the development of the gut immune system indicates that a close relationship between allergic sensitization and the development of the intestinal microflora may occur in infancy. Intestinal micro-organisms could down-regulate the allergic inflammation by counterbalancing type 2 T-helper cell responses and by enhancing antigen exclusion through an immunoglobulin (Ig)A response. The efficacy of probiotics (microbial food additions) in the management of food allergy has been demonstrated, and these data suggest that also prebiotics, food components that target certain indigenous gut bacteria, can possibly be used for this purpose. In conclusion, the developmental pattern of the normal gut microbiota in allergic infants poses an important research avenue, as the role of the gut microflora in the mechanisms of allergy, and thereby the possible targets for efficient bacteriotherapy, are currently undetermined.

Key words: allergy; gastrointestinal immunology; hypersensitivity; intestinal microflora; microbial colonization.

Ann Med 1999; 31: 288–292.

Introduction

The composition of the intestinal microflora differs between individuals and various age groups. Thus, predicting any physiological impact exerted by the flora through composition determinants may be inconclusive. The microflora can be considered 'healthy' if it is in symbiosis with the host, thus fulfilling a role in the host nutrition and physiology. Possible contributions by the microflora towards improved host health and well-being include salvage of energy from carbohydrates which pass undigested through the upper gut, protection against invasive and resident pathogens, participation in vitamin synthesis (particularly vitamins B and K) and the metabolic degradation, or transformation, of xenobiotics (1). Conversely, an aberrant function of the microflora may result in constipation, diarrhoea, flatulence, infections, liver damage and cancer (2).

Because a child has an immature intestinal barrier, it is most vulnerable to allergic sensitization during the first few months of life. The intestinal microflora can potentiate the infant intestinal barrier by contributing towards a nonimmunological defence (3)

From the Department of Biochemistry and Food Chemistry, University of Turku, Turku, Finland; and the [1]Food Microbial Sciences Unit, Department of Food Science and Technology, University of Reading, Reading, UK.

Correspondence: Pirkka V Kirjavainen, MSc, Department of Biochemistry and Food Chemistry, Vatselankatu 2, FIN-20014 University of Turku, Finland. E-mail: pirkka.kirjavainen@utu.fi, Fax: +358 2 3336860.

and by stimulating and directing the development of the gut immune system (4–8). At the same time, however, the intestinal flora is constantly changing and is therefore very sensitive to alterations. Thus, although no direct evidence of the role of intestinal microbes in the mechanism of food allergy exists, factors influencing gut microflora development are of great interest in the context of neonatal allergic sensitization and the potential prophylactic role of microflora modulation, eg by fortification of a certain gut microflora component through the use of probiotics and/or prebiotics.

Factors influencing the formation of gut microflora

Endogenous factors

The gastrointestinal tract of the newborn infant is sterile but is then rapidly colonized by a myriad of micro-organisms originating from the mother, other external contacts (eg nurses) and the surrounding environment. In developed countries high hygienic standards may restrict a full transfer of the protective flora. Moreover, not all bacteria to which the infant is exposed are able to colonize the intestine, as this process is determined by a variety of indigenous factors which may include genetic influence (9). In general, it appears that bacterial strains of maternal origin have a greater tendency to become firmly established in the infant microflora than environmental strains (10, 11) and that the initially colonizing strains persist in the flora longer than those adopted at a later stage (12). The first colonizers must be capable of oxidative metabolizm, as the intestine of a newborn infant has a higher redox potential than that of an adult. Typically, the first inhabitants are enterobacteria, streptococci and staphylococci (13, 14). Such facultative bacteria rapidly metabolize oxygen, which lowers the redox potential thus allowing strictly anaerobic bacteria to flourish and predominate in the ecosystem. Anaerobic bacteria common in early infancy include bifidobacteria, clostridia and bacteroides.

Various control mechanisms may limit proliferation of bacteria in the gut and thus protect the host from microbial overgrowth. These include low pH conditions in the stomach generated by gastric acid and digestive enzymes, such as lysozyme, and pancreatic enzymes which are prevalent in the small intestine. Adhesion of micro-organisms to epithelial cells in the gastrointestinal tract is challenged by the washing action of intestinal peristalsis and mucus. Bacteria which attempt to establish must also compete with the intestinal microflora for colonization sites and essential nutrients and survive in the presence of adverse metabolic end-products (such as acetic, propionic and butyric acid), and antimicrobials (eg peroxides, bacteriocins) that may be produced by the indigenous flora (2). Moreover, the gut immune system can control the development of the intestinal microflora. For example, there is indication that distinct B-cell populations secrete different types of immunoglobulin (Ig)A of varying specificities, which may be involved in controlling the composition and volume of the normal flora. A further influence may be mounted against pathogens entering via the M cells of Peyer's patches (15). In infancy, however, most of these control mechanisms are impaired and operate effectively only with a developing microflora.

Exogenous factors

In a conventional delivery, the first bacteria to colonize the intestine of the infant are those present during passage through the birth canal, ie bacteria which are part of the mother's vaginal and intestinal flora (16–18). In caesarean delivery this contact is avoided, and faecal colonization of an infant delivered in such a manner is substantially delayed. In comparison to vaginally delivered infants, the colonization rate of infants born by caesarean section was seen to be delayed by 10 days and by 1 month for lactobacillus and bifidobacterium-like bacteria, respectively (19). Similarly, Hall and co-workers (20) have demonstrated that conventionally delivered infants were more likely to acquire lactobacilli by the age of 10 days than those delivered through caesarean section. After the age of 30 days, differences were negligible, indicating that contact with the maternal vaginal and intestinal flora was important to initial development of the lactobacilli flora but that other factors were also present during the first weeks of life (20). Some differences, however, appeared to last longer: at 6 months of age 76% of conventionally delivered infants and only 36% of infants born by caesarean section were colonized with bacteria of the *Bacteroides fragilis* group (19).

In modern maternity hospitals, infants are subject to high levels of hygiene during delivery and nursing; however, in developing countries infants are likely to be exposed to lower levels of sterility. A reduced hygienic environment appears to result in hastened initial colonization with a wider spectrum and a less stable population of enterobacterial species. In countries with improved hygienic practices, the enterobacterial flora is generally dominated by one or a few different strains (21, 22). The infants in developing countries are also commonly colonized by pathogenic species such as salmonellae (23). Other factors associated with modern maternity wards which can affect the development of the intestinal microflora in early infancy include antibiotic treatment and nursing in an incubator, both of which delay colonization by lactobacilli. Antibiotic treatment, in particular, delays

formation of the anaerobic flora while isolation in an incubator predisposes to nosocomial enterobacteria (10, 20, 24).

The composition of infant microflora is also substantially influenced by the feeding regime. Faecal populations of breastfed infants are dominated by bifidobacteria, with less than 1% of the total being enterobacteria. Conversely, the microflora of formula-fed infants is more diverse, with bifidobacteria, bacteroides, clostridia and streptococci all being prevalent (14). However, upon the introduction of solid food to the breastfed infants, dramatic changes in the microbiota occur with a sharp increase in enterobacterial and enterococcal counts, as well as colonization by bacteroides, clostridia and anaerobic streptococci. Changes in the formula-fed infants are more moderate (25).

Knowledge on the subsequent health effects of the different colonization patterns is poor. It is likely, however, that the stable and polarized enterobacterial microflora, characteristic of the microbial flora in infants from developed countries, is associated with a lower risk of pathogenic infections (11). From another perspective such a microbiota may not provide sufficient stimulus for the development of the gastrointestinal immune system as the gut is exposed to less immunogenic, and fewer numbers of, bacterial antigens. It also appears that at least a partial tolerance may ultimately be formed to the bacteria belonging to the normal flora (15, 26, 27). Therefore, changes in the microflora which provide new challenging bacterial antigens may sometimes be required to maintain the ability to provoke an immune response and thus 'train' the immature immune system.

Development of the normal intestinal microflora and down-regulation of allergic inflammation

In newborn infants, the type 2 T-helper cell (Th2) cytokines, which are essential mediators in the formation of allergic inflammation, predominate over Th1 cytokines. Th2 cytokines include interleukin (IL)-4, which induces B-cell differentiation into IgE-producing cells, and IL-5, which is important for the activity of eosinophils. Intestinal bacteria have been shown to counterbalance Th2 activity and to be essential for the regulation of the IgE response (28, 29). The former appears to result in the predilection of bacteria in general to promote differentiation of Th1-cell lineage (30). This may be the result of a number of factors, the most significant of which may be a specified CpG motif, which is characteristic of bacterial DNA. This 6-base sequence has been shown to induce polyclonal B-cell activation and secretion of Th1 cytokines IL-6, IL-12 and interferon (IFN)-γ (31). Other factors, such as the lipopolysaccharide (LPS) portion of gram-negative bacteria, may also contribute to this predilection (30, 32).

Intestinal bacteria may also alleviate allergic inflammation by modifying antigen uptake (33), presentation (8) and degradation (34, 35). These modifications may then result in down-regulated IL-4-mediated responses (34) and enhanced antigen exclusion. For example, species common to the gut microbiota, such as bifidobacteria and lactobacilli, have been shown to enhance IgA production in Peyer's patches and potentiate IgA responses towards potentially harmful antigens (36–40). LPS on the other hand may enhance the IgA response mounted to dietary antigen (8). This enhancement may alleviate intestinal inflammation by preventing some potentially allergenic dietary antigens from invading the intestinal mucosa and thus provoking hypersensitivity reactions and also by down-regulating inflammatory responses to pathogenic bacteria by enhancing their exclusion.

Modulation of the intestinal microflora

In the modern environment, because of hygiene measures an infant may be exposed to a narrow spectrum of bacteria. However, the possibility of transfer of pathogens means that the standards of hygiene cannot be relaxed. Instead, the development of the gut microflora can be fortified or its impaired function corrected by administration of microbial cell preparations or components of microbial cells (probiotics) (41) or by dosing with chemicals which stimulate the growth and metabolizm of certain indigenous gut bacteria (prebiotics) (42).

Clinical findings indicate that probiotic supplementation has great potential in the management of food allergy (43). Both lactobacilli and bifidobacteria are common probiotics. Their use is also supported by data from studies performed *in vitro* and in murine models (30). Moreover, these findings attest to the potential of using prebiotics to gain similar results. At present, attention on prebiotic research has mainly been directed towards the use of nondigestible oligosaccharides (44). In particular the feeding of fructo-oligosaccharides (such as inulin-based materials) have been demonstrated in a number of clinical trials to be very efficient at stimulating bifidobacterial growth in the large intestine (45–47). There is, however, currently no firm clinical evidence for the benefit of such treatment.

Conclusion

A wide spectrum of factors may promote the formation of 'abnormal' microflora in infancy. Many

of these elements are associated with modern obstetrical practices and hygienic routines aimed at minimizing the risk of bacterial infections in maternity wards. There is some indication that in early infancy an aberrant array or insufficient number of intestinal micro-organisms may not be able to potentiate the immature gut barrier or to counterbalance a Th2-skewed cytokine profile, ie to reduce two major risk factors for allergic sensitization efficiently enough to avoid allergy. Results on the use of probiotics in the management of food allergy hold much promise and also indicate that prebiotics could potentially be used for the same purpose. Further research should be carried out for comparing intestinal microflora in healthy and allergic infants and for determining whether differences in the microbiota are relevant in the mechanism of allergy. Until reliable information is available, the possible targets for efficient prebiotic therapy and the role of the gut microflora in the mechanisms of allergy can only be speculated upon.

References

1. **Gibson GR, Macfarlane GT, eds.** *Human Colonic Bacteria: Role in Nutrition, Physiology and Pathology.* Boca Raton, FL: CRC Press; 1995.
2. **Simon GL, Gorbach SL.** The human intestinal microflora. *Dig Dis Sci* 1986; 31(9 Suppl): 147S–62S.
3. **Roberfroid MB, Bornet F, Bourley C, Cummings JH.** Colonic microflora: nutrition and health. *Nutr Rev* 1995; 53: 127–30.
4. **Bauer H, Paronetto F, Burns WA, Einheber A.** The enhancing effect of the microbial flora on macrophage function and the immune response. A study in germfree mice. *J Exp Med* 1966; 123: 1013–24.
5. **Crabbé P, Bazin H, Eyssen H, Heremans J.** The normal microbial flora as a major stimulus for proliferation of plasma cells synthesizing IgA in the gut. *Int Arch Allergy Appl Immunol* 1968; 34: 362–75.
6. **MacDonald TT, Carter PB.** Requirement for a bacterial flora before mice generate cells capable of mediating the delayed hypersensitivity reaction to sheep red blood cells. *J Immunol* 1979; 122: 2624–9.
7. **Pulverer G, Ko HL, Roszkowski W, Beuth J, Yassin A, Jeljaszewicz J.** Digestive tract microflora liberates low molecular weight peptides with immunotriggering activity. *Int J Med Microbiol* 1990; 272: 318–27.
8. **Dahlgren UIH, Wold AE, Hanson LA, Midtvedt T.** Expression of a dietary protein in E. coli renders it strongly antigenic to gut lymphoid tissue. *Immunology* 1991; 73: 394–7.
9. **Van de Merwe JP, Stegeman JH, Hazenberg MP.** The resident faecal flora is determined by genetic characteristics of the host. Implications for Crohn's disease? *Antonie Van Leeuwenhoek* 1983; 49: 119–24.
10. **Bennet R, Nord CE.** Development of the faecal anaerobic microflora after caesarean section and treatment with antibiotics in newborn infants. *Infection* 1987; 15: 332–6.
11. **Adlerberth.** Establishment of normal intestinal microflora in the newborn infant. In: Hanson LÅ, Yolken RH, eds. *Probiotics, Other Nutritional Factors, and Intestinal Microflora.* Nestlé Nutrition Workshop Series, Vol. 42. Philadelphia, PA: Lippicott-Raven Publishers; 1999: 63–78.
12. **Kuhn I, Tullus K, Mollby R.** Colonisation and persistence of *Escherichia coli* phenotypes in the intestine of children aged 0 to 18 months. *Infection* 1986; 14: 7–12.
13. **Rotimi VO, Duerden BI.** The development of the bacterial flora in normal neonates. *J Med Microbiol* 1981; 14: 51–62.
14. **Yoshiota M, Fujita K, Sakata H, Murono K, Iseki K.** Development of the normal intestinal flora and its clinical significance in infants and children. *Bifidobacteria Microflora* 1991; 10: 11–7.
15. **Mestecky J, Russell MW, Elson CO.** Intestinal IgA: novel views on its function in the defence of the largest mucosal surface. *Gut* 1999; 44: 2–5.
16. **Mevissen-Verhage EAE, Marcelis JH, de Vos MN, Harmsen-van Amerongen WCM, Verhoef J.** *Bifidobacterium, Bacteroides,* and *Clostridium* spp. in fecal samples from breast-fed and bottle-fed infants with and without iron supplement. *J Clin Microbiol* 1987; 25: 285–9.
17. **Tannock GW, Fuller R, Smith SL, Hall MA.** Plasmid profiling of members of the family Enterobacteriaceae, lactobacilli, and bifidobacteria to study the transmission of bacteria from mother to infant. *J Clin Microbiol* 1990; 28: 1225–8.
18. **Bettelheim KA, Lenox-King SMJ.** The acquisition of Escerichia coli by newborn babies. *Infection* 1976; 4: 174–9.
19. **Grönlund MM, Lehtonen OP, Eerola E, Kero P.** Fecal microflora in healthy infants born by different methods of delivery: permanent changes in intestinal flora after cesarean delivery. *J Pediatr Gastroenterol Nutr* 1999; 28: 19–25.
20. **Hall MA, Cole CB, Smith SL, Fuller R, Rolles CJ.** Factors influencing the presence of faecal lactobacilli in early infancy. *Arch Dis Childhood* 1990; 65: 185–8.
21. **Adlerberth I, Carlsson B, de Man P, Jalil F, Khan SR, Larsson P, et al.** Intestinal colonisation with Enterobacteriaceae in Pakistani and Swedish hospital-delivered infants. *Acta Paediatr Scand* 1991; 80: 602–10.
22. **Adlerberth I, Jalil F, Carlsson B, Mellander L, Hanson LA, Larsson P.** High turnover rate of Escherichia coli strains in the intestinal flora of infants in Pakistan. *Epidemiol Infect* 1998; 121: 587–98.
23. **Bennet R, Eriksson M, Nord CE, Tafari N.** Transient, asymptomatic colonisation of newborn Ethiopian infants by Salmonella. *Infection* 1994; 22: 49–50.
24. **Fryklund B, Tullus K, Burman LG.** Epidemiology of enteric bacteria in neonatal units – influence of procedures and patient variables. *J Hosp Infect* 1991; 18: 15–21.
25. **Stark PL, Lee A.** The microbial ecology of the large bowel of breast-fed and formula-fed infants during the first year of life. *J Med Microbiol* 1982; 15: 189–203.
26. **Shroff K, Meslin K, Cebra J.** Commensal enteric bacteria engender a self-limiting humoral mucosal immune response while permanently colonizing the gut. *Infect Immun* 1995; 63: 3904–13.
27. **Hanson LÅ, Dahlman-Höglund A, Karlsson M, Lundin S, Dahlgren U, Telemo E.** Normal microbial flora of the gut and the immune system. In: Hanson LÅ, Yolken RH, eds. *Probiotics, Other Nutritional Factors, and Intestinal Microflora.* Nestlé Nutrition Workshop Series, Vol. 42. Philadelphia, PA: Lippicott-Raven Publishers; 1999: 217–28.
28. **Gaboriau-Routhiau V, Moreau MC.** Gut flora allows recovery of oral tolerance to ovalbumin in mice after

transient breakdown mediated by cholera toxin or Escherichia coli heat-labile enterotoxin. *Pediatr Res* 1996; 39: 625–9.

29. **Sudo N, Sawamura S, Tanaka K, Aiba Y, Kubo C, Koga Y.** The requirement of intestinal bacterial flora for the development of an IgE production system fully susceptible to oral tolerance induction. *J Immunol* 1997; 159: 1739–45.
30. **Kirjavainen PV, Apostolou E, Salminen SJ, Isolauri E.** New aspects of probiotics – a novel approach in the management of food allergy. *Allergy* (in press).
31. **Klinman DM, Yi A-K, Beaucage SL, Conover J, Krieg AM.** CpG motifs present in bacterial DNA rapidly induce lymphocytes to secrete interleukin 6, interleukin 12, and interferon g. *Proc Natl Acad Sci U S A* 1996; 93: 2879–83.
32. **Weiner HL, Gonnella PA, Slavin A, Maron R.** Oral tolerance: cytokine milieu in the gut and modulation of tolerance by cytokines. *Res Immunol* 1997; 148: 528–33.
33. **Isolauri E, Majamaa H, Arvola T, Rantala I, Virtanen E, Arvilommi H.** Lactobacillus casei strain GG reverses increased intestinal permeability induced by cow milk in suckling rats. *Gastroenterology* 1993; 105: 1643–50.
34. **Sütas Y, Hurme M, Isolauri E.** Down-regulation of anti-CD3 antibody-induced IL-4 production by bovine caseins hydrolysed with Lactobacillus GG-derived enzymes. *Scand J Immunol* 1996; 43: 687–9.
35. **Sütas Y, Soppi E, Korhonen H, Syväoja EL, Saxelin M, Rokka T, et al.** Suppression of lymphocyte proliferation in vitro by bovine caseins hydrolyzed with Lactobacillus casei GG-derived enzymes. *J Allergy Clin Immunol* 1996; 98: 216–24.
36. **Moreau MC, Ducluzeau R, Guy-Grand D, Muller MC.** Increase in the population of duodenal IgA plasmocytes in axenic mice monoassociated with different living or dead bacterial strains of intestinal origin. *Infect Immun* 1978; 21: 532–9.
37. **Kaila M, Isolauri E, Soppi E, Virtanen E, Laine S, Arvilommi H.** Enhancement of the circulating antibody secreting cell response in human diarrhea by a human *Lactobacillus* strain. *Pediatr Res* 1992; 32: 141–4.
38. **Yasui H, Nagaoka N, Mike A, Hayakawa K, Ohwaki M.** Detection of Bifidobacterium strains that induce large quantities of IgA. *Microb Ecol Health Dis* 1992; 5: 155–62.
39. **Majamaa H, Isolauri E.** Probiotics: a novel approach in the management of food allergy. *J Allergy Clin Immunol* 1997; 99: 179–85.
40. **Takahashi T, Nakagawa E, Nara T, Yajima T, Kuwata T.** Effects of orally ingested *Bifidobacterium longum* on the mucosal IgA response of mice to dietary antigens. *Biosci Biotechnol Biochem* 1998; 62: 10–5.
41. **Ouwehand AC, Kirjavainen PV, Shortt C, Salminen S.** Probiotics: mechanisms and established effects. *Int Dairy J* 1999; 9: 43–52.
42. **Gibson GR, Roberfroid MB.** Dietary modulation of the human colonic microbiota: introducing the concept of probiotics. *J Nutr* 1995; 125: 1401–12.
43. **Majamaa H, Isolauri E, Saxelin M, Vesikari T.** Lactic acid bacteria in the treatment of acute rotavirus gastroenteritis. *J Pediatr Gastroenterol Nutr* 1995; 20: 333–8.
44. **Fuller R, Gibson GR.** Probiotics and prebiotics: microflora management for improved gut health. *Clin Microbiol Infect* 1998; 4: 477–80.
45. **Gibson GR, Beatty EB, Wang X, Cummings JH.** Selective stimulation of bifidobacteria in the human colon by oligofructose and inulin. *Gastroenterology* 1995; 108: 975–82.
46. **Buddington RK, Williams CH, Chen SC, Witherly SA.** Dietary supplementation of neosugar alters the fecal flora and decreases activities of some reductive enzymes in human subjects. *Am J Clin Nutr* 1996; 63: 709–16.
47. **Kleessen B, Sykura B, Zunft HJ, Blaut M.** Effects of inulin and lactose on fecal microflora, microbial activity, and bowel habit in elderly constipated persons. *Am J Clin Nutr* 1997; 65: 1397–402.