Carbohydrate Polymers 93 (2013) 263–265

Contents lists available at SciVerse ScienceDirect

Carbohydrate Polymers

journal homepage: www.elsevier.com/locate/carbpol

Evidence-based benefits of specific mixtures of non-digestible oligosaccharides on the immune system

Alma J. Nauta[a,c,*], Johan Garssen[b,c]

[a] Danone Research, Centre for Specialised Nutrition, Singapore
[b] Danone Research, Centre for Specialised Nutrition, The Netherlands
[c] Department of Pharmaceutical Sciences, Utrecht University, Utrecht, The Netherlands

ARTICLE INFO

Article history:
Received 22 November 2011
Received in revised form 2 February 2012
Accepted 13 February 2012
Available online 19 February 2012

Keywords:
Non-digestible oligosaccharides
Immune modulation
Infants
Adults

ABSTRACT

Non-digestible carbohydrates (NDC) are natural constituents of many foods. They are mostly referred to as dietary fibre and are associated with many health benefits mostly connected to gut health. NDC have emerged as a promising nutritional concept to modulate immune function as well. In the world of immunology non-digestible carbohydrates are recognized now as key immunomodulating molecules. Both pharma and food industries realize the enormous potency of these immune active components. Although the mechanisms underlying the effects of NDC on the immune system are not totally clear yet many studies have reported beneficial effects on both mucosal and systemic immunity in humans. The aim of this review is to summarize the available evidence on the immune modulatory effects of specific mixtures of oligosaccharides. Both mechanistic in vitro and in vivo studies have been performed and will be discussed. Finally the potential use of these unique structures will be evaluated.

1. Introduction

Non-digestible carbohydrates (NDC) are natural constituents of many foods and often referred to as dietary fibres that are not (fully) digestible by human or animal digestive tract endogenous enzymes. NDC are associated with many health benefits, including positive effects on fermentation, mineral absorption, barrier function, fat metabolism, glycemic and insulin responses (Meyer, 2004). Plant-derived NDC constitute the majority of dietary fibre in human food. Most NDC are at least partially fermented in the large intestine. A major non-plant-derived source of NDC is human breast milk. Human milk contains 10–12 g oligosaccharides/l making the oligosaccharide fraction a major component of human milk, with levels higher compared to levels of protein in human milk (Boehm et al., 2003). Human milk oligosaccharides (HMOS) are relatively resistant against digestion in the human stomach and intestines, and they fulfil the published criteria for prebiotics. Prebiotics are defined as "food ingredients that beneficially affect the host by selectively stimulating the growth and/or activity of one or a limited number of bacterial species already resident in the colon, and thus attempt to improve host health" (Gibson & Roberfroid, 1995). Accumulating evidence indeed suggest that HMOS can improve gut microbiota composition (prebiotic effect). In addition, HMOS have been shown to improve intestinal functions (integrity/motility), and modulate the immune system. HMOS are structurally very complex and have a huge diversity (Bode, 2006). It has been hypothesized that the large structural variety is to combat the large variety of pathogens that the humans may encounter. Furthermore, HMOS are suggested to modulate the intestinal glycosylation pattern, thereby affecting pathogen adhesion (Angeloni et al., 2005).

2. Alternative oligosaccharides

Due to the enormous complexity of the human milk oligosaccharides it is most unlikely to find natural sources, which contain oligosaccharides 100% identical to human milk oligosaccharides. A mixture of neutral short-chain galacto-oligosaccharides (derived from enzymatic synthesis based on lactose; scGOS) and long chain fructo-oligosaccharides (derived from vegetable plant; lcFOS), comparable to the molecular size distribution of HMOS, has been identified as an alternative (Boehm & Stahl, 2003). This specific oligosaccharide mixture (Immunofortis®) is so far the best studied non-milk derived oligosaccharides for which the prebiotic and linked immune effects have been proven in clinical settings (both adults and infants). A recent review by Robertfroid et al. provide an extensive and clear overview of the state of the art in the research on the health effects of prebiotics, mainly focusing on inulin-type fructans and galacto-oligosaccharides, for which the majority of scientific data has been obtained (Roberfroid et al., 2010).

* Corresponding author at: 11 Biopolis Way, Helios #09-01/02, Singapore 138667, Singapore. Tel.: +65 6830 9404; fax: +65 6830 9410.
E-mail address: alma.nauta@danone.com (A.J. Nauta).

0144-8617/$ – see front matter © 2012 Elsevier Ltd. All rights reserved.
doi:10.1016/j.carbpol.2012.02.021

3. In vivo effects

3.1. Prebiotic function

The beneficial effects on the immune system are commonly ascribed to the stimulation of the growth and metabolism of protective commensal intestinal bacteria which is part of the first defence in the gastro-intestinal tract (Boehm et al., 2005). Different studies have reported that oligosaccharide mixtures were able to modulate the microbiota of bottle-fed infants, making the composition of the microbiota more similar to the bifidobacteria-dominated microbiota in breastfed-infants (Knol, Scholtens, et al., 2005; Moro et al., 2002). The oligosaccharide mixture induced a significant influence on stool characteristics as well such as increasing the stool frequency and softening the stool consistency, alike human milk fed infants. This has been observed in both preterm and term infants (Moro et al., 2002). With regard to metabolism, there was a significant effect of scGOS/lcFOS supplemented formula on the synthesis of short-chain fatty acids (SCFA), resulting in a faecal SCFA pattern similar to that observed in breast fed infants (Knol, Scholtens, et al., 2005). Moreover, it has been demonstrated that stimulation of bifidobacteria reduces the presence of clinically relevant pathogens in the faecal flora, indicating that prebiotic substances might have the capacity to protect against enteral infections (Knol, Lidestri, et al., 2005).

A recent study in non-symptomatic HAART-naive HIV-1-infected adults indicated that with a unique prebiotic mixture containing scGOS/lcFOS and some acidic oligosaccharides mimicking the acidic fraction in human milk, their gut microbiota could, at least partially, be restored by stimulation the growth of bifidobacteria and reduction in faecal pathogenic load (Gori et al., 2011).

3.2. Animal models

Available evidence suggests that HMOS may have systemic effects in infants as they have been found in urine (Obermeier et al., 1999). In line with these observations, the immunomodulatory effects are not restricted to the gastrointestinal tract. In vaccination response studies in mice, oral supplementation with scGOS/lcFOS significantly increased the vaccination response, although the relationship between modulation of the microbiota and immune system was not consistent in all experiments (Vos et al., 2006, 2010). Intervention studies in mouse models for allergy, both cow's milk allergy and experimental allergic asthma, demonstrated that specific mixtures of oligosaccharides inhibited the allergic reaction (Schouten et al., 2009; Vos et al., 2007). Supplementation of a nutritional combination containing scGOS/lcFOS, fish oil, high protein and leucine improved the immune competence and reduced the inflammatory status in a murine model for cancer cachexia (Faber et al., 2008). Interestingly, the same nutritional combination reduced the incidence and severity of pseudomonas aeruginosa translocation in a murine model of chemotherapy-induced immune suppression (Faber et al., 2011). Different mechanisms might have played a role, including modulation of the gut microbiota composition, improved gut barrier and immune function and reduced inflammation.

3.3. Clinical effects

Evidence for the immune modulatory effects of the specific mixtures of oligosaccharides was obtained in different clinical studies. Supplementation with scGOS/lcFOS in infants at a high risk for allergy reduced the cumulative incidence of atopic dermatitis which is one of the first signals for the atopic march in humans (Moro et al., 2006). Interestingly, infants that received scGOS/lcFOS displayed a reduced incidence of infections as well (Arslanoglu, Moro, & Boehm, 2007). This protective effect against atopic dermatitis and infections was still evident at the age of 2 years (Arslanoglu et al., 2008) and preliminary data in a subset indicated protective effects even after 5 years (Arslanoglu et al., 2011). Recent data confirmed the protective effects of supplementation with these oligosaccharides in the development of atopic dermatitis in infants at low risk of developing allergy (Gruber et al., 2010).

Interestingly, recent data showed for the first time the efficacy of specific oligosaccharide mixtures to improve the immune status in HAART-naive HIV-1-infected adults, leading to reduced levels of soluble CD14 (scCD14), CD4(+) T-cell activation, and improved NK cell activity (Gori et al., 2011). All together the immunomodulatory effects induced by non-digestible carbohydrates do have significant biological relevance for allergies, infections as well as inflammation in both adults and infants.

4. In vitro effects

Since microbiota changes are not the only features found other studies were aimed at unravelling direct effect of non-digestible carbohydrates on immune cells. Evidence for direct effects of specific mixtures of oligosaccharides on immune cells have been described, possibly via the interaction with specific sugar receptors (Bode et al., 2004; Eiwegger et al., 2010; Naarding et al., 2005). Oligosaccharides have been shown to interfere with leukocyte recruitment to sites of inflammation and to inhibit cell–cell interactions of leukocytes and lymphocytes via selectins (Eiwegger et al., 2010). Recently, it was demonstrated that scGOS/lcFOS modulated the response of epithelial cells to microbial stimuli, resulting in skewing of underlying PBMCs towards a Th1 phenotype and Treg phenotype (De Kivit et al., 2011). Oligosaccharides have been demonstrated to be able to bind specifically to the lectin-receptor DC-SIGN expressed by dendritic cells. DC-SIGN has been described to interact with a variety of pathogens, including HIV-1, and binding of oligosaccharides inhibited the transfer of HIV-1 to CD4+ T lymphocytes (Naarding et al., 2005). These data suggest that oligosaccharides act systemically and can also modulate immune responses in a microbiota-independent manner, however, more research is needed to identify the underlying mechanisms by which oligosaccharides modulate the immune system.

5. Conclusion

The current body of data on the immune modulatory properties of specific mixtures of oligosaccharides holds great promise for the prevention and or treatment of immune related disorders. Both the development of diseases such as allergies and infections in early life as well as a therapeutic effect in different immune-mediated inflammatory diseases later in life, including cancer and HIV can be affected by adding specific oligosaccharides to the diet. Further understanding and validation of the observed immune modulatory effects of oligosaccharides to realize the full potential of oligosaccharides for a broad range of disorders and treatments is essential and will influence the science of immunology enormously.

References

Angeloni, S., Ridet, J. L., Kusy, N., Gao, H., Crevoisier, F., Guinchard, S., et al. (2005). Glycoprofiling with micro-arrays of glycoconjugates and lectins. *Glycobiology*, *15*, 31–41.

Arslanoglu, S., Moro, G. E., & Boehm, G. (2007). Early supplementation of prebiotic oligosaccharides protects formula-fed infants against infections during the first 6 months of life. *The Journal of Nutrition*, *137*, 2420–2424.

Arslanoglu, S., Moro, G. E., Schmitt, J., Tandoi, L., Rizzardi, S., & Boehm, G. (2008). Early dietary intervention with a mixture of prebiotic oligosaccharides reduces the incidence of allergic manifestations and infections during the first two years of life. *The Journal of Nutrition*, *138*, 1091–1095.

Arslanoglu, S. M., Moro, G. E., Rizzardi, S., Coppola, M., Wiens, F., Stahl, B., et al. (2011). Neutral prebiotic oligosaccharide supplementation early in life and allergy associated with symptoms later on: A five-year follow up. In *ESPGHAN annual meeting*.

Bode, L. (2006). Recent advances on structure, metabolism, and function of human milk oligosaccharides. *The Journal of Nutrition*, *136*, 2127–2130.

Bode, L., Kunz, C., Muhly-Reinholz, M., Mayer, K., Seeger, W., & Rudloff, S. (2004). Inhibition of monocyte, lymphocyte, and neutrophil adhesion to endothelial cells by human milk oligosaccharides. *Thrombosis and Haemostasis*, *92*, 1402–1410.

Boehm, G., Fanaro, S., Jelinek, J., Stahl, B., & Marini, A. (2003). Prebiotic concept for infant nutrition. *Acta Paediatrica Supplement*, *91*, 64–67.

Boehm, G., Stahl, B., Jelinek, J., Knol, J., Miniello, V., & Moro, G. E. (2005). Prebiotic carbohydrates in human milk and formulas. *Acta Paediatrica*, *94*, 18–21.

Boehm, G., & Stahl, B. (2003). Oligosaccharides. In T. Mattilla-Sandholm, & M. Saarela (Eds.), *Functional dairy products* (pp. 201–243). Cambridge: Woodhead Publishing Ltd.

De Kivit, S., Kraneveld, A. D., van de Krant, H. J. G., Schouten, B., Van Esch, B. C., van Kooyk, Y., et al. (2011). Dietary intervention with synbiotics protects against allergic disease via induction of galectin-9 secretion by intestinal epithelial cells. In *ESPGHAN annual meeting*.

Eiwegger, T., Stahl, B., Haidl, P., Boehm, G., Gerstmayr, M., Pichler, J., et al. (2010). Prebiotic oligosaccharides: In vitro evidence for gastrointestinal epithelial transfer and immunomodulatory properties. *Pediatric Allergy and Immunology: Official Publication of the European Society of Pediatric Allergy and Immunology*, *21*, 1179–1188.

Faber, J., van Limpt, K., Kegler, D., Luiking, Y., Garssen, J., van Helvoort, A., et al. (2011). Bacterial translocation is reduced by a specific nutritional combination in mice with chemotherapy-induced neutropenia. *The Journal of Nutrition*, *141*, 1292–1298.

Faber, J., Vos, P., Kegler, D., Luiking, Y., Garssen, J., van Helvoort, A., et al. (2008). Beneficial immune modulatory effects of a specific nutritional combination in a murine model for cancer cachexia. *British Journal of Cancer*, *99*, 2029–2036.

Gibson, G. R., & Roberfroid, M. B. (1995). Dietary modulation of the human colonic microbiota: Introducing the concept of prebiotics. *The Journal of Nutrition*, *125*, 1401–1412.

Gori, A., Rizzardini, G., Van't Land, B., Amor, K. B., van Schaik, J., Torti, C., et al. (2011). Specific prebiotics modulate gut microbiota and immune activation in HAART-naive HIV-infected adults: Results of the COPA pilot randomized trial. *Mucosal Immunology*, *4*, 554–563.

Gruber, C., van Stuijvenberg, M., Mosca, F., Moro, G., Chirico, G., Braegger, C. P., et al. (2010). Reduced occurrence of early atopic dermatitis because of immunoactive prebiotics among low-atopy-risk infants. *The Journal of Allergy and Clinical Immunology*, *126*, 791–797.

Knol, J., Scholtens, P., Kafka, C., Steenbakkers, J., Gro, S., Helm, K., et al. (2005). Colon microflora in infants fed formula with galacto- and fructo-oligosaccharides: More like breast-fed infants. *Journal of Pediatric Gastroenterology and Nutrition*, *40*, 36–42.

Knol, J. B., Lidestri, G., Negretti, M., Jelinek, F., Agosti, J., Stahl, M., et al. (2005). Increase of faecal bifidobacteria due to dietary oligosaccharides induces a reduction of clinically relevant pathogen germs in the faeces of formula-fed preterm infants. *Acta Paediatrica Supplement*, *94*, 31–33.

Meyer, P. D. (2004). Nondigestible oligosaccharides as dietary fiber. *Journal of AOAC International*, *87*, 718–726.

Moro, G., Arslanoglu, S., Stahl, B., Jelinek, J., Wahn, U., Boehm, G., et al. (2006). A mixture of prebiotic oligosaccharides reduces the incidence of atopic dermatitis during the first six months of age. *Archives of Disease in Childhood*, *91*, 814–819.

Moro, G., Minoli, I., Mosca, M., Fanaro, S., Jelinek, J., Stahl, B., et al. (2002). Dosage-related bifidogenic effects of galacto- and fructo-oligosaccharides in formula-fed term infants. *Journal of Pediatric Gastroenterology and Nutrition*, *34*, 291–295.

Naarding, M. A., Ludwig, I. S., Groot, F., Berkhout, B., Geijtenbeek, T. B., Pollakis, G., et al. (2005). Lewis X component in human milk binds DC-SIGN and inhibits HIV-1 transfer to CD4+ T lymphocytes. *Journal of Clinical Investigation*, *115*, 3256–3264.

Obermeier, S., Rudloff, S., Pohlentz, G., Lentze, M. J., & Kunz, C. (1999). Secretion of 13C-labelled oligosaccharides into human milk and infant's urine after an oral [13C]galactose load. *Isotopes in Environmental and Health Studies*, *35*, 119–125.

Roberfroid, M., Gibson, G. R., Hoyles, L., McCartney, A. L., Rastall, R., Rowland, I., et al. (2010). Prebiotic effects: Metabolic and health benefits. *The British Journal of Nutrition*, *104*(Suppl. 2), S1–S63.

Schouten, B., van Esch, B. C., Hofman, G. A., van Doorn, S. A., Knol, J., Nauta, A. J., et al. (2009). Cow milk allergy symptoms are reduced in mice fed dietary synbiotics during oral sensitization with whey. *The Journal of Nutrition*, *139*, 1398–1403.

Vos, A. P., Haarman, M., Buco, A., Govers, M., Knol, J., Garssen, J., et al. (2006). A specific prebiotic oligosaccharide mixture stimulates delayed-type hypersensitivity in a murine influenza vaccination model. *International Immunopharmacology*, *6*, 1277–1286.

Vos, A. P., Knol, J., Stahl, B., M'Rabet, L., & Garssen, J. (2010). Specific prebiotic oligosaccharides modulate the early phase of a murine vaccination response. *International Immunopharmacology*, *10*, 619–625.

Vos, A. P., van Esch, B. C., Stahl, B., M'Rabet, L., Folkerts, G., Nijkamp, F. P., et al. (2007). Dietary supplementation with specific oligosaccharide mixtures decreases parameters of allergic asthma in mice. *International Immunopharmacology*, *7*, 1582–1587.