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Human Milk Oligosaccharides – Nature's Secret Weapon

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Human Milk Oligosaccharides – Background and Metabolism

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Key Messages

Infants receive large amounts of human milk oligosaccharides (HMOs) with a high potential for local effects within the gastrointestinal tract and for systemic functions. In feces and urine, native HMOs and degradation products are present which partly reflect the mothers' specific milk oligosaccharide pattern.

Introduction

In recent years, there has been a tremendous increase in our knowledge regarding specific effects of human milk oligosaccharides (HMOs) which are not or only in trace amounts present in bovine milk (table 1). Concomitantly with these studies, progress in biotechnology nowadays allows to produce at least some HMOs to potentially be added to infant formulas. To decide which compound(s) would be most suitable for supplementation, in which concentrations or combinations, and how long it should be given, studies are needed regarding their metabolic fate as well as their local and systemic effects.

History

Important observations with regard to infants' health have already been made around 1900. The discovery of lactobacilli and bifidobacteria and their relevance for health and disease was an important milestone. At the same time, pediatricians realized that the fecal composition of breast-fed and bottle-fed infants differed. Observations indicated that this difference is particularly linked to the milk carbohydrate fraction. This was the starting point of research on human milk carbohydrates. In the following years, the first HMOs were identified [1]. Studies conducted after 1950 focused on the identification of various HMOs as the 'bifidus factor' in human milk [2]. Since then, about 150 single HMOs have been characterized. It is important to note that the Lewis blood group and the secretor/non-secretor status lead to very specific HMO patterns in milk which are discussed of having an influence on certain diseases [3].

Table 1. Composition of major components in human and bovine milk (in g/l)

	Human milk	Bovine milk
Proteins	10	33
Lipids	40	40
Lactose	65	47
Oligosaccharides	5–15	Not present or only in traces

Structures

Almost all HMOs are based upon lactose which is modified in the mammary gland by the attachment of monosaccharides such as fucose, N-acetylglucosamine, and/or sialic acid (fig. 1). Thus, complex structures with very specific linkages are built, which is the basis for the multifunctionality of HMOs [3, 4].

Physiological Observations as Background to HMO Research

Large amounts of HMOs, i.e. several grams per day, rinse the gastrointestinal tract of a human milk-fed infant, thereby potentially preventing pathogen adhesion to the intestinal mucosa or influencing gut maturation processes [5, 6]. HMOs are considered not to be degraded by human digestive enzymes and transported into the lower parts of the intestine where they may be metabolized by the microbiota or get excreted with feces [7–10]. As about 1–2% of HMOs are excreted via the infants' urine (fig. 2), several hundred milligrams per day may circulate in the infants' blood, which is enough to suppose systemic functions such as anti-inflammatory or anti-infective effects of HMOs.

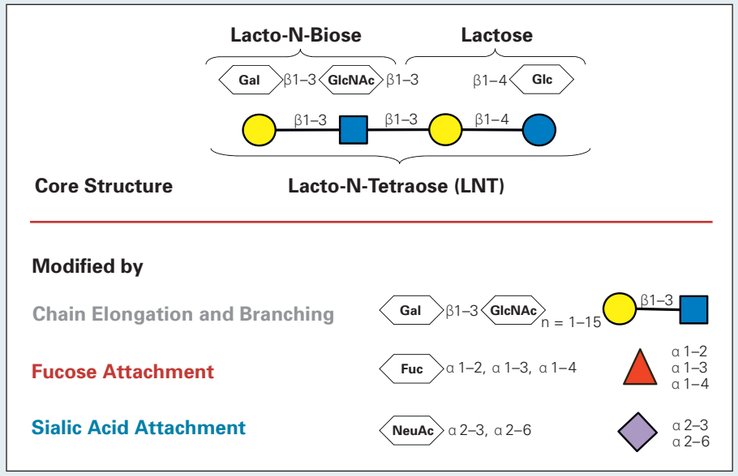

Fig. 1. Composition of HMOs and potential modifications.

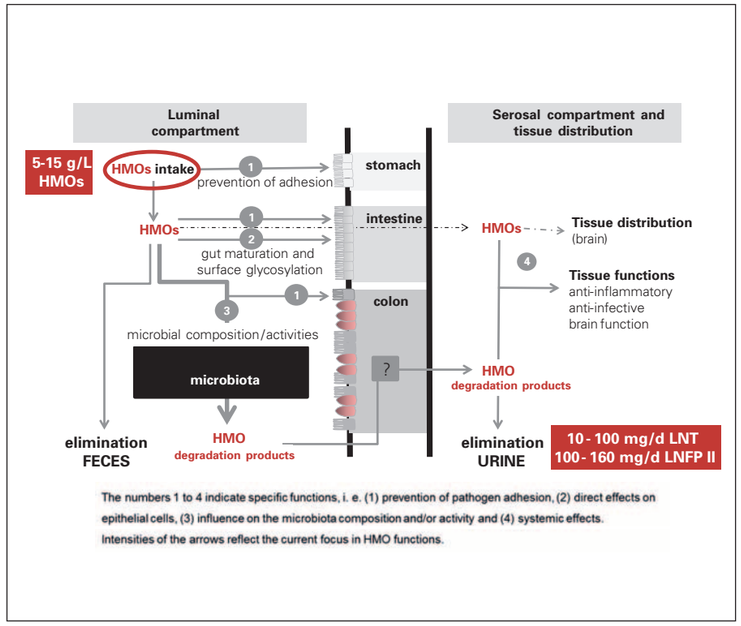

Fig. 2. Intake, metabolism, and potential functions of HMOs.

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Human Milk Oligosaccharides and the Mucosal Immune System

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Key Messages

Human milk oligosaccharides promote the mucosal immune protection through the microbiota and mucosal immune system.

Breastmilk is the recommended and presumed evolutionary adapted nutrition for term-born healthy infants. Numerous meta-analyses showed that breastfeeding for longer periods results in decreased risks primarily for infections, but also for diabetes and overweight, while its effect on allergies is less clear [1]. Among the breastmilk components implicated in immune protection are human milk oligosaccharides (HMOs) [2]. HMOs are elongations of the milk sugar lactose with combinations of galactose, N-acetylglucosamine, fucose, and/or sialic acid. These elongations are structurally similar to glycans exposed on secreted mucins and cell surfaces in the form of glycolipids and glycoproteins. Cell surface glycans stabilize and modulate receptor functions commonly through the interaction with glycan-binding proteins and, due to their dominant, luminal exposure, are often primary docking sites for pathogens. The similarity of HMOs with mucin and cell surface glycans suggests that

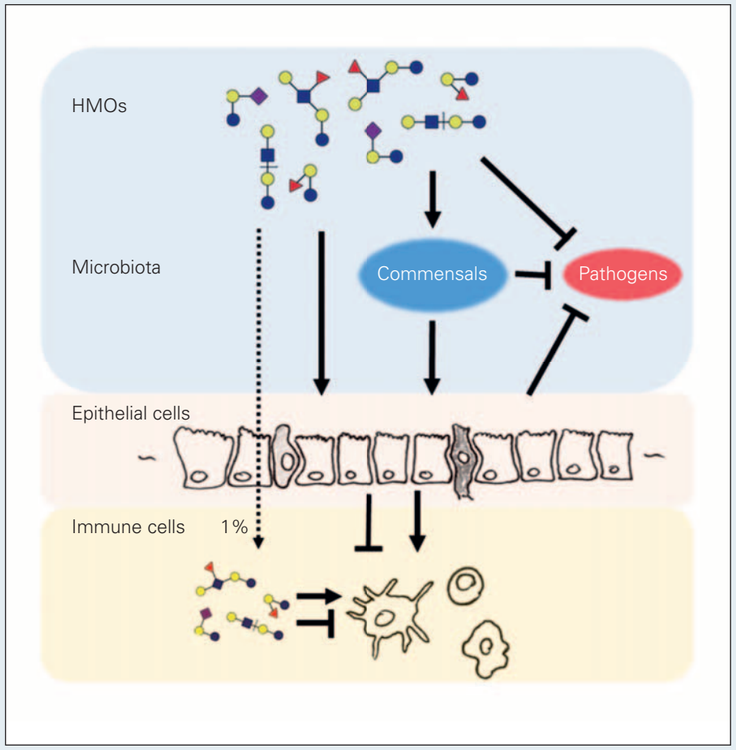

Fig 1. Illustration on how HMOs shape the intestinal environment through their effect on commensals, pathogens, and the mucosal immune system from the epithelium to the underlying immune cells. Arrows indicate 'activation' and bar-headed lines indicate 'inhibition'.

HMOs interfere with glycan-mediated processes that affect (i) the establishment of commensals, (ii) the adherence of pathogens, and (iii) mucosal cell reactivity (fig. 1).

In the intestinal epithelium, enterocytes, enteroendocrine cells, and the chemosensory Tuft cells are candidate signal transducers to the mucosal immune cells. In model systems, HMOs stimulate enterocyte maturation and immune reactivity resulting in improved immune protection [3, 4]. Similarly, HMO exposure of immature human intestinal tissue shifts gene expression profiles towards immune maturation and reduced response to inflammatory stimulation [5].

About 1% of ingested HMOs reach the systemic circulation and are excreted in urine, indicating that HMOs may also directly affect mucosal immune cells lying beyond the epithelial layer. Indeed, HMOs were shown to modulate proliferation, reactivity, and migration of isolated mononuclear cells, T cells, and dendritic cells [6–8].

For such effects, a strong HMO structure-function relation is often observed, meaning that not all HMOs or other oligosaccharides act in the same way. Taken together, an HMO-modulated intestinal environment including both the mucosal immune system and intestinal microbiota might explain at least partly the immune protection provided by prolonged breastfeeding and the proposed link between HMOs and reduced infectious diarrhea [9], lower respiratory tract infection and antibiotic use [10], and delayed onset of allergic eczema in C-section born infants [11].

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Human Milk Oligosaccharides and the Infant Gut Microbiome

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Key Messages

Human milk oligosaccharides (HMOs) are diverse, biologically active components that beneficially modulate the infant microbiota as well as gut, immune, and potentially neurological development. It is now possible to produce large quantities of HMOs enabling supplementation to infant formula, with the goal of supporting the gut microbiota composition and developmental outcomes more similar to that of the breastfed infant.

Factors Influencing Microbial Colonization

Infant gut colonization begins prenatally, continues during the first 2–3 years of life, and is essential for the gastrointestinal, metabolic, neural, and immune development of the infant [1]. Genetic and environmental factors, including mode of delivery, antibiotic use, and diet, shape the colonization process [1, 2]. The gut microbiota of breast- and formula-fed infants differ [3, 4] due, in part, to the high concentrations of human milk oligosaccharides (HMOs) in human milk, which are absent in infant formulas [4].

HMOs Shape the Infant Microbiota

HMOs are resistant to digestion and influence the composition of the infant gut microbiome in several ways: by serving as prebiotics, by acting as substrates for fermentation to short-chain fatty acids, and by reducing pathogens (fig. 1) [3, 4]. The gut microbiota of breastfed infants are typically dominated by bifidobacterial species, with a unique enrichment of *Bifidobacterium longum* spp. *infantis* or *B. infantis* [2, 5]. Most bifidobacterial species that grow on HMOs only metabolize one of the predomi-nant HMOs, namely lacto-N-tetraose, whereas *B. infantis* grows well on several HMOs [6]. Genome sequencing identified that *B. infantis* is unique in that it contains all of the oligosaccharide transport proteins and enzymes needed to transport intact HMOs into the cell, where it is broken down internally [7]. In contrast, other bifidobacterial [8] and *Bacteroides* species [9] have the enzymes that break down the HMOs on their outer cell membrane and then transport the products into the cell for metabolism [7, 8]. If the HMO is hydrolyzed outside the cell, then other bacteria have access to these sugar compounds, which is referred to as cross-feeding [8]. Indeed, different HMOs in milk are both positively and negatively correlated with a number of bacteria in the stool of breastfed infants. Thus, HMOs have broad effects on shaping the infant microbiome, and it is possible to identify which HMO types are prebiotics for specific bacteria in the infant's stool [2, 10].

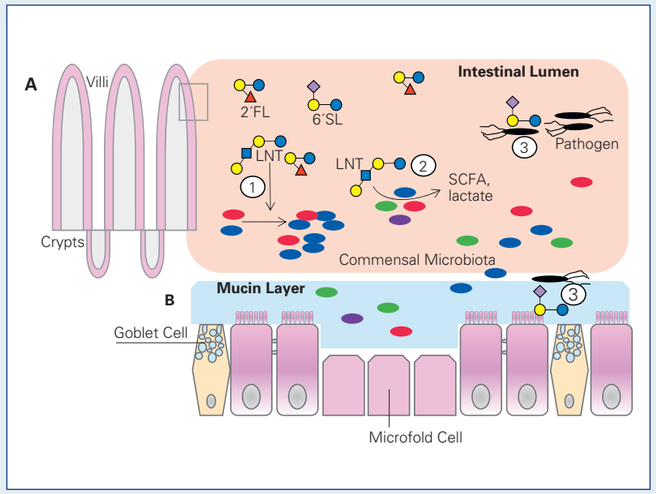

Fig. 1. Potential ways in which HMOs shape the infant gut microbiome. The overall structure of the intestine is shown in **A** and a higher detail of the intestinal lumen is shown in **B**. HMOs can act as prebiotics to increase the growth of *Bifidobacteria* and *Bacteroides* species (1). HMOs are also fermented to produce short-chain fatty acids (SCFAs) acetate, propionate, and butyrate as well as lactic acid. SCFAs reduce the pH of the lumen and can be used by other bacteria and the host (2). Sialylated and fucosylated HMOs can bind to pathogens in the lumen or to receptors on epithelial cells to inhibit binding of pathogens with the host and reduce diarrheal disease (3).

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