# Human Milk Oligosaccharides (HMOs) and Their Health Benefits
## Research Review

## INTRODUCTION

An oligosaccharide is a carbohydrate polymer, or glycan, that contains a small number (typically 3 to 7) of monosaccharides. Human milk is unique for its abundant amounts of oligosaccharides, whereas milk from other mammals, such as cows, only has trace amounts of oligosaccharides (**Table 1**). Human milk oligosaccharides (HMOs) have been the subject of intense research to characterize their structure and function with the hypothesized potential to benefit not only infants but also children and adults.

The building blocks of HMOs are 5 monosaccharides: glucose, galactose, N-acetylglucosamine, fucose, and sialic acid, primarily in the form of N-acetylneuraminic acid. Each HMO has lactose (a disaccharide consisting of galactose and glucose) at the reducing end. Lactose itself can be fucosylated to form isomeric trisaccharides 2'-fucosyllactose (2'-FL) and 3'-fucosyllactose (3'-FL) or sialylated to form 3'-sialyllactose (3'-SL) and 6'-sialyllactose (6'-SL). To form more complex HMOs, lactose at the reducing end can be elongated and branched via the action of glycosyltransferases; elongated chains can further be fucosylated and sialylated in different linkages.[1]

**Table 1.** Approximate Values of Macronutrients and Oligosaccharides in Human Milk vs. Bovine Milk

|  | Human milk | Bovine milk |
|---|---|---|
| Protein (g/L) | 8 | 32 |
| Fat (g/L) | 41 | 37 |
| Lactose (g/L) | 70 | 48 |
| Oligosaccharides (g/L) | 5 - 15 | 0.05 |
| % fucosylated | 50% - 80% | ~1% |
| % sialylated | 10% - 20% | ~70% |
| Number of identified oligosaccharides | >200 | ~40 |

(Chart adopted from Bode.[1])

Although more than 200 HMOs have been identified, the amount/types of HMOs vary depending on a woman's genetic variations. For example, 2'-FL is the most abundant HMO but it is not detected in the milk obtained from women of non-secretor blood type (~20% of the world's population are non-secretors[2]) due to lack of the enzyme α1-2 fucosyltransferase 2 (FUT2).[3]

HMOs resist the low pH in the stomach and enzymatic degradation in the digestive tract, and reach the colon in an intact form, behaving as soluble prebiotic fibers.[4]

## BENEFICIAL PROPERTIES OF HMOs

**Prebiotic effects.** Prebiotics are defined as a selectively fermented ingredient that results in specific changes in the composition and/or activity in the gastrointestinal (GI) microbiota, thus conferring benefits upon host health.[5] HMOs meet the criteria for prebiotic classification.

In an in vitro fermentation model, HMOs supplementation to infant fecal samples significantly increased the number of beneficial *Bifidobacterium longum* spp. *infantis*, whereas the numbers of pathogenic *Escherichia* spp. and *Clostridium perfringens* decreased.[6] Concentrations of lactate and short-chain fatty acids (SCFAs) were significantly higher and the pH values significantly lower in the HMOs-supplemented fecal cultures compared with control cultures. Over 90% of 2'-FL from the HMOs cultures were metabolized by fecal bacteria.[6] The produced SCFAs created an environment favoring the growth of beneficial bacteria over potential pathogens.[7]

**Antiadhesive antimicrobials.** To colonize or infect the host, many bacterial, viral, or protozoan pathogens first adhere to the host by expressing binding proteins that attach to specific glycans on the host's epithelial cell surface.[8] HMOs reduce microbial infections by serving as soluble decoy receptors that prevent pathogen binding to host cells.[9,10]

*Campylobacter jejuni*, one of the most common causes of bacterial diarrhea in humans, binds to intestinal epithelial α1-2-fucosylated glycans.[11] Fucosylated HMOs (including 2'-FL) resemble such cell surface glycans, and inhibited or prevented *Campylobacter* infection or colonization in vivo and ex vivo.[11] This finding was supported by a cohort study; the incidence of *Campylobacter* diarrhea in breastfed infants was inversely associated with the amount of 2'-FL in the mother's milk.[12]

Other in vitro studies found that HMOs inhibited the adhesion to intestinal epithelial cells of pathogens *Escherichia coli* serotype O119, *Vibrio cholerae*, *Salmonella fyris*, and norovirus, and 2'-FL and 3'-FL inhibited the adhesion to respiratory epithelial cells of *Psudomonas aeruginosa*.[13-15]

**Modulators of intestinal epithelial cell responses.** In vitro studies found that HMOs have direct effects on intestinal epithelial cells. For instance, human intestinal epithelial cell lines incubated with 3'-SL had lower gene expression of sialyltransferases and diminished sialylation on cell surface glycans, the attachment sites for the pathogen *E. coli*. As a result, binding of *E. coli* to epithelial cells was significantly reduced.[16] Also, HMOs inhibited proliferation and induced differentiation and apoptosis in cultured human intestinal cell lines by altering corresponding regulator genes.[17,18]

**Immune modulators.** Research using ^{13}C-labeling of HMOs found that ~1% of HMOs are absorbed and reach the systemic circulation and thus may have potential systemic effects.[19,20]

An in vitro study found that, when cord blood mononuclear cells from healthy newborns were co-incubated with sialylated HMOs, the percentage of interferon-γ producing CD3+CD4+ and CD3+CD8+ lymphocytes and the production of IL-13 in CD3+CD8+ cells were significantly increased.[21] This suggests that sialylated HMOs may influence lymphocyte maturation in breastfed newborns. Experimental studies also found that sialylated HMOs modulate selectin-mediated cell-cell interaction. Selectins are adhesion molecules expressed by endothelial cells at the presence of proinflammatory cytokines and mediate leukocyte trafficking and platelet-neutrophil complex formation. Sialylated HMOs reduced leukocyte rolling and adhesion in TNFα-activated human endothelial cells in vitro and reduced platelet-neutrophil complex formation and subsequent neutrophil activation ex vivo.[22,23]

Lipopolysaccharides (LPS) on the membrane of *E. coli* can swiftly trigger secretion of proinflammatory interleukin-8 (IL-8) by epithelial cells.[24] In vitro research demonstrated that pretreatment of intestinal epithelial cells with HMOs or 2'-FL led to reduced adherence and invasion of enterotoxigenic *E. coli* and IL-8 release via attenuation of CD14 induction. In vivo research confirmed the ability of 2'-FL to inhibit inflammation associated with *E. coli* invasion and CD14 expression in *E. coli*-infected mice.[25]

## SUMMARY

HMOs function as prebiotics that promote growth of beneficial gut microbiota (e.g., *Bifidobacterium longum* spp. *infantis*) and the production of SCFAs that create an environment that discourages the growth of potential pathogens. HMOs are antiadhesive antimicrobials, serving as soluble decoy receptors, helping to prevent pathogen adherence to mucosal surfaces, thus reducing the risk for infections. Also, HMOs have been shown to modulate epithelial and immune cell responses. These postulated benefits may explain why human milk has been referred to as the "gold standard" of infant nutrition, and why breastfed infants have a much higher chance of survival and lower chance of infection (including infectious diarrhea) than bottle-fed infants. HMOs' benefits may go beyond infants. Indeed, their effect on the fecal microbiota and GI symptoms in adults has already been the subject of a clinical trial (NCT01927900). HMOs, such as 2'-FL, can now be synthesized via in vitro fermentation—which means they can be produced on a large scale and may be utilized as invaluable dietary ingredients to promote human health.

**References**

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