Prebiotics to manage the microbial control of energy homeostasis

Grootaert, C.[^], Marzorati, M.[^], Van den Abbeele, P.[^], Van de Wiele, T. and Possemiers, S.*

Laboratory of Microbial Ecology and Technology (LabMET), Ghent University, B-9000 Ghent, Belgium.

*: Corresponding author: POSSEMIERS Sam, UGent. Coupure Links 653, B-9000 Gent, Belgium. Phone: +32 9 264 59 76, Fax : +32 9 264 62 48. sam.possemiers@ugent.be

[^]: these authors equally contributed to the review

Key Words: Prebiotics, obesity, homeostasis, diabetes, mucosa

Abstract

The prevalence of obesity is continuously growing and has reached epidemic proportions. It is clear that current methods to combat obesity are not effective enough to reduce the problem. Therefore, further investigation is needed to develop new strategies. Recent research pointed out a potential role of the microbial community associated to the human host in controlling and influencing the energy homeostasis. According to the concept of Gastrointestinal Resource Management, this microbiota and its metabolic potential can be steered with the aim of improving host health. This review therefore focuses on the modulation of the intestinal microbiota through prebiotics with the aim to control of several aspects of metabolic homeostasis. In a first part, the importance of host-microbe cross-talk at the intestinal epithelium is discussed. Yet, energy metabolism, which includes both lipid and glucose metabolism, is also regulated by several key organs including the adipose tissue, brain, liver, muscles, pancreas and gut. Therefore, in a second part, we will discuss the microbial factors that are involved in the communication between these different tissues, and their potential management. Finally, we will give some future prospects of the use of prebiotics in an individualized treatment of metabolic disorders.

Introduction

Humans and microbes co-evolved for several thousands of years. Such an interaction is so bounding that - according to the hologenome theory of evolution - the host organism and its microbiota can be considered as a holobiont, a unit of selection in evolution (Rosenberg and Zilber-Rosenberg, 2011). This complex microbiota can be seen, in economic terms, both as an asset and liability with the capability of influencing the fitness of the host (Possemiers *et al.*, 2009).

In 2004-2005, the first reports about the effect of gut microbiota on the development of obesity and energy metabolism were published by the group of Jeffrey Gordon (Bäckhed *et al.*, 2004; Ley *et al.*, 2005). Since then, the microbial impact on several aspects of metabolic homeostasis was investigated (Burcelin *et al.*, 2009; Cani and Delzenne, 2007; Maurer *et al.*, 2009), such as the effect on lipid metabolism and atherosclerosis (reviewed by Caesar *et al.* (2010)), on metabolic syndrome (reviewed by Cani and Delzenne (2009); Sanz *et al.* (2010); Tilg (2010); Wellen and Hotamisligil (2005)) and insulin resistance and diabetes (reviewed by Delzenne and Cani (2010); Musso *et al.* (2011)). Although the mechanisms of action and the triggering factors are not fully understood, it is commonly accepted that the final health effect is the result of a complex interplay between various bacteria, which interact through various mechanisms with the host.

The capacity and the possibility to interfere in these complex interactions has been defined in the intuitive concept of Gastrointestinal Resource Management (GRM), i.e. the management of the complex gut microbiota and its metabolism with the aim of improving the health of the host (Possemiers *et al.*, 2009).

The use of prebiotics is a possible example of how to try to bring to practice the GRM concept. A prebiotic action is defined as the selective stimulation of growth and/or activity(ies) of one or a limited number of microbial genus(era)/species in the gut microbiota that confer(s) health benefits to the host (Roberfroid *et al.*, 2010).

The focus of this work will be to review the available knowledge on the effect of prebiotics on several aspects of metabolic homeostasis. In a first part, the importance of host-microbe cross-talk at the intestinal surface level is discussed. In a second part, we will discuss the microbial factors that are involved in the cross-talk between different organs of the host, and their potential management. Finally, we will give some future prospects on the use of prebiotics in an individualized approach to control metabolic disorders.

Part 1. The intestinal surface as site of host-microbe crosstalk and its barrier function

Host-microbiota cross-talk at the intestinal surface along the intestinal tract

Around 10^{14} microbes colonize the human gut with a coding capacity exceeding that of the host by a factor 100 (Egert *et al.*, 2006). Throughout evolution, humans co-evolved with this abundant microbiota and an intimate interaction came to existence (Zaneveld *et al.*, 2008). Van den Abbeele *et al.* (2011) recently reviewed the cross-talk at the host-microbial interface, which was crucial during this co-evolution. In addition, this cross-talk is relevant for the disturbances of the host-microbiota association, which can lead to disease states such as allergies (Bjorksten *et al.*, 1999), inflammatory bowel diseases (Garrett *et al.*, 2007) and obesity (Turnbaugh *et al.*, 2006). It should be noted that the latter diseases are currently in the rise (Blaser, 2006). Key in the host-microbe cross-talk mostly is that the host continuously detects microbial signals through strategically localized host receptors (Medzhitov and Janeway, 2002). These microbial signals are referred to as microbe-associated molecular patterns (MAMPs). While fungi and viruses are often recognized through their β -glucans and

nucleic acids, respectively, bacteria are often detected through lipopolysaccharides (LPS), peptidoglycans and teichoic acids (reviewed by Van den Abbeele *et al.* (2011). The host receptors that detect these MAMPs are called pathogen recognition receptors (PRRs) and include a diverse set of transmembrane (e.g. Toll-like receptors) (Takeda *et al.*, 2003), cytosolic (e.g. NOD-like receptors) (Ting *et al.*, 2008) and secreted receptors (e.g. collectins) (Gupta and Surolia, 2007). This allows the host to characterize the nature of the microbial signal and respond appropriately. The resulting host response includes production of antimicrobial peptides (Medzhitov and Janeway, 1997), activation of adaptive immune cells and production of resulting effector molecules including e.g. Immunoglobulin A (IgA) (Macpherson and Uhr, 2004). As a result of the continuous detection of microbes, host defence molecules are continuously secreted and trapped in the overlaying mucus layer, which allows the host to particularly control the composition and abundance of the mucosa-associated microbiota (Figure 1). Specific microbial characteristics such as capacity to adhere to the mucus layer, oxygen tolerance, the ability to degrade host-derived glycans further determine the unique composition of the mucosal microbiota (Van den Abbeele *et al.*, 2011). While mucosal microbes would be crucial for priming the immune system or increasing the bioavailability of beneficial microbial metabolites at the intestinal surface, the luminal microbiota have an important metabolic function.

Given the fact that humans closely interact with their co-evolved luminal and mucosal intestinal microbiota, there is great interest in dietary interventions with e.g. prebiotic compounds that are able to modulate both the luminal and mucosal microbial composition and activity (Langlands *et al.*, 2004; Van den Abbeele *et al.*, 2011). In that way, prebiotics may beneficially steer the host-microbe interactions.

Importance of a proper mucosal barrier and risks in case of increased permeability

Over the last few years, the research group of Nathalie Delzenne produced several groundbreaking papers regarding the onset of obesity and its related metabolic disorders. In several studies, one investigated the impact of a high-fat diet on the intestinal microbiota and rodent hosts (Cani *et al.*, 2007a; Cani *et al.*, 2008; Cani *et al.*, 2006; Cani *et al.*, 2007d; Cani *et al.*, 2009b; Neyrinck *et al.*, 2011). Rodents fed a high-fat diet suffered from impaired gut barrier function. This barrier function is crucial since it forms the basis of the strategic localization of PRRs and subsequent detection of MAMPs. This can be illustrated by the detection of lipopolysaccharides (LPS), a MAMP, through Toll like receptor 4 (TLR-4) (Cario and Podolsky, 2000) and nucleotide-binding oligomerisation domain-1 (NOD-1) (Girardin *et al.*, 2003) (Figure 1). TLR-4 is only in low levels expressed on the apical side of the epithelium while NOD-1 is expressed inside the cell. In this way, LPS is not detected on the apical side where it merely derives from the commensal mucosa-associated microbes but on locations where its presence may derive from potentially dangerous microbes. The increased permeability which is caused by the high-fat diet leads to LPS leakage through the gut wall ultimately leading to increased LPS levels in the blood (endotoxemia). At that point, LPS of commensals is overly detected by the PRRs of the host resulting in inflammatory responses and symptoms of metabolic disorder (Cani *et al.*, 2007a; Cani *et al.*, 2008).

It has been shown that the detrimental effect corresponding to metabolic disorders can be partially reversed by reinforcing the gut barrier function. This can be obtained through modulation of the gut microbiota with (potential) prebiotic compounds such as fructans (Cani *et al.*, 2006; Cani *et al.*, 2007d; Cani *et al.*, 2009b) and long-chain arabinoxylans (Neyrinck *et al.*, 2011). The exact nature of the microbial modulations throughout these experiments remains to be elucidated although strong indications exist that specific microbial groups may play a major role. Firstly, as bifidobacteria decreased during fat feeding (Cani *et al.*, 2007d),

while their abundance increased during supplementation of the (potential) prebiotic compounds (Cani *et al.*, 2006; Cani *et al.*, 2009b; Neyrinck *et al.*, 2011), this genus may have an important protective role towards barrier integrity (Khailova *et al.*, 2009). Also other studies indicate that increased bifidobacteria levels are correlated with normal weight in children (Lundell *et al.*, 2007) and women (Collado *et al.*, 2008), while overweight in these studies corresponded to lower bifidobacteria abundances. Improved gut barrier function has also been attributed to specific *Lactobacillus* spp. through protection of the epithelial tight junctions during external stress (Montalto *et al.*, 2004; Seth *et al.*, 2008).

Besides reinforcing tight junctions between epithelial cells, restricted permeability of the gut wall may also be achieved through elevated secretion of mucin. A future focus may thus be to analyze the mucin composition of the mucus layer, which overlies the epithelium upon prebiotic treatment. This mucus layer normally consists of a double protective layer: a very dense, firmly attached and quite sterile inner mucus layer and a less dense, loosely attached, more strongly colonized outer mucus layer (Johansson *et al.*, 2010; Schreiber, 2010). Prebiotics are typically shown to increase mucin-levels by decreasing the pH (Barcelo *et al.*, 2000; Shimotoyodome *et al.*, 2000), increasing the mechanical stimulation by increased intestinal content and tissue weight (Schmidt-Wittig *et al.*, 1996) or increasing the butyrate production (Barcelo *et al.*, 2000), especially by species residing in the mucosal environment (Van den Abbeele *et al.*, 2011). In contrast, the type of mucin that is produced upon administration of a prebiotic compound is something which has often been neglected. This may be important as specific muc-types, such as the membrane-bound Muc17 (mouse homolog Muc3), have shown to promote epithelial barrier integrity (Resta-Lenert *et al.*, 2011). Further, it has been shown that a mix of *Lactobacillus reuteri* strains is able to reach the epithelium and prevent inflammation and translocation in DSS-treated mice. It was proposed that this might be due to an increased expression of membrane-bound Muc3 (Schreiber, 2010).

In conclusion, the loss of gut barrier integrity leading to increased infiltration of microbial signals may be an important factor at the onset of obesity and its related metabolic disorders. Moreover, prebiotics may be protective by avoiding this loss of barrier integrity.

Part 2. Microbial regulation of host signals involved in lipid metabolism

Energy metabolism is regulated by several key organs including the adipose tissue, brain, liver, muscle, pancreas and the gastrointestinal tract (GIT). The diverse host parameters that are involved in these processes are listed in Table 1. This part of the review will focus on the microbial factors that are involved in the communication between the different tissues, and their potential management with prebiotics.

Impact of prebiotics on gut peptides involved in fat storage

Prebiotics may possibly have an effect on gut peptides that are involved in fat storage. The fasting induced adipose factor (FIAF), also known as angiopoietin-like protein 4 (ANGPTL4), has been thoroughly investigated as a multifunctional signal protein produced by many tissues such as the liver (Kim *et al.*, 2010), adipose tissue (Dutton and Trayhurn, 2008), intestine (Bäckhed *et al.*, 2004) and hypothalamus (Kim *et al.*, 2010). Once secreted in the blood, FIAF inhibits the activity of lipoprotein lipase, an enzyme responsible for the conversion of triglycerides to monoglycerides and fatty acids from circulating lipoproteins (Mandard *et al.*, 2006; Yoshida *et al.*, 2002). As a consequence, these triglycerides cannot be stored in the fat tissue, resulting in a lower body weight (Bäckhed *et al.*, 2004).

A particular feature of the intestinal FIAF gene is that its expression is strongly regulated by the presence of an intestinal microbial community (Bäckhed *et al.*, 2004; Fleissner *et al.*, 2010). Intestinal FIAF expression is significantly repressed in mice with a normal intestinal microbial community compared to germ-free mice. Further, conventionalization of these germ-free animals with intestinal bacteria significantly decreased FIAF levels, resulting in an enhanced fat storage and weight gain (Bäckhed *et al.*, 2004). Moreover, elevated FIAF levels may protect germ-free mice against certain types of high-fat diet-induced obesity through induction of the peroxisome proliferator-activated receptor- γ coactivator-1 α (Pgc-1 α), thereby regulating genes involved in energy metabolism (Bäckhed *et al.*, 2007; Fleissner *et al.*, 2010). Fleissner *et al.* (2010) reported that, despite the intestinal FIAF repression in conventionalized mice, their plasma FIAF levels were not decreased as compared to germfree mice. In contrast, in conventionalized mice, a higher concentration of cleaved FIAF was observed whereas the native FIAF concentration was unchanged. These results suggest that the microbial community increases (cleaved) FIAF in sites of the body other than the intestine. Therefore, the impact of the microbial community on FIAF regulated processes should be further explored, not only in the intestine, but also in other parts of the human body. To the best of our knowledge, only one substrate with impact on FIAF expression was reported. In obese mice fed a high fat diet, it was observed that chitosan from mushrooms significantly decreased FIAF expression in visceral adipose tissue (Neyrinck *et al.*, 2009). It was however not investigated whether this was due to shifts in either microbial fermentation products or in composition of the microbial community.

Grootaert *et al.* (2011) showed that SCFAs such as butyrate and propionate, but not acetate, stimulate FIAF transcription in several colorectal and hepatic cancer cell lines. When investigating the effect of specific intestinal monocultures, differential effects on FIAF expression were identified. An *in vivo* mouse study from Bäckhed *et al.* (2007) demonstrated that FIAF production was more repressed when inoculating germ-free mice with a combination of *Bacteroides thetaiotaomicron* and *Methanobrevibacter smithii*, than with each of them separately. Incubation of HCT-116 cells with *E. coli* resulted in decreased FIAF secretion (Grootaert *et al.*, 2011). In contrast, *in vitro* incubations of intestinal HT-29 and Caco-2 cells with *Enterococcus faecalis* increased FIAF production after a few hours (Are *et al.*, 2008; Grootaert *et al.*, 2011). Similarly, it was shown that *Lactobacillus paracasei* ssp *paracasei* F19, *Lactobacillus rhamnosus* GG and *Bifidobacterium lactis* BB12, and not *Bacteroides thetaiotaomicron*, were able to stimulate FIAF expression in several colonic cell lines including HCT-116, HT-29, LoVo and SW-480 cells (Aronsson *et al.*, 2010). In the case of *Lactobacillus* F19, the FIAF stimulatory effect was attributed to a secreted microbial factor, which was however not identified in the study. In addition, conventionalization of mice with *Lactobacillus* F19 increased native FIAF levels in blood plasma, and resulted in decreased fat storage and increased blood VLDL levels.

Summarized, although FIAF is an interesting molecule to focus on for prebiotic treatment, its functionality largely depends on the site of production, isoform appearance and final target organs. Until now, the most dominating effect of microbial FIAF modulation is not identified yet and needs further investigation with relevant models.

Prebiotics that alter energy intake through satiety signals

Prebiotics may also be used to decrease appetite by modulation of specific hormones involved in appetite and satiety. Leptin is a hormone mainly produced by adipose tissues and inhibits food intake. Prebiotic substrates such as chitosan decrease the production of leptin in adipocytes in high fat diet-induced obese mice (Neyrinck *et al.*, 2009), although the exact mode-of-action is not known. In addition, leptin was also decreased in rats weaned with

inulin-containing high fiber diets (Maurer *et al.*, 2009). Xiong *et al.* (2004) demonstrated that SCFA belonging to C2-C6 fatty acids stimulate leptin in murine adipocyte cell lines and primary adipocyte cell culture. In case of propionate, significantly increased leptin production was attributed to increased GPR41 activation. Leptin stimulation through G-coupled protein receptors was also shown in human adipose tissue (Lahham *et al.*, 2008).

Glucagon-like peptide 1 (GLP-1) and peptide YY (PYY) are satiety stimulating hormones, released in response to nutrient ingestion by L-cells in mainly ileum and colon. GLP-1 promotes insulin secretion and pancreatic β -cell proliferation and controls glycogen synthesis in muscle cells (Delzenne *et al.*, 2007), whereas PYY slows down gastric emptying. In contrast, ghrelin stimulates appetite and is mainly produced by P/D1 cells in the stomach and ϵ -cells of the pancreas (Inui *et al.*, 2004).

Non-digestible carbohydrates, such as oligofructose (Cani *et al.*, 2007b; Maurer *et al.*, 2009; Piche *et al.*, 2003), lactitol (Gee and Johnson, 2005) and resistant starch (Zhou *et al.*, 2008) are effective to induce satiety by modulating the production of the gut peptides GLP-1, PYY and ghrelin through a mechanism that also involves modulation of the intestinal microbial community (Cani *et al.*, 2007b). For instance, rats fed a oligofructose-enriched diet showed a significantly increased GLP-1 and decreased ghrelin production, and doubled the number of GLP-1-expressing cells in the proximal colon (Cani *et al.*, 2004; Cani *et al.*, 2007c). In addition, also human studies showed higher plasma GLP-1, PYY and/or ghrelin levels after intake of oligofructose (Cani *et al.*, 2009a; Parnell and Reimer, 2009; Piche *et al.*, 2003), which may explain the increased satiety feeling and decreased energy intake behaviour of the subjects. Finally, lactitol mainly increased PYY production both in rats and humans (Gee and Johnson, 2005), whereas resistant starch significantly increased both PYY and GLP-1 production (Zhou *et al.*, 2008). The bacterial regulation of gut peptides is mediated by SCFA produced from these indigestible substrates. Physiological concentrations of acetate, propionate and butyrate, but also a pH decrease from 7.5 to 6, significantly increased proglucagon and PYY in the entero-endocrine colon cell line STC-1 (Zhou *et al.*, 2008). In addition, the presence of glucose in the intestine also enhances the GLP-1 production in the L-cells (Egan and Margolskee, 2008). These mechanisms may explain why gut peptide modulation is only observed with highly fermentable fibers (Massimino *et al.*, 1998).

Recently, also FIAF was identified as a potential signal protein with effect on hypothalamic control of appetite. Bacteria by means of LPS are able to induce a low-grade inflammation, as already discussed (Cani *et al.*, 2007a). Brown *et al.* (2009) showed that when mice were treated with LPS, body weight was significantly decreased and increased levels of FIAF were observed in the hypothalamic, pituitary, cortical and adipose tissues. Similar effects on FIAF levels were observed when N-1 neuronal and 3T3-L1 adipocyte cells were treated with LPS (Brown *et al.*, 2009). Therefore, FIAF is considered as one of the mediators of hypothalamic control of appetite and energy metabolism through LPS. In fact, LPS-induced endotoxemia was also associated to an anorectic response via hypothalamic-dependent mechanisms (Huang *et al.*, 1999; Rummel *et al.*, 2008). Yet, it is not desirable to steer this FIAF response by addition of LPS to the host, as LPS stimulates inflammatory responses in other tissues.

In summary, we conclude that the influence of microbiota on satiety hormones is a promising issue for prebiotic treatment, especially for substrates that enhance SCFA production. Indeed, a large part of the SCFAs are transported into the blood stream, thereby targeting several tissues, such as the adipose tissue, which may be induced to produce hormones involved in appetite and satiety.

Prebiotic modulation of cholesterol and lipid metabolism

Several pro- and prebiotics may have a possible effect on serum cholesterol and lipid levels, thereby not only affecting fat storage, but also the development of cardiovascular diseases (reviewed by Ooi and Liong (2010) and Williams (1997)). Several mechanisms have been proposed, including enzymatic deconjugation of bile acids (Bongaerts *et al.*, 2000), assimilation of cholesterol (reviewed by St-Onge *et al.* (2000)), co-precipitation of cholesterol with deconjugated bile, cholesterol binding to bacterial cell walls, incorporation of cholesterol into microbial cell membranes, conversion of cholesterol into coprostanol and production of short-chain fatty acids upon fermentation (reviewed by Ooi and Liong (2010)). Examples of probiotic bacteria influencing cholesterol and lipid metabolism are *Lactococcus lactis* (Nakajima *et al.*, 1992), *Streptococcus thermophilus* (Pulusani and Rao, 1983; Richelsen *et al.*, 1996), *Lactobacillus acidophilus* (Gilliland *et al.*, 1985), *E. faecium* (Richelsen *et al.*, 1996), *Bifidobacterium bifidum* (Beena and Prasad, 1997; Mohan *et al.*, 1996) and *B. longum* (Xiao *et al.*, 2003).

Several human and rodent *in vivo* studies also mention the cholesterol and lipid lowering effects of oligofructose (de Luis *et al.*, 2010; Delzenne *et al.*, 1993; Fiordaliso *et al.*, 1995; Trautwein *et al.*, 1998; Williams and Jackson, 2002), xylo-oligosaccharides (Hsu *et al.*, 2004), chito-oligosaccharides (Li *et al.*, 2007), soybean oligosaccharides (Chen *et al.*, 2010) and resistant starch (Cheng and Lai, 2000; Venter *et al.*, 1990). These effects may be linked to the production of propionate, which inhibits hepatic cholesterol synthesis from acetate (Berggren *et al.*, 1996; Lin *et al.*, 1995; Todesco *et al.*, 1991). Yet, the concentration of propionate that is needed to induce the cholesterol and lipid lowering effect is 10 to 100 fold higher for human than for rat hepatocytes (Lin *et al.*, 1995).

Hepatic lipogenesis is not only regulated by short chain fatty acids, but also by serum glucose and insulin levels (Towle, 2001). Enhanced sugar uptake has been observed in presence of gut bacteria compared to germ-free conditions, which can be explained by several mechanisms. First of all, the presence of an intestinal microbial community leads towards an increase in the amount of capillaries that underlie the small intestinal epithelium (Hooper *et al.*, 2002). Secondly, host monosaccharide transporters are induced by the polysaccharide-processing activity of the microbiota, as was demonstrated by studies with germ-free mice colonized with *B. thetaiotaomicron* (Hooper *et al.*, 2001). The monomers generated from indigestible polysaccharides are delivered as substrates for lipid production in the liver. Besides, they may also activate the lipogenic enzymes in the liver by ChREBP- and SREBP-1- mediated mechanisms (Bäckhed *et al.*, 2004). Hence, the polysaccharide-degrading potential of an intestinal microbial community may be an important determinant for hepatic lipid production. In obese *ob/ob* mice, the intestinal microbial community is enriched for genes that are able to harvest calories from complex plant-derived polysaccharides compared to lean mice. These genes encode for enzymes involved in sugar degradation, sugar transport and acetate and butyrate production (Turnbaugh *et al.*, 2006).

We conclude that the potential influence of intestinal microbiota and prebiotics on lipid and cholesterol production in the host is a complicated process which involves several nutrients, target organs and signalling pathways. More investigation is warranted to identify the dominating mechanism by which prebiotic modulation of the intestinal microbial community may contribute to lipid and cholesterol lowering effects.

Part 3: Prebiotic modulation of glucose and insulin metabolism

As mentioned before, intestinal bacteria can impact food intake and lipid metabolism, but may also be involved in carbohydrate maintenance and disturbances thereof, such as insulin resistance. Insulin resistance is the fundamental defect in type 2 diabetes, a disease that afflicts 6% of adult Americans, up from 3% in the early 1970s (Taubes, 2009). A role of

330 dietary fibers in general and prebiotics in particular has been shown in regulating glucose
331 maintenance in numerous studies and will therefore also be used in this part to illustrate the
332 role of intestinal bacteria.

333 Indeed, consumption of whole grains dietary fibers has been shown to improve blood glucose
334 and insulin responses (reviewed by Gemen *et al.* (2011)). It is now believed that the
335 underlying mechanism is of a multifactorial nature with different activity profiles throughout
336 the gastrointestinal tract (Figure 2). Firstly, inclusion of dietary fiber in dietary products may
337 replace part of the available carbohydrates in the food product, leading to lower glycemic
338 response. For instance, resistant starch lowers the glycemic index, by being indigestible in the
339 upper intestine, as opposed to digestible starch. The resistance to digestion of resistant starch
340 is mainly attributed to particular physical structures, such as the amylose / amylopectin ratio.
341 A higher ratio leads to a more branched polymer structure, which is less susceptible to
342 enzymatic digestion in the small intestine (Brouns *et al.*, 2002; Fassler *et al.*, 2006; Storey *et al.*,
343 2007; Venter *et al.*, 1990).

344 Secondly, depending on their structure, dietary fibers such as arabinoxylans and beta-glucans,
345 form a viscous solution in the stomach, thereby delaying gastric emptying and physically
346 trapping nutrients, such as glucose and thereby reducing their absorption. In addition, the
347 passage of digestive enzymes through the viscous food bolus is limited, which reduces the
348 hydrolysis by digestive enzymes (Mohlig *et al.*, 2005; Regand *et al.*, 2009; Wood *et al.*,
349 2000). The combination of these processes will again lower the glycemic response. Reduced
350 serum glucose concentrations decrease the amount of insulin needed to clear the glucose load.
351 Upon repeated consumption of such fiber, the reduced ambient insulin concentrations may
352 result in an up-regulation of cell surface insulin receptors, thereby increasing insulin
353 sensitivity (Song *et al.*, 2000).

354 As mentioned before, changes in the intestinal bacterial community are involved in obesity,
355 but also in insulin resistance. Interesting work has recently been published on the use of mice
356 genetically deficient in Toll-like receptor 5 (TLR5). These mice spontaneously develop
357 symptoms of the metabolic syndrome, among which insulin resistance (Vijay-Kumar *et al.*,
358 2010). Transfer of the intestinal microbiota of these mice into germ-free wild-type mice
359 allowed transferring the metabolic phenotype into the wild-type mice, including insulin
360 resistance. As this shows that specific microbial community composition may be implicated
361 in insulin resistance, alterations of the community through dietary interventions may also
362 affect glucose and insulin metabolism. A third mode of action of dietary fibers may therefore
363 be related to the modulation of the intestinal microbiota.

364 Dietary fibers are typically non-digestible and therefore reach the colon, where they can be
365 metabolized into SCFA by the intestinal bacteria. There is evidence that hepatocytes may,
366 when exposed to an increase in short-chain fatty acids, increase glucose oxidation, decrease
367 fatty acid release, and increase insulin clearance - an environment conducive to enhanced
368 insulin sensitivity (Frayn *et al.*, 1996; Thorburn *et al.*, 1993; Venter *et al.*, 1990). This would
369 be related with specific interactions with G-protein-coupled receptors GPR41 and GPR43
370 (Delzenne and Cani, 2011; Dewulf *et al.*, 2010). Whereas acetate is typically considered to act
371 as substrate for lipogenesis in the liver, propionate would inhibit *de novo* lipogenesis and
372 gluconeogenesis from lactate, decrease inflammation and improve insulin sensitivity (Al-
373 Lahham *et al.*, 2010; Berggren *et al.*, 1996; Lin *et al.*, 1995). Finally, butyrate has also been
374 linked with improved insulin sensitivity (Gao *et al.*, 2009).

375 As described in the section on the role of the intestinal barrier and low-grade inflammation in
376 obesity and metabolic disorders, specific changes induced in microbial community
377 composition upon (prebiotic) fiber intake may also be involved in improvements of glucose
378 maintenance, through altered host-bacteria interactions, involving improvement of gut barrier
379 function and reduction of LPS leakage (Cani and Delzenne, 2009; Neyrinck *et al.*, 2010;

Neyrinck *et al.*, 2011). In addition, dietary fibers such as beta-glucans, may directly interact with the mucosal immune system and influence insulin sensitivity through immune-modulation (King *et al.*, 2007; Vos *et al.*, 2007).

Delzenne and Cani (2011) recently summarized the current evidence on the relation between specific microbial community composition and diabetes. Changes in community composition seem to involve reduced presence of Firmicutes as opposed to Bacteroidetes (Larsen *et al.*, 2010). Other researchers showed lower representation of the genus *Bifidobacterium* and *Bacteroides vulgatus* (Wu *et al.*, 2010) or the abundance of *Faecalibacterium prausnitzii* (Furet *et al.*, 2010) in microbiota from diabetic individuals and a lower presence of microbiota-related metabolites in the blood and urine of diabetic individuals (Lucio *et al.*, 2010; Zhao *et al.*, 2010).

The interaction between dietary fibers, intestinal microbiota and gut peptide hormones has been described extensively in relation to weight management and lipid metabolism (Cani *et al.*, 2005; Delzenne and Cani, 2011; Delzenne *et al.*, 2007). Interaction of these peptide hormones with glucose metabolism and insulin sensitivity was also shown in numerous animal studies (Delzenne and Cani, 2010). For instance, dietary fiber such as oligofructose can increase the number of endocrine L-cells in the proximal colon of rats (Cani *et al.*, 2007c) and increase the production of GLP-1 and GLP-2, the former being involved in the regulation of insulin sensitivity (Maurer and Reimer, 2011) and the latter in barrier function control (Cani *et al.*, 2009b). Another example is the potential role of adiponectin (Weickert and Pfeiffer, 2008). In a cross-sectional analysis, high intakes of cereal dietary fiber were positively associated with plasma adiponectin after adjusting for lifestyle factors and dietary glycemic load (Qi *et al.*, 2005). Adiponectin may act as a peripheral starvation signal promoting the storage of triglycerides preferentially in adipose tissue (Kim *et al.*, 2007). As a consequence, reduced triglyceride accumulation in the liver and in the skeletal muscle might convey improved systemic insulin sensitivity (Weickert and Pfeiffer, 2006).

Summarized, the positive effects of (prebiotic) dietary fibers on postprandial glucose and insulin response are becoming more and more clear. Recently, Gemen *et al.* (2011) provided a clear overview of the existing scientific literature, in which 39 publications were referred to. Although further research is needed to differentiate the variety of existing fiber sources in their efficacy and specific mode of action, the basic principles of the underlying mechanisms and the intriguing role of the gut microbiota become unravelled.

Future perspectives

The studies described in this review - although they have been conducted on animals - suggest that a successful prebiotic intervention with respect to obesity and its related metabolic disorders could be possible (Cani *et al.*, 2006; Cani *et al.*, 2007d; Cani *et al.*, 2009b; Neyrinck *et al.*, 2011). However, new metagenomic technologies have also pointed out that the interindividual variability of our holobiont is a key factor for the success of a given strategy. For instance, Walker *et al.* (2011) recently showed a strong variation in terms of microbial modulation among human subjects in response to prebiotic supplementation. As a consequence, the management of the intestinal microbiota with the aim of improving human health will optimally require a prior characterization of the microbiota, i.e. the concept of personalized health-care. In this respect, several options may be available. When the necessary genes/species/strains are present, one may target them through specific prebiotics. If not present, the so-called synbiotic approach could be a valuable solution: addition of specific bacteria with a metabolic capability of interest (e.g. probiotics but also microbes beyond the current definition such as butyrate producing species – *Eubacterium rectale*, *Faecalibacterium prausnitzii*, *Anaerostipes caccae*, *Roseburia intestinalis*). A final option

could be the transfer of entire communities (or part of the microbial population) through faecal transplantation (Khoruts *et al.*, 2010), as an extension of the concept of synbiotics. This implies that studies demonstrating the beneficial effect of prebiotics are useful but need a better characterization of the exact modulation of the intestinal microbiota (both luminal and mucosal microbiota) in order to mechanistically explain the beneficial host effect. As an alternative to the individual specific approach, it may be possible that individuals may be grouped and subsequently treated with specific prebiotics based on an enterotype-like classification as proposed by (Arumugam *et al.*, 2011).

References

- Al-Lahham, S.H., Peppelenbosch, M.P., Roelofsen, H., Vonk, R.J. and Venema, K., 2010. Biological effects of propionic acid in humans; metabolism, potential applications and underlying mechanisms. *Biochimica Et Biophysica Acta-Molecular and Cell Biology of Lipids* 1801: 1175-1183.
- Are, A., Aronsson, L., Wang, S., Greicius, G., Lee, Y.K., Gustafsson, J.-Å., Pettersson, S. and Arulampalam, V., 2008. *Enterococcus faecalis* from newborn babies regulate endogenous PPAR γ activity and IL-10 levels in colonic epithelial cells. *Proceedings of the National Academy of Sciences of the United States of America* 105: 1943-1948.
- Aronsson, L., Huang, Y., Parini, P., Korach-André, M., Hakansson, J., Gustafsson, J.A., Pettersson, S., Arulampalam, V. and Rafter, J.J., 2010. Decreased fat storage by *Lactobacillus paracasei* is associated with increased levels of angiopoietin-like 4 protein (ANGPTL4). *PLoS One* 5: e13078.
- Arumugam, M., Raes, J., Pelletier, E., Le Paslier, D., Yamada, T., Mende, D.R., Fernandes, G.R., Tap, J., Bruls, T., Batto, J.M., Bertalan, M., Borruel, N., Casellas, F., Fernandez, L., Gautier, L., Hansen, T., Hattori, M., Hayashi, T., Kleerebezem, M., Kurokawa, K., Leclerc, M., Levenez, F., Manichanh, C., Nielsen, H.B., Nielsen, T., Pons, N., Poulain, J., Qin, J., Sicheritz-Ponten, T., Tims, S., Torrents, D., Ugarte, E., Zoetendal, E.G., Wang, J., Guarner, F., Pedersen, O., de Vos, W.M., Brunak, S., Dore, J., Consortium, M., Weissenbach, J., Ehrlich, S.D., Bork, P., Antolin, M., Artiguenave, F., Blottiere, H.M., Almeida, M., Brechot, C., Cara, C., Chervaux, C., Cultrone, A., Delorme, C., Denariáz, G., Dervyn, R., Foerstner, K.U., Friss, C., van de Guchte, M., Guedon, E., Haimet, F., Huber, W., van Hylckama-Vlieg, J., Jamet, A., Juste, C., Kaci, G., Knol, J., Lakhdari, O., Layec, S., Le Roux, K., Maguin, E., Merieux, A., Melo Minardi, R., M'Rini, C., Muller, J., Oozeer, R., Parkhill, J., Renault, P., Rescigno, M., Sanchez, N., Sunagawa, S., Torrejon, A., Turner, K., Vandemeulebrouck, G., Varela, E., Winogradsky, Y. and Zeller, G., 2011. Enterotypes of the human gut microbiome. *Nature* 473: 174-180.
- Bäckhed, F., Ding, H., Wang, T., Hooper, L.V., Koh, G.Y., Nagy, A., Semenkovich, C.F. and Gordon, J.I., 2004. The gut microbiota as an environmental factor that regulates fat storage. *Proceedings of the National Academy of Sciences of the United States of America* 101: 15718-15723.
- Bäckhed, F., Manchester, J.K., Semenkovich, C.F. and Gordon, J.I., 2007. Mechanisms underlying the resistance to diet-induced obesity in germ-free mice. *Proceedings of the National Academy of Sciences of the United States of America* 104: 979-984.
- Barcelo, A., Claustre, J., Moro, F., Chayvialle, J.A., Cuber, J.C. and Plaisancie, P., 2000. Mucin secretion is modulated by luminal factors in the isolated vascularly perfused rat colon. *Gut* 46: 218-224.
- Beena, A. and Prasad, V., 1997. Effect of yogurt and bifidus yogurt fortified with skim milk powder, condensed whey and lactose-hydrolysed condensed whey on serum cholesterol and triacylglycerol levels in rats. *Journal of Dairy Research* 64: 453-457.
- Berggren, A., Nyman, E.M.G.L., Lundquist, I. and Björck, I.M.E., 1996. Influence of orally and rectally administered propionate on cholesterol and glucose metabolism in obese rats. *British Journal of Nutrition* 76: 287-294.
- Bjorksten, B., Naaber, P., Sepp, E. and Mikelsaar, M., 1999. The intestinal microflora in allergic Estonian and Swedish 2-year-old children. *Clinical and Experimental Allergy: Journal of the British Society for Allergy and Clinical Immunology* 29: 342-346.
- Blaser, M.J., 2006. Who are we? Indigenous microbes and the ecology of human diseases. *European Molecular Biology Organization Reports* 7: 956-960.

489 Bongaerts, G.P., Severijnen, R.S., Tangerman, A., Verrips, A. and Tolboom, J.J., 2000. Bile
 490 acid deconjugation by Lactobacilli and its effects in patients with a short small bowel. *Journal*
 491 *of Gastroenterology* 35: 801-804.
 492 Brouns, F., Kettlitz, B. and Arrigini, E., 2002. Resistant starch and "the butyrate revolution".
 493 *Trends in Food Science and Technology* 13: 251-261.
 494 Brown, R., Imran, S.A. and Wilkinson, M., 2009. Lipopolysaccharide (LPS) stimulates
 495 adipokine and socs3 gene expression in mouse brain and pituitary gland *in vivo*, and in N-1
 496 hypothalamic neurons *in vitro*. *Journal of Neuroimmunology* 209: 96-103.
 497 Burcelin, R., Luche, E., Serino, M. and Amar, J., 2009. The gut microbiota ecology: a new
 498 opportunity for the treatment of metabolic diseases? *Frontiers in Bioscience* 14: 5107-5117.
 499 Caesar, R., Fak, F. and Backhed, F., 2010. Effects of gut microbiota on obesity and
 500 atherosclerosis via modulation of inflammation and lipid metabolism. *Journal of Internal*
 501 *Medicine* 268: 320-328.
 502 Cani, P.D., Amar, J., Iglesias, M.A., Poggi, M., Knauf, C., Bastelica, D., Neyrinck, A.M.,
 503 Fava, F., Tuohy, K.M., Chabo, C., Waget, A., Delmee, E., Cousin, B., Sulpice, T.,
 504 Chamontin, B., Ferrieres, J., Tanti, J.F., Gibson, G.R., Casteilla, L., Delzenne, N.M., Alessi,
 505 M.C. and Burcelin, R., 2007a. Metabolic endotoxemia initiates obesity and insulin resistance.
 506 *Diabetes* 56: 1761-1772.
 507 Cani, P.D., Bibiloni, R., Knauf, C., Neyrinck, A.M., Delzenne, N.M. and Burcelin, R., 2008.
 508 Changes in gut microbiota control metabolic endotoxemia-induced inflammation in high-fat
 509 diet-induced obesity and diabetes in mice. *Diabetes* 57: 1470-1481.
 510 Cani, P.D., Daubioul, C.A., Reusens, B., Remacle, C., Catillon, G. and Delzenne, N.M., 2005.
 511 Involvement of endogenous glucagon-like peptide-1(7-36) amide on glycaemia-lowering
 512 effect of oligofructose in streptozotocin-treated rats. *Journal of Endocrinology* 185: 457-465.
 513 Cani, P.D. and Delzenne, N.M., 2007. Gut microflora as a target for energy and metabolic
 514 homeostasis. *Current Opinion in Clinical Nutrition and Medical Care* 10: 729-734.
 515 Cani, P.D. and Delzenne, N.M., 2009. Interplay between obesity and associated metabolic
 516 disorders: new insights into the gut microbiota. *Current Opinion in Pharmacology* 9: 737-743.
 517 Cani, P.D., Dewever, C. and Delzenne, N.M., 2004. Inulin-type fructans modulate
 518 gastrointestinal peptides involved in appetite regulation (glucagon-like peptide-1 and ghrelin)
 519 in rats. *British Journal of Nutrition* 92: 521-526.
 520 Cani, P.D., Holst, J.J., Drucker, D.J., Delzenne, N.M., Thorens, B., Burcelin, R. and Knauf,
 521 C., 2007b. GLUT2 and the incretin receptors are involved in glucose-induced incretin
 522 secretion. *Molecular and Cellular Endocrinology* 276: 18-23.
 523 Cani, P.D., Hoste, S., Guiot, Y. and Delzenne, N.M., 2007c. Dietary non-digestible
 524 carbohydrates promote L-cell differentiation in the proximal colon of rats. *British Journal of*
 525 *Nutrition* 98: 32-37.
 526 Cani, P.D., Knauf, C., Iglesias, M.A., Drucker, D.J., Delzenne, N.M. and Burcelin, R., 2006.
 527 Improvement of glucose tolerance and hepatic insulin sensitivity by oligofructose requires a
 528 functional glucagon-like peptide 1 receptor. *Diabetes* 55: 1484-1490.
 529 Cani, P.D., Lecourt, E., Dewulf, E.M., Sohet, F.M., Pachikian, B.D., Naslain, D., De Backer,
 530 F., Neyrinck, A.M. and Delzenne, N.M., 2009a. Gut microbiota fermentation of prebiotics
 531 increases satietogenic and incretin gut peptide production with consequences for appetite
 532 sensation and glucose response after a meal. *American Journal of Clinical Nutrition* 90: 1236-
 533 1243.
 534 Cani, P.D., Neyrinck, A.M., Fava, F., Knauf, C., Burcelin, R.G., Tuohy, K.M., Gibson, G.R.
 535 and Delzenne, N.M., 2007d. Selective increases of bifidobacteria in gut microflora improve
 536 high-fat-diet-induced diabetes in mice through a mechanism associated with endotoxaemia.
 537 *Diabetologia* 50: 2374-2383.

538 Cani, P.D., Possemiers, S., Van de Wiele, T., Guiot, Y., Everard, A., Rottier, O., Geurts, L.,
 539 Naslain, D., Neyrinck, A., Lambert, D.M., Muccioli, G.G. and Delzenne, N.M., 2009b.
 540 Changes in gut microbiota control inflammation in obese mice through a mechanism
 541 involving GLP-2-driven improvement of gut permeability. *Gut* 58: 1091-1103.
 542 Cario, E. and Podolsky, D.K., 2000. Differential alteration in intestinal epithelial cell
 543 expression of toll-like receptor 3 (TLR3) and TLR4 in inflammatory bowel disease. *Infection*
 544 and *Immunity* 68: 7010-7017.
 545 Chen, H., Liu, L.-J., Zhu, J.-J., Xu, B. and Li, R., 2010. Effect of soybean oligosaccharides on
 546 blood lipid, glucose levels and antioxidant enzymes activity in high fat rats. *Food Chemistry*
 547 119: 1633-1636.
 548 Cheng, H.-H. and Lai, M.-H., 2000. Fermentation of resistant rice starch produces propionate
 549 reducing serum and hepatic cholesterol in rats. *Journal of Nutrition* 130: 1991-1995.
 550 Collado, M.C., Isolauri, E., Laitinen, K. and Salminen, S., 2008. Distinct composition of gut
 551 microbiota during pregnancy in overweight and normal-weight women. *American Journal of*
 552 *Clinical Nutrition* 88: 894-899.
 553 de Luis, D.A., de la Fuente, B., Izaola, O., Conde, R., Gutierrez, S., Morillo, M. and Teba
 554 Torres, C., 2010. Randomized clinical trial with a inulin enriched cookie on risk
 555 cardiovascular factor in obese patients. *Nutrición Hospitalaria* 25: 53-59.
 556 Delzenne, N.M. and Cani, P.D., 2010. Nutritional modulation of gut microbiota in the context
 557 of obesity and insulin resistance: potential interest of prebiotics. *International Dairy Journal*
 558 20: 277-280.
 559 Delzenne, N.M. and Cani, P.D., 2011. Gut microbiota and the pathogenesis of insulin
 560 resistance. *Current Diabetes Reports* 11: 154-159.
 561 Delzenne, N.M., Cani, P.D. and Neyrinck, A.M., 2007. Modulation of glucagon-like peptide 1
 562 and energy metabolism by inulin and oligofructose: experimental data. *Journal of Nutrition*
 563 137: S2547-S2551.
 564 Delzenne, N.M., Kok, N., Fiordaliso, M., Deboyser, D., Goethals, F. and Roberfroid, M.B.,
 565 1993. Dietary fructo-oligosaccharides modifies lipid metabolism in rats. *American Journal of*
 566 *Clinical Nutrition* 57: 8205.
 567 Dewulf, E., Neyrinck, A.M., Possemiers, S., Deldicque, L., Bindels, L.B., Pachikian, B.D.,
 568 Sohet, F.M., Francaux, M., Cani, P.D. and Delzenne, N.M., 2010. Involvement of receptors
 569 for short chain carboxylic acids and GPR41 GPR43-in the control of adipogenesis by
 570 prebiotics, modulators of selective intestinal microbiota. *Diabetes & Metabolism* 36: A35-
 571 A35.
 572 Dutton, S. and Trayhurn, P., 2008. Regulation of angiopoietin-like protein 4/fasting-induced
 573 adipose factor (ANGPTL4/FIAF) expression in mouse white adipose tissue and 3T3-L1
 574 adipocytes. *British Journal of Nutrition* 100: 18-26.
 575 Egan, J.M. and Margolskee, R.F., 2008. Taste cells of the gut and gastrointestinal
 576 chemosensation. *Molecular Interventions* 8: 78-81.
 577 Egert, M., de Graaf, A.A., Smidt, H., de Vos, W.M. and Venema, K., 2006. Beyond diversity:
 578 functional microbiomics of the human colon. *Trends in Microbiology* 14: 86-91.
 579 Fassler, C., Arrigoni, E., Venema, K., Brouns, F. and Amado, R., 2006. *In vitro* fermentability
 580 of differently digested resistant starch preparations. *Molecular Nutrition and Food Research*
 581 50: 1220-1228.
 582 Fiordaliso, M.F., Kok, N., Desager, J.P., Goethals, F., Deboyser, D., Roberfroid, M.B. and
 583 Delzenne, N.M., 1995. Dietary oligofructose lowers triacylglycerols, phospholipids and
 584 cholesterol in serum and very low density lipoproteins of rats. *Lipids* 30: 163-167.
 585 Fleissner, C.K., Huebel, N., Abd El-Bary, M.M., Loh, G., Klaus, S. and Blaut, M., 2010.
 586 Absence of intestinal microbiota does not protect mice from diet-induced obesity. *British*
 587 *Journal of Nutrition* 104: 919-929.

Frayn, K.N., Williams, C.M. and Arner, P., 1996. Are increased plasma nonesterified fatty acid concentrations a risk marker for coronary heart disease and other chronic diseases? *Clinical Science* 90: 243-253.

Furet, J.P., Kong, L.C., Tap, J., Poitou, C., Basdevant, A., Bouilliot, J.L., Mariat, D., Corthier, G., Dore, J., Henegar, C., Rizkalla, S. and Clement, K., 2010. Differential adaptation of human gut microbiota to bariatric surgery-induced weight loss links with metabolic and low-grade inflammation markers. *Diabetes* 59: 3049-3057.

Gao, Z.G., Yin, J., Zhang, J., Ward, R.E., Martin, R.J., Lefevre, M., Cefalu, W.T. and Ye, J.P., 2009. Butyrate improves insulin sensitivity and increases energy expenditure in mice. *Diabetes* 58: 1509-1517.

Garrett, W.S., Lord, G.M., Punit, S., Lugo-Villarino, G., Mazmanian, S.K., Ito, S., Glickman, J.N. and Glimcher, L.H., 2007. Communicable ulcerative colitis induced by T-bet deficiency in the innate immune system. *Cell* 131: 33-45.

Gee, J.M. and Johnson, I.T., 2005. Dietary lactitol fermentation increases circulating peptide YY and glucagon-like peptide-1 in rats and humans. *Nutrition* 21: 1036-1043.

Gemen, R., de Vries, J.F. and Slavin, J.L., 2011. Relationship between molecular structure of cereal dietary fiber and health effects: focus on glucose/insulin response and gut health. *Nutrition Reviews* 69: 22-33.

Gilliland, S.E., Nelson, C.R. and Maxwell, C., 1985. Assimilation of cholesterol by *Lactobacillus acidophilus*. *Applied Environmental Microbiology* 49: 377-381.

Girardin, S.E., Boneca, I.G., Carneiro, L.A., Antignac, A., Jehanno, M., Viala, J., Tedin, K., Taha, M.K., Labigne, A., Zahringer, U., Coyle, A.J., DiStefano, P.S., Bertin, J., Sansonetti, P.J. and Philpott, D.J., 2003. Nod1 detects a unique muropeptide from gram-negative bacterial peptidoglycan. *Science* 300: 1584-1587.

Grootaert, C., Van de Wiele, T., Van Roosbroeck, I., Possemiers, S., Vercoutter-Edouart, A.-S., Verstraete, W., Bracke, M. and Vanhoecke, B., 2011. Bacterial monocultures, propionate, butyrate and H₂O₂ modulate the expression, secretion and structure of the fasting induced adipose factor in gut epithelial cell lines. *Environmental Microbiology* 13: 1778-1789.

Gupta, G. and Surolia, A., 2007. Collectins: sentinels of innate immunity. *Bioessays* 29: 452-464.

Hooper, L.V., Midtved, T. and Gordon, J.I., 2002. How host-microbial interactions shape the nutrient environment of the mammalian intestine. *Annual Review of Nutrition* 22: 283-307.

Hooper, L.V., Wong, M.H., Thelin, A., Hansson, L., Falk, P.G. and Gordon, J.I., 2001. Molecular analysis of commensal host-microbial relationships in the intestine. *Science* 291: 881-884.

Hsu, C.K., Liao, J.W., Chung, Y.C., Hsieh, C.P. and Chan, Y.C., 2004. Xylooligosaccharides and fructooligosaccharides affect the intestinal microbiota and precancerous colonic lesion development in rats. *Journal of Nutrition* 134: 1523-1528.

Huang, Q.-H., Hruby, V.J. and Tatro, J.B., 1999. Role of central melanocortins in endotoxin-induced anorexia. *American Journal of Physiology - Regulatory, Integrative and Comparative Physiology* 276: 864-871.

Inui, A., Asakawa, A., Bowers, C.Y., Mantovani, G., Laviano, A., Meguid, M.M. and Fujimiya, M., 2004. Ghrelin, appetite, and gastric motility: the emerging role of the stomach as an endocrine organ. *Faseb Journal* 18: 439-456.

Johansson, M.E., Gustafsson, J.K., Sjöberg, K.E., Petersson, J., Holm, L., Sjövall, H. and Hansson, G.C., 2010. Bacteria penetrate the inner mucus layer before inflammation in the dextran sulfate colitis model. *PLoS One* 5: e12238.

Khailova, L., Dvorak, K., Arganbright, K.M., Halpern, M.D., Kinouchi, T., Yajima, M. and Dvorak, B., 2009. *Bifidobacterium bifidum* improves intestinal integrity in a rat model of

637 necrotizing enterocolitis. American Journal of Physiology - Gastrointestinal and Liver
638 Physiology 297: G940-G949.

639 Khoruts, A., Dicksved, J., Jansson, J.K. and Sadowsky, M.J., 2010. Changes in the
640 composition of the human fecal microbiome after bacteriotherapy for recurrent *Clostridium*
641 *difficile*-associated diarrhea. Journal of Clinical Gastroenterology 44: 354-360.

642 Kim, H.K., Youn, B.S., Shin, M.S., Namkoong, C., Park, K.H., Baik, J.H., Kim, J.B., Park,
643 J.Y., Lee, K.U., Kim, Y.B. and Kim, M.S., 2010. Hypothalamic ANGPTL4/Fiaf is a novel
644 regulator of food intake and body weight. Diabetes 59: 2772-2780.

645 Kim, J.Y., De Wall, E.V., Laplante, M., Azzara, A., Trujillo, M.E., Hofmann, S.M., Schraw,
646 T., Durand, J.L., Li, H., Li, G., Jelicks, L.A., Mehler, M.F., Hui, D.Y., Deshaies, Y.,
647 Shulman, G.I., Schwartz, G.J. and Scherer, P.E., 2007. Obesity-associated improvements in
648 metabolic profile through expansion of adipose tissue. Journal of Clinical Investigation 117:
649 2621-2637.

650 King, D.E., Egan, B.M., Woolson, R.F., Mainous, A.G., Al-Solaiman, Y. and Jesri, A., 2007.
651 Effect of a high-fiber diet vs a fiber-supplemented diet on C-reactive protein level. Archives
652 of Internal Medicine 167: 502-506.

653 Lahham, S.H., Weening, D., Rezaee, F., Roelofsen, H. and Venema, K., 2008. Propionic acid
654 stimulates leptin production in human adipose tissue through G-protein coupled receptor(s).
655 10th annual Gut Day: University Medical Centre Utrecht and Gut Flora Foundation, The
656 Netherlands.

657 Langlands, S.J., Hopkins, M.J., Coleman, N. and Cummings, J.H., 2004. Prebiotic
658 carbohydrates modify the mucosa associated microflora of the human large bowel. Gut 53:
659 1610-1616.

660 Larsen, N., Vogensen, F.K., van den Berg, F.W.J., Nielsen, D.S., Andreasen, A.S., Pedersen,
661 B.K., Abu Al-Soud, W., Sorensen, S.J., Hansen, L.H. and Jakobsen, M., 2010. Gut microbiota
662 in human adults with type 2 diabetes differs from non-diabetic adults. PLoS One 5.

663 Ley, R.E., Bäckhed, F., Turnbaugh, P., Lozupone, C.A., Knight, R.D. and Gordon, J.I., 2005.
664 Obesity alters gut microbial ecology. Proceedings of the National Academy of Sciences of the
665 United States of America 102: 11070-11075.

666 Li, X.J., Piao, X.S., Kim, S.W., Liu, P., Wang, L., Shen, Y.B., Jung, S.C. and Lee, H.S., 2007.
667 Effects of chito-oligosaccharide supplementation on performance, nutrient digestibility, and
668 serum composition in broiler chickens. Poultry Science 86: 1107-1114.

669 Lin, Y., Vonk, R.J., Slooff, M.J., Kuipers, F. and Smit, M.J., 1995. Differences in propionate-
670 induced inhibition of cholesterol and triacylglycerol synthesis between human and rat
671 hepatocytes in primary culture. British Journal of Nutrition 74: 197-207.

672 Lucio, M., Fekete, A., Weigert, C., Wagele, B., Zhao, X.J., Chen, J., Fritsche, A., Haring,
673 H.U., Schleicher, E.D., Xu, G.W., Schmitt-Kopplin, P. and Lehmann, R., 2010. Insulin
674 sensitivity is reflected by characteristic metabolic fingerprints - A Fourier transform mass
675 spectrometric non-targeted metabolomics approach. PLoS One 5.

676 Lundell, A.C., Adlerberth, I., Lindberg, E., Karlsson, H., Ekberg, S., Aberg, N., Saalman, R.,
677 Hock, B., Steinkasserer, A., Hesselmar, B., Wold, A.E. and Rudin, A., 2007. Increased levels
678 of circulating soluble CD14 but not CD83 in infants are associated with early intestinal
679 colonization with *Staphylococcus aureus*. Clinical and Experimental Allergy 37: 62-71.

680 Macpherson, A.J. and Uhr, T., 2004. Induction of protective IgA by intestinal dendritic cells
681 carrying commensal bacteria. Science 303: 1662-1665.

682 Mandard, S., Zandbergen, F., van Straten, E., Wahli, W., Kuipers, F., Muller, M. and Kersten,
683 S., 2006. The fasting-induced adipose factor/angiopoietin-like protein 4 is physically
684 associated with lipoproteins and governs plasma lipid levels and adiposity. Journal of
685 Biological Chemistry 281: 934-944.

686 Massimino, S.P., McBurney, M.I., Field, C.J., Thomson, A.B.R., Keelan, M., Hayek, M.G.
687 and Sunvold, G.D., 1998. Fermentable dietary fiber increases GLP-1 secretion and improves
688 glucose homeostasis despite increased intestinal glucose transport capacity in healthy dogs.
689 Journal of Nutrition 128: 1786-1793.

690 Maurer, A.D., Chen, Q., McPherson, C. and Reimer, R.A., 2009. Changes in satiety hormones
691 and expression of genes involved in glucose and lipid metabolism in rats weaned onto diets
692 high in fibre or protein reflect susceptibility to increased fat mass in adulthood. Journal of
693 Physiology 587: 679-691.

694 Maurer, A.D. and Reimer, R.A., 2011. Maternal consumption of high-prebiotic fibre or -
695 protein diets during pregnancy and lactation differentially influences satiety hormones and
696 expression of genes involved in glucose and lipid metabolism in offspring in rats. British
697 Journal of Nutrition 105: 329-338.

698 Medzhitov, R. and Janeway, C.A., Jr., 1997. Innate immunity: the virtues of a nonclonal
699 system of recognition. Cell 91: 295-298.

700 Medzhitov, R. and Janeway, C.A., Jr., 2002. Decoding the patterns of self and nonself by the
701 innate immune system. Science 296: 298-300.

702 Mohan, B., Kadirvel, R., Natarajan, A. and Bhaskaran, M., 1996. Effect of probiotic
703 supplementation on growth, nitrogen utilization and serum cholesterol in broilers. British
704 Poultry Science 37: 395-401.

705 Mohlig, M., Koebnick, C., Weickert, M.O., Lueder, W., Otto, B., Steiniger, J., Twilfert, M.,
706 Meuser, F., Pfeiffer, A.F. and Zunft, H.J., 2005. Arabinoxylan-enriched meal increases serum
707 ghrelin levels in healthy humans. Hormone and Metabolic Research 37: 303-308.

708 Montalto, M., Maggiano, N., Ricci, R., Curigliano, V., Santoro, L., Di Nicuolo, F., Vecchio,
709 F.M., Gasbarrini, A. and Gasbarrini, G., 2004. *Lactobacillus acidophilus* protects tight
710 junctions from aspirin damage in HT-29 cells. Digestion 69: 225-228.

711 Musso, G., Gambino, R. and Cassader, M., 2011. Interactions between gut microbiota and
712 host metabolism predisposing to obesity and diabetes. Annual Review of Medicine 62: 361-
713 380.

714 Nakajima, H., Suzuki, Y., Kaizu, H. and Hirota, T., 1992. Cholesterol lowering activity of
715 rosy fermented milk. Journal of Food Science 57: 1327-1329.

716 Neyrinck, A., Possemiers, S., Verstraete, W., de Backer, F., Cani, P. and Delzenne, N., 2010.
717 Interest of chitin-glucan of fungal origin in the control of metabolic alterations induced by fat
718 diet in mice: involvement of intestinal microbiota. Diabetes and Metabolism 36: A105-A105.

719 Neyrinck, A.M., Bindels, L.B., De Backer, F., Pachikian, B.D., Cani, P.D. and Delzenne,
720 N.M., 2009. Dietary supplementation with chitosan derived from mushrooms changes
721 adipocytokine profile in diet-induced obese mice, a phenomenon linked to its lipid-lowering
722 action. International Immunopharmacology 9: 767-773.

723 Neyrinck, A.M., Possemiers, S., Druart, C., de Wiele, T.V., De Backer, F., Cani, P.D.,
724 Larondelle, Y. and Delzenne, N.M., 2011. Prebiotic effects of wheat arabinoxylan related to
725 the increase in Bifidobacteria, Roseburia and Bacteroides/Prevotella in diet-induced obese
726 mice. PLoS One 6.

727 Ooi, L.G. and Liong, M.T., 2010. Cholesterol-lowering effects of probiotics and prebiotics: a
728 review of *in vivo* and *in vitro* findings. International Journal of Molecular Sciences 11: 2499-
729 2522.

730 Parnell, J.A. and Reimer, R.A., 2009. Weight loss during oligofructose supplementation is
731 associated with decreased ghrelin and increased peptide YY in overweight and obese adults.
732 American Journal of Clinical Nutrition 89: 1751-1759.

733 Piche, T., des Varannes, S.B., Sacher-Huvelin, S., Holst, J.J., Cuber, J.C. and Galmiche, J.P.,
734 2003. Colonic fermentation influences lower esophageal sphincter function in
735 gastroesophageal reflux disease. Gastroenterology 124: 894-902.

Possemiers, S., Grootaert, C., Vermeiren, J., Gross, G., Marzorati, M., Verstraete, W. and Van de Wiele, T., 2009. The intestinal environment in health and disease - recent insights on the potential of intestinal bacteria to influence human health. *Current Pharmaceutical Design* 15: 2051-2065.

Pulusani, S.R. and Rao, D.R., 1983. Whole body, liver and plasma cholesterol levels in rats fed *Thermophilus bulgaricus* and *acidophilus* milks. *Journal of Food Science* 48: 280-281.

Qi, L., Rimm, E., Liu, S.M., Rifai, N. and Hu, F.B., 2005. Dietary glycemic index, glycemic load, cereal fiber, and plasma adiponectin concentration in diabetic men. *Diabetes Care* 28: 1022-1028.

Regand, A., Tosh, S.M., Wolever, T.M.S. and Wood, P.J., 2009. Physicochemical properties of beta-glucan in differently processed oat foods influence glycemic response. *Journal of Agricultural and Food Chemistry* 57: 8831-8838.

Resta-Lenert, S., Das, S., Batra, S.K. and Ho, S.B., 2011. Muc17 protects intestinal epithelial cells from enteroinvasive *E. coli* infection by promoting epithelial barrier integrity. *American Journal of Physiology - Gastrointestinal and Liver Physiology* 300: G1144-G1155.

Richelsen, B., Kristensen, K. and Pedersen, S.B., 1996. Long-term (6 months) effect of a new fermented milk product on the level of plasma lipoproteins - a placebo-controlled and double blind study. *European Journal of Clinical Nutrition* 50: 811-815.

Rijkers, G.T., Bengmark, S., Enck, P., Haller, D., Herz, U., Kalliomaki, M., Kudo, S., Lenoir-Wijnkoop, I., Mercenier, A., Myllyluoma, E., Rabot, S., Rafter, J., Szajewska, H., Watzl, B., Wells, J., Wolvers, D. and Antoine, J.M., 2010. Guidance for substantiating the evidence for beneficial effects of probiotics: current status and recommendations for future research. *Journal of Nutrition* 140: S671-S676.

Roberfroid, M., Gibson, G.R., Hoyles, L., McCartney, A.L., Rastall, R., Rowland, I., Wolvers, D., Watzl, B., Szajewska, H., Stahl, B., Guarner, F., Respondek, F., Whelan, K., Coxam, V., Davicco, M.J., Leotoing, L., Wittrant, Y., Delzenne, N.M., Cani, P.D., Neyrinck, A.M. and Meheust, A., 2010. Prebiotic effects: metabolic and health benefits. *British Journal of Nutrition* 104: 1-63.

Rosenberg, E. and Zilber-Rosenberg, I., 2011. Symbiosis and development: the hologenome concept. *Birth Defects Research Part C: Embryo Today* 93: 56-66.

Rummel, C., Inoue, W., Sachot, C., Poole, S., Hubschle, T. and Luheshi, G.N., 2008. Selective contribution of interleukin-6 and leptin to brain inflammatory signals induced by systemic LPS injection in mice. *Journal of Comparative Neurology* 511: 373-395.

Sanz, Y., Santacruz, A. and Gauffin, P., 2010. Gut microbiota in obesity and metabolic disorders. *Proceedings of the Nutrition Society* 69: 434-441.

Schmidt-Wittig, U., Enss, M.L., Coenen, M., Gartner, K. and Hedrich, H.J., 1996. Response of rat colonic mucosa to a high fiber diet. *Annals of Nutrition and Metabolism* 40: 343-350.

Schreiber, O., 2010. Microcirculation, mucus and microbiota in inflammatory bowel disease. *Acta Universitatis Upsaliensis, Uppsala*, 58 pp.

Seth, A., Yan, F., Polk, D.B. and Rao, R.K., 2008. Probiotics ameliorate the hydrogen peroxide-induced epithelial barrier disruption by a PKC- and MAP kinase-dependent mechanism. *American Journal of Physiology - Gastrointestinal and Liver Physiology* 294: G1060-G1069.

Shimotoyodome, A., Meguro, S., Hase, T., Tokimitsu, I. and Sakata, T., 2000. Short chain fatty acids but not lactate or succinate stimulate mucus release in the rat colon. *Comparative Biochemistry and Physiology Part A: Molecular and Integrative Physiology* 125: 525-531.

Song, Y.J., Sawamura, M., Ikeda, K., Igawa, S. and Yamori, Y., 2000. Soluble dietary fibre improves insulin sensitivity by increasing muscle GLUT-4 content in stroke-prone spontaneously hypertensive rats. *Clinical and Experimental Pharmacology and Physiology* 27: 41-45.

786 St-Onge, M.-P., Farnworth, E.R. and Jones, P.J.H., 2000. Consumption of fermented and
787 nonfermented dairy products: effects on cholesterol concentrations and metabolism. *American*
788 *Journal of Clinical Nutrition* 71: 674-681.

789 Storey, D., Lee, A., Bornet, F. and Brouns, F., 2007. Gastrointestinal responses following
790 acute and medium term intake of retrograded resistant maltodextrins, classified as type 3
791 resistant starch. *European Journal of Clinical Nutrition* 61: 1262-1270.

792 Takeda, K., Kaisho, T. and Akira, S., 2003. Toll-like receptors. *Annual review of*
793 *immunology* 21: 335-376.

794 Taubes, G., 2009. Prosperity's Plague. *Science* 325: 256-260.

795 Thorburn, A., Muir, J. and Proietto, J., 1993. Carbohydrate fermentation decreases hepatic
796 glucose output in healthy subjects. *Metabolism-Clinical and Experimental* 42: 780-785.

797 Tien, E.S., Davis, J.W. and Vanden Heuvel, J.P., 2004. Identification of the CREB-binding
798 protein/p300-interacting protein CITED2 as a peroxisome proliferator-activated receptor a
799 coregulator. *Journal of Biological Chemistry* 279: 24053-24063.

800 Tilg, H., 2010. Obesity, metabolic syndrome, and microbiota multiple interactions. *Journal of*
801 *Clinical Gastroenterology* 44: S16-S18.

802 Ting, J.P., Lovering, R.C., Alnemri, E.S., Bertin, J., Boss, J.M., Davis, B.K., Flavell, R.A.,
803 Girardin, S.E., Godzik, A., Harton, J.A., Hoffman, H.M., Hugot, J.P., Inohara, N., Mackenzie,
804 A., Maltais, L.J., Nunez, G., Ogura, Y., Otten, L.A., Philpott, D., Reed, J.C., Reith, W.,
805 Schreiber, S., Steimle, V. and Ward, P.A., 2008. The NLR gene family: a standard
806 nomenclature. *Immunity* 28: 285-287.

807 Todesco, T., Rao, A.V., Bosello, O. and Jenkins, D.J.A., 1991. Propionate lowers blood
808 glucose and alters lipid-metabolism in healthy subjects. *American Journal of Clinical*
809 *Nutrition* 54: 860-865.

810 Towle, H.C., 2001. Glucose and cAMP: Adversaries in the regulation of hepatic gene
811 expression. *Proceedings of the National Academy of Sciences of the United States of America*
812 98: 13476-13478.

813 Trautwein, E.A., Rieckhoff, D. and Erbersdobler, H.F., 1998. Dietary inulin lowers plasma
814 cholesterol and triacylglycerol and alters biliary bile acid profile in hamsters. *Journal of*
815 *Nutrition* 128: 1937-1943.

816 Turnbaugh, P.J., Ley, R.E., Mahowald, M.A., Magrini, V., Mardis, E.R. and Gordon, J.I.,
817 2006. An obesity-associated gut microbiome with increased capacity for energy harvest.
818 *Nature* 444: 1027-1131.

819 Van den Abbeele, P., Van de Wiele, T., Verstraete, W. and Possemiers, S., 2011. The host
820 selects mucosal and luminal associations of coevolved gut microorganisms: a novel concept.
821 *FEMS Microbiology Reviews* 35: 681-704.

822 Venter, C.S., Vorster, H.H. and Cummings, J.H., 1990. Effects of dietary propionate on
823 carbohydrate and lipid metabolism in healthy volunteers. *American Journal of*
824 *Gastroenterology* 85: 549-553.

825 Vijay-Kumar, M., Aitken, J.D., Carvalho, F.A., Cullender, T.C., Mwangi, S., Srinivasan, S.,
826 Sitaraman, S.V., Knight, R., Ley, R.E. and Gewirtz, A.T., 2010. Metabolic syndrome and
827 altered gut microbiota in mice lacking Toll-like receptor 5. *Science* 328: 228-231.

828 Vos, A.P., M'Rabet, L., Stahl, B., Boehm, G. and Garssen, J., 2007. Immune-modulatory
829 effects and potential working mechanisms of orally applied nondigestible carbohydrates.
830 *Critical Reviews in Immunology* 27: 97-140.

831 Walker, A.W., Ince, J., Duncan, S.H., Webster, L.M., Holtrop, G., Ze, X., Brown, D., Stares,
832 M.D., Scott, P., Bergerat, A., Louis, P., McIntosh, F., Johnstone, A.M., Lobley, G.E.,
833 Parkhill, J. and Flint, H.J., 2011. Dominant and diet-responsive groups of bacteria within the
834 human colonic microbiota. *ISME Journal* 5: 220-230.

Weickert, M.O. and Pfeiffer, A.F., 2008. Metabolic effects of dietary fiber consumption and prevention of diabetes. *Journal of Nutrition* 138: 439-442.

Weickert, M.O. and Pfeiffer, A.F.H., 2006. Signalling mechanisms linking hepatic glucose and lipid metabolism. *Diabetologia* 49: 1732-1741.

Wellen, K.E. and Hotamisligil, G.S., 2005. Inflammation, stress, and diabetes. *Journal of Clinical Investigation* 115: 1111-1119.

Williams, C.M., 1997. Postprandial lipid metabolism: effects of dietary fatty acids. *Proceedings of the Nutrition Society* 56: 679-692.

Williams, C.M. and Jackson, K.G., 2002. Inulin and oligofructose: effects on lipid metabolism from human studies. *British Journal of Nutrition* 87: 261-264.

Wood, P.J., Beer, M.U. and Butler, G., 2000. Evaluation of role of concentration and molecular weight of oat beta-glucan in determining effect of viscosity on plasma glucose and insulin following an oral glucose load. *British Journal of Nutrition* 84: 19-23.

Wu, X.K., Ma, C.F., Han, L., Nawaz, M., Gao, F., Zhang, X.Y., Yu, P.B., Zhao, C.A., Li, L.C., Zhou, A.P., Wang, J.A., Moore, J.E., Millar, B.C. and Xu, J.R., 2010. Molecular characterisation of the faecal microbiota in patients with type II diabetes. *Current Microbiology* 61: 69-78.

Xiao, J.Z., Kondo, S., Takahashi, N., Miyaji, K., Oshida, K., Hiramatsu, A., Iwatsuki, K., Kokubo, S. and Hosono, A., 2003. Effects of milk products fermented by *Bifidobacterium longum* on blood lipids in rats and healthy adult male volunteers. *Journal of Dairy Science* 86: 2452-2461.

Xiong, Y., Miyamoto, N., Shibata, K., Valasek, M.A., Motoike, T., Kedzierski, R.M. and Yanagisawa, M., 2004. Short-chain fatty acids stimulate leptin production in adipocytes through the G protein-coupled receptor GPR41. *Proceedings of the National Academy of Sciences of the United States of America* 101: 1045-1050.

Yoshida, K., Shimizugawa, T., Ono, M. and Furukawa, H., 2002. Angiotensin-like protein 4 is a potent hyperlipidemia-inducing factor in mice and inhibitor of lipoprotein lipase. *Journal of Lipid Research* 43: 1770-1772.

Zaneveld, J., Turnbaugh, P.J., Lozupone, C., Ley, R.E., Hamady, M., Gordon, J.I. and Knight, R., 2008. Host-bacterial coevolution and the search for new drug targets. *Current Opinion in Chemical Biology* 12: 109-114.

Zhao, X.J., Fritsche, J., Wang, J.S., Chen, J., Rittig, K., Schmitt-Kopplin, P., Fritsche, A., Haring, H.U., Schleicher, E.D., Xu, G.W. and Lehmann, R., 2010. Metabonomic fingerprints of fasting plasma and spot urine reveal human pre-diabetic metabolic traits. *Metabolomics* 6: 362-374.

Zhou, J., Martin, R.J., Tulley, R.T., Raggio, A.M., McCutcheon, K.L., Shen, L., Danna, S.C., Tripathy, S., Hegsted, M. and Keenan, M.J., 2008. Dietary resistant starch upregulates total GLP-1 and PYY in a sustained day-long manner through fermentation in rodents. *American Journal of Physiology - Endocrinology and Metabolism* 295: 1160-1166.

Figure legends

Figure 1. Intestinal section with focus on the effect of a fat diet and a prebiotic treatment on LPS detection, through TLR-4 and NOD-1. An increased permeability which is caused by the high-fat diet leads to LPS leakage through the gut wall, PRRs' detection, thus resulting in an inflammatory response.

Figure 2. Schematic overview of the impact of dietary fiber on several parameters involved in energy metabolism. **The mechanism of increased viscosity, decreased absorption of nutrients and replacement of available carbohydrates is relevant for the whole gastrointestinal tract.**

Table 1. Overview of *in vivo* and *in vitro* experiments in which the effect of prebiotics on several parameters involved in fat and sugar metabolism is confirmed. n.r. = not reported.

Signal molecule	Produced by	Function	Bacteria/bacteria-related mode of action	Prebiotics	Experimental design	References
GLP-1	L-cells in ileum/colon	Promotes insulin secretion, pancreatic e-cell proliferation and muscle oxidation	Acetate, propionate and butyrate	Oligofructose, lactitol, resistant starch	Rat and human studies, STC cell line	(Gee and Johnson, 2005; Cani <i>et al.</i> , 2007b; Cani <i>et al.</i> , 2009a; Piche <i>et al.</i> , 2003; Zhou <i>et al.</i> , 2008)
GLP-2	L-cells in ileum/colon	Intestinal barrier	n.r.	Oligofructose	Mice studies	(Cani <i>et al.</i> , 2009b)
PYY	L-cells in ileum/colon	Slows down gastric emptying	Acetate, propionate and butyrate	Oligofructose, lactitol, resistant starch	Rat and human studies, STC cell line	(Cani <i>et al.</i> , 2009a; Gee and Johnson, 2005; Parnell and Reimer, 2009; Zhou <i>et al.</i> , 2008)
Ghrelin	P/D1 cells in the stomach and e-cells of the pancreas	Stimulates appetite	n.r.	Oligofructose, resistant starch	Rat and human studies	(Cani <i>et al.</i> , 2007b; Parnell and Reimer, 2009; Zhou <i>et al.</i> , 2008)
FIAF	Liver, adipose tissue, intestine, brain, thyroid, heart, kidney, skeletal muscles, spleen, pituitary gland, hypothalamus, placenta	Hypothalamic appetite control, fat storage in adipocytes	LPS	n.r.	N1-neuronal cells, 3T3-L1 cell line	(Brown <i>et al.</i> , 2009)
			Propionate, butyrate	n.r.	HT-29, Caco-2, HCT-116, HepG2 cell lines	(Grootaert <i>et al.</i> , 2011)
			<i>Lactobacillus</i> , <i>Bifidobacterium</i> , <i>Bacteroides</i> ,	n.r.	HT-29, Caco-2, HCT-116, LoVo, SW-480 and HepG2	(Are <i>et al.</i> , 2008; Aronsson <i>et al.</i> , 2010; Bäckhed <i>et al.</i> , 2004; Bäckhed <i>et al.</i> , 2007; Grootaert <i>et al.</i> , 2011)

			<i>Methanobrevibacter</i> , <i>Enterococcus</i> , <i>Escherichia coli</i>		cell lines, mice studies	
			CLA	PUFA	COS cells	(Tien <i>et al.</i> , 2004)
			n.r.	Chitosan	High fat diet induced mice	(Neyrinck <i>et al.</i> , 2009)
Leptin	Adipocytes	Stimulates food intake	n.r.	Chitosan	High fat diet induced obese mice	(Neyrinck <i>et al.</i> , 2009)
			C2-C6 SCFA	n.r.	Murine primary adipocytes	(Xiong <i>et al.</i> , 2004)
			Propionate	n.r.	Human adipose tissue, transfected murine adipocyte cell lines	(Lahham <i>et al.</i> , 2008; Xiong <i>et al.</i> , 2004)
Adiponectin	Adipose tissue	Triglyceride storage, insulin sensitivity	n.r.	Cereal fiber	Human study	(Qi <i>et al.</i> , 2005)
Cholesterol	Liver/bile	Lipid transport in GIT and blood	SCFA, mainly propionate	Xylo- oligosaccharides, chito- oligosaccharides, soybean oligosaccharides, resistant starch	Rat, hamster, human, chicken	(Chen <i>et al.</i> , 2010; Cheng and Lai, 2000; de Luis <i>et al.</i> , 2010; Delzenne <i>et al.</i> , 1993; Fiordaliso <i>et al.</i> , 1995; Hsu <i>et al.</i> , 2004; Li <i>et al.</i> , 2007; Trautwein <i>et al.</i> , 1998; Venter <i>et al.</i> , 1990; Williams and Jackson, 2002)