Targeting gut microbiota in obesity: effects of prebiotics and probiotics

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Abstract | At birth, the human colon is rapidly colonized by gut microbes. Owing to their vast number and their capacity to ferment nutrients and secrete bioactive compounds, these gastrointestinal microbes act as an environmental factor that affects the host's physiology and metabolism, particularly in the context of obesity and its related metabolic disorders. Experiments that compared germ-free and colonized mice or analyzed the influence of nutrients that qualitatively change the composition of the gut microbiota (namely prebiotics) showed that gut microbes induce a wide variety of host responses within the intestinal mucosa and thereby control the gut's barrier and endocrine functions. Gut microbes also influence the metabolism of cells in tissues outside of the intestines (in the liver and adipose tissue) and thereby modulate lipid and glucose homeostasis, as well as systemic inflammation, in the host. A number of studies describe characteristic differences between the composition and/or activity of the gut microbiota of lean individuals and those with obesity. Although these data are controversial, they suggest that specific phyla, classes or species of bacteria, or bacterial metabolic activities could be beneficial or detrimental to patients with obesity. The gut microbiota is, therefore, a potential nutritional and pharmacological target in the management of obesity and obesity-related disorders.

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Introduction

The human gut is home to 10^{14} bacteria, which outnumber the total of eukaryotic cells in the human body by an order of magnitude. At birth, the gut of a neonate is sterile. However, at birth, it is immediately colonized by maternal and environmental bacteria, and the complexity of the resulting gut microbiota increases until the weaning to solid foods.[1] The adult microbiota harbors 1,000–1,150 bacterial species, and some experts have suggested that 160 of these species constitute the core microbiota that is present in most individuals.[2] Although many of these species are found in the majority of people, their relative abundance can vary greatly.[3,4] By contrast, studies of microbial coding sequences (called the metagenome) have made it increasingly clear that the functions encoded by the metagenome exhibit great similarity between individuals.[4] The human and mouse gut is dominated by several bacterial phyla including *Bacteroidetes*, *Firmicutes* and *Actinobacteria*.

Some studies have indicated that an altered gut microbiota is associated with several diseases that are particularly prevalent in the 21st century. For example, reduced microbial diversity—a sign of a dysfunctional ecosystem that leads to a decreased stability of the microbiota—has been associated with both inflammatory bowel disease and obesity.[2,4,5] The first studies on the relationship between the composition of the gut microbiota and obesity have shown that the number of *Firmicutes* was increased whereas the number of *Bacteroidetes* was reduced in obese mice and humans compared with lean individuals.[6–9] Interestingly, weight loss achieved by dieting was able to reverse those changes. Although the decrease in the number of *Bacteroidetes* was not observed in all studies,[10,11] changes in this phylum have been suggested to result from an increased energy intake rather than being caused directly by obesity.[9] The bacterial changes at the taxonomic level in individuals with obesity have been described elsewhere.[12,13]

In the first part of this Review, we describe the major factors that could modulate gut microbiota composition, including genetic background, sex, age and diet of the host. In the second part, we describe how gut microbes change the energy metabolism of the host by altering the expression of genes involved in the development of adiposity and obesity-related metabolic disorders, including inflammation. The last part of the Review focuses on the potential role of specific nutrients that target the gut microbiota in the control of obesity and its comorbidities.

Selecting host-adapted gut microbiota

Various components influence the microbial ecology of the gut (Figure 1). In an elegant study, Rawls and coworkers demonstrated that the host can select an optimal microbiota.[14] When germ-free zebrafish were colonized with anaerobic mouse gut microbiota and germ-free mice were colonized with aerobic zebrafish microbiota, the host microbiota reshaped the transplanted microbiota within 2 weeks in both cases.[14] Obviously, physiological characteristics and habitats of fish and mice are very different. However, this observation suggests that the host

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Key points

- The host's intrinsic characteristics, such as genetic factors, the state of the immune system and nutrition are important factors for selecting and shaping the gut microbiota
- Gut microbiota might modulate adiposity by changing the expression of host genes that are involved in fat storage and oxidation, gastrointestinal hormone production and barrier function and in the inflammatory response
- Although all individuals are born with a specific microbiome, the diet can change both the composition and the activity of the microbiota
- Certain fermentable carbohydrates with prebiotic properties can counteract the overexpression of several host targets that are involved in the development of adiposity, metabolic disorders and inflammation

exerts a key influence on the composition of the gut microbiota, although the underlying mechanisms are yet to be clarified.

Genetics

Profiling the gut microbiota of eight different mouse strains—to mimic the genetic diversity of the human population—by DNA fingerprinting suggested that the genetic background of the host has a stronger influence on microbiota composition than the sex of the host.[15] Another study confirmed that the genotype of the host is an important factor in selecting and shaping the gut microbiota.[6]

At birth, the sterile gut of the newborn baby is colonized by bacteria from the mother and the environment.[16] Accordingly, the gut microbiota of both mice pups and human neonates closely resembles that of their mothers.[16] As the mother and the offspring share half of their genes and part of their gut microbiota, whether the development of the gut microbiota is determined by the offspring's genes or the offspring is supplied with an optimal gut microbiota at birth is unknown.

Diet

As mentioned previously, weaning to solid food has a profound effect on the composition and complexity of the gut microbiota. The gut microbiota is greatly responsive to dietary changes in adulthood, too. After switching mice to a high-fat Western diet, drastic changes occur in the composition of their gut microbiota. In particular, the number of bacteria that belong to the class *Erysipelotrichi* of the phylum *Firmicutes* increases dramatically.[17–20] These responses are very rapid; they occur within the first 24 h after changing the animals' diet.[19] Dietary carbohydrates, especially those that are not digested in the upper part of the gut, might also change the composition of the gut microbiota.

The concept of prebiotics was proposed in 1995 on the basis of the observation that certain nondigestible carbohydrates, following fermentation by bacteria, can drive qualitative and selective changes in the composition of the gut microbiota, which have beneficial effects on the host's health.[21] The number of *Bifidobacteria* (phylum *Actinobacteria*) has been shown to increase in the presence of inulin-type fructans with prebiotic properties. This increase occurs within a few days, but rapidly disappears upon withdrawal of the prebiotic compounds (after 1 week). The extent of increase in the number of *Bifidobacteria* is also dependent on their initial number in the gut.[21] Breastmilk also contains oligosaccharides with prebiotic properties that contribute to the increase in the number of *Bifidobacteria* after birth. These findings illustrate that diet has a crucial role in the modulation of the gut microbiota during an individual's lifetime, and the nutrient composition of the diet has to be taken into account when studying the relationship between the composition of the gut microbiota and the host's health.

Immune system

The immune system also seems to be important in the selection of the gut microbiota. Mice that have abnormal Toll-like receptor (Tlr) signaling or produce bactericidal reactive oxygen species have elevated serum titers of antibodies against their commensal microbiota.[24] The increased serum titers are required to maintain the

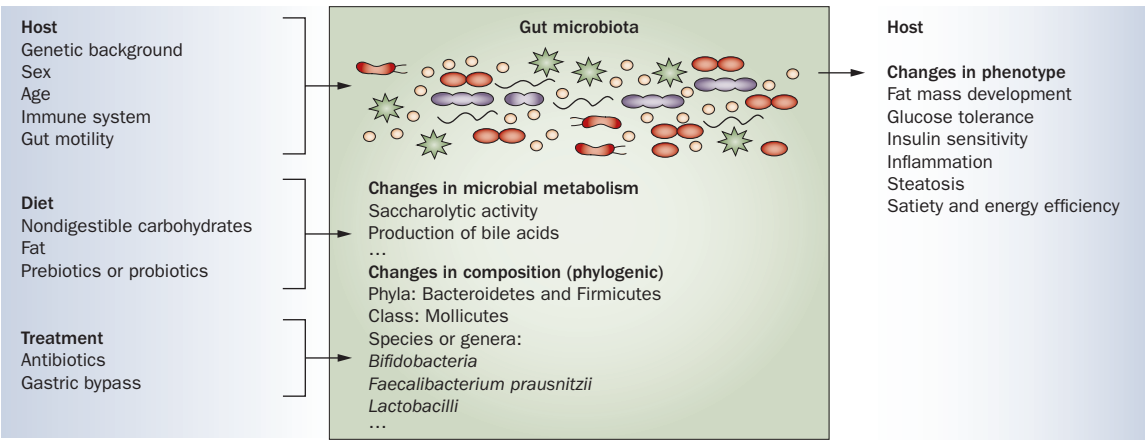

Figure 1 | The gut microbiota is a central component of the host's phenotype. The host's intrinsic characteristics and diet both influence the composition and metabolic activity of the gut microbiota. Changes in the gut microbiota affect the processes involved in energy storage and influence gene expression in various tissues of the host, which contributes to the occurrence of metabolic disorders associated with obesity.

commensal relationship between the gut microbiota and the host. Mutant mice that lack Tlrs (or the signaling components downstream of Tlrs, such as Myd88) have an altered microbial composition in the gut.[25,26]

Characteristics of the gut microbiota contribute to the host's phenotype (Figure 1). If an altered gut microbiota is transplanted into germ-free recipients, the functional characteristics of the donor microbiota can be transferred as well. Non-obese diabetic (NOD) mutant mice, which are prone to develop type 1 diabetes mellitus, were protected from the disease if *Myd88* was ablated, and this protection could be transferred to germ-free recipients.[25] The obesity trait was transmissible by transplanting the 'obese' microbiota of leptin-deficient *ob/ob* mice into germ-free mice,[7] or by transplantation of a Western-diet-associated microbiome to germ-free mice, even if the recipient mice consumed a standard, low-fat, high-carbohydrate diet after transplantation.[17] Furthermore, *Tlr5*-deficient mice develop obesity, inflammation and metabolic syndrome.[26] This phenotype is also transferred upon transplantation of the gut microbiota.

Other factors

In the host's intestines, several factors prevent the overgrowth of the microbiota. These factors include physical mechanisms, such as the rapid movement of epithelial cells from the crypt to the villus, the peristaltic movement of the gut[22] and a thick mucus layer produced by goblet cells that selectively limits the access of bacteria to the epithelium in the colon.[23]

Gut microbiota and fat storage

Compared with their germ-free counterparts, mice with gut microbiota have an increased capability to harvest energy from the gut.[27] Metagenomic analyses of the microbiota performed in obese mice and humans revealed an increased capacity for the degradation (fermentation) of carbohydrates.[4,7] This shift increases the amount of short-chain fatty acids (such as acetate, propionate, butyrate, and L-lactate) that can be used as metabolic substrates by the host to increase the harvest of energy.

In addition to their role as energy substrates, short-chain fatty acids have been proposed to bind to specific G-protein-coupled receptors (GPR41 and GPR43, also called the free fatty acid receptors FFAR3 and FFAR2, respectively), which might promote nutrient absorption and/or adipose tissue mass development. Studies performed in *Gpr41*-deficient mice suggested that the activation of GPR41 by short-chain fatty acids is responsible for the release of the gut hormone PYY. This peptide has been shown to decrease the intestinal transit time, which indicates that it promotes the absorption of nutrients, mostly glucose.[28] Moreover, *Gpr43* is overexpressed in mice that are fed an obesogenic, high-fat diet,[29] which contributes to an increase in adipocyte differentiation and inhibits lipolysis in the adipose tissue. *Gpr43*-deficient mice fed a high-carbohydrate, high-fat diet had a lower body mass and a higher lean mass compared with wild-type mice.[30] The short-chain fatty acids produced by fermentation could then act in different ways: as energy substrates and/or as metabolic regulators.

However, the energy spared by fermentation is not sufficient to explain why mice with normal gut microbiota that were fed with a high-carbohydrate, high-fat diet developed more adipose tissue and exhibited a greater glucose intolerance than germ-free mice that were fed the same diet.[20,31,32] Furthermore, the drastic changes in the gut microbiota's composition that occur after an antibiotic treatment can protect against obesity, glucose intolerance and the insulin resistance induced by a high-fat, carbohydrate-free diet.[33]

The gut microbiota might affect obesity by additional mechanisms beyond energy harvest and the associated short-chain fatty acid production. In accordance with this hypothesis, the gut microbiota also influence the expression of host genes, namely of those that are expressed in the intestine, and control fatty acid absorption, oxidation and storage. One such target is the angiopoietin-related protein 4 (Angptl4), a potent lipoprotein lipase inhibitor.[34] Angptl4 inhibits the uptake of fatty acids from circulating triglyceride-rich lipoproteins in white adipose and muscle tissues and promotes fatty acid oxidation, both in skeletal muscle cells and in adipocytes.[31,35] The overexpression of *Angptl4 i*n white adipose tissue also reduces fat mass.[35] Conversely, germ-free, *Angptl4*-deficient mice exhibit increased lipoprotein lipase activity and adiposity compared with their wild-type counterparts.[36] Interestingly, normal mice exhibit a reduced expression of *Angptl4* in the small intestine compared with germ-free mice, which promotes adipose tissue development.[36] These observations suggest that Angptl4 is a key host protein that is responsive to the gut's microbial environment and can modulate adiposity by controlling fatty acid uptake and metabolism in the tissues.

Colonization of germ-free mice with a typical environmental microbial population stimulates triglyceride synthesis and glycogenesis in the liver. These changes are attributable to a specific microbial family of the phylum *Actinobacteria*, namely *Coriobacteriaceae*.[37] Interestingly, nuclear magnetic resonance spectroscopy of urinary and tissue samples showed changes in bile acid metabolism in the liver of colonized mice, which might contribute to the increase in dietary lipid absorption and the development of hepatic steatosis.[37]

Gut microbiota and obesity

According to a new hypothesis, gut microbes have a role in the host's metabolic homeostasis.[38,39] As type 2 diabetes mellitus and obesity are associated with low-grade inflammation and an altered composition of the gut microbiota, a bacterial compound might act as a triggering factor in the development of obesity, diabetes mellitus and inflammation induced by a high-fat diet. Several experiments indicated that this bacterial compound might be lipopolysaccharide (LPS), a component of the cell wall of gram-negative bacteria.

Mice fed a high-fat diet exhibited enhanced levels of plasma LPS, a state described as metabolic endotoxemia.[40] The elevated serum LPS level is unlikely to be explained

by an increased number of gram-negative bacteria in the gut of individuals with obesity, as such an increase has not been observed in mice nor humans.[12,40,41] Associations between circulating LPS level, consumption of a high-fat diet and the presence of obesity and type 2 diabetes mellitus have been confirmed in humans. Erridge and coworkers found that a high-fat diet induces metabolic endotoxemia in healthy individuals.[42] A link between energy intake (high-fat diet) and metabolic endotoxemia has also been described.[43] Moreover, Creely and colleagues demonstrated that metabolic endotoxemia is a factor associated with the development of type 2 diabetes mellitus.[44]

Associations between endotoxemia and serum levels of insulin and triglycerides and an inverse relationship between endotoxemia and serum HDL cholesterol level were confirmed in patients with type 2 diabetes mellitus and obesity.[45] Furthermore, associations have been proposed between high-fat diet, metabolic endotoxemia and levels of inflammatory markers (TLRs and SOCS3) in mononuclear cells.[46,47] Finally, metabolic endotoxemia is associated with systemic and adipose tissue inflammation in pregnant women with obesity.[48] Altogether, these findings reinforce the hypothesis that fat intake and absorption, obesity and the development of metabolic endotoxemia are related.

Some of the mechanisms that are involved in the development of metabolic endotoxemia seem to be related to the fat content of the diet. Several investigators have shown that intraluminal fat increases intestinal LPS absorption through its incorporation into chylomicrons.[47,50,51] Accordingly, the administration of lipase inhibitors reduces the severity of metabolic endotoxemia.[52] A growing amount of evidence indicates that changes in the integrity of the intestinal barrier occur both in the proximal and the distal part of the gut, which can contribute to the entrance of LPS into the systemic circulation.[33,53–56] Altered distribution and localization of two tight-junction proteins (ZO-1 and occludin) in the intestinal epithelium have been associated with an increased permeability of the intestinal wall in obese and diabetic rodents.[33,53–56] Furthermore, glucagon-like peptide-2 (GLP-2), a gut peptide already known to be involved in the control of epithelial cell proliferation, was confirmed as a regulator of the expression and localization of tight-junction proteins and of the permeability of the intestinal wall in obese mice.[53]

The intestinal endocannabinoid system is expressed differently in germ-free and normal mice and is overactivated in obese mice.[54] The endocannabinoid system is composed of bioactive lipids (including anandamide) that bind to specific receptors (cannabinoid receptors 1 and 2 and peroxisome proliferator-activated receptors) and thereby influences energy homoeostasis and immunity.[57] Activation of the endocannabinoid system in *ob*/*ob* mice contributes to an increased permeability of the intestinal wall, an increased plasma level of LPS and systemic inflammation.[54] A 'crosstalk' between the endocannabinoid system and the gut microbiota also participates in the regulation of adipogenesis directly by acting on the adipose tissue and indirectly by increasing plasma LPS levels.[54] Although correlations have been found between changes in the gut microbiota composition and the elements controlling the gastrointestinal barrier function, such as GLP-2 and the endocannabinoid system, the direct involvement of specific gut microbes and/or of microbial metabolites in this control remains to be elucidated.

In humans, the contribution of changes in the integrity of the gut barrier to obesity and obesity-related inflammation remains to be confirmed. Brignardello and coworkers did not find any substantial difference in the permeability of the intestinal wall in the proximal gut in obese versus lean individuals.[58] Further studies with a greater number of patients are needed to confirm the relationships between the permeability of the gut wall (in the proximal as well as the distal part of the gut), endotoxemia and the metabolic alterations in patients with obesity and type 2 diabetes mellitus.

Few studies have been aimed at finding correlations between the composition of the microbiota and the occurrence of inflammation and metabolic alterations in individuals with obesity.[9,49] An interesting study showed that the number of *Faecalibacterium prausnitzii* decreases in patients with diabetes mellitus and is inversely correlated with inflammatory markers.[9] The low-grade systemic inflammation that characterizes the obese phenotype is controlled by peptides that are produced in the gut. These peptides are influenced by the presence or absence of the gut microbiota.[33,53,55,56] One such protein is the serum amyloid A3 protein (SAA3), which is the most abundant SAA isoform in both the adipose tissue and the colon.[59] SAA3 is upregulated in the adipose tissue of mice fed a high-fat diet and might be a mediator of the chronic inflammation associated with insulin resistance in obesity.[60] The gut microbiota is an important regulator of SAA3 expression. Expression of this peptide was substantially increased in the adipose tissue and colon (by 10-fold and sevenfold, respectively) of conventionalized mice —that is, mice colonized with a normal gut microbiota from a healthy wild-type mouse—compared with germ-free mice. Interestingly, SAA3 expression was mediated through the TLR–MyD88–NFκB signaling pathway.[59]

Taken together, these findings suggest that the gut microbiota modulates the biological systems that regulate the availability of nutrients, energy storage, fat mass development and inflammation in the host, which are all components of the obese phenotype (Figure 2).

Targeted changes in gut microbiota

Effects of prebiotics

In individuals with obesity, changes in the composition of the gut microbiota occur not only at the level of phyla but also at the level of genera or species.[12] For example, a lower number of *Bifidobacteria* at birth has been associated with overweight later in childhood.[61] Furthermore, overweight mothers give birth to neonates that have a decreased number of *Bifidobacteria*, which suggests that obesogenic microbiota is an 'inheritable' trait.[62] In adults, the number of *Bifidobacteria* (and of most groups

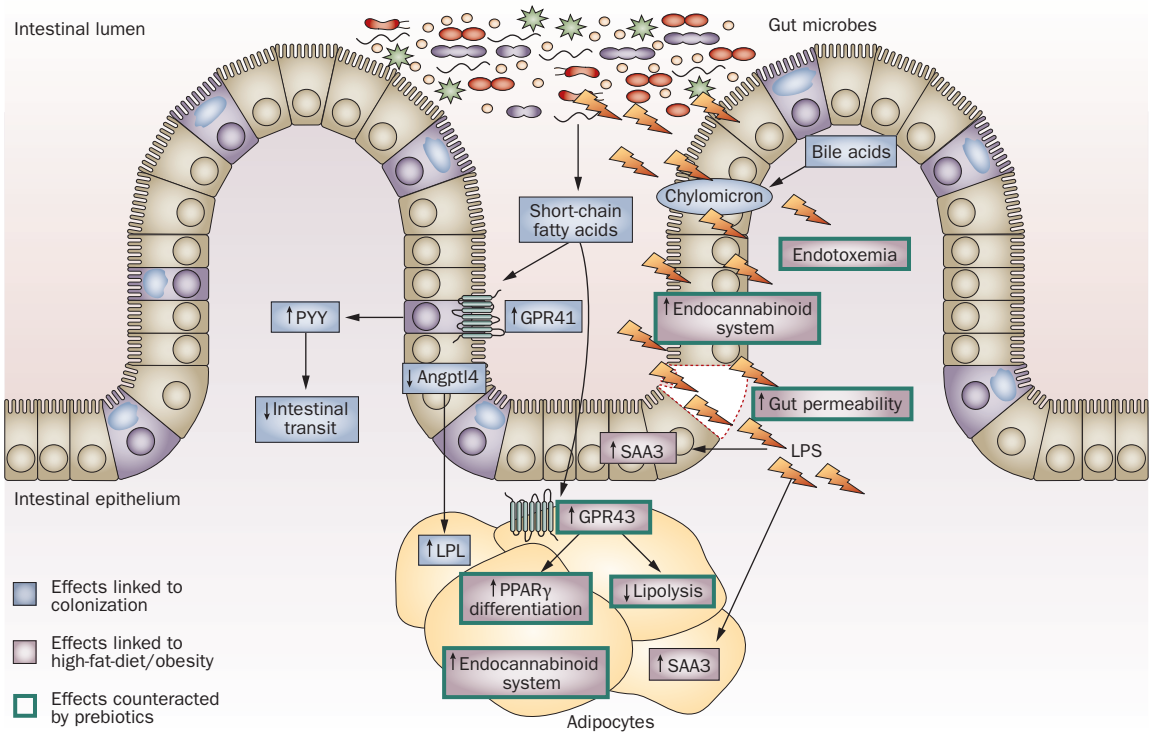

Figure 2 | The host's metabolic responses to changes affecting its gut microbiota, including colonization of the gut in germ-free mice, obesity and high-fat diet, and administration of prebiotics. The short-chain fatty acids produced by the fermentation of carbohydrates bind to GPR41 in the intestine and promote the expression of PYY, which slows down the intestinal transit. Some short-chain fatty acids also activate GPR43—the expression of which is increased by a high-fat diet—in the adipose tissue. This activation decreases lipolysis, increases PPARγ-related differentiation and thereby increases adiposity. The gut microbiota promotes LPL-controlled fatty acid storage in the adipose tissue by blunting the intestinal expression of ANGPTL4. The increased LPS level in the blood is linked to the intestinal activation of the endocannabinoid system that increases the gut's permeability. LPS activates the production of SAA3 peptide in the gut and the adipose tissue. In obese animals, prebiotics increase the production of GLP-1 and GLP-2, decrease the intestinal wall's permeability, inhibit the endocannabinoid system in the gut and the adipose tissue, blunt the overexpression of GPR43 and increase lipolysis in the adipose tissue, thereby decreasing adiposity. Abbreviations: ANGPTL4, angiopoietin-related protein 4; GLP, glucagon-like peptide; GPR, G-protein coupled receptor; LPL, lipoprotein lipase; LPS, lipopolysaccharide; PPARγ, peroxisome proliferator-activated receptor γ; PPY, peptide YY; SAA3, serum amyloid A3 protein.

of *Firmicutes*) is slightly lower in individuals with obesity than in lean people.[11] The number of these bacteria is also decreased in patients with type 2 diabetes mellitus compared with nondiabetic patients.[63] These findings suggest that *Bifidobacteria* play a part in the development of obesity and its related comorbidities.

Bifidobacteria served as a model for the concept of prebiotics,[21] which has been 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".[21] Dietary fructans, which are present in various fruits and vegetables and added to food products, are used as an energy substrate by bacteria, including *Bifidobacterium spp.*, that express β-fructofuranosidase, which promotes their development in the gut. A remarkable increase has been observed in the number of *Bifidobacterium spp.* in mice with diet-induced or genetically determined obesity that were supplemented with inulin-type fructans.[29,41,53] Interestingly, the number of *Bifidobacteria* was inversely correlated with the development of fat mass, glucose intolerance, and LPS level.[41] Moreover, the prebiotic approach prevented the overexpression of several host genes that are related to adiposity and inflammation, which was also associated with the colonization of germ-free mice with gut microbiota (Figure 2). Inulin-type fructans increased the number of endocrine L cells in the jejunum and in the colon of rodents, and promoted the production and release of the active forms of GLP-1 and GLP-2 in the portal vein.[64–67]

Studies on experimental models of GLP-receptor invalidation indicate that GLP-1 participates in prebiotic-driven decreases in appetite, fat mass and hepatic insulin resistance,[67] whereas GLP-2, as mentioned before, contributes to the reduced permeability of the intestinal wall and endotoxemia that are associated with obesity.[53] Interestingly, a 2-week treatment with inulin-type fructans (16 g per day) in healthy volunteers increased the postprandial release of gut peptides (namely GLP-1 and gastric inhibitory peptide), to modify eating behavior (increased satiety and decreased calorie intake) and decrease postprandial glycemia.[68,69] In addition, inulin-type fructans also decreased the activity of the endocannabinoid system (by reducing the expression of cannabinoid receptor 1,

Table 1 | Effects of probiotics or carbohydrates with prebiotic properties in patients with overweight or diabetes mellitus

Microbiota	Study design	*n*	Duration	Treatment	Results
Probiotics					
Lactobacillus acidophilus NCFM[75]	Randomized, double-blind intervention	45 individuals with glucose intolerance and/or diabetes mellitus	4 weeks	Probiotic (10^{10} CFU/day) versus SiO_2/lactose (placebo)	Systemic inflammation upon LPS challenge in both groups Probiotics prevented loss of insulin sensitivity observed in the placebo group
Lactobacillus gasseri SBT2055[76]	Randomized, multicenter, double-blind, placebo-controlled intervention	87 individuals with a BMI of 24.2–37.0 kg/m^2 and visceral adiposity	12 weeks	Fermented milk with probiotics (10^{11} CFU/day) or without probiotics (placebo)	Reduced body weight, BMI, waist and hip circumference, visceral and subcutaneous fat mass in the probiotic versus the placebo group
Prebiotics (nondigestible carbohydrates)					
Arabinoxylan*[77]	Randomized cross-over intervention	15 individuals with type 2 diabetes mellitus	5 weeks	Bread and muffins with 14% arabinoxylan (0% for placebo)	Reduced fasting glycemia, ↓ post-OGTT glycemia and insulinemia No difference in blood lipid level, fat mass and blood pressure
Arabinoxylan[78,79]	Single-blind, controlled, cross-over intervention	11 individuals with impaired glucose tolerance	6 weeks	15 g arabinoxylan supplied daily via bread and powder or isocaloric bread rolls without arabinoxylan (placebo)	Reduced fasting and post-LMCT glycemia and triglyceridemia Reduced total post-LMCT ghrelin No difference in leptin, adiponectin, insulin, resistin and FFA levels
Inulin-type fructans‡[80]	Randomized, double-blind, placebo-controlled intervention	48 individuals with overweight or obesity	12 weeks	21 g per day oligofructose or maltodextrin (placebo)	Reduced body weight, caloric intake, GIP No difference in fasting glucose, insulin, ghrelin, GLP-1, PYY and leptin levels After MTT: reduced glycemia, insulin, AUC for ghrelin, AUC for PYY, AUC for leptin, but no difference in GIP level or AUC for GLP-1
Inulin-type fructans[81]	Randomized, double-blind, cross-over intervention	10 individuals with type 2 diabetes mellitus	4 weeks	20 g short-chain fructans or 20 g sucrose (placebo)	No difference in caloric intake, body weight, levels of glucose, insulin, HDL, LDL and total cholesterol, triglyceride, apolipoprotein A1 and B, lipoprotein(a), FFA, hepatic glucose production, insulin-stimulated glucose metabolism
Inulin-type fructans[82]	Randomized, double-blind, cross-over, placebo-controlled intervention	7 overweight patients with nonalcoholic steatohepatitis	8 weeks	16 g per day oligofructose or maltodextrine (placebo)	Reduced aspartate aminotransferase and fasting insulin levels No difference in levels of triglycerides, fasting glucose and cholesterol

*Arabinoxylans are complex carbohydrates found in the endosperm and the aleurone layer and in pericarp tissues of cereals. Their fermentation is associated with proliferation of *Bifidobacteria* and *Lactobacilli*. Arabinoxylans represent a new class of prebiotics that have a prebiotic index comparable to that of well-established prebiotics.[83] ‡Inulin-type fructans are well-established prebiotics that can selectively stimulate the growth of *Bifidobacteria* and, in some cases, *Lactobacilli*, which markedly changes the composition of the gut microbiota. Most of the potential health benefits associated with their prebiotic effects were discovered and demonstrated using the same food ingredients and/or supplements.[42] Abbreviations: AUC, area under curve; CFU, colony-forming unit; GIP, gastric inhibitory polypeptide; GLP-1, glucagon-like peptide 1; LMCT, liquid meal challenge test; LPS, lipopolysaccharide; MTT, meal tolerance test; FFA, free fatty acids; OGTT, oral glucose tolerance test; PYY, peptide YY.

restoring the expression of anandamide-degrading enzyme and decreasing anandamide levels in the intestinal and adipose tissues), a phenomenon that contributes to an improved barrier function of the gut and adipogenesis.[54] Finally, inulin-type fructan prebiotics counteract the overexpression of GPR43 in the adipose tissue, which is related to a decreased rate of differentiation and a reduced adipocyte size.[29]

Effects of probiotics

Another approach for promoting specific changes in the gut microbiota is the oral delivery of viable strains of bacteria (probiotics) that are then integrated into the gut ecosystem. Interestingly, germ-free mice that are monocolonized with *Lactobacillus paracasei* in this way present an increased level of Angptl4. An increase in Angpt14 could also contribute to the decrease in fat mass observed in normal mice that are fed a high-fat diet supplemented with *L. paracasei*.[70] The relevance of lactobacilli supplementation for the control of adiposity is a matter of debate.[71–73] Some experts suggest that an increased number of intestinal *Lactobacillus spp.* is associated with an increased BMI and elevated level of blood glucose in healthy adults.[74] The genus *Lactobacillus* comprises more than 90 species, and a more complete picture is necessary to discern the relevance of specific subspecies or strains in the control of adiposity.

In individuals with obesity, the administration of different strains of *Lactobacilli* has been shown to decrease fat mass and the risk of type 2 diabetes mellitus and insulin resistance (Table 1). Table 1 also summarizes the few available intervention studies in humans that examined the potential health effect of carbohydrates with prebiotic properties in patients with overweight, obesity or type 2 diabetes mellitus. Unfortunately, none of these studies report changes in the composition of the gut microbiota after probiotic or prebiotic treatment. Therefore, we cannot state at the moment that specific

types of bacteria *per se* are responsible for the improvement of metabolism in individuals with obesity who are treated with probiotic or prebiotic preparations.

Conclusions

Several potential mechanisms might allow gut microbes to interact with the host's tissues and to regulate its energy metabolism. The increase in LPS levels, defined as metabolic endotoxemia, that occurs in individuals with obesity demonstrates that specific components of the gut microbes could trigger metabolic disorders. Experimental studies performed in animals clearly show that the gut microbiota also influences energy metabolism of the host, by regulating systems that have a crucial role in the control of nutrient absorption and metabolism, the integrity of the gut barrier, adipogenesis or hormonal status. These findings indicate that certain molecular targets (namely ANGPTL4, GPR43/41, GLP-2 and the intestinal endocannabinoid system) might be involved in the control of obesity and obesity-related disorders.

Numerous data have been published regarding differences in the composition of the gut microbiota in obese versus lean individuals, in animal models as well as in humans. At present, we cannot conclude that specific genera, classes or species of bacteria are always positively or negatively associated with the obese phenotype. In most cases, however, statistically significant relationships have been established between the presence and/or the amount of specific bacteria and the host phenotype. Integrative approaches using metabolomics and metagenomics should be performed to elucidate the metabolic interactions between the host and the gut microbes in individuals with obesity, and to assess the relevance of prebiotic or probiotic approaches in the control of obesity and obesity-related diseases in humans.

Review criteria

A PubMed search was performed by using the following key words (separately or combined): "gut microb*", "bacteria", "obesity", "adiposity", "energy", "metabolism", "inflammation", "prebiotic*", "probiotic*". Selected papers, including reviews, were published between 1990 and 2011 and chosen on the basis of their content (quality and novelty). The authors focused on specific molecular aspects of the interactions between the host and gut microbes, including endotoxemia. The final list of references was established by adding references suggested by the peer-reviewers.

1. Reinhardt, C., Reigstad, C. S. & Bäckhed, F. Intestinal microbiota during infancy and its implications for obesity. *J. Pediatr. Gastroenterol. Nutr.* **48**, 249–256 (2009).
2. Qin, J. *et al.* A human gut microbial gene catalogue established by metagenomic sequencing. *Nature* **464**, 59–65 (2010).
3. Reyes, A. *et al.* Viruses in the faecal microbiota of monozygotic twins and their mothers. *Nature* **466**, 334–338 (2010).
4. Turnbaugh, P. J. *et al.* A core gut microbiome in obese and lean twins. *Nature* **457**, 480–484 (2009).
5. Ott, S. J. *et al.* Reduction in diversity of the colonic mucosa associated bacterial microflora in patients with active inflammatory bowel disease. *Gut* **53**, 685–693 (2004).
6. Ley, R. E. *et al.* Obesity alters gut microbial ecology. *Proc. Natl Acad. Sci. USA* **102**, 11070–11075 (2005).
7. Turnbaugh, P. J. *et al.* An obesity-associated gut microbiome with increased capacity for energy harvest. *Nature* **444**, 1027–1031 (2006).
8. Ley, R. E., Turnbaugh, P. J., Klein, S. & Gordon, J. I. Microbial ecology: human gut microbes associated with obesity. *Nature* **444**, 1022–1023 (2006).
9. Furet, J. P. *et al.* Differential adaptation of human gut microbiota to bariatric surgery-induced weight loss: links with metabolic and low-grade inflammation markers. *Diabetes* **59**, 3049–3057 (2010).
10. Duncan, S. H. *et al.* Human colonic microbiota associated with diet, obesity and weight loss. *Int. J. Obes. (Lond.)* **32**, 1720–1724 (2008).
11. Schwiertz, A. *et al.* Microbiota and SCFA in lean and overweight healthy subjects. *Obesity (Silver Spring)* **18**, 190–195 (2010).
12. Ley, R. E. Obesity and the human microbiome. *Curr. Opin. Gastroenterol.* **26**, 5–11 (2010).
13. Greiner, T. & Bäckhed, F. Effects of the gut microbiota on obesity and glucose homeostasis. *Trends Endocrinol. Metab.* **22**, 117–123 (2011).
14. Rawls, J. F., Mahowald, M. A., Ley, R. E. & Gordon, J. I. Reciprocal gut microbiota transplants from zebrafish and mice to germ-free recipients reveal host habitat selection. *Cell* **127**, 423–433 (2006).
15. Kovacs, A. *et al.* Genotype is a stronger determinant than sex of the mouse gut microbiota. *Microb. Ecol.* **61**, 423–428 (2011).
16. Dominguez-Bello, M. G. *et al.* Delivery mode shapes the acquisition and structure of the initial microbiota across multiple body habitats in newborns. *Proc. Natl Acad. Sci. USA* **107**, 11971–11975 (2010).
17. Turnbaugh, P. J., Bäckhed, F., Fulton, L. & Gordon, J. I. Diet-induced obesity is linked to marked but reversible alterations in the mouse distal gut microbiome. *Cell Host Microbe* **3**, 213–223 (2008).
18. Hildebrandt, M. A. *et al.* High-fat diet determines the composition of the murine gut microbiome independently of obesity. *Gastroenterology* **137**, 1716–1724 (2009).
19. Turnbaugh, P. J. *et al.* The effect of diet on the human gut microbiome: a metagenomic analysis in humanized gnotobiotic mice. *Sci. Transl. Med.* **1**, 6ra14 (2009).
20. Fleissner, C. K. *et al.* Absence of intestinal microbiota does not protect mice from diet-induced obesity. *Br. J. Nutr.* **104**, 919–929 (2010).
21. Roberfroid, M. *et al.* Prebiotic effects: metabolic and health benefits. *Br. J. Nutr.* **104**, S1–S63 (2010).
22. Macfarlane, G. T. & Macfarlane, L. E. Acquisition, evolution and maintenance of the normal gut microbiota. *Dig. Dis.* **27** (Suppl. 1), 90–98 (2009).
23. Derrien, M. *et al.* Mucin-bacterial interactions in the human oral cavity and digestive tract. *Gut Microbes* **1**, 254–268 (2010).
24. Slack, E. *et al.* Innate and adaptive immunity cooperate flexibly to maintain host-microbiota mutualism. *Science* **325**, 617–620 (2009).
25. Wen, L. *et al.* Innate immunity and intestinal microbiota in the development of type 1 diabetes. *Nature* **455**, 1109–1113 (2008).
26. Vijay-Kumar, M. *et al.* Metabolic syndrome and altered gut microbiota in mice lacking Toll-like receptor 5. *Science* **328**, 228–231 (2010).
27. Wostmann, B. S., Larkin, C., Moriarty, A. & Bruckner-Kardoss, E. Dietary intake, energy metabolism, and excretory losses of adult male germfree Wistar rats. *Lab. Anim. Sci.* **33**, 46–50 (1983).
28. Samuel, B. S. *et al.* Effects of the gut microbiota on host adiposity are modulated by the short-chain fatty-acid binding G. protein-coupled receptor, Gpr41. *Proc. Natl Acad. Sci. USA* **105**, 16767–16772 (2008).
29. Dewulf, E. M. *et al.* Inulin-type fructans with prebiotic properties counteract GPR43 overexpression and PPARγ-related adipogenesis in the white adipose tissue of high-fat diet-fed mice. *J. Nutr. Biochem.* **22**, 712–722 (2011).
30. Bjursell, M. *et al.* Improved glucose control and reduced body fat mass in free fatty acid receptor 2-deficient mice fed a high-fat diet. *Am. J. Physiol. Endocrinol. Metab.* **300**, E211–E220 (2011).
31. Bäckhed, F., Manchester, J. K., Semenkovich, C. F. & Gordon, J. I. Mechanisms underlying the resistance to diet-induced obesity in germ-free mice. *Proc. Natl Acad. Sci. USA* **104**, 979–984 (2007).
32. Rabot, S. *et al.* Germ-free C57BL/6J mice are resistant to high-fat-diet-induced insulin resistance and have altered cholesterol metabolism. *FASEB J.* **24**, 4948–4959 (2010).
33. Cani, P. D. *et al.* Changes in gut microbiota control metabolic endotoxemia-induced inflammation in high-fat diet-induced obesity and diabetes in mice. *Diabetes* **57**, 1470–1481 (2008).
34. Yoshida, K., Shimizugawa, T., Ono, M. & Furukawa, H. Angiopoietin-like protein 4 is a potent hyperlipidemia-inducing factor in mice and inhibitor of lipoprotein lipase. *J. Lipid Res.* **43**, 1770–1772 (2002).
35. Mandard, S. *et al.* The fasting-induced adipose factor/angiopoietin-like protein 4 is physically associated with lipoproteins and governs plasma lipid levels and adiposity. *J. Biol. Chem.* **281**, 934–944 (2006).

36. Bäckhed, F. *et al.* The gut microbiota as an environmental factor that regulates fat storage. *Proc. Natl Acad. Sci. USA* **101**, 15718–15723 (2004).
37. Claus, S. P. *et al.* Colonization-induced host-gut microbial metabolic interaction. *MBio.* **2**, pii: e00271-10 (2011).
38. Cani, P. D. & Delzenne, N. M. The role of the gut microbiota in energy metabolism and metabolic disease. *Curr. Pharm. Des.* **15**, 1546–1558 (2009).
39. Cani, P. D. & Delzenne, N. M. Interplay between obesity and associated metabolic disorders: new insights into the gut microbiota. *Curr. Opin. Pharmacol.* **9**, 737–743 (2009).
40. Cani, P. D. *et al.* Metabolic endotoxemia initiates obesity and insulin resistance. *Diabetes* **56**, 1761–1772 (2007).
41. Cani, P. D. *et al.* Selective increases of bifidobacteria in gut microflora improve high-fat-diet-induced diabetes in mice through a mechanism associated with endotoxaemia. *Diabetologia* **50**, 2374–2383 (2007).
42. Erridge, C., Attina, T., Spickett, C. M. & Webb, D. J. A high-fat meal induces low-grade endotoxemia: evidence of a novel mechanism of postprandial inflammation. *Am. J. Clin. Nutr.* **86**, 1286–1292 (2007).
43. Amar, J. *et al.* Energy intake is associated with endotoxemia in apparently healthy men. *Am. J. Clin. Nutr.* **87**, 1219–1223 (2008).
44. Creely, S. J. *et al.* Lipopolysaccharide activates an innate immune system response in human adipose tissue in obesity and type 2 diabetes. *Am. J. Physiol. Endocrinol. Metab.* **292**, E740–E747 (2007).
45. Al-Attas, O. S. *et al.* Changes in endotoxin levels in T2DM subjects on anti-diabetic therapies. *Cardiovasc. Diabetol.* **8**, 20 (2009).
46. Deopurkar, R. *et al.* Differential effects of cream, glucose, and orange juice on inflammation, endotoxin, and the expression of Toll-like receptor-4 and suppressor of cytokine signaling-3. *Diabetes Care* **33**, 991–997 (2010).
47. Ghanim, H. *et al.* Increase in plasma endotoxin concentrations and the expression of Toll-like receptors and suppressor of cytokine signaling-3 in mononuclear cells after a high-fat, high-carbohydrate meal: implications for insulin resistance. *Diabetes Care* **32**, 2281–2287 (2009).
48. Basu, S. *et al.* Pregravid obesity associates with increased maternal endotoxemia and metabolic inflammation. *Obesity (Silver Spring)* **19**, 476–482 (2011).
49. Larsen, N. *et al.* Gut microbiota in human adults with type 2 diabetes differs from non-diabetic adults. *Plos ONE* **5**, e9085 (2010).
50. Ghoshal, S., Witta, J., Zhong, J., de, V. W. & Eckhardt, E. Chylomicrons promote intestinal absorption of lipopolysaccharides. *J. Lipid Res.* **50**, 90–97 (2009).
51. Laugerette, F. *et al.* Emulsified lipids increase endotoxemia: possible role in early postprandial low-grade inflammation. *J. Nutr. Biochem.* **22**, 53–59 (2011).
52. Dixon, A. N. *et al.* Effect of the orlistat on serum endotoxin lipopolysaccharide and adipocytokines in South Asian individuals with impaired glucose tolerance. *Int. J. Clin.* Pract. **62**, 1124–1129 (2008).
53. Cani, P. D. *et al.* Changes in gut microbiota control inflammation in obese mice through a mechanism involving GLP-2-driven improvement of gut permeability. *Gut* **58**, 1091–1103 (2009).
54. Muccioli, G. G. *et al.* The endocannabinoid system links gut microbiota to adipogenesis. *Mol. Syst. Biol.* **6**, 392 (2010).
55. Brun, P. *et al.* Increased intestinal permeability in obese mice: new evidence in the pathogenesis of nonalcoholic steatohepatitis. *Am. J. Physiol. Gastrointest. Liver Physiol.* **292**, G518–G525 (2007).
56. De La Serre, C. B. *et al.* Propensity to high-fat diet-induced obesity in rats is associated with changes in the gut microbiota and gut inflammation. *Am. J. Physiol. Gastrointest. Liver Physiol.* **299**, G440–G448 (2010).
57. Maccarrone, M. *et al.* The endocannabinoid system and its relevance for nutrition. *Annu. Rev. Nutr.* **30**, 423–440 (2010).
58. Brignardello, J. *et al.* Pilot study: alterations of intestinal microbiota in obese humans are not associated with colonic inflammation or disturbances of barrier function. *Aliment. Pharmacol. Ther.* **32**, 1307–1314 (2010).
59. Reigstad, C. S., Lunden, G. O., Felin, J. & Bäckhed, F. Regulation of serum amyloid A3 (SAA3) in mouse colonic epithelium and adipose tissue by the intestinal microbiota. *Plos ONE* **4**, e5842 (2009).
60. Scheja, L. *et al.* Acute-phase serum amyloid A as a marker of insulin resistance in mice. *Exp. Diabetes Res.* **2008**, 230837 (2008).
61. Kalliomaki, M., Collado, M. C., Salminen, S. & Isolauri, E. Early differences in fecal microbiota composition in children may predict overweight. *Am. J. Clin. Nutr.* **87**, 534–538 (2008).
62. Collado, M. C., Isolauri, E., Laitinen, K. & Salminen, S. Effect of mother's weight on infant's microbiota acquisition, composition, and activity during early infancy: a prospective follow-up study initiated in early pregnancy. *Am. J. Clin. Nutr.* **92**, 1023–1030 (2010).
63. Wu, X. *et al.* Molecular characterisation of the faecal microbiota in patients with type II diabetes. *Curr. Microbiol.* **61**, 69–78 (2010).
64. Cani, P. D., Dewever, C. & Delzenne, N. M. Inulin-type fructans modulate gastrointestinal peptides involved in appetite regulation (glucagon-like peptide-1 and ghrelin) in rats. *Br. J. Nutr.* **92**, 521–526 (2004).
65. Cani, P. D., Neyrinck, A. M., Maton, N. & Delzenne, N. M. Oligofructose promotes satiety in rats fed a high-fat diet: involvement of glucagon-like peptide-1. *Obes. Res.* **13**, 1000–1007 (2005).
66. Cani, P. D., Hoste, S., Guiot, Y. & Delzenne, N. M. Dietary non-digestible carbohydrates promote L-cell differentiation in the proximal colon of rats. *Br. J. Nutr.* **98**, 32–37 (2007).
67. Cani, P. D. *et al.* Improvement of glucose tolerance and hepatic insulin sensitivity by oligofructose requires a functional glucagon-like peptide 1 receptor. *Diabetes* **55**, 1484–1490 (2006).
68. Cani, P. D. *et al.* Gut microbiota fermentation of prebiotics increases satietogenic and incretin gut peptide production with consequences for appetite sensation and glucose response after a meal. *Am. J. Clin. Nutr.* **90**, 1236–1243 (2009).
69. Cani, P. D., Joly, E., Horsmans, Y. & Delzenne, N. M. Oligofructose promotes satiety in healthy human: a pilot study. *Eur. J. Clin. Nutr.* **60**, 567–572 (2006).
70. Aronsson, L. *et al.* Decreased fat storage by Lactobacillus paracasei is associated with increased levels of angiopoietin-like 4 protein (ANGPTL4). *Plos ONE* **5**, (2010).
71. Ehrlich, S. D. Probiotics—little evidence for a link to obesity. *Nat. Rev. Microbiol.* **7**, 901 (2009).
72. Delzenne, N. & Reid, G. No causal link between obesity and probiotics. *Nat. Rev. Microbiol.* **7**, 901 (2009).
73. Raoult, D. Human microbiome: take-home lesson on growth promoters? *Nature* **454**, 690–691 (2008).
74. Stsepetova, J. *et al.* Diversity and metabolic impact of intestinal Lactobacillus species in healthy adults and the elderly. *Br. J. Nutr.* **105**, 1235–1244 (2011).
75. Andreasen, A. S. *et al.* Effects of Lactobacillus acidophilus NCFM on insulin sensitivity and the systemic inflammatory response in human subjects. *Br. J. Nutr.* **104**, 1831–1838 (2010).
76. Kadooka, Y. *et al.* Regulation of abdominal adiposity by probiotics (Lactobacillus gasseri SBT2055) in adults with obese tendencies in a randomized controlled trial. *Eur. J. Clin. Nutr.* **64**, 636–643 (2010).
77. Lu, Z. X., Walker, K. Z., Muir, J. G. & O'Dea, K. Arabinoxylan fibre improves metabolic control in people with type II diabetes. *Eur. J. Clin. Nutr.* **58**, 621–628 (2004).
78. Garcia, A. L. *et al.* Arabinoxylan fibre consumption improved glucose metabolism, but did not affect serum adipokines in subjects with impaired glucose tolerance. *Horm. Metab. Res.* **38**, 761–766 (2006).
79. Garcia, A. L. *et al.* Arabinoxylan consumption decreases postprandial serum glucose, serum insulin and plasma total ghrelin response in subjects with impaired glucose tolerance. *Eur. J. Clin. Nutr.* **61**, 334–341 (2007).
80. Parnell, J. A. & Reimer, R. A. Weight loss during oligofructose supplementation is associated with decreased ghrelin and increased peptide YY in overweight and obese adults. *Am. J. Clin. Nutr.* **89**, 1751–1759 (2009).
81. Luo, J. *et al.* Chronic consumption of short-chain fructooligosaccharides does not affect basal hepatic glucose production or insulin resistance in type 2 diabetics. *J. Nutr.* **130**, 1572–1577 (2000).
82. Daubioul, C. A., Horsmans, Y., Lambert, P., Danse, E. & Delzenne, N. M. Effects of oligofructose on glucose and lipid metabolism in patients with nonalcoholic steatohepatitis: results of a pilot study. *Eur. J. Clin. Nutr.* **59**, 723–726 (2005).
83. Neyrinck, A. M. & Delzenne, N. M. Potential interest of gut microbial changes induced by non-digestible carbohydrates of wheat in the management of obesity and related disorders. *Curr. Opin. Clin. Nutr. Metab. Care* **13**, 722–728 (2010).

Author contributions

All authors contributed equally to all aspects of the article.