OPEN

Clinical & Translational Immunology (2016) 5, e82; doi:10.1038/cti.2016.29

www.nature.com/cti

REVIEW

Dietary metabolites and the gut microbiota: an alternative approach to control inflammatory and autoimmune diseases

James L Richards, Yu Anne Yap, Keiran H McLeod, Charles R Mackay and Eliana Mariño

It is now convincingly clear that diet is one of the most influential lifestyle factors contributing to the rise of inflammatory diseases and autoimmunity in both developed and developing countries. In addition, the modern 'Western diet' has changed in recent years with increased caloric intake, and changes in the relative amounts of dietary components, including lower fibre and higher levels of fat and poor quality of carbohydrates. Diet shapes large-bowel microbial ecology, and this may be highly relevant to human diseases, as changes in the gut microbiota composition are associated with many inflammatory diseases. Recent studies have demonstrated a remarkable role for diet, the gut microbiota and their metabolites—the short-chain fatty acids (SCFAs)—in the pathogenesis of several inflammatory diseases, such as asthma, arthritis, inflammatory bowel disease, colon cancer and wound-healing. This review summarizes how diet, microbiota and gut microbial metabolites (particularly SCFAs) can modulate the progression of inflammatory diseases and autoimmunity, and reveal the molecular mechanisms (metabolite-sensing G protein-coupled receptor (GPCRs) and inhibition of histone deacetylases (HDACs)). Therefore, considerable benefit could be achieved simply through the use of diet, probiotics and metabolites for the prevention and treatment of inflammatory diseases and autoimmunity.
Clinical & Translational Immunology (2016) **5,** e82; doi:10.1038/cti.2016.29; published online 13 May 2016

Over the past few decades, the incidence of inflammatory and autoimmune conditions in Westernised nations has risen sharply.[1] Subsequently, the modern western diet is one environmental factor that has changed with increased overall caloric intake, and changes in the relative amounts of dietary components, including reduced intake of high-calibre nutrients in exchange for more refined and highly processed variants.[2] As such, diet-related inflammatory conditions such as obesity, type 2 diabetes (T2D), cardiovascular disease, chronic kidney disease and autoimmune diabetes (T1D) have become a stigma for Western society.[3–6] It is well established now that our diet influences our gut commensal bacteria or microbiota by creating a paradigm between beneficial and non-beneficial bacterial species.[7] On the other hand, research into what we eat and how it can affect our microbiota is in the early stages. In particular, consumption of dietary fibre and its effects on gut microbiota.[8] During fermentation of fibre, the microbiota produce metabolites or short-chain fatty acids (SCFAs), which can exert beneficial effects in health by maintaining the homeostasis of metabolic function, as well as having profound anti-inflammatory effects by modulating the development and priming of the immune system.[9] The strong anti-inflammatory effects by SCFAs may act via specific G protein-coupled receptors (GPCRs) and/or via inhibiting HDACs; these metabolites promote homeostasis of the gut epithelium, promoting a tightly controlled border between gut microbes and host.[10] Likewise, these metabolites can also influence the immune cells residing closely in the lymphoid compartments of the gut, or can circulate systemically to affect those in peripheral tissues. Here, we provide an overview of the dietary influence on gut microbiota, and how the microbial metabolites produced can alter the outcome of inflammation and autoimmunity. We also discuss dietary SCFA approaches that can be employed to block inflammatory pathways and prevent or treat inflammatory diseases and autoimmunity.

SCFAS IN THE PARADIGM OF GOOD AND BAD NUTRIENTS

Our diet is composed of a variety of dietary macronutrients—carbohydrates, proteins, fats and fibres. Changes in those nutritional components can act as priming triggers for autoimmunity,[11,12] whereas the overconsumption of others can lead to cell damage and inflammation.[13] For instance, the amount of fibre and fat in the diet shapes large-bowel microbial ecology[14] that has been associated with many inflammatory diseases.[15] This is in line with a study showing that consumption of dietary fibre has globally declined below the recommended daily intake, particularly in Westernised societies.[16] Meanwhile, in Mediterranean societies where high intake of fibre from vegetables, fruits and nuts is preferred to intake of highly processed meats and industrialised goods, diet-associated

Department of Biochemistry and Molecular Biology, Monash University, Clayton, Victoria, Australia
Correspondence: Dr E Marino, Department of Biochemistry and Molecular Biology, Monash University, 15 Innovation Walk, Building 75, Clayton, Melbourne, Victoria 3800, Australia.
E-mail: eliana.marino@monash.edu
Received 20 February 2016; revised 4 April 2016; accepted 4 April 2016

complications, such as cardiovascular diseases, have considerably low prevalence.[17] But why is fibre so important? Foods high in fibre provide many health benefits, as it becomes the source of energy for both our own gut cells and the microbial communities that reside there in symbiosis.[18] Industrialised diets might deeply alter the gut microbiota and affect beneficial microbes and their effects on gut, immune and metabolic homeostasis[7,19,20] —a topic that we are going to discuss later throughout this review. The importance of diet and its effects on the gut microbiota are reflected in a recent study showing changes in microbiota diversity through evolution in people following ancestral lifestyles relative to Westernized societies,[21] indicating that changes in the gut microbiota critically shape human biology. Recent studies have shown that resistant starches mediate many of the effects ascribed to fibre, and their supply is critical for optimal gut function.[22] Resistant starches that can be obtained from vegetable, fruits, wheat, corn and nuts are one such form of dietary fibre. They are aptly named because of their strong ability to resist degradation by the body's digestive processes, which continues through to the caecum and large intestine, where they are fermented by the gut microbiota.[22] This property of resistant starches is often utilised in commercial foods to reduce energy density because of the inability of the human body to digest them. In the mammalian gut, primarily the colon, resistant starches are degraded and fermented by gut microbiota that subsequently produce metabolites, the most prominent being SCFA: acetate (two carbons), propionate (three carbons) and butyrate (four carbons).[22] These metabolites are produced at varying ratios, with acetate being the most abundant in the colon (~60%), followed by propionate (~25%) and then, to a much lesser degree, by butyrate (~15%).[23] In addition, acetate may itself fuel the production of fellow SCFA such as butyrate via alternate biochemical pathways. More than 95% of SCFAs are absorbed by the colon, with butyrate being the preferential energy source for colonocytes, as well as having a profound effect on maintaining gut epithelial homeostasis and function.[22]

THE GUT: THE ORIGIN OF INFLAMMATORY DISEASES

A 'leaky gut' in humans and mice, referring to increased gut permeability, disturbed microbial balance and impaired mucosal immunity, has been linked as the preceding step to the initiation of inflammatory diseases and autoimmunity. This is possibly because alteration in microbial ecology and decreased production of SCFAs altered mechanisms of mucosal barrier function.[24,25] For instance, the gut epithelial layer acts as a barrier, preventing the translocation of gut bacteria that can become pathogenic once they reach other organs.[26–29] The SCFA acetate produced by intestinal bacteria reduces gut mucosal permeability.[30] This study inferred that acetate production could be one of the principal features of probiotic bacteria that are thought to provide immune benefits and protection against certain pathogens. A 'leaky' intestinal mucosal barrier underpins the breakdown of immune tolerance and leads to intestinal inflammation and diseases, including coeliac disease, colorectal cancer, allergies, asthma, chronic kidney disease, as well as autoimmune T1D.[1,31,32]

In murine models, colonic epithelial cells can suffer DNA damage from harmful dietary by-products, such as those generated from protein fermentation, which alarmingly can lead to colon cancer.[33] Clarke *et al.* observed that rats with azoxymethane-induced colorectal cancer that were fed diets high in resistant starches have a significantly reduced number of tumour formations compared with rats fed control diets with highly digestible starches.[34] Interestingly, increasing butyrate concentrations in the caecum, as well as in the proximal and distal colon, were negatively associated with tumour formation in the large bowel. In addition, Conlon *et al.* identified an inverse relationship between increased caecal butyrate concentrations and the amount of DNA single-strand breakage in colonocytes.[35] Consequently, epithelial cells treated with butyrate regain gut motility and have reduced intestinal permeability.[36] The diet-derived microbial metabolites accumulating in the gut environment interact with epithelial and immune cells via specific receptors to modulate their respective molecular pathways. Targets of these metabolites include specific GPCRs, which bind free fatty acids such as SCFAs. The effect of metabolite interaction with GPCRs can significantly influence mucosal and immune homeostasis.

GPCRS AND MECHANISMS OF ACTION IN THE GUT

Over the years, a vast number of GPCRs have been identified, some currently with unknown ligands or function; however, only a select few have been characterised as molecular sensors of diet-related microbial products. Of particular interest to this review are the receptors of SCFAs, namely GPR43 (FFA2), GPR41 (FFA3) and GPR109a. GPR43 is activated by SCFAs with varying potency—acetate > propionate > butyrate. Expression of GPR43 has been found on gut epithelial cells and certain immune cells.[37] Similarly, GPR41 also binds the three major SCFAs, but with differing affinities.[38] GPR109a is primarily activated by both niacin and butyrate ligands. Whereas under normal physiological conditions niacin levels are not high enough to activate the receptor, levels of butyrate, obtained from the gut environment, and its oxidised form β-hydroxybutyrate, are sufficient to stimulate a response.[39] Expression of GPR109a has been found on a variety of immune cells, as well as on adipocytes, hepatocytes, gut and retinal epithelium, vascular endothelium and neuronal tissue.[39] Owing to its connection to the NF-κB pathway, GPR109a activation can lead to suppression of pro-inflammatory mediators such as iNOS, COX2, tumour necrosis factor (TNF)-α, interleukin (IL)-1β and IL-6. Thus, focus on GPR109a as a therapeutic target to treat inflammatory diseases has been growing.

In patients suffering from colitis, a form of inflammatory bowel disease, experimental treatment with butyrate enemas has reduced clinical signs of inflammation and even led to remission in some cases.[40] As butyrate potently activates GPR109a, many studies have focused on the effects of GPR109a activation in treating inflammatory conditions of the bowel. Colonic epithelial cells from neonatal mice cultured with butyrate *in vitro* generate an enhanced production of mRNA for anti-inflammatory IL-18; yet colonic epithelial cells from mice lacking GPR109a failed to have this upregulating response.[41] Adding to this, our group demonstrated recently that NLRP3 inflammasome activation in colonic epithelial cells required two signals.[42] First, the priming, induced by gut microbial products and second activation mediated by membrane hyperpolarization. Macia *et al.* showed that dietary fibre had beneficial effects on epithelial integrity by promoting epithelial NLRP3 activation through effects on both signals one and two. By reshaping the gut microbial composition, dietary fibre improved inflammasome priming. The SCFAs, released by anaerobic fermentation by colonic bacteria, activated the inflammasome through their binding to GPR43 and GPR109a.[42] This beneficial role on epithelial integrity was confirmed in a model of dextran sulphate sodium (DSS)-induced colitis *in vivo* in which the protective role of dietary fibre was mediated through NLRP3 activation in the epithelial compartment following GPCR activation.[42]

THE ROLE OF DIET, MICROBIAL METABOLITES AND MUCOSAL IMMUNOLOGY

Interactions with the external environment in vertebrates occur at various sites including the mucosal surfaces of airways, skin and the

gastrointestinal tract.[43] The gut, being the largest immunological organ in our body and in constant contact with food antigens, commensal microbiota and foreign pathogens, has to be extremely adept at innate and adaptive immune regulations.[44] In order to effectively manage these interfaces, the gut has evolved with a highly dynamic anatomy that interacts with the resident microbiota and the mucosal immune system.[45] The gut mucosal immune system consists of three distinct mucosal lymphoid structures: the mucosa-associated lymphoid tissue found in the gastrointestinal tract either in clusters (Peyer's patches) or in isolated lymphoid tissue, the lamina propria where cytokines and immunoglobulins are secreted by effector cells and the epithelium layer in which intraepithelial lymphocytes reside.[43] Another distinct population of immune cells named the innate lymphoid cells (ILCs) is essential for the maintenance of intestinal homeostasis.[47] The ILCs are responsive to microbial composition, and their development and function depends on the specific expression of transcription factors: T-bet for Group 1 ILCs, GATA-3 for Group 2 ILCs or RORγt for Group 3 ILCs.[48] Numerous studies have established a role for ILCs in maintaining a healthy intestinal barrier through the production and secretion of cytokines such as IL-22 and IL-17, or activity of the transcription factor aryl hydrocarbon receptor.[49–51] Whereas more research is needed to fully uncover the role of ILCs in regulating host–microbe interactions, it is clear that ILCs confer another level of protection to the epithelial cells from pathogenic exposure by repairing tissue damage, promoting gut barrier function and preventing systemic inflammation.

Recent studies have shown that SCFAs produced from bacterial fermentation of fibre have anti-inflammatory and immunomodulatory effects through the impact of regulatory T (T_{reg}) cells as an important factor in immune tolerance.[52] The SCFA butyrate promotes inducible T_{reg} (iT_{reg}) number and function in the colon of mice.[53,54] IL-10-producing iT_{regs} develop after TGF-β cytokine exposure in the periphery from naive $CD4^+$ T cells.[53] In addition, adoptive transfer of $CD4^+$ $CD45RB^{hi}$ naive T cells into *Rag1*$^{-/-}$ mice showed their conversion into T_{reg} cells when mice were fed a butyrylated diet.[53] Indeed, it is likely that commensal bacterial species that promote iT_{regs} in the gut[55] do so through production of high amounts of acetate or butyrate. In parallel, Smith *et al.* further expanded on this finding by demonstrating a direct effect of SCFA on colonic T_{regs} through the increased expression of GPR43 mRNA; however, this effect was absent in mice deficient in GPR43.[54]

Importantly, the actions of SCFAs are not limited to intestinal sites. A portion of diet-derived microbial metabolites passes across the mucosa into the lamina propria, where it enters the systemic circulation via the portal vein. Whereas butyrate enacts strong effects in the gut, its levels in circulation throughout the body are often negligible or undetectable. Acetate, the most abundantly produced SCFA, is, however, readily detectible in the peripheral circulation at ~ 50–150 μM. Therefore, the SCFA acetate is one means by which the microbiota may regulate the immune system beyond the gut.

'YOU ARE WHAT YOU EAT': THE ROLE OF MICROBIAL SCFAS IN T2D AND DIABETIC COMPLICATIONS

The Western diet underlies obesity, T2D, as well as asthma and cancer.[56–58] All these conditions are elements of the metabolic disorders, where diet contributes to the chronic inflammation of visceral adipose tissue, insulin resistance and increased intestinal permeability,[59] allowing dissemination of gut bacteria or bacterial products (endotoxaemia). Genetically obese mice had increased intestinal permeability and lipopolysaccharide (LPS) levels in the portal blood, which promote inflammatory liver damage.[60] This is evidenced by the increased levels of TNF-α and reduced zona occludens 1 mRNA in the proximal colon of obese C57BL/6J mice, which correlated with increased macrophage infiltration and levels of inflammatory cytokines TNF-α and IL-6 in the mesenteric fat.[61] In contrast, the gut anti-inflammatory agent 5-aminosalicyclic acid was shown to improve metabolic parameters in diet-induced obesity (DIO) mice, with associated regulation of gut adaptive immunity and reduced gut permeability,[62] thus implicating the role of gut leakiness and inflammation in DIO mice. A similar link between obesity-induced abnormalities in lipid homeostasis, gut permeability and non-alcoholic steatohepatitis was also found in human subjects,[63] similar to increased circulating zonulin and IL-6 in obesity-associated insulin resistance.[64] Two groups[65,66] have demonstrated that induction of IL-22 produced by Group 3 ILCs is impaired in obese mice, and IL-22-deficient mice fed a high-fat diet are more susceptible to developing metabolic disorders. Lymphoid tissue-inducer cells secrete large amounts of IL-22 that maintains gut mucosal barrier integrity and keeps the host–microbial balance.[66,67] In addition, IL-22 is involved in the recruitment of B cells and other lymphocytes to the germinal centres of isolated lymphoid tissues important for pathogen clearance,[68] in line with the theory of endotoxaemia induced by inflammation and increased intestinal permeability. The effects of IL-22 extend beyond the gut as IL-22 is a natural regulator of beta-cell insulin biosynthesis and secretion, protecting beta-cells from stress and preventing insulin hypersecretion, ultimately suppressing islet inflammation in obesity.[65] Administration of exogenous IL-22 to db/db or DIO mice improves obesity-driven insulin sensitivity and gut barrier dysfunction, and reduces chronic inflammation in the liver and adipose tissues.[66]

The potential link between gut microbiota and the obese phenotype was established a decade ago.[69] Since then obese mice treated with prebiotics selectively increased *Bifidobacterium* and showed a decrease in concentrations of LPS and inflammatory cytokines in blood, and this associated with improvements in gut barrier function.[70] Faecal microbiota transferred to germ-free mice from mothers with gestational diabetes induced increased adiposity and insulin sensitivity,[19] thus demonstrating the association between human metabolic disorders and altered microbiota composition. More recently, a study assessing the role of drug effect on gut microbiota of T2D subjects showed that the T2D subjects lacking butyrate-producing gut bacteria could be restored following treatment with metformin, an antidiabetic therapy, suggesting a role for SCFA-producing microbes in disease and health.[71] In addition, SCFAs stimulate the release of the gut hormone glucagon-like-peptide-1 and 2 (GLP-1 and GLP-2),[72] which is responsible for modulating gut barrier function and reducing uptake of inflammatory compounds that may trigger the chronic low-grade inflammation often linked with obesity and cardiovascular disease. Indeed, prebiotic-treated mice show an increased GLP-2 production associated with lower plasma LPS levels and oxidative stress markers.[70] The SCFA acetate has also been demonstrated to regulate production of leptin, an adipose-based hormone crucial for regulating energy homeostasis.[73] Some studies have elucidated the roles of SCFAs and GPCRs and production of leptin *in vitro* and *in vivo*.[74,75] Although concentrations of propionate in serum are quite low or undetectable, treatment of adipose tissue explants with propionate significantly downregulated the production of TNF-α and CCL5 by macrophages, and increased the expression of lipoprotein lipase and GLUT4 (associated with lipogenesis and glucose uptake).[76] Similarly, acetate and propionate stimulated adipogenesis through GPR43.[75,77] Meanwhile, GPR109a promotes lipolysis, as niacin treatment in mice deficient in GPR109a fails to increase the

secretion of adiponectin.[78] In contrast, Tang *et al.* showed that mice with DIO and T2D displayed increased plasma acetate in correlation with higher expression of GPR43 and GPR41 in the islets, and this contributed to compromised capacity of beta-cells to respond to hyperglyceamia.[79] This is in line with increased local glucose-dependent acetate formation by pancreatic islets, also seen in people with diabetes independent of fibre intake.[80] However, several studies show inconsistent results using GPR41- or GPR43-deficient mice.[81–85] Given SCFAs modulate immune responses,[53] the extent to which diet and the gut microbiota account for progression of metabolic syndrome through immune regulation is still poorly understood. Mathis and co-workers[86] showed that low-grade of inflammation in the adipose tissue correlates with reduced T_{reg} cell numbers with downregulated expression of gut-homing markers CD103 and GPR83.

In addition to metabolite-sensing GPCRs, SCFAs also exert activities through epigenetic effects, particularly the HDACs. HDACs regulate chromatin remodelling and gene expression, as well as the function of numerous transcription factors.[1] HDACs are a group of enzymes that remove acetyl groups from the histones that bind DNA.[87] Removing acetyl groups alters the binding of histones to DNA, which changes the expression patterns of different genes.[88] Through this activity, HDACs can have an important role in gene expression. In adipose tissue, a high-fat diet impairs adipogenic differentiation of C/EBPα, PPARγ, FABP4 and adiponectin associated with elevated expression of HDAC 9.[89] The pro-inflammatory obese state can also lead to the development of chronic kidney disease due to a 'leaky' intestinal mucosal barrier,[32] possibly because compromised epithelial integrity allows the dissemination of gut bacteria or bacterial products (endotoxaemia) resulting in kidney damage.[90] Feeding mice with high-fibre diets prompted a reduction in markers of kidney damage including serum concentration of creatinine and urea.[91] Similarly, inhibition of HDAC activity by acetate led to reduced DNA methylation in kidney tissue.[92] Epigenetic modifications are essential for development and proper functioning of the kidney, as they modulate TGFβ signalling, inflammation, profibrotic genes and the epithelial-to-mesenchymal transition, promoting renal fibrosis and progression of chronic kidney disease.[93] As such, HDACs have been shown to have integral roles in the regulation and activity of different immune cells.[94] In leukocyte cells, such as macrophages, neutrophils and eosinophils, HDACs have been linked to controlling cell survival and proliferation, as well as the regulation of cytotoxicity.[95] In B cells, HDACs have been shown to be important for inducing the apoptosis of proliferating cells.[96] HDACs are also important for promoting $CD8^+$ T cells, particularly in regards to increased function and differentiation.[97,98] Besides influencing immune cell survival, HDACs have also been linked to the suppression of cytokine production, having a role in controlling the inflammatory response.[99] HDACs are a very important part of immune regulation, both in promoting and regulating the immune system, and are a potential target for microbial metabolites in influencing the immune system.

DIET, SCFAS AND AUTOIMMUNE CONDITIONS

Impairments in gut barrier function have also been implicated as contributors to autoimmune diseases. Studies into such diseases, including T1D and certain variants of inflammatory bowel disease, emphasise not only genetic factors but also environmental and dietary factors.[31] Twelve-week-old non-obese diabetic (NOD) mice that are pre-diabetic exhibit increased intestinal permeability and, when infected with a bacterial pathogen *Citrobacter rodentium*, show increased activation and proliferation of diabetogenic $CD8^+$ T cells, which accelerate the onset of insulitis.[100] Increased intestinal permeability is associated with clinical diagnoses of T1D,[101] with a link between serum zonulin levels and development of T1D in patients and their relatives.[102] Moreover, diabetes-prone BioBreeding rats fed with hydrolysed casein diet reduced disease incidence by 50%, correlating with decreased lactulose:mannitol ratio and serum zonulin levels, indicative of a tighter intestinal barrier.[103]

Variances in gut microbiota in children diagnosed with T1D, although conflicting, have been widely examined. Children who develop T1D have a less diverse gut microbiota with a decreased presence of *Firmicutes* phylum correlated with decreased fecal butyrate than children with no T1D that presented an increase in *Bacteroidetes* phylum.[104,105] In line with these findings, NOD mice deficient in the adaptor protein myeloid differentiation factor 88 (MyD88), important for the detection of microbial antigens, fail to develop T1D under SPF conditions; yet germ-free conditions lead to an exacerbated development of T1D.[106] In addition, a following study by Markle *et al.* demonstrated the role of the gut microbiota in the marked gender differences that characterise T1D in NOD mice.[107] Similar to humans, male NOD mice display a considerably delayed onset and a reduced incidence of T1D. Remarkably, the female cohort gavaged with male gut microbiota were protected from T1D development, in comparison with female cohorts gavaged with a female gut microbiota or left untreated, which displayed typical disease incidence.[107] Treatment of NOD mice with probiotics coincides with maintenance of beta-cell function and prevention of T1D,[108] and probiotic treatment in genetically susceptible children for the prevention of T1D is currently the focus of the ongoing PRODIA study in Finland.[109] Whereas these studies provide compelling evidence for the role of gut microbiota in modulating T1D development, the specific metabolites responsible for preventing or ameliorating the diabetic immune response remain to be identified.

As alluded to throughout this review, SCFAs may have a major role in prevention of autoimmune diseases, and may underlie at least some microbiota-related associations with human disease. We have shown that SCFAs from the mother cross the placenta and protect against inflammatory asthma in offspring through epigenetic imprinting, mediating changes in gene transcription such as Foxp3 target genes important for tolerance/autoimmunity.[56] Foxp3 is a transcription factor necessary for T_{reg} development and function. SCFAs produced from bacterial fermentation of fibre not only promote iT_{reg} number and function in the colon[53,54] but also induce the promotion of extrathymic generation of T_{regs} via epigenetic effects.[110] This, in turn, allows T_{regs} to better control autoreactive lymphocytes and prevent the development of autoimmune disease.[111] For instance, epigenetic alterations such as histone modifications of the *FOXP3* locus are important for proper Foxp3 expression and the functional activity of $T_{regs.}$[112] Foxp3 also epigenetically modulates transcriptional activity of target gene loci by altering DNA methylation, transcription factor associations and histone modifications. These include the histone acetyltransferases Tip60 and p300 and the HDAC HDAC7.[113,114] T_{regs}, driven by the Foxp3 transcription factor, are particularly important for limiting autoimmunity and chronic inflammation.[114,115]

SCFAs may also exert effects directly on autoreactive cells. B cells have been implicated in the pathogenesis of certain inflammatory diseases, including T1D and lupus, because of their ability to produce autoantibodies, as well as cross present self-antigens to autoreactive T lymphocytes.[116,117] In a mouse model of lupus, treatment with butyrate and synthetic HDAC inhibitors led to the suppression of mechanisms that promote hypermutated antibody responses and class-switching, which culminate in the generation of high-affinity autoantibodies.[118,119] Inhibition of HDACs to limit autoreactive B cells

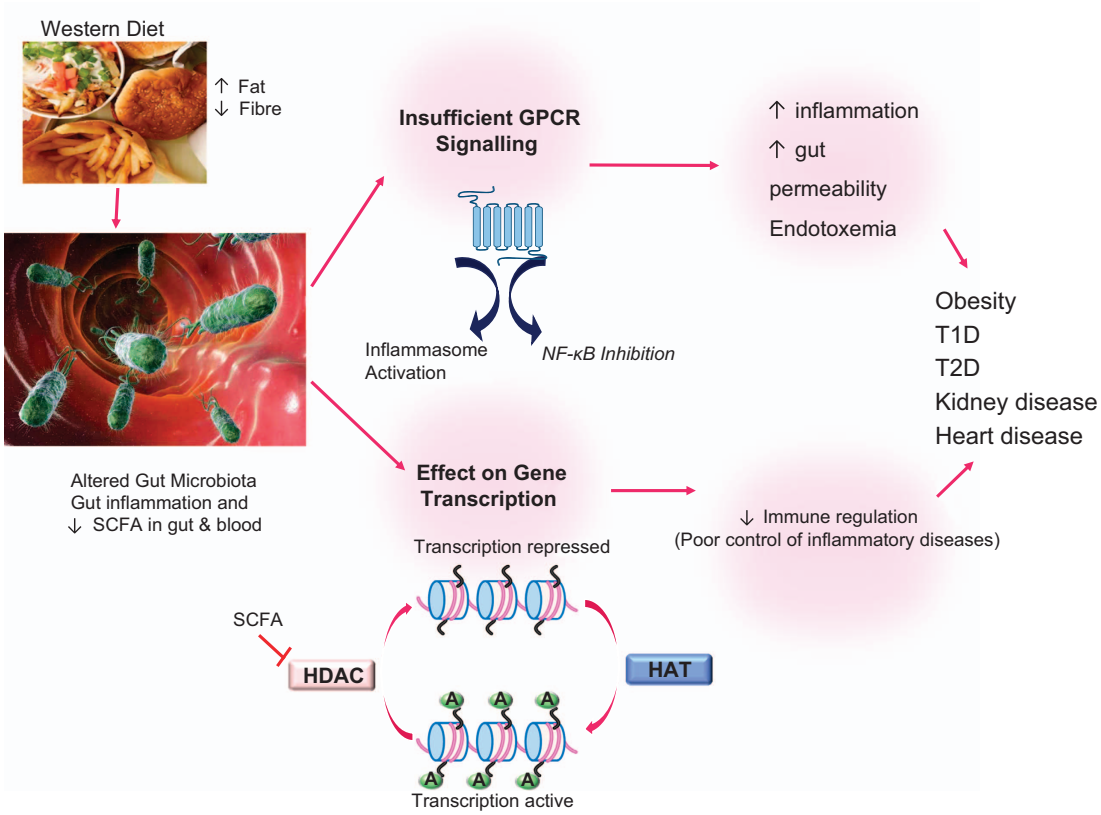

Figure 1 General model of how diet may be contributing to human inflammatory diseases such as obesity, T1D, T2D and kidney and cardiovascular diseases. Diet-induced changes to gut microbiota, and reduced production of SCFAs, lead to changed signalling through GPCRs, changes to gene transcription through HDAC effects and resulting changes to gut homeostasis, Treg biology and regulation of inflammation.

will likely be relevant to other inflammatory diseases such as T1D (our unpublished findings). A potential use for HDAC inhibitors to modify autoreactive B cells relates to individuals diagnosed with T1D, who also develop Celiac disease. Celiac disease is an autoimmune condition involving the inflammation of the small intestine, specifically in response to the presence of gluten food antigen. B cells and gluten-specific $CD4^+$ T cells from the intraepithelial lymphocyte compartment and lamina propria lead the inflammatory response. Apart from priming an immune response, studies in Balb/c and NOD mice have shown a 15% decrease in T_{reg} cell number in response to dietary gluten.[120] This effect is because of the overexpression of IL-15 in Celiac disease, which suppresses T_{reg} activation.[121] Owing to the highly regenerative ability of the small intestine, however, function can be recovered when individuals diagnosed with Celiac disease adhere to a strict gluten-free diet.[122]

GUT MICROBIOTA AND THEIR METABOLITES AS THERAPEUTICS

Targeting the gut microbiota is becoming a revolutionary therapy to correct metabolic dysfunction and inflammatory responses to treat diseases. It is now evident that the Western diets, possibly because of the lack of fibre, contribute not only to the loss of microbiota diversity but also promote an unbalance towards pathogenic gut bacteria associated with many inflammatory diseases. In a recent study, Sonnenburg *et al.*[123] demonstrated that humanised gnotobiotic mice on a Westernised diet (lacking fermentable carbohydrates) related with decreased gut microbiota diversity, particularly Clostridiales and Bacteroidales—predominant producers of SCFAs. Worryingly, feeding with this diet over subsequent generations led to the extinction of those bacterial phyla by the fourth generation, and the missing phyla could only be recovered via faecal microbiota transplant in combination with a diet high in fermentable carbohydrates. Thus, we believe that dietary SCFAs could be an excellent alternative approach to preventing or correcting the deterioration of western gut microbiota, given it is the safest and most cost-effective way to have an impact on the large global patient population.

Targeting the gut microbiota using therapies such as probiotics (treating the individual with healthy bacteria), prebiotics (treating the individual with nutrients to promote good bacteria) and the relatively crude fecal transplant has been largely studied.[124–126] So far, the outcomes from those methods are controversial or not so efficacious, given targeting specific components of the diets or a specific type of bacteria could be beneficial for some but detrimental for others. The advantages of using dietary fibre to target individual microbial metabolites relate to its properties of being highly resistant to human digestion and, therefore, directly modulating the whole microbiota community rather than individual bacteria species. Furthermore, this dietary manipulation of microbial metabolites can be used to naturally tailor a beneficial microbial ecology for each individual based on their personal gut microbial diversity. Figure 1 illustrates the mechanisms of action for the microbial SCFAs, that is, through metabolite-sensing GPCRs and/or HDAC epigenetic remodelling on epithelial cells and/or immune cells such as T_{regs}. One simple model is that reduced production of SCFAs through Western style diet, or antibiotic use and so on, contributes to altered microbial ecology and altered mucosal barrier function, resulting in exposure of the mucosal

immune system to bacteria or their products, which then in turn could affect immune tolerance. Therefore, targeting microbiota through dietary SCFAs could be a promising therapeutic approach to prevent or treat autoimmunity and inflammatory diseases associated with metabolic syndrome, where it has been observed that gut dysbiosis predates the development of the disease.

CONCLUDING REMARKS

In developing our understanding of how dietary components shape the overall panorama of the gut microbiome, and the subsequent metabolite profile, we can identify the likelihood of events leading to inflammation and autoimmunity. Emerging dietary treatments are not only economical, but also offer a non-invasive approach alternate to the risks of surgical procedures for chronic states of inflammation. Although it is in its early days, the implementation of diet and/or microbial metabolites or engineering the gut microbiota as a tool to prevent or treat inflammatory diseases is an exciting prospect that may have a great impact on human health.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

1 Thorburn AN, Macia L, Mackay CR. Diet, metabolites, and "western-lifestyle" inflammatory diseases. *Immunity* 2014; **40**: 833–842.
2 King DE, Mainous AG 3rd, Lambourne CA. Trends in dietary fiber intake in the United States, 1999-2008. *J Acad Nutr Diet* 2012; **112**: 642–648.
3 Cameron AJ, Welborn TA, Zimmet PZ, Dunstan DW, Owen N, Salmon J *et al.* Overweight and obesity in Australia: the 1999-2000 Australian Diabetes, Obesity and Lifestyle Study (AusDiab). *Med J Aust* 2003; **178**: 427–432.
4 Chow FY, Briganti EM, Kerr PG, Chadban SJ, Zimmet PZ, Atkins RC. Health-related quality of life in Australian adults with renal insufficiency: a population-based study. *Am J Kidney Dis* 2003; **41**: 596–604.
5 Kondrashova A, Reunanen A, Romanov A, Karvonen A, Viskari H, Vesikari T *et al.* A six-fold gradient in the incidence of type 1 diabetes at the eastern border of Finland. *Ann Med* 2005; **37**: 67–72.
6 Simpson RW, Shaw JE, Zimmet PZ. The prevention of type 2 diabetes–lifestyle change or pharmacotherapy? A challenge for the 21st century. *Diabet Res Clin Prac* 2003; **59**: 165–180.
7 Turnbaugh PJ, Hamady M, Yatsunenko T, Cantarel BL, Duncan A, Ley RE *et al.* A core gut microbiome in obese and lean twins. *Nature* 2009; **457**: 480–484.
8 De Filippo C, Cavalieri D, Di Paola M, Ramazzotti M, Poullet JB, Massart S *et al.* Impact of diet in shaping gut microbiota revealed by a comparative study in children from Europe and rural Africa. *Proc Natl Acad Sci USA* 2010; **107**: 14691–14696.
9 Macia L, Thorburn AN, Binge LC, Marino E, Rogers KE, Maslowski KM *et al.* Microbial influences on epithelial integrity and immune function as a basis for inflammatory diseases. *Immunol Rev* 2012; **245**: 164–176.
10 Tan J, McKenzie C, Potamitis M, Thorburn AN, Mackay CR, Macia L. The role of short-chain fatty acids in health and disease. *Adv Immunol* 2014; **121**: 91–119.
11 Funda DP, Kaas A, Tlaskalova-Hogenova H, Buschard K. Gluten-free but also gluten-enriched (gluten+) diet prevent diabetes in NOD mice; the gluten enigma in type 1 diabetes. *Diabetes Metab Res Rev* 2008; **24**: 59–63.
12 Lerner A, Matthias T. Changes in intestinal tight junction permeability associated with industrial food additives explain the rising incidence of autoimmune disease. *Autoimmun Rev* 2015; **14**: 479–489.
13 Chassaing B, Koren O, Goodrich JK, Poole AC, Srinivasan S, Ley RE *et al.* Dietary emulsifiers impact the mouse gut microbiota promoting colitis and metabolic syndrome. *Nature* 2015; **519**: 92–96.
14 Kau AL, Ahern PP, Griffin NW, Goodman AL, Gordon JI. Human nutrition, the gut microbiome and the immune system. *Nature* 2011; **474**: 327–336.
15 Clemente JC, Ursell LK, Parfrey LW, Knight R. The impact of the gut microbiota on human health: an integrative view. *Cell* 2012; **148**: 1258–1270.
16 Hartley L, May MD, Loveman E, Colquitt JL, Rees K. Dietary fibre for the primary prevention of cardiovascular disease. *Cochrane Database Syst Revi* 2016; **1**: Cd011472.
17 Estruch R, Ros E, Salas-Salvado J, Covas MI, Corella D, Aros F *et al.* Primary prevention of cardiovascular disease with a Mediterranean diet. *N Engl J Med* 2013; **368**: 1279–1290.
18 Bird AR, Brown IL, Topping DL. Starches, resistant starches, the gut microflora and human health. *Curr Issues Intest Microbiol* 2000; **1**: 25–37.
19 Koren O, Goodrich JK, Cullender TC, Spor A, Laitinen K, Backhed HK *et al.* Host remodeling of the gut microbiome and metabolic changes during pregnancy. *Cell* 2012; **150**: 470–480.
20 Turnbaugh PJ, Ley RE, Mahowald MA, Magrini V, Mardis ER, Gordon JI. An obesity-associated gut microbiome with increased capacity for energy harvest. *Nature* 2006; **444**: 1027–1031.
21 Clemente JC, Pehrsson EC, Blaser MJ, Sandhu K, Gao Z, Wang B *et al.* The microbiome of uncontacted Amerindians. *Sci Adv* 2015; **1**: e1500183.
22 Topping DL, Clifton PM. Short-chain fatty acids and human colonic function: roles of resistant starch and nonstarch polysaccharides. *Physiol Rev* 2001; **81**: 1031–1064.
23 Duncan SH, Holtrop G, Lobley GE, Calder AG, Stewart CS, Flint HJ. Contribution of acetate to butyrate formation by human faecal bacteria. *Br J Nutr* 2004; **91**: 915–923.
24 De Palma G, Nadal I, Medina M, Donat E, Ribes-Koninckx C, Calabuig M *et al.* Intestinal dysbiosis and reduced immunoglobulin-coated bacteria associated with coeliac disease in children. *BMC Microbiol* 2010; **10**: 63.
25 Russell WR, Duncan SH, Flint HJ. The gut microbial metabolome: modulation of cancer risk in obese individuals. *Proc Nutr Soc* 2013; **72**: 178–188.
26 Balmer ML, Slack E, de Gottardi A, Lawson MA, Hapfelmeier S, Miele L *et al.* The liver may act as a firewall mediating mutualism between the host and its gut commensal microbiota. *Sci Tansl Med* 2014; **6**: 237ra266.
27 Cani PD, Amar J, Iglesias MA, Poggi M, Knauf C, Bastelica D *et al.* Metabolic endotoxemia initiates obesity and insulin resistance. *Diabetes* 2007; **56**: 1761–1772.
28 Diehl GE, Longman RS, Zhang JX, Breart B, Galan C, Cuesta A *et al.* Microbiota restricts trafficking of bacteria to mesenteric lymph nodes by CX(3)CR1(hi) cells. *Nature* 2013; **494**: 116–120.
29 Kirkland D, Benson A, Mirpuri J, Pifer R, Hou B, DeFranco AL *et al.* B cell-intrinsic MyD88 signaling prevents the lethal dissemination of commensal bacteria during colonic damage. *Immunity* 2012; **36**: 228–238.
30 Fukuda S, Toh H, Hase K, Oshima K, Nakanishi Y, Yoshimura K *et al. Bifidobacteria* can protect from enteropathogenic infection through production of acetate. *Nature* 2011; **469**: 543–547.
31 Vaarala O, Atkinson MA, Neu J. The "perfect storm" for type 1 diabetes: the complex interplay between intestinal microbiota, gut permeability, and mucosal immunity. *Diabetes* 2008; **57**: 2555–2562.
32 Vaziri ND, Yuan J, Norris K. Role of urea in intestinal barrier dysfunction and disruption of epithelial tight junction in chronic kidney disease. *Am J Nephrol* 2013; **37**: 1–6.
33 Toden S, Bird AR, Topping DL, Conlon MA. Resistant starch attenuates colonic DNA damage induced by higher dietary protein in rats. *Nutr Cancer* 2005; **51**: 45–51.
34 Clarke JM, Topping DL, Bird AR, Young GP, Cobiac L. Effects of high-amylose maize starch and butyrylated high-amylose maize starch on azoxymethane-induced intestinal cancer in rats. *Carcinogenesis* 2008; **29**: 2190–2194.
35 Conlon MA, Kerr CA, McSweeney CS, Dunne RA, Shaw JM, Kang S *et al.* Resistant starches protect against colonic DNA damage and alter microbiota and gene expression in rats fed a Western diet. *J Nutr* 2012; **142**: 832–840.
36 Canani RB, Costanzo MD, Leone L, Pedata M, Meli R, Calignano A. Potential beneficial effects of butyrate in intestinal and extraintestinal diseases. *World J Gastroenterol* 2011; **17**: 1519–1528.
37 Maslowski KM, Vieira AT, Ng A, Kranich J, Sierro F, Yu D *et al.* Regulation of inflammatory responses by gut microbiota and chemoattractant receptor GPR43. *Nature* 2009; **461**: 1282–1286.
38 Brown AJ, Goldsworthy SM, Barnes AA, Eilert MM, Tcheang L, Daniels D *et al.* The Orphan G protein-coupled receptors GPR41 and GPR43 are activated by propionate and other short chain carboxylic acids. *J Biol Chem* 2003; **278**: 11312–11319.
39 Elangovan S, Pathania R, Ramachandran S, Ananth S, Padia RN, Lan L *et al.* The niacin/butyrate receptor GPR109A suppresses mammary tumorigenesis by inhibiting cell survival. *Cancer Res* 2014; **74**: 1166–1178.
40 Scheppach W, Sommer H, Kirchner T, Paganelli GM, Bartram P, Christl S *et al.* Effect of butyrate enemas on the colonic mucosa in distal ulcerative colitis. *Gastroenterology* 1992; **103**: 51–56.
41 Singh N, Gurav A, Sivaprakasam S, Brady E, Padia R, Shi H *et al.* Activation of gpr109a, receptor for niacin and the commensal metabolite butyrate, suppresses colonic inflammation and carcinogenesis. *Immunity* 2014; **40**: 128–139.
42 Macia L, Tan J, Vieira AT, Leach K, Stanley D, Luong S *et al.* Metabolite-sensing receptors GPR43 and GPR109A facilitate dietary fibre-induced gut homeostasis through regulation of the inflammasome. *Nat Commun* 2015; **6**: 6734.
43 Turner JR. Intestinal mucosal barrier function in health and disease. *Nat Rev Immunol* 2009; **9**: 799–809.
44 Flach M, Diefenbach A. Development of Gut-Associated Lymphoid Tissues. In: Mestecky V *et al* (eds). *Mucosal Immunology*. 4th edn, vol. 1-2, Academic press, 2015, pp 31-42.
45 Lepage P, Hasler R, Spehlmann ME, Rehman A, Zvirbliene A, Begun A *et al.* Twin study indicates loss of interaction between microbiota and mucosa of patients with ulcerative colitis. *Gastroenterology* 2011; **141**: 227–236.
46 Gibbons DL, Spencer J. Mouse and human intestinal immunity: same ballpark, different players; Different rules, same score. *Mucosal Immunol* 2011; **4**: 148–157.
47 Brown EM, Sadarangani M, Finlay BB. The role of the immune system in governing host-microbe interactions in the intestine. *Nat Immunol* 2013; **14**: 660–667.
48 Sonnenberg G, Artis D. Innate lymphoid cell interactions with microbiota: implications for intestinal health and disease. *Immunity* 2012; **37**: 601–610.
49 Huber S, Gagliani N, Zenewicz LA, Huber FJ, Bosurgi L, Hu B *et al.* IL-22BP is regulated by the inflammasome and modulates tumorigenesis in the intestine. *Nature* 2012; **491**: 259–263.
50 Quintana FJ, Basso AS, Iglesias AH, Korn T, Farez MF, Bettelli E *et al.* Control of T(reg) and T(H)17 cell differentiation by the aryl hydrocarbon receptor. *Nature* 2008; **453**: 65–71.

51 Sonnenberg GF, Monticelli LA, Alenghat T, Fung TC, Hutnick NA, Kunisawa J *et al.* Innate lymphoid cells promote anatomical containment of lymphoid-resident commensal bacteria. *Science* 2012; **336**: 1321–1325.
52 Bollrath J, Powrie F. Immunology. Feed your Tregs more fiber. *Science* 2013; **341**: 463–464.
53 Furusawa Y, Obata Y, Fukuda S, Endo TA, Nakato G, Takahashi D *et al.* Commensal microbe-derived butyrate induces the differentiation of colonic regulatory T cells. *Nature* 2013; **504**: 446–450.
54 Smith PM, Howitt MR, Panikov N, Michaud M, Gallini CA, Bohlooly YM *et al.* The microbial metabolites, short-chain fatty acids, regulate colonic Treg cell homeostasis. *Science* 2013; **341**: 569–573.
55 Geuking MB, Cahenzli J, Lawson MA, Ng DC, Slack E, Hapfelmeier S *et al.* Intestinal bacterial colonization induces mutualistic regulatory T cell responses. *Immunity* 2011; **34**: 794–806.
56 Thorburn AN, McKenzie CI, Shen S, Stanley D, Macia L, Mason LJ *et al.* Evidence that asthma is a developmental origin disease influenced by maternal diet and bacterial metabolites. *Nat Commun* 2015; **6**: 7320.
57 Turnbaugh PJ, Gordon JI. The core gut microbiome, energy balance and obesity. *J Physiol* 2009; **587**: 4153–4158.
58 Yoshimoto S, Loo TM, Atarashi K, Kanda H, Sato S, Oyadomari S *et al.* Obesity-induced gut microbial metabolite promotes liver cancer through senescence secretome. *Nature* 2013; **499**: 97–101.
59 Gummesson A, Carlsson LMS, Storlien LH, Bäckhed F, Lundin P, Löfgren L *et al.* Intestinal permeability is associated with visceral adiposity in healthy women. *Obesity* 2011; **19**: 2280–2282.
60 Brun P, Castagliuolo I, Di Leo V, Buda A, Pinzani M, Palù G *et al.* Increased intestinal permeability in obese mice: New evidence in the pathogenesis of nonalcoholic steatohepatitis. *Am J Physiol* 2007; **292**: G518–G525.
61 Lam YY, Ha CWY, Campbell CR, Mitchell AJ, Dinudom A, Oscarsson J *et al.* Increased gut permeability and microbiota change associate with mesenteric fat inflammation and metabolic dysfunction in diet-induced obese mice. *PLoS ONE* 2012; **7**: e34233.
62 Luck H, Tsai S, Chung J, Clemente-Casares X, Ghazarian M, Revelo XS *et al.* Regulation of obesity-related insulin resistance with gut anti-inflammatory agents. *Cell Metab* 2015; **21**: 527–542.
63 Farhadi A, Gundlapalli S, Shaikh M, Frantzides C, Harrell L, Kwasny MM *et al.* Susceptibility to gut leakiness: a possible mechanism for endotoxaemia in non-alcoholic steatohepatitis. *Liver Int* 2008; **28**: 1026–1033.
64 Moreno-Navarrete JM, Sabater M, Ortega F, Ricart W, Fernández-Real JM. Circulating zonulin, a marker of intestinal permeability, is increased in association with obesity-associated insulin resistance. *PLoS ONE* 2012; **7**: e37160.
65 Hasnain SZ, Borg DJ, Harcourt BE, Tong H, Sheng YH, Ng CP *et al.* Glycemic control in diabetes is restored by therapeutic manipulation of cytokines that regulate beta cell stress. *Nat Med* 2014; **20**: 1417–1426.
66 Wang X, Ota N, Manzanillo P, Kates L, Zavala-Solorio J, Eidenschenk C *et al.* Interleukin-22 alleviates metabolic disorders and restores mucosal immunity in diabetes. *Nature* 2014; **514**: 237–241.
67 Goto Y, Obata T, Kunisawa J, Sato S, Ivanov II, Lamichhane A *et al.* Innate lymphoid cells regulate intestinal epithelial cell glycosylation. *Science* 2014; **345**: 1254009.
68 Bouskra D, Brezillon C, Berard M, Werts C, Varona R, Boneca IG *et al.* Lymphoid tissue genesis induced by commensals through NOD1 regulates intestinal homeostasis. *Nature* 2008; **456**: 507–510.
69 Ley RE, Backhed F, Turnbaugh P, Lozupone CA, Knight RD, Gordon JI. Obesity alters gut microbial ecology. *Proc Natl Acad Sci USA* 2005; **102**: 11070–11075.
70 Cani PD, Possemiers S, Van de Wiele T, Guiot Y, Everard A, Rottier O *et al.* Changes in gut microbiota control inflammation in obese mice through a mechanism involving GLP-2-driven improvement of gut permeability. *Gut* 2009; **58**: 1091–1103.
71 Forslund K, Hildebrand F, Nielsen T, Falony G, Le Chatelier E, Sunagawa S *et al.* Disentangling type 2 diabetes and metformin treatment signatures in the human gut microbiota. *Nature* 2015; **528**: 262–266.
72 Tolhurst G, Heffron H, Lam YS, Parker HE, Habib AM, Diakogiannaki E *et al.* Short-chain fatty acids stimulate glucagon-like peptide-1 secretion via the G-protein-coupled receptor FFAR2. *Diabetes* 2012; **61**: 364–371.
73 Perry RJ, Zhang XM, Zhang D, Kumashiro N, Camporez JP, Cline GW *et al.* Leptin reverses diabetes by suppression of the hypothalamic-pituitary-adrenal axis. *Nat Med* 2014; **20**: 759–763.
74 Xiong Y, Miyamoto N, Shibata K, Valasek MA, Motoike T, Kedzierski RM *et al.* Short-chain fatty acids stimulate leptin production in adipocytes through the G protein-coupled receptor GPR41. *Proc Natl Acad Sci USA* 2004; **101**: 1045–1050.
75 Zaibi MS, Stocker CJ, O'Dowd J, Davies A, Bellahcene M, Cawthorne MA *et al.* Roles of GPR41 and GPR43 in leptin secretory responses of murine adipocytes to short chain fatty acids. *FEBS Lett* 2010; **584**: 2381–2386.
76 Al-Lahham S, Roelofsen H, Rezaee F, Weening D, Hoek A, Vonk R *et al.* Propionic acid affects immune status and metabolism in adipose tissue from overweight subjects. *Eur J Clin Invest* 2012; **42**: 357–364.
77 Hong YH, Nishimura Y, Hishikawa D, Tsuzuki H, Miyahara H, Gotoh C *et al.* Acetate and propionate short chain fatty acids stimulate adipogenesis via GPCR43. *Endocrinology* 2005; **146**: 5092–5099.
78 Plaisance EP, Lukasova M, Offermanns S, Zhang Y, Cao G, Judd RL. Niacin stimulates adiponectin secretion through the GPR109A receptor. *Am J Physiol Endocrinol Metab* 2009; **296**: E549–E558.
79 Tang C, Ahmed K, Gille A, Lu S, Grone HJ, Tunaru S *et al.* Loss of FFA2 and FFA3 increases insulin secretion and improves glucose tolerance in type 2 diabetes. *Nat Med* 2015; **21**: 173–177.
80 Todesco T, Zamboni M, Armellini F, Bissoli L, Turcato E, Piemonte G *et al.* Plasma acetate levels in a group of obese diabetic, obese normoglycemic, and control subjects and their relationships with other blood parameters. *Am J Gastroenterol* 1993; **88**: 751–755.
81 Bellahcene M, O'Dowd JF, Wargent ET, Zaibi MS, Hislop DC, Ngala RA *et al.* Male mice that lack the G-protein-coupled receptor GPR41 have low energy expenditure and increased body fat content. *Br J Nutr* 2013; **109**: 1755–1764.
82 Bjursell M, Admyre T, Goransson M, Marley AE, Smith DM, Oscarsson J *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* 2011; **300**: E211–E220.
83 Kimura I, Ozawa K, Inoue D, Imamura T, Kimura K, Maeda T *et al.* The gut microbiota suppresses insulin-mediated fat accumulation via the short-chain fatty acid receptor GPR43. *Nat Commun* 2013; **4**: 1829.
84 Lin HV, Frassetto A, Kowalik EJ Jr., Nawrocki AR, Lu MM, Kosinski JR *et al.* Butyrate and propionate protect against diet-induced obesity and regulate gut hormones via free fatty acid receptor 3-independent mechanisms. *PLoS ONE* 2012; **7**: e35240.
85 Samuel BS, Shaito A, Motoike T, Rey FE, Backhed F, Manchester JK *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* 2008; **105**: 16767–16772.
86 Feuerer M, Herrero L, Cipolletta D, Naaz A, Wong J, Nayer A *et al.* Lean, but not obese, fat is enriched for a unique population of regulatory T cells that affect metabolic parameters. *Nat Med* 2009; **15**: 930–939.
87 Mathias RA, Guise AJ, Cristea IM. Post-translational modifications regulate class IIa histone deacetylase (HDAC) function in health and disease. *Mol Cell Proteomics* 2015; **14**: 456–470.
88 De Ruijter AJM, Van Gennip AH, Caron HN, Kemp S, Van Kuilenburg ABP. Histone deacetylases (HDACs): Characterization of the classical HDAC family. *Biochem J* 2003; **370**: 737–749.
89 Chatterjee TK, Basford JE, Knoll E, Tong WS, Blanco V, Blomkalns AL *et al.* HDAC9 knockout mice are protected from adipose tissue dysfunction and systemic metabolic disease during high-fat feeding. *Diabetes* 2014; **63**: 176–187.
90 Ramezani A, Raj DS. The gut microbiome, kidney disease, and targeted interventions. *J Am Soc Nephrol* 2014; **25**: 657–670.
91 Kieffer DA, Piccolo BD, Vaziri ND, Liu S, Lau WL, Khazaeli M *et al.* Resistant starch alters gut microbiome and metabolomics profiles concurrent with amelioration of chronic kidney disease in rats. *Am J Physiol Renal Physiol* 2016; **310**: F857–F871.
92 Andrade-Oliveira V, Amano MT, Correa-Costa M, Castoldi A, Felizardo RJF, De Almeida DC *et al.* Gut bacteria products prevent AKI induced by ischemia-reperfusion. *J Am Soc Nephrol* 2015; **26**: 1877–1888.
93 Wing MR, Ramezani A, Gill HS, Devaney JM, Raj DS. Epigenetics of progression of chronic kidney disease: fact or fantasy? *Semin Nephrol* 2013; **33**: 363–374.
94 Falkenberg KJ, Johnstone RW. Histone deacetylases and their inhibitors in cancer, neurological diseases and immune disorders. *Nat Rev Drug Discov* 2014; **13**: 673–691.
95 Sweet MJ, Shakespear MR, Kamal NA, Fairlie DP. HDAC inhibitors: modulating leukocyte differentiation, survival, proliferation and inflammation. *Immunol Cell Biol* 2012; **90**: 14–22.
96 Yamaguchi T, Cubizolles F, Zhang Y, Reichert N, Kohler H, Seiser C *et al.* Histone deacetylases 1 and 2 act in concert to promote the G1-to-S progression. *Genes Dev* 2010; **24**: 455–469.
97 Fann M, Godlove JM, Catalfamo M, Wood Iii WH, Chrest FJ, Chun N *et al.* Histone acetylation is associated with differential gene expression in the rapid and robust memory CD8+ T-cell response. *Blood* 2006; **108**: 3363–3370.
98 Northrop JK, Wells AD, Shen H. Cutting edge: chromatin remodeling as a molecular basis for the enhanced functionality of memory CD8 T cells. *J Immunol* 2008; **181**: 865–868.
99 Villagra A, Sotomayor EM, Seto E. Histone deacetylases and the immunological network: implications in cancer and inflammation. *Oncogene* 2010; **29**: 157–173.
100 Lee AS, Gibson DL, Zhang Y, Sham HP, Vallance BA, Dutz JP. Gut barrier disruption by an enteric bacterial pathogen accelerates insulitis in NOD mice. *Diabetologia* 2010; **53**: 741–748.
101 Bosi E, Molteni L, Radaelli MG, Folini L, Fermo I, Bazzigaluppi E *et al.* Increased intestinal permeability precedes clinical onset of type 1 diabetes. *Diabetologia* 2006; **49**: 2824–2827.
102 Sapone A, de Magistris L, Pietzak M, Clemente MG, Tripathi A, Cucca F *et al.* Zonulin upregulation is associated with increased gut permeability in subjects with type 1 diabetes and their relatives. *Diabetes* 2006; **55**: 1443–1449.
103 Visser JTJ, Lammers K, Hoogendijk A, Boer MW, Brugman S, Beijer-Liefers S *et al.* Restoration of impaired intestinal barrier function by the hydrolysed casein diet contributes to the prevention of type 1 diabetes in the diabetes-prone BioBreeding rat. *Diabetologia* 2010; **53**: 2621–2628.
104 de Goffau MC, Luopajarvi K, Knip M, Ilonen J, Ruohtula T, Harkonen T *et al.* Fecal microbiota composition differs between children with beta-cell autoimmunity and those without. *Diabetes* 2013; **62**: 1238–1244.
105 Giongo A, Gano KA, Crabb DB, Mukherjee N, Novelo LL, Casella G *et al.* Toward defining the autoimmune microbiome for type 1 diabetes. *ISME J* 2011; **5**: 82–91.
106 Wen L, Ley RE, Volchkov PY, Stranges PB, Avanesyan L, Stonebraker AC *et al.* Innate immunity and intestinal microbiota in the development of Type 1 diabetes. *Nature* 2008; **455**: 1109–1113.

107 Markle JGM, Frank DN, Mortin-Toth S, Robertson CE, Feazel LM, Rolle-Kampczyk U *et al.* Sex differences in the gut microbiome drive hormone-dependent regulation of autoimmunity. *Science* 2013; **339**: 1084–1088.
108 Calcinaro F, Dionisi S, Marinaro M, Candeloro P, Bonato V, Marzotti S *et al.* Oral probiotic administration induces interleukin-10 production and prevents spontaneous autoimmune diabetes in the non-obese diabetic mouse. *Diabetologia* 2005; **48**: 1565–1575.
109 Ljungberg M, Korpela R, Ilonen J, Ludvigsson J, Vaarala O. Probiotics for the prevention of beta cell autoimmunity in children at genetic risk of type 1 diabetes–the PRODIA study. *Ann N Y Acad Sci* 2006; **1079**: 360–364.
110 Arpaia N, Campbell C, Fan X, Dikiy S, Van Der Veeken J, Deroos P *et al.* Metabolites produced by commensal bacteria promote peripheral regulatory T-cell generation. *Nature* 2013; **504**: 451–455.
111 Fontenot JD, Rudensky AY. A well adapted regulatory contrivance: regulatory T cell development and the forkhead family transcription factor Foxp3. *Nat Immunol* 2005; **6**: 331–337.
112 Zhang Y, Maksimovic J, Naselli G, Qian J, Chopin M, Blewitt ME *et al.* Genome-wide DNA methylation analysis identifies hypomethylated genes regulated by FOXP3 in human regulatory T cells. *Blood* 2013; **122**: 2823–2836.
113 Li B, Samanta A, Song X, Iacono KT, Bembas K, Tao R *et al.* FOXP3 interactions with histone acetyltransferase and class II histone deacetylases are required for repression. *Proc Natl Acad Sci USA* 2007; **104**: 4571–4576.
114 Tao R, de Zoeten EF, Ozkaynak E, Chen C, Wang L, Porrett PM *et al.* Deacetylase inhibition promotes the generation and function of regulatory T cells. *Nat Med* 2007; **13**: 1299–1307.
115 Vignali DA, Collison LW, Workman CJ. How regulatory T cells work. *Nat Rev* 2008; **8**: 523–532.
116 Mariño E, Batten M, Groom J, Walters S, Liuwantara D, Mackay F *et al.* Marginal-zone B-cells of nonobese diabetic mice expand with diabetes onset, invade the pancreatic lymph nodes, and present autoantigen to diabetogenic T-cells. *Diabetes* 2008; **57**: 395–404.
117 Marino E, Tan B, Binge L, Mackay CR, Grey ST. B-cell cross-presentation of autologous antigen precipitates diabetes. *Diabetes* 2012; **61**: 2893–2905.
118 Waibel M, Christiansen AJ, Hibbs ML, Shortt J, Jones SA, Simpson I *et al.* Manipulation of B-cell responses with histone deacetylase inhibitors. *Nat Commun* 2015; **6**: 6838.
119 White CA, Pone EJ, Lam T, Tat C, Hayama KL, Li G *et al.* Histone deacetylase inhibitors upregulate B cell microRNAs that silence AID and Blimp-1 expression for epigenetic modulation of antibody and autoantibody responses. *J Immunol* 2014; **193**: 5933–5950.
120 Ejsing-Duun M, Josephsen J, Aasted B, Buschard K, Hansen AK. Dietary gluten reduces the number of intestinal regulatory T cells in mice. *Scand J Immunol* 2008; **67**: 553–559.
121 Zanzi D, Stefanile R, Santagata S, Iaffaldano L, Iaquinto G, Giardullo N *et al.* IL-15 interferes with suppressive activity of intestinal regulatory T cells expanded in Celiac disease. *Am J Gastroenterol* 2011; **106**: 1308–1317.
122 Haines ML, Anderson RP, Gibson PR. Systematic review: the evidence base for long-term management of coeliac disease. *Alimentary Pharmacol Ther* 2008; **28**: 1042–1066.
123 Sonnenburg ED, Smits SA, Tikhonov M, Higginbottom SK, Wingreen NS, Sonnenburg JL. Diet-induced extinctions in the gut microbiota compound over generations. *Nature* 2016; **529**: 212–215.
124 Blümer N, Sel S, Virna S, Patrascan CC, Zimmermann S, Herz U *et al.* Perinatal maternal application of Lactobacillus rhamnosus GG suppresses allergic airway inflammation in mouse offspring. *Clin Exp Allergy* 2007; **37**: 348–357.
125 Khoruts A, Sadowsky MJ, Hamilton MJ. Development of fecal microbiota transplantation suitable for mainstream medicine. *Clin Gastroenterol Hepatol* 2015; **13**: 246–250.
126 Prescott SL. Early-life environmental determinants of allergic diseases and the wider pandemic of inflammatory noncommunicable diseases. *J Allergy Clin Immunol* 2013; **131**: 23–30.