Effects of Oral Glucose-Lowering Agents on Gut Microbiota and Microbial Metabolites

Dongmei Wang¹, Jieying Liu¹,², Liyuan Zhou¹, Qian Zhang¹, Ming Li¹ and Xinhua Xiao¹*

¹ Department of Endocrinology, National Health Commission (NHC) Key Laboratory of Endocrinology, Chinese Academy of Medical Sciences, Peking Union Medical College Hospital, Peking Union Medical College, Beijing, China, ² Department of Medical Research Center, Chinese Academy of Medical Sciences, Peking Union Medical College Hospital, Peking Union Medical College, Beijing, China

The current research and existing facts indicate that type 2 diabetes mellitus (T2DM) is characterized by gut microbiota dysbiosis and disturbed microbial metabolites. Oral glucose-lowering drugs are reported with pleiotropic beneficial effects, including not only a decrease in glucose level but also weight loss, antihypertension, anti-inflammatory, and cardiovascular protection, but the underlying mechanisms are still not clear. Evidence can be found showing that oral glucose-lowering drugs might modify the gut microbiome and thereby alter gastrointestinal metabolites to improve host health. Although the connections among gut microbial communities, microbial metabolites, and T2DM are complex, figuring out how antidiabetic agents shape the gut microbiome is vital for optimizing the treatment, meaningful for the instruction for probiotic therapy and gut microbiota transplantation in T2DM. In this review, we focused on the literatures in gut microbiota and its metabolite profile alterations beneficial from oral antidiabetic drugs, trying to provide implications for future study in the developing field of these drugs, such as combination therapies, pre- and probiotics intervention in T2DM, and subjects with pregestational diabetes and gestational diabetes mellitus.

Keywords: gut microbiota, microbial metabolites, T2DM, antidiabetic drugs, SCFA

INTRODUCTION

The International Diabetes Federation Diabetes Atlas 10th edition shows a continued global increase in diabetes prevalence, estimating that 537 million adults are living with diabetes worldwide, most of which is type 2 diabetes mellitus (T2DM) (1). T2DM is a metabolic disorder with multiple pathogenic factors, including genetic elements, sedentary behaviors, and overeating (2). Once without effective treatment, it might lead to a composite of microvascular or macrovascular complications, for instance chronic kidney disease, diabetic eye disease, and cardiovascular disease (CVD) (3). Differing from insulin-dependent type 1 diabetes mellitus, T2DM is closely interrelated with insulin resistance (IR) and strongly intertwined with obesity, non-alcoholic fatty liver disease, and metabolic syndrome (4). Nowadays, more than 10 types of medicines are approved by the USA Food and Drug Administration for the glycemic treatment (5). Thousands of clinical trials and basic research are proceeding worldwide for diabetes...
pharmacotherapy, including looking for potential intervention targets (6). In addition to the reduction in HbA1c, results from a vast number of clinical and experimental studies have shown the potential effects of glucose-lowering drugs, such as weight reduction, cardiovascular safety, and lipid-lowering and antihypertensive effects; however, the mechanisms behind these benefits need to be further revealed (5, 6).

Gut microbiota has become a hot topic in metabolic disorders in the past decade, including T2DM (7–9). Accumulating evidence confirms that gut microbiota has emerged as a large complex ecological community and a vital regulator of host physical condition, via microbial metabolites and host interactions (10, 11). Among 100 trillion of microorganisms, which is 10 times the number of human body cells, including bacteria, fungi, viruses, and protozoa, the bacterial component is characterized as the most well-investigated group (11, 12) and will be the chief spotlight of this review. There are nearly 500–1,000 species of bacteria within the gastrointestinal tract and more than 90% of the total community are Firmicutes and Bacteroidetes at the phylum level, followed by Proteobacteria, Actinobacteria, Verrucomicrobiota, Fusobacteria, and Firmicutes and Bacteroidetes at the phylum level, followed by Proteobacteria, Actinobacteria, Verrucomicrobiota, Fusobacteria, Cyanobacteria, and Tenericutes (11, 12). The gut microbiota homeostasis is preserved with control of pathogenic microbe growth and protection of beneficial microbes (11, 13). The gut microbiome is considered as a modifiable “new organ” that plays a crucial role in shaping the metabolic and immunological functions of T2DM (14). Although with wide interindividual variation, once the gut microbiota composition was destroyed, an imbalanced gut microbiome community leads to an abnormal production of metabolites, lipid and carbohydrate metabolism disturbance, IR, oxidative stress, and low-grade chronic inflammatory state in T2DM (7, 8, 15–18).

Therefore, understanding how antidiabetes agents influence the gut microbiome might be of importance for optimizing T2DM treatment. Microbiota and host metabolism might interacted with the host as signaling molecules and were further involved in the pathophysiological process of metabolic diseases (29–40) (Figure 1). SCFAs (including butyrate, acetate, and propionate) are the major microbial metabolites produced by dietary fiber fermentation within the intestinal lumen (41). SCFAs were found reduced in T2DM in both clinical and experimental research (42–45). By activation of specific G protein-coupled receptor 41 and 43 (GPR41 and GPR43), SCFAs could stimulate the secretion of peptide tyrosine-tyrosine (PYY) and glucagon-like peptide-1 (GLP-1) from intestinal enteroendocrine L cells (39, 46). PYY is an important neuroendocrine hormone, regulating food intake and energy.

**GUT MICROBIOTA AND METABOLITES ALTERED IN T2DM**

Although the definite microbial signatures linked to T2DM have not been discovered yet, a large number of studies have found that gut microbiota dysbiosis in T2DM is highly associated with specific intestinal microbial taxa or certain enrichment of gene functional pathways (22–28). In a metagenome-wide association study from 345 Chinese individuals, T2DM-related gut flora dysbiosis was characterized by a decreased abundance in a cluster of butyrate-producing bacteria, such as *Roseburia intestinalis*, *Faecalibacterium prausnitzii*, *Clostridiales sp. SS3/4*, and *Eubacterium rectale*, and an increased abundance of opportunistic pathogens, such as *Bacteroides caccae*, *Escherichia coli*, and some *Clostridium* species (*Clostridium symbiosum*, *Clostridium bolteae*, *Clostridium hathewayi*, and *Clostridium ramosum*) (22). Another large-scale metagenome analysis study which recruited a population of 145 70-year-old European women with metagenomic profiles showed increases in the abundance of four *Lactobacillus* species (including *Lactobacillus gasseri*), *Streptococcus mutans*, and *Clostridium hathewayi* and decreases in the abundance of five *Clostridium* species (including *Clostridium beijerinckii*, *Clostridium botulinum*, *Roseburia_272*, and *Bacteroides intestinalis* in the T2DM group (23). Due to the difference in genetic inheritance, diet, and lifestyle factors, the connections among gut microbial communities, microbial metabolites, and T2DM are intricate. Despite the obvious discrepancy in metagenomic clusters between these two populations, the similar microbial functions enriched in T2DM included an increased level in lipid or glucose metabolism-related membrane transport and oxidative stress resistance and a decreased level in metabolism of vitamins and cofactors, butyrate production, and cell motility (22, 23).

To recognize the core gut microbial features of T2DM, a machine learning framework totally recruited more than 9,000 people revealed that a microbiome risk score including 14 microbial features was positively associated with risk of T2DM and the future glucose increment after adjustment for traditional risk factors (such as age, sex, parental history of diabetes, body mass index, systolic blood pressure, and triglycerides) (28). In the meantime, a downward trend of butyrate-producing genus (*Roseburia spp.*) and a rising trend of chronic inflammation-associated genus (*Fibrobacteraceae*) were confirmed in this interpretable machine learning framework (28). Among a substantial body of experimental and clinical research, the genera of *Bifidobacterium*, *Akkermansia*, *Bacteroides*, *Roseburia*, and *Faecalibacterium* were inversely correlated with T2DM, while the genera of *Ruminococcus*, *Blautia*, *Lactobacillus*, and *Fusobacterium* were positively correlated with T2DM (8, 22, 23).

Although the underlying mechanism between complex gut microbiota and T2DM is still unclear, evidence has shown that a variety of metabolites derived from gut flora, including short-chain fatty acids (SCFAs), glycolipid lipopolysaccharides (LPS), bile acids (BAs), trimethylamine-N-oxide (TMAO), indole derivatives, amino acids, vitamins, and one-carbon metabolites, interacted with the host as signaling molecules and were further involved in the pathophysiological process of metabolic diseases (29–40) (Figure 1). SCFAs (including butyrate, acetate, and propionate) are the major microbial metabolites produced by dietary fiber fermentation within the intestinal lumen (41). SCFAs were found reduced in T2DM in both clinical and experimental research (42–45). By activation of specific G protein-coupled receptor 41 and 43 (GPR41 and GPR43), SCFAs could stimulate the secretion of peptide tyrosine-tyrosine (PYY) and glucagon-like peptide-1 (GLP-1) from intestinal enteroendocrine L cells (39, 46). PYY is an important neuroendocrine hormone, regulating food intake and energy.
balance; reduced secretion of GLP-1 in T2DM leads to a reduction of insulin and thus impaired glucose and energy metabolism (47). Besides, SCFAs have been identified as vital mediators in maintaining intestinal immunity and systemic inflammation through upregulating anti-inflammatory regulatory T cells, inhibition of histone deacetylase, and further inhibition of inflammatory signaling pathways and proinflammatory cytokines, such as nuclear factor-kappaB (NF-κB) and tumor necrosis factor alpha (TNF-α) (37, 48).

LPSs, the main compounds of gram-negative bacterial membranes, are known as potent stimulators of inflammation (49). Evidence shows that T2DM subjects possess a high enrichment of gram-negative bacteria, particularly those belonging to Proteobacteria at the phylum level (50). Notably, the Bacteroidetes phylum also belongs to a large part of gram-negative bacteria, but a decreased abundance of Bacteroidetes was found in obesity and diabetes conditions (24, 51–54). This contradiction might be explained by the fact that the LPS produced by the Bacteroidetes phylum has a lower endotoxic activity than other gram-negative bacteria such as the Proteobacteria phylum (55). Subsequently, a high concentration of LPS produced within the gut (metabolic endotoxemia) might lead to chronic low-grade inflammation in diabetic subjects through upregulating inflammatory signaling pathways and proinflammatory cytokine secretion (56, 57). LPSs produced by gut bacteria might damage the intestinal barrier leading to a "leaky gut" syndrome, for instance, a weakened tight junction and reduced gut secretory immunoglobulin A (58).

Originally synthesized from cholesterol in the liver, BAs were revealed to have a reciprocal interaction with gut microbiota via the gut-to-liver axis (40). Primary BAs are converted into secondary BAs by gut microbiota (40). BAs are important...
signaling mediators regulating energy metabolism and systemic inflammation via the nuclear farnesoid X receptor (FXR) and Takeda G protein-coupled receptor 5 (TGR5) (40). In subjects with diabetes and metabolic diseases, BAs’ pool composition altered (60). The altered proportion of FXR antagonistic BAs leads to an altered expression of fibroblast growth factor 19 (FGF19), which were both vital molecules for BAs and glycolipid metabolism (60). Activation of TGR5 by secondary BAs stimulates GLP-1 secretion from L cells to increase insulin secretion and glucose tolerance (61). Evidence shows that modifications of the BA pool presented a beneficial effect in bariatric surgery and antidiabetic treatment (62).

TMAO is predominantly generated from dietary choline, which is transformed to trimethylamine in the gut and then oxidized in the liver (31). Elevated plasma concentrations of TMAO were reported positively related with metabolic dysfunction, such as insulin resistance, CVDs, and T2DM (31, 34, 65), and various bacteria (such as Escherichia fergusonii, Providencia alcalifaciens, and Providencia rustigianii) have been recognized as contributing to the production of TMAO (66). TMAO was found to play a proinflammatory role by activating the nuclear-binding oligomerization domain-like receptor family pyrin domain-containing 3 inflammasome, accelerating reactive oxygen species generation and various proinflammatory cytokines (67). In addition, evidence in experimental research shows that TMAO promoted metabolic dysfunction by directly binding and activating protein kinase-like ER kinase, a key sensor of intracellular stress, and then enhanced transcription activity of forkhead box-O1 in the liver (31).

Indole derivatives are produced from tryptophan by the gut microflora (33). In the recent years, indole derivatives have exhibited anti-inflammatory and anti-diabetic effects (68). Evidence shows that indole derivatives were able to stimulate the secretion of GLP-1 from L cells (32). Various indole derivatives have been synthesized to investigate their bioactivities and biological functions (68). Microbe-specific indoles, such as indole 3-propionic acid, were found to regulate mucosal integrity through activating the xenobiotic sensor, pregnane X receptor, to downregulate enterocyte TNF-α expression and upregulate junctional protein expression (36). In addition to the abovementioned metabolites, vitamins and cofactors produced by probiotics, such as Bifidobacterium and Lactobacillus, yield greater health benefits on patients with T2DM and metabolic diseases (69). Amino acids synthesized by the gastrointestinal microbiota were also vital factors to energy metabolism and glucose homeostasis (70). For instance, Prevotella copri and Bacteroides vulgatus were discovered as the main species mediating the association between biosynthesis of branched-chain amino acids (BCAAs) and IR, and Prevotella copri could induce IR, aggravate glucose intolerance, and increase circulating BCAAs levels (70).

Overall, a vast body of human studies and plentiful animal studies have suggested that T2DM was characterized by gut microbiota dysbiosis and alterations of gut microbiota-derived metabolites, which are important contributors to the pathological injury of T2DM.

### THE EFFECTS OF ORAL ANTIDIABETIC DRUGS ON GUT MICROBIOTA AND MICROBIAL METABOLITES

**Metformin**

Metformin can alleviate patients’ hyperglycemia mainly by significant suppression of glucose production in the liver (71). Activation of the master cellular energy sensor AMP-activated protein kinase (AMPK) is well documented in the mechanism of metformin but may not interpret for its complex beneficial effects (72–75). In fact, metformin was found to modify the intestinal flora community in T2DM in a vast body of clinical research and experimental animal studies (76–80) (summarized in Table 1).

Metagenomics combined with targeted metabolomic data in a randomized, placebo-controlled, double-blind study showed that metformin strongly altered the gut microbiome and its function in individuals with treatment-naive T2DM (79). Subsequently, the authors transplanted fecal samples from three donors (treatment-naive condition compared with 4-month metformin-treated condition) into germ-free mice and observed that glucose tolerance was improved in mice that received 4-month metformin-treated fecal samples, indicating a direct beneficial effect on glucose homeostasis (79). This effect might be mediated by increased SCFA-producing bacteria and the abundance of Akkermansia muciniphila, enriched pathways of the metabolism of vitamins and cofactors, and metalloproteins or metal transporters (79). In line with this research, a large study aimed at disentangling metformin treatment signatures in T2DM recruited 784 subjects from Denmark, Switzerland, and China and illustrated that metformin treatment significantly increased the abundance of Escherichia spp. and reduced that of Intestinibacter spp. The functional enrichment analyses demonstrated that SCFA-producing pathways and enrichment of virulence factors and gas metabolism genes were significantly enhanced, while intestinal lipid absorption and LPS-triggered intestinal inflammation were reduced (77). A randomized clinical trial which recruited 450 T2DM subjects uncovered that metformin altered the gut microbiota composition, increased the beneficial bacteria, such as Blautia and Faecalibacterium, and inhibited potential pathogen-like microbiota, for example, Oscillibacter, Alistipes, and Bacteroides (78).

As summarized in Table 1, most clinical studies revealed that microbes mediated the therapeutic effects of metformin chiefly through improvement in SCFA production, BA pool composition alteration, or reduction in LPS production.

In addition to clinical studies on T2DM patients, a clinical trial which recruited 20 healthy Korean participants found that metformin treatment altered the abundances of Clostridium, Escherichia, Intestinibacter, and Romboutsia, and the relative abundances of metabolites changed including carbohydrate, fatty acid, and amino acid metabolism (95). In experimental animal models, treatment with metformin was revealed to increase SCFA production, to reduce circulation LPS, to inhibit intestinal proinflammatory signaling activities, which was in line with clinical studies (80, 96, 97) (Figure 2). The activation of SCFA receptors, GPR41 and GPR43, stimulated the secretion of...
Clinical research exploring the effects of oral anti-diabetic drugs on gut microbiota in T2DM.

| Anti-diabetic drugs | Subjects | Key results |
|---------------------|----------|-------------|
| Metformin (77)      | 784 subjects from Denmark, Switzerland and China | Escherichia spp.↑ Lactobacillus spp.↓ Functional enrichment: SCFAs producing↑, virulence factors and gas metabolism genes↑, intestinal lipid absorption↓, LPS triggered local inflammation↓ |
| Metformin (78)      | 450 subjects | Simpson’s diversity index↑, Blautia spp. and Faecalibacterium spp↑, Alstes spp.↑, Oscillibacter spp.↑, and Bacteroides spp.↓ |
| Metformin (79)      | 40 treatment-naive T2DM | Firmicutes, Escherichia coli, Bifidobacterium adolescentis, Akkermansia muciniphila↑, SCFA-producing genus↑, Fecal SCFAs and plasma bile acid concentrations↓ |
| Metformin (45)      | 121 subjects | Escherichia coli and Ruminococcus torques↑, Intestinibacter bartlettii↓, Fecal SCFAs increased at 6 months |
| Metformin (81)      | 23 T2DM patients | Enterobacteriaceae↑, Bacteroides fragilis↓, bile acid glycoursodeoxycholic acid↑, Bacteroides caccae, Lachnospiraceae bacterium↑ (Bacteroides uniformis↓, butyrate↑, zinc↓) (microbial butyrate-producing pathways↑) |
| Metformin (82)      | 22 newly diagnosed T2DM | Firmicutes↑, GLP-1↑, lithocholic and deoxycholic acids↑, primary bile acid↓ |
| Metformin (83)      | 14 males with T2DM | Akkermansia muciniphila, Prevotella, Butyrivibrio, Bifidobacterium bifidum, Megasphaera↓, Clostridiales 02d06↓ |
| Metformin (84)      | 112 subjects | Spirochaeta, Turicibacter, and Fusobacterium↑, Taunine and hypotaurine metabolism↑, Several harmful bacteria including Escherichia-Shigella, Bilophila, and Hungatetla↓ |
| Metformin (85)      | 130 T2DM subjects | Bilobacterium↑, SCAA-producing bacterium↑, | |
| Metformin (86)      | 30 T2DM subjects | | |
| Dapagliptin (87)    | 24 subjects | No significant effect on microbial composition |
| Empagliflozin (88) | 67 T2DM with risk factors for CVD | SCFA-producing bacterium↑, Altered plasm BAs pool composition |
| Sitagliptin (89)    | 51 subjects | No significant effect on microbial composition |
| Sitagliptin (90)    | 57 T2DM subjects | Fecal chenodeoxycholic acid↑, cholic acid and ursodeoxycholic acid↑ |
| Vildagliptin (91)   | 30 T2DM subjects | Pseudomonas, Klebsiella, Blautia, Faecalibacterium and Roseburia levels altered |
| Saxagliptin (92)    | 30 T2DM subjects | Megamonas spp↑, Turibacter spp↑ |
| Acarbose (62)      | 51 treatment-naive subjects | Lactobacillus and Bifidobacterium↑ (Bacteroides↓) |
| Acarbose (82)      | 18 subjects | Bilobacterium, Eubacterium, and Lactobacillus↑ (Bacteroides↓) (Bilobacterium longum and Enterococcus faecalis↑) |
| Acarbose (63)      | 96 subjects | Bilobacterium↑, plasma LPS↑ |
| Acarbose (93)      | 30 T2DM subjects | Butyricimonas level increased first and then decreased during treatment |
| Acarbose (94)      | 52 prediabetes patients | Lactobacillus spp.↑ and Dialister spp.↑, Butyricimonas spp., Phascolarctobacterium spp. and Ruminococcus spp.↓ |
| Gliclazide (87)    | 17 subjects | No effect on intestinal microbiota composition |
| Glipizide (62)     | 43 treatment-naive subjects | No effect on intestinal microbiota composition |

SCFAs, short-chain fatty acids; CVD, cardiovascular disease; LPS, lipopolysaccharides; GLP-1, glucagon-likepeptide-1.

PY and GLP-1, inhibiting appetite and improving insulin secretion. At the same time, increased-circulation SCFAs are responsible for improving energy metabolism, suppressing fat accumulation and insulin signaling in adipose tissue, and regulating the intestinal immunity and systemic inflammation (38, 39, 98). Accompanied by decreased LPS produced in the gut, metformin intervention increased goblet cell mass, mucus production, and tight-junction (ZO1 and occludin) proteins in obese gut, thereby relieving intestinal inflammation, decreasing leaky gut, and repairing the intestinal barrier structure (80, 96). In addition, the metabolic benefits of metformin might also be mediated by gut microbiota and bile acid homeostasis (76). Evidence shows that Bacteroides fragilis was decreased in samples from newly diagnosed T2DM patients after metformin treatment for 3 days, meanwhile the BA pool was altered (76). Bile acid glycoursodeoxycholic acid was increased, accompanied by inhibition of intestinal FXR signaling and decreased serum FGF19 levels (76). Reduced circulating FGF19 was found in subjects with metabolic disorders and hepatic steatosis, and FGF19 analogues have been identified as promising therapeutic methods in metabolic improvement (60). However, research associated with FGF19 was inconsistent, and the underlying mechanism still needs further research. Among the numerous gut flora altered during the metformin treatment in both clinical and experimental studies, Akkermansia muciniphila, a mucin-degrading bacterium, is related to healthy intestinal mucosa (79, 84, 96, 99). Furthermore, oral administration of Akkermansia muciniphila to high-fat diet-induced mice without metformin treatment significantly improved glucose homeostasis and reduced visceral adipose tissue inflammation by inducing Tregs, indicating the promising treatment value of Akkermansia spp for T2DM (99).

In brief, in addition to activation of the master cellular energy sensor AMPK (74), metformin might act partly through gut microbiota and its metabolites to improve metabolic health. Notably, the metformin concentration in the gastrointestinal lumen is 30–300 times higher than in the circulation (100). High concentrations of metformin in the gastrointestinal lumen...
can increase glucose uptake and inhibit mitochondrial oxidative phosphorylation in enterocytes then accelerate glucose utilization through glycolysis and overproduction of lactate, the reason why metformin might contribute to gastrointestinal intolerance in a minority of people (71, 101, 102). Previous studies also hint that overproduction of lactate might also be microbially mediated (71, 103). Therefore, the potential mechanisms and contradiction of gastrointestinal intolerance and gut microbiota-related benefits need further investigation.

**SGLT2 Inhibitors**

Sodium-glucose cotransporter 2 (SGLT2) inhibitors improve glycemic control by increasing renal glucose excretion, accompanied by pleiotropic non-glycemic properties, such as reductions in body weight and cardiovascular and renal protection effects (104–107). However, the underlying mechanism of the pleiotropic benefits was still not clear. Evidence shows that the protective effect might be explained for increased ketone body production in CVD, a clear fuel to improve the cardiac function of the energy-starved myocardium (108). As an orally ingested antidiabetic agent, experimental animal studies have found that SGLT2 inhibitor intervention slightly altered the microbiota composition in experimental animal studies (109–111) (summarized in Table 2).

Dapagliflozin treatment showed minor beneficial alterations of gut microbiota in T2DM mice, a trend for decreased *Oscillospira* spp. and *Firmicutes/Bacteroidetes* ratios and increased *Akkermansia muciniphila* in the treatment group (109). In the butyrate-supplemented diet-fed db/db mice, the dapagliflozin-treated mice were also characterized by a decreased trend in Firmicutes/Bacteroidetes ratios, as well as a decreased trend in *Adlercreutzia* spp. and *Alistipes* spp. and an increased trend in *Streptococcus* spp (111). In addition to slight alterations in gut microbiota, SGLT2 inhibitor intervention significantly improved intestinal SCFA production in animal models (110, 113). However, the results were inconsistent, and dapagliflozin treatment was found to have no beneficial effects on gut bacteria in diabetic rats (112). Only two clinical studies explored the alteration of fecal microbiome with SGLT2 inhibitor treatment (87, 88). Seventy-six treatment-naive T2DM with risk factors for CVD were included in a randomized, open-label, two-arm clinical trial (88). After a 3-month intervention, empagliflozin improved glucose metabolism and reduced CVD-related risks, while it significantly altered the gut microbiota, including an increase in SCFA-producing bacteria and a reduction in several harmful bacteria such as *Escherichia–Shigella, Bilophila*, and *Hungatella* (88). However, another clinical study found no significant effect on microbial alpha diversity or composition.
Another experimental study discovered that microbial metabolites, for example, hippurate and indole-3-ethanol, were decreased by pioglitazone intervention in iNOS knockout mice (122). These experiment research suggested that TZDs might have mild protective effects on gut microbiota, mainly focused on lipid and carbohydrate metabolism and inflammation. However, no clinical study focused on gut microbiota and microbial metabolites alterations with TZDs treatment in T2DM subjects; further research is still needed.

**Dipeptidyl Peptidase-4 Inhibitors**

Dipeptidyl peptidase-4 (DPP-4) inhibitors inhibit the degradation of glucagon-like peptide-1 (GLP-1) and glucose-dependent insulintropic polypeptide to stimulate insulin secretion, reserve β-cell function, and maintain glucose homeostasis (123). A series of experimental studies have shown that DPP-4 inhibitors might be able to improve energy metabolism through shaping the gut microbial composition and increasing fecal SCFAs (124–127) (summarized in Table 3). In high-fat diet-induced obesity mice, DPP-4 inhibitors exerted an important impact on gut microbial composition and fecal metabolites, particularly the increased abundance of Bacteroidetes (124). Researchers then transplanted the fecal microbiota of DPP-4 inhibitor-treated patients to germ-free mice and observed an improved glucose intolerance (124). Compared with that in GLP-1 receptor agonist liraglutide-treated mice, the gut microbiota differed substantially in mice treated with DPP-4 inhibitors, indicating that the hypoglycemic mechanism of DPP-4 inhibitors on gut microbiota is at least not primarily by GLP-1 and the other potential benefit of DPP-4 inhibitors needs further research (124, 128). In addition to increment of SCFA-producing flora, DPP-4 inhibitors were found to reduce Toll-like receptor ligands and improve the production of antimicrobial peptides, exerting immunomodulatory and anti-inflammatory effects and maintaining intestinal homeostasis in obese mice, as well as

### Thiazolidinedione Insulin Sensitizers

Thiazolidinedione (TZD) drugs are effective oral agents for T2DM in improving insulin sensitivity (117). TZDs are ligands of peroxisome proliferator-activated receptor gamma (PPAR-γ), leading to the activation of various pathways related to glycemic homeostasis and lipid metabolism (117, 118). The expression of PPAR-γ is abundant in the intestinal tract; thus, it is possible that PPAR-γ agonists straightly impact on gut microbiome homeostasis to improve energy metabolism (119, 120). However, only a few experimental animal studies explored whether TZD treatment can modify gut microbiota homeostasis (119, 121, 122). In a high-fructose-fed mouse model, pioglitazone partly altered gut microbiota and relieved the intestinal inflammation and epithelial barrier impairment, such as preventing the increment of the pathogenic bacteria *Deferribacteresceae* (*Mucispirillum*) (121). In diabetic mice, treatment with rosiglitazone promoted insulin sensitivity without modifying the composition of gut flora but improved the gene expression related to lipid and carbohydrate metabolism as well as immune regulation in the ileum and colon (119).
cross talk with the liver and the whole-host health (126, 127). Some studies exhibited a decreased trend in the Firmicutes/Bacteroidetes ratio with treatment of DPP-4 inhibitors (124, 125, 127), while one experimental animal study found an enlarged abundance of Firmicutes and increased ratios of Firmicutes/Bacteroidetes (94). Although the relation between metabolic disorders and the Firmicutes/Bacteroidetes ratio is currently contradictory, more literatures considered it as a characteristic of obesity and T2DM (55).

There existed a few clinical studies that explored the gut flora modifying the effect of DPP-4 inhibitors (89–91). However, in a clinical study which included 51 T2DM patients, the advantageous effect of sitagliptin on glucose control, weight loss, and BA metabolism was not related to alterations in the gut microbiota (89, 90). No significant effect on microbial composition was found, which is possibly due to the fact that these subjects previously used metformin or sulphonylureas as hypoglycemic therapies, and it might have covered the possible effects of DPP-4 inhibitors (89, 124). Another clinical study which included 90 T2DM subjects found that both vildagliptin and saxagliptin altered the composition of gut microbiota, respectively (91). Thus, the microbiota-shaping effects of DPP-4 inhibitors in clinical studies and its additional hypoglycemic mechanism need further investigation.

**α-Glucosidase Inhibitors**

α-Glucosidase inhibitors are antidiabetic drugs, including acarbose, miglitol, and voglibose, which delay the absorption of carbohydrates in the intestinal tract to inhibit the rise in postprandial plasma glucose concentration (131). α-Glucosidase inhibitors are inhibitors of both human and bacterial α-glucosidases, and because of its high intestinal drug concentration, α-glucosidase usually has noticeable impacts on the intestinal flora (132, 133). Large amounts of research revealed that α-glucosidase inhibitors could shape the composition of the gut microbiome in both animal studies and clinical studies (62, 92–94, 134). Evidence shows that acarbose modulated the gut microbiota and corresponding shaped fecal and plasma BA composition, which may improve host energy metabolism (62, 135). A clinical study which recruited 51 treatment-naïve T2DM patients showed that a three-mouth treatment with acarbose increased *Lactobacillus* and *Bifidobacterium* abundances and reduced *Bacteroides* abundances, along with altered plasma BA composition (62). Another clinical study which included 95 T2DM patients found that acarbose treatment improved the abundance of *Enterococcus faecalis* and *Bifidobacterium longum*, along with the reduction of plasma inflammatory factors, such as prothrombin activator inhibitor-1 and LPS levels (93). As summarized in Table 4, intervention with α-glucosidase inhibitors in experimental animal studies also confirmed significant impacts on gut microbiota and relevant metabolites. In addition to their glucose-lowering and energy metabolism-improving effects, α-glucosidase inhibitors were found to reverse joint inflammation on collagen-induced arthritis mice and the underlying mechanism might be due to the alteration of host-commensal interactions, which have been confirmed to be

| Anti diabetic drugs | Animal model | Dose | Duration | Key results | Mechanism of action |
|---------------------|--------------|------|----------|-------------|---------------------|
| DPP-4 inhibitor (124) | HFD-fed C57BL/6 | 300 mg/kg/day of saxagliptin or 4 g/kg of sitagliptin | 4 weeks | The changes of 65.6% genera induced by HFD were rescued by the DPP-4 inhibitor. Bacteroidetes↑ Firmicutes↓ Bacteroides S24-7 group, Bacteroidaceae, Ruminococcaceae, Desulfovibrionaceae and Streptococcaceae↑ Fecal SCFAs (especially succinate) ↑ Lactobacillus spp., Ruminococcaceae↑ Cecal propionate↑ Cecal TLR ligands↑ | Increasing the production of succinate contributed to the hypoglycemic effect of DPP-4 inhibitor |
| DPP-4 inhibitors (125) | HFD-fed C57BL/6 | 15 mg/kg/day | 12 weeks | Ruminococcus, Dorea, Verrucomicrobiota↑ Plasma sphingomyelin, phosphatidylcholine and lysophosphatidylcholine entities↑ Lactobacillus spp.↑ Cecal propionate↑ Cecal TLR ligands↑ | Elevated levels of butyrate-producing flora Reduced levels of certain plasma sphingomyelin, phosphatidylcholine and lysophosphatidylcholine entities Promoted antimicrobial peptide production and increased crypt depth in the ileum Indirectly reduced the expression of proinflammatory cytokines in the liver |
| Vildagliptin (127) | WD-fed C57BL/6 | 50 mg/kg/day | 8 weeks | Oscillibacter spp., Ruminococcaceae↓ Lactobacillus spp.↑ Firmicutes to Bacteroidetes ratios↑ | Promoted antimicrobial peptide production and increased crypt depth in the ileum |
| Sitagliptin (94) | Zucker diabetic fatty rats | 10.76 mg/kg/day | 4 weeks | Lactobacillus spp.↑ Firmicutes↑ | No significant effect on microbial composition |
| Saxagliptin (128) | STZ-induced ApoE−/− C57BL/6 mice | 80 mg/kg/day | 8 weeks | No significant effect on microbial composition | |
| Linagliptin (126) | HFRU-fed C57BL/6 | 15 mg/kg/day | 5 weeks | Bacteroidetes spp.↑ Proteobacteria spp.↓ Zo-1 mRNA, Mucin mRNA↑ | Attenuated hepatic steatosis by gut-liver axis modulation |
| Vildagliptin (129) | STZ-induced diabetic Sprague-Dawley rats | 20 mg/kg/day | 12 weeks | Firmicutes/Bacteroidetes ratios↑ Bacteroides and Erysipelotrichaceae↑ | Increased SCFAs production |
| Sitagliptin (130) | HFD/HC-STZ Sprague-Dawley rat | 10 mg/kg/day | 12 weeks | Firmicutes↓ Bacteroidetes, Tenericutes↑ | Increased SCFAs-producing bacteria and probiotic |

STZ, streptozocin; HFD, high-fat diet; WD, Western diet; HFRU, high-fructose diet; HF/HC, high fat or high carbohydrate; SCFA, short-chain fatty acids; TLR, Toll-like receptors.

### Table 3: Experimental animal studies analyzing the effects of DPP-4 inhibitors on gut microbiota.

### Table 4: Intervention with α-glucosidase inhibitors in experimental animal studies.
correlated with rheumatoid arthritis, such as several butyrate-producing species, *Lactobacillus* spp. and *Oscillospira* spp (48, 138, 141). These results suggested a promising prospective of α-glucosidase inhibitors due to its potential antiarthritis effect mediated by the gut microbiome (134, 138).

In fact, over 95% of the acarbose dose was not absorbed in the gut, coupled with its feature to inhibit microbial α-glucosidases, and subjects’ treatment response to acarbose is dependent on several factors, such as dietary intake, genetic factor, and microbiota composition before treatment (also named enterotypes) (62, 91–93, 136, 142, 143). The acarbose-shaped gut microbial composition might be related to the dietary intake in a small Japanese population with T2DM (92). Moreover, hierarchical clustering showed that the habitual dietary intake of sucrose, fat, and carbohydrates was associated with three distinct microbial clusters, and even the abundance alteration of *Faecalibacterium* was positively related to dietary rice intake but negatively related to bread intake (92). A previous study also found that patients with a gut flora driven by *Bacteroides* displayed more beneficial modifications in gut microbiota, plasma BA composition, and more metabolic metabolism enhancement after acarbose treatment than those with *Prevotella* (62). In addition, researchers revealed that acarbose resistance has spread in certain host gut microbiomes, which contributed an emerging layer to the multifaceted network of carbohydrate-mediated cross talk among various human microbiomes (132, 144). Besides, in antibiotic pretreatment mice, whose gut microbial enzyme activities have been weakened, the metabolism of voglibose was reduced and more significantly glucose-lowering effects were presented (143). In brief, from currently clinical and experimental studies, α-glucosidase inhibitors have obvious effects on gut microbiota and its effects significantly depend on host diet and the original composition of the gut microbiome.

**Other Oral Glucose-Lowering Agents**

Other less researched oral antidiabetes medications, such as sulfonylurea and glinide insulin secretagogues, have been noticed to cross talk with probiotic bacteria or microbial metabolic profiles (145–147). Nevertheless, two clinical studies which were designed to assess the effects of sulfonylurea in T2DM subjects found no beneficial impacts on gut microbiota composition even in treatment-naïve subjects, but with enhanced glycemic control (62, 87). At the same time, acarbose showed beneficial effects on the composition of the gut microbiome, suggesting that the detected metabolic modifications of sulfonylureas might not be intermediated by their impacts on the gut microbiota (62, 87). Recently, a few newly invented oral anti-glucose agents were discovered and used in clinical application, such as chigitazar and imeglimin (148, 149). Activating as a peroxisome proliferator-activated receptor pan-agonist for glucose control, chigitazar was found to improve insulin sensitivity and lipid homeostasis and reduce circulating levels of inflammatory parameters (150, 151).

**TABLE 4 | Experimental animal studies analyzing the effects of α-glucosidase inhibitors on gut microbiota.**

| Anti-diabetic drugs | Animal model | Dose | Duration | Key results | Mechanism of action |
|---------------------|--------------|------|----------|-------------|---------------------|
| Acarbose (94)       | Zucker diabetic rats | 32.27 mg/kg/day | 4 weeks | Actinobacteria† | Selectively increased the beneficial flora |
| Acarbose (134)      | Old mice      | 1,000 ppm | 8 months | Munibacilaceae † SCFA† | Modulated the fermentation products of the gut flora |
| Acarbose (136)      | HS or PP-fed mice | 400 ppm | 28 days | Diet-dependent gut community structure alteration and SCFA increasing | Increased SCFA production |
| Acarbose (137)      | STZ-induced HFHSD-fed SD rats | 30 mg/kg/day | 7 weeks | Escherichia-Shigella† Munibacilaceae, Lachnospiraceae, Bifidobacterium, Ruminococaceae, UCG-014, Ruminococcus_1, Romboutsia, Eggerthellaceae, Alitipes, Faecalibaculum, Ruminococaceae, UCG-013 and Peptococcaceae† | Beneficial composition of gut microbiota restored |
| Acarbose or miglitol (139) | Collagen-induced arthritis mice | 500 mg/kg/day | 55 days | Lactobacillus spp., Anaeroplasmata spp., Adlercreutzia spp., and RF36 spp.† | Regulated immunity via Th17/Treg cells in the intestinal lamina propria |
| Voglibose (135)     | HFD-fed C57BL/6 mice | 1 mg/kg/day | 12 weeks | the ratio of Firmicutes to Bacteroidetes† Plasm taurocholic, cholic acid and deoxycholic acid† | Downregulated gene expression of CYP8B1 and HNF4a Upregulated gene expression of PGC1α Reduced LPS levels in portal plasma |
| Miglitol (139)      | HFHSD-fed rats | 0.04% miglitol plus in diet | 12 weeks | Enysipelotrichaceae and Coriobacteriaceae† Plasm LPS† | Increased cecal lactate contents and altered intestinal flora |
| Miglitol (140)      | ChREBP-knockout mice | 0.08% miglitol plus in diet | 8 weeks | Lactobacillales and Bifidobacterium† clostridium cluster XIVa† Fecal lactate† | Increased cecal lactate contents and altered intestinal flora |

**Notes:** STZ, streptozocin; HFD, high-fat diet; HS, high-starch; PP, plant polysaccharides; HFHSD, high-fat, high-sucrose diet; SCFAs, short-chain fatty acids; HNF4a, hepatocyte nuclear factor 4α; PGC1α, peroxisome proliferator-activated receptor-yco-activator-1α; LPS, lipopolysaccharide.

**References:**

(48, 138, 141, 62, 91–93, 136, 142, 143)
Imeglimin was confirmed to have the effects of modulating mitochondrial bioenergetics, enhancing mitochondrial function, improving insulin sensitivity, and preserving β-cell function (152–154). However, the associations of these anti-glucose agents and gut microbiota composition were still lacking.

In addition, newly identified exciting targets, including glucokinase activators and G-protein-coupled receptor 40 agonists, have also been researched, although not clinically usable (155, 156). Therefore, with the development of novel glucose-lowering agents, further research is still needed to uncover the complex interaction among gut microbiota, glucose-lowering agents, and the microbial-host metabolic cross talk.

**CONCLUSIONS AND FUTURE PROSPECTS**

Antidiabetic agents modify the gut flora and thereby alter gastrointestinal and plasma metabolite profiles, further improving metabolic health. Knowledge and studies so far indicate that oral antidiabetes drugs, including metformin, DPP-4 inhibitors, and α-glucosidase inhibitors, have obvious effects on gut microbiota and microbial metabolites, while SGLT2 inhibitors and TZDs have slighter effects (62, 77, 87, 89). Even if the definite microbial signatures linked to certain antidiabetic agents have not been discovered yet, understanding how antidiabetes drugs influence the gut microbiome might be vital for identifying their potential mechanisms and optimizing their treatment. Although different hypoglycemic drugs shape gut microbiota differently, they have been confirmed to have some similar effects in regulating microbiota and metabolites. Among various microbiota and metabolites derived from gut flora, metformin, SGLT2 inhibitors, DPP-4 inhibitors, and α-glucosidase inhibitors have been demonstrated to have similar effects on increased SCFA-producing bacteria and SCFA production, which may partly explain their beneficial effects in the regulation of insulin sensitivity enhancement, energy metabolism, and systemic inflammation (77, 78, 110, 124, 134). Notably, among various SCFA-producing bacteria, *Akkermansia muciniphila* has been proven increased particularly during the metformin treatment in both clinical and experimental studies, which also related to healthy intestinal mucosa and anti-inflammatory action (79, 84, 96, 99). In addition, alteration of the BA pool was commonly displayed in both metformin and α-glucosidase inhibitors, corresponding with decreased *Bacteroides fragilis* in metformin-treated individuals and increased *Lactobacillus* and *Bifidobacterium* abundances and reduced *Bacteroides* abundances in α-glucosidase inhibitor-treated individuals, respectively (62, 76). In addition, reduction of opportunistic pathogen and attenuated intestinal inflammation could be seen in intervention research on metformin, DPP-4 inhibitors, and α-glucosidase inhibitors (77, 126, 138). Therefore, manipulation of gut microflora composition could be a potential and promising target to improve metabolic outcomes in subjects with T2DM. The microbiota–host cross talk might convey novel and potential ideas of generally used oral glucose-lowering drugs.

Firstly, combination therapy might have additional benefits, due to the fact that different antidiabetes drugs shape gut microbiota with distinct effects (62, 94, 112). For example, dapagliflozin increased the abundance of *Desulfovibrioaceae* in a T2DM rat model, which is an unfriendly sulfate-reducing bacteria in the gut, while metformin reduced it on the contrary, revealing a rationality and complementary action of combined pharmacotherapy between dapagliflozin and metformin (112). However, the definite combination effects of metformin and SGLT2 inhibitors need further investigation. T2DM is a chronic disease with progressive features and possible complex complications; a satisfactory treatment effect is hard to achieve with monotherapy. Besides metformin and SGLT2 inhibitor combined treatment, combination therapies, such as metformin with pioglitazone or metformin with DPP-4 inhibitors, might exhibit a synergistic role in gut microbiome benefits (157, 158). Further investigations in both experimental and clinical are needed to figure out the combined pharmacotherapy effects on gut microbiota.

Secondly, pre- and probiotics could be a promising treatment for T2DM in the modulation of gut microbiota (159). For example, *Actinoplanes* spp. and *Lactobacillus* spp. have been definitively demonstrated to effectively inhibit the alpha-glucosidase activity to reduce glucose levels (160, 161). The combination of hypoglycemic agents and certain probiotics or prebiotics may further enhance the glucose-lowering effects (82, 162). Prebiotics, such as inulin and galacto-oligosaccharide, could be fermented by the gut flora, leading to modulation of intestinal microbiota and the production of various microbial metabolites including SCFAs (163–165). Besides, evidence shows that combination of metformin and gastrointestinal microbiome modulator (consisting of inulin, betaglucan, and polyphenols) treatment significantly relieved metformin tolerance than the placebo combination (166). Notably, for patients with pregestational diabetes and gestational diabetes mellitus (GDM), the dominating pharmacotherapy is insulin, while only metformin and glyburide are used in some countries (167, 168). Other oral hypoglycemic agents are limited in these patients. Hyperglycemia during pregnancy is associated with significantly increased maternal and fetal metabolic disturbance and morbidity (167). Therefore, dietary modification and physical activity are particularly important for glycemia control (167). A systematic review and meta-analysis revealed that probiotic supplementation in GDM could significantly reduce homeostasis model assessment of the insulin resistance index with no adverse effects reported (169). Evidence shows that inulin-type fructan supplementary improved glucose and lipid metabolism in HFD-induced GDM mouse models associated with gut flora modification (170). Our research team found that maternal inulin treatment improved glucose metabolism in adult male offspring via regulation of the hepatic long non-coding RNA profile (164). However, results are inconsistent showing that probiotics, including *Lactobacillus rhamnosus* and *Bifidobacterium animalis* subspecies *lactis*, did not prevent GDM in overweight and obese pregnant women (171). Thus, more clinical studies are needed to verify these results and explore the ideal bacterial composition of pre- and probiotics that might positively alter glucose metabolism in GDM or pregestational diabetes.

Thirdly, FMT from normal glucose tolerance or antidiabetes treatment subjects to mice revealed a significant improvement in gut microbiota composition, glucose homeostasis, and metabolic
health (17, 124, 172). Despite that this promising treatment was still in its infancy (173–175), FMT combined with antidiabetes drugs might bring novel interventions and perspectives in T2DM management. The effects and mechanisms underneath these potential treatment schedules are still unclear, and it is vital to further develop meaningful and applicable interventions combined with intestinal microbiota in the future study.

**AUTHOR CONTRIBUTIONS**

DW: writing—original draft preparation. JL, LZ, QZ, ML, and XX: writing—review and editing. XX: supervision. XX and QZ: funding acquisition. All authors contributed to the article and approved the submitted version.

**FUNDING**

This work was supported by the grants from the National Natural Science Foundation of China (No. 82170854, 81870579, 81870545, 81507015, 81170376), Beijing Natural Science Foundation (7202163), Beijing Municipal Science & Technology Commission (Z20110000520011), CAMS Innovation Fund for Medical Sciences (CIFMS2021-12M-002).

**REFERENCES**

1. Sun H, Saeedi P, Karuranga S, Pinkepank M, Ogurtsova K, Duncan BB, et al. IDF Diabetes Atlas: Global, Regional and Country-Level Diabetes Prevalence Estimates for 2021 and Projections for 2045. *Diabetes Res Clin Pract* (2022) 183:109119. doi: 10.1016/j.diabres.2021.109119

2. Stumvoll M, Goldstein BJ, van Haften TW. Type 2 Diabetes: Principles of Pathogenesis and Therapy. *Lancet* (2005) 365(9467):1335–46. doi: 10.1016/s0140-6736(05)61302-x

3. Gregg EW, Sattar N, Ali MK. The Changing Face of Diabetes Complications. *Lancet Diabetes Endocrinol* (2016) 4(6):537–47. doi: 10.1016/s2213-8587(16)30010-9

4. Ferguson D, Finck BN. Emerging Therapeutic Approaches for the Treatment of NAFLD and Type 2 Diabetes Mellitus. *Nat Rev Endocrinol* (2021) 17(8):484–95. doi: 10.1038/s41574-021-00507-z

5. Draznin B, Aroda VR, Bakris G, Benson G, Brown FM, Freeman R, et al.9.

6. Masters of Host Development

7. Bordalo Tonucci L, Dos Santos KM, De Luces Fortes Ferreira CL, Ribeiro SM, De Oliveira LL, Martino HS. Gut Microbiota and Probiotics: Focus on Metabolic Syndrome Traits in the METSIM Cohort. *Genome Biol* (2021) 51:102590. doi: 10.1038/ebiomed.2019.11.051

8. Turnbaugh PJ, Ley RE, Hamady M, Fraser-Liggett CM, Knight R, Gordon JI. *Nature* (2006) 449(7164):804–10. doi: 10.1038/nature06244

9. Canfora EE, Meex RCR, Venema K, Blaak EE. Gut Microbial Metabolites in Obesity, NAFLD and T2DM. *Nat Rev Endocrinol* (2019) 15(5):261–73. doi: 10.1038/s41574-019-01516-z

10. Louzopone CA, Stombaugh JI, Gordon JI, Jansson JK, Knight R. Diversity, Pathogenesis and Therapy. *Lancet* (2005) 365(9467):1335–46. doi: 10.1016/s0140-6736(05)61302-x

11. Lozupone CA, Stombaugh JI, Gordon JI, Jansson JK, Knight R. Diversity, Pathogenesis and Therapy. *Lancet* (2005) 365(9467):1335–46. doi: 10.1016/s0140-6736(05)61302-x

12. Sommer F, Bäckhed F. The Gut Microbiota in Prediabetes and Diabetes: A Population-Based Cross-Sectional Study. *Diabetes Care* (2021) 44(Suppl 1):S111–s124. doi: 10.2337/dc21-0454-8

13. Gou W, Ling CW, He Y, Jiang Z, Fu Y, Xu F, et al. Interpretable Machine Learning Framework Reveals Robust Gut Microbiome Features Associated with Type 2 Diabetes. *Nat Commun* (2021) 12:803. doi: 10.1038/s41467-021-03260-7

14. Tilg H, Moschen AR. Microbiota and Diabetes: An Evolving Relationship. *Gut* (2014) 63(9):1513–21. doi: 10.1136/gutjnl-2014-306928

15. Zhao L, Zhang F, Ding X, Wu G, Lam YY, Wang X, et al. Gut Bacteria Selectively Promoted by Dietary Fibers Alleviate Type 2 Diabetes. *Science* (2018) 359(6380):1151–6. doi: 10.1126/science.aao5774

16. Jia J, Dou P, Gao M, Kong X, Li C, Liu Z, et al. Assessment of Causal Direction Between Gut Microbiota-Dependent Metabolites and Cardiometabolic Health: A Bidirectional Mendelian Randomization Analysis. *Diabetes* (2019) 68(9):1747–55. doi: 10.2337/db19-0153

17. Zhang PP, Li LL, Han X, Li QW, Zhang XH, Liu JI, et al. Fecal Microbiota Transplantation Improves Metabolism and Gut Microbiome Composition in Db/db Mice. *Acta Pharmacol Sin* (2020) 41(5):678–85. doi: 10.1038/s41401-019-0330-9

18. Vangipurapu J, Fernandes Silva L, Kaulasamaa T, Smith U, Laakso M. Microbiota-Related Metabolites and the Risk of Type 2 Diabetes. *Diabetes Care* (2020) 43(6):1319–25. doi: 10.2337/dc19-2533

19. Hu N, Zhang Q, Wang H, Yang X, Jiang Y, Chen R, et al. Comparative Evaluation of the Effect of Metformin and Insulin on Gut Microbiota and Metabolome Profiles of Type 2 Diabetic Rats Induced by the Combination of Streptozotocin and High-Fat Diet. *Front Pharmacol* (2021) 12:794103. doi: 10.3389/fphar.2021.794103

20. Ng SC, Xu Z, Mak JWY, Yang K, Liu Q, Zuo T, et al. Microbiota Engraftment After Fecal Microbiota Transplantation in Obese Subjects With Type 2 Diabetes: A 24-Week, Double-Blind, Randomised Controlled Trial. *Gut* (2022) 71(4):716–23. doi: 10.1136/gutjnl-2020-332617

21. Hartsva AR, Boutier KE, Bäckhed F, Nieuwoudt M. Insights Into the Role of the Microbiome in Obesity and Type 2 Diabetes. *Diabetes Care* (2015) 38(1):159–65. doi: 10.2337/dc14-0769

22. Qin J, Li Y, Cai Z, Li S, Zhu J, Zhang F, et al. A Metagenome-Wide Association Study of Gut Microbiota in Type 2 Diabetes. *Nature* (2012) 490(7418):55–60. doi: 10.1038/nature11450

23. Karlsson FH, Tremaroli V, Nookaew I, Bergström G, Brehm CJ, Fagerberg B, et al. Gut Metagenome in European Women With Normal, Impaired and Diabetic Glucose Control. *Nature* (2013) 498(7452):99–103. doi: 10.1038/nature12198

24. Medina-Vera I, Sanchez-Tapia M, Noriega-Lopez L, Granados-Portillo O, Guevara-Cruz M, Flores-Lopez A, et al. A Dietary Intervention With Functional Foods Reduces Metabolic Endotoxaemia and Attenuates Biochemical Abnormalities by Modifying Fecal Microbiota in People With Type 2 Diabetes. *Diabetes Metab* (2019) 45(2):122–31. doi: 10.1016/j.dib.2018.09.004

25. Org E, Blum Y, Kasela S, Mehrabian M, Kuusisto J, Kangas AJ, et al. Relationships Between Gut Microbiota, Plasma Metabolites, and Metabolic Syndrome Traits in the METSIM Cohort. *Genome Biol* (2017) 18(1):70. doi: 10.1186/s13059-017-1194-2

26. Li SC, Xiao Y, Wu RT, Xie D, Zhao HH, Shen GY, et al. Comparative Analysis of Type 2 Diabetes-Associated Gut Microbiota Between Han and Mongolian People. *J Microbiol* (2021) 59(7):693–701. doi: 10.1007/s12275-021-0454-8

27. Wu H, Tremaroli V, Schmidt C, Lundqvist A, Olsson LM, Krämer M, et al. The Gut Microbiota in Prediabetes and Diabetes: A Population-Based Cross-Sectional Study. *Cell Metab* (2020) 32(3):379–390.e3. doi: 10.1016/j.cmet.2020.06.011

28. Gou W, Ling CW, He Y, Jiang Z, Fu Y, Xu F, et al. Interpretable Machine Learning Framework Reveals Robust Gut Microbiome Features Associated
Wang et al. Gut Microbiota and T2DM

29. Aguilar, Clément K, Sokol H. Gut Microbiota-Derived Metabolites as Central Regulators in Metabolic Disorders. Gut (2021) 70(6):1174–82. doi: 10.1136/gutjnl-2020-323071

30. Krautkramer KA, Fan J, Bäckhed F. Gut Microbial Metabolites as Multi-kingdom Intermediates. Nat Rev Microbiol (2021) 19(2):77–94. doi: 10.1038/s41579-020-0438-4

31. Chen S, Henderson A, Petriello MC, Romano KA, Gearing M, Miao J, et al. Trimethylamine N-Oxide Binds and Activates PERK to Promote Metabolic Dysfunction. Cell Metab (2019) 30(6):1141–1151.e5. doi: 10.1016/j.cmet.2019.08.021

32. Chimerel C, Esmery E, Summers DK, Keyser U, Gribble FM, Reimann F. Bacterial Metabolite Indole Modulates Incretin Secretion From Intestinal Enteroendocrine L Cells. Cell Rep (2014) 9(4):1202–8. doi: 10.1016/j.celrep.2014.10.032

33. Galligan JJ. Beneficial Actions of Microbiota-Derived Tryptophan Metabolites. Neurogastroenterol Motil (2018) 30(2):e13283. doi: 10.1111/nmm.13283

34. Heianza Y, Ma W, Manson JE, Rexrode KM, Qi L. Gut Microbiota Metabolites and Risk of Major Adverse Cardiovascular Disease Events and Death: A Systematic Review and Meta-Analysis of Prospective Studies. J Am Heart Assoc (2017) 6(7):e004947. doi: 10.1161/JAHA.116.004947

35. Saad MJ, Santos A, Prada PO. Linking Gut Microbiota and Inflammation to Obesity and Insulin Resistance. Physiol (Bethesda) (2016) 31(4):283–93. doi: 10.1152/physiol.00014.2015

36. Venkatesh M, Mukherjee S, Wang H, Li H, Sun K, Benechet AP, et al. Gut Microbial Metabolites as Multi-kingdom Intermediates. Nat Rev Microbiol (2021) 504(7480):451–5. doi: 10.1038/nature12726

37. Ratajczak W, Rytel M, Kus C, Leszczyńska S, Strakowska M. Bile Acid Control of Inflammation in Obesity, Type 2 Diabetes, Dyslipidemia, and Gut Immunity. Cell Metab (2019) 60(5):2343–60. doi: 10.1016/s1097-6110(19)30157-5

38. Yang W, Yu T, Huang X, Bilotta AJ, Xu L, Lu Y, et al. Intestinal Microbiota-Bacterial Metabolite Indole Modulates Incretin Secretion From Intestinal Enteroendocrine L Cells. Cell Rep (2014) 9(4):1202–8. doi: 10.1016/j.celrep.2014.10.032

39. Kimura I, Ozawa K, Inoue D, Imamura T, Kimura K, Maeda T, et al. The Gut Microbiota and Short-Chain Fatty Acids in the Faeces of T2DM Rats Should be One of Antidiabetic Cations and Their Consequences. Front Nutr (2021) 80(1):37–49. doi: 10.1017/s0029665120006916
110. McCreight IJ, Bailey CJ, Pearson ER. Metformin and the Gastrointestinal Tract. *Diabetologia* (2016) 59(3):426–35. doi:10.1007/s00125-015-3844-9

111. Bhat M, Molyneux S, Batteux B, Gras-Champel V, Masmoudi K, Maizel J, et al. A Study of Associations Between Plasma Metformin Concentration, Lactic Acidosis, and Mortality in an Emergency Hospitalization Context. *Crit Care Med* (2020) 48(12):e194–202. doi:10.1097/CCM.0000000000004589

112. Lazarus B, Wu A, Shin JI, Sang Y, Alexander GC, Secora A, et al. Association of Metformin Use With Risk of Lactic Acidosis Across the Range of Kidney Function: A Community-Based Cohort Study. *JAMA Intern Med* (2018) 178(7):903–10. doi:10.1001/jamainternmed.2018.0292

113. Mishima E, Fukuda S, Kanemitsu Y, Saigusa D, Mukawa C, Asaji K, et al. The Gut Microbiota Drives the Impact of Bile Acids and Fat Source in Diet on Microbial Cellular Volume in Patients With Type 2 Diabetes. *Diabetes* (2021) 70(12):2810–22. doi:10.2337/db21-0270

114. Li L, Xu S, Guo T, Gong S, Zhang C. Effect of Dapaglifozin and Cardiovascular Outcomes in Type 2 Diabetes. *N Engl J Med* (2019) 380(4):347–57. doi:10.1056/NEJMoai1812389

115. Pilkovic V, Jardine MJ, Neal B, Bompunt S, Heerspink HJL, Charytan DM, et al. Canaglifozin and Renal Outcomes in Type 2 Diabetes and Nephropathy. *N Engl J Med* (2019) 380(24):2295–306. doi:10.1056/NEJMe1911713

116. Osataphan S, Macchi C, Singhal G, Chimene-Weiss J, Sales V, Kozuka C, et al. SGLT2 Inhibition Reprograms Systemic Metabolism via FGFr21-Dependent and -Independent Mechanisms. *JCI Insight* (2019) 4(5): e123310. doi:10.1172/jci.insight.123310

117. Ahmadian M, Suh JM, Hahn N, Liddle O, Atkins AR, Downes M, et al. Pparγ Signaling and Metabolism: The Good, the Bad and the Future. *Nat Metab* (2013) 19(5):557–66. doi:10.1038/nm.3159

118. Yang H, Suh DH, Kim DH, Jung ES, Liu KH, Lee CH, et al. Metabolomic and Lipidomic Analysis of the Effect of Pioglitazone on Hepatic Steatosis in a Rat Model of Obese Type 2 Diabetes. *Br J Pharmacol* (2018) 175(17):5610–25. doi:10.1111/bph.14434

119. Madsen MSA, Gronhøj MD, Eid J, Christensen-Dalsgaard M, Sommer M, Rigbolt K, et al. Characterization of Local Gut Microbiome and Intestinal Transcriptome Responses to Rosiglitazone Treatment in Diabetic Db/Db Mice. *BioMed Pharmacother* (2021) 133:110966. doi:10.1016/j.biopha.2020.110966

120. Lefèvre M, Paulweber B, Fajas L, Woods J, McCrory C, Colombel JF, et al. Peroxisome Proliferator-Activated Receptor Gamma Is Induced During Differentiation of Colon Epithelium Cells. *J Endocrinol* (1999) 162(3):331–40. doi:10.1677/joe.01620331

121. Li JM, Yu R, Zhang LP, Wen SY, Wang SJ, Zhang XY, et al. Dietary Fructose-Induced Gut Dysbiosis Promotes Mouse Hippocampal Neuroinflammation: A Benefit of Short-Chain Fatty Acids. *Microbiome* (2019) 7(1):98. doi:10.1186/s41689-019-0713-7

122. Aggarwal H, Pathak P, Kumar Y, Jagavelu K, Dikshit M. Modulation of Inulin Resistance, Dyslipidemia and Serum Metabolome in iNos Knockout Mice Following Treatment With Nitrite, Metformin, Pioglitazone, and a Combination of Ampicillin and Neomycin. *Int J Mol Sci* (2021) 23(1):195. doi:10.3390/ijms23010195

123. Thornberry NA, Gallwitz B. Mechanism of Action of Inhibitors of Dipeptidyl-Peptidase-4 (DPP-4). *Best Pract Res Clin Endocrinol Metab* (2009) 23(4):479–86. doi:10.1016/j.beem.2009.03.004

124. Lian X, Song L, Zeng B, Liu B, Qiu Y, Qu H, et al. Alteration of Gut Microbiota Induced by DPP-4 Treatment Improves Glucose Homeostasis. *ElBioMedicine* (2019) 44:665–74. doi:10.1016/j.ebiom.2019.03.057

125. Ryan PM, Patterson E, Carafa I, Mandal R, Wishart DS, Dinan TG, et al. Metformin and Dipeptidyl Peptidase-4 Inhibitor Differentially Modulate the Intestinal Microbiota and Plasma Metabolome of Metabolically Dysfunctional Mice. *Curr Diabetes Rev* (2020) 4(2):146–155.e2. doi:10.2174/1742287619666180912143434

126. Silva-Veiga FM, Miranda CS, Martins FF, Daleprane JB, Mandarim-de-Lacerda CA, Souza-Mello V. Gut-Liver Axis Modulation in Fructose-Fed Mice: A Role for PPAR-Alpha and Linaglitin. *J Endocrinol* (2020) 247(11):1–24. doi:10.1530/joe-20-01319

127. Olivares M, Neyerick AM, Pötgens SA, Beaumont M, Salazar N, Cani PD, et al. The DPP-4 Inhibitor Vildagliptin Impacts the Gut Microbiota and Prevents Disruption of Intestinal Homeostasis Induced by a Western Diet in Mice. *Diabetologia* (2018) 61(8):1838–48. doi:10.1007/s00125-018-4647-6

128. Wang L, Li P, Tang Z, Yan X, Feng B. Structural Modulation of the Gut Microbiota and the Relationship With Body Weight: Compared Evaluation of Liraglutide and Saxagliptin Treatment. *Sci Rep* (2016) 6:33251. doi:10.1038/srep33251

129. Zhang Q, Xiao L, Li M, Yu M, Yu P, Zheng J, et al. Vildagliptin Increases Butyrate-Producing Bacteria in the Gut of Diabetic Rats. *Plos One* (2017) 12(10):e0184735. doi:10.1371/journal.pone.0184735

130. Yan X, Feng B, Li P, Tang Z, Wang L. Microflora Disturbance During Progression of Glycemic Intolerance and Effect of Sitagliptin: An Animal Study. *J Diabetes Res* (2016) 2016:2093171. doi:10.1155/2016/2093171

131. Dirir AM, Daou M, Yousef AF, Yousef LF. A Review of Alpha-Glucosidase Inhibitors and Gut Bacteria -Glucosidase Inhibitors From Plants as Potential Candidates for the Treatment of Type-2 Diabetes. *Phytochem Rev* (2021) 1–31. doi:10.1007/s11101-021-09773-1

132. Balaich J, Estrella M, Wu G, Jeffrey PD, Biswas A, Zhao L, et al. The Human Microbiome Encodes Resistance to the Antidiabetic Drug Acarbose. *Nature* (2021) 508(7537):110–5. doi:10.1038/s41586-021-04091-0

133. Tan K, Tesař C, Wilton R, Jedrzejczak RP, Joachimiak A. Interaction of Antidiabetic α-Glucosidase Inhibitors and Gut Bacteria α-Glucosidase. *Protein Sci* (2018) 27(8):498–508. doi:10.1002/pro.3444

134. Smith BJ, Miller RA, Ericson AC, Harrison DC, Strong R, Schmidt TM. Changes in the Gut Microbiome and Fermentation Products Concurrent With Enhanced Longevity in Acarbose-Treated Mice. *MBio Microbiol* (2019) 19(1):130. doi:10.1128/mBio.00986-19

135. Do H, Lee YS, Ha MJ, Cho Y, Hwang Y, Yoon JY, et al. Beneficial Effects of Voglibose Administration on Body Weight and Lipid Metabolism via Gastrointestinal Bile Acid Modification. *Endocr J* (2016) 63(8):691–702. doi:10.1507/endoj.115-0747

136. Baxter NT, Lesniak NA, Sinani H, Schloss PD, Koropatkin NM. The Glucosylamine Inhibitor Acarbose Has a Diet-Dependent and Reversible Effect on Body Weight, Lipid Metabolism, and Insulin Sensitivity. *Am J Physiol Gastrointest Liver Physiol* (2021), 1–31. doi:10.1152/ajpgi.00141.2021

137. Wang et al. Gut Microbiota and T2DM
Conflict of Interest: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher’s Note: All claims expressed in this article are solely those of the authors and do not necessarily represent those of their affiliated organizations, or those of the publisher, the editors and the reviewers. Any product that may be evaluated in this article, or claim that may be made by its manufacturer, is not guaranteed or endorsed by the publisher.

Copyright © 2022 Wang, Liu, Zhou, Zhang, Li and Xiao. This is an open-access article distributed under the terms of the Creative Commons Attribution License (CC BY). The use, distribution or reproduction in other forums is permitted, provided the original author(s) and the copyright owner(s) are credited and that the original publication in this journal is cited, in accordance with accepted academic practice. No use, distribution or reproduction is permitted which does not comply with these terms.