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The Effects of Synbiotic Supplementation on Metabolic Status in Diabetic Patients Undergoing Hemodialysis: a Randomized, Double-Blinded, Placebo-Controlled Trial

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Abstract

This study was conducted to evaluate the effects of synbiotic supplementation on metabolic profiles in diabetic patients undergoing hemodialysis (HD). This randomized, double-blinded, placebo-controlled clinical trial was performed in 60 diabetic HD patients. Participants were randomly assigned into two groups to receive either synbiotic capsule, containing *Lactobacillus acidophilus*, *Lactobacillus casei*, and *Bifidobacterium bifidum* (2×10^9 CFU/g each), plus 0.8 g/day of inulin ($n = 30$) or placebo ($n = 30$) for 12 weeks. Synbiotic supplementation significantly decreased fasting plasma glucose ($\beta - 13.56$ mg/dL; 95% CI, $- 23.82, - 3.30$; $P = 0.01$), insulin levels ($\beta - 5.49$ µIU/mL; 95% CI, $- 6.92, - 4.05$; $P < 0.001$), and insulin resistance ($\beta - 2.25$; 95% CI, $- 3.02, - 1.48$; $P < 0.001$), while increased the quantitative insulin sensitivity check index ($\beta 0.02$; 95% CI, 0.01, 0.02; $P < 0.001$) compared with the placebo. Additionally, synbiotic intake resulted in a significant reduction in high-sensitivity C-reactive protein ($\beta - 2930.48$ ng/mL; 95% CI, $- 3741.15, - 2119.80$; $P < 0.001$) and malondialdehyde levels ($\beta - 0.60$ µmol/L; 95% CI, $- 0.99, - 0.20$; $P = 0.003$). Moreover, we found a significant increase in total antioxidant capacity ($\beta 142.99$ mmol/L; 95% CI, 61.72, 224.25; $P = 0.001$) and total glutathione levels ($\beta 131.11$ µmol/L; 95% CI, 89.35, 172.87; $P < 0.001$) in the synbiotic group compared with the placebo group. Overall, synbiotic supplementation for 12 weeks had beneficial effects on glycemic control, biomarkers of inflammation, and oxidative stress in diabetic patients under HD. This study was registered in the Iranian website (www.irct.ir) for registration of clinical trials (http://www.irct.ir: IRCT2017090133941N17). http://www.irct.ir: IRCT2017090133941N17.

Keywords Synbiotic supplementation · Diabetes · Hemodialysis · Metabolic status

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Introduction

Diabetes mellitus is associated with a chronic inflammatory state and accounts as a major cause of end-stage renal disease (ESRD) [1]. The risk of cardiovascular events is higher in diabetic patients compared to non-diabetics with ESRD [2]. Several factors including insulin resistance, elevated reactive oxygen species (ROS), decreased antioxidant defense, infection, and comorbid conditions such as hypertension exacerbate the inflammatory status in patients undergoing hemodialysis (HD) [3]. Recently, it has stated that the intestinal tract and microbiota may play an important role in the systemic inflammation detected in patients under HD [4].

Probiotics are "live strains of strictly selected microorganisms, when administered in adequate amounts can confer a health benefit in the host" [5]. Prebiotics are nonviable food component that confer a health benefit through modulating the host's microbiota [5]. Synbiotics describe as a combination of synergistically acting probiotics and prebiotics [6]. The use of synbiotic for a number of metabolic disorders, including metabolic syndrome and diabetes, has recently received much attention. Therapeutic interventions to target the intestinal microbiota are promising areas of investigation in diabetic patients undergoing HD. In addition, diabetic HD patients are in a particular state of health and this can be affected by the beneficial effects of synbiotics or any medical intervention. A study conducted in ESRD patients under HD therapy has shown that synbiotic compounds could balance the intestinal microbiota through increasing the number of Bifidobacterium [7]. The beneficial effects of synbiotic supplementation have been demonstrated on insulin metabolism in patients with type 2 diabetes mellitus (T2DM) [8], non-alcoholic fatty liver disease (NAFLD) [9], rheumatoid arthritis (RA) [10], and overweight and obesity [11]. In addition, we have also reported that synbiotic consumption in diabetic patients significantly improved glucose homeostasis parameters [12], although other researchers failed to find any glycemic improvement in response to ingesting synbiotic supplements in diabetic patients [13, 14].

The effects of synbiotics on lipid profiles in patients with metabolic disorders are controversial. Eslamparast et al. [15] reported that synbiotics supplementation for 28 weeks improved triglycerides and total- and HDL-cholesterol in patients with metabolic syndrome (MetS), yet did not affect LDL-cholesterol levels. Moreover, synbiotic intake in healthy young volunteers significantly reduced triglycerides and total- and LDL-cholesterol, while increased HDL-cholesterol levels [16]. Conversely, we did not observe any beneficial effect on lipid profiles following synbiotic supplementation in patients with RA [10]. The favorable effects of synbiotics and probiotics on metabolic profiles may be due to the microbial production of short-chain fatty acids (SCFA), which in turn results in increasing glucagon-like peptide-1 (GLP-1) and

inhibits postprandial hyperglycemia [17]. Moreover, probiotics may suppress inflammation and oxidative stress by inhibiting lipid peroxidation, modulating the pattern of cytokine secretion from a pro-inflammatory to an anti-inflammatory state and preserving the activity of antioxidant enzymes [18]. However, data investigating the impact of synbiotic on glycemic control, lipid profiles, biomarkers of inflammation, and oxidative stress in diabetic patients undergoing HD are scarce. Therefore, this study was aimed to determine the effects of synbiotic supplementation on metabolic profiles of diabetic patients under HD.

Methods

Trial Design and Study Participants

This study was registered in the Iranian website for clinical trials (http://www.irct.ir: IRCT2017090133941N17). It was a randomized, double-blinded, placebo-controlled clinical trial. This investigation was conducted in 60 diabetic patients undergoing HD, 18 to 80 years old, and had been referred to the Akhavan Clinic in Kashan, Iran, between November 2017 and February 2018. The investigation was performed following the Declaration of Helsinki and informed consent was taken from all participants. This study was approved by the ethics committee of Kashan University of Medical Sciences (KAUMS). Pregnant women, taking probiotic and/or synbiotic supplements, antioxidant and/or anti-inflammatory supplements within the last 3 months prior to the enrollment in the study; patients who required medications adjustment during the study; and those who had recently been diagnosed with T1DM or T2DM were excluded from the study.

Study Design

At first, participants were matched based on sex, duration of dialysis and diabetes, BMI, and age. Patients were requested not to change their routine physical activity or usual diets throughout the study and not take any anti-inflammatory and antioxidant medications or supplements during the 12-week intervention which might affect the results of the study. Consumption of synbiotic supplements and placebos was monitored through asking subjects to return the medication containers. Furthermore, a short message was being sent to all participants' cell phones every day to remind them using the supplements. A 3-day food record (one weekend day and two weekdays) and physical activity records were completed by all participants. The individual's nutrient intake was then calculated and averaged at weeks 0, 3, 6, 9, and 12 of the intervention using Nutritionist IV software (First Databank, San Bruno, CA), the version which is modified for Iranian foods.

Intervention

To have more appropriate function, the strains used in probiotic supplements for human consumption should be derived from the human intestinal tract, well characterized, able to survive the rigors of the digestive tract and colonized, biologically active against the target, and stable and amenable for commercial production and distribution [19]. These are the basic minimal criteria that should be considered in a high-quality probiotic supplement. Little was known about the ideal choice and the dosage of synbiotic used for diabetic patients undergoing HD, so we selected the supplement and its dose based on previous published studies in diabetic patients with coronary heart disease [20] and overweight children [21]. In these trial cases ($n = 30$) received a synbiotic capsule containing *Lactobacillus acidophilus*, *Lactobacillus casei*, and *Bifidobacterium bifidum* (2×10^9 CFU/day each) plus 0.8 g/day of inulin for 12 weeks. Synbiotic supplements and placebos (corn starch) were produced by Tak Gen Zist Pharmaceutical Company, Tehran, Iran, and approved by the Food and Drug Administration.

Assessment of Anthropometric Measures

A trained staff took anthropometric measurements at the clinic at baseline and after 12 weeks of intervention. Body weight was measured after an overnight fast, using a calibrated digital scale (Seca, Hamburg, Germany).

Assessment of Outcomes

In the current study, the homeostasis model of assessment-insulin resistance (HOMA-IR) was considered as the primary outcome. Lipid profiles, biomarkers of inflammation, and oxidative stress were assessed as secondary outcomes. Fasting blood samples were collected at the beginning and 12 weeks after the intervention (10 mL/sample) at Kashan reference laboratory and centrifuged to separate serum. Then, the samples were stored at − 80 °C until analysis. Serum insulin concentrations were measured using an ELISA kit (DiaMetra, Milano, Italy) with inter- and intra-assay coefficient variances (CVs) of less than 5%. HOMA-IR and the quantitative insulin sensitivity check index (QUICKI) were determined according to the standard formulas [22]. Enzymatic kits (Pars Azmun, Tehran, Iran) were used to quantify fasting plasma glucose (FPG), serum triglycerides, VLDL-, total-, LDL-, and HDL-cholesterol concentrations with inter- and intra-assay CVs of less than 5%. Serum hs-CRP concentrations were measured using a commercial ELISA kit (LDN, Nordhorn, Germany) with inter- and intra-assay CVs of less than 7%. Plasma nitric oxide (NO) using Griess method [23], total antioxidant capacity (TAC) by ferric reducing antioxidant power method, developed by Benzie and Strain [24], total glutathione (GSH) using Beutler et al.'s method [25], and malondialdehyde (MDA) concentrations were measured using thiobarbituric acid

reactive substances spectrophotometric test [26] with inter- and intra-assay CVs of less than 5%.

At baseline, subjective global assessment (SGA) questionnaire was explained for the participants and completed through face-to-face interview by the main investigator of this study (Z.A). This process was repeated by the same person at the end of the intervention. Then, SGA classifications were converted into numerical equivalents: a score of < 10 points was regarded as well nourished; 10–17 points, at risk for malnutrition or mildly to moderately malnourished; and scores of more than 17 points, severely malnourished [27].

Sample Size

We used randomized clinical trial sample size calculation formula, where type one (α) and type two errors (β) were considered as 0.05 and 0.20 (power = 80%), respectively. According to the previous published trial [28], we used 1.80 as the SD and 1.45 as the mean change (d) of HOMA-IR which was the primary outcome. Using sample size calculation formula, 25 participants were required in each group; allowing for 5 probable dropouts in each group, the final sample size was calculated as 30 participants in each intervention group.

Randomization

Participants were randomized using computer-generated random numbers. Randomization and allocation were concealed from both researchers and patients until the completion of final analyses. The randomized allocation sequence, enrolling participants, and allocating them to interventions were conducted by a trained nutritionist in dialysis clinic.

Statistical Methods

The Kolmogorov-Smirnov test was conducted to determine the normality of data. To detect the differences in anthropometric measures and dietary intakes between treatment groups, we used independent-samples t test. Multiple linear regression models were applied to assess treatment effects on study outcomes, after adjusting for confounding parameters including age and BMI. The effect sizes were presented as the mean differences with 95% confidence intervals. P values < 0.05 were considered statistically significant. All statistical analyses were conducted using the Statistical Package for Social Science version 18 (SPSS Inc., Chicago, IL, USA).

Results

All 60 participants (30 in each group) completed the trial (Fig. 1). The compliance rate was high, since more than 90% of

synbiotic and placebo capsules were taken by study participants during the course of the intervention in both groups. No side effects were reported following the consumption of synbiotic in diabetic patients undergoing HD throughout the study.

At the beginning of the trial, study participants in two intervention groups were comparable in terms of their gender, mean age, height, weight, BMI, and years of dialysis. Moreover, there was a significant difference between two groups in their weight ($P = 0.02$) and BMI ($P = 0.02$) change (Table 1). The reported consumption of anti-diabetic or anti-lipidemic drugs, angiotensin-converting enzyme inhibitors, aldosterone receptor blockers, phosphate binders, residual renal function at baseline and rate of CVD, cerebrovascular disease, hypertension, kidney stone, and cancer at baseline were not statistically different between two intervention groups at baseline or end of the trial.

We found no significant change in dietary macronutrient intakes, total dietary fiber, and total sugar intake throughout the intervention, using 3-day dietary records (Supplemental file 1).

After 12-week intervention, synbiotic supplementation significantly decreased FPG ($\beta - 13.56$ mg/dL; 95% CI, $- 23.82$, $- 3.30$; $P = 0.01$), serum insulin levels ($\beta - 5.49$ μIU/mL; 95% CI, $- 6.92$, $- 4.05$; $P < 0.001$), HOMA-IR ($\beta - 2.25$; 95% CI, $- 3.02$, $- 1.48$; $P < 0.001$), and HbA1c ($\beta - 0.44\%$; 95% CI, $- 0.79$, $- 0.09$; $P = 0.01$) and significantly increased QUICKI (β 0.02; 95% CI, 0.01, 0.02; $P < 0.001$) compared with the placebo (Table 2). Moreover, synbiotic supplementation resulted in a significant reduction in serum hs-CRP ($\beta - 2930.48$ ng/mL; 95% CI, $- 3741.15$, $- 2119.80$; $P < 0.001$) and plasma MDA levels ($\beta - 0.60$ μmol/L; 95% CI, $- 0.99$, $- 0.20$; $P = 0.003$), while a significant increase in plasma TAC (β 142.99 mmol/L; 95% CI, 61.72, 224.25; $P = 0.001$) and

GSH levels (β 131.11 μmol/L; 95% CI, 89.35, 172.87; $P < 0.001$) was observed compared with the placebo. Synbiotic supplementation did not affect other metabolic parameters.

Discussion

In the current study, we investigated the effects of synbiotic supplementation on metabolic profiles in diabetic patients undergoing HD. We found that taking synbiotic for 12 weeks by diabetic patients undergoing HD significantly improved health parameters, including glycemic control, biomarkers of inflammation, and oxidative stress, but did not significantly affect lipid profiles. We have previously reported beneficial effects of probiotic supplementation containing *Lactobacillus acidophilus*, *Lactobacillus casei*, and *Bifidobacterium bifidum* in diabetic patients under HD [29]; however, the synergistic immunomodulatory and anti-inflammatory effects of probiotics and inulin might improve the impact of probiotics on clinical and metabolic symptoms of these patients. Indeed, the effect sizes of 0.47, 0.35, 0.41, 0.11, 0.48, 0.15, 0.05, and 0.14 for FPG, insulin, HOMA-IR, QUICKI, HbA1c, hs-CRP, TAC, MDA, and SGA scores in this study were generally higher than the effects observed in the previous study with probiotics alone, with the effect sizes of 0.12, 0.29, 0.30, 0.29, 0.08, 0.07, 0.08, 0.11, and 0.10, respectively [29].

Effects on Glycemic Control and Lipid Profiles

The findings of this study showed that taking synbiotic supplements for 12 weeks by diabetic patients under HD

Fig. 1 Summary of patient flow diagram
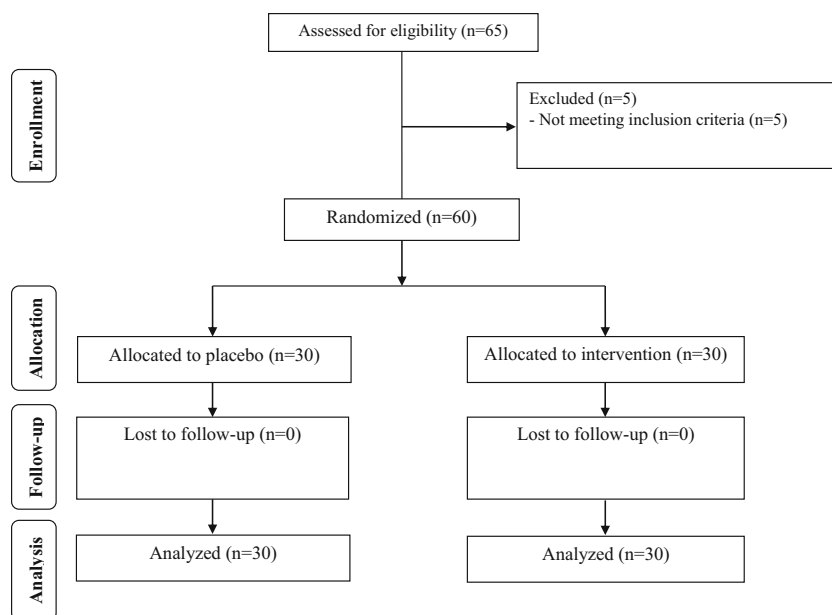
Table 1 General characteristics of study participants

	Placebo group ($n = 30$)	Synbiotic group ($n = 30$)	P^1
Gender (%)			
Male	21 (70.0)	21 (70.0)	1.00[†]
Female	9 (30.0)	9 (30.0)	
Type of diabetes (%)			
Type 1	2 (6.7)	2 (6.7)	1.00[†]
Type 2	28 (93.3)	28 (93.3)	
Age (years)	62.8 ± 14.8	62.8 ± 12.7	0.98
Height (cm)	161.4 ± 10.5	164.8 ± 11.0	0.22
Weight at study baseline (kg)	70.3 ± 14.3	71.9 ± 15.8	0.68
Weight at end-of-trial (kg)	69.8 ± 13.9	72.6 ± 15.4	0.47
Weight change (kg)	− 0.5 ± 1.9	0.7 ± 2.0	0.02
BMI at study baseline (kg/m^2)	26.9 ± 4.7	26.4 ± 5.4	0.70
BMI at end-of-trial (kg/m^2)	26.7 ± 4.6	26.7 ± 5.4	0.96
BMI change (kg/m^2)	− 0.2 ± 0.7	0.3 ± 0.8	0.02
Years on dialysis	3.9 ± 1.2	3.6 ± 1.1	0.50

Data are means ± SDs

[1] Obtained from independent t test

[†] Obtained from Pearson's chi-square test

significantly increased weight and BMI. In a meta-analysis conducted by Million et al. [30], *Lactobacillus acidophilus* administration led to significant weight gain in both humans and animals models. In an animal study conducted by Heo et al. [31], gut microbiota, modulated by probiotics and *Garcinia cambogia* extract, was associated with weight gain. The significant association between protein-energy wasting leading to weight loss and mortality in patients undergoing maintenance HD therapy is well documented [32, 33]. Moreover, there is an increased risk of hospitalization among patients under HD who are underweight [34]. Probiotic supplementation might promote weight gain by altering intestinal flora [35]; however, current evidence is still controversial [36]. In the current study, synbiotic supplementation for 12 weeks to diabetic patients under HD resulted in a significant reduction in FPG, insulin, and HOMA-IR and an increase in QUICKI score, but did not affect lipid profiles. These improvements in glucose homeostasis might be contributed to the observed weight gain in our study. Consistent with our findings, a 28-week synbiotic supplementation with 2×10^8 CFU/day of seven strains of bacteria (*Lactobacillus casei*, *Lactobacillus rhamnosus*, *Streptococcus thermophilus*, *Bifidobacterium breve*, *Lactobacillus acidophilus*, *Bifidobacterium longum*, and *Lactobacillus bulgaricus*) and prebiotics (250 mg fructooligosaccharide) significantly improved glucose metabolism in patients with MetS [15]. Furthermore, in a meta-analysis conducted by Hadi et al. [9], synbiotic supplementation to NAFLD patients decreased fasting glucose and insulin levels, yet did not change HOMA-IR. In contrast to what we found, Rajkumar et al.

[16] reported that probiotic intake containing 2×10^9 CFU/day *Lactobacillus salivarius alone* or in combination with fructooligosaccharide (10 g/day) to healthy volunteers for 6 weeks decreased triglycerides and total- and LDL-cholesterol and increased HDL-cholesterol levels. Furthermore, consumption of a synbiotic containing *Lactobacillus acidophilus* and *Bifidobacterium bifidum* (10^8 CFU/day) plus oligofructose by elderly patients with T2DM increased HDL-cholesterol concentrations [37]. The absence of beneficial effects of synbiotics on lipid profiles in our study might be due to the difference in study participants, baseline levels of measured markers, dosage, and type of probiotic and prebiotic used.

Insulin resistance and hyperglycemia might increase the risk of protein-energy malnutrition, as well as cardiovascular morbidity and mortality in patients with ESRD [38, 39]. Therefore, improved glucose homeostasis might prevent malnutrition, inflammation, and atherosclerosis syndrome, the latter being one of the most important phenomenon in patients undergoing HD treatment. Promising effects of synbiotics on glycemic control might be due to increased production of SCFA, especially butyrate that increases fatty acid beta-oxidation and GLP-1 secretion [17], and modulates the expression of lipogenic and glucogenic gene such as peroxisome proliferator-activated receptor gamma, glucose transporter type 4, and glucose-6-pohspahtase [18]. Probiotics also might decrease toll-like receptor activity, which in turn reduces muscle insulin resistance caused by inflammatory signaling pathway [40].

Table 2 Metabolic profiles, biomarkers of inflammation, and oxidative stress at study baseline and after the 12-week intervention in patients with diabetic hemodialysis that received either synbiotic supplements or placebo

Variables	Placebo group (n = 30)		Synbiotic group (n = 30)		Difference in outcome measures between synbiotic and placebo treatment groups[1]	
	Baseline	Week 12	Baseline	Week 12	β (95% CI)	P^2
FPG (mg/dL)	121.5 ± 43.9	128.2 ± 47.4	123.2 ± 57.6	115.9 ± 43.0	− 13.56 (− 23.82, − 3.30)	0.01
Insulin (μIU/mL)	17.7 ± 9.1	18.9 ± 8.3	18.7 ± 6.1	14.2 ± 6.5	− 5.49 (− 6.92, − 4.05)	< 0.001
HOMA-IR	5.5 ± 3.9	6.1 ± 4.1	6.1 ± 4.4	4.4 ± 3.3	− 2.25 (− 3.02, − 1.48)	< 0.001
QUICKI	0.30 ± 0.02	0.30 ± 0.03	0.30 ± 0.02	0.32 ± 0.02	0.02 (0.01, 0.02)	< 0.001
HbA1c (%)	6.2 ± 0.9	6.1 ± 1.1	6.4 ± 1.3	5.9 ± 1.1	− 0.44 (− 0.79, − 0.09)	0.01
Triglycerides (mg/dL)	137.5 ± 56.6	141.6 ± 64.3	131.5 ± 52.8	123.3 ± 44.7	− 12.52 (− 27.62, 2.57)	0.10
VLDL-cholesterol (mg/dL)	27.5 ± 11.3	28.3 ± 12.9	26.3 ± 10.5	24.7 ± 8.9	− 2.50 (− 5.52, 0.51)	0.10
Total cholesterol (mg/dL)	140.2 ± 41.7	143.0 ± 42.4	142.6 ± 46.1	138.8 ± 30.7	− 5.41 (− 16.98, 6.16)	0.35
LDL-cholesterol (mg/dL)	75.4 ± 37.7	79.3 ± 38.6	84.2 ± 42.8	85.0 ± 30.6	0.18 (− 11.40, 11.77)	0.97
HDL-cholesterol (mg/dL)	37.3 ± 8.5	35.4 ± 9.7	32.0 ± 8.9	29.1 ± 7.7	− 1.96 (− 4.88, 0.94)	0.18
Total-/HDL-cholesterol ratio	3.9 ± 1.3	4.4 ± 2.5	4.6 ± 1.5	5.0 ± 1.5	− 0.10 (− 0.86, 0.66)	0.79
hs-CRP (ng/mL)	5091.0 ± 2700.0	5627.5 ± 2906.6	6008.3 ± 3133.7	3396.7 ± 2562.9	− 2930.48 (− 3741.15, − 2119.80)	< 0.001
NO (μmol/L)	62.2 ± 16.7	62.6 ± 22.6	63.5 ± 13.5	65.9 ± 13.4	2.88 (− 4.89, 10.66)	0.46
TAC (mmol/L)	1233.3 ± 295.8	1183.7 ± 231.9	1223.2 ± 224.5	1318.8 ± 236.7	142.99 (61.72, 224.25)	0.001
GSH (μmol/L)	564.4 ± 221.2	513.8 ± 172.4	654.9 ± 32.6	702.9 ± 56.4	131.11 (89.35, 172.87)	< 0.001
MDA (μmol/L)	3.1 ± 1.0	3.2 ± 1.1	2.4 ± 0.7	2.1 ± 0.6	− 0.60 (− 0.99, − 0.20)	0.003
SGA score	10.5 ± 2.3	10.6 ± 2.3	9.7 ± 2.7	8.6 ± 2.3	− 1.5 (− 2.31, − 0.76)	< 0.001

Data are mean ± SDs

[1] "Outcome measures" refer to the change in values of measures of interest between baseline and week 12. β (difference in the mean outcome measures between treatment groups (synbiotic group = 1 and placebo group = 0))

[2] Obtained from multiple regression model (adjusted for baseline values of each biochemical variables, age, and baseline BMI)

FPG, fasting plasma glucose; *GSH*, total glutathione; *HOMA-IR*, homeostasis model of assessment-insulin resistance; *HDL-cholesterol*, high-density lipoprotein-cholesterol; *Hs-CRP*, high-sensitivity C-reactive protein; *LDL-cholesterol*, low-density lipoprotein-cholesterol; *MDA*, malondialdehyde; *NO*, nitric oxide; *QUICKI*, quantitative insulin sensitivity check index; *VLDL-cholesterol*, very low-density lipoprotein-cholesterol; *SGA*, subjective global assessment; *TAC*, total antioxidant capacity

Effects on Biomarkers of Inflammation and Oxidative Stress

Current findings showed that the consumption of synbiotic by diabetic patients under HD significantly reduced hs-CRP and MDA and significantly increased TAC and GSH, but did not significantly change NO levels. There are discrepant results reporting different effects of synbiotic supplementation on biomarkers of inflammation and oxidative stress. Consistent with our findings, Kleniewska et al. [41] observed that taking synbiotic supplements containing 4×10^8 CFU/day *Lactobacillus casei* and 400 mg inulin for 7 weeks by healthy volunteers significantly improved MDA and GSH concentrations. Moreover, in a meta-analysis conducted by Hadi et al. [9], synbiotic supplementation to NAFLD patients led to a significant reduction in hs-CRP levels. Others have reported that prebiotic and probiotic supplementation decreased oxidative stress [42, 43], while some of them failed to detect a significant effect of probiotics on antioxidant status [44, 45]. Our previous study indicated that synbiotic supplementation

containing three probiotic bacteria species *Lactobacillus acidophilus*, *Lactobacillus casei*, and *Bifidobacterium bifidum* (2×10^9 CFU/g each) plus 800 mg inulin significantly reduced hs-CRP and MDA and increased NO levels in diabetic patients, but did not affect other antioxidant markers [20]. In contrast to our findings, synbiotic supplementation did not decrease hs-CRP levels in women with polycystic ovary syndrome [46] or overweight children [21]. High serum levels of CRP in patients under HD are associated with all-cause and CVD mortality [47]. Inflammation and elevated oxidative stress presented in ESRD patients aggravate disease condition and are associated with a large number of ESRD complications, including protein-energy wasting and atherosclerosis [48]. Current evidence has demonstrated that probiotic and synbiotic intake may decrease the production of pro-inflammatory cytokines, inhibit lipid peroxidation such as MDA, and increase the activity of antioxidant enzymes [18]. In the current study, we observed a significant reduction in inflammation after synbiotic ingestion which was accompanied by the activation of G protein-coupled receptor 41 and

GPR43 [49], and reduction of the activity of nuclear factor-κB [50]. The up-regulation of gene expression of interleukin-18 by SCFA [51] and increased production of methylketones family in the colon, following the intake of synbiotic, [52] might explain its anti-inflammatory effects. Unfortunately, we did not measure SCFA levels in the current study as a mechanism for the observed effects, but this measurement is recommended in future studies.

This study had several limitations. We were not able to measure fecal bacteria loads before and after probiotic supplementation. Due to lack of appropriate funding, we could not quantify circulating interleukin-6 and tumor necrosis factor-α levels and gene expression related to insulin and inflammation signaling pathway in diabetic patients under HD. In addition, we did not measure SCFA and GLP-1 levels as a mechanism for the observed effects. In the current study, all participants were from a limited area and similar genetics. Renal and metabolic health, among all health parameters, was associated with many factors including genetics. This should be considered in our findings interpretation.

Conclusions

Overall, we found that taking synbiotic for 12 weeks by diabetic patients undergoing HD significantly improved health parameters, including glycemic control and biomarkers of inflammation and oxidative stress, but did not significantly affect lipid profiles. Our findings suggest that synbiotic supplementation may be relevance valuable therapeutic agent for diabetic patients undergoing HD. Further research is recommended to confirm the beneficial effects of synbiotic supplementation in other populations.

Compliance with Ethical Standards

Ethics Approval and Consent to Participate All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and national research committee and with the 1964 Helsinki declaration and its later amendments.

Consent for Publication Not applicable.

Availability of Data and Material The primary data for this study is available from the authors on direct request.

Competing Interests The authors declare that they have no conflict of interest.

Abbreviations ESRD, end-stage renal disease; FPG, fasting plasma glucose; GLP1, glucagon-like peptide-1; GSH, total glutathione; HD, hemodialysis; HOMA-IR, homeostasis model of assessment-insulin resistance; HDL-cholesterol, high-density lipoprotein-cholesterol; hs-CRP, high-sensitivity C-reactive protein; LDL-cholesterol, low-density lipoprotein-cholesterol; MDA, malondialdehyde; NO, nitric oxide; QUICKI, quantitative insulin sensitivity check index; SCFA, short-chain fatty acids; SGA, subjective global assessment; T2DM, type 2 diabetes mellitus; TAC, total antioxidant capacity; VLDL-cholesterol, very-low-density lipoprotein-cholesterol

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