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Meta-analyses

Probiotic supplementation for management of cardiovascular risk factors in adults with type II diabetes: A systematic review and meta-analysis

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SUMMARY

Background & aims: The effectiveness of probiotics in control of hypertension and dyslipidemia in diabetic patients remains unclear. Therefore, we systematically reviewed relevant data to elucidate the effects of probiotics on blood pressure and lipid profile of type 2 diabetic patients.
Methods: We searched PubMed, ISI Web of Knowledge, Scopus, The Cochrane Library, ClinicalTrials.gov, ProQuest Dissertations and Theses databases until May 2016. The primary outcomes were systolic blood pressure (SBP) and diastolic blood pressure (DBP), total cholesterol (TC), low-density lipoprotein cholesterol (LDL-C), high-density lipoprotein cholesterol (HDL-C) and triglycerides (TG). Other biochemical response and adverse effects were considered as secondary outcomes. Data was extracted from included studies and pooled in meta-analysis whenever possible (both standardized mean difference (SMD) analysis and weighted mean difference (WMD) analysis were performed).
Results: Eleven eligible randomized controlled trial (n = 641) were identified. Pooling data from these trials demonstrated probiotic consumption significantly decreased SBP (WMD, −3.28 mmHg; 95% confidence interval [CI], −5.38 to −1.18), DBP (WMD, −2.13 mmHg; 95% CI, −4.5 to 0.24), LDL-C (WMD, 8.32 mg/dl; 95% CI, −15.24 to −1.4), TC (WMD, −12.19 mg/dl; 955 CI −17.62 to −6.75) and TG (WMD, −24.48 mg/dl; 95% CI, −33.77 to −11.18) in type 2 diabetic patients compared with placebo. The methodological quality varied across trials included in this study.
Conclusion: This systematic review suggests probiotics supplementation may be helpful for control of dyslipidemia and hypertension in type 2 diabetic patients. Conducting more trails with large sample size and long follow-up time still is necessary to develop clinical practice guidelines for management of cardiovascular risk factors in patient with type 2 diabetes.

1. Introduction

Diabetic patients encounter several challenges due to insulin deficiency or insulin resistance. They are at higher cardiovascular risks such as dyslipidemia and raised blood pressure [1]. Triglyceride level is higher in diabetic patients because of impaired action of insulin and is one of the reasons for raised low density lipoprotein cholesterol (LDL-C) production. Furthermore LDL-C particles can be glycated in diabetics increasing LDL-C half-life while, glycation of high density lipoprotein cholesterol (HDL-C) decrease its half-life; and all these contribute to cardiovascular events [2]. Insulin resistance decreases nitric oxide bioavailability in endothelial cells and hyperglycemia inhibits nitric oxide production in arterial endothelial cells, these events leads to vasoconstriction and consequent hypertension and also release of pro-inflammatory cytokines [3,4]. Considering complexity of diabetic conditions and its raising prevalence, many attempts have been made for finding new effective agents.

Recent evidence shows altered intestinal microbiome in patients with type 2 diabetes [5]. The gut dysbiosis can lead to metabolic disturbances because more than 1000 phylotypes in the human gut, cooperates in dietary metabolism with the host [6,7]; animal models also imply the importance of gut microbiota in

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regulation of fatty acid uptake and hepatic lipogenesis [8,9]. In this regard, several experiments explored the effects of probiotics consumption on controlling cardiovascular risk factors in patients with type 2 diabetes, and reported interesting results [10–12]. Therefore, this review was conducted to systematically review human trials in which effects of probiotics supplementation were investigated on management of blood pressure and plasma lipid levels among patients with type 2 diabetes. The data were then properly synthesized by meta-analysis.

2. Methods

This systematic review was conducted and reported according to Cochrane [13] and PRISMA [14] guidelines, respectively. Pre-specified protocol of this review was published in PROSPERO with a registration number of CRD42015025517.

2.1. Data sources and search strategies

Search strategies were defined according to specified PICOT criteria (presented in Supplementary Table 1) and adjusted for each of the following electronic databases: PubMed, ISI Web of Knowledge Scopus,, The Cochrane Library, ClinicalTrials.gov, ProQuest Dissertations and Theses. The last update was done at 06/05/2016 (no language restriction was applied).

2.2. Eligibility criteria and study selection

Obtained results after applying above strategies were entered in EndNote software. Duplicates were removed and final library was prepared for further screening.

Clinical trials in which probiotics in the form of any pharmaceutical formulations or dairy products administered to adult diabetic patients were included after title and abstract screening. In the next step full texts were reviewed for inclusion of eligible studies; all clinical trials that divided patients into intervention and placebo (same product as intervention excluding probiotics content) groups, and measured any of the following items were included in the systematic review: the plasma levels of total cholesterol (TC), LDL-C, HDL-C, triglycerides (TG), systolic blood pressure (SBP) and diastolic blood pressure (DBP) as primary outcomes. Other biochemical response and adverse events were considered as secondary outcomes.

All steps were performed independently by the two authors (F.H and V.A) and any disagreement was discussed for final decision.

2.3. Data items and data collection process

Following data was collected and documented by two reviewers (F.H and V.A) independently: 1. Study identification and design information including author name, year of publication, trial registration number, country, study design, duration of intervention and follow-up, number of participants in each group, interventions (type and dose of probiotics), 2. Characteristics of the participants including, mean age and BMI, 3. Their findings including primary outcomes, secondary outcomes and adverse effects. In the case of deficient data or no access, corresponding authors were contacted properly.

2.4. Assessment of risk of bias

We assessed risk of bias of individual studies using Cochrane Risk of Bias tool [15]. Following criteria were evaluated by the two authors (F.H and V.A): random allocation, masking or blinding, selection bias caused by attrition, selective reporting of outcomes, assessment of patient's compliance, and monitoring food intake and basic parameters.

2.5. Data synthesis and analysis

Extracted data from eligible studies were entered into the STATA 11.2 software (Stata Corp, College Station, TX) for statistical analysis. Mean differences in outcomes between baseline and after intervention which were reported in at least 3 qualified studies were used for meta-analysis. Due to high heterogeneity, random effect analysis was performed [16] to obtain standardized mean differences (SMD); and $P < 0.05$ was considered as significant result. Weighted mean differences (WMD) were also calculated to present data pooling results in original units of measurements for more applicable interpretation of results in clinical settings.

Heterogeneity was evaluated by Cochran's Q-test [17] and reported as I square (I^2) [18]. The I^2 value has a range of 0–100%, which can be classified into following groups: $I^2 < 25\%$, $25\% < I^2 < 75\%$ and $I^2 > 75\%$ which represent a low, moderate and high degree of heterogeneity, respectively. When I^2 value was more than 25%, possible reasons for heterogeneity was investigated using one or more of the following ways: 1. Sensitivity analysis (to identify which trial(s) is causing the heterogeneity and how each trail contributes to the overall analysis) 2. Sub-group analysis (according to the characteristics of patients, intervention or study) 3. Random effect mete-regression (to identify which trial-level variables give a reason for the heterogeneity).

To assess possible publication bias in each analysis, we used Begg and Egger's tests (P value less than 0.05 was reported as significant), and visual inspection of funnel plots [19].

3. Results

3.1. Included studies and their characteristics

3964 records were detected, from which 800 was removed after duplicate deletion. Screening steps resulted in entering 13 eligible studies in systematic review and 11 in the meta-analysis (Fig. 1) including 10 randomized controlled trials (RCTs) [10–12,20–26] and 1 single blinded trial [27] and 2 crossover trials [28,29]. These articles were published from Iran [10–12,23–27], Brazil [21], Malaysia [20], Denmark [22], Saudi Arabia [28], and USA [29]. Reasons for exclusion of some studies after full text screening were presented in Supplementary Table 2. Data from 641 type 2 diabetic patients entered in analysis including 352 in intervention and 358 in placebo groups.

Probiotics were administered in different forms including fermented milk or yogurt [10–12,21–23,25], bread [24], tablet [26,29], and freeze-dried powder in the capsule [27,28] or sachet [20]. Microorganism daily dose were reported as cfu/gram or cfu/litter, or germ/volume of the supplement. Three studies administered *Saccharomyces cerevisiae* [26,28,29], other studies administered one bacterial species [12,22,24], two [10,21], three [23], four [11,27], five [25], or six [20] bacterial species (Table 1).

3.2. Risk of bias of individual studies

Two studies were of poor quality and therefore excluded from meta-analysis [27,29]. Two were fair [25,28] and the rest were good [10–12,20–24,26]. Randomization performed appropriately in 10 [10–12,20–24,26,27], in 2 studies no method was mentioned for randomization [25,28], and in one study patients were distributed according to clinical history and observations [29]. Blinding was applied in 12 studies and design of one study was not proper to conserve blinding [25]. Attrition bias was detected in 3 studies due

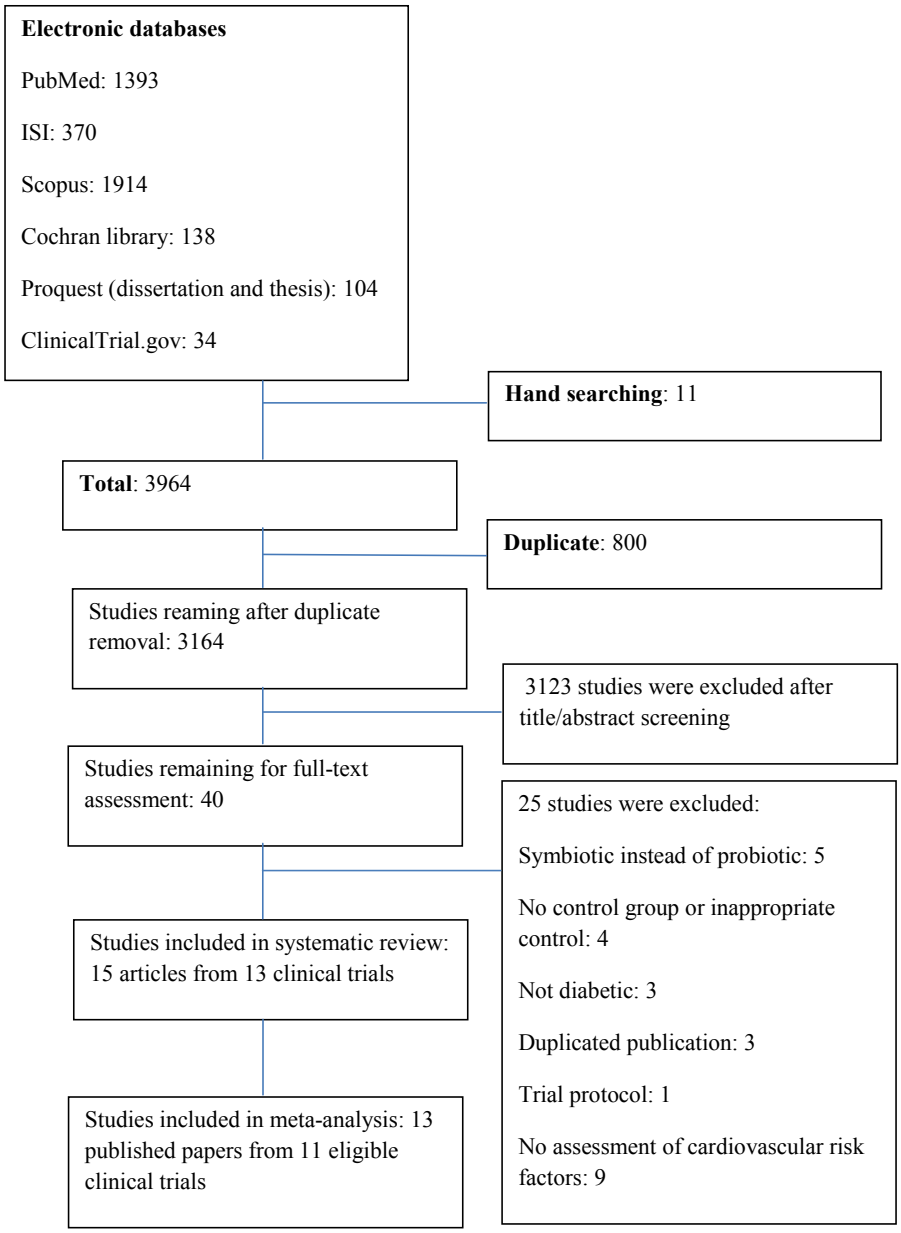

Fig. 1. Research flow diagram of systematic review.

to missing several patients during study period [27–29]. Almost all studies control patients' dietary intake except one [27]. In two studies adherence of patients to experiment plane was not monitored [25,27]. Nine studies were registered as clinical trials from which 3 did not present outcomes completely [22,23,29], 3 was not registered anywhere [11,27,28], and registration number that was presented by one study [21] was not found in related address (Table 2).

3.3. Meta-analysis, meta-regression, sensitivity analysis

3.3.1. Plasma lipid profile (TG, TC, LDL-C and HDL-C level)

Ten studies reported plasma TG level. Data pooling showed significant effect of probiotic use on reducing plasma TG level (n = 10; SMD = −1.028; 95% CI: −1.669, −0.387; $P = 0.002$) (Fig. 2). Heterogeneity test results however was not acceptable (n = 10; $I^2 = 92.9\%$; $P = 0.000$) and needed more investigation. A sensitivity analysis in which studies with fair quality [25,28] were omitted, reduced heterogeneity up to 53.2%, while TG reduction effect was not affected (n = 8; SMD = −0.528; 95% CI: −0.8, −0.256; $P = 0.000$). Age seemed to be important factor; with increasing mean age of participants, in 8 studies that reported this item, TG lowering effect became less significant (n = 8; $tau^2 = 0$; $P > t = 0.029$; $I^2_{res} = 13.35\%$). Meta-regression analysis regarding both age and BMI removed heterogeneity (n = 8; $tau^2 = 0$; $Prob > f = 0.043$; $I^2_{res} = 0\%$) (Fig. 2).

Ten studies reported plasma total cholesterol level. Probiotic administration leaded to significant TC reduction (n = 10; SMD = −0.860; 95% CI: −1.247, −0.472; $P = 0.000$) (Fig. 3). Data were again heterogenic (n = 10; $I^2 = 81.8\%$; $P = 0.000$). Sub-group analysis according to type of microorganism removed heterogeneity in yeast group without affecting TC lowering effect (n = 2; SMD = −0.600; 95% CI = −0.866, −0.333; $P = 0.000$. $I^2 = 0.0\%$; $P = 0.459$); however data in bacteria group was still highly heterogenic (n = 8; $I^2 = 85.5\%$; $P = 0.000$), further sub-grouping by separating studies in which one bacterial species was used showed

Table 1
Characteristics of included studies in systematic review.

Author/date	N. of participants (intervention/placebo)	Administered probiotics	Targeted outcomes	Type of study	Trial number	Overall quality[a]
Firouzi/2016 (Firouzi et al. [20])	136 (48/53)	Each sachet containing 3×10^{10} *Lactobacillus acidophilus*, *Lactobacillus casei*, *Lactobacillus lactis*, *Bifidobacterium bifidum*, *Bifidobacterium longum* and *Bifidobacterium infantis* (10^{10} cfus/day).	serum lipid profile, SBP, DBP	Randomized, double-blind, parallel-group, controlled clinical trial	NCT01752803	Good
Tonucci/2015 (Tonucci et al. [21])	50 (23/22)	120 g/d of fermented milk containing *Lactobacillus acidophilus LA5* and *Bifidobacterium lactis BB12,* for 6 weeks.	serum lipid profile, TC/HDL-C, LDL-C/HDL-C, adiponectin	Randomized, double-blind, parallel-group, placebo controlled trial	ensaiosclinicos.gov.br/rg/RBR-219644	Good
Hariri/2015 (Hariri et al. [12])	48 (20/20)	Soy milk enriched with *L. plantarum A7* (2×10^7) (200 ml/day)	SBP, DBP	Randomized double-blind, placebo-controlled	IRCT201405265062N8	Good
Hove/2015 (Hove et al. [22])	41 (23/18)	Milk fermented with *L. helveticus* (Cardi04 yogurt) 300 ml/day for 12 weeks.	plasma lipids, SBP, DBP	Randomized, double-blinded, prospective, placebo-controlled study	NCT00699426	Good
Ostadrahimi/2015 (Ostadrahimi et al. [23])	60 (30/30)	Probiotic fermented milk (*Lactobacillus casei, Lactobacillus acidophilus, Bifidobacteria*) 600 ml/day for 8 week	serum lipid profile	Randomized double-blind placebo-controlled clinical trial	IRCT201307092017N14	Good
Shakeri/2014 (Shakeri et al. [24]; Bahmani et al. [44])	81 (26/26)	Probiotic bread (*Lactobacillus sporogenes*: 1×10^8 cfu/gr), 120 g/day for 8 weeks.	serum lipid profile, TC/HDL-C, Non-HDL-C, SBP, DBP	Randomized, double-blinded, controlled clinical trial	IRCT201311215623N13	Good
Mohamadshah/2014 (Mohamadshahi et al. [11])	42 (21/21)	Probiotic yogurt (*Lactobacillus acidophilus La-5*, *Bifidobacterium lactis Bb-12,* 3.7×10^6 cfu/mg of both, 300 g/day for 8 weeks	serum lipid profile	Randomized double-blind, controlled clinical trial	–	Good
Bayat/2014 (Bayat et al. [25])	80 (20/20)	Probiotic yoghurt (*Strep. thermophilus, Lactobacillus thermophilus, L bulgaricus, L acidophilus, bifidobacterium,* at least 10^6bacteria/g of yogurt) 150 g/day	Serum lipid profile, SBP and DBP	Parallel randomized clinical trial	IRCT2013041311763N7	Fair
Hosseinzadeh/2013 (Hosseinzadeh et al. [26])	84 (42/42)	*Saccharomyces cerevisiae,* 1800 mg/day (6 tablets) for 12 weeks	serum lipid profile, SBP, DBP	Double-blind, randomized, clinical trial	RCT138807062513N1	Good
Mazloom/2013 (Mazloom et al. [27])	34 (16/18)	Probiotic capsules.(*L. acidophilus, L. bulgaricus, L. bifidum, L. casei.*) 3000 mg/day for 6 weeks	plasma lipid profile,	Single-blinded clinical trial	–	Poor
Ejtahed/2012 (Ejtahed et al. [10,45])	60 (30/30)	probiotic yogurt enriched with *B. lactis Bb12, L. acidophilus La5*, 300 g/day for 8 weeks	serum lipid profileSBP, DBP	Randomized, double-blind, controlled clinical trial	IRCT 138903223533N1	Good
Bahijiri/2000 (Bahijiri et al. [28])	78 (74/69)	Brewer's yeast as capsule (23.3 μg Cr/day), for 8 weeks	serum lipid profile.	Randomized, double blind cross-over	–	Fair
Rabinowitz/1983 (Rabinowitz et al. [29])	43 (28/58)	brewer's yeast extract (tablet) for 4 month	TC, TG,	Double-blind, random crossover	–	Poor

T2DM: type 2 diabetes mellitus, SBP: systolic blood pressure, DBP: diastolic blood pressure, HDL-C: high density lipoprotein cholesterol, LDL-C: low density lipoprotein cholesterol.
[a] Overall quality of studies with available full texts were obtained after risk of bias assessment according to the criteria provided in Cochrane Handbook. Details are presented in Table 2.

Table 2
Risk of bias assessment of included studies according to Cochrane Risk of Bias tool.

Study	Random allocation	Blinding	Selection bias due to attrition	Monitoring food intake and basic parameters	Compliance assessment	Selective report	Overall quality
Rabinowitz 1983 [29]	−	+	+	−	−	+	poor
Bahijiri 2000 [28]	?	+	+	−	?	+	Fair
Ejtahed 2012 [45]	+	+	+	+	+	+	Good
Hosseinzadeh 2013 [26]	+	+	+	+	+	+	Good
Mazloom 2013 [27]	+	+	−	−	?	−	Poor
Mohamadshahi 2014 [11]	+	+	+	+	?	+	Good
Bayat 2014 [25]	?	−	−	+	+	+	Fair
Shakeri 2014[24]	+	+	+	+	+	+	Good
Ostadrahimi 2015 [23]	+	+	+	+	−	+	Good
Hove 2015 [22]	+	+	+	+	−	+	Good
Hariri 2015 [12]	+	+	+	+	+	+	Good
Tonucci 2015 [21]	+	+	+	+	?	+	Good
Firouzi 2016 [20]	+	+	+	+	+	+	Good

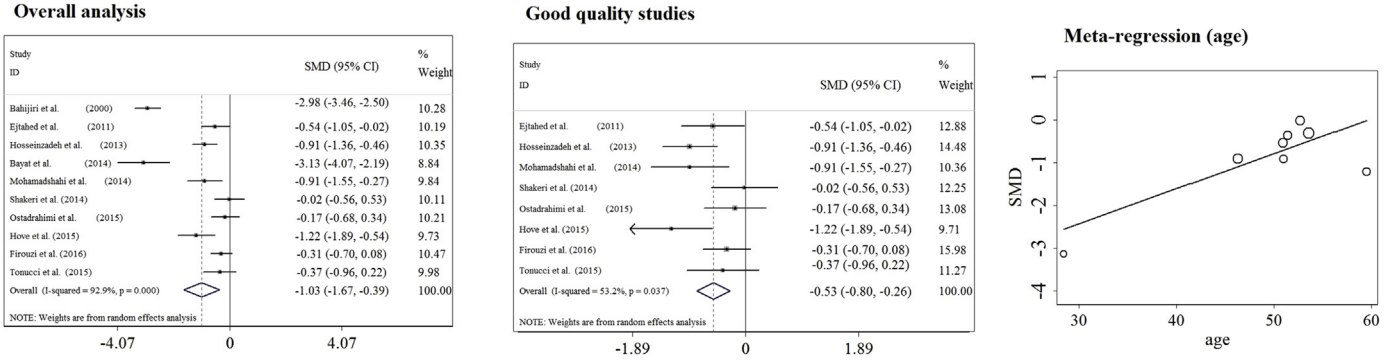

Fig. 2. Forest plot for overall data pooling of TG level form eligible studies (left), Forest plot for pooling data of TG level form good quality studies (middle), meta-regression graph regarding age (right).

that single bacterium cannot reduce TC level significantly (n = 2; SMD = −0.219; 95% CI = −0.629, 0.191; $P = 0.296$), multiple species group still contained heterogenic data. Meta-regression analysis displayed that mean age and basic total cholesterol level were also related with response to probiotics supplementation; TC lowering effect decreased by increasing age (n = 8; $tau^2 = 0.0588$; $P > t = 0.063$; $I^2_{res} = 21.54\%$) and basic total cholesterol level (n = 9; $tau^2 = 0.0473$; $P > t = 0.160$; $I^2_{res} = 26.57\%$) (Fig. 3).

Nine studies reported LDL-C level. Overall meta-analysis showed significant difference between LDL-C level in placebo and probiotic groups (n = 9; SMD = −0.869; 95% CI: −1.685, −0.053; $P = 0.037$) (Fig. 4), but highly heterogenic data ($I^2 = 94.4\%$; $P = 0.000$) necessitated further analysis. In a sensitivity analysis one study with fair quality was removed [25] and then meta-regression according to mean age of participants was performed; results displayed negative relationship between age and LDL-C lowering effect of probiotics and heterogeneity reduced to 31.25% (n = 7; $tau^2 = 0.1488$; $P > t = 0.008$) (Fig. 4).

Ten studies reported HDL-C level. Whole data pooling resulted in non-significant difference between HDL-C change in control and intervention groups (n = 10; SMD = 0.913; 95% CI: −0.2, 2.027; $P = 0.108$) (Fig. 5); while significant heterogeneity observed between studies (n = 10; $I^2 = 97.3\%$; $P = 0.000$). Subgroup analysis according to BMI revealed that beneficial effect of probiotic supplementation on HDL-C level was significant in patients with BMI equal or more than 29 (n = 5; SMD = 1.873; 95% CI = 0.236, 3.509; $P = 0.025$), in opposite no difference was observed within group with BMI less than 29 (n = 5; SMD = −0.063; 95% CI = −1.321, 1.195; $P = 0.922$). However heterogeneity is still high within groups (Fig. 5). Different BMI of studies within these two groups was again effective in producing high heterogeneity as detected by meta-regression; after removing one heterogeneous study [11], meta-regression according to BMI reduced heterogeneity to 67.35% (n = 9; $tau^2 = 0.5856$; $P > t = 0.001$). In another sensitivity analysis after removing one study [25] and carrying out meta-regression using mean age of participants, significant effect of this parameter

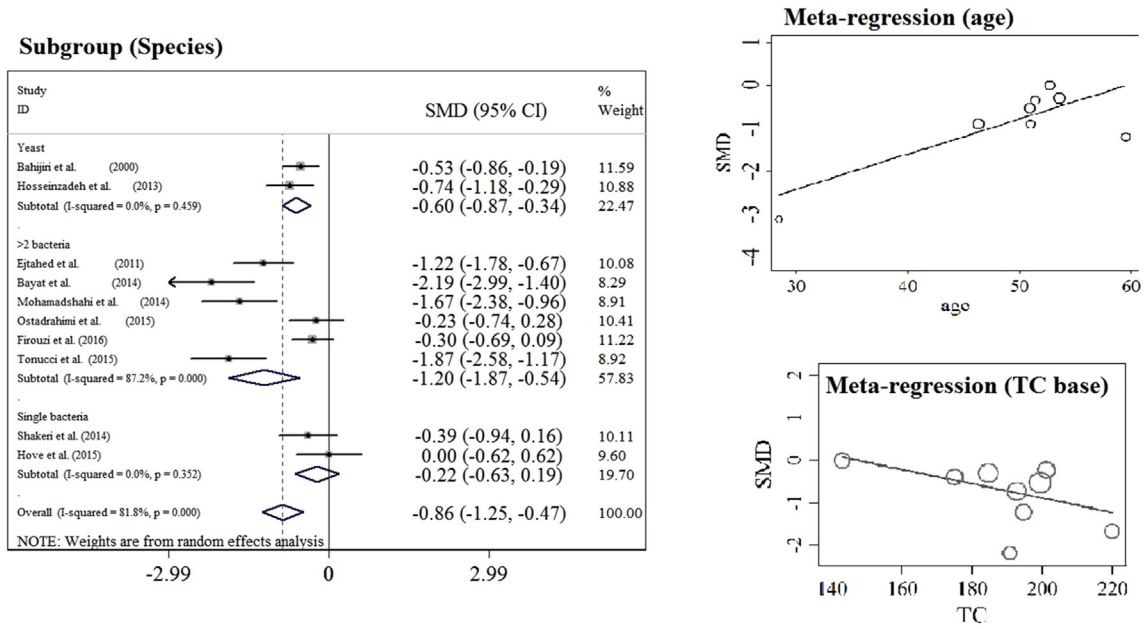

Fig. 3. Forest plot for overall data pooling of TC level form eligible studies and sub-grouping according to type of microorganism (left), meta-regression graph regarding age (middle), meta-regression regarding TC base of participants (right).

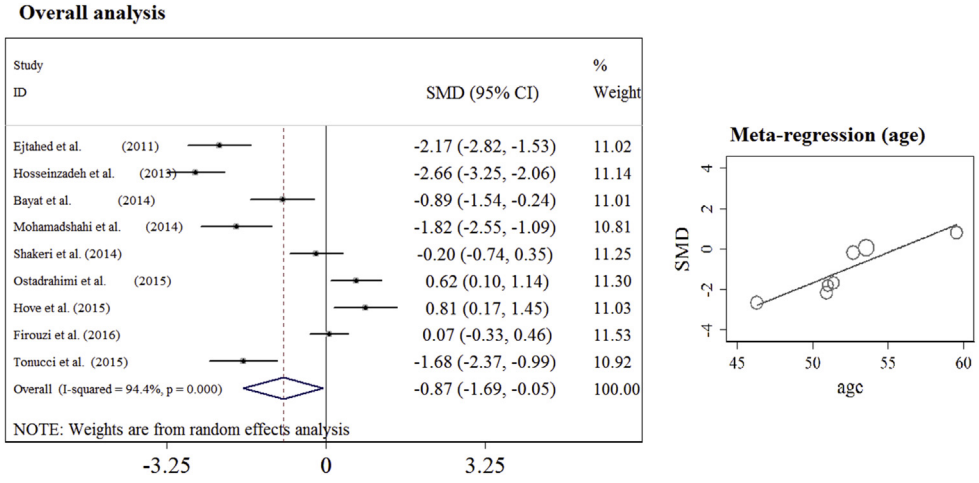

Fig. 4. Forest plot for overall data pooling of LDL-C level form eligible studies (left), meta-regression graph regarding age (right).

was also appeared (n = 7; tau^2 = 0.6032; $P > t$ = 0.016; I^2_{res} = 64.95%) (Fig. 5).

3.3.2. Systolic and diastolic blood pressure

Seven studies reported SBP. Data pooling detected significantly reduced SBP post-intervention (n = 6; SMD = −0.928; 95% CI = −1.582, −0.274; P = 0.005) with high heterogeneity (n = 7; I^2 = 89.6%; P = 0.000) (Fig. 6). One of seven studies used *S. cerevisiae*, and others used bacterial species. When meta-regression regarding BMI was performed for bacteria group, heterogeneity reduced to 28.52%; that showed the effect of BMI difference on the blood pressure lowering of bacterial probiotics (n = 6; tau^2 = 0.084; P = 0.057) (Fig. 6). Actually SBP in more obese patients showed less significant change. Sub-group analysis according to BMI also confirmed this result; changes of SBP was not significant among patients with BMI equal or more than 29 (n = 3; SMD = −0.093; 95% CI = −0.361, 0.175; P = 0.497), however the effect was significant in patients with BMI less than 29 (n = 3; SMD = −1.622; 95% CI = −2.572, −0.671; P = 0.001).

Seven studies reported DBP. Data pooling of these 7 trials displayed significant difference between placebo and intervention groups (n = 7; SMD = −0.882; 95% CI = −1.758, −0.007; P = 0.048); however high heterogeneity exists between studies' results (n = 7; I^2 = 94%; P = 0.000) (Fig. 7). Sub-group analysis regarding basic BMI removed heterogeneity and revealed significant effect of probiotic supplementation in patients with BMI less than 29 (n = 3; SMD = −1.212; 95% CI = −1.820, −0.604; P = 0.000; I^2 = 57.7%) in opposite non-significant results obtained for patients with BMI equal or more than 29 (n = 4; SMD = −0.638; 95% CI = −1.989, 0.713; P = 0.355; I^2 = 96.4%); removing the study with yeast as intervention reduced heterogeneity in high BMI group (n = 3; SMD = 0.126; 95% CI = −0.393, 0.645; P = 0.092; I^2 = 71.6%). Meta-regression in bacteria group regarding BMI also confirmed results from sub-group analysis (n = 6; tau^2 = 0.3049; $P > t$ = 0.251; I^2_{res} = 51.41%)(Fig. 7).

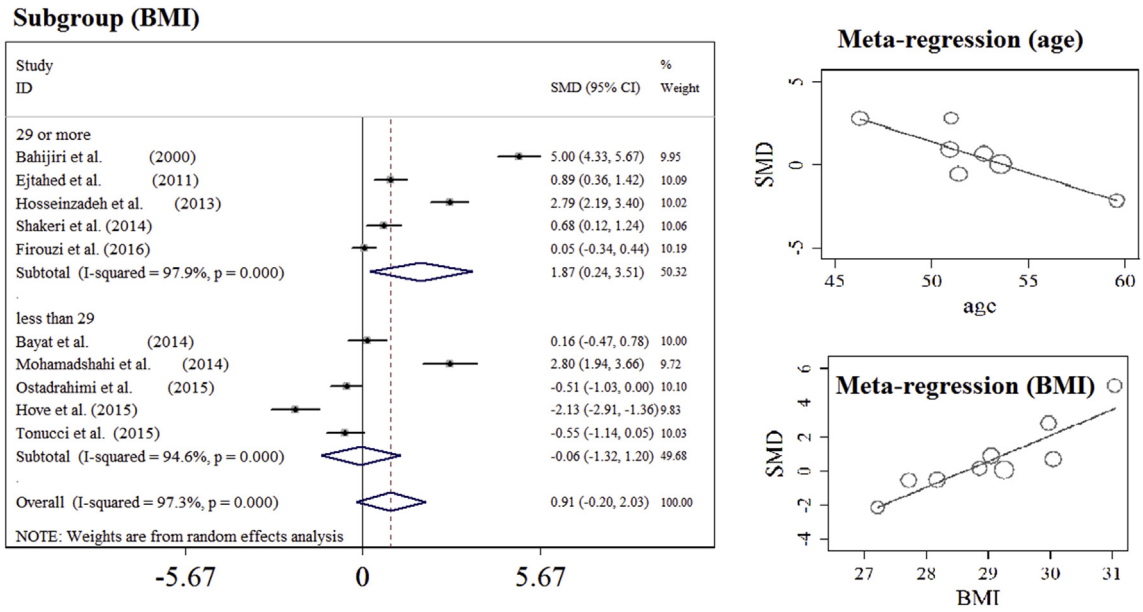

Fig. 5. Forest plot for overall data pooling of HDL-C level form eligible studies and sub-grouping according to BMI (left), meta-regression graph regarding age (upper right), meta-regression graph regarding BMI (lower right).

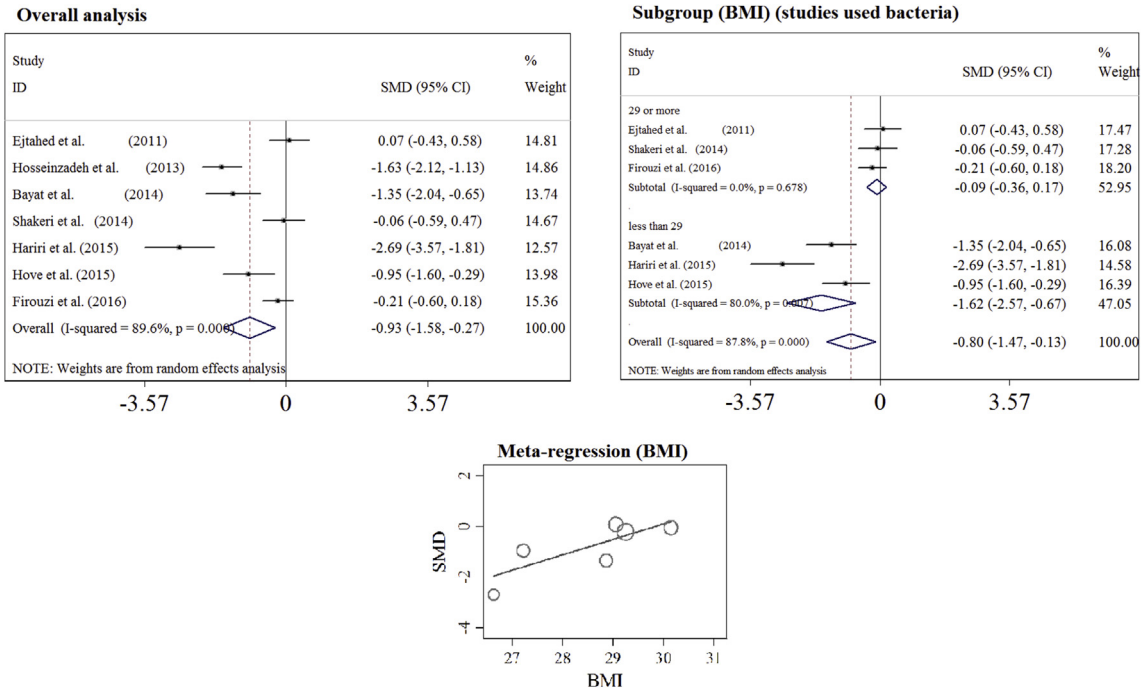

Fig. 6. Forest plot for overall data pooling of SBP level form eligible studies (left), sub-grouping studies that used bacteria according to BMI (middle), meta-regression graph regarding BMI (right).

3.3.3. Publication bias

Funnel plots were displayed in Supplementary Fig. 1 Begg and Egger's tests did not show significant publication bias in any of the analysis.

3.4. Other laboratory data

Very low density lipoprotein (VLDL) cholesterol level was decreased after probiotic supplementation [24], resistin also decreased significantly [21] while adiponectin fall was non-significant [21,30], fat distribution and liver steatosis were also reported unchanged [30]. Fat cell size tended to be decreased; however the change was non-significant [30].

3.5. Adverse events

Just five studies gave reports of adverse events in their results. Three studies reported minor gastrointestinal discomfort such as diarrhea and flatulence [20–22], which leaded to withdrawal of one participant from intervention group in one of the studies [21].

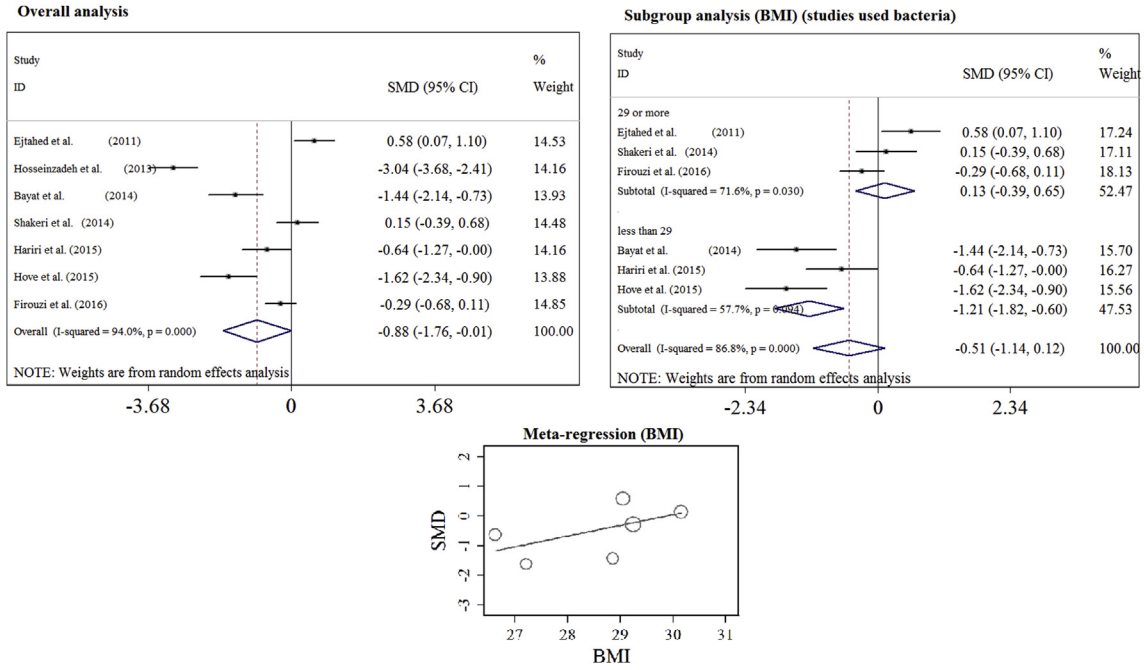

Fig. 7. Forest plot for overall data pooling of DBP level form eligible studies (left), sub-grouping studies that used bacteria according to BMI (upper right), meta-regression graph regarding BMI (lower right).

In two trials it was just stated that probiotics were well tolerated by diabetic patients and no serious event was detected [24,29].

4. Discussion

Results of our meta-analysis found that administration of probiotics can be helpful in reduction of blood lipids and blood pressure in patients with type 2 diabetes. We found that probiotics can significantly reduce TG and TC and LDL-C blood level; however, the impact of probiotics on increasing HDL-C was not statically significant and it depended on patient BMI and age. In addition, pooling analysis of seven trails showed that probiotic supplementations significantly reduced SBP and DBP.

WMD analysis revealed that probiotics consumption reduced TG level up to 24.479 mg/dl, TC level up to 12.188 mg/dl, and LDL-C up to 8.32 mg/dl in diabetics. Our findings were in agreement with another study reported that probiotic supplementation did not exhibit a significant effect on lipid profile of patients with type 2 diabetes [31]. They could find only nine trials in their review; although have included seven of the thirteen studies included in our review. They included trails which their intervention was symbiotics, a supplement containing both probiotics and prebiotics, and also studies with poor quality (identified by Cochrane's risk of bias assessment tool). Furthermore, Le et al. included only studies published in English language journals which potentially results in language bias. Finally, they did not report effect of probiotic supplementation on blood pressure of type 2 diabetic patients.

High blood pressure (hypertension) contributes to high prevalence of cardiovascular diseases in type 2 diabetic patients. Diabetic adults are two times more likely to have increased blood pressure than adults without diabetes [32]. There is a positive correlation between hypertension and insulin resistance [33]. Furthermore, the coexistence of both conditions enhances the incidence of cardiovascular disease [34]. The results of current review showed that consuming probiotics could significantly reduce SBP by 3.278 mmHg and DBP by 2.131 mmHg in patients with type 2 diabetes. The reduction observed in the present study was mild; however, even a small reduction of blood pressure may associate with decrease in incidence of stroke and coronary heart disease [35].

The antihypertensive mechanisms of probiotics are complex and not fully understood. Blood pressure lowering effect of some probiotics may be mediated by releasing angiotensin converting enzyme inhibitory peptides [36]. Other mechanisms such as biotransformation of phytoestrogens (which can act as vasodilatory factors) and improvement of insulin sensitivity and lipid profile may also explain the antihypertensive effect of probiotics [37].

Our sub-group analysis according to BMI also showed that effectiveness of probiotics consumption on blood pressure in more obese diabetic patients was less significant. Previous study also reported high blood pressure in obese participants (BMI >30 kg/m^2) were less controlled (approximately 30%) compared with non-obese ones (BMI <25 kg/m^2) [38]. These observations, along with ours, suggest that obesity may cause more difficulty in controlling high blood pressure and weight loss could improve antihypertensive effect of probiotics.

The current meta-analysis indicated a stronger effect of probiotics on TC, TG and LDL-C than on HDL-C. Our finding was similar to that reported in a recent meta-analysis trying to assess efficacy of probiotics in reduction of lipid plasma levels in people with mild hypercholesterolemia [39]. They reported HDL-C and TG levels were not significant different between the probiotic intervention and control groups. It has been suggested that probiotics may reduce cholesterol in the form of cholesteryl esters, via alteration of lipid transporters rather than affecting cholesterol synthesis in the liver [37]. Although the mechanisms by which probiotics may improve lipid profile have not been fully explained, reduction of cholesterol absorption and bile acid reabsorption in intestine could be involved [40].

Our study suggested the improvement lipid profile in patients with type 2 diabetes depended on a variety of factors including patient's characteristics (age and BMI) and probiotic dosage.

Meta-regression using mean age of participants showed that improvement of lipid profile by probiotic supplementation in younger individuals was more significant than that in the elderly. Considering higher cholesterol baseline value in the elderly, it could be more difficult to address high cholesterol in old patients.

According to sub-group analysis the effectiveness of probiotic on management of lipid profile is influenced by patient's BMI Probiotic supplementation showed more beneficial effect on lipid profile (TG, HDL-C) of obese patients which may be due to restoring balance and harmony to the gut microbiome. Dysbiosis, alterations of the collection of microbes in the gut which mostly observed in obese and dyslipidemic patients, has previously been demonstrated to be restorable by probiotics or prebiotics consumption. Vrieze et al. [41] performed a randomized double-blind controlled trial and reported that intestinal microbiota transplantation from non-obese donors significantly reduced fasting triglyceride levels in obese patients with metabolic syndrome. For future studies it would be useful to perform stool samples before and after probiotic administration to determine modifications of gut microbiota.

Based on the sub-group analysis multi-strain probiotic supplements exhibited more effectiveness (*e.g.*, TC) compared with single-strain formulas. In agreement with our founding, other groups reported better beneficial health effect for supplement with different probiotic strains compared to single probiotic strain [42]. The underlying mechanism may be synergistic and cooperative interactions between different probiotic strains. Our results indicated that in addition to the number of strains, the type of strains (*e.g.*, *Saccharomyces*, *Bifidobacterium* and *Lactobacillus*) determines the effectiveness of probiotics because different types of probiotic strain have different therapeutically activities and mode of actions [43]. However, more well-conducted trails are still required to propose which type of probiotics should be used in multi-strain supplement.

The major limitation of present study is the variation in design, methodology and data reporting among included studies. Some studies did not report sufficient information about sequence generation, adherence of patients to experiment plane, and incomplete outcome data. Different probiotic interventions (*e.g.*, strains, dosages and duration), participant's characteristics and sample size were stated in included studies. These might be the potential sources of heterogeneity of findings. Almost all included studies did not perform reliable microbiological tests to confirm the viability of microorganisms. Only Firouzi et al. [20] carried out fecal analysis in order to quantify the amount of *Lactobacillus* and *Bifidobacterium* spp before and after the supplementation. They reported that *Lactobacillus* spp was significantly increased in probiotic groups. Further studies are still required to demonstrate the relationship between gut microbiota composition and change in lipid profile due to probiotic supplementation.

In summary, probiotics supplementation could be helpful in reduction of type 2 diabetic patient's risk for cardiovascular diseases. However, blood pressure and lipid lowering effects of probiotics is not enough strong to consider them as a non-pharmacologic alternative. Our study suggests that administration of probiotics could be helpful in management of dyslipidemia and hypertension in type 2 diabetic patients. More clinical trials with good design, large sample size and long follow-up time must be conducted in the future to develop clinical practice guidelines Additionally, confounders influencing microbiome composition and lipid metabolism (*e.g.*, ethnicity/race, use of medicine, dietary habits, physical activity and baseline comorbidities) must be considered in future studies.

Conflict of interest

There is no conflict of interest.

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Appendix A. Supplementary data

Supplementary data related to this article can be found at http://dx.doi.org/10.1016/j.clnu.2017.02.015.

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