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Effect of Berberine on C-reactive protein: A systematic review and meta-analysis of randomized controlled trials

Mohammad Beba[a], Kurosh Djafarian[a], Sakineh Shab-Bidar[b,*]

[a] Department of Clinical Nutrition, School of Nutritional Sciences and Dietetics, Tehran University of Medical Sciences (TUMS), Tehran, Iran
[b] Department of Community Nutrition, School of Nutritional Sciences and Dietetics, Tehran University of Medical Sciences, Tehran, Iran

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ABSTRACT

Objectives: Clinical evidence suggests the beneficial effects of Berberine (BER) on inflammatory markers. However, these results are controversial. The aim of this systematic review was to assess the effects of BER on C-reactive protein (CRP) using clinical trials in adults.
Design: Systematic review and meta-analysis.
Main outcome measures: We searched randomized controlled trials in PubMed and Scopus up to November 2018. The mean differences (MD) and confidence interval (CI) of CRP (mg/L) concentrations were pooled with a random- or a fixed-effects model depending on the results of heterogeneity tests.
Results: Of 1242 studies identified, 5 were included in the meta-analysis. Pooled analysis showed that serum levels of CRP were decreased after BER supplementation (MD:-0.64 mg/L, 95% CI(-0.67 to -0.61) $P < 0.001$)) without any significant heterogeneity ($I^2 = 0.0\%$ and $P = 0.984$).
Conclusion: This meta-analysis showed BER supplementation may ameliorate the state of chronic inflammation. Patients with cardiovascular disease and diabetes are two important groups which may benefit from BER supplementation. Further well-designed investigations with larger samples are needed to ascertain the long-term effects of BER on chronic inflammation.

1. Introduction

Low-grade systemic inflammation is a common pathogenic condition in many diseases. Increasing systemic inflammatory markers such as C reactive protein (CRP),[1] play a key role in developing chronic diseases such as obesity, type 2 diabetes (T2D), metabolic syndrome (MS), cardiovascular disease (CVD)[2,3] and nonalcoholic fatty liver disease(NAFLD).[4] Several anti-inflammatory medications have emerged during last decades to manage and control the related chronic inflammatory diseases but the observed improvements appear rather modest, and long-term safety and high-priced prescription drugs have been challenged.[5,6] High level of cholesterol is a key factor which could induce/promote inflammation in patients suffering from CVD, statins are the most frequent drugs which is used to lower cholesterol concentrations, BER might be considered as an alternative or add-on therapy to statins, although there is still insufficient evidence available with respect to long-term safety and effectiveness on cardiovascular disease prevention and treatment. These nutraceuticals could exert significant lipid-lowering activity and might present multiple non-lipid-lowering actions, including improvement of endothelial dysfunction and arterial stiffness, as well as anti-inflammatory and antioxidative properties.[7,8,9] The anti-inflammatory effects of BER have been investigated in a number of clinical trials over the past 20 years. However, the findings have been inconsistent. Most studies have reported that BER had beneficial effects on inflammation, while a few studies found that BER had no effect on inflammatory marker concentrations.[10,11] In this paper, we performed a meta-analysis of randomized controlled trials (RCTs) to systematically review the effects of BER on serum levels of CRP.

2. Methods

The meta-analysis was conducted according to the Preferred Reporting Items for Systematic
Reviews and Meta-analyses (PRISMA) Guidelines.[12]

2.1. Search strategy

Systematic search was conducted in the PubMed and Scopus (up to November 2018) for identifying eligible studies by two authors (MB

* Corresponding author.
E-mail address: s_shabbidar@tums.ac.ir (S. Shab-Bidar).

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and SS-b). The following combination of search terms was used: ("Berberine" OR "umbellatine" AND "inflammation" OR "inflammatory markers" OR "CRP" OR "C-reative protein" OR "hs-CRP" OR "High sensitive C-reactive protein). Of all articles, RCTs were checked by reviewing titles, abstracts, population and study design in order to select relevant publications for inclusion and exclusion criteria. Literature searches were downloaded into EndNote (version X7, for Windows, Thomson Reuters, Philadelphia, PA, USA) to manage and to facilitate the review process.

2.2. Study selection

All titles and abstracts were screened to evaluate eligibility for inclusion. For a study to be included in the systematic review, it had to be [1]: RCTs [2]; participants aged 18 years or older [3]; intervention with Berberine [4]; minimum intervention period of four weeks [5]; assessment of serum CRP [6]; data with normal distribution and report of mean and standard deviation (SD) or available data (standard errors, 95% confidence interval, *p*-values) to calculate these values [7]; included patients must not receive any other agents which could affect the final results.

2.3. Data extraction

Data extraction was performed to find publication year, daily dose of BER supplementation, study design, intervention duration, country, design details, health status, age and number of participants, means and SD data of each parameter before and after the intervention.

2.4. Data analysis

Pooled effect size was expressed as weighted mean differences (MD) and its corresponding 95% Confidence interval (CI) for each parameter in this meta-analysis. The heterogeneity of the included studies was examined by $\chi 2$ tests and the degree of heterogeneity was estimated using I^2 statistic. The fixed-effect model (I^2 was below 50%)[13] or the random-effects model (I^2 was above 50%) [14] was chosen for the meta-analysis. Subgroup analysis was done based on baseline values of CRP (< 2 mg/day versus > 2 mg/day) and trial duration (less than 8 weeks versus > 8 weeks). Potential publication bias was explored by using Egger line regression test (Egger's test) [15] and Begg rank correlation test (Begg's test).[16] All analyses were performed by STATA software (version 12.0). $P < 0.05$ was considered as statistically significant.

3. Results

3.1. Description of studies

The initial search identified 1242 articles, of which 572 articles were removed after duplication and 664 were excluded based on their titles and abstracts. After a full-text review of the remaining 6 potentially relevant articles, one article was further excluded because we could not find full text. Finally, 5 papers with RCT design were selected for meta-analysis [17–21] (Fig. 1).

3.2. Study characteristics

Five trials involving 317 subjects were published from 2012 to 2016, in which four were conducted in China[17,18,20,21] and one in USA,[19] which involved patients with acute coronary syndrome, hypertension, T2DM, obesity and ischemic stroke. The intervention duration lasted from 1 to 24 months. The daily dose of BER varied from 100 to 500 mg three times a day. All of our studies reported plasma CRP levels (mg/L).[17–21] The characteristics of included studies are shown in Table 1.

3.3. Risk of bias

The assessment of risk of bias was presented in Fig. 2. All studies had randomized trial design, while two of our five trials had detailed information about random sequence generation [17,18] and just two studies described the methods of allocation concealment. [17,18] Blinding of participants and personnel were considered to be unclear in all trials.[17–21]

3.4. Meta-analysis

The effect of BER supplementation on the levels of plasma CRP is shown in Fig. 3. The pooled analysis showed that the BER supplementation reduced CRP [MD= -0.64, 95% CI (-0.67 to -0.61) mg/ml, $I^2 = 0.0\%$] compared to the control group.

3.5. Subgroup analysis

We did subgroup analysis based on baseline values of CRP and trial duration. The sub-group analysis showed that there was no significant difference between categories of CRP baseline values ($p = 0.87$) (Fig. 4) and trials duration ($p = 0.48$) (Fig. 5).

3.6. Publication bias

Egger's test showed no evidence of publication bias ($p = 0.25$) for the included studies.

3.7. Side-effects

Just one of our five trials provided information on side-effects. Only three patients from the BER-treated group complained of minor side-effects, such as abdominal pain, rash and constipation, and so discontinued treatment.[17]

4. Discussion

In this systematic review and meta-analysis, we summarized published evidence from five RCTs that investigated the effects of BER supplementation on CRP levels. To the best of our knowledge, this is the first meta-analysis that examined the effect of BER supplementation on CRP level. The main finding of the review was that the serum level of CRP significantly decreased in BER supplementation group compared with placebo group. The point is that BER supplementation could significantly decrease CRP concentrations in both sexes including men and women with inflammatory status. Dose and duration are two important factors which can affect final results in clinical trials and in case of BER supplementation, duration seems to be more effective than dose. One of our studies which lasted 24 months[18] could lower CRP levels more effective than studies with shorter period of intervention even by using higher doses of BER.[19,20]

It is generally accepted that inflammation is a precursor of several chronic diseases such as hypertension, diabetes, renal and cardiovascular disease.[22] Elevated levels of certain inflammatory biomarkers, such as vascular cell adhesion molecule 1 (VCAM1) and CRP have been observed in patients with hypertension and diabetes.[23,24]. It was reported that low levels of hs-CRP (< 2 mg/L) is associated with reduced risk of incident stroke, incident CHD, and CHD death, whereas low LDL-C (< 70 mg/dL) was not protective.[25] Therefor finding a substance which could improve the inflammation status especially in those patients with chronic diseases associated with inflammatory condition like diabetes and CVD, are of great important. Investigating the effect of nutraceuticals on cardiovascular risk factors has been the focus of interest for many years.[7,26] Animal studies have shown the pharmacological activities of BER on cardiac function in which it could improve the cardiac contractility[27] pressure overload-induced cardiac hypertrophy

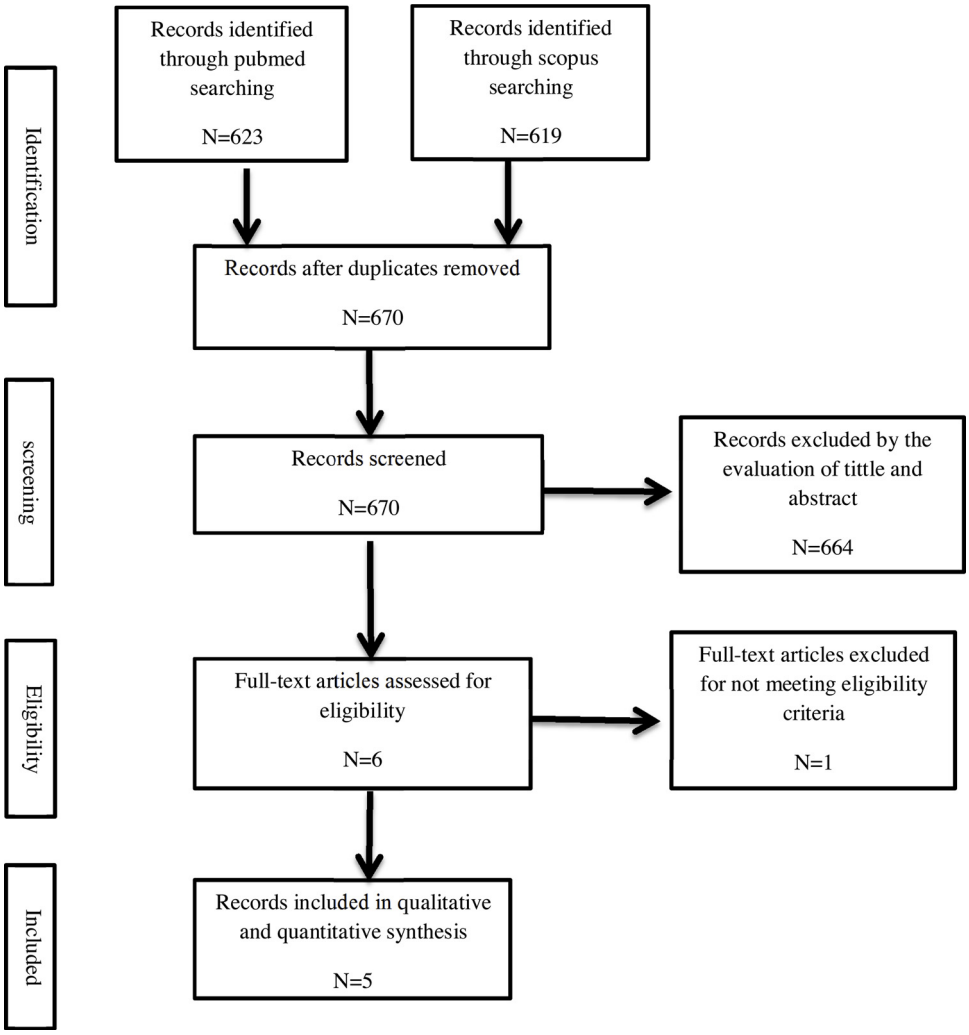

Fig. 1. Flow diagram of the study selection process.

Table 1
The characteristics of included studies in this study.

First author	Shu Meng	Peifeng dai	Yueshan Hu	ZHU Fei-qi	Lingling Chen
Country	China	China	USA	China	China
Year	2012	2015	2012	2015	2016
Problem	Acute coronary syndrome	Hypertensive patients with type 2 diabetes mellitus	Obesity	Ischemic stroke	Type 2 diabetes
Study population	130	69	7	55	30
Sex	Both	Both	Both	Both	Both
Sample size (intervention)	61	36	7	16	30
Dose(mg/day)	900	300	1500	1200	300
Duration of treatment(month)	1	24	3	3	2
Baseline CRP (intervention)	1.8	1.93	0	4.72	0
SD1	2.65	0.29	0	6.31	0
Final CRP(intervention)	1.2	1.58	3.63	2.56	2.88
SD2	2.25	0.07	1.47	2.97	1.93
N(pelacebo)	69	33	7	28	30
Base line CRP(placebo)	1.9	1.95	0	4.92	0
SD3	2.15	0.35	0	7.96	0
Final CRP (placebo)	1.5	2.22	4.14	3.23	3.45
SD4	2.65	0.05	2.02	5.67	2.32

SD: standard deviation, CRP: C-reactive protein. CRP values are mg/L.
1: standard deviation for baseline CRP in intervention group.
2: standard deviation for final CRP in intervention group.
3: standard deviation for baseline CRP in placebo group.
4: standard deviation for final CRP in placebo group.

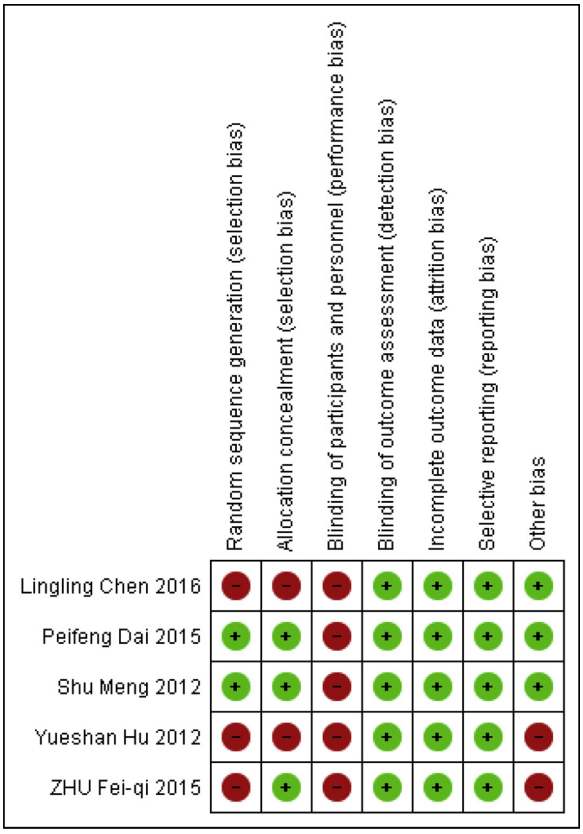

Fig. 2. Risk of bias assessment of included studies.

and dysfunction [28] and prevent the development of left ventricular hypertrophy.[28] Human studies have also demonstrated that BER may improve endothelial function by reducing oxidative stress.[29] Patients with diabetes also may benefit from BER consumption due to its anti-oxidant and anti-inflammatory activity.[30] The potential benefits of BER are attributed to its interaction with multiple proteins in the body,[31] changing metabolic pathways of lipid[7,32,33] and glucose.[7] The anti-inflammatory effect of BER using acute phase proteins and proinflammatory cytokines as markers has been studied in multiple in *vitro* and in *vivo* studies.[34–37] In different cell lines such as immune cells, hepatocytes, adipose tissues, islets β-cells, BER could suppress the production of MCP1 (monocyte chemoattractant protein 1), interleukin (IL)-6, IL-1β, CRP and HP (hepatoglobin). Despite multiple in *vitro* studies, the numbers of human studies evaluating the effect of BER on inflammatory biomarkers are rare. Of human studies, most of them have been conducted on patients with diabetes. It seems BER suppresses inflammation through various mechanisms, such as mitogen-activated protein kinase- (MAPK-) mediated pathways, the nuclear factor-kappa B (NF-κB), and the Rho GTPase signaling pathway.[38–40] Based on

In terms of toxic and side effect of BER, our data shows that the incidence of toxic side effects may be related to the doses of BER. When the dose of BER increases, the risk of toxic side effect also increases. Currently, the dispute about drug toxicity of BER has not been resolved. Studies on the toxicity of BER also lacks systematic and complete research, so a conclusion as to whether BER is toxic or non-toxic cannot be made. Based on the included 5 trials, BER can produce a few side effects. The incidence of adverse reactions is low, without occurrences or serious adverse reactions that affect important organs occurred in the course of treatment.

Our meta-analysis included only 5 randomized controlled trial studies with a small population, thus performing further studies with a bigger population are needed to determine whether BER is effective on controlling/lowering CRP levels. Moreover, we had high level of heterogeneity with fixed effect model which was removed after applying random effect models.

The present systematic review provides evidence that BER supplementation may significantly improve the process of inflammatory status. Patients with cardiovascular disease and diabetes are two important groups which could benefit the most from BER supplementation. Further studies, especially with larger sample size and well-designed RCTs, are needed to confirm the effectiveness of BER supplementation on inflammation.

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Declaration of Competing Interest

We wish to confirm that there are no known conflicts of interest associated with this publication and there has been no significant financial support for this work that could have influenced its outcome.

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MB and SSb contributed to conception/design of the research; MB

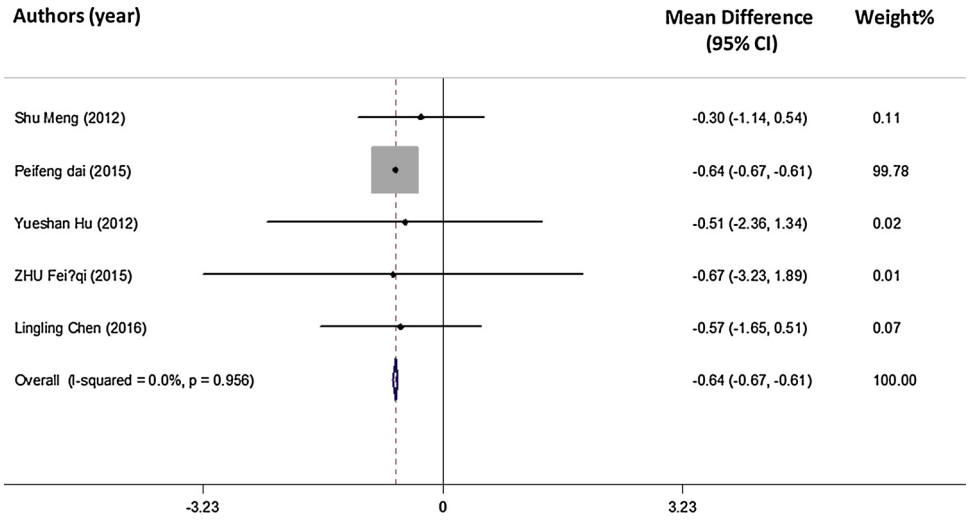

Fig. 3. Forest plot of comparisons of BER supplementation versus placebo.

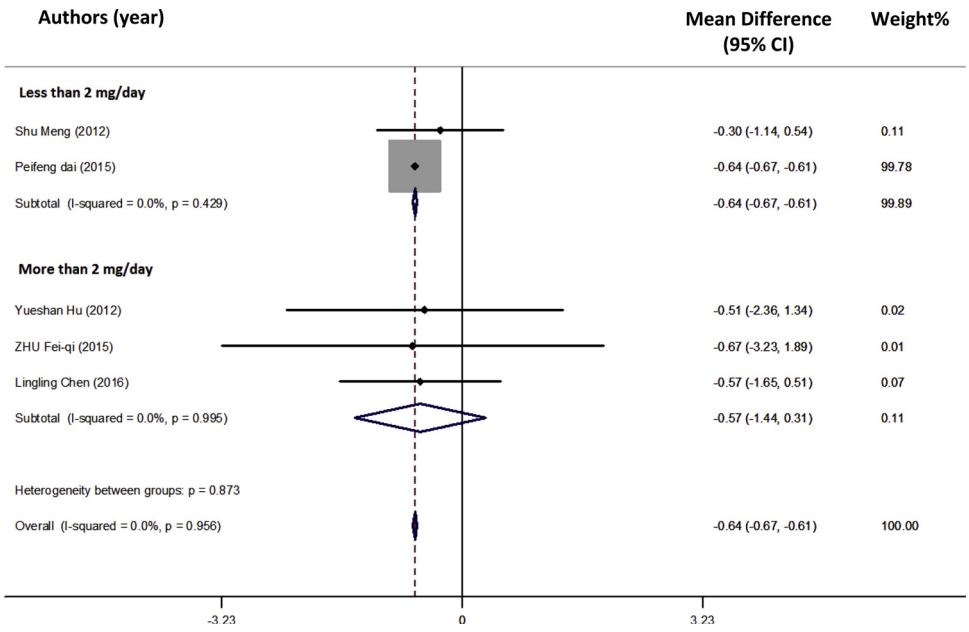

Fig. 4. Effect of BER supplementation on CRP based on baseline values.

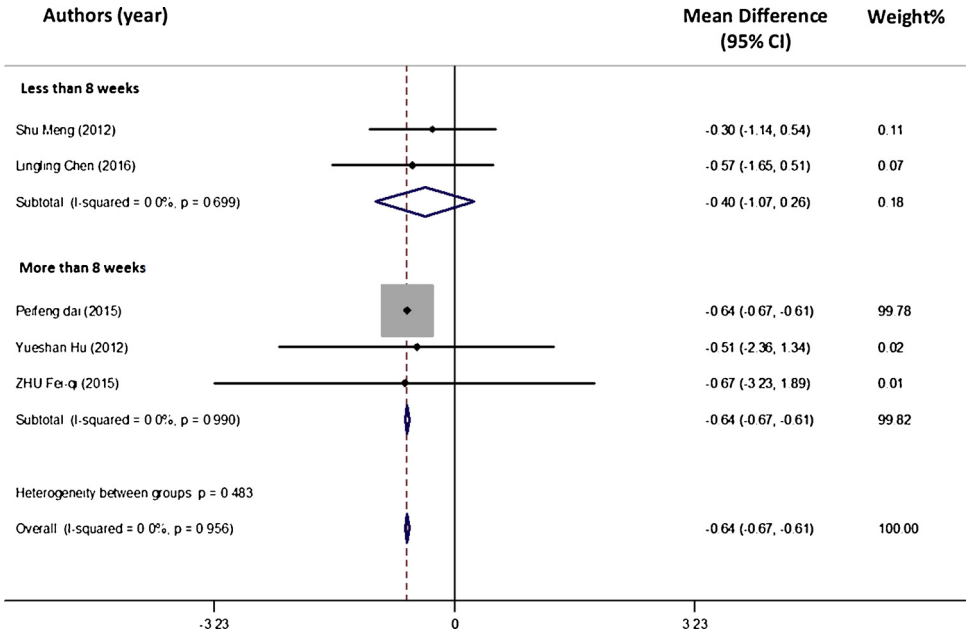

Fig. 5. Effect of BER supplementation on CRP based on duration.

contributed to acquisition, analysis, or interpretation of the data; MB drafted the manuscript; KD and SSb critically revised the manuscript; and SS-b agrees to be fully accountable for ensuring the integrity and accuracy of the work. All authors read and approved the final manuscript.

References

1. Kawano J, Arora R. The role of adiponectin in obesity, diabetes, and cardiovascular disease. *J Cardio Metab Syndr.* 2009;4(1):44–49.
2. Pradhan A. Obesity, metabolic syndrome, and type 2 diabetes: inflammatory basis of glucose metabolic disorders. *Nutr Rev.* 2007;65(suppl_3) S152-S6.
3. Guarner V, Rubio-Ruiz ME. *Low-grade systemic inflammation connects aging, metabolic syndrome and cardiovascular disease.* 2015; 2015.
4. Basaranoglu M, Ormeci N. Nonalcoholic fatty liver disease: diagnosis, pathogenesis, and management. *Turk J Gastroenterol.* 2014;25(2):127–132.
5. Esser N, Paquot N, Scheen AJ. Anti-inflammatory agents to treat or prevent type 2 diabetes, metabolic syndrome and cardiovascular disease. *Expert Opin Investig Drugs.* 2014;24(3):283–307.
6. Barrios V, Escobar C, Cicero AFG, et al. A nutraceutical approach (Armolipid Plus) to reduce total and LDL cholesterol in individuals with mild to moderate dyslipidemia: review of the clinical evidence. *Atheroscler Suppl.* 2017;24:1–15.
7. Banach M, Patti AM, Giglio RV, et al. The role of nutraceuticals in statin intolerant patients. *J Am Coll Cardiol.* 2018;72(1):96–118.
8. Cicero AF, Colletti A, Bajraktari G, et al. Lipid-lowering nutraceuticals in clinical practice: position paper from an International Lipid Expert Panel. *Nutr Rev.* 2017;75(9):731–767.
9. Vuddanda PR, Chakraborty S, Singh S. Berberine: a potential phytochemical with multispectrum therapeutic activities. *Expert Opin Investig Drugs.* 2010;19(10):1297–1307.
10. Chen F, Yang Z, Liu Y, et al. Berberine inhibits the expression of TNFα, MCP-1, and IL-6 in AcLDL-stimulated macrophages through PPARγ pathway. *Endocrine.* 2008;33(3):331–337.
11. Jeong HW, Hsu KC, Lee J-W, et al. Berberine suppresses pro-inflammatory responses through AMPK activation in macrophages. *Am J Physiol – Endocrinol Metab.* 2009.
12. Adamska-Patruno E, Ostrowska L, Golonko A, et al. Evaluation of energy expenditure and oxidation of energy substrates in adult males after intake of meals with varying fat and carbohydrate content. *Nutrients.* 2018;10(5):627.

13. Mantel N, Haenszel W. Statistical aspects of the analysis of data from retrospective studies of disease. *J Natl Cancer Inst.* 1959;22(4):719–748.
14. DerSimonian R, Kacker R. Random-effects model for meta-analysis of clinical trials: an update. *Contemp Clin Trials.* 2007;28(2):105–114.
15. Egger M, Smith GD, Schneider M, Minder C. Bias in meta-analysis detected by a simple, graphical test. *BMJ.* 1997;315(7109):629–634.
16. Begg CB, Mazumdar M. Operating characteristics of a rank correlation test for publication bias. *Biometrics.* 1994:1088–1101.
17. Meng S, Wang LS, Huang ZQ, et al. Berberine ameliorates inflammation in patients with acute coronary syndrome following percutaneous coronary intervention. *Clin Exp Pharmacol Physiol.* 2012;39(5):406–411.
18. Dai P, Wang J, Lin L, Zhang Y, Wang Z. Renoprotective effects of berberine as adjuvant therapy for hypertensive patients with type 2 diabetes mellitus: evaluation via biochemical markers and color Doppler ultrasonography. *Exp Ther Med.* 2015;10(3):869–876.
19. Hu Y, Ehli EA, Kittelsrud J, Ronan PJ, Munger K, Downey T, et al. Lipid-lowering effect of berberine in human subjects and rats. *Phytomedicine.* 2012;19(10):861–867.
20. Zhu FQ, Chen L, Zhu JH. Influence of berberine combining with atorvastatin on serum high-sensitivity C-reactive protein and adipocyte fatty acid-binding protein in patients with acute ischemic stroke. *Chin J Contemp Neurol Neurosurg.* 2019;15(1):43–47.
21. Chen L, Lu W, Li Y. Berberine ameliorates type 2 diabetes via modulation of Bifidobacterium species, Tumor necrosis factor-α, and lipopolysaccharide. *Int J Clin Exp Med.* 2016;9(6).
22. Dai P, Wang J, Lin L, Zhang Y, Wang Z. Renoprotective effects of berberine as adjuvant therapy for hypertensive patients with type 2 diabetes mellitus: evaluation via biochemical markers and color Doppler ultrasonography. *Exp Ther Med.* 2015;10(3):869–876.
23. Liu L, Liu Y, Qi B, Wu Q, Li Y, Wang Z. Nicorandil attenuates endothelial VCAM-1 expression via thioredoxin production in diabetic rats induced by streptozotocin. *Mol Med Rep.* 2014;9(6):2227–2232.
24. Suzuki H, Sakamoto M, Hayashi T, et al. Effects of co-administration of candesartan with pioglitazone on inflammatory parameters in hypertensive patients with type 2 diabetes mellitus: a preliminary report. *Cardiovasc Diabetol.* 2013;12(1):71.
25. Penson PE, Long DL, Howard G, et al. Associations between very low concentrations of low density lipoprotein cholesterol, high sensitivity C-reactive protein, and health outcomes in the Reasons for Geographical and Racial Differences in Stroke (REGARDS) study. *Eur Heart J.* 2018;39(40):3641–3653.
26. Ward N, Sahebkar A, Banach M, Watts G. Recent perspectives on the role of nutraceuticals as cholesterol-lowering agents. *Curr Opin Lipidol.* 2017;28(6):495–501.
27. Zhao H, Hong Y, Xie J, Xie X, Wang J, Fan J. Effect of berberine on left ventricular remodeling in renovascular hypertensive rats. *Yao xue xue bao= Acta Pharm Sin.* 2007;42(3):336–341.
28. Li M-H, Zhang Y-J, Yu Y-H, Yang S-H, Iqbal J, Mi Q-Y, et al. Berberine improves pressure overload-induced cardiac hypertrophy and dysfunction through enhanced autophagy. *Eur J Pharmacol.* 2014;728:67–76.
29. Cheng F, Wang Y, Li J, et al. Berberine improves endothelial function by reducing endothelial microparticles-mediated oxidative stress in humans. *Int J Cardiol.* 2013;167(3):936–942.
30. Ni W-J, Ding H-H, Tang L-Q. Berberine as a promising anti-diabetic nephropathy drug: an analysis of its effects and mechanisms. *Eur J Pharmacol.* 2015;760:103–112.
31. Ma X, Chen Z, Wang L, et al. The pathogenesis of diabetes mellitus by oxidative stress and inflammation: its inhibition by berberine. *Front Pharmacol.* 2018;9(782).
32. Dong Y, Chen YT, Yang YX, et al. Metabolomics study of type 2 diabetes mellitus and the antidiabetic effect of berberine in zucker diabetic fatty rats using Uplc-ESI-Hdms. *Phytother Res.* 2016;30(5):823–828.
33. Liu Y, Zhang L, Song H, Ji G. Update on berberine in nonalcoholic fatty liver disease. *Evid Based Complement Altern Med.* 2013;2013.
34. Choi B-H, Ahn I-S, Kim Y-H, et al. Berberine reduces the expression of adipogenic enzymes and inflammatory molecules of 3T3-L1 adipocyte. *Exp Mol Med.* 2006;38(6):599.
35. Lou T, Zhang Z, Xi Z, et al. Berberine inhibits inflammatory response and ameliorates insulin resistance in hepatocytes. *Inflammation.* 2011;34(6):659–667.
36. Hsu YY, Chen CS, Wu SN, Jong YJ, Lo YC. Berberine activates Nrf2 nuclear translocation and protects against oxidative damage via a phosphatidylinositol 3-kinase/Akt-dependent mechanism in NSC34 motor neuron-like cells. *Eur J Pharm Sci.* 2012;46(5):415–425.
37. Pang B, Zhao LH, Zhou Q, et al. Application of berberine on treating type 2 diabetes mellitus. *Int J Endocrinol.* 2015;2015:905749.
38. Li Z, Geng YN, Jiang JD, Kong WJ. Antioxidant and anti-inflammatory activities of berberine in the treatment of diabetes mellitus. *Evid Based Complement Alternat Med.* 2014;2014:289264.
39. Wang Y. Attenuation of berberine on lipopolysaccharide-induced inflammatory and apoptosis responses in beta-cells via TLR4-independent JNK/NF-kappaB pathway. *Pharm Biol.* 2013.
40. Xie W, Du L. Diabetes is an inflammatory disease: evidence from traditional Chinese medicines. *Diabetes Obes Metab.* 2011;13(4):289–301.