Title: Association of plasma osteoprotegerin levels with the severity of lower extremity arterial disease in patients with type 2 diabetes

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Author's response to reviews: see over
Dear Editor:
Our manuscript has been extensively revised in accordance with the enclosed reviewer’s comments. All changes in manuscripts emphasized by red characters.

Reviewer's report:
This is a study on a potential association between plasma OPG and lower extremity arterial disease in T2DM patients. As described by the authors there are not many studies on peripheral artery disease, and this study, which do find a significant association between plasma OPG and a Doppler investigation, therefore seems relevant. Overall, I therefore find the study interesting and well-described.

Major compulsory revisions:
In the Statistical section it is stated how data is expressed depending on their distribution: How was the distribution tested? This should be stated in the manuscript. Also, OPG concentrations are throughout the manuscript reported as mean +/- SD. But OPG concentrations almost always have a skewed distribution – which also looks to be the case for these data when looking at Figure 1: Please explain this discrepancy.

Answers:
We sincerely appreciate reviewer’s constructive comments. In our study, normal distribution of data was tested by the Kolmogorov-Smirnov test. According to reviewer’s advice, we have added the description in the manuscript. Also, as reviewer’s comments, the OPG concentrations have a skewed distribution in this study. We are very grateful for reviewer’s comment, at the same time we sincerely apologize for any confusion may have been caused by our negligence. According to reviewer’s suggestion, we have changed the expressed the OPG concentrations as median (inter-quartile range) in the manuscript.

Minor essential revisions:
In general, the manuscript is well-written, but there are some language flaws, especially in the Discussion section, which must be re-read.

Answers:
We thanks for reviewer’s reminder. According to reviewer’s instruction, we have re-read the article and revised the language in the manuscript.

Background
At the end of the Background section, the authors state that little is known about the association between plasma OPG levels and lower extremity arterial disease. The information may be scarce, but there are a number of studies looking at the association between OPG and peripheral artery disease (PAD) that must be mentioned, namely a review by Hosbond et al (2012), a study by Poulsen et al on T2DM patients (2011) and finally a study by O'Sullivan et al (2010). Please
incorporate these in the Background and later in the Discussion when comparing with earlier findings.

**Answers:**
We thanks for reviewer’s reminder. According to reviewer’s suggestion, we have incorporated previous studies in the Background and the Discussion.

**Methods**
It is stated that the T2DM patients was selected. How was this done?

**Answers:**
Thank you for your advice. According to the reviewer’s suggestion, we have revised our manuscript and described how the T2DM patients were selected in the manuscript.

Subjects were recruited from the Department of Endocrinology at Xinhua Hospital Affiliated to Shanghai Jiaotong University between 2013 and 2014. All unrelated subjects with T2DM who attended the Diabetes Clinic at the Xinhua Hospital were recruited consecutively to participate in a prospective study to identify the risk factors predisposing to the development of diabetic complications. Each visit comprised clinical assessments and laboratory investigations to determine the control of diabetes and related cardiovascular risk factors, and the presence of diabetic complications. Diabetes was defined according to the 2008 American Diabetes Association diagnostic criteria (MM). Subjects with arteriovenous grafts/shunts, vasculitis, chronic kidney disease, cerebral infarction, coronary artery disease, malignancies, or with immunological diseases, osteoporosis and subjects receiving systemic glucocorticoids or immunosuppressants were excluded from the study. A total of 712 T2DM subjects, who attended regular visits at least twice a year, with the latest follow-up in or before October 2014, were enrolled in the study. Of these, 505 patients with lower extremity arterial stenosis and 207 patients without lower extremity arterial stenosis. Written informed consent was obtained from all the participants. The study was approved by the Institutional Review Board of Xinhua Hospital Affiliated to Shanghai Jiaotong University School of Medicine.

The authors erroneously use the term Triacylglycerol, where I suppose they should have used Triglycerides (page 6 and Table 3). Please correct this.

**Answers:**
We thanks for reviewer’s reminder. It’s just a description error, the right expression should as “Triglycerides”, according to reviewer’s instruction, we have changed it in manuscript.

**Discussion**
Line 171: I do not see how the demonstrated association between OPG and the severity of lower arterial extremity stenosis provides an explanation for the increased vascular events in patients with high plasma OPG. As we know nothing certain about the mechanism through which OPG increases the risk (is it just an epiphenomenon?), I find the authors must settle with the potential use of plasma OPG as a biomarker for this condition, not an explanation for any forthcoming
events. Please adjust this.

If the Tables are not a part of the manuscript, main numbers must be presented in the text in order to facilitate the reading of the paper.

**Answers:**
We are sincerely appreciative of reviewer’s profound and thought-provoking comments. As reviewer’s opinion, the association of OPG and severity of lower arterial extremity stenosis may be just an epiphenomenon. Our findings only suggest that OPG is a potential biomarker of lower extremity arterial stenosis in type 2 diabetes. Measurement of plasma OPG could be useful for lower extremity arterial stenosis stratification of type 2 diabetes. According to reviewer’s advice, we have revised its in the manuscript. Also, as reviewer’s instruction, we have added the main numbers in the text.

Discretionary revisions:
In the Methods chapter, section “Determination of lower …” (p. 6), the phrase “Doppler examination can accurately …” should be erased. This is a statement, which does not belong in the method section.

**Answers:**
We thanks for reviewer’s reminder. According to reviewer’s instruction, we have erased it in the manuscript.

Also, please omit the phrase “We compared the degree of …”, which if necessary can be incorporated in the statistical section. The final sentence belongs to the section regarding the Patients (p. 4).

**Answers:**
We thanks for reviewer’s reminder. According to reviewer’s instruction, we have erased it in the manuscript.

Also, please correct the pluralis form used here (histories (history), backgrounds (background), medications (…)).

**Answers:**
We thanks for reviewer’s reminder. According to reviewer’s instruction, we have changed it in the manuscript.

Results, Baseline characteristics, line 1: … patients with … (not had).

**Answers:**
We thanks for reviewer’s reminder. According to reviewer’s instruction, we have changed it in the manuscript.