PEER REVIEW HISTORY

BMJ Open publishes all reviews undertaken for accepted manuscripts. Reviewers are asked to complete a checklist review form and are provided with free text boxes to elaborate on their assessment. These free text comments are reproduced below.

ARTICLE DETAILS

TITLE (PROVISIONAL)	Do bisphosphonates and RANKL inhibitors alter the progression of coronary artery calcification? A systematic review.
AUTHORS	Saunders, Samantha; Chaudhri, Kanika; McOrist, Nathan; Gladysz, Karen; Gnanenthiran, Sonali; Shalaby, Grant

VERSION 1 - REVIEW

REVIEWER NAME	***Cai, Xiaoling***
REVIEWER AFFILIATION	Peking University People's Hospital
REVIEWER CONFLICT OF INTEREST	Na
DATE REVIEW RETURNED	12-Feb-2024

GENERAL COMMENTS	Samantha L Saunders BMedSci et al. conducted a systematic review of the progression of coronary artery calcification in patients receiving bisphosphonates and RANKL inhibitors. The topic is of interest since there is a physiological link between lipids and bone. However, only five studies (n=377) of heterogeneous papers eligible for inclusion in the review were included. I have several comments regarding the manuscript: Please provide the research question using PICO formulation. I encourage authors to be more explicit about the population included. Please add follow up time in Table 1. In this paper, the strong heterogeneity, few included studies, and short follow-up time, which are not enough to verify the hypothesis.

REVIEWER NAME	***Minisola, S.***
REVIEWER AFFILIATION	Biomarker Design Forsch GmbH
REVIEWER CONFLICT OF INTEREST	Na
DATE REVIEW RETURNED	12-Mar-2024

GENERAL COMMENTS	This is an interesting metanalysis, trying to address an important question in clinical practice. However, the number of studies reported is so small that no conclusion can be made with certainty. To increase the value of this paper, please address the following points. The issue of etidronate is an interesting one. Perhaps you should expand this section, in terms of dose used, what could be considered an optimal dose balancing risk and benefits and so on. Regarding denosumab, there are some “in vivo” studies addressing the effect on molecules of cardiovascular risk that could be important to report to give a wider view of the problem.

REVIEWER NAME	***Zeng, Irene***
REVIEWER AFFILIATION	Auckland University of Technology Faculty of Health and Environmental Sciences, Health Falculty research office
REVIEWER CONFLICT OF INTEREST	Na
DATE REVIEW RETURNED	03-May-2024

GENERAL COMMENTS	It is an important topic reviewing alternative therapies for CVD targeting a reduction in Coronary artery calcium (CVC). Please find my review summary: 1. Abstract While the result summary is comprehensive, there is room for improvement. I suggest a more structured approach, starting with a description of the types of studies (i.e. the number of RCTs, observational studies, and NonRCTs) , followed by the range of effect sizes from the studies measured by delta CAC. Separating the positive and negative studies could also enhance the clarity of the summary. 2. Search Strategy How were these keywords included in the search? Has the search been conducted by a librarian? 3. Studies selection What are the inclusion and exclusion criteria? 4. Outcomes The study population needs to be defined separately from the outcomes. 5. Data synthesis and analysis Has the effect size from each study been collected apart from statistical significance measured using the P value? The writing of this paragraph could be improved. For example, the outcomes of each study were extracted with their effect size measured by OR and RR with their confidence interval. The statistical significance reported by their P values is also collected. The sentence “Raw data collected” is confusing- raw data normally is referred to unit record observation. 6. Primary outcomes All the mentioned studies need to include the reference in the text. Reference papers must be included so the reader can identify the described studies in the Bibliography. Minor: AU need the full description at their first-time quotation. 7. Secondary outcomes A 95% confidence interval needs to be included in the referred effect size, for example, on page 11. Line 297. 8. Discussion It is worthwhile to mention that there is only one RCT conducted and all the other evidence are from observational studies. 9. Places of referring P value can be replaced by “any statistical hypothesis result (i.e. p value) “. For example page 12 , line 323.

VERSION 1 – AUTHOR RESPONSE

Reviewer 1

1. **Please provide the research question using PICO formulation.**

Response: The authors would like to thank the reviewer for this suggestion – initially, we had cited the review protocol which contains the PICO formulation. However, per the reviewer's suggestion, we have now included it explicitly in the methods section of the paper. Please see lines 125 - 127 and Table 1 on page 20.

2. **I encourage authors to be more explicit about the population included.**

Response: Thank you for this suggestion. The inclusion criteria now inform of the participants who were eligible for inclusion (as discussed above). Please see lines 125-127 and Table 1 on page 20. Furthermore, the "Demographical Data" section of the Results has been adjusted to "Demographical Data and Population" and highlights characteristics of the included population, such as prevalence of CKD and osteoporosis. Please see lines 184 – 185.

3. **Please add follow up time in Table 1.**

Response: Thank you for this suggestion. Data on follow-up time can now be found in column 9 of Table 2 on pages 21 – 22.

4. **In this paper, the strong heterogeneity, few included studies, and short follow-up time are not enough to verify the hypothesis.**

Response: The authors agree with this statement. While a conclusion was not able to be drawn on this occasion, we strongly hope that our systematic review will bring attention to the lack of studies available in the literature on this topic, and therefore will highlight the important need for further research in this area.

Reviewer 2

1. **The issue of etidronate is an interesting one. Perhaps you should expand this section, in terms of dose used, what could be considered an optimal dose balancing risk and benefits and so on.**

Response: Thank you for this insight. The authors agree that this is a fascinating area which could potentially lead to future research in the field. We have since expanded this section in the "Bisphosphonates and CAC" section of the Discussion, citing additional relevant reviews which delve into the dosing and side effect profile of etidronate. Please see lines 240 – 246.

2. **Regarding denosumab, there are some "in vivo" studies addressing the effect on molecules of cardiovascular risk that could be important to report to give a wider view of the problem.**

Response: We would like to thank the reviewer for this suggestion. As per our inclusion criteria, *in vitro* and *in vivo* animal studies were not included in the review. However, we have looked further into this topic and have cited three additional papers which were relevant to the discussion. This has been updated in lines 252 – 260 of our Discussion.

Reviewer 3

1. **Abstract – I suggest a more structured approach, starting with a description of the types of studies (i.e. the number of RCTs, observational studies, and NonRCTs),**

followed by the range of effect sizes from the studies measured by delta CAC. Separating the positive and negative studies could also enhance the clarity of the summary.

Response: We would like to thank the reviewer for this insight. We have since reformatted the abstract in line with the Journal's recommendations. We have since added in the inclusion criteria under "Eligibility Criteria". We have now outlined the number of observational studies and RCTs and have included the n values of each individual study so that the effect sizes are clear to the reader. We have separated the results section based on intervention used; three studies investigating bisphosphonate use, one study investigating RANKL inhibitor use, and one study investigating both. We believe this separation of intervention will help readers understand the pertinent findings more clearly. Please see page 2.

2. **Search Strategy – how were these keywords included in the search? Has the search been conducted by a librarian?**

Response: The search strategy was originally reported in the Review Protocol (Supplemental File 1). However, for clarity, we have now added the search strategy to the manuscript as its own supplementary file (Supplementary File 2). The search strategy was developed by a medical librarian with search syntax altered according to each database's subject headings and thesaurus. This has been updated in the "Search Strategy" section of the Methods. Please see lines 131 – 133.

3. **Studies selection – what are the inclusion and exclusion criteria?**

Response: Thank you for suggesting that this is included. We have now included the inclusion criteria using the PICO formulation (see Methods section, "Eligibility criteria", also previously discussed in response to Reviewer 1, question 1). Please see lines 125 – 127 and Table 1 on page 20.

4. **Outcomes – the study population needs to be defined separately from the outcomes.**

Response: Thank you for this suggestion. This has been discussed previously in response to Reviewer 1, question 2. The inclusion criteria now inform of the participants who were eligible for inclusion (as discussed above). Please see lines 125 – 127 and Table 1 on page 20. Furthermore, the "Demographical Data" section of the Results has been adjusted to "Demographical Data and

Population" and highlights characteristics of the included population, such as prevalence of CKD and osteoporosis. Please see lines 184 – 185.

5. **Data synthesis and analysis – has the effect size from each study been collected apart from statistical significance measured using the P value? The writing of this paragraph could be improved. For example, the outcomes of each study were extracted with their effect size measured by OR and RR with their confidence interval. The statistical significance reported by their P values is also collected. The sentence "Raw data collected" is confusing- raw data normally is referred to unit record observation.**

Response: The authors would like to thank the reviewer for this insight. Per this recommendation we have since re-reviewed the papers included in the study and extracted the effect sizes, where available. Only one paper reported effect size. Additionally, we have rewritten the "Data Synthesis and Analysis" section of the Methods to reflect this, and to increase clarity on outcomes extracted. Furthermore, we have removed the line "raw data was collected" per the reviewer's recommendation. Please see lines 155-167.

6. **Primary outcomes – all the mentioned studies need to include the reference in the text. Reference papers must be included so the reader can identify the described studies in the Bibliography. Minor: AU need the full description at their first-time quotation.**

Response: Thank you for identifying this oversight. The included papers are now cited in the "Study Inclusion" section of the Results as well as in Table 2. Furthermore, the "Primary Outcomes" section of the Results section has been re-written to separate the bisphosphonate results from the denosumab results for enhanced clarity and easier interpretation of the results. Likewise, the paragraphs in each of these new sections have also been separated to discuss positive results first, followed by negative results, again for improved clarity. The *n* values of each paper to demonstrate sample size have also been included here. Agatston Units have been written in full at first time of quotation in the Introduction. Please see lines 189 – 208.

7. **Secondary outcomes – a 95% confidence interval needs to be included in the referred effect size, for example, on page 11. Line 297.**

Response: An effect size was not reported in this paper so unfortunately, could not be included.

8. **Discussion – It is worthwhile to mention that there is only one RCT conducted, and all the other evidence are from observational studies.**

Response: Thank you for this recommendation. The first paragraph of the Discussion has been updated to reflect this information. Please see lines 220 – 221.

9. **Places of referring P value can be replaced by "any statistical hypothesis result (i.e. p value) ". For example, page 12, line 323.**

Response: Thank you for this suggestion. This line has since been corrected to align with the reviewer's suggestion. Please see line 231.

The authors would like to express their sincere gratitude to the Editorial team and those who reviewed the paper for their comprehensive comments and feedback. We hope that you will consider our review for publication in your journal following the amendments made.

VERSION 2 – REVIEW

REVIEWER NAME	***Zeng, Irene***
REVIEWER AFFILIATION	Auckland University of Technology Faculty of Health and Environmental Sciences, Health Falculty research office
REVIEWER CONFLICT OF INTEREST	Na
DATE REVIEW RETURNED	25-Jul-2024

GENERAL COMMENTS	Authors have addressed all my comments.