PEER REVIEW HISTORY

BMJ Open publishes all reviews undertaken for accepted manuscripts. Reviewers are asked to complete a checklist review form (**http://bmjopen.bmj.com/site/about/resources/checklist.pdf)** and are provided with free text boxes to elaborate on their assessment. These free text comments are reproduced below.

ARTICLE DETAILS

TITLE (PROVISIONAL)	Association between serum magnesium concentration and metabolic syndrome, diabetes, hypertension and hyperuricemia in knee osteoarthritis: a cross-sectional study in Hunan Province, Chin
AUTHORS	Wang, Yi-lun; Wei, Jie; Zeng, Chao; Yang, Tuo; Li, Hui; Cui, Yang; Xie, Dong-xing; Xu, Bei; Liu, Zhi-chen; Li, Jia-tian; Jiang, Shi-de; Lei, Guanghua

VERSION 1 – REVIEW

REVIEWER	PROF O B FAMILONI OLABISI ONABANJO UNIVERSITY TEACHING HOSPITAL, NIGERIA
REVIEW RETURNED	01-Sep-2017

GENERAL COMMENTS	The statement in the Result section and the Discussion about the relationship between serum Mg and metabolic syndrome needs qualification. Whereas there might be association with 2nd and 5th quartile, the overall tendency did not show significant association. In line 231, a P trend of 0.120 is not significant and even Table 1 line 26, a P of 0.059 is at best a weak association

REVIEWER	Reda Morsy Tanta University, Egypt
REVIEW RETURNED	23-Sep-2017

GENERAL COMMENTS	1. There are papers for researchers and also for others have been published concerning the association between Mg and Mg intake on osteoarthritis and they found the association is inverse. 2. There are previous papers for researchers, having the same style, about association between Mg, osteoarthritis, and hyperuricemia. 3. What is the file number of ethics approval 4. What is about the reference sample of study? Is the reference normal people or patients having only osteoarthritis?. If they were patients only having osteoarthritis, what was the association with Mg?

REVIEWER	John Sayer Newcastle University, United Kingdom
REVIEW RETURNED	24-Nov-2017

GENERAL COMMENTS	This is a cross-sectional study of a population with osteoarthritis of the knee and an examination of the prevalence of metabolic syndrome, hypertension, hyperuricaemia and diabetes mellitus and the serum magnesium level. It included 962 patients.

	Major comments The abstract is difficult to read, with the comparison of different quintiles for each association. I wondered if the second quintile was the second highest or second lowest etc. Please clarify. Please comment on the GFR in the first part of the results, page 9 as I believe this produced signicant differences in Mg levels, and hence vital to correct for this is any further comparisons (table 1 p<0.001). In fact, given this factor, I do not think any of the models that do not include GFR are valid. I do not think it is a fair conclusion that MetS is associated in a linear way with serum magnesium levels. The quintiles with a lower prevalence were 2 and 5, with trends not showing much significance. Based on this I don't support the conclusion (page 11 line 272) that Mets is negatively associated with serum Mg. The data does support DM and HU in association with Mg levels in a linear fashion. The conclusions that modifying Mg levels for therapeutic benefit are over interpreting the data. This is too speculative as it is only an association that has been shown, rather than causality. The authors need to be more clear on how these results can be interpreted. Mechanisms are discussed but are not explored in any detail. Figure is too basic and adds nothing to the understanding of the data. Minor There are numerous English language problems Page 4 line 73 – “adusted by a” change to “adjusted for a” Page 4 line 76 “Kidney is” change to “The kidney is” Page 4 line 82 “which was not the best” change to “which may not be” Page 5 line 96 should read “injury” Page 5 line 101 change “take measures” to “adopt measures” Page 5 line 107 change “examine” to “examining” Page5 line 109 change “another study of ours” to “we have previously shown” Page 5 line 110 change “it is reasonably speculated” to “we speculate” Page 6 line 123 – please give name of ethics committee Page 9 line 209 – please state whether this is MDRD or another variation of eGFR formulae Page 9 line 214 – test – should be tests, which tests specifically Page 12 line 289-90 – doesn't make sense Page 12 line 298 – this is too speculative as it is only an association that has been shown, rather than causality.

REVIEWER	Neslihan GOKCEN Cukurova University Faculty of Medicine, Adana/TURKEY
REVIEW RETURNED	26-Nov-2017

GENERAL COMMENTS	To Authors, I do thank you for your valuable well-design study. Some diseases such as diabetes mellitus and some drugs such as loop diuretics, thiazide diuretics can cause hypomagnesemia. The medical history of patients may be suggested to evaluate whether the drugs are a confounder. Despite all these confounding factors, authors analyzed the outcomes by multivariable methods and this method strengthened the study. The article is well written and well

	presented. I have some minor comments. Minor Comments: 1. The medical history of patients may be suggested to evaluate whether the drugs are a confounder. 2. You may extend the limitations. 3. Please check the table. You used the abbreviation for metabolic syndrome as MS. 4. Please arrange the figure more intelligible.

VERSION 1 – AUTHOR RESPONSE

Responses of every comment raised by four reviewers are listed as follows:

Reviewer: 1
Reviewer Name: PROF O B FAMILONI
Institution and Country: OLABISI ONABANJO UNIVERSITY TEACHING HOSPITAL, NIGERIA

The statement in the Result section and the Discussion about the relationship between serum Mg and metabolic syndrome needs qualification. Whereas there might be association with 2nd and 5th quartile, the overall tendency did not show significant association. In line 231, a P trend of 0.120 is not significant and even Table 1 line 26, a P of 0.059 is at best a weak association.
Response: Thank you very much for your kind comments. We agree with the reviewer's comment that the overall tendency about the relationship between Mg and MetS was not significant. It suggests that the dose-response-relationship between Mg and MetS was failed to be observed in this study. However, it is true that there were negative findings in the relationship between the 2nd and 5th quintiles of serum Mg and MetS, even after adjustments of confounders. To some extent, we believe that Mg is inversely associated with MetS in knee OA patients, but this association was not in a dose-response-relationship manner. We appreciate your kind comments, and thus we modified relevant statements about the association between Mg and MetS in the Result, Discussion and Abstract sections in our revised manuscript. We hope that it could make our manuscript more qualified. Thanks again! (Clean version: Page 2 line 32-45; Page 8 line 197-208; Page 10 line 252-253; Page 12 line 314-315)

Reviewer: 2
Reviewer Name: Reda Morsy
Institution and Country: Tanta University, Egypt

1. There are papers for researchers and also for others have been published concerning the association between Mg and Mg intake on osteoarthritis and they found the association is inverse.
2. There are previous papers for researchers, having the same style, about association between Mg, osteoarthritis, and hyperuricemia.

Response: Thank you very much for your kind comments. We agree with the reviewer's comment that there are already papers that examined the association between Mg and OA and hyperuricemia. Furthermore, our previous studies have also examined the aforementioned associations (1. Zeng C, et al. Relationship between Serum Magnesium Concentration and Radiographic Knee Osteoarthritis. Journal of Rheumatology, 2015, 42:1231-6; 2. Zeng C, et al. Association between Dietary Magnesium Intake and Radiographic Knee Osteoarthritis. Plos One, 2015, 10:e0127666; 3. Zeng C, et al. Association between low serum magnesium concentration and hyperuricemia. Magnesium Research, 2015, 28:56. 4. Wang Y L, et al. Association between Dietary Magnesium Intake and Hyperuricemia. Plos One, 2015, 10(11):e0141079.) However, this is the first study examining the associations

between serum Mg and the prevalence of metabolic syndrome, diabetes mellitus, hypertension and hyperuricemia in radiographic knee OA patients. We think the study purposes are totally different.

3. What is the file number of ethics approval

Response: Many thanks for your reminder. We have modified relevant sentences in the Method section in our revised manuscript. The file number is 201312459. There we can further provide the original copy of ethics approval, if it is necessary. (Clean version: Page 5 line 95-98)

4. What is about the reference sample of study? Is the reference normal people or patients having only osteoarthritis?. If they were patients only having osteoarthritis, what was the association with Mg?

Response: We set the participants in the 1st quartile of serum Mg (≤0.85) as the reference sample. The serum Mg in the manuscript was classified into five categories based on the quintile distribution: ≤0.85, 0.86-0.89, 0.90-0.92, 0.93-0.96 and ≥0.97 mmol/L. Similar distribution of serum Mg concentration was observed in some previous studies (1. Markovits N, et al. Database evaluation of the association between serum magnesium levels and the risk of atrial fibrillation in the community. International Journal of Cardiology, 2016, 205:142-6; 2. Chen C, et al. Low serum magnesium levels are associated with impaired peripheral nerve function in type 2 diabetic patients. Scientific Reports, 2016, 6. 3. Zhan Y, et al. Association between serum magnesium and anemia: china health and nutrition survey. Biological Trace Element Research, 2014, 159:39-45). Since all included participants were knee OA patients, thus the reference sample of this study is also knee OA patients. In addition, we wonder if there is any misunderstanding because the association between Mg and OA has already been examined in our previous studies (1. Zeng C, et al. Relationship between Serum Magnesium Concentration and Radiographic Knee Osteoarthritis. Journal of Rheumatology, 2015, 42:1231-6; 2. Zeng C, et al. Association between Dietary Magnesium Intake and Radiographic Knee Osteoarthritis. Plos One, 2015, 10:e0127666).

Reviewer: 3
Reviewer Name: John Sayer
Institution and Country: Newcastle University, United Kingdom

This is a cross-sectional study of a population with osteoarthritis of the knee and an examination of the prevalence of metabolic syndrome, hypertension, hyperuricemia and diabetes mellitus and the serum magnesium level. It included 962 patients.

Response: Many thanks!

Major comments

The abstract is difficult to read, with the comparison of different quintiles for each association. I wondered if the second quintile was the second highest or second lowest etc. Please clarify.

Response: Thank you very much for your kind comments. The serum Mg in the manuscript was classified into five categories based on the quintile distribution: ≤0.85, 0.86-0.89, 0.90-0.92, 0.93-0.96 and ≥0.97 mmol/L. The “second quintile” means the second quintile of serum Mg (0.86-0.89mmol/L). Similar distribution of serum Mg concentration was observed in some previous studies (1. Markovits N, et al. Database evaluation of the association between serum magnesium levels and the risk of atrial fibrillation in the community. International Journal of Cardiology, 2016, 205:142-6; 2. Chen C, et al. Low serum magnesium levels are associated with impaired peripheral nerve function in type 2 diabetic patients. Scientific Reports, 2016, 6. 3. Zhan Y, et al. Association between serum magnesium and anemia: china health and nutrition survey. Biological Trace Element Research, 2014, 159:39-45). We are very sorry for the misunderstanding. Thus we have modified the Abstract section in our revised manuscript. We hope that it will be easier to read (Clean version: Page 2 line 20-48).

Please comment on the GFR in the first part of the results, page 9 as I believe this produced signicant differences in Mg levels, and hence vital to correct for this is any further comparisons (table 1 p<0.001). In fact, given this factor, I do not think any of the models that do not include GFR are valid.
Response: We agree with the reviewer's comment that the GFR is an important factor. Therefore, we have conducted a sensitivity analysis by adding GFR into multivariable logistic regression models in our manuscript (Model 3), and the reverse associations remained significant. However, it could not deny the necessity of controlling other potential confounders.

I do not think it is a fair conclusion that MetS is associated in a linear way with serum magnesium levels. The quintiles with a lower prevalence were 2 and 5, with trends not showing much significance. Based on this I don't support the conclusion (page 11 line 272) that Mets is negatively associated with serum Mg. The data does support DM and HU in association with Mg levels in a linear fashion. The conclusions that modifying Mg levels for therapeutic benefit are over interpreting the data. This is too speculative as it is only an association that has been shown, rather than causality. The authors need to be more clear on how these results can be interpreted. Mechanisms are discussed but are not explored in any detail.
Response: We agree with the reviewer's comment that Mets did not associate with serum Mg levels in a linear way. It suggests that the dose-response-relationship between Mg and MetS was failed to be observed in this study. However, it is true that there were negative findings in the relationship between the 2nd and 5th quintiles of serum Mg and MetS, even after adjustments of confounders. To some extent, we believe that Mg is inversely associated with MetS in knee OA patients, but this association was not in a dose-response-relationship manner. We appreciate your kind comments, and thus we modified relevant statements about the association between Mg and MetS in the Result, Discussion and Abstract sections in our revised manuscript (Clean version: Page 2 line 32-45; Page 8 line 197-208; Page 10 line 252-253; Page 12 line 314-315). Moreover, we added a paragraph about potential mechanisms of our results in the Discussion section in our revised manuscript. (Clean version: Page 9-10 line 266-280) We hope that it could make our manuscript more qualified.

Figure is too basic and adds nothing to the understanding of the data.
Response: Many thanks for your advice. After discussion, we decided to delete this figure in our revised manuscript.

Minor
There are numerous English language problems
Response: We are very grateful to you for your kind and elaborate corrections, very grateful! We are very sorry for these language problems, and we have polished our manuscript.

Page 4 line 73 – "adusted by a" change to "adjusted for a"
Response: Done accordingly (Clean version: Page 3 line 54).

Page 4 line 76 "Kidney is" change to "The kidney is"
Response: Done accordingly (Clean version: Page 3 line 57).

Page 4 line 82 "which was not the best" change to "which may not be"
Response: Done accordingly (Clean version: Page 3 line 63).

Page 5 line 96 should read "injury"
Response: Done accordingly (Clean version: Page 4 line 69).

Page 5 line 101 change "take measures" to "adopt measures"
Response: Done accordingly (Clean version: Page 4 line 73).

Page 5 line 107 change “examine” to “examining”
Response: Done accordingly (Clean version: Page 4 line 80).

Page5 line 109 change “another study of ours” to “we have previously shown”
Response: Done accordingly (Clean version: Page 4 line 82).

Page 5 line 110 change “it is reasonably speculated” to “we speculate”
Response: Done accordingly (Clean version: Page 4 line 84).

Page 6 line 123 – please give name of ethics committee
Response: Done accordingly (Clean version: Page 5 line 95-98).

Page 9 line 209 – please state whether this is MDRD or another variation of eGFR formulae
Response: Done accordingly (Clean version: Page 7-8 line 183-185).

Page 9 line 214 – test – should be tests, which tests specifically
Response: Done accordingly (Clean version: Page 8 line 188).

Page 12 line 289-90 – doesn't make sense
Response: Many thanks for your advice. We have deleted this sentence.

Page 12 line 298 – this is too speculative as it is only an association that has been shown, rather than causality.
Response: Many thanks for your advice. We have modified relevant statements (Clean version: Page 11 line 281-289).

Reviewer: 4
Reviewer Name: Neslihan GOKCEN
Institution and Country: Cukurova University Faculty of Medicine, Adana/TURKEY

I do thank you for your valuable well-design study. Some diseases such as diabetes mellitus and some drugs such as loop diuretics, thiazide diuretics can cause hypomagnesemia. The medical history of patients may be suggested to evaluate whether the drugs are a confounder. Despite all these confounding factors, authors analyzed the outcomes by multivariable methods and this method strengthened the study. The article is well written and well presented. I have some minor comments.
Response: Thank you very much!

Minor Comments:
1. The medical history of patients may be suggested to evaluate whether the drugs are a confounder.
Response: Many thanks for your advice. In fact, relevant medication status (including anti-hypertensive, anti-hyperuricemia and anti-diabetes drugs) have already been assessed during the face-to-face interview in this cross-sectional study, which served as diagnostic criteria of hypertensive, hyperuricemia and diabetes in our study (Clean version: Page 6 line 136, 144-150).

2. You may extend the limitations.
Response: Done accordingly. We have extended the limitations in our revised manuscript (Clean version: Page 12 line 306-307).

3. Please check the table. You used the abbreviation for metabolic syndrome as MS.
Response: Thank you so much for your correction (Clean version: Page 20 line 352).

4. Please arrange the figure more intelligible.

Response: Thanks for your kind comments. After discussion, we decided to delete this figure in our revised manuscript because it is too basic and adds nothing to the understanding of the data.

VERSION 2 – REVIEW

REVIEWER	PROFESSOR O B FAMILONI DEPT OF MEDICINE OLABISI ONABANJO UNIVERSITY TEACHING HOSPITAL SAGAMU, NIGERIA
REVIEW RETURNED	12-Feb-2018

GENERAL COMMENTS	The authors have complied significantly with the issues raised in the original manuscript

REVIEWER	Neslihan GOKCEN Cukurova University Faculty of Medicine, TURKEY
REVIEW RETURNED	28-Jan-2018

GENERAL COMMENTS	To Authors, I do thank you for your valuable well-design study. I think the present study will contribute the literature in terms of showing the association between magnesium and osteoarthritis.

REVIEWER	Teeranan Angkananard Division of Cardiovascular Medicine, Department of Medicine, Faculty of Medicine, HRH Princess Maha Chakri Sirindhorn Medical Center, Srinakharinwirot University, Nakhon Nayok, Thailand
REVIEW RETURNED	15-Feb-2018

GENERAL COMMENTS	1. Why did the authors estimate eGFR with MDRD formula, whether the results were the same with CKD-EPI: 2009 formula? 2. It 's not clear about covariates used in measurement of serum Mg concentration. Those covariates used as described in citations, they were analyzed with metabolic syndrome and DM, not for Mg itself.

REVIEWER	Miguel de Carvalho University of Edinburgh
REVIEW RETURNED	20-Mar-2018

GENERAL COMMENTS	Statistical Review The authors employ multivariable logistic regression methods so to examine if serum magnesium is negatively related with a few diseases; to streamline the report I will focus on DM. The statistical analysis is not sound and it would need to be substantially revised and further clari ed. Here are some concerns: Why partitioning serum Mg (S_{mg}) into ve categories? : The inquiry of interest could be conducted with a simple logistic regression model with p denoting the probability of a subject su ering from DM.

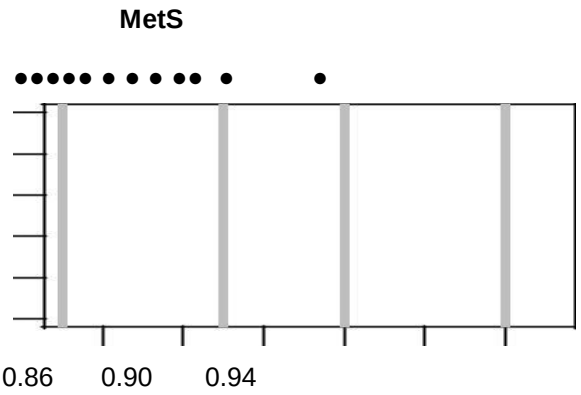
$$\log \frac{1\ p}{p} = + S_{mg};$$

Mann{Whitney statistics: It would be appropriate to report Mann{Whitney statistics. I presume these should be smaller than 0.5 given that from Table 2 a lower value of serum Mg is less indicative of DM. However, for HP I would suspect that the value should be around 0.5 given the values reported in Table 4.

Writing: On the section on statistical analysis I recommend starting from the onset with the models \Logistic regression was conducted...". Also, revise \Model 1 were adjusted for age and sex" and be more concrete about what you mean here; I presume that the authors meant that they consider age and sex as covariates, but this should be clari ed.

Visualization: Resorting to visualization could facilitate communicating the statistical analysis. The R code (R Development Core Team, 2016) below could be used to obtain a chart such as the attached gure.

```
## R code

plot(x, y, xlab =
Serum Mg , ylab    =    , main = "MetS", pch = 16)

abline(v = 0.85,
col = "grey", lwd    = 3)

abline(v = 0.89,
col = "grey", lwd    = 3)

abline(v = 0.92,
col = "grey", lwd    = 3)

abline(v = 0.96,
col = "grey", lwd    = 3)
```

	 Serum Mg BMI : In Table 1 the authors report descriptive statistics on BMI. So why considering in model 2 a dummy variable for BMI instead of using the raw data? References R Development Core Team (2016), R: A Language and Environment for Statistical Computing, Vienna, Austria: R Foundation for Statistical Computing.

REVIEWER	Maria Sanchez University of Oxford
REVIEW RETURNED	26-Mar-2018

GENERAL COMMENTS	The statistical analysis is very long and it might be streamlined. It currently includes a lot of repeated information. For example, covariates included in the model 2 are repeated for each outcome. Type of variable (continuous, binary, etc.) could be described in “methods” in the “Assessment of other exposures” section, so the section would be easy to follow.

VERSION 2 – AUTHOR RESPONSE

Reviewer: 4

Reviewer Name: Neslihan GOKCEN

Institution and Country: Cukurova University Faculty of Medicine, TURKEY

To Authors,

I do thank you for your valuable well-design study. I think the present study will contribute the literature in terms of showing the association between magnesium and osteoarthritis.

Response: We are thankful to you for pointing out some important modifications needed in our previous manuscript.

Reviewer: 1

Reviewer Name: PROFESSOR O B FAMILONI

Institution and Country: DEPT OF MEDICINE, OLABISI ONABANJO UNIVERSITY TEACHING HOSPITAL, SAGAMU, NIGERIA

The authors have complied significantly with the issues raised in the original manuscript

Response: We are thankful to you for pointing out some important modifications needed in our previous manuscript.

Reviewer: 5

Reviewer Name: Teeranan Angkananard

Institution and Country: Division of Cardiovascular Medicine, Department of Medicine, Faculty of Medicine, HRH Princess Maha Chakri Sirindhorn Medical Center, Srinakharinwirot University, Nakhon Nayok, Thailand

1.Why did the authors estimate eGFR with MDRD formula, whether the results were the same with CKD-EPI: 2009 formula?

Response: Many thanks for your professional comments. We chose the MDRD equation, primarily because it is common in the clinic, as well as epidemiological studies. However, the CKD-EPI equation has recently been established as the most accurate method through validation studies. Thus, we believe that it is reasonable and acceptable for the reviewer to raise this comment. Therefore, we used the CKD-EPI 2009 to estimate eGFR and re-did the relevant statistical analyses in the revised manuscript (track version: line 175-177; clean version: 173-174). Finally, the results did not change materially.

2.It's not clear about covariates used in measurement of serum Mg concentration. Those covariates used as described in citations, they were analyzed with metabolic syndrome and DM, not for Mg itself.

Response: Covariates in our manuscript were chosen referring to some of the previous similar studies. The objectives of these studies were also to investigate associations between Mg and some metabolic diseases. Thus, we believe that it is not uncommon to choose these covariates for analyses. Moreover, we agree with the reviewer that we should include some covariates about Mg itself. Therefore, in view of the kidney is the key organ in maintaining Mg homeostasis, we have conducted a sensitivity analysis by adding eGFR into multivariable logistic regression models which showed that the reverse associations remained significant.

Thanks again for pointing out important modifications needed in our manuscript.

Reviewer: 6

Reviewer Name: Miguel de Carvalho

Institution and Country: University of Edinburgh

See attached pdf.

1.Why partitioning serum Mg (Smg) into five categories? The inquiry of interest could be conducted with a simple logistic regression model

log(p/(1-p))=α+βSmg

with p denoting the probability of a subject suffering from DM.

Response: Many thanks for your kind comments. Actually, we believe that it is not an uncommon practice in similar observational studies that partitioning serum Mg into five categories [1-6]. Meanwhile, we also believe that it is helpful for health care providers to facilitate the clinical decision making because the odds ratios in that environment are more telling and easy to be understood.

References:

[1].Xiao Y, Soohoo M, Streja E, et al. Serum Magnesium Levels and Hospitalization and Mortality in Incident Peritoneal Dialysis Patients: A Cohort Study. American Journal of Kidney Diseases, 2016, 68(4):619-627.

[2].Markovits N, Kurnik D, Halkin H, et al. Database evaluation of the association between serum magnesium levels and the risk of atrial fibrillation in the community. International Journal of Cardiology, 2015, 205:142.

[3].Zeng C, Wang Y L, Wei J, et al. Association between low serum magnesium concentration and hyperuricemia. Magnesium Research Official Organ of the International Society for the Development of Research on Magnesium, 2015, 28(2):56.

[4].Jr E L, Wang W, Ma L, et al. Serum Magnesium and Mortality in Hemodialysis Patients in the United States: A Cohort Study. American Journal of Kidney Diseases, 2015, 66(6):1056-1066.

[5].Lutsey P L, Alonso A, Michos E D, et al. Serum magnesium, phosphorus, and calcium are associated with risk of incident heart failure: the Atherosclerosis Risk in Communities (ARIC) Study. American Journal of Clinical Nutrition, 2014, 100(3):756.

[6].Joosten M M, Gansevoort R T, Mukamal K J, et al. Urinary and plasma magnesium and risk of ischemic heart disease. American Journal of Clinical Nutrition, 2013, 97(6):1299-306.

2.Mann-Whitney statistics: It would be appropriate to report Mann-Whitney statistics. I presume these should be smaller than 0.5 given that from Table 2 a lower value of serum Mg is less indicative of DM.

However, for HP I would suspect that the value should be around 0.5 given the values reported in Table 4.

Response: Many thanks for your kind consideration. However, we are very sorry that we do not quite understand why it would be appropriate to report Mann-Whitney U statistics. We believe that although this kind of statistical method is effective but only can provide an unadjusted OR. The followings are the results of Mann-Whitney U tests in the present manuscript (please see attached file-Cover letter-20180426). We can find these results are similar with the results of model 1 or 2 or 3 in our manuscript. Anyway, if it is necessary for us to add these results, we are willing to do further revision next time.

3.Writing: On the section on statistical analysis I recommend starting from the onset with the models "Logistic regression was conducted...". Also, revise "Model 1 were adjusted for age and sex" and be more concrete about what you mean here; I presume that the authors meant that they consider age and sex as covariates, but this should be clarified.

Response: We are very sorry for our incorrect and convoluted writing in this part. Thus, we have re-written this part according to your suggestion. We hope that the revision will be streamlined and meet with your approval (track version: line 152 to 207; clean version: line 150 to 180).

4.Visualization: Resorting to visualization could facilitate communicating the statistical analysis. The R code (R Development Core Team, 2016) below could be used to obtain a chart such as the attached figure.

Response: Thank you very much for your thoughtful and thorough guidance. One figure including 4 charts according to the R code provided by you (Figure 1, please see attached file-Cover letter-20180426) and relevant statements were added in the revised manuscript (track version: 188-189, 205, 221-222, 234-235, 251-252, 258-259, 812; clean version: 156-157, 178, 194-195, 207-208, 224-225, 231-232, 567).

5.BMI: In Table 1 the authors report descriptive statistics on BMI. So why considering in model 2 a dummy variable for BMI instead of using the raw data?

Response: Special thanks to you for your professional comments. We are very sorry for our negligence of the inconsistency. Thus, we re-did relevant statistics by using BMI (continuous data) in the revised manuscript (track version: line 164, 817-823, 828-829, 834-835; clean version: 161-162, 577, 583, 589). Finally, the results did not change materially.

Thanks again for pointing out important modifications needed in our manuscript!

Reviewer: 7

Reviewer Name: Maria Sanchez

Institution and Country: University of Oxford

The statistical analysis is very long and it might be streamlined. It currently includes a lot of repeated information. For example, covariates included in the model 2 are repeated for each outcome. Type of variable (continuous, binary, etc.) could be described in “methods” in the “Assessment of other exposures” section, so the section would be easy to follow.

Response: Many thanks for your kind comments. We are very sorry for our convoluted writing in this part. Thus, we have re-written this part according to your suggestion. We hope that the revision version will be streamlined and meet with your approval. Special thanks again to you for your good comments. (track version: line 152 to 207; clean version: line 150 to 180)

VERSION 3 – REVIEW

REVIEWER	Miguel de Carvalho University of Edinburgh
REVIEW RETURNED	28-Apr-2018

GENERAL COMMENTS	The revision has improved substantially, but there are two key aspects with the analysis that are critical and that require further clarification and revisions. My main concern is that the Mann-Whitney statistics reported by the authors in the letter, along with Table 2, and the new Fig. 1 in the paper, cast doubts on the the strength of association between serum magnesium and Met. This should be revised in the manuscript including the section on results; the claims on inverse association (if any) are definitely too strong, compared with those on DM. In addition to this, I presume there are overlapping points in Figure 1, and if that is the case this needs to be clarified in the text. Indeed, if that was not the case, Figure 1 would actually suggest that the levels of Serum MG would have a low ability to differentiate between diseased and non-diseased subjects (for any of the conditions under analysis).

REVIEWER	Maria Sanchez-Santos, MSc University of Oxford, Oxford, UK
REVIEW RETURNED	14-May-2018

GENERAL COMMENTS	My recommendation is accept with no changes. The authors have satisfactorily made the necessary changes to the manuscript.

VERSION 3 – AUTHOR RESPONSE

Reviewer(s)' Comments to Author:

Reviewer: 6
Reviewer Name: Miguel de Carvalho
Institution and Country: University of Edinburgh
Please state any competing interests or state ‘None declared’: None declared

Please leave your comments for the authors below
The revision has improved substantially, but there are two key aspects with the analysis that are critical and that require further clarification and revisions. My main concern is that the Mann-Whitney

statistics reported by the authors in the letter, along with Table 2, and the new Fig. 1 in the paper, cast doubts on the strength of association between serum magnesium and Met. This should be revised in the manuscript including the section on results; the claims on inverse association (if any) are definitely too strong, compared with those on DM. In addition to this, I presume there are overlapping points in Figure 1, and if that is the case this needs to be clarified in the text. Indeed, if that was not the case, Figure 1 would actually suggest that the levels of Serum MG would have a low ability to differentiate between diseased and non-diseased subjects (for any of the conditions under analysis).
Response: We appreciate your professional comments. Thus we referenced similar statements in a previous high-quality study (Bleys J, Navas-Acien A, Guallar E. Serum selenium and diabetes in U.S. adults. Diabetes Care, 2007, 30(4):829), and modified corresponding statements about the association between Mg and MetS in the Results, Discussion and Abstract sections in our revised manuscript (clean version: line 40-41, 200-208, 254-256; track version: line 40-41, 201-210, 257-262). We hope that the revision will be reasonable and meet with your approval.
In addition, to show the overlapping points, we colorized the Fig. 1 based on the prevalence of MetS under different quintiles of serum Mg concentration, and added detail instructions in the figure legend (clean version: line 569-573; track version: line 574-578). Thanks again for pointing out important modifications needed in our manuscript!

Reviewer: 7
Reviewer Name: Maria Sanchez-Santos, MSc
Institution and Country: University of Oxford, Oxford, UK
Please state any competing interests or state ‘None declared’: None

Please leave your comments for the authors below
My recommendation is accept with no changes. The authors have satisfactorily made the necessary changes to the manuscript.
Response: We are thankful to you for pointing out some important modifications needed in our previous manuscript.

VERSION 4 – REVIEW

REVIEWER	Miguel de Carvalho University of Edinburgh
REVIEW RETURNED	04-Jul-2018

GENERAL COMMENTS	Thank you for carefully revising the manuscript and for taking on board these final concerns.

VERSION 4 – AUTHOR RESPONSE

Reviewer(s)' Comments to Author:
Reviewer: 6
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Please state any competing interests or state ‘None declared’: None declared

Please leave your comments for the authors below
Thank you for carefully revising the manuscript and for taking on board these final concerns.
Response: Many thanks for your kind comments!