PEER REVIEW HISTORY

ARTICLE DETAILS

TITLE (PROVISIONAL)	Prevalence and associated factors of microalbuminuria in Chinese individuals without diabetes: cross sectional study
AUTHORS	Xiao, Jianzhong; Xing, Xiaoyan; Lu, Juming; Weng, Jianping; Jia, Weiping; Ji, Linong; Shan, Zhongyan; Liu, Jie; Tian, Haoming; Ji, Qiuhe; Zhu, Dalong; Ge, Jiapu; Chen, Gang; Chen, Li; Guo, Xiaohui; Zhao, Zhigang; Li, Qiang; Zhou, Zhiguang; Yang, Zhao-Jun; Shan, Guang-Liang; He, Jiang; Yang, Wenying

VERSION 1 - REVIEW

REVIEWER	Juliana CN Chan Chair Professor Department of Medicine and Therapeutics The Chinese University of Hong Kong The Prince of Wales Hospital Shatin, Hong Kong I do not have conflict of interest relevant to the paper.
REVIEW RETURNED	07-Jul-2013

THE STUDY	Using a large dataset which had previously reported the prevalence of diabetes in Chinese population, the authors examined the prevalence of microalbuminuria (MAU) in over 34,000 subjects without diabetes (based on 75g OGTT) and reported a prevalence of 16% in men and 25% in women. They further reported that SES factors (education, farmer, female, income levels) were independently associated with risk of MAU. This is a potentially interesting and informative analysis. However, the hypthesis has not been well defined and as such, the analysis has not addressed the research question systematically. Motivated by previous reports on the association between SES and MAU, the author aimed to validate the observation in this large community-based cohort. However, since many of the risk factors for MAU (e.g. BG, BP, lipid, smoking and obesity) are also associated with SES, it will be more appropriate to first test the risk association between MAU and SES and then adjust for demographic factors (e.g. age, sex, area of residence) and then (BP, BP, lipids, obesity and smoking) and then less conventional risk factors (insulin resistance index) and see if the risk associations of MAU with SES will be progressively attenuated. If so, the results would suggest that the risk association was partly or wholly mediated by these risk factors. If the risk association between MAU and SES persists after these adjustments, other unmeasured variables e.g. dietary factors, exercise, psychosocial stress might contribute to the association.

	It is not usual to consider women and famer as having low SES which is often defined by education and income which nevertheless may not be correlated (i.e. highly educated people may have low income or vice versa). Besides, farmer can be categorised under manual worker if occupation is used to define SES. Since male and female have major biological differences while the farmer status is heavily linked to area of residence (urban versus rural) which in turn have many other confounders, e.g. access to care, nutritional factors etc), these factors should be adjusted separately rather then considered as a SES factor. Thus, it is important to clearly define SES (e.g. low education and/or low income) with appropriate references. Apart from analysing the risk association of SES with MAU as a category, it will be interesting to examine the dose effects of SES (0, 1, 2) on such risk using both univariable and multivariable analysis. Was MAU measured in a central laboratory? 95% confidence intervals should be given with prevalence. There are multiple grammatical and spelling errors and the paper will benefit from pofessional editing to increase the flow of arguments and and clarity of presentation.

REVIEWER	Prof. Dr. Dr. h.c. Walter F. Riesen Director emeritus Center for Laboratory Medicine Kantonsspital CH-9000 St. Gallen
REVIEW RETURNED	09-Jul-2013

GENERAL COMMENTS	Spot collection of urine: was it the second morning urine, which was collected?

REVIEWER	MacIsaac, Richard University of Melbourne, Endocrinology & Diabetes
REVIEW RETURNED	23-Jul-2013

REPORTING & ETHICS	This study documents the prevalence of microalbuminuria in 35,430 Chinese individuals without diabetes. Diabetes was excluded on the results of an oral glucose tolerance test. The prevalence of microalbuminuria was related to a number of socioeconomic factors and metabolic disorders. The prevalence of microalbuminuria was 16.9% and 25.1% in mean and women, respectively. A multivariate logistic analysis demonstrated that microalbuminuria was strongly associated with female sex, low education level, low occupation level, high blood pressure, obesity, old age and higher blood glucose levels within the normal range. It was suggested that the results of this study could encourage the development strategies to lower the risk of microalbuminuria in susceptible populations. The methodology of the study appears to be sound. The results also appear to be plausible. My recommendation is that the paper would be acceptable for publication after some major revisions. In particular, my concerns are:

	1. Only one measurement of the urinary albumin to creatinine ratio was made to document the presence of microalbuminuria. There is a large variability in albumin excretion rates and the author should acknowledge this limitation. Even for diabetic patients with established microalbuminuria, spontaneous regression or remission is a common occurrence. 2. Gender specific cut offs for microalbuminuria have not been used. Possibly, the lack of gender specific ACR cut-offs may have influenced the high prevalence of microalbuminuria reported for females. Could the authors re-analyse their data using recommended sex-specific cut-offs for ACR and report these results? Other studies have documented a higher prevalence of microalbuminuria in males. 3. Is any information available regarding the smoking status of the participants in this study? Smoking history has been associated with a high prevalence of microalbuminuria in other studies. 4. Higher ACR levels are seen in individuals with sub-clinical inflammation/infection. I imagine that no information has been collected regarding the dental health of participants in this study. However, I would postulate that the relatively high presence of microalbuminuria may be related to periodontal disease, which would be expected to have a higher prevalence in economically underdeveloped areas. Would the authors like to make any comments about this suggestion? 5. The authors state that the results of this study should encourage the development of strategies to lower the risk of microalbuminuria in susceptible populations. Do they have any specific suggestions as to how this may be achieved? Minor points: • In the abstract, under the primary and secondary outcome measures, the units for ACR should be mg/g. • Could the authors please re-align the tables, so that the last bracket is not on a new line in tables 1 and 2 • I found Figure 1 difficult to read. Could the authors please supply an enlarged and clearer version of this figure? • The authors should quote the results of the PREVEND Study. This was a large study documenting the prevalence of microalbuminuria in Groningen in the Netherlands. A prevalence of microalbuminuria of 6.6% was reported for subjects without diabetes or vascular risk factors. For hypertensive subjects, the prevalence was 11%. Over 40,000 subjects were initially screened for microalbuminuria in the study. (Hillege et al, Circulation 2002, 106-1777-82) • The authors have quoted a sub-study from the 'LIFE STUDY' (Wachtell, Journal of Hypertension, 2002, 20, 405-412) to support their statement that 'A recent study demonstrated methods that decrease microalbuminuria, may reduce the risk for end stage renal disease' (See page 12). I feel that this is not an appropriate reference to use in this setting. The results reported from this particular study by Wachtell, are only observational in nature and suggest that a higher level of urinary albumin, is associated with a higher serum creatinine level. I don't think patients will follow through to end stage renal disease in this study. Could the authors please check this and think about another reference to support this statement? Possibly they could use results of the Steno 2 Study (the last study reported from this group of patients, which documents clinical end points, such as progression to end stage renal disease and cardiac death. It was published in the New England Journal of Medicine)

VERSION 1 – AUTHOR RESPONSE

To Professor Juliana CN Chan:

This is a potentially interesting and informative analysis. However, the hypothesis has not been well defined and as such, the analysis has not addressed the research question systematically.
Motivated by previous reports on the association between SES and MAU, the author aimed to validate the observation in this large community-based cohort. However, since many of the risk factors for MAU (e.g. BG, BP, lipid, smoking and obesity) are also associated with SES, it will be more appropriate to first test the risk association between MAU and SES and then adjust for demographic factors (e.g. age, sex, area of residence) and then (BP, BP, lipids, obesity and smoking) and then less conventional risk factors (insulin resistance index) and see if the risk associations of MAU with SES will be progressively attenuated. If so, the results would suggest that the risk association was partly or wholly mediated by these risk factors. If the risk association between MAU and SES persists after these adjustments, other unmeasured variables e.g. dietary factors, exercise, psychosocial stress might contribute to the association.
It is not usual to consider women and famer as having low SES which is often defined by education and income which nevertheless may not be correlated (i.e. highly educated people may have low income or vice versa). Besides, farmer can be categorised under manual worker if occupation is used to define SES.
Since male and female have major biological differences while the farmer status is heavily linked to area of residence (urban versus rural) which in turn have many other confounders, e.g. access to care, nutritional factors etc), these factors should be adjusted separately rather then considered as a SES factor.
Thus, it is important to clearly define SES (e.g. low education and/or low income) with appropriate references. Apart from analysing the risk association of SES with MAU as a category, it will be interesting to examine the dose effects of SES (0, 1, 2) on such risk using both univariable and multivariable analysis.

We thank the reviewer for these valuable insights and suggestion. We re-defined the low SES and adopted a gender-specific cut-off value for microalbuminuria. We agree that MAU is associated with many factors so we catgorised the factors into 3 distinct groups that included 1)conventional demographic factors and metabolic factors and factors; 2)SES factors including personal education levels, family income levels, and occupation; and 3)residence including rural/urban residence, and regional economic development level. We then reran our analyses to show more clearly the unique affect of these covariates. The revised results are shown in figure 1-2 and table 3.

Was MAU measured in a central laboratory? 95% confidence intervals should be given with prevalence.

The ACR was measured in a commercialized lab in Beijing. The urine specimens were refrigerated during transport. We included these detail, along with the 95% CIs in the revised manuscript.

There are multiple grammatical and spelling errors and the paper will benefit from pofessional editing to increase the flow of arguments and and clarity of presentation.
We received help from a professional English speaking editor to help the language issues.

To Prof. Dr. Dr. h.c. Walter F. Riesen

1.Spot collection of urine: was it the second morning urine, which was collected?

The spot urine was collected in the morning, but not necessarily the second or first morning urine.

2. Only one measurement of the urinary albumin to creatinine ratio was made to document the presence of microalbuminuria. There is a large variability in albumin excretion rates and the author should acknowledge this limitation. Even for diabetic patients with established microalbuminuria, spontaneous regression or remission is a common occurrence.
We agree that this limitation of method of study (one time spot urine sample) and included this limitation in the revised manuscript.

3. Gender specific cut offs for microalbuminuria have not been used. Possibly, the lack of gender specific ACR cut-offs may have influenced the high prevalence of microalbuminuria reported for females. Could the authors re-analyse their data using recommended sex-specific cut-offs for ACR and report these results? Other studies have documented a higher prevalence of microalbuminuria in males.
Thank you for your constructive suggestion. We agree that the lack of gender specific ACR was likely to have overestimated the prevalence in women. Therefore, we adopted 22.1 to 299 mg/g for men and 30.9 to 299 mg/g for women as cut-off value and made a new calculation. Amazingly, there was no difference of MAU prevalence between men and women. We re-calculated all data and made new figures 1-2 and table 3 accordingly.

4. Is any information available regarding the smoking status of the participants in this study? Smoking history has been associated with a high prevalence of microalbuminuria in other studies.
We did have data on smoking, but we did not find smoking status to be associated in either men or women with MAU in multivariate analyses. The prevalence of MAU in no-smokers versus smokers were 24.9%, and 23.9% in men, and 24.6% and 22.0%in women.

5. Higher ACR levels are seen in individuals with sub-clinical inflammation/infection. I imagine that no information has been collected regarding the dental health of participants in this study. However, I would postulate that the relatively high presence of microalbuminuria may be related to periodontal disease, which would be expected to have a higher prevalence in economically underdeveloped areas. Would the authors like to make any comments about this suggestion?
Yes, it is highly likely that the association between microalubminuria, periodontal disease, and low SES played a part in our results. Unfortunately we did not have data on periodontal disease. We included a discussion of this issue in the revised manuscript.

6. The authors state that the results of this study should encourage the development of strategies to lower the risk of microalbuminuria in susceptible populations. Do they have any specific suggestions as to how this may be achieved?
We appreciate this suggestion and included specific suggestions to lower the risk of microalbuminuria in susceptible populations – i.e. reforming the healthcare system, improving access to health facilities, promoting health education especially in population with low SES in under-developed region, and preventing periodontal diseases.
Minor points:
7.In the abstract, under the primary and secondary outcome measures, the units for ACR should be mg/g.
Thank you very much. We made this correction in the manuscript.

8 Could the authors please re-align the tables, so that the last bracket is not on a new line in tables 1 and 2

We realigned the tables as recommended.

9. I found Figure 1 difficult to read. Could the authors please supply an enlarged and clearer version of this figure?
We enlarge all figures and made changes in figure 2 in response to suggestions from Reviewer #1 and the change of definition of MAU.

10.The authors should quote the results of the PREVEND Study. This was a large study documenting the prevalence of microalbuminuria in Groningen in the Netherlands. A prevalence of microalbuminuria of 6.6% was reported for subjects without diabetes or vascular risk factors. For hypertensive subjects, the prevalence was 11%. Over 40,000 subjects were initially screened for microalbuminuria in the study. (Hillege et al, Circulation 2002, 106-1777-82)
Thank you for pointing out this important study. We cited the paper as suggested in the revised manuscript.

11.The authors have quoted a sub-study from the 'LIFE STUDY' (Wachtell, Journal of Hypertension, 2002, 20, 405-412) to support their statement that 'A recent study demonstrated methods that decrease microalbuminuria, may reduce the risk for end stage renal disease' (See page 12). I feel that this is not an appropriate reference to use in this setting. The results reported from this particular study by Wachtell, are only observational in nature and suggest that a higher level of urinary albumin, is associated with a higher serum creatinine level. I don't think patients will follow through to end stage renal disease in this study. Could the authors please check this and think about another reference to support this statement? Possibly they could use results of the Steno 2 Study (the last study reported from this group of patients, which documents clinical end points, such as progression to end stage renal disease and cardiac death. It was published in the New England Journal of Medicine)
Thank you for pointing this out. We used two new references, one is from the PREVEND-IT study and another one you suggested from steno-2 study. We made changes in the discussion section as a result.

VERSION 2 – REVIEW

REVIEWER	Professor Richard MacIsaac, Bsc(HONS), PhD, MBBS, FRACP Director, Department of Endocrinology & Diabetes, St Vincent's Hospital, Melbourne Professorial Fellow, University of Melbourne Senior Principal Research Associate, St Vincent's Institute of Medical Research Acting Director of Research, St Vincent's Hospital, Melbourne I have no conflict of interest to declare
REVIEW RETURNED	26-Aug-2013

The reviewer completed the checklist but made no further comments