PEER REVIEW HISTORY

ARTICLE DETAILS

TITLE (PROVISIONAL)	Is self-rated health associated with cardiovascular risk factors and disease in a low-income setting? A cross-sectional study from the Amazon Basin of Brazil
AUTHORS	Holm, Anna Engell; Gomes, Laura; Wegener, Alma; Lima, Karine; Matos, Luan; Vieira, Isabelle; Kaagaard, Molly; Pareek, Manan; Medeiros de Souza, Rodrigo; Romero Farias Marinho, Claudio; Biering-Sorensen, Tor; Silvestre, Odilson; Brainin, Philip

VERSION 1 – REVIEW

REVIEWER	Gholamrezanezhad, Ali USC Keck School of Medicine, Radiology
REVIEW RETURNED	12-Dec-2021

GENERAL COMMENTS	I had the great pleasure to review the manuscript entitled "Association between self-rated health, cardiovascular risk factors and echocardiography: A study from the Amazon Basin, Brazil". The manuscript is well designed and organized. Although the study is important, I believe there are significant limitations with this paper. More specifically, the studied population is only from a certain part of the world, which limits generalizability of the findings to the rest of the world. Several aspects of such questionnaires can be affected by cultural factors. I suggest performing a large population based study from different cultures, if the authors would like to publish it in an international journal. Otherwise, this manuscript is well suited for a local journal. - No information about the second questionnaire has been provided. Was it validated? Who filled the questionnaires? The participant or the interviewer? My other main concern about this study is the fact that there is not much novelty. In fact ,this study does not add that much to our current knowledge.

REVIEWER	Islam, AKM Monwarul National Institute of Cardiovascular Diseases, Department of Cardiology
REVIEW RETURNED	15-Dec-2021

GENERAL COMMENTS	Good work. However, there are some observations. 1. In this study, the echocardiographic parameters have been termed “Subclinical cardiac disease by echocardiography”, but a proportion of the study subjects had LVEF <45%. Were they ‘subclinical’?

	2. In the REFERENCE section, some references need correction and completion, e.g., Ref. 33, 36, 38, 39, as follows” (33) Salomon JA, Tandon A, Murray CJ. Comparability of self rated health: cross sectional multi-country survey using anchoring vignettes. BMJ. 2004 Jan 31;328(7434):258. doi: 10.1136/bmj.37963.691632.44. Epub 2004 Jan 23. PMID: 14742348; PMCID: PMC324453. (36) May M, Lawlor DA, Brindle P, Patel R, Ebrahim S. Cardiovascular disease risk assessment in older women: can we improve on Framingham? British Women's Heart and Health prospective cohort study. Heart. 2006 Oct;92(10):1396-401. doi: 10.1136/hrt.2005.085381. Epub 2006 Mar 17. PMID: 16547204; PMCID: PMC1861043. (38) McFadden E, Luben R, Wareham N, Bingham S, Khaw KT. Occupational social class, risk factors and cardiovascular disease incidence in men and women: a prospective study in the European Prospective Investigation of Cancer and Nutrition in Norfolk (EPIC-Norfolk) cohort. Eur J Epidemiol. 2008;23(7):449-58. doi: 10.1007/s10654-008-9262-2. Epub 2008 May 29. PMID: 18509727. (39) Kaplan GA, Keil JE. Socioeconomic factors and cardiovascular disease: a review of the literature. Circulation. 1993 Oct;88(4 Pt 1):1973-98. doi: 10.1161/01.cir.88.4.1973. PMID: 8403348.

REVIEWER	Moe, Kjartan Baerum Hospital
REVIEW RETURNED	31-Dec-2021

GENERAL COMMENTS	General comments Holm et al present a very interesting dataset from the Amazon Basin. Cardiovascular disease is still one of the most important causes of mortality, especially in low- and middle-income countries. Contrasting this, most data are collected in high-resource settings, possibly limiting their generalization to low- and middle-income areas. Thus, studies from such settings are highly welcome The current study examines the associations between self-rated health (SRH) and cardiovascular risk factors (CRF). However, neither the abstract, the introduction, nor the discussion explains clearly why this is interesting to study. In the discussion (p.13, lines 14-23) it is stated that “Considering the close relationship we found with several cardiovascular risk factors, self-rated health could be obtained by non-medical personnel and enable screening of remote communities. Consequently, selected individuals, i.e., persons with low self-rated health and no known cardiovascular risk factors, could be referred for risk factor optimization in healthcare facilities.» But as the outcomes associated with SRH are rather crude (which of course is often the case in low-resource settings), would it not be quite easy to register these in a low-resource setting as well, providing more specific information about cardiovascular risk than the SRH? Furthermore, I do not think your data support that SRH could be used to detect unknown cardiovascular disease. On the contrary, SRH failed to associate with echocardiographic parameters. What could be interesting to

	look at in this context, are possible associations between SRH and blood pressure levels. The inclusion process should be described more thoroughly, preferably with the support of a flow chart. What was the purpose of including a second group of patients (p.7 lines 15-20). Could this affect generalization of your results? Would the results be the same if this group was excluded? Obviously only persons who responded to all questionnaires were included (p.7 line 5), but how many were there, and do you have reasons to believe that these were different from the rest of your population? There are also some concerns and questions to the statistics in the paper: Were multivariate regression analysis conducted by forced entry of all the variables, or did you apply any selection criterion (p. 10 lines 46-55)? Table 1 and supplemental table 1 are lacking information on the statistical methods applied and information about what is presented (mean/median etc.). Obviously, the median should be presented for not normally distributed values. To me, it seems like this is not the case. Please elaborate and specify. I guess a sample size analysis was not conducted as this was a secondary study? Please, specify in your statistics section. Low sample size might explain the lack of any associations between SRH and subclinical cardiac disease. Please comment in your discussion part. Specific comments p.2 lines 4-7 Title: Does not adhere to journal guidelines. Should include study design. p. 4 lines 23-27 Article summary: You state “strengths and limitations”, but what follows is a summary of major results and a conclusion, not strengths and limitations. Please correct. p.5 line 7 “Cardiovascular disease is the leading cause of mortality worldwide”. Please find a suitable reference for this. p. 8 lines 25-29 Physical activity was defined as physical activity during leisure time without any time limit or threshold. But in what time frame (e.g. the last week, the last month, the last year, ever?) p.9 lines 9-12 Why was CRP only available in a subset of the population? p.14 line 30 “an poorly balanced” correct to “a poorly balanced”. p.15 lines 28-34 But in your material, self-reported health does not seem to associate with socioeconomic factors? This should be commented on here. Table 2 “Multivariable models were mutually adjusted other cardiovascular risk factors in addition to age, sex, work, family income, living area (rural/urban) and prior heart disease”. I guess something is lacking here. Please correct. Contributor statement

	Most of the authors are not qualified to be authors according to ICMJE (Vancouver) regulations. Please remove (and possible add to the acknowledgements section)

VERSION 1 – AUTHOR RESPONSE

Reviewer #1

1. I had the great pleasure to review the manuscript entitled "Association between self-rated health, cardiovascular risk factors and echocardiography: A study from the Amazon Basin, Brazil". The manuscript is well designed and organized.

Response:

We thank Reviewer #1 for the kind words and for the insightful comments.

2. Although the study is important, I believe there are significant limitations with this paper. More specifically, the studied population is only from a certain part of the world, which limits generalizability of the findings to the rest of the world. Several aspects of such questionnaires can be affected by cultural factors. I suggest performing a large population-based study from different cultures, if the authors would like to publish it in an international journal. Otherwise, this manuscript is well suited for a local journal.

Response:

Thank you for this important comment. We certainly agree that our findings are from a specific part of the world which may limit their overall generalizability. However, we argue that the presented findings are important for several reasons:

1. First, we examined a sample from the Amazon Basin of Brazil, a region with approximately 30 million inhabitants [1]. Importantly, the Amazon Basin extends into eight other neighboring countries (Peru, Bolivia, Colombia, Venezuela, Guyana, Suriname, French Guyana and Ecuador). While national and cultural differences may exist, these countries have populations who live in the same environment and share the same unique features of the Amazon Basin.

2. The EQ-5D-5L instrument is globally recognized and is designed to mitigate country-specific differences in language and culture, enhancing generalizability to other parts of the world. However, and as noted by Reviewer #1, cultural differences could still have influenced the results.

3. A way to mitigate cultural differences is to establish country specific norms and index values. Unfortunately, such values for rural parts of the Amazon Basin have yet to be established. This study therefore represents an important first step for establishing index values of self-reported health, which will allow for future comparisons with the rest of the world.

4. While cardiovascular disease is one of the most important causes of mortality, there is a paucity of studies investigating this in low- and middle-income settings (please refer to comment #1 from Reviewer #3). This study contributes to fill this gap.

5. The results from this study may be considered hypothesis-generating for future studies, both in the Amazon Basin of Brazil, and in other parts of the world.

Based on the above, we believe that our findings are of significance beyond Brazil and that our results contribute to scientific progress in terms of patient reported health measures and research in low-income settings. We have updated the Strength and Limitations section to emphasize these points.

Strengths and Limitations, page 16, line 3-4:

'Data from this study represents an important first step in establishing EQ-5D-5L index values for the rural parts of the Amazon basin.'

Strengths and Limitations, page 16, line 7-10:

'While the generalizability of our findings to other regions in the world may be disputed, the Amazon Basin covers eight other countries in addition to Brazil. Hence, our findings are likely to be applicable to populations in these areas or to populations who share similar environment and culture.'

3. No information about the second questionnaire has been provided. Was it validated? Who filled the questionnaires? The participant or the interviewer?

Response:

Thank you for commenting on the second questionnaire, which was specifically created for this study. This questionnaire was administered by trained personnel, who filled in all answers based on interviews with the participants. This has now been added to the Methods section.

Methods, page 6, line 9-10:

*'Two different questionnaires were administered by trained interviewers **(i.e., study personnel). These interviewers also filled out the questionnaires.'***

Unfortunately, the second questionnaire has not been validated in other studies or settings prior to commencement of this study. This is now mentioned as a limitation.

Strengths and Limitations, page 15, line 21-22:

'Furthermore, it is a limitation that the questionnaire for health behavior has not been validated in other studies or settings'

4. My other main concern about this study is the fact that there is not much novelty. In fact, this study does not add that much to our current knowledge.

Response:

Thank you for raising this important point. While prior studies have already explored associations between self-rated health and cardiovascular disease, this is the first study to specifically examine this in the Amazon Basin of Brazil.

Furthermore, the authors are not aware of prior studies investigating self-rated health in the general population and (i) the relationship with echocardiographic parameters or (ii) sex differences in this domain with respect to cardiac disease.

We do acknowledge that some of our associations are negative; however, we believe this still complies with the aims and scope of BMJ Open:

All research study types are considered. [...] This includes specialist studies and studies reporting negative results.

Reviewer #2

1. Good work. However, there are some observations.

Response:

We thank Reviewer #2 for acknowledging the value of our work.

2. In this study, the echocardiographic parameters have been termed "Subclinical cardiac disease by echocardiography", but a proportion of the study subjects had LVEF <45%. Were they 'subclinical'?

Response:

Thank you for this observation. For this specific study population, no participants had recognizable symptoms or signs of heart disease during the examination. Consequently, we classified left ventricular ejection fraction <45% as 'subclinical'. We have elaborated on this in the Methods section.

Methods, page 7, line 1-3:

'A medical doctor (PB) evaluated all patients. None of them displayed clinical signs or symptoms of heart disease (absence of shortness of breath, chest pain, swelling of legs and irregular heart rhythm).'

3. In the REFERENCE section, some references need correction and completion, e.g., Ref. 33, 36, 38, 39, as follows

(33) Salomon JA, Tandon A, Murray CJ. Comparability of self rated health: cross sectional multi-country survey using anchoring vignettes. BMJ. 2004 Jan 31;328(7434):258. doi: 10.1136/bmj.37963.691632.44. Epub 2004 Jan 23. PMID: 14742348; PMCID: PMC324453.

(36) May M, Lawlor DA, Brindle P, Patel R, Ebrahim S. Cardiovascular disease risk assessment in older women: can we improve on Framingham? British Women's Heart and Health prospective cohort study. Heart. 2006 Oct;92(10):1396-401. doi: 10.1136/hrt.2005.085381. Epub 2006 Mar 17. PMID: 16547204; PMCID: PMC1861043.

(38) McFadden E, Luben R, Wareham N, Bingham S, Khaw KT. Occupational social class, risk factors and cardiovascular disease incidence in men and women: a prospective study in the European Prospective Investigation of Cancer and Nutrition in Norfolk (EPIC-Norfolk) cohort. Eur J Epidemiol. 2008;23(7):449-58. doi: 10.1007/s10654-008-9262-2. Epub 2008 May 29. PMID: 18509727.

(39) Kaplan GA, Keil JE. Socioeconomic factors and cardiovascular disease: a review of the literature. Circulation. 1993 Oct;88(4 Pt 1):1973-98. doi: 10.1161/01.cir.88.4.1973. PMID: 8403348.

Response:

All references have now been checked for their completeness and updated to conform to the requirements of the journal. Thank you for your help with this.

Reviewer #3

1. Holm et al present a very interesting dataset from the Amazon Basin. Cardiovascular disease is still one of the most important causes of mortality, especially in low- and middle-income countries. Contrasting this, most data are collected in high-resource settings, possibly limiting their generalization to low- and middle-income areas. Thus, studies from such settings

are highly welcome

Response:

We are grateful to Reviewer #3 for acknowledging our effort in conducting an important study from a low-resource setting.

2. The current study examines the associations between self-rated health (SRH) and cardiovascular risk factors (CRF). However, neither the abstract, the introduction, nor the discussion explains clearly why this is interesting to study. In the discussion (p.13, lines 14-23) it is stated that "Considering the close relationship we found with several cardiovascular risk factors, self-rated health could be obtained by non-medical personnel and enable screening of remote communities. Consequently, selected individuals, i.e., persons with low self-rated health and no known cardiovascular risk factors, could be referred for risk factor optimization in healthcare facilities.» But as the outcomes associated with SRH are rather crude (which of course is often the case in low-resource settings), would it not be quite easy to register these in a low-resource setting as well, providing more specific information about cardiovascular risk than the SRH?

Response:

Thank you for raising this crucial point. It is, of course, necessary to properly describe the scientific motivation for conducting this investigation. The abstract, introduction and discussion have been updated. Accordingly, Please see below.

Abstract, page 2, line 1-2:

'Objective: Prior studies have suggested that *self-rated health* ~~*is associated with*~~ ***may be a useful indicator of cardiovascular disease. Consequently,*** *we* ***aimed to*** *assess*~~*ed*~~ *the relationship between self-rated health, cardiovascular risk factors, and subclinical cardiac disease in the Amazon Basin.'*

Introduction, page 4, line 16-18:

'~~*The aim of this study was to assess whether self-rated health is related to*~~ ***To understand whether self-rated health may be used to screen for cardiac disease in low-income settings, we aimed to investigate the relationship with*** *cardiovascular risk factors and disease in the general population from the Amazon Basin of Brazil.'*

Discussion, page 13, line 6-7:

'These findings indicate that in a low-income setting, self-rated health may to some extent provide information on cardiac risk profiles.'

3. Furthermore, I do not think your data support that SRH could be used to detect unknown cardiovascular disease.

Response:

We fully agree with this interpretation. While we found a significant relationship with cardiac risk factors, our results did not show a significant relationship with subclinical cardiovascular disease determined by echocardiography. We believe this is explicitly mentioned in our Conclusion:

Conclusion, page 16, line 13-14:

'*Conversely, self-rated health was not associated with cardiac disease by echocardiography.'*

If the reviewers and editors believe we need to expand on this even further, we would be happy to do so. Thank you.

References

1 Santos D, Salomão R, Veríssimo A. Fatos da Amazônia 2021. *Amaz 2030* 2021;86.https://amazonia2030.org.br/wp-content/uploads/2021/04/AMZ2030-Fatos-da-Amazonia-2021-3.pdf

VERSION 2 – REVIEW

REVIEWER	Islam, AKM Monwarul National Institute of Cardiovascular Diseases, Department of Cardiology
REVIEW RETURNED	04-Apr-2022

GENERAL COMMENTS	I like to convey thanks to Holm et al. for their efforts to deal with the issues raised by the Reviewers and make the manuscript better.

REVIEWER	Moe, Kjartan Baerum Hospital
REVIEW RETURNED	04-Apr-2022

GENERAL COMMENTS	General comments

	I thank Holm et al for clarifications provided in the rebuttal letter to the editors. For some reason they have however only replied to some of my comments. I therefore repeat the comments which were left unanswered in the first review and also use the opportunity to follow-up on some topics. 1. Firstly, the authors still claim in their conclusion that SRH could be used for screening for cardiovascular risk factors. ”Healthcare policies could potentially utilize self-rated health for screening or as a target to improve health behavior.” I do not think this is supported by their design or data because, firstly: SRH was not associated with subclinical CVD (as Holm et al state clearly), but more importantly: the other significant outcomes (hypertension, hypercholesterolemia, diet, and physical activity) could all be defined by the patient herself. Screening is used to detect something, but these risk factors were already known to the person in question. Hence, screening for this risk factor itself is as easy as using the SRH, and provide more specific information about the cardiovascular risk profile, and thus better guidance to preventive measures. Furthermore, I wonder if the SRH may in this case be biased as persons who know that they have adverse levels of cardiovascular risk factor(s) are more prone to have lower SRH? If this is the case, the authors may in fact be exploring whether cardiovascular risk factors could be used to screen for SRH-levels. 2. The inclusion process should be described more thoroughly, preferably with the support of a flow chart. What was the purpose of including a second group of patients (p.7 lines 15-20). Could this affect generalization of your results? Would the results be the same if this group was excluded? Obviously only persons who responded to all questionnaires were included (p.7 line 5), but how many were there, and do you have reasons to believe that these were different from the rest of your population? 3. There are also some concerns and questions to the statistics in the paper: Were multivariate regression analysis conducted by forced entry of all the variables, or did you apply any selection criterion (p. 10 lines 46-55)? Table 1 and supplemental table 1 are lacking information on the statistical methods applied and information about what is presented (mean/median etc.). Obviously, the median should be presented for not normally distributed values. To me, it seems like this is not the case. Please elaborate and specify. I guess a sample size analysis was not conducted as this was a secondary study? Please, specify in your statistics section. Low sample size might explain the lack of any associations between SRH and subclinical cardiac disease. Please comment in your discussion part. Specific comments p. 1st paragraph “Cardiovascular disease is the leading cause of mortality worldwide”. Please find a suitable reference for this. p. 7 2nd paragraph

	Physical activity was defined as physical activity during leisure time without any time limit or threshold. But in what time frame (e.g. the last week, the last month, the last year, ever?) p.8 1st paragraph Why was CRP only available in a subset of the population? p.15 2nd paragraph But in your material, self-reported health does not seem to associate with socioeconomic factors? This should be commented on here. Table 2 “Multivariable models were mutually adjusted other cardiovascular risk factors in addition to age, sex, work, family income, living area (rural/urban) and prior heart disease”. I guess something is lacking here. Please correct. Contributor statement Most of the authors are not qualified to be authors according to ICMJE (Vancouver) regulations. Please remove (and possible add to the acknowledgements section)

VERSION 2 – AUTHOR RESPONSE

Reviewer #2

I like to convey thanks to Holm et al. for their efforts to deal with the issues raised by the Reviewers and make the manuscript better.

Response:

We thank Reviewer #2 for the kind words and for the insightful comments throughout this process. Thank you.

Reviewer #3

1. I thank Holm et al for clarifications provided in the rebuttal letter to the editors. For some reason they have however only replied to some of my comments. I therefore repeat the comments which were left unanswered in the first review and also use the opportunity to follow-up on some topics.

Response:

We thank Reviewer #3 for their insightful and important comments, which have helped improve the quality of the manuscript. We deeply apologize if we had left any comments unanswered, and we are grateful for receiving a second opportunity to respond to these.

2. Firstly, the authors still claim in their conclusion that SRH could be used for screening for cardiovascular risk factors. "Healthcare policies could potentially utilize self-rated health for screening or as a target to improve health behavior." I do not think this is supported by their design or data because, firstly: SRH was not associated with subclinical CVD (as Holm et al state clearly), but more importantly: the other significant outcomes (hypertension, hypercholesterolemia, diet, and physical activity) could all be defined by the patient herself. Screening is used to detect something, but these risk factors were already known to the person in question. Hence, screening for this risk factor itself is as easy as using the SRH, and provide more specific information about the cardiovascular risk profile, and thus better guidance to preventive measures.

Response:

Thank you for this important comment on the applicability of self-rated health as screening tool. We fully agree that this study cannot position self-rated health as an independent instrument for screening. However, we would claim that this was not the purpose of the study.

In this particular study, we aimed to elucidate the relationship between self-rated health and cardiovascular risk factors – and based on our findings, we proposed that self-rated health may *potentially* be used for screening. Implementing self-rated health as a potential future screening instrument would require re-examination of a validation cohort where risk factors were not known to research participants. Consequently, this study represents a first step to conduct such a 'validation' study, and our findings should therefore be regarded as hypothesis generating.

To clearly emphasize this, we have updated our wording throughout the manuscript. We believe, this more clearly reflects the aim of the study.

Abstract, Subsection 'Conclusion', page 2-3:

'Our findings are of hypothesis generating nature and future studies should aim to determine whether assessment of self-rated health may be useful for health screening related to policymaking or lifestyle interventions. ~~*Assessment of self-rated health could be useful for screening or as a target in healthcare policies for lifestyle interventions.'*~~

Introduction, page 4:

'To understand whether self-rated health ***in future studies*** *may be used to screen for cardiac disease in low-income settings, we aimed to investigate the relationship with cardiovascular risk factors and disease in the general population from the Amazon Basin of Brazil.'*

Conclusion, page 16-17:

'On a hypothesis-generating basis, *healthcare policies could potentially utilize self-rated health for screening or as a target to improve health behavior.* ***Nevertheless, this should be investigated in future validation studies****'*

As stated in the Discussion section, we perceive that the most likely reason as to why we did not find a relationship with subclinical cardiovascular disease is that this was a relatively healthy and young population (mean age 41 years). Moreover, the sample size was limited.

3. Furthermore, I wonder if the SRH may in this case be biased as persons who know that they have adverse levels of cardiovascular risk factor(s) are more prone to have lower SRH? If this is the case, the authors may in fact be exploring whether cardiovascular risk factors could be used to screen for SRH-levels.

Response:

Thank you for this question. We have several considerations with respect to this.

(i) According to the sequence of our questionnaires, participants were first asked to provide information about their self-rated health. Following this, we asked about cardiovascular risk factors. We believe this sequence may have contributed to lower the risk of bias pointed out by Reviewer #3.
(ii) As this was a secondary study, participants were informed about the overall purpose of the study, but were not directly informed about the secondary hypothesis, i.e., the assessment of self-rated health and cardiovascular risk factors.
(iii) We had a clearly defined hypothesis prior to commencing data analysis, thus minimizing the risk of establishing relationships by chance.
(iv) We agree that there may be a risk of reverse causation when assessing self-rated health and cardiovascular risk factors. This is an overall risk which applies to all studies addressing this topic. This has already been commented on in our Strengths and Limitations section (page 15-16): '*We adjusted our models for cardiac disease at baseline in an attempt to limit reverse causation'.*

We have incorporated more considerations on this in the Strengths and Limitations section, page 16:

'To reduce bias, we had a clear and predefined hypothesis prior to commencing data analyses and a rigorous design for the sequence of questionnaires.'

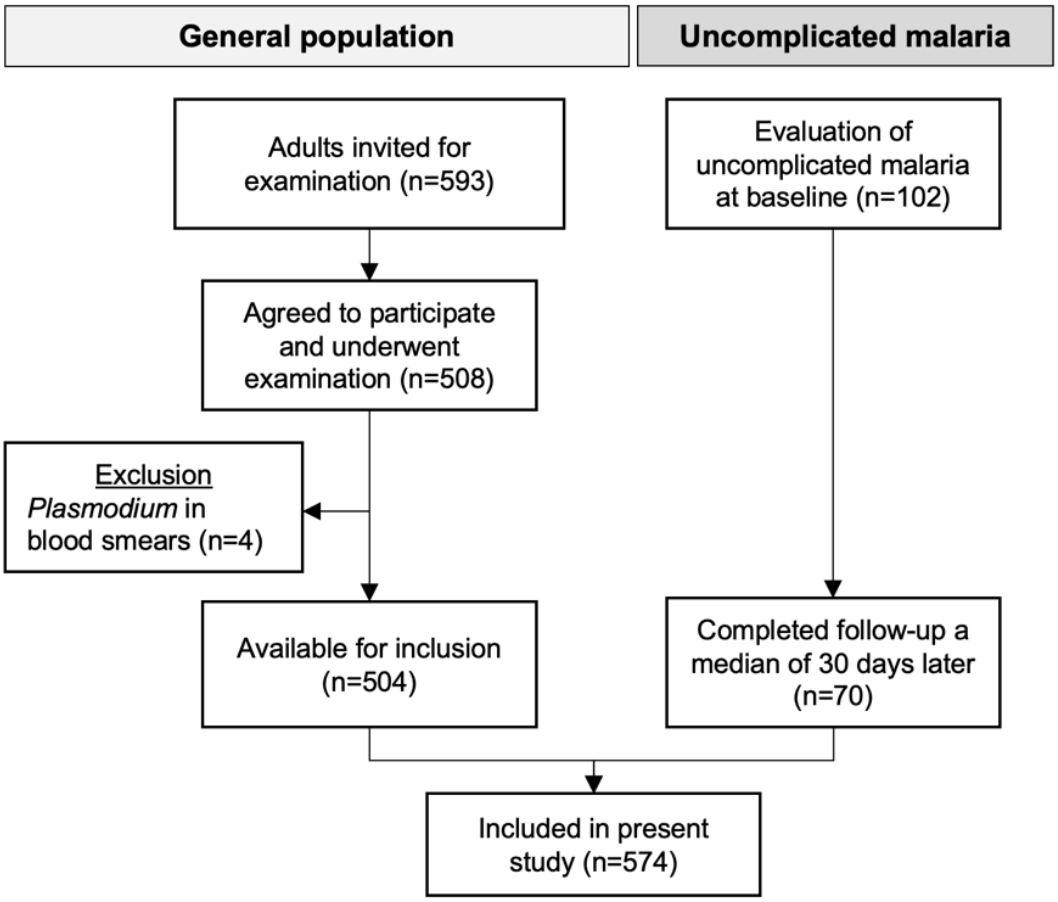
4. The inclusion process should be described more thoroughly, preferably with the support of a flow chart.

Response:

Thank you for this observation. We have added a flowchart in a novel Figure 1 (see below) and contributed more details related to the second group of participants who were included (recently treated for uncomplicated malaria).

Figure 1.

Figure 1.



Methods, subsection 'Sampling', page 5:

'Local healthcare agents provided lists of persons associated with each clinic, who we invited to participate in the study ***(Figure 1).'***

Methods, subsection 'Sampling', page 6:

'A total of 504 participants ***from the general population*** *were included from healthcare clinics.* ***As a part of the main study, we also examined patients diagnosed with uncomplicated malaria in healthcare clinics. This group of participants underwent a follow-up examination a median of 30 days later, when they had completed treatment and had no symptoms of malaria. According***

to the above-mentioned inclusion and exclusion criteria, a total of 70 participants from this group were eligible for inclusion (Figure 1). ~~*A second group of 70 participants from the general population who had recently completed anti-malarial treatment and for whom the above-mentioned inclusion and exclusion criteria also applied, were included as well*~~*'.*

5. What was the purpose of including a second group of patients (p.7 lines 15-20). Could this affect generalization of your results?

Response:

Thank you for asking. The purpose of including the second group was to increase the number of participants and thereby achieve greater statistical power. As this group of participants had already completed anti-malarial treatment, were free from malaria symptoms, and came from the same population as the main study group, we have no reason to believe that this could have affected the generalizability of the study.

This has been added to the Strengths and Limitations section, page 16:

'To increase the sample size, we included a subgroup of participants recently treated for malaria (n=70). As this group was derived from the same population, had no symptoms of malaria, and all associations remained unchanged when excluded, we do not believe its inclusion affects the generalizability of our results.'

6. Would the results be the same if this group was excluded?

Response:

In a sensitivity analysis, we excluded this group from all analyses and found that all positive associations remained significant. Please refer to the new Supplemental Table 2 and Supplemental Figure 1 (see below).

Supplemental Table 2.
Association between self-rated health (per 10-point increase), cardiovascular risk factors and disease in the study population excluding recently treated malaria patients (n=504).

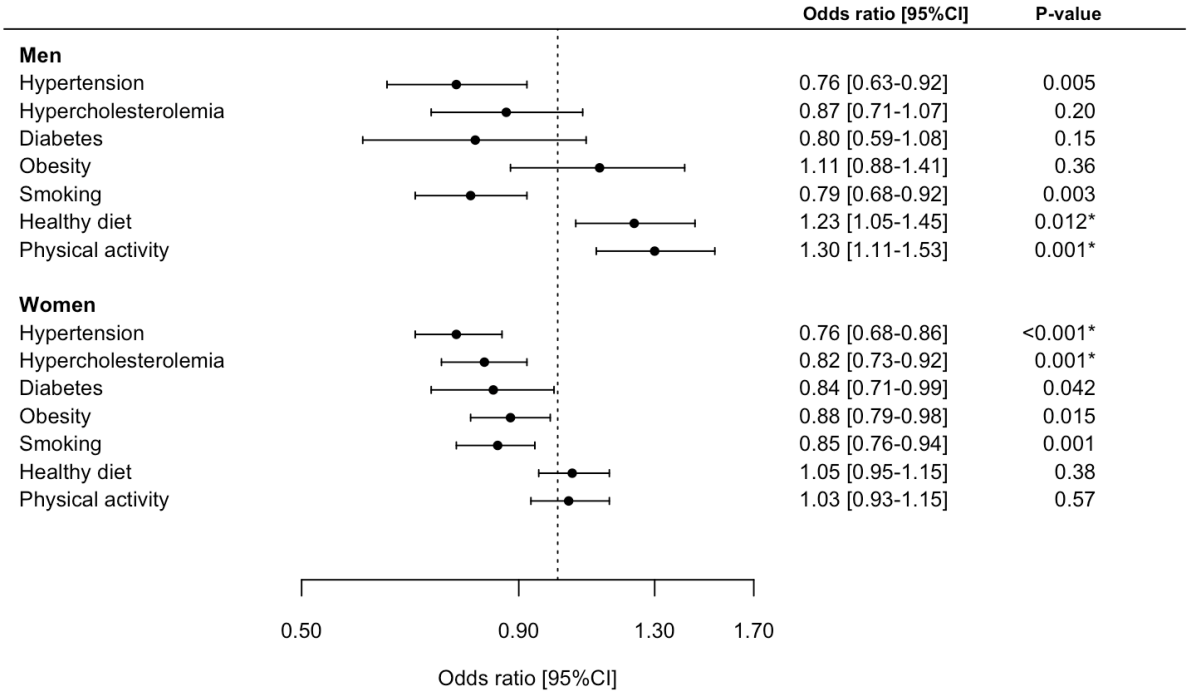
	Unadjusted odds ratio [95%CI]	*P*	*Adjusted odds ratio [95%CI]**	*P*	*P interaction sex*

Risk factors					
Hypertension	*0.76 [0.69 to 0.84]*	*<0.001*	*0.86 [0.77 to 0.96]*	*0.007*	*0.005*
Hypercholesterolemia	*0.83 [0.75 to 0.92]*	*<0.001*	*0.90 [0.80 to 1.00]*	*0.05*	*0.20*
Diabetes	*0.82 [0.71 to 0.95]*	*0.009*	*0.98 [0.83 to 1.17]*	*0.86*	*0.15*
Obesity	*0.91 [0.83 to 0.99]*	*0.036*	*0.96 [0.87 to 1.06]*	*0.40*	*0.36*
Smoking	*0.83 [0.76 to 0.91]*	*<0.001*	*0.93 [0.84 to 1.03]*	*0.16*	*0.003*
Heathy diet	*1.19 [1.00 to 1.18]*	*0.047*	*1.09 [1.00 to 1.19]*	*0.049*	*0.012*
Physical activity	*1.12 [1.03 to 1.22]*	*0.010*	*1.06 [0.96 to 1.17]*	*0.22*	*0.001*
Subclinical cardiac disease					
LV ejection fraction <45%	*0.92 [0.73 to 1.16]*	*0.49*	*1.02 [0.79 to 1.31]*	*0.88*	*0.91*
LV hypertrophy	*0.92 [0.74 to 1.15]*	*0.47*	*1.10 [0.84 to 1.44]*	*0.48*	*0.33*
Diastolic dysfunction	*0.85 [0.68 to 1.08]*	*0.18*	*1.05 [0.77 to 1.43]*	*0.77*	*0.23*

**Multivariable models were mutually adjusted for cardiovascular risk factors (hypertension, hypercholesterolemia, diabetes, obesity, smoking, healthy diet, physical activity) in addition to age, sex, work, family income, living area (rural/urban) and prior heart disease*
LV: left ventricular

Supplemental Figure 1. *Forest plot*

*Association between self-rated health (per 10-point increase) and cardiovascular risk factors stratified by sex. *indicates that the association remained significant in multivariable models.*



We have updated the manuscript accordingly.

Results section, subsection ‘Cardiovascular risk factors’, page 11:

‘The associations remained unchanged when we excluded participants recently treated for malaria (Supplemental Table 2 and Supplemental Figure 1).’

Results section, subsection ‘Interactions with sex’, page 12:

‘The associations remained unchanged when we excluded participants from the malaria group (Supplemental Table 2 and Supplemental Figure 1).’

7. Obviously only persons who responded to all questionnaires were included (p.7 line 5), but how many were there, and do you have reasons to believe that these were different from the rest of your population?

Response:

Thank you for this question. Unfortunately, we are unsure whether this question refers to the group of participants recently treated for malaria or people from the general population who were invited and did not participate.

According to Response 6 to Reviewer #3, all associations remained unchanged when patients recently treated for malaria were excluded. The overall participation rate from the population was high (85%), and we therefore believe that the included sample is representative of the general population in this region of the Amazon Basin.

8. There are also some concerns and questions to the statistics in the paper:
Were multivariate regression analysis conducted by forced entry of all the variables, or did you apply any selection criterion (p. 10 lines 46-55)?

Response:

Thank you for an important consideration on our multivariable models. All variables in the model were carefully selected prior to commencing data analyses. The selection was based on prior studies assessing associations between self-rated health, clinical data [1], socioeconomic data [2], and chronic illness [3].

Our Statistics section on page 10 has been updated with this information:

'Included variables were selected based on prior studies of self-rated health [1–3] and were defined prior to commencing data analyses.'

9. Table 1 and supplemental table 1 are lacking information on the statistical methods applied and information about what is presented (mean/median etc.). Obviously, the median should be presented for not normally distributed values. To me, it seems like this is not the case. Please elaborate and specify.

Response:

Thank you for making us aware of this. We have meticulously examined all variables presented in Table 1 and Supplemental Table 1. We identified that C-reactive protein and bilirubin were non-normally distributed, and that creatinine was normally distributed. Accordingly, we have updated the tables such that they are properly presented. Furthermore, we have provided details as to how we calculated P-values for trend/difference, and how normally distributed and non-normally distributed variables are presented. Please refer to updated versions of the tables below, where novel information is marked in red.

Table 1. *Baseline clinical characteristics by tertiles of self-rated health*

	Tertiles of self-rated health			
	1st tertile (n=231)	*2nd tertile (n=226)*	*3rd tertile (n=117)*	*P trend**
	0 to 70	*71 to 90*	*91 to 100*	
Baseline				
Age, years	*46 ± 16*	*38 ± 13*	*39 ± 15*	*<0.001*
Female, %	*154 (67%)*	*127 (56%)*	*69 (59%)*	*0.06*
Self-reported race, %				*0.51*
White	*33 (14%)*	*24 (11%)*	*20 (17%)*	
Mixed	*163 (71%)*	*175 (77%)*	*77 (66%)*	
Black	*32 (14%)*	*26 (12%)*	*18 (15%)*	
Indigenous	*2 (1%)*	*1 (<1%)*	*1 (1%)*	
BMI, kg/m²	*28 ± 6*	*27 ± 5*	*26 ± 4*	*0.002*
Abdominal circumference, cm	*90 ± 14*	*87 ± 12*	*84 ± 11*	*<0.001*
Asthma	*11 (5%)*	*8 (4%)*	*2 (2%)*	*0.36*
COPD, %	*3 (1%)*	*3 (1%)*	*1 (1%)*	*0.92*
History of MI, %	*2 (1%)*	*2 (1%)*	*1 (1%)*	*1.00*
Heart failure, %	*3 (1%)*	*2 (1%)*	*0 (0%)*	*0.47*
Rheumatic heart disease, %	*7 (3%)*	*7 (3%)*	*4 (3%)*	*0.97*
SBP, mmHg	*134 ± 20*	*131 ± 20*	*131 ± 19*	*0.29*
DBP, mmHg	*83 ± 12*	*81 ± 11*	*82 ± 12*	*0.17*
Risk factors				
Hypertension, %	*66 (29%)*	*32 (14%)*	*14 (12%)*	*<0.001*
Hypercholesterolemia, %	*52 (23%)*	*23 (10%)*	*14 (12%)*	*<0.001*
Diabetes, %	*21 (9%)*	*6 (3%)*	*6 (5%)*	*0.012*
Obesity, %	*68 (29%)*	*45 (20%)*	*20 (17%)*	*0.012*
Smoking, %	*106 (46%)*	*65 (29%)*	*46 (39%)*	*<0.001*
Healthy diet, %	*87 (38%)*	*130 (58%)*	*59 (50%)*	*<0.001*
Physical activity, %	*64 (28%)*	*94 (42%)*	*53 (45%)*	*<0.001*

Socioeconomic status				
Work status, %				*0.09*
Employed	*77 (33%)*	*98 (43%)*	*53 (45%)*	
Self-employed	*20 (9%)*	*23 (10%)*	*9 (8%)*	
Other	*134 (58%)*	*105 (47%)*	*55 (47%)*	
Family income, BRL	*1250 [800, 2000]*	*1500 [1000, 3000]*	*1200 [800, 2000]*	*0.11*
Rural living area, %	*92 (40%)*	*78 (35%)*	*55 (47%)*	*0.08*
Biochemistry				
Blood sugar, mg/dL	*110 ± 74*	*100 ± 27*	*110 ± 49*	*0.10*
Bilirubin, mg/dL	*0.3 [0.2, 0.5]*	*0.4 [0.2, 0.5]*	*0.4 [0.2, 0.5]*	*0.55*
Platelets, mm^3	*229± 76*	*240 ± 67*	*234 ± 66*	*0.28*
Leukocytes, mm^3	*6349 ± 1991*	*6383 ± 1723*	*6532 ± 1915*	*0.68*
Reticulocytes, %	*0.75 ± 0.19*	*0.80 ± 0.22*	*0.77 ± 0.22*	*0.44*
Hemoglobin, g/dL	*14 ± 1*	*14 ± 1*	*14 ± 1*	*0.13*
C-reactive protein, mg/L	*0 [0, 0]*	*0 [0, 0]*	*0 [0, 0]*	*0.44*
Creatinine, mg/dL	*0.9 ± 0.3*	*0.9 ± 0.2*	*0.9 ± 0.2*	*0.59*
INR	*1.02 ± 0.12*	*1.01 ± 0.10*	*1.02 ± 0.10*	*0.30*
Echocardiography				
LV ejection fraction<45%, %	*9 (4%)*	*6 (3%)*	*3 (3%)*	*0.69*
LV hypertrophy, %	*9 (4%)*	*4 (2%)*	*4 (3%)*	*0.39*
Diastolic dysfunction, %	*7 (3%)*	*5 (2%)*	*1 (1%)*	*0.43*
LV ejection fraction, %	*57 ± 6*	*57 ± 5*	*58 ± 5*	*0.48*
LV mass index, g/m^2	*71 ± 18*	*68 ± 17*	*70 ± 16*	*0.11*
E/e'	*7.3 ± 2.6*	*6.7 ± 2.1*	*6.9 ± 2.3*	*0.014*
E/A	*1.2 ± 0.5*	*1.3 ± 0.4*	*1.3 ± 0.4*	*0.003*
Left atrial volume index, mL/m^2	*20 ± 6*	*19 ± 5*	*19 ± 4*	*0.025*
TR velocity, m/s	*2.3 ± 0.3*	*2.3 ± 0.3*	*2.3 ± 0.2*	*0.34*

COPD: chronic obstructive pulmonary disease, SBP: systolic blood pressure, DBP: diastolic blood pressure, BMI: body mass index, INR: international normalized ratio, LV: left ventricular, TR: tricuspid regurgitation
**P for trend was calculated using linear regression models for normally distributed variables and Cuzick's nonparametric test for trend for non-normally distributed variables.*
Normally distributed variables are displayed as mean ± standard deviation.
Non-normally distributed variables are presented as median [interquartile range].
Proportions are displayed as n (%).

Supplemental Table 1. *Baseline clinical characteristics by sex*

	Men *n=224*	*Women* *n=350*	*P difference**
Baseline			
Age, years	*40 ± 15*	*42 ± 15*	*0.28*
Race, %			*0.41*
White	*33 (15%)*	*44 (13%)*	
Mixed	*153 (68%)*	*262 (75%)*	
Black	*36 (16%)*	*40 (11%)*	
Indigenous	*1 (<1%)*	*3 (1%)*	
BMI, kg/m^2	*26 ± 4*	*28 ± 5*	*0.001*
Abdominal circumference, cm	*87 ± 13*	*88 ± 13*	*0.45*
Asthma	*4 (2%)*	*17 (4%)*	*0.06*
COPD, %	*2 (1%)*	*5 (1%)*	*0.57*
History of MI, %	*2 (1 %)*	*3 (1%)*	*0.96*
Heart failure, %	*3 (1%)*	*2 (1%)*	*0.33*
Rheumatic heart disease, %	*8 (4%)*	*10 (3%)*	*0.18*
SBP, mmHg	*133 ± 16*	*131 ± 22*	*0.18*
DBP, mmHg	*82 ± 12*	*82 ± 12*	*0.73*
Risk factors			
Hypertension, %	*37 (17%)*	*75 (21%)*	*0.15*
Hypercholesterolemia, %	*26 (12%)*	*63 (18%)*	*0.039*
Diabetes, %	*9 (4%)*	*24 (7%)*	*0.15*
Obesity, %	*31 (14%)*	*102 (29%)*	*0.001*
Smoking, %	*98 (44%)*	*119 (34%)*	*0.019*
Healthy diet, %	*99 (44%)*	*177 (51%)*	*0.14*
Physical activity, %	*107 (48%)*	*104 (30%)*	*0.001*

Socioeconomic status			
Work status, %			*0.001*
Employed	*70 (31%)*	*224 (64%)*	
Self-employed	*125 (56%)*	*103 (29%)*	
Other	*29 (13%)*	*23 (7%)*	
Family income, BRL	*1700 [1000, 2750]*	*1200 [800, 2000]*	*0.001*
Rural living area, %	*99 (44%)*	*126 (36%)*	*0.050*
Biochemistry			
Blood sugar, mg/dL	*100 ± 24*	*110 ± 67*	*0.047*
Bilirubin, mg/dL	*0.4 [0.3, 0.6]*	*0.3 [0.2, 0.4]*	*<0.001*
Platelets, mm^3	*220 ± 81*	*244 ± 62*	*0.001*
Leukocytes, mm^3	*6040 ± 1590*	*6632 ± 2002*	*0.001*
Reticulocytes, %	*0.75 ± 0.19*	*0.80 ± 0.22*	*0.44*
Hemoglobin, g/dL	*15 ± 1*	*13 ± 1*	*0.001*
C-reactive protein, mg/L	*0 [0, 0]*	*0 [0, 0]*	*0.59*
Creatinine, mg/dL	*1.0 ± 0.3*	*0.8 ± 0.2*	*<0.001*
INR	*1.03 ± 0.09*	*1.00 ± 0.11*	*0.001*
Echocardiography			
LV ejection fraction<45%, %	*11 (4.9%)*	*7 (2.0%)*	*0.05*
LV hypertrophy, %	*6 (2.7%)*	*11 (3.1%)*	*0.75*
Diastolic dysfunction, %	*3 (1.3%)*	*13 (3.7%)*	*0.09*

COPD: chronic obstructive pulmonary disease, SBP: systolic blood pressure, DBP: diastolic blood pressure, BMI: body mass index, INR: international normalized ratio, LV: left ventricular

**P difference was calculated using the chi-square test, Student's t-test, and the Wilcoxon rank-sum test.*
Normally distributed variables are displayed as mean ± standard deviation.
Non-normally distributed variables are presented as median [interquartile range].
Proportions are displayed as n (%).

10. I guess a sample size analysis was not conducted as this was a secondary study? Please, specify in your statistics section. Low sample size might explain the lack of any associations between SRH and subclinical cardiac disease. Please comment in your discussion part.

Response:

We have now clearly indicated in the Statistics section (page 10) that no sample size calculation was conducted:

'As this was a secondary study, no sample size calculation was conducted.'

In addition, we now mention this consideration in the Discussion section (page 13), when commenting on the lack of association with echocardiographic parameters:

'Another potential reason could be low statistical power due to the limited size of the study population.'

11. p. 1st paragraph

"Cardiovascular disease is the leading cause of mortality worldwide". Please find a suitable reference for this.

Response:

Thank you for commenting on this. We have now replaced the previous reference with the following:

> Roth, G.A.; Mensah, G.A.; Johnson, C.O.; Addolorato, G.; Ammirati, E.; Baddour, L.M.; Barengo, N.C.; Beaton, A.Z.; Benjamin, E.J.; Benziger C.P.; et al. Global Burden of Cardiovascular Diseases and Risk Factors, 1990–2019: Update From the GBD 2019 Study. *J Am Coll Cardiol* **2020**, 76, 2982–3021, doi:10.1016/J.JACC.2020.11.010

We believe that this is a more suitable reference covering global mortality related to cardiovascular disease.

12. p. 7 2nd paragraph
Physical activity was defined as physical activity during leisure time without any time limit or

threshold. But in what time frame (e.g. the last week, the last month, the last year, ever?)

Response:

Thank you for pointing this out. We asked study participants whether they participated in physical activity on a weekly basis. We have now updated the Methods section (page 7) with this information:

'Physical activity was defined as participation in any kind of physical activity, ***on a weekly basis,*** *during leisure time. We did not apply any time limit or threshold.'*

13. p.8 1st paragraph
Why was CRP only available in a subset of the population?

Response:

Thank you for this question. As describe, C-reactive protein was only available in 436 study participants. The lack of measurements was due to a failure in the supply chain, not permitting delivery of analytical kits to our laboratory in this region of the Amazon Basin. The main reason behind this was the corona virus pandemic in Brazil 2020-2021, during which flights to this part of the Amazon were prohibited from operating.

14. p.15 2nd paragraph
But in your material, self-reported health does not seem to associate with socioeconomic factors? This should be commented on here.

Response:

Thank you for commenting on our Strengths and Limitations section, where we point out that socioeconomic status, generally, is perceived to be associated with cardiac risk factors.

We agree that according to our Table 1, socioeconomic characteristics such as work status, family income, and living in rural areas, did not vary significantly according to the P for trend test. We do not believe this is adequate to completely rule out a potential association between socioeconomic status and self-reported health. However, we have updated this section with a brief comment.

Strengths and Limitations section, page 15:

'Socioeconomic status is ***perceived to be*** *associated with self-rated health and cardiovascular risk factors* [4,5]*, and despite our multivariable adjustment, residual confounding may still exist.* ***Interestingly, parameters of socioeconomic status did not vary significantly across tertiles of self-reported health (Table 1), indicating that this relationship may differ in this region.'***

15. Table 2
"Multivariable models were mutually adjusted other cardiovascular risk factors in addition to age, sex, work, family income, living area (rural/urban) and prior heart disease". I guess something is lacking here. Please correct.

Response:

We have now updated this description such that it includes all variables in the model. We have updated the description in both Table 2 and Table 3. Please see below:

'Multivariable models were mutually adjusted for other cardiovascular risk factors ***(hypertension, hypercholesterolemia, diabetes, obesity, smoking, healthy diet, physical activity)*** *in addition to age, sex,* ***race****, work, family income, living area (rural/urban) and* ~~*prior*~~ ***known*** *heart disease* ***(prior myocardial infarction, heart failure, rheumatic heart disease).'***

16. Contributor statement
Most of the authors are not qualified to be authors according to ICMJE (Vancouver) regulations. Please remove (and possible add to the acknowledgements section)

Response:
Thank you for this observation. Accordingly, we have updated the author contribution statement such that this rightfully reflects the contributions from all co-authors. Indeed, all authors have provided substantial contributions in the four domains of the ICMJE criteria. Please refer to an updated list below (page 18-19):

'AEH: Conception of study, planning and design, funding, data acquisition in the field and laboratory, data analysis, statistics, writing, critical review.

LCG: ***Substantial contribution to*** *data acquisition primarily in the laboratory, planning****, critical review***

LOM: ***Substantial contribution to*** *data acquisition****, critical review***

AW: ***Substantial contribution to*** *data acquisition, data analysis****, critical review***

KOL: ***Substantial contribution to*** *data acquisition****, critical review***

MDK: ***Substantial contribution to*** *data acquisition, data analysis****, critical review***

MP: ***Substantial contribution to*** *interpretation of data****, critical review***

IVMV: ***Substantial contribution to*** *data acquisition****, critical review***

RMS: ***Substantial contribution to*** *interpretation of data****, critical review***

CRF: ***Substantial contribution to*** *interpretation of data****, critical review***

TBS: ***Substantial contribution to*** *conception of study****, critical review***

*OMS: **Substantial contribution to** conception of study**, critical review***

PB (responsible for the overall content as guarantor): Conception of study, planning and design, funding, data acquisition in the field and laboratory, data analysis, statistics, writing, critical review.'

In addition to these contributions, all authors have provided final approval of the version to be published and agree to be held accountable for all aspects of the work. Based on this, we believe that all of the above listed members of the research group qualify for co-authorship according to the ICMJE criteria

References

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3 Dong W, Wan J, Xu Y, *et al.* Determinants of self-rated health among Shanghai elders: A cross-sectional study. *BMC Public Health* 2017;**17**:1–12. doi:10.1186/S12889-017-4718-5/TABLES/5

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5 Kaplan GA, Keil JE. Socioeconomic factors and cardiovascular disease: a review of the literature. *Circulation* 1993;**88**:1973–98. doi:10.1161/01.CIR.88.4.1973

VERSION 3 – REVIEW

REVIEWER	Moe, Kjartan Baerum Hospital
REVIEW RETURNED	22-Jun-2022

GENERAL COMMENTS	Thank you for your comprehensive and thorough review of the manuscript. The paper is in my opinion well-written and conclusions are soundly justified by your data. A couple of grammar remarks remain which you can probably correct in the proof if the manuscript is accepted by the Editor(s) Abstract - Setting «We performed questionnaires, physical examination, and echocardiography. Logistic and linear regression models were applied to assess self-rated health, cardiac risk factors and cardiac disease by echocardiography."

	There is inconsistent use of “Oxford comma” in these two sentences. As far as I have observed, you have otherwise applied throughout the rest of the paper. Discussion 1st Paragraph “, we found that self-rated health was significantly associated with cardiovascular risk factors and that these association were modified by sex.” Plural: "associations".